

CONTROLLING BROWN LOCUST HOPPER BANDS IN SOUTH AFRICA WITH A MYCO-INSECTICIDE SPRAY

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ABSTRACT

Serious outbreaks of the Brown locust, *Locustana pardalina* regularly occur in the semi-arid Karoo region of South Africa. Current preventive control strategies require the extensive use of broad spectrum contact insecticides to control locust outbreaks. However increasing pressure from land holders and conservation bodies, concerned about the high use of conventional chemicals and their possible negative impact on the Karoo ecosystem, has created an urgent need for examining alternative non-chemical and environmentally safer methods of control.

Following initial screening tests, an oil-based ULV formulation of the entomopathogenic fungus, *Metarhizium flavoviride* (isolate IMI 330189), was applied at a rate of $3.7-4.7 \times 10^{12}$ conidia/l using a hand-held spinning disc sprayer to treat 4th-5th instar hopper bands in the Hanover District of Cape Province during a brown locust outbreak.

First mortality from fungal infection was recorded at 3 days and this rose to >95% after 14 days for samples taken 0 and 24 hours from 4 out of 5 bands treated with the myco-insecticide. Median time to kill averaged 10.3 days. Poorer control was achieved in one band due to application problems and mortality was lower (82-95%) in samples taken 72 hours after treatment. Further trials are planned for the 1994-95 season.

INTRODUCTION

Four species of locusts may be the subject of control operations in southern African region: the brown locust *Locustana pardalina* (Walker), the African migratory locust *Locusta migratoria migratorioides* (Reiche & Fairmaire), the southern African desert locust *Schistocerca gregaria flaviventris* (Burmeister) and the red locust *Nomadacris septemfasciata* (Serville).

Of these the brown locust is the most important for southern Africa. Its short life cycle with 3-4 generations per year, a drought resistant egg stage, intense phase polymorphism and an ability to swarm simultaneously over large areas makes it a formidable pest problem. Repeated outbreaks are the rule; in only five out of the past 45 years has no control been required in the Karoo. Endemic to the semi-arid areas of South Africa (especially the Karoooid parts), Namibia and Botswana (Faure & Marais, 1937) the area subject to its invasions extends up to 16°S latitude and encompasses nine southern African countries (Lea, 1964).

Current preventive control strategies involve the extensive use of broad spectrum insecticides to control brown locust outbreak populations in the Karoo before they can migrate to the principal grain producing areas. However, Karoo landholders and conservation bodies are concerned about the high use of conventional chemicals (e.g.

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58,000 bands sprayed in the past season) and their possible negative impact on the unique Karoo ecosystem.

The Locust Research Unit of the Plant Protection Research Institute (PPRI) of South Africa has evaluated new and promising compounds for effective locust control for many years, using laboratory bioassays followed by field trials of selected compounds. Entomopathogenic fungi, particularly the deuteromycetes are especially attractive for development as biological control agents of acridids (Prior and Greathead, 1989). In response to a request by the PPRI, *Metarhizium flavoviride* Gams & Rozsypal inoculum was supplied by IIBC and is currently under investigation in a collaboration between PPRI and the LUBILOSA programme*.

The approach taken by LUBILOSA has been to formulate the aerial conidia of these fungi in vegetable or mineral oils, thereby increasing their efficacy and decreasing reliance on conditions of high humidity (Bateman *et al.* 1993). They have a contact action, which allows direct penetration of the host cuticle without ingestion. Being oil-based these formulations are suitable for application with the ultra-low volume (ULV) equipment normally in use by locust control organizations. Biopesticides based on these fungi could be produced in many countries, using plant with a level of technology equivalent to that of a brewery. This paper describes the laboratory testing in Pretoria and the first field trial conducted with the myco-insecticide against gregarious bands of brown locust nymphs. This was carried out on the Nuwefontein Farm (30° 56'S, 24° 11'E), Hanover District, Cape Province, South Africa in the Karoo outbreak region of this species of locust.

MATERIALS AND METHODS

Laboratory bioassay

A strain of *Metarhizium flavoviride* (IMI 330189ss) isolated from a grasshopper, *Ornithacris cavroisi* (Finot), in 1988 in Niamey, Niger was selected by LUBILOSA as a "standard isolate" for laboratory and field research, due to its virulence against African locusts. Imported under quarantine into South Africa it was first assayed in 1993 against fifth instar brown locust hoppers and adults in Pretoria. Successfully cultured in the PPRI laboratories on potato dextrose agar, conidia were harvested and the spores filtered and separated by centrifuge. Spore counts were made by means of a haemocytometer and diluted with groundnut oil to the concentrations needed for bioassay.

Individual drops, containing 3.0×10^4 and 6.0×10^4 conidia were applied to the neck membrane of individual insects ($n=30$), using an electrically driven micro-applicator. Controls were treated with oil only. Following inoculation, locusts were kept in a quarantine glasshouse, supplied with green feed, bran and water, and their mortalities recorded over the next 21 days. Air temperature and humidity were constantly monitored throughout this period.

Field Trials

Field trials were undertaken against 4th-5th instar gregarious bands on Nuwefontein farm from 17th-21st February 1994. Following good rains, a full-blown locust outbreak was in progress in the central Karoo at the time and numerous bands were present

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throughout the Hanover District. These were migrating across undulating, open country where the vegetation comprised typical mixed Karoo veld, with scattered low Karoo-bushes (e.g. *Pentzia*, *Chrysocoma*, *Lycium* etc.) and knee high grasses (e.g. *Eragrostis*, *Stipagrostis*) present. Vegetation was green at the time and cover was estimated at approximately 30%.

