

NATURAL ENEMIES AS CONTROL AGENTS FOR STORED-PRODUCT INSECTS

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Introduction

Insect pests of stored products are attacked by a variety of natural enemies, including vertebrates, insects, mites, and pathogenic microorganisms. Some of these are of potential value as control agents for storage pests. The present paper outlines briefly how they might be applied for this purpose, illustrates their efficacy by presenting experimental results for representative examples, and concludes with a few remarks on what needs to be done and on the current direction of research.

Application of biological control involves one or more of the following: (1) introduction and establishment of exotic natural enemies (classical biological control), (2) conservation and augmentation of resident species, and (3) periodic colonization by inundative or inoculative releases. Use of the first approach in storages will probably be limited, because many natural enemies of storage pests have been widely distributed in shipments of insect-infested commodities and are now cosmopolitan. Biological control of storage pests is envisioned as comprising four primary approaches: (1) conservation by altering the environment to minimize adverse effects on natural enemies, such as limiting the use of chemical pesticides, (2) augmentation of natural enemies to make them more effective, as for example, development of insecticide-resistant strains or providing supplemental food to sustain biological agents during periods of low pest density, (3) inoculative releases, that is, serial releases of small numbers of predators or parasites, and (4) application of microbial insecticides.

Use of natural enemies to control insect pests in storage situations is not a new concept, but it has only recently received serious attention. This can be attributed, at least in part, to the often expressed objection that use of predators and parasites in and around stored products would increase contamination of the products with insect remains and to the observation that natural enemies appear in significant numbers only after a product has become heavily infested and serious damage has already occurred. Although the first criticism certainly has some validity, blanket rejection of biological control on the basis of sanitary requirements is unwarranted, because insect remains are of comparatively little concern in some products such as seed grain,

animal feed, and raw commodities that will be cleaned during processing. Also, because predaceous and parasitic insects have essentially no ability to penetrate packages, they could be used to advantage in warehouses containing packaged commodities to reduce pest populations and thereby decrease the chances of commodity infestation. With regard to the second objection, it is true that when nature is allowed to take its course, populations of predators and parasites are slow to overtake and suppress pest populations. However, the situation is quite different when natural enemies are deliberately released during the early stages of infestation, as will be illustrated by examples presented in this paper.

Predators

We know that stored-product insects are attacked by a variety of predators such as birds, rodents, spiders, and insects of various orders, but we know very little about their impact on pest populations. Only Xylocoris flavipes (Reuter), one of several predatory anthocorid bugs commonly found in storages, has been carefully studied in this regard. Its effect on prey populations was first reported by Jay et al. (1968) who showed that it could suppress small laboratory populations of several beetle species. Its efficacy was later confirmed by several experiments on a larger scale, the results of which serve to illustrate the potential value of predators as control agents.

The effect of X. flavipes on populations of the red flour beetle, Tribolium castaneum (Herbst), was demonstrated by Press et al. (1975) in an experiment with infested lots of inshell peanuts (farmers' stock). They placed each lot (about 210 liters) in a plywood bin with a capacity of about 1800 liters and added adult beetles at the rate of 4.5/liter of peanuts. One week later they added various numbers of adult predators to all but one bin. Samples taken 15 weeks after the peanuts were infested showed that the predator had suppressed population growth of the beetle and had reduced the number of damaged peanut kernels by 66% (Fig. 1). The beetle population in the control bin, which received no predators, reached a level of 38/liter. This represents a population increase in excess of 800%. The damage level was 37 worm-cut kernels/liter. In comparison, the beetle population in the remaining bins increased by only 20 to 80%, depending upon the initial density of predators, and damage to the peanuts was correspondingly less.

X. flavipes also had a pronounced impact on populations of the sawtoothed grain beetle, Oryzaephilus surinamensis (L.), in small lots (about 32 liters each) of shelled corn contained in 98-liter fiber drums (Arbogast 1976). Suppression in this case ranged from 97 to 99% (Fig. 2). Populations that were free of predation increased more than 1900-fold in 15 weeks - from 1.2 insects/liter

initially to more than 2500/liter. When predators were added to the drums at a rate of 0.3/liter one week after the beetles, population increase was only 65-fold, and when predators were added at higher rates, increase was reduced to about 20-fold.

