Christos G. Athanassiou
Frank H. Arthur *Editors*

Recent Advances in Stored Product Protection

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Preface

"Recent Advances in Stored Product Protection" was based on discussions that initiated in 2012, on whether there is a need for one more book in durable food protection. Despite the fact that there are numerous developments during the last years toward the introduction of new techniques in stored product protection, these developments are not summarized as a whole, and there are "advances" that have not reached the wide audiences or the research communities associated with stored products. As unique, man-made ecosystems, the various postharvest environments provide unique characteristics that for the most part do not exist in field crop or orchard pest control systems. These characteristics are closely tied with the existence of partially or fully closed environments, such as bulk storages, food storage and production facilities, and even retail environments where processed grain products are stored and sold to consumers. In this regard, several major novel techniques can be used only in storage and processing facilities, warehouses, and silos, and not before or after those stages.

Innovations in stored product entomology do not only refer to nonchemical control but also to chemicals that are an essential part of pest management. In an effort to include wider subjects, many different aspects are analyzed here regarding pest management on bulk grains and in processing and storage facilities, but also to other stored products such as dried fruits, nut products, and spices, i.e., "high value commodities". At the same time, chemical control is presented along with the phenomenon of resistance and resistance management, which should be an essential part of integrated pest management in stored product protection. Biological control is also addressed, by discussing both "microbials" and "macrobials". Recent advances, to a large extent, include control associated with emerging pests in stored product protection, such as invasive species, and also highlight the renewed interest for the importance of stored product arthropods as public health pests. In this sense, pests of museums and related facilities can be also considered as "storage" pests, and their control is largely based on the same techniques that are applied to agricultural commodities. One additional key element in recent advances is the economics of stored products and stored product protection, which is also discussed in detail in the last chapter.

Our sincere thanks to all contributors for their time and effort that made this book possible, their willingness to share their knowledge and their data that are presented in the individual chapters, and their support in innumerable ways during the preparation of this book. Finally, we would like to express our gratitude to our families for their continuous support during this adventure in science.

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Chapter 1
Importance of Stored Product Insects

Manoj K. Nayak and Gregory J. Daglish

Introduction

With the continuous development of human civilization over the last 10,000 years or so, methods to produce food and other materials and to store them for future use have developed enormously. Moreover, significant population growth over the last few centuries aligned with increasing life expectancy and industrialisation has led to the gradual loss of arable land to make way for housing, industries and transportation network. To feed the world's bulging population, food security has become one of the most important priorities for both the developed as well as developing nations across the globe. The concept of food security emerged only in the 1970–90s due to the deepening of global food crisis, specifically affecting the poor in the under developed world. At the World Food Summit of 1996, issues of famine, hunger and food crisis were extensively examined and the behaviour of potentially vulnerable and affected people was identified as a critical aspect, based on which the initial focus, was on the volume and stability of food supplies. Food security was defined in the 1996 World Food Summit (FAO 1996) as: 'availability at all times of adequate world food supplies of basic foodstuffs to sustain a steady expansion of food consumption and to offset fluctuations in production and prices'. The leaders attending the conference also solemnly proclaimed that 'every man, woman and child has the inalienable right to be free from hunger and malnutrition in order to develop their physical and mental faculties'. The universal understanding now is that, apart from targeting to produce more food, we must protect what we produce.

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Grain has been a major source of food for humans since the beginning of agriculture and settled communities. Archaeological evidences suggest that grain was grown and stored in bulk at least 7000 years ago, much before the great civilizations of the Orient and Mesopotamia (Roberts 1976). Ancient Egyptian records indicated storage of grains in pits lined with straw during 2000 B.C. (Lee 1960), and archaeological remnants in India showed communal granaries comprised of mud-brick houses in 2500 B.C. (Mellart 1961). Effects of colonisation, international trade and industrialisation have resulted in many advances in storage structures since these descriptions from the ancient history. Although the old mud-brick structures are still in practice in several parts of Africa, now we have large structures of steel and concrete for storing commodities in most of the developed countries (Reed 1992). Several commodities are being stored, that may include but not limited to durable food and materials for livelihood such as grain (cereals, pulses, oilseed and nuts), dried tubers, dried fruits, herbs and spices; dried fish and meat products; museum and herbarium artefacts and hides skins and wool.

The storage environment, with rich sources of food as described above, is a very attractive place for a range of insects to thrive, and show preference for the stored commodities over their previous natural habitats. The threat to biosecurity of stored food, specifically the grain and its products from insect infestations has been a well-established phenomenon. A recent forecast estimated that the food production would need to increase by 60% to feed an estimated global population of 10.5 billion in 2050 (Alexandratos and Bruinsma 2012). To meet this demand and to ensure future global food security, apart from increasing production and improving distribution; a major focus should be on reducing post-harvest food losses.

A significant proportion of the post-harvest losses occur due to infestations from insect pests. Of the 32 taxonomic Orders of insects, species belonging to only three Orders, Coleoptera (beetles), Lepidoptera (moths) and Psocoptera (psocids) are considered as pests of stored commodities. In addition, a few species belonging to the Orders Hemiptera (bugs) and Hymenoptera (wasps) are also being reported to be associated with stored commodities, but only as predators or parasites of the species belonging to the three major orders mentioned here (Rees 2004; Heaps 2006) Most of the stored products pests are considered as opportunists; several beetles (Coleoptera) were initially recorded under the bark of trees; several moths (Lepidoptera) were supposedly originated from dead and ripening fruits; whereas several psocids (Psocoptera) were originated from leaf litters (Rees 2004). A rare exception is the granary weevil *Sitophilus granarius* (L.) (Coleoptera: Curculionidae), which is the only species that has never been detected outside the storage environment (Plarre 2010). The oldest record of storage pests associated with human beings goes back to ancient Egypt, where *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae), *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae) and *S. granarius* (Coleoptera: Curculionidae) were reported (Rees 2004).

In the recent past, there have been comprehensive books and chapters written on the classification, identification and general biology of major pests of stored commodities and their management (e.g. Rees 2004; Hagstrum et al. 2012). The aim of this chapter is to highlight the importance of the storage pests affecting the durable commodities. Apart from a brief listing of major stored product pests, we aim to, broadly, discuss their economic impact. Unless there is an absolute need, our focus will be on published reports from the last decade.

Overview of Insect Pest Species Associated with Stored Products

Stored product pests can be defined as organisms that are injurious to stored commodities of all types including grains, pulses, fruits, seeds and plant and animal materials (Hill 2002). The impact can be in the simplest form of physical damage to the commodity and can extend to a broader sense of economic loss in terms of quality and market access. Due to its global importance as a major stored commodity, grain will constitute a significant part of the discussion in this chapter. Rees (2004) has given a comprehensive account of all stored product pests along with their host range, distribution and life cycles. We provide an overview of insects in stored products under the following six categories: pests of grain and their products; pest of dried fruits and nuts; pests of durable herbs, spices, tobacco and pet food products; pests of dried fish and other animal products; predators and parasitoids; and scavengers and foragers.

Pests of Grain and Grain Products

Pests of grain and grain products constitute the major group of insects occurring in the stored product environment. They are further categorised into primary and secondary pests. Primary pests attack whole grain and are capable of penetrating an undamaged seed coat and pod to feed on the embryo, endosperm or cotyledons. Secondary pests feed on grain products or grain that has already been damaged by the primary pests or as a result of harvesting, handling and transporting. The primary pests typically have a narrow range of food preferences such as cereals and pulses. The secondary pests, however, have a wide host range including damaged whole grains, milled products such as flour and processed and manufactured food products such as breakfast cereals, chocolates and compound animal foods. There is a distinct difference in the life cycle of pests belonging to these two categories. The life cycle of a primary pest involves lodging of the eggs inside or on the outer coat of grain, followed by development within the grain, making the immature stages difficult to detect. Because the entire life cycle (egg, larva and pupa) often takes place inside the kernel these primary pests are also called as internal feeders. In contrast, the eggs of secondary pests are laid in a scattered manner in or near the food source where the developing larvae can easily be seen. As the entire life cycle takes place outside the whole grain, the secondary pests are called as external feeders. Due to the major difference in life cycles, damage to whole grain by primary pests is very distinctive in form and pest recognition is easier compared to the secondary pests.

Major Primary Pests

The major primary pests of cereals include the beetles of the genus *Sitophilus* [the rice weevil *S. oryzae* (L.), the maize weevil *S. zeamais* Motschulsky, the granary weevil *S. granarius* (L.)], the borers [the lesser grain borer *Rhyzopertha dominica* (F.) and the larger grain borer *Prostephanus truncatus* (Horn)] and the Angoumois grain moth *Sitotroga cerealella* (Olivier).

The major primary pests of legumes (peas, beans, grams etc.) are bruchids including the bean weevil *Acanthoscelides obtectus* (Say), the cowpea weevils belonging to the genus *Callosobruchus* [*C. maculatus* (F.), *C. chinensis* (L.) and *C. analis* (F.) and *C. phaseoli* (Gyllenhal)], the groundnut bruchids *Caryedon serratus* (Olivier) and the Mexican bean weevil *Zabrotes subfasciatus* (Boheman).

Major Secondary Pests

The major secondary pests include two species of *Tribolium*, the rust red flour beetle *T. castaneum* (Herbst) and *T. confusum*; a group of *Cryptolestes* species [the rusty grain beetle *C. ferrugineus* (Stephens) and flat grain beetles *C. pusillus* (Schönherr) and *C. pussilloides* (Steel and Howe)]; two *Trogoderma* species [the Khapra beetle *T. granarium* Everts and the warehouse beetle *T. variabile* Ballion]; the saw-toothed grain beetle *O. surinamensis*; the warehouse moth *Cadra cautella* (Walker); the rice moth *Corcyra cephalonica* (Stainton); the Indian meal moth *Plodia interpunctella* (Hübner) and a group of *Liposcelis* psocid species [*L. bostrychophila* Badonnel, *L. entomophila* (Enderlien), *L. decolor* (Pearman) and *L. paeta* Pearman].

Pests of Dried Fruits and Nuts

Major insect pests under this category include the dried fruit beetle *Carpophilus hemipterus* (L.) and several moths including the Indian meal moth *P. interpunctella*, almond moth *C. cautella* (Walker), tobacco moth *Ephestia elutella* (Walker) and the raisin moth *Cadra figulilella* (Gregson).

Pests of Durable Herbs, Spices, Tobacco and Pet Food Products

Key pests under this category include the cigarette beetle *Lasioderma serricorne* (F.), the drugstore beetle *Stegobium paniceum* (L.), the carpet beetles *Trogoderma glabrum* (Herbst) and *Trogoderma ornatum* (Say), the lesser mealworm *Alphitobius diaperinus* (Panzer) and the clothes moth *Tineola bisselliella* (Hummel).

Pests of Dried Fish and Other Animal Products

Species particularly belonging to the genus *Dermestes* called hide beetles (*D. maculatus* Degeer, *D. frishii* Kugelann) and the red legged ham beetle *Necrobia rifipes* (Degeer) are considered as major pests of stored dried fish, hides, skins and other dried animal products such as processed meat and cheese.

Predators and Parasitoids

The predators include mainly the bugs such as the stack bug *Lyctocoris campestris* (F.), the cereal bugs *Xylocoris* species, and the assassin bugs *Amphibolus venator* (Klug) and *Peregrinator biannulipes* (Montrouzier). These insects do not damage the commodities, but prey on other insects, which can be an advantage from pest management point of view. However, their presence can be considered as a contamination issue and a health issue because they may also irritate the storage workers.

Parasitoids that are associated with stored products are mostly wasps that attack the juvenile stages of beetle and moth pests that are already in the commodity, and do not feed on the commodity. The major parasitic wasps include *Trichogramma* species; *Habrobracon* species and *Venturia canescens* (Gravenhorst) parasitising several moth species; *Anisopteromalus calandrae* (Howard) parasitising several beetles and moth species; *Uscan* species parasitising eggs of bruchids; and *Choetospila elegans* Westwood parasitising several beetle pests.

Scavengers and Foragers

These are insects that occur in stored products but don’t feed directly on them. They rather feed on the residues or the dead bodies of insects and other animals. Some of the notable scavengers include cockroaches, earwigs and silverfish.

Implications from Insect Infestations

Economic Implications from Quantitative Loss Due to Physical Damage

The term ‘post-harvest loss’ (PHL) is being used over last two decades in relation to both quantitative and qualitative food loss in the post-harvest system (de Lucia and Assennato 1994; Hodges et al. 2011). A significant quantitative loss can occur to the stored products due to physical damage caused as a result of direct consumption by primary pests followed by a series of invasions from a range of secondary pests. A cumulative weight loss of up to 56.9% of wheat was reported as a result of feeding by *R. dominica* over a period of 2 months (Rao and Wilbur 1972). Few reports are also available from laboratory evaluations on the direct physical damage caused to grain by psocids. McFarlane (1982) estimated a weight loss of 5% and of milled rice as a direct result of heavy infestations of *L. bostrychophila* over a 6-month period, whereas Kücerová (2002) measured an average weight loss of 9.7% of broken wheat kernels due to infestations from the same species over a 3 months period. Weight loss was 0.17% during the 4 months of storage from a stable infestation of 4000 psocids/kg. In a recent study, it was revealed that although weight loss due to *L. entomophila* and *L. paeta* infestations was low in intact kernels (0.2 and 0.4%, respectively) compared with damaged wheat seeds (8.5 and 3.3%, respectively), germination in intact kernels was reduced by 32% by *L. paeta* infestation (Gautam et al. 2013).

In terms of post-harvest losses during storage, there were species-specific reports available from several countries in the sub-Saharan Africa. *C. maculatus*, alone was found to be responsible for up to 24% losses in stored pulses in Nigeria (Tapondjou et al. 2002); whereas about 23% losses were recorded in stored maize due to combined infestations of *S. zeamais* and *P. truncatus* in Benin (Meikle et al. 2002).

Physical damage to grain during post-harvest handling such as threshing, drying and transporting make the grain vulnerable to rapid and extensive damage and decay by insects and mould (Rowley 1984). The most common methods to measure weight loss assessment include the standard volume weight, thousand-grain mass and count and weigh methods (Reed 1986). Our discussion, however, will be focussed on reported losses irrespective of the methods used to measure them. Although figures on actual economic damage are difficult to obtain due to the ‘commercial in confidence’ nature of the information, we present here the published reports.

A comprehensive review of the assessment of losses caused by insects to stored food commodities was undertaken way back in 1955 (Parkin 1956). According to that review, the annual losses to grains as a direct result of insect infestations from major stored grain pests in the USA over a decade (1951–60) was 325 million bushels, valued at $454.8M (USDA 1965). It is noted that the currency values reported here and elsewhere in the text are at the time of the respective reporting period, it may vary significantly if calculated at the present time. Moreover, a further loss of $8.8M incurred due to insect infestations in processed cereal products (USDA 1965). We have seen similar figures on post-harvest losses due to insects even after 30 years after that report, where the losses in the USA were estimated to be $500M per year during 1990 (Herein and Meronuck 1991). In contrast, based on a study over 1961–71, Bourne (1977) reported the losses arising from insect infestation in stored grain in central storage and handling systems in Australia to be insignificant. Although there has been no recent report from Australia on the current losses from insect infestations in stored post-harvest commodities, research in the USA has estimated that in developed countries, the average annual losses may go to 10% (Mason and McDonald 2012).

For the Indian subcontinent, the annual losses of grain during storage are estimated at $1B (INR 50B) (Singh 2010; Nagpal and Kumar 2012), and losses due to insect problems alone are estimated around $364M (INR 17B) (Boxall 2001). In the developing countries, the estimated losses are being reported to be up to 20% by several authors (Mason and McDonald 2012). In sub-Saharan Africa, the losses are reported to be around $4B annually (World Bank and FAO 2011).

In a recent study, Abass et al. (2014) assessed the post-harvest handling practices and food losses in a maize-based farming system in semi-arid areas of Central and Northern Tanzania during two harvest seasons in 2012. Based on the major crop maize, these researchers have estimated the quantitative post-harvest losses during storage to be 15–25%, mostly attributed to damages caused by the larger grain borer, *P. truncatus*), the grain weevil *S. granarius* and, the lesser grain borer *R. dominica*.

Among the range of pests, *S. zeamais* is considered as the most destructive in stored maize grain in tropical and subtropical regions worldwide and causes grain yield losses of 15–30% in developing countries (Bergvinson 2001). In sub-Saharan Africa, *S. zeamais* along with *P. truncatus* is reported as the major pests of stored maize and significantly impact household food security in the smallholder sector (Vowotor et al. 2005).

Effect on Quality

Serious biological deterioration of stored commodities, specifically grain can occur between the initial storage period and first processing as a direct result of activities from insects and related fungi (Fleurat-Lessard 2002). The effect of insect activities in grain mass can be multifold so far as quality is concerned. The grain loses value and receives a lower grading due to simple contamination from dead insect bodies, waste products, frass and dusts as a result of insect activities (Fleurat-Lessard 2002). The insect feeding activities can also add to the fatty acid content of the grain and leave high quantities of uric acid that lead to grain rancidity (Mason and McDonald 2012).

Significant effect on seed germination due to direct feeding by insects has been well demonstrated. In Brazil, Santos et al. (1990) showed that the presence of *S. zeamais* and *S. cerealella* in maize grains led to a reduction in germination with increasing developmental stage of the insects. In Nigeria, Okiwelu et al. (1987) recorded high level of moisture, combined with a decrease in germination ability of maize due to infestation by *S. zeamais*, while Mbata (1994) showed that infestation of bambara groundnuts (*Vigna subterranea*) with *C. subinnotatus* reduced seed viability and increased free fatty acids and peroxides, which are indices used in measuring biochemical deterioration. In a recent study, Keskin and Ozkaya (2013) revealed that infestation from *S. granarius* had significantly reduced thiamine and riboflavin contents in wheat over a period of 6 months storage. Sudesh et al. (1996) found that infestation of wheat, maize and sorghum grains with single or mixed populations of *T. granarium* and *R. dominica* resulted in substantial reductions in the contents of total lipids, phospholipids, galactolipids, and polar and nonpolar lipids, while Kumar et al. (1996) recorded a substantial reduction in starch in parboiled cassava chips due to infestation with *S. oryzae* and *R. dominica* as compared to the uninfested chips.

Apart from the quality affected by the devouring of grain by both adults and immatures of the pests, the effect of quality can be severely affected by secondary infestation from a range of fungi. The presence of insects raises the product temperature, due to their feeding activity, resulting in ‘hot spots’, sometimes reaching up to 57 °C (Mills 1989). These spots, in turn, lead to concentrating of humidity within the product, thus stimulating seed deterioration and further fungal activity. Fungal infestation results in change in colour, taste, smell, reduction in nutritional value, increase in free fatty acids (FFA) and reduction of germination ability (Sauer et al. 1992). Preferential attack of the embryo by *Eurotium* species, can result in 50–100% reduction in germination, reduced amino acid contents of the grain that leads to loss of the characteristic grain odour and flavour (Sauer et al. 1992). In a heavily infested grain bulk, the natural odour is replaced with a musty or mouldy odour. Mixing of off-odour grain with a good batch of grain fails to mask the off-odour and it can be expensive to overcome this problem through the use of ozone (Mendez et al. 2003).

Several species of *Aspergillus* and *Penicillium* are associated with mycotoxins (e.g. aflatoxins in groundnuts, sorghum and rice, and orchratoxin A in corn, oats and barley) that can inflict major health hazards to human and animals (Sauer et al. 1992) (see Sect. ‘Work Place Health and Safety Implications’, below for more).

Work Place Health and Safety Implications

Infestation of insects in grain mass can indirectly lead to several workplace health and safety issues, particularly to those handling grain from harvest to storage and transport.

Although psocids are considered as one of the smallest among all stored product insects, they have emerged as a major concern in the stored grain environment in recent years and in large infestations, they can have significant health and safety impacts (Nayak et al. 2014). In severe infestation situations, psocids have been reported to have swarm over storage walkways, ladders, etc., making them slippery and exposing workers to risk of injury (Rajendran 1994; Jiang et al. 2008). Psocids have also been implicated in the development of allergic conditions in workers caused by transmission of microorganisms (Turner et al. 1996) and may be responsible for transmission of bacterial diseases (Obr 1978), although there is no direct evidence for this. There have been reported cases of delusory parasitosis caused by psocids (Turner 1987). Lis et al. (2011) highlighted the serious health hazard of defensive secretions produced by *T. castaneum* and *T. confusum*. Through a literature review, they have reported the carcinogenic effects of benzoquinones, secreted by these two stored product pests.

As mentioned in the preceding section, elevation in heat and moisture in the grain mass due to insect feeding encourages the development of several species of fungi. Mycotoxins are metabolites that are produced by these fungi can cause several animal and human health problems. A flatoxin from *Aspergillus* is of the greatest concern due to its high carcinogenic properties, and therefore, it is being regulated in the grain trade across the globe (CAST 1989). In several developing countries, high intake of aflatoxins was shown to have a positive link with high incidences of primary liver cancer and hepatitis B in human populations (CAST 1989). Moreover, in animals, aflatoxin was shown to cause acute or chronic diseases in poultry, swine, cattle and many other farm animals (CAST 1989).

Rejection by Consumers and Loss of Market

The consumer preference for safe and clean food that is free from insects and chemical residues has taken an unprecedented momentum over the last few decades. In a comprehensive review recently, Stejskal et al. (2015) have highlighted the filth contamination of flour and pest risk trends in stored food and feed products in Europe based on reported cases in the past 80 years. The two demanding aspects of safe and clean food of the consumer can be conflicting. Several contact insecticides that have been used by industry to provide long-term protection from a range of pests leave residues that attract a lower price and restrict markets. Fumigations help in disinfesting stored commodities only and it is difficult to provide long-term protection from insects without applying contact pesticides. Moreover, increasing pressure from environmental movements has seen the phase out of several treatments including one of the most effective fumigants, methyl bromide (Johnson et al. 2012).

To have a competitive edge and meet the consumer preference for insect-free grain, Australia has adopted a ‘Nil tolerance’ policy for live insects in grain destined for international markets, and this principle is recently being implemented at the domestic market (GTA 2017). Currently, this policy applies to all life stages of 12 pest species comprising beetles, moths and psocids. In the USA, strict laws are also in place limiting the number of permissible live insects in commodities and insect-damaged kernels (IDKs), both of which can incur losses in the market. U.S. Department of Agriculture rejects a grain consignment for sale for human consumption, if two or more live insects are detected in a kilogram of grain sample (Adam and Alexander 2012). This issue can be overcome through fumigation to kill the live insects, but at a cost that reduces the value of the consignment. Discounts are also imposed at the time of sale if grain bulk is detected with more than 32 IDKs in a 100 g sample.

Even if exporting countries try to maintain the 'insect-free' status of their grain consignments, sometimes, due to the failure of the phytosanitary measures, live insects are detected at the importing ports that can lead to rejection of the whole shipment of grain or can incur a demurrage cost along with costs for disinfestation. In 2007, Egypt rejected a US$84M load of US soft red wheat due to detection of live insects (Farm Futures 2007).

Strict legislations and regulations are also in place at the international level to restrict the movement of certain pests that are considered exotic to some countries. Quarantine regulations have been imposed on such insect pests and standardised phytosanitary certifications have been developed to restrict their movements for global trading of grain and processed commodities (Tyler and Hodges 2002). Notable among the quarantine pests is the Khapra beetle, *T. granarium* (Stibick 2007). This pest is notorious for its destructive nature and so far reported on 96 commodities across the globe. Due to its high level of tolerance to major treatments including fumigant methyl bromide, in the USA, the cost of its eradication in the 1950s was estimated to be US$8.4M (Klassen 1959).

Costs Associated with Pest and Resistance Management and Research

Since the realisation of the importance of the insect pests in stored commodities and their economic implications, there have been ongoing efforts to develop new treatments and pest management strategies to reduce their impact. Post-harvest storage environments across the globe have witnessed the emergence and demise of several contact insecticides and fumigants over the last century. Typically a fumigant gas is used as a disinfestant to control pest populations in an already infested grain bulk, whereas contact insecticides are being used as residual treatments of freshly harvested uninfested grain for protecting it from insect attacks, hence these treatments are named as 'grain protectants'. Apart from these major treatments, there are several other aspects of pest management that include hygiene, aeration cooling, drying and controlled atmospheres. There is a significant literature available on the history of different pest management options developed and used in the stored grain systems across the world (see Chapter 'Insect Pest Management in Stored Grain' by Daglish et al.).

The cost associated with managing a pest infestation varies greatly depending on the treatment used. We cite a few examples here on the economics of the current pest management practices in developed countries. In Australia, the current costs for a range of pest management options calculated in Australian dollars per tonne of grain stored (incorporating labour and materials) are: on-farm hygiene (AU$0.23/t), an aeration cooling system (AU$0.91/t), aeration drying (AU$17.21/t), phosphine (AU$0.35/t), sulfuryl fluoride (AU$4.00), silo bags (AU$4.00/t), and a protectant treatment of chlorpyrifos-methyl, s-methoprene and spinosad (AU$3.40/t) (GRDC 2017). Adam and Alexander (2012) outlined a comprehensive account of economics around the integrated pest management (IPM) decisions that are currently being practiced in the USA. These authors, calculated the cost components of sampling for live insects to be approximately at US$0.40/t including the cost of amotorised equipment and labour required to separate and count insects. They have also undertaken a costing for fumigation with turning to be approximately at US $1.20/t (Adam and Alexander 2012).

It is important to note here that the estimated cost of discovery, development and registration to bring a new pesticide to the market for use by industry exceeds US $180M and may take 8 to 10 years (Whitford et al. 2017), and the stored products is likely to be a small market for new pesticides. Development of resistance in target pest species to a particular chemical, therefore, can be a very costly affair, both in terms of losing the market for that product and developing an alternative. Even if there have been several contact insecticides and fumigants introduced to the storage systems across the world, resistance in key pest species have been a regular phenomenon (Nayak et al. 2015). In the last two decades, a major emphasis has been given to the management of resistance in key pest species across the globe to enhance the longevity of established products (Nayak et al. 2015). In the following paragraphs, we will cite few examples to highlight the costs associated with pest and resistance management and related research.

Recently, an AU$30M research initiative was launched under the umbrella of Cooperative Research Centre for National Plant Biosecurity (CRCNPB) to protect Australia's post-harvest grain (approximate annual value of AU$9B) (CRCNPB 2007). Over a 5 year period (2007–12), the CRCNPB brought together the skills of industry, government and scientific institutions in a unified national approach to develop new technologies, training and biosecurity safeguards. Key CRCNPB partners included federal and state governments, universities, three major bulk grain-handling companies, and the Grains Research and Development Corporation (representing grain growers). A significant portion of the AU$30M was allocated to the development of new phosphine fumigation protocols to manage strongly resistant genotypes in key pest species; development of alternatives such as nitrogen and sulfuryl fluoride, and other research to underpin an integrated approach for pest and resistance management for growers and bulk grain handlers. A follow-up initiative, the Plant Biosecurity Cooperative Research Centre (2012–18), invested approximately AU$42M over a 5 year period focused on the development and adoption of new technologies and tools to protect the post-harvest grain from major pests. A significant part of the investment was allocated towards an ongoing national monitoring programme for pests to key fumigant phosphine (PBCRC 2012).

In 2014, with USAID support of US$8.2M, several universities in the USA formed a unique consortium called ‘Feed the Future Innovation Lab for the Reduction of Post-harvest Loss’ (KSU 2014). A major goal of this initiative is to improve storage conditions and pest management practices in developing countries including Ethiopia, Ghana, Guatemala and Bangladesh.

The high cost of research investments to tackle insect problems in the grain storages in Australasia region and the benefits from such investments have been well demonstrated in a report from the Australian Centre for International Agricultural Research (ACIAR) (Francisco et al. 2009). As part of the regional cooperation and development programmes, ACIAR supported a series of four research projects during 1983–2005 on developing best practices in use of pesticides for protection of post-harvest grain in the tropical areas of Australia, the Philippines, Malaysia, Thailand and China (Francisco et al. 2009) The main aim of these projects was to use grain protectants in combination and in rotation at lowest effective dose rates to mitigate the serious resistance problem in several pest species to the organophosphate malathion. At the time of an impact assessment undertaken in 2007, the total investment in these projects was AU$9.6M. The adoption of pest management technologies developed through these projects had resulted in a reduction of losses in stored paddy in the Philippines from 9.5 to 4.8% per year (Francisco et al. 2009).

Conclusion

In this chapter, we attempted to give an overview of the importance of stored products insects that highlighted their economic implications in terms of quantitative loss, the effect on quality and consumer and market sensitivity. The impact of insect pests on post-harvest losses contributes significantly towards the overall food loss of approximately 1.3B tonnes of food every year across the world (FAO-World Bank 2010). We also tried to demonstrate the costs involved in research and development towards managing them to emphasise the indirect costs associated with these pests. To conclude, we will outline several approaches that we suggest would help in reducing the impact of stored product pests, specifically in the stored grain environment.

Across the globe, most of the post-harvest grain that is stored in traditional storage structures are vulnerable to insect infestations and mould growth, specifically during long-term storage. The first step in protecting post-harvest grain is the availability of modern storage systems that are sealable (airtight), to be suitable for using fumigants; currently, a common method used for disinfestation of bulk grain. In Australia, sealable silos are currently being sold through adherence to a legalised standard AS2628 (GRDC 2014). Silos meeting this standard pass a 5-minute half-life pressure testing, ensuring a high level of air-tightness. Investing in such type of storage structures would play a critical role in reducing post-harvest losses and preserving grain quality, which in turn would yield in increased revenue in the long term. For farmers and small holders of grain in Africa, there are new modern small storage options (from 20 to 3000 kg capacity) available including UV-stabilised polypropylene bags, small metal silo bins and hermetic bags (FAO 2014).

Early detection of pests through regular monitoring of their populations in and around storages and diagnosis of their resistance towards different treatments are critical components for implementation of appropriate strategies to manage them and preventing their spread. In Australia, a national monitoring programme is in place since last three decades that helps in early detection and management of strongly phosphine resistant stored grain pests across both on farms and bulk handling storages (Nayak et al. 2017). Development of alternatives to traditional pesticides and biorational approaches to disinfest and protect grain are also being promoted recently as ways to address consumer demand for insect and residue free grain and processed food. In a recent review Phillips and Throne (2010) highlighted the importance of biorational tools in stored commodity pest management that include sanitation, management of temperature in stored grain, use of natural enemies of storage pests, computer-assisted decision-making system for pest control and insect sampling. In Australia, recently sulfuryl fluoride has been developed as a suitable alternative to phosphine (Nayak et al. 2016) to manage strongly phosphine resistant populations of rusty grain beetle *C. ferrugineus*, that has recently become a major problem in the bulk grain storages (Nayak et al. 2013). Binary combinations of currently registered products with a new biopesticide spinosad (Daglish 2008) are being used by industry as a way to mitigate multiple resistances to grain protectants in major stored grain pests in Australia; whereas a method is in place for early detection of resistance to spinosad in its target pest *R. dominica* (Nayak and Daglish 2017).

While we are developing modern pest and resistance management strategies that are aimed at meeting the cost-benefit expectations and market access requirements, the role of extension specialists cannot be ignored. In Australia, a well-established national grain storage extension team works as an interface between the researchers and end-users, specifically the growers. This network facilitates extension programmes to growers across the country that include bulletins, fact sheets, on-site demonstrations and workshops emphasising the benefits of adoption of best pest and resistance management practices along the post-harvest grain value chain.

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Chapter 2

Human Health Problems and Accidents Associated with Occurrence and Control of Storage Arthropods and Rodents

Vaclav Stejskal, Jan Hubert and Zhihong Li

Introduction

The UN Food and Agriculture Organization (FAO) estimates that the global human population will reach 9.8 billion in the year 2050, that is over 70% higher than the previous estimate cited by Hagstrum and Phillips (2017). The recent scientific estimates also predict that human population and global demand for food will be increasing for at least another 40 years (Godfray et al. 2010), which is a huge challenge for agriculture to increase its overall gross field production. However, human food security depends not only on primary agricultural production but also on efficient post-harvest storage and distribution of agricultural commodities and food products. Thus, future public resources investments to fund research on Integrated Pest Management (IPM) of stored products will be essential to protect the world's food supply (Hagstrum and Phillips 2017).

The enormous amount of human food resources stored all around the globe becomes spoiled due to many pest organisms including insects, mites, rodents and birds, fungi, bacteria, and viruses. A recent review by Hagstrum and Subramanyam

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(2009) lists more than 1600 insect species associated with stored products. In China, the national survey revealed 270 species of stored grain insects (Yan et al. 2008). The occurrence of so many arthropod and rodent species in the vicinity of stored commodities represents the potential for establishment of pest infestations. The complex web of negative interactions of animals with humans is traditionally divided into two broad categories: economical and human health related. The pest negative effects on human health may be classified as direct or indirect (Table 2.1 and Fig. 2.1). Food

Table 2.1 Categories of negative effects of stored product pests on human health

Main categories of negative effects	Sub-categories of negative effects
Direct effects	**(a) Effects on food security** – Food contamination and deterioration **(b) Effects on food safety** – Physical irritants of digestive tracts – Alimentary contamination by arthropod allergens – Food toxins and poisonings – Carcinogens and mutagens in food **(c) Effects on home- and workplace safety and occupational disease issue** – Pest effect on human psychic health – Aero and contact-arthropod allergen contamination of environment – Parasitoses (myases; acariases) – Insect and mites bites and itches – Rodent bites – Slip and fall accidents – Human injuries from fires, explosion and mechanical collapse of damaged store structure
Indirect effects	**(a) Transmission and host of pathogens** – Transmission of fungi – Transmission of bacteria – Transmission of viruses **(b) Changes of temperature and humidity, physical grain destruction by pests and treatments: facilitation of fungi growth and production** of mycotoxins – Physical grain destruction by pest accelerating fungal colonization and growth – Rapid increasing of temperature (heating) and humidity (hot spots formation) caused by pest respiration in stored commodities – Increased grain humidity in stored commodities caused by insecticide sprays **(c) Poisoning by toxic insecticides and rodenticides** – Dust and silicosis – Insecticide residues – Fumigants – Rodenticides **(d) Intoxication by anoxic atmospheres (CO_2, N_2, NO_2)** **(e) Human entrapment, engulfment and drowning in grain mass during pests sampling or pesticide application** **(g) Agrochemicals as sources of explosion and fire in stores**

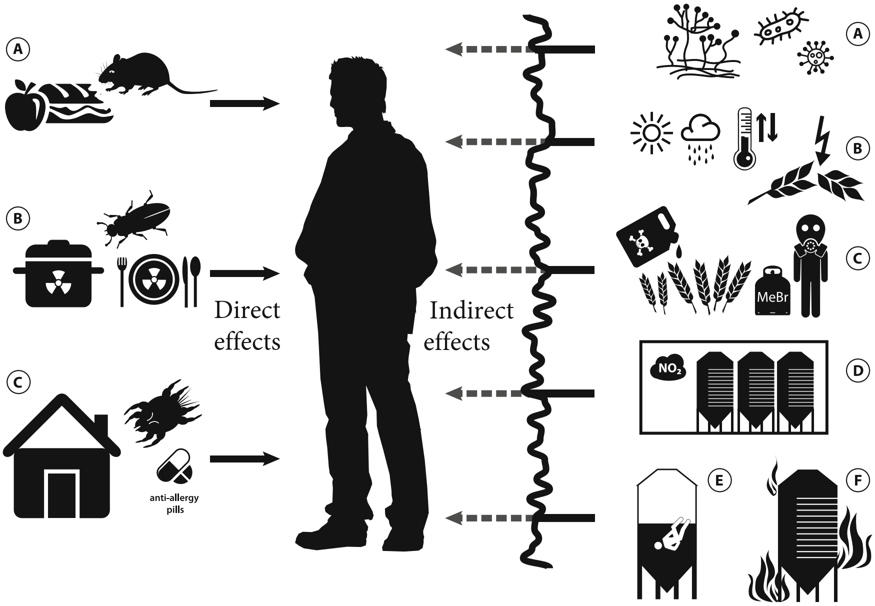

Fig. 2.1 Graphical visualisation of the direct and indirect effects caused by stored product pests to human listed in Table 2.1

contaminated by mite allergenic proteins (i.e. faeces, dead mite bodies) is a typical example of the direct negative impact of storage pests on human health. The presence of pests requiring chemical treatment may accidently result in excessive pesticide residues in human food. An accidental human intoxication during pesticide application is an example of the indirect effect of pests on human health. Arthropods can be used detrimentally (Lockwood 2012), and the chemicals selected for pest control (e.g. fumigants such as phosphine) can be misused for suicide, criminal or terroristic purposes. It is important to note that some storage arthropods may be beneficial from economic standpoints, but have negative impacts on human health (Stejskal 2000). For example, as documented beow, several species of parasitic bethylidae wasps and predatory mites are able to control pests in stores as natural bioagents but they may also attack and sting humans.

Because of the above-mentioned complexity of storage pest–human interactions, this chapter focuses more on reviewing of pest key negative impacts of storage pests on human health and providing selected illustrative examples, rather than a comprehensive ‘list of stored arthropod and vertebrate species of medical importance’.

Direct Effects on Human Health

Direct effects of storage pests on human health include reduction or spoilage of the food supply (i.e. effect on food security), food resource contamination (effect on food safety) or direct pest attack on human (stings, parasitism) caused by the presence of pests in homes or working environments (i.e. effects on home and workplace safety). This section is divided according to these areas of direct negative effects.

Effects on Food Security

Food security and national state reserves. National food security is one of the most important political priorities of federal and state governments. To ensure an adequate food supply, many governments support gathering strategic food resources to provide food to citizens in the unexpected crisis situations [natural disasters, wartimes (Stejskal et al. 2015)]. For example, to address this problem the State Administration of Grain (SAG) was established in 1999 on the basis of the former State Administration of Grain Reserves in the People's Republic of China. There are currently 196,000 people engaged in the Chinese grain storage nationwide (Grain 2002; Gu 2016). To protect state reserves, technologically advanced storage facilities are built and equipped with automatic ventilation appliances, computer-controlled temperature measuring systems and closed-loop fumigation devices.

Food removal/deterioration as a cause of starvation and malnutrition. Various international agencies (e.g. FAO, WHO) estimate that there are more than 900 million people suffering from malnutrition at the worldwide scale. While most citizens of developed countries have not suffered from malnutrition since post WWII, it is estimated that currently more than 20 million people in developing countries face starvation and famine (BBC 2017). According to the United Nations (UN), estimates show that about 1.4 million children are at risk annually from starvation, which means that we are facing the largest worldwide humanitarian crisis since 1945. Such estimates clearly demonstrate that for many developing countries, food security is still a serious issue. The main food security problems in these countries are caused by ongoing war, political instability, environmental fluctuations or disasters coupled with the lack of economic resources, proper agricultural technology knowledge and availability. In the agricultural arena, food losses occur not only during the cropping stage but also, and sometimes more importantly, during the storage of the commodity and food processed from raw commodities and stored. When a certain percentage of stored harvest is lost, the same percentage of the resources contributed to the crop production is also wasted. Moreover, the nutritional value of stored food can be significantly reduced as well. The consumption of pest-damaged cereals could cause malnutrition, leading to immune system weakening and consequently higher risk of infectious diseases outbreaks and/or its severity enhancement and disease course worsening. The negative impact of pest-damaged food consumption has been observed in a mice model proving reduced weight gain compared to mice fed with undamaged corn (Santos 2007).

Extent of food/commodity losses during storage. Feeding activity of rodents, birds, insects and mites causes significant loss of stored commodities and food resources. There are different estimates of stored commodities worldwide losses ranging from 3 to 60%. Phillips and Throne (2010) documented that stored product insects can cause post-harvest losses of around 9% in developed countries and more than 20% in developing countries. Reduction of food and feed storage losses can be accomplished through the use of pesticides. The availability of insecticides led to the historical decrease of the amount of infested wheat cargos imported to Great Britain during 1953–1970. Infestations of export cargoes declined from more than 95% to nearly zero within a decade of using malathion in Australia (Freeman 1973; Muir and White 2000). It is difficult to prevent losses in places where proper storage technology and protective chemicals are not available. Tefera et al. (2011) demonstrated that smallholder farmers in developing countries are not equipped with effective grain storage technologies that would help prevent losses caused by post-harvest insects and pathogens. In the case of agricultural commodities produced and stored in Asia, it was documented that rodents and arthropods are more damaging (responsible for approximately 5% annual losses) than birds (responsible for approximately 0.8% losses) (Ahmed 1983). The losses are mainly caused by their high rate of multiplication resulting in extensive populations under optimal conditions. Losses caused by rodents are associated with damage that occurs from direct food consumption but also from food spoilage by rodent urine and removal of food, described as resource hoarding or caching behaviour (Smith and Reichman 1984).

Effects on Food Safety

Arthropods are major contaminants of agricultural commodities and food. For example, Orriss and Whitehead (2000) analysed food samples obtained from 4795 USA-FDA detention offices of imported agricultural food products, and showed that the most frequent problem of non-complying imported food was arthropod filth (32%), followed by under-processing and spoilage of canned food (12%) and microbial contamination (11%). Arthropod pests are a hazard to food safety since they not only vector various diseases and toxicogenic fungi (moulds), but directly impact food safety via production of allergens and potential carcinogens (Gorham 1979; Arlian 2002; Stejskal and Hubert 2006). The main criterion defining risk of food contaminants occurrence is the probability of stored commodity/food contamination. To calculate this risk, estimation of at least three basic risk parameters are needed: (i) contamination rate, (ii) distribution of contaminants, and (iii) the efficacy of cleaning and disinfestation treatments. Although negative effects of storage pest on human health are known, there is surprisingly little experimentally substantiated information [e.g. Hodges et al. (1996), Stejskal and Aulicky (2014)] that establish risk of food contamination according to the parameters mentioned above. An alternative approach to food contaminants risk management was proposed by Stejskal (2001). This includes agricultural products quality and safety damages penalization and inclusion of this parameter of contamination into the economic injury level (EIL) equation. In the USA, a legislatively established maximal threshold for pest contaminants called the defect action level (DAL) was implemented (FDA 1988; Olsen 1998). Flinn and Hagstrum (2001) described one of the few biological control studies that showed reductions in insect fragment counts. The European Union has zero tolerance for live pests in food, but surprisingly no action levels or thresholds for pest contaminants are established in the EU legislation.

Physical irritants of digestive tracts. Many larvae from the family Dermestidae (Coleoptera) are equipped with sharp and curly setae that upon ingestion can irritate the human digestive tract. Consequently, they cause enteric problems (Lillie and Pratt 1980) such as diarrhoea and perianal itch (Judd 1956; Okumura 1967; Goddard 2003). Although larvae of some moths have been documented to cause dermatitis and urticaria; we could not find any records for pyralid moths from the genus *Plodia* sp. and *Ephestia* sp. In addition, it is still unclear whether the primary effect on the digestive tract results from physical irritation of epithelia by arthropod setae or allergic reaction to insect proteins.

Arthropod allergen in the food. Many arthropod and vertebrate species are documented as allergen producers. Arthropod contamination may negatively affect human health via pest allergenic proteins contained in faeces or dead bodies. A variety of allergic diseases have been described that are associated with food arthropod contamination, such as asthma, rhinitis and eczema. Currently, these allergies are reaching epidemic proportions in human population worldwide (Holgate 1999). In sensitive individuals, the consumption of contaminated food might cause cutaneous symptoms (urticaria, facial angioedema or both) or exacerbate respiratory diseases (rhinitis, asthma or both) immediately after the ingestion of mite-contaminated meal. This effect was documented as a result of eating mite-contaminated pancakes (Gonzalez-Perez et al. 2013). In some cases, this has led to anaphylactic reactions known as 'pancake anaphylaxis' (Matsumoto et al. 1996; Blanco et al. 1997; Sanchez-Borges et al. 1997, 2013). House dust mites (*Dermatophagodies pteronyssinus* and *D. farinae*), as well as stored product mites (i.e. *Acarus siro*, *Aleuroglyphus ovatus*, *Blomia tropicalis*, *B. freemani*, *Tyreophagus entomophagus*) are among the most common species responsible for anaphylactic reactions (Sanchez-Borges and Fernandez-Caldas 2015). Despite these reactions, allergen contamination is not usually monitored in stored food commodities.

Food toxins, intoxications, mutagens and carcinogens. Some insects produce defensive substances that are, after ingestion, toxic to vertebrates including humans. The most dangerous are blister beetles (Meloidae, Coleoptera), also known as

'Spanish flies', that produce the toxin cantharidine (Goddard 2003). These insects may occur on field crops and may thus appear in stored agricultural commodities as contaminants after harvest. Although cases of domestic animal fatal intoxications have been cited (Bahme 1968), no cases of human poisoning via food contaminated by cantharidine have been reported. However, these insects or their toxins present significant food safety dangers if used for criminal purposes (Lockwood 2012). The secretion of the red flour beetle, *Tribolium castaneum* (Herbst), contains at least 13 different quinones (Howard 1987) usually defined as 'defense and antimicrobial chemicals' (Yezerski et al. 2007). Contaminated food has not only unpleasant smell but also was documented to be responsible for liver and spleen tumours in small vertebrates (el-Mofty et al. 1988, 1989, 1992). These chemicals seem to be thermostable, since the mutagenic effects on mice were still evident after feeding of bakery products prepared with use of contaminated flour (el-Mofty et al. 1992). Biscuits artificially infested by *T. castaneum* contain 1,4-benzoquinone and induce neoplastic lesions in mice. However, so far no validation of the negative impact on human health of naturally available levels of *T. castaneum* contaminants in human food has been verified. Hodges et al. (1996) expressed the opinion that low quinone contamination rates naturally observed in rice are unlikely to pose a serious health threat. Future research in this field is necessary to identify the 'danger levels' of these compounds.

Effects on Home- and Workplace Safety and Occupational Disease Issue

Stored product insects, rodents and their natural enemies commonly occur in households, supermarkets, commodity stores, mills, bakeries and farms. Occasionally, pests and beneficial arthropods may reach high population densities in household pantries, but usually more frequently on farms and in industrial premises. Not surprisingly, many case studies describing various professional and occupational diseases among storekeepers, cereal workers and bakers connected to storage pest have been documented. Pest management professionals, and also scientists and technicians working with pest insects are at risk from associated occupational diseases.

Arthropod allergens and physical irritants. Contamination from allergens and physical irritants (e.g. spikes and setae) associated with stored product arthropods have been documented by several authors (Herling et al. 1995; Alvarez et al. 1999). Respiration allergies may be triggered by inhalation of spikes and setae (*hastisetae*) from larvae of Dermestid beetles. The most frequent genera include *Anthrenus* sp., *Attagenus* sp., *Trogoderma* sp. and *Dermestes.* Loir and Legangneux (1922) described a severe pruritic, erythematous and vesiculating eruptions on the exposed parts of the skin, and dry cough in stevedores who were unloading a cargo of bones infested by dermestids. Rhinitis and asthma symptoms after Dermestidae exposure in personnel in museums storing these species have been reported by Sheldon and Johnston (1941). Similarly, Siegel et al. (1991) described occupational disease symptoms such as conjunctivitis, sneezing, cough, dyspnoea, wheezing and dermatitis after exposure to dermestids. Brito et al. (2002) described a case of occupational rhinoconjunctivitis and asthma in a wool worker caused by dermestids in Spain. Case studies involving hypersensitive reactions to different insect orders and genera have been described by multiple authors (e.g. Lunn 1966; Herling et al. 1995; Alvarez et al. 1999; Alanko et al. 2000; Makinen-Kiljunen et al. 2001; Marraccini et al. 2007). In the meat industry, allergic diseases caused by mites can occur as well (Armentia et al. 1994).

Allergenic reactions are also reported for predatory mites (e.g. *Cheyletus* spp.), that might be utilized as a means of biological control of stored product pests (Musken et al. 2000). For example, the predatory mite C*heyletus eruditus* produces a respiratory allergen associated with persistent non-occupational allergic rhinitis (Poza Guedes et al. 2016), in addition, the protein extract of the mites caused skin reactions (Neto et al. 2002). Due to the feeding biology of *Cheyletus* on stored product mites and insect eggs (Cebolla et al. 2009), it is not clear whether the compounds responsible for immunoglobulin E (IgE) reactivity originates from the ingested prey pest-mites or *Cheyletus* themselves.

There are no worldwide acceptable threshold levels for arthropod numbers in stored commodities or limits regarding allergens and chemical contaminants. Stejskal and Hubert (2008) attempted to estimate the relationship between mite densities found in grain stores situated in the Czech Republic and potential risk of allergens. The estimation of mite allergy-risk levels (ARLs) was based on published mite densities that affect human health (Lau et al. 1989; Platts-Mills et al. 2000). Exposure to more than 100 mites/g of dust (1 mite g^{-1} grain) increases the risk of sensitization and clinical symptoms of allergic disease, while exposure to more than 500 mites g^{-1} dust (5 mites g^{-1} grain) increases the risk of acute asthma attacks (Lau et al. 1989; Platts-Mills et al. 2000). Based on these data, five classes of allergy risk level (ARL) have been estimated (Stejskal and Hubert 2008). Further research is recommended to identify new allergens and allergen producers among stored product pests and further effort is needed to quantify the risks associated with allergens.

Parasitoses (myases; acariases). Mites are reported to cause human ascariases (i.e. presence of living mites) of human internal organs (Li and Wang 2000; Cui 2014). The evidence for ascariasis occurrence mainly originates in Asia. Li et al. (2003) studied 1994 patients sensitive to mites, and among them 8% had mites in stool, urine samples or both. Li and Wang (2000) demonstrated a high risk of pulmonary ascariasis among staff of medicinal herb storehouses, rice storehouse and mills. In addition, the presence of stored product and house dust mites on human bodies is used in forensic acarology (Perotti et al. 2009; Solarz 2009). Various types of ascariases are widely distributed, but currently is not the main consideration in research.

Insect and mite bites and itches. Parasitoids and predators are often associated with stored product prey arthropods and can be utilized as biocontrol agents, but they occasionally directly attack humans. Hymenoptera stings cause the majority of severe toxic and/or allergic reactions to humans, as the venoms may contain several toxic constituents, including histamine, serotonin, acetylcholine, dopamine, and noradrenaline. Large wasps endanger workers when they fly indoors, while other wasps can nest in hidden areas within internal structures. Less visible, but medically important are attacks inflicted by small wasps. Hatsushika et al. (1990) described cases of sting dermatitis caused by the bethylid parasitic wasp *Cephalonomia gallicola* (Bethylidae, Hymenoptera) in Japan. They reported four human cases of sting dermatitis (28-year-old woman, 27-year-old woman, 43-year-old woman and 15-year-old boy) from Tatami-mats in the Okayama Prefecture. The cigarette beetle *Lasioderma serricorne* was listed as being associated with the wasp. Recently, Lee et al. (2014) described the first clinical reports of stings caused by *C. gallicola* in Korea. All patients developed painful erythematous papules at the sting sites and had a past history of wasp attack. There are also reports of several cases of *C. gallicola* attacks in the Czech Republic and elsewhere in Europe (Mazanek 2002). Parasitic or predatory mites may cause dermatitis. For example, mites in the family Pymotidae are ectoparasites of stored product insects caused skin reactions in laboratory workers in Brazil (da Cunha et al. 2006). Skin rashes were reportedly caused by an outbreak of *Pyemotes herfsi* in the Midwestern United States (Broce et al. 2006). Skin lesions were also observed after bites from *C. malaccensis* (Yoshikawa 1985). These findings are of importance for determining if predatory mites can be used in biological control programs for stored product arthropods.

Rodent bites. Another public health issue of concern is bites inflicted by rodents in farm and urban environments (Battersby et al. 2008). Hirschhorn and Hodge (1999) examined reports of 622 victims of rodent bites in the USA. Rodent bites can also help transmit disease pathogens (Elliott 2007).

Pest effect on human psychic health. Arthropod effects on human psyche have both medical and social dimensions (Anderson 1993). Insect encounters in food have a negative emotional effect that is compounded in the event of a large infestation (Baker and Swan 2013). Repeated exposures to arthropod or vertebrate pests can elicit serious symptoms related to entomophobia or pathological anxiety. Evolutionary psychologists claim that the fear or phobic response from a pest encounter is a natural defensive mechanism (Nesse and Williams 1995). However, many human behavioural defences seem to be expressed too readily or too intensely as shown by models based on so-called smoke-detector principle (‘defences with graded responses expressed to the optimal degree when the marginal cost equals the marginal benefit’) (Nesse 2001). This potential emotional harm or damage may be one explanation as to why in most countries there is a zero tolerance threshold of arthropods in food or in human habitat (Stejskal 2002). Fear resulting from pest encounters may also be utilized in criminal attacks, as shown in the case of using white mice during a robbery in a Czech supermarket in 2011 (CT24 2011).

Slip and fall accidents due to insect and mite floor contamination. One of the most common causes of injuries in the warehouse and retail markets is slips on a floor. Although there are reports of arthropod infestations causing accidents of this type there no published studies of such incidences. The general recommendations are to simply us proper cleaning and sanitation practices to prevent slips and falls inside structures where there is human traffic.

Human injuries connected to fires and explosions associated with pest activities. There are various causes of fires and explosions of commodities in stores and silos (Kimball 1997; Persson 2013) including the biological explanations [e.g. Meijer and Gast (2004)]. Several records of fatal incidents of staff or firefighters associated with fire, blaze, explosion or mechanical collapse of grain silo elevators structure and building constructions on farms have been published (e.g. Stusinski 2013). Rodents can chew through electric wiring and insulation, insects and microorganisms may produce moisture and heat in an infested and unventilated commodity, and feeding by insects produce dust and frass that can contribute to dust in silos and mills.

Indirect Health Effects

Indirect negative effects of storage pests include transmission of pathogens, parasites and increases in commodity temperature or moisture that contribute to the growth of mycotoxin or allergen-producing fungi. Another type of indirect negative effect is secondary exposure to insecticides, including fumigants, which contribute to health risks. Indirect effect also includes accidents or criminal use of pesticides due to their widespread availability in most countries.

Transmission and Hosting of Pathogens (Storage Pest Role of Vectors, Hosts)

Transmission of fungi. Any microbial infestation renders the stored product unsuitable for human or animal consumption. Infestations of stored product insects can contribute to microbial infestations, most commonly caused by the genera *Aspergillus* and *Penicillium.* Hanuny et al. (2008) found that *Rhizoglyphus robini* enhanced the fungal infestation of onion by *Fusarium oxysporum* dissemination. Franzolin et al. (1999) conducted an experiment with sterilized and contaminated maize and showed that *Tyrophagus putrescentiae* contributed to the growth of *Aspergillus flavus*, and also vice versa, suggesting a synergistic effect and mutual benefit between these two species mycological analysis of the house mice (*Mus musculus*) faeces revealed 35 isolated species of fungi (Stejskal et al. 2015).

Transmission of parasites, pathogenic viruses and bacteria and risk of antibiotic resistance genes distribution. Rodents produce faeces that contaminate agricultural commodities and processed food products (Stejskal and Aulicky 2014; Aulicky et al. 2015). It is general public health knowledge that rodent urine and faeces may contain parasites, pathogenic bacteria and viruses such as *Hantavirus* spp., *Salmonella, Staphylococcus aureus*, *Enterococcus* spp., *Pseudomonas* *aeruginosa, Klebsiella pneumoniae, Escherichia coli, Serratia* sp., *Proteus* sp. *Toxoplasma gondii*, etc. In comparison with rodents, less attention has been paid to for the potential of arthropod pests to host and transmit bacteria, but recent studies have documented the potential (Zurek and Gorham 2010). The faeces-to-food is the main route of pathogen transmission by arthropods (Zurek and Gorham 2010). The microorganisms in arthropods may be embedded in the gut and disseminated via faeces, or transmitted from the body surface, as shown by an interaction between *Salmonella* and the lesser mealworm *Alphitobius diaperinus* (Crippen et al. 2012; Zheng et al. 2012). Beetles can carry enterococci that have genes for antibiotic resistance (Yezerski et al. 2005). For example, *T. castaneum* can acquire antibiotic-resistant enterococci from animal feed and transfer them to uninfested feed (Channaiah et al. 2010). More emphasis should be placed on research to identify the communities of bacteria in the gut of stored product arthropods, and determine their role in pathogen transmission.

Pest Temperature and Humidity Changes and Physical Grain Destruction and Treatments: Facilitation of Fungal Growth and Mycotoxin Production

Physical grain destruction and grain tunnelling by primary pest. Primary moth and beetle pests are able to attack and feed on intact grain, which disrupts the grain layers and enables entrance and growth of fungi. Consequent fragmentation of the grain provides new substrates for fungal growth and other fungal vectors (e.g. mites, psocids) that disseminate the fungi on their bodies and faeces.

Pest heat and humidity production in stored commodities. Cotton et al. (1960) were among the first scientists who experimentally showed that uncontrolled storage pest population increase in stored grains led to increases in temperature and humidity, which later became known as ‘hot spots’. In the stored grain mass, water and heat are produced chemically through respiration by the pest populations (Fleurat-Lessard 2002). Adler (2013) described a laboratory experiment demonstrating that within a month, an initial population of 20 *Sitophilus granarius*, in 200 g of triticale increased moisture content to levels favouring microbial development which can lead to grain deterioration. Fourar-Belaifa et al. (2011) found that when the insect density exceeds 1000 insects per kg, a rapid increase in grain moisture occurs, and after 160 when insect populations reached 2000–3000 insects per kg moisture content increased by 2–5%. Dharmaputra et al. (1994) observed similar results in dense populations of *S. zeamais* in corn. The use of grain protectants may also cause increases in moisture content. Vasquez-Castro et al. (2008) evaluated the effect of spray volume on the moisture content of corn and wheat grains, and showed 0.8% increase related to initial values. Although the grain moisture should increase minimally when grain protectants are used according to the label, there is some level of risk especially if an incorrect amount of water volume is used.