The spray formulation was made up on site as follows: dried conidia were suspended in a mixture of 30% groundnut oil and 70% paraffin ('Jet A1'). Separate batches of formulation were prepared for each occasion; the measured spore concentrations were 3.7×10^{12} and 4.7×10^{12} conidia/litre respectively. Approximately 70% of the conidia germinated after 24 hours of incubation on potato dextrose agar plates.

A Micron 'Ulva plus' spinning cup sprayer, fitted with an orange restrictor was used to apply the formulations. This produced an emission rate of approximately 50 ml/min, which with a track spacing of 3m and a walking speed of 1.4 m/s, gave a localised volume application rate equivalent to 2 litres/hectare. With 5 alkaline batteries inserted the rotational speed was 7000 RPM. A VMD of $68 \mu\text{m}$ was measured (using a Malvern 2600 particle size analyser) when this equipment was assessed in the laboratory with a similar blank formulation.

Nymphal bands, identified for treatment, were sprayed during the early morning (06:45-09:00) while still densely aggregated on their overnight roosts. Target bands varied in size, the largest being about 0.3 hectares and probably comprised approximately 2.5×10^5 individuals. Five bands were treated with the myco-insecticide and two control bands were sprayed with the formulating oil only. Spray plots were equivalent to operational spot treatments and were normally approximately 30 x 30 m (covered by 10 swaths). Air temperature, soil surface temperature and relative humidity during spray application and during the post application period were continuously logged.

Immediately after spraying, a portion of each band was corralled by placing out an open-topped 1.5 x 1.5 m enclosure ("boma") consisting of four 0.3 m high, smooth-sided, plastic walls mounted on metal frames. Samples of hoppers (Table 1) were subsequently transferred from each enclosure into 250 x 250 x 440 mm metal gauze and galvanized steel cages after 0, 24 and 72 hours post application, for further evaluation. After being transported back to Pretoria the cages of infected hoppers were maintained outdoors for daily inspection.

RESULTS

Laboratory bioassays

Doses of 3 and 6×10^4 spores killed 90-93% of brown locust hoppers after 21 days with median lethal times (MLT) of 9.8 and 7.5 days respectively. First mortality occurred after 3-4 days. Air temperature in the quarantine glasshouse over the entire period averaged 27.2°C (range $24-34^\circ\text{C}$) with 19% R.H. (range 13-23%). The same doses assayed against adult brown locusts gave 100% kill after 12 and 9 days (MLT = 10.1 and 4.4 days) respectively. Here the mean air temperature was 28°C (range $24-34^\circ\text{C}$) with 40% R.H. (range 28-52%).

Both life stages assumed a characteristic bright red coloration just before death. This was also unusual in that locusts died suddenly *in situ*, either still clinging to the sides of the cage or to the vegetation. Apart from reduced feeding activity, there were few outward signs of infection making early diagnosis of diseased hoppers difficult.

Following these encouraging results we evaluated the myco-insecticide in the Karoo. Since band control, especially of the two final "red coat" nymphal instars, is the main

method of control employed by the locust organisation in the Karoo, we targeted this stage for our field tests. It is also more tolerant than the adults to chemical control.

Field Results

Maximum air and soil surface temperatures over the four days reached 28.1° and 44.6° C respectively (Figure 1); humidities were generally high at night and in the early morning, but low in the afternoon, falling to a minimum of 24.5%.

All bands treated with the formulation showed a higher rate of mortality than the controls with >95% mortality after 14 days for caged hoppers taken from four of the bands, 24 hours after treatment (Figure 2). Caged samples maintained in Pretoria experienced temperatures ranging from 14-26° and 60-90% r.h.. Low efficacy in band 2 can almost certainly be attributed to poor application due to very little wind (see Table 1). The bushes were denser in this plot than in most of the others and it is possible that little spray drifted onto the sampling zone. Disturbance during the laying out of the plot, which was unduly long in this case, could also have caused hoppers to retreat within the shelter of the bushes.

TABLE 1. Measured spraying conditions, Median Lethal Times and hoppers sampled

Band no.	Treatment	Time	Wind Speeds (m/s)	Temp. r.h.		MLT: days (sample size, n=)		
				°C	%	0 h.	24 h.	72 h.
1	Control	06.50	0.8 S, variable	18.5	92	- (63)	- (70)	- (42)
2	Fungus	07.35	(0) E, very poor	22	80	- (85)	17 (66)	- (81)
3	Fungus	07.50	0.8 E, good	22.5	57	8 (176)	9 (42)	10 (64)
4	Fungus	08.10	1.7 E	22	80	10 (84)	10 (61)	10 (72)
4b	Fungus					11 (53)	10 (44)	12 (46)
5	Control	07.15	2.1 E	19	80	- (75)	- (67)	- (59)
6	Fungus	07.30	2.1 E	19	80	15 (89)	10 (39)	8 (61)

NB: MLT values not assigned if mortality is <50%

FIGURE 1 Meteorological data recorded during the myco-insecticide trial on Nuwefontein Farm.