LeCato et al. (1977) demonstrated that *X. flavipes* can effectively suppress residual insect populations in empty storage facilities. They introduced 15 pairs each of *O. surinamensis*, *T. castaneum*, and the almond moth, *Ephestia cautella* (Walker), into each of two warehouse rooms that were empty except for a small quantity of rolled oats scattered on the floor to simulate food debris and two boards placed on the floor to provide hiding places. A week later, they added 30 pairs of adult *X. flavipes* to one of the rooms. Changes in the almond moth population that was subjected to predation are compared in Fig. 3 with changes in the population that was free of predation. The almond moth, and in fact all three pest species, increased steadily in the room that

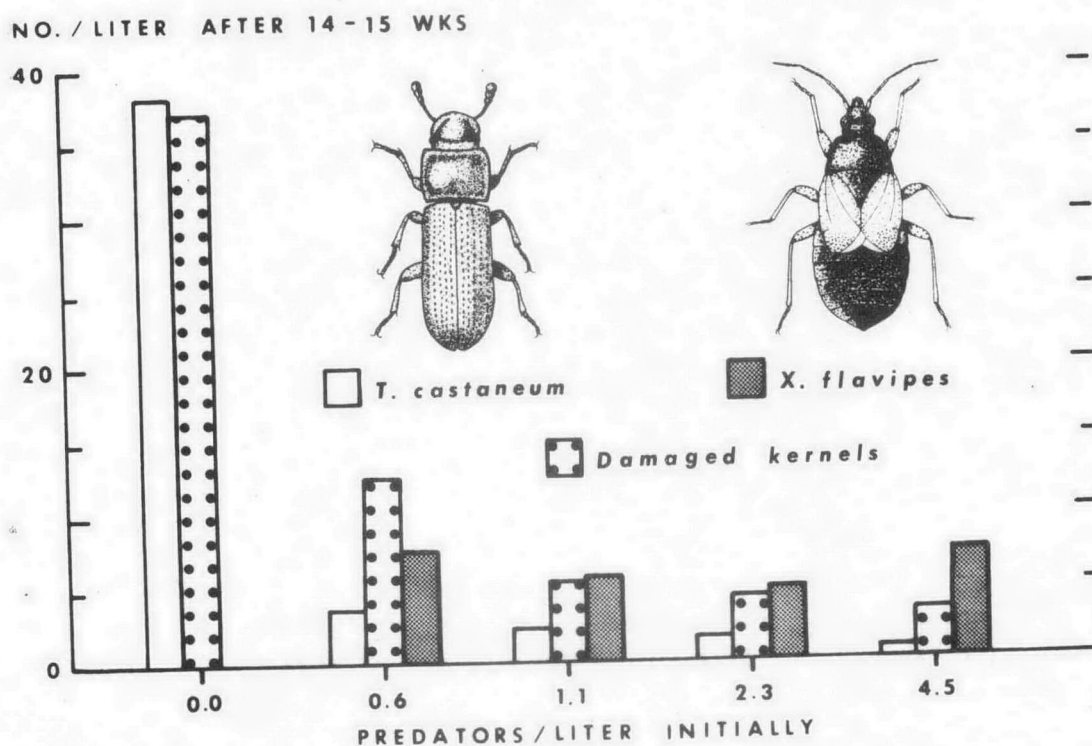


Figure 1. Effect of *Xylocoris flavipes* on populations of the red flour beetle infesting stored farmers' stock peanuts and on the amount of damage caused by the beetles. (Data from Press et. al. 1975).

received no predators and reached population levels in excess of 2500 after 100 days. When predators were present, the pest populations increased over the first 25 or 50 days and then declined to levels below those at which they were introduced.

