International Journal of Pest Management
Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/ttpm20

Development of nanoalumina dust as insecticide against Sitophilus oryzae and Rhyzopertha dominica

M. Buteler[ab], S.W. Sofie[c], D.K. Weaver[b], D. Driscoll[c], J. Muretta[c] & T. Stadler[d]

[a] Instituto de Investigaciones en Biodiversidad y Medio Ambiente (INIBIOMA) CCT Comahue CONICET, Bariloche Rio Negro, Argentina

[b] Department of Land Resources and Environmental Sciences, Montana State University, Bozeman, MT, USA

[c] Department of Mechanical and Industrial Engineering, Montana State University, MT, USA

[d] Instituto de Medicina yBiologia Experimental de Cuyo (IMBECU) CCT Mendoza CONICET, Mendoza, Argentina

Published online: 03 Feb 2015.

To cite this article: M. Buteler, S.W. Sofie, D.K. Weaver, D. Driscoll, J. Muretta & T. Stadler (2015) Development of nanoalumina dust as insecticide against Sitophilus oryzae and Rhyzopertha dominica, International Journal of Pest Management, 61:1, 80-89, DOI: 10.1080/09670874.2014.1001008

To link to this article: http://dx.doi.org/10.1080/09670874.2014.1001008

International Journal of Pest Management, 2015
Vol. 61, No. 1, 80–89, http://dx.doi.org/10.1080/09670874.2014.1001008

Development of nanoalumina dust as insecticide against *Sitophilus oryzae* and *Rhyzopertha dominica*

M. Buteler[a,b]*, S.W. Sofie[c], D.K. Weaver[b], D. Driscoll[c], J. Muretta[c] and T. Stadler[d]

[a]*Instituto de Investigaciones en Biodiversidad y Medio Ambiente (INIBIOMA) CCT Comahue CONICET, Bariloche Rio Negro, Argentina;* [b]*Department of Land Resources and Environmental Sciences, Montana State University, Bozeman, MT, USA;* [c]*Department of Mechanical and Industrial Engineering, Montana State University, MT, USA;* [d]*Instituto de Medicina yBiología Experimental de Cuyo (IMBECU) CCT Mendoza CONICET, Mendoza, Argentina*

(*Received 28 September 2014; final version received 15 December 2014*)

This study aims to characterize and improve the insecticidal activity of nanostructured alumina dusts. To accomplish these goals, multiple solution based synthesis routes utilizing standard aluminum salt precursors were utilized to synthesize three unique types of alumina dust. These were compared with regards to morphology, particle size and surface area using electron microscopy and dynamic light scattering particle size analysis. Insect toxicity of the various dusts was assessed using two insect species that are pests of stored grain, *Sitophilus oryzae* and *Rhyzopertha dominica*. The dust synthesized using a modified glycine-nitrate combustion process consistently yielded greater mortality rates, and all dust types were more effective on *S. oryzae* than on *R. dominica*, although the difference varied across dust types. The data show that insecticidal activity is dependent on particle size, particle morphology and surface area but also indicated that minimizing particle size and maximizing surface area are not the sole dominant factors influencing efficacy. This study does however suggest that pesticide dusts can be engineered through modified synthesis to better target different insect species.

Keywords: aluminum oxide; surface area; particle size; stored product pest; nanopesticide

1. Introduction

Nanotechnology applications have largely consisted of technology advancements in the biomedical and electronic fields, although numerous research efforts focus on their applications in a wide range of industries. Recently, nanoparticles have shown promise in different fields of agricultural biotechnology (Barik et al. 2008; Rahman et al. 2009). Nanocarriers are being designed to reduce the volume of application and slow down the release kinetics of agrochemicals (Perez de Luque & Rubiales 2009). Nanoparticulate dusts could also be used to enhance the efficiency of traditional pesticides by improving the coverage and stability of pesticide formulations (Liu et al. 2008; Werdin González et al. 2014).

Several studies have demonstrated the potential of some nanomaterials as insecticides, such as: silver nanoparticles (Ki et al. 2007; Arjunan et al. 2012; Marimuthu et al. 2011), nanosilica (Debnath et al. 2011; Barik et al. 2012) and nanostructured alumina (NSA)(Stadler et al. 2010b, 2012).

Nanoparticles, as well as other inert dusts, may provide an alternative strategy to traditional broad spectrum insecticides, to manage pests which have become resistant to conventional pesticides in integrated pest management programs (Korunic 1999).There is no record of resistant insect populations to inert dusts, and given their mode of action is based on physical phenomena, development of resistance to dusts has been considered unlikely in storage insects (Golob 1997). However, newer studies suggest that development of tolerance may be possible through adaptations in the cuticle hydrocarbon profile (Korunic & Ormesher 1999; Riguax et al. 2001; Shah & Khan 2014). Thus, to prevent development of resistance and to use inert dusts in the framework of integrated management programs, inert dusts could be used in combination with other management tools such as entomopathogenic fungi (Lord 2001; Kavallieratos et al. 2006; Michalaki et al. 2006), insecticides (Athanassiou et al. 2004; Athanassiou & Kavallieratos 2005; Athanassiou et al. 2008), or natural enemies (Palyvos et al. 2006).

The most widely accepted explanation for the mode of action of inert dusts is that they kill arthropods by removing or absorbing the epicuticular lipid layers by capillarity causing excessive water loss through the cuticle (Ebeling 1971; Mewis and Ulrich 2001; Stadler et al. 2010a). Therefore, their efficacy is hypothesized to be strongly related to their specific surface area (in units of square meters per gram) as well as particle size. Activity of these dusts is assumed to be based on the capillary draw of epicuticular waxes and this is supported by the fact that their performance is reduced as ambient humidity increases and temperature decreases. Thus, adsorbed water can influence their viscosity and therefore the NSA insecticidal efficacy (Ebeling 1971; Arthur 2000, 2001; Fields & Korunic 2000).

In the case of NSA, its efficacy is comparable, and in some cases greater than that of commercial insecticidal dusts such as enhanced diatomaceous earth like Protect-It

*Corresponding author. Email: lpe@mendoza-conicet.gob.ar

(Stadler et al. 2012). The action mechanism of NSA has not yet been fully elucidated but it is thought to act similarly to inert dusts like kaolinite, silica gel and diatomaceous earth. Previous studies have shown that humidity affects the efficacy of NSA in a similar way as it does on inert dusts (Stadler et al. 2010b, 2012). This study aims to further improve the insecticidal activity of NSA while generating new insight into the relationship between processing variables and the type of dusts generated. For this purpose, we tested various synthesis methodologies that yield different particle morphologies and hence, different surface areas and particle sizes. The synthesis process focused on a standard aluminum nitrate precursor through different processing routes to develop three unique types of dust. Insecticidal aluminum oxide dusts were synthesized by chemical solution routes and characterized by scanning electron microscopy (SEM), particles size analysis and nitrogen-absorption-based surface area analysis. The insecticidal effect of these dusts was assessed at different concentrations, relative ambient humidity level and using two insect species that are pests of stored grain, *Sitophilus oryzae* (Coleoptera: Curculionidae) and *Rhyzopertha dominica* (Coleoptera: Bostrichidae). These two insect species were chosen for the experiments given that they vary greatly in their susceptibility to inert dusts. Chemical makeup of epicuticular waxes varies across insect species (Nawrot et al. 1994), and this should translate into differences in susceptibility to nanoalumina and inert dusts due to differences in wetting. According to Subramanyam and Roesli (2000), *S. oryzae* is among the most susceptible species to diatomaceous earth and *R. dominica* is among the least susceptible ones. The three types of dusts were also compared to a commercially available nanoalumina powder and a diatomaceous earth (silica) based insecticide dust.

