# PROJECT PROPOSAL

## AGRICULTURAL ENGINEERING

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**EFFICACY OF LOCALLY MINED DIATOMACEOUS EARTHS AS MAIZE GRAIN PROTECTANTS AGAINST MAJOR STORAGE INSECT PESTS IN SMALLHOLDER STORES UNDER SUB-HUMID CONDITIONS.**

**BY**

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# 1.0. INTRODUCTION

## 1.1. Background

Insects are a major cause of post harvest grain losses (Hardcastle, 1969). By boring within the kernels and feeding on the surfaces they remove food material and in most cases selecting highly nutritive fractions (Subramanyam *et al*, 1994). In addition to destroying stored products directly by feeding, storage insects can cause heating and moisture accumulation in stored grain because of the respiration of large populations. This can cause 'hotspots' and can result in the lowering of grain quality and spoilage by reducing germinability, increasing fungal and bacterial infection, charring of seeds and increasing the free fatty acids levels of the stored products. The extent of food damage varies with type of grain, insect species, and its rate of increase, the voracity of the larvae and the temperature, moisture and the nutritive value of the stored product. According to (Giga *et al*, 1991), the most prevalent storage pests of maize grain are *Sitophilus* spp., *Sitotroga cerealella* and *Tribolium castaneum*.

Unless insect control measures are applied, grain quality and value are likely to be reduced since the grain becomes unsaleable, goes mouldy and sometimes can be reduced to dust by the insects feeding on it. Small-scale farmers may lose up to 15% of their stored maize due to insects during one storage season of 8-10 months (Giga *et al*, 1991). Such losses can be directly and indirectly translated into monetary losses with considerable socio-economic and political repercussions. Prevention of these losses would result in improved household food security, control over sales and more food for the non-farming populations. The use of chemical insecticides if properly applied at the right time can cause a rapid death to most insects thereby minimizing insect problems (Hill, 1978). However, Quarles (1996) revealed that resistance has developed world wide to the chemical method. Fumigation also minimizes insect problems but small-scale farmers fail to adopt the system because their storage facilities are not air tight, they lack the equipment required for application and they are not trained on how to apply the fumigants.

Mvumi *et al* (1995) noted that in Zimbabwe at least 75% of the small-scale farmers use synthetic insecticides to protect their grain from attack by storage insect pests. However the rising cost of synthetic insecticides, the development of insecticide resistance (Giga *et al*, 1986), contamination of food stuffs with residues and exposure of users to toxic chemicals (Hill, 1978) contribute to failure by farmers to appreciate their relevance.

## 1.2. Diatomaceous earth as grain protectants

Inert dusts particularly diatomaceous earths offer an alternative to synthetic insecticides (Golob, 1997). Diatomaceous earth is obtained from deposits of diatomite fossilized sedimentary layers of tiny phytoplankton single celled small plants called diatoms many of them originating twenty million years ago in lakes and seas (Fields *et al*, 2000). The inert dust absorbs the oily or waxy outer cuticle layer by direct contact resulting in dehydration and death. In addition to its desiccant action, it works abrasively to rupture the cuticle of insects rendering it permeable to water and insects die if they lose sixty percent (60 %) of their water, which is approximately thirty percent (30 %) of their body weight (Ebeling, 1971).

The insecticidal properties of diatomaceous earth vary depending upon its geological origin, application rate, particular pest problems, grain moisture content, relative humidity and length of storage period; hence their efficacy may vary with geographical location and climatic conditions (Stathers *et al*, 2002b)

In Zimbabwe deposits of diatomaceous earth identified in the Zambezi and Limpopo valleys have shown from preliminary tests to have activity against storage insect pests. However, Stathers *et al* (2002a) reported that information on its efficacy in smallholder stores under sub-humid conditions is still very limited despite successful trials on imported diatomaceous earth Protect-It and Dryacide. Consequently, effectiveness of the local diatomaceous earth may spearhead mining and processing of the products in Zimbabwe.

