

# Disinfestation of Wheat Using Liquid Nitrogen Aeration

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**Abstract**—A study was undertaken to investigate the effect of liquid nitrogen aeration on mortalities of adult *Cryptolestes furrugineus*, rusty grain beetles, in a prototype cardboard grain bin equipped with an aeration system. The grain bin was filled with Hard Red Spring wheat and liquid nitrogen was introduced from the bottom of the bin. The survival of both cold acclimated and unacclimated *C. furrugineus* was tested. The study reveals that cold acclimated insects had higher survival than unacclimated insects under similar cooling conditions. In most cases, mortalities of as high as 100% were achieved at the bottom 100 cm of the grain bin for unacclimated insects for most of the trials. Insect survival increased as the distance from the bottom of the grain bin increased. There was no adverse effect of liquid nitrogen aeration on wheat germination.

**Keywords**—Cold acclimated, liquid nitrogen aeration, mortalities, rusty grain beetles.

## I. INTRODUCTION

CANADA is the second largest exporter of grain in the world with an annual production of 57.8 Mt (million tonnes) [1]. More than 50% of the grains produced are exported every year [4]. Grains are usually stored on farm for a considerable time and then moved to the terminal elevators for shipment to domestic or export markets. However, consumption of cereal grains by insects, mites, rodents and microbial spoilage or contamination during storage causes losses of up to 10% in terms of both quality and quantity. Zero tolerance for live insect pests in grain bulks is required by the Canada Grain Act and has earned Canada a worldwide reputation in providing good consistent end-use quality grains and oilseeds each year. Therefore, maintaining good grain quality and preserving grain from infestation is a prime task for the Canadian grain handling and export industry.

Chemical insecticides such as phosphine, malathion, and methyl bromide have widely been used for controlling insect pests in grain because these chemicals are inexpensive and fast-acting. On the downside, chemical insecticides not only pose health risks to people who apply them, but can also pose risk to humans and animals who consume the chemically

treated grain [4]. In addition, the frequent use of chemical insecticides and fumigants helps insects develop resistance, making higher doses of these chemicals necessary. With consumers beginning to become more concerned about insecticide residues in foods, many markets no longer accept grain that has been treated with chemical insecticides. As a result, alternate control methods need to be developed and applied.

The current focus of research in this area has been on developing physical methods of insect control. Physical control methods are based on the ecology of stored-product insects, and the realization that biological limitations exist for all insect species. These methods that involve manipulation of the physical environment, like temperature, composition of atmospheric gases and moisture content; mechanical impact, inert dusts and ionizing radiation, are used to modify the environment to such a degree that it becomes lethal to the stored-products pests.

It has been established that most insect species cannot survive at temperatures lower than  $-15^{\circ}\text{C}$  for more than a few hours [3]. Low temperature control methods have several advantages over chemical insecticides as they are effective against pesticide resistant populations, pose no hazards to applicators and leave no harmful residues [2]. For creating extremely low temperature conditions liquid nitrogen is widely used in the medical and seed storage. However, little work has been done on the use of liquid nitrogen as a low temperature agent to disinfest stored-products. It is, therefore, of interest to explore how effective liquid nitrogen is as a disinfestation agent. The objectives of this study were to: 1. determine the effect of a liquid nitrogen induced cooling front on grain bulk temperature; 2. study the effect of liquid nitrogen aeration on mortalities of rusty grain beetle adults in a grain bulk; 3. study the effect of liquid nitrogen aeration on grain germination.

## II. MATERIALS AND METHODS

A prototype grain bin 2 m in height and 0.61 m in diameter was built with a hollow cardboard tube and filled with red hard spring wheat (cultivar AC Barrie) grown in Manitoba in 2006. The bin space was divided into four vertical zones by five levels of thermocouples. Each level was equally spaced 0.50 m apart from its adjacent level(s) along the vertical direction. Four sampling holes, through which grain samplers containing caged insects (Fig. 1) were inserted into the grain bin, were drilled at heights of 0 m, 0.5 m, 1.0 m, and 1.5 m separately from the bottom of the bin. Because it was convenient to bury the grain sampler in grain sample at the top layer, i.e., the level at the height of 2.0 m, no sampling hole was drilled. Plastic tubes with

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a diameter of 38 mm and rubber stoppers were installed on the entrance of sampling holes.

