EFFECTS OF FLAMELESS CATALYTIC INFRARED RADIATION ON STORED-WHEAT INSECTS AND WHEAT QUALITY

by

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Abstract

Stored-grain insects were managed traditionally with grain protectants and the fumigant phosphine. Protectant use leads to undesirable pesticide residues on grain. Many stored-grain insects are resistant to grain protectants and phosphine. Therefore, novel technologies are needed in the future to replace traditionally used methods. Preliminary laboratory and pilot scale field trials have shown catalytic infrared radiation of 2.8 to 7 µm wavelength to be effective in killing insects associated with stored wheat. The effectiveness of catalytic infrared radiation in killing life stages of three economically-important stored-grain insects in hard red winter wheat were evaluated. Wheat (113.5 or 227.0 g) infested with eggs, various ages of larvae, pupae, and adults were exposed for 45 or 60 sec at a distance of 8.0 or 12.7 cm from the catalytic infrared emitter. Infested wheat samples unexposed to infrared radiation served as the control treatment. Temperatures attained by the wheat samples during infrared exposure were monitored continuously using a non-contact infrared thermometer. The three insect species tested were the lesser grain borer, Rhyzopertha dominica (F.); rice weevil, Sitophilus oryzae L.; and red flour beetle, Tribolium castaneum (Herbst). The life stages of R. dominica and S. oryzae developing within wheat kernels were age-graded using radiographic techniques. The mean temperatures attained by wheat at the various treatment combinations ranged from 80° to 114°C. Both the time-dependent temperature profiles and mean wheat temperatures were greater in 113.5 g of wheat, exposed at a distance of 8.0 cm from the infrared emitter for 60 sec. The most heat tolerant stage in R. dominica was the older larvae, whereas in S. oryzae it was the egg, and in T. castaneum it was the pupa. In general, older larvae of all three species were less susceptible to infrared radiation than young larvae. The differences in susceptibility among life stages of all species to infrared radiation decreased with an increase in the mean temperature attained, and temperatures ≥105°C were needed to obtain 98 to 100% mortality of all life stages. Exposure to catalytic infrared radiation at the temperatures used to disinfest wheat did not adversely affect wheat, flour, and baking quality.

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Dedication

I dedicate this thesis with love to my mother, Pilistina Jokudu Mada, and my father, Eliaba Mada. They have been a guiding light and inspiration for me throughout my life.

CHAPTER 1 - Rationale for evaluating flameless catalytic infrared radiation for management of insects in stored wheat

Wheat production in the United States and Kansas

The United States ranks third in the world in wheat production and produces about 64 million metric tons annually. About 31 million metric tons are exported to 100 countries while about 33 million metric tons are consumed domestically (www.uswheat.org). A large volume of wheat is used to make breakfast cereals, breads of different types, pasta, and noodles. Wheat is also used as a raw material for the manufacture of starch. Kansas is a leading producer of wheat in the United States. In 2006, Kansas harvested 9.5 million metric tons of wheat from 9.5 million acres with an estimated value of about US \$ 1.2 billion. Nearly 10% of the wheat is stored on the farm and 70% is stored off the farm. Wheat in storage is susceptible to spoilage by insects, molds, and bacteria. Depending on the length of storage, grain moisture, and level of pest management, stored wheat is attacked by a variety of insect species (Storey et al. 1984, Reed and Pedersen 1987, Kenkel et al. 1992, and Martin et al. 1997). The most common and damaging insect species associated with wheat stored on farms and at elevators in Kansas and neighboring states is the lesser grain borer, Rhyzopertha dominica (F.) (Reed et al. 1991, Dowdy and McGaughey 1994, Vela-Coiffier et al. 1997, Reed et al. 2003). Rhyzopertha dominica is also an important species of stored wheat in the United States and the world (Sinha and Watters 1985). The rice weevil, Sitophilus oryzae (L.), and red flour beetle, Tribolium castaneum (Herbst), are found to a lesser extent in Kansas stored wheat (Reed et al. 1991, Reed et al. 2003). Activity of all three species of insects has also been observed outside farm bins (Dowdy and McGaughey 1994) and elevator silos (Dowdy and McGaughey 1997). Rhyzopertha dominica is typically found in wheat received at milling facilities (Good 1937, Perez-Mendoza et al. 2004). Both R. dominica and S. oryzae immatures complete development inside kernels of wheat (Sinha and Watters 1985), and the immature stages contribute to insect fragments when the wheat is milled. Although T. castaneum is found in wheat, it prefers dockage (McGregor 1964), and is an important pest associated with flour mills (Good 1937).

Management of stored-wheat insects

Insects in stored wheat are managed primarily by chemical means (Kenkel et al. 1992, Martin et al. 1997). About 12% of farmers (n = 446) and 39% of elevator managers (n = 293) surveyed fumigated their stored wheat with phosphine (Martin et al. 1997). About 50% of the same farmers and elevator managers applied the organophosphate malathion or chlorpyrifosmethyl to their stored wheat (Martin et al. 1997). A 1999 Kansas survey (Kansas Agricultural Statistics Service [KASS] 2000) indicated that 71% of farm-stored wheat was treated with insecticides (7,273 kg). The National Agricultural Statistics Service (NASS) survey of elevator managers indicated that 11.6% of wheat stored at the elevators was furnigated with phosphine, 1.4% was treated with chlorpyrifos-methyl, and 1.5% with malathion (NASS 1999). The use of grain protectants such as malathion and chlorpyrifos-methyl on wheat results in the presence of residues, which may be unacceptable to some domestic or foreign buyers. The USDA's Pesticide Data Program survey (USDA 1998a,b) found detectable residues (<0.3 ppm) of malathion and chlorpyrifos-methyl in a majority of wheat samples (56 to 73%) received at elevators, and 40-48% of the samples had both malathion and chlorpyrifos-methyl residues, which suggests some level of blending at receiving elevators. In addition, the 1997 USDA's Pesticide Data Program data for wheat indicated that 23 out of the 623 samples analyzed had illegal residues of pirimiphos-methyl, a product not registered for use on wheat.

There is documented resistance to the traditionally used organophosphate grain protectants and the fumigant phosphine in *R. dominica*, *T. castaneum*, and *S. oryzae* (Subramanyam and Hagstrum 1996), although a direct link between application rates and control failures has not been established.

In the United States sale and distribution of chlorpyrifos-methyl (Reldan® at 6 mg (AI)/kg) ceased as of December 31, 2004, under the 1996 Food Quality Protection Act, which set tougher standards for reviewing the safety of registered pesticides, especially organophosphates. However, chloropyrifos-methyl (3 mg (AI)/kg) is currently available in Storcide II® formulation in combination with deltamethrin (0.5 mg (AI)/kg). This product was registered in 2004.

Methoprene, first registered in 1992, is currently available as Diacon II® formulation for grain treatment. This insect growth regulator does not kill the adults, but affects growth and development of immatures. The presence of two or more live adults can result in wheat being classified as infested according to the Federal Grain Inspection Standards (GIPSA 1997). Several formulations of diatomaceous earth dusts can be applied to grain, but these dusts adversely affect grain physical properties and hence are not widely used (Subramanyam and Roesli 2000). In January 2005, spinosad received United States Environmental Protection Agency's approval as a grain protectant at 1 mg (AI)/kg on barley, millets (foxtail, proso, and pearl), oats, rice, sorghum (milo), triticale, wheat, and birdseed (Federal Register 2005, Vol. 70: 1349-1357). The maximum residue limits for spinosad on grain were approved by The CODEX Committee on Pesticide Residues in 2006. The tolerance for spinosad in the United States is 1.5 mg(AI)/kg while the CODEX tolerance is 1 mg(AI)/kg. The registrant of spinosad, Dow AgroSciences (Indianapolis, IN), has been working with the grain industry and various countries for approval of spinosad tolerances on grain. Launch of commercial products will be delayed until international tolerances are in place. None of these grain protectants, except for diatomaceous earth dusts, can be used on organic wheat.

New and innovative technologies should be explored for effective disinfestation of wheat, because of problems associated with pesticide residues on grains, resistance development in insects, and to meet quality demands (e.g., wheat free of pesticide residues) of domestic and foreign buyers. Furthermore, limited pest management options are available for managing insect pests in stored organic wheat. My thesis research was therefore designed to explore and evaluate flameless catalytic infrared radiation as an effective alternative to currently used grain protectants and the fumigant phosphine for controlling insects, especially *R. dominica*, *S. oryzae*, and *T. castaneum*, in stored hard red winter wheat.

Infrared radiation for disinfesting grains

Infrared radiation has wavelengths (0.075 to 1000 µm) longer than visible light (380 -750 nm) but shorter than microwaves (0.1 to 100 cm) (Penner 1998) (Figure 1:1). In this range of 3 to 7 µm, bonds between oxygen and hydrogen atoms in the water molecule are stretched asymmetrically and symmetrically (Wehling 1998). This characteristic of water to absorb infrared radiation (Figure 1:2) has been used for rapid drying of cereal commodities, especially wheat (Bradbury et al. 1960) and rice (Schroeder 1960, Schroeder and Rosberg 1960, Faulkner et al. 1969). Since insects have a higher moisture content (>60%) than stored grain (<15%), it is probable that insects will absorb higher infrared radiation dose than the grain and therefore heat up more rapidly. This differential heating makes the use of infrared radiation an attractive technology for stored-grain insect pest management. Frost et al. (1944) measured internal temperatures of stored-grain insects exposed to various wavelengths, intensities, and exposure time, and he attributed insects' mortality to increased body temperature. Infrared radiation used in killing different stages of stored product insects developing within kernels was evaluated about four decades ago (Tilton and Schroeder 1963, Cogburn 1967, Cogburn et al. 1971, Kirkpatrick 1973, Kirkpatrick and Tilton 1972, Kirkpatrick et al. 1972, Tilton et al. 1983). In these earlier tests, infrared radiation was generated by combusting natural gas or propane over ceramic tiles in the presence of oxygen. Kirkpatrick and Tilton (1972) exposed 100 adults of 12 stored-product insect species in 150 g of soft red winter wheat (13.5% moisture) placed below the gas-fired heater in a single kernel thickness layer, 65 cm from the radiation source. The grain attained a final temperature of 49°C in 20 sec, 57°C in 32 sec, and 65.5°C in 40 sec. In these tests, temperatures were not measured during exposure, but immediately after exposure with a banjo thermometer (Yellow Springs Instrument Model 435F, Yellow Springs, OH). A 40 sec exposure time resulted in 99.6% mortality of adults of the 12 insect species. Immature stages of species developing within kernels needed higher temperatures than those required for killing exposed adults. Schroeder and Tilton (1961) reported total control of internal insects namely S. oryzae and R. dominica life stages in rough rice at final grain temperatures of 56° and 68°C,

respectively. The final rough rice temperatures (41° to 63°C) were inversely related to distance from the infrared emitter (50.8 to 15.2 cm), and temperatures increased with an increase in exposure time at any given distance from the emitter (Tilton and Schroeder 1963). For all species (*R. dominica*, *S. oryzae*, and the Angoumois grain moth, *Sitotroga cerealella* (Olivier)), mortality estimates were based on number of adults that emerged from infested samples exposed to infrared relative to infested untreated samples. Mortality varied among the species, and some of the treatments did not completely prevent the emergence of adults. Species susceptibility (from high to low) was *S. oryzae>S. cerealella>R. dominica*. The authors further extrapolated the adult emergence curves plotted against the final temperatures attained and recommended a final grain temperature of 65° to 70°C for 100% mortality of immature stages developing within kernels. Kirkpatrick et al. (1972) compared infrared to microwave radiation in disinfesting soft red winter wheat infested with *S. oryzae*. They concluded that infrared radiation was superior to microwaves in killing both immatures and adults of *S. oryzae*.

In all of the past research, gas-fired heaters of high intensity were used. Such high temperatures and an open flame cannot be used in any grain handling facility because of explosion hazards. Catalytic Drying Technologies LLC, a company in Independence, KS, has developed a proprietary flameless infrared technology for diverse drying applications (http://www.catalyticdrying.com). This company received an award from the United States Environmental Protection Agency's Pollution Prevention Program for developing the flameless catalytic infrared radiation source, under the "Environmentally Preferable Products" category. Unlike flame infrared emitters, in flameless infrared emitters, propane or natural gas chemically react at the surface of a platinum catalyst below gas ignition temperatures, delivering peak radiant energy in the 2.8 to 7 µm range, and the resulting temperatures are below 500°C.

Bench top infrared heater

The Catalytic Drying Technologies company donated a bench-top flameless catalytic infrared emitter. The heating elements are housed in a circular stainless steel casing (Figure 1:3).

The diameter of the emitter surface is 27.94 cm (surface area, 613.36 cm²). To initiate the reaction, heated coil is heated with a 110 volts electricity supply for 15 min. Propane gas was supplied by a hose to the heating elements, surrounding the platinum catalyst, from a 473 ml cylinder (Ozark Trail Propane Fuel, Bentonville, AR). The gas pressure was kept constant at 27.9 cm of water column (0.4 psi). The total heat energy output of the bench-top model is 1.46 kW/h (5,000 BTU/h). The reaction producing infrared energy is shown below:

$$C_3H_8 + 5O_2$$
 \longrightarrow $3CO_2 + 4H_2O + infrared energy$

Advantages and limitations of infrared radiation for grain disinfestation

The catalytic infrared emitters are environmentally friendly, and since they do not to use any flame these emitters do not produce any NOx or CO. The co-products of catalytic oxidation-reduction reaction are infrared radiation, carbon dioxide, and water vapor. It is a viable pesticide alternative for those storing organic or non-organic wheat. Rice and wheat can resist infrared temperatures up to 80°C without adverse effects on baking or milling qualities (Kirkpatrick 1974). However, we were unable to find quantitative data to support these statements. The limitation of using infrared radiation is that it is a responsive tactic, and therefore, there is no residual effectiveness. Disinfested grain may get reinfested after exposure to infrared radiation. However, this is not a limitation if wheat, soon after disinfestations, is subjected to milling. A drop in moisture content (0.5 to 0.6%) can be expected (Kirkpatrick 1974), with some loss in germination. A slight drop in moisture may be beneficial because it may discourage mold germination. However, there is very limited work on the adverse effects of infrared radiation on wheat quality or quality of end-use products made from exposed wheat, and these adverse effects may be negligible with the low intensity flameless catalytic infrared emitters.

Research objectives

The research reported in this thesis focuses on exploring infrared radiation as a tool for managing insects in stored wheat as a replacement to grain protectants and phosphine which are

no longer available for use (e.g., chlorpyrifos-methyl), or which are partially effective due to the development of insect resistance (e.g., malathion, chlorpyrifos-methyl, and phosphine). Furthermore, this work aims to develop new and innovative integrated pest management (IPM) strategies and tactics that can be implemented by end-users to reduce pesticide inputs while maintaining stored wheat quality standards. The proposed work will be conducted using hard red winter wheat produced in Kansas. Specific objectives of the work are:

- (1) To determine effectiveness of infrared radiation against eggs, larvae, pupae, and adults of *R. dominica*, *S. oryzae*, and *T. castaneum* in wheat using a laboratory bench top infrared emitter.
- (2) To evaluate changes in physical, chemical, rheological, and end-use properties of wheat exposed to infrared radiation.

The thesis has four chapters excluding this chapter. Chapters two through four deal with the effects of infrared radiation on various life stages of *R. dominica*, *S. oryzae*, and *T. castaneum*, respectively. The fifth chapter deals with effects of infrared radiation on wheat quality. Radiographic equipment at the USDA's Grain Marketing and Production Research Center in Manhattan, KS, milling equipment and the Wheat Quality Laboratory facilities in the Department of Grain Science and Industry, as well as the services of Nanoscale Corporation, Manhattan, KS, were instrumental in accomplishing these two project objectives.

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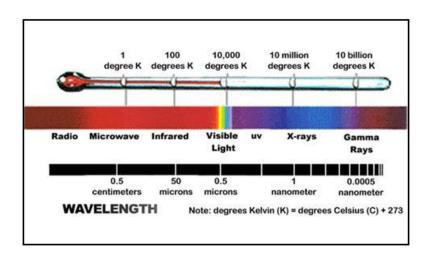


Figure 1:1. Electromagnetic spectrum showing the relative position of infrared radiation.

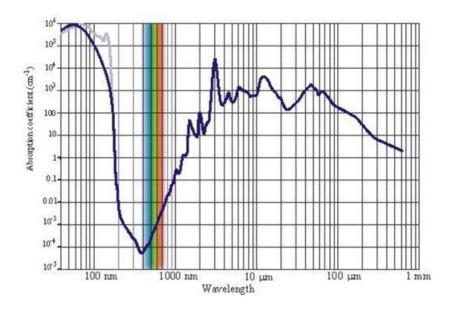


Figure 1:2. Absorption spectra for water showing peak radiation absorption bands at 3, 4.5, and 6 µm (www.catalyticdrying.com)





Figure 1:3. Bench top model of infrared emitter (left) showing the propane bottle and pan with hard red winter wheat. The non-contact thermometer (Raytek®, Model MX4) is used for continuous measurement of temperatures attained by wheat (right) during exposure to infrared via a RS-232 cable connected to a laptop computer. Photos by Bh. Subramanyam.