## 1.3. Justification

The rising cost of synthetic insecticides, the development of insecticide resistance (Giga *et al*, 1986), safety of consumers, and availability of sources of local diatomaceous earth make it necessary to evaluate the effectiveness of the local diatomaceous earth (Stathers *et al*, 2002b).These would be exploited to provide a most cost effective source of inert dust as well as well documented information on their formulations which is currently lacking.

Diatomaceous earths provide a safer alternative to chemical insecticides since they are less toxic, inert and contain no major active ingredients (Golob, 1997). They are also easy to apply and they require no specialized equipment during treatments. They can be used on grain that can be used as feed or food. However, they tend to reduce grain bulk density and fluidity (Fields et al, 2002), making them unsuitable and unbeneficial to highly mechanized storage but at the same time they can be of benefit to small-scale on farm storage .Grain treated with diatomaceous earths can be separated easily from the dust. Thus the grain can be consumed at will implying that there is no withholding period. In theory the dust can be reusable because once separated it can be reapplied in storage bins.

The study will reinforce the previous study by Janga (2004, unpublished) since it will factor in variability between seasons i.e. differences in climatic conditions and grain moisture content for the two previous levels of 0.20% and 0.25% for Chemutsi and the possibility to use a lower admixture rate of Chemutsi 0.15%, which was not included in the previous study. Another local diatomaceous earth Beitbridge will be evaluated. The storage period will also be lengthened from the previous October to May to the normal storage period (July to April) so as to obtain more reliable information on persistence of the diatomaceous earth.

Anticipated effectiveness of the local diatomaceous earth may help to initiate the registration process. The effectiveness and possible adoption of the local diatomaceous earth stand not only to benefit small holder farmers but the nation at large since it may also influence the private sector into mining and processing of the diatomaceous earth resulting in a bout of the local industry, creation of employment and the scarce foreign currency from exports to other sub-humid countries in Africa where grain is stored on farm.

Apart from protecting maize grain, diatomaceous earth can be used in the purification of water, as a filtering agent for swimming pools, filtration of commercial fluids and clarification of liquors and juices (Korunic, 1997).

## 1.4. Objectives of the study

The overall objective of the study is to evaluate the efficacy of the locally mined diatomaceous earths against major storage insect pests under sub-humid conditions. The specific objectives are:
a) To determine the efficacy of local diatomaceous earths and compare it with commercial diatomaceous earth.

b) To identify the most appropriate application rate.

c) To assess the effect of environmental conditions on the efficacy of the diatomaceous earths.

## 1.5. Hypotheses

The study tested the following hypotheses:
a) Locally mined diatomaceous earths are as effective in protecting bulk maize grain against storage insect pests as commercial diatomaceous earth Protect-I t.

b) Increase in relative humidity reduce efficacy of diatomaceous earth.

# 2.0. LITERATURE REVIEW

## 2.1. Evolution of the diatomaceous earth

It has been known for many years that stored grain can be kept free from infestation by the common grain weevil (*Calandra granaria*) by mixing it with small amounts of certain mineral powders such as silica (Kitchener *et al*, 1943). The Chinese used diatomaceous earth for pest control 4000 years ago and the American Indians often stored grain with dust to protect it. Over the years diatomaceous earth have been used on both laboratory and field experimental basis for grain protection (White *et al*, 1975), and have been found to have a toxic effect on insects due to their ability to adsorb the lipid layer from the insect cuticle leading to dehydration (Ebeling, 1971).

Diatomaceous earths are registered as grain protectants in Australia, United States of America, Canada, China, Germany and many Asian counties under various trade names such as Dryacide and Perma Guard (Quarles and Winn, 1996) after extensive research on their efficacy. The use of these diatomaceous earths is slowly creeping into many developing countries in Africa where deposits of the earth are lying idle with no exploration and realization of their potential as grain protectants. Thus diatomaceous earth marketing and research companies have opportunities to incorporate these deposits into newer and more effective products.