Poisoning by Toxic Insecticides and Rodenticides

Many stores, farms and food industry facilities rely on the use of pesticides, which if applied according to label directions impose minimal risks. But, if label directions are not followed, workers are at risk from acute or chronic health risks, or even death. The fumigant phosphine is very toxic and there are occupational health issues in developing countries resulting from incorrect usage or mis-application. Language barriers are present difficulties for users in developing countries (Eddleston et al. 2002).

Spray admixtures and risk of pesticide residues in food. Regular official controls on pesticide residues in food commodities are practiced in most countries to ensure compliance with legal limits (Maximum Residue Levels—MRLs). Data from the European Food Safety Authority (EFSA 2015) by European Food Safety Authority (EFSA) for Europe showed that 97.4% of the tested food samples fell within the legal limits and 54.6% of the samples contained no quantifiable residues. In general, higher prevalence of residues exceeding the maximum residue levels (MRL) was seen in imported products (5.7%) versus domestic products (1.4%) for domestic products. The risk of pesticide residues in stored grain accounts for little overall residue concerns since there is a limited number of insecticides worldwide that can be used on stored grains. In the EU, deltamethrin and pirimiphos-methyl are the most commonly used grain protectants. When protectants are used in stored grain, the residues are expected to give control during the storage period (Arthur 1994). Degradation of organophosphates increases as temperature increases, while pyrethroids are more stable (Arthur et al. 1992, 1994). However, there are exceptions, the half-live of pirimiphos-methyl was 23.9–28.9 d, and that of deltamethrin was 23.9–24.8 d in stored rice in Beijing and Jiangsu in China (Yu et al. 2014). Protectant residues usually decline as the commodity is processed Fleurat-Lessard et al. (2007), however different types of food products, for example baby food and adult food, may have differing standards for residues (Balinova et al. 2007).

Oil-seeds cross-contamination by insecticides used on cereal grain. There are risks of cross-contamination of grain products, particularly during transport (Dauguet 2007, 2009). This hazard rarely results from direct use of inappropriate pesticide for the particular commodity but rather from: (i) treatment of cereals at their receipt during the same period as of another crop; (ii) storage in bins previously containing insecticide-treated cereals; and (iii) loading commodity into empty bins using the handling equipment treated by insecticide before the receipt of the alternate crop (Dauguet et al. 2010, 2011).

Inert dusts and risk of pulmonary silicosis. Inert dusts (e.g. diatomaceous earth, silica) are used as an alternative to organic chemicals. They absorb lipids from the waxy outer layer of insect integument and are also abrasive. Similarly, zeolite dusts represent a broad range of microporous, crystalline aluminosilicates of natural or synthetic origin that are applied as particle films (De Smedt et al. 2015). These dusts do not leave any organic residues in the stored commodities but if they are used improperly, the inhalation of dust particles poses serious health risks, because crystalline silica dust may enter terminal respiratory pathways (Heyder et al. 1986) and cause silicosis and consequently silica dust-associated tuberculosis. The association of silicosis and silico-tuberculosis with diatomaceous earth dust has been recognized for decades (Vigliani and Mottura 1948). Furthermore, silica has been classified as a potential human carcinogen (class 1—IRAC) (Sherson 2002). Silicosis is an occupational illness reported in silica mill workers, stone workers and in the mining industry. No published epidemiologic data are available regarding grain dust exposure causing toxic effects on workers, but silicosis in flour mill workers has been reported (Athavale et al. (2011). This author also explained that in India, flour production is predominantly performed on a small scale, where milling is accomplished by grinding grain between stones and a steel wheel. In order to maintain the rough surface texture of flour, 'Agra' (80% silica content) and the buff stone (81% silica) for grinding and regular chiselling is used, and worker exposure to silica dust is common (Athavale et al. 2011). Nevertheless, silicosis is generally considered to be a preventable occupational disease and usually develops through years of exposure to crystalline silica-containing dust.

Fumigation insecticides and respiration poisonings. The acute or chronic toxic effects of fumigants may result in a variety of illnesses (Mehler et al. 1992; Burgess et al. 2000). For example, chromosome rearrangements were found in phosphine applicators and epidemiological studies of similarly exposed workers indicated increased occurrences of non-Hodgkin's lymphoma (Garry et al. 1992). The extensive usage of methyl bromide in the past for phyto-quarantine of stored commodities, soil and flour mill fumigations is responsible for more than 300 cases of poisoning reported in the literature (Alexeeff and Kilgore 1983). Langard et al. (1996) described a fatal accident resulting from methyl bromide leakage through sewage pipes. Deschamps and Turpin (1996) described methyl bromide intoxication during grain store fumigation in two fumigation workers, because they entered a building when the concentration of methyl bromide exceeded the labelled rate. Historically, various formulations of hydrogen cyanide gas were used for fumigation of mills, grain store warehouses and ships (Sherrard 1928; Williams 1931). It was documented by LeMay (2015) that during 1919–1922 several crew members were intoxicated and some of them died during fumigation of a ship with sodium cyanide. Although no intoxication related to the stores and mills data are available for the fumigant sulfuryl fluoride, Calvert et al. (1998) provided a report on health effects based on a study of fumigant applicators. Contrary to the universal belief that risks associated with phosphine fumigation are low because of the slow release from tablets or pellets, the rate of phosphine poising at a global scale is still very high because of malpractice, misuse or accidents. Also, bystander risk is a concern. A study from Germany (Lauterbach et al. 2005) summarized and elucidated details of phosphide poisonings based on a 20-year data collection (1983–2003). Out of the total 188 cases, 65% were unintentional residential, 28% attempts of suicide commitments (intentional), 5% occupational and 2% undetermined. In the majority of the intentional intoxication cases with PH_3, the poison was ingested, whereas the inhalation exposure dominated in unintentional poisonings. The broader phosphide availability and its wider use in Asia increase risks in comparison with Europe or the USA. Bumbrah et al. (2012) warned recently that there was an increase in the number of phosphide poisoning cases and deaths caused by suicidal ingestion and he added that due to broad-spectrum applications it cannot be ignored. A retrospective analysis (Gupta et al. 2003) of the child poisoning (1999–2002) recorded by the National Poisons Information Centre in India (NPIC) showed a total of 2720 poisonings; for 9.1% were responsible agricultural pesticides. Among them aluminium phosphide was the most common, followed by organochlorines and organophosphates (Gupta et al. 2003). Gargi et al. (2006) evaluated data of poisonings recorded in Amritsar, India (1997–1998) and found that majority (76.5%) of the victims of intoxications were suicide commitments, in 20.9% cases accidents and in 1.8% cases homicide was reported. The most common poison was aluminium phosphide (38%) followed by organophosphorus compounds (17.6%). The dangerous exposure might occur during grain fumigation (Jones et al. 1964; Zaebst et al. 1988). NIOSH (1999) described several cases of phosphine poisonings during fumigation of grain or during rodent pest fumigations in the USA. In 1992, the intoxication of the family followed by the death of one child caused by phosphine was reported also in the Czech Republic (Lastovickova 1992). The recent reports suspect that in Thailand agro-pesticide phosphine has been used for control of bed bugs in hotels leading to fatal incidents of foreign tourists (Blum 2014).

Chemical inhalation exposure from freight containers—an emerging risk. Freight containers are the main tools for transportation of commodities and materials around the globe. The containers may be partly hermetically sealed which contributes to higher efficacy of pest-related fumigations but also for retention and accumulation of various vapours and fumigant gases. This creates a risk to workers to inhalation of multiple toxic chemicals during transport or particularly during inspection and manipulation of freight containers in ports (Spijkerboer et al. 2008; Preisser et al. 2011, 2012; Svedberg and Johanson 2013; Kloth et al. 2014; Baur et al. 2015). For example, Baur et al. (2010a, b) published data of freight containers study performed in the second largest EU port terminal in Hamburg, Germany in 2006. The authors investigated 2113 freight containers arriving to Hamburg over a 10-week period. They determined that 1478 (70%) containers were contaminated with toxic chemicals above chronic reference exposure levels; 761 (36%) even exceeded the higher acute reference exposure level thresholds. Similar results were obtained in studies performed in Italy (Tortarolo 2011) and Australia (Wagstaffe et al. 2012). The New Zealand Customs Service carried out an extensive study concerning 9 fumigant or volatile chemicals (benzene, chloropicrin, ethylene dibromide, ethylene oxide, formaldehyde, hydrogen cyanide, methyl bromide, phosphine, and toluene) risks associated with import containers in 2011 at the Port of Tauranga (Service 2012). This study found that at least 1 of the 9 selected fumigants was present in 89.7% of the air samples collected and over 18% of the air samples were found to be above the safe reporting level (some concentrations were found to be 100 times excessive to legal limits.

Exposure to slow-release evaporation insecticides (e.g. DDVP strips). DDVP (=dichlorvos, an organophosphate insecticide) evaporation formulations are based on either solid plastic or resin matrix formed as pellets or strips from which DDVP continually evaporates into an enclosed space (Bengston 1976). In some countries, the slow-release DDVP strips are still allowed for protection of homes, farms and storages—including bulk storage of raw grains. In a grain storage structure, the DDVP strips can be hung in the headspace. However, strips must not be used in any area where people will be present for extended periods of time. There is a serious international debate on the health risks of DDVP use. A total of 31 cases of acute DDVP pest strip-related illnesses have been reported in the USA and Canada (Tsai et al. 2014). A majority of these illnesses were found to have resulted from the use of the product in violation of label directions.

Rodenticide ingestion and/or inhalation. Two types of rodenticides either with acute or chronic toxicity are currently used in farms, stores and food production facilities. The rodenticides with acute toxicity are most dangerous for human intoxication. They are represented by zinc phosphide (Zn_3P_2) that after ingestion rapidly releases toxic phosphine (PH_3) gas that immediately results in intoxication with high death rate. For example, Chugh et al. (1998) described that out of 20 zinc phosphide poisonings cases recorded in Asia, 5 patients died. The rodenticides with chronic toxicity are mainly anticoagulants (vitamin K mimetics), having high chronic but low acute toxicity. The human death rate caused by anticoagulants is generally low also due to the availability of efficient treatment of long-term oral 'antidote' vitamin K administration. The US poison surveillance system reported more than 16,000 cases of intoxication by anticoagulant rodenticides. Most of these intoxications were not associated with improper pests control interventions but with suicidal attempts or with accidental ingestions by children (Bronstein et al. 2009). However, the endangered groups are those pest-control professionals or farmers who administer anticoagulants regularly in high doses or take medically prescribed anticoagulants. Svendsen et al. (2002) reported a case of coagulation derangement in a pest-control operator applying rodenticides 3 h a week using gloves, but without a mask and not washing his hands between applications. He developed a symptomatic coagulation disturbance, probably caused by absorption of the poisons during work. In addition, anticoagulants pose an environmental risk if used around stores and food facilities that can cause secondary poisoning of wildlife after ingestion of rodents intoxicated by anticoagulants [e.g. Newton et al. (1990)].

Negative Impact of Anoxic Atmospheres and Toxic Fermentation Silo Gases

Anoxic, modified or controlled atmospheres (CO_2; N_2,). Modified and controlled atmospheres are anoxic atmospheres based on the elevated concentration of inert gases, such as carbon dioxide (CO_2) or nitrogen (N_2), and decreased oxygen (O_2) concentration that are present in the silos where commodities are stored (Navarro 2006). Exposure to toxic-modified atmosphere will kill pests but is also toxic to humans. Carbon dioxide CO_2 has been recognized as a significant workplace hazard for over 100 years through risks of asphyxiation (McGillivray and Wilday 2009). Harper (2011) claimed that to reduce the oxygen concentration in air down to a level immediately dangerous to life the CO_2 concentration would need to be at least 50% v/v. However, CO_2 might create an immediate threat to life at a concentration of only 15% in air due to the toxicological impact it has on the body when inhaled at this concentration (Harper 2011). It is believed that nitrogen atmospheres containing less than 14% oxygen or more than 5% CO_2 may be dangerous to human life. The hidden danger rests in the fact that inert gases do not contain any smells and the physiological states of sleepiness and unconsciousness may come after exposure very quickly without any warning signs. Personnel entering a nitrogen atmosphere containing less than 10% oxygen may collapse without warning and become unconscious; light-headedness and nausea, and unconsciousness may occur in less than 5 min in 9% CO_2 (Bond 1984). If the person is not immediately removed to fresh air it dies due to intoxication and suffocation. Carbon dioxide may furthermore be dangerous after its release/ ventilation at the end of the treatment. It may accumulate at the floor level and in the lower part of technology since it is heavier than air. In these areas, carbon dioxide endangers all of the personnel not equipped with self-breathing apparatus. This effect is well known from the famous Italian Cave of dogs near Naples (Grotta del Cane: Phlegraean Fields at Pozzuoli), where volcanic fumarole releases carbon dioxide that tends to accumulate in the deeper parts of the cave being particularly dangerous to small animals like a dog.

Silo gases (e.g. NO_2; CO)—a hidden danger. Silo gases, such as nitrogen dioxide (NO_2), are formed by the natural fermentation of chopped silage or by spoiled grain in the silo. These gases might be risky for the silo staff and pest-control operators during pest sampling/monitoring, cleaning or application of pesticides (Murphy 2015). Another risky gas that might be generated by stored grain in silo and other enclosed stores is carbon monoxide (CO) (Whittle et al. 1994).

Human Accidents During Sampling or Pesticide Application in Stores and Silos

There are multiple hazards associated with grain storage (Dennis 1973). An analysis of 236 fatalities involving workers handling grain in bulk storages between 1985 and 1989 showed that 21% of the deaths were caused by suffocation, 12% by entanglement in the auger (6 workers/year), 11% by a fall from an elevation and 9% by electrocution (Snyder et al. 1992). Monitoring of storage pests is done through a repeated sampling of the grain surface using spears and/or traps, which requires physical contact with the grain surface. When fumigations are done using phosphine-generating tablets, it is necessary to walk on the grain surface to dispense those tablets. Thus, personnel are prone to the entrapment by commodity (i.e.

victims are partially submerged but cannot remove themselves) or engulfment in it (i.e. engulfment occurs when victims are completely buried within the grain) (Wagner 2014). If there is no iron safety-mash on the top of the upright silos, workers may accidentally fall into the silo while taking samples for pest monitoring or while applying phosphine tables from the top of the silo during grain loading.

Fires, Explosion and Intoxication Due to Fumigation Control of Storage Pests

Phosphine may spontaneously ignite at concentrations above a safety threshold or in the presence of water or high moisture. O'Malley et al. (2013) described a case where a fumigation stack containing aluminium phosphide became soaked with rainwater and caught fire at a pistachio processing plant in California (USA). The personnel were exposed to pyrolysis byproducts, particulates and extinguisher ingredients. Six of the 10 hospitalized workers suffered from respiratory distress. NIOSH (1999) published two case studies of phosphine fumigation-related accidents in the USA. In the first, case 2 explosions from 62 trays of aluminium phosphide occurred during disposing of unused material. The second case was caused by an accidental explosion after placement of phosphide pellets under a tarpaulin. Several cases of phosphine-elicited explosions caused in connection with the transit of fumigated commodities on ships were also reported. For example, the phosphine fumigation-related explosion—caused by high humidity on bulk carrier ship Theofylaktos anchored at Rio Grande—happened in December 2012 (MSIU 2013). On the Safety4SEA portal, information about explosions occurred on the board of bulk carrier ships were reported caused by the improper distribution of fumigant pellets on the top of the cargo instead of applying tablets subsurface across the entire cargo (Safety4SEA 2017). Another case connected with fire and explosion concerning accumulated disposal of phosphide residues after ship cargo fumigation was published by (Club 2014).

The additional risk connected to storage pest management poses structural fumigation and hermetic isolation of the building or store. Furthermore, in case of malpractice or technical accident of pipelines distributing heating and cooking toxic gas may be physically damaged. Gas is then released and accumulated and the explosion risk connected to accidental open fire use that is illegal during all fumigating procedures or sparking or glittering light following with explosion might occur (CNN 2002).

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Chapter 3
Insect Pest Management in Stored Grain

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Introduction

Once cereal grain is harvested and put into storage, it provides a resource for a range of insect pests of stored grain. With the exceptions of *Sitophilus zeamais* Motschulsky (the maize weevil) (Giles and Ashman 1971), *Prostephanus trunctatus* (Horn) (the larger grain borer) (Tigar et al. 1994), *Sitotroga cerealella* (Olivier) (Lepidoptera: Gelechiidae) (the Angoumois grain moth) (Trematerra 2015), these insects rarely attack grain in the field before harvest, but once grain is in storage there is a degree of inevitability that insect infestation will occur. Insects can be carried into storage via infested harvesters or other machinery (Sinclair and White 1980), and infested grain can be moved from storage to storage during the postharvest

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handling of grain (Perez-Mendoza et al. 2004). Even without human help, field studies have demonstrated the importance of flight in some species, and so flying insects represent another source of infestation (e.g. Mahroof et al. 2010; Ridley et al. 2011a, b). This means that methods are needed to either disinfest grain or to protect it from infestation during storage. The aim of this chapter is to review recent advances in insect pest management in stored grain, focussing primarily on research published in the last 10 years on chemical and non-chemical methods, ranging from methods that are well established to those that are still being evaluated. As many papers have been published on this broad topic, sometimes we cite only a few papers from the many published to illustrate particular recent advances.

Disinfestation

Insecticides

For many years, the dichlorvos was available for grain disinfestation, but this organophosphate (OP) is being phased out because of concerns about its chronic health effects. Dichlorvos had the advantage over other OPs used on stored grain of having considerable vapour action (Desmarchelier and Banks 1977), so it was a useful treatment in situations where insufficient gas tightness was achievable for fumigation to be effective, though resistance in *Rhyzopertha dominica* (F.) (the lesser grain borer) has long been a concern (Zettler and Cuperus 1990). Dichlorvos served a valuable purpose, and the loss of this insecticide has created a need for a rapid disinfestant for situations where fumigation is not feasible because of poor gas tightness or resistance to phosphine.

Fumigation

Phosphine and methyl bromide continue to be the main fumigants used in stored products after many decades of use and despite issues facing each. For phosphine, there has been a continued development of resistance in a range of species, and much of the recent research on phosphine has been about understanding the impact of resistance and providing a basis for improved fumigation ensuring control of resistant types. In the case of methyl bromide, the restricted access to this fumigant has stimulated the search for alternatives (Bell 2000), some of which have been registered for use on stored grain.

Although phosphine resistance was detected in many countries around the world in the 1970s, it was relatively uncommon at that time (Champ and Dyte 1976), but studies since then have shown that the problem is increasing. This is demonstrated by the fact that populations with resistant individuals can be common (e.g. Opit et al. 2012; Daglish et al. 2014, 2015), and that newer or stronger types of resistance are emerging (e.g. Lorini et al. 2007; Pimentel et al. 2009; Jagadeesan et al. 2012; Opit et al. 2012; Nayak et al. 2013). Obviously, the detection of phosphine resistance raises concerns about its impact on fumigations, so, many recent papers have investigated the practical significance of phosphine resistance. Daglish et al. (2002) fumigated mixed-age populations of *Sitophilus oyzae* (L.) (the rice weevil) to determine times to population extinction at constant concentrations. An important finding was that the expression of resistance (i.e. time to population extinction) depended on the concentration used. They tested a susceptible strain, a weakly resistant strain and a strongly resistant strain, and found that differences between strains were greatest at low concentrations and negligible at high concentrations such as 1.5 mg L^{-1} (≈1000 ppm). For at least one species, therefore, resistant insects would be phenotypically susceptible in short fumigations with sufficiently high concentrations. To a lesser extent, this was also the case with mixed-age populations of susceptible, weakly resistant and strongly resistant *R. dominica* (Lorini et al. 2007). In recent years, researchers have also investigated times to population extinction in psocids of the species *Liposcelis bostrychophila* Badonnel (Nayak and Collins 2008) and *Cryptolestes ferrugineus* (Stephens) (the rusty grain beetle) (Kaur and Nayak 2015). By far the greatest threat is strongly phosphine resistant *C. ferrugineus*, with an estimated time to population extinction of 10 days at 2 mg L^{-1} (≈1400 ppm) and 25 °C. Nayak et al. (2003) demonstrated the unique ability of the eggs of the psocid *Liposcelis bostrychophila* Badonnel to delay its development under phosphine fumigation as a way to survive a fumigation and emerge to adults, several days after the end of fumigation. These authors have recommended that to control these psocids using phosphine, a relatively low concentration of phosphine should be applied for extended exposure times (e.g. 0.05 mg L^{-1} for 16 days) that should allow all eggs to hatch to the much less tolerant nymph stage.

Several studies have been published recently on phosphine fumigation trials and these have had a focus on controlling resistant insects. Rajendran and Muralidharan (2001) carried out trials on bagged paddy rice stored outdoors and inside warehouses in southern India. There was considerable loss of phosphine from the sheeted bag stacks, despite using new sheets and ensuring that sheets were weighed down with sand snakes, and this was particularly so for the outdoor stacks which had lost phosphine at about twice the rate that indoor stacks did. The authors attributed the loss of phosphine to several factors including sorption by the paddy rice, and permeation and leakage through the fumigation covers, and they suggested that the registered application rate of 3 g t^{-1} of phosphine (released from aluminium phosphide) was probably insufficient to control resistant populations. In another paper, Wang et al. (2006) reported the results of a Chinese study on phosphine fumigation of paddy rice in sheeted indoor bag stacks. The application rate was 1.67 g m^{-3} of phosphine (released from aluminium phosphide) based on the enclosed bag stack volume but the mean maximum concentration was about 50% lower, and the authors attributed this mainly to sorption. Although all natural and caged insects were dead after 21 days of fumigation, the authors raised the possibility that resistant pupae might have survived had they been tested. Recently, Ridley et al. (2011a, b) reported Australian trials of phosphine fumigation of wheat stored in silo bags fumigated with an application rate of 2.1 g t^{-1} of phosphine (released from aluminium phosphide). The fumigations were judged to be effective in controlling strongly phosphine resistant *R. dominica* based on examination of cages containing all developmental stages after the 17-day fumigation period. Carpaneto et al. (2016) conducted silo bag trials in Argentina showing that silo bags were often insufficiently gas-tight for phosphine fumigation and made a recommendation for improving gas retention.

Phosphine sorption by grain has been suggested to play a role in the loss of phosphine observed during fumigation field studies (e.g. Rajendran and Muralidharan 2001; Wang et al. 2006; Ridley et al. 2011a, b), and several recent laboratory studies provide some insights into the practical impact of this process. By fumigating a range of cereal grains, Reddy et al. (2007) showed that sorption reduced the likelihood of target terminal concentrations being achieved. Daglish and Pavic (2008) investigated effect of dose (based on the volumetric capacity of fumigation vessel) on sorption in wheat. They found that the daily percentage decline in gaseous phosphine was negatively correlated with dose, and that re-fumigated wheat was less sorptive. In a subsequent study, they showed that wheat was less sorptive the longer it was stored before being fumigated for the first time (Daglish and Pavic 2009). The sorption was faster at 25 °C than 15 °C, and the decline in sorptive capacity was greater when grain was stored at 25 °C than at 15 °C. They confirmed that re-fumigated wheat tended to be less sorptive, but they concluded that re-fumigated wheat was less sorptive because it had been in storage longer. The practical implications of these studies are that the impact of sorption on phosphine fumigations will be greatest in low dose fumigations or fumigations of freshly harvested grain.

Methyl bromide has received very little research attention in recent years, and this may be partly because of the focus on reducing its use because of its status as an ozone-depleting substance. One exception is the work of Athanassiou et al. (2015) investigating methyl bromide efficacy under laboratory conditions against five psocid species: *Liposcelis paeta* Pearman, *L. entomophila* (Enderlein), *L. decolor* (Pearman), *L. bostrychophila* and *Lepinotus reticulatus* Enderlein. They showed that eggs were more tolerant than mobile stages and identified concentrations of methyl bromide needed to control psocids in 48 h fumigations in the absence of significant amounts of grain. Further laboratory or field investigation of methyl bromide efficacy in the presence of grain would be valuable because sorption by grain can greatly reduce the fumigant concentration (e.g. Cherif et al. 1985).

Sulfuryl fluoride, although initially used for fumigating mills and other structures to control stored product insects, is now being used to fumigate grain. Recent research has focused on control of insects and sorption in laboratory studies. Although some laboratory efficacy studies were conducted with structural fumigation in mind, the results are useful with regard to sulfuryl fluoride fumigation of grain. Bell and Savvidou (1999) estimated combinations of concentration and exposure period needed to control eggs of *Ephestia kuehniella* Zeller (the Mediterranean flour moth) and Baltaci et al. (2009) did the same for eggs and other stages of *E. elutella* (Hübner) (the warehouse moth). They found that tolerance to sulfuryl fluoride varied greatly with egg age and that sulfuryl fluoride was less effective at lower temperatures. Athanassiou et al. (2012) investigated sulfuryl fluoride efficacy against five psocids (*L. paeta*, *L. entomophila*, *L. decolor*, *L. bostrychophila* and *L. reticulatus*) and showed that eggs were more tolerant than nymphs or adults. Jagadeesan et al. (2015) compared the responses of susceptible and strongly phosphine resistant strains of *Tribolium castaneum* (Herbst) (the red flour beetle) to sulfuryl fluoride. They found that eggs were the most tolerant stage and there was no cross-resistance between phosphine and sulfuryl fluoride, indicating the potential for the latter in managing phosphine resistance. Studies like these help build a picture of sulfuryl fluoride efficacy against stored grain insects but carefully monitored field trials are needed. Although many field trials have been conducted evaluating the efficacy structural fumigations (e.g. Campbell et al. 2010) they do not provide data helpful to understanding sulfuryl fluoride efficacy against insects in grain fumigations. Opit et al. (2016) conducted trials in the USA in small metal bins containing wheat and assessed efficacy against *R. dominica* and *T. castaneum* in the wheat or in muslin bags in the wheat. The target dose was a concentration x time product of 1500 mg-h L^{-1} and the fumigations lasted up to 1 day. High levels of control were achieved against both species but sulfuryl fluoride was more effective against *R. dominica* (complete control) than *T. castaneum* (some survival). Nayak et al. (2016) conducted trials in Australia in bunkers (sheeted grain piles) and a concrete silo, and assessed efficacy against natural infestations and caged containing mixed-age populations. The target dose was a concentration x time product of 1500 mg-h L^{-1} and the fumigations lasted up to 14 days. Complete control was achieved of natural infestations and caged populations. The species controlled included *R. dominica*, *T. castaneum*, *C. ferrugineus* and *S. oryzae*; although not all species were present in all fumigation trials.

Two recent studies investigated the potential role of sulfuryl fluoride sorption by grain. Sriranjini and Rajendran (2008) fumigated a range of grain types with sulfuryl fluoride in the laboratory and showed that sorption reduced the amount of fumigant gas in all cases. Subsequently, experimentation on wheat showed that sulfuryl fluoride is sorbed faster than phosphine but slower than methyl bromide, and temperature is a major factor affecting sorption with faster sorption occurring at higher temperature (Hwaidi et al. 2015). These studies show that, as with other fumigants, sorption by grain can be expected to contribute to gas loss during commercial fumigations.

Two other fumigants that have progressed from research to registration for grain are ethyl formate and carbonyl sulphide. In the case of ethyl formate, recent research comprised Australian research on ethyl formate applied as a liquid and ethyl formate applied as a vapour with carbon dioxide. Caged insects were inserted into the grain to assess fumigant efficacy. Trials in farm bins showed that ethyl formate applied as a liquid had the potential for use on stored grain (Ren and Mahon 2006). In that study, ethyl formate was applied in two stages to avoid the problem of rapid sorption reducing exposure of insect to lethal concentrations. Complete control of key pests such as *Sitophilus* and *Tribolium* species was achieved. Laboratory research was also conducted on combining ethyl formate with carbon dioxide to improve penetration through the grain mass, reduce sorption, increase efficacy and reduce flammability (Haritos et al. 2006; Damcevski et al. 2009; Dojchinov et al. 2009). Ren et al. (2008) demonstrated the efficacy of carbonyl sulphide applied as a liquid to wheat in a large concrete silo in Australia, with complete control possible of caged of mixed-age populations of the key pests *R. dominica*, *T. castaneum* and *S. oryzae*. Research on ethyl formate and carbonyl sulphide has led to the registration of cylinderised formulations of carbonyl sulphide and ethyl formate + carbon dioxide. To our knowledge, however, only the latter is available in the market.

Several recent field studies have focussed on determining the strengths and weakness of fumigating bulk grain with ozone gas. The major challenge in using ozone is to optimise its application so as to maximise its penetration through the grain bulk so that high levels of mortality can be achieved before it degrades into oxygen. A pilot study using recirculation showed the rapid decay of ozone as it passed through bulk wheat highlighting a practical challenge (Hardin et al. 2010). Kells et al. (2001) evaluated ozone against caged adults in maize at doses of up to 50 ppm and 5 days of exposure. They found that efficacy varied across species and complete mortality was difficult to achieve. *Tribolium castaneum*, for example, was harder to control than *S. zeamais*. Similarly, Bonjour et al. (2011) evaluated ozone against bags of insects in wheat at doses of up to 70 ppm and 4 days of exposure. They found that efficacy varied across species. *Rhyzopertha dominica*, *C. ferrugineus* and *O. surinamensis*, for example, were harder to control than *S. oryzae*. These studies show that ozone has potential but that complete control of all species may not be possible.

Other Methods

Considerable research has been undertaken on heat disinfestation of stored products. The concept of the application of heat treatment is simple: temperature is increased until it reaches a lethal level for insects; this level is considered to be 50 °C (Mahroof et al. 2003a, b; Yu et al. 2011). Nevertheless, reaching this lethal threshold is not always easy, as different structures have different physicochemical properties that are expected to negatively influence the overall efficacy of the application (Yu et al. 2011). Quantification of the responses of different species and developmental stages within species to flameless catalytic infrared radiation has been the focus of several recent studies (Khamis et al. 2010, 2011a, b), and another study suggests that there are no negative effects of this treatment on grain (Khamis et al. 2010, 2011c). Elevated temperatures have been also used tested successfully in combination with other methods, such as diatomaceous earths (Dowdy and Fields 2002), contact insecticides (Kljajic et al. 2009) or nitrogen (Athanassiou et al. 2016a).

Although there are numerous techniques to implement controlled or modified atmospheres, nitrogen seems to be the most promising agent for disinfestation of stored grain. Given that approximately 80% of the atmosphere contains nitrogen, usually there is no need to use gas in cylinders, but only nitrogen generators or pumps that can take the nitrogen from the air and introduce the gas in the area that is to be treated (Navarro et al. 2012a, b). The basis of this application is to reduce oxygen, usually to less than 1% (Adler et al. 2000; Athanassiou et al. 2016b). In practice, nitrogen can be applied either on commodities, in designated areas that are made for this purpose, i.e. nitrogen chambers, or on target facilities, e.g. a silo or a warehouse, either empty or with products (Navarro 2006, 2012; Navarro et al. 2012a). For some commodities, it is now known that the application of nitrogen does not affect some key organoleptic properties, while, under certain circumstances, can also reduce microbial load (Navarro 2012; Navarro et al. 2012b; Athanassiou et al. 2016b).

There are numerous other methods that have been utilised for disinfestation. One method that gains in importance is the use of pheromones for moth suppression through mating disruption. This concept has been successfully tested in storage and processing facilities in various types of commodities and facilities in Europe, and found effective in suppressing populations of *Ephestia* spp. or *Plodia interpunctella* (Hübner) (the Indian meal moth) (Pyralidae). Currently, there are several formulations that are commercially available for moths (Trematerra et al. 2011, 2013; Athanassiou et al. 2016c), while there are recent efforts to develop the first formulation for beetles, using *Lasioderma serricorne* (F.) (the cigarette beetle) (Anobiidae) as a target species (Mahroof and Phillips 2014).

Apart from heat treatment, there is a renewed interest for the use of ‘cold treatments’. Insects are generally tolerant to cold, so care should be taken to calculate the target temperature and the required exposure interval that is needed to obtain complete control, without affecting the commodity (Fields 1992; Flinn et al. 2015). Generally, the most difficult to control life stage is the egg (Fields 1992; Johnson and Valero 2003).

Many papers have been published on the potential of essential oils extracted from a wide range of plant species, and the vast majority of these report preliminary screening. Rajendran and Sriranjini (2008) reviewed the literature on essential oils, highlighting their potential but also a number of constraints, including lack of data on sorption, tainting and residues in food. Research on allicin, one the components of garlic essential oil, is an example of research that that has gone beyond preliminary screening. Lu et al. (2013) investigated fumigant toxicity of allicin in the laboratory against different developmental stages of three insect species, showing that the presence of wheat reduced efficacy because of sorption. Laboratory studies like this one provide more practical information than preliminary screening studies, but studies on bulk grain stored under realistic are also needed.

Other disinfestation treatments such as carbon dioxide, changes in pressure, and micro-biocontrol agents, are briefly discussed in other chapters of this book.

Protection

Cooling

Temperature management is an important component for insect pest management in stored bulk grains, particularly in temperate regions of the world. Aeration is described as using ambient air to cool the grain mass to temperatures that will limit insect population growth. A common threshold is 15 °C, which is the lower limit of development for most stored product insects (Howe 1965; Fields 1992). Typical airflow rates used are 0.0515–0.31 m^3/min/metric tonne (t), depending on the specific commodity and harvest date (Reed and Arthur 2000; Navarro et al. 2012c). Aeration is not to be confused with grain drying, which uses airflow rates several orders of magnitude higher than those used for aeration, with the emphasis on drying grain to moisture contents required for long-term storage (Reed and Arthur 2000). Aeration does not necessarily eliminate an existing infestation, and the most optimal use of aeration involves usage with other components in a management plan.

There are several established methodologies for utilising aeration, and most involve using some type of controller to activate aeration fans thresholds (Arthur and Casada 2010). In Australia, aeration controllers were developed that used web-bulb temperatures for precise activation and optimization of moisture management (Wilson and Desmarchelier 1994). In the USA, more simple controllers are used that activate fans when temperatures fall below a specified threshold (Ranalli et al. 2002; Arthur et al. 2008; Arthur and Casada 2005, 2010). There are several recent papers that give a detailed review of aeration strategies, one, in particular, is Navarro et al. (2012c), which is a chapter in the book "Stored Product Management", available from the Kansas State University (www.k-state.edu). Other publications can be found that also give a more detailed review of aeration, along with numerous references, that readers can consult for more information (Navarro et al. 2012c).

There are several recent advances in utilising aeration for insect pest management, including historical weather data to predict hours available for cooling stored grains in different geographic regions (Arthur and Johnson 1995; Arthur and Siebenmorgen 2005). These studies examined the various hours necessary for cooling grains at different airflow rates at specified temperature thresholds, and predicted hours needed for cooling. Analysis was done by using a q-Basic code that used daily high and low temperatures, and sunrise and sunset data, to predict temperature each hour (Arthur and Johnson 1995), and was subsequently utilised for a number of successive studies based on the same procedures. These procedures can be adapted for any site in the world that has similar temperature and sunset data, and is an excelled method for examining the feasibility of aeration.

Analysis of weather data as described above can be incorporated into insect pest population models to predict the impact of aeration on population development (Arthur et al. 1998, 2001, 2011a, b; Arthur and Flinn 2000). It can also be used to predict a number of insect generations that could occur in a given time period (Arthur et al. 2003). Results of these model simulation studies show the benefits of using aeration, particularly in warm regions where winter temperatures are not normally low enough to cool the grain mass. In the USA, a web-based management system was developed for stored rough rice (also known as paddy rice) that includes aeration along with other management options (Arthur et al. 2011a, b). Although the recent modelling efforts have mainly been done in the USA, historical weather data are available worldwide, and can be utilised in a similar manner to incorporate aeration into management plans for bulk grains.

Another recent advance in aeration is using defined cycles to cool a grain mass, and incorporating an initial cooling cycle in advance of what can be accomplished in waiting for temperatures to cool so that the specified threshold of 15 °C can be achieved. Arthur and Casada (2005) conducted studies in experimental grain bins and showed that using a summer cooling cycle at 22 °C resulted in lower pest populations in stored wheat compared to waiting to use the standard first autumn cooling cycle of 15 °C. Another aspect for consideration is using suction aeration, which pulls the cool air down through the grain mass, rather than the standard pressure aeration, which pushes the cool air upward through the grain mass (Arthur and Casada 2010, 2017). Cooling the top portion resulted in lower temperatures, which in turn led to lower insect populations, as assessed through the use of probe traps (Arthur and Casada 2010, 2017). However, in the Arthur and Casada (2017) study, high infestation rates during the third year of a 3-year study mitigated the positive effects of aeration. One possible reason for those high infestations was the fact that the grain could have been infested when it was loaded into the bins for the third year of the study. Thus, aeration should be considered as a component of integrated management plans, not as a direct killing agent such as fumigation. It is unlikely that aeration alone will eliminate an existing infestation.

Grain Protectant Insecticides

Grain protectants continue to play a major role in protecting grain during storage. The scientific literature on grain protectants is extensive and can only be reviewed very selectively here. As with the fumigant phosphine, the continued development of resistance in a range of species has provided much of the impetus for research on protectants. Malathion resistance was already of concern in the 1970s (Champ and Dyte 1976) and other resistances have developed since then. Pyrethroid resistance has developed in *R. dominica* (Lorini and Galley 1999), *T. castaneum* (Collins 1990) and *S. zeamais* (Guedes et al. 1995); and methoprene resistance has developed in *R. dominica* (Daglish et al. 2013).

Despite extensive research on potential new grain protectants in recent years (see below), only one new grain protectant from a new chemical group has been registered and adopted. Spinosad was developed from the fermentation products of a bacterium found in soil (Salgado 1998), before being evaluated as a grain protectant. Since 2002, many papers have been published on the susceptibility of a wide range of stored grain insects to grain treated with spinosad, including strains that were resistant to other grain protectants (e.g. Fang et al. 2002a, b; Nayak et al. 2005; Vayias et al. 2009). These studies showed that *R. dominica* is much more susceptible than other major pest species, including strains of *R. dominica* that are resistant to other grain protectants. Other laboratory studies showed that spinosad remains active against this species for long periods of storage (Fang and Subramanyam 2006; Daglish and Nayak 2006). Long-term efficacy of spinosad on bulk-stored wheat was demonstrated in silo-scale studies in the USA and Australia, with wheat treated at an application rate of 1 mg kg^{-1} which ultimately became the registered rate for this protectant (Fang et al. 2002a, b; Flinn et al. 2004; Subramanyam et al. 2007; Daglish et al. 2008). Spinosad residues were shown to be relatively stable on stored wheat in field studies in the USA and Australia (Fang et al. 2002a, b; Subramanyam et al. 2007; Daglish et al. 2008), which matched the results from the laboratory (Daglish and Nayak 2006). Due to its widespread use by the Australian grain industry, a discriminatory dose of 1 mg kg^{-1} has been established to monitor for potential resistance development in *R. dominica*, based on an investigation of the base-line susceptibly of field populations of this species (Nayak and Daglish 2017).

A major and long-standing challenge with grain protectants is finding a single protectant or combination of protectants that will provide protection against the pest species of concern. One reason is that there can be large interspecific differences in susceptibility to grain protectants, and another is the development of resistance. This problem can be illustrated by a laboratory study from Australia looking at the efficacy of three grain protectants applied alone or in binary combinations against resistant strains of five pest species (Daglish 2008). The five species were *R. dominica*, *T. castaneum*, *C. ferrugineus*, *S. oryzae* and *Oryzaephilus surinamensis* (L.) (the saw-toothed grain beetle). The three grain protectants were chlorpyrifos-methyl (an organophosphorus compound), s-methoprene (a juvenile hormone analogue) and spinosad (a biopesticide). At the time of the study, chlorpyrifos-methyl and s-methoprene were registered in Australia and spinosad was under consideration. The most effective combinations were spinosad (1 mg kg^{-1}) + chlorpyrifos-methyl (10 mg kg^{-1}) which controlled all strains except for OP-resistant *O. surinamensis*, and chlorpyrifos-methyl (10 mg kg^{-1}) + s-methoprene (0.6 mg kg^{-1}) which controlled all strains except for methoprene-resistant *R. dominica*. The result is that spinosad is usually applied in Australia in combination with chlorpyrifos-methyl and s-methoprene. In another study, Nayak and Daglish (2007) have shown the advantage combined treatment of spinosad and chlorpyrifos-methyl to control four *Liposcelis* psocid species that have shown variable levels of resistance to a range of currently registered grain protectants in Australia.

There are several commercial formulations available that combine more than one substance. One of the oldest ones in the market is Storicide II, which contains the OP chlorpyrifos-methyl with the pyrethroid deltamethrin. This combination has been found to be effective against stored-product insects (e.g. poscids) that are tolerant to other substances (Athanassiou et al. 2009a). In that study, the authors noted that psocids were susceptible to this combination, and hypothesised that this was mostly due to the presence of chlorpyrifos-methyl in the mixture, as psocids were found also susceptible to pirimiphos-methyl.

There have been many studies published on potential new grain protectants from a range of chemical groups, with most of these studies undertaken in the laboratory. Spinetoram, a spinosyn-based insecticide, has been evaluated in both short-term and long-term laboratory studies (Vassilakos et al. 2012, 2015). Based on experience against insect pest of field crops, spinetoram was expected to have greater potency than spinosad but these studies show that spinetoram efficacy against stored grain beetles is broadly similar to that of spinosad. Despite the promising results for spinetoram, evaluation has not progressed to field trials. Other compounds that have been evaluated in the laboratory include ethiprole (a phenyl-pyrazole) (Arthur 2002), imidacloprid (a neonicotinoid) (Daglish and Nayak 2012), thiamethoxam (a neonicotinoid) (Arthur et al. 2004) and indoxacarb (an oxadiazine) (Daglish and Nayak 2012). In general, all tested compounds show potential in the laboratory at varying doses depending on the species tested, but none have progressed to field trials.

There are several combinations of two or more active ingredients that can be used as grain protectants. For example, a combination of diatomaceous earth with natural pyrethrum and piperonyl butoxide was found more effective and with higher ‘speed of kill’ than other formulations that were based on diatomaceous earth alone (Athanassiou et al. 2004; Athanassiou and Kavallieratos 2005). Tucker et al. (2015) found that the insect growth regulator methoprene could be used successfully with synergized pyrethrin aerosols for the control *T. castaneum*. Arthur (2002) tested different combinations of ethiprole with other insecticides with satisfactory results for wheat and maize. Liu et al. (2016) showed the feasibility of using aeration with methoprene against a wide range of insect species. In general, there are numerous studies that showed that certain combinations may have some benefits in comparison with the application of a single active ingredient, as the range of species that can be controlled can be expanded (Daglish 2008; Arthur 2012; Athanassiou et al. 2009a). In this context, currently, there are several commercial formulations available for admixture with the grains, which combine two active ingredients with different modes of action, such as one OP, either pirimiphos-methyl or chlorpyrifos-methyl, and another substance, usually a pyrethroid (e.g. deltamethrin or cypermethrin) or a neonicotinoid (thiamethoxam). These combinations can also be used in order to mitigate insect resistance to certain insecticides, as these populations will be exposed to substances that have different modes of action (Daglish 2008).

Field studies were completed in Australia on bifenthrin (a pyrethroid) (Daglish et al. 2003) and diflubenzuron (a chitin synthesis inhibitor) (Daglish and Wallbank 2005) providing data on efficacy against key pest species and stability of residues. Bifenthrin which targeted *R. dominica* was tested in combination with chlorpyrifos-methyl so that the combination treatment would give broad spectrum control. Bifenthrin was seen as a potential replacement for bioresmethrin which had been withdrawn from the Australian market, but the results showed that bifenthrin failed to control pyrethroid-resistant *R. dominica*. Diflubenzuron targeted *S. oryzae* and proved to be effective against this species. Surprisingly, the diflubenzuron + s-methroprene resistant combination controlled methoprene-resistant *R. dominica*. Neither bifenthrin nor diflubenzuron progressed to registration, despite the promising results.

Plant products have been widely evaluated for stored product protection, but registered products for this use are extremely few. Detailed lists of the plant species that have been evaluated so far are given by Prakash and Rao (1997) and Weaver and Subramanyam (2000). There are different ways that these compounds can be used, but the most common evaluations refer to their repulsive activity and their insecticidal effect. For example, Arthur et al. (2011a, b) reported that catmint oil could be utilised further as a repellent of *T. castaneum* and *T. confusum*. Neem oil, which is produced by the neem tree, *Azadirachta indica*, has also some repulsive activity to insects (Athanassiou et al. 2014) but most of the studies available are about its insecticidal value. The major active ingredient of neem, azadirachtin, is now registered as an insecticide on various crops. Athanassiou et al. (2005) found that azadirachtin was effective for the control of *S. oryzae*, *R. dominica* and *T. confusum*, but at dose rates that were much higher than the currently used grain protectants, which constitutes such an application unrealistic. Nevertheless, the future of plant extracts for stored product protection, especially for use as grain protectants, remains uncertain and should be regarded on the basis of additional research that goes far beyond their insecticidal value (Athanassiou et al. 2015). Hence, even if a plant extract is proved effective as an insecticide, there will be always additional requirements that are needed for registration purposes, such as toxicological and ecotoxicological data. In this effort, the economics of the development of such plant product-based formulations, i.e. its cost-effectiveness, should always be examined in more detail.

One other parameter that should be taken into account when grain protectants are applied is their uneven distribution. In fact, uneven distribution may lead to the occurrence of zones within the grain bulks that are under-dosed or even untreated areas, which allow insect colonisation and progeny production. Daglish and Nayak (2010) demonstrated that the efficacy of s-methoprene against *R. dominica* was negatively influenced by an uneven application on wheat. Moreover, in that study, the authors suggested that this phenomenon may be related with selection of resistance. Similar results for the uneven distribution have also been reported in the case of spinosad (Athanassiou et al. 2009b) and spinetoram (Vassilakos and Athanassiou 2012).

Conclusions

The aim of this chapter was to review recent advances in insect pest management in stored grain, ranging from methods that are well established to those that are still being evaluated. This topic has been the subject of considerable laboratory and field research as evidenced by the large and growing body of published studies. Resistance to phosphine and various insecticides, as well as the phase out of methyl bromide as an ozone-depleting substance, continue to be major drivers for research on management of insects in stored grain. Other research has focussed on improving basic understanding of various technologies or ways of improving methods currently in use. Despite extensive research on a wide variety of chemical and non-chemical treatments, very few have been commercialised. Two examples are spinosad which has been registered as a grain protectant and sulfuryl fluoride which is now available as a grain fumigant. The interest in non-chemical treatments, especially aeration cooling, is encouraging. In general, integrated pest management is seen as the goal of entomologists, requiring the strategic integration of multiple methods to provide maximum effect, with minimal health and environmental risks. Some of the chemical and non-chemical treatments reviewed in this chapter have great potential to be used as part of an integrated approach.

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Chapter 4
Structural Pest Management for Stored Product Insects

Frank H. Arthur

Introduction

Stored product insects represent a diverse group of species that can infest raw grains but also can infest structures associated with the milling, processing, storage, and distribution of finished grains and grain-based products. One of the recent developments in managing these insects is documenting the extensive presence of these insects in and around milling, processing, and warehouse facilities (Semaeo et al. 2013), which represents a new awareness of the infestation potential. Stored product insects cause damage through direct consumption of food products, through contamination due to the presence of insects, insect parts (fragments, hairs, cast skins, etc.), and can harbor allergens that are potentially by-products associated with infestations (Larsen 2008). Equipment and machinery can become infested, resident populations can persist in wall voids, floor cracks, and in hidden areas inside a structure. These infestations are often hidden and difficult to treat with insecticides. The mere presence of insects inside a structure can be a cause for concern. Although exact quantitative losses are difficult to determine, the loss through product rejections, consumer complaints, and potential legal actions resulting from infestations all contribute to economic losses that can be passed on to the consumer. Thus, the various storage industries encountered in the food distribution channel are aware of the importance of insect infestation, potential losses from those infestations, and of integrated management options for control. In this chapter, we will review recent advances in the various components associated with structural pest management of stored product insects, including insecticides,

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temperature manipulation, environmental controls, and insect-resistant packaging. The chapter is not meant to be an exhaustive list of control options, a complete review of control strategies that are discussed, or an extensive listing of references for each topic. The chapter is meant to guide readers to sources of information for further review, and will focus on the entomological aspects of stored product insects associated with storage of processed grain and grain-based products, topics regarding management of bulk grains in bins and elevators will be found in other chapters of this book. Selected research papers will be referenced, but not described in detail.

Pre-binning Treatments for Stored Grains

An empty grain bin or elevator silo can be considered as a structure, and managing that structure, and the area around the structure, is an important aspect of integrated control programs for stored grains. Grain spillage in and around bins and silos support resident populations of stored product insects, which can quickly infest newly-harvested grains loaded into those bins and silos (Reed et al. 2003; Arthur et al. 2006). While cleaning and removing old grain from a bin or silo is preferred, areas in the bottom portions of a bin, such as underneath the false flooring of a metal bin, are often difficult to clean out without removal of that floor. Fumigation is usually employed as a treatment for those areas, but another option could be some type of heat treatment for disinfestation. A recent series of studies showed the potential for using this strategy (Tilley et al. 2014, 2015), though further research is needed on some aspects, such as the economic advantage for using heat instead of fumigation (Tilley et al. 2007a). In the study evaluating different types of heaters, propane heaters were superior to electric heaters, both in terms of cost and insect control (Tilley et al. 2007b). A field trial in an empty elevator silo also showed potential of using heat, but results also showed how even small amounts of grain can provide insulation from lethal high temperatures (Opit et al. 2011).

Residual insecticides are often used to treat the flooring surface of a grain bin or silo prior to loading new grain into the bin. Residual surface treatments will be discussed later in the chapter, but it should be noted that there are insecticides that be used as a pre-binning treatment on the flooring surface, but not on the grain itself. An example in the United States (US) is the pyrethroid cyfluthrin (Tempo®). In contrast, the organophostate pirimiphos-methyl (Actellic®) is labeled for direct application to corn and sorghum that is to be stored in a bin, but not as a pre-binning flooring treatment. Persistence of a residual insecticide will vary depending on the flooring surface. For example, concrete is a porous surface, and insecticides are not as persistent on concrete compared to metal, which is a relatively non-porous surface (see citations in Arthur 2009). Persistence on any surface can also vary depending on the insecticide (Arthur 2009; Wijayaratne et al. 2012).

Fumigants

Phosphine is the predominant fumigant used world-wide to control insects in stored bulk grains, but is not extensively used as a structural treatment in the milling and processing industries due to the corrosive effects on electrical wiring (Bond et al. 1984). There are mixtures of phosphine with carbon dioxide, and other forms of phosphine, including cylinderized phosphine (Eco2Fume®) and pure phosphine produced through a generating system. These advances enable more usage of phosphine as a structural treatment outside of the traditional uses in stored grains. Perhaps the biggest issue with phosphine today is the concern regarding development of resistance in several important stored product insect species (Opit et al. 2012a, b; Nayak et al. 2013; Holloway et al. 2016; Gautam et al. 2016). Strategies being implemented to mitigate resistance development include resistance monitoring, extending the fumigation time to increase the CT (concentration × time) product, and using alternative strategies to decrease reliance on phosphine. Resistance development may be of more concern in bulk grains than as a structural treatment because of the different usage patterns.

Methyl bromide (MB) was historically a major component of insect pest management programs for the milling industries. It was identified as an ozone depleting agent, and an international agreement called the Montreal Protocol was signed by a number of developed and developing countries (Fields and White 2002). This agreement mandated the phase-out of MB starting in 2005 for developed countries, with provisions made for Continuing Use Exemptions (CUEs) for mills and processing plants until 2015 (Fields and White 2002; Baltaci 2009). MB is still allowed for quarantine and pre-shipment (QPS). Alternatives to MB were broadly defined by the Methyl Bromide Technical Options Committee (MBTOC) to include alternative fumigants, heat treatments, improved insect monitoring, integrated control strategies, and other insecticides, including the use of residual surface treatments and application of aerosol insecticides. In the US, the Annual International Research Conference on Methyl Bromide Alternatives and Emissions Reductions, provides a forum for International researchers to present results on alternatives to methyl bromide, and complete copies of the Proceedings of this conference is available from the website of the sponsoring organization, the Methyl Bromide Alternatives Outreach (www.mbao.org). Similarly, the Controlled Atmospheres and Fumigation Conference, held every four years, has a complete Proceedings, with full-length articles (www.caf.org). The International Working Conference on Stored Product Protection (IWCSPP), also held every four years, has a Proceedings with many papers on fumigation.

The fumigant sulfuryl fluoride (SF), under the trade name Profume®, has been introduced and labeled for use in many countries as an alternative to MB, including the US. It was originally registered by DOW AgroSciences, but in 2015 the marketing rights were sold to Douglas Products (www.douglasproducts.com, Liberty, MO, USA). Campbell et al. (2015a, b) conducted a meta-analysis of structural treatments in wheat and rice mills, using MB, SF, and heat as whole-plant treatments. Results showed that both MB and SF gave similar levels of control, with greater initial reductions in flour beetle populations in wheat mills compared to rice mills. This paper also gave a detailed analysis of the metrics associated with how the beetle populations were assessed using trap catch data.

Fumigants can penetrate into hidden areas inside structures, equipment, and packaged food products and eliminate existing pest infestations. Recent research has documented that while fumigants are effective, population re-colonization and rebound after treatment can affect overall results (Campbell et al. 2010a, b; Buckman et al. 2013; Campbell et al. 2015a, b). The recolonization aspect of fumigation treatments is a topic that has received little historical attention until recently. Since there can be extensive insect populations in and around milling, processing, and storage facilities (Arthur et al. 2015a, b, 2016), potential re-colonization after fumigation is a factor that should be considered in management plans, especially during the warmer months of the year.

Variation among insect species and life stages will also affect fumigation efficacy. There is a general consensus among researchers that the egg stage is the most difficult life stage to kill with any fumigant (Bell and Savidou 1999; Baltaci et al. 2009; Jagadeesan 2014). Monitoring and sampling studies with *Tribolium* species have shown that most of a resident population within a facility is in the immature stages, and there are also areas where mobile adults may be able to escape exposure, thus contributing to survival and potential population rebound (Campbell et al. 2010a, b). There are also concerns regarding toxicity of the different fumigants to the egg stage, however, multiple factors, including temperature, exposure time, and species variation contribute to fumigant efficacy, so precise comparisons of fumigants are sometimes difficult and conclusions can be erroneous if these other factors are not taken into account. The structure of eggs may also play a role in susceptibility to fumigants. The egg chorion can contain channels known as micropyles and aeropyles, which could enable greater penetration of fumigants compared to eggs with a more solid structure of the chorion, and can partially explain differences in susceptibility to phosphine on eggs among different species (Gautam et al. 2014).

Modified Atmospheres

Extensive historical research has been conducted using modified atmospheres to kill insect pests of stored products (Navarro et al. 2012). In general, modified atmospheres use a mixture of gases, which can include nitrogen and carbon dioxide, to reduce the oxygen content down to levels of around 1–2% (Navarro et al. 2012). Other recent developments include the use of ozone for disinfestation of bulk grains (McDonough et al. 2011; Jian et al. 2013; Isikber and Athanassiou 2015; Savi et al. 2015). Although modified atmospheres are an effective control strategy, the costs for sealing and application often limit their use for structural management of bins containing raw grains. When modified atmospheres are used in structural control inside mills and warehouses, it is often in conjunction with fumigations as more of an additive or supplemental control. Large bags or cocoons can be effectively utilized for hermetic of storage grains that are stored on the ground on in bulk bags (DeGroote et al. 2013; Moussa et al. 2014). Hermetic storage of cowpeas and other crops has proven to be effective in storing grains in sub-Saharan Africa using the Purdue Improved Cowpea Storage (PICS) bags (Njoroge et al. 2014; Sudini et al. 2015; Martin et al. 2015).

Heat and Cold

The use of heat as a control strategy for mills and warehouses was first mentioned by Dean (1911, 1913), however; there was comparatively little scientific research in the US until the late in the twentieth century (Subramanyam et al. 2011). Much of the recent research includes work on field applications of heat to control insects in flour mills (Brijwani et al. 2012a, b; Campolo et al. 2013), and also research to identify the life stages of stored product insects that are most tolerant to heat (Mahroof et al. 2006; Yu et al. 2011). Models have also been constructed to predict temperature/exposure times necessary to control life stages of different species (Boina et al. 2008; Yan et al. 2014).

In the USA, the use of cold is more restricted to small-scale applications within a facility, rather than as a whole-plant treatment (Johnson and Valero 2003; Arthur et al. 2015a, b). In other parts of the world where there is an extensive winter season, it may be possible to use cold as a whole-plant treatment. However, the effect of freezing temperatures on equipment has not been investigated. Also, the even distribution of the cold temperatures throughout the facility may be an issue, and supplemental air movement may be required. A recent review by Andreadis and Athanassiou (2017) discusses the cold tolerance of stored product insects and provides examples of different species.

Recent research has also focused on examining the susceptibility of different insect life stages to cold, similar to work done for heat. Life stages of individual species vary in susceptibility to cold; Arthur et al. (2016) examined susceptibility of *T. castaneum* and *T. inclusum*, and showed that the latter species was far more tolerant, particularly in the larval stage. Also, if cold is being considered as a disinfestation strategy, for example to eliminate infestations inside bagged processed food such as flour, penetration of cold will depend on a number of factors, including the bulk mass of the commodity. One study of distribution of cold temperature through a stack of palletized flour bags showed that the outer peripheral bags cooled much more quickly that the center bags, which also affected resulting efficacy on eggs of *T. castaneum* (Flinn et al. 2015).