2. Materials and methods

2.1 Synthesis of nanostructured dusts

Three methods were utilized to obtain NSA for comparison with the commercial dust. All methods were based on chemical solution methods utilizing a common metal salt precursor. The commercial dust was purchased from Advanced Materials (Catalog # 26N-0802A) as a baseline aluminum oxide material. The first nanodust referred to as "combustion" dust, which was synthesized using the glycine nitrate combustion process. Aluminum nitrate nonahydrate (Al(NO3)3 ·9H2O) in a saturated water solution was used as an oxidizer combined with glycine as the fuel in a glycine/nitrate ratio of 0.45. The solid products of combustion were calcined for 1 hour at 1000 °C to decompose any remaining organics prior to characterization and toxicity assessment.

The second process yielded what is referred to as the "polymeric precursor" dust, and employed a chelation process in which the aluminum ions are bound in a polymeric network thus allowing decomposition into a fine inorganic dust after thermolysis (Buyukaksoy et al. 2012a, 2012b). As before, aluminum nitrate nonahydrate was saturated in an aqueous solution. The solution was added to ethylene glycol in a molar ratio of 0.4 Al+3/ethylene glycol and heated to evaporate the water. As the water was evaporated, the aluminum and nitrate ions were incorporated into the polymer matrix. The resulting gel was heated to 400 °C allowing the nitrates to slowly oxidize the ethylene glycol. The product was calcined at 1000 °C for 1 hour to decompose residual organics.

The final technique to synthesize nanostructured aluminum oxide was derived via a co-precipitation technique, and is therefore referred as the "co-precipitation" dust. This process began with a reservoir of commercial buffer solution with pH set at 8.1. Saturated aluminum nitrate solution was then titrated into the buffer forming insoluble aluminum oxy-hydroxide which precipitated. Concentrated ammonium hydroxide was simultaneously titrated into the solution to maintain a pH between 8.1 and 8.2 as monitored by a digital pH meter. After the precipitate was washed and the supernatant poured-off, the gel was dried and then calcined at 1000 °C for 1 hour.

Following calcination, all dusts were ball milled for 24 hours in an aqueous suspension with yttria-stabilized zirconia grinding media to disperse agglomerates which may have formed during calcination. After ball milling, the aqueous suspensions were flash frozen using liquid nitrogen and the water sublimed through lyophilization (Advantage EL80, Virtis Co.) to prevent re-agglomeration of particulates through capillarity during the drying process.

2.2 Electron microscopy

Upon completion of all the powder synthesis steps, initial characterization of particle size and morphology for all four NSA dusts was completed using SEM (FE-SEM, Zeiss SUPRA 55VP) to observe the general packing and morphology of the bulk powders. Dusts were again suspended in an aqueous mixture and aerosol sprayed onto a conductive carbon mounting tape before being allowed to dry. In addition to application on carbon tape, dusts were also sputter coated with iridium prior to imaging.

2.3 Particle size and surface area

A dynamic light scattering system was used (Nano S-90, Malvern Instruments) to determine an accurate particle size distribution of the powders. Samples were prepared in an aqueous solution at 12.7 vol% solids loading with polyammonium methacrylate (Darvan C-N, RT Vanderbilt) as a dispersant. Samples were ultrasonically dispersed for 2 minutes at 80% power to ensure that any soft agglomerates were broken. Standard nitrogen absorption, Brunauer-Emmett-Teller or BET, surface area analysis was performed on calcined and dispersed dusts (Particle Technology Labs, Downers Grove, IL).

2.4 Insect toxicity bioassay – test insects

The species tested were adults of grain pests, the rice weevil *Sitophilus oryzae* (L.), and the lesser grain borer,

Rhyzopertha dominica (F). Insects were obtained from colonies with no history of exposure to insecticides, reared at Montana State University, according to procedures outlined in the rearing manual for stored-product insects first used by United States Department of Agriculture (USDA) Stored-Product Insect Research and Development Laboratory in Savannah, GA (1969). Insects were reared at 28° ± 1°C and 70% ± 5% relative humidity (RH) in dark. The desired relative humidity (RH) was maintained by using saturated salt solutions (Winston & Bates 1960).

Adults used in all experiments were 0–6 weeks old, of unknown sex and mating status. Wheat used for the experiments was hard white spring wheat (Clearine cv.). The moisture content of the wheat was determined by a Foss Infratec 1241 Grain Analyzer(Foss North America, Silver Spring, Maryland).

2.5 *Insect toxicity bioassay - toxicity assessment*

Laboratory bioassays were conducted to test the toxicity of NSA (commercial, combustion, co-precipitation and polymeric precursor dusts) using dry dust applications and evaluated at two different concentrations of the product: 125 ppm and 250 ppm.

The dusts were tested at two different humidity levels. The different levels of ambient humidity were 43% and 75% RH, and were achieved using chambers containing saturated solutions of potassium carbonate or sodium chloride, respectively, in water (Winston & Bates 1960). These humidity chambers were created in 20 cm × 30 cm × 11 cm plastic boxes, with false floors made using plastic waffle-type grids on the bottom with the salt solution below the grid (Arthur 2000). Temperature and humidity inside the chambers were monitored with HOBO data recorders (Onset Computer, Bourne, MA, USA). The experiments were conducted at 25 ± 1°C with a constant photoperiod of 12:12 (L:D). At 250 ppm concentration and 75% RH, a silica based diatomaceous earth dust was further included as a positive control. This diatomaceous earth, Protect-It®, was included for comparison purposes given its similar efficacy to NSA in previous studies (Stadler et al. 2012).

For each ambient humidity treatment, the wheat was acclimatized in the corresponding chamber at 25 ± 1°C for several weeks until it achieved the desired constant grain moisture content. The target grain moisture levels for the low and high humidity treatment were 10.7 and 14.7, respectively (Wicklow et al. 1998). The bioassay methodology was the same as that reported in Stadler et al. (2010b). Briefly, once the grain achieved the target moisture content, it was mixed with NSA or Protect-It® and shaken for 30 seconds to allow an even distribution of the dust through the entire grain mass. Then, dilutions were conducted to achieve the desired concentrations in 30 g of wheat, and placed in Petri dishes. Dilutions were achieved by adding untreated wheat to the treated one and shaking vigorously for 30 seconds. A Petri dish containing 30 g of untreated wheat was used as control. Ten insects were placed in each Petri dish. Each experiment consisted of five replicates and was run twice with each insect species and concentration of NSA. Adult mortality was assessed 3, 6 and 9 days after continuous exposure to the treated wheat in the corresponding humidity chamber.

2.6 *Data analysis*

The data were analyzed using the Mixed Procedure (PROC MIXED) of the Version [9.3] of the SAS System (SAS Institute, 2011) with mortality as the response variable and concentration, product, time and their interaction as main effects. Date of observation was the repeated measure in the analysis. Another separate analysis of the mortality at the end of the bioassay was conducted with PROC MIXED, where mortality was the response variable and product, concentration and their interaction were the main effects. In all analyses, Petri dish replicates were included as a random factor and each humidity level was analyzed separately. The variance–covariance structure was modeled as compound symmetry. Control mortality was corrected using Abbott's formula (Abbott 1925). LSMEANS comparisons were conducted with the Tukey option in SAS.