## 2.2. Advantages of diatomaceous earths

Diatomaceous earth has low toxicity to vertebrates; $LD_{50}$ > 5000mg/kg (Subramanyam *et al*, 1994). Quarles (1996) further indicated that a wide variety of wildlife (except insect pests) have been exposed to the diatomaceous earth for millions of years without developing chronic skin, eye, digestive or reproductive problems. The only problem noted by the same research scientist is silicosis which is due to long term inhalation of the dust by mine workers. Quarles (1996) cited MacDonald (1989) to indicate that the exposure of the diatomaceous earth in storage structures and facilities can be solved by use of dust masks.

Quarles (1996) described the diatomaceous earth as chemically stable. The implication is that the dust does not dissipate with time and its effectiveness does not diminish for as long as it is dry. As a result, diatomaceous earth products can be relatively inexpensive compared to other synthetic insecticides which might require repeated application to achieve long term protection from insects (Quarles, 1996). Furthermore, diatomaceous earths have no adverse effects on milling, baking and malting qualities at application rates below 300 ppm (Korunic, 1996). They are also easy to apply and they require no specialized equipment during treatments. They can be used on grain that can be used as feed or food (Golob, 1997).

Subramanyam *et al* (1994) indicated that insecticide resistance in stored grain to date has not been documented. Ebeling (1971) attributed the unlikelihood of resistance to diatomaceous earth to the basis that the physical and not chemical properties render it effective. Strains of the red flour beetle showed 240 – fold resistance to deltametrin, a chemical insecticide (Subramanyam and Hagstrum, 1996). Resistance is less of a problem with fumigants e.g. methyl bromide and phosphine but these are dangerous toxic gases which are a threat to the ozone layer (Taylor, 1996). However, insects may avoid deposits or products with diatomaceous earths but Quarles (1996) associated it to the repellency properties of the diatomaceous earth and not the behavioural resistance of the insects.

## 2.3. Disadvantages of diatomaceous earths

Korunic *et al* (1996) in Quarles (1996) attributed high application rates to excessive dust production during handling. This may result in flowable properties of grain being affected, test weight reduced, grain quality lowered, and high costs of maintenance of machinery ( due to wear and tear) being incurred (Fields, 2002). As a result, both the grain and milling industry may not accept grain treated with diatomaceous earth (Subramanyam *et al*, 1994). Therefore high rates may not be recommended for commercial grain but rather for smallholder grain storage where grain handling is not highly mechanized since low application rates despite reducing dust problems reduce efficacy. However experiments in recent years as described in Quarles (1996) reveal that wet application (dust applied as slurry instead of powder) and top dressing of the diatomaceous earth with combinations of aeration reduces the problems associated with dust.

## 2.4. Mode of action of diatomaceous earth

Despite considerable research on the use of mineral dusts as insecticides, the mode of action is not satisfactorily established. There has been considerable controversy as to whether the powders cause lethal dehydration by the adsorption of the oil or wax layer or by abrasion of the cuticle (Ebeling, 1961). A number of investigators proposed that when insects crawl over layers of finely divided particles of dry dusts, they are susceptible to desiccation (Zacher and Kunicke, 1931, Cotton and Frankenfeld, 1949). Ebeling and Wagner (1959) indicated that this desiccation results from the removal by these fine dry small particles of some of the very thin lipid layer of the cuticle that protects insects from abnormal rapid water loss.

However, Wigglesworth (1945) attributed the desiccating action of the finely divided powders entirely to their ability to remove the protective lipid layer by abrasion. Therefore if the protective wax can be removed only by abrasion as noted by Wigglesworth (1945) and Beament (1945), the use of inert dusts for the control of insects through desiccation must be confined to those species primarily granary weevils which abrade their bodies as they crawl among the kernels. Highly sorptive dusts have however proved to be superior for such insects (Cotton and Frankenfeld, 1949). Korunic (1998) also stated that the other mode of action is the insect repellence caused by the physical presence of the dust. Despite these conflicting results, Ebeling's (1971) hypothesis of the physical chemistry of the wax remains speculative. The objective of the study however will be focused on effectiveness of the diatomaceous earths irrespective of their mode of action.

