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Mycopesticides as part of integrated pest management of locusts and grasshoppers

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Abstract

The search for alternatives to the widespread use of chemicals to control locusts and grasshoppers has led to the development of mycoinsecticides based on *Metarhizium anisopliae* var. *acridum*. Two products are available commercially: Green Muscle® in Africa and Green Guard® in Australia. Only Green Guard® has been used in large control operations, with nearly 80,000 ha treated since operational use began in 2000. Research, though important, was not sufficient to lead to operational use of Green Guard®: the critical factor was the formation of the Locust and Grasshopper Biocontrol Committee that consisted of research providers, end-users and a commercial partner who together favored the rapid development of a commercial product.

In Australia, *M. a. var. acridum* is used in environmentally sensitive areas such as near waterways, or where there are rare and endangered species, and on the many properties, in locust-source areas in the interior, that produce organic beef for export. However, an increasingly important use is by landholders, who are now required to list all the chemicals used for pest control when they sell their products. This, potentially very large, use is limited by the myopesticide's slower action and higher price.

Slower action is not a major problem when *Metarhizium* is used in preventive control programs. The higher price is, in part, a consequence of its narrow host range. *M. a. var. acridum* is specific to locusts and grasshoppers, so research, development and registration costs must be divided amongst 1 or 2 users, not dozens, as is common with chemical pesticides.

Yet one of the greatest problems is the sporadic nature of locust and grasshopper outbreaks, which results in an intermittent need for *Metarhizium* and a lack of ready availability when outbreaks do occur. Use of *Metarhizium* in a number of countries, including those that have more regular outbreaks, may be one way of ensuring high-volume production at a reasonable price. Only then will there be a regular operational use to prove that this mycoinsecticide works under a wide variety of environmental conditions, so that it can take its place as part of the integrated pest management of locusts and grasshoppers throughout the world.

Key words

mycopesticides, *Metarhizium*, locusts, grasshoppers

Introduction

Locusts and grasshoppers are major pests of agriculture that can cause substantial damage to pastures and crops. With the Australian plague locust, *Chortoicetes terminifera* (Walker), Wright (1986) reported that locust control during the 1984 plague limited damage to A\$5 million instead of the A\$103 million (1984 Australian dollars throughout) that would have occurred without control. Control costs of A\$3.4 million saved an estimated A\$98 million, giving a benefit:cost ratio of about 29:1. Damage and measurable benefits of control in dollar terms may be less in countries with subsistence

farming, but outbreaks of a number of species often follow drought and crop failure. Food aid is often required during drought and when rains do come and a good crop promises adequate food supplies, farmers hardly appreciate the crop being eaten by locusts or grasshoppers. Consequently, there is usually the political will to control locust outbreaks, particularly when faced with highly visible bands and swarms.

However, control of locusts has relied on the use of large amounts of chemical pesticides. The widespread use of chemicals to control plagues of grasshoppers in North America and of locusts in Africa during the 1980's, led to the decision to provide funding to investigate biological alternatives to chemicals. A variety of potential biological control agents has been investigated in recent years and the reasons why *Metarhizium anisopliae* var. *acridum* (Driver and Milner) was chosen for study in a number of countries will be outlined, including the progress to the development of 2 commercial products Green Muscle® in Africa and Green Guard® in Australia. A number of factors currently limiting operational use will be discussed, as will possible strategies to overcome these limitations, so that use of *M. a. var. acridum* can be expanded to become a significant part of locust and grasshopper management worldwide.

Locusts as a target for biological control agents

Locusts are highly mobile pests, and most have the ability to migrate hundreds of kilometers. Long-distance migration means that a locust problem that begins in one region often suddenly migrates to another region, or even another country, hundreds or thousands of kilometers distant. As a result, effective management requires preventive control of locusts in one region or country to protect crops of another. Such treatments need to begin early in outbreaks and require coordination on a national or international scale. Where countries are large enough in geographical area for a locust problem to be contained within their borders, a national organisation is sufficient, but often the problems are so large that international organisations are required.