3. Results and discussion

3.1 *Particle size and morphology*

Examination of the obtained micrographs indicates that the individual synthesis route heavily influences particle morphology (Figures 1 and 2) although there may not be as significant size difference among the various dusts synthesize in this study (Figure 3). Despite efforts to prevent agglomeration, both the combustion synthesis and co-precipitation dusts appear at high magnification to be composed of very small particulates of <50 ηm (ηm = nanometers) as is supported by particle size analysis using the Zetasizer (Figure 3). This generates a bimodal distribution of pore sizes associated with the individual particles in contrast to agglomerate contact points. The commercial alumina and polymeric precursor are fully densified with larger particles. The combustion synthesis dust has a platelet morphology contrasting with the other nearly spherical dust. This may be a result of the batch-based process yielding skinning and evaporation processes associated with precursor sticking to the walls of the combustion vessel.

At lower magnifications, the bulk view of the combustion dust shows a highly porous structure. The co-precipitated dust yielded substantial porosity of the cluster, while the polymeric precursor showed larger particulates, with much less porosity. The polymeric precursor and the commercial alumina dust showed a much more dense particulate structure, indicating a reduced range of pore gradients. SEM images qualitatively indicate that the combustion synthesized dusts tend to form highly porous agglomerates with highly tortuous surfaces.

A well-established theory for the mode of action of nanoalumina is that it kills arthropods by adsorbing the epicuticular lipid layers through capillarity, causing

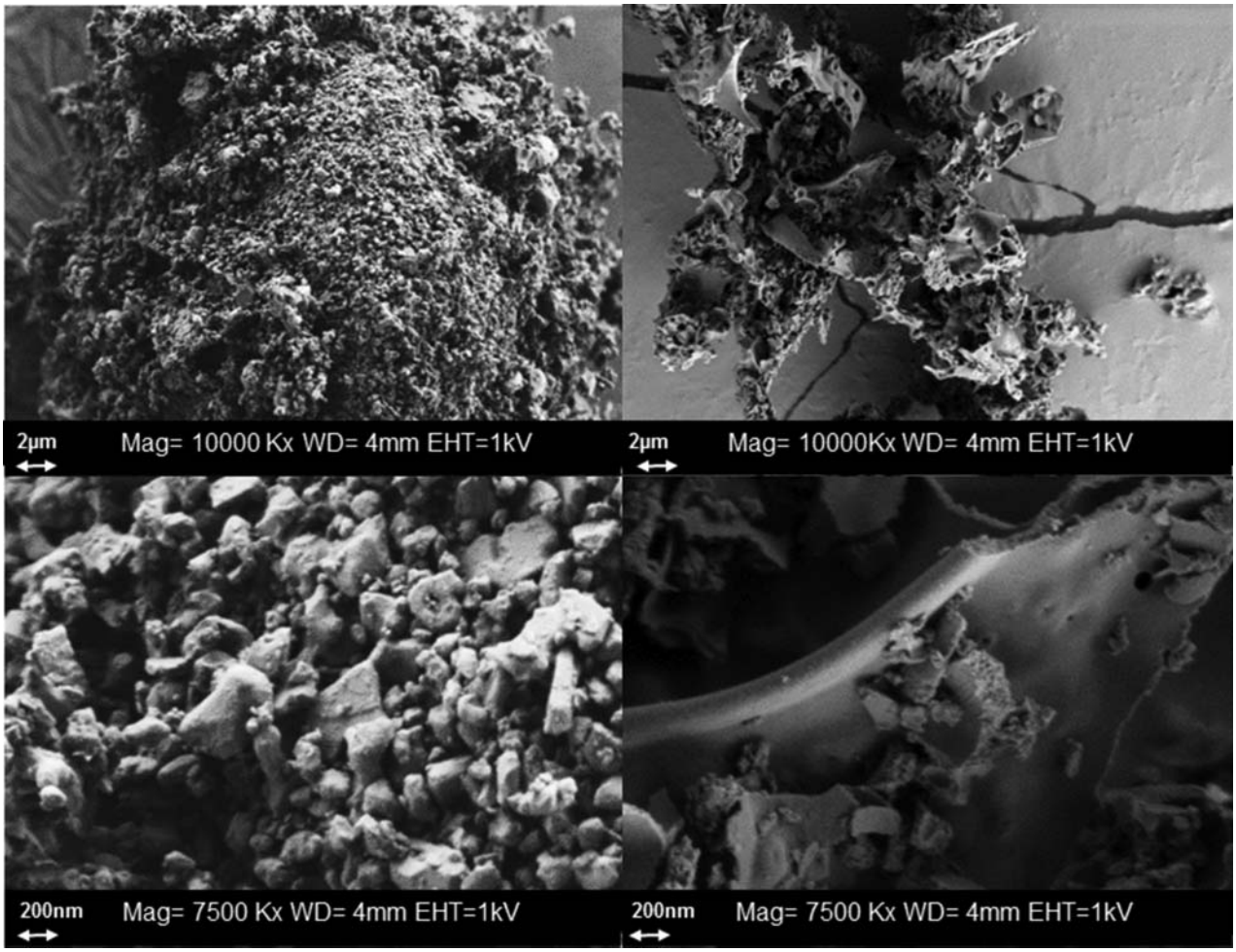

Figure 1. Paired low (bottom) and high (top) magnification SEM images of commercial nano-scale aluminum oxide (left) and combustion synthesized dust (right).

excessive water loss through the cuticle, (Stadler et al. 2010a). This would suggest that the most effective dusts would have a small particle size with high surface areas as well as being of a composition conducive to wetting of the specific hydrocarbons present on the surface of the insect. A key question is whether the porosity of the individual particulate or the capillaries formed at the interface between adjacent particles is the primary mechanism of adsorption. Capillary pressure is defined by Equation 1 below in which it is readily apparent that across species interfacial energy and hence wetting is a key factor in tailoring specific insecticide dusts. The interfacial energy based on the epicuticular liquids present on a single chemical compound is assumed to constant given the identical precursors used, except for changes in humidity, thus dictating that pore radius will influence the extraction of the hydrocarbons.

$$P = \frac{2\gamma\cos\theta}{r} \quad (1)$$

P - capillary pressure
γ - interfacial energy
θ - wetting angle
r - interface radius.