An aeration fan was used to blow liquid nitrogen into the grain bin more rapidly and uniformly during the experiments and to achieve uniform grain temperature during experiment. The liquid nitrogen tank was placed on an electronic weighting scale and this scale was used to measure the weight loss rate of liquid nitrogen. The foundation of the bin was made of wood 0.61 m tall and had a square cross-section (0.76 m × 0.76 m). The foundation was covered by plastic foam for heat insulation and a metal perforated floor with 1mm holes was inserted between the bin body and its foundation to form the entire grain bin. The perforations allowed air to pass through freely but prevented the wheat kernels from falling through. The nitrogen container was connected with the grain bin through an insulated inlet at the bottom and liquid nitrogen was introduced into the grain bin by the aeration fan. Liquid nitrogen was introduced inside the bin through a set of four nozzles that spread it uniformly into the bin plenum in four different directions perpendicular to one another.

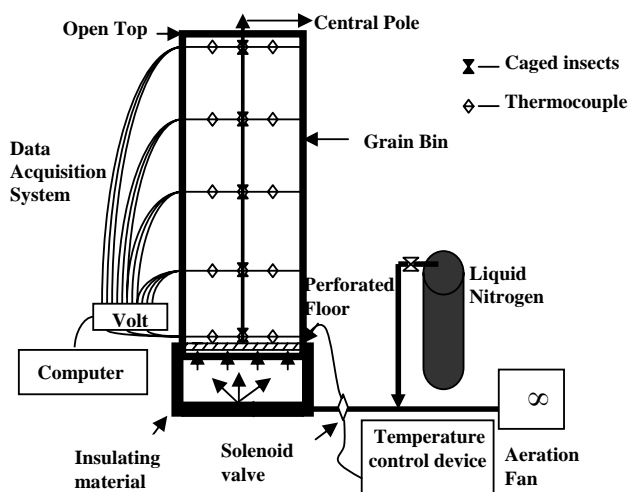


Fig. 1 Schematic of liquid aeration system

The thermocouple on the perforated floor was connected with a temperature control device connected with the data acquisition system. The temperature control device was connected to the solenoid valve on the inlet. This valve controlled the on/off state of the solenoid valve and therefore controlled the liquid nitrogen intake from the container. The threshold temperature that controlled the solenoid valve was set at  $-15^{\circ}\text{C}$ .

In this study, rusty grain beetles (*Cryptolestes ferrugineus*), the most common and cold-hardy insects of stored grain in Canada were used to evaluate the potential of liquid nitrogen as a disinfestation method. The unacclimated adults of *C. ferrugineus* used in the experiments were obtained by sieving daily the jar content with a sieve of 20.0 mm. Cold acclimated adults of *C. ferrugineus* were obtained by sieving daily the jar content with a sieve of 20.0 mm. The insects were held at  $15^{\circ}\text{C}$  for one week then transferred to  $10^{\circ}\text{C}$  for one week before the

experiment. Fifty adult rusty grain beetles were put in  $80\text{ mm} \times 20\text{ mm}$  screen bags and placed at the five different levels in the bin. In order to get insects in and out of the bin easily, standard grain probe traps (samplers) were used and these screen bags containing insects were put in the traps. A total of 200 insects (100 acclimated and 100 unacclimated) were placed in each sampler. Rubber stoppers were used to seal the sampling ports once the traps consisting of the insects were inserted in the bin. The number of live insects was counted before and after each trial to calculate the mortality. A control group of 100 insects (50 non-acclimated and 50 acclimated insects) at each level was placed in the grain bin before running the experiment trials described above. No liquid nitrogen was introduced into the grain bin for the two consecutive days when the control group was tested in the grain bin for survival rates.

Germination of the wheat seeds subjected to liquid nitrogen treatment was assessed by incubating 25 seeds on no. 3 filter paper in a 90 mm diameter Petri dish saturated with 5.5 mL of distilled water. The dishes were placed in a plastic bag to prevent desiccation of the filter paper and kept at  $25 \pm 1^{\circ}\text{C}$  with a humidity of  $70 \pm 5\%$  for 7 d. On the seventh day the germinated seeds were counted and the germination percentage was calculated.