CHAPTER 2 - Susceptibility of *Rhyzopertha*dominica (F.) (Coleoptera: Bostrichidae) life stages
exposed to a flameless catalytic infrared emitter

Abstract

In laboratory experiments, a flameless catalytic infrared emitter was used to kill different life stages of the lesser grain borer, *Rhyzopertha dominica* (F.), an economically important insect species associated with stored wheat in Kansas. The heater emits infrared in the 3 to 7 μm range. A non-contact infrared thermometer obtained real-time grain temperatures during exposures of uninfested and infested wheat containing various life stages of *R. dominica*. The grain temperatures attained were influenced by wheat quantity, distance from the emitter, and exposure time, which in turn influenced effectiveness against various life stages of *R. dominica*. In general, higher grain temperatures were attained in 113.5 g of wheat as opposed to 227.0 g, at 8.0 cm from the emitter surface rather than at 12.7 cm, and during a 60-sec exposure compared to a 45-sec exposure. Logistic regression indicated that the probability of death of various life stages of *R. dominica* was temperature-dependent. The log odds ratios showed older larvae were less susceptible to infrared than younger larvae. About 99 to 100% mortality of *R. dominica* life stages occurred when using 113.5 g of wheat, exposed for 60 sec at a distance of 8.0 cm from the emitter resulting in mean wheat temperatures of 108 to 114°C. These promising results show flameless catalytic infrared technology to be a viable option for disinfestation of stored wheat.

Introduction

Stored-grain insects have been primarily managed by chemical methods. However, many consumers are increasingly demanding grain and grain products free of any pesticide residues. The prolonged use of pesticides has resulted in stored-grain insects developing resistance to traditionally used pesticides (Sinha and Watters 1985, Subramanyam and Hagstrum 1996). Furthermore, current government laws such as the 1996 Food Quality Protection Act have resulted in revoking uses of previously registered pesticides. For example, the uses of the organophosphate chlorpyrifos-methyl as a grain protectant were cancelled as of December 31, 2004. Therefore, newer and environmentally benign technologies need to be explored as an alternative to traditionally used pesticides for managing insects associated with stored grain.

The use of infrared radiation for disinfestation of grain was explored between 60s and 80s. Previous research has shown that grains exposed for less than 60 sec to infrared radiation (3 to 7 µm range) to be effective in killing primary (internal developers) and secondary insects (external developers) in both soft wheat and paddy rice (Tilton and Schroeder 1963, Cogburn 1967, Cogburn et al. 1971, Kirkpatrick and Tilton 1972, Kirkpatrick et al. 1972, Kirkpatrick 1975, Tilton et al. 1983). The infrared radiation used in these tests was generated when propane gas was burnt over ceramic tiles producing more than 14.07 kW/h (48,000 BTU/h) of heat energy (Tilton et al. 1961, Kirkpatrick and Cagle 1978). With these gas-fired infrared emitters, temperatures as high as 930°C at the emitter surface were attained. Such high temperatures and flames are not safe in grain storage and handling facilities due to explosion hazards. In previous research, grain temperatures were not measured in "real-time", and temperatures were measured after infrared exposure possibly resulting in underestimating the temperatures attained. Furthermore, life stages of insects, especially internal developers, exposed to infrared radiation were not accurately confirmed.

Flameless catalytic infrared radiation is a new technology developed by Catalytic Drying Technologies LLC in Independence, KS (www.catalyticdrying.com). The flameless catalytic infrared radiation is produced when propane or natural gas chemically reacts in the presence of a

platinum catalyst at temperatures below ignition (Gabel et al. 2006, Pan et al. 2008). This technology may be suited for drying various commodities, and one paper examined the combined benefits of drying and disinfesting rough rice (Pan et al. 2008). Infrared can be used for enzyme inactivation, for baking, and for inactivating microorganisms (Sandu 1986, Gabel et al. 2006). At the mid-infrared range (3 to 7 μ m), water, protein, sugars and nucleic acids have maximum infrared absorption (Sandu 1986, Pan et al. 2008). The absorbed energy causes these chemicals to vibrate at a frequency of 8.8 x 10^7 to 1.7 x 10^8 MHz, resulting in an increase in temperature (Fasina et al. 1999). In infested grain, insects have greater moisture content than the grain, and it is hypothesized that the former will receive a lethal dose of infrared energy.

In the present investigation, the effectiveness of a bench top flameless catalytic infrared emitter was evaluated against all life stages of the lesser grain borer, *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae), a pest that is common and severe in stored wheat (Sinha and Watters 1985).

Materials and Methods

Insect Rearing. Cultures of *R. dominica*, obtained from the USDA-ARS, Stored-Product Insect Research Unit, Manhattan, KS, were reared in a laboratory growth chamber (Model I-36 VL; Percival Scientific, Perry, IA) in the Department of Grain Science and Industry, Kansas State University, at 28°C, 65% RH, and 14:10 (L:D) h photoperiod on organic hard red winter wheat (var. Jagger) purchased from Heartland Mills in Marienthal, KS.

Identifying Various Life Stages of *R. dominica***.** Wheat was equilibrated to 12% moisture by placing it at 28°C and 65% RH for one week in 0.45-liter glass jars with wire-mesh screened lids. Equilibrated wheat (5 g) was placed in each of five 24-ml plastic vials, and the wheat in vials was infested with 20 unsexed adults of *R. dominica*. After 72 h, all adults were removed, and the grain contents, including eggs, were reintroduced back into the vials. The 5 g samples in the five vials were subjected to radiographic analysis (X-rays), 72 h after adult

infestation (day 0), and on days 7, 14, 21, 24, and 28 following day 0. Adults that emerged daily from wheat in the vials were counted until adult emergence has completely ceased.

Kernels from each vial were spread into a single layer on a sample holder and subjected to X-rays from a Faxitron X-ray device (Model 43855A; Faxitron X-Ray Corporation, Lincolnshire, IL). The voltage setting was 28kV and the time to acquire images using a digital camera, connected to the Faxitron, took 10 sec. All images were saved in the tagged image file (TIF) format. The stage of development was characterized by measuring the length and width of tunnels inside kernels made by larvae. A total of 48 kernels on each of the observation days (0 to 28 d) were examined, and width within kernels were measured. The tunnel width for each kernel was determined by averaging width of the tunnel at the top, middle, and the bottom portions.

The captured images were coded and given to six graduate students in the Department of Grain Science and Industry with varying levels of expertise in stored-product entomology. None of the students had experience with Faxitron or in determining the ages of *R. dominica* immatures inside kernels. The purpose of this exercise was to determine if a naïve observer was able to accurately age-grade insects based on images alone. The scores of 0 (false identification) and 1 (correct identification) by students were tallied to determine accuracy in determining various ages of *R. dominica*.

Grain Infestation and Infrared Treatments. Three factors that influence infrared intensity, as indicated by the measured temperature, were evaluated. These factors include grain quantity (113.5 and 227.0 g), distance from the emitter (8.0 cm and 12.7 cm), and exposure time (45 and 60 sec). The 133.5 g and 227.0 g of wheat at 12% moisture were placed in individual 0.45-liter glass jars with wire mesh and filter paper lids. One hundred unsexed adults of *R. dominica* from cultures were added to each jar. After 3 d of infestation, the adults were removed by sifting the contents, and all grain contents including eggs of *R. dominica* that were laid within the 3 d were returned to the jars. The 3-d old jars with eggs represented age 0 of *R. dominica*, and jars that were held in the growth chamber at 28°C and 65% RH for 7, 14, and 21 and 24-d represented larvae in different developmental stages, and jars held for 28 d represented pupal and

teneral adults within kernels. For adult exposures, ~100 unsexed 2-wk-old adults from cultures were introduced into jars holding 113.5 and 227.0 g of wheat prior to infrared exposures.

A bench top model was used to expose wheat infested with various life stages of *R*. *dominica* to infrared radiation. The bench top model, donated by Catalytic Drying Technologies LLC, has a circular heating surface of 613.36 cm², and is fueled by a 473-ml container of propane (Ozark Trail Propane Fuel, Bentonville, AR) at 28.0 cm of water column pressure. The total heat energy output of this unit is 1.47 kW/h (5,000 BTU/h). Infrared is emitted when the propane reacts with oxygen in the presence of a platinum catalyst.

A steel pan of 27.94 cm diameter and 3.8 cm deep with a steel handle (43-cm long) was used below the heater to expose infested wheat in a single layer to infrared. Infested wheat was exposed for 45 and 60 sec to infrared at 8.0 and 12.7 cm from the emitter. Temperature of exposed wheat was measured continuously at the center of the pan using a non-contact infrared thermometer (Raynger MX4 Model 4TP78 Raytek®, Santa Cruz, CA). The infrared thermometer works in the 8 to 14 µm range and has a rapid response time of 250 milliseconds. The thermometer was connected via an USB port to a laptop computer using a RS-232 cable to record "real-time" grain temperatures every second (LabVIEW (National Instruments Corporation, Austin, TX).

The infrared thermometer, with emissivity set at 0.95, was calibrated against a mercury thermometer using 12% moisture organic hard red winter wheat (1400 g in glass containers) as the substrate. Calibration was done at 13 different temperatures between 25 and 126.5°C, where it took approximately 7.5 to 23 h for the wheat in glass containers to uniformly reach the set growth chamber (25 to 38°C) or electric oven temperatures (Blue M, Blue island, IL) (41.5 to 126.5°C) temperatures. The mercury thermometer was placed on the surface of the wheat. Temperature with the infrared thermometer was measured at the same location as the mercury thermometer. Calibration experiments at each of the constant temperatures were replicated twice. The mean mercury temperatures were regressed against the mean infrared thermometer

temperatures using linear regression (SAS Institute 2002), and the slope was tested for significant deviation from one using a *t*-test at $\alpha = 0.05$ (SAS Institute 2002).

Data recorded by the infrared thermometer were saved in a Microsoft Excel® file for further analysis. The thermometer was mounted on a tripod away from the infrared heater and was directed in such a way to record only the wheat sample being heated below the infrared emitter.

Each life stage, grain quantity, distance, and time combination were replicated three times and all replicates were tested on the same day. Wheat, 113.5 and 227.0 g, infested similarly, but unexposed to infrared treatment, served as the control treatments, and there were four replicates for each grain quantity.

Assessment of Insect Mortality. Wheat exposed to infrared radaiation was placed back in jars and the mortality of *R. dominica* adults was determined after infested wheat in jars was held for 24 h in a growth chamber at 28°C and 65% RH. For all immature stages, after infrared exposure, the wheat was placed back in the jars and incubated at 28°C and 65% RH until emergence of adults. Adult emergence was checked after 42-d from day 0. Untreated wheat replicates (controls) were handled similarly. As *R. dominica* larvae and pupae develop within kernels, it would be difficult to determine survival of these stages in infrared-exposed and control wheat samples (Wadley's problem) (Finney 1971). Therefore, these stages were reared to adulthood to confirm mortality, and the number of adults that emerged from eggs, larvae, and pupae in untreated wheat indicated both the validity and robustness of our experimental approach and the degree of control due to infrared exposure.

Experimental Design and Data Analysis. The experiment was run as a completely randomized design. The mean temperature was plotted as a function of time for 113.5 and 227.0 g of wheat exposed for 45 and 60 sec at 8.0 and 12.7 cm from the infrared emitter. There were eight temperature profiles for each insect age. A comparison of temperature profiles across various ages showed that for any given quantity of grain, distance from emitter, and exposure time, the profiles were essentially similar.

The temperature profile for each replicate was averaged over time. The mean wheat temperature attained for any given insect age, wheat quantity, and exposure time combination between 8.0 cm and 12.7 cm distance from the emitter was compared using two-sample *t*-tests for equal variance (SAS Institute 2002). Two-sample *t*-tests were also used to compare mean temperatures attained after a 45-sec and 60-sec exposure or in 113.5 and 227.0 g of wheat when all other factors were fixed. In order to determine if the mean wheat temperature for a given quantity of grain, distance from emitter, and exposure time combination across the various ages tested (eggs [day 0], 7, 14, 21, 24, 28, and 42 d [adult emergence]) was similar, a linear regression of temperature versus insect age was performed and the slope was tested for deviation from zero (SAS Institute 2002).

The number of adults that emerged from untreated wheat and those exposed to infrared in the various treatment combinations was recorded. The main effect of insect age, wheat quantity, distance from emitter, and exposure time and their two-way interactions on the probability of death were determined using logistic regression at $\alpha = 0.05$ (SAS Institute 2002). Odds ratios from logistic regression were used to show differences in susceptibility (odds of dying) of various life stages exposed to infrared. The odds ratio for adults (1) was used as a reference and a ratio >1 showed that a life stage was more susceptible than adults to infrared while a ratio <1 showed that a stage was less susceptible than adults. Differences in susceptibility of various life stages was also determined by plotting probability of death as a function of mean wheat temperature averaged over wheat quantity, distance from heater, and exposure time.

Results

Identifying R. *dominica* **Life Stages.** Wheat infested by adults for 3 d represented the egg stage (day 0). Larvae that hatch from eggs enter kernels to continue development (Arbogast 1991). The first sign of larvae within kernels was noticed 8-9 d after day 0 or 11-12 d after adult infestation. Larvae in various stages of development were evident at 14 and 21-d (Figure 2:1). A

majority of kernels had pupae at 24-d, and both pupae and teneral adults were observed within kernels at 28-d.

As larvae developed, the tunnel width within kernels, showed a corresponding increase. It is surprising to find the width show an increase at 28 d when compared to 24-d, because at 24-d many of the kernels had pupae. Even though 48 kernels from the same set of five vials were observed over time, the same 48 kernels were not measured on different observation days. Therefore, variations in development of larvae among kernels may have contributed to the differences observed between 24 and 28 d. All six students (100.0%) accurately scored *R. dominica* that were 21, 24, and 28 d old. However, only four out of the six students (66.7%) were able to accurately score *R. dominica* stages that were 7 and 14 d old. Adults of *R. dominica* started to emerge at 30 d, and the peak emergence occurred at 33 d and no adults emerged after 36 d (Figure 2:4).

Calibration of Infrared Thermometer. The actual wheat temperature, as measured by the mercury thermometer (y), relative to the infrared thermometer (x) was best described by the following linear regression equation: y = 0.86 (SE, 0.28) + 0.99 (0.004)x (n = 13; $R^2 = 0.999$) (Figure 2:2) The slope was not significantly different from one (t = 2.5; df = 11; P = 0.985); therefore, no corrections were necessary when using infrared thermometer for measuring wheat temperature during infrared exposures.

Temperature Profiles During Infrared Exposure. Thirteen measurements, taken at different points of the emitter surface with the infrared thermometer, showed temperature variations in range from 335 to 474°C. This suggested that the temperature output was not uniform across the emitter surface. The low temperatures recorded were at the edges as compared to the center, and therefore, during infrared exposure temperature of the wheat in the center of the pan was measured.

The temperatures attained by wheat were greater when grain was exposed for longer time periods at same grain quantity and distance from the emitter. Figure 2:3 is a representative temperature profile obtained for wheat infested with eggs exposed to infrared in 113.5 and 227.0

g of wheat at distances of 8.0 and 12.7 cm from the emitter for 45 and 60 sec. The temperature profiles for other ages for a given wheat quantity, emitter distance, and exposure time followed a similar trend. In general, temperatures attained by wheat were greater in 113.5 g of wheat, at 8.0 cm from the emitter, and after a 60 sec exposure. The time-dependent temperature profile was highest for 113.5 g of wheat exposed for 45 or 60 sec at a distance of 8.0 cm from the emitter followed by 113.5 g of wheat exposed at 12.7 cm. In 227.0 g samples of wheat exposed to infrared radiation, recorded grain temperatures were consistently lower than in 113.5 g of wheat, regardless of emitter distance or exposure time. The presence of twice as many kernels in the 227.0 g of wheat than in the 113.5 g of wheat may have resulted in some kernel surfaces not being heated uniformly.

Two sample *t*-tests for each life stage or *R. dominica* age group (0, 7, 14, 21, 24, 28, and 42 d) have shown that the mean temperature attained by 113.5 or 227.0 g of wheat during a 45-or 60-sec exposure was significantly greater at 8.0 cm from the emitter when compared with mean temperature attained by wheat at 12.7 cm from the emitter (*t*, range among ages, grain quantities, and exposure times = 7.48 to 61.12; df = 4; P < 0.002). The mean temperature attained by wheat after a 60-sec exposure was significantly and consistently higher than those attained after a 45-sec exposure at a given insect age, grain quantity, and distance from the emitter (*t*, range = -3.89 - -16.80; df = 4; $P \le 0.0176$). In general, grain quantity did not influence the mean grain temperatures attained for any given insect age, distance from emitter, and exposure time. In 21 out of the 28 comparison, the difference in mean temperature attained by 113.5 and 227.0 g of wheat was not significant (*t*, range = -2.75 - 2.51; df = 4; P > 0.5130). In the 7 other cases, mean temperatures attained by wheat were higher with 113.5 g than with 227.0 g.

The slope of the linear regression between mean temperature attained by 113.5 or 227.0 g of wheat at 8.0 or 12.7 cm from the emitter after a 45 or 60 sec exposure and the insect age was not significantly different from zero (t, range among grain quantities, distance from emitter, and exposure times = -1.62 – 0.19; n = 7; $P \ge 0.1656$).

Adults of *R. dominica* Observed in Infrared-Exposed Wheat Samples. Logistic regression analysis showed that the probability of death of *R. dominica* was influenced significantly (P < 0.0001) by insect age ($\chi^2 = 642.65$; df = 6), wheat quantity ($\chi^2 = 323.10$; df = 1), distance from the emitter ($\chi^2 = 342.67$; df = 1), and exposure time ($\chi^2 = 223.79$; df = 1). All two-way interactions (insect age x wheat quantity, insect age x distance from the emitter, insect age x exposure time [df = 6]; wheat quantity x distance from the emitter, wheat quantity x exposure time, and distance from the emitter x exposure time [df = 1]), were all highly significant (χ^2 range = 47.11 – 565.57; P < 0.0001).

