## **Insect Pests: Microbial Control**

A seminar on progress in the microbial control of insects was held in Fukuoka, Japan, 21–23 April 1967 under the auspices of the United States-Japan Co-operative Science Program and sponsored by the Japan Society for the Promotion of Science and the National Science Foundation. The general theme of past accomplishments, present state, and future possibilities of utilizing insect pathogens for effective pest management was selected. The potential of viruses, bacteria, fungi, protozoa, and nematodes were discussed in detail.

H. Aruga (Tokyo University) emphasized the importance of pathogens that cause disease in the management of beneficial insect species, such as the silkworm and the honeybee. He pointed out the similarity of principles and stated that information on hostpathogen relationships in beneficial host species could be useful to the understanding of host-pathogen interactions in pest species. The silk industry has been very important in the Orient for many centuries and problems concerning pathogens in the management of the silkworm have been encountered frequently. Accordingly, a large proportion of the studies of insect pathogens concern the silkworm. These studies, neverthelesss, are very useful to pathologists investigating other insect species.

S. Kawase (Nagoya University), in discussing the infection process and chemical nature of the cytoplasmicpolyhedrosis virus of the silkworm, showed that its multiplicative pattern appears to be similar to that of other viruses and the viral nucleic acid is double-stranded RNA with a sedimentation coefficient of 17S. The nucleus in the midget epithelium appears to be the site of protein synthesis. T. Hukukara (Tokyo University) discussed genetic variation among the cytoplasmicpolyhedrosis viruses of which two variants have been isolated. One causes

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the formation of hexahedral, and the other, icosahedral inclusions. The virus particles, however, are morphologically and serologically similar and they are equally virulent for *Bombyx mori* and *Malacosoma neustria testacea*. H. Watanabe (Tokyo University) reported that selected strains of silkworm are variable in their susceptibility to cytoplasmic-polyhedrosis virus and suggested that resistance may be controlled by a single dominant gene.

K. Yamafuji (Kyushu University) reported his latest observations on inducing nuclear-polyhedrosis virus infections in the silkworm. He concluded that a previral DNA component is contained in the normal silkworm genome and that inducing agents such as hydroxylamine cause the viral component to multiply and cause disease. The previral DNA appears to be highly resistant to shearing forces with a sedimentation constant of 35S. These studies pose interesting questions regarding nuclearpolyhedrosis virus infections in other host species. For example, is the virus transmitted throughout a host population in the same manner as the host genetic material? G. R. Stairs (Ohio State University) presented evidence that the nuclear-polyhedrosis viruses are not transmitted through the host population in the same manner as are genes. He suggested that they are essentially parasitic and that simple numerical and spatial relationships play a major role in the survival of these viruses in their host populations.

More than one virus often is found in a host population. The army worm, *Pseudoletia unipuncta*, is an example. Y. Tanada (University of California, Berkeley) discussed his studies in this area. A nuclear-polyhedrosis virus and a granulosis virus occur simultaneously in field populations of this species. In laboratory studies he found that the granulosis virus appears to enhance the effect of the nuclear-polyhedrosis virus. The granulosis virus retained its effect after inactivation by heat. The specific synergistic activity appears to be located in the virus rod and not in the inclusion body protein. The interaction of viruses in insects was discussed further by H. Aruga (Tokyo University) who stated that the silkworm is susceptible to two types of cytoplasmic-polyhedrosis virus and that these viruses interfere with one another. The virus of flacherie also interferes but the nuclear-polyhedrosis virus does not interfere with the cytoplasmic-polyhedrosis viruses.

An assessment of the nuclear-polyhedrosis virus of Heliothis species as a factor in the management of these pests was made by G. E. Allen (Mississippi State University). He suggested that the virus can be successfully utilized in the management of Heliothis on cotton crops. The introduction of virus does not interfere with parasites and predators but appears to be most effective when the density of these parasites and predators is relatively high. Similar studies by K. Akutsu (Tokyo Agricultural Experiment Station) showed that the cabbage army worm, Memestra brassicae, and the cabbage butterfly, Pieris rapae crucivora, may be controlled by inclusion-type viruses, the former with a nuclear-polyhedrosis and the latter with a granulosis. Similarly, R. Koyama and K. Katagiri (Asakawa Forest, Government Forest Experiment Station, Tokyo) reported that the pine caterpillar, Dendrolimas spectabilis, has been successfully controlled by a cytoplasmic-polyhedrosis virus that was disseminated in a water suspension from helicopters at the rate of 10<sup>11</sup> polyhedra per hectare.

The safety of insect pathogens was discussed by A. M. Heimpel (U.S. Department of Agriculture) who stated that numerous pest insect species are susceptible to one or more pathogens and that these pathogens are not infectious for plants, beneficial insects, vertebrates, or man. The artificial dissemination of these pathogens for control purposes does not constitute a hazard to humans or livestock because they are continuously present in the pest populations as a part of the natural microbiota. The frequency or concentration relative to the susceptible pest species is simply increased by the artificial dissemination of a pathogen so that the level of damage caused by the pest population can be suppressed below the economic threshold for the crop in question. Heimpel's remarks were dirceted particularly toward the use of the inclusion-type viruses.

