Overview of North American stored product research

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Abstract

Major locations for stored product research in North America are in Winnipeg, Manitoba, Canada, and Manhattan, Kansas, USA. Recent personnel changes and research areas are reviewed. One of the pressing research areas in the U.S. is reducing the need for fumigations in flour mills and evaluating alternative treatments. Long-term studies are beginning to show efficacy of better IPM practices, including use of aerosol treatments, for reducing the need for fumigation. Heat treatment as an alternative to fumigation continues to be refined through research. Models have been developed for optimizing heat treatments and fumigations. Studies at grain elevators are optimizing insect management at these large storage facilities, including better sampling methods and computer programs that aid in decision making. Recent studies are beginning to look at insect populations in feed mills and their association with microbes. A number of studies have investigated the biology and control of psocids, which are emerging pests of stored products in the U.S. There have been major research efforts in both Canada and the U.S. to develop better sampling and detection methods, including thermal imaging, automated digital x-rays, molecular methods, and development of better attractants and improved interpretation of trap catches. The red flour beetle, *Tribolium castaneum*, is the first agricultural pest to have its genome sequenced, and mining the genome has produced vast knowledge of various physiological processes that might be exploited for control of this and other pests. Expected future research trends are on aerosol treatments of structural facilities, improved methods for detecting internal insect pests, psocid biology, improved attractants, improved interpretation of trap catches for making pest management decisions, and application of genomic technologies for insect control.

1. Locations for stored product research

The main center for stored product research in Canada is in Winnipeg, Manitoba. Entomologists Paul Fields and Noel White are at the Agriculture and Agri-Food Canada Cereal Research Centre, and biosystems engineers Digvir Jayas and Jitendra Paliwal are at the University of Manitoba. Most of the storage research is at the Canadian Wheat Board Centre for Grain Storage Research on the University of Manitoba campus.

The main center for stored product research in the United States is in Manhattan, Kansas. The Stored Product Insect Research Unit (previously the Biological Research Unit) at the Center for Grain and Animal Health Research (previously the Grain Marketing and Production Research Center), which is part of the United States Department of Agriculture's Agricultural Research Service (USDA-ARS), has seven scientists working on basic and applied aspects of stored-product entomology. These are Frank Arthur, Dick Beeman, Jim Campbell, Paul Flinn, Jeff Lord, Brenda Oppert, and Jim Throne. During the past four years, Tom Phillips moved from Oklahoma State University, where he had faculty responsibility for stored-product entomology, to be Head of the Department of Entomology at Kansas State University, which has a long history of training students in stored-product entomology; George Opit moved from a postdoctoral position at the USDA-ARS Stored Product Insect Research Unit to take Tom Phillips' position at Oklahoma State University; and Dirk Maier moved from Purdue University to be Head of the Department of Grain Science and Industry at Kansas State University. Bhadriraju Subramanyan continues his work on stored-product entomology in the Department of Grain Science and Industry at Kansas State University. Other North American university faculty with major assignments in stored products are Linda Mason at the Purdue University Department of Entomology, Charles Woloshuk at the Purdue University Department of Botany and Plant Pathology, and Carol Jones at the Oklahoma State University Department of Biosystems Engineering. A number of other state, federal, and university researchers in North America have partial appointments in stored products.

2. Research highlights and trends in North America during the last four years

2.1. Reduced use of methyl bromide for structural fumigations

Use of methyl bromide for structural fumigations has decreased steadily in North America since its phase-out was proposed. Most facilities have reduced fumigations from three calendar-based fumigations per year to one or fewer per year. This has been achieved through better pest management practices, such as improved monitoring and sanitation, and through use of aerosols or spot treatments with heat or insecticides. Heat is also used for insect control in some facilities, instead of fumigation. Sulfuryl fluoride is also being used for fumigation of some facilities, rather than using methyl bromide.

Efforts have been made to model structural fumigations to try to improve efficacy. Chayaprasert et al. (2008, 2009) developed a model for structural fumigations, and they used it to investigate fumigation leakage rates from flour mills under different weather conditions. They showed that leakage rates varied within the same facility at different times of the year, indicating that leakage rate should be calculated from sealing factors for the particular facility and using historical local weather conditions for the actual fumigation period. Their results showed that leakage rates for methyl bromide and sulfuryl fluoride did not differ when simulations were run under the same weather conditions for a facility, when the fumigants were applied at the same dosages. Cryer (2008) also found that predicted leakage rates for the two fumigants were similar.