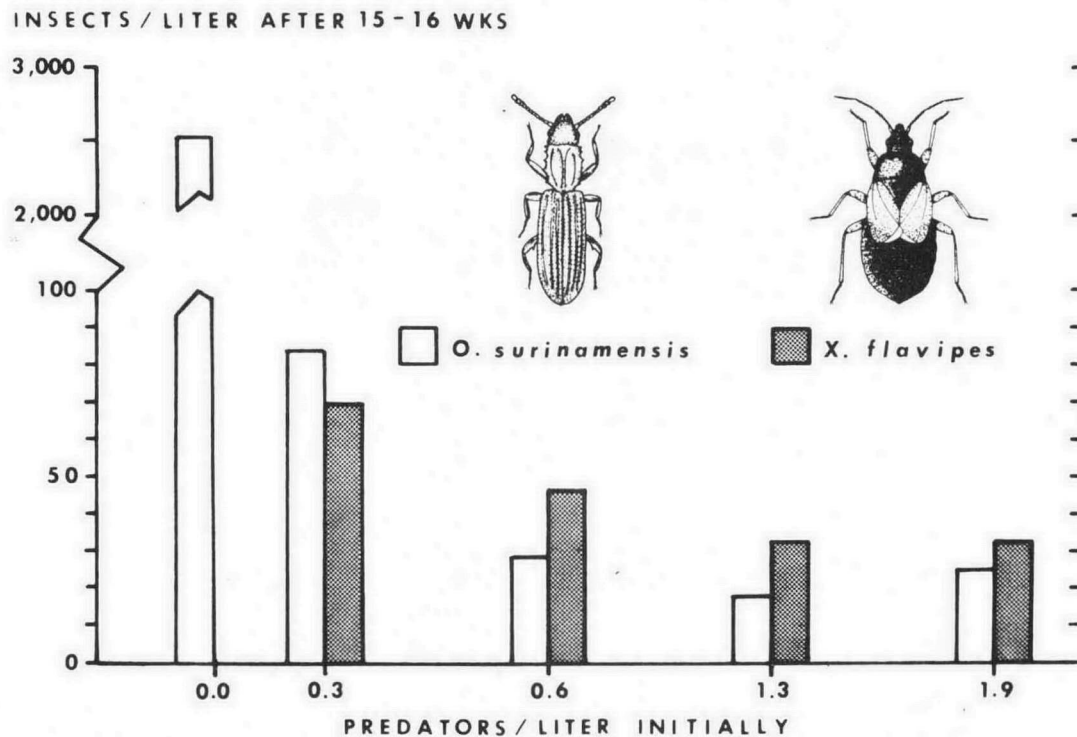


Figure 2. Effect of *Xylocoris flavipes* on populations of the sawtoothed grain beetle infesting stored corn. (Data from Arbogast 1976).

Parasites

The impact of hymenopteran parasites on populations of stored-product insects has been illustrated by a number of studies. Williams and Floyd (1971) showed that the pteromalid wasps *Anisopteromalus calandrae* (Howard) and *Choetospila elegans* Westwood can reduce population growth of the maize weevil, *Sitophilus zeamais* Motschulsky, in stored corn. They conducted experiments on 250-g replicates of shelled corn, each contained in a 1.9-liter round fiber carton and infested by the addition of 40 adult weevils. The cartons were held either in the laboratory or in a farm-type storage bin exposed to the elements. Five female