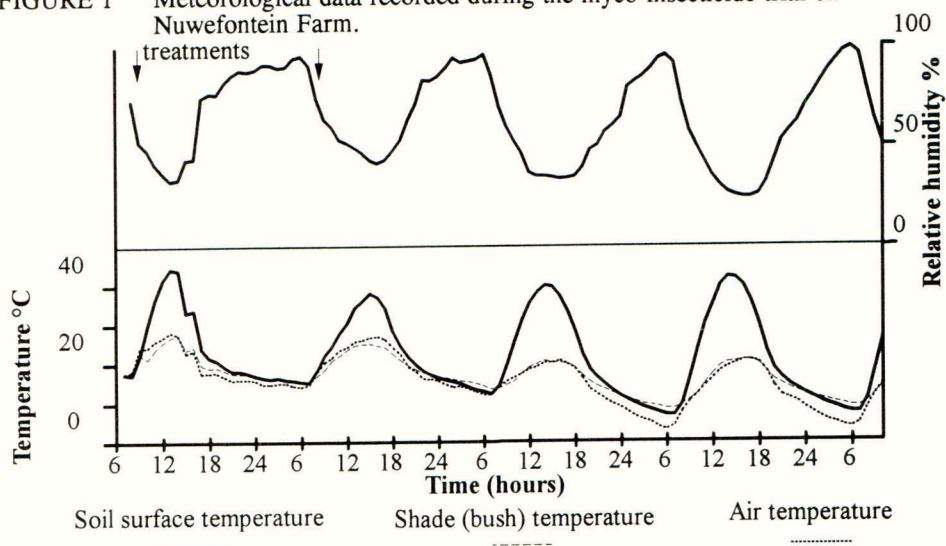


FIGURE 2a Brown locust mortality after treatments on 17/2/1994

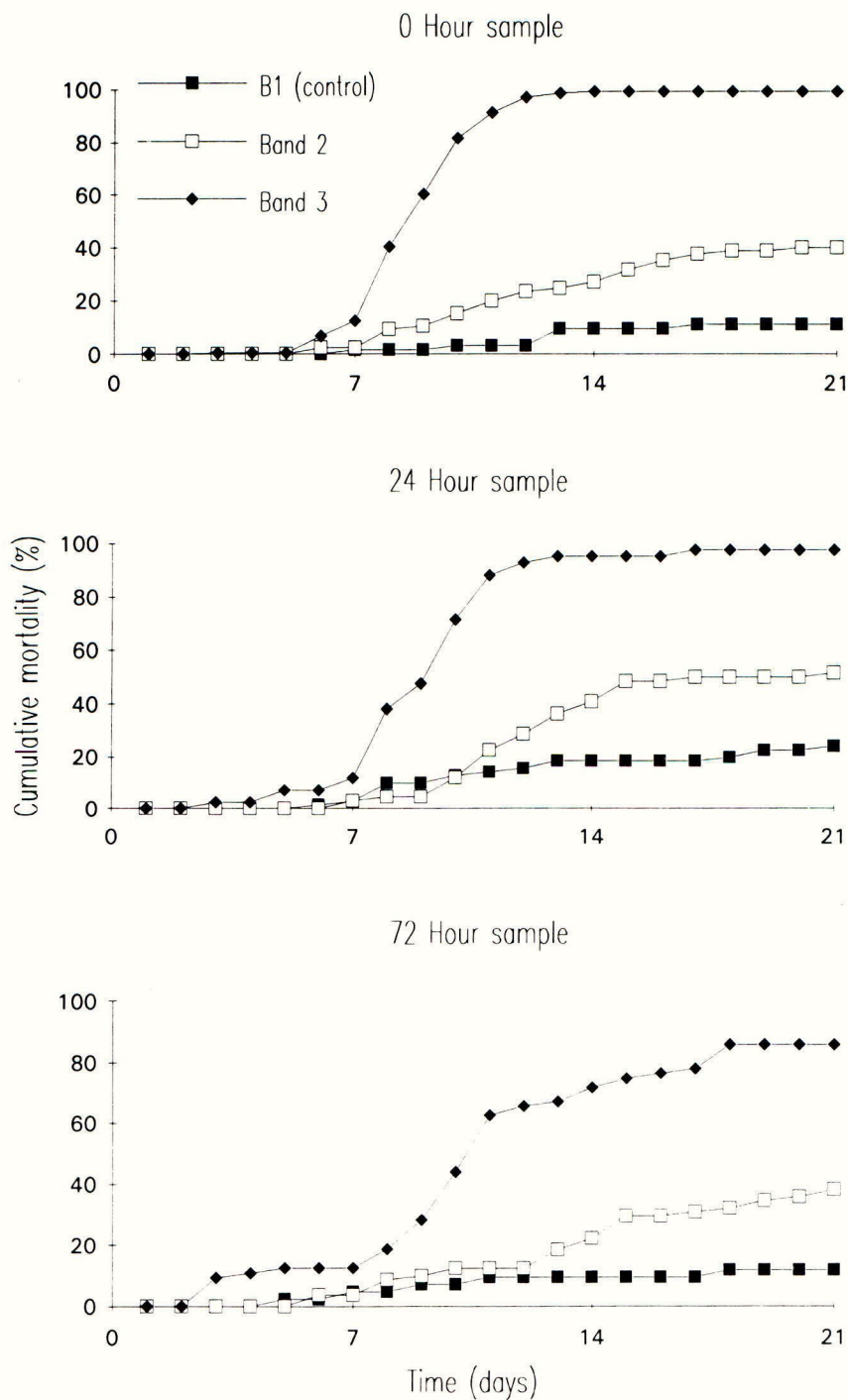
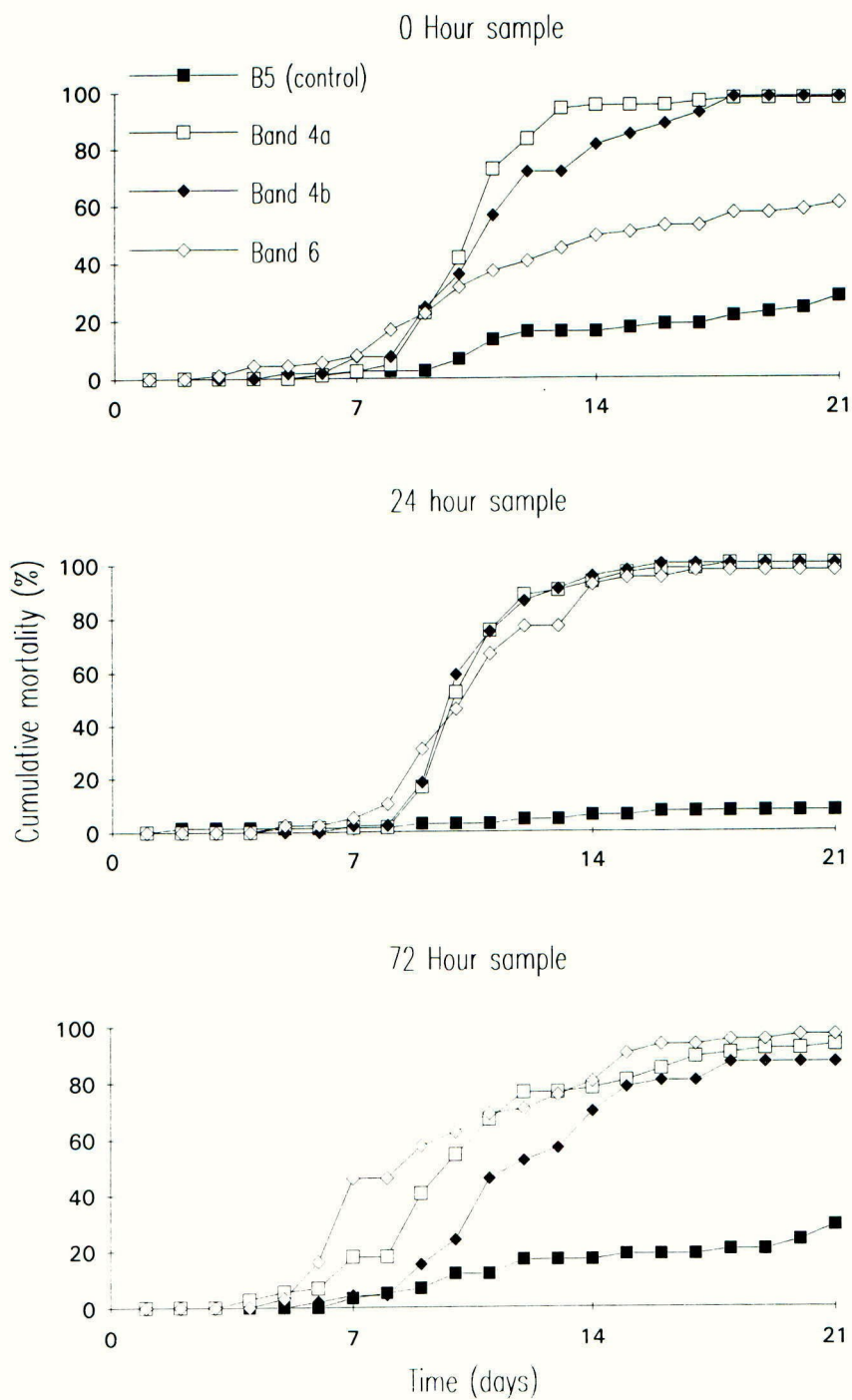