Particle size analysis (Figure 3) clearly delineated the commercial dust from the remaining three in terms of average particle size. The commercial dust had an average particle size of 783 ηm with a relatively wide distribution and a small secondary peak at approximately 6 μm. SEM images of the commercial dust clearly show individual particles smaller than 150 ηm. However, based on particle size analysis, even with the use of the polymeric dispersant, this material has formed rather large agglomerates that are not easily broken up by the ultrasonic mixing process. The combustion synthesized, co-precipitated and polymeric precursor dusts had substantially smaller average particle sizes of 9.5, 11.2 and 6.5 ηm, respectively. The NSA prepared through co-precipitation was significantly larger in particle size than the other laboratory synthesized dusts showing a bimodal particle size distribution indicating larger particles or agglomerates. This is likely due to the inherent agglomeration process that occurs during drying of the precipitated oxy-hydroxide in the co-precipitation methodology. Interestingly, the combustion synthesized dust observed immediately after the synthesis indicated the formation of relatively large platelets when transferred directly from the combustion chamber to the SEM mount. However, during both the subsequent handling of the powder and the preparation of samples for

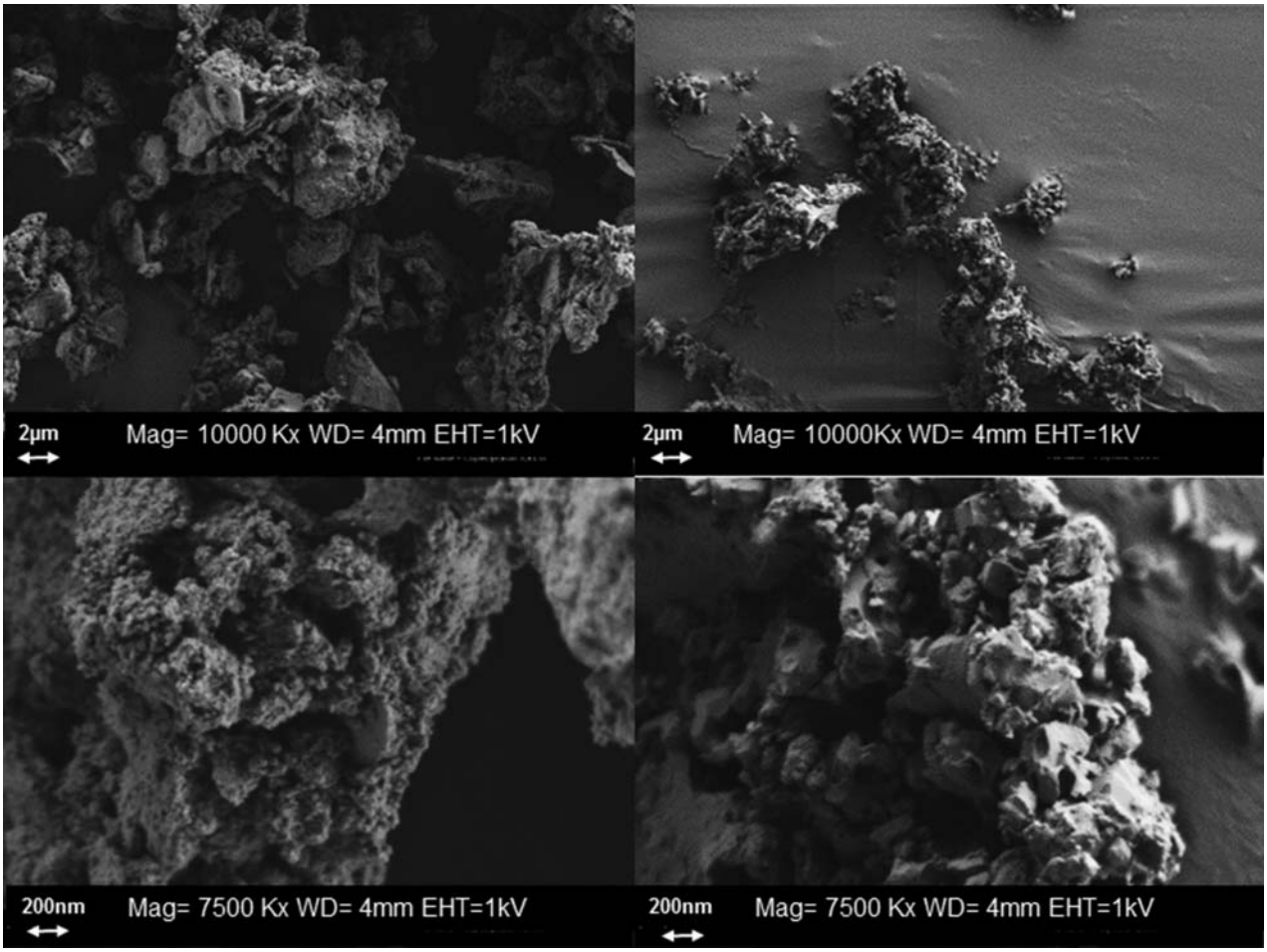

Figure 2. Paired low (bottom) and high (top) magnification SEM images of co-precipitate dust (left) and polymeric precursor dust (right).

particle size analysis it is shown that these platelets form extremely weak agglomerates that easily breakup into their constituent nanoparticles (Figure 1). The variation between the particle size observed in the SEM images and that reported by laser scattering particle size analysis indicates that it is possible to obtain a level of particle dispersion in a water suspension that is not matched by the dry dusts, indicating that either method used independently may not capture the outcome associated with optimum pesticide activity.

Results of BET analysis are combined with average particle size for comparison (Figure 4). As supported by SEM and particle size results, the commercial NSA exhibited a very small specific surface area. While particle size is easily influenced by agglomerates, BET methods can measure surface area whether agglomerated or dispersed. The low surface area of the commercial dust is consistent with both larger particles that are also much smoother and of higher density. The polymeric precursor and co-precipitation routes produced specific surface areas of 74.42 and 74.13 $m^2.g^{-1}$, respectively, indicating a rough surface texture of the individual particles. The combustion synthesized dust however showed a strong deviation in surface area at 16.76 $m^2.g^{-1}$. Based on the small average particle size of 9.54 ηm, the combustion synthesized dusts are also relatively smooth compared to the other synthesis methods. A schematic representation of the dusts to identify the apparent trends in size and morphology based on particle size and surface area analysis is provided in Figure 5.

3.2 Dust insecticide efficacy

In *R. dominica*, mortality due to NSA dusts increased with time (125 ppm low humidity: $F = 18.85$, df = 2, 40, $P < 0.0001$; 250 ppm low humidity: $F = 11.41$, df = 2, 90, $P < 0.0001$; 250 ppm high humidity: $F = 47.29$, df = 2, 108, $P < 0.0001$) and was different among dusts (125 ppm low humidity: $F = 28.57$, df = 4, 20, $P < 0.0001$; 250 ppm low humidity: $F = 69.80$, df = 4, 45, $P < 0.0001$; 250 ppm high humidity: $F = 18.54$, df = 5, 54, $P < 0.0001$) for all humidity levels and doses tested. There was also a significant interaction between time and treatment for the 250 ppm dose (high humidity: $F = 3.68$, df = 10, 108, $P = 0.0003$; low humidity: $F = 3.34$, df = 8, 90, $P = 0.0022$) (Figures 6 and 8). At the end of the bioassay after nine days of continuous exposure, there was a treatment effect on mortality ($F = 23.17$, df = 5, 54, $P < 0.0001$) and the combustion synthesized dust was more effective than the rest of the dusts and as effective as the diatomaceous earth Protect-It (Tukey contrasts $P < 0.05$) to *R. dominica* in

