

# Diatomaceous Earths – Natural Insecticides

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## SUMMARY

The regulatory issues for diatomaceous earth (DE) cover three fields: consumer safety, worker safety, and proof of efficacy against pests. For consumer safety, regulatory issues are similar to those for other additives, and a principal benefit of DEs is their removal by normal processing methods. For worker safety, regulatory issues are similar to those for other dusts, such as lime. The proof of potential insecticide values of DE may be assessed by using the analysis of physical and chemical properties of DE and its effect on grain properties and the proof of efficacy may be regulated by bioassay of standard design. Integrated pest management (IPM), a knowledge-based system, is rapidly providing a framework to reduce dependence on synthetic chemical pesticides. The main principle of post-harvest IPM is to prevent problems rather than to react to them. The specific curative measures using synthetic pesticides should be applied only when infestation occurs. DE and enhanced diatomaceous earth (EDE) formulations hold significant promise to increase the effectiveness and broaden the adoption of IPM strategies, thereby reducing the need for synthetic pesticides. By incorporating DE in an effective IPM program, grain is protected against infestation, loss caused by insects is prevented and grain quality is maintained until the grain is processed. Cases study data on the use of DE for commodity and structural treatment show that DE is already a practical alternative to synthetic pesticides in some applications.

**Keywords:** Diatomaceous earths; Safety; Insecticides; Stored products

## INTRODUCTION

Diatomaceous earth (DE) is a dust varying in color from white, grey and yellow to red. Dust is formed from fossilized diatoms, single-celled algae of various shapes and sizes which are composed almost entirely of amorphous silicon dioxide.

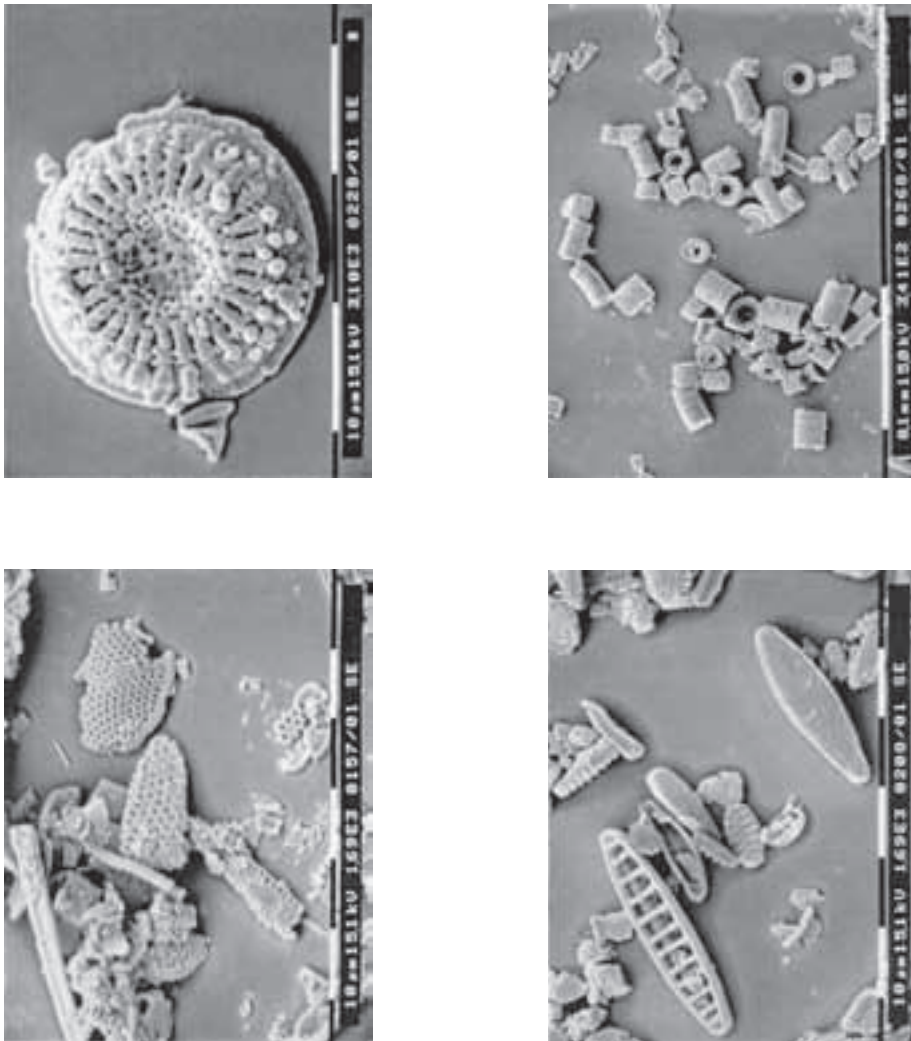
The specific mass varies, depending on the type and source of DE, from about 220-230 g/l up to about 670 g/l, while the value of pH differs from 4.4 to above 9 (Korunic, 1997, 1998). DE is without smell, its moisture content is about 2-6%, it is insoluble in water, non-inflammable with no risk of dust explosion. In addition to amorphous silica (from about 60 to about 93%), the major ingredient is calcium, but there are numerous other elements such as aluminium, magnesium, sodium, iron, phosphorus, sulphur, nickel, zinc, manganese and others (Subramanyam, 1993, Subramanyam and Roesli, 2000). Until now, no data have been obtained regarding a connection between the insecticidal action of dusts and any of these elements. After processing (digging, drying, milling) particles are 1 to about 150 microns in diameter, with median particle size between 2.5 to

30 microns. All particles contain very small inner pores which have the physical ability to absorb wax (lipids) molecules from the epicuticle of insects (Ebeling, 1971). DE adheres to the insect body and damages the protective waxy layer of the insect cuticle by sorption, and to a lesser degree by abrasion. The result is loss of water from insect body resulting in death (Ebeling, 1971). DE is also known to repel insects (White et al., 1966).

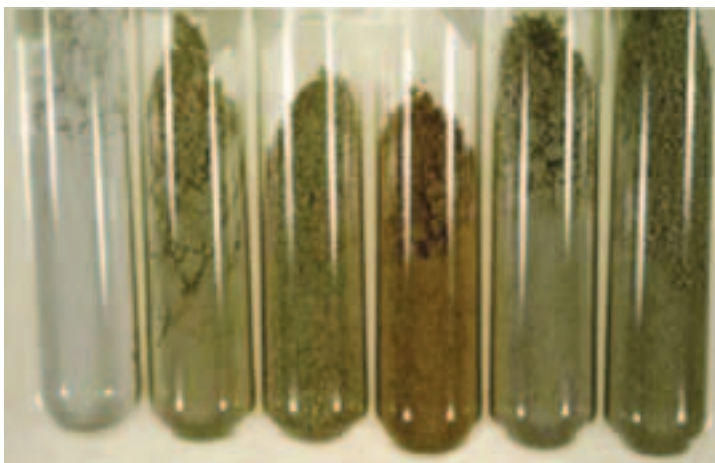
DE has long been known as a potentially useful grain protectant because it is safe to use, does not affect grain end-use quality, provides long-term protection and is comparable in cost to other methods of grain protection (Korunic et al., 1996a, 1996b). There are several review papers published in international journals describing different aspects of DEs use in insect control (Ebeling, 1971; Aleksander et al., 1944a, 1994b, 1994c; Korunic, 1994, Golob, 1997; 1998; Subramanyam and Roesli, 2000; Nikpay, 2006). Over the years, the use of DE has been limited because the required dose rates of 1000 to 3500 ppm (parts per million) for most DE products was found to reduce significantly the grain bulk density and flowability, and left visible dust residues (Subramanyam et al., 1994; Golob, 1997).