Preventive control also requires a good forecasting and survey system that allows outbreaks to be found early, and there must be a range of proven, readily available products and techniques in place, to control outbreaks as soon as they are found. Having an environmentally friendly option is important so that locusts in environmentally sensitive areas can be treated without having to resort to chemical pesticides. Worldwide there is a general increase in constraints on chemical insecticide use, but in Australia, there is the additional limitation of significant parts of locust-source areas in the interior now producing organic beef for the Japanese market. Without a biological alternative, locusts on organic properties or

in environmentally sensitive areas in Australia would have to be left untreated, substantially reducing the effectiveness of preventive control programs.

The search for potential biological control agents for locusts

The most commonly used and widely sold biological control agent in the world is *Bacillus thuringiensis* (Berliner), but this agent has not been effective against locusts and grasshoppers (Zelazny *et al.* 1997). In the search for a biological for locusts, it was clear that, most likely, a successful agent would, like *B. thuringiensis*, need to be produced *in vitro* in order to ensure the production of large quantities at a reasonable price. *Paranosema locustae* (Canning) was already available, but was a poor candidate: it caused low mortality at field doses and had high production costs because of its *in vitro* production within locusts and grasshoppers.

A major breakthrough was the finding that fungi like *Beauveria* and *Metarhizium* had much greater efficacy if applied in oil compared to applications in water (Prior *et al.* 1988, Bateman *et al.* 1993), and this advantage was ideally suited for use in locust control where nonwater-based ULV applications are common. *Beauveria bassiana* (Balsamo) Vuillemin had initial success in field tests in Canada (Johnson & Goettel 1993), but subsequent field applications were often unsuccessful when weather conditions allowed grasshoppers to increase their body temperatures to high enough levels to effectively stop development of the fungus (Inglis *et al.* 1996, 1997).

Following the desert locust outbreak in Africa during the 1980's, Canada and a number of countries in Europe funded the LUBILOSA (LUtte Biologique contre les LOcustes et les SAuteriaux) program, a long term project to find a biological alternative to chemicals. The LUBILOSA program demonstrated that *M. a. var. acridum* killed locusts and grasshoppers in a wide variety of conditions, including in deserts (Langewald *et al.* 1997). This initial success led to similar programs in a number of other countries, including Australia, Brazil, Madagascar and Mexico, and has led to the development of the commercial products Green Muscle®, used against locusts and grasshoppers in Africa, and Green Guard® used in Australasia.

In Africa, Green Muscle® has been tested in many field trials and registered for use in a number of countries, but as yet has had limited operational use, even during the 2003-2005 desert locust outbreak. On the other hand, in Australia, Green Guard® has been used operationally since 2000, when over 20,000 ha of bands of the Australian plague locust, *C. terminifera*, were treated. Operational use has continued when outbreaks occurred: 15,000 ha were treated during the 2003-04 season and 35,000 ha treated during 2004-05. The factors that led to the rapid development of *Metarhizium* to operational use in Australia will be examined, along with progress in other regions in moving to similar operational use.

Factors contributing to the use of *Metarhizium* as part of locust control in Australia

For some years, locust control in Australia has involved a strategy of preventive control, but this program initially relied entirely on the use of chemical pesticides (Hunter 2004). While constraints on the use of chemicals had been increasing for some time, particularly with the broad-acre spraying required to control large infestations of locusts, it was the switch, during the mid 1990's, to production of organic beef for Asian markets, that spurred an intense search for a biological alternative. Many properties in the locust-source areas of western Queensland and northern South Australia became

organic: chemicals could no longer be used in a significant locust-source area, which threatened the continuing success of preventive control. But these locust-source areas were in pastoral regions far from crops (Hunter 2004), so the locusts could readily be treated with a biological, because the immediate mortality that is desirable when pests are already damaging crops is not required. By the mid 1990s, it was clear that the most promising biological control agent was the fungus *M. a. var. acridum* that had been developed by LUBILOSA in Africa and then by CSIRO and the APLC in Australia (Lomer *et al.* 1993, Hooper *et al.* 1995, Langewald *et al.* 1997).