Although aerosol insecticide applications are being used more frequently in flour mills and other structures, there are few recent reports on their efficacy. Toews et al. (2006b) monitored insect populations in an operating flour mill during a 22-month period. They found that insects were almost always captured inside the mill in the first trapping period after fumigation or aerosol application with dichlorvos+pyrethrin, but it wasn't clear if the insects survived treatment or immigrated from outside. Arthur and Campbell (2008) showed that survival of the confused flour beetle, *Tribolium confusum* (Jacquelin du Val) (Coleoptera: Tenebrionidae), increased when food was provided after treatment with a pyrethrin-CO2 aerosol, emphasizing the importance of sanitation in facility pest management programs. This aerosol treatment seemed to have no long-term effect on resident insect populations in the study warehouse. Arthur (2008) investigated efficacy of synergized pyrethrin and methoprene aerosols for control of the red flour beetle, *Tribolium castaneum* (Herbst), and *T. confusum* in a commercial food storage facility, and in this study found no effect of presence of food on efficacy of the synergized pyrethrin aerosol. *Tribolium confusum* adults were less susceptible to the synergized pyrethrin aerosol than *T. castaneum* adults. Few treated larvae or pupae of either species survived to the adult stage. Few larvae of either species exposed to the insect growth regulator methoprene survived to become adults, and only 13% of Indianmeal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), larvae embedded in food media and exposed to methoprene survived to the adult stage. Overall, results of aerosol studies show good efficacy of aerosols and good promise for the use of aerosols to reduce the need for structural fumigations.

Much research has been conducted on efficacy of heat for controlling insects in structures, but recent efforts have begun to synthesize these data to provide tools to users for optimizing heat treatments. Boina et al. (2008) developed a model for predicting survival of *T. confusum* during heat treatments. The model was validated with independent data from nine heat treatments in structures, and the model predicted the observed mortality well.

2.2. Insect management at grain elevators

Efforts continue to improve insect management at large, central grain storages. Arthur et al. (2006) sampled grain residues at nine elevators over a two-year period and found that 80% of the insects in grain residues were in the genera Sitophilus and Cryptolestes. Numbers of Cryptolestes in samples increased rapidly in spring, and then remained fairly constant. Numbers of Sitophilus in samples increased through warm months, and then decreased. Pest insects were present in 42% of samples, while beneficial insects were found in 5% of samples. The parasitoid *Anisopteromalus calandrae* (Howard) (Hymenoptera: Pteromalidae) comprised 90% of beneficial insects found. Pest insects were most prevalent in samples taken from elevator boot pits and tunnels. The large numbers of insects in these samples indicate the need for good sanitation around grain storages.

Flinn et al. (2007) extended their previous expert system to aid in pest management decision making for farm-stored grain to be applicable to elevator storages. Users can input current grain temperatures and moistures and numbers of insects from grain samples, and the expert system uses built-in models to make predictions about future insect populations and to recommend management actions. The validity of the expert system was tested in a four-year project at commercial grain elevators. The program failed to identify unsafe storage conditions in only 2 of 399 bins in Kansas, and it correctly identified unsafe storage conditions in all 114 bins in Oklahoma. The number of bins fumigated was reduced because only bins with higher densities of insects were fumigated, rather than treating all bins on a calendar basis. The expert system is being used by a scouting company that was developed as a result of this project. That company contracts with elevators to sample their grain and provide management recommendations.

Insect populations in bins were determined during the elevator study (Flinn et al., 2010). In these bins that were 30-35 meters tall, the lesser grain borer, *Rhyzopertha dominica* F. (Coleoptera: Bostrichidae), *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Laemophloeidae), and *T. castaneum* made up 44, 36, and 19% of the insects in the top 3.7 meters of grain and 84, 8, and 8% from 3.8 to 12.2 meters, respectively. Few insects were found below 12 meters, so this suggested that sampling the center of the top 12 meters of grain provided the best estimate of insect populations in the bins. Sampling was done with a vacuum sampler. *Cryptolestes ferrugineus* was prevalent near the start of the storage period in June, *C. ferrugineus* and *R. dominica* were found in approximately equal numbers in autumn, and then *R. dominica* was prevalent in winter. Infestation in the bins began at the top and moved down throughout the storage season.