The number of *R. dominica* adults that emerged from infested wheat samples exposed to infrared radiation in various treatment combinations is shown in (Table 2:2). In general, the number of adults that emerged, or the probability of death, estimated by the logistic regression appeared to be influenced by grain quantity, distance from the emitter, and exposure time, which indirectly influenced temperatures attained by the wheat. Increasing the grain quantity or increasing the emitter distance resulted in lower probability of insect death. The relationship between insect age, wheat temperature, and probability of death was best depicted in Figure 2:5. Irrespective of insect age, the best treatment appeared to be 113.5 g of wheat exposed for 60 sec at a distance of 8.0 cm from the emitter, because in these treatments the mean temperatures attained ranged from 107.9 to 113.5°C, and the probability of death ranged from 0.99 to 1.00 (99.0 to 100.0% mortality).

The mean temperatures attained for a given grain quantity, emitter distance, and exposure time across the various insect ages were found not to be significantly different from one another. Therefore, mean temperature data over all the ages were averaged and the probability of death of various insect ages were plotted as a function of this average temperature (Figure 2:5) The graph revealed that at a given temperature, there was variation in how the various ages of insects responded to infrared, but differences in susceptibility to temperature tended to become smaller at temperatures >105°C. In general, based on the probability of death, 7-d old insects (young larvae) were the most susceptible whereas 21-d old insects (old larvae) were least susceptible.

The odds ratios also provided similar information. For example, the odds ratio for 21-d old insects was 0.94 when compared with the adults (odds ratio, 1.00), which were the next least susceptible stage to infrared. Odds ratios for the other stages in increasing order were eggs (1.45), 24-d old insects (2.35), 14-d old insects (2.41), and 7-d old insects (3.80).

Discussion

The experimental protocols used for grain infestation and infrared exposures were successful because adult progeny emerged from untreated wheat infested with various R. dominica ages. The eggs of R. dominica, which are laid outside the kernels (Arbogast 1991), take about 7-d to hatch (Hagstrum and Milliken 1988). This explains why we were unable to identify any first instars in the X-ray scans taken at 7-d. However, larvae inside kernels were visible after 8 d onwards. Potter (1935) opened wheat kernels and extricated larvae in different developmental stages and measured their head capsule widths. We did not use this destructive method, but instead used a non-destructive method, which involved measuring tunnel lengths and widths in kernels infested with developing larvae. These measurements have been reported previously for R. dominica by Potter (1935). Tunnel widths were also used to characterize the four instars of three stored-grain insects that develop within kernels of wheat such as the granary weevil, Sitophilus granarius (L.) (Kirkpatrick and Wilbur 1965); maize weevil, Sitophilus zeamais Motschulsky (Sharifi and Mills 1971a); and rice weevil, Sitophilus oryzae (L) (Sharifi and Mills 1971b). These authors showed four periods of tunnel widening indicative of four instars. In our study, the measured tunnel widths indicated five periods of tunnel widening indicative of five instars. Both Potter (1935) and Elek (1994) have reported R. dominica to go through four to five instars. The fact that we observed several adults within kernels in scans during the 28-d observation period, supports the view that R. dominica may have four instars. Schatkzi and Fine (1988) also noted that some adults remained inside kernels after complete development. Nevertheless, the X-ray confirmation and our experimental protocol used

provided a valid basis to gauge the impact of infrared treatments on various life stages of *R*. *dominica*.

The egg-to-adult development of *R. dominica* takes about 900 degree-days (DD) (Subramanyam et al. 1990). At 28°C and 65% RH, adults of *R. dominica* emerged between 30 and 36 d, which translates to 840 to 1008 DD. The mean egg-to-adult development in our study was 33 d or 924 DD. At 30°C and 56% RH, the mean egg-to-adult development of *R. dominica* on kibbled (cracked) wheat was 33.5-d (Elek 1994), whereas Howe (1965) at 28°C reported the mean egg-to-adult development to be 37-d.

In our study, it was surprising to observe consistently more adult progeny emerging from 227.0 g than 113.5 g of untreated wheat that was infested with 100 unsexed adults for only 3-d, and further studies are warranted to understand this phenomenon. The lowest number of progeny emerged was 212 adults while the highest was 581 adults. Toews et al. (2000) reported on progeny production when 100 g of each of eight United States wheat cultivars were infested with 50 unsexed *R. dominica* adults for 7 d. In their study, they carried out three separate experiments and progeny production was determined at 27° and 34°C and 70% RH. They found large differences in progeny production, which varied from a low of 123 to a high of 940 adults. It is hard to understand why more adults emerged from 227.0 g than 113.5 g of wheat, although obviously there were twice the number of kernels in 227.0 g of wheat. These differences could be related to differences in kernel size which may vary with the cultivar, environmental conditions (Toews et al. 2000), and differences in sex ratio of adults originally used to infest the wheat, since unsexed adults were used in our study.

The temperature measured by the infrared thermometer may include some errors, because temperature attained by the grain in the pan center was measured. The temperature differences across the emitter surface could have resulted in differential heating of grain, with the maximum heat being near the center (see Materials and Methods). It is possible that the steel pan was radiating and reflecting heat during infrared exposure or absorbed radiation may have been different between the two grain quantities. Given our experimental set up, it would be difficult to

eliminate some of these sources of error in temperature measurements and subsequent impact on insect survival and progeny production.

Of the factors examined, distance of wheat from the emitter and exposure time influenced mean temperatures attained. The quantity of grain used had minimal impact on mean temperatures attained. Fewer adults of *R. dominica* emerged in infrared treatments where the mean temperatures ranged from 108 to 114°C compared to treatments where the mean temperatures were below 105°C. The temperatures we observed were twice as high as those reported by Tilton and Schroeder (1963), Kirkpatrick (1975), Kirkpatrick and Tilton et al., (1972), Kirkpatrick and Cagle (1978), Tilton and Vardell (1983), and Pan et al., (2008) in various stored commodities, including wheat. Our results cannot be directly compared with work done by these authors for several reasons. In all of the previous work, except for Pan et al. (2008), the stage of development of insects within kernels that were exposed to infrared was unknown, and different from stages exposed in our study. Furthermore, the infested samples in previous work were subdivided (pseudoreplicates) for infrared exposure, and the type of infrared unit used was gas-fired and not a flameless catalytic emitter that we used. All authors, including Pan et al. (2008) measured grain temperatures immediately after infrared exposure, and hence their temperatures were lower than those reported in this paper.

In our laboratory tests, we noticed that the temperature of wheat drops rapidly soon after exposure to infrared radiation (data not shown). Pan et al. (2008) used a flameless catalytic emitter on rough rice of 20.6 and 25.0% moisture infested for 4 d with 100 adult *R. dominica* and they exposed it in a single layer (250 g) for 25, 40, 60, and 90 sec. Temperatures were measured using a Type T thermocouple (Omega Engineering Inc., Stamford, CT) immediately after heated rice was placed in a container. In 20.6 and 25.0% moisture rice, the temperatures attained ranged from 49.0 to 69.4°C. A 90-sec exposure was necessary for 99.5 to 100% mortality adults and/or eggs present in the infested samples.

The purpose of testing various ages of insects to infrared was to identify the most heat tolerant stage, because controlling the most heat tolerant stage may control all other stages. In

our study, susceptibility differences among the various ages of *R. dominica* were noted. The 21-d-old insects (old larvae) were more tolerant to infrared relative to the other stages, and the 7-d-old insects (young larvae) were more susceptible. Reasons for susceptibility differences among the life stages to infrared radiation should be explored further. The variation in susceptibility among *R. dominica* stages, especially those developing within kernels, may be related to the adverse effects of infrared on the insect's physiological processes. Except for the egg and adult stages, all other stages of *R. dominica* are spent within kernels. In the 113.5 and 227.0 g of infested wheat, it was difficult to know how the infestation was distributed among kernels relative to the portion of grain being heated. This could have had an effect on insect mortality or the number of adults that emerged. The location of the insect within the kernel may also influence its susceptibility to heat (Beckett and Morton 2003). In the case of adults, their ability to move away from areas that are hotter to seek cooler areas may make them less susceptible to heat. Similarly, some of the eggs could have escaped infrared treatment, perhaps by being shielded by kernels.

In conclusion, of all the infrared treatments, 113.5 g of wheat exposed for 60 sec at a distance of 8.0 cm from the emitter, was most effective treatment in disinfesting wheat containing eggs, larvae, pupae, and adults of *R. dominica*. Our laboratory results show that the flameless catalytic infrared technology is a viable tool for managing *R. dominica* life stages in stored wheat, provided the short exposures resulting in grain temperatures of 108 to 114°C do not adversely affect the end-use qualities of wheat.

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Table 2:1. Tunnel width (mm) for different ages of *R. dominica*.

Age (d)	Faxitron (Mean ± SE)	Width of thorax (abdomen) ^a
7 ^b	0.24 ± 0.03	0.198 (0.172)
14	0.34 ± 0.01	0.287 (0.242)
21	0.40 ± 0.02	0.635 (0.512)
24	0.53 ± 0.01	0.859 (0.776)
28	0.61 ± 0.02	0.845 (0.962)

^aPotter (1935; see text for details) showing actual thoracic and abdominal width of extricated immatures.

^bData based on observations on day 8.

Table 2:2. Emergence of adults from infested wheat unexposed to infrared containing various ages of R. $dominica^a$.

Insect age (d)	Mean \pm SE $(n = 4)$ number	er of adults in:
	113.5 g	227.0 g
0 (eggs)	437.3 ± 5.4	581.0 ± 14.3
7	300.0 ± 20.8	570.0 ± 29.0
14	228.3 ± 53.3	517.5 ± 68.2
21	211.5 ± 8.7	384.5 ± 39.9
24	241.5 ± 31.2	530.3 ± 29.0
28	284.8 ± 34.8	415.0 ± 42.1

 $^{^{\}mathrm{a}}$ The ages correspond to when R. dominica stages were exposed to infrared treatments.

Table 2:3. Emergence of *R. dominica* adults from infested wheat in various infrared-exposed treatments, mean temperature attained by wheat, and probability of insect death.

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec)	Mean temperature (°C)	Mean no. adults	Probability of death
0	113.5	8.0	45	102.3 ± 1.0	6.0 ± 0.8	0.97
			60	113.5 ± 0.5	0.7 ± 0.2	0.99
		12.7	45	87.6 ± 0.8	52.7 ± 5.4	0.88
			60	91.9 ± 0.8	14.7 ± 2.0	0.97
	227.0	8.0	45	102.1 ± 1.0	119.3 ± 8.7	0.83
			60	110.5 ± 1.4	18.3 ± 0.6	0.96
		12.7	45	85.4 ± 0.3	214.0 ± 12.3	0.57
			60	90.7 ± 0.8	190.0 ± 6.4	0.83
7	113.5	8.0	45	99.9 ± 0.3	0.3 ± 0.3	0.99
			60	111.5 ± 0.2	0.3 ± 0.3	1.00
		12.7	45	83.8 ± 0.3	10.7 ± 0.7	0.95
			60	91.4 ± 0.9	0.7 ± 0.3	0.99
	227.0	8.0	45	101.8 ± 0.6	28.3 ± 6.2	0.94
			60	109.3 ± 1.0	1.0 ± 0.6	0.99
		12.7	45	84.1 ± 0.3	187.0 ± 22.5	0.78

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec)	Mean temperature (°C)	Mean no. adults	Probability of death
			60	87.9 ± 0.7	51.7± 13.2	0.92
14	113.5	8.0	45	102.4 ± 1.0	0	1.00
			60	107.9 ± 0.7	0	1.00
		12.7	45	86.3 ± 0.3	14.7 ± 6.1	0.94
			60	90.8 ± 0.2	0	1.00
	227.0	8.0	45	100.6 ± 1.4	49.3 ± 6.3	0.90
			60	108.4 ± 0.7	5.0 ± 2.3	0.98
		12.7	45	82.9 ± 0.3	220.7 ± 15.6	0.68
			60	87.8 ± 0.3	65.7 ± 8.1	0.88
21	113.5	8.0	45	99.9 ± 0.5	3.0 ± 0.4	0.95
			60	108.5 ± 0.5	0	1.00
		12.7	45	84.6 ± 0.1	27.7 ± 6.9	0.85
			60	89.4 ± 0.5	20.3 ± 4.1	0.95
	227.0	8.0	45	100.3 ± 0.0	118.7 ± 5.2	0.77
			60	108.1 ± 0.6	22.7 ± 3.0	0.94
		12.7	45	83.9 ± 0.5	288.3 ± 9.2	0.47

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec)	Mean temperature (°C)	Mean no. adults	Probability of death
			60	88.8 ± 0.1	117.7 ± 11.6	0.76
24	113.5	8.0	45	102.9 ± 0.6	0.7 ± 0.2	0.98
			60	109.4 ± 0.3	0	1.00
		12.7	45	85.7 ± 0.3	28.3 ± 10.5	0.93
			60	90.4 ± 1.0	0.7 ± 0.2	0.98
	227.0	8.0	45	101.7 ± 0.4	49.7 ± 10.1	0.90
			60	109.0 ± 0.3	15.7 ± 2.1	0.97
		12.7	45	83.9 ± 0.3	173.3 ± 19.1	0.68
			60	89.2 ± 0.7	73.0 ± 3.6	0.88
28	113.5	8.0	45	76.2 ± 24.1	5.7 ± 1.0	0.95
			60	110.2 ± 0.4	0.3 ± 0.2	0.99
		12.7	45	84.9 ± 0.3	53.0 ± 10.6	0.87
			60	91.4 ± 0.7	2.7 ± 0.9	0.97
	227.0	8.0	45	100.7 ± 0.8	10.3 ± 1.8	0.81
			60	107.6 ± 1.4	37.7 ± 9.0	0.94
		12.7	45	82.6 ± 0.3	194.7 ± 11.3	0.50

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec)	Mean temperature (°C)	Mean no.	Probability of death
			60	88.4 ± 0.3	125.0 ± 13.7	0.79
42 d (adults)	113.5	8.0	45	102.3 ± 1.0	0.3 ± 0.2	0.96
			60	109.7 ± 0.9	0	1.00
		12.7	45	85.9 ± 0.3	7.7 ± 1.0	0.86
			60	91.6 ± 0.9	0.7 ± 0.2	0.96
	227.0	8.0	45	100.8 ± 0.9	8.3 ± 0.6	0.81
			60	110.2 ± 0.2	0	1.00
		12.7	45	84.0 ± 0.4	91.0 ± 1.1	0.45
			60	90.1 ± 0.2	21.3 ± 1.4	0.77

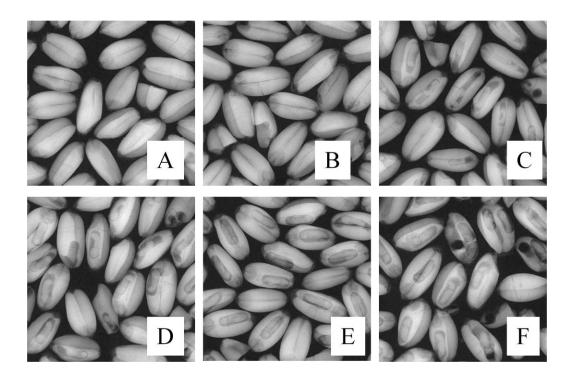


Figure 2:1. Radiographic images of *R. dominica* life stages within kernels on days 0 (A), 7 (B), 14 (C), 21 (D), 24 (E), and 28 (F).

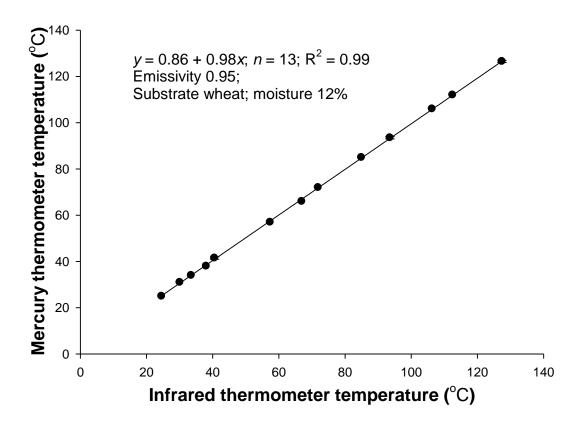


Figure 2:2. Calibration curve for infrared thermometer.

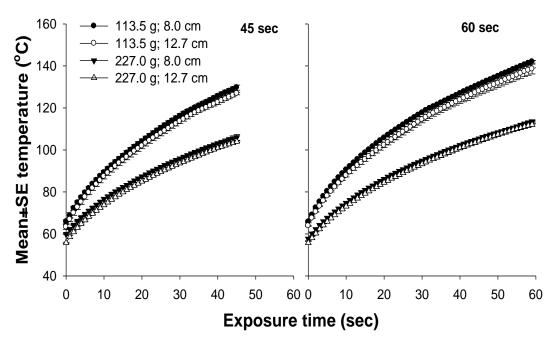


Figure 2:3. A generalized time-dependent temperature profile attained with different quantities of wheat exposed at 8.0 and 12.7 cm from the emitter for 45 or 60 sec.