## 2.5. Factors affecting efficacy of diatomaceous earth

A number of factors affect insect desiccation and thus the efficacy of the diatomaceous earth. The properties of the dust generally correlated with insect desiccation are their specific surface area, their pore size, oil absorption capacity, species of diatoms and their chemical compositions (Quarles, 1992, Le Patourel and Singh, 1984). Finely divided powders increased in their ability to kill insects as they decreased in particle size down to the optimum size of about 1 micrometer (Alexander *et al*, 1944). However, comparative efficacy studies in the laboratory by Subramanyam (1995) on commercial diatomaceous earths (Protect-It, Insecto, Dryacide and Perma-Guard) indicated that efficacy did not correlate with particle size. Therefore further studies are needed to clearly reveal the correlation between efficacy and particle size.

Published research by Ebeling (1961), indicate that the primary mode of insecticidal action of the diatomaceous earth is the adsorption of the lipids from insect epicuticles. To achieve a lethal rate of desiccation, sorptive dusts need to remove the lipid at a faster rate than it can be replaced by insect’s synthesis. The rate of lipid removal thus is a function of both the capacity of the dust to adsorb the lipid and the rate at which saturated aggregates are lost and fresh dust picked up by the grain (Le Patourel, 1986, Ebeling, 1971). Therefore the greater the adsorption capability of the diatomaceous earth, the greater the efficacy is likely to be. However there is little research published correlating any single physical parameter and efficacy although oil adsorption capacity is an easily measured parameter (Quarles, 1992a). Further work therefore need to be done on the correlation between the physical parameters and efficacy as this can be used to identify diatomaceous earths deposits that will be more effective for insect control.

The lethal effect of the diatomaceous earth is also dependent upon the ambient temperature, relative humidity of the environment and the grain moisture content (Le Patourel, 1986). The effectiveness of the diatomaceous earth according to Le Patourel (1986) is limited to admixture with relatively dry grain (< 14% moisture content) and regions of low relative humidity (< 70%). This does not imply that the diatomaceous earth becomes less effective as a desiccant, but because the insects have a constant source of water to replace their losses (Korunic, 1996). Higher temperatures make the diatomaceous earth more effective, as water loss is enhanced (Le Patourel, 1986).

Insect physiology and morphology also affect the efficacy of the diatomaceous earth. Quarles (1992) reported that treatment was most effective for hairy insects with large surface area to volume ratios such as the rice weevil, maize weevil and the granary weevil. Larger application rates were needed for smooth beetles such as the confused flour beetle and red flour beetle.

The species of the diatoms, freshwater or marine diatoms determine the physical properties of the diatomaceous earth and ultimately the efficacy. According to Subramanyam (1995), freshwater diatoms are more uniform in shape i.e. smaller range in shape and size than marine diatoms. Diatomaceous earth products from marine diatoms have a larger number of smaller irregular particles than freshwater products; hence they are more effective (Quarles, 1996). Katz (1991) noted that it is a belief among many pest control specialists that freshwater diatoms have a higher electrostatic attraction than marine diatoms hence they are more effective. However, as indicated by Quarles (1992a) and Subramanyam *et al* (1994), the hypothesis needs further investigation because the registered commercial diatomaceous earths Insecto, a marine diatomaceous earth product have an oil adsorption value of 175 % while a freshwater diatomaceous earth shows adsorption of about 116 %. Furthermore, electrostatic attraction is probably less important than oil adsorption capacity since pest insects in stored products are always exposed to the dust.

In general, the higher the application rate of the diatomaceous earth, the more effective, as the chances of the insect encountering the desiccant are increased (Ebeling, 1971). The type of grain determines the application rate since the dust must adhere to the grain before giving protection. Thus, a higher application rate is required to protect maize grain than wheat (Ebeling, 1971, Aldryhim, 1993).