2.3. Insect sampling and detection

The Insector® electronic probe trap is a commercial system for monitoring insects in stored grain by automatically counting the insects falling into traps and identifying the insects falling into the traps to species based on size. Flinn et al. (2009) tested the system in bins of wheat in Kansas to determine accuracy of counts and species identification. They developed models that could be used to convert trap catch to density (insects/kg of grain), and they adapted the Stored Grain Advisor expert system to input trap catches and make management recommendations. Management decisions made using the expert system were similar to those made based on grain samples. However, the accuracy of the species identifications depended on the species involved. The identifications were accurate for C. ferrugineus, but the system could not accurately differentiate *R. dominica* and *T. castaneum* because of their similarity in size. In the same study, Opit et al. (2009a) found that the Insector® system worked well for electronically counting the psocids *Liposcelis decolor* (Pearman) (Psocoptera: Liposcelididae) and *L. entomophila* (Enderlein), although the system can't differentiate psocid species.

Manickavasagan et al. (2006) tested the use of thermal imaging of a small (1.5-m diameter by 1.5-m high) experimental bin of barley to detect hot spots, but found that the method was not accurate. Manickavasagan et al. (2008) tested the use of thermal imaging to detect all stages *C. ferrugineus* inside wheat kernels, and found around 80% accuracy in determining whether or not a kernel was infested.

There have been expanded efforts to use molecular methods to detect insects in samples of grain. Atui et al. (2007) tested the myosin ELISA method originally developed by Quinn et al. (1992) because there were concerns by industry that the myosin breaks down over time if the insects are dead, resulting in lower predictions of insect density than actually present. They found that the myosin level was reduced 58% in the first two weeks after R. dominica died, but that there was no further degradation after that time. Balasubramanian et al. (2007) showed that DNA fingerprinting could be used to detect fragments of *T. castaneum* or *T. confusum* in flour at concentrations of 1% (2 mg of insect parts in 200 mg flour), but not at lower concentrations.

Pearson and Brabec (2007) developed an electronically conductive roller mill that measures electrical conductance as a grain sample is crushed, and can detect fourth instar and older *R. dominica* or the rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), in the grain with 83% accuracy based on detection of water in the insects. The accuracy of the method is similar to that for other rapid detection methods, but the method has the advantage that it can process one kilogram of wheat in one minute. Toews et al. (2006a) investigated the use of computed tomography (multiple x-rays taken across a sample) to detect S. oryzae pupae in wheat kernels, and they found 87-94% accuracy of the method for detecting infested kernels, depending on infestation level. Given cost and processing time involved to detect this relatively large insect stage, the method does not look promising for use by industry at this point. Singh et al. (2009) used near-infrared (NIR) hyperspectral imaging for the detection of wheat kernels damaged by *C. ferrugineus*, *R. dominica*, *S. oryzae*, and *T. castaneum*, and found 85-100% accuracy of the method. Narvankar et al. (2009) used x-rays and automated analysis software to detect infection of wheat kernels by the storage fungi Aspergillus niger, *A. glaucus* group, and *Penicillium* spp., and they were able to differentiate healthy and infested kernels with 93-99% accuracy. Haff and Pearson (2007) used x-rays and automated analysis software to detect granary weevil, *Sitophilus granarius* (L.), larvae in wheat kernels with about 85% accuracy, which was similar to accuracy obtained when the x-rays were examined by lab personnel.

2.4. Insects in feed mills

Little work has been done on insects in feed mills, but insects can be a major problem in feed mills and can potentially transmit diseases in the feed. Larson et al. (2008b) surveyed eight feed mills for a year in the midwestern U.S., and they found that the largest number of insects (38,933) was found in the mill interior, followed by the mill exterior (3776), the load-out area (1068), and the receiving area (620). In the mill, 49% of the insects were *T. castaneum*; and 90% of the insects in the mill exterior, 67% in load-out area, and 38% in the receiving area were the warehouse beetle, *Trogoderma variabile* (Ballion) (Coleoptera: Dermestidae). Species presence depended on the crop type processed and was influenced by the degree of pest management and sanitation practiced. Also, lures for *T. castaneum*, *T. variabile*, and the cigarette beetle, *Lasioderma serricorne* F. (Coleoptera: Anobiidae), were placed in the traps, which presumably influenced the species captured.