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The bacteria of insects have been used successfully for control of pest species in many countries. T. Watanabe (Kvushu University) discussed his studies on the chemistry of the toxic crystal of Bacillus thuringiensis. The crude toxin was fractionated into two active protein components with Sephadex G-200 but the elution patterns of both components appeared to be identical in DEAE-cellulose columns, K. Aizawa and N. Fujijoshi (Kyushu University) reported success in selecting strains of Bacillus thuringiensis that are highly pathogenic for the pest species, Pieris rapae crucivora, Hyphantria cunea, Plutella maculipennis, and Chilo suppressalis, but much less pathogenic for the silkworm, Bombyx mori. Thus, these strains can be used without fear of damaging the important silkworm population. They also reported on the isolation of a spore-forming bacterium, Bacillus moritai, that is highly pathogenic for the housefly, Musca domestica. The bacterium is nonpathogenic for the silkworm, mouse, chicken, pig, canary, or fish. Field tests have been very successful.

The status of growth and spore production on artificial media of *Bacillus popilliae* and *Bacillus lentimorbus* pathogens of the Japanese bettle was discussed by R. Rhodes (U.S. Department of Agriculture). Considerable progress had been made in the production of viable spores on artificial media but these media are still not as satisfactory as the hemolymph of the host larvae.

J. D. Briggs (Ohio State University) pointed out that commercial preparations of *Bacillus thuringiensis* have been used successfully in the United States but extensive and comprehensive education of entomology students in the ecology of insect pathogens will be necessary before the most effective exploitation of these organisms can be made. Appreciation of the ecosystem concept and epizootiology of insect diseases are essential components in the full understanding of insect biology and successful management of insect populations.

The assessment of important ecological factors in the utilization of pathogens was demonstrated in a paper on fungus disease of mosquitoes by J. M. Couch (University of North Carolina). The fungi, *Coelomomyces*, are obligate parasites of mosquitoes and there appears to be a close time correlation between hatching of eggs and germination of sporangia. Apparently, a very delicate relationship between the fungus and its host has evolved and careful ecological studies are necessary before these pathogens can be used effectively for pest management.

The infection mechanism of insect pathogenic fungi was discussed by J. Aoki (Tokyo University) who suggested that the moisture level at the intersegmental membrane was higher than at other areas of the cuticle. Consequently, conidiospores germinate more readily at the intersegmental areas. Also the tanning action in the insect cuticle may inhibit the penetration of hyphae and, in highly pathogenic strains, protease activity appeared to be stronger. A summary of the various species of fungi causing "muscardine" diseases in the silkworm and other insects was presented by K. Aoki (Ministry of Agriculture and Forestry, Tokyo). The pathogenicity and ecology of some of these fungi were discussed in respect to their interaction with the silkworm. The possible utilization of the fungus, Aschersonia aleyrodis, for control of the citrus whitefly was considered by N. Oho (Horticultural Research Station, Hiratsuka). D. W. Roberts (Boyce Thompson Institute for Plant Research, New York) explained that the possibilities of utilizing fungi for pest suppression is very promising but a good deal of basic study remains to be done before this end can be realized. For example, the toxins produced by insect fungi may be purified and these may eventually become useful "insecticides."

J. P. Kramer (Cornell University) reviewed some of his work on the ecological relationships of the protozoa with their host insect species and emphasized the need for comprehensive ecological studies before successful utilization of this group of pathogens can be accomplished.

An appraisal of the DD136 nematode for the control of insect populations was made by S. R. Dutky (U.S. Department of Agriculture). The nematode and its associated bacterial insect pathogen are capable of entering and killing many insect species. The complex can be grown on artificial media and in various, easily reared, insect hosts; so large quantities are not difficult to obtain. It has been artificially disseminated with success into populations of the codling moth, boll weevil, and housefly larvae.

The isolation and characterization of immunity principles in silkworm larvae were reported by T. Kawarabata and K. Aizawa (Kyushu University). A bacteriolytic substance in the hemolymph with a proteinaceous nature has been verified and a viral inhibitory factor has been detected in silkworm hemolymph after infection by nuclearpolyhedrosis virus. Investigations such these provide valuable ecological information about host-pathogen relationships.

An interesting account of the interactions between pathogens, hosts, and host parasites was presented by M. Tamashiro (University of Hawaii). Pathogens infecting a host usually do not infect the parasites but the suitability of the host as a substrate for the development of the parasite is decreased by infections. The adult hymenopterous parasites were also capable of transmitting virus from host to host on their ovipositors.

I. M. Hall discussed integrated microbial control and chemical control and concluded that future population management will necessitate expansion of integration procedures where different types of pathogens and chemicals will be used for the suppression of the total pest population. E. A. Steinhaus (University of California, Berkeley) emphasized the ecological approach to the study of insect pathogens and pointed out that studies of the pathogens of the silkworm and honey bee may contribute much to the utilization of microbes in pest population management.

It was obvious from the papers and the ensuing discussions that the use of pathogens in the management of insect populations has great potential. Microorganisms definitely should be exploited by entomologists in their continuing battle to prevent crop damage and spread of infectious diseases of man and animals. There was general agreement that extensive ecological studies are most essential to the understanding of both simple and complex pathogen-host relationships. The understanding of these relationships will be necessary before the pathogens can be used successfully and effectively. Much worthwhile work has been done but a strong and expanding effort must be made in order to fully realize the potential usefulness or, in beneficial species, destructiveness of microorganisms in population management.

Co-organizers of this seminar were K. Aizawa (Kyushu University) and G. R. Stairs (Ohio State University).

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