## 2.6. Laboratory trials on efficacy of diatomaceous earth.

Stathers *et al* (2000a) studied two commercially registered diatomaceous earths (Dryacide and Protect-It) for their efficacy and persistence against four common tropical storage pests (*Prostephanus truncatus, Sitophilus zeamais, Callosobruchus maculatus* and *Acanthoscelides obtectus*). The diatomaceous earths products were admixed at different application rates and relative humidities under controlled temperatures using host commodities. Dryacide was tested at 0.1% w/w (recommended Australian application rate) and an untreated control was also included. Protect-It at 0.1% and 0.15% w/w for *P.truncatus* and at 0.05% and 0.1% w/w for *S.zeamais* were used for maize commodity. All treatments were tested at both 50% and 60% relative humidity.

Persistence was considered after 3 and 6 months of storage by assessing adult insect mortality rate. At both humidities, Protect-It at either application rate caused higher mortality of both *P.truncatus* and *S.zeamais* than Dryacide. However, the diatomaceous earth products were more effective at 50% relative humidity than at

60% relative humidity confirming reports by Carlson and Ball (1962), Le Patourel (1986), and Maceljski and Korunic (1971), that increase in grain moisture content or relative humidity reduces efficacy. Korunic (1998) indicated that laboratory trials are conducted under rigidly controlled experimental conditions and provide little information on the effectiveness and persistence of the diatomaceous earth in small scale farm storage situations. Thus further studies of the long term persistence of diatomaceous earth applied to host commodity are needed. Stathers et al (2002a) also noted that storage method used (bulk or bag) storage conditions and quantity of commodity, are likely to affect the efficiency of diatomaceous earths.

## 2.7. Evaluation of efficacy of diatomaceous earth outside Zimbabwe

Inert dusts such as diatomaceous earth and silica aerogel dusts have been tested for stored grain insect control by numerous researchers (Parkin, 1944, White *et al*, 1966). The dust like any other dust damage or adsorb the oily or waxy layer of the cuticle of insects inducing death (Ebeling, 1971).

In USA diatomaceous earth dusts have been evaluated for suppressing insects in stored shelled maize (Redlinger and Womack, 1960). The formulations tested, Perma-Guard and Kenite revealed variable effectiveness on various insect species and high rates of 2-4g of dust / kg of grain (0.2 -0.4% w/w) were necessary to obtain more than 95% mortality.

Further notable research  on efficacy of diatomaceous earths was done by Subramanyam *et al* (1994) by testing the effectiveness of Insecto (commercial diatomaceous earth registered in 1984 in USA) to control six economically important stored grain insect species during an 8.2 month test period. The test was carried out using an application rate of 0.05% w/w (0.5g of dust / kg of grain) in 12 metal barrels using 109kg of wheat grain against *Rhyzopertha dominica*, *S.oryzae*, *Cryptolestes ferrugineus*, *Cryptolestes pusillu*s and *T. castaneum*. The results revealed Insecto to be equally effective to synthetic insecticides in suppressing all the insect pests although the suppression levels varied with insect species. Susceptibility to inert dusts is dependent upon body surface area and thickness of the cuticle which vary with insect species (Quarles, 1992). Therefore different application rates are required for different insect species.

The physical characteristics (particle size distribution and surface area) of Insecto and older formulations, Perma-Guard and Kenite (White *et al*, 1975) and the environmental conditions of both trials were essentially similar. This, as a result fails to explain the greater effectiveness of Insecto at low application rates 0.05% compared to higher application rates of 0.2%-0.4% for older formulations. However, Le Patourel (1986) explained that under tropical conditions higher application rates of at least 0.10% are considered effective to compensate for loss in insecticidal activity due to adsorption of atmospheric moisture.

The field test results of Insecto is of little value to field studies on efficacy of diatomaceous earth in Zimbabwe since the artificial introduction of known insect species is not reflective and comparable to natural infestation as in our on farm storage season in Zimbabwe. Furthermore, the storage conditions (relative humidity and ambient temperature) in metal barrels differ with those in on farm granaries.