FIGURE 2b Brown locust mortality after treatments on 18/2/1994



Application conditions were better for bands 4-6 and samples taken after 24h died faster than those taken at 0h; this was especially apparent in band 6. This could be due to secondary pick-up of the conidia. ULV locust spraying trials with pyrethroids have shown that 5th instar hoppers may acquire a further 1.5-3.5 times active ingredient during the two hours after initial contact with the spray (H.D. Brown, unpublished). Nguyen (1980) similarly measured a linear increase in acquisition of fenitrothion by *Chortoicetes terminifera* (Walker), during the first two hours after application. Although mortality commenced earlier in samples taken after 72h, the final mortality level was lower than in the 24h sample. Some individuals may be able to overcome the disease by prolonged exposure to full sunlight.

DISCUSSION

An array of insect enemies is associated with the brown locust in the Karoo (Greathead, 1963). Although some of these have claimed to exercise high levels of control in the past, this has not been validated during recent plague cycles. Thus, the sarcophagid parasite of the active stages, *Wohlfahrtia pachytyli* Townsend, regarded as important in the 1920s (Potgieter, 1929), has been found in recent unpublished work by PPRI to attain a maximum of only 6% parasitism; while the *Systoechus* egg predator (Bombyliidae) destroyed no more than 10% of egg pods in egg beds. Other than a single record of a bacterium, *Serratia marcescens* Bizio, found on hoppers, we have little evidence of natural control attributable to pathogens in the Karoo. Stragglers from bands are very rare.

