In this study, 4800 adults of rusty grain beetle, *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Laemophloeidae), were introduced in a 4.7 m diameter and 8 m high hopper-bottom bin holding 48 t of wheat at 9.6±0.3 % moisture content (wet basis) and at 22 to 27ºC. Twenty Insector® (pitfall traps that electrically count captured insects and estimate the insect densities) were installed inside the bin at four layers. The grain was fumigated by adding dry ice after six week storage. Carbon dioxide (CO₂) concentrations at these Insectors® locations were measured every week during the grain storage period and every day during fumigation period. Insect activity at each Insector® location was monitored and their densities were measured by the Insector® system. It was found that adults moved down in the first week after they were introduced. Their distribution followed the temperature gradient with most adults being captured from the warmer layer. Descendants of the introduced adults followed the temperature gradients except in the top layer which had warmer grain. Adults did not produce enough CO₂ to be detected by the CO₂ analyzer. Adults were captured in 84 h after high concentration CO₂ were introduced into the bin.

**Key words:** Insect activity, insect density, fumigation, Insector®, *Cryptolestes ferrugineus*

INTRODUCTION

Spatial distribution is one of the most important ecological properties of a species. There has been some effort to describe the spatial distribution of *Cryptolestes ferrugineus* (Stephens) in stored grain; however, the published studies are based on either small sample units of grain (<2.75 kg) or small containers (less than 1.5 t grain). Their distribution inside the grain was also studied in a short time period (say less than one month). Therefore, the distribution of the descendants of the introduced insects inside stored-grain bins is not known.

Fumigation by CO₂ is one of the methods of disinestation (Mann et al., 1997). Controlled laboratory experiments have shown that a lethal environment for the rusty grain beetle can be achieved by elevating the CO₂ concentration. To create an environment lethal to insects, >40% CO₂ concentrations must be maintained for at least 4 d (White et al., 1990; 1993). This requirement poses a problem because most stored bins used in the world are not
airtight and CO₂ can leak out even in a well-sealed bin (Mann et al., 1997). Therefore, monitoring insect activity during fumigation might be an effective way to verify whether the pests are eradicated.

Traps to detect insects in stored grain are effective and sensitive tools for grain management. Sampling rusty grain beetles using modified pitfall traps is well documented (Loschiavo, 1974; Toews and Phillips, 2002). A version of an electronic pit-fall trap, produced and known as the Insector® (OPI Systems Inc. Calgary, Canada), is commercially available. The Insector® system also identifies species to groups and measures insect densities of the captured insects.

The aim of this study was to monitor insect activity and distribution of the introduced adults and their descendants inside a farm-sized bin during storage and fumigation periods by using the Insector® system.

**MATERIALS AND METHODS**

Experiment was conducted in a metal hopper bin (4.7 m diameter, 8 m high, and 4.0 m cylinder part) filled with about 48 t wheat (hard red spring No 1) (Fig. 1). There were 20 Insectors® and 20 CO₂ tubes installed inside the bin (Fig. 1). One extra CO₂ tube was located inside the head space of the bin. The wheat had less than 0.7±0.4% dockage which included 0.6±0.1% smaller than wheat determined by sieving using 1.651 mm opening sieve. Test weight of the wheat was 846.7±3.3 kg m⁻³. The moisture content of the wheat was 9.8±0.1%. The hopper bin was located inside a building and the room temperature was 25±5°C. To prevent the insects from climbing out of the bin, the vent, loading hole, and the aeration duct were taped during experimental period. The manhole was closed except during the sampling period.

*Cryptolestes ferrugineus* were reared at 30±5°C and 75±5% r.h. on cracked wheat plus wheat germ (95:5 wt/wt), and were held in the dark during rearing and experiments. Eggs of *C. ferrugineus* hatch in 4–5 d and the complete life cycle takes about 3 weeks at optimum conditions of 35°C and 70% r.h. (Smith, 1965). *Cryptolestes ferrugineus* have an average life span of 6–9 mo. Adults of mixed sex were 1 d to 2 mo at the start of the experiment. Before introducing beetles into the grain bin, 4800 adults were selected using a gentle vacuum and were kept inside four 4-L glass bottles with about 3 kg of wheat in each bottle. The bottles were kept inside the bin for at least 24 h to allow insects to acclimate to the experimental conditions.

To warm up the grain (using the warm room air), grain was aerated for about 6 d and temperature was recorded by the Insector® system. After the grain reached the room temperature (25±5°C), the aeration fan was stopped and air at each Insector® location was pulled out through a CO₂ detector (Carbon Dioxide Analyzer, Model: series 9519, Alpha Omega Instruments, Rhode Island, USA) and the CO₂ concentration was measured. After this measurement, 4800 adults of *C. ferrugineus* were introduced at the top of the grain and half radii of the bin. The CO₂ concentrations at each Insector® location were measured every three or four days during the following 6 wk (the storage period).
After this storage period, grain was fumigated by introducing CO$_2$ into the bin (Fig. 1) using the method of Mann et al. (1997). To watch the response of insects to low CO$_2$ concentration, CO$_2$ concentration inside the bin during the first fumigation period was less than 5%. During the second fumigation period, CO$_2$ concentration was higher than 52.9% at any location. During fumigation period, CO$_2$ concentration was measured every day.