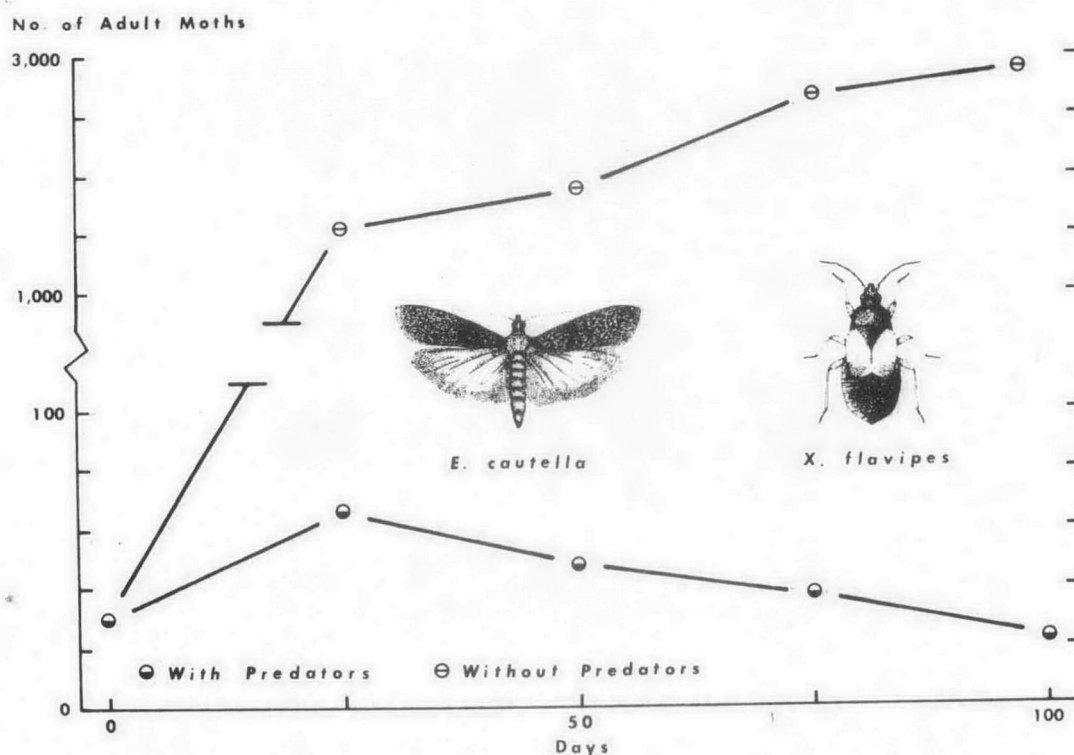


Figure 3. Trends in an almond moth population exposed to predation by *Xylocoris flavipes* and a population free of predation. (Data from LeCato et al. 1977).

parasites were added to each carton 21 days after the weevils were added, and the number of weevils was determined after 2 months and again after 4 months. Population growth of the weevil was reduced 25 to 50% by *C. elegans* and more than 50% by *A. calandreae*.

Some insight into the effectiveness of the bethylid wasp *Holepyris sylvanidis* (Brêthes) in regulating populations of *Oryzaephilus* can be gained from information reported by Spitler and Hartsell (1975). In order to evaluate pirimiphos-methyl (Actellic (R)) as a protectant for inshell almonds, they placed treated and untreated nuts in drums and stored them in a room where they were subjected to infestation by several species of insects, including the merchant grain beetle, *O. mercator* (Fauvel). When the almonds were first placed in storage, and at monthly intervals thereafter, 10,000 merchant grain beetles were released in the room. The beetle population in the untreated nuts reached a peak of about 1,000 adults/sample after 6 months of storage (Fig. 4). At that time, the control drums were invaded by *H. sylvanidis*, and the number of living beetles fell

abruptly. After 8 months there were only 200 beetles/sample and the parasite was no longer detectable. The beetle population then began to increase again, but leveled off following a resurgence of the parasite. Although, the test was terminated after 12 months, it appears that after overtaking its host population, the parasite was beginning to hold it at an equilibrium density of about 250-300/sample, even though a large source of immigrant beetles was provided at monthly intervals.

The braconid wasp Bracon hebetor Say, which parasitizes late-stage larvae of various moths, can effect significant natural control of moth populations in warehouses (Hagstrum and Sharp 1975), suggesting that it may have considerable potential as a biological control agent. The ichneumonid wasp Venturia canescens (Gravenhorst) has also shown potential for control of stored-product moths. The value of both species has been confirmed by laboratory and small scale warehouse tests. A single introduction of B. hebetor into laboratory cultures of the Indianmeal moth, Plodia interpunctella (Hübner), reduced emergence of adult moths by 74% (Press et al. 1974). Semiweekly releases of B. hebetor into a 42.5-m³ room containing food debris infested with E. cautella resulted in 97% suppression of the moth population and semiweekly releases of V. canescens resulted in 92% suppression (Press et al. 1982).