While research was important in demonstrating the initial potential of *M. a. var. acridum*, it was the Locust and Grasshopper Biocontrol Committee (LGBC) that rapidly brought this control agent to commercial reality in Australia. The LGBC was formed in 1997, and moved *M. a. var. acridum* from scientific promise to operational use by the year 2000, when over 20,000 ha of locust bands were treated. The LGBC consisted of research providers, a commercial partner and end users (Australian Plague Locust Commission, Departments of Agriculture and a farmer organisation), who pooled their financial resources and technical expertise to rapidly advance the development of a commercial product, Green Guard®.

A major use was to be on properties producing organic beef, so the oils used with Green Guard® were chosen following consultations with the National Association for Sustainable Agriculture Australia (NASAA), an organic farm-certifying organisation. A thicker oil was required to reduce settling during transport, so the spores were mixed in corn oil as Green Guard® ULV concentrate; but the concentrate had to be diluted in a thinner oil prior to application, leading to the choice of Summer Spray Oil® as a diluent. NASAA advised that both oils were suitable for use on certified organic properties.

This type of close liaison between researchers, the commercial partner and end users, occurred at each stage of development of Green Guard®, and continues today. Research and development is clearly focussed on producing the product that each end user wants, formulated as they want and controlling locusts or grasshoppers effectively in their respective situations.

But critical to the success of the LGBC was that an important end user, the Australian Plague Locust Commission, agreed to purchase about A\$200,000 worth of Green Guard® per year for 3 y. The agreed purchase contract led to the commercial partner spending significant amounts of money in improving production, leading to higher and higher yields of a consistently high-quality product. Higher yields mean lower costs of production and yields have improved in recent years, such that 120 g of spores are produced per kg of rice substrate, much higher than reported by manufacturers of other isolates such as the LUBILOSA isolate in Africa (Cherry *et al.* 1999) and the isolates in Mexico (Cepeda-Puente *et al.* 2005).

Contributing to the success of Green Guard® are a number of biological characteristics of the FI-985 isolate, the active ingredient in this product. In addition to higher yield, laboratory tests have shown that FI-985 survives longer in ultraviolet light than other common isolates of *M. a. var. acridum* (Fargues *et al.* 1996): an important factor in increasing survival time in the field. Scanlan *et al.* (2001) demonstrated the critical importance of secondary pickup of spores from the vegetation: at lower, more affordable doses, many of the locusts in a treated area do not pick up sufficient spores during spraying, but do pick up additional spores from the vegetation over the next day or so. Isolates more susceptible to UV may not survive long enough in the field to be picked up and may require a higher initial dose, increasing costs.

But it is the ability to survive under high temperatures in the field

that was a major contributor to the uptake of *M. a. var. acridum* in Australia. Locust treatments often occur during midsummer and on the organic chemical-free properties in the locust source areas of the arid interior, maximum temperatures during summer are commonly above 40°C. While the FI-985 strain used in Green Guard® does develop at slightly higher temperatures than some other isolates of *Metarhizium* (Milner *et al.* 2003), even FI-985 does not develop at much above 36°C. In spite of a lack of development in the laboratory at high temperatures, mortality of locusts treated with Green Guard® is most rapid when it is very hot, with >90-95% mortality being reached in 7 to 9 d. On very hot days, *Metarhizium* development is inhibited by high locust body temperatures in the middle of the day, but temperatures are in the 23-33°C range for rapid *Metarhizium* development all night, as well as early and late in the day, leading to rapid mortality. The 7 to 9-d mortality when using *Metarhizium* where it is very hot, is acceptable with the preventive control programs against nymphs of the Australian plague locust: even fipronil applied at wider-track spacings outside cropping areas takes a number of days to kill nymphal bands.

