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Detection techniques for stored-product insects in grain

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Abstract

Cereal grains are the major source of food for humans and most domesticated animals. In many developing countries, overall postharvest losses of cereals and legumes of about 10–15% are fairly common. Consumption of cereals and legumes by pests such as insects during storage and microbial spoilage or contamination may make these totally inedible. On farms, manual samples, traps, and probes have been used to determine the presence of insects. Manual inspection, sieving, cracking-floatation and Berlese funnels are being used at present to detect insects in grain handling facilities. These methods are not efficient and are time consuming. Acoustic detection, carbon dioxide measurement, uric acid measurement, near-infrared spectroscopy, and soft X-ray method have the potential for use at the industry level to detect insects in grain samples as their usefulness has been demonstrated in the research laboratories. Researchers have developed image analysis programs to automatically scan X-ray images to detect insect infestations. The use of near-infrared (NIR) spectroscopy has been investigated to detect hidden insects in wheat kernels. X-ray and NIR spectroscopy methods are cost prohibitive and current NIR instrumentation requires complex operating procedures and calibrations. The advantages and limitations of these insect detection methods are evaluated and the advantages of one technique over the other are described in this paper. © 2005 Elsevier Ltd. All rights reserved.

Keywords: Insect detection; Insect trap; Pheromones; Acoustical method; Electrical conductance; Berlese funnel; NIR spectroscopy; Machine vision; X-ray imaging

1. Introduction

Cereal grains, oilseeds and legumes (hereinafter referred to as grains) are the major source of food for humans and most domesticated animals in the world. Annual world production of main cereal grains in millions of metric tones (Mt) is: wheat—554; rice—530; maize—508; barley—162; and sorghum—58 (CGC, 1998). Annual world production of the major oilseed crops in millions of metric tonnes is: soybeans—115; cottonseed—33; rapeseed (canola)—27; sunflower seed—23; and groundnut—18 (CGC, 1998). Worldwide production of main legumes is 55 Mt (FAO, 2000). Grain production in any country varies from year to year hence grain should be stored strategically from years of over-production for use in years of under-production. Also, grains must be stored for several other reasons such as point of production is not the point of consumption and time of production is not the time of consumption. Stored grain can have losses in both quantity and quality. Losses occur when the grain is attacked by insects, mites, rodents, birds, and microorganisms.

Insect infestations in grain cause quantity and quality losses and lower crop values. Insects not only consume grain but also contaminate it with their metabolic byproducts and body parts. Insects produce heat and moisture due to their metabolic activities that can lead to growth of microflora and the development of hotspots in grain. Heavily infested grains are unfit for seed purposes and its products are unsuited for human consumption. In many developing countries, overall post-harvest losses of grains of about 10–15% are fairly common (Lucia & Assennato, 1994). Infestation of grains by pests such as insects during

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storage may make grains totally inedible through associated microbial spoilage and contamination. Taking the commercial section of grain storage, where large volumes of grain are held for long periods, the potential for loss is extremely high and is directly summable in financial terms. The total economic losses in Canada due to stored-product pests and microorganisms in grains and oilseeds can be in the millions of dollars annually (White, 1993).

The standards of quality for grains have been established in the majority of countries to satisfy customers, among whom the awareness for clean grain and its products are increasing. With consumers demanding that food be of the highest possible quality, the contaminants such as insects, rodent droppings, and ergot (a toxic fungal body) in post-harvest grains must be minimized. There is an increasing trend among grain buyers towards zero-tolerance to these contaminants. Countries such as Canada have a legally defined zero tolerance for stored-grain insects (Canada Grain Act, 1975).

Grain destined for domestic and export markets is inspected to take preventative actions to reduce quality and quantity losses that might occur during storage and transport. Stored grain is vulnerable to both external and internal damage by insects, but internal infestations are the most difficult to detect (Pederson, 1992). Detection and removal of internal insects from grains are important control measures for ensuring storage longevity, seed quality and food safety. Inspecting for insect-damaged kernels is labour intensive and many infested kernels may be undetected where an immature insect has not emerged from the kernel. Grain inspectors at milling facilities need to know the quantity of hidden insect infestation so that loads with excessive infestations can be cleaned or diverted for other uses.