Aerosols

The use of aerosols (also called fogging, space sprays, or ultra-low volume sprays), involves dispensing a liquid insecticide formulation through a mechanical device in the form of fog or mist (Peckman and Arthur 2006). These systems can either be installed in the headspace of a milling facility or portable systems brought into the facility. Aerosols do not penetrate through commodities, so they confer control by the deposition of particles generated by the application system onto surfaces (Arthur et al. 2014a, b). Aerosols are expected to disperse throughout the area where they are applied, and hence can be considered as structural treatments. The cost of aerosol applications is less compared to fumigation or heat treatment, and there is increasing interest in aerosols as a replacement for methyl bromide (Boina and Subramanyam 2012).

Common aerosols used as part of management programs in the US include dichlorvos and pyrethrins applied alone or with an insect growth regulator (IGR) (Arthur 2012). The IGR gives residual control of immature stages (Sutton et al. 2011; Arthur 2015a, b). Dichlorvos is an organophosphate insecticide, with excellent vapor toxicity and disperses well in structures, but is highly volatile and only gives short residual control of insects (Subramanyam et al. 2014). New label restrictions on the use of this product in the USA may limit applications to mechanical release from the outside. Pyrethrin is a natural insecticide produced by grinding dried flowers of certain species of the chrysanthemum plants. Synergized pyrethrins can be applied alone or with IGRs to provide increased residual activity (Sutton et al. 2011).

Most previous published research with aerosol and particle size focused on aerosols applied for mosquito control, which evaluated particle sizes in terms of the ability of aerosols to control flying mosquitoes (Bonds 2012). Optimum particle size estimates ranged from 5 to 30 μm, but there were no data translating this estimate to control of stored-product insects. Arthur et al. (2015a, b) conducted studies in a vertical-flow aerosol application chamber to examine differences between particles of 1% active ingredient [AI] dispensed at 2 μm versus 16 μm, using different post-exposure techniques and adults of *T. confusum* as the target species and life stage. The smaller particle size was largely ineffective even though in some trials the actual concentration of insecticide was equal to or greater than the concentration dispensed at 16 μm.

The importance of cleaning and sanitation in conjunction with insecticide application is receiving more emphasis. Accumulated food dusts and spillage can compromise the effectiveness of residual surface treatment and aerosols (Arthur 2008; Arthur and Campbell 2008; Toews et al. 2010), while providing shelter and nutrition and enable survivors of treatments to recover from exposure. Recent studies show the presence of food material dramatically increased survival of different life stages of confused flour beetles exposed to aerosols in a simulated field exposure (Kharel et al. 2014a, b). In addition, in Arthur et al. (2014a, b), efficacy of the 16 μm pyrethrin aerosol was reduced when adult confused flour beetles were provided with a food source after they were exposed. Therefore, it is important in assessing insecticide efficacy to also consider the interaction with level of sanitation.

Field tests with pyrethrin aerosols indicated that they were an effective control strategy (Arthur 2008; Arthur and Campbell 2008; Arthur et al. 2013b), but there were no assessments of how aerosols were distributed in actual practice. Simulated field studies in experimental sheds show that barriers and obstructions can reduce dispersal of aerosols (Tucker et al. 2014, Tucker et al. 2015 Kharel et al. 2015). Similar results were obtained in a field study utilizing multiple floors of a flour mill, utilizing the same techniques as the paper cited above (Campbell et al. 2014). Also in the field, there was considerable variation in efficacy, depending on where bioassay arenas were located in the mill. Results showed that there were zones within each floor that received less aerosol compared to the more open areas on each floor. Further research is needed to quantify deposition of aerosol particles in field sites.

Contact Surface Sprays

Residual liquid-based insecticides can be applied as sprays for general surface treatment to a flooring surface, but must leave sufficient residues to kill immature or adult life stages of insects encountering those treated surfaces. Common insecticides used in the US are the pyrethroids cyfluthrin and deltamethrin, along with the insect growth regulators methoprene and pyriproxyfen (Arthur 2012). The effectiveness of these spray treatments depends on a variety of factors, including the composition of the specific treated surface. There is a considerable body of research on efficacy of residual treatments on different surfaces, using a variety of stored product insects as the test species (Arthur 2012; Wijarantne 2012). Residual efficacy is usually greater on non-porous surfaces such as metal or floor tile, compared to more porous surfaces such as concrete (Wijarantne 2012). In addition, it is rare that an entire flooring surface would be treated, thus residual spray treatments may offer limited control of species or life stages that may not have direct or limited access to the treated surface.

Research with adult flour beetles show that the presence of food material, either while the adult is exposed or after it is removed from a treated surface, severely compromises the efficacy of contact insecticides (Arthur 2009, 2013, 2015a, b). Flour residues within milling facilities can be correlated with increased capture of *T. castaneum* (Semaeo et al. 2012). Adults can often be knocked down after exposure on a treated surface, but exhibit varying levels of recovery and survival, if removed from that treated surface (Arthur 2015a, b; Agrafioti et al. 2015). The level of knockdown, and resulting recovery, are often correlated, and can be measured by indices that attempt to relate knockdown and potential recovery (Agrafioti et al. 2015). Recent studies also show there is variation between standard laboratory strains used in research studies and field strains when they are exposed on a treated surface, in general; the field strains are more tolerant and there is variation in the response of those field strains to residual surface treatments (Seghal et al. 2014).

Current research with contact insecticides on treated surfaces is focusing more on aspects such as knockdown and recovery of adult insects exposed for short time intervals on treated surfaces (Athanassiou et al. 2011a, b, 2013; Agrafioti et al. 2015), and residual efficacy of IGRs on different surfaces (Arthur et al. 2009; Fontenot et al. 2013). The IGRs present a challenge, since they do not generally affect adult insects, thus immature stages must be exposed on a treated surface to evaluate the IGRs. Food material has to be provided to those exposed immatures. However, the IGRs methoprene and pyriproxyfen, which are used as residual surface treatments in the US in addition to being used as aerosols, offer excellent residual control (Arthur et al. 2009). One caution is that there is evidence that the presence of a flour food source will tend to absorb residues from a treated surface, thus reducing potential residual efficacy towards exposed adults (Arthur et al. 2015c). However, in studies in which larvae have been exposed on a surface treated with an IGR, and a flour food source provided to those larvae, it is apparent that the flour absorbs residues from the treated surface (Arthur and Fontenot 2012; Arthur 2015b, 2016). Some of the recent advances with residual treatments applied to various surfaces are new data on insects that can be considered emerging pests of stored products, including psocids (Nayak et al. 2014). Studies indicate that psocids are harder to kill with contact insecticides and aerosols compared to stored product beetles, and application rates that will kill stored product beetles do not give the same level of control with psocids (Nayak et al. 2014).

Repellents

Repellents are often used for personal protection against biting insects, but the odor associated with these types of products may likely limit their uses around processed food material. Repellants for stored product insects have been examined frequently over the years, often by simply treating half of a treatment arena with a repellent, and recording presence/absence of insects on either the treated or untreated portions of the arena at selected post-treatment intervals. New research with repellents focuses more on behavioral aspects of repellents, rather than traditional methodologies such as recording presence or absence (Arthur et al. 2011). There are several excellent reviews that discuss using of natural product tested for either direct control of stored product insects or use as a repellent (Isman et al. 2011; González et al. 2013; Kedia et al. 2015), however; to date there has been limited commercial development of natural products/essential plant oils for use in structural applications to control stored product insects.

Mating Disruption Strategies

Pheromones are chemical secreted by insects to attract members of their own species. They can be aggregation pheromones, which attract males and females, or sex pheromones, which are generally produced by the female to attract the male for mating. Several pheromones have been synthesized and commercialized, and one of the primary products is the sex attractant *Plodia interpuctella*, the Indianmeal moth, commonly known as ZETA (Athanassiou et al. 2016). It also attracts four other moth species as well. It has been extensively used in monitoring programs for *P. interpunctella* (Athanassiou et al. 2016).

This pheromone is now being used as a control strategy, under the broad term of mating disruption. The concept is that a number of pheromone dispensers are placed inside a structure, and multiple pheromone plumes are generated, thus confusing the males and making it difficult for them to locate the female. There are several recent studies that show success using this strategy, as assessed by moth populations before and after a disruption effort has been initiated (Savodelli and Trematerra 2011; Trematerra et al. 2013; Athanassiou et al. 2016). There are several commercial products available in the US, all with different release rates of the pheromone, variation in recommendations for numbers and spacings, and other instructions for usage, but they are all based on the standard ZETA pheromone. There is excellent potential for the expansion of this strategy for control of *P. interpunctella* in a variety of situations. Currently, research is being conducted on mating disruption for other stored product insects (Mahroof et al. 2014), and other products could be introduced into the market for use in control programs.

Insect Resistant Packaging

This chapter focuses on structural pest management for stored product insects, but one overlooked aspect of management is insect-resistant food packing for protection and even control of insects. Stored product insects that can infest packaged goods can be categorized as penetrators, those insect species that can penetrate through a package, and invaders, species that required a flaw in the package to gain access to the contents (Highland 1991). Examples of penetrators are larvae of *P. interpunctella* (Borwditch 1997), adults of *Sitophilus* spp. and *Rhyzopertha dominica* (F.), larvae and adults of *Lasioderma serricorne* (F.), and *Stegobium paniceum* (L.), and dermestids, including *Trogoderma variabile Ballion*, the warehouse beetle (Highland 1991; Mowery et al. 2002; Hou et al. 2004; Mullen et al. 2012). Many research papers cite the Highland (1991) book chapter as the source of the classification system for invaders and penetrators. These insects produce different types of damage, such as scars, scratches, and holes, which vary with species and with packaging materials, (Ruidavets et al. 2007; Chung et al. 2011). Entrance and exit holes can often be determined, and the papers cited above have electron micrographs showing the different types of damage indicators produced by invaders and penetrators.

Physical measures of package strength, such as tensile strength, thickness of the film, differentiation between layers, and elongation values are important measures regarding susceptibility to penetration by insects. Chung et al. (2011) evaluated four different types of packaging materials of varying thickness, in which pinholes had been made in the films to provide access. *Tribolium castaneum* adults could not penetrate any of the materials, while penetration of *P. interpunctella* larvae varied with physical characteristics of the film. The film with the highest elongation value and lowest tensile strength gave the best protection against *P. interpunctella*. The specific type of package can also be an important factor in assessing susceptibility of a package. In tests by Lu and Ma (2015), eggs and adults of *L. serricorne* were released on wheat flour packaged in five common bag types: vacuum plastic bags, Kraft paper bags, nonwoven cloth bags, aluminum foil bags, and woven plastic bags. The greatest penetration of adults was in nonwoven cloth bags. Little penetration occurred in aluminum foil and plastic bags. Flaws in packaging sealing and air holes in materials can also facilitate entry by stored product insects. Studies with weevils and penetration of packaging containing pasta showed that weevils could enter carton boxes that were not well sealed, and air vents in polypropylene facilitated entry (Trematerra and Savoldelli 2015).

Different repellents have been added to packaging films, with varying degrees of success and efficacy, but to date there has been limited commercial applications utilizing repellents. Hou et al. (2004) designed a laboratory testing methodology for evaluating different repellents applied to paper envelopes to simulate a package, and assessing penetration through the treated paper. DEET was the most effective repellent, followed by Neem, but protein pea flour did not deter insect entry into the envelopes. However, in a test in which solutions of protein-enriched pea flour was applied to polyethylene sheeting at concentrations ranging from 0 to 10%, the highest concentration of 10% prevented penetration of adult *R. dominica* and *S. oryzae* through the sheeting (Mohan et al. 2007). There have been several recent tests using different forms of cinnamon oil as a repellent. Micro-encapsulated cinnamon oil imprinted on polypropylene film repelled *P. interpuctalla* larvae, and in addition had no effect on physical properties of the packaging or on sensory perception of packaged candy products (Kim et al. 2013; Jo et al. 2015). Cinnamon extract was also an effective repellent for *P. interpunctella* (Na et al. 2008). Other compounds tested for repellent activity in packaging include propryonic acid and (E)-2-hexenal. Both reduced populations of *Sitophilus granarius* (L.), the granary weevil, in treated packaging relative by 85–90% in treated packaging relative to controls (Germinara et al. 2010, 2012).

There have been several recent research studies utilizing various commercial insecticides that have been either incorporated into or impregnated onto various packaging materials. Packaging treated with the insect growth regulator (IGR) methoprene is commercially available in the US (Arthur 2016). Initial trials with different materials showed when late-stage larvae of *T. castaneum* or *T. confusum* were exposed on packaging that had been treated with concentrations of 0.1–0.5% active ingredient (AI) of methoprene, they could not emerge as morphologically-normal adults. Development was arrested at either the larval stage or the pupal stage, and both the outside and inside surfaces of the packaging showed activity (Arthur 2016). Further expanded tests confirmed susceptibility of *T. castaneum* to the treated packaging at a 0.1% AI concentration of methoprene, however, late-stage larvae of *T. variabile*, were less susceptible to the methoprene-treated packaging compared to *T. castaneum* (Scheff et al. 2016). If the egg stage was exposed on the treated packaging, few adults of either species were able to complete development, and again development was arrested at either the larval or pupal stages. New research also shows efficacy of the pyrethrin deltamethrin incorporated into packaging material manufactured by Vestergaard (Lausanne, Switzerland). The material has a broad range of toxicity on a variety of stored product beetle species (Paudyal et al. 2016, 2017), and can offer long-term protection from entry into the packaged product. However, regardless of the effectiveness of repellents or chemical treatments incorporated into the packaging, faulty and inadequate sealing of the package is a primary mode of entry for insect and mite pests (Ruidavets et al. 2007; Hubert et al. 2014; Athanassiou et al 2011c).

Conclusion

This chapter summarizes the various control strategies that can be used for structural management of stored product insects. It is not mean to be an exhaustive review of those various components, and focuses more on broad trends and applications rather than detailed examinations of research studies. Similarly, references are included for specific examples, and inclusion of a reference does not necessarily denote preference over similar studies that could potentially be included. Many of the cited references in turn contain citations of historical research and more detailed information than can be presented in this chapter. Readers are provided with means to conduct detailed literature searches on a specific topic.

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Chapter 5
Bacterial Insecticides and Inert Materials

Christos G. Athanassiou and Frank H. Arthur

Introduction

The term “novel insecticides” can be regarded as a category that includes the insecticides with a novel mode of action, but also insecticides that are novel in terms of their low mammalian toxicity but also their environmental-friendly profile. Under this context, it is difficult to identify active ingredients that are novel and separate them from the “traditional” substances, which include insecticides such as organophosphorous (OP) compounds, carbamates, and pyrethroids, since many of them, such as some pyrethroids, have low mammalian toxicity. Hence, there are certain substances that are compatible with these profiles, but, in terms of their discovery and use are not new, but their use fulfills certain low-risk requirements. In this chapter, we will focus on the recent development of contact insecticides, i.e., the insecticides that cause death to insects through contact or digestion, regardless of the way that these insecticides are applied e.g., in the surfaces or directly on the grain. Although there are numerous substances that fall into the category of “novel insecticides” in postharvest applications, in the following we will refer to those which are already registered for this purpose, are close to registration or they are

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closely related with already registered active ingredients. These can be summarized in two broad categories: bacterial-based insecticides and inert materials. Other major categories of insecticides are covered in other chapters of this book.

Spinosad

Although spinosyns are considered relatively novel active ingredients, their evaluation for stored product protection was started more than a decade ago (Hertlein et al. 2011 and references therein). Spinosyns are metabolites of the naturally occurring actinomycete *Saccharopolyspora spinosa* Mertz and Yao (Bacteria: Actinobacteria). Spinosad is based on spinosyns A and D, and it is by far the most commonly used spinosyn-related insecticide globally. It acts neurotoxically through contact of digestion on the nicotinic acetylcholine and gamma aminobutyric acid (GABA) receptors (Sparks et al. 1995). It has no mammalian toxicity, a friendly environmental profile and so far there are no reports of cross-resistance (Salgado and Sparks 2005; Hertlein et al. 2011). Furthermore, spinosad is considered as an organic insecticide. Spinosad has been proven effective against a wide range of stored product insects in numerous types of commodities/crops (Hertlein et al. 2011).

Spinosad has been found highly effective against the stored product Bostrychidae, *Rhyzopertha dominica* (F.), the lesser grain borer, and *Prostephanus truncatus* (Horn), the larger grain borer. Early studies documented that spinosad could provide 100% kill of *R. dominica* adults on grain treated with dose as low as 0.1 ppm (Fang et al. 2002a, b; Athanassiou et al. 2008a, b, c). Similarly, on maize, *P. truncatus* was very susceptible to 0.1 ppm (Hertlein et al. 2011), which is considerably lower in comparison with the doses that are used for older grain protectants. The same doses are effective against adults of the rusty grain beetle, *Cryptolestes ferrugineus* (Stephens) and the flat grain beetle, *Cryptolestes pusillus* (Schonherr) (Coleoptera: Laemophloeidae) (Vayias et al. 2009a; Hertlein et al. 2011). Higher doses, such as 0.5 ppm, are effective against stored product moths, such as larvae of the Indianmeal moth, *Plodia interpunctella* (Hubner), and the Mediterranean flour moth, *Ephestia kuehniella* (Zeller) (Lepidoptera: Pyralidae) (Hertlein et al. 2011). Saglam et al. (2011) in laboratory bioassays found that spinosad has no ovicidal effect against the confused flour beetle, *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae), but it was very effective against newly hatched larvae. Similarly, Vayias et al. (2009b) showed that spinosad was effective for the control of larvae of *T. confusum*, on various types of commodities. However, there are species for which spinosad was moderately effective. In the case of weevils of the genus *Sitophilus* (Curculionidae), the maize weevil, *Sitophilus zeamais* Motschulsky, 100% kill was achieved at doses of 1 ppm or lower, but higher doses are needed for the rice weevil, *Sitophilus oryzae* (L.) (Hertlein et al. 2011). Based on the available literature, *T. consusum* and *T. castaneum* are the most tolerant stored product insects to spinosad. One other category of species that are tolerant to spinosad are psocids (Psocoptera). Most of the major stored product psocids of the family Liposcelididae were tolerant to spinosad, with the exception of *Liposcelis entomophila* (Enderlein) (Athanassiou et al. 2009). At the same time, spinosad is very toxic to stored product Hymenopteran parasitoids (Toews and Subramanyam 2004).

Spinosad has been registered for direct application on grains, as a typical grain protectant. Despite its high degree of dissipation under the effect of light, spinosad has a remarkable persistence. Fang and Subramanyam (2003) showed that spinosad had the same efficacy against *R. dominica*, during a 4-month storage period and that spinosad performance was not affected by moisture or temperature. Similar reports have been reported for other commodities and longer residual storage times (Hertlein et al. 2011). Although variations in spinosad performance may occur at different temperature and moisture/relative humidity levels, generally the effect of these factors on spinosad efficacy is less important in comparison with other insecticides (Fang and Subramanyam 2003; Athanassiou et al. 2008b). Still, one factor that seems to have a certain impact on spinosad efficacy is the type of the commodity. Generally, spinosad is less effective on maize than on other grains. According to Chintzoglou et al. (2008), dissipation of spinosad occurs more rapidly on maize, which suggests a possible interaction of this ingredient with the exterior of the maize kernel. Fang et al. (2002a) noted that there are considerable differences in spinosad performance among different classes of wheat and that it was more effective on durum wheat than on hard red spring, hard red winter, and soft red winter wheat.

Despite the fact that spinosad is not currently registered for use on surfaces (i.e., floors, walls, etc.), it has been evaluated with success for this purpose at a wide range of application scenarios. Toews et al. (2003) examined its performance on concrete, waxed and unwaxed tile, and galvanized steel, against eight stored product insect species, and found that despite variations among surfaces spinosad was effective at 0.05 mg of active ingredient [AI]/cm^2. Spinosad is more slow-acting in comparison with other active ingredients, such as pyrethroids or pirimiphos-methyl (Pozidi-Metaxa and Athanassiou 2013), but exposed insects are immobilized right after contact with the treated substrate, which has a certain effect in the concomitant oviposition (Hertlein et al. 2011). For the most susceptible species, such as *R. dominica*, even a short exposure of hours to spinosad-treated wheat or maize can result in high mortality (Athanassiou et al. 2010). Moreover, for the same species, mortality can occur even if only a part of the grain is treated, which means that this active ingredient can be used in partial treatments of the grain mass (Athanassiou et al. 2010).

Spinetoram

One additional insecticide that is based on the spinosyn family is spinetoram. Spinetoram has spinosyn J and L as active ingredients and acts on insects with the same way that spinosad does (Dripps et al. 2011). In comparison with spinosad, spinetoram is considered more stable (Chloridis et al. 2007). During the recent years, a series of bioassays have shown that spinetoram is effective against stored product insects on raw grains and on different surfaces (Saglam et al. 2013; Vassilakos and Athanassiou 2013, 2015; Vassilakos et al. 2014a, b, 2015). In fact, in some of the cases tested, spinetoram was more effective than spinosad, but in general, spinetoram and spinosad were found to be equally effective for the control of *P. truncatus, R. dominica, T. confusum,* and *S. oryzae* adults (Athanassiou and Kavallieratos 2014). At the same time, the combined use of spinetoram with spinosad did not reveal any additive or detrimental effect (Athanassiou and Kavallieratos 2014).

Abamectins

One other metabolite-based insecticide that has been tested for stored product protection is abamectin. Abamectin belongs to the avermectins group of insecticides, which are produced by the soil microorganism *Streptomyces avermitilis* Burg et al., (Actinomycetales: Streptomycetaceae) (White et al. 1997; Ahmad et al. 2003). Abamectin acts on insects as an agonist to GABA receptors (White et al. 1997). Hussain et al. (2005) tested abamectin for the control of larvae of *T. castaneum*, with good results. Later, Athanassiou and Korunic (2007), tested abamectin in combination with diatomaceous earth and found that this combination was effective for the control of several stored product beetle species. In a more recent effort, Kavallieratos et al. (2009) found that abamectin was effective for the control of *S. oryzae*, *R. dominica,* and *T. confusum*. Moreover, in that study, the authors reported that the efficacy of abamectin was not influenced by key biotic or abiotic factors, such as temperature and commodity. Still, it is uncertain if abamectin will be eventually registered for stored product protection, including its use as a grain protectant.

Inert Materials and Nanoparticles

The use of the nanoparticles in crop protection and also against public health pests has been tested in detail, but only a few nanoparticles have been evaluated for stored product protection. Nanoparticles (NPs) in crop protection and production can be defined as ultrafine particles, ranging between 1 and 100 nm, and have properties that are not shared by non-nanoscale particles with the same chemical composition (Auffan et al. 2009). Another category of materials of bigger size, which do not fall into the broad nanoparticle family, is diatomaceous earths (DEs) and zeolites. To a lesser extent, there are other materials that have been tested for this purpose, such as kaolin and different types of minerals.

Diatomaceous earths (DEs) are probably the definition of “natural products” in stored product protection. DEs are formed by the fossils of unicellular cells, known as diatoms (phytoplanktons), which existed during the Cretaceous and mainly during the Eocene and Miocene periods (Korunic 1998). When diatoms were fossilized, they produced an amorphous chalky rock, called diatomaceous earth. Today, DEs have been utilized in numerous different industrial uses, such as in water purification or the filtration of other liquids, such as alcohol and beverages, separation of oils and chemicals, but also in making explosives, removing tastes and odors etc. (Korunic 1998). According to the US Environmental Protection Agency (USEPA), DEs are classified in the General Recognized as Safe (GRAS) category, which means that they can be used as food and feed additives (Subramanyam and Roesli 2000).

Historically, the use of DEs as insecticides were started before WWII, but the first formulations were applied at high dose rates and had a high crystalline silica content, which posed some risks for human health (Korunic 1998; Subramanyam and Roesli 2000). When mined, DE contains usually >50% moisture and most of the solids contain silica, while particles vary in their size (usually between 0.5 and 100 μm) (Korunic 1997, 1998). Particle size is negatively related to efficacy (Subramanyam and Roesli 2000; Vayias et al. 2009c). Hence, the preparation of DE samples is regarded as a cheap procedure, since it involves only drying and particle separation (milling/sieving). As an inert material, DE has little interaction with the environment, provided that the conditions are generally dry (Athanassiou et al. 2005). At the same time, the presence of DEs does not affect the bread- and pasta-making properties of amylaceous commodities (Korunic et al. 1996).

Several theories concerning the mode of action of DEs have been proposed. The most widely accepted theory is that death is caused by absorption that results in water loss, as the DE particles adhere to the insect cuticle (Subramanyam and Roesli 2000). However, there is an evidence of secondary modes of action, with abrasion being the most widely accepted (Subramanyam and Roesli 2000). Mode of action through abrasion may be related with the shape of the particles and generally round-shaped are less abrasive than angle-shaped DE particles (Korunic 1998).

As compared with the neurotoxic insecticides, DEs are much more “slow-acting” (Athanassiou et al. 2005b; Vayias et al. 2009c). Subramanyam and Roesli (2000) indicated that this is one of the main drawbacks on the use of DEs, given that delayed mortality can cause oviposition before death, which may increase grain damage. Nevertheless, the main disadvantage on the use of DEs is the fact that the application of DEs negatively affects the bulk density of the grain (tests weight or volume to weight ratio). Korunic et al. (1996, 1998) found that the reduction of the presence of DE in the grain increases bulk density, and that slurry DE formulations (suspensions) had less negative effects than dry DEs. However, even the minimal presence of DE, to a rate as low as 10 ppm, reduced bulk density. Consequently, this effect by the use of DEs should be considered as unavoidable. Generally, smaller particles reduce the bulk density to a higher extent than larger particles (Korunic 1998; Korunic et al. 1998; Vayias et al. 2009c). Vayias et al. (2009c) found that, for several types and formulations of DEs, particles with size

<45 µm were generally more effective against stored product insect species than particles that were >45 µm. In that study, the authors suggested that for many naturally occurring DEs, with particle sizes between 0 and 150 µm, most of the insecticide properties should be attributed to the portion of the particles that were <45 µm. Still, there were some types of DEs for which particles that were >45 µm had similar efficacy with particles that were <45 µm. Moreover, apart from efficacy reduction, larger (>150 µm) particles that contain rocks, sand, and very large diatoms, should be avoided during production since they can reduce the insecticidal effect.

There are several commercially available formulations that are based on either DE alone or the combination of DE with several other compounds, such as low mammalian toxicity insecticides, botanicals, or food-grade additives. Older commercially available formulations had relatively high application rates, 1000 ppm (1 g per kg of grain) or higher, which negatively affected bulk density and flowability. As a result, despite the high efficacy (Vayias and Athanassiou 2004, Athanassiou et al. 2005a, b), the wider adoption of DEs in a large-scale use was impractical (Subramanyam and Roesli 2000; Athanassiou et al. 2004). Recently, there are several DE formulations that can be used at considerably low concentrations (400 ppm or lower), which include mixtures of DEs with natural pyrethrum (Athanassiou et al. 2004; Athanassiou and Kavallieratos 2005; Vayias et al. 2006), silica aerogel (Fields and Korunic 2000); botanicals (Athanassiou and Korunic 2007), metabolites (Athanassiou and Korunic 2007; Vayias et al. 2009b) and low doses of OPs or pyrethroids (Arthur 2004). Moreover, there are several research reports that underline the synergistic or additive effect of DEs with several other ingredients. For instance, Lord (2001) found that the simultaneous use of DEs greatly enhanced the insecticidal effect of the entomopathogenic fungus *Beaveria bassiana* (Balsamo) Vuillemin (Hyphomycetes: Moniliales) against several major stored product beetle species. This work initiated a series of publications with the same combinations but with different species, with similar promising results (Akbar et al. 2005; Lord 2005; Vassilakos et al. 2006; Athanassiou and Steenberg 2007). Lord (2001) suggested that DEs are likely to inactivate the fungistatic and fungicidal epicuticular lipids which enhance the germination of the fungal conidia. Moreover, abrasion caused by DEs on the insects' cuticle may increase conidia germination and penetration in the insects' body (Akbar et al. 2004; Lord 2005). However, for another fungal species, *Metarhizium anisopliae* (Metschnikoff) Sorokin (Ascomycota: Hypocreales) the simultaneous use of DEs had no effect, and in some cases, had to slight detrimental effect on activity (Michalaki et al. 2006, Kavallieratos et al. 2006). Still, for this fungal species, other inert materials (chalk powder, charcoal, ash etc.), may additive effects (Batta 2003, 2004).

One of the most promising means to reduce the total amount of DEs used is the combined use of DEs with pyrethroids. In Brazil, Lazzari et al. (2001) obtained long-term protection of paddy rice with the use of DE with deltamethrin. Similar results have also been reported by other researchers for various pests and commodities (Ceruti and Lazzari 2005). Arthur (2004) tested a DE formulation that contained chlorpyriphos-methyl, piperolyl butoxide (PBO), deltamethrin, and mineral oil, and found that efficacy was high at doses as low as 50 and 100 ppm. For a commercial DE that was enhanced with natural pyrethrum and PBO, Athanassiou et al. (2004) found that the "speed of kill" was more rapid than other DEs that were based only on SiO_2. Moreover, Vayias et al. (2006) found that the same pyrethrum-enhanced DE was effective against *T. confusum*, even at the pupal stage, while the DEs alone killed only the adults after their emergence. At the same time, the application of natural pyrethrum alone provided the same results (Vayias et al. 2006). Overall, given that pyrethroids have low mammalian toxicity, this combination seems to be the most promising combination with DEs.

Apart from the application with pyrethroids, that DEs can be used with botanicals (plant extracts). One example is the combination of DE with the extract bitterbarkomycin (BBM) in a single formulation (DEBBM). Athanassiou and Korunic (2007) found that DEBBM was very effective against several stored product insect species, at doses as low as 75–150 ppm. Yang et al. (2010) combined essential oils of *Allium sativum* L. with DE against *S. oryzae,* and *T. confusum* and found that this application was much more effective than the application of either the oil or the DE alone. Moreover, Islam et al. (2014) reported that DE enhanced the insecticidal effect of the monoterpenoids eugenol and cinnamaldehyde for *S. oryzae* and that this combination produced greater mortality than the application of DE alone. Plant extracts, particularly those that are active as essential oils, have a rapid breakdown. The absorption of these compounds into amorphous silicaceous materials is likely to prolong the insecticidal activity of these oils, and at the same time, to accelerate the mortality caused by DEs.

Other combinations of DEs include different types of insecticides. Wakil et al. (2013) found that the efficacy of the neonicotinoid thiamethoxam when applied alone was lower than the application of thiamethoxam with DEs, at least for some species, commodities, and exposure intervals. Also, Wakil et al. (2013) noted that these two compounds could be used with success with *B. bassiana*. Moreover, a DE formulation that contained abamectin (DEA) was very effective against a wide range of species at very low dose rates (75 ppm) (Athanassiou and Korunic 2007).

There are considerable differences among species, regarding their susceptibility to DEs, but the data available are often contradictory. From the stored product species tested, all data indicate that the species of the genus *Tribolium* are the least susceptible (Subramanyam and Roesli 2000; Arthur 2000a, b; Vayias and Athanassiou 2004). Arthur (2004) found that S. *oryzae* and *T. castaneum* could survive at doses that killed *R. dominica*. Moreover, with a formulation that is based on SiO_2 alone, Athanassiou et al. (2005) reported that *T. confusum* adults were much more tolerant than *S. oryzae* in wheat treated with DE. In fact, when comparing *T. castaneum* with *T. confusum*, Arthur (2000a, b) noted that *T. confusum* was more tolerant than *T. castaneum.* Regarding the other beetle species, many studies show that *C. ferrugineus* is probably the most susceptible species to DEs (Korunic 1998; Korunic and Athanassiou 2007). Korunic (1998) reported that water loss for *C. ferrugineus* is likely to occur more rapidly than other species, due to its body characteristics, which is very flat. Among the other major stored product beetle species *O. surinamensis, S. oryzae*, *S. granarius,* and *S. zeamais* are considered as susceptible to DEs (Subramanyam and Roesli 2000, Arthur 2001). Conversely, according to Fields and Korunic (2000) *R. dominica* should be considered as tolerant to DEs. In that paper, the authors indicated that *R. dominica* adults are generally slow-moving, so the possibility of picking up DE particles is reduced. However, there are studies where *R. dominica* was found susceptible to DEs (Kavallieratos et al. 2005; Athanassiou and Kavallieratos 2005). As indicated for almost all grain protectants, there are differences in the susceptibility among different strains (populations) of the same species. For example, Vayias et al. (2006) found considerable differences in susceptibility to DEs among populations of *T. confusum* originating from different areas of Europe. Vayias and Athanassiou (2004) found that newly emerged *T. confusum* adults were more susceptible to DEs than older ones. Nevertheless, regarding beetle larvae, the differences are wider than in adults. Generally, most larvae are expected to be more susceptible than adults.

Regarding moths, most of the data available are for larvae. As in the case of beetle larvae, young moth larvae are more susceptible than older ones, especially those that enter the pre-pupation period (Subramanyam et al. 1998; Nielsen 1998; Mewis and Ulrichs 2001; Athanassiou 2006). For *P. interpunctella* larvae, Mewis and Ulrichs (2001) noted that the effectiveness of a DE was considerably higher in first instar larvae than against third to fifth instar larvae. In comparison with *T. confusum* larvae, *E. kuehniella* larvae were less susceptible to DE (Vayias and Athanassiou 2004; Athanassiou 2006). Mewis and Ulrichs (2001) also indicated that *P. interpunctella* larvae were less susceptible than larvae of *T. confusum.* In contrast with beetle larvae, moth larvae produce webbing, which is likely to reduce their contact with the DE particles (Nielsen 1998).

Given that soft-bodied arthropods are expected to be more susceptible after exposure to DEs, several DEs have been evaluated against other stored product pest categories, such as mites and psocids. In the case of mites, DEs are very effective against Astigmata, which are considered as the most soft-bodied mite species. Cook and Armitage (1999) and Cook et al. (1999) noted that DE was effective for the complete control of *Acarus siro* L. (Astigmata: Acaridae) while high doses are needed for the control of *Lepidoglyphus destructor* (Schrank) (Astigmata: Glycyphagidae). In this context, Palyvos et al. (2006) found that the cheese mite, *Tyrophagus putrescentiae* (Schrank) (Astigmata: Acaridae) was very susceptible to DEs, with mortality that was 100% or close to 100% even after 2 days of exposure to the treated substrate.

Zeolites

Natural zeolites are often referred as "the magic rock" due to their remarkable range of use which not only includes agriculture but also food elements. Their mode of action is probably the same as that of DEs (Rumbos et al. 2016; Eroglou et al. 2017). There are different types of zeolites, such as clinoptilolite, analcime, chabazite, laumontite, mordenite, and phillipsite (Mumpton 1999). In a recent review, Eroglou et al. (2017) highlighted the importance of zeolites in agriculture and food protection, underlying several paradigms of using zeolites in stored product protection. The European Food Safety Authority (EFSA) lists clinoptilolite in the substances that are safe for utilization as food and feed additives (EFSA 2013).

Paradoxically, there are disproportionally fewer studies that focus on the evaluation of zeolites for stored product protection in comparison with those that are available for DEs. However, there is a renewed interest in the investigation of zeolites for this purpose, probably due to the recent legislation that highlights their safety. One earlier study for natural zeolite application for stored product insects is that of Haryadi et al. (1994). Overall, recent works showed that zeolites are effective against a wide range of target species, such as *S. zeamais*, *S. oryzae, R. dominica, T. confusum*, *T. castaneum*, and *O. surinamensis* (Kljajic et al. 2010; Rumbos et al. 2016). Moreover, zeolites are excellent mycotoxin binders (Kovac et al. 1995).

In general, the performance of zeolites regarding their influence on specific properties on the treated commodities is similar to that reported for DEs; hence zeolites do not affect bread-making properties when applied on grains (Bodroža-Solarov et al. 2012). At the same time, zeolites notably reduce the bulk density of the grains, even if they are applied at low dose rates (Bodroža-Solarov et al. 2012; Rumbos et al. 2016). Recently, Rumbos et al. (2016) reported that the reduction that zeolites cause to the bulk density of the grains was related with their particle size, with lower particles having a stronger effect on bulk density reduction. In that study, the authors noted that the degree of attachment of the zeolite particles among grains was different, and less particles were found on maize than on other grains, probably due to the same patterns that have been previously reported for DEs (Kavallieratos et al. 2005).

Mineral-Based Particles

Metal NPs are usually synthesized chemically, but there are cases where living organisms are involved in their formation (Dubey et al. 2009). Some metal NPs have shown remarkable properties as insecticides. Examples of these types of NPs are aluminum oxide, zinc oxide, titanium oxide, and silver. Their mode of action for these materials is primarily absorption of epicuticular lipids through, which means that they work in a similar way with other inert materials, such as DEs (Stadler et al.

2009, 2012; Buteler et al. 2015). Moreover, as in the case of DEs and zeolites, their insecticidal effect is heavily influenced by their particle size, morphology, and certain key biotic and abiotic factors (Stadler et al. 2012; Buteler et al. 2015). Ki et al. (2007) tested nanosilver and found high efficacy for the control of the case-bearing clothes moth, *Tinea pellionella* (L.) (Lepidoptera: Tineidae). These NPs have been evaluated for the control of different stored product insect species, especially primary colonizers, such as *R. dominica* and *S. oryzae* (Stadler et al. 2009, 2012; Debnath et al. 2011; Abduz-Zahir et al. 2012; Buteler et al. 2015). Rouhani et al. (2013) showed that silicon dioxide-based nanoparticles were generally more effective than silver-based nanoparticles against the cowpea seed weevil, *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae). Moreover, some of these agents can also be used as viable binders for certain reduced-risk agents, such as antifeedants (Usha Rani et al. 2014).

Delivery Systems

One of the most novel approaches in stored product protection is the use of NPs and related agents for the so-called “auto-dissemination” of active ingredients. The term auto-dissemination has been initially formed to describe the use of electrostatically charged dusts for the delivery of pheromones through contaminated adults in order to cause insect control through mating disruption, a technique also known as “auto-confusion”. In this context, this technique has been successfully used for the control of stored product moths, such as *P. interpunctella* and *E. kuehniella*, in various types of facilities in Europe (Trematerra et al. 2013). The basic difference between auto-confusion and “classical” mating disruption is that there is no air permeation with the pheromone and contamination is carried out through baiting stations. The most widely used material that is used for this purpose is Entostat, based on carnauba wax, but there are also other substances that have been evaluated for this purpose (Baxter 2008; Baxter et al. 2008).

Electrostatically-charged dusts have been tested for the delivery of insecticides for the control of stored products. Recently, Athanassiou et al. (2016) tested a combination of Entostat with the OP pirimiphos-methyl and found that with this combination the efficacy of pirimiphos-methyl was increased, in combination with the use of pirimiphos-methyl alone, regarding the control of *S. oryzae* and *O. surinamensis*. Generally, the application of Entostat did not affect the bulk density of the grains and they had no effect on their bread and pasta-making properties (Athanassiou et al. 2016). Conversely, the adherence of the dust particles on the grains was high, apparently due to their electrostatic nature. Wakefield et al. (2010) noted that the presence of Entostat had no repulsive effect on *O. surinamensis* adults.

The combination of micro-biocontrol agents with electrostatic powders has been examined for different stored product insects. For *P. interpunctella*, Baxter (2008) tested different types of dusts for the delivery of *B. bassiana*, and found that Entostat worked better than others. These results were also confirmed for stored product beetles by Athanassiou et al. (2017), where a formulation that combined a specific strain of *B. bassiana* (Bb38), Entostat and kaolin clay provided a satisfactory level of control in both laboratory and semi-field conditions, without loss in efficacy through time. It is estimated that this technique may serve for a wider range of species and uses (i.e., both for admixture with the grain and for surface treatments) in the near future. In addition some NPs, including charged powders may limit breakdown of active ingredients (Athanassiou et al. 2016, 2017).

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Chapter 6
Insect Pest Management of Oilseed Crops, Tree Nuts and Dried Fruits

Shlomo Navarro and Hagit Navarro

Introduction

Many storage insects are polyphagous and some attack specific groups of products making it difficult to classify them. Storage insects have generally a global distribution, and very seldom are they specific to a certain climatic zone or a specific crop. Stored product pest management literature is detailed in reference to cereals and their products but less information exists in relation with the three groups of commodities referred in the chapter, namely oilseed crops, tree nuts and dried fruits.

The preparation of this chapter is largely based on the experience reported in the Hebrew book on Insects of Stored Products (Navarro et al. 1991) and the long-term experience gained on sampling for the presence of insects in a variety of stored products imported to Israel from various countries, at the former Department of Stored Products in the Israel Agricultural Research Organization. This chapter is dedicated to two colleagues who largely contributed to the understanding and identification of stored product pests in Israel and worldwide: Prof. Moshe Calderon (Navarro 2000) and Dr. Jonathan Donahaye (Navarro 2008).

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Commodities

Oilseed Crops

Worldwide, the major sources of edible seed oils are soybeans, sunflowers, rapeseed, cotton and peanuts. The primary scope of cultivation of those crops is for the oil contained in the seeds. The oil content of cereal grains (e.g. wheat) is only 1–2% and that of oilseeds ranges from about 20% for soybeans to over 40% for sunflowers and rapeseed (canola) (Stefansson 2009; Weiss 2000). For industrial purposes, oils from flax (linseed) and castor beans are used. At room temperature, fats are solid and oils are liquid, and edible fats and oils are similar in molecular structure.

A significant portion of the calories in the human diet is fats and oils that are essential nutrients. In salads or as cooking oils, edible vegetable oils are used or may be solidified (by a process called hydrogenation) to make margarine and shortening. In this chapter, the following crops were considered as oilseeds: coconut, cottonseed, peanut, safflower, sesame, soybean and sunflower oil.

Tree Nuts

A wide variety of dried seeds enclosed in a hard shell, which is generally edible, are termed as nuts. The shell does not open to release the seed. In a botanical context, only those fruits that are considered true nuts and tree nuts are fruits that grow on trees.

Nuts like hazelnuts, chestnuts and acorns have hard shell walls and originate from a compound ovary; many other seeds come from fruits that naturally free themselves from the shell. Since the common usage of the term is not restrictive, in a botanical sense, many nuts such as almonds, pecans, pistachios, walnuts and Brazil nuts (Alasalvar and Shahidi 2008) are not nuts. Common usage of the term often refers to any hard-walled, edible kernel as a nut (Bewley et al. 2006). Tree nuts have been part of the diet of humans since Paleolithic times (O'Neil et al. 2015). In this chapter, the following crops were considered as tree nuts: almond, Brazil nut, cashew, hazelnut (filbert), macadamia nut, pecan, pine nut, pistachio and walnut.

Dried Fruits

Fruits may be dried by reducing their water content to a level that eliminates deterioration due to microfloral activity. The water content can be reduced either naturally, through sun drying, or through the use of specialized dryers or dehydrators. Dried fruit is prized because of its sweet taste, nutritive value and extended shelf life (Hui 2006). Historically food preservation of grapes, dates and figs would be dried in direct sunlight. People observed that after drying, fruit took on an edible form and valued them for their stability as well as their concentrated sweetness (Trager 1995). Today, dried fruit consumption is widespread. Nearly half of the dried fruits sold are raisins, followed by dates, prunes, figs, apricots, peaches, apples and pears (Hui 2006). The date palm was one of the first cultivated trees. Figs were also dried in the Middle East where their daily use was probably greater than or equal to that of dates.

In this chapter, the following crops were considered as dried fruits: apple rings, apricots, banana chips, cantaloupe, coconut-white chips, currants, dates, Medjool, dates—pitted whole, dried blueberries, dried cherries, dried cranberries, dried fancy strawberries, dried peaches, dried pears, dried whole figs, ginger—crystallized, kiwi, sliced, mango—dried slices, nectarines, orange peels glazed, papaya spears, pineapple rings, prunes—pitted and raisins.

Insects of Oilseed Crops, Tree Nuts and Dried Fruits

In this section, the authors made an attempt to classify the presence and preference of storage insects in each group of commodities. This attempt quickly revealed that several of the storage insect species identified as pests have a wide preference due to their polyphagous nature. Therefore, in this section, storage insects are presented according to their families in alphabetical order and the major preference of each insect is given according to the nature of the damage they cause on the group of commodities or specific product. For each family, a short description of the family of storage insects and each species is described in relation to its behavioural and feeding habits. Family names, species and their occurrence in three different types of commodities they damage are given in Table 6.1. Since some groups of mites also cause damage on some products, the most common mites were also included in this section. We are aware that not all insects or mite species are presented in this section. The preference in selecting the storage insects is based on literature surveys and experiences of the authors with those insect species.

Family Anthribidae—Coleoptera (Beetles)

The Anthribidae is a large family of beetles, most of which feed on dead wood and fungi. Only one species, *Araecerus fasciculatus*, is of economic importance on stored products, particularly on cocoa beans.

Table 6.1 Insect families, species and their occurrence in three different types of commodities

	Family	Insect and mite species	Oilseed crops	Tree nuts	Dried fruits
1	Anthribidae	*Araecerus fasciculatus*		x	
2	Bruchidae	*Caryedon serratus*	x(peanuts)		
3	Cleridae	*Necrobia rufipes*	x(coconut)		
4	Cucujidae	*Cryptolestes spp.*	x	x	x
5	Dermestidae	*Trogoderma granarium*	x		
		Trogoderma inclusum			x
6	Mycetophagidae	*Typhea stercorea*			x
7	Nitidulidae	*Carpophilus mutilatus*	x		x
		Carpophilus hemipterus			x
		Carpophilus mutilatus			x
		Carpophilus humeralis			x
8	Silvanidae	*Oryzaephilus surinamensis*	x	x	x
		Oryzaephilus mercator	x	x	x
		Ahasverus advena	x		
9	Tenebrionidae	*Tribolium castaneum*	x	x	x
		Tribolium confusum			
10	Trogossitidae	*Tenebroides mauritanicus*	x	x	x
11	Phycitidae	*Ephestia cautella*	x	x	x
		Ephestia elutella	x		x
		Plodia interpunctella			x
		Ephestia calidella			x
		Ephestia figulilella			x
		Spectrobates ceratoniae		x	x
12	Carpoglyphidae (Acarina)	*Carpoglyphus lactis*			x

Stored product insects occurrence was based on most common commodities. It is possible that some additional insects may occur on those commodities, but to the best knowledge of the authors those listed in the table were identified as the most common

Araecerus fasciculatus Degeer, The coffee bean weevil.

The adult is moderately large (3–5 mm) in comparison with many pests of stored products. The ground colour is dark brown or greyish brown, and the prothorax and elytra have many patches of light-coloured setae giving a mottled appearance. The elytra are slightly shorter than the abdomen usually leaving one abdominal segment exposed. The three terminal segments of the antennae are longer than the other segments, forming a loose club (Fig. 6.1).

It is well known as a pest of stored high-value commodities such as coffee and cocoa beans in most tropical and subtropical regions. Coffee cherries may become infested in the field but large populations of the beetle are usually only found in

Fig. 6.1 Adults of the coffee bean weevil, *Araecerus fasciculatus* (with permission)

association with beans that have been in storage for considerable periods. The beetle is of no real significance as a pest of good quality stored beans. The contamination caused by the presence of the beetle is usually of greater importance than the actual damage it is able to cause. In contrast, dried cassava may be severely damaged particularly in the initial phase of storage. Other commodities known to be infested by the beetle include maize, groundnuts, Brazil nuts, spices, roots and various processed foods. The larvae feed initially on the pulp of coffee cherries and then attack the seed. In stored coffee beans, each female lays about 50 eggs, and the life cycle takes 46–66 days at 28 °C and 70% r.h. On maize, the insect develops most quickly at a high moisture content and is severely affected by low humidity. All stages, except pupae, die when the r.h. is lower than 60%, and at 27 °C the developmental period increases from 29 to 57 days. When the r.h. is reduced from 100 to 60%, the adults may live for more than 17 weeks, but their longevity is reduced at low humidities (Navarro et al. 1991).

Family Bruchidae

Beetles of this family feed on plant seeds. They are easily recognized by the short hairs that cover their body, and the elytra do not cover all the abdomen, leaving the last abdominal tergum (the pygidium) exposed. Most stored products Bruchidae feed on seeds of legumes. They damage a wide range of stored pulses. The only member of this family that attacks groundnuts or tamarind is *Caryedon serratus*.

Caryedon serratus (Olivier), Groundnut seed beetle

C. serratus [*Caryedon gonagra* (Fabricius)] is an important pest of in-shell stored groundnuts (Arachis) in West Africa. *C. serratus* is a large robust insect (Fig. 6.2) reddish brown in colour with dark, irregular markings on the elytra.

Oviposition and the length of adult life in the Bruchid beetle *C. serratus* were investigated by da Fonseca (1965) and Donahaye et al. (1966). Adult lifespan increases with decreasing temperature and increasing relative humidity. Adults survived for 3–4 days at 45 °C and 70% r.h. Optimum conditions are probably about 27–30 °C 70–90% r.h., under which the adult length of life was about 21 days. The adults become sexually mature on emergence from the cocoons, and mating takes place within 24 h of emergence. The pre-oviposition period is

Fig. 6.2 Adult of the groundnut seed beetle, *Caryedon serratus* (with permission)

between 24 and 48 h at 27.5 and 30 °C. About 80% of the eggs were laid in crevices in the shell of the nut, where they are difficult to find. The largest mean numbers (106–115) were laid at 27.5 and 30 °C and 70–90% r.h.; the absence of nuts did not influence the numbers of eggs laid, but caused some irregularity in the oviposition pattern. More than one copulation appeared to be necessary for the female to lay a full complement of eggs. Under conditions of alternating periods of light and darkness, there was a daily rhythm of oviposition, with a strong correlation between numbers of eggs laid and the periods of darkness. *C. serratus* is of Asian origin and it is prevalent in the warm parts of Asia, North-Eastern and West Africa, the West Indies and parts of South and Central America, but it is serious pest of groundnuts only in West Africa (Dobie et al. 1985).

Family Cleridae

Members of this family are characterized by sparse hairs on their body with some long stiff hairs, especially on the lateral margins of the prothorax. In some species, there is a distinct neck between the prothorax and the elytra. Some adults are of metallic blue colour, green or bronze. Most species are predators but those that can survive on stored products include the *Necrobia rufipes*, red-legged ham beetle, which is also a pest of coconuts (Ashman 1963; Dobie et al. 1985).

Necrobia rufipes (Degeer), Red-legged ham beetle, copra beetle.

The red-legged ham beetle adults are shiny green or bluish with the basal segments of the antenna and the legs red, about 4.5 mm long (Fig. 6.3), can survive on copra alone, especially when it is mouldy.

The adults are surface feeders; the larvae bore into dry or smoked meats and do most damage. The red-legged ham beetle also attacks bones, hides, copra (dried coconut kernels from which coconut oil is extracted), dried egg, cheese, guano, bone meal, dried figs and palm nut kernels. Although refrigeration has reduced the impact of beetle on meats, they are the most significant pest of dried and salt fish including herring.

N. rufipes is active in the warmer parts of temperate areas and in the tropics in South America and Africa (Dobie et al. 1985). The beetle will develop in warm

Fig. 6.3 Adult of the red-legged ham beetle or copra beetle, *Necrobia rufipes* (with permission)

(30–34 °C) and damp conditions, but will not breed below 20.5 °C. Its presence in high-value commodities such as processed meats and cheese, even in small numbers, cause rejection. It is attracted to stearic and palmitic acids produced by rotting copra.

Family Cucujidae

Cryptolestes is the only genus with species that are important as stored product pests in the family of Cucujidae. *Cryptolestes* species are some of the most common and destructive secondary pests of cereal grains, cereal products, nuts, oil cake, dates, dried fish and a variety of other materials in both temperate and tropical regions, often the following infestation by other insects such as moths and weevils. Small larvae cannot enter totally intact cocoa beans but are able to penetrate if the seed coat is imperfect or slightly damaged by poor handling or damage by other pests. They also attack the germ, thus reducing the percentage germination as well as causing weight loss and loss of quality.

Beetles of this family are characteristically flattened and generally only about 1.5–2 mm in length. The antennae are usually relatively long, sometimes about half the body length. Most members of this family have been found under the bark of trees, in damaged seed pods or in the tunnels made by other beetles. Many are predatory during at least one stage of their life cycle. Identification of *Cryptolestes* species usually requires preparation of the male or female genitalia. These beetles are sometimes overlooked in stores because they are very small and can hide in small cracks and in the seams of sacks. The adults are winged but rarely fly.

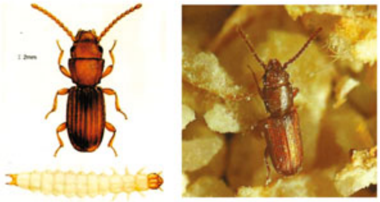

Fig. 6.4 Adult and larvae of the rusty grain beetle, *Cryptolestes ferrugineus* (left) and adult of the flat grain beetle, *Cryptolestes pusillus* (right) (with permission)

Cryptolestes ferrugineus (Stephens) and *C. pusillus* (Schonherr)

Description: The adults are very small, about 2.5-mm-long, elongate, very flat, light brown beetles (Fig. 6.4). The head and prothorax are very conspicuous and account for half the body length. The antennae are filiform and in males are very long (at least 1.5 mm), often longer than half the length of the body; in females, the antennae are usually a little shorter than half the body length. The adults walk with a characteristic swaying movement. Mixed populations of more than one species of *Cryptolestes* often occur.

Being small and highly flattened, they are able to enter very small cracks and crevices in grains and enter otherwise well-packaged processed food. Eggs (up to about 200 per female) are laid on or amongst the infested product. The life cycle of *C. ferrugineus* at 75% r.h. varies from 69–103 days at 21 °C to 17–26 days at 38 °C; below 50% r.h. mortality is high. Optimum conditions are 33 °C, 70% r.h., when the mean duration of the life cycle is 23 days. This species can survive overwintering in temperate climates (Navarro et al. 1991). *Cryptolestes pusillus* prefers a higher humidity than *C. ferrugineus*. Development at above 50% r.h. can take place between 17 and 37 °C. Under conditions of 33 °C, 80% r.h., the life cycle takes 27–30 days (Navarro et al. 1991).

Family Dermestidae

There are approximately 700 species worldwide that are members of the family Dermestidae. They can range in size from 1 to 12 mm. Key characteristics for adults are round oval-shaped bodies covered in scales or setae. The (usually) clubbed antennae fit into deep grooves. The hind femora also fit into recesses of the coxa. Larvae are scarabaeiform and also have setae. Dermestids have a variety of habits; most genera are scavengers that feed on dry animal or plant material, such as skin or pollen, animal hair, feathers, dead insects and natural fibres.

***Trogoderma Granarium* Everts, Khapra Beetle**

T. granarium larvae feed on a wide variety of stored products and dried foods including cottonseed meal, dried fruits, ground nuts and coconuts. They prefer whole

Fig. 6.5 Adult of the khapra beetle, *Trogoderma granarium* (with permission)

grain and cereal products such as wheat, barley and rice, but larvae have been recorded on the following: oats, rye, corn, dried blood, dried milk, fishmeal, ground nuts, flour, bran, malt, flax seed, alfalfa seed, tomato seed, pinto beans, blackeyed cowpeas, sorghum seed, grain straw, alfalfa hay, noodles, cottonseed meal, dried fruits, lima beans, coconuts, garbanzos, lentils, powdered yeast and many other products as well (Lindgren and Vincent 1959; Lindgren et al. 1955). This pest is cosmopolitan in distribution. It is a major pest of wheat in Indian subcontinent and subject to quarantine regulations in most developed countries. The origin of this species is stated to be India and was first reported from the states of Punjab and Haryana. Adult Khapra beetles have wings (Fig. 6.5), but apparently do not fly and feed very little. Mated females live from 4 to 7 days, unmated females from 20 to 30 days and males from 7 to 12 days.

Mating occurs about 5 days after emergence, and egg laying begins almost immediately at 40 °C. Egg laying may begin at 1–3 days at cooler temperatures, but no eggs are produced at 20 °C. Eggs hatch in 3–14 days and the female lays an average of 50–90 eggs that are loosely scattered in the host material. Complete development from egg to adult can take 26–220 days, depending upon temperature. Optimum temperature for development is 35 °C. If the temperature falls below 25 °C for a period of time or if larvae are very crowded, they may enter diapause. They can survive temperatures below −8 °C. In diapause, the larvae can moult but are inactive and may remain in this condition for many years (Dobie et al. 1985). Development can occur at a relative humidity as low as 2%. High relative humidity may be the limiting factor in the survival of introduced Khapra beetles (Howe and Lindgren 1957).

Fig. 6.6 Adults of the larger cabinet beetle, *Trogoderma inclusum* (with permission)

Trogoderma Inclusum LeConte, Larger Cabinet Beetle

T. inclusum is a well-known pest of insect collections and has been recorded infesting dried milk. The larvae will also develop on a variety of plant materials like grain, seeds of many kinds, nuts and dried fruits (Hinton 1945) (Fig. 6.6). Adult females deposit up to 45 eggs within the food source and these hatch in 8–12 days. The larval stage lives about 5 months and moults every 10–14 days. The entire life cycle is completed in about 6 months and adults live up to 1 month afterward to mate and lay eggs. Adults are mottled or bi-coloured in appearance, oval in shape, with clubbed antennae and are 3.5 mm in length. Adults may be distinguished from other *Trogoderma* species by the presence of a distinct notch in the inner margin of the eyes. Larvae are cream coloured with tufts of hair and spines present, typical of the *Trogoderma* and are also called the mottled dermestid (Navarro et al. 1991).

Mycetophagidae

Members of this family are mostly fungus feeders, those associated with stored products live in damp and mouldy commodities. Although they are not considered serious pests, they serve to indicate the presence of damp and mouldy storage conditions.