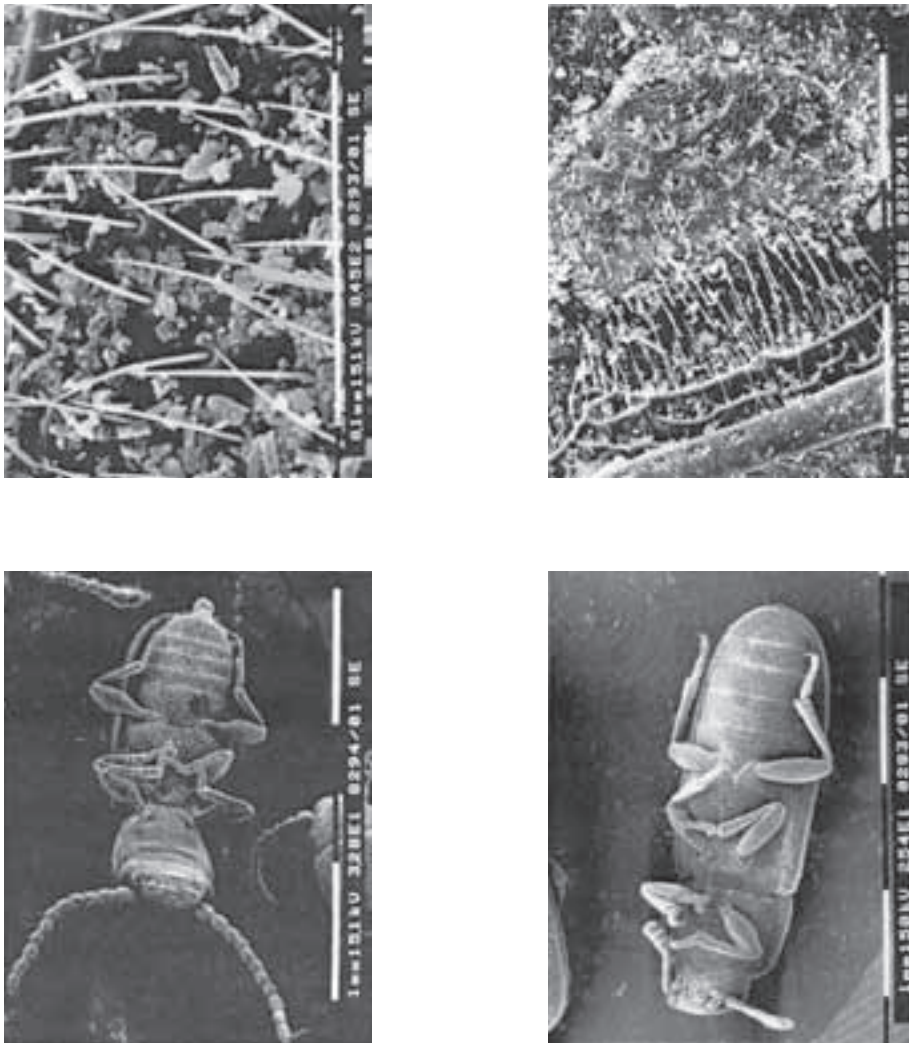
Figure 1. Different shapes of diatoms (Source: Internet)



**Figure 2.** Fossilized bodies of different shapes of diatoms and broken particles of body walls (Photo Korunic)



**Figure 3.** DE of various colors (Photo Korunic)



**Figure 4.** *Cryptolestes ferrugineus*

Upper row – part of abdomen covered with DE particles

Lower row – bodies of insects covered with DE particles

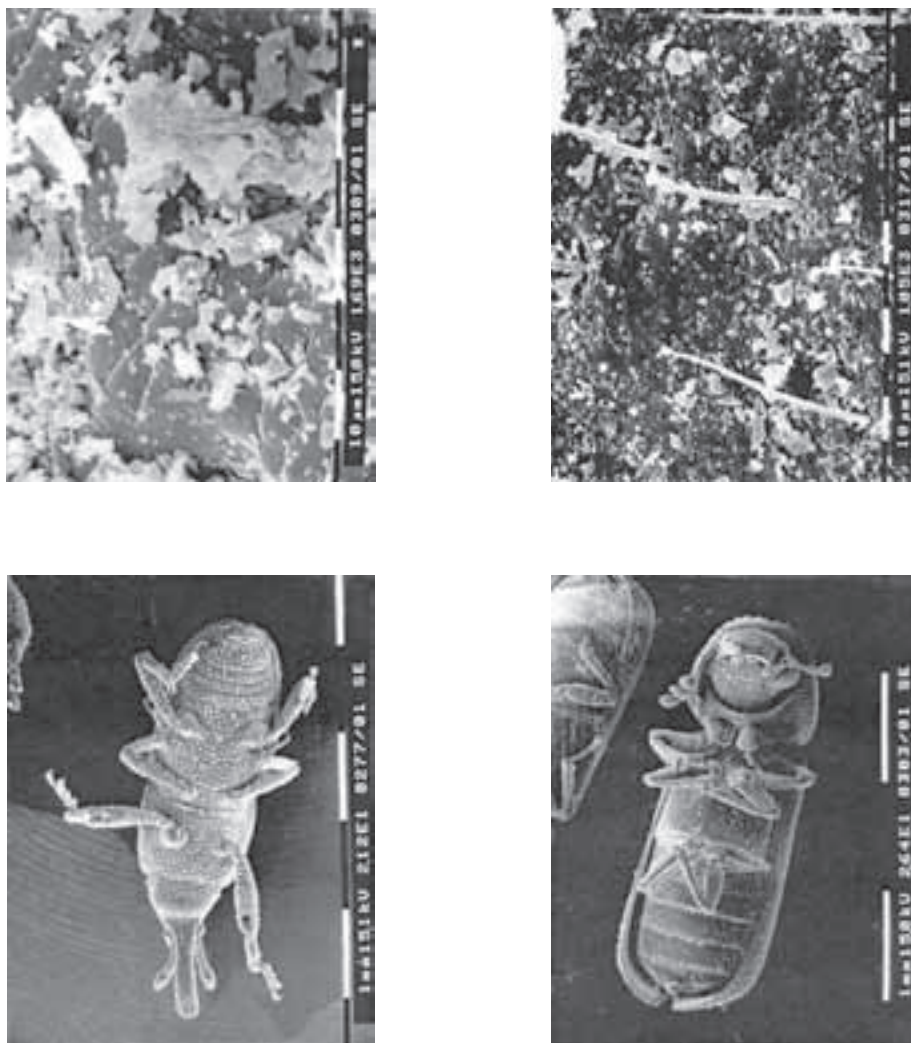
(Photo Korunic)

*Tribolium castaneum*

Newer diatomaceous earth-based insecticides, for example Protect-It, can be used at lower concentrations with acceptable efficacy against insects and with reduced adverse effects on grain handling and bulk density (Korunic and Fields, 1998; Korunic et al., 1998). Other effective formulations of DE have also been developed in the past several years that can be used at concentrations from 300 ppm to 1000 ppm to control stored grain insect pests (Dryacide, Protect-It, Silicosec, Insecto, Celatom DE, etc.) (Korunic, personal communication).