## 2.8. Field trials in Zimbabwe

Confirmation of laboratory trials that diatomaceous earth, Protect- It and Dryacide successfully reduced populations of some major Zimbabwe storage insect pests under constant conditions (Giga and Chinwada, 1994, Stathers *et al*, 2002a) formed the basis for on farm field trials of the two diatomaceous earth in Zimbabwe. Stathers *et al* (2002b) conducted on farm field trials for two consecutive seasons using host commodities maize (Z*ea mais*) and sorghum (S*orghum bicolar*) against storage insect pests for up to 8 months storage in three agro-ecological zones (AEZ) in Zimbabwe. The two formulations were successful in reducing the populations of the insect pests in all the three AEZ at application rates of 0.1% for Dryacide and 0.2% Protect-It in both seasons. The trial period of two storage seasons make the results more reliable since it factors in variability between seasons i.e. differences in climatic conditions and grain moisture content

Recent work by Janga (2004) revealed that the local diatomaceous earth provided significant protection against maize storage insects pests. The trial was conducted over 8 month storage period using the local diatomaceous earth Chemutsi at three levels 0.1%, 0.2% and 0.25%, commercial diatomaceous earth Protect-It at 0.1% and a synthetic insecticide ASD. The trial results revealed that ASD was the most effective grain protectant in terms of low damage levels of maize grain compared to all the diatomaceous earth though it was not statistically significant. This indicate that systematic insecticides have a slight edge over desiccant dusts. Protect-It at 0.1% had significantly lower damage compared to the local diatomaceous earth at all three levels. However, Chemutsi at 0.20% and Chemutsi at 0.25% provided significant protection against insect damage although excessive damage was noticed in Chemutsi at 0.1%. Thus the local diatomaceous earth was not as effective at lower application rates as was in commercial diatomaceous earth Protect-It but rather at higher application rates. This can be attributed to differences in geological origin of the diatoms, sorptive capacity and particle size of the diatomaceous earth as reported by Alexander *et al* (1944).

The storage period used in the trial was short (October to May) and does not reflect the period in which the formulations are effective, thus a longer storage period (July to May) would aid in reflecting the persistence of the diatomaceous earth. Moreover, a trial over one storage season may not be enough to assert the consistency of the diatomaceous earths' performance over time.

# 3.0. MATERIALS AND METHODS

## 3.1. Trial Site

On station trials will be conducted at the Institute of Agricultural Engineering (IAE) in Hatcliffe which is natural region 2a. The trial will be carried out over 40 week's storage season period starting from July 2004 and ending in April 2005 which coincides with normal storage season in Zimbabwe. Rainfall, ambient temperature and relative humidity data will be recorded at IAE during the storage period.

## 3.2. Storage Facility

Four granaries which are already constructed at the research station will be used. Each granary will be used as a separate replicate and consists of six compartments. The compartments were plastered with mud on the internal surfaces to prevent hidden infestations. A half-inch mesh wire was installed on the top of the compartments in order to prevent rodents from attacking the grain but at the same time exposing the grain to a great number of insects as possible. The base of the granaries is made up of cement slabs and raised approximately fifty centimeters on columns above the ground. Thorough cleaning and replastering of the store is vital to eliminate residual infestation and cross contamination.

## 3.3. The Grain

Pioneer white PHB 30 H83 obtained from a farmer in Domboshava will be used. Approximately four tonnes were procured with 960kg for each granary and 1060kg for each compartment. The grain will be thoroughly mixed prior to treatments to eliminate variations in grain moisture content before loading it as bulk grain in the stores.

## 3.4. Preparation of raw diatomaceous earth samples

The diatomaceous earths, mined at Chemutsi and Beitbridge in the Zambezi and Limpopo valleys respectively and collected with the assistance from Dorowa Minerals Ltd a subsidiary of Zimbabwe Phosphate Industries Ltd will be crushed and pounded in the laboratory using a pestle and mortar. The pulverized diatomaceous earth will be sieved using a 150 micrometer sieve to obtain very fine dust for increased sorptive and porous qualities.