***Typhea stercorea* (L.)** The hairy fungus beetle.

Adults *T. stercorea* are 3-mm-long, oval, brown, flattened, and have hairy elytra with parallel lines of fine hairs. It takes 21–33 days for the species' eggs to hatch and the eggs are laid or loosely attached to grain. The larvae are able to move easily and the adults can fly (Fig. 6.7). This species infests grain and may be confused with minute mould beetles. The species eats stored products such as mouldy cereal, tobacco, peanuts and hay, and also eats fungi that grow on damp food. It may infest cereal grains, wheat, barley, oats, bran, rice, flour, bread, dried fruit, mouldy plant and animal materials.

Fig. 6.7 Adult of the hairy fungus beetle, *Typhaea stercorea* (with permission)

Family Nitidulidae

About 155 genera and 2150 species of nitidulidae are known, most of which feed on the sap of trees, flowers, fermenting fruit, fungi or carrion. They are known pests in date orchards and carry the infestation to dates in stores. The adults are flattened ovate to oblong beetles, 2–5 mm in length. In most stored product species, the elytra are shortened, leaving two or three segments of the abdomen exposed (Fig. 6.8). They are light brown to black in colour, and many species have yellow or red markings on the elytra. The antennae are 11-segmented, the last three segments forming a compact oval club. Identification to species is often difficult and genitalia preparations may be needed. The larvae may grow to about 6–7 mm long and are elongate, parallel-sided and slightly flattened.

Carpophilus hemipterus (L.) (Fig. 6.9) and *C. dimidatus* (F.) are cosmopolitan. *Carpophilus maculatus* (Murray) has been recorded in the Indo-Australian region and Pacific Islands, and in Ghana, Nigeria and Zambia, while *C. obsoletus* (Erichson) is widespread in the tropics and subtropics. *C. hemipterus* has distinctive yellow spots on its elytra.

Carpophilus dimidiatus (F.), *C. hemipterus* (L.), *C. maculatus* (Murray) and *C. obsoletus* (Erichson) occur as pests in both in the field in date plantations and

Fig. 6.8 Adult of the dried fruit beetle, *Carpophilus hemipterus* (with permission)

Fig. 6.9 Adult of the corn sap beetle, *Carpophilus dimitiatus* (with permission)

stores. Many other species of *Carpophilus* are field pests and also occur incidentally in stores. *Carpophilus* spp. persist and become a problem in stored commodities that have a relatively high moisture content such as dried fruit, cocoa and oil cake and in other commodities such as cereals that have been poorly dried. *C. hemipterus* can be an important pest on dried fruit, especially if it is mouldy. Therefore, *Carpophilus* species act as indicators of damp, mouldy conditions if they persist long after harvest.

Each female (under favourable conditions) lay on average about 1000 eggs which hatch in 2–3 days. The larval stage lasts 6–14 days during which time it moults three times, feeding in or on the stored product and on any mould present. It does, however, have difficulty entering undamaged products. The pupa is formed within the commodity or on the surface of bags and lasts between 5 and 11 days. The adults are active flyers, and flight distances of up to 3 km have been reported. The adults can live for a year but 3 months is more normal. Many generations can occur in a year due to the insect's short generation time. In *C. hemipterus,* the generation time varies from 42 days at 18.5 °C to 12 days at 32 °C, when the r.h. is above 70% (Dobie et al. 1985).

Family Silvanidae

Members of this small family are found under the bark of trees. They are generally 2–4 mm long, flattened and parallel-sided. Most species feed on plant materials, but some are associated with wood-boring beetles. A few are associated with stored products as secondary pests. Of these, the saw-toothed grain beetle, O*ryzaephilus surinamensis* (L.) and the merchant grain beetle *O. mercator* (Fauvel) are the most important. Adults of both species are slender, flattened dark brown beetles 2.5–3.5 mm long. Both larvae and adults attack grain, cereal products, nuts, copra, dried fruits, botanical drugs, tobacco, candy, dried meats and other commodities (Perez and Nelson 1975).

Oryzaephilus Surinamensis **(L.) Saw-Toothed Grain Beetle (Fig. 6.10)**

Adults are small (2.5–3.0 mm), brown, dorsoventrally compressed elongated beetle, recognizable by tooth-shaped serrations along sides of the prothorax.

It is able to survive on a wide range of foodstuffs including cereals and cereal products, dried fruit, oilseeds and oilseed products but will not breed below 17.5 °C or above 40 °C. Females lay an average of 375 eggs, at a rate of 6–10 per day over a period of 2 months at 30–33 °C but egg laying may be prolonged and females may live for over a year at lower temperatures. Development from egg to adult ranges from 20 days at 30–35 °C and 70–90% r.h. to 80 days at 20 °C and 50–60% r.h. The optimum conditions for development are 32.5 °C and 90% r.h. at which a population can multiply itself 50 times in 1 month. If initial storage temperature is

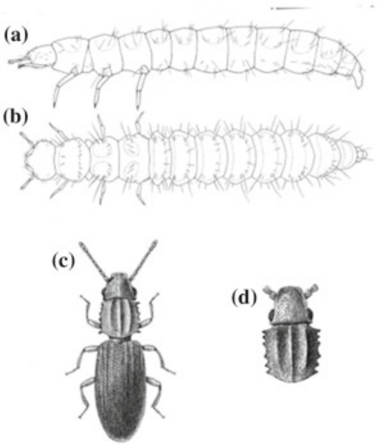

Fig. 6.10 Larva and adult of the saw-toothed grain beetle, *Oryzaephilus surinamensis* (a and b: larvae, c: adult), and adult of the merchant grain beetle, *O. mercator* (d) (Gorham 1991)

near to 35 °C and heat loss is slow enough for product temperature to remain above 25 °C for 3 months, the population is capable of increasing 10,000 times in this period. The larvae are generally free living, although they enter the seeds from time to time to feed particularly on the germ. However, they are unable to attack sound seeds (secondary pests). The adults of *O. surinamensis* do not fly. Dust encourages the development of large populations (Navarro et al. 1991). *O. surinamensis* is cosmopolitan in both tropical and temperate regions. An extremely serious pest in many stored products, causing heating in cereal grains and contamination of packaged products. The Far East is a serious pest of rice.

Oryzaephilus mercator Fauv., The Merchant Grain Beetle

O. mercator is also cosmopolitan but is more abundant in dried fruit, oilseeds and oil cake than in cereal grains. It is less common in temperate regions due to its sensitivity to low temperatures. The merchant grain beetle can be differentiated by the narrower head margin behind the eyes. Both the larval and adult stages of the merchant grain beetle attack all foods of vegetable origin; their preferred foods are oilseed products such as nuts and sunflower seeds. The habits and development of the two species are similar. The merchant grain beetle, however, is less cold tolerant and lays only about one-half to two-thirds as many eggs as does the saw-toothed grain beetle. The adult merchant grain beetles are strong flyers and may originate from other areas; they also are introduced into new grain from contaminated grain. Adults live an average of 6–10 months, but can live as long as 3 years. The females lay between 43 and 285 eggs during their lifetime. Eggs are dropped loosely among grain kernels or tucked into a crevice in a kernel. The tiny eggs are slender and white, and hatch in 3–5 days when environmental conditions are optimal (26.7–35 °C). The larvae emerge and crawl freely above the grain to feed on broken kernels. Larger larvae may tunnel into kernels to feed. Larvae mature in about 2 weeks and construct cocoon-like coverings by joining together small grains or pieces of grain. Within these structures the larvae pupate. The pupal stage lasts about a week. Total development from egg to adult requires about 3–4 weeks.

Ahasverus advena (Waltl), The Foreign Grain Beetle (Fig. 6.11)

This beetle is approximately 2 mm in length. It can be distinguished chiefly by slight projections or knobs on each front corner of the pronotum, and its club-shaped antennae. The larvae are worm-like, cream coloured and often reach a length of 3 mm before pupating into darker adults. Males and females are identical in appearance both as larvae and adults. The adult is usually reddish brown, or sometimes black. The foreign grain beetle is found in tropical and temperate regions, and is a scavenger feeding on moulds, dead insects and damaged foodstuff; the beetles are uncommonly found in figs. It can complete development at temperatures between 20 and 35 °C.

The beetle can only survive if relative humidity exceeds 70%, so it emerges in higher humidity conditions. Its diet is entirely fungi. It can often be found in grain storage facilities, where it feeds on the mould growing on the products. It can be found in other moist locations such as the walls of houses around plumbing systems. The adult female can begin laying eggs around 3–4 days after emerging from the pupa. It can lay up to 8 or 12 eggs per day, but generally produces 1–4. Eggs are laid singly or in clusters of two or three, and they hatch in 4–5 days. The larval stage is completed in 11–19 days, and pupation takes 3–5 days. Larval development takes longer in dried conditions. Mated males have an average life span of 159 days, and mated females live about 208 days. Unmated beetles live longer, males up 275 days and females up to 300.

Fig. 6.11 Adult of the foreign grain beetle, *Ahasverus advena* (with permission)

Family Tenebrionidae

This is a very large cosmopolitan family. Of some 80 species recorded on stored products, less than 10 are major secondary storage pests (do not attack the whole beans). Two species that attack cocoa beans are the rust-red flour beetle and the confused flour beetle. They can develop large populations causing heating and moisture migration within the cocoa bulk, as well as damage to cocoa beans originally attacked by primary pests. Most are brown or black elongated beetles with flattened dorsoventrally. The larvae are active, cylindrical, yellowish in colour, and commonly termed wireworms.

Tribolium castaneum Herbst, Rust-Red Flour Beetle

Adults are small (2.3–4.4 mm), reddish brown and have antennae with three segmented club. A close relative is *T. confusum* that has antennae that broaden towards tip but without club (Fig. 6.12). It is a cosmopolitan but particularly serious secondary pest of stored cereals, cereal products, legumes, oilseeds, nuts, spices and cocoa beans in warmer climates. Quinones secreted by the adult particularly at high population densities produce an unpleasant taint on the produce. The eggs are usually covered with adhering particles of cocoa flour which are laid among the substrate. The cylindrical white to yellow larvae and the pupae may be found living freely in the food medium. Newly emerged adults mate and females start to lay within about 4 days. Adults live for about 9–14 months and females produce up to 18 eggs per day with oviposition decreasing with age (Navarro et al. 1991). Temperature limits for life cycle are 20 and 40 °C but optimum is about 32 °C. Development from egg to adult takes 22 days at 34 °C and 72% r.h. and 50 days at 24 °C 76% r.h. The adults fly readily under warm conditions but distribution of the

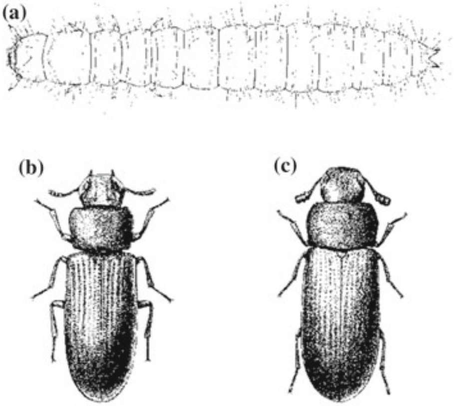

Fig. 6.12 Larva and adult of the confused flour beetle, *Tribolium confusum* (a: larva, b: adult), and adult of the rust-red flour beetle, *T. castaneum* (c) (Gorham 1991)

pest depends mainly upon passive transport in trade. Aggregation is often concentrated at the grain surface (Navarro et al. 1991).

Tribolium confusum J. du Val. Confused Flour Beetle

It is similar to *T. castaneum* with no flying ability but distinguishable by uniform broadening of antennal segments instead of three segmented terminal clubs (Fig. 6.12).

It is an important secondary pest of cereals and other products including cocoa beans. This species has a very similar life cycle to *T. castaneum* but under identical conditions of temperature and humidity it develops more slowly. Also, it is less susceptible to low temperatures. Therefore, although it is cosmopolitan in distribution and can be found together with *T. castaneum*, it tends to replace *T. castaneum* in temperate regions and is less common than *T. castaneum* in the tropics (Dobie et al. 1985). The confused flour beetle *T. confusum* and the red flour beetle *T. castaneum* are sometimes found in dried fruits (Perez and Nelson 1975).

Family Trogossitidae

Tenebroides mauritanicus (L.) The cadelle beetle

The cadelle is the only member of the Trogossitidae family that occurs in stored products. It damages grains, oilseeds, nuts, flour and other grain products, dried fruit and spices (Dobie et al. 1985). A slim, flat, 5–11-mm-long beetle, dark brown to black; ventral side, antennae and legs are red-brown. A particular feature is the head distinctly separated from rest of the body (Fig. 6.13). The larvae prefer the germ of the cereals. The last instar may bore into soft wood top to create a pupation chamber. In the tropics, it is found in mills, silos and warehouses, on grain, mill products and feeds. Irregular borings are found in kernels; germs are preferred.

The dirty-white larva has a black head, behind this a black shield, two black hooks at the end of the body and long body hairs. It has a cosmopolitan distribution and is a minor pest of stored products. It has been found associated with a wide range of commodities like oilseeds and their products, cereals, also found in mills, granaries and storehouses. This species is commonly called as ‘Cadelle’ or ‘Yellow mealworm’ or the Bolting cloth beetle because of its common habit of cutting bolting-silk-net cloths and redressing machines in flour mills. A female may continue laying eggs for a period of 3–15 months and on an average a single female lay about 500 eggs during this period. However, it has been reported that a female is capable of producing over 1700 eggs. The eggs are elongated, cigar-shaped, white, measuring 1.5–2.0 mm in length.

The egg hatches in 1–2 weeks. The larva is easily recognizable due to its greater length (3.4–8.63 mm) and is thus one of the largest grain-infesting insects. It is pale

Fig. 6.13 Adult of the Cadelle, *Tenebroides mauritanicus* (with permission)

white or greyish white in colour with prominent black head, thoracic shield and the two short dark horny projections at the posterior end of the body. The rate of the development of larva is very slow (10–20 months) and under unfavourable conditions of food and climate, the larval period may last for 3–4 years. Pupation takes place either inside the infested grain or between some grains fastened together. Both the larvae and adults can live for considerable periods without food.

Family Phycitidae—Lepidoptera (Moths)

Subfamily Phycitinae

About 70 species of moths in the families Pyralidae, Tineidae, Oecophoridae and Gelechiidae are associated with stored products. However, only the tropical warehouse moth (Almond moth) *Ephestia cautella* (Walker) (Pyralidae), the Mediterranean flour moth, *Ephestia kuehniella* (Zeller) (Pyralidae), the Tobacco or warehouse moth, *Ephestia elutella* (Hubner) (Pyralidae), the Indian meal moth, *Plodia interpunctella* (Hubner) (Pyralidae), the rice moth *Corcyra cephalonica* (Stainton) (Pyralidae) and the Angoumois grain moth, *Sitotroga cerealella* (Olivier) (Gelechiidae) and *Spectrobates ceratoniae* (Zeller) (Pyralidae) are considered widely distributed and major pests of stored products.

Adults of stored product moths have two pairs of fragile wings covered with scales, and often with delicate colourings and markings, and have coiled mouthparts forming a coiled tube (suctorial proboscis) with which they drink. Their bodies and legs are also covered with scales, which rub off easily when touched. The adults do not feed in store; live for about only 1 week and are the reproductive stage in the life cycle. The damage to stored products is caused by the larvae (caterpillars).

Ephestia cautella Walker—Tropical Warehouse Moth, The Dried Current Moth, Almond Moth or Fig Moth

Dried fruits are the preferred food of *E. cautella*, but it is also a serious pest of a wide range of commodities especially cereal flours and cereal products, nuts, dates, oilseeds, cocoa beans and spices. Feeding by the larvae is the main type of damage, but frass and silk-filled galleries cause significant contamination of the infested commodity. The larvae preferentially feed on seed germs and cause reduction in seed viability and quality. It occurs outdoors on figs, dates and probably other hosts which act as reservoirs for infestation in storage.

E. cautella is found throughout the tropics, subtropics and warmer regions of the world. It is absent from more temperate regions where other species such as *E. elutella* and *P. interpunctella* are dominant. However, it can be a serious pest in heated storage facilities in temperate regions. Females produce sex pheromone to attract potential mates for copulating. In heavy infestation, the mature larvae leave the produce in search of pupation sites, such as the walls of the store or spaces between bags. The dispersal behaviour is mediated by a pheromone secreted from the larval salivary glands. The adults are most active during the twilight hours and rest or hide during daytime.

The forewings of the adults are greyish brown with indistinct wavy markings. Both wings are rounded and the hind wings are fringed with pale border. The wingspan and body length are about 15–20 and 10 mm, respectively. It is very difficult to distinguish this moth from closely related species unless the genitalia are dissected and examined under the microscope (Fig. 6.14).

Mating takes place shortly after emergence and egg laying starts 24 h later. Most of the eggs are laid during the first 4 days and hatch in about 4 days between 10 and 38 °C, the optimum conditions being 25 °C and 80–90% r.h. A female can lay

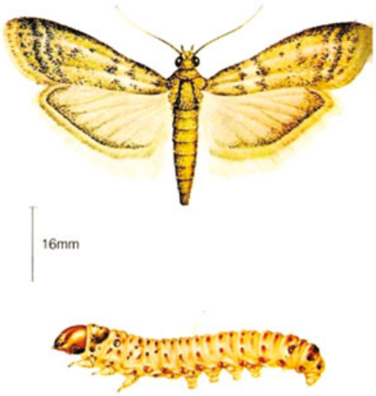

Fig. 6.14 Adult (top) and larva (bottom) of the tropical warehouse moth, *Ephestia cautella* (with permission)

between 114 and 350 eggs. Newly hatched larvae wander in search of food and are able to penetrate very small cracks in packages. They spin silk webbing which they attach to the food substrate forming a loose tunnel in which they develop. The contamination caused by webbing is often more serious than the actual damage caused by larval feeding on the product. The larvae have five instars and when fully grown is 12–14 mm long. Before pupating, the larvae enter a wandering stage when they leave the food substrate and search for suitable nooks and crevices for pupation. The pupal period lasts 7–12 days. The life cycle is from 1 to 3 months under field conditions depending on climate and food substrate, and in the laboratory under constant temperature/humidity conditions is 6 weeks at 24 °C and 75% r.h. (Navarro et al. 1991).

Ephestia elutella (Hübner), The Tobacco Moth, the Warehouse Moth, Cocoa Moth

E. elutella can cause significant damage in the production and storage of tobacco. This species infests cereal, fruit, shelled nuts, cocoa beans, fish, spices and tobacco. It is a common stored product insect in temperate and tropical regions. The adult *E. elutella* has brownish grey forewings crossed with two light bands (Fig. 6.15). The hind wings are paler and plain grey. The wingspan is 14–20 mm. Adult female lays 120–150 eggs on or near the products. Eggs hatch within 10–12 days. Life cycle takes 50–90 days under optimum temperature conditions. Young larvae are a creamy-white colour with dark spots on their sides. Larvae pass through 4–5 moults to attain full growth when they are 10–12 mm long. The larvae go to diapauses stage throughout the winter before pupation. The pupae are light brown turning black before the adult emerges.

Plodia interpunctella (Hübner), The Indian Meal Moth

P. interpunctella feeds on many stored products and is found in the home or in grocery stores worldwide and particularly in warm climates. The larvae are general feeders, as they can be found in grain products, seeds, dried fruit, dog food and spices.

Eggs of *P. interpunctella* appear greyish white and range in length from 0.3 to 0.5 mm. Eggs are oviposited singly or in clusters, and are generally laid directly on

Fig. 6.15 Adult of the tobacco moth, or warehouse moth or cocoa moth, *Ephestia elutella* (with permission)

Fig. 6.16 Adult of the indianmeal moth, *Plodia interpunctella* (with permission)

the larval food source. There are five to seven larval instars. Their colour is usually off-white, but has been observed to be pink, brown or almost greenish, depending on the food source. The mature larvae are about 12.5 mm in length. The larvae pupate either in a silken cocoon or unprotected. The pupae are 6–11 mm and are pale brown in colour. Pupation takes place away from the infested material. In fact, the late instar larvae can travel such distances that they are often mistaken for clothing pests. Within the pantry, the small larvae often climb to other shelves before pupating. This misleads people trying to find the source of the infestation.

Adults are a common sign of an infestation (Fig. 6.16). Adults do not feed. However, even though not necessary for egg production, adults have been reported to be interested in fruit juice and sugar baits. Adults have wingspan of about 16–20 mm. The forewings of this moth are reddish brown with a copper sheen on the outer two-thirds and grey on the inner third. At rest, the wings are held roof-like over the body. The head and thorax of the moth appear grey and the posterior brown, with a coppery sheen. The damage to stored products due to the contamination by *P. interpunctella* exceeds the amount of food eaten by the insects.

Ephestia calidella (Guenée) *Cadra calidella* (Guenée), The Date Moth

E. calidella is a pest of stored dried fruit, particularly dates, and of carobs. Unlike *E. cautella*, however, both attack appropriate crops before harvest (Cox 1974; Prevett 1968). The importance of *E. calidella* comes from that it attacks the ripening and/or fallen date fruits, and may fly into stores or be carried there with the product (Fig. 6.17).

E. calidella could develop and reproduce in somewhat wide conditions as a pest in the field and warehouse. In addition, the incipient diapause elevates the importance of this pest, since provision of a diapause enables a species to survive periods of extreme conditions and act as a mechanism to synchronize adult emergence. Diapause may also protect larvae from pesticides used in their control. On the other

Fig. 6.17 Adult of the date moth, *Ephestia calidella* (with permission)

hand, for control purposes, especially in warehouses, it might be possible to deter oviposition by increasing incidence of diapause early in the season or to reduce success in overwintering by reducing the incidence of diapause late in the season.

Laboratory observations were made on the development and reproduction of *E. calidella* at six different conditions of temperature and light, approximately simulated to the environmental phases in the field. Conditions for lowest mortality and shortest developmental period were 29 °C and 16 h photoperiod. The developmental periods, in general, were completed without incidence of diapause at all conditions above 20 °C/14 h L (light), though it was the longest (51.79 days) under this regime. The critical condition for induction of diapause was 15 °C/12 L; however, survival of larvae was good in such conditions. At 10 °C/10 L, there was no egg hatched and all neonates larvae which transferred to this regime died soon; later larval stages could survive and developed up to last instar and eventually entered diapauses (Alrubeai 1987).

Ephestia figulilella Gregson (*Cadra figulilella*), The Raisin Moth

E. figulilella has a nearly cosmopolitan distribution. The larvae attack various drying and dried fruits, fallen figs and damaged or mouldy clusters of grapes on vines. Raisins are attacked until they become too dry. Other recorded food includes cottonseed cake, cacao beans, cashew kernels and fallen mulberries. Adults' wingspan is 12–20 mm (Fig. 6.18). There are about three overlapping generations per year and a partial fourth. Larvae are white with four rows of purple spots along the back. Full-grown larvae are about 10 mm long (Perez and Nelson 1975).

These insects live and develop primarily out-of-doors; they are often brought into storages with infested commodities. Female raisin moths deposit eggs on all common varieties of drying and dried fruits. From egg to adult, the elapsed time is about 43 days at 28.3 °C. At that temperature, eggs hatch in 3–6 days, and larvae reach full growth in about 32 days. Larvae moult several times, usually six, with a range of from four to eight times. In raisin storages, any larvae that escape fumigation continue to feed, and in the spring they pupate and emerge. In vineyards, most of the larvae pass the winter in cocoons in the upper few inches of soil near the vine trunks and along under the wires or under the rough bark of the grapevines. In fig orchards, many larvae overwinter in a 15 cm band of soil around the tree trunks. The overwintering larvae pupate in the spring. The pre-pupal period lasts 1 day and the pupal period, about 10 days. Emergence of the adult moths begins in April and

Fig. 6.18 Adult of the raisin moth, *Ephestia figulilella* (with permission)

reaches a peak in May. No adults or eggs are found during winter. The males live for an average of 11 days and the females for 16 days. The raisin moth is a prolific insect. In summer, mated females provided with water averaged 351 eggs. The record was 692. Most eggs are laid in the first few hours of darkness during the daily flight period. On warm nights, these moths are in the air from about one-half hour after sunset until sunrise, chiefly during the earlier part of that period (Perez and Nelson 1975).

Spectrobates (Ectomyelois) Ceratoniae (Zeller), Carob Moth

The geographical distribution of *S. ceratoniae* is from the Mediterranean basin to Iran, South Africa, Australia and the Americas. *S. ceratoniae* is polyphagous. In the field, the pest develops mainly in *Acacia farnesiana* Linnaeus, carobs, dates and figs. In storage, it infests almonds and diverse nuts (Navarro et al. 1986). The adult moth is about 0.8–1.0 cm in length, dark grey, and the forewings bear two transverse whitish stripes (Fig. 6.19). The fully grown larva is 16–18 mm long, pink, and has a brown head with segmental protuberances brown head that bear small setae.

Females of the first generation lay their eggs on the developing pods of *A. farnesiana* and of carobs, later ovipositing also on dates, figs and citrus fruit. A female lays 100–350 eggs in about 1 month, and it fecundity being affected by the host plants. The hatched larvae enter into any available openings or cracks in the fruit, wherein they feed without harming the seeds. They often remain there even after harvest, thus invading storage facilities. The pest develops (especially in storage) throughout the entire year, pupating where they had fed. During summer, the pest oviposits on citrus fruit, preferring grapefruits, especially when infested by mealybugs, to whose honeydew the moths are attracted, or on fruits that touch each other. The larvae burrow into the fruit but cannot complete their development and die. Almonds are attacked only in mid-summer, after their hulls had split and/or they had become. The pest completes 4–5 annual generations, each requiring 1.5–5.0 months. *Acacia* pods and left-over almonds serve as overwintering sites, within which the larvae hibernate. Females attract males by a pheromone that is usually produced from about midnight till dawn, and most oviposition occurs right after sunset.

The larvae are serious pests of dry almonds, carobs, dates as well as various nuts in storage (Navarro et al. 1986). Damage is due to burrowing within these fruits, with attendant webs, frass, moults and moulds. As the pest can live in stores the year around, the longer the certain infested commodity is stored, the greater the injury. In the field, *S. ceratoniae* may be a pest of citrus, figs and dates. Injury is due to larval burrowing around the calyx (button), which causes premature yellowing and fruit drop (up to 10%). Infested fruits secrete a sticky gum that kills the larvae. Thus, the carob moth is a fruit moth (Gerson and Applebaum 2004).

Fig. 6.19 Adults of the carob moth, *Spectrobates (Ectomyelois) ceratoniae* (with permission)

Astigmata: Family Carpoglyphidae

Carpoglyphus lactis (Linnaeus) The Dried Fruit Mite

C. lactis is known as the most important pest species from dried apricots among the stored product pests. According to Güldali and Çobanoğlu (2011), the duration of immature stage is 5–12 days in the different combinations of temperature and relative humidities. Increases in temperatures and relative humidities accelerated life cycle. Adult longevity ranged from 11 to 68 days in the different combinations of temperature and relative humidities. The longest longevity of *C. lactis* was 68 days for females and 67 days for males at 18 °C and 65% r.h. The duration of oviposition period is 6.8–4.5 days in different temperatures and relative humidities. Average egg laying capacities of females were ranged from 26.62 to 41.23 depending on the temperatures and relative humidities. The highest number of *C. lactis*'s eggs was obtained as 41.23 at 28 °C and 80% r.h. Çobanoğlu (2009) collected samples of apricots during 2000–2003 from the most important apricot producing areas (Malatya and Elazig) and the exporting centre (Izmir). A total of 16 mite species belonging to 13 genera and 11 families were identified during the surveys. The pest species *C. lactis* was the most abundant species recorded (69.14% of samples). The highest infestation value recorded was 15,534 mites per kg (Çobanoğlu 2009).

Massive and frequent infestations of dried fruit imported from the Mediterranean region by *C. lactis* has been found in 180 samples taken from supermarkets, 13% were contaminated; the contamination levels ranged from 0 to 660 mites per g of dried fruit. The contamination was found in dried apricots, figs, plums and raisins. The mites were able to enter every dried fruit packing material tested, including polypropylene and aluminium foils. This indicates that mites can move from package to package in supermarkets. Mites are known as allergen producers and vectors of mycotoxin-producing fungi. These findings indicate that an increased risk of *C. lactis* contamination exists in dried fruit (Hubert et al. 2011) (Fig. 6.20).

Insect Pest Management Methods

Chemical Methods—Fumigants

Fumigants are widely used for pest elimination in all stored products that include cereals, oilseeds, pulses, spices, dried fruits, tree nuts and their processed foods, to prevent economic and quality losses due to insect pest attack. Fumigants may be used (a) as a hygienic measure during storage; (b) to provide wholesome food for the consumer; and (c) as a mandatory requirement in trade and in quarantine (Rajendran 2001). Of the 16 fumigants listed in common use some 32 years ago by Bond (1984), only very few remain today. Most of these fumigants have been

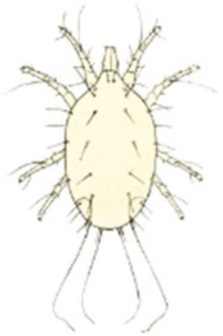

Fig. 6.20 Adult of the dried fruit mite, *Carpoglyphus lactis* (with permission)

withdrawn or discontinued on the grounds of environmental safety, cost, carcinogenicity and other factors. Methyl bromide (MB) has been phased out in developed countries since 2005 and since 2015 in developing countries, because of its contribution to stratospheric ozone depletion (UNEP 2014). Although there are exemptions for quarantine and pre-shipment purposes, as well as the possibility to apply for exemptions where no alternative exists, the applicant has to demonstrate that every effort is being made to research alternative treatments. Phosphine remains popular, particularly in developing countries; however, many insects have developed resistance to phosphine over the last decade (Cao et al. 2003; Savvidou et al. 2003).

Food industries and particularly exporters are dependent on fumigation as a quick and effective tool for insect pest control in food commodities. Following on the WTO and Free Trade Policies, trade traffic of foodstuffs across the world has been considerably increased. Consequently, fumigation for disinfesting stored food commodities has been playing a significant role. Some developed countries have adopted the approach of zero tolerance of insect pests in food commodities. At the same time, the fumigation technology that is required to obtain this zero infestation has been facing threats/constraints because of regulatory implementation and the development of resistance (Arthur and Rogers 2003).

The aim of the present section is to elucidate the practical aspects to the use of gaseous treatments for the control of oilseed crops, tree nuts and dried fruits.

The gaseous treatments

These treatments may be categorized into three groups: (a) residue-leaving fumigants that are synthetically produced volatile chemicals; (b) volatile essential oils of botanical origin; and (c) non-residual modified atmospheres (MAs). In this section, only synthetically produced volatile chemicals will be discussed.

Fumigants and their current status:

The chemical treatments discussed in this section are categorized under structural treatments and commodity fumigations. Among the synthetic chemical treatments, the list today is limited to MB, phosphine, sulfuryl fluoride, propylene oxide, ethyl formate, hydrogen cyanide, ozone and carbon dioxide.

The current most commonly used fumigants

Methyl bromide

MB has been used to fumigate oilseeds, tree nuts and dried fruits. One of the main features that makes MB a commercially desirable fumigant is its speed of action. In addition, MB has a number of additional desirable features including its recognition by quarantine authorities, and its broad registration for use; it also has good penetration ability, and the commodity airs rapidly after exposure. When considering alternatives, the above properties need to be viewed against a background of MB as a highly toxic, odourless gas with substantial ozone-depleting potential and adverse effects on a number of durables, particularly loss of viability, quality changes, taint and residues.

MB plays an important role in pest control in durable and perishable commodities and particularly in quarantine treatments. The Montreal Protocol, an international treaty developed to protect the earth from the detrimental effects of ozone depletion and signed by 175 countries, is now phasing out ozone-depleting compounds including MB on a worldwide basis. Accordingly, legislative changes have been made in different countries to control the use of MB, which has an average ozone-depleting potential of 0.4. The ban on MB currently exempts quarantine and pre-shipment (QPS) treatments, emergency uses and certain critical uses where no alternatives have yet been developed (TEAP 2006). However, these exemptions are being reviewed periodically in international meetings and they might not continue forever. In the absence of suitable alternatives, this loss of MB as a fumigant could seriously affect the protection of stored and exported food commodities from pest organisms. To combat this situation, one approach has been to accept the use of MB where no alternatives exist, but, after fumigation, to absorb the gas for recycling or to destroy it instead of releasing it to the atmosphere. There has been some limited implementation of recovery and recycling for MB, mainly in North America and Europe. Recovery and recycling systems are generally complex and expensive to install compared with the cost of the fumigation facility itself. These systems would also require a level of technical competence, not normally found at fumigation facilities. Therefore, examples of recovery and recycle in current commercial use are few.

Phosphine

Phosphine has been in use for oilseeds, tree nuts and dried fruits in solid preparations of aluminium or magnesium phosphide and in cylinders containing carbon dioxide ECO_2 FUME® or nitrogen FRISIN®. Lately, on-site phosphine generators that can release the fumigant up to the rate of 5 kg h^{-1} are available in some countries (Argentina, Chile, China and USA). Metal phosphide formulations with slow or altered rates of phosphine release have been developed and tested in Australia (Waterford and Asher 2000) and India (Rajendran 2001). Improved application techniques such as the 'Closed Loop System' in the USA, SIROFLO® and SIROCIRC® in Australia and PHYTO EXPLO® in Europe have been developed for application in different storage situations. Insect resistance is a serious concern that threatens the continued effective use of phosphine. Phosphine fumigation protocols have been revised in different countries to tackle the problem of insect resistance to the fumigant. Two major restrictions of phosphine are that it requires 7–10 days of exposure to achieve the same level of control as that of MB and that it corrodes copper and its alloys and therefore electrical and electronic items need protection from exposure to the fumigant. Phosphine also reacts to certain metallic salts, which are contained in sensitive items such as photographic film and some inorganic pigments.

New fumigants:

Sulfuryl fluoride

Sulfuryl fluoride has been used as a structural fumigant for dry wood termite control for the past 55 years, but it also can be used to disinfest flour mills and food factories (Bell et al. 1999). Although it can be used effectively for insect pest control in dry tree nuts and food grain, data are lacking regarding the effect of sulfuryl fluoride on quality of the treated commodity and persistence of residues. The fumigant is more penetrative into treated commodities than MB. Insect eggs are the most tolerant stage for sulfuryl fluoride. The relative egg tolerance can be overcome by increasing the exposure period and by raising the treatment temperature (Bell et al. 1999). Sulfuryl fluoride has been registered and used as a structural fumigant in Germany, Sweden and the USA. Sulfuryl fluoride is available under the trade name 'Vikane' containing 99.8% sulfuryl fluoride and 0.2% inert materials. Apart from the USA, China has been producing sulfuryl fluoride (trade name 'Xunmiejin') since 1983 (Guogan et al. 1999). Also, sulfuryl fluoride can be applied under reduced pressure so that the exposure period can be drastically reduced (Zettler and Arthur 2000). The fumigant was noted as highly toxic to diapausing larvae of the codling moth, *Cydia pomonella* in stored walnuts (Zettler et al. 1999). Sulfuryl fluoride is now registered under the new trade name 'ProFume®' for the protection of stored food commodities (Schneider et al. 2003). ProFume® is registered in the US to allow virtually all mills and food processing facilities to test, adapt and consider adoption as an alternative to MB. Additionally, registration coverage in EC countries for numerous milling and food processing applications is broad and increasing (TEAP 2006).

Propylene oxide

Propylene oxide (PPO) is a fumigant registered and used in the USA as a sterilant for commodities such as dry and shelled walnuts, spices, cocoa powder and nutmeats (Griffith 1999). It is a colourless and flammable liquid, and is used as a food emulsifier, surfactant, cosmetics and starch modifier. Under normal temperature and pressure, PPO has a relatively low boiling point (34 °C) and a noticeable ether odour (Weast et al. 1986). A disadvantage of PPO is that it is flammable at from 3 to 37% in air, and therefore, to avoid flammability it should be applied under low pressure or in CO_2 -enriched atmospheres. Griffith (1999), in preliminary tests on some stored product pests, indicated that PPO has insecticidal properties under vacuum conditions as a fumigant. Navarro et al. (2004b) studied the relative effectiveness of PPO alone, and in combination with low pressure or CO_2.

Ethyl formate

Currently, ethyl formate is being used for the protection of dried fruits in Australia. It has been found suitable for in-package treatment of dried fruits. It is registered in Israel for dried dates (Finkelman et al. 2010). Ethyl formate is known as a solvent and is used as a flavouring agent in the food industry. It is naturally present in certain fruits, wine and honey. In India, extensive laboratory tests against insect pests of food commodities and field trials on bagged cereals, spices, pulses, dry fruits and oil cakes have been carried out on the fumigant (Muthu et al. 1984). Studies in Australia indicate that, unlike phosphine, ethyl formate is rapidly toxic to storage insects including psocids (Annis et al. 2000).

Physical Methods

Temperature Manipulation

Low-Temperature Manipulation

Ambient air aeration

Aeration is a suitable treatment for cooling oilseeds, but not in current use for tree nuts and dried fruits. Among the oilseeds aerated are soybeans, rapeseeds, cotton seeds, sunflower, peanuts and corn. Aeration technology is used to modify the bulk microclimate to reduce or eliminate the development of harmful or damaging organisms in the stored commodity by reducing and maintaining the commodity temperatures at safe levels below humidity levels which support microflora activity. Thus, aeration helps sustain favourable storage conditions for the safe preservation of oilseeds' quality.

Aeration is the forced movement of ambient air by fan power through a grain bulk (in this context, the term grain is used in reference to any granular seed or commodity like corn, cotton seeds, soybeans, peanuts {decorticated or in-shell}, rapeseeds and sunflower) to improve grain storability. Aeration is primarily for cooling, but there are additional objectives of aeration, such as to equalize grain temperature throughout the bulk, limited drying and removal of fumigant residues and odours.

Substantial storage losses can be caused by microflora that flourish in moist grain and insects that can be destructive if preventative control measures are not taken. These losses should be considered a result of interactions between the components of the ecosystem, affected by the grain and ambient weather conditions. The interactions between damaging pests, the grain and other physical components of the system form a dynamic infrastructure, with each component continuously affecting the others. The role of aeration in this ecosystem is to uniformly ‘condition’ the stored grain to a desirable low temperature and maintains desirable conditions in the grain bulk by moving the sufficient air volumes of suitable quality through the grain mass (Navarro and Noyes 2002b).

Cooling for pest suppression—Stored product insects are of tropical or subtropical origin and require fairly high temperatures, typically 24–32°C for development. Grain-infesting insects are sensitive to low temperatures. Stored product insect development is generally stopped below 16 °C; there is little insect survival above 43 °C. Temperatures of commodities at harvest can range from 32 to 43 °C, depending on the specific crop and location.

At temperatures lower than 21 °C, population growth of most storage insects is significantly suppressed. Product temperatures in the range of 16–21 °C are considered ‘safe’ for insect management, as feeding and breeding are slow. Complete life cycles at these temperatures take 3 months or more, so insect population growth remains insignificant. Consequently, insect damage caused under these low-temperature conditions is minimal (Flinn et al. 1997).

The crucial control parameter for mite pests is not temperature, but establishing an equilibrium relative humidity (ERH) below about 65%. About 12.5% moisture content (MC) for corn at 25 °C suppresses mite development. Temperatures required to suppress the development of mites in damp grain (14–16% MC wet basis) are obtainable in temperate climates, but maintaining low uniform grain temperatures is too expensive at the bulk periphery when mean ambient temperatures are favourable for mite development. Although cooling moist grain is unlikely to prevent moderate mite infestation, aeration is expected to minimize ‘hot spots’ and heavy mite populations associated with hot spots.

Suppression of microfloral growth—Low temperatures are required to prevent mould damage in moist grain. Temperatures lower than 5 °C are needed for the suppression of most mould development (for *Penicillia* moulds, below 0 °C). Most fungi do not grow at relative humidity below 65%, which is equivalent to roughly 12.5% MC for corn, 11% for soybeans, 9% for cotton seeds and 6% for shelled peanuts at typical storage temperatures (lower MCs for oilseeds). In practice, mould growth is dependent mainly upon interstitial air humidity. Although cooling grain may not seem like an efficient method for controlling mould, at lower seed temperatures, mould damage is reduced.

Maintenance of seed and grain quality—Low kernel temperatures are desirable for better maintenance of seed and grain quality. Studies have shown that the lower the temperature (within certain limits), the longer the seeds maintain full viability. A rule of thumb (Harrington 1973) states that a seed’s lifespan in storage is doubled for each 5 °C decrease in temperature (within the range of 0–50 °C and for each 1% decrease in seed moisture (within the range of 5–14%). Seeds are commonly stored with equilibrium relative humidity from 30 to 40% with good results.

Equalization of temperature throughout the grain bulk—Because of self-insulating properties, grain placed in storage during summer harvest retains initial harvest temperatures for a long time before cool weather arrives in the fall (except for grain near bin walls, exposed conical base or the surface). As the ambient temperature drops during the cool season, the surface (and peripheral) layers of the grain become considerably cooler than the internal grain mass. It is recommended that harvest heat be removed by nighttime suction aeration as soon as ambient temperatures are 8–11 °C below internal grain mass temperatures to minimize insect activity at or near the grain surface. The initial cooling should be followed by additional aeration when generally lower ambient temperatures will allow cooling the entire grain mass below 21 °C. Natural convection currents in the grain bulk alone are sufficient to cause large amounts of moisture to 'migrate' to cooler layers or the cooler surface grain, where the air cools to 'dew point' and deposits excess moisture, slowly increasing the grain MC in the upper parts of the grain bulk. Equalizing the temperature in the grain bulk using aeration prevents moisture migration on the top layers of the bulks and prevents headspace and downspout condensation.

Prevention of biological heating of dry grain—In corn bulks where infestation is localized, insect populations develop in small pockets of corn. The lesser grain borer and the three primary weevil species found in corn in the U.S.—the rice weevil, the maize weevil and the granary weevil—are characteristic species that develop localized infestations in bulk grains, creating hotspots. Temperatures of heavily infested corn undergoing widespread heating are typically about 38–43 °C. When heavy infestations are discovered, the grain should be fumigated immediately to stop insect activity. Then, aeration should be used to cool the corn bulk.

Prevention of spontaneous heating—In warm moist grain (ERH > 70%), respiration can become very intensive due to mould development. High levels of respiration produce a phenomenon called 'spontaneous heating'. Heating of the grain bulk is detrimental to grain quality. In spontaneous heating, hot spot temperatures can easily reach 57–60 °C creating steep temperature gradients between heated and surrounding cool grain. In bulks containing oil-rich seeds such as cottonseeds, soybeans and sunflower seeds at sufficiently high moisture conditions, very high temperatures are generated and 'spontaneous combustion' can occur, starting a fire. Do not operate aeration fans if fire is detected (by the smell of smoke or burning grain in the exhaust air stream) in a grain bulk.

Chilling grain with refrigerated air

There are some storage situations where ambient air conditions are not suitable to cool grain. For these situations, refrigerated air units for chilling grain have been developed for commodities that justify the added expense of refrigerated aeration. In refrigerated aeration, ambient air is passed through the evaporator coil and a secondary reheat coil of the refrigeration unit and then is blown into the grain bulk using the existing aeration system. Passage through the secondary reheating coil adjusts the air relative humidity to 60–75% to match the target MC of the dry grain. The amount of reheating and the final air temperature are adjustable by the operator to achieve the desired aeration conditions.

High-Temperature Manipulation

High-temperature manipulation has served as a disinfestation method for dates. No other commodities among the oilseed crops, tree nuts and dried fruits are known to be treated using thermal disinfestation. For disinfestation of dates, fumigation using methyl bromide is carried out in small chambers. Dates received at the packing stations are first fumigated, then stored in cold chambers, dried, sorted and then packed. It is important to mention that fumigation using MB has an important role to remove the adults and larvae from inside the dates. If this fumigation is not carried out, low temperatures kill the insects but they remain inside the fruit.

Field infestations of nitidulid beetles serve as a source to carry the infestation into storage. MB; however, has been phased out since 2005 under the Montreal Protocol for developed countries (Non-Article 5), and since 2015 for the developing countries (Article 5), excluding treatments before shipment and quarantine (TEAP and MB 2003). Instead of using fumigants, heat treatment is now considered the most efficient way of controlling nitidulid beetles in stored dates.

In the drying process of dried fruits and nuts, temperatures are kept usually moderate (35–55 °C) to avoid commodity damage. Temperatures used for drying Madjoul dates in Israel should be kept within the range of 45–55 °C to avoid the blistering effect that separates the skin from the pulp of the fruit. However, studies that consider not only control but also emigration of nitidulid beetles from dates using heat during the drying process were lacking in the literature until now. Emigration that causes the insects to abandon the fruit is perhaps more important than killing them, since it actually disinfests the fruit, thereby improving its quality. With this approach in mind, it was considered possible that heat treatment may be effective in producing emigration and control of nitidulid beetles of dates. The effectiveness of heat in causing emigration and mortality of *Carpophilus* spp. larvae from dates were compared by Navarro et al. (2003, 2004a). The average disinfestation value obtained was greatest at exposure to 50 °C (92.3%) and this differed highly significantly from disinfestations levels at 40 and 55 °C (Navarro et al. 2004a). Previous data that reported disinfestation levels obtainable by using methyl bromide indicated that the highest disinfestation did not exceeded 90% (Donahaye et al. 1991, 1992). Navarro et al. (2004a) showed that the highest mortality values reaching 100% were obtained at 50 and 55° C.

Commercial application of thermal disinfestation:

The drying facility consisted of a polyethylene clad hothouse 40 m long × 10 m wide × 3 m high, specially prepared for large-scale commercial drying of dates. The hothouse can accommodate up to 12 rows of stacked dates positioned in parallel across the hothouse and covered over their top and sides with polyethylene liners to form drying ducts. Each row consists of 10 pallets, arranged five pallets lengthwise and two pallets across. Each pallet holds crates stacked 20 layers high with five crates (40 × 60 × 10 cm) per layer. Each crate holds 3 kg of dates, one layer deep of the cultivar Medjool. Thus, a standard row consisting of 10 pallets holds 3 tonnes of dates.

Since disinfestation and control were most effective at 50 °C, these findings were examined at a commercial drying facility. It was shown that between 1 and 2 h were required for the dates to reach the set temperature of 50 °C. During the following 3 h, dates were exposed to heated air (50 °C), after which an examination of infested dates inserted into the drying ducts, and natural infestations showed that successful emigration and control were obtained. This method produced results comparable with those obtained with MB fumigation and was suitable as a replacement strategy for infestation control. The drying facility served for heat disinfestation of Medjool, to the full satisfaction of the processors (Finkelman et al. 2006).

Integration of heat treatment in the disinfestation processes of dates:

The conventional handling practice for dates, when disinfestation is based on fumigation, permits the use of small fumigation chambers. The raw dates follow the sequence of receipt of dates at the packing stations, first fumigation, and then storage, sorting and drying. The initial quick disinfestation based on MB precedes the storage and drying for two main reasons: to prevent insect contamination in the dates and the packing station, and to limit the size of drying facilities. Drying time may be extended to a few days depending on the initial moisture content and the selected drying temperature. In the attempt to integrate heat treatment for disinfestation, the conventional sequence should be reversed to incorporate first the disinfestation in the drying process and then storage whether at ambient or cold storage (depending on the cultivar and moisture content).

An aspect that necessitated further research and development was related to the application of heat on date varieties other than Medjool. For this purpose to study the implementation of the method to other date varieties, trials were carried out on Deglet-Noor in branches and Zahidi in bulk (inside Dolev-type crates), and Halawi in factory-type boxes (Navarro et al. 2009). Laboratory and field tests of Deglet-Noor in branches were very successful and encouraging to indicate the possibility of implementing thermal disinfestation of this date cultivar in Dolev-type crates. In commercial-scale trials, temperature increase indicated the successful application of the method. Laboratory- and commercial-scale disinfestation trials failed to cause changes in the colour of the branches and the dates. In commercial-scale trials with Halawi date cultivar stored in factory boxes (12 kg), air flow rate and temperature increase rate, suitable for thermal disinfestation, could be achieved only after reduction in the quantity of the dates in the ducts (Navarro et al. 2009). These promising results encouraged expansion of the thermal disinfestation method to Deglet-Noor on branches, and Zahidi and Halawi varieties in factory boxes. The transition period to thermal disinfestation is involved in changing the accepted practices and investment in new systems. It is most desirable that these investments are carried out in a professional manner for reducing the cost of thermal disinfestation, particularly by preferring the version that uses solar energy (Navarro et al. 2010a, b).

Modified Atmospheres

The development of this technology has come about mostly over public concern for the adverse effects of pesticide residues in food and the environment. Although this method has become well established for control of storage pests, its commercial use is still limited to a few countries. More recent investigations have attempted to integrate modified atmosphere application into the twenty-first-century version of raw product and manufactured food storage and transportation (Navarro 2006).

MA is proposed to serve as the general term, including all cases in which the atmospheric gases composition or their partial pressures in the treatment enclosure have been modified to create in it conditions favourable for the control of insects. In an MA treatment, the atmospheric composition within the treated enclosure may change during the treatment period. In a CA treatment, atmospheric composition within the treated enclosure is controlled or maintained at a level and duration lethal to insects. The result in either case is the creation of a safe and environmentally benign process to manage food preservation (Navarro 2006).

Modified Atmosphere/Hermetic Storage

A type of MA that can be applied for the protection of grain is ‘hermetic storage’, also termed as ‘sealed storage’ or ‘airtight storage’ or ‘sacrificial sealed storage’. This method takes advantage of sufficiently sealed structures that enable insects and other aerobic organisms in the commodity or the commodity itself to generate the MA by reducing oxygen (O_2) and increasing carbon dioxide (CO_2) concentrations through respiratory metabolism. Respiration of the living organisms in storage (insects, fungi and grain) consume oxygen (O_2), reducing it from near 21% in air to 1–2% while production of carbon dioxide (CO_2) rises from an ambient 0.035% to near 20% (White and Jayas 2003). This environment kills insect and mite pests and prevents aerobic fungi from growing (Weinberg et al. 2008). Elevated CO_2 and depleted O_2 levels will generally maintain stored grain quality for long periods of time. Grain with excessive moisture may be invaded by lactate-forming bacteria and yeasts (White and Jayas 1993). Hermetic storage has been in use for several thousand years preserving grains in airtight pots or containers (Adler et al. 2000). The key to successful hermetic storage is air tightness and control of condensation. In modern times, storage size has increased from small family storages to large bulks representing many producers or a portion of a country's total production. In the 1960s and 70s, large above-ground hermetic storage in some African and Asian countries was discredited because of severe condensation problems particularly in metal structures (Navarro et al. 1994). Semi-underground storage has been used successfully in Argentina, Kenya and Cyprus; Australia and Israel have successfully used bunker storage systems from the 1980s. With recent improvements in materials and construction of flexible, nonporous bags and liners, a variety of size options offer protection for products from 25 to 1000 kg up to 10,000–15,000 tonnes (Navarro 2010). Commodities including cereals, oilseed grains, pulses, cocoa and coffee can be stored safely for many months, maintaining high quality and limiting moulds and mycotoxins (Navarro et al. 2010b). Plastic structures suitable for long-term storage systems, as well as intermediate storage of grain in bags or in bulk, have been developed and applied. These storage systems based on the hermetic principle are (1) Bunker storage in gastight liners for conservation of large bulks of 10,000–15,000 tonnes; (2) Flexible gastight silos supported by a weld-mesh frame of 50–1000 tonnes capacity for storage of grain in bulk or in bags; (3) Gastight liners for enclosing stacks of 5–1000 tonnes capacity termed storage cubes or Cocoons^{TM}, and designed for storage at the farmer-cooperative and small trader level or larger commercial and strategic storage facilities; (4) Silo bags of 200 tonnes capacity for on-farm grain storage directly in the field. This technique was originally used for grain silage and involves storing dry grain in sealed plastic bags; and (5) Small portable gastight containers of 25 kg–2.5 tonne, called SuperGrainbags™ which are suitable for seed storage and man-portable and bagged commodities. These structures enabled the application of modern MA technology to provide quality preservation and insect control (Navarro et al. 1994, 1998).

CA Under Normal Atmospheric Pressure

Gas supply from pressurized cylinders—CA is a modified gas composition, usually produced artificially, and maintained unchanged by adding desired gases (CO_2 or nitrogen [N_2]), supplied from pressurized cylinders or otherwise. This supplementary introduction of gases is carried out when their concentration in the sealed container drops to below the desired level.

The objective of CA treatment is to attain a composition of atmospheric gases rich in CO_2 and low in O_2, or a combination of these two gases within the storage enclosure or treatment chamber. These set concentrations are maintained for the time necessary to control the storage pests. A widely used source for production of such atmospheric gas compositions is tanker-delivered liquefied CO_2 or N_2, when the target CA gas composition is <1% O_2 or high CO_2 concentration. For large-scale application of N_2 or CO_2, vaporizers are essential. These vaporizers consist of a suitably designed receptacle with a heating medium (electricity, steam, diesel fuel or propane), a super-heated coil with hot water jacket, and forced or natural draught.

Combustible gases—For on-site generation of CAs by combustion of hydrocarbon fuel to produce a low-O_2 atmosphere containing some CO_2, commercial installations—termed exothermic gas generators or gas burners—are available. Their CA composition is designed to allow the presence of approx. 2–3% O_2 with CO_2 removed through scrubbers. Several adaptations are required for their use in the grain industry, i.e. tuning equipment to obtain an O_2 level of <1%, utilizing to full advantage the CO_2 generated and removing excessive humidity from the atmosphere generated. Combustion of propane and butane yields approximately 13 and 15% CO_2, respectively. The CA generated is more toxic than an N_2 atmosphere deficient in O_2 due to the presence of CO_2 in the MA, causing hypercarbia, which together with hypoxia are synergistic in their effect on insect mortality.

On-site N_2 generators—Commercial equipment, termed also 'pressure-swing adsorption' systems, uses the process of O_2 adsorption from compressed air passed through a molecular sieve bed. For continuous operation, a pair of adsorbers is provided that operate sequentially for O_2 adsorption and regeneration. Nitrogen at a purity of 99.9% can be obtained through regulation of inlet airflow; this method of N_2 generation is an expanding new approach to CA generation technology. Equipment is now being manufactured that is rated to supply an outlet flow of 120 m^3/h at an outlet purity of 98% N_2.

Ozone—Ozone can be generated and used to kill insects although it reacts with caulking in bins and may bleach grain. Ozone also lowers levels of microflora on seed. It is suggested for use in railcars at low temperatures and low humidity (McClurkin and Maier 2010). It is also effective in killing insects at 1800 ppm for 120 min and can be applied in specially modified augers (McDonough et al. 2010).

CA Under Altered Atmospheric Pressure

Vacuum or low pressures—In a low-pressure environment, there is a close correlation between the partial pressure of the remaining O_2 and the mortality rate. Until recently this treatment could only be carried out in specially constructed rigid and expensive vacuum chambers. A practical solution has been invented that uses flexible liners. To achieve the low pressures in the flexible liners, sufficiently low pressures (25–50 mm Hg absolute pressure) can be obtained (using a commercial vacuum pump) and maintained for indefinite periods of time by continued operation of the pump.

High-pressure carbon dioxide treatment—CO_2 treatments can be significantly shortened to exposure times that may be measured in hours using increased pressure (10–37 bar) applied in specially designed metal chambers that stand the high pressures. Because of the high initial capital investment, these high-pressure chamber treatments may be practical for high-value products such as spices, nuts, medicinal herbs and other special commodities.

Effects of CA on Product Quality

Germination of seeds—Seeds below their critical moisture content are not significantly affected at high CO_2 or low O_2 atmospheres. However, with increasing grain moisture contents, carbon dioxide-rich atmospheres could reduce the physiological quality of grain by interfering with the enzymatic activity of glutamine decarboxylase. The adverse effect of CO_2 on germination of rice, maize and wheat becomes more pronounced at temperatures higher than 47 °C and, from observations carried out so far, this adverse effect may not be detectable at all below 30 °C. Therefore, if preservation of germination is of primary importance, the use of CO_2-free low O_2 atmospheres is preferred if expected temperatures are significantly above 30 °C.

Viability of corn stored under hermetic (148 days storage) and non-hermetic (120 days storage) conditions in the Philippines did not indicate significant changes between the initial and final samples (Navarro and Caliboso 1996; Navarro et al. 1998). In same trials, viability of paddy stored under hermetic conditions did not change significantly. To test viability of wheat stored under hermetic conditions in Israel, two trials were carried out with storage periods of 1440 and 450 days only under hermetic conditions. Viability of wheat changed slightly from an initial 99–97% after 1440 days and from 97 to 91% after 450 days, respectively. In both trials, insect populations were successfully controlled and the average CO_2 concentrations ranged between 10 and 15%.

Product quality preservation—Donahaye et al. (2001) reported on quality preservation of 13.4–31.9 tonne lots of paddy, stacked in flexible enclosures and stored outdoors for 78–183 days. The quality of the paddy was compared with that of three control stacks (5.3–5.6 tonnes capacity) held under tarpaulins in the open for 78–117 days. Percent milling recovery and levels of yellowing in the gastight stacks showed no significant change. In a study on quality preservation of stored cocoa beans by bio-generated modified atmospheres, respiration rates of fermented cocoa beans were tested at equilibrium relative humidities of 73% at 26 °C in hermetically sealed containers. The O_2 concentration was reduced to <0.3%, and CO_2 concentration increased to 23% within 5.5 days. The free fatty acid (FFA) content of cocoa beans at 7.0, 7.5, and 8.0% moisture content under hermetic conditions of 30 °C remained below or close to 1.0% after 90 and 160 days of storage (Navarro et al. 2010a, b).