Several methods have been developed to detect hidden insects in whole kernels. Infestation of grains may be detected by staining of kernels to identify entrance holes for eggs, floatation, radiographic techniques, acoustic techniques, uric-acid measurement, nuclear magnetic resonance imaging and immunoassays (Pederson, 1992). The Berlese funnel method is currently used in Canada to detect infestations of live insects in incoming and export grain at terminal elevators (CIGI, 1993). Acoustic monitoring and near-infrared reflectance (NIR) spectroscopy methods have been used to detect infestations of the lesser grain borer, Rhyzopertha dominica (F.) in wheat. All these methods show varying degrees of efficiency in detecting different stages of insects. Some techniques are time consuming, require trained personnel and are difficult to implement in real time. Most of these techniques have not been found feasible to be implemented in the grain inspection system because of their cost, unreliability, and the varying degrees of success obtained in detecting infestations. Most of the countries in the world use probes and traps as a common detection technique for stored-product insects in grain. Canada uses Berlese funnel method while United States uses visual reference images along with the probes for insect detection in stored grain.

The objectives of this work were

- (1) to integrate information related to various types of insect detection techniques in stored grains;
- (2) to discuss the working principle of detection methods; and
- (3) to discuss the advantages of one method over the other.

2. Insect detection techniques

2.1. Grain probes and insect traps

Insect populations in stored grain are often monitored using traps. An alternative is to take grain samples with a grain trier, deep bin cup, or vacuum probe (Hagstrum, Flinn, Subramanyam, Keever, & Cuperus, 1990) which are sifted on screens. A brass probe trap developed by Loschiavo and Atkinson (1973) for detecting insects in stored grain has been redesigned several times using newer and less expensive plastic materials (White et al., 1990). Pitfall and probe traps are commercially available traps (Treite Inc., Salinds, CA, USA) that are used for detecting adult insects in stored grain (White et al., 1990). Probe traps are cylindrical tubes with perforations in the upper section through which insects drop into the trap and are unable to escape because of the shape of the receptacle. These traps have a pointed tip for easy insertion into the grain. Traps must be removed from the grain bin and inspected periodically to determine the number and type of insects that have been captured.

For studies on insect ecology and evaluation of the effectiveness of pest management, estimation of insect densities is generally required. Probe traps are used for finding insect density. White and Loschiavo (1988) developed a stacked version of the probe trap for investigating insect activity at different grain depths. Shuman, Coeffelt, and Weaver (1996) developed instrumentation for counting probe trap catches electronically and they are now commercially available (Stormax Insector, OPI systems, Calgary, AB, Canada). Probe traps detect insects when no insects are detected by standard grain sampling methods such as grain trier method (Barak & Harein, 1982) because they remain in the grain for long periods.

This method of detecting insects is labour intensive, limits the temporal availability of data, and restricts placement of the probe traps in easily accessible locations. Interpretation of trap catch is difficult because many factors influence trap catch. The catch increases proportionally with an increase in trapping duration (Fargo, Epperley, Cuperus, Clary, & Noyes, 1989). Capture rate is influenced by insect species and grain temperature and type of grain (Wright & Mills, 1984).

2.2. Pheromones

The most common use of aggregation or sex pheromones is in traps to monitor insect populations. For use in monitoring, chemical attractants usually are impregnated or encased in a rubber or plastic lure that slowly releases the active components over a period of several days or weeks. Traps containing these lures are constructed of paper, plastic, or other materials. Most traps use an adhesive-coated surface or a funnel shaped entrance to capture the target insect. Traps for some pests are coated with an adhesive that also contains the chemical attractant.

Stored-product insects can be detected with a variety of traps, some using food attractants or synthetic insect pheromones (Vick et al., 1990). Three stored-product insects that commonly occur together, and for which pheromones are available, are the lesser grain borer, *R. dominica* (Williams, Silverstein, Burkholder, & Khorramshahi, 1981) and the red flour beetle *Tribolium castaneum* (Herbst) (Suzuki & Mori, 1983), which use an aggregation pheromone, and the warehouse beetle, *Trogoderma variabile* (Ballion) (Cross et al., 1976), which uses a sex pheromone.