## 3.5. Experimental Design

A randomised complete block design with four replicates will be employed for the trial. The four stores will be treated as 4 separate blocks. Allocation of treatments to compartments will be randomized within blocks to eliminate intentional or unintentional bias and ensure independence amongst observations.

## 3.6. Grain Treatments

The treatments include two locally mined diatomaceous earths, Chemutsi and Beitbridge; imported diatomaceous earth Protect-It, and an untreated control. The

treatments will be admixed at the following application rates calculated on mass basis, i.e., mass of insecticide/ mass of grain:
A – Untreated (negative control)
B - Protect-It 0.1% w/w (positive control)
C – Chemutsi 0.15% w/w
D – Chemutsi 0.20% w/w
E – Chemutsi 0.25% w/w
F – Beitbridge 0.20% w/w
Thorough admixing of the maize grain with the respective grain protectants will be done on a clean floor surface using clean shovels.

## 3.7. Grain Sampling

Initial samples for laboratory analysis, approximately 1.5kg will be collected using a scoop. Subsequent samples of approximately 1.5kg will be collected every eight weeks for forty weeks storage period using multi-compartmented sampling spears (one for each treatment so as to avoid mix up of the treatments) and measured using a field scale. The sampled grain will then be placed in the polythene bags that will be made intact using flexible rubber bands to reduce moisture absorption which would affect moisture measurements. Samples will be analyzed in the Soil Science and Agricultural Engineering (SSAE) laboratory.

## 3.8.0. Sample analyses

### 3.8.1. Moisture content determination

The samples in each sample will be thoroughly mixed by shaking the bags to reduce variability and approximately 200 grams will be removed using a scoop from each of the samples for moisture content analysis. The moisture content will be determined using the standard oven method at 113 $^{0}$C for 4 hours using 3 sub-samples so as to obtain accurate results. Moisture content (mc) will be calculated on wet basis using the following formula:

$$\% \text{ mc} = (W_{wet} – W_{dry})*100/W_{wet}$$

Where $W_{wet}$= weight of wet sub-sample, and $W_{dry}$= weight of dry sub-sample

### 3.8.2. Sieving

The remaining sample will be sieved using the appropriate sieves to obtain trash. Mechanically damaged grain, unhealthy grain and any foreign materials will be removed. Insects in the trash, dead and live will be identified and counted per species within seven days from the sampling date when the conditions will still be similar to the store conditions.

### 3.8.4. Damage assessment

The clean healthy grain will then be reduced to more workable equal portions of grain which are representative sub-samples using the riffle divider and any three working samples of approximately 200-300g used for damage assessment.
Weight loss will be related to damage. The holes on grains (damaged grain) will be identified and the damaged grain and undamaged grain counted and weighed. The converted percentage mass method will be used to assess weight loss.

### 3.8.5. Data Entry and Analysis

Microsoft excel software will be used for all data processing and the statistical analysis will done using Minitab. Analysis of variance (ANOVA) will be used to compare the two controls with the diatomaceous treatments as well as their application rates.

# TIME PLAN

**MONTH (2004-2005)**

| **Activity** | **Resources** | JUL | AUG | SEP | OCT | NOV | DEC | JAN | FEB | MAR | APR | MAY |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Trial set-up** | 4granaries 4 t maize, respirator masks, shovels, de formulations | ▬ | | | | | | | | | | |
| **Sampling** | 180 polythene bags, scoop, markers and labels field scale, sampling spears | ▬ | | ▬ | | ▬ | | ▬ | ▬ | | ▬ | |
| **Sample analysis** | Sieves, trays, tares, oven, riffle divider, 180 poly-bags,180 data sheets, forceps | ▬ | | ▬ | | ▬ | | ▬ | ▬ | | ▬ | ▬ |
| **Data processing and analysis** | Computer, Diskettes, Bond paper | ▬ | | ▬ | | ▬ | | ▬ | ▬ | | ▬ | ▬ |
| **Write-up** | Bond paper | | | | | | | | | | | |
| **Dissertation binding and submission** | Bond paper | | | | | | | | | | | ▬ |

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