As a result of this high, moderately rapid, mortality at high temperatures, a major use of Green Guard® has been on organic properties in locust source areas in the interior. A second important use has been in environmentally sensitive areas, such as near waterways or where there are rare and endangered species. But an increasingly important use is by individual landholders who want to limit the amount of chemicals used on their products. In Australia, as in a number of other countries, landholders selling products must present a Vendor Declaration that lists chemicals used for pest control on their products: more and more landholders prefer the use of Green Guard® because of a perceived lower price received for products on which chemical use has been declared.

Factors currently limiting the use of *Metarhizium* for locust control

(i) *Slow action in mild to cool conditions.*—The use of Green Guard® as part of preventive control and as a way of avoiding having to declare chemical use in their products, is potentially very large, but is limited by its slower action, particularly in the agricultural zone during mild spring temperatures (maxima 20 to 30°C). Locust mortality then takes 10 to 14 d, or about 2 nymphal instars. To ensure mortality occurs before the flying adult stage, the main use during spring has been against mid to late-instar bands in pastures. While treatment of last-instar nymphs sometimes occurs, there have been only a few treatments of bands in crops or of adult swarms anywhere near crops, because of the damage that could be caused during the 2 w the locusts would take to die. However, spraying of adults is possible, with locust species that have a long, often diapausing, adult stage during the dry noncrop season. This occurs with the spur-throated locust *Austracris guttulosa* (Walker) in Australia, *Schistocerca piceifrons* (Walker) in Latin America and *Nomadacris septemfasciata* (Audinet-Serville) in Africa, and could be particularly important with the latter species which is common in areas subject to flooding.

(ii) *Significant numbers survive when used against dense infestations.*—Control operators and landholders are used to the >99% mortality common with chemical pesticides. Mortality of 95 to >99% can occur after application of *M. a. var. acridum*, especially when it is very hot; but mortality of 90% or even 80-85% is more common, which is acceptable when pest densities are moderate (10 to 20 m⁻²),

as then only a few locusts m⁻² survive. But when very dense bands of several thousand m⁻² cover a significant percentage (*eg.*, >5%) of the area, a 90% decline could still leave sufficient numbers to form an adult swarm of 10 to 20 m⁻². Consequently, while a 90% reduction is a significant contribution to preventive control away from crops, such a reduction is often not considered to be sufficient with dense infestations near crops.

(iii) *Higher price for the product.*—Higher price can be a major problem limiting the use of biologicals. For Green Guard®, the price difference has been reduced substantially by the higher yield of the FI-985 isolate during production. Costs could be further reduced by locating production in developing countries. But a significant contributor to the higher cost is a direct consequence of its specificity: research, development and registration costs must be divided amongst 1 or 2 users, not dozens, as is common with chemical pesticides. And the requirements for registration are becoming so costly in some countries that it is just not economically viable to register such a specific product for a sporadic pest like locusts and grasshoppers.

It is a fallacy, albeit a common one, to compare costs of control based solely on the cost of the product. With most locust control programs, the cost of staff, administration, survey, search and aerial application is much more than the cost of the product. The real cost per hectare is actually the total budget for the control program divided by the number of hectares treated; so even with a product like Green Guard®, costing 2 to 3 times as much as some of the older-generation chemicals, when total costs are factored in, the cost difference is actually < 2:1.

And for preventive control to be successful, cost alone cannot be the major factor that determines if treatment occurs. With the Australian plague locust, the cost of treating swarms is high, because helicopters, which are expensive, are used to locate and mark the swarms. Bands cost much less to treat, especially when bands are treated with chemicals like fipronil, that can be applied at wide-track spacings of 300-500 m. But the much higher cost of treating swarms does not mean that they are not treated: no matter how successful band treatments are, swarms almost always form and these must be treated if preventive control is to succeed.