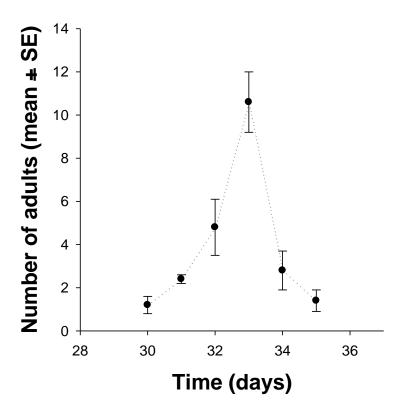


Figure 2:4. Adult emergence patterns of *R. dominica* in 24-ml wheat samples (n = 5).

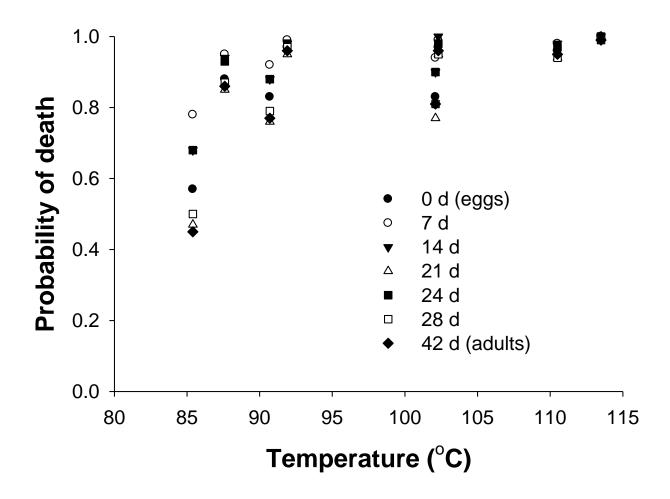


Figure 2:5. Probability of death of different life stages of *R. dominica* at various mean wheat temperatures attained.

CHAPTER 3 - Effect of flameless catalytic infrared emitter on *Sitophilus oryzae* (L.) Life Stages

Abstract

A flameless laboratory bench top flameless catalytic infrared emitter was evaluated against life stages of the rice weevil, *Sitophilus oryzae* (L.), an insect species associated with stored wheat. The infrared radiation emitted was in the 3-7 µm range. A non-contact infrared thermometer measured grain temperatures instantaneously during exposures of infested wheat. Insect mortality was a function of final grain temperature attained. In general, higher grain temperatures were attained when using 113.5 versus 227.0 g of wheat, at 8.0 cm from the heater versus 12.7 cm during a 60 sec exposure versus a 45 sec exposure. Complete mortality of all life stages of *S. oryzae* was achieved at 8.0 cm from the emitter using 113.5 g of wheat, during a 60-sec exposure, and the mean grain temperatures attained ranged from 108.4 to 111.8°C. The log odds ratio tests showed that eggs (0-d old) were the least susceptible to infrared, followed by adults within kernels (28-d old), pupae (24 d- old), young larvae (7-d-old), larvae that were 14 to 21-d-old, and adults (42-d-old). These data indicate infrared to be a viable option for disinfesting wheat containing various ages of *S. oryzae*.

Introduction

There is an increasing need for evaluating alternatives to traditional pesticides for managing insect pests associated with grain and grain products in light of the changing regulatory climate and demand for pesticide-residue free foods by consumers (Hagstrum and Subramanyam 2000, Hagstrum and Subramanyam 2006). In addition, some stored-grain insects have developed resistance to existing pesticides (Subramanyam and Hagstrum 1996, Sinha and Watters 1985). Therefore, there is a need to explore new and effective alternatives to pesticides. New government laws such as the 1996 Food Quality Protection Act have made registration of new pesticides more stringent to meet current food and public health safety standards.

More than two decades ago, several scientists evaluated gas fired infrared radiation to disinfest rice and other cereal grains (Tilton and Schroeder 1963, Cogburn 1967, Cogburn et al. 1971, Kirkpatrick and Tilton 1972, Kirkpatrick et al.1972, Tilton et al. 1983). They achieved 100% mortality of 12 adult insect species associated with grain where the final grain temperatures attained were 56 to 63°C. The gas fired infrared radiation used was generated with propane combusted on ceramic tiles producing more than 14.07kW/h (48,000 BTU/h) of heat energy (Tilton et al. 1961, Kirkpatrick and Cagle 1978). However, these gas-fired infrared generators had an open flame and produced temperatures that exceeded 900°C. Such high temperatures and open flames are not suitable for use in dusty grain storage and handling facilities due to explosions hazards. In previous evaluations of infrared radiation, the different life stages of insects developing with kernels were not mentioned, and the grain temperatures were measured after infrared exposure and not in "real time", which resulted in under reporting actual temperatures attained by the grain.

Flameless catalytic infrared radiation is a new technology developed by Catalytic Drying Technologies LLC, Independence, KS (www.catalyticdrying.com). It has been used primarily in the natural gas industry to heat pipes and in the automobile industry to dry paints. The flameless infrared radiation is emitted when propane or natural gas is combusted in the presence of a platinum catalyst resulting in temperatures of about 400°C at the emitter surface. The only other co-products of this chemical reaction are water and carbon dioxide (Gabel et al. 2006, Pan et al. 2008). Infrared radiation has been successfully used to inactive enzymes and pathogenic bacteria, dehydrate food commodities, and disinfest commodities (Sandu 1986, Gabel et al. 2006, Pan et al. 2008).

In the present investigation, the effectiveness of the flameless catalytic infrared radiation was evaluated against different life stages of the rice weevil, *Sitophilus oryzae* (L.), an economically important internal pest of stored wheat (Sinha and Watters 1985). Specific objectives were to examine the influence of grain quantity, distance from the infrared emitter, and exposure time on susceptibility of various *S. oryzae* life stages.

Materials and Methods

Insects Rearing. Cultures of *S. oryzae* were reared on 12% moisture organic wheat (var. Jagger), obtained from Heartland Mills, Marienthal, KS, at 28°C, 65% RH, and 14:10 (L:D) h photoperiod in a growth chamber (Model I-36 VL, Percival Scientific, Perry, IA).

Identifying Life Stages of S. oryzae. In 24-ml vials, 5 g of 12% moisture wheat were infested with 20 unsexed adults of S. oryzae. There were five such vials. After 3 d, the adults were removed and the samples were incubated at 28°C and 65% RH to obtain life stages of a specific age. Wheat samples infested for 3 d represented the egg stage (day 0), and samples infested and incubated for 7, 14, 21, 24, and 28 d after day 0 represented various ages of S. oryzae. Each 5-g wheat sample in a vial was subjected to X-ray analysis (Model 43855A, Faxitron X-ray Corporation, Lincolnshire, IL) to obtain images of insects at age 0, 7, 14, 21, 24, and 28 d. These samples were spread in a monolayer on the sample holder and subjected to Xrays for 10 sec at 28kV. X-ray images were acquired using a digital camera. The digital images were used to score ages of insects developing within wheat kernels. The digital images were given to six graduates students in the Department of Grain Science and Industry, Kansas State University, with varying experiences with S. oryzae. The students were asked to classify the ages of S. oryzae based on the images. An inaccurate classification resulted in a "0" score and an accurate classification resulted in a "1" score, and these values were tallied to determine the accuracy (expressed as a percentage of total classifying correctly) among the six students in correctly age-grading S. oryzae. In addition, ages of each stage were determined by measuring the tunnel width within kernels made by developing immature stages. To measure the tunnel width, the top, middle, and bottom potions of the tunnel from 48 infested kernels at a given insect age were measured and averaged.

Grain Infestation and Infrared Treatment. The factors influencing effectiveness of infrared against *S. oryzae* life stages included grain quantity exposed (113.5 and 227.0 g),

distance from the emitter (8.0 cm and 12.7 cm), and exposure time (45 sec and 60 sec). Wheat (113.5 or 227.0 g) of 12% moisture was placed in 0.45-liter glass mason jars fitted with filter paper and mesh lids, and each jar was infested with ~100 unsexed *S. oryzae* adults of mixed ages. After 3 d, all introduced adults were separated from the wheat and the wheat was incubated at 28°C and 65% RH for various time periods to obtain different *S. oryzae* ages for infrared exposure. The 3-d old jars represented the egg stage (day 0) of *S. oryzae*, while those held for 7, 14, 21, 24 and 28 d following day 0 represented various life stages. The 28 d infested sample represented both adults that emerged and those that did not emerge from the kernels. Adults of *S. oryzae* were not separately added to wheat and exposed to infrared; however, wheat that was infested as explained above and held for 42 d from day 0 represented the adult stage.

A bench top model, donated by Catalytic Drying Technologies LLC, was used for infrared exposure. The bench top model has a circular heating surface of 613.36 cm², and propane from a 473-ml container (Ozark Trail Propane Fuel, Bentonville, AR) was the fuel used to start the initial reaction delivered at 27.9 cm of water column pressure. The total heat energy output of the unit is 1.47kW/h (5,000 BTU/h).

Wheat samples were exposed to infrared in a 3.8 cm deep steel pan of 27.94 cm diameter with a 43 cm long handle. Each infested grain quantity was exposed at 8.0 and 12.7 cm from the emitter surface for 45 or 60 sec. Independent samples were exposed for each treatment combination, and each treatment combination was replicated three times. Wheat infested similarly, but unexposed to infrared served as the control treatment, and the control treatments were replicated four times.

The temperature of wheat in pan during infrared exposure was measured continuously at the center of the steel pan using a non-contact thermometer infrared thermometer (Raynger MX4 Model 4TP78, Raytek®, Santa Cruz, CA), placed away from interference from the emitter surface. The infrared thermometer works in 8 to14 µm range. The non-contact thermometer was connected to a laptop computer via a RS-232 cable to record "real time" grain temperatures using a data acquisition program developed by the Electronic Design Laboratory at Kansas State University in LABView (National Instruments Corporation, Austin, TX). The emissivity of the infrared thermometer was set to 0.95, and the temperatures recorded by the infrared thermometer were as accurate as those measured by a mercury thermometer based on calibration experiments (Khamis 2009).

Assessment of Insect Mortality. Insect mortalities for 0, 7, 14, 21, 24, and 28 d samples in untreated wheat and wheat exposed to infrared were based on counting adults that emerged from kernels at ~42 d from time 0. Since *S. oryzae* completes development within kernels, the effectiveness of infrared can be ascertained by examining the number of adults that emerged in infrared-exposed wheat relative to emergence in untreated wheat. In 42-d-old infested untreated samples *S. oryzae* adults had already emerged from kernels. Therefore, mortality of adults exposed to infrared for this age group was determined directly by counting number of live and dead adults. Adults were counted 24 h after exposure in untreated control and infrared-exposed wheat samples.

Experimental Design and Data Analysis. The experiment was run as a completely randomized design. The time-dependent temperature profile, averaged every second from replicated data, was plotted as a function of time for 113.5 and 227.0 g of wheat exposed for 45 and 60 sec at 8.0 and 12.7 cm from the infrared emitter. There were eight temperature profiles for each insect age. A comparison of temperature profiles across various ages showed that for any given quantity of grain, distance from emitter, and exposure time, the profiles were essentially similar.

The time-dependent temperature profile for each replicate was averaged over time to obtain a mean temperature attained by wheat during the exposure period. The mean wheat temperature attained for any given insect age, wheat quantity, and exposure time combination between 8.0 cm and 12.7 cm distance from the emitter surface was compared ($\alpha = 0.05$) using two-sample *t*-tests for equal variances (SAS Institute 2002). Two-sample *t*-tests were used to compare differences in mean temperatures attained between a 45 and 60 sec exposure at any given insect age, grain quanity and distance from heater. Additionally, comparisons were also made of mean temperatures attained between 113.5 and 227.0 g of grain at any given insect age, distance from heater, and exposure time.

In order to determine if the mean wheat temperature attained for a given quantity of grain, distance from emitter, and exposure time combination was consistent across the various ages tested (eggs [day 0], 7, 14, 21, 24, 28, and 42 d [adult emergence]) a linear regression of temperature versus insect age was performed and the slope was tested for deviation from zero (SAS Institute 2002).

The main effect of insect age, wheat quantity, distance from emitter, and exposure time and their two-way interactions on the probability of death were determined using logistic regression at $\alpha = 0.05$ (SAS Institute 2002). Odds ratios from logistic regression were used to show differences in susceptibility (odds of dying) of various life stages exposed to infrared. The odds ratio for adults (1) was used as a reference. A ratio >1 showed that a life stage was more susceptible than adults to infrared while a ratio <1 showed that a stage was less susceptible than adults. Differences in susceptibility of various life stages was also determined by plotting probability of death as a function of mean wheat temperature averaged over wheat quantity, distance from heater, and exposure time.

Results

Identifying *S.oryzae* Life Stages. Immatures of *S.oryzae* complete development inside the kernels, and therefore, the different life stages had to be confirmed using radiographic techniques. Eggs and different instars of *S.oryzae* were difficult to identify and distinguish in the kernel with the Faxitron, except at higher magnifications (Figure 3:1). Larvae in various phases of development were predominant between days 7 and 21, whereas pupae stages were predominant on day 24. In addition to pupae, a few old larvae were occasionally observed within kernels on day 24. A few *S. oryzae* adults were observed on day 28 inside the kernels; however by this time, most had already emerged as adults (Figure 3:3). Adult emergence ceased after day 30.

Tunnel widths (mean \pm SE) due to egg deposition (day 0) or developing larvae that are 7, 14, and 21-d-old larvae were measured. Tunnel widths for 0, 7, 14, and 21 d insect ages were 0.26 ± 0.01 , 0.57 ± 0.03 , 0.72 ± 0.01 , and 1.39 ± 0.01 mm, respectively. All six students were able to accurately identify the various insect ages from digital images.

Adult Emergence in Untreated Samples. Consistently more *S. oryzae* adults emerged in 227.0 g of untreated, infested wheat compared with 113.5 g of wheat (Table 3:1). The mean number of adults that emerged in 113.5 g of wheat across the various insect ages ranged from 216.8 to 281.3 whereas in 227.0 g of wheat it ranged from 380.0 to 447.3.

Temperatures Attained During Infrared Exposure. Higher temperatures were attained by the same grain quantity exposed at same distance but at longer exposure times. Temperature profiles for a given grain quantity, distance from emitter, and exposure time were similar,

irrespective of the insect age. Therefore, it was deemed necessary to show typical temperature profiles attained for the two grain quantities, two distances from the emitter, and the two exposure times (Figures 3:2). The mean grain temperature attained was greatest (112.8°C) in 113.5 g of wheat exposed for 60 sec at a distance of 8.0 cm from the emitter (Table 3:3). The lowest mean temperature attained (79.2°C) was in 227.0 g of wheat exposed for 45 sec at a distance of 12.7 cm from the emitter.

Two sample *t*-tests for each life stage or *S. oryzae* age group (0, 7, 14, 21, 24, 28, and 42 d) showed that the mean temperature attained by 113.5 or 227.0 g of wheat during a 45 or 60-sec exposure was significantly greater at 8.0 cm from the emitter when compared with mean temperature attained by wheat at 12.7 cm from the emitter (*t*, range among ages, grain quantities, and exposure times = 7.12 - 56.58; df = 4; P < 0.0001). The mean temperature attained by wheat after a 60-sec exposure was significantly and consistently greater than those attained after a 45-sec exposure at a given insect age, grain quantity, and distance from the emitter (*t*, range = -2.89 - -39.50; df = 4; $P \le 0.0447$), except in four instances where mean temperature attained after a 45 sec and 60 sec exposure were similar (*t*, range = -1.19 - -2.39; df = 4; $P \ge 0.0749$).

In general, grain quantity had the least influence on the mean grain temperatures attained for any given insect age, distance from emitter, and exposure time. In 20 out of the 28 comparisons, the difference in mean temperature attained by 113.5 and 227.0 g of wheat was not significant (t, range = -2.75 – 2.51; df = 4; $P \ge 0.5130$). In eight other cases, mean temperatures attained by wheat were higher with 113.5 g than with 227.0 g (t, range = -4.29 – 8.10; df = 4; t = 0.0018).

The slope of the linear regression between the mean temperature attained by 113.5 or 227.0 g of wheat at 8.0 or 12.7 cm from the emitter after a 45 or 60 sec exposure and insect age was not significantly different from zero (t, range among grain quantities, distance from emitter, and exposure times = -1.86 – 1.54; n = 7; $P \ge 0.1222$).

Adults of *S. oryzae* were not observed in 20 out of the 56 infrared treatment combinations (Table 3:2), indicating complete mortality. In the remaining 36 infrared treatment combinations, the mean number of adults that emerged ranged from 0.3 to 295.7. Insect mortality in most cases was directly related to mean grain temperatures attained. Complete mortality of all insect ages was achieved in 113.5 g of grain, exposed for 60 sec, at 8.0 cm from the emitter surface.

Exposure of all insect ages in 227.0 g of wheat for 45 sec at a distance 12.7 cm from the emitter surface resulted in 46 to 99% mortality.

Logistic regression indicated that insect age ($\chi^2 = 1438.81$; df = 6 P < 0.0001), wheat quantity ($\chi^2 = 5.88.10$; df = 1 P < 0.0153), distance from emitter ($\chi^2 = 114.68$; df = 1 P < 0.0001), and exposure time ($\chi^2 = 5.055$; df = 1 P < 0.0246) influenced the probability of death of *S. oryzae*. All two-way interactions (insect age x wheat quantity, insect age x distance from emitter, insect age x exposure time [df = 6]; wheat quantity x distance from emitter, wheat quantity x exposure time, and distance from emitter x exposure time were highly significant (χ^2 range = 7.80 - 211.61; P = < 0.0001 - 0.0052).