This trial was not the first attempt at manipulating fungi for locust control in South Africa. There are several old records of natural fungal epizootics decimating swarms of locusts in South Africa (Pole Evans, 1911; Potgieter, 1929). These all refer to the entomophthoralean fungus, *Entomophaga grylli* (Fresenius) (= *Empusa grylli*), which was regularly associated with red locust invasions of the warmer and wetter parts of South Africa (and Namibia) at the beginning of the century. Because these fungi are wholly dependant on high humidity, attempts to artificially induce epizootics by their release have not met with success. The famous "South African Locust Fungus" was commercially distributed in Grahamstown in 1899, but instead of *E. grylli*, the packages contained a common mucor pin-mould fungus (Skaife, 1925). This sealed the fate of any further fungal work at the time and it is still not feasible to commercially mass produce products based on the entomophthorales.

With mortality levels of >95%, *M. flavoviride* affords the opportunity of killing a large number of locusts for the first time by means of an introduced pathogen. Such final levels of kill are as good as those achieved by any of the conventional insecticides in current use in southern Africa, and are comparable with other myco-insecticide field tests with acridids carried out in western Africa (e.g. Lomer *et al.*, 1993). They were achieved in spite of high levels of ultra violet radiation experienced in the Karoo and an apparent slow rate of kill for fifth instar brown locust hoppers in the laboratory; similar topical applications of $6.5-7.4 \times 10^4$ conidia to fifth instar nymphs of *S. gregaria* gave MLTs of 3.9-4.7 days (Prior *et al.*, in press). The time taken to kill locusts is of course a disadvantage, but reduced feeding behaviour is an additional symptom of mycoinsecticide treatment. Thus feeding in *S. gregaria* has been shown to be more than halved during the first five days of fungal infection (Moore *et al.*, 1992).

The trial described here was preceded by good rains and the Karoo was particularly verdant. Although the results are encouraging, further testing under different climatic conditions is required and trials are planned for the 1994-95 season, possibly under different climatic conditions. The fate and behaviour of marching bands treated with the myco-insecticide will also be investigated.

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DISCOVERY AND INVESTIGATION OF A NOVEL NEMATODE PARASITE FOR BIOLOGICAL CONTROL OF SLUGS

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ABSTRACT

Phasmarhabditis hermaphrodita is a parasite which inhibits feeding and causes mortality in a wide range of pest species of slugs and snails, but is harmless to other invertebrates and is rapidly killed at 35°C, so is unable to survive in homoeothermic animals. It can be readily reared in culture, formulated, and stored for use as a biocontrol agent, using technology similar to that developed for entomopathogenic nematodes. It is well adapted to the relatively low temperatures at which slugs are active and troublesome as pests. When applied to moist soil, it is able to protect a range of crops from slug damage, including subterranean damage which is especially difficult to control. The dependence of the nematode on moist soil is probably less of a constraint for its use than for entomopathogenic nematodes, as the target pests, slugs, are similarly dependent on moisture.

INTRODUCTION

Slugs belonging to the families Arionidae, Limacidae and Milacidae are important molluscan pests of a wide range of crops, especially in the UK (South, 1992), but chemical control is relatively ineffective compared to that of insect pests (Henderson & Parker, 1986) and presents risks to a range of non-target organisms (South, 1992). Thus, there is a need for more effective and environmentally safe methods of preventing slug damage, which could be provided by biological control. However, few potential biological control agents for slugs are recorded in the literature and it is unlikely that any of them would meet the criteria for use as an inundative biological molluscicide. These criteria include: effectiveness against the full range of pest species of slugs; ease of production; ease of handling and storage; suitable persistence after application; and lack of harmful effects on non-target organisms. For example, the protozoan parasite *Microsporidium novacastrisensis* (Jones & Selman, 1984) probably has limited potential as a biocontrol agent because it parasitises only the field slug, *Deroceras reticulatum*, and can be reared only *in vivo*.

For this reason, a search was initiated in 1987 at Long Ashton Research Station for a biological control agent which would be active against the full range of pest species in the cool moist conditions where slugs cause most damage. In contrast to insect pests, slugs require moist conditions for survival, activity, growth and reproduction and they are adapted to relatively low temperatures. For example, the field slug, *D. reticulatum*, which is the commonest and most widespread species of pest slug in the UK and also world-wide, has an optimum temperature for growth of only 18°C and is able to grow at temperatures as low as 5°C (South, 1992). As slugs are usually most troublesome as pests in autumn and spring, when soil temperatures are typically *c.* 10°C, slugs were collected from the field and maintained in crowded conditions at 10°C to allow any disease or parasites present to be expressed at a temperature at which an effective biocontrol agent would be required to operate in the field. Early in 1988, the rhabditid nematode, *Phasmarhabditis hermaphrodita*, was discovered parasitising the field slug, *D. reticulatum*. This paper summarises laboratory investigations, mostly at 10°C, and field studies of the potential of this bacterial-feeding nematode as a biological control agent. As a result of this research, the Agricultural Genetics Company Ltd has applied for a patent covering the use of this nematode for control of mollusc pests (Wilson *et al.*, 1993c) and a commercial product was launched in spring 1994 for use in home gardens in the UK. Current research is also briefly outlined.

BIOLOGY AND MODE OF ACTION

P. hermaphrodita has previously been recorded as non-feeding dauer larvae (third stage larvae with mouth and anus closed, inside a retained second stage cuticle) living in the body cavity of slugs and snails, but later stages were not found in live slugs and snails (Maupas, 1900; Morand, 1988). This species and the closely related *P. neopapillosa*, found as dauer larvae in living slugs by Mengert (1953) and in living slugs and snails by Morand (1988), were not considered to be parasites of slugs. However, they were thought to show a degree of adaptation to life with slugs, in that dauer larvae entered the body cavity of living slugs but did not develop until after the slug died, then grew and reproduced on the decaying slug cadaver, producing dauer larvae once again when the food supply became depleted (Mengert, 1953). Dauer larvae are believed to survive in the soil ready to repeat the cycle by entering the body of slugs when the opportunity arises.

