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Thermal death kinetics of adult *Sitophilus oryzae* and effects of heating rate on thermotolerance

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ABSTRACT

Information on thermal death kinetics of targeted stored insects under different heating conditions is essential for developing postharvest disinfestation treatment protocols. Using a heating block system, the thermal death kinetics of adult rice weevil, *Sitophilus oryzae* (L.), were determined at temperatures from 44 to 50 °C at 2 °C intervals and a heating rate of 5 °C/min. The effects of heating rates (0.1, 0.5, 1, 5 and 10 °C/min) on mortality were also examined. The results showed that thermal death curves of *S. oryzae* followed a 0-order kinetic reaction model. The required holding times for achieving 100% mortality were 130, 50, 12, and 4 min at 44, 46, 48, and 50 °C, respectively. The activation energy for killing *S. oryzae* was 505 kJ/mol and the *z* value obtained from the thermal-death-time curve was 3.9 °C. Insect mortality after a 20 min exposure to 46 °C at low heating rates (0.1 or 0.5 °C/min) was significantly lower than that at high heating rates (1–10 °C/min). The information provided by thermal death kinetics for *S. oryzae* is useful in developing effective postharvest thermal treatment protocols.

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1. Introduction

Infestations of grain by various stored product pests may occur at any time from harvest to consumption (Lee et al., 2001). It is estimated that the overall damage caused by such pests is about 10–40% of the annual worldwide production of stored grains (Mishra et al., 2013), with annual losses of about 27% of the total rice yield (Alfonso-Rubí et al., 2003). Among these pests, the rice weevil, *Sitophilus oryzae* (L.), is one of the most destructive and widespread in stored grains and legumes, causing reduction in weight, quality, commercial value, and seed germination, and increased susceptibility to fungal infestation (Jian et al., 2012). The adult female *S. oryzae* (L.) commonly bores a shallow cavity in the kernel in which to deposit eggs, sealing the egg cavity with a gluey secretion (Feng et al., 2004). Although pest management practices often target the most visible adult stage, postharvest treatments are often required to prevent additional product damage during

storage and avoid re-infestation of products before shipment to domestic and international markets.

Traditional chemical fumigations using methyl bromide and phosphine are common to control stored grain pests. Despite its effectiveness, global use of methyl bromide is being phased out due to its listing as an ozone depleter by the Montreal Protocol (USEPA, 2001). In China, populations surveyed over 8 years from 55 locations showed resistance to phosphine increased 10.7% for *S. oryzae* and 71.2% for lesser grain borer (*Rhyzopertha dominica*) (Yan et al., 2004). Therefore, it is urgent to develop an alternative non-chemical method to completely control *S. oryzae* in grain.

As an efficient and safe method, heat treatments have been widely studied to control insect pests in agricultural products (Shellie and Mangan, 1994; Jin, 2011; Purohit et al., 2013). Thermal mortality data of targeted insects have been obtained by directly exposing insects in a water bath (Thomas and Mangan, 1997; Wang et al., 2009), heating insects in glass or metal tubes (Thomas and Shellie, 2000; Lurie et al., 2004) or heating infested fruits in water baths (Hansen et al., 2004). Due to large variations in insect mortality data resulting from these traditional methods, a heating block system (HBS) was developed and used successfully to obtain reliable thermal death kinetic data for several insect pests of fruits and nuts (Wang et al., 2002a,b; Johnson et al., 2004; Armstrong et al., 2009). When compared with the two water immersion

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methods, the HBS yields lower mortality data with less variation, resulting in more conservative treatment recommendations (Wang et al., 2009). Results obtained by the HBS were confirmed for radio frequency heat treatment of walnuts infested with fifth-instar codling moth, *Cydia pomonella* (Wang et al., 2001a) or fifth-instar navel orangeworm, *Amyelois transitella* (Mitcham et al., 2004), and with hot water treatments for cherries infested with third-instar *C. pomonella* (Feng et al., 2004; Hansen et al., 2004).

Thermal death kinetic models derived from thermal mortality data may be useful in predicting the efficacy of different treatment conditions and designing new treatment protocols. A 0.5th-order kinetic model was successfully applied to data from *C. pomonella* (Wang et al., 2002a), Indian meal moth, *Plodia interpunctella* (Johnson et al., 2003), *A. transitella* (Wang et al., 2002b), Mediterranean fruit fly, *Ceratitis capitata* (Gazit et al., 2004; Armstrong et al., 2009), red flour beetle, *Tribolium castaneum* (Johnson et al., 2004), and Mexican fruit fly, *Anastrepha ludens* (Hallman et al., 2005). To develop practical heat treatments for *S. oryzae*, it is desirable to develop a similar model based on HBS data.

