I.12 The Biological Control Potential of Parasites, Predators, and Fungal Pathogens

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Introduction

Grasshoppers, like all other animals, are subject to a large number of parasites, predators, and pathogens, including fungi, protozoa, and viruses (Henry et al. 1985, Prior and Greathead 1989, Streett and McGuire 1990). Parasites, predators, and pathogens can be used as "classical" biological control agents. Classical biological control is defined as "the importation and release of an organism outside its natural range for the purpose of controlling a pest species" (Howarth 1991). Another approach, "augmentative" biological control, uses native or exotic organisms that are released periodically to enhance mortality in a targeted pest population. Insect pathogens generally fall in this category because many can be mass-multiplied and applied as biological pesticides (Prior and Greathead 1989).

Insect Parasites and Predators

Classical Introduction Approach.—According to a review article by Prior and Greathead (1989), the classical biological control of a pest grasshopper using an insect parasite or predator as the beneficial agent has been attempted on nine occasions: there were two cases using bombyliids or bee flies, three cases using sarcophagid flies, two cases using meloid beetles, and two cases using scelionid wasps. Only two of these nine attempts resulted in the establishment of the introduced beneficial, a meloid beetle in Corsica and a scelionid wasp in Hawaii. However, the only project that has been claimed as a success was the introduction of a *Scelio* sp. from Malaysia, released against the rice grasshopper in Hawaii.

As suggested by Greathead (1992) and by Siddiqui et al. (1986), the possibilities for classical work certainly have not been exhausted, particularly with any scelionid egg parasites having an acceptable degree of host specificity. A controversy surrounding the request by Richard J. Dysart for permission to release a species of *Scelio* from Australia against pest grasshoppers in the United States seemed to pivot around the issue of host specificity. In spite of the constraints involved in the classical biological control approach, there are even more problems to consider in the augmentative approach.

Augmentative Approach.—Using insect parasites or predators as substitutes for chemical insecticides is not considered feasible for the control of grasshoppers. In his recent review of biological control options for tropical locusts and grasshoppers, Greathead (1992) expressed the same sentiments. In order for this approach to be workable, the natural enemy to be used must have a number of attributes:

- An acceptable level of host specificity, assuring some degree of safety to nontarget organisms,
- The ability to be easily reared in a laboratory situation and be produced in large quantities, and
- Costs of production and delivery to the target areas low enough so that the cost of using the biocontrol organism is competitive with the cost of using chemicals.

Concerns about host specificity would eliminate several groups of natural enemies, for example, the meloid and carabid beetles, whose larvae wander through the soil in search of a wide range of hosts. Similarly, certain beneficial groups can be eliminated from consideration because they are not amenable to handling in captivity, for example, the egg predators (Bombyliidae, Meloidae) and the nemestrinid parasites (Greathead 1992).

Although certain scelionid egg parasites can be reared easily in the laboratory, the rearing process is dependent on a constant supply of grasshopper eggs of a certain age. Considering the immense areas that would require release of parasites, plus the logistics of rearing and delivery, it is certain that the costs of using *Scelio* sp. parasites in an augmentative approach would be unacceptable.

Classical Introduction Approach to the Use of Fungi

One of the first documented reports of attempting to use *Entomophaga* (= *Empusa) grylli* Fresenius (Batko) as a classical biological agent occurred in South Africa in 1896 (Howard 1902). A man named Arnold Cooper, of Richmond, Natal (South Africa), noticed grasshoppers dying apparently from a fungous disease. He took specimens to the Bacteriological Institute at Grahamstown, where a fungus capable of infecting healthy grasshoppers was isolated. Subcultures of the isolate were made, and vials containing them were distributed to planters in areas

where grasshoppers were abundant. Planters such as H. H. Wells chronicled the situation in 1899: "I dipped captured adult grasshoppers into fluid containing the fungus then released them into the swarm over a period of two to three days...to my profound astonishment I found grasshoppers hanging in clusters all over my farm...millions of them." Many other equally favorable reports were received by the Bacteriological Institute, and distribution of the culture tubes continued.

Questions concerning the precise "nature" of the fungus were raised in 1899 and 1900. Specimens sent to the Royal Botanic Gardens, Kew, England, were identified as a *Mucor* sp. The same determination had been made simultaneously in Victoria, Australia, from similar specimens received from Natal. Circumstantial evidence suggests that perhaps two different fungi were in fact distributed. *Mucor* sp., which is easily cultivated and was readily identified by the authorities of the day, could have been contaminated with resting spores of *Entomophaga* sp. This scenario would support the reports of "clusters of diseased grasshoppers" by planters such as H. H. Wells and early photographs showing dead grasshoppers hanging from the tops of foliage. That phenomenon provides strong evidence of infection by *Entomophaga* sp. It is also apparent that "mixtures of fungal cultures" originating in South Africa were freely distributed to Australia and North America during the period 1899–1901 (Howard 1902).