Our studies have shown that *P. hermaphrodita* is a parasite capable of killing slugs (Wilson *et al.*, 1993a,c). Dauer larvae enter the shell cavity of *D. reticulatum*, introducing bacteria on which they feed and grow to autogamous hermaphroditic adults which reproduce mainly oviparously, with eggs laid in an advanced stage of development. Infection of the shell cavity by *P. hermaphrodita* causes a characteristic swelling of the mantle and slug feeding is strongly inhibited within a few days of infection until the slug dies. In bioassays at 10°C, feeding inhibition and mortality of *D. reticulatum* have been shown to be directly related to the dose of infective larvae, with a given dose of nematodes causing greater inhibition of feeding than mortality. Most bioassays have been done using a soil-based system which was designed to bring slugs into contact with infective dauer larvae of the nematode under conditions similar to those that would occur in the field. Plastic boxes (135 x 75 x 60 mm) are filled with air-dried clay-loam soil aggregates (12 - 24 mm across). As the aggregates are introduced to the boxes, each layer is moistened with

water (30 ml water 100 g⁻¹ soil) and 10 slugs are introduced to each box when it is half-filled with soil. Boxes are left for 5 days at 10°C, then slugs are transferred individually to Petri dishes where survival and food consumption are monitored. This bioassay system has been used to demonstrate that *P. hermaphrodita* is able to kill all pest species of slugs tested, belonging to the main families containing pest species (Arionidae, Limacidae and Milacidae), as well as snails (Wilson *et al.*, 1993a,c). However, other invertebrates have not been affected by exposure to high doses of the nematodes, including earthworms (*Lumbricus terrestris*), carabid beetles (*Pterostichus melanarius*) tenebrionid larvae (*Tenebrio molitor* and *Zophobas morio*) and wax moth larvae (*Galleria mellonella*).

The optimum temperature for growth of *P. hermaphrodita* is c. 17°C and it is able to infect and kill *D. reticulatum* at temperatures as low as 5°C. Thus, it responds to temperature in a similar manner to its slug hosts. There has been a recent report of Polish strains of entomopathogenic nematodes of the genera *Steinernema* and *Heterorhabditis* killing *D. reticulatum* and *D. agreste* under laboratory conditions (Jaworska, 1993). However, tests of the ability of UK strains of *S. feltiae* and *Heterorhabditis* sp. to kill *D. reticulatum* were negative (Wilson *et al.*, 1994b) and, in any case, it is likely that the dependence of entomopathogenic nematodes on relatively high temperatures would restrict their usefulness in protecting crops from slug damage in most commercially situations. Georgis (1992) points out that successful field trials with entomopathogenic nematodes have been in the temperature range 16 - 28°C.

REARING, FORMULATION AND STORAGE

P. hermaphrodita has been cultured on nutrient-rich kidney-based media, in Petri dishes, in foam-chip cultures and in liquid cultures, using techniques similar to those developed for entomopathogenic nematodes. Initially, cultures were xenic, containing an unknown mixture of bacteria (Wilson *et al.*, 1993b). Nematodes grown in xenic cultures were shown to be able to kill slugs and prevent slug damage in the laboratory (Wilson *et al.* 1993a) and in mini-plot field experiments (Wilson *et al.*, 1994d). However, it is desirable for reliable use of the nematode as a biocontrol agent to culture it using a selected strain of bacterium. Indeed, experiments have shown that the strain of bacterium used to culture this bacterial feeding nematode has a profound influence on its growth and ability to cause mortality in slugs (Wilson *et al.*, 1994c). Interactions between the nematode, the bacterium and the host slug are clearly important, but the nature of these interactions is not fully understood. A strain of the bacterium *Moraxella osloensis* has been selected which produces high yields of infective dauer larvae which are consistently capable of inflicting mortality on all pest species of slugs tested (Wilson *et al.*, 1994c).

In current commercial production, nematodes are reared in monoxenic liquid cultures with *M. osloensis* in fermenters. Yields of infective dauer larvae are influenced by the composition of the growth medium and the environment within the fermenter, where yields in excess of 100,000 infective dauer larvae ml⁻¹ have been achieved. Infective dauer larvae are harvested from the fermentation medium by centrifugation followed by repeated washing in water. Dauer larvae can be stored in aerated water at 5°C and 10°C, but survival is relatively poor, and larvae are killed rapidly at 35°C (Wilson *et al.*, 1993b). In

commercial production, nematodes are concentrated to give between 0.1×10^6 and 2.0×10^6 nematodes g^{-1} and mixed with calcium montmorillonite clay to produce a water-dispersable friable formulation containing 0.05×10^6 to 1.8×10^6 larvae g^{-1} (wet weight), which is stored in bags of 8 μm high density polyethylene. *P. hermaphrodita* has been maintained in this formulation, under refrigeration, for 6 months. When required, the formulation is mixed with water and applied to soil using conventional spray equipment or a watering can with rose.

FIELD TRIALS

Since 1989, the effects of nematodes applied to soil have been tested in a number of field trials at Long Ashton (Wilson *et al.*, 1994a) and, since 1993 in a number of independent tests, including successful small-scale trials in Switzerland (Speiser & Andermatt, 1994). In most trials at Long Ashton, the effects of the nematodes have been compared with methiocarb bait pellets ('Draza', Bayer Ltd), a standard product for slug control, broadcast on soil at the recommended field rate of 5.5 kg product ha^{-1} . Details of three representative field trials are outlined below.