Heating rate may have an important effect on the mortality of treated insects (Evans, 1986). Neven (1998) reported that *C. pomonella* larvae may experience thermal conditioning and acclimation at relatively slow heating rates (0.13–0.2 °C/min). Thomas and Shellie (2000) reported that the exposure times needed to achieve 99% mortality of *A. ludens* at 44 °C are 62 and 42 min when heated at 0.175 and 1.4 °C/min, respectively, suggesting that slower heating rates require longer exposures to the target temperature to achieve the same mortality. For conventional heating, heating rates at the center of the product mass may range between 0.05 and 2 °C/min, depending on heating method, product size, and final temperature (Wang et al., 2001b). Target insects may have adequate time to adapt to the heat and increase their thermotolerance (Waddell et al., 2000; Garczynski et al., 2011). As a result, conventional heat treatments typically require long treatment times to achieve adequate mortality levels. Wang et al. (2002a,b) confirmed that heating rates between 5 and 15 °C/min, which correspond to that used in microwave and radio frequency heating, did not increase thermotolerance of fifth-instar *C. pomonella* and *A. transitella*. Therefore, understanding the effect of different heating rates on the mortality of *S. oryzae* would be useful in the development of a treatment protocol.

The HBS system is more difficult to use with internal stages such as *S. oryzae* larvae and pupae. Because removal of internal stages from the seeds causes high mortality, they must be treated within the seed. However, insulation by the seed slows the heating rate and makes it difficult to quantify. The treatment response of internal stages is normally measured by adult emergence, which complicates direct comparisons between stages. The heating rates for treating external, mobile adult stages are easier to quantify, and evaluation is immediate and consistent. For these reasons, we selected the adult stage for this initial study.

Our objectives were to 1) determine the thermal mortality of adult *S. oryzae* at 4 selected temperatures as a function of holding time using the heating block system, 2) develop the thermal death kinetic model of the adult *S. oryzae*, 3) predict the holding time needed to achieve the required mortality at given populations, and 4) explore the effects of heating rates on the thermal mortality of the adult *S. oryzae*.

2. Materials and methods

2.1. Heating block system (HBS)

The HBS was composed of top and bottom aluminum blocks (254 × 254 × 18 mm) which fit together to form the insect

treatment chamber (214 × 214 × 3 mm), heating pads, and a data acquisition/control unit (Fig. 1). Calibrated type-T thermocouple sensors were used to monitor the temperatures of the top and bottom blocks. Heating rates (0.1–15 °C/min) and the set-point temperature were controlled by Visual Basic software via a solid-state relay. Two PID controllers (I32, Omega Engineering, Inc., Stamford, CT) regulated the two block surface temperatures separately. The high thermal capacitance of the blocks provided smooth temperature profiles over the heating and holding periods with temperature deviations from the set point (≤ 60 °C) of less than 0.3 °C (Wang et al., 2002b). More detailed descriptions of the HBS can be found in Gazit et al. (2004) and Johnson et al. (2003).

2.2. Test insects

Adult *S. oryzae* were obtained from the College of Plant Protection, Northwest A&F University, Yangling, China. The adults were kept in a glass jar containing 10 g of wheat grains covered with nylon-mesh-screen (Tilley et al., 2007; Zhao et al., 2007) and were reared under ambient conditions of 27 ± 2 °C, 65% relative humidity and a photoperiod of 14:10 (L:D) h with artificial light.

2.3. Treatment procedures

Based on the thermal-death-time curves for *P. interpunctella* (Johnson et al., 2003) and *T. castaneum* (Johnson et al., 2004), four or five exposure times (1–130 min) at 44, 46, 48 and 50 °C and a heating rate of 5 °C/min were selected to provide a wide range of mortality levels including 100% for adult *S. oryzae*. To determine the effect of heating rate on *S. oryzae* mortality, heating rates of 0.1, 0.5, 1, 5 and 10 °C/min were used. Based on previous test results, a treatment temperature of 46 °C with an exposure of 20 min was selected to provide mortality levels of below 100%. Heating rates ≤ 1 °C/min simulate conventional hot air and hot water heat treatments and fast heating rates ≥ 5 °C/min simulate rapid heating methods using microwave and RF energies.

Fifty actively moving adults were randomly selected for each temperature–time combination test. Because of their small size and speed, *S. oryzae* adults were treated in the heat treatment chamber in a nylon-mesh bag. Since the insect chamber in the HBS was only 3 mm in height, the heat transfer effect of the bag on insect mortality was negligible. For all treatments, the HBS began at a pretreatment temperature of 20 °C. Control insects were placed in the unheated HBS for the longest exposure time at each temperature. Immediately upon completion of the exposure time, the block was opened and the bag was removed from the block. Treated adults were gently brushed into a glass jar containing wheat. The tested *S. oryzae* adults were held for 6 days after treatment for observation. The pests were considered dead if no movement was observed. All trials were repeated three times.

Mean values and standard deviations were calculated from three replicates for each treatment. The mean values for insect mortality under different heating rates were separated at $P = 0.05$ level using least significant difference (LSD) *t*-test.

2.4. Insect thermal kinetic modeling

The thermal death kinetic response of heat treated insects can be described by a temperature–time model, using mean survival ratios as a function of exposure time for each temperature. The model has been previously used for third-instar *C. capitata* (Gazit et al., 2004; Hallman et al., 2005) and fifth-instar *C. pomonella* (Wang et al., 2002b). The general equation for the kinetic model is as follows:

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