Documents indicate that fungus cultures were obtained from South Africa by Dr. L. O. Howard in 1900 for subculture and release against grasshoppers in Colorado. A total of 223 "probable releases" were made in 24 States plus the Philippine Islands and Cuba during the period 1901–02 (Howard 1902). Howard further states that "No effort was made to determine the exact nature of the fungus contained in the culture tubes received from South Africa in the spring of 1900, but subsequent events indicate plainly that the Bacteriological Institute at Grahamstown is sending out more than one kind of fungus."

Professor L. Bruner (1901) also reported on a fungus, *Sporotrichum* sp. (= *Beauveria* sp.) he discovered infecting locusts in Argentina in 1897–98. He noted that "it is quite similar to the fungus which is used in destroying chinch-bugs in some portions of the United States." Bruner also stated "that [although] considerable time has been spent in experimenting with this South American fungus upon our North American grasshoppers, thus far the results have all been negative since not a single insect has died from the disease."

These early attempts to use entomopathogenic fungi as "classical" biological control agents set the precedent for introduction and distribution of exotic pathogens in North America. It is apparent that numerous releases of unknown species from a wide variety of locations were made with little concern for environmental consequences beyond reduction of the pest species of the day.

For more than 100 years, the literature on grasshopper fungi has documented the evolution of a wide range of biological facts and observations. Habitat and climatic requirements are most often alluded to as dampening factors for the expression of fungus disease. The initial association between cool, wet, spring weather and an ensuing fungus epizootic plus other observations led to the current data base.

Many entomologists have reported the importance of microhabitats and macrohabitats for the development and expression of fungus epizootic among grasshopper populations. Reports indicate that fungus-infected grasshoppers are often restricted to roadside ditches; perimeters of cropland; low-lying, moist swales and intermittent waterways in pastures and hayfields; and various other noncultivated habitats (Hostetter et al. 1992 unpubl., Packham et al. 1993, McDaniel 1987).

A review of the accumulated information suggests that perhaps entomopathogenic fungi can be exploited in a "classical" sense through novel manipulations and applications already existing in North American agroecosystems.

The theoretical basis for the use of pathogens in biological control has been thoroughly discussed by many authors; most notably by Anderson (1980, 1982) and Hochberg (1989).

A mathematical model derived by Hochberg (1989) shows that host populations may be regulated to low and relatively constant densities if sufficient numbers of

pathogens are translocated from reservoirs to habitats where transmission can occur. The model accounts for host–pathogen interactions based on heterogeneity; pathogen populations are not uniform. Transmissibility and lifespan of the pathogen differ among individuals or life stages in the environment. Pathogens are considered as two distinct subpopulations; one as transmissible and short lived, and one as nontransmissible and long lived (e.g., *Entomophaga macleodii* and *E. grylli* pathotype 3, conidia and resting spores).

Infective entities of the pathogen can cause infection only when they are translocated (abiotically or biotically) from the reservoir to the susceptible host. Hochberg suggests that, to increase the efficacy of indigenous pathogens of insects, the focus should be on the identification and manipulation of pathogen reservoirs between nontransmissible and transmissible subpopulations.

The model suggests that for the introduction of exotic pathogens as classical biological control agents, the conditions for the likelihood of success are (1) long lifespan of pathogen stages residing in reservoirs and (2) the propensity of these stages to be translocated to the habitat of the host for transmission.

Two practical applications of this model would be the use of existing Conservation Reserve Program (CRP) land and Federal and State highway rights-of-way as reservoirs or "refugia" for hosts, pathogens, parasites, and predators (Parker 1971).

The CRP program, which was devised in accordance with Title XII of the Food Security Act of 1985 (P.L. 99–198), provides for farmers to enter voluntarily into multiyear (10-year minimum) contracts with USDA to take specified highly erodible cropland out of annual production and put it into some other permanent vegetation. CRP acreage has been identified, quantified, and mapped for each county in each State by personnel of USDA's Agricultural Stabilization and Conservation Service. Blocks of land most often occur in multiples of 40 acres and will be available as a stabilized system (for a minimum of 10 years).

It may be feasible to isolate grasshopper populations on CRP acreage with timely applications of chemical agents or mechanical barriers followed by inoculation/suppression with biological agents utilized in concert with naturally occurring parasites. Geographical imaging systems (GIS) are in place and could be used to delineate graphically and link strategic release areas based on ecological requirements of natural enemies across vast acreages. Host–pathogen reservoirs could be maintained and manipulated by augmentative releases of pathogens, parasites, and predators.

Manipulation of the habitat could be effected in a variety of ways: (1) clearcutting or stripcutting of foliage, which forces susceptible stages of the target species to concentrate in an area favorable to pathogens and arthropod natural enemies; (2) regulation of irrigation practices to create optimum habitat (cover crops) within the reservoir; (3) timely use of disruptive techniques (cultivation, spring-tooth harrow, mowers) to facilitate movement of pathogens from the soil (reservoir) to the host habitat (transmission–infection arena).