The probabilities of death plotted as a function of mean temperatures showed variation among stages. Increasing the temperatures resulted in higher probabilities of death, irrespective of the life stage, and these probabilities were close to 1 (100% mortality). Generally, a temperature of about 100°C was needed to achieve 100% mortality, irrespective of insect age (Figure 3:4). In general, eggs were the least susceptible stage followed by adults within kernels (28-d-old), pupae (24-d-old), young to old larvae (7- to 21-d old), and 42-d-old adults.

Discussion

Females of *S. oryzae* after mating chew a shallow hole on the kernel to lay a single egg. After egg laying, the hole is sealed with a gelatinous plug (Kirkpatrick and Wilbur 1965, Sharifi and Mills 1971). Larvae hatch from eggs and complete development within kernels (Howe 1965, Arthur and Throne 2003, Pearson et al. 2003). Instars of *S. oryzae* within wheat kernels can be identified by head capsule width or by tunnel widths produced by developing larvae (Sharifi and Mills 1971). Sharifi and Mills (1971) reported both head capsule widths and tunnel widths for the four *S. oryzae* instars. The mean tunnel widths, indicative of each instar, reported by Sharifi and Mills (1971) was 0.31 (range, 0.30 - 0.37), 0.51 (0.43 - 0.62), 0.83 (0.70 - 0.97), and 1.34 (1.20 - 1.48) mm, respectively. The measured mean tunnel width on day 0 in our study was 0.26. At this time only eggs were present and not first instars, which is in agreement with that of Sharifi and Mills (1971) who reported the diameter of the egg cavity to be 0.25 to 0.30 mm. The tunnel widths for 7, 14, and 21-d-old larvae in our study were within the ranges reported by Sharifi and Mills (1971) for second through fourth instars.

Radiographic analysis is one of many methods (Dennis and Decker 1962, Haff and Slaughter 2004) to determine internal infestation. The age or stage of internally developing insects, including *S. oryzae*, was unknown in previous research with infrared treatments of grain (Tilton and Schroeder 1962, Cogburn 1967). Therefore, we used radiographic analysis to determine insects stage subjected to infrared treatments. The accurate scoring by students also indicated that the digital images alone were sufficient to age-grade *S. oryzae* by visual inspection.

Campbell (2002) reported that *S. oryzae* laid more eggs in kernels that were ≥ 20 mg. The consistently greater progeny production in 227.0 g of wheat compared with 113.5 g could be due to the availability of more kernels that weighed ≥20 mg, or just natural variability in the number of eggs laid by the mixed age adults used in our study. For example, Soderstrom (1960) infested 150 g of wheat grain with 150 unsexed *S. oryzae* adults (1:1 ratio) for 3 d, the same time period as ours. The mean number of adults that emerged ranged from 162 to 258. Cogburn (1967) infested 2,200 g of rice (infestation density of 1 insect/g) with *S. oryzae* for 7 d. This amount of rice was then divided into 50 g lots. The number of adults that emerged from untreated wheat lots ranged from 144 to 285. The fact that large number of adults emerged from infested untreated wheat samples indicated that our experimental protocol was robust, and can be used to gauge the effectiveness of infrared radiation by comparing adults emergence in infrared exposed versus untreated samples.

Exposure time and distance from emitter had a greater influence on the mean temperatures attained than grain quantity. Generally, mortality of insect stages was directly related to mean temperature attained. However, there were instances where the same mean temperature did not result in same mean probability of death for a given insect stage. For example, in Table 3:2 the mean temperature attained by 113.5 and 227.0 g of wheat exposed for 45 sec at 12.7 cm from the emitter was essentially similar (81°C). However, the probability of death was 0.89 in 113.5 g of wheat and 0.46 in 227.0 g of wheat. In other cases, the probability of death for a given stage was the same, but the mean temperatures attained were different. The plot of probability of death as a function of insect age also showed variability in how different stages responded to the same mean temperatures, especially below 100°C. These findings suggest that mean temperature alone may not be a good predictor of insect death. An understanding of the amount of infrared radiation that is reflected, scattered, and absorbed at the

two grain quantities, exposure times, and distances may shed some light on the insect responses observed.

We found eggs to be the most tolerant to infrared. Kirkpatrick (1975) reported only 8% percent mortality of eggs and first instars exposed to infrared radiation. Flameless catalytic infrared radiation is a viable tool for disinfesting organic and non-organic stored wheat, and our study showed that temperatures of 90 to 112°C were necessary to produce 99 to 100% mortality of all life stages of *S. oryzae*. Future work should focus on developing customized on-line systems capable of rapid disinfestation of wheat at farm bin sites, grain elevators, and grain-processing facilities.

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Table 3:1. Emergence of adults from infested wheat unexposed to infrared containing various ages of *S. oryzae*^a.

Insect age (d)	Mean \pm SE ($n = 4$) number of adults is				
	113.5 g	227.0 g			
0 (eggs)	281.3 ± 22.0	403.3 ± 11.6			
7	242.3 ± 39.2	403.3 ± 35.0			
14	244.8 ± 29.6	380.0 ± 21.5			
21	225.8 ± 12.8	398.3 ± 24.3			
24	226.0 ± 9.9	395.0 ± 16.4			
28	246.3 ± 44.4	447.3 ± 24.1			
42	216.8 ± 17.6	385.5 ± 18.6			

^aThe ages correspond to when *S. oryzae* was exposed to infrared treatments.

Table 3:2. Emergence of *S. oryzae* adults from infested wheat in various infrared-exposed treatments, mean temperature attained by wheat, and probability of insect death.

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec)	Mean temperature (°C)	Mean no. adults	Probability of death
0	113.5	8.0	45	103.2 ± 0.4	0	0.98
			60	108.4 ± 1.4	0	1.00
		12.7	45	81.1 ± 1.2	49.0 ± 2.6	0.89
			60	90.5 ± 0.8	0	0.99
	227.0	8.0	45	99.9 ± 0.1	139.3 ± 7.8	0.80
			60	107.5 ± 1.4	26.0 ± 3.2	0.92
		12.7	45	81.3 ± 0.4	295.7 ± 15.0	0.46
			60	87.6 ± 0.4	210.7 ± 3.9	0.69
7	113.5	8.0	45	100.9 ± 0.9	0	1.00
			60	109.9 ± 0.6	0	1.00
		12.7	45	82.7 ± 2.4	0	0.99
			60	86.1 ± 1.5	0	0.99
	227.0	8.0	45	102.1 ± 0.6	0	0.98
			60	109.1 ± 0.6	0	1.00
		12.7	45	81.3 ± 1.3	12.0 ± 4.3	0.98

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec	Mean temperature (°C)	Mean no adults	Probability of death
			60	84.1 ± 0.5	0	0.95
14	113.5	8.0	45	106.2 ± 0.5	0	1.00
			60	112.3 ± 1.0	0	1.00
		12.7	45	84.9 ± 0.2	0	0.96
			60	88.6 ± 0.8	0	1.00
	227.0	8.0	45	102.3 ± 0.1	3.0 ± 2.1	0.98
			60	108.4 ± 0.2	0.3 ± 0.0	1.00
		12.7	45	81.8 ± 0.6	20.7 ± 5.5	0.68
			60	86.7 ± 1.1	3.0 ± 0.0	0.96
21	113.5	8.0	45	100.5 ± 0.8	0	1.00
			60	110.1 ± 0.8	0	1.00
		12.7	45	84.6 ± 0.1	0.7 ± 0.0	0.99
			60	92.4 ± 1.1	0	1.00
	227.0	8.0	45	101.4 ± 0.7	4.7 ± 2.7	0.98
			60	111.8 ± 0.6	0.3 ± 0.0	1.00
		12.7	45	81.7 ± 0.7	37.0 ± 7.0	0.92
			60	88.6 ± 0.5	6.0 ± 1.0	0.97

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec	Mean temperature (°C)	Mean no adults	Probability of death
24	113.5	8.0	45	104.7 ± 1.5	0.3 ± 0.0	0.99
			60	110.1 ± 0.8	0	1.00
		12.7	45	82.8 ± 0.5	4.3 ± 0.7	0.97
			60	89.7 ± 0.5	1.3 ± 0.7	0.99
	227.0	8.0	45	102.5 ± 0.0	47.7 ± 22.8	0.95
			60	109.8 ± 0.6	8.0 ± 3.8	0.98
		12.7	45	82.9 ± 0.5	114.3 ± 23.3	0.97
			60	87.4 ± 0.5	55.0 ± 15.9	0.99
28	113.5	8.0	45	102.2 ± 0.7	0	0.99
			60	110.7 ± 0.9	0	1.00
		12.7	45	84.9 ± 0.5	16.8 ± 7.2	0.97
			60	90.8 ± 0.6	1.0 ± 0.6	0.99
	227.0	8.0	45	102.7 ± 0.8	21.2 ± 12.1	0.92
			60	110.4 ± 0.4	0.5 ± 0.0	0.97
		12.7	45	83.2 ± 0.3	114.8 ± 6.9	0.71
			60	87.9 ± 0.3	17.2 ± 7.7	0.79
42 (adults)	113.5	8.0	45	106.0 ± 0.8	0	1.00

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec	Mean temperature (°C)	Mean no adults	Probability of death
			60	111.8 ± 1.1	1.0 ± 0.0	1.00
		12.7	45	86.1 ± 1.6	12.3 ± 1.5	1.00
			60	91.0 ± 0.7	0	1.00
	227.0	8.0	45	101.0 ± 1.5	10.7 ± 5.8	1.00
			60	111.8 ± 1.1	1.0 ± 0.0	1.00
		12.7	45	82.1 ± 1.3	60.0 ± 12.3	0.94
			60	88.7 ± 0.6	48.0 ± 3.6	0.98

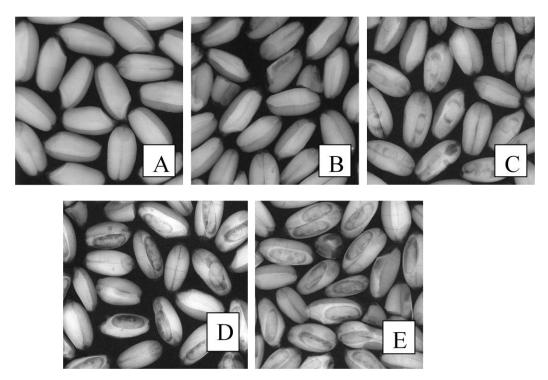


Figure 3:1. Radiographic images of *S. oryzae* life stages within kernels on days 0 (A), 7 (B). 14 (C), 21 (D) and 24 (E).

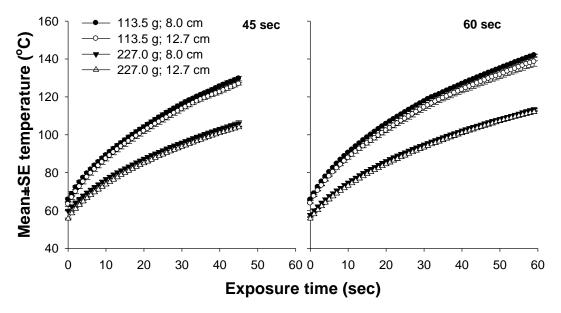


Figure 3:2. A generalized time-dependent temperature profile attained with different quantities of wheat exposed at 8.0 and 12.7 cm from the emitter for 45 or 60 sec.

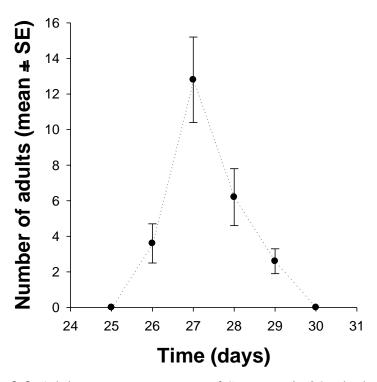


Figure 3:3. Adult emergence patterns of *S. oryzae* in 24-ml wheat samples (n = 5).

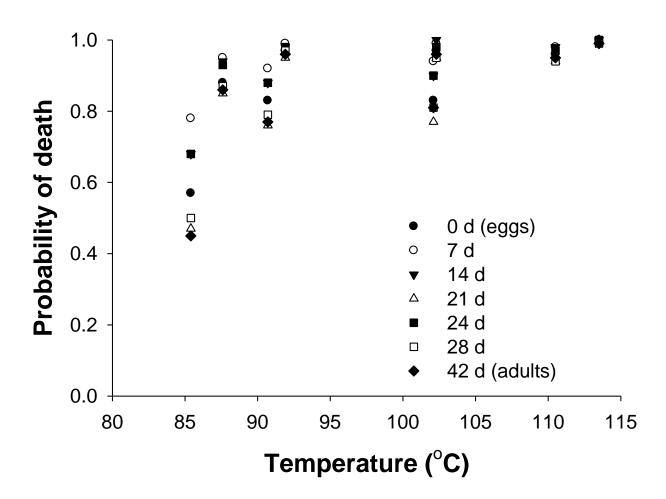


Figure 3:4. Probability of death of different life stages of *S. oryzae* at various mean wheat temperatures attained.

CHAPTER 4 - Evaluation of catalytic infrared radiation against life stages of *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae)

Abstract

The susceptibility of various life stages of *T. castaneum* to flameless catalytic infrared radiation in the 3 to 7 μm range was evaluated in the laboratory. Eggs (day 0), 7-, 14-, and 21-d-old larvae; pupae, and 2-wk-old adults (42-d-old from day 0), were exposed for 45 or 60 sec in 113.5 or 227.0 g of wheat at a distance of 8.0 or 12.7 cm from the emitter. The mean temperatures attained during exposures were measured continuously using a non-contact infrared thermometer. The mean grain temperatures attained increased with an increase in exposure time, and were inversely related to distance from the emitter. Grain quantity did not greatly influence mean temperatures attained. Pupae were the least susceptible stage and 7-day-old larvae were the most susceptible stage. Variation in probability of death of various life stages tended to decrease with an increase in mean grain temperatures attained, and complete control of all life stages occurred at mean grain temperatures between 108 and 111°C. All life stages were killed after a 60 sec exposure at 8.0 cm from the emitter in 113.5 g of wheat. Our laboratory results showed flameless catalytic infrared radiation to be a valuable tool to manage insects in stored organic and non-organic wheat.

Introduction

Wheat in storage is susceptible to spoilage by insects, molds, and bacteria. The damage contributed by insects to stored wheat depends on the length of storage, grain moisture, and the effectiveness of pest management tactics (Storey et al. 1984, Reed and Pedersen 1987, Kenkel et al. 1993, Martin et al. 1997). The most common and damaging insect species associated with wheat stored on farms and at elevators in Kansas and neighboring states is the lesser grain borer, *Rhyzopertha dominica* (F.) (Dowdy and McGaughey 1994, Reed et al. 1991, Vela-Coiffier et al. 1997, Reed et. al. 2003). The rice weevil, *Sitophilus oryzae* (L.), and red flour beetle, *Tribolium castaneum* (Herbst), are found to a lesser extent in wheat stored on farms and at elevators in Kansas (Reed et al. 1991, Reed et al. 2003); however, the latter species is more common than the former. Activity of all three species of insects was observed outside farm bins (Dowdy and McGaughey 1994).

There has been documented resistance to traditionally used organophosphate grain protectants and the fumigant phosphine in *R. dominica*, *S. oryzae*, *T. castaneum* (Subramanyam and Hagstrum 1996). In addition, the use of grain protectants leads to presence of residues, which may unacceptable to consumers and foreign buyers of US grains. Therefore, there is a need to explore effective alternatives, especially environmentally-benign technologies, to replace or complement currently used insecticides for protecting stored grains.

Infrared radiation in the 3 to 7 µm range is one such technology that has shown promise against insects associated with stored grains in laboratory tests (Schroeder 1960, Schroeder and Rosberg 1960, Tilton and Schroeder 1963, Cogburn 1967, Cogburn et al. 1971, Kirkpatrick 1973, Kirkpatrick and Tilton 1972, Kirkpatrick et al. 1972, Tilton et al. 1983). The infrared radiation in these previous evaluations was generated when natural gas or propane combusted over ceramic panels in the presence of oxygen. These gas-fired radiation sources were of high intensity producing 14.07 kW/h (48,000 BTU/h) of heat energy and temperatures were close to 930°C. The open flame and high temperatures are undesirable for use in grain-handling establishments or mills for continuous on-line disinfestation of grain because of explosion hazards. In all of the previous studies reported above, grain temperatures were measured after infrared exposure, resulting in underestimating the temperatures actually attained by grains. Additionally, the exact life stages of insects exposed were not confirmed.

Flameless catalytic infrared radiation is a new technology developed by Catalytic Drying Technologies, LLC (Independence, KS) (http://www.catalyticdrying.com). In flameless infrared emitters, propane or natural gas chemically react with oxygen in the presence of a platinum catalyst below flame temperatures (500°C), delivering peak infrared radiant energy in the 3 to 7 µm range. The only co-products of this reaction are carbon dioxide and water. Catalytic Drying Technologies, LLC, received an award from the United States Environmental Protection Agency's Pollution Prevention Program for developing flameless infrared radiation sources, under the "Environmentally Preferable Products" category. The objective of this research was to evaluate the susceptibility of different life stages of *T. castaneum* exposed to flameless catalytic infrared radiation energy.