Parasitic mites have also demonstrated a marked facility for controlling storage pests, and their potential for this purpose was recently reviewed by Bruce (1983). One of the most promising species is the straw itch mite, Pyemotes tritici (Lagrèze-Fossat and Montané), which was evaluated by Bruce and LeCato (1979). They found that, in the laboratory, this mite inflicted 100% mortality on eggs, larvae, and pupae of O. mercator, and 97% mortality on adults. Although some stages of T. castaneum and of the cigarette beetle, Lasioderma serricornis (Fabricius), were resistant to attack, the mite inflicted 100% mortality on early-instar larvae of both species. Eggs, early-instar larvae, and adults of E. cautella and P. interpunctella were susceptible to attack and suffered mortality ranging from 91 to 100%. Late-instar larvae were apparently immune and pupae suffered only about 50% mortality. Several attributes of P. tritici make it especially attractive as a biocontrol agent: (1) high reproductive potential, (2) short life cycle (4-7 days), (3) females give birth to mature offspring, (4) 95% of offspring are females, (5) females mate at birth and begin host seeking immediately, (6) populations can easily be reared and synchronized, and (7) cosmopolitan distribution. The straw itch mite has one shortcoming as far as biological control is concerned. It does bite humans and produces a rashlike dermatitis. However, this drawback is not as serious as it may seem, because the mites usually live no longer than about 10

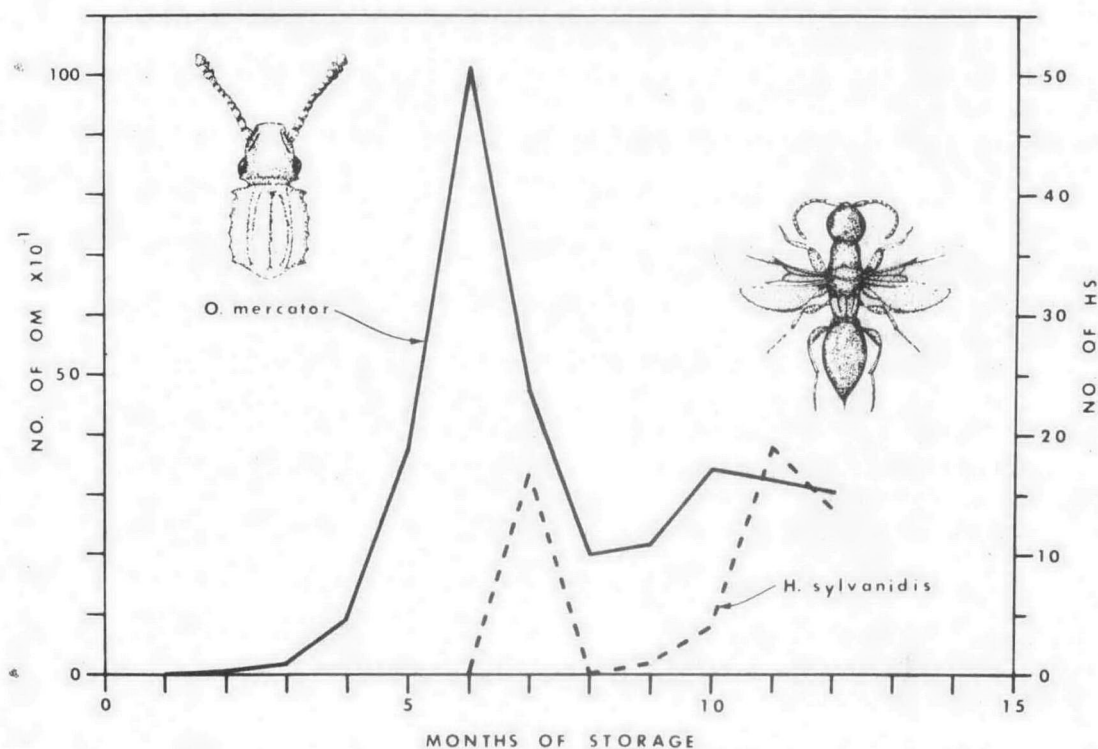


Figure 4. Relationship between population levels of the parasitic wasp *Holepyris sylvanidis* and its host *Oryzaephilus mercator* on stored inshell almonds. (Data from Spitler and Hartsell 1975).

days without hosts and so will die out soon after a pest infestation has been eliminated.