Two initial trials were done in mini-plots (each 0.5 m^2) surrounded by slug-proof barriers, with introduced populations of *D. reticulatum*. Nematodes reared in xenic cultures were applied as a drench to soil (Wilson *et al.*, 1994d) immediately before planting Chinese cabbage seedlings (and wheat seeds in the second experiment). Plots were irrigated as necessary to maintain moist soil conditions suitable for slugs and nematodes. In the first trial, plots treated with nematodes ($2 \times 10^{10} ha^{-1}$) or methiocarb pellets had significantly less slug damage than untreated plots, and from the third week onwards, there was significantly less slug damage on plots treated with nematodes than on methiocarb-treated plots. When the experiment ended after 6 weeks, there were significantly fewer *D. reticulatum* in soil samples from nematode-treated and methiocarb-treated plots than in soil from untreated plots. In the second experiment, six nematode doses (1×10^8 to $2 \times 10^{10} ha^{-1}$) were applied. In plots treated with nematodes, protection from slug damage improved over the first 3 weeks then stabilised; protection also improved with increasing nematode dose between 1×10^8 and $8 \times 10^8 ha^{-1}$, but showed little or no further improvement at doses higher than this (2×10^9 to $2 \times 10^{10} ha^{-1}$). Doses of 8×10^8 and above provided similar protection to that provided by methiocarb. Slug numbers were not measured in this experiment.

A field experiment was set up in winter wheat in autumn 1991. The wheat crop accounts for the majority of molluscicide use in the UK. Surveys in the mid 1980s of the perception, by UK growers and advisors, of crop protection problems in cereals demonstrated that slugs were the pests causing most concern in wheat and that slugs were by far the greatest perceived crop protection problem in first wheat crops after oilseed rape (Glen, 1989). The most severe damage is done when slugs kill seeds and seedlings below ground before emergence, but damage continues after emergence, with characteristic grazing of leaves.

Infective larvae of *P. hermaphrodita*, produced in monoxenic liquid cultures with *M. osloensis*, were sprayed onto soil at five dose rates (10^8 to $10^{10} ha^{-1}$) in 1100 l water ha^{-1} immediately after seed sowing. Seedling survival increased and slug grazing damage

declined linearly with increasing log nematode dose. An index of the amount of plant material surviving slug damage in each treatment was obtained (Figure 1) by multiplying the number of plants per 0.5 m of drill row by the proportion of the area of these plants remaining after leaf grazing by slugs. This index of undamaged plant material increased linearly with increasing logarithm of nematode dose and 6 weeks after treatment it was found that significantly more plant material had survived slug damage in plots treated with the two highest nematode doses than in untreated plots (Figure 1). Damage on methiocarb treated plots was not significantly different from untreated plots, but was similar to that on plots treated with the second highest dose of nematodes ($3 \times 10^9 \text{ ha}^{-1}$).

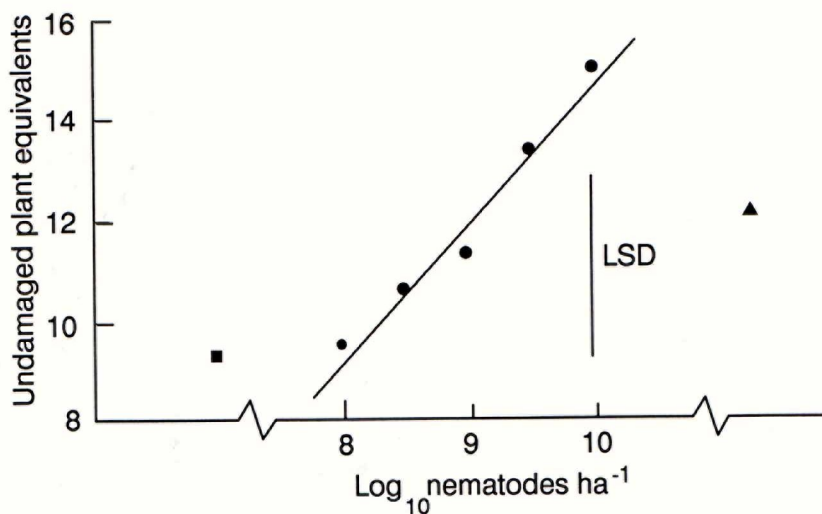


FIGURE 1. Index of the number of wheat seedlings and the area of leaves surviving slug damage, 6 weeks after drilling wheat in plots left untreated (■), or sprayed with one of five doses of nematodes (●), or treated with methiocarb pellets (▲), immediately after drilling.

Despite relatively dry and cool (mean air temperature 7.6°C) conditions during the first week after nematode application in this trial, nematodes were effective in decreasing slug damage to seeds, as well as later damage to seedlings. This confirms laboratory findings that *P. hermaphrodita* is able to find, penetrate and cause disease in slugs at the low temperatures in which slugs are active and troublesome pests. However, despite the clear effect of nematode application in reducing slug damage, it had little or no impact on slug populations (mainly *D. reticulatum*) in soil samples taken at five intervals over a 27 week period following treatment (Figure 2A shows slug numbers up to 13 weeks after treatment). In contrast, during the first weeks after treatment, significantly more slugs were recorded under refuge traps on plots treated with nematodes than under traps on untreated plots (Figure 2B) and a higher percentage of slugs on nematode-treated plots showed symptoms of nematode infection than slugs on untreated plots (Figure 2C & D). Moreover, a higher percentage of slugs recorded in traps (Figure 2C) showed symptoms of nematode infection than was found for slugs in soil samples in the same treatment (Figure 2D). This evidence indicates that infection of slugs by *P. hermaphrodita* altered slug behaviour,