The most extensive research of DE has been undertaken in the field of protection of stored agricultur-

al products. Although different and sometimes contradictory results have been obtained, general conclusions about the sensitivity of stored product insects to DE were in agreement with results obtained by Korunic et al. (1996a, 1996b) and Korunic et al. (1997). Given the same commodity, there is a significant variation in the susceptibility of different insect species to enhanced DE Protect-It. The insects, in order of most susceptible to least, are *Cryptolestes ferrugineus* Steph., rusty grain beetle > *Oryzaephilus surinamensis* L., saw toothed grain beetle > *Sitophilus oryzae* L., rice weevil > *Sitophilus granarius* L., granary weevil > *Ryzoperthe dominica* F., lesser grain borer > *Tribolium castaneum*



**Figure 5.** *Sitophilus oryzae* Upper row – part of abdomen covered with DE particles  
*Rhyzopertha dominica* Lower row – bodies of insects covered with DE particles  
 (Photo Korunic)

Herbst., red flour beetle and > *Prostephanus truncatus* (Horn), larger grain borer. The concentrations required to achieve a 90% reduction in offspring were similar, or in some cases significantly higher than the concentrations required to reduce parent populations by 90%. In addition to different insect species, DE efficacy also varies by commodity. There is a significant variation in the efficacy of DE on different types of commodities against the same insect species (Aldryhim, 1990). The commodities, in order of highest to lowest doses for LD<sub>50</sub> (lethal dose that kills 50% of insect population) are: milled rice>corn>oats>barley>wheat (Korunic et al., 1997; Korunic, 2007a). Amorphous silicon dioxide

is non-toxic to mammals (Anon., 1991) and has been registered in many countries as a food additive (Anon., 1981, 1991). The regulatory issues for DEs cover three fields, namely consumer safety, worker safety, and proof of efficacy against pests. For consumer safety, regulatory issues are similar to those for other additives, and a principal benefit of DEs is their removal by normal processing methods (Desmarchelier and Allen, 2000). For worker safety, regulatory issues are similar to those for other dusts, such as lime. The proof of efficacy may be regulated by performing rapid assessment of potential insecticide values of DE (Korunic, 1997) and bioassay of standard design (Fields et al., 2003).

## HEALTH, SAFETY AND REGULATORY ISSUES

A good overview of occupational health and safety issues concerning silica is presented in the publications of NOHSC (1995), USA EPA (1996) and IARC Monograph (WHO, 1997).

Generally, there are two main health and safety issues relating to the use of DE. One concerns consumers, both humans and animals, and the other concerns workers. Health hazard of DE to consumers is reduced by two factors. Firstly, silica does not accumulate in mammals but is excreted as silicate in urine. In addition, daily intakes of silicates from water and plants and from soil by ruminants, is high. Secondly, DE is removed (> 99%) to below limits of detection by "traditional" washing of food and also by processes used in modern mills to clean grain prior to milling. The main hazard to workers is from inhalation because dusts cause respiratory problems. The issues of importance are: the amount of dust, its particle size and extent to which any given DE product contains crystalline silica. In broad terms, exposure safety limits for amorphous DE are similar to those for common materials such as cement and lime. Two main methods of reducing intake of various dusts are the procedures for reducing their amounts in workspace and the use of equipment to protect against inhalation (e.g. masks). DEs also dry the skin; this can be prevented by wearing hats, glasses, gloves and overalls. There is evidence that, in some situations, DE can reduce hazard from grain dust because a large number of small respirable particles of grain dust can adhere to non-respirable particles of DE, thus greatly reducing the amount of respirable dust in a workspace (Desmarchelier and Allen, 2000). DE poses no local threat to the environment, as approximately 50% of the Earth's crust is silica (e.g. sand) or a silicate (e.g. soil). DE residues do not degrade on stored grain, but can be readily removed by processing. However, residues before processing can cause problems in grain movement and to worker safety (airborne dust). Exposure standards for DE may vary slightly among countries, and users of DE should consult local regulations. Exposure standards such as the Time Weighted Average (TWA) specify a maximum exposure level for workers for an 8 h day and a 5-day working week. The US standard (OSHA 1991) for DE containing < 1% of crystalline silica is 6 mg/m<sup>3</sup>. The Australian TWA for DE can be compared with those for kaolin and starch (each 10 mg/m<sup>3</sup>), lime (5 mg/m<sup>3</sup>), wood dust (1-5 mg/m<sup>3</sup>, depending on type), cotton dust (0.2 mg/m<sup>3</sup>), white asbestos (1.0 fibre per ml of air) and blue asbestos (0.1 fibre per ml of

air). The Australian standard (NOHSC, 1995) for uncalcined DE is the same as for precipitated silica or silica gel, no value for the respirable fraction and a value of 10 mg/m<sup>3</sup> for the inspirable fraction. In contrast to synthetic pesticides, DE cannot penetrate through the skin or through clothing. DE can easily be removed from clothing by regular washing (Desmarchelier and Allen, 2000).

A good background to possible problems caused by DE in diet is given by Iler (1977; 1979) and Villota and Hawkes (1986). Silica is an important component of many plants, and is often present in outer sheaths (e.g. rice hulls, where the ash is almost pure silica). Iler (1977, 1979) describes many interesting examples, e.g., the inner part of bamboo (bamboo sugar or tabashir) has an ash content which is pure silica, and bamboo sugar has been a part of Chinese medicine for at least 700 years (and doubtless for much longer). The silica content of malt hulls is so high that beer is a saturated solution of silica. Grains contain 0.02-0.07% silica by weight. Human blood contains 20-40 mg silica/l, urine 36 mg/l and the aortic wall 120-150 mg/kg. Silica is used as a thickener in ointments and suppositories, as filler in tablets and is used to prevent clogging in hygroscopic powders (Martindale, 1972). It is also used in dentifrices (Budavari, 1989). The combination of removability by processing, inherent low toxicity, natural presence in foods, and water solubility supports the general acceptance of the active ingredient of DE in diet as "generally regarded as safe" (GRAS). In a monograph on evaluated carcinogenic risks of chemicals to humans (1997), the International Agency for Research on Cancer of the World Health Organization (IARC, WHO) listed amorphous silica dioxide (diatomaceous earth) into Group 3, explaining that "there is inadequate evidence for the carcinogenicity of amorphous silica to humans". The USA Occupational Safety and Health Administration (OSHA) of the U.S Department of Labor, gave in a memorandum on April 5, 1999, under the Hazard Communication Standard (HCS) for uncalcined diatomaceous earth (amorphous silica), an interpretation about the status of DE regarding the health effects of silicosis and carcinogenicity. DE has been tested as a whole and evaluated as a Group 3 carcinogen by IRAC. A Group 3 listing indicates that DE is not classifiable as to carcinogenicity to humans since definitive conclusions cannot be drawn from the research conducted to date. Therefore, there is no requirement under the HCS to state a definitive finding of carcinogenicity on labels or in material safety data sheets (MSDS) for DE products containing less than 1% crystalline silica. However, this policy does not apply to DE products containing more than 1% crystalline silica.

## REGULATIONS FOR EFFICACY AGAINST INSECTS

When DE Dryacide first became widely used in Australia, the problem arose of certification of batch performance. As it was not possible, at the time, to devise a chemical or physical test for the certification, a bioassay procedure was developed to evaluate the efficacy of each batch of that DE. This test involved exposure of *S. granarius* on wheat according to a defined protocol. This procedure made possible product certification but was time-consuming. An internationally-accepted standard test would be useful to facilitate comparison between various diatomaceous earths (Desmarchelier, personnel communication, 1996).