Insect Monitoring

Monitoring Pests with Attractants

Pheromone Lures and Traps

Traps baited with synthetic pheromones are used in food processing and storage facilities for the detection of infestation and monitoring. Numerous traps and lures have been developed for use in monitoring programmes. Lures are designed to release pheromones at a near-constant rate over time, usually for several weeks or months. The slow release of pheromone is achieved by incorporating the compound into a plastic matrix from which it is slowly released, or by the pheromone passing from a reservoir through a semi-permeable membrane. The most common trap design for flying insects employs plastic or wax-coated paper covered with insect-trapping glue on one or more surfaces. Such sticky traps usually require the insect to orient to a lure placed on or near the sticky trapping surface within a part of the trap protected from dust and debris. A variety of sticky trap designs have been used almost exclusively for monitoring storage moths, but anobiid, bostrichid and dermestid beetles, which can orient in flight to point sources of sex pheromone, can also be monitored with sticky traps. The sticky traps have a relatively short service life because they are made of paper. Non-sticky bucket and funnel traps can be used for flying insects and are considered both non-saturating, due to their large collection reservoirs, and reusable due to durable plastic construction.

Grain Probe Traps

Traps for monitoring beetle populations in bulk-stored grain do not require the use of pheromones. Traps, such as grain probe traps or pitfall cone traps, are placed at or below the surface of grain masses. They capture beetles that are simply walking over or through the grain (Barak et al. 1990). The principle behind probe trap operation is that insects move through mass of grain, walk into the holes of the probe shaft, drop through the void inside the probe and are directed by a funnel into a collection vial. Grain probe and pitfall traps are important tools in detecting the presence of beetles populations in grain masses at densities lower than what can be detected by regular sampling the grain and examining for their presence. Therefore, these traps detect insects when conventional grain sampling methods fail to do so (Hagstrum et al. 1990, 1998). A most recently developed technology for monitoring insects is a probe trap equipped with an electronic device to count insects and send counts back to a computer (Shuman et al. 1996; Litzkow et al. 1997).

To determine the effectiveness of the use of attractants in probe traps (unpublished data by Navarro 1992) installed in shelled and unshelled peanuts in a pilot plant experiments, no significant differences were observed in the presence of the attractants in the probes. Also according to Phillips et al. (2000), there is some evidence suggesting that pheromones and food attractants should not be used in devices intended for monitoring insects in bulk-stored grain. Additionally, one may increase the risk of infestation from flying beetles if synthetic aggregation pheromones are installed with traps in the grain.

Suppressing Pest Populations with Pheromones

Since the availability of the synthetic sex pheromones, the idea of trapping all the insects in a population and causing a population to be suppressed was reported in various agricultural systems by Lanier (1990). For aggregation pheromones that attract females, mass-trapping may have significant impact on a population if substantial females are removed. For sex pheromones that attract only males, a critical number of males must be removed to ensure that an effective number of females go unmated. Several studies report successful population suppression of storage moths following deployment of a high density of traps (Levinson and Levinson 1979, Suss and Trematerra 1986; Trematerra and Battaini 1987; Trematerra 1988, 1990), but typically no evaluation of the success of the treatment is conducted except for continued monitoring with pheromone traps. Pierce (1999) reported on long-term mass-trapping of *L. serricorne*, with its sex pheromone in a commercial bakery over a 9-year period, and inferred population suppression from reduced trap catch in later years of the study. Field studies such as these are difficult to perform and validate because of the lack of proper controls, the inability to carry out sufficient replications and without a means to assess the insect population independent of trapping males. Athanassiou et al. (2016) evaluated the application of mating disruption (MD) for stored product Pyralidae in a large storage facility of amylaceous products in central Greece. Pyralidae populations were monitored by using pheromone-baited sticky traps and Petri dishes containing semolina. In the infesting species *E. kuehniella*, after the application of MD, the numbers of adult males found on the pheromone-baited traps were reduced in the MD-treated facility, as compared with the untreated (UTC) facility. Similarly, the numbers of larvae in the oviposition traps were also reduced, as compared with the UTC facility. During the successive years of MD deployment, there was a noticeable decrease in the number of *E. kuehniella* male adults in the MD-treated facility, but the high-density areas, and the infestation foci, were different among years. In 2014, the majority of the male adults were found in the areas where some years earlier, during the beginning of MD, there were no individuals, or their numbers were low. This suggests that, despite the effectiveness of the method, may be a spatial displacement to other areas.

Mating disruption is another method for population suppression whereby the atmosphere of the pest's environment is saturated with a high level of synthetic pheromones; males are unable to locate and mate with females due to the elevated pheromone levels. Unmated females do not reproduce and the population declines the next generation (Jones 1998a, b). Nearly, all mating disruption research and application has been with moths. Atmospheric permeation is usually achieved by some broadcast distribution of many pheromone releasers throughout the environment that maintain stable and relatively high levels of pheromone for an extended period of time.

Current Usage

The main use of pheromones for stored product insects remains as tools for monitoring and detection, although research continues on methods to apply pheromones for control purposes. Use of pheromone-baited traps by the food industry has increased greatly in the last two decades and use patterns may have shifted slightly. However, no data was encountered of any commercial use of pheromones to control populations of stored product pests. Work on lure-and-kill methods has not progressed beyond that discussed above.

Traps are among of the most sensitive methods to detect insect pests in bulk-stored grain. However, probe traps have not been widely adopted by the grain industry. Key factors that are likely responsible for the disinterests in probe traps by the grain industry are the need for periodic servicing of the traps in the grain and the lack of knowledge on how to interpret trap catch data. Most commercial grain managers are reluctant or forbidden to enter bulk storage structures and have workers walk on the surface of the grain because of safety concerns regarding entrapment in grain, dust exposure, low oxygen exposure and other issues surrounding work in confined spaces. An alternative to manually counting insects in probe traps was recently developed. This would have reduced the labour and time required for servicing the traps. The electronic grain probe insect counter (EGPIC) utilizes a tubular probe trap body and an electronic counting device to count insects that fall through the trap (Shuman et al. 1996; Litzkow et al. 1997). Counted insects pass completely through EGPIC and are not retained; hence, no emptying of the device is required. Commercial development of EGPIC is presently underway and the manufacturer envisions coupling EGPIC units together with temperature cables permanently installed in grain storage facilities.

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Chapter 7
Microbial Biopesticides

M. E. Wakefield

Introduction

Control of stored product insects and mites in grain storage and processing facilities has been dominated by the use of synthetic chemical insecticides. Both contact insecticides and fumigants have been used, but in the past few decades concerns have been raised over the use of some of these chemical classes. In particular, the commonly used fumigant methyl bromide was shown to be an ozone depleter and its use was banned under the Montreal Protocol with only a few exemptions outside of the stored product area. There is also increasing concern over the use of other chemical classes such as organophosphates. Within the European Union the introduction of the Plant Protection Products Regulation 1107/2009, which introduced hazard based cut-off criteria rather than risk-based evaluations may further reduce the number of active ingredients that are registered for use. Added to this, there is evidence for resistance to the major classes of chemicals used for stored product insect control and it is possible that as the products that can be used become more limited, so the incidence of resistance will increase. Alternative methods for the control of storage insects and mites have examined physical methods of control e.g. cooling, drying, heat and the potential for biological control has received more interest.

Biological control agents can be divided into macroorganisms (parasitoids and predators) and microorganisms. Microorganisms that are specific to insects can be used as the basis of biopesticides. Some microorganisms such as the baculoviruses tend to be highly specific and in general will only infect a single genus or species of insect. Others, such as entomopathogenic fungi, may show a broader host range. In this chapter microorganisms that are used for the control of stored product insects and mites are described. In addition, entomopathogenic nematodes are also considered.

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Microbial Biopesticides

Microbial biological control agents are available as commercial products to control insect and mite pests in various agricultural and horticultural sectors. These biological control agents include insect pathogens (bacteria, in particular *Bacillus thuringiensis*, entomopathogenic fungi, baculoviruses, protozoa) and, in some definitions (and as used here), also include entomopathogenic nematodes. The potential for the use of these biological control agents for the control of stored product insects and mites has been reviewed previously (Cox and Wilkin 1996; Moore et al. 2000). The main advantages of these agents is that they are naturally occurring; they pose minimal risks to users, consumers and the environment due to their specificity; they fit into an integrated pest management approach and can be used with other control measures (Cox and Wilkin 1996; Moore et al. 2000). The main disadvantage is that they tend to be slow acting in comparison to conventional chemical insecticides and the specificity that reduces the associated risks, may also limit the target host and life stages that may be affected. Research to examine the potential of these organisms for control of stored product insects and mites has increased in the past decade and these findings are considered below.

Bacillus thuringiensis

Bacillus thuringiensis (B.t.) is a gram-positive bacterium that has been studied extensively for the control of a wide range of insect pests, primarily coleopteran and lepidopteran species. During sporulation the bacterium produces a protein crystal. When ingested, the alkaline PH and enzyme activity in the insect gut causes cleavage of the crystal into smaller units known as endotoxins. The toxin acts directly on the membranes of midgut epithelium cells resulting in an increase in membrane permeability followed by swelling and eventual cell lysis. The resulting ionic imbalance in the haemolymph can cause paralysis and death in some species. In other species, vegetative propagation of B.t. is possible due to the changes in the gut and haemolymph conditions, resulting in septicemia that may contribute to or cause the death of the insect. The toxins produced by B.t. have been studied extensively and more than 400 genes encoding toxins have been identified. Most of these Cry proteins have a distinctive insecticidal spectrum and therefore toxins that are effective against for example lepidopteran species may not act against coleopteran or dipteran species.

Bacillus thuringiensis is the most successful commercial microbial insecticide, and is utilized globally. The development of recombinant DNA technology significantly increased the exploitation of genes/proteins derived from B.t. from the 1980s onwards through incorporation of the genes encoding various Cry toxins in genetically modified insect–resistant crops. *Bacillus thuringiensis* has been registered for use against stored product Lepidoptera for several decades in the USA, but it is not used extensively as it is not able to control coleopteran pests (Phillips and Throne 2010). Research to identify new strains of *B. thuringiensis* with potential for control of stored product moths and beetles continues (e.g. Blanc et al. 2002; Ozturk et al. 2008; Bozlagan et al. 2010; Yilmaz et al. 2012), but more recently focus has also been placed on the mechanism of action of the toxins as a means to identify other potential control methods for stored product pests (Contreras et al. 2013).

Improved activity against coleopteran pests is a requirement for more widespread use in stored product protection. In an attempt to improve the effect of Cry3A against *Tribolium castaneum* recombinant modifications of Cry3A protoxins were prepared and tested against *T. castaneum* larvae. The modified protoxins were not as effective as the natural or the recombinant Cry3A and it was concluded that structural modifications are unlikely to improve the toxicity of this toxin for *T. castaneum* (Mostafa et al. 2013). In addition to control of insect species, the potential for B.t. to control stored product mite species has been also reported (Erban et al. 2009).

The commercial availability of transgenic maize expressing B.t. toxins potentially offers protection while the grain is in store, in addition to protection against pests in the growing crop. However, this needs to be balanced against the fact that B.t. toxins tends to have a narrow specificity which means that protection is unlikely to be found against all insect species that may be found in storage premises. Sedlacek et al. (2001) demonstrated in a laboratory study that transgenic maize varieties expressing Cry1Ab affected emergence and fecundity of *Plodia interpunctella* and emergence of *Sitotroga cerealella*. The MON810 variety of transgenic maize was found to cause significant mortality for four species of stored product moth (100% for *Ephestia elutella. E. cautella* and *P. interpunctella* and 65% for *E. kuehniella*) with *P. interpunctella* being the most sensitive of the species tested (Hubert et al. 2008a). Two transgenic maize varieties (MON810 and TC 1507) showed a significant effect on survival of and a significant increase in development time. Avoidance behaviour for the TC 1507 variety, which expresses the Cry1FA protein from B.t. var. aizawai, was demonstrated, but there was no avoidance behaviour towards the MON810 variety, which expresses the Cry1Ab protein derived from B.t. var. kurstaki HD-1(Gryspeirt and Gregoire 2012). Riudavets et al. (2006) examined the effect of transgenic rice varieties on *P. interpunctella, Sitophilus oryzae* and *Liposcelis bostrychophila.* Lethal and/or sublethal effects were found for all three species, but these were dependent on the gene expressed (Cry1B or Cry1Aa). Transgenic crops therefore may offer improved resistance to attack by some stored product species depending on the cry protein expressed, but larger scale trials are required to ascertain the level of control that could be exerted in a practical situation.

One disadvantage of the use of B.t. is the rapid development of resistance in lepidopteran pests. Resistance to B.t. was first noted in *P. interpunctella* (McGaughey 1985). Shortly after, resistance in *E. cautella* was also reported

(McGaughey and Beeman 1988). Strategies for B.t. resistance management have been considered and within the stored products area recent research has examined the potential of diflubenzuron, soybean trypsin inhibitor and chitinase (Hubert et al. 2008b) or protease inhibitors (Oppert et al. 2011) for use in control strategies with Cry proteins as part of a resistance management approach.

Entomopathogenic Fungi (EPF)

There are approximately 1000 species of fungi that are pathogenic to insects. Of these only a few species have been studied extensively, mainly due to the amenability of certain fungi to mass production and formulation. The most extensively studied entomopathogenic fungi (EPF) include *Metarhizium anisopliae* (Metchnikoff) Sorokin and *Beauveria bassiana* (Balsamo) Vuillemin. Of the species studied, only a few isolates have been commercialized for use as insect pest control agents. For example, currently the EU pesticides database lists only six isolates of EPF (two *Beauveria bassiana* isolates, two *Paecilomyces fumosoroseus*, one *Metarhizium anisopliae* and one *Lecanicillium muscarium*) that are approved for use as insecticides in the EU, with approval of two other *Beauveria bassiana* isolates pending.

In contrast to other microbial pathogens, EPF are able to cause disease in insects through contact, rather than having to be ingested. Infection of an insect can be divided into three parts: adhesion of the fungal spore to the insect cuticle, germination of the spore and penetration through the cuticle and proliferation within the host. The death of the insect is likely a result of starvation and physiological/biochemical disruption, but the production of toxic secondary metabolites may also play a role. Under appropriate environmental conditions the mycelium breaks out of the insect and spores are produced that can infect other suitable hosts.

Research to examine the potential for the use of EPF to control stored product insects and mites has expanded considerably in the past decade. The majority of the studies have focussed on laboratory experiments, but some field scale evaluation has also been reported (Cherry et al. 2007; Meikle et al. 2001; Sabbour 2012). Laboratory studies have focussed primarily on isolates of *B. bassiana* and *M. anisopliae*, but other species such as *B. brongniartii*, *Isaria fumosorosea* and *Paecilomyces* species have also been evaluated (Table 7.1). The laboratory studies have examined the effect of EPF when applied to surfaces (e.g. Cox et al. 2004; Wakefield et al. 2010), directly to the commodity/diet (e.g. Dal Bello et al. 2001; Khashaveh et al. 2011) or by direct immersion of the insects (e.g. Cherry et al. 2005; Faraji et al. 2013; Golshan et al. 2013). The range of species that have been tested has also expanded and effectiveness against coleopteran, lepidopteran, psocoptera and acarine species has been reported (Table 7.1).

Table 7.1 A summary of laboratory studies to examine the efficacy of entomopathogenic fungi against stored product insects and mites

Entomopathogenic fungus species	Stored product insect/mite species	Authors
Beauveria bassiana	*Acanthoscelides obtectus*	Searle and Doberski (1984)
		Khashaveh et al. (2011)
	Acarus siro	Cox et al. (2004)
	Callosobruchus maculatus	Cherry et al. (2005)
		Lawrence and Khan (2002)
		Lawrence and Khan (2009)
		Mahdneshin et al. (2011)
		da Paz Junior et al. (2012)
		Shams et al. (2011)
		Vanmathi et al. (2011)
	Cryptolestes ferrugineus	Cox et al. (2004)
	Ephestia cautella	Sabbour et al. (2012)
	Ephestia elutella	Liu et al. (2008)
		Liu et al. (2009)
	Ephestia kuehniella	Anagnou-Veroniki et al. (2005)
		Cox et al. (2004)
		Faraji et al. (2013)
		Bischoff and Reichmuth (1997)
		Sabbour et al. (2012)
	Lasioderma serricorne	Liu et al. (2009)
	Lepinotus patruelis	Cox et al. (2004)
	Oryzaephilus surinamensis	Cox et al. (2004)
		Frydocva et al. (1989)
		Searle and Doberski (1984)
		Wakefield et al. (2013)
	Plodia interpunctella	Bischoff and Reichmuth (1997)
		Sabbour et al. (2012)
	Prostephanus truncatus	Dhuyo and Selman (2007)
		Kassa et al. (2002)
		Smith et al. (1998)
	Rhyzopertha dominica	El-Khayat (2000)
		Mahdneshin et al. (2009)
		Moino et al. (1998)
		Padin et al. (1996)
		Rice and Cogburn (1999)
	Sitophilus granarius	Cox et al. (2004)

(continued)

Table 7.1 (continued)

Entomopathogenic fungus species	Stored product insect/mite species	Authors
		Frydocva et al. (1989)
		Hluchy and Samsinakova (1989)
		Khashaveh et al. (2011)
		Levchenko and Melashchenko (1979)
		Marcu (1990)
		Shams et al. (2011)
		Wakefield et al. (2013)
	Sitophilus oryzae	Dal Bello et al. (2001)
		El-Khayat (2000)
		Kavallieratos et al. (2012)
		Moino et al. (1998)
		Padin et al. (2002)
		Padin et al. (1996)
		Rice and Cogburn (1999)
		Zidan (2013)
	Sitophilus zeamais	Adane et al. (1996)
		Kassa et al. (2002)
		Moino et al. (1998)
		Pimentel and Ferreira (2012)
		Ruelas-Ayala et al. (2013)
		Smith et al. (1998)
		Lourencao et al. (1993)
		Teshome and Tefera (2011)
	Tribolium castaneum	Golshan et al. (2013)
		Khashaveh et al. (2011)
		Padin et al. (2002)
		Padin et al. (1996)
		Padin et al. (1997)
		Rice and Cogburn (1999)
		Wakefield et al. (2013)
	Tribolium confusum	Cox et al. (2004)
		Frydocva et al. (1989)
		Wakefield et al. (2013)
	Trogoderma granarium	Draganova et al. (2012)
	Tyrophagus longior	Cox et al. (2004)
Metarhizium anisopliae	*Acanthoscelides obtectus*	Rodrigues and Pratissoli (1990)
	Callosobruchus maculatus	Cherry et al. (2005)

(continued)

Table 7.1 (continued)

Entomopathogenic fungus species	Stored product insect/mite species	Authors
		Lawrence and Khan (2002)
		Mahdneshin et al. (2011)
	Ephestia cautella	Sabbour et al. (2012)
	Ephestia kuehniella	Faraji et al. (2013)
		Sabbour et al. (2012)
		Bischoff and Reichmuth (1997)
	Oryzaephilus surinamensis	Khashaveh and Chelav (2013)
	Plodia interpunctella	Bischoff and Reichmuth (1997)
		Sabbour et al. (2012)
	Prostephanus truncatus	Kassa et al. (2002)
	Rhyzopertha dominica	Mahdneshin et al. (2009)
		Padin et al. (1996)
		Batta (2005)
		Wakil et al. (2010)
	Sitophilus granarius	Khashaveh et al. (2008)
	Sitophilus oryzae	Dal Bello et al. (2001)
		Kavallieratos et al. (2012)
		Padin et al. (1996)
		Batta (2004)
		Zidan (2013)
	Sitophilus zeamais	Kassa et al. (2002)
		Rodrigues and Pratissoli (1990)
		Ruelas-Ayala et al. (2013)
		Ahmed (2010)
		Ekesi et al. (1999)
		Lourencao et al. (1993)
		Teshome and Tefera (2011)
	Tribolium castaneum	Padin et al. (1996)
		Khashaveh and Chelav (2013)
	Trogoderma granarium	Khashaveh et al. (2011)
Beauveria brongniartii	*Sitophilus zeamais*	Rodrigues and Pratissoli (1990)
	Ephestia elutella	Liu et al. (2008)
	Oryzaephilus surinamensis	Cox et al. (2004)
	Ephestia kuehniella	Cox et al. (2004)
	Lepinotus patruelis	Cox et al. (2004)
	Acarus siro	Cox et al. (2004)

(continued)

Table 7.1 (continued)

Entomopathogenic fungus species	Stored product insect/mite species	Authors
Isaria fumosorosea	*Sitophilus oryzae*	Kavallieratos et al. (2012)
	Callosobruchus	Lawrence and Khan (2002)
	Ephestia elutella	Liu et al. (2008)
	Plodia interpunctella	Sabbour et al. (2012)
	Ephestia cautella	Sabbour et al. (2012)
	Ephestia kuehniella	Sabbour et al. (2012)
Nomuraea rileyi	*Rhyzopertha dominica*	Padin et al. (1996)
	Sitophilus oryzae	Padin et al. (1996)
	Tribolium castaneum	Padin et al. (1996)
Paecilomyces cateniobliquus	*Ephestia elutella*	Liu et al. (2008)
Paecilomyces farinosus	*Sitophilus oryzae*	Dal Bello et al. (2001)
	Plodia interpunctella	Bischoff and Reichmuth (1997)
	Ephestia kuehniella	Bischoff and Reichmuth (1997)
Paecilomyces lilicanus	*Sitophilus zeamais*	Ahmed (2010)
Paecilomyces sp.	*Sitophilus zeamais* *Prostephanus truncatus*	Kassa et al. (2002)
Lecanicillium lecanii	*Sitophilus zeamais*	Ahmed (2010)
	Sitophilus oryzae	Dal Bello et al. (2001)
		Padin et al. (1996)
	Rhyzopertha dominica	Padin et al. (1996)
	Tribolium castaneum	Padin et al. (1996)

Isolates of EPF have different characteristics and therefore not all isolates are as effective against a particular insect species. For example, Adane et al. (1996) assessed the virulence of ten isolates of *B. bassiana* against *Sitophilus zeamais* and found that the mortality of adult insects ranged from 38 to 89% when corrected for control mortality, whilst median lethal times ranged from a mean of 2.7–8.7 days. Other examples are reported for *Tribolium castaneum* (nine isolates of *B. bassiana* with mean mortality ranging from 15.55 to 60.0% (Golshan et al. 2013)) and *Callosobruchus maculatus* (seven isolates of *B. bassiana* with mean mortality ranging from 12.24 to 100% mortality (da Paz Junior et al. 2012)) amongst others. The factors that affect the virulence of a particular isolate are not fully understood, but are likely to include the production of enzymes that facilitate penetration of the cuticle (Khan et al. 2012) and possibly the production of secondary metabolites. Overexpression of protease and chitinase genes has resulted in an increase in the virulence of *M. anisopliae* and *B. bassiana* isolates respectively demonstrating the critical involvement of these enzymes in the infection process (St Leger et al. 1996; Fang et al. 2005; Fan et al. 2007).

Studies have also shown that some species are more susceptible to infection by EPF than others. For example, Cox et al. (2004) demonstrated that for the four isolates of *B. bassiana* examined, *Tribolium confusum* was less susceptible to infection than *Oryzaephilus surinamensis* or *Cryptolestes ferrugineus*. A difference in susceptibility for *S. zeamais* and *Prostephanus truncatus* has also been shown, with *P. truncatus* showing a greater mean percentage mortality to the *B. bassiana* isolates tested (Kassa et al. 2002). *Sitophilus granarius* was found to be more susceptible than *O. surinamensis* or *T. castaneum* to the *B. bassiana* isolates tested (Khashaveh et al. 2011). It is clear that the interactions between specific EPF isolates and different insect species are complex and a greater understanding is required. Some studies have started to address this by looking at the cuticular lipids (Lord and Howard 2004) and defensive secretions (Wakefield et al. 2013) of less susceptible species.

Differences in the effect of EPF on larval and adult stages have been observed. Treatment with EPF resulted in a higher mortality of the larval stage compared to the adults for *Tribolium castaneum* and *P. truncatus* (Akbar et al. 2004; Dhuyo and Ahmed 2007), although the reverse was found in a study to examine the effect of *B. bassiana* isolates on *Trogoderma granarium* larvae and adults (Draganova et al. 2012). A reduction in egg hatch as a result of *B. bassiana* infection was observed for *Rhyzopertha dominica* and *T. castaneum*, although eggs of *O. surinamensis* and *C. ferrugineus* were not affected (Lord 2009a). It is therefore likely that treatment with an EPF will suppress population development by affecting multiple life stages, although the relative effectiveness on each stage will depend on the insect species.

The efficacy of entomopathogenic fungi used in conjunction with other materials, in particular diatomaceous earths, has been studied for several different insect species (Lord 2001, 2005, 2007a, b, c; Akbar et al. 2004; Dal Bello et al. 2006; Kavallieratos et al. 2006; Michalaki et al. 2006; Vassilakos et al. 2006; Athanassiou et al. 2007, 2008a, b; Batta 2008; Ramaswamy et al. 2009; Wakil et al. 2010, 2011; Tahira et al. 2011; Sabbour et al. 2012; Nabaei et al. 2012; Riasat et al. 2013). A synergistic effect of application of *B. bassiana* and a diatomaceous earth against adult *R. dominica* and *O. surinamensis* at all doses tested was first reported by Lord (2001). Other studies have since examined the application of *B. bassiana, M. anisopliae, P. fumosoroseus* with different diatomaceous earths and have reported improved efficacy at different temperatures and humidities. It has been shown that desiccation, which is a result of treatment with a diatomaceous earth, increases the efficacy of *B. bassiana* (Lord 2007a, b, c). Similarly other forms of stress, for example, dietary (Lord et al. 2010) and reduced oxygen or elevated carbon dioxide levels (Lord 2009b) have also been shown to increase the efficacy of *B. bassiana*.

In the UK, a succession of research projects have culminated in the development of a dry powder product based on the entomopathogenic fungus *Beauveria bassiana* and an electrostatically chargeable wax powder (Entostat™) for the structural treatment of grain stores. The research programme has progressed from the isolation and identification of EPF from insect cadavers in UK grain stores, through assessment of the host range and effective concentrations, development of mass production methods, formulation and assessment of storage stability and field scale testing (Cox et al. 2003, 2004; Wakefield et al. 2013). Recent field-scale efficacy trials of structural treatment using the formulated product has resulted in a maximum of 64, 98 and 99% mortality of the stored-product beetles *Sitophilus granarius, Oryzaephilus surinamensis* and *Cryptolestes ferrugineus* respectively 14 days after treatment (M. Wakefield, unpublished). The application for the Annex I listing of the isolate with the grain store surface treatment formulation as a representative product (Annex III) will be submitted by the commercial partners to the EU rapporteur in mid-2014, with the approval of the active and submission of the EU product dossier anticipated in 2016.

The continued research on EPF for use against stored product pests, in conjunction with a greater understanding of the mechanisms of interaction, potential sub-lethal effects (Pedrini et al. 2010) and novel application methods (Baxter 2011; Wakefield et al. 2013) means that a commercial product for stored product insect control is likely in the near future.

Baculoviruses

Baculoviruses are a large group of viruses with double stranded DNA, which are pathogenic to invertebrates and, in particular, to lepidopteran species. They are not able to act through contact and must be ingested to be effective. Once ingested the occlusion bodies produced by the baculovirus are dissolved in the alkaline environment of the midgut, releasing the baculovirus particles. This leads to infection of the gut cells, followed by the fat body tissues. The insect will stop feeding within a few days of ingestion and virus particles are released into the environment by gut expulsion and following the death and disintegration of the insect. Baculoviruses can also be transmitted from an infected female to her progeny via the egg.

Baculoviruses offer potential for the control of stored product moths. However, they tend to be host specific and therefore need to be identified from the pest species of interest. For example, tests of commercial products based on nuclear polyhedrosis virus or granulosis virus from *Spodoptera exigua, Mamestra brassicae, Cydia pomonella* or *Helicoverpa zea* resulted in very low mortality (maximum 31.4%) for *E. kuehniella* (Anagnou-Veroniki et al. 2005). However, granulovirus has been isolated from *P. interpunctella and E. kuehniella.* The *P. interpunctella* granulovirus has been well studied (Moore et al. 2000; McVean et al. 2002a, b) and has shown potential for control of this species. The effects of sublethal infection and dietary stress in combination with granulosis virus infection have been investigated for this species (Burden et al. 2002; McVean et al. 2002a, b). A research project in the US has led to registration of a product based on the *E. kuehniella* granulovirus for the control of this species in stored nuts (Vail et al. 2003).

Protozoa

Protozoa are single celled organisms that, as for the baculoviruses, enter the host by ingestion or vertical transmission from the female to her progeny. The potential of protozoa for the control of stored product insects and mites has been reviewed (Cox and Wilkin 1996; Moore et al. 2000). These reviews highlighted the potential of *Mattesia* and *Nosema* species. Building on the studies reported in these reviews, factors determining the virulence of *Nosema whitei* in *T. castaneum* have been studied and the importance of host age at the time of infection has been demonstrated (Blaser and Schmid-Hempel 2005).

Mattesia oryzaephili, a pathogen of *Oryzaephilus surinamensis* has also been found in *C. ferrugineus*. Its ability to control *C. ferrugineus* and *C. pusillus* larvae has been demonstrated (Lord 2003). It was also found that this protozoan could infect *Cephalonomia tarsalis* and *C. waterstonii*, parasitoids of *O. surinamensis* and *C. ferrugineus* respectively (Lord 2006). Infection resulted in a significant reduction in the number of progeny produced and a significant reduction in survival. The author concluded that as both parasitoid species remained alive and continued to oviposit, albeit with a reduced number of progeny, there was potential for *M. oryzaephili* and the parasitoid species to be used as independent natural controls (Lord 2006).

An indirect sandwich enzyme-linked immunosorbent assay (ELISA) was developed for detection of *M. oryzaephili* in epizootiological studies and was used to screen colonies of *O. surinamensis*, *C. ferrugineus*, *C. pusillus* and *C. turcicus*. Although the pathogen was not detected in colonies of *C. turcicus*, in laboratory colonies of *C. ferrugineus* the percentage positive ranged from 0.2 to 83.9%. The colonies with the highest percentage were on the verge of collapse. A colony of *C. pusillus* showed a prevalence of 16.5% and an *O. surinamensis* colony showed a prevalence of 7.6% (Lord 2007a, b, c).

More recently, the first microsporidian infection of *C. ferrugineus* has been reported (Lord et al. 2010). The isolate was identified as *Nosema oryzaephili* and based on phylogenetic analysis of the small subunit ribosomal DNA, designation as *Paranosema oryzaephili* was proposed (Lord et al. 2010). The isolate was most infectious to *C. ferrugineus*, but infection of *O. surinamensis, T. castaneum* and *E. kuehniella* was also demonstrated. The author also compared infection to that of *Paranosema whitei*, a microsporidia that infects *T. castaneum*, and found that *P. whitei* did not infect *C. ferrugineus* at the highest dose used. It was suggested that infectivity of *C. ferrugineus* could be used to distinguish between *P. whitei* and *N. oryzaephili,* which are morphologically similar and difficult to distinguish (Lord et al. 2010). Although protozoa have caused population crashes in laboratory cultures, the potential for these organisms to suppress naturally occurring populations remains to be established.

Nematodes

Several species of entomopathogenic nematode (EPN) are available commercially for the control of agricultural insect pests. The most studied entomopathogenic nematode species for biological control are members of the Steinernematidae and Heterorhabditidae families. The infective stage (third-stage juvenile) of entomopathogenic nematodes actively seek out their host and enter through the mouth, anus or spiracles. Entomopathogenic nematodes carry symbiotic pathogenic bacteria that are released into the haemocoel causing the death of the insect within a few days as a result of septicemia or toxemia. The species of bacteria carried by nematodes is related to the genus with *Steinernema* associated with bacteria from the *Xenorhabdus* genera and *Heterorhabditis* associated with bacteria from the *Photorhabdus* genera (Boemare 2002). The bacteria are consumed by the developing nematodes, providing a source of nutrition. The nematodes reproduce in the insect cadavers and infective juveniles are released to infect new hosts. Within the past decade the suitability of entomopathogenic nematodes for the control of stored product insects has been examined more extensively. Efficacy has been shown for different species of entomopathogenic nematode against various species and stages of stored product insect (Table 7.2).

Different studies have shown differing degrees of efficacy for the same nematode species. For example, Laznik et al. (2010) examined three strains of *S. feltiae* against *S. oryzae* and found that at 20 °C and the highest concentration tested (2000 infective juveniles per adult) a maximum of 65.3% mortality after 8 days exposure was observed. In contrast, Athanassiou et al. (2010) using the same temperature and nematode concentration found very little mortality (1.7%) with the strain of *S. feltiae* used. The selection of the strain of nematode is therefore a highly important factor in the development of an effective product.

It is also recognized that a single strain of nematode is unlikely to be equally effective against several species of stored product pest. For example, de Carvalho Barbosa Negrisoli et al. (2013) found that adult *A. obtectus* were more susceptible to infection to eight different species/strains of entomopathogenic nematode (mortality range 10–68%) compared to either adult *S. oryzae* (mortality range 2–26%) or *S. zeamais* (mortality range 0–34%). Similar findings have been reported for other nematode and insect combinations (Ramos-Rodriguez et al. 2006; Athanassiou et al. 2008a, b, 2010). This could necessitate the use of combined strains and/or species for effective control where it is likely that more than one species is present as is commonly found in storage premises.

Different life stages have shown variation in the susceptibility to infection by entomopathogenic nematodes. Ramos Rodriguez et al. (2006) found that larvae and adults of *E. kuehniella* and *P. interpunctella* were highly susceptible to infection by *S. feltiae, S. carpocapsae and S. riobrave.* The pupal stage of these moth species was less susceptible, in particular to *S. feltiae.* Larvae of *T. castaneum and T. confusum* were found to be more susceptible than adults (Ramos Rodriguez et al. 2006; Athanassiou et al. 2008a, b, 2010). This may be due to the greater mobility of

Table 7.2 Recent studies to assess the efficacy of entomopathogenic nematode species against stored product insects

Nematode species	Stored product species	References
Heterohabditis bacteriophora *Heterohabditis indica* *Heterohabditis marelatus* *Heterohabditis megidis* *Heterohabditis zealandica*	*Plodia interpunctella*	Mbata and Shapiro-Ilan (2005)
Heterorhabditis bacteriophora *Heterorhabditis megidis* *Steinernema carpocapsae* *Steinernema feltiae*	*Sitophilus granarius*	Trdan et al. (2005)
Steinernema carpocapsae *Steinernema feltiae* *Steinernema riobrave*	*Ephestia kuehniella* *Oryzaephilus surinamensis* *Plodia interpunctella* *Rhyzopertha dominica* *Sitophilus oryzae* *Tenebrio molitor* *Tribolium castaneum* *Trogoderma variabile*	Ramos-Rodriguez et al. (2006)
Heterorhabditis bacteriophora *Heterorhabditis megidis* *Steinernema carpocapsae* *Steinernema feltiae*	*Oryzaephilus surinamensis* *Sitophilus granarius*	Trdan et al. (2006)
Steinernema riobrave	*Plodia interpuncella* *Tribolium castaneum*	Ramos-Rodriguez et al. (2007)
Steinernema feltiae	*Ephestia kuehniella* *Tribolium confusum*	Athanassiou et al. (2008a, b)
Heterorhabditis bacteriophora *Heterohabditis indica* *Steinernema. abbasi* *Steinernema. asiaticum* *Steinernema. feltiae* *Steinernema pakistanense* *Steinernema siamkayai*	*Callosobruchus chinensis*	Shahina and Salma (2009)
Heterohabditis bacteriophora *Steinernema feltiae* *Steinernema carpocapsae*	*Ephestia kuehniella* *Rhyzopertha dominica* *Sitophilus oryzae* *Tribolium confusum*	Athanassiou et al. (2010)

(continued)

Table 7.2 (continued)

Nematode species	Stored product species	References
Steinernema feltiae	*Sitophilus oryzae*	Laznik et al. (2010)
Heterohabditis indica	*Habrobracon hebetor* *Plodia interpunctella*	Mbata and Shapiro-Ilan (2010)
Heterorhabditis bacteriophora *Heterohabditis indica* *Steinernema. abbasi* *Steinernema. asiaticum* *Steinernema. feltiae* *Steinernema pakistanense* *Steinernema siamkayai*	*Sitophilus oryzae*	Shahina and Salma (2010)
Heterorhabditis bacteriophora *Heterohabditis indica* *Steinernema. abbasi* *Steinernema. asiaticum* *Steinernema. feltiae* *Steinernema pakistanense* *Steinernema siamkayai*	*Tribolium castaneum*	Shahina and Salma (2011)
Heterorhabditis bacteriophora *Heterorhabditis megidis* *Steinernema carpocapsae* *Steinernema feltiae*	*Lasioderma serricorne* *Tribolium confusum*	Rumbos and Athanassiou (2012)
Heterohabditis bacteriophora *Steinernema carpocapsae* *Steinernema rarum* *Steinernema riobravis*	*Acanthoscelides obtectus* *Ephestia (Anastrepha) kuehniella* *Sitophilus oryzae* *Sitophilus zeamais* *Tenebrio molitor*	de Carvalho Barbosa Negrisoli et al. (2013)

the adult stage being able to avoid contact with the nematodes (Athanassoiu et al. 2010) or due to differences in the thickness of the cuticle between adults and larvae (Rumbos and Athanassiou 2012).

Temperature is also a significant factor for the efficacy of a nematode species/strain. In a study using *S. feltiae,* efficacy against *S. granarius* and *O. surinamensis* was greater at 20 and 25 °C than at 15 °C. Athanassiou et al. (2010) found that of three nematode species tested *H. bacteriophage* was more effective against *E. kuehniella* larvae at 30 °C, but at 20 °C *S. feltiae* was more effective.

The feasibility of using entomopathogenic nematodes outside of the laboratory has received less attention. Ramos-Rodriguez et al. (2007) examined the effect of *S. riobrave* against mixed life stages (larvae, pupae and adults) of *Plodia interpunctella* and *Tribolium castaneum* in empty grain bins following laboratory studies and concluded that this nematode species had potential for the control of all life stages of *P. interpunctella*, in particular larvae. Although fewer *T. castaneum* larvae survived compared to the controls (mean of 14.6 and 72% for the nematode treatment and control respectively), this was largely due to the effect on larvae and there was no significant difference in survival of pupae and adults. The authors concluded that improvements in formulation or application in combination with other treatments could improve the overall efficacy.

Development of an effective product requires advances in the production and application of entomopathogenic nematodes. Currently entomopathogenic nematodes may be produced in vivo or in vitro. In vivo methods for commercial application generally use *Galleria mellonella* or *Tenebrio molitor* as the host species and these methods are generally only suitable for small scale production, for laboratory testing or production for small niche markets (Shapiro Ilan et al. 2012). In vitro methods are based on the introduction of nematodes to a pure culture of their symbiotic bacteria contained in a nutritive medium. The methods may use either solid or liquid culture. The advantages and disadvantages of the different production systems have been recently reviewed by Shapiro Ilan et al. (2012). Formulation of EPNs has been improved and led to enhanced efficacy. The stored product environment may be viewed as more conducive to the application of nematodes as some of the factors that affect success in other environments, for example UV radiation, soil additives and competing organisms, may be absent. This is however, offset by the comparatively dry environment of a store in comparison with soil. Advancements made in the production and formulation of EPNs for agricultural and horticultural use could benefit application in the stored product area if suitable strains are identified.

Although formulation may permit application and survival of the nematodes at the low moisture content level found in stored grain, it is less likely that persistence or recycling of the entomopathogenic nematodes will occur (Athanassiou et al. 2010). It has also been suggested that EPN will not be suitable for application to grain doe to consumer acceptability (Rumbos and Athanassiou 2012) and there use should therefore be for the prophylactic treatment of stores, in particular for crack and crevice treatments or applied in bait stations in a lure and kill approach (Rumbos and Athanassiou 2012; de Carvalho Barbosa Negrisoli et al. 2013).

Compatibility of Microbial Biopesticides with Other Control Measures

Recent research has continued to increase the possibility of the development of effective microbial biopesticides for use in the protection of durable stored products. The nature of these agents makes them good candidates for use in integrated pest management strategies. Within these strategies it is likely that more than one control measures would be used. Therefore, it is important to understand the compatibility of the microbial biological control agent with other control measures that are likely to be used. This includes biological, chemical and physical control measures.

There have been several studies that have examined the compatibility of the use of microbial biological control agents and parasitoids. A possible additive or synergistic effect of the entomopathogenic nematode *H. indica* and the parasitoid *Habrobracon hebetor* for the control of *P. interpunctella* was demonstrated in laboratory studies (Mbata and Shapiro-Ilan 2010). The high mortality with the nematode or parasitoid alone did not permit a firmer conclusion to be made. The study also demonstrated that female parasitoids did not differentiate between nematode-infected and uninfected larvae, but infective juvenile nematodes preferentially infected parasitized host larvae. Significantly fewer *H. hebetor* progeny were produced in treatments in combination with the nematode than when the parasitoid was used alone. The authors concluded that there is potential to use the two biocontrol methods in combination, as there is no effect on the adult parasitoid, but the detrimental effect on the parasitoid larvae should be considered. It was suggested that timing of the nematode applications should be considered to reduce impact on the parasitoid and enhance the success of the combined treatment (Mbata and Shapiro-Ilan 2010).

Examination of the effect of four isolates of *M. anisopliae* and *B. bassiana* on *P. truncatus* and its parasitoid *Teretriosoma nigrescens* showed that although significant levels of mortality of the parasitoid occurred after 6 and 14 days, mortality of *P. truncatus* was greater (Bourassa et al. 2001). The effect on parasitism of infected hosts was not examined in that study. Lord (2001) examined the effect of *B. bassiana* on *Cephalonomia tarsalis*, an ectoparasitoid of *O. surinamensis*. The parasitoid did not avoid fungus treated grain and would oviposit into infected *O. surinamensis* larvae, although this resulted in the death of the parasitoid larva. Adult wasps were also susceptible to the fungus and it was concluded that grain treated with the fungus would have a detrimental effect on *C. tarsalis*.

Treatment of wheat with *B. bassiana* also negatively affected survival of *Lariophagus distinguendus* and *Anisopteromalus calandrae,* two parasitoids of *S. granarius* (Hansen and Steenberg 2007). The combined effect of the fungus and the parasitoid on suppression of a developing *S. granarius* population was less than when the parasitoid species were used alone. The authors concluded that the combined use of the entomopathogenic fungus and the parasitoid species may be possible in a targeted treatment if the fungus could be placed in a trap with an attractant for *S. granarius*, thus minimizing exposure to the fungus for the parasitoids (Hansen and Steenberg 2007).

The compatibility of the protozoan *M. oryzaephili* and the parasitoids *C. tarsalis* and *C. waterstonii* has been examined and it was concluded that *Cephalonomia* species could be used to inoculate *M. oryzaephilli* into beetle populations (Lord 2006). Female wasps became infected when paralyzing or feeding on infected *O. surinamensis* larvae and infection reduced female wasp longevity.

The potential for the combined use of microbial control agents and parasitoids is very much dependent on the nature of the microbial agent and the degree to which it can infect the parasitoid species. Entomopathogenic fungi may be less compatible, as shown in the studies described above, than agents such as protozoa and nematodes that have a more specific host range.

Compatibility of entomopathogenic fungi with physical control agents, such as diatomaceous earths has been extensively researched, as highlighted above. Compatibility with other physical control methods such as modified atmospheres has also been studied. Reduction of the oxygen concentration to 5% for 72 h significantly increased the mortality of *T. castaneum* larvae, but an elevation in the carbon dioxide level to 40% for 72 h had no significant effect (Lord 2009b). This was potentially explained by the effect that the modified atmospheres had on the germination and growth of the fungus; reduced oxygen had no effect on germination whilst elevated carbon dioxide negatively affected germination (Lord 2009b).

There have been studies to examine the effect of chemical insecticides on various species of entomopathogenic fungi and whilst some insecticides do not affect the fungi, others can have detrimental effects (see for example Cuthbertson et al. 2005). A recent study has demonstrated some compatibility with insecticides and *M. anisopliae,* particularly if the insecticides are used at sub-lethal levels (Apoorva and Ramaswamy 2013). Further research to examine the compatibility of various control measures is needed, but the potential for more than one control option within an IPM approach potentially offers improvements in overall efficacy with a reduced input of chemical insecticides.

Further Development of Microbial Biopesticides for Stored Product Protection

Research on the use of microbial biological control agents for the control of stored product insects and mites has increased in the past decade. However, the majority of the research remains at the scale of laboratory studies under controlled environmental conditions. Therefore, larger field scale evaluations over complete storage seasons are required to ascertain the potential for the use of microbial biopesticides. Developments in formulation and application are likely to be needed to optimize the use of these agents and to achieve the level of efficacy required by end users. This may also be realized through the use of combined treatments that include the microbial biological control agent. Use of such control measures may necessitate a greater understanding of these products by pest control operators and other end users, in particular if combination treatments with different timings are required.

The variation in the virulence of different strains or isolates of insect pathogens is well documented. The characterization of the different strains/isolates should continue, although this can be a time consuming operation. New strains are continually being discovered but not assessed. For example, it is reported that less than 20% of the entomopathogenic nematode species discovered since 2001 have been evaluated for biocontrol efficacy (Shapiro Ilan et al. 2012).

Research in other fields also offers potential for novel uses of microbial biological control agents in the future. The recent publication of the genome sequences of *M. anisopliae* and *M. acridium* (Gao et al. 2011) offers a tool for the greater understanding of virulence and methods to investigate the interactions between species together with the possibility of determining factors that could be manipulated to improve control. Genetic engineering has been used to improve the virulence of entomopathogenic fungi by reducing the spore load required to kill or shortening the time to kill (Fang et al. 2012). The majority of studies have utilized genes from entomopathogenic fungi themselves for strain improvement, for example, additional copies of the cuticle degrading protease in *M, anisopliae* (St Leger et al. 1996) or a chitinase gene from *B. bassiana* (Fang et al. 2005). A genetically engineered *M. anisopliae* isolate has also been used to deliver a scorpion toxin resulting in a 22 fold decrease in the spore concentration required to kill *Manduca sexta* (Wang and St Leger 2007). Genetic engineering has also been used to improve the tolerance of fungi to abiotic stresses (Fang et al. 2012).

The proteins secreted by entomopathogenic fungi are also attracting interest and have been shown to have insecticidal properties. The genome sequences of *M. anisopliae* and *M. acridum* have a large proportion of genes encoding secreted proteins, of which approximately 30% have no functionally characterized homologs, and these may have potential as novel control agents (Gao et al. 2011). The proteinaceous secretions of *M. anisopliae* and *B. bassiana* for the control of *C. maculatus* have been examined (Murad et al. 2006, 2007), although the insecticidal properties of the proteins remains to be elucidated.

Strains and isolates of insect pathogens can also be improved through non-molecular techniques. Selection and hybridization have been used to improve traits such as host finding for entomopathogenic nematodes (Shapiro Ilan et al. 2012) whilst increased virulence of *B. bassiana* isolates was observed when grown in media containing a hydrocarbon source (Pedrini et al. 2011).

Although different insect pathogens are at varying stages of development for the control of stored product insects and mites, the continued interest in this area combined with advances in knowledge of host pathogen interactions and other biotic and abiotic factors increases the likelihood of significant developments in the coming years.

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Chapter 8
Insecticide Resistance

Patrick J. Collins and David I. Schlipalius

Introduction

Insecticides, including contact chemicals and fumigants, are essential components of the majority of stored product protection systems. Their use enables the implementation of effective quarantine systems, ensures food security and facilitates domestic and international trade. Insecticides have many advantages. They can be integrated easily into grain handling logistics; they reliably provide the freedom from insect infestation demanded by many markets; and they are relatively inexpensive to apply. Despite their central importance, however, there are a surprisingly small number of chemicals used in the protection of stored products. Chemical residue levels are tightly regulated as stored products are usually foods. In addition, because of the often large volumes of commodity involved and convenience of application, fumigants are frequently the preferred treatments, rather than liquid insecticides. However, fumigant use requires strict workplace health and safety precautions and must comply with stringent environmental constraints. These factors, coupled with toxicological considerations, limit the range of materials available for application to grain and make them costly to develop. For these reasons, loss of any one chemical treatment will have a significant impact on pest management. Consequently, the development of resistance in stored product pests to any registered insecticide is a particularly significant problem that requires urgent solutions.

The purpose of researching resistance phenomena is ultimately to develop strategies to prevent or delay its development or to combat it once it is manifest.

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The development of resistance to insecticides is an evolutionary phenomenon and best understood from a genetics perspective. As we will see, both molecular and population genetics approaches have driven the most recent advances in our understanding of resistance, and this has been fundamental to progress in its management.

Insecticides act on genotypic variation (through mutation, changes in chromosome structure, recombination and gene flow) to select for resistant phenotypes. Thus, our first step in understanding resistance is to understand its genetic basis. We can then examine how the selection process occurs. It is important to note that control of the selecting agent is wholly in the hands of humans. The rate at which insecticide resistance occurs is a result of human intervention (i.e. application of insecticide) that interacts with a range of factors inherent to the insects and their environment. In the presence of insecticides, individuals with a particular genotype (resistant) enjoy an advantage in survival and reproduction over other genotypes. These advantages lead to an increased genetic contribution to future generations. An individual's genetic contribution to future generations is called its fitness. Any potential fitness advantage possessed by individuals with resistance genes, however, is only apparent when the insecticide is being applied against the insect population. Insecticides are used intermittently, so we need to ask the question: what happens to gene frequencies when the insecticide is no longer being applied? Furthermore, populations of insects are rarely completely isolated. They are part of a larger ecosystem that contains other populations of the same species with different resistance gene frequencies. Therefore, factors such as insect movement and mating systems may also impact on the rate of resistance selection.

In this review, we will discuss important advances in our understanding of the genetics of resistance and the process of selection for resistance genes, and how this knowledge may help us combat resistance. We also recommend that the reviews by Boyer et al. (2012), Opit et al. (2012) and Nayak et al. (2015) to the reader to gain a comprehensive perspective of insecticide resistance in insect pests of stored products.

The Genetic Basis of Resistance

Molecular Genetics

New sequencing technologies now make it affordable to sequence whole genomes and transcriptomes (all the expressed genes, or RNA sequencing) of most organisms. This has allowed the construction of multiple insect genomes that can be used as references for research. Of the insect pests of stored products, only the genome of *Tribolium castaneum* has been sequenced with a reference sequence being made available publicly in 2008 (Richards et al. 2008), while a transcriptome of *Liposcelis bostrychophila* has also been sequenced (Dou et al. 2013; Wei et al. 2013).

Considering the increasing availability and the decreasing costs of new sequencing technologies, it is expected that many more pest insect transcriptomes and genomes will be available in the near future. Access to published reference genomes available in databases, such as GenBank or EMBL, will enable a broad range of molecular investigations into the genetic basis of an insect's physiology and ecology. The genome of *T. castaneum* has been used to identify genes responsible for pyrethroid resistance (Zhu et al. 2010) and phosphine resistance (Schlipalius et al. 2012; Jagadeesan et al. 2013).

Pyrethroid Resistance

Zhu et al. (2010) characterised a strain from Australia that was highly resistant to deltamethrin (Collins 1998). They found that the resistance was most likely metabolic and that high levels of a cytochrome P450, CYP6BQ9, a detoxification gene expressed primarily in the brain and central nervous system, were responsible for the majority of the resistance.

Phosphine Resistance

Advances in molecular genetics and DNA sequencing have enabled the identification and characterisation of the underlying genetics of phosphine resistance. The major species in which phosphine resistance has been investigated are *T. castaneum* (Jagadeesan et al. 2012, 2013) and *R. dominica* (Schlipalius et al. 2002, 2008, 2012; Kaur et al. 2012), with other species such as *Sitophilus oryzae* also starting to be investigated (Daglish et al. 2014; Nguyen et al. 2015). Resistance to phosphine has been found to be highly conserved and conferred primarily by two autosomal (i.e. not sex-linked) recessive genes, *rph1* and *rph2* (*r*esistance to *ph*osphine *1* and *2*) (Schlipalius et al. 2008). The *rph1* gene confers a weak resistance (20–30x) when homozygous and is thought to have arisen first (Schlipalius et al. 2008). The *rph2* gene also confers weak resistance when homozygous (12–20x), but acts synergistically with *rph1* to give rise to phenotypes that are strongly resistant (>250x). This two-gene requirement for strong resistance has been shown to be the case in three species investigated to date, *R. dominica* (Schlipalius et al. 2002, 2008), *T. castaneum* (Jagadeesan et al. 2012, 2013) and *S. oryzae* (Nguyen et al. 2015).

The *rph2* gene in both *R. dominica* and *T. castaneum* codes for the dihydrolipoamide dehydrogenase (DLD) gene (Schlipalius et al. 2012), which is a subunit of major metabolic enzyme complexes involved in energy metabolism, such as the TCA cycle and amino acid metabolism. These complexes are mostly mitochondrial and include pyruvate dehydrogenase, alpha-ketoglutarate dehydrogenase, branched-chain amino acid dehydrogenase and the glycine cleavage system.

In insects, the alleles conferring resistance appear to be clustered around the active site of the protein (Schlipalius et al. 2012), which suggests that it may be a possible target site for phosphine. The observation that strains of insects with the *rph2* (DLD) resistance locus were also hypersensitive to arsenic in the form of arsine gas (Schlipalius et al. 2012), an effect that was previously observed in highly resistant *R. dominica* from Bangladesh (Chaudhry and Price 1991), supports this hypothesis. Arsenic has been shown to bind to the dihydrolipoamide cofactor of the enzyme complexes that contain DLD (Bergquist et al. 2009), which implies that phosphine is having a direct effect on the DLD enzyme. This insight into the mode of action of phosphine may be used in the future to find synergists with phosphine or to overcome resistance.

Molecular Markers

The sequence information of the *rph2* phosphine resistance gene has been used to develop molecular markers for known resistance alleles. DNA markers for phosphine resistance have already been deployed against populations from Australia (Kaur et al. 2013a), India (Kaur et al. 2015), the USA (Chen et al. 2015) and Turkey (Koçak et al. 2015). In the USA, Chen et al. (2015) reported that the marker for *rph2* resistance correlated well with the frequency of strong resistance detectable by bioassay.

Interestingly, although multiple alleles in DLD causing phosphine resistance were reported from strains isolated in Australia (Schlipalius et al. 2012), only one allele has been detected so far in other countries. This allele is the P45/49S allele (Kaur et al. 2015), which is a similar change in the protein shared at the same homologous position in both *T. castaneum* and *R. dominica*. Surveys reported so far from India (Kaur et al. 2013a), the USA (Chen et al. 2015) and Turkey (Koçak et al. 2015) show that high frequencies of this particular allele of *rph2* are common in grain storages. Although the sequences of DLD from each strain have not been reported from all these countries, it is highly likely that the allele has arisen independently in each case (Kaur et al. 2015).

There are major advantages associated with the use of molecular markers for resistance testing over the classical bioassay technique (Schlipalius et al. 2008). These include the fact that heterozygotes, or carriers of resistance that are phenotypically susceptible, can be accurately detected, and therefore, the potential for an insect population to develop strong resistance to phosphine can be assessed at a very early stage. Molecular testing has no requirement for live insects or a particular life stage, so eggs, larvae and pupae, live or dead can be assayed just as easily as live adults. This removes the need to maintain cultures of live insects caught in the field and removes the need for culturing facilities and the associated labour costs. The data generated from molecular tests are unambiguous and can be compared between surveys and laboratories easily without the requirement to develop local bioassays. There is also no requirement for a minimum number of insects per sample, as individuals can be tested. This is useful for when only one or two individuals may be detected during sampling.

Molecular markers for phosphine resistance genes now have application in routine monitoring, ecological research and evaluation of resistance management practices. Information on the frequency of resistance, the actual alleles present and their geographical distribution can be delivered in a relatively short amount of time, making these molecular markers highly valuable in resistance decision-making and contributing significantly to the preservation of phosphine as a routine treatment.

Factors Contributing to the Rate of Selection

Advances in Our Understanding of Fitness

Resistance is generally the result of the selection of relatively rare mutations that confer a fitness advantage to the insect possessing the resistance genes in the presence of insecticides. New mutations such as those coding for resistance, however, can be disruptive to the genome and may have detrimental pleiotropic effects. In the absence of insecticide, the resistance gene provides no advantage, and depending on the nature of the mutation, may be deleterious. That is, there may be a fitness cost associated with the resistance gene in the absence of insecticide. The actual effects may be physiological or even behavioural. Identifying fitness costs associated with resistance is important in designing resistance management strategies because the higher the fitness cost, the longer it is likely to take for resistance to spread in the population (Klior and Ghanim 2012). Differences in fitness between resistant and susceptible genotypes are assumed in the design of key resistance management tactics such as alternation of insecticides (Onstad 2008).

Pyrethroid Resistance

The nature of fitness costs has been investigated in maize weevil strains, *Sitophilus zeamais*, from Brazil, with high levels of resistance to pyrethroid insecticides. The Juiz de Fora strain had reduced and delayed emergence, reduced population growth and consumed less maize compared with two susceptible strains. That is, there appeared to be a fitness cost associated with resistance to pyrethroids in this strain. This was in contrast to the Jacarezinho strain, in which there was no apparent cost associated with resistance. This strain showed similar population growth rate and development time to susceptible strains (Fragoso et al. 2005). The dominant resistance mechanism in both strains was target site insensitivity (Guedes et al. 1995). The contrast between these two strains presented an opportunity to identify the physiological basis for fitness costs (Guedes et al. 2006), that is, to answer the question: was there an energy trade-off between insecticide resistance and other processes associated with development and reproduction? To do this, Guedes et al. (2006) measured respiration rate and fat body morphology as indicators of energy requirements and capacity in these two strains and compared their rates of development. They found that the Jacarezinho strain (no-cost) had higher body mass and larger fat body providing higher energy reserves than Juiz de Fora, and in addition, this strain had higher respiration rate indicating better mobilisation of energy. They postulated that the extra energy available to the insect compensates for the additional energy requirements needed to support resistance mechanisms without compromising demographic performance. In contrast, the Juiz de Fora strain was smaller in size, had lower respiration rate and had lower demographic performance indicating a physiological cost associated insecticide resistance. Other adaptations shown by the Jacarezinho strain included higher amylase activity, which could contribute to more efficient breakdown of starches (Araujo et al. 2008a), and more efficient digestive enzymes (Araujo et al. 2008b).