Pathogens

Stored-product insects are subject to infection by numerous pathogenic organisms including protozoa, bacteria, fungi, and viruses. These organisms provide some natural control of insect pests in storage facilities and some show considerable promise as biological control agents.

Protozoa. - Protozoa of the Class Sporozoa (Orders Gregarinida, Coccidia, and Microsporidia) are common and widespread among natural populations of stored-product insects. They generally produce chronic debilitating diseases that reduce population growth by increasing mortality, slowing development, and lowering fecundity.

Among the gregarines, most members of the Suborder Eugregarina do little damage to the host tissue and can be considered commensals, although they may become mildly pathogenic under conditions of dietary or other environmental stress. Harry (1967), for example, showed that when larvae of the yellow mealworm, Tenebrio molitor L., infected with the eugregarine Gregarina polymorpha (Hammerschmidt) (Stein) are reared under optimal conditions, they complete larval development as rapidly and achieve the same pupal weight as noninfected larvae. When infected larvae are grown on a suboptimal diet, however, their ability to complete development and the pupal weight they achieve are reduced. Another example is provided by the work of Dunkel and Boush (1969), who reported that larvae of the black carpet beetle, Attagenus megatoma (F.), infected with the eugregarine Pyxinia frenzeli Laveran & Mesnil lose weight almost twice as rapidly during starvation as do gregarine-free larvae. They suggested that since feral populations of A. megatoma are frequently infected with P. frenzeli and commonly subjected to partial starvation, P. frenzeli may serve as a continuous check on population growth. Schwalbe and Baker (1976), however, found little difference in weight loss during starvation between A. megatoma larvae infected with this organism and uninfected larvae. They suggested that the inconsistency between their results and those of Dunkel and Boush may have resulted from an improved nutritional state of the larvae in their tests or from a difference in the degree of infection.

In contrast to eugregarines, gregarines of the Suborder Neogregarina (Schizogregarina) are virulent pathogens. Finlayson (1950) reported that when adult female flat grain beetles, Cryptolestes pusillus (Schoenherr), are placed in a culture medium containing spores of Mattesia dispora Naville, all of their offspring die in the larval stage, so that the adult population declines rather than increases. He also found that this organism infects and kills the larvae of the rusty grain beetle, C. ferrugineus (Stephens), and a third unidentified, species of Cryptolestes, but that the larvae of C. turcicus (Grouvelle) are apparently immune. Species of Mattesia also infect various species of Trogoderma and are thought to be responsible for the erratic population trends in these beetles. Schwalbe et al. (1973) showed that the virulence of one of these pathogens, Mattesia trogodermae Canning, infecting larvae of T. glabrum (Herbst) is influenced by dosage and temperature. When they exposed larvae to 100 mg of spore powder per gram of culture medium for 24 h, the median survival time ranged from 20 (35°C) to 29 days (25°C). Ashford (1970) found that another neogregarine, Lymphotropha tribolii Ashford, prevents normal larval development and increases larval mortality in T. castaneum. Furthermore, although the longevity and fecundity of 75% of the survivors in his tests were normal, the remainder of the survivors were weakened and sterilized.