The efficacy of diatomaceous earth (DE) against insects depends greatly on several physical properties of diatom particles. Ideally, active DE should have high amorphous silicon dioxide content with a uniform particle size (less than 10 microns), a high oil sorption capacity, a large active surface, and very little clay and other impurities (Korunic, 1998). The analysis of physical and chemical properties of DE is time-consuming and expensive, and can be conducted only by experts at specially equipped laboratories. Therefore, in the past, bioassays were considered the most important criterion for assessment of the efficacy of DE against insects. These methods are relatively expensive and time-consuming also, since they require an expert and a well-equipped entomological laboratory.

Korunic (1997) has made a contribution to the study of DE by correlating biological efficacy against some simple physical methods. After conducting numerous experiments with 36 different diatomaceous earths or formulations collected from the USA, Mexico, Canada, Australia, Japan, China, and Macedonia, the results indicated that the efficacy of DE against insects depends on different properties of the diatom particles. For example, he correlated insecticidal activity with the effect of DE on the bulk density of wheat admixed with DE, with tapped density of DE, with the ability of DE to adhere to wheat kernels and with DE pH value. It is possible to evaluate and to predict the insecticidal value of diatomaceous earth mainly by very simple and low-cost analyses of these properties of DE without bioassays or extensive physical and chemical analyses. This approach has considerable potential, both for evaluation of alternative sources of DE and for checking batch performance. As there are over 10,000 known varieties of diatoms, quicker methods of evaluating potential insecticidal activity of their fossilized bodies are most welcome (Korunic, 1997).

At the 8<sup>th</sup> International Working Conference on Stored Product Protection, York, U.K., (2002) there was a discussion on problems encountered in testing the effectiveness of diatomaceous earths. A protocol was developed, and the efficacy of four diatomaceous earth samples was tested against laboratory reared cultures of 7 to 21 day old unsexed adults of *S. oryzae* and *T. castaneum*. Four independent testing centres followed the protocol and, based on the results, a standard protocol was developed. This standard protocol for evaluating DEs will make it easier to compare DEs by different laboratories. The recommendations for standardized DE testing are: to use the following concentrations of DE; 0, 300, 500, 700, 900 and 1100 ppm against *S. oryzae* and *T. castaneum* on wheat (100 g/replicate, 3 replicates, 50 adults that are 7 to 21 days old) at 13% m.c., 25° C and 60 ± 10 r.h. The mortality assessment should be taken at 7 and 14 days, and offspring assessment for *S. oryzae* taken 7 weeks and for *T. castaneum* 10 weeks after the beginning of the experiment (Fields et al., 2003).

## DIATOMACEOUS EARTH FROM DIFFERENT GEOLOGICAL LOCATIONS AND EFFICACY AGAINST INSECTS

The efficacy of DEs from different sources (mines) on insects is not the same (Katz, 1991; Snetsinger, 1988; Korunic, 1998). DE from salt water is more common, cheaper and supposedly more efficacious as an insecticide (Snetsinger, 1988). However, the results of Korunic's research (1997, 1998) show that the efficacy of DE against insects depends on different physical and morphological characteristics of diatoms rather than on its origin. Formulations of DE collected from different parts of the world, in spite of their similar mode of action against insects, are significantly different in their efficacy against insects, physical properties and in diatom species forming each DE. These are important findings because numerous registered formulations of DE are currently available under different names and a common belief is that they are equally efficacious (Korunic, 1996a, 1996b).

Some of the formulation names available as insecticides on the market are: Dryacide, Dicalite, Diacide, DiaFil, Insecolo, Insectigone, Insecto, Kenite, Melocide, Organic Plus, Perma-Guard, Protect-It, Silicosec, Shellshock, etc. (Subrammanuam and Roesli, 2000). Certain formulations are composed not only of DE. They contain a percentage of some other insecticide, most frequently pyrethrum (0.1-0.2%) and 1% piperonyl butoxide (1.0%) (Diacide Homeguard, Diatetect,

Perma Guard D-20, Perma-Guard D-21, etc.) (Quarles and Winn, 1996). Many DE-based insecticides have been applied to control pests in various areas, but most frequently for the protection of agricultural stored products and against pests in homes and gardens. Application to growing plants is occasionally done (fruit plants, vine and vegetable). DE has gradually replaced some synthetic insecticides, and in the near future it will probably replace even more, especially in the stored-product protection field (Korunic, 1994, 1998; Quarles and Winn, 1996; Golob, 1997; Subramanyam and Roesli, 2000).

There are numerous reports from around the world on a widespread resistance of several stored-product insects to grain protectants from the groups of synthetic pyrethroids, organophosphates, carbamates, chlorinated hydrocarbons, *Bacillus thuringiensis*, botanicals and fumigants (Subramanyam and Hagstrum, 1995; Kljajić and Perić, 2005). Since only a physical method of controlling insects is involved in DE, genetic resistance is unlikely (Ebeling, 1971). However, a potential tolerance or resistance development should be considered when choosing DE to replace existing grain protectants (Korunic, 1998; Rigaux et al., 2001; Fields, 2003; Vayias et al., 2008). The development of resistance in stored-grain insects to some commonly used grain protectants and fumigants, coupled with consumers' concern for pesticide residues in processed cereal grains, has prompted an exploration of newer diatomaceous earth-based insecticides as alternatives to commonly used grain protectants.

DE is one of the safest and most efficacious pesticides used in agriculture, public health and veterinary areas. DE can be incorporated in an integrated pest management (IPM) strategy for the grain and food processing industry as a grain protectant and a residual insecticide (general, spot or crack and crevice treatment) to reduce insect problems (Korunic et al., 1996a, 1996b, 1998; Subramanyam and Roesli, 2000).

## BACKGROUND OF DIATOMACEOUS EARTH USE AND RESULTS

“Bathing in sand” is commonplace with birds and poultry protecting themselves against mites and other parasites. Four thousand years ago, observation of this natural phenomenon probably led the Chinese to use DE to control pests (Allen, 1972). In 1880, it was noticed in the USA that road dust killed caterpillars of the cotton moth (Stelle, 1880). Until the 1950s, clay dusts, sand or silica gels had been used more extensively in practice and in research than was DE. In the early 1950s, DE was used to

fight fruit moths, cucumber beetles, Mexican bean beetle larvae, stored-products pests and cockroaches (Bartlett, 1951). Generally, dusts including DE are insect repellents. Their repellent quality depends on dosage. High dosage of DE increases insect repellence, and because of this phenomenon reduces the negative influence of DE on parasites and predators (Flanders, 1941; Bartlett, 1951). The most extensive research of DE conducted by numerous researchers has been made in the field of protection of stored agricultural products (Korunic, 1998). The use of DE for structural treatment in stored product facilities was studied by Desmarchelier et al. (1992), Wright (1990), McLaughlin (1994) and Bridgeman (1994).

In the last decade a great number of papers have been published in various scientific journals dealing with the effectiveness of various DEs against several stored product insect pests. Research groups from Greece and the USA published the highest number of these papers, mainly reporting on laboratory research of DE effectiveness against stored product insect pests with extensive reference sections (Arthur, 2003; Atahanassiou et al., 2005; Kavallieratos et al., 2012). Although different and often completely contradictory results were reported, it is possible to draw a general conclusion on the sensitivity of stored product insects to DE. The most sensitive are insects of the genus *Cryptolestes*, and the most tolerant are those of the genus *Prostephanus*. All other species fall between these two genera, ranging from the most sensitive *Cryptolestes* to the less sensitive *Oryzaephilus*, tolerant *Sitophilus* and the most resistant *Tribolium* and *Rhyzopertha*, (Maceljski and Korunic, 1972; Desmarchelies and Dines 1987; Fields and Muir, 1996; Korunic et al., 1997; Korunic and Fields, 1998, 2006). The difference among species regarding their tolerance is considerable. Under certain conditions and using a DE with high efficacy against insects, a dosage of 300 ppm (0.3 g/kg) applied over 24 hours was required to achieve a 100% mortality of *C. ferrugineus*. However, applying the same dosage under the same conditions to *T. castaneum*, 100% mortality of the insects did not occur after 21 days (Korunic and Fields, 1998).