Similarly, even though treatment with Green Guard® is more expensive than treating either bands or swarms with chemicals, Green Guard® still needs to be applied in areas where chemicals cannot be used to ensure a successful control program. Besides, even if biologicals are slightly more expensive, it is actually cheaper to use a moderate amount of a biological early, as part of preventive control, than to wait and use a much larger amount of chemical a generation or two later, when locust numbers have increased substantially.

There are ways of reducing costs of locust control: in northern Mexico, aircraft and chemical costs have been reduced dramatically by applying fipronil at 100-m intervals instead of the 20-m intervals used with other chemicals in the past (Barrientos *et al.* 2005). Similarly, costs of applying *M. a. var. acridum* can be reduced by applying at the 100-m track spacings, from an aircraft flying at a height of 10 m, as used by the APLC when applying Green Guard®.

(iv) *Sporadic nature of outbreaks leading to problems of supply.*—Yet one of the greatest problems for the sustained use of *M. a. var. acridum* is a direct result of one of its greatest virtues: its specificity in only attacking locusts and grasshoppers. The specificity to a pest with

sporadic outbreaks, means that there is only an intermittent need for the product in a given region. Consequently, even though a number of countries in Latin America, Asia and Africa have tested local isolates against locusts or grasshoppers, outbreaks have rarely lasted long enough to lead to the development of a readily available commercial product. In Brazil, which is a large user of biologicals against a number of pests (Magalhaes & de Faria 2005), a local isolate, CG423, was successfully tested against *Rhammatocerus schistocercoides* (Rehn) during the late 1990s (Magalhaes *et al.* 2000); but the subsequent collapse of outbreaks has meant no further progress. In Peru, production of Brazilian *M. a. var. acridum* was included as part of an FAO program to control an outbreak of *Schistocerca piceifrons peruviana* (Walker) during 2000-01, but the outbreak collapsed before the product could be used operationally (Solano-Morales 2005). In Madagascar, local isolates were tested against the migratory locust, and in Indonesia a local isolate and the Australian isolate (Sudarsono & Hunter, unpub. data) were tested, but not used subsequently, following the collapse of the outbreaks. However, a recent increase in locust numbers in Indonesia during the past year may lead to the opportunity for further tests there.

In Mexico, *M. a. var. acridum* has caused high mortality of *Schistocerca piceifrons piceifrons* in field trials (Hernandez-Velasquez *et al.* 2003). Local commercial production has followed, but the switch from production in the laboratory to production on a commercial scale, has resulted in variable quality, low yields and a high price, so that little has been used operationally. In Africa, there have been many successful trials of Green Muscle® against a wide variety of locust and grasshopper pests. However, no regular market has been developed, and delays in funding, combined with no dedicated production and no proven operational use, led to Green Muscle® being unable to take full advantage of the 2003-2005 desert locust outbreak. The first swarms were seen in December 2003, but the first moderate scale (1400 ha) use of Green Muscle® did not occur until June 2005, by which time the outbreak was already in decline.

The overall trend with *M. a. var. acridum* is that, while local isolates have been tested in many regions, the sporadic nature of outbreaks has limited the development and use of commercial products. Sporadic outbreaks in a given region mean there is little incentive for companies to have systems geared to specifically produce local *M. a. var. acridum*. Production systems are set up for other products and may not be naturally amenable to obtaining the highest possible yields of *M. a. var. acridum*. Lower yields and costs associated with switching from one product to another result in a higher price.

A second problem with the irregular requirement for product is that when there is an outbreak, by the time funds are made available, a decision made to order product, and then it is produced, many months will have passed and significant opportunities for preventive control will have been missed. As well, many locust species are common near waterways or other environmentally sensitive areas. Locusts in such areas are likely to be treated and if a biological is not readily available, chemicals will be used to ensure locusts do not escape and reach crops, particularly in developing countries where food security is of paramount importance. Consequently, local political or regulatory authorities that insist on the use of local isolates of *M. a. var. acridum* often will inadvertently ensure the continued substantial side effects of using of chemicals near waterways and other environmentally sensitive areas in their regions.