Larson et al. (2008a) surveyed enterococci bacteria associated with stored-product insects in feed mills in the U.S. These bacteria are usually not a direct disease threat to humans, but they are considered as reservoirs of antibiotic resistance. A number of enterococci bacteria were found associated with the beetles from the feed mills, and intermediate to complete resistance to the antibiotics chloramphenicol, ciprofloxacin, erythromycin, neomycin, streptomycin, tetracycline, and vancomycin was found.

2.4. Psocids as emerging pests of stored products

Psocids became pests of increasing concern in Australia and China in the 1990's, but were not of concern in the U.S. until the 2000's. In a two-year study of psocids in bins of wheat stored in Kansas, Opit et al. (2009b) found only *L. entomophila* in 2005 and only *L. decolor* in 2006. Psocid density increased quickly after the grain was put into storage in July, and peaked in October before dropping to almost zero in December as temperatures decreased. But, some live psocids were found throughout the winter. Cardboard refuges placed on the surface of the grain or on the hatch and Insector® probe traps were good ways to sample psocids (Opit et al., 2009a). Placing cardboard refuges on the hatch of the bin is probably the simplest and least expensive way to sample psocids, and catches correlated well with numbers of psocids found in grain samples. The numbers of psocids caught using the various methods varied greatly; for example, in 2005 there were 77,502 psocids in the Insector® probe traps, 33,615 in surface refuges, 3395 in hatch refuges, and 547 in trier samples.

The biology of many psocid species has been poorly studied, so there have been efforts to expand our knowledge of biology of psocid species found in the U.S. Opit and Throne (2008b) characterized the population growth and development of the psocid *Lepinotus reticulatus* Enderlein (Psocoptera: Trogiidae) at constant temperatures. *Lepinotus reticulatus* did not survive at constant relative humidities of 32 to 55%, but populations increased at 75% relative humidity at 22.5 to 32.5°C. Populations declined at 35°C. Population growth of the psocid *Liposcelis brunnea* Motschulsky was similar to that for *L. reticulatus*, except that *L. brunnea* populations could develop from 55 to 75% relative humidity and developed more rapidly at 65% than 75% relative humidity (Opit and Throne 2009).

Opit and Throne (2008a) compared suitability of six grains for development of *L. reticulatus* and *L. entomophila*. Numbers of *L. reticulatus* produced were greatest on oats (175 progeny produced by five females in 32 days) followed by rice (134), barley (120), milo (115), wheat (115), and maize (18). Much larger numbers of *L. entomophila* were produced: wheat (502), barley (490), milo (275), rice (172), oats (154), and maize (146). This may help explain the relative prevalence of *L. entomophila* worldwide. In a similar study, Athanassiou et al. (2010) showed that milo was the most suitable crop for development of Liposcelis bostrychophila Badonnel, *L. decolor*, *L. paeta* Pearman, and *L. entomophila*, followed by wheat and rice. Maize was the least suitable for all species. They also showed that all four species could develop on whole grain, although progeny production was maximized when some cracked grain was present. In addition, they showed that progeny production was greater on durum wheat than on hard red winter, hard red spring, soft white winter, soft white spring, soft club, soft red winter, or hard white wheats.

Most control technologies for stored-product insects have been developed for beetles, and psocids don't always respond the same as beetles to these control technologies. Recent studies have investigated control technologies that are in common use in the U.S. for control of psocids. Guedes et al. (2008b) showed that *L. entomophila* and *L. reticulatus* are susceptible to heat, although it took about twice as long to kill *L. entomophila*. They investigated the production of heat-inducible proteins when these two species of psocids were exposed to high temperatures, and they found heat-inducible proteins only in *L. entomophila*. This may help explain the more widespread distribution of this species worldwide because these heat-inducible proteins may have a protective function when the insect is stressed.