Research has also been done on the effect of DE on numerous other insects, such as ants, bedbugs, textile pests, various caterpillars in agriculture, crickets, termites, earwigs, June beetles, potato beetles, silverfish, fleas, centipedes, pillbugs, snails, as well as poultry mites, ticks, etc. (Wilbur et al., 1971; De Crosta, 1979; Snetsinger, 1982, 1988; Rambo, 1992).

The efficacy of DE primarily depends on the physical properties of its dust, and not on its chemical composition (Korunic, 1997). The dust is physically stable



and will affect insects as long as it is dry and in sufficient concentration to ensure that insects will come in contact with enough diatom particles. Everything that reduces the ability of particles to absorb wax from insect cuticles directly reduces the efficacy of DE. An increase in moisture content of treated grain or air relative humidity (r.h.) will considerably reduce the efficacy of DE, especially if moisture content is more than 14% or relative humidity exceeds 70% (Korunic, 1994; Fields and Korunic 2000). It means that products with high moisture content should not be treated with DE or, having no other solution, higher dosages should be applied. Increased temperature increases the efficacy of DE, an exception being species of the genus *Tribolium* (*T. castaneum* and *Tribolium confusum* Jacquelin du Val). These species show greater tolerance of a temperature of 30°C than of 22–24°C (Maceljski and Korunic, 1972; Aldryhim, 1990; Fields and Korunic, 2000). Athanassiou et al. (2005) found out that the DE formulation Silicosec caused lower mortality of *T. confusum* at 32°C than at 30°C.

Korunic (1998) showed that the biological activity of one silica - Celite - increased significantly with reduced particle size, whereas no correlation was found between particle size and activity of another silica - DE 'Macedonia'. There are considerable advantages in having DEs that do not contain inspirable particles but are still acceptably efficacious against insects. These findings indicate a possible influence of DEs' particle sizes on the effectiveness against insects, which was confirmed by Vayias et al. (2009).

Some insects have greater sensitivity to DE because of their anatomy and physiology. Generally, insects with large surface area in relation to their body volume (i.e. smaller insects) are more sensitive because they lose greater amounts of water from their body. Insects with rough or hairy body surface collect more DE particles per unit area, consequently causing greater cuticle damage. Therefore, such insects are more sensitive than others (Carlson and Ball, 1962). Insects with a thin epicuticle or thin wax coat are more sensitive than those with thicker wax coats (Bartlett, 1951). Insects with soft wax coats, such as cockroaches, are more sensitive than those with hard wax coats (Ebeling, 1971). Insects that can recover the water lost, such as sucking insects and mites, are more resistant than those that must metabolized water from their food (Flanders, 1941). DE formulations can be more effective against insects if they contain high purity amorphous silica with uniformly sized diameter particles (less than 10 microns), with minimal impurities and with less than 1% crystalline silica (Calvert, 1930; Allen, 1972; Katz, 1991).

An addition of DE to grain does not change its health status, the baking qualities of flour or the quality of final products (La Hue, 1978; Desmarchelier and Davies, 1987; Aldryhim, 1990; Korunic et al., 1996b).

## THE STATUS OF DIATOMACEOUS EARTH IN INTEGRATED STORED GRAIN PEST MANAGEMENT

Integrated Stored Grain Pest Management (ISGPM) involves an understanding of how insect populations respond to grain moisture and temperature, the relationship between insect numbers and storage losses, the complex nature of the stored grain ecosystem, monitoring and sampling methods, the effects of various insecticides, and so on. For a successful introduction of DE into Integrated Pest Management, it is crucial to know factors such as grain moisture content and temperature, amounts of dockage (chaff, weed seeds) and broken kernels, grain type and quality, availability of food to insects and insect species present. These factors will often have a greater influence on the efficacy of DE than on other (synthetic) grain protectants. It is also very important to understand that there is no single control measure that is effective for an extended period of time in controlling stored-grain insects at 100% mortality level.

Certain factors have generated renewed interest in using DE as a component of ISGPM. These are:

- ⇒ consumer demand for food free of pesticide residues
- ⇒ development of insect resistance to synthetic insecticides
- ⇒ potential loss or restricted application of currently available stored-grain pesticides due to new regulations.

In the Proceedings of the 6<sup>th</sup> International Working Conference on Stored-product Protection (Inert Dusts Workshop Summary), Canberra, Australia, 17-23 April 1994, one of the conclusions was that inert dusts, mainly diatomaceous earth, should be a part of the mainstream of stored product protection. Therefore, when determining how to solve stored-grain pest problems, DE should be considered along with other tools, such as fumigants, trapping and physical methods. At the working conference three areas of DE use were outlined:

- ⇒ admixture of DE with grain
- ⇒ use of DE as a structural treatment on walls and floors
- ⇒ addition of DE to the surface of grain bulks.

It is generally thought that DE should be used as a preventive measure for grain protection and not as a

curative measure or means of disinfestation. The attributes of DE as a preventive grain protectant include:

- ⇒ prevention of pest infestations
- ⇒ reduction (over 90% to 100%) in low level grain infestations. Low level infestation is defined as an insect population too low to influence grain temperature and moisture content
- ⇒ prevention of loss of grain quantity and quality, without leaving harmful residues.

The role of DE as a component of ISGPM should be addressed in the light of these general considerations. In an effective ISGPM program, methods of prevention and control are integrated to give maximum protection of grain at the lowest possible cost.

It is important to point out that for full and effective protection of stored grain, DE should be used in conjunction with other measures, such as good housekeeping and proper management. To prevent insect infestations in uninfested grain, suppress pre-existing insect populations (over 90% mortality) and avoid loss of grain quality during storage, it is very important to know how, where and when to apply DE. DE may be successfully used to protect stored grain against insect infestations only if every step of a suitable Integrated Pest Management (IPM) program is fully implemented. Empty granaries should be cleaned and treated with DE (dusting) about 2 weeks before filling them with grain. After loading a granary, the top layer and/or surface of the grain mass should be treated with DE as a minimum treatment. It is important to note that treating only portions of the grain mass will not provide complete protection if the grain is already infested. It means that this type of treatment should be used on newly harvested grains or grains that have been fumigated. If grain is already infested in the field (maize with maize weevil *Sitophilus zeamais* M., beans and soybean with Bruchidae, etc.) this method of protection should not be implemented. In this situation, a combined treatment would be more effective. For example, the treatment of empty granaries with DE combined with phosphine fumigation and a DE surface application of the grain, or the combination of cooling and a surface application of DE.

## CASE STUDIES OF FIELD AND SMALL-SCALE FIELD COMMODITY AND STRUCTURAL TREATMENTS WITH DIATOMACEOUS EARTH

Case study data on the use of DE for commodity and structural treatment show that DE may be used as a practical alternative to some conventional pesticides in

certain applications. The case studies selected below refer to the use of DE under field conditions. Numerous other experiments conducted in laboratories in several countries are not included in the case studies.