### III. RESULTS AND DISCUSSION

#### A. Cooling Patterns

For all the trials, cooling patterns at the three measurement locations on the same level were similar while cooling patterns at different levels were quite different. For example, take the temperature change recorded for experiment trial 1. The temperatures at all three measurement locations on the bottom level reach their lowest readings in 11 to 17 minutes after the injection of liquid nitrogen started (Fig. 2). After 37 minutes from the start of the experiment, temperature rose to a value where it gradually stabilized. After 8.3 h the second liquid nitrogen tank was connected. This brought drastic temperature changes, but the temperature stabilized again within 40 min. The temperature stabilized at  $-22^{\circ}\text{C}$  to  $-28^{\circ}\text{C}$ . When liquid nitrogen was stopped at 16 h, the temperature began to rise at a rate of  $0.5^{\circ}\text{C}/\text{h}$ .

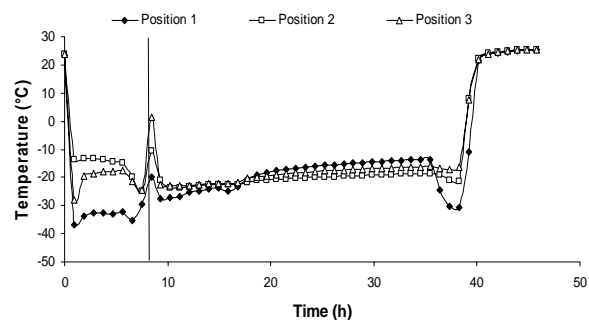


Fig. 2 Temperature change at level 1 inside the grain bin during trial 1

On levels 3, 4 and 5, temperature changes were much more gradual than levels 1 and 2. Interestingly, temperature drop was observed on level 5 even after the liquid nitrogen supply was

disconnected from the foundation of the grain bin while the aeration fan was kept running. This was because the cooling front was still moving upward through the bin even though the liquid nitrogen supply had been cut off. When both the liquid nitrogen supply and aeration fan were stopped simultaneously, temperature at all levels seemed to maintain well.

#### B. Insect Mortality

Test results from the control trial indicated that the survival rates for both unacclimated and acclimated insects were 99.6% when no nitrogen was blown through the bin. Samplers were removed from grain bins and insects were manually counted to calculate mortality at least 8 h after liquid nitrogen injection stopped. On level 1, mortalities of 100% were achieved in all experimental trials. On level 2, the combination of low temperature and sufficiently long duration of cooling period resulted in mortalities of 100% for unacclimated insects and over 90% for acclimated insects. On level 3, mortalities of over 98% were achieved in all experimental trials for unacclimated insects. For acclimated insects, temperature conditions achieved on level 3 were less effective as compared to the insect mortalities for layers 2 and 3. On levels 4 and 5, the mortalities in all five experimental trials decreased greatly as the average temperatures hovered around  $-15^{\circ}\text{C}$  or warmer and that too for shorter periods of time. The mortalities for acclimated and unacclimated insects were 6% and 20%. Therefore, this temperature condition was not effective against either acclimated or unacclimated insects.

#### C. Germination

Germination levels of wheat kernels before and after each cooling process ranged between 97.3% and 100%. Statistical analysis indicates that the difference in the mean values of the two groups in each trial was not large enough to accept that the effect of cooling process was significant. Therefore, it can be concluded that aeration with liquid nitrogen does not affect the germination level adversely.

### IV. CONCLUSION

The results establish that liquid nitrogen can effectively be used for cooling the grain to temperatures that are unsuitable for insect survival. The lower levels of grain reached temperatures much lower than the upper layers when liquid nitrogen was introduced from the bottom of the bin. Insect mortalities for cold acclimated and unacclimated insects were different under the same cooling conditions. Cold acclimated insects survived better than unacclimated insects. In general, long exposure time ( $>2$  h) is required to achieve effective killing of insects. Cooling had no effect on the germination levels of the grain.

#### ACKNOWLEDGMENT

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