Although pheromone traps give an indication of pest density, several factors make the interpretation of density estimates complex and difficult. Environmental factors affect trap catches. Temperature, rainfall, and wind speed and direction influence attractant release from lures and insect flight (Fields, Van Loon, Dolinski, Harris, & Bunkholder, 1992). Loschiavo et al. (1986) determined that many insects fly and respond to semiochemicals only at certain time (dawn, midday, dusk, or night), and temperatures (10– 15 °C).

2.3. Visual lures

Visual lures used in insect management fall into the following categories: lights (incandescent, fluorescent, and ultraviolet) that attract insects from dark or dimly lit surroundings (e.g. in warehouses, mills, and elevators); coloured objects that are attractive because of their specific reflectance and shapes or silhouettes that stand out against a contrasting background.

A great number of insect species are attracted to light of various wavelengths. Although different species respond uniquely to specific portions of the visible and invisible spectrum, most traps or other devices that rely on light to attract insects use fluorescent bulbs or bulbs that emit ultraviolet wavelengths. Hundreds of species of moths, beetles, flies, and other insects, are attracted to artificial light. They may fly to lights throughout the night or only during certain hours.

Insect electrocutors can be effective in certain indoor situations, especially in food warehouses and processing plants. Electrocutors are placed in dimly lit areas where their light is not visible from outdoors. In such locations the trap does not lure insects into the building, yet it does attract and kill certain flies, moths, and beetles that are pests of stored products or nuisances in food production areas (Gilbert, 1984). Light traps have been used for several decades to monitor the presence of insects and to determine seasonal patterns of pest density. But because pheromone traps are much more specific (they catch only one or a few pest species instead of many) and more convenient, light traps are no longer as widely used.

2.4. Acoustical methods

Acoustical detection methods use insect-feeding sounds to automatically monitor both internal and external grain feeding insects. Insects hidden inside kernels of grain can be detected acoustically by amplification and filtering of their movement and feeding sounds. Hagstrum, Webb, and Vick (1988) demonstrated that sounds of R. dominica larvae can be used to estimate larval population densities without removing grain samples.

Hagstrum, Flinn, and Shuman (1996) detected the presence of one infested kernel in a 650 g of grain. The effective use of an acoustic method to detect insects in grain requires a quantitative understanding of several physical and biological factors that affect sound production, insect distribution, and detection. The physical factors include the intensity, duration and spectral characteristic of the sound at the source, the distance to the receiver, the receiver's spectral sensitivity (Beranek, 1988) and the background noise (Mankin, Shuman, & Coffelt, 1996). Biological factors include unfavourable environment, insect behaviour and insect inactivity (Mankin et al., 1996). One of the disadvantages with acoustic methods is that they cannot detect dead insects in grain and infestation by early larval stages of insects.

2.5. Electrical conductance

Pearson, Brabec, and Schwartz (2003) detected hidden internal insect infestations in wheat kernels using electrical conductance. Their studies showed that the identification accuracies for all wheat samples were 88% for large sized larvae, and 87% for pupae, and there was no sound kernel misclassified as infested.

A single kernel characterization system is commonly used to measure grain kernel weight, moisture content, diameter, and hardness. This system works on the principle of electrical conductance and compression force. The kernel acts as one resistor in a two-resistor and voltage-divider circuit of the single kernel characterization system. Conductance is monitored by measuring the voltage across the kernel. A low voltage measurement corresponds to low kernel resistance, which is typical of high moisture-content kernels. If a live insect is present inside a kernel, there is likely to be a large downward slope in the conductance signal. Based on the signal characteristics of the system and by computing the range of voltage levels in the conductance signal, infested kernels are differentiated from sound kernels.

Though this method is inexpensive, inspecting single grain kernels is time consuming. Infested kernels with insect eggs and young larvae may be undetected because of the low moisture content. This method cannot detect kernels with dead internal insects. The insect detection rates by this method are very low when compared with the inspection by soft X-rays (Pearson et al., 2003). The sample input is another problem as this method applies to single kernels only.