Materials and Methods

Insects Rearing. Laboratory cultures of *T. castaneum* were reared under controlled conditions in the Department of Grain Science and Industry, Kansas State University, Manhattan, KS, on 95% bleached wheat flour and 5% (by wt) of brewer's yeast (Lesaffre Yeast Corporation, Milwaukee, WI) in growth chambers at 28°C, 65% RH, and 14:10 (L:D) h.

Obtaining Life Stages (Ages) of *T. castaneum.* To obtain various life stages or ages of *T. castaneum*, 100 unsexed 2-wk-old adults from cultures were used to infest several jars containing 100 g of flour plus yeast diet. Prior to use in tests, the diet was sifted through a 250 μm sieve (Seedburo Equipment Company, Chicago, IL). The procedures for extracting eggs, young larvae, old larvae, pupae, and adults of *T. castaneum* for use in tests were similar to those described by Mahroof et al. (2003). To collect eggs, contents in jars with adults were sifted after one day using 840 μm and 250 μm sieves. Adults were retained on the 840 μm sieve and eggs on the 250 μm sieve. Eggs were counted and gently removed with a hairbrush onto 9 cm glass Petri dishes. The eggs represented age 0 (day 0) for *T. castaneum*. To extract other life stages, jars infested with adults were held at 28°C and 65% RH for 7, 14, and 21 d after day 0 to obtain different ages of larvae. Jars held for 24 d represented the pupal stage. Adults were evident on day 28. Adults from these jars were held for another two weeks to obtain 2-wk-old or 42 d-old insects. Adults were obtained by sifting the jar contents over a 840 μm sieve.

Grain Infestation and Infrared Treatments. Three factors that influence grain temperatures attained and consequently insect responses were evaluated. The factors explored

included grain quantity (113.5 and 227.0 g), distance from the emitter (8.0 cm and 12.7 cm), and exposure time (45 and 60 sec). Organic wheat (Heartland Mills, Marienthal, KS) was weighed (113.5 g or 227.0 g) and placed in individual 0.45-liter glass jars and covered with wire mesh and filter paper lids. These jars were placed in a growth chamber at 28°C and 65% RH to equilibrate the moisture content to 12%. Equilibrated wheat for the various treatments received 100 *T. castaneum* individuals of a specific age, except in the case of eggs where 50 individuals were added to the wheat. Infested wheat was exposed to the bench top infrared emitter for 45 or 60 sec at a distance of 8.0 or 12.0 cm (see below). Each of the infrared treatment combinations was replicated three times. Control wheat included wheat infested similarly but unexposed to infrared. For each treatment combination, there was a corresponding control, and each control treatment was replicated four times.

Benchtop Infrared Heater. The bench top infrared emitter used in this study was donated by Catalytic Drying Technologies LLC. The infrared emitter elements are housed in a circular stainless steel casing with a surface area of 613.36 cm². Propane (473 ml cylinder; Ozark Trail Propane Fuel, Bentoville, AR) was supplied to the heating element via a hose at a pressure of 28.0 cm of water column. Grain samples for infrared exposure were placed in 27.9 cm internal diameter and 3.8 cm deep stainless steel pan with a 48 cm long handle. Thirteen measurements taken with an infrared thermometer (Bhadriraju Subramanyam, unpublished data), showed the temperature at emitter surface to range from 335 to 474°C, with higher temperatures recorded near the center of the emitter.

Temperature Measurements During Infrared Exposure. The temperature of infested grain during infrared exposures was continuously recorded using a non-contact infrared thermometer (Raytek Ranger® MX4TM, Model 4TP78, Santa Cruz, CA). This thermometer works in the 8 to 14 µm range, and the emissivity was set at 0.95. The infrared thermometer was previously calibrated with a mercury thermometer (Khamis et al., 2009), and found to record temperatures as accurately as a mercury thermometer. The infrared thermometer was connected to a laptop via RS232 cable to acquire "real time" temperature data every second. The infrared thermometer recorded temperature of grain at the center of the pan. The temperature data acquisition program was written by the Electronic Design Laboratory, Kansas State University, in LABView (National Instruments Corporation, Austin, TX).

Assessment of Insect Mortality. After infrared exposure, the grain and any grain debris were returned to the original glass jar, and incubated at 28°C and 65% RH. Adult mortality was assessed 24 h after infrared exposure based on number of dead adults out of the total exposed. Immature stages were reared to adulthood on grain, and their mortality was based on number of adults that failed to emerge out of the total exposed. Similar assessments were made on infested grain that was not exposed to infrared.

Experimental Design and Data Analysis. The experiment was run as a completely randomized design. The time-dependent temperature profile for each replicate was averaged over time to obtain a mean temperature attained by wheat during the exposure period. The mean wheat temperature attained for any given insect age, wheat quantity, and exposure time combination between 8.0 cm and 12.7 cm distance from the emitter surface was compared ($\alpha = 0.05$) using two-sample *t*-tests for equal variances (SAS Institute 2002). Two-sample *t*-tests were used to compare differences in mean temperatures attained between a 45 and 60 sec exposure at any given insect age, grain quantity and distance from heater. Additionally, comparisons were also made of mean temperatures attained between 113.5 and 227.0 g of grain at any given insect age, distance from heater, and exposure time.

In order to determine if the mean grain temperature attained for a given quantity of grain, distance from emitter, and exposure time combination was consistent across the various ages tested (eggs [day 0], 7, 14, 21, 24, and 42 d [2-wk-old adults]), a linear regression of temperature versus insect age was performed and the slope was tested for deviation from zero (SAS Institute 2002).

The main effect of insect age, wheat quantity, distance from emitter, and exposure time and their two-way interactions on the probability of death were determined using logistic regression at $\alpha = 0.05$ (SAS Institute 2002). Odds ratios from logistic regression were used to show differences in susceptibility (odds of dying) of various life stages exposed to infrared. The odds ratio for adults (1) was used as a reference. A ratio >1 showed that a life stage was more susceptible than adults to infrared while a ratio <1 showed that a stage was less susceptible than adults. Differences in susceptibility of various life stages was also determined by plotting probability of death as a function of mean wheat temperatures attained averaged over wheat quantity, distance from heater, and exposure time.

Results

Emergence of *T. castaneum* Adults from Immatures on Untreated (Control) Wheat. On untreated 113.5 and 227.0 g of wheat, 71-72% of the introduced eggs became adults (Table 4:1). All of the individual larvae and pupae added to untreated wheat emerged as adults and the survival of these stages was $\geq 99\%$.

Temperature Profiles During Infrared Exposure. The time-dependent temperature profile, averaged every second from replicated data, was plotted as a function of time for 113.5 and 227.0 g of grain exposed for 45 and 60 sec at 8.0 and 12.7 cm from the infrared emitter. There were eight temperature profiles for each insect age. A comparison of temperature profiles across various ages showed that for any given quantity of grain, distance from emitter, and exposure time, the profiles were essentially similar. Therefore, 2 out of the 12 grapsh were selected to show a typical time-dependent temperature profile (Figure 4:1).

Irrespective of the grain quantity, higher mean grain temperatures were attained when grain was exposed at 8.0 cm from the emitter. Similarly, high grain temperatures were attained after 60 sec compared to a 45 sec exposure. Irrespective of the distance from emitter and exposure time, mean temperatures attained tended to be higher in 113.5 g of wheat than in 227.0 g. The highest mean grain temperature attained (113.1°C) was in 113.5 g of wheat exposed for 60 sec and distance of 8.0 cm from the emitter, and the lowest mean temperature attained (80.9°C) was in 227.0 g of grain exposed for 45 sec at a distance of 12.7 cm from the emitter surface (Table 4:2).

Two sample *t*-tests for each life stage or *T. castaneum* age (0, 7, 14, 21, 24, and 42 d) have shown that the mean temperature attained by 113.5 or 227.0 g of wheat during a 45 or 60 sec exposure was significantly greater at 8.0 cm from the emitter when compared with mean temperature attained by wheat at 12.7 cm from the emitter (t, range among ages, grain quantities, and exposure times = 7.27 – 33.46; df = 4; <math>P < 0.0001). The mean temperature attained by wheat after a 60 sec exposure was significantly and consistently higher than those attained after a 45 sec exposure at a given insect age, grain quantity, and distance from the emitter $(t, range = -3.94 - 15.63; df = ; P \le 0.0170)$ for 20 of 24 comparison. In four comparisons the mean grain temperatures attained between 45 and 60 sec exposures were similar $(t, range = -1.73 - 2.63; df = 4; P \le 0.0580)$. In general, grain quantity did not influence the mean grain temperatures attained for any given insect age, distance from heater, and exposure time. In 17 out of the 24

comparisons, the difference in mean temperature attained by 113.5 and 227.0 g of wheat was not significant (t, range = -1.73 – 2.70; df = 4; P > 0.0520). In 7 other cases, mean temperatures attained by 113.5 g of grain was significantly higher than that attained by 227.0 g of grain (t, range = 3.11 – 16.33; df = 4; $P \le 0.0360$).

The slope of the linear regression between mean temperature attained by 113.5 or 227.0 g of grain at 8.0 or 12.7 cm from the emitter after a 45 or 60 sec exposure and the insect age was not significantly different from zero (t, range among grain quantities, distance from emitter, and exposure times = -1.09 – 1.70; n = 6; $P \ge 0.1637$).

Responses of *T. castaneum* Life Stages to Infrared. The mortality of *T. castaneum* life stages increased with an increase in the mean grain temperature attained (Table 4:2). One hundred percent mortality was achieved for all stages in 113.5 g of grain, exposed for 60 sec at a distance of 8.0 cm from the emitter. At the same distance and exposure time, mortality of *T. castaneum* life stages in 227.0 g of grain was 98 to 100%. Exposure of 227.0 g of grain for 45 sec at a distance of 12.7 cm from the emitter resulted in 78 to 95% mortality. Lower mortalities occurred across life stages in 227.0 g of grain despite observed high grain temperatures. Different grain quantities resulted in different insect mortalities, despite reaching the same temperature, indicating that temperature alone was not a factor determining insect lethality.

Logistic regression analysis showed that the probability of death of *T. castaneum* was influenced significantly by insect age ($\chi^2 = 26.7$; df = 5 P < 0.0001), grain quantity ($\chi^2 = 67.9$; df = 1 P < 0.0001), distance from the emitter ($\chi^2 = 51.3$; df = 1 P < 0.0001), and exposure time ($\chi^2 = 97.7$; df = 1 P < 0.0001). All two-way interactions (insect age x wheat quantity, insect age x distance from heater, insect age x exposure time [df = 5]; wheat quantity x distance from heater, wheat quantity x exposure time, and distance from heater x exposure time [df = 1]) were also highly significant (χ^2 range = 8.3 – 44.3; P < 0.0001).

Both the insect mortality (probability of death data) and odds ratios showed that pupae were the least susceptible stage, followed by eggs, adults, old larvae (21-d-old), mid-to-old larvae (14-d-old), and young larvae (7-d-old). The variation in probability of death of various life stages of *T. castaneum* was evident at mean grain temperatures below 105°C (Figure 4:2). Across grain quantities, exposure times, and distance from emitter, all life stages were killed when the mean grain temperatures attained were between 108 to 111°C.

Discussion

Nearly 71 to 99% of the adults emerged from immature stages on untreated grain. This suggested that the methods employed infesting grain with immatures to gauge effectiveness of infrared radiation was valid.

The fact that the slopes of regressions of mean temperatures attained as a function of insect age for various treatment combinations indicated very little variation in our experimental approach. This was expected because all insect ages were exposed on the same day. Two sample *t*-tests showed that the distance from emitter and exposure time had a greater influence on mean temperatures attained than grain quantity. The mean temperatures attained were inversely related to distance from the emitter, but positively related to exposure time.

In general, at mean grain temperatures above 90°C, fewer *T. castaneum* individuals completed developmental into adults in infrared-treated grain. Although temperatures attained by the two quantities of grain exposed for a specific time were similar, the mortality of *T. castaneum* was different by at least 10%. For example, the mortality of the egg stage in 113.5 g of wheat exposed for 45 sec at a distance of 12.7 cm from the emitter was 89% and the mean temperature attained by the grain was 83°C (Table 4:2). The mean grain temperature was also 83°C when 7-d-old larvae were exposed to infrared under the similar conditions; however, the mortality was 78%. This suggested that temperature alone was not a factor influencing insect susceptibility. Among the life stages, pupae were less susceptible to infrared while young larvae were highly susceptible, and the variation in susceptibility among stages decreased at temperatures above 105°C. Plausible reasons for the observed differences among life stages to infrared warrant further study.

All life stages were killed when mean grain temperatures attained were between 108 to 111°C, irrespective of the grain quantity, distance from emitter, and exposure time. However, exposure of 113.5 g of grain to infrared at a distance of 8.0 cm from the emitter for a duration of 60 sec consistently produced 100% mortality irrespective of insect age. To our knowledge, this is the only paper that systematically shows the effects of infrared radiation on various life stages of *T. castaneum* in stored wheat. One other paper with little or no data on effectiveness of infrared on *T. castaneum* was that of Kirkpatrick and Cagle (1978). In summary, our tests show the bench top flameless infrared emitter to be a viable tool for disinfesting stored wheat containing

various life stages of *T. castaneum*. Large-scale tests need to be conducted to verify our laboratory findings to make this technology commercially viable to grain industry stakeholders.

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Table 4:1. Emergence of *T. castaneum* adults from immature on untreated wheat^a.

Insect age (d)	Mean \pm SE ($n = 4$) number of adults in:				
	113.5 g	227.0 g			
0	35.3 ± 4.9	36.0 ± 5.4			
7	101.0 ± 1.0	99.5 ± 0.5			
14	100.0 ± 0.0	100.0 ± 0.0			
21	100.0 ± 0.0	99.5 ± 0.5			
24	99.3 ± 0.5	100.0 ± 0.0			

^aAdults (100) from cultures were directly added to wheat.

Table 4:2. Emergence of *T. castaneum* adults from infested wheat in various infrared-exposed treatments, mean temperature attained by wheat, and probability of insect death.

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec	Mean temperature (°C)	Mean no. adults	Probability of death
0	113.5	8.0	45	101.3 ± 1.1	0	0.98
			60	107.6 ± 1.2	0	1.00
		12.7	45	83.1 ± 2.0	4.0 ± 1.7	0.89
			60	89.2 ± 0.3	14.7 ± 2.0	0.98
	227.0	8.0	45	97.3 ± 0.9	119.3 ± 8.7	0.87
			60	108.1 ± 0.6	0.3 ± 0.3	0.98
		12.7	45	81.9 ± 1.0	12.7 ± 3.2	0.47
			60	85.9 ± 1.6	3.3 ± 1.3	0.87
	113.5	8.0	45	103.5 ± 0.2	0	1.00
			60	111.4 ± 0.5	0	1.00
		12.7	45	83.7 ± 0.1	0.3 ± 0.3	0.78
			60	87.3 ± 0.6	0	1.00
	227.0	8.0	45	100.1 ± 0.1	0	0.96
			60	108.5 ± 0.8	0	1.00

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec	Mean temperature (°C)	Mean no adults	Probability of death
		12.7	45	83.7 ± 0.6	0.3 ± 0.3	0.78
			60	87.3 ± 0.6	0	0.96
14	113.5	8.0	45	103.2 ± 1.3	0.3 ± 0.3	0.99
			60	110.6 ± 0.6	0	1.00
		12.7	45	85.0 ± 0.5	0.3 ± 0.3	0.95
			60	91.3 ± 0.4	0	0.99
	227.0	8.0	45	101.8 ± 0.6	4.3 ± 1.9	0.95
			60	109.3 ± 0.7	0	0.99
		12.7	45	83.7 ± 1.0	31.7 ± 8.4	0.69
			60	87.8 ± 0.3	65.7 ± 8.1	0.88
21	113.5	8.0	45	103.5 ± 0.3	0	0.99
			60	111.4 ± 0.5	0	1.00
		12.7	45	84.4 ± 0.9	2.3 ± 0.9	0.95
			60	92.0 ± 1.6	1.0 ± 0.6	0.99
	227.0	8.0	45	100.6 ± 0.9	8.7 ± 4.1	0.94
			60	109.8 ± 0.7	0.7 ± 0.3	0.99
		12.7	45	82.7 ± 0.6	66.3 ± 2.3	0.67

Insect age (d)	Grain quantity (g)	Distance from emitter (cm)	Exposure time (sec	Mean temperature (°C)	Mean no adults	Probability of death
			60	87.4 ± 0.3	12.7 ± 1.8	0.94
	113.5	8.0	45	103.3 ± 1.3	0	0.99
			60	108.5 ± 2.7	0	1.00
		12.7	45	80.9 ± 1.7	16.0 ± 6.7	0.90
			60	86.4 ± 1.4	0	0.99
	227.0	8.0	45	103.1 ± 1.4	21.7 ± 11.2	0.89
			60	113.1 ± 0.1	0	0.98
		12.7	45	82.9 ± 0.7	59.3 ± 9.8	0.5
			60	84.8 ± 0.8	13.7 ± 0.8	0.88
42 (adults)	113.5	8.0	45	102.3 ± 0.3	0	0.99
			60	108.0 ± 0.3	0	1.00
		12.7	45	82.6 ± 1.6	14.3 ± 6.4	0.92
			60	87.3 ± 0.9	2.0 ± 1.2	0.99
	227.0	8.0	45	100.0 ± 0.5	6.3 ± 0.9	0.91
			60	106.6 ± 0.9	0	0.99
		12.7	45	82.2 ± 0.3	42.3 ± 2.8	0.56
			60	86.9 ± 0.7	9.0 ± 3.5	0.91

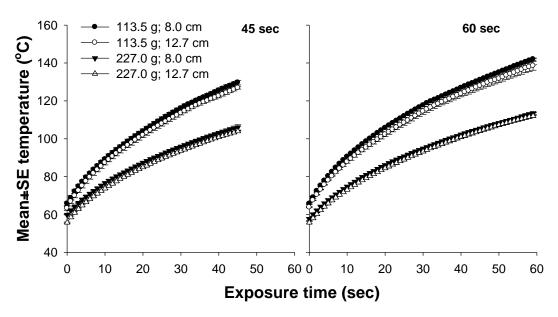


Figure 4:1. A generalized time-dependent temperature profile attained with different quantities of wheat exposed at 8.0 and 12.7 cm from the emitter for 45 or 60 sec.