Ready supply through production for regions with regular outbreaks

Lomer *et al.* (1999) suggested that to ensure sustainability and a readily available supply of Green Muscle®, markets needed to be developed against locusts and grasshoppers that have regular outbreaks. Becker Underwood has taken up this suggestion for its product Green Guard®: it has been developing markets in countries that have a chronic locust and grasshopper problem. To ensure food security for its large population, China engages in preventive control of locusts and grasshoppers: 300,000 to 1,000,000 ha are treated per year, mainly in pastures before the insects reach crops.

The oriental migratory locust (*Locusta migratoria manilensis*) (Meyen) and some grasshopper species in China, are common near water, so a biological agent (*Paranosema locustae*) has been used in recent years; this is in spite of the fact that there is often only a 30 to 60% reduction in the first month after treatment, although higher reductions are sometimes evident in subsequent generations (Zhang *et al.* 1998, 2001). In trials with Green Guard® in 4 Chinese provinces over the past 3 y, low to moderate doses (25 to 50 g ha⁻¹ of spores) resulted in 76 to 97% mortality of migratory locusts in 8 to 11 d, and in one case, 14 d. Green Guard® treatments were found not to affect the numbers of several common locust natural enemies: the generalist robber flies (Diptera: Asilidae) (*Ommatius* spp., *Promachus* spp) and Bombyliid flies (*Anastoechus chinensis*). At a site treated at 50 g ha⁻¹, there were 0.85 ± 0.15 m⁻² natural enemies at treatment and 0.80 ± 0.23 m⁻² 11 d later. At a site treated at 125 g ha⁻¹, there were 0.84 ± 0.16 m⁻² natural enemies at treatment and 1.00 ± 0.19 m⁻² 11 d later. The demonstrated efficacy of Green Guard® and the lack of effect on natural enemies, led the China Ministry of Agriculture to apply Green Guard® against locust and grasshopper infestations during June 2005. In parallel with these applications, moves are being made to register Green Guard® in China, which will lead to production and supply for this substantial regular market.

The supply of *M. a. var. acridum* to countries with irregular outbreaks

To ensure that *M. a. var. acridum* is continuously available in the Australasian region, the aim is to have one or more factories that will regularly produce Green Guard® and will be able to supply product at short notice to both countries with regular outbreaks (China, and to a lesser extent Australia), as well as to countries whose outbreaks are sporadic. With Green Muscle®, a similar mix of uses between species with regular and sporadic outbreaks is being tested.

When production for regular markets is fully in place, commercial products will be in a position to supply Australia, Asia and Africa. North and South America have the problem of not having at least one species that has large regular outbreaks. There is some production of a local isolate in Mexico, but the normal annual requirement for enough *M. a. var. acridum* to treat a few thousands of hectares is hardly sufficient to lead to commercial production large enough to address a large outbreak when it does occur. And there may not even be a ready market for the Mexican isolate in the USA and Canada when outbreaks occur there: current attitudes of regulatory authorities suggest that even the Mexican isolate will be considered foreign and would have to go through many regulatory hurdles before being used. If such attitudes had been held in Africa, Green Muscle®, based on an isolate from Niger, would not have been used in neighboring countries, let alone countries in the south of

the continent, like South Africa (Price *et al.* 1997) and Tanzania.

Overall, the use of products containing *M. a. var. acridum* is still in its infancy. Even in Australia, where operational use of Green Guard® has generally proven successful, further widespread use and testing is required. Only as it is applied operationally against various species and its efficacy monitored carefully in different environmental conditions will the full benefits of *M. a. var. acridum* in the preventive control and integrated pest management of locusts and grasshoppers be realised.

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