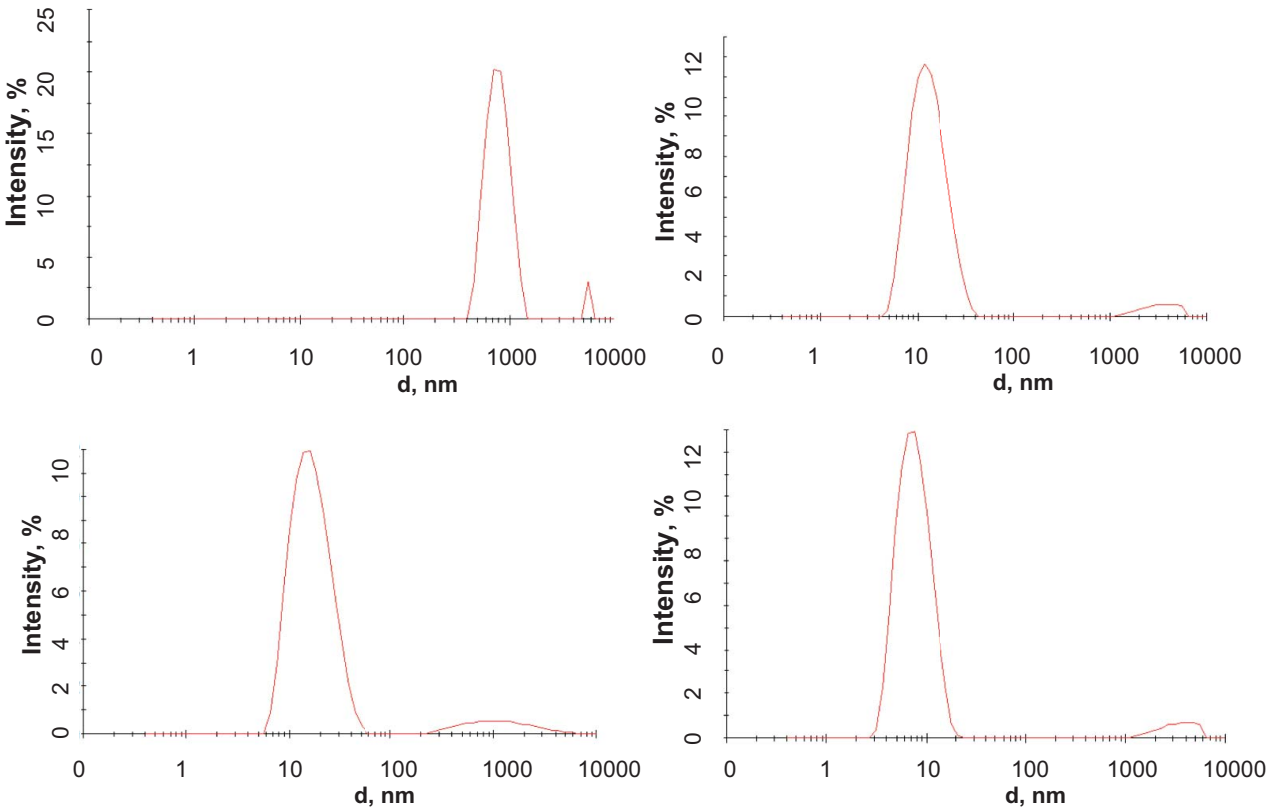

Figure 3. Particle size distributions of commercial (top left), combustion synthesis (top right), co-precipitation (bottom left) and polymeric precursor (bottom right) dusts.

treated wheat held at the high humidity. At 125 ppm under low humidity, there was also a difference in mortality among treatments (F = 24.61, df = 4, 20, $P < 0.0001$), where the combustion dust was more effective than the rest of the tested dusts followed by the co-precipitation and polymeric precursor dusts (Tukey contrasts $P < 0.05$). At 250 ppm under low humidity, there was also a difference in mortality among dusts (F = 9.89, df = 4, 45, $P < 0.0001$), where the combustion synthesized dust was as effective as the co-precipitation and polymeric precursor dusts, and followed by the commercial dust (Tukey contrasts $P < 0.05$). These results suggest that *R. dominica* is most affected by very small, yet smooth particulate dusts (Figure 5).

In *S. oryzae*, mortality due to NSA dusts also increased with time (125 ppm low humidity: F = 350.61, df = 2, 45, $P < 0.0001$; 250 ppm low humidity: F = 5.05 df = 2, 40, $P < 0.0001$; 250 ppm high humidity: F =

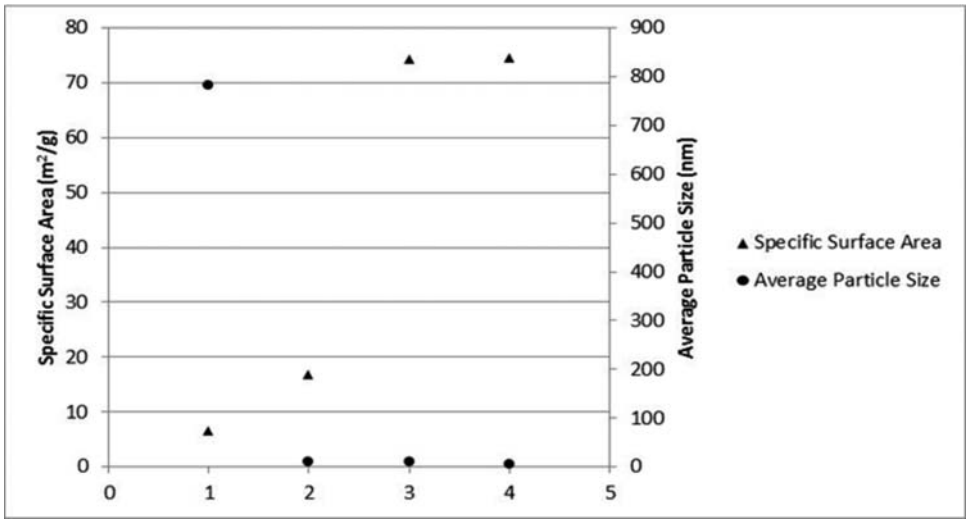

Figure 4. Plot of specific surface areas and average particle sizes by composition. 1: commercial, 2: combustion synthesis, 3: co-precipitation, 4: polymeric precursor.

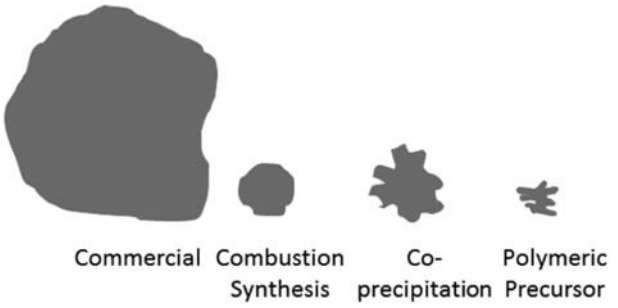

Figure 5. Comparative schematic representation of the aluminum oxide dusts.

237.17, df = 2, 108, $P<0.0001$) and was different among dusts (125 ppm low humidity: $F = 58.11$, df = 4, 45, $P < 0.0001$; 250 ppm low humidity: $F = 87.52$, df = 4, 20, $P < 0.0001$; 250 ppm high humidity: $F = 92.74$, df = 5, 54, $P < 0.0001$) for all humidity levels and doses tested. There was also a significant interaction between time and treatment 125 ppm low humidity: $F = 21.28$, df = 4, 45, $P < 0.0001$; 250 ppm low humidity: $F = 19.57$, df = 8, 40, $P < 0.0001$; 250 ppm high humidity: $F = 39.65$, df = 10, 108,$P<0.0001$) (Figures 7 and 8). Early in these toxicity tests, the combustion synthesis dust had a greater efficacy at both humidity levels, with 100% mortality after three days of exposure under low humidity.