### Commodity treatment

*Case study 1.* Field tests conducted by Agriculture and Agri-Food Canada (AAFC), Cereal Research Centre, Winnipeg, Canada

In 1994 and 1995, the Cereal Research Center in Winnipeg conducted laboratory and field tests on the efficacy and various other properties of the enhanced diatomaceous earth (EDE) Protect-It®.

In 1994, field trials were conducted using three metal granaries at the AAFC Experimental Farm in Glenlea, Manitoba. At the beginning of August, 40 tons of Hard Red Spring wheat was dusted with 300 ppm of Protect-It. After treatment, *C. ferrugineus* and *T. castaneum* adults were introduced onto the top surface of wheat bulk. In October, the *C. ferrugineus* population was reduced to zero and the *T. castaneum* population was reduced by over 95%, as compared to the untreated grain.

During the fall of 1995, field trials were conducted at three sites in southern Manitoba. The trials were conducted in 27 to 80 tons metal granaries with 16 to 20 tons of wheat in each granary. The grain used was Hard Red Spring wheat harvested in August and September of 1995. To insure infestation, *C. ferrugineus* and *T. castaneum* were released on the surface of the grain mass after treatment with Protect-It®. Insect populations were measured by using both probe pitfall traps and by extracting insects from wheat samples taken from the granaries. In addition, there were two indirect measures of efficacy: the mortality of insects confined to jars held on treated wheat in the granaries, and bioassays conducted in the laboratory on wheat taken from the granaries. At each site there was an untreated control granary and 75 ppm and 100 ppm dust applications. There was also an aqueous spray application of 100-ppm Protect-It at two sites.

In general, *C. ferrugineus* populations were consistently controlled by all three treatments. *C. ferrugineus* populations in the treated bins, as measured by probe pitfall traps, were reduced by 87.5-99.8% in comparison to the control populations. *T. castaneum* populations were reduced by 0-81.9% in comparison to the control populations. When measured by sieving samples from the granaries, rusty grain beetles were reduced by 92-100% and red flour beetles by 70-100%. *C. ferrugineus* and *T. castaneum* populations were reduced by

99-100%, and 56-100%, respectively, as determined by Berlese funnel extractions. Regarding the jars held in the granaries, rusty grain beetle mortality ranged from 93% to 100% and red flour beetle from 1% to 89%.

It was concluded that *C. ferrugineus* was consistently controlled by all three treatments (75 and 100 ppm dust and 100 ppm spray). *T. castaneum* populations were reduced but not controlled (control defined by at least 90% mortality) by the most effective treatment, which was the 100 ppm dust application. To control *T. castaneum* on Grade 1 Hard Red Spring wheat, at least 300 ppm must be applied (Fields and Timlick, 1995; Fields et al., 1996).

*Case study 2.* Tests conducted in Croatia by the Croatian Plant Protection Institute

In 1996, Protect-It was tested under field conditions at two sites in Croatia (Bertovic, 1997; Hamel, 1997). A preventive treatment of wheat was performed at one site immediately after harvest. At another site, wheat was treated after the first cleaning, one and a half months after harvest. DE was applied as a dust (100 ppm) and as an aqueous spray (150 ppm). The results indicated that 100 ppm controlled *Cryptolestes* spp., and greatly reduced *S. oryzae* numbers. The applied concentrations were not high enough to provide acceptable control of *T. castaneum* or *R. dominica*.

*Case study 3.* Large-scale laboratory tests with EDE Protect-It in Italy

Large-scale laboratory tests were carried out at Regione Emilia Romagna, Servizio fitosanitario, Ravenna, during 1997. Wheat was dusted with 300 and 600 ppm of Protect-It. After treatment, rice weevil, lesser grain borer and red flour beetle were exposed to treated grains for 4 weeks. Based on the results, the authors recommended a dose of 300 ppm for rice weevil and 600 ppm for lesser grain borer and *T. castaneum* (Contessi and Mucolini, 1997).

*Case study 4.* Field and laboratory experiments with EDE Protect-It in China

The effectiveness of Protect-It was studied under field and laboratory conditions in Sichuan and Guangdong provinces, P.R. China, in 1997. The tests were conducted using three stored-grain insect pests in paddy rice and wheat. Insect species used in the experiments were *S. zeamais*, *T. castaneum* and *R. dominica*. The test grains were: soft wheat, produced locally, with moisture content of 12.0% (Sichuan); No. 1 Canada Western Hard Red Spring wheat with 12.0% moisture content (Guangdong) and paddy rice, produced locally at each site, with 12.9% moisture content. Two pesticides

were tested: Protect-It, produced in Canada, and Fenitrothion 65% EC (fenitrothion), produced in China.

Conditions at the Sichuan grain storehouse ranged from 24.0 to 28.0°C and from 66 to 76% r.h. At the Guangdong site, air temperatures in the wheat storehouse ranged from 26.0 to 30.5°C, with 69 to 88% r.h. Air temperatures in the paddy rice storehouse ranged from 27.0 to 31.0°C, with relative humidity from 69 to 92%.

Bioassay results both for the field-treated and laboratory-treated grain demonstrated a good residual activity of Protect-It. An analysis of adult and progeny mortality showed that the following concentrations of Protect-It were comparable in efficacy to 8 ppm of fenitrothion, and caused 90 to 100% mortality of test insects:

1. 300-500 ppm to control *S. zeamais* and *T. castaneum* on wheat and paddy rice
2. 500 ppm to as much as 700 ppm to control *R. dominica* on wheat
3. 300-500 ppm to control *R. dominica* on paddy rice.

Protect-It concentrations that controlled adult insects also substantially reduced the production of progeny (by over 90 to 100%) in all three species tested and on both commodities (Zeng et al., 1998).

*Case study 5.* Insect control on stored malting barley with diatomaceous earth in Southern Brazil

A field test was carried out in 90 ton metallic granaries using a salt water diatomaceous earth formulation from Brazil. DE was applied over the malting barley on the conveyor belt as a dust and as a slurry at the concentrations of 1500 ppm and 300 ppm, respectively. Malting barley was stored for 6 months. The malting barley treated with DE had very low numbers of insects (*Sitophilus* spp.). However, insect populations were very high on untreated barley. The results indicated that DE can be used to control insects on malting barley, either as dust or slurry, with an advantage of being non-toxic and without leaving any toxic residues (Rupp et al., 1998).

*Case study 6.* Layer treatment – a small-scale field test conducted in Canada

Dosage rates of DE required to control most economically important stored product pests have undesirable effects on the handling properties of grain when applied to the entire grain mass. A more acceptable strategy is to only apply DE to a portion of the grain mass. The objectives of a layer treatment study conducted in Ontario, Canada, by Korunic and Mackay (2000) were to evaluate the efficacy of DE Protect-It, applied as a surface layer treatment of Hard Red Spring wheat (HR-SW), against three insect species and to determine the

optimum dilution ratio of treated and untreated grain in order to mitigate adverse effects of DE on grain bulk density (test weight). All treatments (750 ppm, 50 cm and 100 cm layers; 500 ppm, 50 cm and 100 cm layers) were successful in significantly reducing the numbers of *S. oryzae*, *T. castaneum* and *R. dominica* by more than 98 percent relative to their respective controls.

Treatment with 500 and 750 ppm of Protect-It reduced the test weight of HRSW by 4.9 and 5.2 kg/hl, respectively. The test weight of a 1:4 (treated : untreated) dilution of grain treated with 500 ppm was comparable to the test weight of grain treated with 750 ppm. The test weight of a 1:9 dilution of grain treated with 750 ppm was comparable to the test weight of grain treated with 500 ppm.