2.6. Berlese funnel method

The Berlese funnel method is commonly used in terminal elevators in Canada to detect infestations due to live insects in grain. A Berlese funnel works on the principle that insects move away from heat. The Berlese funnels are 49–79% efficient in recovering free-living adults of *Cryptolestes ferrugineus* (Stephens) in wheat samples (Smith, 1977). This method is often slow and inaccurate in detecting infestations. It takes 5–6h to determine the presence of insects in 1 kg grain samples and during this time, the grain would have been loaded into bins or ships. The performance of a Berlese funnel depends on insect stage, size of grain sample, and moisture content of grain (Smith, 1977). Furthermore, this method cannot be used for hidden infestation in grain kernels.

2.7. Near-infrared reflectance (NIR) spectroscopy

The NIR spectroscopy has evolved as a fast, reliable, accurate and economical technique available for compositional analysis of grains (Kim, Phyu, Kim, & Lee, 2003). This technique can be used for both qualitative and quantitative analysis. The NIR technique provides information based on the reflectance properties of different substances present in a product. The NIR is based on the absorption of electromagnetic wavelengths in the range 780-2500 nm. The concentrations of constituents such as water, protein, fat, and carbohydrate can be determined using classical absorption spectroscopy. Elizabeth, Dowell, Baker, and Throne (2002) determined that a NIR system is the best method to detect single kernels of wheat that contained live or dead internal rice weevils at various life stages. They also classified sound kernels and kernels containing live pupae, large larvae, medium-sized larvae, and small larvae with an accuracy of 94%, 92%, 84% and 62%, respectively.

The NIR method has been used to identify several Coleopteran species (Dowell, Throne, Wang, & Baker, 1999), to detect parasitized weevils in wheat kernels (Baker, Dowell, & Throne, 1999), and to detect external and internal insect infestation in wheat (Dowell, Throne, & Baker, 1998; Ghaedian & Wehling, 1997; Ridgway & Chambers, 1996). The NIR system used to detect insects in kernels can scan 1000 kernels per second (Dowell et al., 1999). Perez-Mendoza, Throne, Dowell, and Baker (2003) compared a NIR system and the standard floatation method for detecting insect fragments in wheat flour. They determined that the standard floatation method is time consuming (about 2 h/ sample) and expensive. In contrast, a NIR system is rapid (<1 min/sample), does not require sample preparation and could easily be automated for a more sophisticated sampling protocol for large flour bulks.

The NIR method cannot detect low levels of infestations in bulk samples, or differentiate between live and dead insects (Dowell et al., 1999). The NIR method is very sensitive to moisture content in samples and the instrument requires frequent calibration (McClure, 1987). Ridgway and Chambers (1998) captured NIR images of infested wheat samples by using a NIR vidicon camera. They compared the NIR images with the image of the sample acquired using X-rays and concluded that NIR images can detect the insects in the grain sample by the changes in kernel composition as a result of infestation. The NIR imaging method is an indirect method and cannot detect larvae because of the movement of larvae in the cavity due to heat generated from the lighting which obscured the details of NIR images.

The chemical information of the food materials is obscured by changes in the spectra caused by physical properties such as the particle size of powders. This means that NIR spectroscopy becomes a secondary method requiring calibration against a reference method for the constituent of interest. As a consequence of the physics of diffuse transmittance and reflectance and the complexity of the spectra, calibration is normally carried out using multivariate mathematics.

2.8. Machine vision

In the United States, visual reference images and interpretive line prints are used as the inspection system for insect infestation and grain grading (Federal Grain Inspection Service, 1997). In this system individual grain kernels are compared with the photographic print of the representative sample. The disadvantage with this technique is that this method is subjective, time consuming and internal infestations cannot be identified.