The current soil conservation program under the aegis of P. L. 99–198 will probably be succeeded by another "idle acres" program that may provide an exceptional opportunity for demonstrating the principles of IPM.

Federal and State highway rights-of-way could be manipulated to become "beltway reservoirs" for beneficial organisms across entire States. Millions of dollars are spent each year throughout the rangeland States for highway beautification and maintenance programs (e.g., landscaping, mowing, spraying). Monies diverted into development and conservation of habitat may be a wise investment toward long-term stability in the agrosystem. Perhaps a highly visible program of conservation and manipulation of "reservoirs of natural enemies" along the Nation's roadways would pique public interest and support.

Augmentative Approach.—Presently, entomopathogenic fungi have the greatest probability of exploitation as microbial control agents for managing grasshopper populations. The wide range of orthopteran hosts and environments from which fungi have been isolated has revived interest in this group over the last decade.

Worldwide, at least 10 genera of fungi are known to be entomopathogens of grasshoppers and locusts (Prior and Greathead 1989). Use in the initial phase will be "augmentative": "insecticidal" formulations and applications will used to augment natural enemies in the target area (Foster et al. 1991–94 unpubl.).

The most promising candidates are found among the *Beauveria* spp., *Metarhizium* spp., and *Entomophaga* spp. *Beauveria* spp. and *Metarhizium* spp. have host-specific strains and are purported to be nonhazardous to nontarget organisms (Prior and Greathead 1989). Conidia, or spores (the infective entity), are easily produced on commercially available solid substrates or in fermentation processes and can be formulated and applied similarly to other contact chemical pesticides (Foster et al. 1991–94a and b unpubl.).

Because they are lipophilic, the conidia of *Beauveria* spp. and *Metarhizium* spp. can be formulated with oil carriers and applied via ultralow-volume techniques. Oil droplets have the advantage in that droplets of smaller volume (mean diameter) can be generated at the nozzle (time of release), and the oil prevents evaporation during travel to impact on the target (grasshopper cuticle). Oil formulations have the advantage of spreading over the also lipophilic insect cuticle, thereby carrying conidia to intersegmental membranes and joints. Delivery to those areas increases the probability of penetration and infection of the insect (Prior and Greathead 1989).

Vegetable, soybean, or corn oils produced within or near insecticide-application areas could provide sustainable, nontoxic, environmentally safe formulation bases. The use of vegetable oils could decrease reliance on petroleum-based carriers.

The augmentative application of *Entomophaga grylli*, pathotype 1 (= *E. calopteni* [Bessey] Humber), was attempted in South Dakota (McDaniel 1987). McDaniel noticed the presence of *E. grylli* while conducting grasshopper surveys in 1979–80. Among other observations, he noted that the majority of grasshoppers dying from the fungus were found in areas not subject to cultivation (e.g., field borders, roadside ditches, alfalfa fields) and from the edges of corn and soybean fields.

McDaniel reported that he "triggered two fungus outbreaks in the spring of 1982 in plots in Hughes county near Blunt, SD and at a location 21 miles west on the Bad River road in Stanley county." The triggering was accomplished by collecting 4,468 plant sections, each of which had a fungus-killed grasshopper attached; taking them to an area known to be free of the fungus disease; and taping them to the tops of tall grasses and alfalfa plants.

Fungus-killed grasshoppers were observed 15 days after inoculation and a 53-percent reduction of the population occurred within 45 days. McDaniel also reported that the fungus continued to kill grasshoppers at these plots through 1986 with no additional inoculum of spores.

McDaniel developed a method of extracting resting spores from cadavers for inoculation of field plots. He extracted 2 gal of pure spores from 38 gal of hand-picked, dead, fungus-killed grasshoppers. He was able to effect disease in release plots using infected grasshoppers or by applying with a grass-seed spreader ground-up bodies of *Melanoplus differentialis* (Thomas), *M. bivittatus* (Say), and *M. sangunipes* (F.) that had been treated with fungal spores.

McDaniel (1987) attributed the unsuccessful inoculations done with pure resting spores to the fact that they had been stored for several months at room temperature between collection in late fall and application in early spring.

Entomophaga spp.—particularly the Australian isolate, *Entomophaga grylli* pathotype 3—may be best utilized as "classical biological control agents." Members of this complex cannot be produced easily on axenic substrates or in large enough quantities to be used as insecticidal treatments. Current ideology views this as a limitation of the present state of technology; however, perhaps not all entomopathogenic fungi or other microbial agents are best used as insecticides.

The best utilization of entomopathogens will evolve over time along with increased understanding of the ecology and the systems that regulate it. The many avenues of availability are just beginning to be explored. Exploitation will require long-term commitment, innovative

approaches, and the willingness to tailor management practices within the principles of ecology.

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