After nine days of exposure of *S. oryzae* adults to treated wheat under high ambient humidity, there was a treatment effect on mortality ($F = 95.89$, df = 5, 54, $P < 0.0001$), where the combustion synthesized and the polymeric precursor dusts were more effective than the others, followed in efficacy by the diatomaceous earth Protect-It and the co-precipitation dust (Tukey contrasts $P < 0.05$) (Figure 7). At 125 and 250 ppm under low humidity, there was also an effect of type of dust on mortality (125 ppm: $F = 32.31$, df = 4, 45,$P<0.0001$; 250 ppm: $F = 108.31$, df = 4, 20,$P<0.0001$), where the combustion dust, as well as the co-precipitation and polymeric precursor dusts were more effective than the commercial dust and the control (Tukey contrasts $P < 0.05$) (Figures 7 and 8). As expressed by previous studies, particle size itself is not enough to explain the difference in efficacy among inert dusts (Arnaud et al. 2005).

The reduced mortality observed at the higher humidity level supports the notion that dusts can adsorb either water or cuticle waxes and thus atmospheric water reduces the effectiveness of all the tested dusts by competing with the cuticle hydrocarbons. Interestingly, the combustion

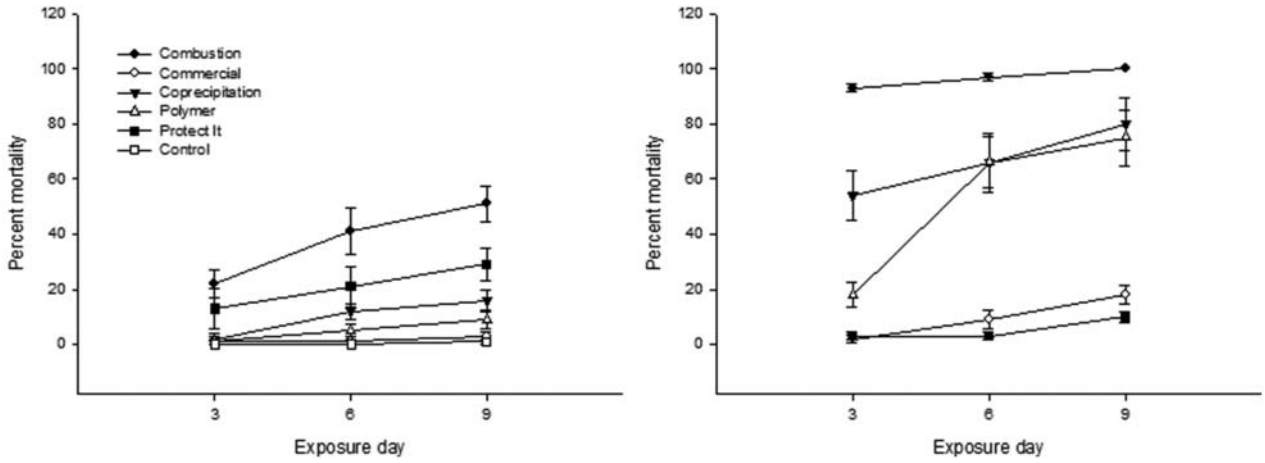

Figure 6. Efficacy of pesticide dusts on *Rhyzopertha dominica* at 250 ppm under low (43%) (right) and high (75%) humidity (left).

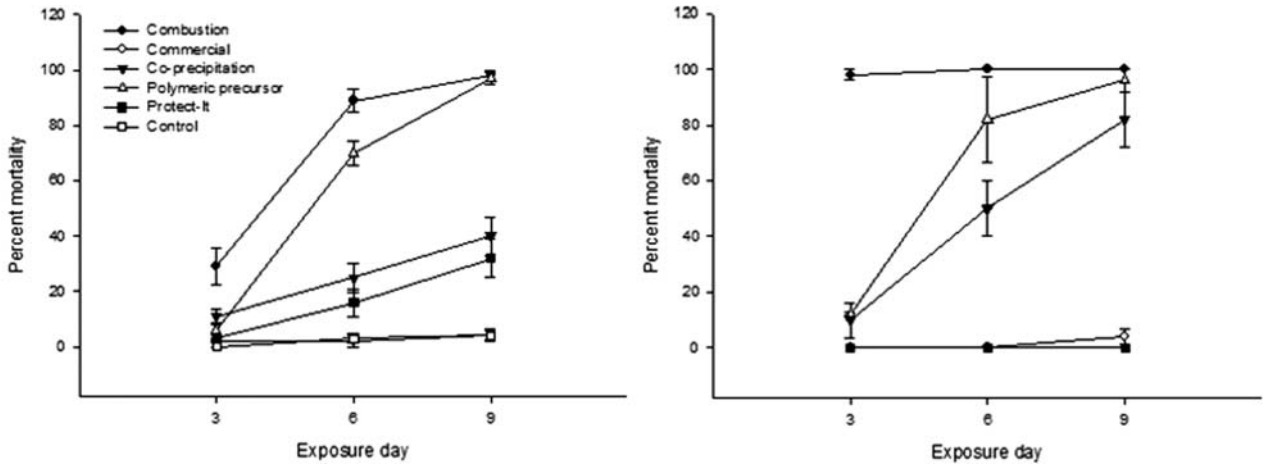

Figure 7. Efficacy of pesticide dusts on *Sitophilus oryzae* at 250 ppm under low (43%) (right) and high (75%) humidity (left).

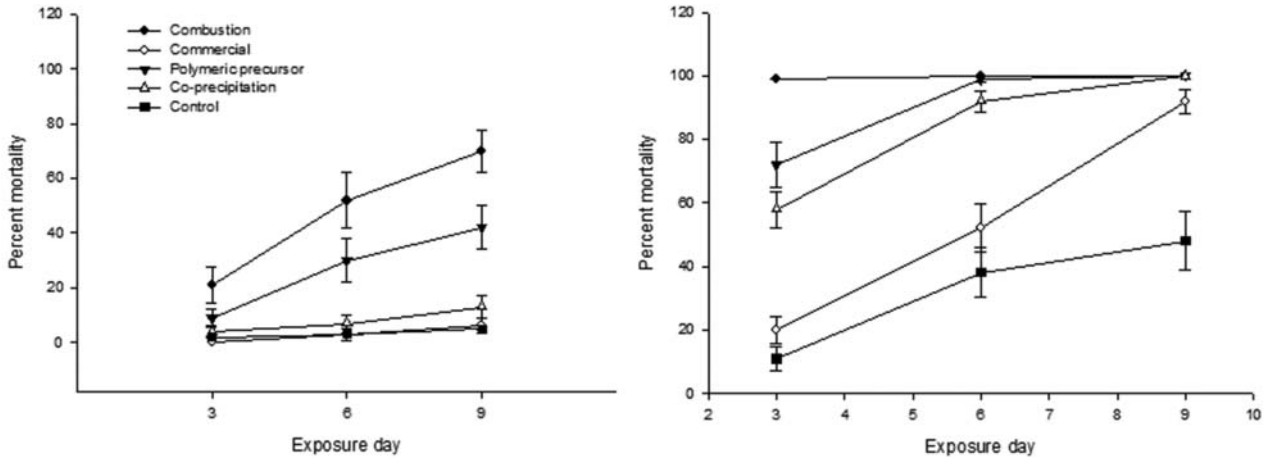

Figure 8. Efficacy of pesticide dusts on *Rhyzopertha dominica* (left) and *Sitophilus oryzae* (right) at 125 ppm under low (43%) humidity.

synthesized dusts yielded the best performance under both moisture treatments. The significant insecticidal performance difference between the laboratory synthesized NSA and commercial NSA as well as the corresponding particle size and morphology differences clearly support the dependence of insecticidal activity on physical properties of the nanoparticulate dust. The results obtained in this study support the hypothesis that inert dusts absorb epicuticular hydrocarbons by capillary forces and that dusts with smaller particle size will cause greater insect mortality. However, relatively small variations in particle size and surface area yield significant differences in efficacy that are not fully understood. While the three laboratory synthesized dusts indeed support this notion when compared to the commercial dust, it is interesting that the combustion synthesized NSA was significantly more effective than the other dusts despite its relatively small specific surface area. Based on SEM results, it appears that the very small particulate dust (~10 ηm) tends to agglomerate in the dry state to form micron-scale structures that easily breakup during any mixing or agitation process. SEM images qualitatively indicate that the combustion synthesized dusts form highly porous agglomerates with highly tortuous surfaces. This type of morphology would indeed seem most conducive to a capillarity driven adsorption of an epicuticular lipid layer.