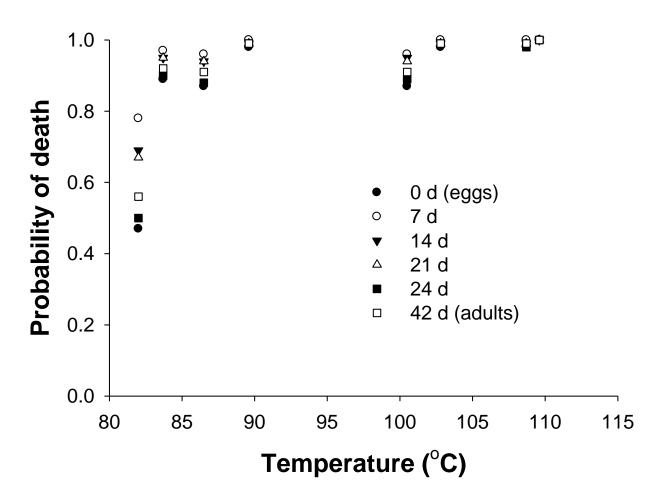


Figure 4:2. Probability of death of different life stages of *S. oryzae* at various mean wheat temperatures attained.

CHAPTER 5 - Effect of flameless catalytic infrared radiation on wheat quality

Abstract

Laboratory tests have shown infrared radiation to be effective against internal and external insects associated with stored grain. However, the impacts of infrared radiation on grain quality are poorly understood. Laboratory tests with a flameless catalytic infrared emitter in infested hard red winter wheat (113.5 or 227.0 g) exposed for 45 or 60 sec at a distance of 8.0 or 12.7 cm showed it to be effective in killing all life stages of two internal and one external insect species, especially at mean grain temperatures of 108° to 111°C. This study investigated the effects of flameless catalytic infrared emitter at temperatures used for disinfesting grain on wheat physical, chemical, rheological, and end-use qualities. The test weight of wheat was unaffected when exposed to infrared radiation. However, a slight drop in kernel moisture increased kernel hardness, but these effects were reversed when the wheat was tempered prior to milling. Flour, shorts, and bran yields, and chemical properties (protein and ash) of these fractions between untreated wheat and infrared-treated wheat showed minor differences that were inconsequential. Mixograph and bake test results did not reveal any consistent adverse effects of infrared radiation on the quality parameters studied. Our result show that infrared treatments that provide effective control of stored-grain insects do not affect the physical, chemical, rheological, and end-use qualities of hard red winter wheat.

Introduction

The United States is third in the world in wheat production behind China and India, and in 2004 produced 63.8 million metric tons of wheat. About 31 million metric tons is exported to 100 countries while about 32.7 million metric tons is consumed domestically (www.uswheat.org). A large volume of wheat is utilized to make breakfast cereals, breads of different types, pasta, and noodles. Wheat is also used as a raw material for the manufacture of starch. Wheat stored on farms and at elevators in Kansas is susceptible to spoilage by insects, molds, and bacteria. Depending on the length of storage, grain moisture, and use and level of pest management, stored wheat is attacked by a variety of insect species (Storey et al. 1984, Reed and Pedersen 1987, Kenkel et al. 1992, Martin et al. 1997). The most common and damaging insect species associated with wheat stored on farms and at elevators in Kansas and neighboring states is the lesser grain borer, Rhyzopertha dominica (F.), rice weevil, Sitophilus oryzae (L.), and red flour beetle, Tribolium castaneum (Herbst) (Reed et al. 1991, 2003). Activity of all three species of insects has also been observed outside farm bins (Dowdy and McGaughey 1994) and at elevators (Dowdy and McGaughey 1997). Both R. dominica and S. oryzae immatures complete development inside kernels of wheat and the immature stages contribute to insect fragments when the wheat is milled.

Grain protectants are routinely used to manage insect populations on the farms while the fumigant phosphine is used on grain after it leaves the farm (Kenkel et al. 1992, Martin et al. 1997). Insects in stored wheat are managed primarily by chemical means and non-chemical methods are underutilized. The use of chemicals, especially traditionally used organophosphate grain protectants such as malathion and chlorpyrifos-methyl have resulted in development of insect resistance (Subramanyam and Hagstrum 2006).

New and innovative technologies need to be constantly explored for effective disinfestations of wheat, because of problems associated with pesticide residues, resistance development in insects, and to meet quality demands (e.g., wheat free of pesticide residues) of domestic and foreign buyers. Furthermore, limited pest management options are available for managing insect pests for stored organic wheat.

Infrared (non-ionizing radiation) radiation is electromagnetic energy with wavelengths (0.075-1000 μ m) longer than visible light (380-750 nm) and shorter than microwaves (0.1-100 cm) (Penner 1998). This energy is transferred to whatever material absorbs it, and the absorbed

energy causes a measurable change in the material's temperature. This radiant "heat energy transfer" depends on how readily the molecularly bonded atoms in the material convert the incident radiant energy into molecular vibrational energy that in turn raises the temperature of the absorbing material and its surroundings. Water readily absorbs mid-infrared radiant energy by the symmetric and asymmetric stretching of molecular bonds between oxygen and hydrogen atoms and by bending of the same bonds (Wehling 1998). The wavelengths most associated with these absorption mechanisms fall between about 2.8 and 7 µm.

The unique nature of absorption of infrared radiation by water has been used for rapid drying of cereal commodities, especially wheat (Bradbury et al. 1960) and rice (Schroeder 1960; Schroder and Rosberg 1960, Faulker and Wratten 1969). In addition to drying commodities, infrared has been used successfully to kill immature insect stages adults of stored-grain insects (Tilton and Schroeder 1963, Cogburn 1967, Cogburn et al. 1971, Kirkpatrick and Tilton 1972, Kirkpatrick et al. 1972, Tilton et al. 1983). In all these tests, infrared radiation sources used natural gas or propane combusted over ceramic panels in the presence of oxygen. These gasfired radiation sources were of high intensity producing 14.067 kW/h (48,000 BTU/h) resulting in temperatures close to 926°C. The infrared radiation wavelength produced was 2.5 µm; small amounts of carbon dioxide and water vapor were also produced. These high intensity infrared emitters had an open flame and may not be suitable for use in dusty grain-handling facilities due to explosion hazards.

Catalytic Industrial Technologies, LLC (Independence, KS) has developed a proprietary flameless infrared technology for various drying applications (http://www.catalyticdrying.com). In flameless infrared emitters, propane or natural gas chemically react with oxygen in the presence of a platinum catalyst delivering radiant energy in the 3 to 7 µm range resulting in temperatures below 500°C. Temperature to start the self-sustaining catalytic reaction is achieved by an electric heating element embedded in the emitter unit. The catalytic infrared heaters are environmentally friendly and since they do not use any flame these heaters do not produce any NOx or CO. The co-products of catalytic oxidation-reduction reaction are infrared radiation, carbon dioxide and water vapor. Pan et al. (2008) reported the catalytic emitter to be effective in drying and disinfesting paddy rice. In the laboratory, we evaluated a bench top catalytic infrared emitter from Catalytic Drying Technologies LLC using 113.5 and 227.0 g of wheat exposed for 45 or 60 sec at a distance of 8.0 and 12.7 cm from the emitter in killing all life stages three insect

species associated with hard red winter wheat. These species included two internal developers, *R. dominica*, *S. oryzae*, and one external developer, *T. castaneum*. The tests showed catalytic infrared radiation to be effective in killing all life stages of the three species, especially when the grain attained temperatures between 108° and 111°C during a 60 sec exposure (Khamis 2009). However, very little information is available in literature on the adverse effects of these short exposures to infrared on wheat physical and chemical properties. Only Kirkpatrick and Cagle (1978) reported a drop of less than 0.5% of moisture of infrared-exposed wheat. The lack of information on the adverse effects of infrared radiation on wheat quality and the positive results obtained in our laboratory prompted us to explore the effects of infrared radiation of wheat physical, chemical, rheological, and end-use qualities.

Materials and Methods

Uninfested organic hard red winter wheat was procured from Heartland Mills in Marienthal, KS. Wheat (113.5 and 227.0 g) was placed in separate 0.45-liter glass jars with wire mesh and filter paper lids. These jars were incubated at 28°C and 65% RH to equilibrate the wheat moisture to 12% (wet basis). A large number of such jars were set up for infrared exposures. Each grain quantity was exposed to a bench top flameless catalytic emitter at 8.0 or 12.7 cm from the emitter for either 45 or 60 sec (see Table 5:1 for treatment combinations). For each treatment combination, a total of 2 kg was accumulated by repeated exposures at the parameters specified. The only two treatment combination that were not used because of poor insect kill from our previous laboratory tests (Khamis 2009) were 113.5 and 227.0 g of wheat exposed at 12.7 cm from the emitter for 45 sec, as these treatments produced less than 90% mortality of *R. dominica*, *S. oryzae*, and *T. castaneum* life stages. Each treatment combination was replicated three times. Six 2 kg lots of wheat unexposed to infrared radiation served as the control treatment.

A 1000 g wheat sample from each treatment combination was used to determine test weight (bulk density) of wheat using the Winchester bushel apparatus (AACC 1991, Method 55-10). Kernel moisture (% wet basis) and kernel hardness (a dimensionless index called hardness index) of infrared-exposed and unexposed wheat before and after tempering to 16% moisture prior to milling were determined using the Perten 4100 Single Kernel Characterization System (SKCS, Perten Instruments North America Inc., Springfield, IL), based on developments by

Martin et al. (1993). The SKCS tests 300 kernels per sample and provides frequency distribution data for kernel hardness and moisture.

Infrared exposed and unexposed wheat were tempered for 16 h. The tempered wheat was weighed and milled in a Buhler mill (Buhler MLU-202, Uzwil, Switzerland) in the Department of Grain Science and Industry, Kansas State University, Manhattan, KS. The gaps in the roller mills were adjusted according to the recommendation of the manufacturer. Before milling, the machine was warmed for 30 min by milling wheat. The feed rate was 130 to 150 g of wheat/min. The break and reduction flours were combined. Shorts were collected and resifted through 132 µm aperture cloth sieve for 2 min to extract any residual flour. The flour extracted from shorts and previous flour fractions were blended for five minutes using a laboratory blender. Total flour yield, bran, shorts and losses were calculated and reported on a percentage basis based on the weight of the original wheat and the weights of each of the three fractions. The ambient temperature and relative humidity during milling were 14°C and 40%, respectively.

The flour particle size distribution of wheat flour particles was determined using a Malvern Mastersizer 2000 (Malvern Instruments Ltd, Worcestershire, UK), which uses light diffraction to determine the size of dry powders. This analysis was conducted by NanoScale Coproration, Manhattan, KS. Two samples of each treatment combination were analyzed to provide information on 10%, 50% and 90% of the particles below a certain diameter.

The Agtron M-45 Color Meter (Agtron Inc, Reno, NV) housed in the Department of Grain science and Industry at Kansas State University was used to determine bran contamination of flour samples. The Agtron M-45 color meter is a direct-reading reflectance spectrophotometer designed to measure the relative spectral qualities of product samples. The meter, set on the green wavelength (546 nm), is calibrated using a 67 calibration tile for the "0% Relative Spectral Reflectance" reading and a 97 calibration tile for the "100% Relative Spectral Reflectance" reading. When using the green wavelength light, relative spectral reflectance is inversely proportional to the degree of bran contamination of flour (Gillis 1963, Shuey 1975). The higher the relative spectral reflectance the brighter (better, low ash) the flour.

The AACC 44-15A air oven method for the bran and shorts, while ash and protein were measured using AACC 08-01 muffle furnace method and AACC 46-30 Leco combustion method, respectively (AACC 1991). The moisture content, protein, and ash of flour were determined using Perten DA 7200 near infrared analyzer (Perten Instruments, Springfield, IL).

Protein and ash contents were reported on 14% moisture basis. A Megazyme total starch enzymatic assay kit (Megazyme International Ireland Ltd., Wicklow, Ireland) was used to measure residual starch in bran (AACC Method 76.13). The flour was subjected to AACC Method 56-81B to determine the falling number (AACC 1991), a measure of enzyme activity or sprout damage to wheat. The AACC Method 54-40A was employed to determine gluten strength or resistance of dough to mixing with pins and flour water absorption using a mixograph. AACC Method 10-10B was used to determine bake water absorption, mixing time, and load volume. The falling number test, mixograph, and bake tests were done at the Wheat Quality Laboratory, Department of Grain Science and Industry, Kansas State University.

Data Analysis. Data on each of the physical, chemical, and rheological properties obtained were subjected to one-way analysis of variance (ANOVA) to determine significant differences among the seven treatments at $\alpha = 0.05$ using the GLM procedure (SAS Institute 2002). Means among treatments were separated using Ryan-Einot-Gabriel-Welsch Multiple Range (REGWQ) test. None of the data was transformed for analysis because Levene's test (SAS Institute 2002) showed variances among treatments to be homogeneous for each of the quality factors. The kernel moisture and kernel hardness frequency distributions before and after tempering were plotted using SigmaPlot software (Scientific Graphing Software, version 11, Jandel Corporation, San Rafael, CA).

Results

The mean test weight of wheat not exposed to infrared radiation was 79.6 kg/hl, and those exposed to infrared ranged from 79.9 to 81.0 and differences in test weight among the treatments were not significant (F = 2.84; df = 6, 14; P = 0.0504). The kernel hardness and moisture before and after tempering among treatments showed some variation that was significant (Table 5:2). The kernel hardness of kernels exposed to infrared were consistently and slightly higher than wheat that was not exposed to infrared radiation, and differences among treatments were significant (F = 3.74; df = 6, 14; P = 0.0197). The kernel moisture in infrared treatments was 0.6 to 1.7% lower than that of untreated wheat (F = 9.52; df = 6, 14; P = 0.0003). Except for treatment C, kernel hardness after tempering also showed differences among treatments (F = 8.14; df = 6, 14; P = 0.0006), but unlike hardness prior to tempering there was no

trend. Tempered wheat among treatments equilibrated to the same moisture content (F = 1.17; df = 6, 14; P = 0.3734).

The effect of infrared radiation on kernel hardness and kernel moisture is obvious by observing the frequency distribution of these parameters (Figures 5:1-5:4). The kernel hardness frequency distributions were less variable than kernel moisture distributions. The kernel moisture was affected to a greater extent when 113.5 g of wheat was exposed to infrared radiation compared with 227.0 g of wheat. However, tempering resulted in both kernel hardness and moisture distributions of infrared-exposed wheat approximating that of untreated wheat.

Both the flour and bran yield were slightly lower than the control treatment (A), but the flour loss was higher with infrared-treated wheat compared with control wheat (Table 5:3). The flour yield, bran yield, and flour loss during mill were all significantly different among the treatments (F, range among quality factors = 2.91 - 3.65; df = 6, 14; $P \le 0.0469$). The flour lost during milling was about 6 to 9%. However, the yield of shorts was not significantly different among the treatments (F = 2.73; df = 6, 14; P = 0.0572).

The flour particle diameters of untreated wheat were slightly lower than particles from infrared-treated wheat (Table 5:4). About 90% of the flour particles in untreated wheat were below 116 μ m, whereas particles from infrared-treated wheat ranged from 120 to 126 μ m. However, these differences were not significant (F = 2.58; df = 6, 14; P = 0.0672). The differences in sizes of 10% and 50% of the particles among treatments was smaller than sizes for 90% of the particles. However, significant differences among treatments in particle sizes at both these percentages was detected (F, range = 3.35 – 4.17; df = 6, 14; $P \le 0.0291$).

The bran contamination of flour, based on Agtron spectrometer was not significantly different among the treatments (F = 2.23; df = 6, 14; P = 0.1013), and the agtron reflectance units among treatments ranged from 64.3 to 67.0.

The moisture content of flour among the treatments ranged from 13.2 to 14.0% (Table 5:5) and these differences were non-significant (F = 2.30; df = 6, 14; P = 0.0940). The ash content values were rounded off to one decimal place in the table, but the actual values among treatments ranged from 0.56 to 0.63%. The protein content among treatments differed by 0.1 to 0.2%. However, the flour ash and protein values were different among the treatments (F, range = 4.24 - 4.32; df = 6, 14; P < 0.0122), and the differences observed did not follow any pattern. The moisture and ash of shorts, and ash of bran were significantly different among the treatments (F,

range = 3.64 - 6.73; df = 6, 14; $P \le 0.0216$). The protein for both shorts and bran, and bran moisture and total starch were not different among the treatments (F, range = 1.40 - 2.71; df = 6, 14; $P \ge 0.0584$). In cases where differences were observed, there were no discernable trends.

The falling number values for untreated wheat flour was 537.3 and similar values for infrared-treated flour was 557.0 to 592.3, and differences among treatments were not observed (F = 0.95; df = 6, 14; P = 0.4927).