It was concluded that a 100 cm surface layer treatment with 500 ppm of Protect-It is sufficient to control *S. oryzae*, *R. dominica* and *T. castaneum* (population reduction over 95%) and that no more than 20 percent of the total grain mass should be treated in order to minimize bulk density reduction.

## Structural treatment

*Case study 7.* Application of DE Protect-It to empty grain bins

Protect-It is recommended for surface treatment of empty grain bins to eliminate or reduce residual pest insect populations that may contribute to infestation of newly harvested and stored grain. The objective of this study was to determine the efficacy of Protect-It against grain insects when applied to empty farm bins at two different rates, 3 and 7 g/m<sup>2</sup>. Tests were conducted during the late summer of 1997 in central Oklahoma, a major wheat-producing state of the USA. The insect species tested were *S. oryzae* and *R. dominica*. The general approach to this study was to apply different rates of Protect-It to two identical grain bins. Test insects were put in a number of test cages which were placed on the bin floor. Before insects were introduced into the cages, the entire bins, including open test cages, were treated with Protect-It as prescribed on the product label. The placement and treatment of cages in the granaries exposed the insects to environmental conditions found in the field. Insect mortality in treated cages was compared with that of insects held in untreated cages.

Temperatures and relative humidity were continually recorded in each bin using a recording hygrothermograph placed in the center of granary floor. Temperature and humidity inside the two bins were very similar throughout the study. Temperature recorded at the

centers of bin floors ranged from 16.5 C to 39°C, and relative humidity ranged from 34 to 100%.

The lower tested dose of Protect-It (3 g/m<sup>2</sup>) caused between over 90% and as much as 100% mortality of test insects after one week under experimental conditions. The application of Protect-It to empty bins in this study was relatively easy and effective (Phillips and Bonjour, 1997).

*Case study 8.* Activity of Protect-It in empty bins against two stored product pests

The objective of this test was to determine the efficacy of Protect-It as a general structural insecticide for grain bins. The test insects were *T. confusum* and *S. zeamais*. Although *T. confusum* is not found in grain bins, it was chosen because of the availability of suitable lab cultures and because it would be a stringent test for Protect-It in the field. Lab tests had shown that *T. confusum* was one of the species most tolerant to diatomaceous earth pesticides.

Two commercial 8000-bushel (approximately 218 tons) bins, measuring 30 feet (9.1 meters) in diameter by 14 feet (4.3 meters) high, were used in the experiment. The bins were located near Purdue University (West Lafayette, Indiana). The bins were equipped with full floor aeration systems. DE Protect-It was applied on September 10, 1997, by blowing a measured amount through the aeration fan. The deposition rates of 3 g/m<sup>2</sup> for bin 1 and 7 g/m<sup>2</sup> for bin 2 were based on calculations of bin interior surface areas.

Protect-It controlled (100% mortality) *S. zeamais* at the rate of 3 g/m<sup>2</sup>, and *T. confusum* (94% mortality) at 7 g/m<sup>2</sup>. *S. zeamais* was found more susceptible to Protect-It than *T. confusum*. The product was able to accommodate the ranges of temperature and humidity encountered during the test. It was also able to overcome the added pressure of having food available to the insects. Blowing Protect-It into the bins using the aeration fans posed no difficulty and was a suitable method of application (Mason, 1997).

*Case study 9.* Structural treatment with DE slurry: an integrated component of GRAINCO's IPM strategy

GRAINCO, the central grain handling organization in Queensland, Australia, conducted a series of trapping trials in several bulk grain stores to assess the effectiveness of slurry applications of the DE insecticide Dryacide as a structural treatment strategy to replace residual insecticides traditionally used to disinfest storehouses. The results confirm that DE slurry treatment is an effective replacement for residual chemicals as a structural treatment. Structural treatment with a

DE slurry is now an integral component of GRAINCO's IPM plan, which incorporates insecticide resistance management and is characterized by a reduced reliance on residual grain protectant chemicals (Bridgeman, 1994; Bridgeman and Collins, 1994).

*Case study 10.* Diatomaceous earth combined with heat to control insects in structures

The objective of the study was to examine a combined impact of high temperatures and DE on the mortality of *T. confusum*. Based on a success in combining phosphine, carbon dioxide and heat, the effects of combining heat and diatomaceous earth treatments were recently investigated (Fields et al., 1997). A field trial was conducted in Peterborough, Ontario, Canada, at a breakfast food manufacturer during their regular heat treatment.

The trial employed several different methods to apply diatomaceous earth to a food processing plant that routinely uses heat to control infestations. The most effective method was an electric powered duster that uses a blower to create a cloud of dust. The duster left a fine, evenly distributed coat of 1 to 2 g/m<sup>2</sup> of DE on the floor in the mill. *T. confusum* adults exposed to DE in the power dusted area died at an average temperature of 40±1°C, whereas insects in the same area, but not exposed to DE, died at an average temperature of 46±1°C. The combination of DE and heat also gave more rapid control, with the median time of death occurring 17±1 hours after the beginning of heat treatment of DE treated insects, compared with 35±1 hours for insects in the area that only received heat. It should be noted that the conditions were ideal for DE activity. Relative humidity in the plant was very low, between 10 and 20%. The lower the relative humidity, the better DE works. Also, insects were confined to rings on the floor, and were not able to escape the treated area. Although this trial was only done on a small scale, further tests are to be conducted on a larger scale. The authors concluded that the use of DE Protect-It and heat was a promising alternative to methyl bromide (Fields et al., 1997).

*Case study 11.* Combination of cooling with surface application of DE to control insects

Cooling with ambient air has been regarded in some regions of the world as useful, but in other cases insufficient for complete insect control. One of the key elements of an improved strategy may include supplementing cooling with other control methods. Nickson et al. (1994) discussed a number of cases where a surface application of the DE Dryacide to grain in several Australian

storehouses was supplemented with cooling. In Victoria in 1992, aeration in silos controlled insects except for the top 10 cm of grain bulk. A subsequent application of DE eradicated the remaining insects. Experiments were also conducted in 1993 and 1994 at other locations and no live insects (*S. oryzae*) were found in grain probe traps 2 months after surface application of DE. The combination of cooling and DE surface treatment is currently in commercial use although careful management is required for success (Nickson et al., 1994).

*Case study 12.* The use of DE grain surface treatment as a gas (PH<sub>3</sub>) barrier

Phosphine (PH<sub>3</sub>) is a well-known chemical alternative to methyl bromide that is used for commodity and structural fumigation. In order to develop and improve phosphine-based processes for pest control in stored commodities, the use of Dryacide as a surface treatment in combination with phosphine fumigation was evaluated (Winks et al., 1994). The results achieved in the majority of situations showed that DE was effective in gas tight tarpaulin. A trial conducted in a 3800 t horizontal storage at Kingsvale in late 1992 indicated that a layer of DE on the surface of the grain was as effective as it had been in many vertical silos (Winks et al., 1994).

## LIMITATIONS IN USE

One of the conclusions of the 6<sup>th</sup> International Working Conference on Stored-product Protection (Inert Dusts Workshop Summary), Canberra, Australia, 17-23 April 1994, was that inert dusts, mainly diatomaceous earth, should be a part of mainstream stored product protection. However, nowadays, because of a significant effect on grain handling and quality, the status of DE is rather different.

Despite numerous advantages of diatomaceous earth, its use to control stored-product insects remains limited because of some very serious obstacles and disadvantages. The main limitations of DE are: a reduction in grain flowability, reduction in bulk density (test weight) of grain, ineffectiveness in some situations, workers' discomfort due to airborne dust and health concerns over the presence of crystalline silica.