A full toxicological profile and a more detailed analysis of the efficacy of NSA dust as insecticide for stored product pests would require testing it against other insect species, and exceeded the scope of this study. However, using two species that differ in their susceptibility to inert dusts in general (Subramanyam & Roesli 2000), allowed us to conduct a quick screening of different types of NSA dusts, and showed which physical characteristics to aim for to increase insecticide effectiveness of these dusts, like particle size in combination with porosity. It also provided information on which synthesis processes provide the most effective NSA, which in this case was the combustion synthesis technique. This quick and simple method can be used to synthesize highly pure, homogeneous and crystalline oxide ceramic powders, including ultrafine nanoparticles with broad particle size distributions (Mukasyan et al. 2007; Timmaji et al. 2011). Some of the parameters that control the combustion reaction include fuel-to-oxidizer stoichiometry, solvent concentrations and heating rates, as well as doping strategies to increase the wettability of the alumina surface by changing the chemical composition at the surface (i.e. silica) (Toniolo et al. 2005, 2007). Further research will focus on investigating and optimizing these process variables to achieve a more effective NSA dust.

Excellent efficacy of the dusts in general for both insect species tested was observed at low humidity, which decreased significantly at elevated humidity (Figures 6 and 7). To establish the rate at which the NSA adsorbs water, a gravimetric analysis was performed for the combustion synthesized dust at 75% RH over a period of 7 hours. The weight gain from adsorbed water in a humidity controlled environment shows that adsorbed water content reaches a nearly constant value after only 60 minutes of exposure (Figure 9). Therefore, even short exposures to high humidity can rapidly influence the efficacy of these materials, however, long term storage may not mitigate the performance of the nanoalumina as indicated by the efficacy tests that were performed well beyond 60 minutes of exposure. The powder studied by

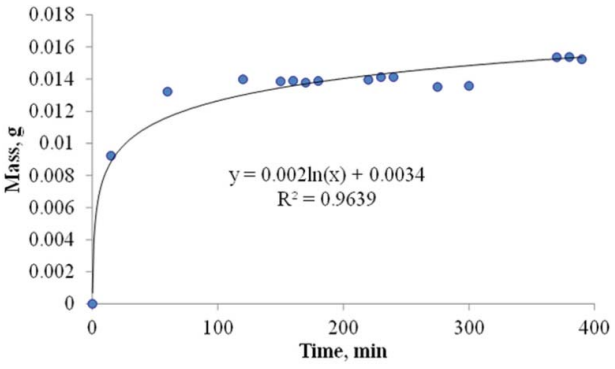

Figure 9. Weight gain of combustion synthesized alumina as a function of time, measured at 75% RH.

gravimetry was the combustion synthesized dusts, (Figure 9) which had a relatively low surface area when compared to the other dusts tested, and it is anticipated that higher surface area dusts may take substantially longer to reach an equilibrium water content. Interestingly, the efficacy data still show the combustion powder to be most effective even in a humid environment such that a lack of complete saturation in the high surface area particles does not appear to improve performance. In this manner, we hypothesize that the nearly persistent positive curvature of the small combustion synthesized particles (in contrast to the high surface area particles that consist of far more dramatic surface undulation, Figure 5) enhances the formation of a capillary between the insect and particle.

4. Conclusions

Three solution synthesized nanoalumina powders were fabricated, characterized and tested for efficacy as insecticides. The combustion synthesized dust consistently yielded greater mortality rates, and all the dusts were more effective on *S. oryzae* than on *R. dominica*, yielding 100% mortality after three days under low ambient humidity. The differences in toxicity observed for the two insect species across the various types of dusts are a strong indication that dusts can be optimized or tailored for different insect species. The defining characteristic of the inert dusts that renders maximum efficacy does not solely reside in particle size, and this study indicates that surface area and hence morphology have a great impact in addition to surface chemistry, which was designed to be constant for this study. Future studies will examine compositional variations in nanodusts ranging from pure alumina to a combination of alumina and silica to tailor the wetting behavior of epicuticular waxes.

Acknowledgements

We are very grateful to Megan Hofland for technical assistance and to the anonymous reviewers whose suggestions highly improved an earlier version of the manuscript.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

This work was supported by the CONICET [grant number PIP N°11220100100360]; NC-213 Marketing and Delivery of Quality Grains and Bioprocess Coproducts: multistate research project.

References

Abbott WS. 1925. A method of computing the effectiveness of an insecticide. J Econ Entomol. 18:265–267.

Arjunan NK, Murugan K, Rejeeth Ch, Madhiyazhagan P, Barnard DR. 2012. Green synthesis of silver nanoparticles for the control of mosquito vectors of malaria, filariasis, and dengue. Vector Borne Zoonotic Diseases. 12:262–268.

Arnaud L, Thi Lan HT, Brostaux Y, Haubruge E. 2005. Efficacy of diatomaceous earth formulations admixed with grain against populations of *Tribolium castaneum*. J Stored Prod Res. 41:121–130.

Arthur FH. 2000. Toxicity of diatomaceous earth to red flour beetles and confused flour beetles (Coleoptera: Tenebrionidae): effects of temperature and relative humidity. J Econ Entomol. 93:526–532.

Arthur FH. 2001. Immediate and delayed mortality of *Oryzaephilus surinamensis* (L.) exposed on wheat treated with diatomaceous earth: effects of temperature, relative humidity, and exposure interval. J Stored Prod Res. 37:13–21.

Athanassiou CG, Kavallieratos NG. 2005. Insecticidal effect and adherence of PyriSec in different grain commodities. Crop Prot. 27:703–710.

Athanassiou CG, Kavallieratos NG, Andris NS. 2004. Insecticidal effect of three diatomaceous earth formulations against adults of *Sitophilus oryzae* (Coleoptera: Curculionidae) and *Tribolium confusum* (Coleoptera: Tenebrionidae) on oat, rye and triticale. J Econ Entomol. 97:2160–2167.

Athanassiou CG, Kavallieratos NG, Basileios JV, Vassiliki KS. 2008. Evaluation of a new, enhanced diatomaceous earth formulation for use against the stored products pest, *Rhyzopertha dominica* (Coleoptera: Bostrychidae). Int J Pest Manag. 54:43–49.

Barik TK, Kamaraju R, Gowswami A. 2012. Silica nanoparticle: a potential new insecticide for mosquito vector control. Parasitol Res. 111:1075–1083.

Barik TK, Sahu B, Swain V. 2008. Nanosilica-from medicine to pest control. Parasitol Res. 103:253–258.