Computerized image analysis has been shown to have great potential for detecting and identifying various nongrain particles and insects in wheat. A machine vision system for detecting insects in grains consists of a high speed integrated machine vision software package used with a monochrome CCD (charge coupled device) camera and a personal computer. Zayas and Flinn (1998) detected *R. dominica* adults in wheat bulks with higher than 90% accuracy using structural and colour information. (Ridgway, Davies, Chambers, Mason, & Bateman, 2002) developed a rapid machine vision method for the detection of adult beetles and determined that detection rates were 89% for commercial samples containing several insect species.

A major obstacle to the development of commercially useful machine vision systems for insect detection in grain is the limited rate of sample throughput. Simple, fast and reliable algorithms are needed to allow a statistically significant portion of a grain sample to be inspected in the short time available. Furthermore, device complexity, particularly in terms of camera type, computer specification and sample delivery system, must be minimized to make this approach cost effective. Another disadvantage is that it can

161

Table 1
Advantages and disadvantages of various detection techniques for stored-product insects in grain

Insect detection methods	Pros	Cons
Grain probes and insect traps	Widely used, inexpensive, used for finding insect density	Labour intensive, limits the temporal availability of data, cannot detect internal insects, restriction in the placement of traps
Pheromones	Gives an indication of pest density	Environmental factors affects trap catches
Visual lures	Can be effective in indoor situations	Not very effective
Acoustical methods	Internal infestation can be detected	Cannot detect dead insects and infestations by early larval stages
Electrical conductance	Hidden internal infestation can be identified	Kernels with insect eggs and young larvae cannot be detected, efficiency is low compared to soft X-rays
Berlese funnel method	Cheap and commonly used method at elevators	Very slow and internal infestations cannot be identified
Near-infrared spectroscopy (NIR)	Rapid method, no sample preparation	Cannot detect low levels of infestation, sensitive to moisture content in samples, calibration of equipment is complex and frequent
Machine vision	Effective in detecting external insects	Cannot detect internal insects
X-ray imaging	Non-destructive, highly accurate, detect both internal and external insects, able to detect both live and dead insects inside grain kernels	Cannot detect insect eggs

detect only external insects in the grain bulks whereas NIR spectroscopy and X-ray methods can detect internal insects.

2.9. X-ray imaging

Soft X-ray is the only non-destructive, direct method that can detect insect infestations in grain kernels (Haff & Slaughter, 1999; Karunakaran, Jayas, & White, 2003; Milner, Lee, & Katz, 1952). Karunakaran et al. (2003) correctly identified wheat kernels infested by *Sitophilus oryzae* (L) larvae and pupae-adults with more than 97% accuracy from the soft X-ray images. They also identified sound kernels with 99% accuracy and also indicated that in the future an automated line-scan X-ray system could inspect 1 kg grain in about 15 min compared to 5–6 h using a Berlese funnel.

A soft X-ray system consisting of a fluoroscope operated at 15 kV potential and $65 \mu A$, produces real-time images (Karunakaran et al., 2003). Single kernels were placed on plastic wrap on a platform between the X-ray tube and detection system and were scanned. Images formed on the detection screen were captured by a CCD black and white camera and digitized by a digital video creator. The digital images were processed to detect insect-infested kernels.

Of all the available methods to detect insects in grains, soft X-ray imaging is the only non-destructive and timesaving technique. The soft X-ray technique can be effectively used to identify mechanical damage in grains destined for seed purposes. Small larvae and weevil egg plugs are difficult to distinguish from denser portions of the grain by using the soft X-ray method (Karunakaran et al., 2003). Table 1 shows the advantages and disadvantages of various detection techniques for stored-product insects in grain.

3. Conclusion

Insect populations in grains can be generally monitored by several methods. Probe traps can be effective but it is difficult to interpret the type and size of insect catch. Pheromone traps are affected by several factors like wind speed, temperature and direction. Acoustical methods can detect only live insects while electrical conductance can detect internal insects in grains. The NIR spectroscopy method is very sensitive to moisture content in samples and the instrument requires frequent calibration. Machine vision systems cannot detect internal infestations. The soft X-ray method is the only non-destructive and direct method to detect insect infestations both by internal and external grain feeding insects. Thus the soft X-ray is the method which appears to have the greatest potential among the insect detection techniques.

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