DE adheres to the surface of kernels and increases friction between grains. This causes increased angles of repose and decreased bulk densities (Korunic et al., 1998). DE applied at a rate of 500 ppm causes a decrease of about 6 kg/hl in bulk density of wheat, barley, oats, rye or corn. Also, the source of DE affects how much the

bulk density is reduced. There can be as much as a four-fold difference in reduction in bulk densities between DE sources (Korunic et al., 1998). Unfortunately, DEs that are the most effective insecticides are also the ones that reduce bulk densities the most (Korunic, 1997).

As desiccation is the mode of action, diatomaceous earth does not control insects in moist grains as well as in dry grains (La Hue, 1978). Unlike a fumigant, it will not control the immature stages that remain within the kernel, e.g. *Sitophilus* spp (Subramanyam and Roesli, 2000).

Application of inert dusts can be undesirable because of the dust generated. To alleviate this, aqueous applications for surface treatments are used in Australia (Bridgeman, 1994), although this somewhat reduces the effectiveness of inert dusts (Maceljski and Korunic, 1972).

DE can be used as a mild abrasive and there is concern over an increased wear on grain handling machinery. However, DE is relatively soft, having an index of 1 to 1.5 on Moh's hardness 1 to 10 scale, and it is softer than silver (2.5-4), copper (2.5-3), nickel and iron (5), quartz (7) and diamond (10) ([http://www.tedpella.com/company\\_html/hardness.htm](http://www.tedpella.com/company_html/hardness.htm)).

Tests need to be conducted to determine if DE does increase the actual wear of grain handling and milling equipment.

Depending on the source and processing, DE can contain anywhere from 50 to 0.1% crystalline silica, although DEs registered as insecticides generally have less than 6% crystalline silica, or in some countries less than 1%. Crystalline silica has been shown to be carcinogenic if inhaled (IARC, 1997). However, the use of proper dust masks, or the use of low crystalline silica DE can protect against this health risk (Desmarchelier and Allen, 2000).

## FUTURE OF DIATOMACEOUS EARTH

In the last decade, there has been an increase in the use of diatomaceous earth because of its low mammalian toxicity, worker safety, low risk food residues and the occurrence of resistant insect populations associated with the use of chemical insecticides.

In order to reduce DE dosages that have adverse effect on grain quality, DE is often mixed with other compounds such as silica gel, dry honey, unactivated yeast and sugar to increase the efficacy (Quarles and Winn, 1996; Korunic and Fields, 1998; Subramanyam and Roesli 2000). However, high doses of these mixtures still have a significant negative effect on grain bulk density and

flowability (Jackson and Webley, 1994; Korunic et al., 1998). Possible solutions for the implications of high doses of DEs include a combined use of DE and other low-risk methods, such as extreme temperatures (Fields et al., 1997; Dowdy 1999), grain cooling with surface treatment with DE (Nickson et al., 1994), or a mixture with entomopathogenic fungi (Lord, 2001; Akbar et al., 2004; Kavallieratos et al., 2006; Vasilakos et al., 2006; Michalaki et al., 2007), a mixture with synthetic insecticides (Korunic, 2001; Stathers, 2003; Arthur, 2004a, 2004b; Athanassiou, 2006; Chanbang et al., 2007; Korunic and Rozman, 2010) or a mixture with plant extracts (Korunic, 2007b, Athanassiou and Korunic, 2007). Experimentation with other components often revealed synergistic or enhanced effectiveness (Korunic, 2001; Lord, 2001; Stathers, 2003; Athanassiou and Korunic, 2007; Korunic, 2007b; Korunic and Rozman, 2010).

If these newer enhanced formulations can respond to the limitations of diatomaceous earth, there will be a wider adoption of diatomaceous earth to control stored-product insect pests. To address the respiratory health concerns associated with crystalline silica, we expect that these new formulations will have less than 1% crystalline silica and only a minor fraction of particles in the range of up to 10 microns (respirable dust) or preferably even less of 5 micron particles (able to enter the lungs). Diatomaceous earth will come from deposits that have been rigorously tested to insure high efficacy and safety and should be combined with additives to enhance activity and reduce dosage.

Resistance to residual insecticides has been one of the reasons to search for alternatives to chemical insecticides. Laboratory experiments have shown that several stored-product pests can have up to 2-fold reduction in susceptibility when exposed to diatomaceous earth for 5-7 generations. Although there are no reports on cases of insects developing resistance to diatomaceous earth in commercial stores, these results suggest that it will be necessary to use resistance management strategies to prevent widespread resistance to diatomaceous earth products.

## CONCLUSIONS

Although DEs have some disadvantages, there are considerable practical advantages in their use to control insects.

The advantages include a possibility of dust removal before consumption, long lasting effect (persistence), acceptable margins for safety if proper precautions are

taken, and a mode of action against pests that is different to the mode of action of conventional insecticides. Some of the very important disadvantages include an undesirable effect on bulk density (test weight) and grain handling properties.

With the approaching of stricter regulations dealing with the use of synthetic grain protectants, we must turn to a more systemic approach to pest control. The use of Integrated Pest Management is becoming the primary systemic approach used by stored product managers.

While the use of DE-based insecticides cannot be interpreted as a sole replacement or alternative to other more toxic synthetic conventional grain protectants, it is an integral part of many IPM strategies. Effective IPM strategies that include a common use of modified and enhanced DE-based products offer an alternative to synthetic insecticides.

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## Dijatomejske zemlje – prirodni insekticidi

### REZIME

Zakonskom regulativom za dijatomejsku zemlju (DZ) pokrivena su tri osnovna područja: sigurnost potrošača, sigurnost radnika u proizvodnji i dokazi efektivnosti na insekte. U području sigurnosti potrošača zakonski propisi su slični kao i za ostale aditive, s time da je osnovna prednost upotrebe DZ mogućnost njenog uklanjanja s tretirane robe tokom procesa obrade. U području sigurnosti radnika u proizvodnji propisi su slični onima koji se primenjuju i za ostala prašiva, na primer za kreč. Efektivnost insekticida može se dokazati primenom standardizovanih bioloških oglada, kao i analizom fizičkih i hemijskih osobina DZ i delovanjem DZ na zrnaste biljne proizvode. Sistem integralnog suzbijanja štetočina baziran na znanju omogućava postepeno smanjenje upotrebe sintetičkih, konvencionalnih insekticida. Glavni princip integralnog sistema u zaštiti uskladištenih poljoprivrednih proizvoda se primarno zasniva na prevenciji, tj. sprečavanju infestacije proizvoda, a posebne mere primene sintetičkih insekticida primenjuju se samo u slučaju kada dođe do infestacije robe. DZ i modifikovane, tj. efikasnije DZ imaju sve važnije mesto u sistemu integralne zaštite uskladištenih poljoprivrednih proizvoda i važnu ulogu u postepenom smanjivanju upotrebe sintetičkih insekticida. Uvođenjem DZ u sistem integralne zaštite, zrnasta roba je zaštićena od infestacije, gubitak uzrokovan insektima je smanjen, a kvalitet robe je očuvan sve do trenutka tehnološke obrade. Već brojni opisani slučajevi korišćenja DZ za tretiranja zrnaste robe, kao i raznih praznih prostora pokazuju da je DZ u pojedinim slučajevima moguća alternativa za sintetičke insekticide.

**Ključne reči:** Dijatomejske zemlje; bezbednost; insekticidi; uskladišteni proizvodi