Buyukaksoy A, Petrovksy V, Dogan F. 2012. Redox stability of Ni-YSZ cermets prepared by polymeric precursor infiltration. ECS Trans. 45:509–514.

Buyukaksoy A, Petrovsky V, Dogan F. 2012. Optimization of redox stable Ni-YSZ anodes for SOFCs by two-step infiltration. J Electrochem Soc 159:F841–F848.

Debnath N, Dipankar D, Seth R, Chandra R, Bhattacharya SCh, Goswami A. 2011. Entomotoxic effect of silica nanoparticles against *Sitophilus oryzae* (L). J Pestic Sci. 84:99–105.

Ebeling E. 1971. Sorptive dusts for pest control. Annu Rev Entomol. 16:123–158.

Fields P, Korunic Z. 2000. The effect of grain moisture content and temperature on the efficacy of diatomaceous earths from different geographical locations against stored-product beetles. J Stored Prod Res. 36:1–13.

Golob P. 1997. Current status and future perspectives for inert-dusts for control of stored-product insects. J Stored Prod Res. 33:69–80.

Kavallieratos NG, Athanassiou CG, Michalaki MP, Batta YA, Rigatos H, Pashalidou FG, Balotis GN, Tomanovic Z, Vayias BJ. 2006. Effect of the combined use of *Metarhizium anisopliae* (Metschinkoff) Sorokin (Deuteromycotina: Hyphomycetes) and diatomaceous earth for the control of three stored product beetle species. Crop Prot. 25:1087–1094.

Ki HY, Kim JH, Kwon SC, Jeong SH. 2007. A study on multifunctional wool textiles treated with nano-sized Silver. J Mater Sci. 42:8020–8024.

Korunic Z. 1999. Enhanced diatomaceous earth: an alternative to methyl bromide. Aust J Tech. 2:95–104.

Korunic, Z. Ormesher, P. 1999. Evaluation and standardised testing of diatomaceous earth. In: Zuxun J, Quan L, Yongsheng L, Xianchang T, Lianghua G, editors. Stored product protection. Proceedings of the 7th International Working Conference on Stored-product Protection; 1998 Oct 14–19, Beijing. Chengdu (China): Sichuan Publishing House of Science and Technology; p. 738–744.

Liu Y, Tong Z, Prud'homme RK. 2008. Stabilized polymeric nanoparticles for controlled and efficient release of bifenthrin. Pest Manag Sci. 64:808–812.

Lord JC. 2001. Desiccant dusts synergize the effect of *Beauveria bassiana* (Hyphomycetes: Moniliales) on stored-grain beetles. J Econ Entomol. 94:367–372.

Marimuthu S, Rahuman AA, Rajakumar G, Santhoshkumar T, Vishnu Kirthi A, Jayaseelan A, Bagavan A, Zahir AA, Elango E, Kamaraj C. 2011. Evaluation of green synthesized silver nanoparticles against parasites. Parasitol Res. 108:1541–1549.

Mewis I, Ulrichs Ch. 2001. Action of amorphous diatomaceous earth against different stages of the stored product pests *Tribolium confusum*, *Tenebrio molitor*, *Sitophilus granarius* and *Plodia interpunctella*. J Stored Prod Res. 37:153–164.

Michalaki M, Athanassiou CG, Kavallieratos NG, Batta YA, Balotis JN. 2006. Effectiveness of *Metarhizium anisopliae* (Metschinkoff) Sorokin applied alone or in combination with diatomaceous earth against *Tribolium confusum* Jacquelin du Val larvae: influence of temperature relative humidity and type of commodity. Crop Prot. 25:418–425.

Mukasyan AS, Epstein P, Dinka P. 2007. Solution combustion synthesis of nanomaterials. Proc Combustion Inst. 31:1789–1795.

Nawrot J, Maliński E, Szafranek J. 1994. Function and composition of cuticular hydrocarbons of stored-product insects. In: Highley E, Wright EJ, Banks HJ, Champ BR, editors. Stored product protection. Proceedings of the 6th International Working Conference on Stored-Product Protection; 1994 Apr 17—23; Canberra, Australia. Wallingford (UK): CAB International..

Palyvos NE, Athanassiou CG, Kavallieratos NG. 2006. Acaricidal effect of a diatomaceous earth formulation against *Tyrophagus putrescentiae* (Astigmata: Acaridae) and its predator *Cheyletus malaccensis* (Prostigmata: Cheyletidae) in four grain commodities. J Econ Entomol. 99:229–236.

Perez de Luque A, Rubiales D. 2009. Nanotechnology for parasitic plant control. Pest Manag Sci. 65:540–545.

Rahman A, Seth D, Mukhopadhyaya SK, Brahmachary RL, Ulrichs C, Goswami A. 2009. Surface functionalized amorphous nanosilica and microsilica with nanopores as promising tools in biomedicine. Naturwissenschaften. 96:31–38.

Rigaux M, Haubruge E, Fields PG. 2001. Mechanisms for tolerance to diatomaceous earth between strains of *Tribolium castaneum.* Entomologia Experimentalis Applicata. 101: 33–39.

SAS Institute Inc. 2011. Base SAS® 9.3 Procedures Guide. Cary (NC): SAS Institute Inc. ISBN 978-1-60764-895-6.

Shah MA, Khan AA. 2014. Use of diatomaceous earth for the management of stored-product pests. Int J Pest Manag. 60:100–113.

Stadler T, Buteler M, Weaver DK. 2010a. Nanoinsecticidas: Nuevas perspectivas para el control de plagas [Nanoinsecticides: New perspectives for pest management]. Rev Soc Entomol Argent. 69:149–156.

Stadler T, Buteler M, Weaver DK, Sofie S. 2010b. Novel use of nanostructured alumina as an insecticide. Pest Manag Sci. 66:577–579.

Stadler T, Buteler M, Weaver DK, Sofie S. 2012. Comparative toxicity of nanostructured alumina and a commercial inert dust for *Sitophilus oryzae* (L) and *Rhyzopertha dominica* (F) at varying ambient humidity levels. J Stored Prod Res. 48:81–90.

Subramanyam BH, Roesli R. 2000. Alternatives to pesticides in stored-product IPM. Boston (MA): Kluwer Academic Publishers; Chapter 12, Inert dusts; p. 321–379.

Timmaji H, Chanmanee W, de Tacconi N, Rajeshwar K. 2011. Solution combustion synthesis of BiVO4 nanoparticles: effect of combustion precursors on the photocatalytic activity. J Adv Oxidation Technol. 14(1):93–105.

Toniolo J, Lima M, Takimi A, Bergmann C. 2005. Synthesis of alumina powders by the glycine-nitrate combustion process. Mater Res Bull. 40:561–571.

Toniolo J, Takimi A, Andrade M, Bonadiman R, Bergmann C. 2007. Synthesis by the solution combustion process and magnetic properties of iron oxide (Fe3O4 and-Fe2O3) particles. J Mater Sci. 42:4785–4791.

Werdin González JO, Gutiérrez MM, Ferrero AA, Fernández Band B. 2014. Essential oils nanoformulations for stored-product pest control – characterization and biological properties. Chemosphere. 100:130–138.

Wicklow DT, Weaver DK, Throne JE. 1998. Fungal colonists of maize grain conditioned at constant temperatures and humidities. J Stored Prod Res. 34:355–361.

Winston PW, Bates DH. 1960. Saturated solutions for the control of humidity in biological research. Ecology. 41:232–237.