Insect daily density at each Insector® location (four layers and five locations at each layer, Fig. 1) was monitored by the Insector® system. The Insector® system classified the captured insects into six groups. During testing, the density of insects (RGB group in Insector®) was measured and interpreted as the density of rusty grain beetle because there was no other insect species. These densities at each location were used to analyze the adult activity and distribution in the following time periods: the first week, four weeks (the second, third, and fourth weeks), descendant distribution (the fifth and six weeks), during the first fumigation period (the seventh and eighth weeks), and during the second fumigation period (the ninth and tenth weeks).
RESULTS AND DISCUSSIONS

Grain temperature
The average temperature of the grain during the entire experimental period was 25.1±4.6°C. In any layer, there was no difference in the temperature at any location. Grain temperature slightly changed during the entire experimental period (Fig. 2). This change was caused by the temperature gradient in the vertical direction inside the building. The temperature at the upper-layers was always higher than that at the lower-layers (Fig. 1 and 2). There was about 1.0°C/m temperature gradient in the vertical direction inside the bin and there was no gradient in the horizontal direction (Tukey test for each layer and all the P>0.5). Temperature gradient at the beginning of the experiment between top and the third layer was about 3.0°C/m and this gradient was gradually decreased (Fig. 2).

![Fig. 2- Grain temperature at different layers of the bin.](image)

Adult distribution in the first week
Adults were captured inside the entire bin in less than 24 h after they were introduced (Fig. 3). This indicated that the adults could move down more than 4 m in less than 1 d. In about 1wk of the adult introduction, adults moved down because the bottom layer had highest insect density (Fig. 3). This indicated that lots of adults did not stay at the warm top layers. The downward movement might be caused by the drift effect (Jian et al., 2009), and the crowding effect at the introduction location. This study found the same trend during that time period as has been reported in the literature that more than 70% of adults are found in the bottom half of small columns after they were introduced (Jian et al., 2009).
Fig. 3- Management action and adult density at each Insector® location.
Adult distribution in four weeks

Introduced adults gradually moved up because the top layer had the highest insect density in between 1 and 4 wk. Their distribution followed temperature distribution because the top grain was warmer than that in any other layer (Fig. 2 and 3). This result was consistent with the fact that adults move to higher temperature regions inside a grain bulk in response to temperature gradients (Jian et al., 2009).

Descendant distribution

Descendants of the introduced insects were mainly found inside the third layer (Fig. 3). The insect density inside this layer during this time period was significantly higher than that inside any other layer (Tukey test and all $P<0.0001$). This result indicated that the distribution of the descendant adults followed temperature gradient except the top layer, which was the warmer layer, and did not move down. The descendants might also not move very often or very far because adult densities at other locations were significantly low (Fig. 3). This result indicated that the descendant adults might have different movement behavior than that of the introduced adults. If they did not move often or far away from the egg laying location, their distribution might relate to the egg laying behavior of the introduced adults.

Adult distribution in horizontal direction

There was no significant difference in insect densities at each half-radius locations in all 4 layers (Tukey test and all $P>0.05$). However, insect density at the center location was higher than that at other locations (Fig. 4) with more than 37% chance. The grain was loaded without using spreader and the dockage at the center location was about 8 times higher than at other locations. This high dockage at the center location might explain the higher insect densities at the center locations inside any layer (Jian et al, 2009).

Adult activity during the first fumigation period

Before grain was fumigated, CO$_2$ concentration was less than 0.05% which was the same as that without insect infestation. This indicated that adults of *C. ferrugineus* could not produce enough CO$_2$ to be detected by using the CO$_2$ Analyzer. During the first fumigation period, the CO$_2$ concentration was less than 5% at any location and there was no difference between different locations. At the end of the first fumigation, CO$_2$ concentration was less than 0.05% at any location. During this fumigation period, the measured insect densities at each location were significantly lower than that without fumigation (Fig. 3, Tukey test and all $P<0.0001$). However, some adults were still moving because there were some captured adults during this time period (White et al., 1993).

Adult activity during the second fumigation period

After 44 h of the CO$_2$ introduction, the CO$_2$ concentration reached 73.4% at the center of the second layer. This was the highest concentration measured and achieved at the end of the CO$_2$ introduction. At the end of the CO$_2$ introduction, the mean of the CO$_2$ concentration was 60.6±2.3%. The lowest CO$_2$ concentration was 52.9%, which was at the center bottom. Two weeks later, CO$_2$ concentration decreased to 34.2±2.3%. During the second fumigation period, some adults were captured in the first 84 h. This indicated that not all adults were killed in 84 h. There were no captured adults after 4 d of the CO$_2$ introduction.
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