The Orders Coccidia and Microsporidia also include virulent pathogens of stored-product insects. Park (1948), for example, noted that an unidentified species of the coccidian genus Adelina limits growth of T. castaneum populations by increasing mortality, especially among the immature stages, and pointed out that it may also act by reducing fecundity. The microsporidian Nosema whitei Weiser is a pathogen of Tribolium species, and is also capable of infecting O. surinamensis but not T. molitor, Palorus ratzeburgi (Wisemann) (smalleyed flour beetle), Gnathocerus cornutus F. (broadhorned flour beetle), or P. interpunctella (Milner 1972b). Milner (1972a) showed that infection with this organism drastically reduces the rate of development of T. castaneum and delays molting after the second molt. Infected adults lay few eggs, although egg viability is unaffected. The pathogenic effect of N. whitei on T. castaneum is increased by dietary stress. George (1971) showed that mortality is higher and death occurs sooner among larvae on diets deficient in protein and cholesterol than among larvae reared on optimal diets. Other species of Nosema, N. plodiae Kellen and Lindegren and N. heterosporum Kellen and Lindegren, are pathogens of P. interpunctella (Kellen and Lindegren 1974). When P. interpunctella was reared on a diet containing spores of these Nosema, the number of insects that survived to the adult stage decreased as the spore concentration increased. Nosema heterosporum, with a median lethal concentration (LC₅₀) of 4.52×10^3 spores/g of diet, is more virulent than N. plodiae, which has an LC₅₀ of 8.09×10^6 spores/g of diet. All surviving adults of P. interpunctella reared on a diet containing N. plodiae were infected. Such adults transmit the organism to other adults during mating and through the ovaries to the next generation (Kellen and Lindegren 1971).

Research to evaluate the efficacy of protozoa in controlling stored-product insects has focused primarily on using a combination of pheromone and protozoan spores to attract and inoculate the insects (Burkholder and Boush 1974). Males attracted and contaminated in this manner return to their natural habitat where they infect others of their kind. Shapas et al. (1977), who evaluated pheromone-baited, spore-transfer sites for suppression of T. glabrum with M. trodogermae, found that a single introduction of spores by this method into dense populations of T. glabrum produced 81% suppression of the F₁ generation and nearly 100% suppression of the F₂ generation.

Bacteria. - Numerous species of bacteria occur in association with insects, but relatively few can be considered insect pathogens. Perhaps the best known insect-pathogenic bacterium is Bacillus thuringiensis Berliner, a rod-shaped, spore-forming organism that produces a disease of lepidopterous larvae. Sporulating cells of this organism contain an ovoid endospore and a crystalline parasporal body which are eventually released by autolysis.

Insects normally contract the disease by ingesting the spores and crystals. Ingested crystals are transformed to an active toxin that damages the cells of the insect midgut, thus inhibiting feeding and giving entrance to the spores.

Susceptible species may be affected by the spores alone or by the crystals alone, but in most species a mixture of the two produces the greater mortality. Thus crystals are about 30x more toxic to E. cautella than spores, and the toxicity of a mixture is directly related to the number of crystals in the mixture. On the other hand, crystals are only about 3x more toxic to P. interpunctella than spores, and maximum toxicity is obtained with a 50:50 mixture (McGaughey 1978a).

Bacillus thuringiensis provides control of E. cautella and P. interpunctella when applied to grain as an aqueous suspension, as a dust, or as a bait. It is effective as a bulk treatment (all grain uniformly treated) or surface layer application (uniformly treated grain layered over untreated grain) and persists under storage conditions without noticeable decrease in insecticidal activity for at least a year (McGaughey 1976, 1978b, Kinsinger and McGaughey 1976). Larvae of the Augoumois grain moth, Sitotroga cerealella (Olivier), are also susceptible and although application of B. thuringiensis is less effective in achieving control, application at rates sufficient to control P. interpunctella and E. cautella can be expected to substantially reduce population growth of S. cerealella and may obviate the need for other control measures (McGaughey 1976, McGaughey and Kinsinger 1978).

Bacillus thuringiensis is compatible with several grain fumigants (McGaughey 1975a). The toxicity of a formulation consisting of spores and crystals was not reduced by treatment with phosphine, carbon tetrachloride-carbon bisulfide, ethylene dichloride-carbon tetrachloride, or methyl bromide, although methyl bromide killed or otherwise prevented germination of the spores.

Viruses. - Insects are affected by a diverse group of viruses, some of which are important pathogens of storage pests. Among these pathogens are granuloses of P. interpunctella (PGV) and E. cautella (CGV) and a nuclear polyhedrosis of E. cautella. Granuloses are rod-shaped nuclear viruses in which each virus particle is enclosed in a protein capsule. Nuclear polyhedroses form cubic, hexagonal or nearly spherical polyhedra, each containing numerous virus rods, in the nuclei of host cells. When capsules or polyhedra are ingested by a susceptible insect, the protein surrounding the virus particles is digested and the particles enter the host tissue.