Mixograph results show both water absorption and mixing time to be different among the treatments (F, range = 4.53 - 5.83; df = 6, 14; P \leq 0.0032) (Table 5:6). The bake test results (Table 5:7) showed that the baking absorption values, mixing time and loaf volume to vary significantly among the treatments (F, range = 3.13 - 4.53; df = 6, 14; $P \leq$ 0.0369). However, the trends did not show a consistent pattern.

Discussion

The test weight of hard red winter wheat varieties in the United States range from 73.7 to 83.2 kg/hl (Maghirang et al. 2006). The untreated and infrared-treated wheat test weight in our study was 79 to 81 kg/hl. Maghirang et al. (2006) reported that wheat grain test weights greater than 71.7 kg/hl to be correlated with higher flour extraction potential.

The lower kernel hardness values of infrared-exposed wheat relative to untreated wheat may be purely a result of moisture loss upon infrared exposure. The frequency distributions of kernel moisture and hardness support this view. Kirkpatrick and Cagle (1978) reported a drop of 0.5% moisture of 13.5% of wheat exposed to infrared radiation. This is expected, because infrared radiation is used for drying grains (Pan et al. 2008). This drying effect could have reduced both the flour and bran yield and increased flour loss during milling, and may have resulted in the larger particle sizes observed. Flour yield is influenced by many factors such as milling conditions, condition of the machine, and type of wheat (Maghirang et al. 2006). Flour yields among the treatments were within the range stated by the Buhler Mill manufacturer (64 – 70%). However, rehydration during tempering normalized the moisture contents of infrared-treated wheat to that of untreated wheat. Therefore, changes in kernel moisture and hardness upon infrared exposures are reversible at all the treatment combinations. Osborne et al. (1997), Osborne and Andersen (2003), and Ohm et al. (1998) reported that kernel hardness to be directly related to flour yield and quality.

Equilibration of grain moisture contents during tempering perhaps did not result in any bran contamination of the flour. Whole wheat has 9.2-15.8% of protein (Maghirang et al. 2006). A 1% loss of protein through the bran and shorts during milling is expected (Halverson and Zeleny 1988). Desirable flours should have between 11 to 12% protein, and despite minor differences the protein content among the treatments were close to 12%. Shorts are composed of some bran and germ. Ash content is 10 times in bran than in flour. Bakers normally specify flour ash content of 0.48 to 0.52%, and the ash content in our flour averaged around 6%, very close to commercial specifications. Despite differences in some chemical constituents in flour, shorts, and bran, the lack of consistent trends make it difficult to explain the statistical differences observed.

Falling numbers for wheat flour can range from 278 to 861 (Maghirang et al. 2006), and the falling numbers we observed fell within these ranges. The higher falling number value for infrared-treated wheat compared with untreated wheat, despite lack of significant differences, may suggest that exposing wheat to infrared may have inactivated the alpha amylase enzyme. Falling number values below 278 may indicate sprout damage or increased enzymatic activity (Atwell 2001).

In the United States, mixograph is a common method for evaluating physiochemical properties of dough and bread-making potential (Ingelin and Lukow 1999, Maghirang et al. 2006). Desirable flours should have water absorption in the range of 62 to 66%, and the flours from untreated and infrared-treated wheat in our study averaged about 68%. Good flour should have medium to mid-medium mixing time since it has a positive correlation with dough proof height and volume (Maghirang et al. 2006). Flour from infrared-treated wheat and untreated wheat had a moderate mixing time. Moisture absorption during baking was slightly higher than the flour absorption, probably due to inclusion of other additives during baking. However, the flour and bake mixing times were essentially similar. Maghirang et al. (2006) reported baking absorption, mixing time, and loaf volume for hard red winter wheat cultivars to be 58.2 to 66.4%, 2.33 to 6.75 min, and 685 to 1060 cc. The baking absorption values in our study fell outside this range, but the mixing time and loaf volumes were well within ranges specified by Maghirang et al. (2006). Although several physical, chemical, and rheological parameters were statistically different among untreated and infrared-treated wheat samples, the differences observed were not too large to be of any practical concern. Our results suggest that infrared

radiation used for disinfesting stored wheat, under our test conditions, did not adversely affect wheat physical, chemical, rheological, and end-use qualities.

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Table 5:1. Treatment combinations used for exposing organic wheat to flameless catalytic infrared radiation.

Infrared treatment ID	Factors evaluated:				
	Grain quantity	Distance from emitter	Exposure time		
	(g)	(cm)	(sec)		
A (Control)	113.5	0	0		
	227.0	0	0		
В	113.5	8.0	45		
C	227.0	8.0	45		
D	113.5	8.0	60		
Е	227.0	8.0	60		
F	113.5	12.7	60		
G	227.0	12.7	60		

Table 5:2. Changes (mean \pm SE) in kernel hardness and moisture of untreated and infrared-treated wheat before and after tempering.

Treatment	Before tempe	Before tempering		After tempering	
	$\overline{\mathbb{H}^{a,b}}$	Moisture (%) ^b	HI ^{a,b}	Moisture (%) ^{b,c}	
A	77.1 ± 0.6 b	$11.5 \pm 0.1a$	74.9 ± 0.6 bc	15.4 ± 0.0	
В	82.2 ± 1.4 ab	10.4 ± 0.1 bc	$80.4 \pm 0.5a$	15.3 ± 0.2	
C	$79.1 \pm 3.2ab$	10.9 ± 0.1 ab	$72.5 \pm 0.5c$	15.2 ± 0.1	
D	$85.3 \pm 0.3a$	$9.8 \pm 0.1c$	78.7 ± 1.4 ab	14.9 ± 0.4	
E	$85.9 \pm 0.8a$	10.4 ± 0.1 bc	$78.0 \pm 0.7 ab$	15.7 ± 0.1	
F	$82.9 \pm 0.8ab$	10.3 ± 0.0 bc	78.9 ± 1.6 ab	15.4 ± 0.4	
G	$79.7 \pm 2.3ab$	10.2 ± 0.6 bc	$75.2 \pm 0.7 bc$	15.3 ± 0.4	

^aHI, kernel hardness index.

^bMeans within a column followed by different letters are significantly different (P < 0.05, REGWQ test).

 $^{^{\}rm c}$ Means among treatments were not significantly different from one another (P > 0.05, one-way ANOVA).

Table 5:3. Milling yields of various fractions and flour yield loss in various treatment combinations.

Treatment	Mean \pm SE (%) ^a				
	Flour	Bran	Shorts ^b	Loss	
A	$68.3 \pm 0.8a$	$17.5 \pm 0.7a$	8.3 ± 0.3	$6.0\pm0.4b$	
В	$68.0 \pm 0.6 ab$	15.7 ± 1.3 ab	9.0 ± 0.6	$7.3 \pm 0.6ab$	
C	$67.5 \pm 0.7ab$	16.8 ± 0.6 ab	9.3 ± 0.4	$6.4 \pm 0.5b$	
D	$66.8 \pm 0.7 ab$	16.3 ± 0.5 ab	9.3 ± 0.2	7.6 ± 1.0 ab	
E	$65.2 \pm 0.7b$	$16.7 \pm 0.7ab$	9.1 ± 0.3	$9.1 \pm 0.3a$	
F	$67.1 \pm 0.2ab$	$15.2 \pm 0.5 ab$	9.6 ± 0.1	$8.1 \pm 0.3 ab$	
G	$66.3 \pm 0.7a$	$13.3 \pm 1.0b$	10.0 ± 0.3	8.4 ± 0.7 ab	

^aMeans within a column followed by different letters are significantly different (P < 0.05, REGWQ test).

^bMeans among treatments were not significantly different from one another (P > 0.05, one-way ANOVA).

Table 5:4. Particle size distribution of flour from untreated and infrared-treated wheat.

Infrared Treatment ID	Mean ± SE percentage of particles below a certain diamet			
	10 ^a	50 ^a	90 ^b	
A (Control)	$12.6 \pm 0.1b$	49.6 ± 0.5 b	115.8 ± 1.6	
В	13.3 ± 0.1 ab	$53.5 \pm 0.2ab$	120.1 ± 1.2	
C	$13.0 \pm 0.3 ab$	52.2 ± 1.8 ab	120.1 ± 1.6	
D	$13.7 \pm 0.3a$	$56.5 \pm 2.3a$	125.6 ± 3.5	
E	$13.6 \pm 0.1a$	56.5 ± 0.6 ab	122.3 ± 0.0	
F	$13.5 \pm 0.0a$	$54.0 \pm 0.5 ab$	119.8 ± 1.0	
G	$13.5 \pm 0.0a$	$54.0 \pm 0.3ab$	120.5 ± 0.6	

^aMeans within a column followed by different letters are significantly different (P < 0.05, REGWQ test).

 $^{^{\}rm b}$ Means among treatments were not significantly different from one another (P > 0.05, one-way ANOVA).

Table 5:5. Proximate analysis values for flour, shorts, and bran from untreated and infrared-treated wheat.

Fraction	Treatment	Mean \pm SE (%	6) ^a		
	Moisture	Ash Protei	n Starch		
Flour A	14.0 ± 0.3^{b}	$0.6 \pm 0.0a$	$12.0 \pm 0.0a$		
В	13.2 ± 0.2	$0.6 \pm 0.0a$	$11.8 \pm 0.0b$		
C	13.9 ± 0.3	0.6 ± 0.0 ab	$12.0 \pm 0.0 ab$		
D	13.4 ± 0.2	0.6 ± 0.0 ab	$11.8 \pm 0.0 ab$		
Е	13.6 ± 0.3	$0.6 \pm 0.0 b$	$11.8 \pm 0.0a$		
F	13.5 ± 0.2	$0.6 \pm 0.0a$	11.9 ± 0.0 ab		
G	13.4 ± 0.2	0.6 ± 0.0 ab	$11.8 \pm 0.0 ab$		
Shorts A	$11.4 \pm 0.1a$	$2.7 \pm 0.1c$	14.6 ± 0.2^{b}		
В	10.6 ± 0.1 b	$3.1 \pm 0.1a$	15.3 ± 0.0		
C	11.3 ± 0.3 ab	2.7 ± 0.0 bc	15.2 ± 0.5		
D	10.6 ± 0.1 b	2.9 ± 0.1 abc	15.1 ± 0.2		
E	11.2 ± 0.1 ab	$2.7 \pm 0.0c$	14.5 ± 0.1		
F	11.1 ± 0.1 ab	3.0 ± 0.1 abc	14.6 ± 0.1		
G	$10.9 \pm 0.0 ab$	$3.1 \pm 0.0 ab$	15.3 ± 0.0		
Bran A	13.7 ± 0.2^{b}	5.3 ± 0.1 ab	17.1 ± 0.1^{b}	15.2 ± 0.4^{b}	
В	12.8 ± 0.2	$5.3 \pm 0.0a$	16.3 ± 0.1	15.8 ± 0.4	
C	13.3 ± 0.3	5.3 ± 0.0 ab	16.9 ± 0.1	16.7 ± 0.1	
D	12.7 ± 0.2	5.3 ± 0.0 ab	16.6 ± 0.2	16.9 ± 0.3	
Е	13.2 ± 0.1	5.2 ± 0.0 ab	16.4 ± 0.3	15.4 ± 0.6	
13.3 ± 0.5	$5.4 \pm 0.0 ab$	16.6 ± 0.2	15.5 ± 0.8		
G	13.0 ± 0.3	$5.4 \pm 0.0a$	16.5 ± 0.2	14.4 ± 1.0	

 a For each fraction, means within a column followed by different letters are significantly different (P < 0.05, REGWQ test).

 $^{\rm b}$ Means among treatments were not significantly different from one another (P > 0.05, one-way ANOVA).

Table 5:6. Mixograph results of flour from untreated and infrared-treated wheat.

Treatment	$Mean + SE^a$			
	Water absorption (%)	Mixing time (min)		
A	68.7 ± 0.3 ab	3.8 ± 0.0 ab		
В	67.3 ± 0.3 c	3.8 ± 0.2 ab		
C	68.3 ± 0.3 abc	$3.7 \pm 0.1 ab$		
D	67.7 ± 0.3 bc	$3.7 \pm 0.2ab$		
E	$69.0 \pm 0.0a$	$4.5 \pm 0.5a$		
F	$68.0 \pm 0.0 abc$	$3.0 \pm 0.3b$		
G	67.7 ± 0.3 bc	$2.8 \pm 0.0b$		

^aMeans within a column followed by different letters are significantly different (P < 0.05, REGWQ test).

 Table 5:7. Baking test results of flour from untreated and infrared-treated wheat.

Treatment	$Mean \pm SE^a$				
	Water absorption (%)	Mixing time (min)	Loaf volume (cc)		
A	72.7 ± 0.3 ab	3.5 ± 0.0 ab	995.0 ± 17.3a		
В	$71.4 \pm 0.3c$	$3.6 \pm 0.2ab$	$873.7 \pm 22.7ab$		
C	72.4 ± 0.3 abc	3.5 ± 0.1 ab	$959.0 \pm 3.5ab$		
D	71.7 ± 0.3 bc	$4.2 \pm 0.6a$	$822.7 \pm 71.4b$		
E	$73.1 \pm 0.0a$	$4.4 \pm 0.6a$	860.0 ± 50.6 ab		
F	72.1 ± 0.0 abc	3.0 ± 0.3 ab	$954.3 \pm 5.8ab$		
G	71.7 ± 0.3 bc	$2.6 \pm 0.1b$	924.7 ± 11.7 ab		

^aMeans within a column followed by different letters are significantly different (P < 0.05, REGWQ test).

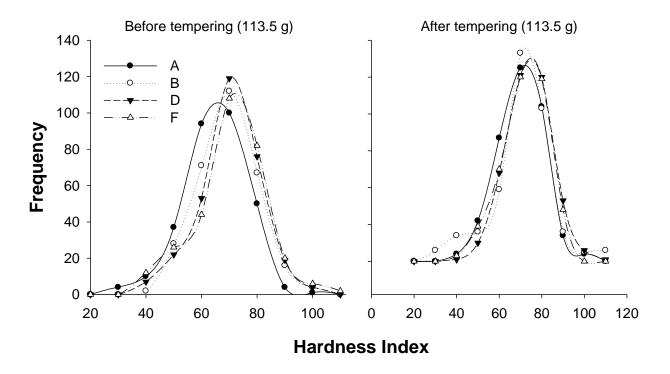


Figure 5:1. Frequency distribution of kernel hardness in 113.5 g of wheat exposed to A, B, D, and F treatment combinations (see Table 5:1 for treatments).

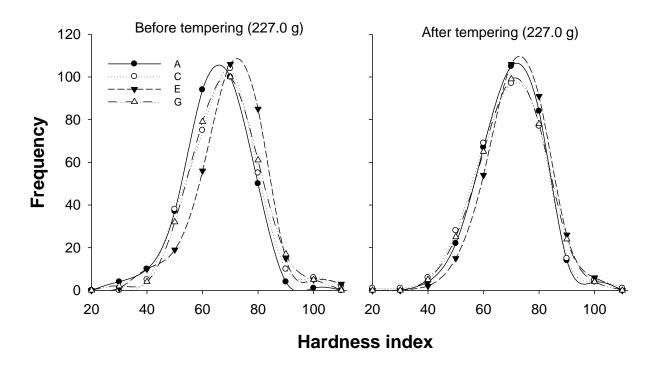


Figure 5:2. Frequency distribution of kernel hardness in 227.0 g of wheat exposed to A, C, E, and G treatment combinations (see Table 5:1 for treatments).

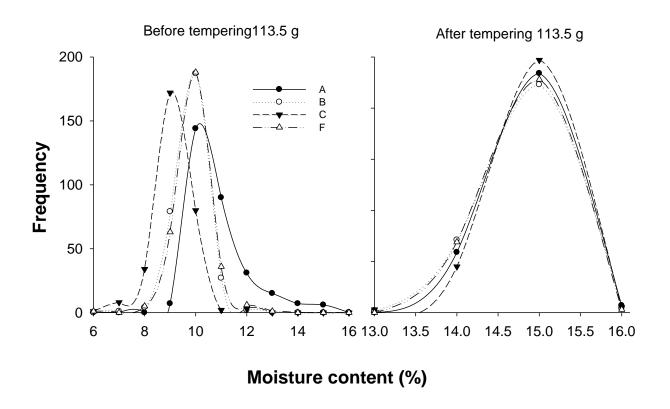


Figure 5:3. Frequency distribution of kernel moisture in 113.5 g of wheat exposed to A, B, C, and F treatment combinations (see Table 5:1 for treatments).

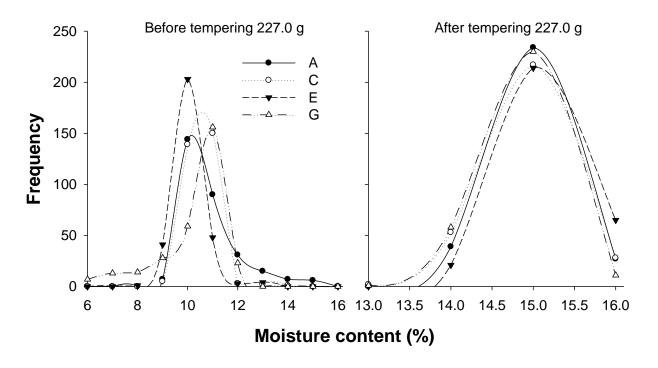


Figure 5:4. Frequency distribution of kernel moisture in 227.0 g of wheat exposed to A, C, E, and G treatment combinations (see Table 5:1 for treatments).