In addition to physiological differences, a resistance genotype may also affect insect behaviour. Pyrethroid-resistant *S. zeamais* were found to better detect the presence of deltamethrin than susceptible insects, being less likely to feed on deltamethrin sprayed grain when given a choice (Guedes et al. 2009a). In no-choice experiments, one pyrethroid-resistant strain continued to feed on treated grain at higher concentrations but another ceased feeding. Guedes et al. (2009a) suggested that the continued feeding in the former strain may have been to compensate for energy expended protecting against the insecticide. In several other studies, no correlation between physiological resistance to insecticides and other behavioural parameters, such as flight initiation and walking, could be found in *S. zeamais* (Periera et al. 2009; Guedes et al. 2009b; Braga et al. 2011; Corrêa et al. 2014), although it appears that higher walking activity and flight initiation may be associated with increased insect weight in this species (Guedes et al. 2009b).

Phosphine Resistance

An indication of possible fitness deficit associated with phosphine resistance was provided by Pimentel et al. (2007) who reported correlations between respiration rate, rate of reproduction and phosphine resistance ratios at the LC_{50} in field-collected samples of *T. castaneum*, *R. dominica* and *Oryzaephilus surinamensis*. For all species, respiration rate decreased as resistance ratio increased, while instantaneous rate of population growth decreased as resistance factor increased. More definitive evidence was provided by Kaur et al. (2012) who identified increased delays in development of immature stages after fumigation with phosphine in the strong phosphine resistance strain of *R. dominica*. The delay was inherited in a similar manner to the toxicity response and appeared to be a pleiotropic effect of phosphine resistance.

A possible link between movement and phosphine resistance in *R. dominica* was proposed by Pimentel et al. (2012) who found that walking activity was significantly reduced after exposure to phosphine in one phosphine-resistant strain compared with a susceptible strain but not in another. In contrast, Kaur et al. (2013a, b) found no evidence of any link between phosphine resistance and duration of walking or flight initiation in genetically characterised strong resistant, weak resistant and susceptible strains.

A simple approach to detecting fitness differences associated with resistance genes is to establish a ‘population cage’. Typically in these experiments, homozygous resistant and susceptible strains are crossed and then bred through a series of generations without exposure to the insecticide. A representative sample of insects is tested at intervals to detect any change in the frequency of resistant and susceptible genotypes or phenotypes. Using this method revealed no evidence of fitness deficits in *R. dominica* over 20 generations (Schlipalius et al. 2002) or *S. oryzae* (Daglish et al. 2014) or *T. castaneum* (Daglish et al. 2015) over seven generations. Jagadeesan et al. (2012) crossed homozygous weak resistant and strong resistant strains of *T. castaneum* with a susceptible strain. They found no fitness deficit associated with *rph1* but there was an indication of a fitness cost linked with *rph2*. This was confirmed in a follow-up study (Jagadeesan et al. 2013) where the authors observed a significant decrease in the frequency of *rph2* homozygous resistant genotype in *T. castaneum* over 18 generations with a corresponding increase in heterozygote and susceptible genotypes indicating a selective fitness disadvantage for homozygotes at the *rph2* locus. In contrast, the authors observed a significant increase in the frequency of the homozygous *rph1*, suggesting a fitness advantage of weakly resistant homozygotes compared to susceptible genotypes.

What Have We Learned About Fitness?

Fitness deficits associated with resistance genes can, theoretically, impact on the rate of selection of resistance in insect populations, and are therefore important to consider when developing resistance management strategies. Recent research demonstrates that there may be fitness differences between resistant and susceptible insects but the evidence is generally inadequate. There appears to be a deficit associated with phosphine resistance in some species, perhaps an allele of *rph2*, expressed as delayed immature development (Pimentel et al. 2007; Jagadeesan et al. 2012; Kaur et al. 2013a, b). It is yet to be determined, however, if this effect would be significant in the practical management of resistance to phosphine. Evidence from population cage experiments suggests that it may not (Schlipalius et al. 2002; Daglish et al. 2014). Severe fitness deficit has also been associated with a pyrethroid-resistant strain of *S. zeamais* (Guedes et al. 2006); however, it appears to be quite limited in frequency. It should be noted evolution is not static and deleterious effects associated with resistance may be reduced by the selection of modifier genes that may interact with the resistance genes producing a positive epistasis. Thus, fitness disadvantage associated with resistance alleles, perhaps obvious in the initial stages of selection, may become undetectable over generations as selection for an optimum phenotype occurs.

The methods used to detect and characterise fitness traits in insects are critical. Many factors can affect life history and behavioural parameters and the expression of these characteristics can vary significantly from strain to strain, independent of resistance status. This is particularly apparent when comparing long maintained laboratory reference strains with recently derived field strains which have been under quite different selection pressures. These pressures can result in significant differences in life history traits and other factors such as disease contamination. The best way to minimise genetic and other differences between strains is to compare strains that share a similar genetic background except for the character of interest. These can be created by either isolating two or more lines from one field strain, preferably one line with the resistance gene(s) and one without, usually through single pair mating and selection, or by creating isogenic or introgressed lines. The latter are created through repeated back-crossing of a resistant parent into a susceptible strain with selection. Many studies suffer from poorly defined genotyping and from the possibility of unknown background strain effects influencing experimental results. In addition, laboratory-based studies may not account for fitness aspects that are important under field conditions, such as disease susceptibility or temperature fluctuations.

Population Structure and Gene Flow

The development of molecular resistance gene markers (Schlipalius et al. 2012; Jagadeesan et al. 2013; Kaur et al. 2013a) is facilitating much more accurate assessments of resistance gene frequencies within populations of insect pests of stored products (Jagadeesan et al. 2013; Kaur et al. 2013a; Chen et al. 2015; Koçak et al. 2015); however, these markers have not yet been used to investigate resistance gene flow. Nevertheless, gene flow may be inferred from studies of population dispersal supported by neutral DNA marker studies. As well as being carried along transportation routes, the major pest species *T. castaneum*, *C. ferrugineus* and *R. dominica* are active flyers and are known to readily disperse relatively long distances (Mahroof et al. 2010; Semeao et al. 2010; Ridley et al. 2011; Daglish et al. 2014). Comparisons of individual insects across landscapes using neutral DNA markers indicate gene flow over very broad areas (Drury et al. 2009; Ridley et al. 2011) supporting the need for implementation of management strategies on a regional scale. Biological factors may also contribute significantly to rates of resistance selection. For example, one study revealed that 97% *T. castaneum* and *R. dominica* emigrating from grain storages had mated, and most with more than one male (Walter et al. 2014), increasing the likelihood that their progeny carries resistance genes. Furthermore, experimental evidence (Kaur et al. 2013a, b) indicates that phosphine-resistant insects can disperse as actively as susceptible insects. Sublethal effects are also likely to be important as it has been demonstrated that prior exposure to relatively low concentrations of phosphine reduces fecundity in phosphine-resistant *T. castaneum* (Ridley et al. 2012a) and *R. dominica* (Ridley et al. 2012b). It is now clear that biological and ecological factors and their interactions can impact significantly on resistant gene flow and on insecticide selection in populations of insect pests of stored products. The results of these studies are central to our understanding of the selection and distribution of resistance and, therefore, provide valuable information for the development of resistance management strategies.

Managing Resistance—Modelling the System

Laboratory studies can reveal important information about the factors that drive resistance development such as its inheritance, dominance and any fitness aspects associated with it, and field studies can provide insights into the dispersal of insects in the landscape and gene flow. However, because of the very large numbers of insects involved, the considerable number of potential variables, including those imposed by humans, and the long time frames required to include realistic numbers of insect generations, controlled experiments designed to test resistance management strategies are necessarily quite limited in scope. Simulation modelling, however, offers a method of evaluating a range of tactics and strategies under various scenarios that can produce credible and useful results if based on realistic data.

Rather than taking a theoretical approach based on general assumptions as was common in the past, recent model development has been based on information about real resistance phenomena, in particular, resistance to phosphine in the lesser grain borer, *R. dominica*. Resistance to phosphine in this species is controlled by two major genes (Collins et al. 2002; Schlipalius et al. 2002) and represents a serious resistance threat (Collins et al. 2000). Both Lilford et al. (2009) and Shi et al. (2012a) independently demonstrated that models based on the real-life two-gene situation in this species, rather than the single-locus resistance assumption used in previous modelling, much more accurately matched theoretical predictions of genotypes which subsequently affected model predictions, thus supporting the need for accurate experimental analysis of resistance genetics. Both models also used published phosphine mortality responses for *R. dominica* (Collins et al. 2002; Daglish 2004; Collins et al. 2005) to estimate resistance factors for various genotypes and to model survival rates (Lilford et al. 2009; Shi et al. 2011).

The Lilford two-locus model (Lilford et al. 2009) consisted of nine subpopulations, corresponding to nine genotypes, modelled by a system of nonlinear ordinary differential equations. Although the model was relatively simple, being based on responses of a single life stage and homogeneous fumigant concentrations, temperature, etc., simulations of fumigations matched field observations and expected gene frequencies. The model was expanded to include the response of all life stages (Thorne et al. 2010) providing insights into gene frequencies and population recovery between fumigations and strategic timing of treatments.

A different approach to modelling phosphine resistance in *R. dominica* was taken by Shi and collaborators who developed a stochastic, individual-based model (Shi et al. 2011, 2012a, b, 2013). These models represent the fact that insect populations consist of individual beetles, each of a particular genotype and life stage (Shi et al. 2012a). The advantage of this approach over simpler population-based models is that it allows more aspects of individual variability and biological reality to be included. The Shi model also used a daily time step, which can capture real conditions in more detail and obtain more precise results than a weekly time step. In the real world, conditions are more likely to change day to day than week to week (Shi et al. 2012b). Using this model, Shi et al. investigated management tactics relating to both single and successive fumigations. They concluded that fumigation for a longer period and at lower concentration is more effective than a shorter fumigation at a higher concentration. This is because under the former, eggs and pupae have time to develop to less tolerant stages (this assumes no delay in development under fumigation, or effect on fecundity). In addition, in a two-gene system where dilution of resistance genes through immigration has a greater effect, extending fumigation times will have a significant impact on delaying the development of resistance. Shi et al. (2012b) also examined the impact of initial resistance gene frequency and initial number of insects on fumigation success. They found that the rate of insect survival increases proportionally with initial genotype frequency and that if the original frequency of homozygous resistant insects is increased *n* times, then the fumigation needs to be extended *n* days to achieve the same level of control. Likewise, to achieve a similar level of control of a population that is *n* times larger than another population, the fumigation time needs to be increased by approx. *n* days. This demonstrates that increasing the duration of fumigation is an efficient way to increase efficacy, a conclusion matched by experimental evidence (Collins et al. 2005).

The authors then examined the impact of two other important factors and their interaction: fumigation dosage consistency within a storage and the effect of insect immigration into the storage, on resistance frequencies and insect numbers. This analysis was over 732 days and included a series of six fumigations within that time. The consistency of dosage, that is, distribution of fumigant, was the key factor in avoiding evolution of resistance and suppressing population increase. When consistency was high (even distribution of fumigant) there was no increase in the frequency of resistance or population numbers, regardless of immigration rate. When immigration is excluded, selection of resistance occurs faster in storages with moderate dosage inconsistency than in storages with low dosage inconsistency. In moderate dosage inconsistency, overall numbers increase but this increase is slow. In storages with low dosage consistency, that is, a leaky storage or one within which the gas is very unevenly distributed, population numbers increase because more insects survive, and resistance frequencies increase with every fumigation because of selection. Immigration of susceptible insects has a dilution effect but not enough to counteract the increase in resistance frequencies, even at the highest immigration rates (100 insects/day). The practical significance of this analysis is that storages should be well sealed before fumigation and active mechanisms used to distribute phosphine evenly to achieve high dosage consistency. In addition, insect movement into storages should be reduced as much as possible.

Ideally, models of resistance to insecticides should allow simulation and analysis of real-life storage situations in which variables can be changed to create a range of scenarios to test resistance management assumptions and tactics. The models described here incorporated genotype response data and life table information. The predicted outcomes matched the experimental evidence and industry experience demonstrating their validity and potential to be used to evaluate resistance management tactics. The models also highlighted key gaps in our knowledge thus helping to guide the direction of research. Nevertheless, these models are relatively simple and to adequately simulate fumigations of stored grain many more factors, such as physical characteristics of the fumigant (e.g. leakage, distribution and sorption) and biological influences, such as the effects of temperature and sublethal exposure on insect biology, need to be included, as well as the response of insects to various management interventions.

Conclusions

The most important recent advances in our knowledge of insecticide resistance have been in our understanding of genetics of resistance, particularly to phosphine, and the development of advanced molecular tools for diagnosing resistance. This has also led to the discovery of the underlying mechanisms involved in phosphine resistance and new knowledge of the mode of action of phosphine, which was not previously well understood. These insights will provide a basis for the development of new chemical tactics. Our understanding of selection of resistance genes and gene flow processes in populations, however, is at an early stage. Some progress has been made in our knowledge of relative fitness in particular; however, the importance of this issue is not resolved. The research on biological factors that may influence selection rates and gene flow emphasise the importance of these studies but there are still many gaps in our knowledge. Simulation modelling offers a method of integrating our knowledge of insect biology and ecology, genetic processes and insect responses to insecticides with the action of the insecticide on the commodity and within the storage. Models will become more powerful and predictive as more information is added but their usefulness relies on the quality of experimental data available, and there are still many gaps. However, models have the potential to be very valuable tools for understanding resistance development, testing resistance management tactics and devising resistance management strategies.

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Chapter 9
Biological Control of Stored-Product Insects

Matthias Schöller, Sabine Prozell, Pompeo Suma and Agatino Russo

Introduction

Previous reviews on biological control of stored-product pests published during the last 30 years reported results of laboratory experiments, collected examples for practical application in grain stores and in processing companies, and summarized the literature on the relevant control agents dating back to the beginning of the last century (Haines 1984; Arbogast 1985; Burkholder and Faustini 1991; Brower et al. 1996; Schöller et al. 1997; Schöller 1998, 2010; Schöller and Flinn 2000, 2012; Schöller and Prozell 2006). In this chapter, information on the biology of the most studied natural enemies is updated. Moreover, some aspects of biological control that were not considered previously are addressed: for example, information on molecular characters for determination of natural enemies of stored-product pests. These allow potentially the determination of species by people who are not trained taxonomists, and determination based on fragments or immature stages of the beneficials. A table summarizing these data is presented here, along with an overview of the application sites for biological control in Europe is given. Finally, stored-product pests attack also cultural heritage, i.e. museum items and historic buildings, and control examples for this field of application are summarized.

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Biology of the Natural Enemies of Stored-Product Pests

Predators

Xylocoris flavipes (Reuter) (Hemiptera—Anthocoridae)

Approximately, 50 species are comprised into the single genus of the tribe Xylocorini, *Xylocoris* Dufour, 1831. About 30 and 10 species of *Xylocoris* species are distributed in the Palaearctic and Nearctic regions, respectively (Yamada et al. 2013). All members feed on various small arthropods in such environments (Lattin 2000) and species of *Xylocoris* are known to occur in various habitats. They are frequently found in litter layers of shaded forest, beneath the tree bark and where plant debris accumulates. A few of them, *X. flavipes* (Reuter 1875), *X. galactinus* (Fieber 1836) and *X. sordidus* (Reuter 1871), were collected in stored food facilities, especially in grains.

The warehouse pirate bug *X. flavipes* is probably one of the most studied species from an ecological, ethological and economic point of view. This insect is indigenous to warm areas of the world (it was described for the first time from Algeria), and it is actually widespread also in subtropical and temperate regions, e.g. Northern Africa, Western Europe, South America, the USA, Eastern Asia and Australia (Pericart 1972; Lattin 2000). It is known as an efficient polyphagous predator of eggs and early developmental stages of beetles and moths. Its occurrence and release as beneficial for biocontrol against stored-product pests under a variety of environmental conditions is well documented (Jay et al. 1968; Awadallah and Tawfik 1972; Abdel-Rahman et al. 1983; Arbogast 1985; Brower and Mullen 1990). The predator is frequently associated with stored-product beetles such as *Tribolium confusum* Jacqueline du Val, *T. castaneum* (Herbst), *Sitophilus zeamais* Motschulsky, *S. granarius* (L.), *Lasioderma serricorne* (F.) (Press et al. 1975; Schöller and Prozell 2011a, b) and moths like *Plodia interpunctella* (Hübner), *Sitotroga cerealella* (Oliver) and *Corcyra cephalonica* Stainton (Reichmuth 2000; Reichmuth et al. 2007; Rabinder and Singh 2011; Suma et al. 2013). Recent studies showed potential of the pirate warehouse bug to reduce populations of seed beetles (Coleoptera: Bruchinae) (Berger 2017). Most effective control occurred when *X. flavipes* was added as soon as possible after the legumes were stored (Sing and Arbogast 2008a, b). Its developmental cycle has been examined in laboratory studies (Arbogast et al. 1971; Awadallah and Tawfik 1972), and studies have also been conducted that focused on the efficacy of *X. flavipes* in relation to the size of the host. The bug will attack eggs and generally small-sized insects, but not larger insects or internal grain feeders (Lecato and Davis 1973; Imamura et al. 2008; Sing and Arbogast 2008a, b). It has been found in storages throughout the world (Loko et al. 2013; Basumatary et al. 2013). Its optimal temperature range is of 29–31 °C, but can complete its life cycle at a temperature higher than 35 °C (Arbogast 1975; Abdel-Rahman et al. 1983) with an optimum relative humidity at 70% (Saha et al. 2012). All active stages of this predatory bug search for hosts. Studies have been conducted to determine the effects on different prey populations through the analysis of functional and numerical responses to predict the maximum number of prey attacked, and predator development, survival and reproduction. Holling (1959a, b) studied the predator–prey relationship from a mathematical point of view, showing that the behaviour of *X. flavipes* is of type II. This means that the predatory bug responds to an increase in the number of prey by preying until its overabundance is reached. This was confirmed by other authors (Brower et al. 1996; Rahman et al. 2009). Other biological observations showed a Mediterranean strain of the predator to be more adapted to high temperatures compared to the strain tested in the USA by Arbogast (1975) (Russo et al. 2004). In temperate areas, summer temperatures can exceed 30 °C; thus, the use of this predator would represent a good strategy within an IPM approach (Schöller and Prozell 2011a, b). Other recent studies showed *X. flavipes* would successfully control *Tribolium* spp. (Murata et al. 2007) and *C. cephalonica* (Rabinder and Singh 2011) in combination with other predators or combined with the use of insect growth regulators (Ferdous et al. 2010).

Parasitoids

Anisopteromalus calandrae (Howard) (Hymenoptera—Pteromalidae)

Anisopteromalus calandrae is one of the most frequently found parasitoids in stored grain, and it is widely distributed. It has been reported as a natural enemy of the following pests: *S. granarius, S. zeamais*, *Rhyzopertha dominica* (F.), *Stegobium paniceum* (L.), *L. serricorne*, *Acanthoscelides obtectus* (Say) and *Callosobruchus maculatus* (F.) (Williams and Floyd 1971; Arbogast and Mullen 1990; Reichmuth 2000; Reichmuth et al. 2007; Ngamo et al. 2007). *Anisopteromalus calandrae* is a primary, idiobiont ectoparasitoid attacking the late larval stages and early pupae of beetles inside seeds and cocoons (Shin et al. 1994). The female first paralyses the host larva, and second, lays an egg on the host body surface. Successively, the newly emerged larva will feed on the host's haemolymph (Ahmed 1996) and only one parasitoid emerges from each parasitized larva (Arbogast and Mullen 1990). Olfactometric studies showed female wasps to prefer products infested by *S. oryzae*, *R. dominica*, *L. serricorne* and *T. confusum* larvae-infested products compared to uninfested product (Belda and Riudavets 2010).

Gokhman et al. (1998) reported the existence of two closely related *Anisopteromalus* species summarized in applied literature as *A. calandrae*. They have contrasting reproductive strategies, one being r-strategist and its sibling species a k-strategist. The two are similar in external morphology, but differ in chromosome number and are consequently completely reproductively isolated. There are also some differences in life history (Gokhman et al. 1998, 1999; Timokhov and Gokhman 2003). The main hosts for the r-strategists are weevils that occur in high population density (Timokhov and Gokhman 2003), compared with the Anobiidae, which usually occur in small and isolated patches that are considered as putative hosts for the sibling species (Gokhman et al. 1998; Sasakawa et al. 2012). The DNA barcode sequences of *A. calandrae* and the sibling species were recently identified and can enable accurate identification of these morphologically similar species (Sasakawa et al. 2012). Recent studies (Sasakawa et al. 2013) showed a better learning capacity related to oviposition of the r-strategist versus the sibling species; thus, this aspect is really important for their application in biological control programmes. However, the taxonomy of the two species is not set.

Anisopteromalus calandrae can develop when temperatures range from 30 to 36 °C (Niedermayer et al. 2013). Other studies showed extreme temperatures to negatively affect the developmental cycle. Nguyen et al. (2013) found the male pupal stage to be temperature sensitive (38 °C), with negative effects on emergence and overall male reproduction ability. High temperatures during the early pupal stage had deleterious effects, i.e. male emergence and survival, and reproductive capacity, depending directly on temperature exposure. However, not all were equally affected by high temperature, the main impact being on sperm production. Temperature treatments (30–38 °C) had no effect on emergence of males, but temperatures exceeding 40 °C could be lethal. A temperature of 39 °C was considered to be sub-lethal when applied for 3 days at the early pupal stage.

Most studies addressed the effectiveness of *A. calandrae* in bulk grain. However, the release of *A. calandrae* in five types of bags (i.e. jute, calico (Americana cloth), polypropylene, polythene and nylon) containing wheat infested with *R. dominica* larvae showed this parasitoid to be able to suppress host populations in all types of bags, except in those made of polythene (Mahal et al. 2005).

Theocolax elegans (Westwood) (Hymenoptera—Pteromalidae)

Theocolax elegans is a widely distributed parasitoid occurring in stored wheat. It has been reported as a parasitoid of beetles, i.e. *S. granarius, S., oryzae, S. zeamais*, *R. dominica*, *S. paniceum*, *Callosobruchus* spp. and of the grain moth *S. cerealella* (Reichmuth et al. 2007). For an extensive list of potential hosts, see Sharifi (1972). *Theocolax elegans* is a solitary ectoparasitoid that parasitizes beetle larvae located within the seeds (Toewes et al. 2001). The parasitoid larva develops within the grain kernel on the host larva (Assem and Kuennen 1958). Long-range host finding is thought to be mediated by cereal volatiles. Recently, a positive response of the wasp towards such signals was reported (Germinara et al. 2009). This species has been well-studied with different cereal pests as hosts, e.g. *S. zeamais* (Williams and Floyd 1971; Imamura et al. 2004) and *R. dominica* (Toews et al. 2001; Flinn and Hagstrum 2002) with different results. This parasite reduced *S. zeamais* populations by up to 50% (Williams and Floyd 1971), and showed a preference for pupae and fourth instar larvae of the host (Sharifi 1972). Field studies showed a significant decrease in the number of *R. dominica* fragments in flour that was milled from this wheat with parasitoids present (Flinn and Hagstrum 2001). With this insect species as a host, the level of parasitization ranged from 50 to 99% depending on temperature (Flinn et al. 1996; Flinn 1998) and the parasitoid larval survivorship was highest on fourth instar larvae (Reichmuth et al. 2007). On the contrary, on the larger grain borer, *Prostephanus truncatus* (Horn), the wasp had a reduced impact, probably because the ovipositor was not long enough to reach the beetle larva inside the maize seed (Helbig 1998). The life cycle of *T. elegans* is about 22 days at 27 °C (Sharifi 1972) and it is more or less one-half of that of its host at the same temperature (Toews et al. 2001). The temperature has an important effect on the efficiency of parasitism for *T. elegans* and different studies have been undertaken in order to evaluate its effect on parasitizing *S. zeamais* larvae (Imamura et al. 2004) and *R. dominica* (Flinn and Hagstrum 2001). Both studies revealed that the wasp performed better when the temperatures were lower than 30 °C. Laboratory trials showed that *T. elegans* was ten times more effective at suppressing populations of *R. dominica* in grain that was cooled to 25 °C compared to grain that was stored at 32 °C (Flinn 1998). According to Flinn and Hagstrum (2001), this is due to a combination of two factors: the population growth rate of *R. dominica* is reduced by half at the lower temperature, and the functional response of *T. elegans* parasitizing *R. dominica* is the same at both temperatures.

Lariophagus distinguendus (Förster) (Hymenoptera—Pteromalidae)

Lariophagus distinguendus has been reported as potential agent for biological control for a wide number of beetles that infest stored agricultural products (Steidle and Schöller 1997): *S. oryzae* (Lucas and Riudavest 2002), *S. granarius* (Wen and Brower 1994; Steidle and Schöller 2002), *R. dominica* (Menon et al. 2002), *L. serricorne* (Steidle et al. 2006), *S. paniceum*, *A. obtectus* and *Ptinus clavipes* (Reichmuth 2000). It is a solitary ectoparasitoid of larvae and prepupae. *L. distinguendus* females produce sex pheromones (Ruther et al. 2000) inducing males to produce a characteristic wing fanning. The pheromones have an arresting effect and are active only when the female is very close at the male (0–5 mm) (Steidle and Reinhard 2003). The performance of male wing fanning influences the mating success in *L. distinguendus* (Benelli et al. 2013a). A role as contact sex pheromone has been played by the female cuticular hydrocarbons, Kuehbandner et al. (2012) showed that the composition of these compounds has been influenced by the host, recording similar profiles on strain reared on the same host, even if spatially isolated, and different profiles in strain reared on different host, suggesting that host shift in *L. distinguendus* might lead to reproductive isolation of host races, due to the modification on the cuticular semiochemistry.

Some publications showed the strong influence of temperature on parasitism (Hong and Ryoo 1991; Ryoo et al. 1991) and by intra- and interspecific competition (Ryoo et al. 1996). Gonen and Kugler (1970) considered *L. distinguendus* as a low fecundity species therefore not suitable as an agent for biological control. Successively, Steidle (1998) showed that the fecundity depended on strains, hosts and rearing conditions. The wasp is able to parasitize its host in the presence of low temperatures, which affects biological control programmes in Northern Europe (Hansen 2007; Niedermayer et al. 2013). Another behavioural factor with a great practical importance is the capacity of *L. distinguendus* to find its hosts inside the substrate or product. Steidle and Schöller (2002) found that females of *L. distinguendus* can localize their hosts at a depth of 4 metres from the grain surface, but the level of parasitization decreases with increasing depth. Adarkwah et al. (2012) showed for the first time the ability of *L. distinguendus* to penetrate bulk-stored maize and locate and parasitize larvae of *S. zeamais* at various depths. The searching capacity is facilitated by kairomonal substances originating from faeces of *Sitophilus* spp. (Steiner et al. 2007). Recently, larval faeces of *S. paniceum* have been also demonstrated as a key olfactory cue in host location by *L. distinguendus* (Benelli et al. 2013b). This is an important, practical element which needs further study.

Habrobracon hebetor (Say) (Hymenoptera—Braconidae)

Habrobracon hebetor is a cosmopolitan idiobiont gregarious ectoparasitoid. It develops on larvae of many Lepidoptera, mainly members of the family Pyralidae (Schöller 1998) and it has been studied as a control agent in Europe (Kovalenkov and Meschcheryakova 1983; Balevski 1984), Middle East (Gerling 1971; Amir-Maafi and Chi 2006; Shoukat 2012), Africa (Payne et al. 2011; Farag et al. 2012; Ba et al. 2013), China (Huang 1986) and the USA (Hopper 2003; Grieshop et al. 2006). Recently, Ghimire and Phillips (2010) provided biological data for *H. hebetor* with hosts other than Phycitinae, including the Galleriinae *Galleria mellonella* L. and *Corcyra cephalonica* (Stainton) and of families Gelechiidae, such as *Phthorimaea operculella* (Zeller) and *Sitotroga cerealella* (Olivier). The number of hosts may have even increased in recent years (Amir-Maafi and Chi 2006), but this is probably due to the presence of different strains in fields and warehouses (Heimpel et al. 1997). Today, *H. hebetor* is recommended for biological control and it has been extensively studied from the biological and demographical points of view (Amir-Maafi and Chi 2006; Forouzan et al. 2008; Akinkurolere et al. 2009). Carrillo et al. (2005) examined the cold hardiness of *H. hebetor* and reported that it is a chill-susceptible insect, and demonstrated that pupae and adults do not tolerate tissue freezing, aspects that have a great importance in biological control programmes in cold areas. Conversely, Chen et al. (2011) reported that it is possible to store the wasp for 20 days in a refrigerator at 5 ± 1 °C with no negative effect on survival and reproductive capabilities. Chen et al. (2012) also recently clarified that the photoperiod has a great influence in inducing the reproductive diapause. The braconid can develop in a wide range of temperatures and studies have shown that *H. hebetor* has an inverse time of manipulation of its host when temperature increases from 17 to 29 °C, but from 32 °C, this handling time becomes proportional (Zhong et al. 2009).

As reported by Adarkwah et al. (2010), the wasp is not a valid tool for biological control of *C. cephalonica* in rice inside jute bags, due to the inability of *H. hebetor* to invade the bagging material. Field observations conducted in Central and South European bakeries and mills showed that the species occurs simultaneously with the solitary endoparasitoid *Venturia canescens* (Gravenhorst) (Hymenoptera: Ichneumonidae). This parasitoid seems to be more common than *H. hebetor* in stored-product environments that have populations of the Mediterranean flour moth, *Ephestia kuehniella* Zeller. In trials conducted in small boxes, the reproduction of *V. canescens* was significantly reduced by the presence of *H. hebetor* but after additional scaled-up research showed that there was no such effect in *V. canescens*. This result suggests a better long-range host finding in *V. canescens*; therefore, the combination of the two parasitoids may improve the biological control of *E. kuehniella* (Paust et al. 2006). Adarkwah and Schöller (2012) used the same parasitoid combination in controlling *P. interpunctella* infestations, and reported the combination could be as effective as *H. hebetor* alone. However, large numbers of parasitoids would have to be applied in warehouses or silos with bulk-stored grain, and the parasitoid releases would need to be integrated with other control methods. Others studies have been conducted that combined the use of the parasitoid with the entomopathogenic nematode *Heterorhabditis indica* Poinar, Karunakar and David (Homl strain) (Rhabditida: Heterorhabditidae). These studies highlighted the importance of improving the compatibility of the two control agents in terms of the ability of *H. indica* to orientate preferentially to parasitized *P. interpunctella* larvae, and also in determining if there was a detrimental effect on *H. hebetor* development (Mbata and Shapiro-Ilan 2010).

Molecular Identification of Beneficials by Molecular Characters

Molecular data for six natural enemies of stored-product pests (Table 9.1) are available in GenBank®, a genetic sequence database and annotated collection of all publicly available DNA sequences (Benson et al. 2013). These are low numbers out of the 57 (Schöller 1998) identified natural enemies of stored-product pests; however, currently molecular data of most of the commercially available species is available.

Application Sites for Biological Control

Biological control in stored products has been commercialized since 1998 (Prozell and Schöller 2003). Most applications were against stored-product moths in bakeries, retail trade and private households, and against weevils in grain on farms. Table 9.2 lists the applications by target pests, site and beneficials. Fifty percent of the types of application are control of pyralid stored-product moths. The reasons for this might be the fact that biological control of pyralids was the first commercialized application and is best known in the public, and/or the fact that *Trichogramma* spp. are hardly visible under practical conditions due to their small size.

Table 9.1 Molecular identification of beneficials by molecular characters

Species	Sequence	Base pairs	GenBank accession number	Remarks	Authors
Anisopteromalus calandrae	Mitochondrial COI gene for cytochrome oxidase subunit I, partial cds, DNA linear	652	AB690356	Additional notes on *Anisopteromalus* sp. (Hymenoptera: Pteromalidae), the sibling species of a parasitic wasp of stored-product pests, *Anisopteromalus calandrae* (Howard)	Sasakawa, K. and Shimada, M.
	Carboxylesterase mRNA, complete cds. *Anisopteromalus calandrae* malathion resistant	1963	AF064524		Zhu, Y.C., Dowdy, A.K. and Baker, J.E., Insect Biochem. Mol. Biol. 29 (5), 417–425 (1999)
	Clone 2 mitochondrial RAPD marker	584	AY366905		Zhu, Y.C., Dowdy, A. and Baker, J. Unpublished
	Clone 1 mitochondrial RAPD marker	365	AY366904		Zhu, Y.C.
	28S Ribosomal RNA gene, partial sequence	452	DQ080093		Krogmann, L., Strecker, U. and Abraham, R. unpublished
Lariophagus distinguendus	Cytochrome oxidase subunit I (CO1) gene, partial cds; mitochondrial	664	JX658774	Pforzheim strain	Hacker, K., Brose, S., Gantert, C., Ruediger, C., Krogmann, L., Steidle, J.L.M.
	28S Ribosomal RNA gene, partial sequence	452	DQ080097		Krogmann, L., Strecker, U. and Abraham, R.
	cytochrome oxidase subunit I (CO1) gene, partial cds; mitochondrial	664	JX658774	Host races, good species and early adult experience in a parasitic wasp, Unpublished	Hacker, K., Brose, S., Gantert, C., Ruediger, C., Krogmann, L. and Steidle, J.L.M.
Habrobracon hebetor	16S Ribosomal RNA gene, partial sequence	458	AF003483	Insect Mol. Biol. 7(2), 129–150 (1998) Evolutionary relationships among the Braconidae (Hymenoptera: Ichneumonoidea) inferred from partial 16S rDNA gene sequences	Dowton, M., Austin, A.D. and Antolin, M.F.
	28S rRNA gene	434	AJ245691	Proc. R. Soc. Lond., B, Biol. Sci. 267(1442), 491–496 (2000) Estimating ancestral geographic distributions: a Gondwanan origin for aphid parasitoids?	Belshaw, R., Dowton, M., Quicke, D.L.J. and Austin, A.D.

(continued)

Table 9.1 (continued)

Species	Sequence	Base pairs	GenBank accession number	Remarks	Authors
	Partial 28S rRNA gene	624	AJ231498	Biol. J. Linn. Soc. Lond. 73(4), 411–424 (2001) Paraphyletic taxa and taxonomic chaining: evaluating the classification of braconine wasps (Hymenoptera: Braconidae) using 28S D2-3 rDNA sequences and morphological characters	Belshaw, R., Lopez-Vaamonde, C., Degerli, N. and Quicke, D.L.J.
	18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5.8S ribosomal RNA gene, and internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence	1197	EF491261		Paul, A., Malathi, V.G. and Thomas, A.
	16S mitochondrial ribosomal RNA, mitochondrial gene, partial sequence	404	U68145	Naturwissenschaften 84 (11), 502–507 (1997) Molecular and morphological data suggest a single origin of the polydnaviruses among Braconid wasps	Whitfield, J.B. and Guerber, C.
Cephalonomia tarsalis	Isolate KOR-H7 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	658	KC510135	Study on the identification methods of food pest insects using DNA barcodes Unpublished	Cho, S.Y., Suh, K.I., Kim, K.H. and Bae, Y.J.
Holepyris sp.	MC-2010 28S large subunit ribosomal RNA gene, partial sequence	619	GU213970	Insect Syst. Evol. 41, 55–73 (2010) Phylogeny of bethylid wasps (Hymenoptera: Bethylidae) inferred from 28S and 16S rRNA genes	Carr, M., Young, P.J.W. and Mayhew, P.J.

(continued)

Table 9.1 (continued)

Species	Sequence	Base pairs	GenBank accession number	Remarks	Authors
Trichogramma evanescens euproctidis	Tt4 internal transcribed spacer 2, complete sequence	376	AF043613	Biol. Control 16, 177–184 (1999) Molecular differentiation of five *Trichogramma* species occurring in Portugal (as *T. turkestanica*)	Silva, I.M.M.S., Honda, J.Y., Van Kan, F.J.P. M., Hu, J., Neto, L., Pintureau, B. and Stouthamer, R.
	Tt1 internal transcribed spacer 2, complete sequence	372	AF043615	Biol. Control 16, 177–184 (1999) Molecular differentiation of five *Trichogramma* species occurring in Portugal (as *T. turkestanica*)	Silva, I.M.M.S., Honda, J.Y., Van Kan, F.J.P. M., Hu, J., Neto, L., Pintureau, B. and Stouthamer, R.
	Trichogramma euproctidis isolate PRO9-486 TF10/09-11 internal transcribed spacer 2, partial sequence	377	JF415945	*Trichogramma* of Canary Islands, unpublished	Polaszek, A., Rugman-Jones, P., Stouthamer, R., Hernandez- Suarez, E., Cabello, T. and Pino Perez, M.
	SQG internal transcribed spacer 2, complete sequence	514	DQ088062	BioControl (2013) 58:483–491 Molecular identification of *Trichogramma* species from Pakistan, using ITS-2 region of rDNA	Muhammad Farooq Nasir, Gregor Hagedorn, Carmen Büttner, Christoph Reichmuth, Matthias Schöller
	5.8S ribosomal RNA gene, partial sequence; internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence	415	JF920443	Iranian *Trichogramma*: ITS2 DNA characterization and natural *Wolbachia* infection, unpublished	Poorjavad, N., Goldansaz, S.H., Machtelinckx, T., Tirry, L., Stouthamer, R. and Van Leeuwen, T.
	Isolate 033 5.8S ribosomal RNA gene, partial sequence; internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence	428	JF920453	Iranian *Trichogramma*: ITS2 DNA characterization and natural *Wolbachia* infection, unpublished	Poorjavad, N., Goldansaz, S.H., Machtelinckx, T., Tirry, L., Stouthamer, R. and Van Leeuwen, T.
	Internal transcribed spacer 2, complete sequence	376	DQ389076	Biol. Control 40(1), 48–56 (2007) Egg parasitoids of the genus *Trichogramma* (Hymenoptera, Trichogrammatidae) in olive groves of the Mediterranean region	Herz, A., Hassan, S.A., Hegazi, E., Khafagi, W.E., Nasr, F.N., Youssef, A.I., Agamy, E., Blibech, I., Ksentini, I., Ksantini, M., Jardak, T., Bento, A., Pereira, J.A., Torres, L., Souliotis, C., Moschos, T. and Milonas, P.
	FO5 internal transcribed spacer 2, partial sequence	377	DQ137263	Biol. Control 37(3), 375–381 (2006) Natural occurrence of *Wolbachia*-infected and uninfected *Trichogramma* species in tomato fields in Portugal	Goncalves, C.I., Huigens, M.E., Verbaarschot, P., Duarte, S., Mexia, A. and Tavares, J.

(continued)

Table 9.1 (continued)

Species	Sequence	Base pairs	GenBank accession number	Remarks	Authors
Xylocoris flavipes	Cytochrome c oxidase subunit I (co1) gene, partial cds; mitochondrial	621	KF365462	Molecular characterization of anthocorid predator *Xylocoris flavipes* using cytochrome oxidae subunit I (COI) gene, unpublished	Reetha, B., Venkatesan, T., C, B.R. and Jalali, S.K.
	Cytochrome oxidase subunit I gene, partial cds; mitochondrial	527	JQ782835	Evolution of traumatic insemination, Unpublished	Jung, S.
	28S ribosomal RNA gene, partial sequence	379	JQ782795	Evolution of traumatic insemination, unpublished	Jung, S.
	18S ribosomal RNA gene, partial sequence	1507	JQ782790	Evolution of traumatic insemination, Unpublished	Jung, S.
	16S ribosomal RNA gene, partial sequence; mitochondrial	364	JQ782756	Evolution of traumatic insemination, unpublished	Jung, S.

Table 9.2 Application sites for biological control of stored-product and museum pests in Europe

Site	Target pest	Beneficial	Organic production (yes, no, does not apply)
Persipan production	*Cadra cautella*	*Trichogramma evanescens*	No
Citrus pulp store	*Cadra cautella*	*Trichogramma evanescens*	No
Tobacco company	*Ephestia elutella*	*Habrobracon hebetor*	No
Medical plants	*Ephestia elutella, Plodia interpunctella*	*Trichogramma evanescens*	No
Tea store	*Ephestia elutella, Plodia interpunctella*	*Habrobracon hebetor*	Yes
Hay storage	*Ephestia elutella*	*Habrobracon hebetor*	No
Spices company	*Ephestia elutella*	*Trichogramma evanescens, Habrobracon hebetor*	No
Mill	*Ephestia kuehniella*	*Habrobracon hebetor*	Yes and no
Bakery	*Ephestia kuehniella*	*Habrobracon hebetor, Trichogramma evanescens*	Yes and no
Wholesale trade stored products	*Ephestia kuehniella*	*Trichogramma evanescens*	Yes and no
Bee keepers	*Galleria mellonella*	*Trichogramma evanescens*	Yes and no
Historic building	*Gibbium psylloides*	*Lariophagus distinguendus*	Does not apply
Herb store, herb production farm	*Idaea inquinata*	*Trichogramma evanescens*	Yes
Wine Cellar	*Nemapogon granellus*	*Trichogramma evanescens*	No
Historic building	*Niptus hololeucus*	*Lariophagus distinguendus*	Does not appy
Chocolate company	*Oryzaephilus mercator*	*Cephalonomia tarsalis*	No
Retail trade	*Oryzaephilus surinamensis*	*Cephalonomia tarsalis*	Yes
Grain store on farm	*Oryzaephilus surinamensis*	*Cephalonomia tarsalis*	Yes
Retail trade	*Plodia interpunctella*	*Trichogramma evanescens*	Yes
Health shop	*Plodia interpunctella*	*Trichogramma evanescens*	No

(continued)

Table 9.2 (continued)

Site	Target pest	Beneficial	Organic production (yes, no, does not apply)
Tea retailer	*Plodia interpunctella*	*Trichogramma evanescens*	Yes and no
Tea wholesale	*Plodia interpunctella*	*Trichogramma evanescens*	Yes
Pet food wholesale, pet food retail trade	*Plodia interpunctella*	*Habrobracon hebetor*	Does not apply
Private households, food	*Plodia interpunctella*	*Trichogramma evanescens*	Does not apply
Herb store	*Plodia interpunctella*	*Habrobracon hebetor*	Yes
Pigeon rearing	*Plodia interpunctella*	*Habrobracon hebetor*	Does not apply
Fishermen's shop	*Plodia interpunctella*	*Trichogramma evanescens*	No
Pharmacy	*Plodia interpunctella*	*Trichogramma evanescens*	No
Pet cages, private households	*Plodia interpunctella*	*Habrobracon hebetor*	Does not apply
School/daycare kitchen	*Plodia interpunctella*	*Trichogramma evanescens*	Does not apply
Catering service	*Plodia interpunctella*	*Trichogramma evanescens*	Yes
Wholesale trade stored products	*Plodia interpunctella*	*Trichogramma evanescens*	Yes and no
Restaurant/café	*Plodia interpunctella*	*Trichogramma evanescens*	Yes and no
Nuts	*Plodia interpunctella*	*Trichogramma evanescens*	Yes
Seed wholesale, seed research institute	*Plodia interpunctella*	*Trichogramma evanescens*	No
Historic building	*Pyralis farinalis*	*Trichogramma evanescens*	Does not apply
Bakery	*Sitophilus granarius*	*Lariophagus distinguendus*	Yes and no
Grain store on farm	*Sitophilus granarius*	*Lariophagus distinguendus*	Yes and no
Brewery	*Sitophilus granarius*	*Lariophagus distinguendus*	Yes
Pasta company	*Sitophilus zeamais*	*Anisopteromalus calandrae*	No
Tea wholesale	*Stegobium paniceum*	*Lariophagus distinguendus*	Yes

(continued)

Table 9.2 (continued)

Site	Target pest	Beneficial	Organic production (yes, no, does not apply)
Tea store	*Stegobium paniceum*	*Lariophagus distinguendus*	Yes
Library	*Stegobium paniceum*	*Lariophagus distinguendus*	Does not apply
Import of South American woollen textiles	*Tineola bisselliella*	*Apanteles carpatus*	No
Carpet store, furniture store, dress-coat rental	*Tineola bisselliella*	*Trichogramma evanescens*	Does not apply
Daycare	*Tineola bisselliella*	*Trichogramma evanescens*	Does not apply
Historic mustard company	*Tineola bisselliella, Tinea* sp.		Does not apply
Technical museum	*Tineola bisselliella*	*Trichogramma evanescens*	Does not apply
Art museum	*Tineola bisselliella*	*Trichogramma evanescens*	Does not apply
Art museum	*Tineola bisselliella*	*Baryscapus tineivorus*	Does not apply
Pet food production company	*Tineola bisselliella*	*Trichogramma evanescens*	No
Wholesale trade stored products	*Tribolium castaneum*	*Xylocoris flavipes*	No
Art museum	*Trogoderma angustum*	*Laelius pedatus*	Does not apply
Art museum	*Trogoderma angustum*	*Xylocoris flavipes*	Does not apply

Stored-Product Pests in Cultural Heritage

Stored-product pests may destroy materials as well, either through travel to pupation sites or because the materials contain ingredients suitable for development. This initiated the interest in biological control of these pests in museums and other environments with cultural heritage items, as well as research in specific natural enemies of museum pests. Table 9.3 summarizes the natural enemies currently investigated.

Biological Control of the Drugstore Beetle *Stegobium paniceum* and the Tobacco Beetle *Lasioderma serricorne*

The drugstore beetle *Stegobium paniceum* (L., 1758) may develop in historic books with covers filled with pulp made from linen scraps. A trial on host finding of the store chalcid *Lariophagus distinguendus* (Förster 1841) in boxes containing books was carried out in an Israeli library, where *Lariophagus distinguendus* was shown to find host larvae both between and inside infested books (Wilamowski et al. 2008). In Halle/Saale, Saxony-Anhalt, Germany, a historic library became infested by *S. paniceum*. The beetles were thriving both below the floorboard on wheat straw used as insulation, and in books dating back to the sixteenth to eighteenth century. *Stegobium paniceum* developed in the pulp, produced the characteristic exit holes and therewith destroyed irreplaceable cultural heritage. The books were moved to a fumigation chamber and treated with nitrogen. However, some re-infestation was detected after the books were moved back to the library, presumably originating from the floorboard. *Lariophagus distinguendus* was released on the shelves, 2000 individuals in October and June respectively. The release successfully suppressed the re-infestation of the library (Schöller 2010). More recently, chalcids were regularly released against *S. paniceum* in the Kunsthistorisches Museum Vienna (Querner et al. 2013). A bibliography of the natural enemies of *S. paniceum* and the tobacco beetle *Lasioderma serricorne* (F. 1792), a species with similar biology, is given in Schöller (1998).

Biological Control of Spider Beetles (Anobiidae, Ptininae)

Spider beetles are mainly scavengers feeding equally on plant or animal materials. Besides their natural habitats, a number of species infest historic houses feeding on organic insulation materials and become a nuisance in residences (Howe 1953). Moreover, spider beetles were found to infest historic books and herbaria (Gamalie 2006). A number of spider beetle species were found to be suitable hosts for *L. distinguendus*, such as *Ptinus fur* L., 1758 (Herold 1933; Hüsing 1935), *Ptinus tectus* (Boieldieu 1856; Kaschef 1955), *Gibbium psylloides* (Czenpinski 1778) (Kaschef 1961) and *Niptus hololeucus* (Faldermann 1835); (Schöller and Prozell 2014). Spider beetles are difficult to control in houses because the larvae develop hidden within the walls and in dead floors, and no monitoring devices are available. In recent years, *L. distinguendus* was released against the hump beetle *G. psylloides* and the golden spider beetle *N. hololeucus* in Germany by pest control companies and became a regularly applied control technique (Kassel 2008). However, due to the lack of appropriate monitoring techniques, the optimal timing of the parasitoid releases has not been determined. In this case, modelling the pest and parasitoid's population dynamics might be useful. As a first step, a modelling software 'SITOPHEX' (Roßberg et al. 2004) was programmed for the system *Sitophilus*

Table 9.3 Natural enemies evaluated for biological control of moths and beetles attacking cultural heritage

Parasitoid species	Pest species attacked	Host stage	References	Field trial/ commercial application
Moths				
Trichogramma evanescens	*Tineola bisselliella*	Egg	Zimmermann (2005)	Yes/yes
	Tinea pellionella	Egg	Hase (1919)	No/no
Trichogramma piceum	*Tineola bisselliella*	Egg	Zimmermann (2005)	No/no
Apanteles carpatus	*Tineola bisselliella*	Larva	Plarre et al. (1999)	Yes/no
	Tinea pellionella	Larva	Plarre et al. (2000)	No/no
Baryscapus tineivorus	*Tineola bisselliella*	Larva	Matzke and Plarre (2013)	Yes/yes
	Tinea pellionella	Larva	Matzke and Plarre (2013)	No/yes
Beetles				
Anobiidae				
Lariophagus distinguendus	*Stegobium paniceum*	Larva	Kaschef (1955)	Yes/yes
	Lasioderma serricorne	Larva	Steidle et al. (2006)	No/yes
	Niptus hololeucus	Larva	Kassel (2008)	Yes/yes
	Gibbium psylloides	Larva	Kaschef (1961), Kassel (2008)	Yes/yes
	Mezium affine	Larva	Schöller unpubl.	No/no
Anisopteromalus calandrae	*Stegobium paniceum*	Larva	Schöller and Prozell (2014)	Yes/yes
	Lasioderma serricorne	Larva	Schöller and Prozell (2014)	Yes/yes
	Niptus hololeucus	Larva	Kassel (2008)	Yes/yes
	Gibbium psylloides	Larva	Kassel (2008)	Yes/yes
	Mezium affine	Larva	Schöller unpubl.	No/no
Spathius exarator	*Anobium punctatum*	Larva	Becker (1954)	Yes/yes
Cephalonomia gallicola	*Anobium punctatum*	Larva	Paul et al. (2007)	No/no

(continued)

Table 9.3 (continued)

Parasitoid species	Pest species attacked	Host stage	References	Field trial/ commercial application
Dermestidae				
Laelius pedatus	*Trogoderma angustum*	Larva	Al-Kirshi (1998)	No/no
	Anthrenus verbasci	Larva	Al-Kirshi (1998)	No/no
Xylocoris flavipes	*Trogoderma angustum*	Larva	Schöller and Prozell (2014)	Yes/no
	Anthrenus verbasci	Egg, larva	Schöller and Prozell (2014)	No/no
	Attagenus unicolor	Larva	Schöller and Prozell (2014)	No/no
	Attagenus smirnovi	Egg, larva	Schöller and Prozell (2014)	No/no
	Anthrenocerus australis	Egg, larva	Schöller and Prozell (2014)	No/no

granarius—*L. distinguendus*. The biological data of the granary weevil *S. granarius* (L., 1758) was replaced by those of the golden spider beetle *N. hololeucus* (Schöller and Prozell 2011a, b). One of the major differences in biology of the granary weevil compared to the golden spider beetle is the low reproduction of the latter at temperatures higher than 25 °C.

The current release strategy by pest control companies is the monthly release when temperature conditions are favourable. The susceptible old larval stages and the pupae are controlled within 1 month and the population is suppressed in a way that few or no adults enter the living rooms. If the number of releases is reduced to four, one in beginning of July, September, March and May, respectively, an increase in young larvae is predicted for June. However, if the timing of the four releases is changed to beginning of July, March, May and June, this increase in young larvae can be suppressed (Schöller and Prozell 2011a, b). This effect can be explained by the poor development of *N. hololeucus* during high temperatures in September, indicating the importance of population suppression early in spring in temperate climates. Even though these models cannot be validated at present due to the lack of monitoring devices, simulation models are thought to give some decision support for parasitoid releases.

Biological Control of Larder Beetles

Larder beetles (Dermestidae) are among the cultural heritage pests most difficult to control with insecticides. Two approaches for biological control were tested: the control by a parasitoid naturally occurring in houses, and the control by a generalist predator transferred from the stored-product environment. The parasitoid *Laelius pedatus* (Say 1836) (Hymenoptera: Bethylidae) is a gregarious ectoparasitoid of several larder beetle species including *A. verbasci* and *T. angustum* (Amante et al. 2017). The shiny black wasps measure 2–3 mm in length (Reichmuth et al. 2007). During its life span, a female wasp paralysed 74 ± 20 larvae of *A. verbasci* (Al-Kirshi 1998). The average number of eggs per female wasp and day was 1.42 ± 0.2 if larvae of *T. angustum* were used as a host. Most egg-laying activity was observed at temperatures between 25 and 28 °C, while no oviposition occurs at 15 °C. A mated female lives 6–8 weeks at room temperature (Al-Kirshi 1998). This parasitoid occurs in Central Europe in buildings, but there are no studies on the biological control potential of laboratory-reared wasps in field trials. Larder beetles in the genera *Attagenus* are not parasitized (Al-Kirshi 1998), as well as *Anthrenocerus australis* (Hope 1843); (Schöller and Prozell 2014).

The predatory pirate bug *X. flavipes* (Reuter 1875) is a natural enemy of various pest Coleoptera and Lepidoptera (Arbogast 1978). The potential for biological control of larder beetles attacking cultural heritage was only recently evaluated (Schöller and Prozell 2014). Eggs and larvae of five larder beetle species in the genera *Anthrenus*, *Attagenus*, *Trogoderma* and *Anthrenocerus* were found to be accepted as prey (Schöller and Prozell 2014).

Biological Control of Clothing Moths

Several parasitoid Hymenoptera were evaluated for biological control of the webbing clothing moth *Tineola bisselliella* (Hummel 1823) and the case-bearing clothes moth *Tinea pellionella* Linné, 1758 (Table 9.3), including both egg and larval parasitoids.

Biological Control of the Cloth Moth *Tineola bisselliella*

Several species of *Trichogramma* have been shown to parasitize eggs of *T. bisselliella* including *T. evanescens euproctidis*, the species currently applied against stored-product moths in Central Europe (Zimmermann et al. 2003). *Trichogramma* spp. were shown to traverse distances of at least 15 m on smooth surfaces within 1 h, equalling 30,000 times a wasp's body length in 1 h, comparable to a vehicle 2 m in length driving at 60 km/h (Quednau 1958). However, for *Trichogramma* spp., the surface structure of textiles and carpets is comparably large. In order to test if a large surface will reduce the effectiveness of released parasitoids on cloth, Zimmermann (2005) placed cloth (25 cm × 45 cm) in cages (100 cm × 50 cm 65 cm) previously used for simulated field trials with *Trichogramma* spp. on green plants. He compared three types of cloth: (1) Finely woven cloth 1.5 mm in thickness without long distant strand (fibres) (2) medium-finely woven cloth ca. 3.0 mm in thickness with long distant strand and (3) tanned sheepskin rug ca. 2.5 cm in thickness. Five batches of eggs with 120 *T. bisselliella* eggs each were placed at 10, 20, 30 and 40 cm from the release point as baits. The number of female *T. evanescens* active on the cloth was increasing with an increasing number of parasitoids released. The number of *Trichogramma* females active on the cloth was decreasing with increasing thickness of the cloth. The conclusions drawn for the release recommendations were: *Trichogramma* spp. have to be applied inundatively, in most cases, all year round, and the release units have to be placed directly on the cloth or shelf to be protected. Currently, 1000 *T. evanescens* per square meter and week are recommended.

Recent promising results have been obtained for the mass release of *T. evanescens* in museums (Querner and Biebl 2011). First practical applications of *T. evanescens* were performed in a depot of an ethnographical museum in South-West Germany, and in the Jewish Museum in Berlin. In depots, the identification of infested items might be very time consuming, but the parasitoids actively search for moth eggs. In the Jewish Museum, the parasitoids helped to suppress a residual cloth moth population feeding on fluff balls formed by wear debris of the visitor's wool clothes in cracks and crevices and was integrated with an improved cleaning procedure. Historic cars in museum exhibition rooms were treated in Vienna (Austria) (Querner et al. 2013), Munich and Bochum (Germany) (Biebl 2009). Felt mats within the cars were infested by *T. bisselliella*. The surface area of felt is relatively small compared to other woollen materials, and monitoring of the moths with the help of pheromone-baited sticky traps showed a breakdown of the moth population after parasitoid release. The number of *T. evanescens* released per week on a total of 60 cars was 45,000 (Biebl 2013). Art by Joseph Beuys containing felt was protected from infestation by *T. bisselliella* in an exhibition room of the Neue Galerie Kassel (Germany).

Considerable research has also been done on the braconid wasp *Apanteles carpatus* (Say 1836), however, this parasitoid is not yet commercially available. *Apanteles carpatus* is a solitary koinobiont endoparasitoid of *T. bisselliella* and *T. pellionella* L., 1758 larvae, i.e. one wasp develops per moth host larva and the parasitoid larva will eventually kill the moth larva. *A. carpatus* is capable of complete development in all larval stages of *T. bisselliella* (Plarre et al. 2000). For a field study in a heavily infested rug store, Plarre et al. (1999) released laboratory-reared *A. carpatus* on a monthly basis. The release of *A. carpatus* alone had no suppression effect on the *T. bisselliella* population, only the combination of *A. carpatus* release with a sanitation programme significantly reduced the pest. Another larval parasitoid, the eulophid *Baryscapus tineivorus* was recently studied with *T. bisselliella* and *T. pellionella* as hosts (Matzke and Plarre 2013). Two to 36 parasitoid progeny developed on late instars of the host. Developmental time from egg to adult is about 3 weeks at 28 °C and 75% RH. This parasitoid has a sex ratio very favourable for biological control, i.e. 85% females.