Granuloses are usually host species specific, although some show cross infectivity. At a concentration of 8.6×10^4 capsules/g of a bran diet, PGV killed 96% of exposed P. interpunctella larvae in 25 days (Hunter 1970). CGV killed 74% of E. cautella larvae exposed to a concentration of 1.2×10^5 capsules/g and 100% of those exposed to a concentration of 1.2×10^6 capsules/g (Hunter and Hoffman 1970). Ephestia cautella, E. elutella (Hübner) (tobacco moth), and Cadra figulilella (Gregson) (raisin moth) were found to be nonsusceptible to PGV (Hunter 1970), but P. interpunctella was moderately susceptible to CGV (Hunter and Hoffman 1972). Although capsules of CGV from infected P. interpunctella larvae were generally abnormal in form and contained multiple virus particles, the virus remained virulent and developed normally in the natural host.

The efficacy of PGV as a microbial insecticide was investigated by Hunter et al. (1973, 1977, 1979). An aqueous suspension of this virus effectively protected peanuts, almonds, walnuts, and raisins against infestation by P. interpunctella. Inshell almonds were protected for 134 days and feeding damage was so reduced that the number of rejected nuts dropped by as much as 88%. McGaughey (1975b) found that aqueous and dust applications of PGV gave effective control of P. interpunctella on stored corn and wheat when the material was applied as a bulk treatment, a mixed treatment (treated grain mixed with untreated grain), or as a surface layer treatment. PGV was found to be compatible with malathion, and a mixture of the virus and chemical provided better control of P. interpunctella on almonds than either material alone (Hunter et al. 1975).

In contrast to granuloses, many nuclear polyhedroses are cross infective among lepidopterous species. A nuclear polyhedrosis isolated from E. cautella showed an LC_{50} of between 0.25 and 0.50 polyhedra/mm² of surface of an agar base diet (Hunter et al. 1973a). Most larvae died of the disease in 8 to 10 days after exposure to the virus at a concentration of 4 polyhedra/mm². On a bran diet, the LC_{50} was slightly greater than 3.2×10^4 polyhedra/g, and higher concentrations were required when the inoculum was mixed with the diet than when it was layered on the surface. Plodia interpunctella was also found to be susceptible to this virus, although susceptibility was lower than in the almond moth and the infection developed more slowly. A concentration of 6.4×10^4 polyhedra/g of a bran diet killed 45% of exposed larvae (Hunter et al. 1973b).

Concluding remarks

The potential value of biocontrol as a method of protecting stored commodities from insect damage has been clearly demonstrated, but technologically, the method is still

rudimentary. Further development of biological control as a storage procedure will require field trials to answer questions concerning: (1) efficacy under actual conditions of commercial storage, (2) compatibility with other storage procedures, (3) cost effectiveness, and (4) undesirable side effects. Routine application of the method will also require improved techniques for mass producing natural enemies suitable for use in the storage environment.

Some progress has already been made along these lines. Bacillus thuringiensis, for example, is produced commercially and has been used in the control of lepidopterous pests for some time. It has recently been registered as a wettable powder and dust formulation for moth control on stored grains, soybeans, sunflower seeds, crop seeds and peanuts. The granulosis virus of the Indianmeal moth (PGV) is still under investigation at USDA's Stored-Product Insects Research Laboratory, Fresno, California. The next step in evaluating this pathogen will probably include large scale applications of the virus under commercial warehouse conditions, but pursuit of safety data for registration will depend upon future interest by firms engaged in producing biocontrol agents (W. R. Kellen, in litt.). No registration is required for use of predators and parasites, but up until now none of these agents have been evaluated under commercial warehouse conditions. A pilot test is currently underway at our laboratory to evaluate B. hebetor and X. flavipes for controlling pests of stored farmers' stock peanuts. This test will provide the first data on the efficacy and cost effectiveness of predators and parasites in actual storage conditions.

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