Athanassiou et al. (2009a) tested all insecticides registered for stored wheat, rice, and maize in the U.S. for effectiveness against psocids, and found that the organophosphate class of insecticides was most effective for psocid control. Effectiveness of methoprene, spinosad, and pyrethrin varied with the grain on which they were tested and with psocid species and stage (egg, nymph, or adult). Efficacy of three diatomaceous earth formulations for control of psocids varied greatly, with adult mortality sometimes exceeding 90%, but progeny production was never reduced more than 39% indicating that diatomaceous earth alone would not be suitable for psocid control (Athanassiou et al. 2009b). Guedes et al. (2008a) tested surface insecticides that are used for structural treatments for control of psocids, and they found that the insecticides ß-cyfluthrin and chlorfenapyr, but not pyrethrins, were effective for control of *L. bostrychophila* and *L. entomophila*. *Liposcelis bostrychophila* was slightly more tolerant than *L. entomophila*, and behavioral studies showed that this may have been due to less movement and that *L. bostrychophila* tended to keep their abdomen raised which may have resulted in less contact with the insecticide. Interestingly, psocids were able to move at a rate of 0.5 cm per second, which is quite rapid for an insect that is only about 1 mm in length.

2.5. Attractants and trap interpretation

Monitoring insects is an important component of an integrated pest management (IPM) program, and this is routinely practiced as part of structural IPM. But much work remains to be done in this area, including improvement and development of attractants and better interpretation of trap catches for pest management decision making. Edde and Phillips (2006a) investigated the impact of male age on rate of pheromone emission in *R. dominica*, which produce an aggregation pheromone, and found that emission was about twice as high in younger males (12-weeks old) than older ones. Edde and Phillips (2006b) investigated the response of *R. dominica* adults to volatiles from different plant materials and found that response was greater to volatiles from wheat than from acorns, cowpeas, peanuts, or dried potato tubers. Males feeding on wheat also produced greater quantities of pheromone (Edde et al., 2007).

Mahroof and Phillips (2007) found that volatiles from *Capsicum* spp. were most attractive to *L. serricorne*, among a number of plant volatiles tested, and mated females exhibited the highest response to these compounds. Mahroof and Phillips (2008) found that *L. serricorne* were equally attracted to three commercial pheromone lures, but more beetles were captured when volatiles from *Capsicum* spp. were included with the lures.

Nansen et al. (2006) investigated ways to improve implementation of traps and interpretation of trap catches of *P. interpunctella*. They investigated response of males in a very simple environment where there was no food or females present except when used as lures. They found that male catches were similar when using concentrations from 1 to 2,000 µg of pheromone, captures of males decreased with distance from the release point, and the greatest numbers of insects were caught within three meters of the release site.

2.6. Genomics

Tribolium castaneum was the first agricultural pest to have its genome sequenced (*Tribolium Sequencing* Consortium, 2008). RNAi has been a useful tool in *T. castaneum* for knocking out genes to determine their functions. Much of the postgenomic work has concentrated on genes involved in cuticle breakdown and synthesis during molting because this is a very vulnerable stage for insects, making it a target for new insect control tools. A number of chitinase genes have been found with varying functions, and these are often specialized to act in a certain part of the body at a certain molt (e.g., Zhu et al., 2008). Another potentially vulnerable process in stored-product insects is water regulation because these insects live in a relatively dry environment, and a number of genes involved in osmoregulation have been identified (Park and Beeman, 2008).

3. Future trends in research

The number of stored product researchers in North America seems to have stabilized, after a decade of reductions. Recently, positions have been filled as people leave them.

Use of aerosol treatments in structural facilities continues to increase, and further studies will be required as new aerosolized insecticides and new application technologies are introduced.

Studies on insect detection technologies will continue because raw grain handlers and processors are very interested in improved detection technologies. Ideally, they would like detection systems that can detect all stages of internal insects automatically, accurately, and quickly, such as processing a 1-kg sample of grain in one minute.

Studies on psocid biology will continue, as there are many species of psocids about which we know little, and the biology of the different species appears to vary greatly. Industry personnel have expressed interest in psocid attractants to monitor and possibly mass trap psocids, and almost nothing is known about psocid attractants.

There is still a need for improved attractants for the major pests of food processing and warehouse facilities, and for better interpretation of monitoring data to aid in pest management decision making.

Although we have learned much about the genes involved in many physiological processes of *T. castaneum*, there is a need to take this technology to the next step of applying genomics for insect control.

✧Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

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