Conclusions

Research on natural enemies of stored-product pests continues to benefit from basic research using parasitoids and predators of stored-product pests as model organisms, but recommendations for practical biological control are steadily increasing as well. Interest in integrated stored product protection is growing, and biological control became one of its components in Central Europe. Tables 9.2 and 9.3 showed a wide range of fields of application. However, availability of natural enemies is still limited as well as expertise for practical application. Consequently, research in mass production and more data from field trials are future tasks in this research area.

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Chapter 10
Emerging Pests in Durable Stored Products

Christos G. Athanassiou and Christos I. Rumbos

Background

The term "emerging pest" has been used many times with different meanings in the pre-harvest, as well as in the post-harvest stages of agricultural production. Often, emerging is confused with "alien" or "invasive" which illustrate different patterns regarding the spatiotemporal perception on pest distribution in different geographical areas. Emerging pests are, by definition, already known or newly described species, whose incidence or geographical distribution is increasing (Roy 2011). Conversely, alien species are the ones that are introduced outside of their natural range either intentionally or unintentionally by human activity (IUCN 2000), whereas invasive pests are nonnative, alien species whose introduction and/or spread threaten biological diversity (Kelly et al. 2013).

The majority of the data that are available for emerging species of agricultural importance are mostly focused on arthropod species that are present in crops and orchards. There are disproportionally fewer data for emerging species in stored products, probably due to the fact that there are fewer species that are related to the stored-product ecosystem and can be classified as emerging. Nevertheless, often stored-product insects are more likely to spread through international trade of durable agricultural commodities than field pests, due to the fact that they can easily survive within the commodity for certain periods of time, which may not be feasible in the case of crop/orchard pests. Hence, these species can travel through vehicles, etc., and this is why they have been named as "insect travelers" (Aitken 1975). In

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this regard, many stored-product arthropods do not overwinter, while in fact, low temperatures may be beneficial for their development. For example, stored-product mites can develop better at low temperatures, especially when these are combined with elevated relative humidity (r.h.) conditions (Athanassiou et al. 2003, 2005, 2011). Other species are able to enter diapause for a very long period and start to build high population densities when the conditions prevailing are suitable. One good example is this of the khapra beetle, *Trogoderma granarium* Everts (Coleoptera: Dermestidae), which remains at diapause for years and can even interrupt diapause with "foraging excursions" (Burges 1959, 1963; Banks 1977).

In the following, we will refer to those arthropod species that can be considered as emerging pests in stored products. Other species can be also considered, but the species that are mentioned here can serve as a baseline for further research for additional cases of low prevalence or expansion. In general, the pests that are shaped here under the characterization of "emerging" can be classified into four broad categories as follows:

1. *Quarantine*. Here, we will give examples of species that are of quarantine importance in several countries of the world, for which there are multiple interceptions in zones where they are not present, and therefore, there is a danger of establishment or low prevalence. These cases are directly related to phytosanitary regulations and export key scenarios.
2. *Invasive*. Here, we have species that are not of quarantine importance, but they are intercepted in areas that were not recorded before. For example, there are species in the European Union that are established in the south, but now their presence is recorded in the north, indicating expansion and low prevalence. In other words, "native" species for some zones may become "invasive" for neighboring zones. Climate change, monitoring techniques and control practices may play a role in this rate of expansion.
3. *Frequently noticed*. By this definition, we want to characterize stored-product insect species that were not recorded often in the past, but they are currently frequently recorded. These species were already common in several areas and most of them have a global distribution, but their presence was either negligible or they were not considered as major pests. In this category, we can include stored-product psocids (Psocoptera) and mites (Acari), which, at least in the majority of surveillance data that are available, their presence was underestimated. Again, as above, monitoring techniques play an important role in the detection of these species.
4. *Potentially emerging*. Under this category, we have included some paradigms of species that are currently not important in most parts of the world but may play an important role in the future. Moreover, there are species that can infest other than stored products and have been found to "switch" to durable stored products.

Stejskal et al. (2015) classify "emerging pests" and "emerging risks" by using similar definitions. In particular, in this study, the authors give the definitions of five different categories: emerging species, new levels of population density, emerging biotypes, new negative effects and risks, and newly endangered and packaged commodities. Based on the above, many of the pests that we will address as emerging in the following text may become emerging through changes in the way that they are monitored and controlled and not necessarily due to a noticeable increase in their presence, dominance, and frequency of detection. For instance, withdrawal of specific insecticides that were effective for the control of specific species might have contributed to their spread, or changes in detection and estimation methods (i.e., identification techniques) might have contributed to improved monitoring and the development of revisited decision support systems that also include these pests as well. These biotic invasions in stored products, even if they are from species that are already established, may obviously have some global consequences, while, in the case of the post-harvest ecosystems, have mainly humans as dispersal agents, due to the fact that stored-product arthropods are "synanthropic species". Nevertheless, in newly introduced species, there may be a lag stage between immigrant and invader (Mack et al. 2000), but often in stored products, this stage does not last long, as the conditions that prevail in storage environment may not be that diverse in comparison with field crops. For example, the larger grain borer, *Prostephanus truncatus* (Horn) (Coleoptera: Bostrychidae), was able to establish in Central Africa right after its introduction (Hodges 1986). For some species that are already present and until recently were regarded as minor nuisance pests in stored products, their classification to major pests can occur due to newer developments and more research on their biology, that reveal their high importance. For example, the psocids of the genus *Liposcelis* are now known that can easily infest undamaged grain kernels which gave another dimension to their importance (Nayak et al. 2014).

The Khapra Beetle

Trogoderma granarium Everts (Coleoptera: Dermestidae) is a noxious stored-product insect and quarantine insect species in several countries worldwide (Myers and Hagstrum 2012; EPPO 2013). Although its ability to spread actively is limited because adults do not fly (EPPO 2013), today's extensive international trade transactions offer an efficient means of spreading *T. granarium* worldwide. Despite the quarantine restrictions, this pest, which originated in India, is already present in Central America, Western Africa, and the Middle East, as well as in some countries of Asia and Europe (EPPO 2013). Apart from the records of established infestations, the number of interceptions of *T. granarium* in non-khapra beetle countries has increased the recent years (Myers and Hagstrum 2012; Day and White 2016; EPPO 2016a). *Trogoderma granarium* is characterized by several ecological and behavioral traits that render it a successful invader, such as its feeding behavior, the facultative larval diapause, as well as the resistance that several populations have developed to commonly used insecticides.

Its ability to infest and reproduce in a huge variety of commodities classifies *T. granarium* into the generalist species (Lindgren et al. 1955; Lindgren and Vincent 1959; Aitken 1975; Hagstrum and Subramanyam 2009; Borzoui et al. 2015; Athanassiou et al. 2016). Finding a suitable food source easily helps the establishment and the subsequent spread of *T. granarium*, rendering it in several cases more competitive than native species. It has been found infesting 96 different commodities, including animal (dead mice, dried blood and dried insects) and vegetal stored products, mainly grains, cereals, and related amylaceous commodities (Hagstrum and Subramanyam 2009). Recently, Athanassiou et al. (2016) studied the population growth of *T. granarium* on a variety of grains and amylaceous commodities. Although some commodities were more preferred and favored the rapid population growth, *T. granarium* could survive and develop in all tested commodities, and in all cases maintained a basic population (Athanassiou et al. 2016). This ability to survive, even in low numbers in unfavorable commodities, gives an extra advantage to *T. granarium*, compared to specialist species. Apart from being a generalist species, *T. granarium* takes advantage of its ability to feed on products with low moisture content (up to 2%) (Ahmedani et al. 2007), when most storage insects require a grain moisture content of at least 8% (Mason and McDonough 2012).

The larvae of *T. granarium* can undergo facultative diapause, which can last for years until conditions become favorable again for the insect (Burges 1962; Nair and Desai 1973; Bell 1994; Wilches et al. 2016). Diapausing larvae are particularly tolerant to several chemical and nonchemical methods (EPPO 2013; Wilches et al. 2016). This makes the control of this species very difficult and simultaneously helps the pest to overcome harsh conditions following the introduction in a new region, until conditions become favorable again. Moreover, *T. granarium* can have up to 10 generations per year, depending on the quality and availability of the food source and the environmental conditions (EPPO 2013). Its ability to complete a life cycle at an optimum temperature range (32–35 °C) in only 26 days (EPPO 2013), and its high reproductive rate helps *T. granarium* establish rapidly high populations following infestation. *Trogoderma granarium* can survive at a wide range of abiotic conditions, i.e., under extreme temperature and moisture regimes (Burges 2008; Wilches et al. 2016). At the same time, *T. granarium* can build up heavy infestations under hot and dry conditions that are unfavorable for development of most common storage insects (Hadaway 1956).

Once established, the eradication of *T. granarium* can be a difficult and costly procedure. Its ability to hide in protected locations (cracks, crevices, etc.) (Aitken 1975; Myers and Hagstrum 2012) and enter into diapause may negatively affect control measures. The success of eradication programs can be also impeded by the development of resistance to commonly used residual and fumigant insecticides, including phosphine, which has been demonstrated for several *T. granarium* populations (Bell et al. 1984; Bell and Wilson 1995; Kumar et al. 2010; Athanassiou et al. 2015; Kavallieratos et al. 2016). Although eradication is difficult, there are several historically recorded cases of effective eradication of *T. granarium* infestations. For instance, *T. granarium* was introduced and established in USA several times in the past decades, and in all cases, it was effectively eradicated, due to extensive eradication programs involving fumigation and surveillance (Myers and Hagstrum 2012).

The accurate identification of interceptions is a critical point in the attempt to keep invasive species out of a region (Armstrong and Ball 2005). Identification of *T. granarium* requires the expertise of a stored-grain entomologist, as many *Trogoderma* species that occur in storage facilities resemble *T. granarium* (Pasek 1998; EPPO 2013; ISPM 2016). In the 1940s, *T. granarium* was incorrectly identified as the black carpet beetle, *Attagenus piceus* Olivier (Coleoptera: Dermestidae), resulting in the spread of *T. granarium* in the west coast of the United States (Myers and Hagstrum 2012). Moreover, adult specimens are rare, whereas larvae, which are more often encountered in infected commodities, can only be identified after special preparation for microscopic identification, which is not in all cases available (ISPM 2016). Apart from the morphological identification of *T. granarium* individuals, immunological (Stuart et al. 1994) and molecular techniques (Yan et al. 2010; Olson et al. 2014) have been developed for this purpose. Although promising, these techniques cannot yet ensure fast and reliable distinction of *T. granarium* from the other *Trogoderma* species present in storage facilities, therefore, they cannot be exploited as quarantine diagnostic tools.

The Larger Grain Borer

Similar to other bostrychids, the story of *P. truncatus* is typical of a wood boring insect that abandons its natural forest hosts and adopts new feeding habits and habitats (Cogburn et al. 1984; Borgemeister et al. 1998; Nansen et al. 2002). *Prostephanus truncatus* switched from starchy roots, tubers and woods (Nansen et al. 2002 and references therein) to agricultural stored products (Hodges et al. 1985; Markham et al. 1991). It has morphological characteristics that are typical of xylophagous insects, i.e., strong mandibles and powerful mandibular muscles (Li 1988; Nansen and Meikle 2002), thus infestation by *P. truncatus* can be devastating for stored products, as it completely destroys the commodity (Hodges 1986; Kumar 2001). Losses due to the conversion of the grain into flour by adult feeding can be high, depending on the environmental conditions. Hodges (1986) stated that *P. truncatus* can be more destructive than “traditional” storage pests and reported losses up to 40% for maize cobs and up to 70% for dried cassava roots after 6 and 4 months of storage, respectively. Similar, high dry weight losses have been reported by other researchers for stored maize (Pantennius 1988; Muatinte et al. 2014) and dry cassava (Hodges et al. 1985; Borgemeister et al. 1997). This shows the devastating potential of *P. truncatus* and highlights its ability, once introduced into a new region, for extensive population development.

The origins of *P. truncatus* can be traced back to Central America and Mexico, where the species is native and probably coevolved with maize (Hodges 1986; Dunkel 1992). Early and recent records of the species from countries like

Nicaragua, Guatemala, Brazil, Peru, Colombia (Hodges 1986 and references therein) reveal the wide distribution of the pest in the New world, particularly in traditional maize growing regions. Outside the American continent, *P. truncatus* was introduced and reported for the first time in sub-Saharan Africa in the early 1980s, due to infested shipments of maize to Tanzania (Dunkel 1992), where it caused heavy infestations to locally stored maize and cassava stored production (Dunstan and Magazini 1981; Golob and Hodges 1982; Hodges et al. 1983). Since then, the pest has spread and established to at least other 16 African countries (Nansen and Meikle 2002; EPPO 2016b), and has been intercepted in several countries worldwide, such as Israel (Calderon and Donahaye 1962), Iraq (Al-Sousi et al. 1970), Germany (Scholler 2013), France (EPPO 2016b) and USA (EPPO 2016b). Relatively recently, adults of *P. truncatus* have been located in flours of a Sicilian bakery in Italy (Suma and Russo 2005), which were all immediately destroyed, and thereafter, no new record of its presence in Italy has been detected.

Prostephanus truncatus has been reported to cause heavy infestations and economically significant damage to stored maize and dry cassava (Hodges 1986; Muatinte et al. 2014). Particularly for maize, it can infest the stored cobs and grains, as well as the maize cobs in the field (Hodges 1986). In laboratory studies, it can develop also on sweet potato, white yam, as well as grains (Hodges 1986 and references therein, Markham et al. 1991; Roux 1999). Moreover, apart from crop host-plant species and agricultural products, *P. truncatus* has been reported to attack and reproduce on seeds and wood of various forest plant species (Makundi 1987; Nang'ayo et al. 1993; Nansen et al. 2002). These data suggest that *P. truncatus* may be able to utilize other potential hosts in warehouses and storage facilities, or even exploit, upon its invasion into a new region, non-crop hosts for its survival and establishment.

Although *P. truncatus* develops best at high temperatures and relatively high humidity levels (30 °C, 70–80% r.h.), where it can complete a life cycle in around 25 days (Bell and Watters 1982), the pest can develop in a wide range of climatic conditions. Its ability to establish in the cool highland areas of Mexico (Tigar et al. 1994) to the hot and dry sub-Saharan Africa (Golob and Hodges 1982; Hodges et al. 1983) highlights the potential of the pest to become established throughout the tropics and subtropics. Hodges (1986) suggests that the ability of *P. truncatus* to tolerate very dry conditions and develop in low moisture grains (Hodges and Meik 1984), similarly to *R. dominica* (Birch 1945), helps *P. truncatus* outcompete other storage insects which are more susceptible to dry environments.

Mites

Stored-product mites are often overlooked as stored-product pests, despite the fact that their presence is very common in various types of durable agricultural commodities, often associated with stored-product insects (Athanassiou et al. 2003, 2011). For example, apart from grains and amylaceous commodities where many species of mites are considered very common (Athanassiou et al. 2003; Stejskal et al. 2015), the mite *Carpoglyphus lactis* L. (Acarina: Carpoglyphidae) was recorded in 13% of dried fruit packages in the Czech Republic, often in extremely high densities (Hubert et al. 2011). Similar data have been reported from dried apricots from Turkey (Cobanoglou 2009). Moreover, Athanassiou et al. (2002) found that mites were also present in seed and ginned cotton, and in addition, there were some species in the final product that were associated with human health problems. Overall, it is generally considered that even if insects are not present, mites are very likely to occur (Athanassiou et al. 2005).

Stored-product mites have a wide range of conditions that favor their development, but generally, they can develop better in conditions of high r.h. and relatively low temperatures (Sinha 1995). Although their presence is associated with the presence of insects, their optimum development is different from that for insects (Sinha 1995; Athanassiou et al. 2003). Moreover, their spatial association with insects is directly related to their ability to feed on insect eggs and other life stages (Athanassiou et al. 2011). Mite predators, such as the *Blattisocius keegani* Fox (Mesostigmata, Ascidae) and *Cheyletus malaccensis* Oudemans (Prostigmata, Cheyletidae) feed on eggs of stored-product moths, such as the Mediterranean flour moth, *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) (Athanassiou and Palyvos 2006). Even plant-feeding, non-predatory stored-product mites have been reported to be opportunistic predators on insects. For example, the cheese mite, *Tyrophagus putrescentiae* (Schrank) (Astigmata: Acaridae), has been found to feed on eggs of the cigarette beetle, *Lasioderma serricorne* (F.) (Coleoptera: Anobiidae) (Papadopoulou 2006).

Apart from the infestation per se, stored-product mites can contribute to the dispersal of fungi in stored products, many of which are aflatoxicogenic (Rodriguez et al. 1980, 1984; Sinha 1995; Hubert et al. 2003, 2004a, b, 2006, 2011, 2013, 2014). More recent studies show specific interactions of stored-product mites with bacteria (Hubert et al. 2012a, b). In this context, the importance of stored-product mites for human health is high. Furthermore, stored-product mites are also allergens (Mariana et al. 2000; Thomas et al. 2002; Arlian 2002; Arlian et al. 2008). Apart from the European house dust mite, *Dermatophagoides pteronyssinus* Trouessart (Astigmata: Pyroglyphidae), and the American house dust mite, *Dermatophagoides farina* Hughes (Astigmata: Pyroglyphidae), there are major stored-product mite species that are allergens, such as several species of the genus *Suidacia* (Astigmata: Saproglyphidae), *T. putrescentiae* and the storage mite, *Lepidoglyphus destructor* (Astigmata: Glycyphagidae) (Mariana et al. 2000; Athanassiou et al. 2002; Hubert et al. 2003; Danielsen et al. 2004; Arlian et al. 2008).

There are numerous studies in the scientific literature that established the importance of stored-product mites in relation to infestations and quantitative degradation of stored products (Sinha and Wallace 1973; Sinha 1974, 1995; Sinha and Harasymek 1974; Cusack and Brennan 1975; Athanassiou et al. 2001, 2003; Hubert et al. 2003; Stejskal et al. 2015). Given that mites are considerably smaller in size than major insect species, they often remain undetected, unless their numbers are extremely high and their presence becomes visible. Moreover, most of the studies that are related to mites are based on mite sampling by absolute methods, i.e., by extracting the mites from a sample of a commodity (Cusack and Brennan 1975; Athanassiou et al. 2003; Stejskal et al. 2015). The most commonly used method for mite sampling in bulk grains is the Berlese–Tullgren method, whereby grain samples are placed under a light source and mites are extracted in a collecting vial through a funnel (Cusack and Brennan 1975; Athanassiou et al. 2003). The available techniques for trapping of stored-product mites are few and also labor-intensive. Wakefield and Dunn (2005) tested a trap that was effective for the detection of *Acarus siro* (L.) (Astigmata: Acaridae), *Lepidoglyphus destructor* Schrank (Astigmata: Glycyphagidae) and *Tyrophagus longior* Gervais (Astigmata: Acaridae) and found that this trap was not equally effective for the detection of all species, probably due to interspecific interactions.

Mites are generally considered more important than insects in counties with a moist climate. In a surveillance on stored grains in the UK, Wildey (2002) reported that the most frequently recorded arthropods were mites. In the Czech Republic and the former Czechoslovakia, Stejskal et al. (2015) noted that the frequency of detection in grain stores was higher for mites than for insects. In that study, the authors also noted that the composition of mite fauna has gradually changed and that *T. putrescentiae* is recorded more often than before, while the opposite is true for *Glycyphagus domesticus* De Geer (Astigmata: Glycyphagidae) (Stejskal et al. 2015). *Tyrophagus putrescentiae* is considered a mite species that can also develop at dryer conditions than other major stored-product mites, such as *A. siro* and *L. destructor* (Athanassiou et al. 2005). Earlier studies from southeastern Europe, where grains and conditions are drier, showed that this species is very common, and it is usually much more numerous than *A. siro* (Palyvos 2000; Athanassiou et al. 2003, 2005). Due to its increased frequency of detection in stored grains and related commodities, *T. putrescentiae* is now considered as an important pest (Stejskal et al. 2003, 2015; Hubert et al. 2009). This species is also common in other commodities of high protein content, such as ham (Abbar et al. 2016; Amoah et al. 2016). It should be mentioned that eggs of this species are very difficult to control with some of the aerial insecticides that are effective for other major species (Phillips et al. 2008; Zhao et al. 2012, 2016), which also contributes to the importance of this species as a pest of stored products.

Psocids

Psocids have been characterized for more than a decade as “emerging pests” (Nayak 2006, 2014; Throne et al. 2009). Psocids have been long regarded as pests of stored products and they were often detected, but their importance was underestimated. Overall, psocids were considered as secondary colonizers and less important than other secondary species of stored-product beetles and moths (Nayak et al. 2014). Nevertheless, currently, it is well established that psocids are able to colonize sound kernels of different grain commodities (Stejskal et al. 2006;

Athanassiou et al. 2010). According to Kučerová (2002) losses on grains by an infestation by psocids can exceed 10%. Although psocids can develop better in mixtures of intact with cracked kernels, they can develop and persist on whole kernels for an extended time (Athanassiou et al. 2010, 2014). Moreover, they are relatively long-lived at the adult stage and can survive to some extent without food (Nayak et al. 2014 and references therein). Finally, they are also capable of surviving in debris and frass (Athanassiou et al. 2010, 2014).

One other key characteristic on psocid biology is that many species have the ability to survive, develop and infest stored products at moderate relative humidity levels, contrary to established perceptions, thus some species can develop high populations in a relatively short time on grains and related amylaceous commodities (Opit and Throne 2008a, b; Athanassiou et al. 2014). However, probably the most important characteristic that classifies psocids as emerging pests of stored products is that they are tolerant to many insecticides used to control beetles and moths, which should not be considered as an outcome of previous exposure, but more as a natural phenomenon (Nayak et al. 1998, 2002a, b, 2003, 2014; Collins et al. 2000; Daglish et al. 2003). For example, *Liposcelis entomophila* Enderlein, *L. bostrychophila* Badonnel and *L. paeta* Peamlan (Psocoptera: Liposcelididae) were found to be tolerant to the pyrethroid bioresmethrin plus piperonyl butoxide (Nayak et al. 1998). In other studies, piperonyl butoxide + chlorpyrifos-methyl did not control *L. entomophila* (Daglish et al. 2003), whereas deltamethrin was not able to provide long-term protection against *L. bostrychophila*, *L. paeta* and *L. entomophila* (Nayak et al. 2002a, b). Organophosphorous compounds are generally more effective than other chemistries to control psocids (Nayak et al. 1998; Collins et al. 2000; Athanassiou et al. 2009). In contrast, insect growth regulators are not considered to be effective control agents (Bucci 1994; Nayak et al. 1998; Athanassiou et al. 2010). Currently, some psocid species and strains are tolerant to phosphine (Nayak et al. 2003, 2014; Dou et al. 2006) and sulfuryl fluoride (Athanassiou et al. 2012). As noted for mites, phosphine resistance contributes to the increased importance of psocids as stored-product pests. In addition, difficulties in detection complicate the assessment of psocid populations in storage and processing facilities (Stejskal et al. 2008; Throne et al. 2009; Opit et al. 2009a, b).

Currently, the increasing importance of stored-product psocids has been documented in several countries (Nayak et al. 2014). According to Stejskal et al. (2015) we can consider psocids as the most important emerging taxonomic group of stored-product pests in the last decade. Apart from the infestation to stored products, psocids are associated with the presence of fungi in various commodities, and they can cause allergic asthma to humans (Perotin et al. 2011; Nayak et al. 2014).

Future Emerging Pests

It is not easy to predict the future species that will be of increased interest at the post-harvest stages of agricultural commodities. Given that stored-product pests are in some sense urban and suburban pests, as well as agricultural storage pests, there is an increased concern about the presence of various species in urban food warehouses, retail facilities and even homes. For example, stored-product insects and mites become more important and frequently detected in pet food, which is a considerable source of infestation at the urban environment (Stejskal et al. 2015 and references therein). Finally, misidentification can occur in many cases, as for *T. granarium*, which is very similar to other species of the same genus (Adler 2013).

The gradual change of a species' status in its food preferences is not an unusual occurrence, as there are considerable interactions of the post-harvest ecosystems with other ecosystems (Dunkel 1992; Mack et al. 2000). There are examples of species that can infest products at the pre-harvest stages and then continue infestation in the store, such as the bean weevil, *Acanthoscelides obtectus* (Say) (Coleoptera: Bruchidae), or the dried fruit beetle, *Carpophilus hemipterus* (L.) (Coleoptera: Nitidulidae) (Aitken 1975; Hill et al. 2002). The forest system is the natural habitat for both *R. dominica* and *P. truncatus* (Cogburn et al. 1984; Borgemeister et al. 1998; Nansen et al. 2002). One of the most interesting examples of a species transition is that of the larger black flour beetle, *Cynaeus angustus* LeConte (Coleoptera: Tenebrionidae) (Dunkel et al. 1982; Dunkel 1992). This species was present in desert ecosystems in Nearctic and Neotropical areas, infesting plants (cactus and others). Less than eighty years ago, this species was first recorded in flour, grains, and other amylaceous commodities, and subsequently became more prevalent (Dunkel et al. 1982; Dunkel 1992). This ability to switch from the desert ecosystem to the stored-product ecosystem is an example of a transition to a new food source, and thus *C. angustus* is one of the most recent introductions to stored products.

Another example is species of the genus *Dinoderus* (Coleoptera: Bostrychidae). Buchelos (1991) recorded *Dinoderus brevis* Horn and the bamboo powder-post beetle, *D. minutus* (F.), in imported bamboo sticks in Greece. In that study, the author suggested that a further expansion could occur to other commodities, especially for *D. minutus*, which has a wider range of hosts, including stored products. Although the interception was on wood and grains, the vehicle for expansion may not be the commodity on which the pest can cause high economic damage (Buchelos 1991). Today, species of the genus *Dinoderus* are often reported from grains (Kumar and Okonronkwo 1991; Rees 1991) and their presence on cereals requires additional investigation and risk assessment. Similarly, *T. granarium* develops much easier on wheat than on rice, but the majority of the interceptions in US are on rice (Myers and Hagstrum 2012). If we include in the above pest categories the "emerging biotypes" that refer to strains with certain characteristics, e.g., resistance to phosphine, there are considerable "emerging" issues in global post-harvest ecosystems which must be considered. This challenge may require action at the species level, but, above all, also at the political level, to obtain a unified approach to mitigate the consequences of further expansion of the species/strains mentioned above. Harmonization of key export scenarios can play a role here, as there are different perceptions per county/ecozone that consist additional implications. For example, *T. granarium* is a quarantine species for countries such as USA, Canada, and Australia, but at the same time and during the same period, the species was excluded from the list of quarantine species in Europe.

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Chapter 11
Museum Pests–Cultural Heritage Pests

Pasquale Trematerra and David Pinniger

Introduction

Many types of artefacts are vulnerable to deterioration from biological, physical and chemical sources. Artefacts that consist of organic materials, such as fur, hides, linen, plant material, wood, wool, etc., can be infested by a range of insects. Most heritage areas have collections which are at risk, including archaeology, prints and drawings, contemporary art installations, folk art, fine arts, ethnography, books and archives, industrial and technical heritage and natural sciences. The major factors causing deterioration are the agents of decay such as mould, insects and rodents and the environmental effects of humidity, temperature and light (Zycherman and Schrock 1988; Child 2001; Montanari et al. 2008; Pinniger 2009).

In the past, there was widespread use of many toxic or persistent materials, such as arsenic and mercuric chloride and DDT to prevent textiles and natural history specimens from being destroyed by pests. However, many treasures have been lost over the years to pests such as carpet beetle *Anthrenus* spp., webbing clothes moth *Tineola bisselliella*, and common furniture beetle, *Anobium punctatum* (Florian 1997; Carter and Walker 1999). In recent years, conservators and other museum staff have worked to develop alternative strategies for preventing and controlling pests. Integrated Pest Management (IPM) strategies have been developed and adopted with success. IPM strategies based on detection and prevention of pests have been successful in many small and large museums, museum stores, historic houses, galleries, libraries and archives worldwide (Rossol and Jessup 1996;

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Pinniger 2011a). There are many parallels with IPM in stored product protection and in the food industry. However, in the heritage sector, these methods have been modified to take account of the special needs of the historic collections and the buildings in which they are housed.

As a general concept, IPM emphasizes the integration of disciplines and control measures including biological enemies, cultural management, sanitation, proper temperature utilization and targeted pesticide use into a total management system aimed at preventing pests reaching damaging levels. For cultural heritage collections, prevention is better than cure, and any action which retards pest attacks is to be preferred to remedial treatments. Failing to prevent pest attacks will lead to the loss of objects, information and knowledge. Keeping optimal storage or exhibition conditions is part of the conservation approach known as 'preventive conservation', as defined by the European Confederation of Conservator-Restorers' Organisations (E.C.C.O. 2002): 'Preventive Conservation consists of indirect action to retard deterioration and prevent damage by creating conditions optimal for the preservation of cultural heritage as far as is compatible with its social use'.

The concept of IPM has been applied in museums and historic houses since the 1980s (see, for example, Story 1986; Linnie 1996; Albert and Albert 1988; Bennett et al. 1988). Strategies for museums and IPM concepts were described by Florian (1997), Kingsley et al. (2001), Pinniger (2001), Pinniger and Winsor (2004), Strang and Kigawa (2006), Querner and Morelli (2010) and Xavier-Rowe and Lauder 2011. Recent and complete works on IPM in museums were written by Brokerhof et al. (2007) and Pinniger (2009). The most important part of IPM is the prevention of pest attack. The control of the indoor environment plays a central role in preservation of cultural heritage collections. This is implemented by sealing the building, regulating the climate, periodical general cleaning, introducing quarantine and regular monitoring the collection with traps.

Beetles belonging to the families Anobiidae and Dermestidae and moths belonging to the family Tineidae are major pests of collections in art and natural history museums. Pests such as the webbing clothes moth, *T. bisselliella*, the case-bearing clothes moth *Tinea pellionella*, the drugstore beetle *Stegobium paniceum*, the common furniture beetle *A. punctatum*, different carpet beetles (*Attagenus* spp., or *Anthrenus* spp.) and silverfish *Lepisma saccharina* regularly cause problems in museums. Mice, pigeons and mould can also present a risk to collections in certain circumstances (Mound 1989; Strang and Dawson 1991; Pinniger 1994, 2009; Florian 1997; Ranacher 1998; Child 2001; Trematerra and Süss 2007). Also, there are many additional wood borers that can seriously damage the wooden artefacts or books, paper etc., such as termites and beetles such as Lyctidae, Bostrychidae and Cerambycidae.

This chapter will provide information on advanced and sustainable pest prevention and control strategies, to be used in worldwide cultural heritage institutions. The focus will be on alternative pest management strategies of insect pests.

Materials Most at Risk from Pest Attack

Materials most at risk from pest attack are animal skins, papers, dried food, dried plants and seeds, feathers, freeze-dried natural history specimens, fur, hair, insect specimens, materials rich in starch, parchment and vellum, sapwoods, silk, wool, and any damp organic material. Entire collections of books and entomological specimens have been lost; more common is damage such as holes or 'grazing' on the surface of textiles; an object's value for display can be lost and important decorative or aesthetic features destroyed. In general, dirty and neglected objects in dark places will be more at risk than those that are clean and in well-lit areas. The rate at which insect pest consume food is strongly affected by environmental conditions, the nature of the food materials themselves (e.g. age, nutritional value and specific composition). Furthermore, it seems that presence of organic dust is important for survival of some insect species. Improved knowledge about these factors may be an important tool for predicting the development of insect pest damage in various types of museum artefacts. Although moths (Lepidoptera) beetles (Coleoptera) and termite (Isoptera) species cause most insect pest problems, other insects, such as cluster flies (Calliphoridae), ants and cockroaches can also cause a great nuisance. Vertebrates (rodents and birds) can damage collections by eating them, shredding them for nesting material and staining from their urine and faeces.

Developing an IPM Strategy

In the past, chemical methods were used for control of pests in cultural heritage collections. However, this has proved problematic with regard to deposition of unwanted chemicals on the artefacts, and unwanted side effects on humans. In recent years most museums have turned to non-chemical methods as alternatives, these control methods include the use of high or low temperatures, or low oxygen atmospheres (Rossol and Jessup 1996; Florian 1997; Ranacher 1998; Pinniger 2001).

The key to avoiding pest infestations is to understand the conditions under which they thrive. The four principal needs for these pests are food, warmth, humidity and harbourage. These factors are often inter-linked. It is also important to develop procedures, such as quarantine of incoming material, so that a pest is not introduced as a result of normal collection activities. It is also important to identify degrees of risk from pests to collections on display and in store. The concept of 'Risk Zones' is based on the principle of assessing each area of a museum or historic house and assigning a Risk Zone, either Very High (red), High (orange), Low (yellow) or Very Low (green). Procedures for monitoring housekeeping and action plans are then based on the appropriate zone (Doyle et al. 2007). This Risk Zone approach is now being used successfully as the basis of IPM programmes in a number of museums in the UK (Pinniger 2011a).

Developing an IPM strategy:

- prevent entry of pests (insects, birds, and rodents) into buildings;
- develop good exterior building maintenance and appropriate landscaping;
- avoid practices and habits that attract pests;
- moderate interior high humidity and temperatures;
- develop good interior housekeeping practices;
- maintain appropriate food/trash removal practices;
- assess all display and storage areas for risk from pests and assign appropriate risk zones;
- set up and implement measures to detect and monitoring pests;
- inspect all incoming museum objects;
- inspect stored collections periodically for pest activity;
- take actions that reduce the source and spread of the pest infestation;
- isolate infested materials, and choose the most appropriate safe control method (s) for eradication.

Reviewing the IPM procedures, with periodic assessment of the effectiveness of the strategy and modifying in order to improve it.

Cleaning is probably the most important part of any IPM programme. The material that attracts insects is normally organic, such as leather and wool, paper, wood, though some insects can make do with dust and fluff derived from these. A close examination will usually show accumulations of organic dirt and debris in corners, wall/floor angles and behind fittings that will support and feed insect pests. Unused rooms and storage areas are often neglected and dirt and debris will provide food and an ideal harbourage for pests. Periodic deep cleaning of stores is recommended. All horizontal surfaces where dust and rubbish can accumulate should be cleaned, including the tops of storage and display units, light fittings and ledges, etc. Special care should be taken in less accessible areas.

Common sites where pests hide within buildings undisturbed by human activity include: unused or little frequented rooms and cupboards; cracks between floorboards; cavity walls and floors; gaps between walls and floors; dead spaces behind and under storage cabinets; heating and ventilation ducts; felt sealing strips on doors; old or discarded display material; deaccessioned material which has not been removed from the site, etc. Many of these sites are difficult to inspect.

Temperature

Cool conditions will discourage insects from breeding and any areas with temperatures of 20 °C and above will encourage insect breeding. Temperatures of 24–25 °C will allow insects to complete their development much more quickly. This may result in pests such as clothe moths completing two or more life cycles in a year. Although it may not be possible to lower temperatures in public areas, object stores should be at as low a temperature as is practical. However, ensure that the relative humidity levels are not permitted to rise to unacceptable levels. Direct sunlight can cause local hotspots even in cool areas, and preventive measures such as shading should be installed. Also, uneven temperatures can result in localized condensation on cool walls. Vertebrate pests are much more tolerant of high or low temperatures than insects. They have much better temperature regulatory systems and so have more potential nesting sites available to them.

Humidity

Many insect species can tolerate a wide range of relative humidity (r.h.) and some will survive for long periods in a dry environment, for example, the biscuit beetle, *S. paniceum,* but other species, such as furniture beetle *A. punctatum*, need a damper environment. Silverfish, *L. saccharina*, will only breed rapidly and cause serious problems in conditions of above 70% RH. Booklice (Psocoptera), also need a higher level of humidity, they are often found in damp basements or in localized damp areas. However, there are species that can survive at r.h. levels that are close to 60% (Opit and Throne 2008). Some other insects have a strong preference for environments with a higher r.h., this is because at these levels moulds and fungi have begun to attack their food sources making it easier to digest. Vertebrate pests need water to live, so it is important to eliminate potential sources of drinking water such as leaking pipes and taps, and cover all open drains with securely fitted wire mesh screens. By establishing appropriate environmental conditions, it is possible to limit the threat from a large number of potential insect pest species. This can be achieved by mechanical means but you also need to ensure that there are not localized areas of high humidity where some pests will thrive. These can be caused by condensation, poor damp proofing, broken or missing damp-proof courses, or leaks from gutters or water, sewage and heating pipes.

Inspection Area/Quarantine Area

An essential part of any pest prevention policy in a museum is to keep pests out of collections. Insects can be introduced from many sources, including new acquisitions, objects on loan from other collections and items returned from loan. A quarantine area must be established which is physically isolated from display areas and collection storage. Objects must be checked for infestation before being allowed into the main collection areas, whether storage or display. Inspection may reveal insect damage and clothes moth webbing, but insect eggs or small larvae are more difficult to see. Similarly, the emergence holes of wood boring insect may be obvious but any developing larvae will be hidden in the wood. If it is suspected that there may be an active infestation, the objects should be isolated and allowed to incubate over a period. If adult insects are seen to emerge, then remedial action can be taken. Pests may also be living in non-collections objects such as figures or other ‘props’ in displays. Packing materials may also be infested and will need to be checked and possibly treated or disposed of. Some museums have established procedures that treat all appropriate items, when they are acquired or on return from loan (Pinniger 2011b).

Keeping Pests Out

Good building construction practice and subsequent maintenance are essential if insects are to be kept out of a building. Excluding pests from older and historic buildings is often far more difficult. Most modern buildings successfully prevent the entry of pests by careful design and detailing of potential entry points such as doors, windows and vents (Scott 1991; Trematerra and Fleurat-Lessard 2015). All unnecessary openings should be blocked. Check brickwork for cracks and damaged pointing and initiate remedial work as soon as possible. Doors and windows can be fitted with unobtrusive sealing barrier strips. Windows and doors can be fitted with fly mesh screening; splits and holes in wooden doors and windows or their frames should be caulked. Cupboards, cabinets and drawers, which may appear to sound, should be inspected because they may have hidden cracks that can allow insect access. Particular attention needs ground, basement and roof areas. Where it is aesthetically acceptable, it is worthwhile cutting back vegetation for at least 3 m around the perimeter of buildings.

Specific measures to exclude rodents include wire mesh screens and grills fitted to ventilation louvre or other openings that cannot be sealed; sheet-metal cladding fixed to the base of wooden doors, windows or walls in high-risk areas or where there are signs of gnawing; insert steel mesh into gaps around pipes and in eaves to prevent access but without restricting ventilation; ensure that exterior drain covers and rodding caps are sound and close fitting; and fitting metal cones or screens around exterior pipework, cables and poles (Doyle et al. 2007). Measures to discourage birds from nesting or roosting include netting or wire screens fitted over balconies, alcoves, light wells and other openings. Steel mesh fitted into the gaps in eaves will prevent access, but without restricting ventilation. Prevent birds perching on ledges and sills by fitting sharp metal wire spikes and strip; anti-perching gel. Spikes (pins) and wires are more expensive but more durable. Cap chimneys to prevent birds getting inside and nesting.

Identifying Pests and Pest Activity

Although it is often the adult insects that are found, it is the larval stage that does the most serious damage. Adults will be more active and obvious during the warmer summer months but the larvae will feed and grow throughout the rest of the year. Signs of activity may include emergence holes and dead adults, but also silk webbing and cast skins of larvae and frass (Moud 1989; Pinniger 1994; Florian 1997; Chiappini et al. 2001; Trematerra and Süss 2007; Trematerra 2016). Some insect species are widespread and are a serious threat to cultural heritage items as they feed on a large range of organic materials.

Typical signs of a rodent problem are as follows: gnaw marks near the base of doors and cabinets. They can gnaw through wood, particleboard, plaster and asphalt, as well as soft metals such as zinc, lead and aluminium. Faecal droppings and urine stains may also be seen. Rats do not normally have a distinctive odour but mice have a musty smell reminiscent of stale biscuits. The house mouse, *Mus domesticus*, is the main cause of concern, although in many collections and historic houses the fear of infestation is often far greater than the real risk. Functions, cafes and restaurants which provide ample supplies of food for rodents are the main source of infestation problems. Brown or Norway rats, *Rattus norvegicus* and black rats, *Rattus rattus*, are rare in museums and usually only found associated with considerable and accessible sources of human or animal food (Strang and Dawson 1991).

The most common bird pests are pigeons (*Columba livia*). Roosting or nesting birds can be a nuisance on windowsills, ledges and other architectural features. Their droppings, which are mostly urine through which they excrete uric acid, cause unsightly stains and can damage the building fabric. These droppings pose a health hazard to humans as pigeons carry parasites and spread disease. The nests, feathers and other debris attract insect pests such as clothes moths and carpet beetles, and these can move into the building and cause damage to the collection. Birds can also become trapped in chimney flues, where they quickly die and become an attractive food supply for several insect pest species.

Museum Pests

Any insect pest with chewing mouthparts is a risk to museum specimens. Carpet beetles, clothes moths, powderpost beetles, cockroaches and others pose threats to specimens through feeding damage, frass, faeces and excretions. Some pests pose indirect risks such as fires (rodents gnawing on wires) and secondary infestations (dead cluster flies in attics can attract carpet beetles). Any insect pest that infests houses, restaurants or other buildings may at some time also become a pest in a museum. They can loosely be grouped into five categories: textile pests; wood pests; stored product pests; pests associated with mould and high humidity; general pests. The first step in solving any pest problem is to identify the pest and learn about its biology and habits. The identification of the insects present in heritage sites is essential in order to undertake preventive and curative measures. Frequently, the accurate identification of the insects requires complex keys often based on very specific morphological characters. This identification becomes even more difficult when considering the larval stages of insects. Therefore, accurate identification very often requires verification by entomologists.

Textile Pests

Most insect damage to textiles is caused by carpet beetles (family Dermestidae) or clothes moths (families Tineidae). The adults may be attracted to lights and windows, but this is not the stage that does the damage, as adults feed outside on pollen or not at all (Aitken 1975). It is the larva stage that feeds on fabric, fur, feathers or virtually anything made of animal fibres.

Carpet Beetles

Immature carpet beetles feed on dried animal products such as wool, silk, felt, hair, fur, feathers, dead animals, and taxidermy. The adults are often seen in spring crawling up walls and congregating on window ledges. Carpet beetle larvae are repelled by light and are usually found burrowed deeply into the infested material or in little-used drawers, cases and storage bins. Four species of carpet beetle are most likely to be found in museums (Fig. 11.1a).

Anthrenus (carpet beetles)

Adult *Anthrenus* are 2–3 mm long and covered with grey and gold scales; they fly in warm weather and may frequently be found on windowsills. The eggs hatch into short, fat, hairy larvae which are extremely small (less than 1 mm) and can get through very small cracks. As the larvae grow, they moult leaving empty hairy, cast skins which may be the first signs of attack. The larvae will wander widely and chew holes in textiles, bindings and mounts with animal glue. They eat other protein-rich material such as wool, fur, feathers, silk and skins. They will also feed on dead insects and are often found in birds' nests. One of the most common species worldwide is the varied carpet beetle, *Anthrenus verbasci*, a destructive pest of textiles and natural history specimens. There are a number of other species of *Anthrenus* including *Anthrenus flavipes, Anthrenus sarnicus* and *Anthrenus scrophulariae*.

Anthrenus verbasci (varied carpet beetle) is primarily a scavenger, and it is common in the nests of birds and on dead animals. It can damage woollens, carpets, wall hangings, hides, horns, taxidermy and bone artefacts, and insect collections. Small populations often go unnoticed behind furniture or along baseboards feeding on

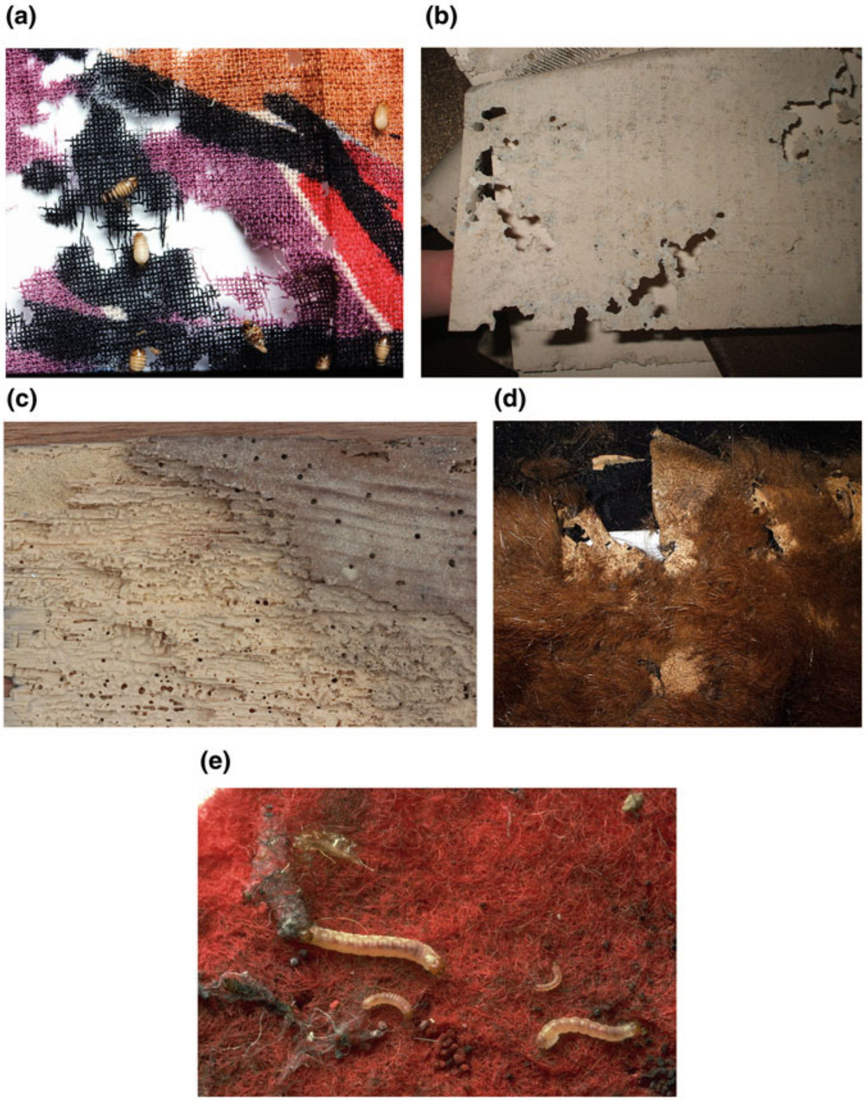

Fig. 11.1 **a** Infestation of *Anthrenus verbasci* carpet beetle larvae eating wool textile © DBP Entomology **b** Damage to paper documents by silverfish *Lepisma saccarhina* © DBP Entomology **c** Damage by furniture beetle *Anobium punctatum* to a wood floorboard from a historic house © DBP Entomology **d** Damage to a fur coat lining by webbing clothes moth *Tineola bisselliella* © DBP Entomology **e** Larvae of webbing clothes moth *Tineola bisselliella* eating wool felt textile © DBP Entomology

accumulated lint, hair, food crumbs, dead insects and other organic debris. The adult is 2–4 mm, oval to round, with patches of white, yellow and black scales on its back. The larva is teardrop-shaped and covered with rows of spiky brown hairs.

Anthrenus flavipes (furniture carpet beetle) attacks furniture (particularly old horsehair-stuffed furniture) and items made from wool, fur, feathers, silk, horns and tortoiseshell. It is commonly found in the USA and warmer parts of Europe. The adult is 2–4 mm and is rounded and blackish with variable mottling of yellow and white scales on the back and yellow scales on the legs. The larva is much darker and more hairy than the varied carpet beetle.

Anthrenus scrophulariae (common carpet beetle) attacks carpets, woollens and animal products such as feathers, furs, leather, silks, mounted museum specimens and pressed plants. It is mainly found in the USA. The adult is 2–4 mm with a band of orange scales down the middle of its back. The larva is reddish brown and covered with brown or black hairs.

Attagenus (fur and carpet beetles)

The genus *Attagenus* are mainly scavengers on the dried material of animal origin. Adults are oval, hairy beetles 3–5 mm long. Several species are uniform black in colour. Larvae are tapered, hairy and elongate, and when grown are about 10 mm long. Household infestations typically start from birds' nests and dead birds or rodents in roof spaces or wall cavities. They may also be found in the nests of bees and wasps. Adults can also be found feeding on nectar on flowers.

Attagenus unicolor (black carpet beetle) is the most abundant and destructive of these carpet beetles in the USA and some European countries. The adult is 3.5–5 mm long, a solid dark brown or dull black colour, and more elongate than *Anthrenus* carpet beetles. The larva is tapered from the front carrot-shaped. It is covered with bands of golden brown hairs and has a characteristic 'tail' of long hairs at the rear end.

Attagenus pellio (the two-spot carpet beetle) is found in the cooler parts of North America and Northern W Europe. The adult is 3.5–5 mm long, a dull black colour with two distinct white spots on each wing case. The larva is tapered carrot-shaped covered with bands of dark brown hairs and has a characteristic 'tail' of long hairs at the rear end.

Attagenus smirnovi (brown carpet beetle or vodka beetle) is a recently introduced pest to Europe, but has spread rapidly to many countries including Denmark, Sweden and the UK (Pinniger 2011a). The adult is 3.5–5 mm long, dark brown with a black thorax and head. The larva is tapered carrot-shaped covered with bands of brown hairs and has a characteristic 'tail' of long hairs at the rear end.

Dermestes

Dermestids are 2–12 mm long oval or elongate oval beetles with short, clubbed antennae. They usually have a distinct colour pattern, and many are covered with fine setae or scales. Larvae and adults of *Dermestes* species feed on many dry substances of animal origin such as bones, dried meats, feathers, fur, glue, leather and skins; they may also attack food products of vegetable origin. Some museums use cultures of *Dermestes* for cleaning animal carcasses to prepare skeletons and as they can cause havoc if they are allowed to spread into other areas, they should never be used on the museum site.

Dermestes maculatus and ***D. peruvianus*** (hide and leather beetles). The hide or leather beetles, as the name suggests, will attack leather and skins but they will not feed on tanned leather in good condition. The larvae can be found on dead animals, and they also attack book bindings with animal glue. The adults are black or dark brown and larger (6–10 mm) than carpet beetles. The larvae are also dark brown and large and very hairy and they have two distinctive spikes at the back end. The larvae have a very destructive habit of tunnelling into objects to pupate and they make big holes.

Clothes Moths

These are small, silvery-gold moths with a wingspan of 10–15 mm. They have narrow wings fringed with long hairs. They avoid light and are rarely seen flying, they prefer dark corners, closets, and storage areas, and usually remain out of sight. A number of species of moth will attack and cause serious damage, the larval stage causes damage to woollen clothes and objects such a feather hats, dolls and toys, bristle brushes, weavings, and wall hangings, they very rarely damage books and papers (Fig. 11.1d, e).

Tineola bisselliella, the webbing clothes moth, and ***Tinea pellionella***, the case-making clothes moth, are the two most common clothes moths found in museums both have a wide distribution. The larvae feed on the surface of the material infested. The webbing clothes moth larvae produce feeding tunnels of silk and patches of silken webbing on the fabric's surface. The case-making clothes moth larva is rarely seen since it constructs a cylindrical case of fabric, which it carries around to hide and feed in. The grain moth ***Tinea granella*** was found on dried plants, corks dried animals and fungus (Trematerra and Süss 2007; Trematerra 2016). Other moth species such as the white-shouldered house moth, ***Endrosis sarcitrella***, and the brown clothes moth, ***Hofmannophila pseudospretella***, are very common in old houses where they can be often found in bird nests in blocked chimneys. The larvae will occasionally tunnel into bindings of books but very rarely cause damage to clean, dry materials. Larvae of Indian meal moth ***Plodia interpunctella*** live on foods, dried herbs and spices and may be found in food areas.

Wood Pests

Materials made of wood are susceptible to attack by a number of wood infesting pests. The culprits in museums in cooler countries are furniture beetles and in hotter climates, it is usually powderpost beetles or drywood termites, but often other termite categories, such as the subterranean termites. All can severely damage valuable artefacts because the larvae are hidden away in the wood.

Powderpost Beetles

The term powderpost beetle applies to any of two related families: Lyctidae (true powder pest beetles that cannot digest cellulose and usually attack the sapwood of hardwood), and Bostrychidae (false powderpost beetles that attack both softwood and hardwood). The term ‘powderpost’ comes from the very fine, powder-like frass (excrement and bits of wood) produced by the feeding process. Powderpost beetles infest wooden artefacts and materials such as frames, books, toys, furniture, tool handles, gunstocks, bamboo, lumber, panelling, and crating, as well as girders, studs, flooring, and other wooden building components. Look for exit holes where the talc-like beetle frass will be found.

Adult powderpost beetles, ***Lyctus brunneus, L. linearis*** and often ***L. africanus***, leave exit holes which are circular 0.8–1.6 mm diameter, the frass is loose and has a very fine powdery consistency. There are many species of Bostrychid beetle ranging from the very small bamboo borers ***Dinoderus minutus*** or ***D. brevis*** to the large ***Heterobostrychus***. The adults leave exit holes which are circular 1.5–7 mm in diameter. The frass has a tendency to stick together in clumps, feeling more gritty to the touch powderpost beetle spend most of their lives unseen as larvae tunnelling within wood, so their frass and the exit holes they make in the wood’s surface as they emerge as adult beetles are the primary signs of their presence. In warm conditions, the life cycle can be completed in a year. The adult beetles are short-lived and seldom seen.

Furniture Beetles

The common furniture beetle or woodworm, ***Anobium punctatum***, is widespread in most temperate countries where it infests buildings, furniture and wooden objects and may also attack books. The larvae make tunnels in compressed paper and take 2–5 years to complete development depending upon the food and the conditions of temperature and moisture content. Adults emerge in spring leaving small 1.5–2 mm exit holes with a pile of gritty frass below the hole. *Anobium* infestations will survive in cool, damp conditions but do not thrive in hot, dry conditions with humidities below 65%. Outbreaks of woodworm activity are usually confined to collections which have been brought in from damper storage in cellars, outbuildings or areas where there are leaks, condensation and poor air circulation. *Anobium* larvae will attack wooden shelving and wooden boards in books, preferring starchy hardwood and softwood, they will not attack sound heartwood. Plywood or bindings with animal glue are particularly susceptible, because of the added protein, and can be severely damaged (Fig. 11.1c).

Other Anobiidae beetle species which have recorded as attacking timber and books are ***Xestobium rufovillosum*** (death watch beetle), ***Catorama herbarium*** (the Mexican book beetle), ***Nicobium castaneum***, ***Ptilinus pectinicornis***, ***Ernobius mollis***, ***Oligomerus ptilinoides*** and ***Gastrallus imarginatus***.

Termites

Termites are the world's most serious and destructive pests of structural timber. In many museums, libraries and archives, termite infestation of the buildings has spread to display and storage furniture, archives and book collections, which are then seriously damaged. There are many different species of termites and they are generally divided into two pest types with distinctly different lifestyles—drywood and subterranean. Termites are not a serious problem in cool temperate countries and the most severe problems are encountered in countries with warm temperate and tropical climates (Daniel 2001).

Cryptotermes and ***Kalotermes*** (drywood termites). Drywood termites, which include the genera *Cryptotermes* and *Kalotermes*, bore tunnels and galleries in wood in many directions, and also live in compacted paper and books. Although some faecal pellets are scattered in the passages, large quantities are often stored in chambers or thrown out of the wood through 'toilet holes'. Drywood termites attack wooden items of all kinds and establish colonies in dry, sound wood with low levels of moisture and do not require contact with the soil. Most infestations are in building structures but they may spread into furniture and wooden objects in museum collections which are then attacked. The excavated galleries or tunnels feel sandpaper-smooth. Dry and loose, their distinct six-side faecal pellets are found in piles where they have been kicked out of the chambers. Other than the pellets, there is very little external evidence of drywood termite attack in wood as they tend to work just under the surface of the wood. Swarming winged adults may also be a sign of an infestation. Winged termites have four wings of equal length and when the wings, are shed after swarming they may be visible. However, given that usually swarming occurs only once per year, termites usually remain unseen during most of the time.

Reticulitermes, ***Coptotermes*** and ***Macrotermes*** (subterranean termites). Subterranean termite colonies need to live in contact with soil and some species, including *Reticulitermes*, *Coptotermes* and *Macrotermes*, will spread from the natural environment of soil and trees to the woodwork of buildings. Many species require fungi in their diet, which is produced on decaying wood or paper within the nest. They need to maintain high levels of moisture in the colony for the development of fungi and to prevent the desiccation of the nymphs and workers. This leads to the characteristic tube-building habitat of subterranean termites. These tubes, which may be some metres in length, are constructed of soil and faecal material which protect the termites as they pass between nests in the soil to the food sources of wood or paper. Subterranean termites are found near or below ground level and seldom spread above the lower floors, this means that collections in basement areas are particularly at risk. They may invade libraries and archive stores and completely destroy the inner parts of books and bound archives, just leaving an outer skin of bindings or packaging. The eradication of termites and their subsequent exclusion can be extremely difficult and often requires specialist expertise.

Pests of Dried Food

Many museums include items made in part of seeds, nuts, grains, spices, dried fruits and vegetables, and other foods. A long list of pests, traditionally called stored product pests, can infest items containing these foods. Probably the most common of such pests in museums are the cigarette beetle and the drugstore beetle.

Stegobium paniceum (Biscuit beetles or drugstore beetles). *Stegobium* belongs to the Anobiidae, the same family as the common furniture beetle or woodworm, *A. punctatum,* although it will not attack wood. When it is warm (above 22 °C), the adults are very active and will fly to light sources. *Stegobium paniceum* is a major pest of dried plant specimens, and it is also a serious pest of books and manuscripts where it usually lives in the starchy bindings. It has also been found living in dried animal specimens and mummies larvae feed by tunnelling through a wide variety of foods and spices (particularly paprika or red pepper and has been known to chew through tin foil and lead sheeting). The adult drugstore beetle is rounded, reddish brown and hairy it has a three-segmented antennal club.

Lasioderma serricorne, cigarette beetle, is named for the fact that it is a pest of stored tobacco, but is also a serious pest of flax, spices, crude drugs, seeds, and, most importantly for museums, books and dried plants. This beetle has been called the ‘herbarium beetle’ in hotter countries because of the damage it can cause to dried herbarium specimens. It has also been found infesting rodent baits. The adult beetle is very similar to the cigarette beetle but smaller and much shinier antennae with 14 elements.

Pests Associated with Mould and High Humidity

Moulds are fungi that can cause damage or disintegration of organic matter. When moisture and other environmental conditions are right, mould spores can germinate and develop and cause significant damage to wood, textiles, books, fabrics, insect specimens, and many other items in a collection.

Psocids or Booklice

They feed on microscopic mould growing on paper and in the starchy glue in the binding. Psocids also infest such items as dried plants in herbaria, insect collections, manuscripts, cardboard boxes, and furniture stuffed with flax, hemp, jute or Spanish moss. Psocids are so small they rarely cause serious damage. However, their presence often indicates a moisture problem and the likely presence of damaging moulds. They are tiny insects, less than 1–2 mm, and range in colour from clear to light grey or black. Most indoor psocids are wingless.

Liposcelis bostrychophila and other species of *Liposcelis* (booklice). There are a number of different species of booklice which have very different habits and needs. *Liposcelis bostrychophila* is the most common species in heated buildings (Florian 1997). The adult is wingless and very small (less than 1 mm). They develop through a series of nymphal stages which feed on microscopic moulds on a range of substrates including flour, paper and cardboard, dried plants, herbaria, insect collections, manuscripts, cardboard boxes, furniture stuffed with flax, hemp, jute or moss. Populations of *Liposcelis* can increase very rapidly if temperatures rise above 25 °C (Opit and Throne 2009), and this gives rise to apparent population explosions. Although damaged by a few booklice may be negligible, large numbers of booklice will graze the surface of books and papers. In addition, squashed bodies will stain materials and may encourage moulds.

Adistemia spp., ***Cryptophagus*** spp. and ***Mycetophagus*** spp. (plaster beetles and fungus beetles). There are many species of these small brown beetles, 1–2 mm long, including ***Adistemia watsoni***, ***Cryptophagus nitidulus***, ***Mycetophagus quadriguttatus***. They feed on microscopic moulds and are often found in large numbers when papers and books are stored in damp areas. As they graze, they may cause some superficial damage to the surface of the paper.

Crustacea (Woodlice)

Woodlices are not insects but belong to the Crustacea, which also includes shrimps and crabs. There are a number of species that come into buildings, most are greyish-brown and range in size from a few millimetres to 15 mm. Woodlice live in damp, rotting vegetation and wood and may graze on damp paper and cardboard. They are often found in basements or near doors and windows where they have wandered in from damper outdoor environments. As they cannot survive for long in dry conditions, most soon die of desiccation without causing any damage. If there is a persistent problem of live woodlice, the area should be investigated as there is probably local high humidity and rotting wood.

General Pests

Any household pest may become a pest if it gets into a museum. Cockroaches, silverfish, ants, rodents and other common pests can invade and cause problems in museums.

Cockroaches

Cockroaches are omnivorous and feed on leather, paper, glues, animal skins, hair and wool fabrics, especially if the item is stained with food and sweat. German cockroaches ***Blattella germanica*** (10–16 mm) are found indoors in warm, humid areas, preferring crevices near food and water in bathrooms and kitchens. The brown-banded cockroach ***Supella longipalpa*** (11–14 mm) is also found indoors and requires less moisture. The German and brown-banded cockroaches are the only known domestic cockroach species that depend on human activities for survival. The oriental cockroach ***Blatta orientalis*** (18–28 mm) prefers decaying food, is cold tolerant and prefers damp areas with temperatures below 29 °C. In warmer countries, it can be found in bark mulch around the perimeter of buildings. The American cockroach ***Periplaneta americana*** (28–53 mm) requires a water source and prefers fermented foods. It can be found in sewers and basements, particularly around drains and pipes. Cockroach faecal pellets can resemble small mouse droppings without pointed ends. All species lay their eggs in egg cases (oothecae), and then these hatch into tiny nymphs which develop rapidly in the right conditions. Identification of the insect adults, nymphs and egg cases and an understanding of its life cycle are critical to determine what are the risks and assess the most appropriate control measures. ***Blaptica dubia*** was reported by Montanari et al. (2008) as pest of photographs.

Lepisma and ***Ctenolepisma*** (Silverfish). Silverfish are always associated with damp conditions and they require localized humidity above 70–80% to breed and multiply. They are primitive, scaly, wingless insects (10–15 mm) with three bristles (cerci) at the tail end. Silverfish feed on starch, glue, ink and microscopic moulds paper, paper products and textiles (cotton or artificial silk), they will also eat organic glue on wallpaper. There are a number of silverfish species including ***Lepisma*** and the larger ***Ctenolepisma***. Silverfish damage can be recognized by the ragged, scraped surface areas and irregular holes in paper. They are serious pests in humid countries, but in temperate climates, they are usually confined to damp rooms and basements (Fig. 11.1b).

The related firebrat ***Thermobia*** will also damage paper, photographs and bindings but needs hotter and drier conditions.

Ptinus tectus and ***Niptus hololeucus*** (spider beetles). Spider beetles are common in birds' nests and general debris in attics, basements and stores where they will feed on a wide range of vegetable and animal detritus. The adults are 3–5 mm, hairy and superficially spider-like. The brown, hairy Australian spider beetle, ***P. tectus***, and the golden spider beetle, ***N. hololeucus***, are common in many temperate countries. The larvae are similar in appearance to those of the biscuit beetle and they will also boreholes and cavities in paper and wood before pupating in a globular silk cocoon.

Rodents and Birds

Rodents and particularly mice, *M. domesticus*, will seriously damage paper when the female mice collect and shred paper to make nests. They will also damage books by their habit of gnawing hard objects to keep their teeth sharp. Additionally, mouse urine and droppings can stain paper and also present a disease hazard.

Birds (like pigeons) will rarely directly damage books but droppings can be unsightly and corrosive.

Monitoring

Regular checking is needed to look for live adults and larvae and the presence of shed larval skins or faeces. The presence of fresh feeding debris or frass around or below specimens is an indication of an active infestation. Exit holes, feeding holes, hair falling from fur or pelts, mats of fibres, silken feeding tubes or cases, or moth or beetle pupae are also signs of activity. Windowsills and the inside of ceiling light fixtures should be checked on a regular basis as many pests fly or crawl to light. Pests may be found behind baseboards, under furniture, behind mouldings, in cracks in floors, behind radiators, or in air ducts.

The Use of Insect Traps

Traps are used to detect the presence of insects and rarely to control them. Trap catches can show an increase in insect numbers in a specific area; the spread of a pest from one area to another; an invasion of the adult insects in summer; or the failure of a control treatment. Traps should be used as a supplement to visual inspection but not as a replacement. The information they provide can then be used to identify what preventive and remedial measures are required and to establish priorities.

A range of sticky traps is available that work on the principle of the wandering insect blundering into the trap and becoming stuck on the non-toxic adhesive surface (Pinniger 2009; Child 2011). The traps for Coleoptera are designed to be placed on the floor and are most effective when placed in corners and wall/floor angles, traps for Lepidoptera are designed to be suspended (at about 2.50 m from the floor) in the environments. In historic houses, place a trap in fireplaces to check for pests in blocked and disused flues.

Most traps will remain effective for at least a year and need to be checked at regular intervals. It is better to check regularly every two months than to start by checking every week and then finding that the workload is too great. A minimum regime would be to check traps four times a year, in March, June, September and December. It is also important to check the stickiness of the trap at the same time as you check for pests. Dust can build up on the adhesive which renders it useless. The greater the number of traps used, the greater is the chance of finding insects. However, the workload should not be underestimated and trapping programmes should be designed to be manageable. Traps should be placed in a regular grid pattern and all traps date labelled and their position marked on a plan.

Insects caught in traps should be identified and the information recorded in a log. Record whether the insects caught are larvae or adults as an adult beetle may simply have wandered in from outside. If it is a larva then it is almost certain that the species is breeding within the building. Over a period of time, careful monitoring of the traps enables a picture to be built up of insect distribution. Additional traps can be placed in areas where pests need to be more accurately pinpointed.

Large numbers of non-pest insects may be caught on traps, especially if they are near an outside door. When this happens, the traps should be replaced as the trapped insects can act as a food source for pest species. Moreover, even in pheromone traps, the capture of non-target species is very common, hence, it is essential that the insects captured should be identified, at least up to the group of species level noted above (wood borers, psocids, etc.).

Pheromones

Pheromone traps are one of the most valuable new tools for pest management in museums. Traps useful in museum settings include those for cigarette beetles, drugstore beetles, Indian meal moths, and warehouse beetles (*Trogoderma*). Trapping procedures vary depending on whether the objective is monitoring or control. Pheromone traps are generally effective when pest numbers are very low and they can be qualitatively used to provide an early warning of pest incidence. They are useful in defining areas of pest infestation, particularly in cases when the overall distribution and life cycle are poorly understood. Simple mathematical models are needed to interpret pheromone trap catches and to provide predictions of pest population dynamics and distributions (Hagstrum and Subramanyam 2006).

Multiple pheromones for different species can be employed in single traps where no interspecific influence of the semiochemical attractants has been shown. Pheromone traps are currently available for *T. bisselliella*, *T. pellionella, Trogoderma* spp., *L. serricorne*, *S. paniceum*, *B. germanica*, *A. verbasci* and *Attagenus* spp. The lures are extremely effective, but will only attract the males of the target species and have no effect on other insect species. Some species, such as *S. paniceum*, are only attracted to pheromones when there is a light source and are not effective in the dark.

Various studies have reported success in the mass trapping and attracticide method in the control of *L. serricorne* and *P. interpunctella*. Against *Trogoderma glabrum* attracticide method utilized pheromones in an inoculation device contaminated with some kind of pathogen (*Mattesia* spp.), or in the control of *P. interpunctella* with granulosis virus. In mating disruption, several successful experiments have been reported, such as for *A. unicolor* (*megatoma*), *L. serricorne, P. interpunctella*, *S. paniceum* and *Trogoderma inclusum* (Trematerra 2012). Trial evaluation of the Exosex CL tab™ pheromone disruption system for *T. bisselliella* has shown that populations of moths can be suppressed when it is used as part of an IPM programme (Lauder 2011). A preliminary field study in the USA suggests that release of the synthetic sex pheromone serricornin can significantly inhibit proper orientation of male cigarette beetles *L. serricorne* to females and result in reduced reproduction (Trematerra 2012).

Ultraviolet light traps can be useful for detecting and controlling some flying insects, particularly flies and some moths. These traps must be checked and emptied periodically or the dead insects will themselves attract dermestid beetles and other scavengers. For these traps, there are designs that are very discreet, in order not to be visible from the visitors.

For most pests in the immediate museum area, the action level will be one live specimen. Presence of live adults or larvae indicates ongoing infestations, which should be investigated immediately and treated as a priority. Shed larval skins and feeding damage may have resulted from old infestations, but in regularly monitored and cleaned collections, these should be regarded as an indication of an active infestation.

Control Methods

If a serious insect infestation occurs, or if insect problems do not respond to the preventive techniques, direct treatment for insect infestation may be necessary. This strategy should be used as a last resort.

Chemical Treatments

Pesticides used in museum pest control are generally similar products used for household or other structural pest control (Pinniger and Child 1996). Insecticides should only be used as a targeted treatment and not as a routine. They should not normally be applied directly to objects unless this is approved by a conservator. Museums are potentially good sites in which to use non-conventional pesticides such as repellents and insect growth regulators (IGRs) for controlling cockroaches, cigarette beetles, and certain other stored product pests.

Common chemical treatments used to control insects include the following:

- aerosol sprays;
- baits and pellets (which are eaten by the insects);
- contact and residual sprays (normally sprayed into cracks and crevices, these kill on contact and/or by absorption of the pesticide when the insect walks through the residue);
- cold fogging concentrates (these use equipment that suspends a pesticide and oil formulation in the air);
- fumigants (these expose infested material to a lethal gas);
- residual and vapor pest strips (the insect absorbs pesticide by walking across residual pest strips, while pesticide evaporates from vapor pest strips to become a fumigant);

- dusts (e.g. boric acid or silica dust, which dehydrates insects or interferes with internal water regulation);
- attractants (which lure insects into traps, sometimes killing them);
- repellents are also sometimes used; these are meant to discourage rather than kill insects.

Repellents

Paradichlorobenzene and naphthalene have been commonly used as repellents in museum cases. These materials do not eliminate infestations, but may be useful in preventing them. Paradichlorobenzene and naphthalene may cause damage to certain plastics (bakelite, for example), and may soften and shrink resins, adhesives, and paints. They are now banned in some countries.

Cedar wood chests are often recommended to protect fabrics from clothes moths and carpet beetles. However, only freshly cut cedar wood is toxic or repellent to fabric pests, and then only in an airtight container. By the time the wood is two years old, there is no toxic effect left. Lavender and lavender oil has been shown to have a repellent effect on adult clothes moths but has little effect on larvae (Pinniger pers. com.).

The knowledge of these substances and their potential to repel or attract insects could be utilized in pest management by either using them by masking attractive artefacts or in attractive traps (Shaaya and Kostyukovsky 2006). Some of these compounds have been identified, especially pheromones of relevant pest species like cigarette beetles, drugstore beetles, Indian meal moths and warehouse beetles (*Trogoderma*).

Fumigants

If non-chemical treatment of infested materials is not practical, some materials can be treated with standard insecticides. However, in most situations, infested museum specimens should be fumigated. Fumigation is hazardous and it requires professional training to do it safely and effectively. Fumigation of museum specimens is normally conducted in special fumigation chambers, vaults or ‘bubbles’. Some fumigation is done under tarpaulins. In severe and extensive infestations, an entire building may have to be ‘tented’ and fumigated.

There are a number of different fumigants to choose from. The choice will depend mostly on the objects and materials to be fumigated, since different fumigants are best suited for certain uses. Some fumigants cannot be used on certain materials because they may react with them. The most commonly used fumigants for museum specimens in the past were methyl bromide and ethylene oxide. More recently, sulfuryl fluoride and carbon dioxide have been introduced as replacements. Fumigation treatments do not provide a residual effect that will prevent reinfestation.

Methyl bromide was a very effective fumigant for pests. However, it has been identified as an ozone-depleting chemical and is no longer available in many countries. It has also been used in the past for fumigation of commercial aircraft.

Objects are sometimes fumigated using phosphine. This is done either in a special chamber or under gas-proof sheeting. The temperature must be above 20 °C to avoid over long exposure to the chemical. At high relative humidity, phosphine can corrode metal objects (gold, silver, copper and brass) and it is therefore only suitable for treatment of wood, some textiles or natural history specimens. On painted plasters, this gas induces the highest colour alterations after treatment, especially on gold gildings. It should not be recommended for the disinfestation of heritage premises in the presence of metal artefacts in silver, copper (or alloy containing copper), tin or lead.

Sulfuryl fluoride is one of the most suitable substitute gases for methyl bromide. It is very effective for termite control and wood borers but its cost remains very high as the required active substance has to be at least double the concentration, to achieve the same effect against other pest species as with methyl bromide. Sulfuryl fluoride fumigations have been used to control wood infesting beetles in structures, in museums and churches without damaging materials. However, eggs are less susceptible to sulfuryl fluoride.

Ethylene oxide (ETO) was commonly used in libraries and archives until the 1980s, and many libraries had their own ETO chambers. ETO is effective against insect adults, larvae and eggs. There is evidence that ETO can change the physical and chemical properties of paper, parchment and leather. Because of health issues, it has now been banned in many countries.

In general, fumigants and other pesticides can cause long- and short-term health problems, ranging from nausea and headaches to respiratory problems to cancer. Many chemical treatments will leave residues and may be absorbed into the body to cause health problems years later.

Non-chemical Treatments

Non-chemical management includes cultural controls (temperature and humidity control, sanitation and lighting), pest-proofing (pest-proof containers or display cases, screening and caulking, etc.), trapping (mechanical, sticky, pheromone and light traps) and vacuuming. Treatment of objects can be by freezing or heating, and, in rare cases, 'radiation' such as microwave ovens and gamma irradiators.

Cultural Control

Many heritage buildings have an endemic population of insects living in voids and dead spaces. Poor sanitation encourages pests, food debris, grease, loose hairs and other organic debris in and around specimens, storage areas, and in cracks and crevices in floors and furniture attracts and feeds pests. Good sanitation, particularly regular vacuuming, of display and storage areas removes potential food and newly arrived foraging pests.

Light shields, curtains and closed doors can reduce the numbers of flying insects attracted to the museum. Windows in areas where specimens are kept should be tightly screened or kept closed at all times to prevent pest entry. Caulk or otherwise seal cracks and holes in walls and floors, holes around pipes and other utility lines, and other points of pest entry. Install door sweeps where necessary. Air vents and hot air registers can be equipped with filters to trap potential incoming pests. Filters should be changed on a regular basis.

Lowered humidity and temperatures reduce the chance of infestation and slow down the growth of existing pest populations. For some pests, such as psocids and silverfish, reducing humidity can eliminate a pest problem.

The most effective way to prevent damage from dermestid beetles, clothes moths, and many other museum pests is to prevent the establishment of infestations in the first place. All incoming specimens should be examined carefully for damage and live insects and any showing signs of infestation should be isolated and disinfested. All actions should be recorded.

Adult dermestid beetles and other museum pests feed on pollen and nectar, so decorative cut flowers should be kept out of specimen areas to reduce the chance of accidental infestation. Those specimens at high risk of insect damage should be kept in insect-proof cases and examined on a regular basis.

Low Temperature

Strang (1992) reports on the efficacy of cold treatments against cultural heritage pests and with correct procedures, this method will kill all stages of an insect's life cycle. Low-temperature treatments are used routinely by many museums and are sometimes used for large-scale programmes of disinfestation, particularly when moving collections from one building to another (Berkouwer 1994). Low-temperature treatments are generally used for new objects and specimens coming into the museum to prevent pests being introduced on incoming collections.

Many types of infested museum collections can be disinfested by freezing them in a chest freezer or a large commercial freezer (Bergh et al. 2006). Textiles, furniture, herbarium specimens, books, mammal and bird collections, as well as various ethnographic materials, have been successfully frozen for insect control. Low-temperature treatment schedules should be clearly documented and recorded. The objects must be sealed in polythene (other plastic films such as polyester can be used) and exposed to temperatures of −30 °C for three days or −18 °C for at least 14 days. Objects should not be removed from the bag until they have returned to room temperature and there is no risk of condensation. Freezing is most often conducted at temperatures that are lower, e.g. −35 °C, or at more moderate temperatures but for very long time, e.g. more than 2 weeks. Materials can be treated in household or commercial freezers, blast freezers, or controlled-temperature and humidity freezers.

Freezing provides no residual protection from attack. If collections are not returned to a well-maintained storage area, reinfestation will almost certainly occur. Very fragile objects, those made from a combination of materials, and artefacts with friable media should probably not be frozen. Note that freezing poses a significant risk of damage to certain wood veneers, bone, lacquers, some painted surfaces and leather. Generally, low temperature is not advisable for canvas and wood-panel paintings, painted or inlaid wooden objects, finished furniture, lacquered wooden objects, objects under tension (e.g. drums, strung parchments), composite objects containing ivory or teeth and inorganic materials, such as glass, high-fired ceramics and metal.

Low, but above-freezing temperatures, usually 5 °C, will stop insects feeding and reproducing and can be used to protect items in storage. The best example is low-temperature storage of valuable furs, skins and costumes.

Elevated Temperature

Heat can effectively exterminate insects and it has been used widely in food processing (Xavier-Rowe et al. 2000; Hagstrum and Subramanyam 2006). The risk of damage to objects is greatest at higher temperatures, but studies have shown that heating can be successful at more moderate temperatures (around 50–52 °C) (Ackery et al. 2004). With any heat treatment of vulnerable collections, it is essential to maintain a constant humidity so that the objects do not dry out and shrink or crack. Safe treatments can be carried out in a controlled humidity heat chamber such as Thermo Lignum (Nicholson and von Rottberg 1996). Objects are placed in the chamber and the temperature is gradually raised to 52 °C and then lowered back down to ambient over a cycle of 18–20 h. The control system keeps the RH at a set level of, for example, 50% for the whole cycle. Some less sensitive objects can be treated in an oven at 52 °C if they are bagged to keep the RH around the object stable. Large objects can be treated in a simple hot box (Xavier-Rowe et al. 2000) or by using solar heating (Daniel 2001; Brokerhof 2002). Studies must be conducted to determine whether repeated treatment with low or high temperatures make the artefacts more attractive to insects and thus more vulnerable to infestation (Strang 2001).

Modified Atmospheres

Modified atmospheres have been used widely in the agricultural and food industries to control insect infestation and are now being used by some museums. The term refers to several processes: increased carbon dioxide, decreased oxygen (anoxia), by the use of inert gases (primarily nitrogen) and use of oxygen scavengers to decrease oxygen levels. Both nitrogen anoxia and carbon dioxide fumigation can be very effective in killing insects in objects. The technique is particularly useful for fragile and very vulnerable objects, which might be damaged by low- or high-temperature treatments (Selwitz and Maekawa 1998; Pinniger 2001).

Modified atmospheres can be applied (1) in a traditional fumigation chamber or a portable fumigation bubble or (2) in low-permeability plastic bags. With a chamber or a bubble, materials are prepared for treatment, air is evacuated from the chamber and carbon dioxide (generally about 60% concentration) or nitrogen (to achieve an atmosphere of less than 0.1% oxygen) is introduced. Long exposures of three weeks or more may be needed to kill all pests. Once treatment is finished, the vacuum is released, the carbon dioxide or nitrogen is removed and the chamber is aerated. The process for treating materials in low-permeability plastic bags is similar, except that materials are sealed in bags with an oxygen scavenger that will reduce the oxygen level in the enclosure to less than what is needed for insect respiration. In some cases, the bags are purged with nitrogen before sealing.

Different species of insects, as well as different stages, differ in their tolerance to low oxygen atmospheres. As with carbon dioxide, exposure times need to be longer at lower temperatures and treatments may not be effective below 20 °C. The larvae of some wood boring beetles are particularly tolerant of low oxygen and may survive for many weeks.

Use of controlled atmospheres against pests in museums has received an increasing amount of interest during the last twenty years (Maekawa and Elert 2003; Pinniger 2011b). At moment, the recommended protocol validated for *Anthrenus vorax, L. serricorne*, *S. paniceum* and *T. bisselliella* suggests an oxygen percentage below 0.1% for at least three weeks at above 20 °C. The most common problems are the long treatment time that, together with difficulty in achieving and maintaining such low oxygen concentrations.

Modified atmospheres show great promise, but additional research is needed to determine optimum exposure times and methods for particular types of insects (Rust et al. 1996; Selwitz and Maekawa 1998; Binker 2001; Warren 2001). The use of carbon dioxide as fumigant gas is very effective and is used by a number of museums in North America (Selwitz and Maekawa 1998; Warren 2001).

A Museum example

The Kunsthistorisches Museum in Wien is one of the largest museums in Europe with numerous exhibitions and storage rooms housed mainly in historic buildings and with a large variety of object types. Formerly, all kinds of chemicals were applied in the collections against insect pests or fungi, for example, DDT, naphthalene, methyl bromide, lindane (up to 1982), pyrethroids (until 1998) or ethylene oxide. Eulan was sprayed or objects submerged with until 1990, thymol applied to remove mould or xylamon was used to combat wood destroying insects. In addition, natural crystalline camphor, patschuli, lavander flowers, essential oils like clover in alcohol or lemongrass were used to prevent infestation. From 1996, fumigations with nitrogen were tested in the Picture Gallery collection (Ranacher 1998) and the construction of the walk-in nitrogen chamber initiated for the whole museum. In 1998, this 32 m^3 chamber was built and since then, all infested objects from the museum, but also from other museums, institutions and private collections are successfully treated in 5-week cycle.

Future Developments

Because of the effects of some chemicals on staff, objects and the environment, there has been pressure to move away from persistent and toxic insecticides. Some alternatives could be found in the use of semiochemicals, radiation, essential oils, biological control.

Semiochemicals. The development of semiochemical-based systems for population suppression is exciting, but success will depend upon the understanding of pest behaviour and the availability of economically priced lures. In the management of museums, insect pheromones can be used to monitor and to suppress and control the pest populations by means of mass trapping, attracticides and mating disruption methods, as well as acting as repellents and as specific behavioural stimulants or deterrents (Phillips and Throne 2010; Trematerra 2012). During recent years, computer-assisted decision support systems have also been developed that estimate insect population growth and the spatial distribution of insects as a function of environmental factors (Brenner et al. 1998; Trematerra and Sciarretta 2004; Baslé et al. 2011).

Radiation. Microwaves are used successfully in the food, agricultural and textile industries to control insects. Their effectiveness depends on the type of insect and the intensity and frequency of the radiation. The average infested book is microwaved for 20–30 s. According to Chmielewska et al. (2011), this method is safe for most hardback books printed after 1950 and high-quality soft-cover books with sewn bindings. Method on valuable old editions, older books with metallic dyes, inexpensive soft covers (it will melt the glue) or books bound in leather. X-rays, gamma rays and electron beams could be applied for disinfestation of cultural heritage objects. A dose as low as 0.3 kGy of gamma radiation completely inhibits the development of immature stages and sterilizes adults of *L. serricorne* and *S. paniceum* (Chmielewska et al. 2011). When it is needed, the effectiveness of gamma irradiation can be increased by application of additional treatment that would predispose insects to become easily damaged: high temperature, chemical treatment and infrared or microwave radiation could be additional applied.

Essential oils. Certain plant essential oils and their active constituents, mainly terpenoids, have potentially high bioactivity against a range of insect and mites. They are also highly selective to insects, since they are probably targeted to the insect receptor, a non-mammalian target (Shaaya and Kostyukovsky 2006). The ultimate goal is the introduction of these phytochemicals with low toxicity as alternatives to methyl bromide and phosphine fumigations. The use of bioactive compounds, for example, essential oils with CO_2, or isothiocyanates, especially methylthio-butyl ITC can be used as fumigants.

Biological control. While little information is available on natural enemies of the more specific cultural heritage pests, and almost none on biological control, a lot of information is available on natural enemies and biological control of stored product pests. There are parasitoid species that are associated with human-based habitats and their stored product insect hosts and also parasitoid species that accept stored product insects as hosts, but were transferred from agricultural ecosystems to indoor habitats. Natural enemies are known from many cultural heritage pests, but evaluation of their potential for biological control has been limited to, for example, biological control of *T. bisselliella* with *Trichogramma* spp.; *T. bisselliella* and *T. pellionella* with *Apanteles carpatus*; *S. paniceum* and *L. serricorne* with *Lariophagus distinguendus*; *A. punctatum* with *Spathius exarator*. Use of parasitoid wasps *L. distinguendus* and *Trichogramma evanescens* as part of an IPM concept in museums against *S. paniceum* and *T. bisselliella* in Austria and in Germany is reported by Schöller and Prozell (2011).

Conclusions

There are new developments that are now becoming available for the detection, prevention and control of museum pests.

Application of pesticides to control insect pests in museums has been shown to be accompanied by unwanted side effects on humans as well as on the items themselves. Regulations are continually being revised to restrict or ban the use of many chemicals. Certain alternative strategies (e.g. freezing and anoxia) have therefore been developed and used for a number of years (Bergh et al. 2003, 2006; Child and Pinniger 2008). Low-temperature treatments are now used for quarantine prevention and infestation control in many museums worldwide. The use of high temperatures is less common at the moment, but offers a rapid and safe alternative for many objects. The ability to use solar heating is of particular interest and value for developing countries with limited access to expensive equipment and technology. The further development and adoption of treatment regimes based on anoxia and low and high temperatures should ensure that historic collections will be safely preserved for the future.

An important part of using various methods for control of museum pests is an analysis of potential unwanted side effects. The exact effects of each treatment method need to be known to allow an informed decision balancing the effect of the treatment against the continued attack by insect pests (Kigawa and Strang 2011). Modern conservation ethics determine that where possible, any effect on artefacts must be minimal or reversible (Caple 2000). Methods in use must also be safe for persons using them, for persons handling the artefacts after treatment and for museum visitors (Carter and Walker 1999).

Changes in the distribution of a number of animal species in the northern hemisphere have been documented during the last decades. This includes insect pests in agriculture and forestry general and it has implications for urban and museum pests as well. There is evidence of the spreading of museum pests to new sites and the likelihood of this increase in the future must be considered (Hansen et al. 2011). Evaluation of the future risks and the distribution of the insect pests will also have to include global climate change predictions.

Pests of Museum—Cultural Heritage Pests

Wood pests		Distribution	
		North America	Europe
Coleoptera			
Anobium punctatum	Furniture beetle/woodworm	+	+
Callidium violaceum	Longhorned beetle		+
Dinoderus minutus	Bamboo powderpost beetle	+	
Hylotrupes bajulus	Old house borer/house longhorn	+	+
Lyctus brunneus	Powderpost beetle	+	+
Lyctus linearis	Powderpost beetle	+	+
Nicobium castaneum	Library beetle		+
Pentarthrum huttoni	Wood boring weevil	+	+
Xestobium rufovillosum	Death watch beetle	+	+
Isoptera			
Cryptotermes brevis	Drywood termite	+	
Kalotermes flavicollis	Yellow-necked drywood termite	+	
Reticulitermes lucifugus	Subterranean termite/destructive European termite	+	+

General pests		Distribution	
		North America	Europe
Dictyoptera			
Blattella germanica	German cockroach	+	+
Blatta orientalis	Oriental cockroach	+	+
Periplaneta americana	American cockroach	+	+

(continued)

(continued)

General pests		Distribution	
		North America	Europe
Coleoptera			
Lasioderma serricorne	Cigarette beetle	+	+
Niptus hololeucus	Golden spider beetle	+	+
Ptinus tectus	Australian spider beetle	+	+
Stegobium paniceum	Drugstore beetle/biscuit beetle	+	+
Lepidoptera			
Hofmannophila pseudospretella	Brown house moth	+	+
Endrosis sarcitrella	White-shouldered house moth	+	+
Thysanura			
Lepisma saccharina	Silverfish	+	+
Thermobia domestica	Firebrat	+	+
Psocoptera			
Liposcelis bostrychophila	Common booklouse	+	+
Trogium pulsatorium	Deathwatch booklouse	+	+

Pests of textiles and natural history		Distribution	
		North America	Europe
Coleoptera			
Anthrenus flavipes	Furniture carpet beetle	+	+
Anthrenus verbasci	Varied carpet beetle	+	+
Anthrenus museorum	Museum beetle		+
Anthrenus sarnicus	Guernsey carpet beetle		+
Attagenus pellio	Two-spotted carpet beetle		+
Attagenus smirnovi	Brown carpet beetle/vodka beetle		+
Attagenus unicolor	Black carpet beetle	+	+
Dermestes lardarius	Larder beetle		+
Dermestes peruvianus	Peruvian hide beetle	+	+
Dermestes maculatus	Leather beetle	+	+
Reesa vespulae	Museum nuisance	+	+
Thylodrias contractus	Odd beetle	+	
Trogoderma angustum	Berlin beetle		+
Lepidoptera			
Tineola bisselliella	Webbing clothes moth	+	+
Tineola pellionella	Case-making clothes moth	+	+
Trichophaga tapetzella	Carpet moth/tapestry moth	+	+

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Chapter 12
Economic Theory Versus Reality in Stored Grain IPM: Theory and Practice in Stored Product Management

Phil Kenkel and Brian D. Adam

Introduction

It is humbling for both economists and entomologists that their recommendations are not always followed. Individual decisions often seem to be at odds with society's best interests, at least from the perspective of the scientist making the recommendation. Frequently, those decisions seem to be at odds with the individual's best interest. Mumford and Norton (1984) address this "disconnect" between the recommendations of scientists and what decision makers actually do:

> Entomologists should also be aware that actual pest management decisions are based on normative considerations and therefore are subjective, no matter how carefully the costs and benefits have been assessed. Consequently, the economics of decision making in pest management is not just concerned with the dollars and cents of pest damage and control but with the goals and behavior of those who make pest management decisions (p. 157).

Of course, one could just as easily substitute the word "economists" for "entomologists" in that paragraph.

This chapter attempts to explain and illustrate some of the reasons why recommendations by entomologists and economists for stored product management seem correct in theory, but are not followed in practice. The reasons fit into four broad categories. First, the decision makers may simply not understand the recommendation or the potential benefits to their business of adopting the recommendations, or they may be subject to human inertia; for example, "stuck in their ways." This category is what many researchers typically assume when their

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recommendations are not followed, believing that their educational/outreach efforts are inadequate, and that they need to do a better job of explaining their recommendations.

Second, the scientist may not understand the nuances of the commercial storage system or the risks and constraints faced by the decision maker. This can be due both to lack of communication among scientists across disciplines and a failure of the scientists to understand the commercial storage environment. For example, an economist may not understand that fumigation has different efficacy on different life stages of insects, or an entomologist may not recognize that capital and labor have opportunity costs if diverted from other uses to managing an Integrated Pest Management (IPM) program. Fortunately, most of these misunderstandings or oversights can be resolved in effective multidisciplinary cooperation.

A third possibility is that we are incorrect in our assessment of the degree of IPM adoption. IPM is a multifaceted strategy involving scouting/monitoring as well as chemical-based and nonchemical-based control strategies. It is often assumed that IPM adoption will reduce pesticide use. This could be the case if IPM provides alternative nonchemical control options, or eliminates redundant applications. However, it is also possible that the decision maker was underestimating pest damage, in which case IPM adoption and increased sampling could lead to more chemical use. Measuring the degree of adoption of a multifaceted strategy is complex. It is possible that many of the components of stored product IPM have been adopted without resulting in reduced chemical applications.

In this chapter, we present an economic approach, with several variations, for decision making in stored grain. Through this discussion, we examine possible explanations for the perceived lack of adoption of IPM in stored product systems.

Description of a Stored Grain IPM System

In order to discuss the degree of IPM adoption in the context of stored products, and possible barriers to adoption, it is useful to first describe a conceptual IPM-based decision framework. While various definitions for IPM have been proposed, they share similar themes. IPM is typically described as a systematic approach, involving multiple strategies to prevent or reduce pest damage by the most economical means. IPM is also generally considered to include regular monitoring to determine if and when controls, including pesticide treatments, are justified. The IPM approach is often described in terms of the economic threshold model that was introduced by Stern et al. (1959), who defined the economic threshold as the density of a pest population that will justify treatment. Mumford and Norton (1984) expanded the definition to describe the economic threshold as the density of a pest population where the benefit of treatment just exceeds its cost.

To illustrate the economic threshold model in a stored product context, Figs. 12.1 and 12.2 show the tradeoff between treatment cost and cost resulting from insect infestation. In Fig. 12.1, insect population grows exponentially (with the rate of growth depending on many factors, but especially temperature and humidity). As the insect population grows, cost of insect damage grows (e.g. in stored wheat, if the insect is *Rhyzopertha dominica*, or lesser grain borer, discounts due to both insect-damaged kernels (IDK) and live insect infestation result). If a one-time treatment (e.g., fumigation) occurs that is effective, insect population is reduced to nearly zero, and the insect population resumes its exponential growth, from the new, much lower, population base. As the insect population grows in this new cycle, as before, the cost of insect damage grows as well. Throughout the storage period, this cycle could repeat several times, depending on temperature, humidity, and other factors, requiring several treatments.

Figure 12.2 shows that the optimal time for treatment is when cost of treatment is equal to the predicted cost of not treating.[1] At the left edge of Fig. 12.2, the cost of treatment with zero treatments is zero, but the cost of insect damage is high. Moving to the right of the figure, increasing the number of treatments raises the treatment costs while reducing the cost of insect damage. In between, where the two curves intersect, cost of treatment and cost of insect damage are equal at the optimal number of treatments.

Adam et al. (2010a, b) provide a more detailed description of how the economic threshold model could be applied to stored grain and it is useful to summarize that approach. Specifically, the model estimates the costs and benefits of alternative strategies for controlling lesser grain borer *R. dominica*. A calendar-based fumigation timing strategy was compared with a sampling-based fumigation strategy, in a simulation over 20 years of historical weather. In the calendar-based strategy, the manager fumigated every year on the same date, and in the sampling-based strategy, the manager fumigated when the number of insects in a sample exceeded a predetermined threshold.

The economic threshold was based on three components. The first component was to estimate cost of each component of insect control in grain storage using an economic engineering approach. The second component was to predict the insect population that would result under various environmental conditions and under alternative insect control strategies. The insect growth model described by Flinn et al. (2007) was used to simulate insect growth under the historical weather years and the two strategies, as well as under alternative assumptions about fumigation effectiveness and insect immigration rate into the grain storage facilities. This deterministic model predicts daily populations of *R. dominica* in the larval, pupal, and adult stages as a function of the previous day's population, insect immigration rate, mortality rate due to fumigation and natural death, and grain temperature. Grain temperature is the most important variable for predicting insect population at various locations within a bin. The *R. dominica* model is coupled with a two-dimensional grain storage model that uses a thermal transfer equation to predict changes in grain temperature as a function of hourly observations of solar radiation,

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[1] This characterization abstracts from risk considerations. Taking risk into account may change the optimal timing of control. See, for example, Mbah et al. (2010).

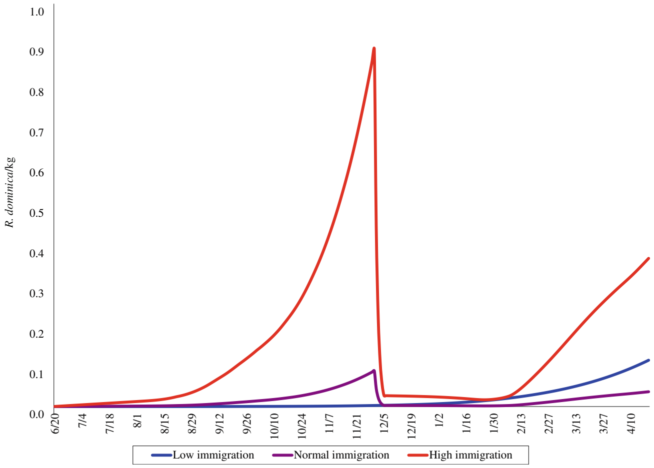

Fig. 12.1 Growth of *R. dominica* in stored wheat with one fumigation (data from software model described in Flinn et al. 2010)

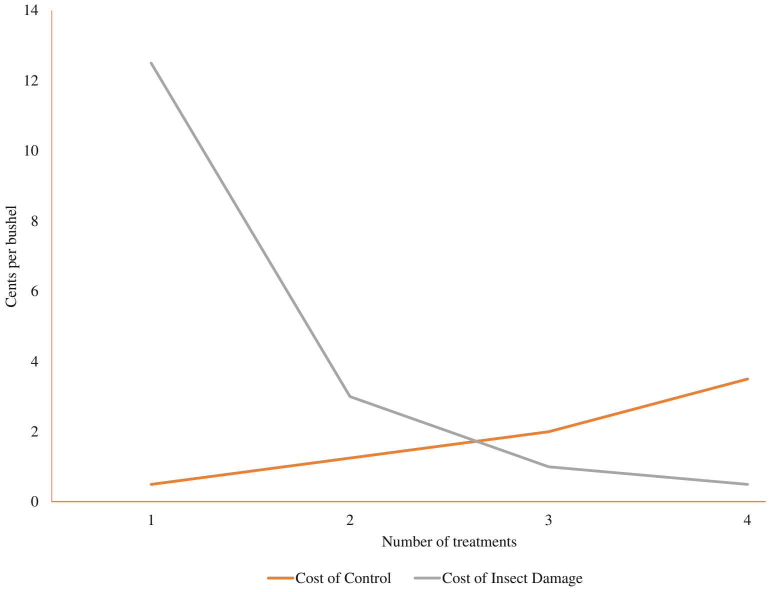

Fig. 12.2 Economic tradeoff between cost of insect damage and cost of insect control

cloud opacity, dew point temperature, dry bulb temperature, relative humidity, barometric pressure, and wind speed.

The final component was to use the predicted insect numbers to predict economic damage. The elevator manager was assumed to choose the insect control strategy that minimized expected total cost of both insect control and insect damage.

$$\min{}_j E\left(C_j\right) = \left[TC_j + E\left(D_j\right) + E(L_j)\right] \tag{12.1}$$

where $E(C_j)$ is the expected cost of insect control strategy j, TC_j is the treatment cost associated with the jth insect control strategy; D_j is the discount due to damaged grain, and L_j is the discount due to live insects at time of marketing. In this way, the model explicitly considers the tradeoff between treatment cost and damage cost, including opportunity costs.

While the work by Adam et al. (2010a, b) is an attempt to rigorously apply IPM's economic threshold model to stored grain systems, it abstracts from many of the informational constraints facing commercial grain managers. The described decision framework revolved around predicted insect populations which were generated from an insect growth model. In a commercial setting, a manager must estimate insect populations from sampling data. Sampling is difficult and provides imprecise estimates of the current pest population. Insects are not distributed evenly in the structure, making it difficult to interpret sample results. Conceivably, the insects could all be in the top of the bin, the bottom of the bin, or in some pocket of high moisture grain in the center of the bin. That creates great uncertainty in predicting the true insect population from the sample results.

The second component of the stored grain IPM model was the economic consequences from insect presence, which were represented as a simple market-determined price discount. In reality, the consequence of insects depends upon both on the grain buyer sampling procedures (which could fail to detect insects or overestimate the true population) and the grain buyer's standards and policies. In many cases, grain buyers reject a load when a single insect (live or dead) is detected. The resulting economic consequence depends upon alternative markets, transportation costs, and fumigation costs required to successfully remarket the grain. If the grain was intended for a high-value market, for example a flour mill specifying high protein wheat, the differential relative to the next best market may be quite significant. Grain managers typically market to a wide variety of outlets making it impossible to predict the economic consequence of insect presence at that point in time that treatment decisions are considered. The "discount" for insect presence may range from pennies to dollars per bushel and is unknown at the time storage decisions are made. There is also a reputational effect that occurs when a preferred grain buyer detects pest or pest damage in one load of grain and elects not to accept future loads from the supplier. In addition to these constraints which make it difficult to implement an economic threshold model, there are other general factors which make it difficult to implement IPM in commercial grain storage systems.

Other Constraints to IPM Adoption in Commercial Grain Storage Systems

Undefined Storage Period

When grain is delivered to a commercial elevator, the storage period is undefined. In fact, it is not even clear whether the grain will be stored. The producer delivering the grain may decide to sell some of all of the grains immediately or place it in open storage (storage for an undetermined period of time). At any point in time, the producer can decide to sell the grain. At that point in time, the grain merchandiser can decide to place a storage hedge and continue to store the grain as company-owned inventory, or sell the grain down the marketing channel. Grain held under a storage hedge also has an undetermined storage period. The grain merchandiser may subsequently roll over the storage hedges, increasing the storage time or reverse the hedge when market conditions are favorable. The elevator superintendent (the individual typically in charge of stored grain management) may not know if the grain being placed in a particular bin is for sale in a short time or for storage, or the length of the storage period. This uncertainty in storage time makes it difficult to apply IPM because the results of Adam et al. (2010a, b) indicate that duration of grain storage may be a key factor affecting the relative costs of sampling-based and calendar-based fumigation. Sampling-based fumigation is more likely to be profitable for shorter storage periods.

Prevention Is Challenging

A standard component of IPM is to manage the system to prevent pests from becoming a threat. In field crop situations, this might include crop rotation or pest resistant varieties. Sanitation and sealing are typically described as a stored grain IPM strategy to prevent pest populations (e.g., Hodgson and Holscher). In a food processing environment, pests can often be controlled through exclusion and by sanitation practices that deny the pests the food and water needed to survive. A commercial grain storage environment is more challenging. The commodity is stored in an unpackaged form, at ambient temperatures with constant flows into and out of the facility. Even the cleanest grain elevator is a far cry from the sanitary conditions of a processing facility. Similarly, while seams and cracks in grain facilities can and should be sealed, even the best-sealed grain elevator is accessible to insects.

Sanitation and sealing have important roles in stored grain management. Ignoring basic measures such as removing old grain before filling a storage structure or cleaning up spilled grain around the bins clearly escalates pest losses. However, even the most intensive sanitation program is unlikely to eliminate pest infestation. Intensive sanitation often requires entry into bins and aeration ducts and access into dead areas of conveying equipment. The point of diminishing returns to sanitation, particularly in light of increasing concern over worker entry in confined space, is an important topic for further research.

Insect Treatment Costs Are a Small Portion of Department Budget

Grain superintendents can monitor pest treatment costs, which is another key element of IPM. Superintendents are also concerned with electricity, maintenance, and labor costs as well as controlling shrinkage (loss of weight) within accepted levels. Kenkel (2010) published a grain storage cost calculator that provides baseline storage and handling cost for various grains. Monthly variable cost for storing wheat was estimated at $13/bu. with total costs at $26/bu (bu. is the abbreviation for bushel, which is the storage unit used in the US. A bushel of wheat in the 60 lbs or 27.2 kg). Fumigation costs were estimated at $0.005/bu., which represented 2% of total costs and 4% of variable costs. A grain superintendent would have to achieve significant reductions in insect treatment costs before it would create noticeable impacts on the department expense report. It should also be noted that even if the elevator tracks treatment costs on a per bushel basis, few grain accounting systems have the sophistication to relate cost to days of storage. Fumigation costs per bushel are low in years when grain moves quickly after harvest and high in years when more grain is stored for longer periods of time.

Multiple Responsibilities

The previous discussion has alluded to the multiple responsibilities of the grain superintendent but this point is worth expanding on. Most elevator superintendent job descriptions describe activities in grain operations, safety and compliance, and facility maintenance. Grain operations include procurement and customer relations, grain receiving, conditioning, blending, logistics, and quality management. In many operations, the grain superintendent will be expected to assist other departments (e.g., the fertilizer department) during seasonal peak periods. The grain elevator superintendent is responsible for the elevator department budget with personnel, electricity, and maintenance representing the largest categories. Multiple responsibilities do not necessarily diminish the focus on stored grain management but they do put it in context of the overall objectives. Every grain superintendent would like to decrease pest-related costs. They would also like to decrease maintenance and repair costs, improve worker safety, and increase throughput. All of these management areas compete for the superintendent's limited time and attention.

Pressure to Reduce Personnel

The ratio of personnel expense to gross margin has long been recognized as a key performance metric for grain elevator firms. Personnel costs are typically the largest single expense line item for the firm. Managers continually strive to reduce their per bushel labor costs by increasing the scale of storage and increased automation. In addition to the financial pressure, it is becoming increasing difficult to recruit, hire, and retain high-quality personnel in the small rural communities where grain facilities are located.

Worker Safety Issues Dominate

Worker safety issues are also a major concern for grain firms. In August of 2010, the U.S. Department of Labor's Occupational Safety and Health Administration (OSHA) sent a letter to 3300 grain elevator operators advising them of their responsibilities for grain elevator worker safety and of penalty citations of almost $4 M that had been issued to three grain firms in the preceding months. OSHA also added the grain handling industry to its so-called Severe Violator Enforcement Program, which subjected facilities to enhanced inspection and enforcement with possible criminal penalties. In 2009, OSHA issued a letter of interpretation on the use of sweep augers that led many in the industry to conclude that it was difficult if not impossible to completely empty the old grain out of most bins while remaining in compliance with the standard (Feed and Grain). In response to this regulatory environment, grain firms have been rapidly revising procedures to reduce and eliminate bin entry. Stored grain IPM with its emphasis on monitoring, detection, and pest identification, is in many ways at odds with the trends of decreased personnel and avoidance of entry into storage structures and confined spaces.

Are We Underestimating the Adoption of Stored Product IPM?

As mentioned in the introduction, another very relevant question is whether the level of adoption of IPM in grain storage systems has been underestimated. In other systems, information on pesticide usage and IPM adoption is somewhat plentiful. For example, USDA-NASS publishes annual reports on agricultural chemical usage in field crops, fruits, and vegetables. A 2001 review by the Environmental Protection Agency found that some level of IPM had been adopted on about 75% of US crop acres. Numerous studies have examined IPM adoption in specific cropping systems. For example, Farrar, Baur, and Elliot (2015) examined IPM adoption in the Western states of the USA. They examined pesticide use and IPM practices on various field crops, fruits, vegetables, nuts, and ornamentals in Arizona, California, Oregon, Hawaii, Utah, and Washington. While the results varied across crops and location, they also suggested relatively high levels of IPM adoption. For example, in 1995, California growers applied 8.6 lbs of pesticide active ingredient per $1000 of gross crop value, a figure which fell to 3.8 lbs in 2012. The authors concluded that the simultaneous trends of increasing crop yields and reduced pesticide use was "achieved in part by widespread adoption of IPM in California." Interestingly enough, the same study reported that the use of fumigants in California increased from 39 to 45 million lbs (a 15% increase) over the same 1995–2012 timeframe.

Unfortunately, information on both pesticide use and IPM adoption in stored grain systems is quite limited. Impressions as to the degree of IPM adoption are often based on indirect evidence such as the California fumigant data or limited survey data. The consensus opinion is that the degree of IPM adoption in stored grain systems has been much lower relative to the adoption in cropping systems. This impression is likely based on the limited information on fumigant use which indicates flat or increasing trends.

However, a number of studies, including Maupin and Norton (2010), have indicated that IPM adoption may lead to continued or even increased pesticide use. While this outcome initially seems counterintuitive, it is quite reasonable. A core component of IPM is scouting and sampling to gain a better understanding of pest presence and damage. That information can cause the decision makers to conclude that they were either overestimating or underestimating the consequences of pest populations. Expectation that IPM will decrease pesticide use is based on the assumption that decisions makers overestimate pest presence and damage. It is easy to see how, in some settings, the decision maker may have been underestimating pest populations with the result that the increased information from IPM can lead to additional treatments. While it would be hoped that IPM adoption increases profitability, the impact on pesticide use is ambiguous. Observations of continued pesticide use do not necessarily indicate that IPM has not been adopted.

This is consistent with the previously described simulation results in Adam et al. (2010a, b). That study modeled wheat storage in the Southern Great Plains under fairly typical storage environments. Based on the prevailing weather patterns, the model predicted substantial increases in insect populations over the summer storage period which required fumigation at least once per year, every year. In this storage environment, a sampling-based IPM approach would lead to the same outcome as a calendar-based fumigation program. In that situation, the degree of IPM adoption obviously could not be inferred from data on fumigant usage. Of course, the lack of an economic benefit to IPM adoption would also serve as deterrent to adoption. In other storage environments, where there are lower rates of insect population growth and/or in which grain is stored for a shorter length of time, an IPM sampling program could potentially be economical because it would identify instances where fumigations could be avoided.

Many of the components of stored grain IPM including monitoring grain quality, sanitation, and aeration, are part of standard operating procedures at commercial elevators. In most cases, the procedures are not specially targeted at reducing pest damage or reducing fumigation. They are, instead, components of a more general goal of maintaining grain quality. Conclusions as to whether the glass of stored grain IPM adoption is half full or half empty are somewhat subjective. Future research should include surveys of food processing firm managers to determine the range of insect control measures they are actually using, including measures that could be considered IPM. Better understanding of managers' actual practices would help researchers make more precise recommendations that are consistent with firms' objectives and constraints.

Addressing the Gaps

There are two basic strategies for bridging these gaps between stored grain IPM theory and commercial grain storage practices. The first approach is to reduce the uncertainties and imperfections in the systems. Sampling and detection procedures could be improved (and hopefully automated) to the point that managers have accurate real-time measures of pest populations throughout all areas of storage facilities. Insect population projection models could be similarly improved to provide accurate estimates of populations and damage for various storage periods and specific commercial storage environments. Sampling procedures could be improved to the point that storage damage is precisely measured. Grain accounting software could be modified to decompose grain sales, quality measures, and discount schedules to accurately measure the economic consequences of pest damage. After all, of those advances had been achieved, the cost effectiveness of specific practices could be analyzed. For example, it might be conclusively shown that sweeping empty bins were cost effective while the costs of vacuuming aeration plenums exceeded the benefits.

An alternative strategy is to better integrate the principles of stored grain IPM into the overall objective of managing the grain elevator. IPM information could be integrated with worker safety and preventive maintenance information. This could help to create a set of best management practices for a grain handling operation that could be readily extended. For example, in the context of preventative maintenance for machines, monitoring refers to bearing temperature, belt alignment, belt speed, and plug detection, in the context of IPM, monitoring focuses on grain temperature, moisture problems, and insect numbers, while for OSHA regulations, monitoring focuses on grain dust accumulation. From the viewpoint of the elevator superintendent, all of these areas need to be incorporated into the monitoring section of standard operating procedures.

Integrating stored grain management into grain facility management would also help to identify logical synergies. For example, preharvest preventative maintenance procedures that include checking gearbox oil levels, and testing bearing temperature and belt alignment monitors could be integrated with cleaning conveying equipment and checking grain temperature cables, which are IPM-related strategies.

A more commercial view of stored grain IPM might indicate that some components must be modified or abandoned. Grain elevator managers strive for systems which eliminate any entry into bins and confined spaces. As they make facility upgrades, they are opting for "zero entry" sweep augers and self-unloading floors. Grain elevator managers are clearly willing to make substantial investments to eliminate bin entry. It may be time to declare any IPM procedure that requires bin or confined space entry as impractical.

Labor requirements and costs could also be more explicitly integrated into stored grain IPM recommendations. Stored grain IPM sanitation strategies treat every area as a reservoir for pest populations. In the context of the elevator operation, there are drastic differences in the cost and practicality of cleaning different areas. Sanitation procedures that require the removal of access plates, the disassembly of equipment, or the entry into bins and confined spaces obviously have a much higher cost. More research is needed to determine the payoff for specific sanitation procedures and whether electing not to clean some of the more difficult areas negates the benefits of the overall sanitation program.

Similarly, recommendations on pest monitoring include a wide range of absolute sampling methods (diverter samplers, Ellis cups, grain trier, vacuum probe, and pelican) and relative sampling measures (sticky traps, corrugated harborage traps, pitfall cone traps, and probe traps). The rationale is that monitoring will alert a manager to treat a bin which they might not have planned on treating and eliminate treatments in bins where it is not justified. However, the various monitoring procedures vary drastically in cost and accuracy. Some of the most practical methods, such as probe traps, do not provide clear thresholds for treatment. IPM strategies attempt to weigh tradeoffs between pest control costs and pest damage. There is currently very limited information on the costs and benefits of specific subcomponents of the stored grain IPM system.

Implementation of stored grain IPM could also benefit from improved facility design. As elevator managers reinvest in infrastructure to generate speed and space, more information should be available on IPM compatibility. In food processing environments, sanitation requirements are clearly defined, and equipment with "clean in place" (CIP) capabilities has been widely adopted. There has been much less work in automating sanitation procedures in the grain elevator environment. Furthermore, there is little information comparing the degree of cleanout, ease of cleaning, or effectiveness of sealing of alternative systems. In the food processing industry, there are numerous reports and fact sheets describing how to evaluate CIP equipment without endorsing any particular equipment brand. Similar guides could be developed for the grain industry if automated sanitation systems were developed.

It would also appear logical to integrate sampling capabilities into facility design. The major challenge of pest sampling in the stored grain environment is the difficulty in accessing the entire grain mass. Technologies such as powered vacuum probes are one solution, but they require significant labor and the equipment must be transported from bin to bin. It would seem logical to integrate sampling systems into storage structure design so that samples could be automatically drawn from various locations within the bin.

As previously discussed, it is difficult to develop a clear economic threshold for stored grain pest treatment. Part of the challenge relates to the US grain marketing system. Grain quality assessment in the USA is primarily based on visual observation and physical measurement. In an overall sense, the US grain marketing system is extremely efficient in communicating to buyers and sellers the properties of the commodity being marketed (Kenkel and Adam 2012). It also channels grain with particular quality levels to buyers willing to pay for those traits. Part of the cost of pest damage is the opportunity cost of the sale to a higher priced but more quality-conscious buyer. For this reason, it is inherently difficult to accurately project the impact of varying degrees of pest damage on each lot of grain stored.

In light of these challenges, it might be more appropriate to promote stored grain IPM in the context of supply chain management. Food industry firms have been active participants in quality control systems such as the ISO 9000 family of standards and in pursuing preferred supplier and strategic supplier relationships. All of these concepts involve to some degree a philosophical commitment to quality rather than precise balancing of costs and benefits. The highest value of stored grain IPM may be more in demonstrating a commitment to grain quality and a philosophy to eliminate unnecessary pesticide use. In other words, stored grain IPM may fit into the “triple bottom line” concept more easily than into the “financial bottom line.” IPM certification programs have been developed in other segments of the food industry. Developing a certification program for stored grain IPM would lead to some interesting discussion on the key practices and components. It would also provide another avenue for benefits.

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Erratum to: Human Health Problems and Accidents Associated with Occurrence and Control of Storage Arthropods and Rodents

Vaclav Stejskal, Jan Hubert and Zhihong Li

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In the original version of the book, Fig. 2.1 has to be inserted along with the citation in Chap. 2. The erratum chapter and the book have been updated with the change.

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