making them more prone than uninfected slugs to rest under refuge traps. Thus, the reduction in slug damage following nematode treatment, coupled with a change in slug behaviour and a lack of apparent impact on slug numbers in soil, suggests that the effect of the nematode in protecting wheat from slug damage was probably through inhibition of feeding by infected slugs, which has been shown to be important in laboratory bioassays, rather than through death of infected slugs. The incidence of nematode infection in slugs in untreated plots in this trial probably reflected the background level of infection in this field; slugs in traps 20 - 30 m from the experimental site showed similar levels of infection to slugs in traps on untreated plots; also, in February and March 1988, 17% of *D. reticulatum* in this field were found to be infected by *P. hermaphrodita*.

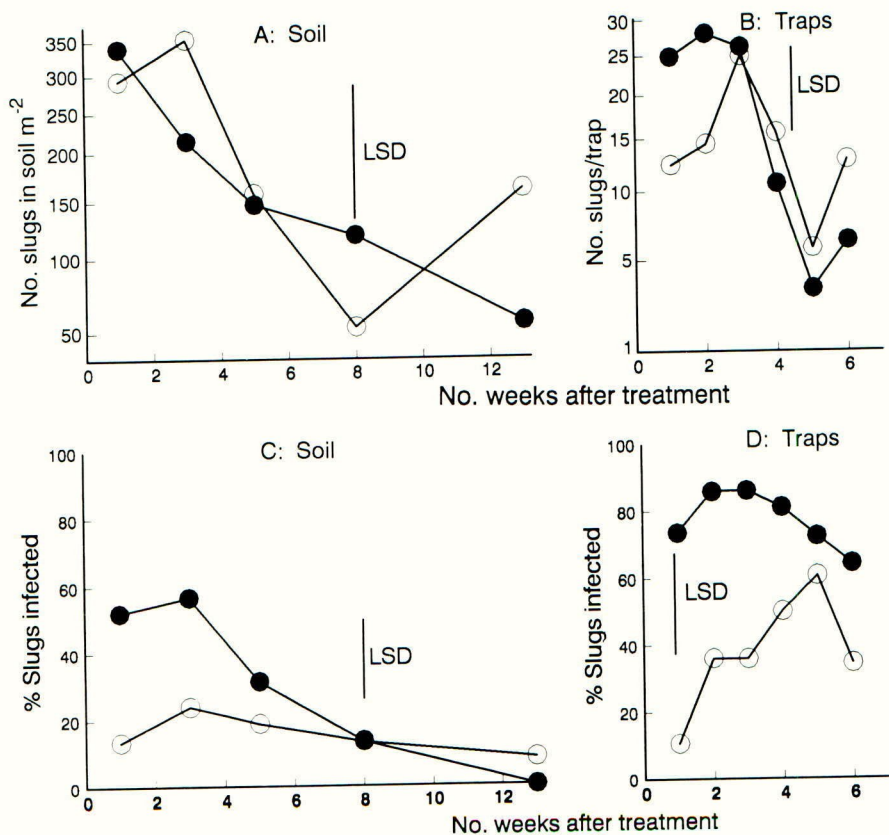


FIGURE 2. Effect of treatment with 1×10^{10} nematodes ha^{-1} (●) immediately after drilling wheat compared with untreated plots (○), on slug numbers (A) in soil samples, (B) in surface refuge traps (all numbers are shown on a square root scale), and on the percentage of slugs with signs of nematode infection (C) in soil samples and (D) in surface refuge traps. Five different doses of nematodes were applied (see Figure 1), but only the highest nematode dose and untreated plots are shown for clarity.

Despite the consistent effect on slug damage in the three experiments described, there are some important differences between experiments. In the mini-plot experiment where the effect of dose rate was investigated, there was little or no improvement in damage reduction at doses greater than $8 \times 10^8 \text{ ha}^{-1}$, whereas in the wheat experiment, crop protection improved linearly with logarithm of dose up to a dose of $1 \times 10^{10} \text{ ha}^{-1}$. Moreover, in the first mini-plot experiment, slug numbers in soil were reduced following nematode application, whereas in the wheat trial, there was no apparent effect on slug numbers in soil, although the nematode apparently affected slug behaviour on treated plots.

In all three field trials described here, nematodes applied at doses of $3 \times 10^9 \text{ ha}^{-1}$ and greater were as effective as, or more effective than, methiocarb in protecting Chinese cabbage and wheat from slug damage. These and other field experiments indicate that *P. hermaphrodita* is effective as a biocontrol agent for use against slugs over the relatively low range of temperatures at which slugs are damaging as pests, provided that soil is reasonably moist at and after the time of application. In experiments where the nematode has been applied one or two weeks before sowing or planting a susceptible crop, it has been found that application of nematodes in moist soil conditions is probably more important than timing of application and there has been no indication of a clear and consistent benefit from early application. It is likely that the inhibition of feeding associated with nematode infection plays an important part in its rapid action in the field. However, further studies are required to determine the best timing and method of application in different situations.

COMMERCIALISATION AND FUTURE RESEARCH

A commercial product based on *P. hermaphrodita* (Nemaslug^(R)) was launched for sale to UK gardeners in spring 1994. Each pack contains a minimum of 6×10^6 nematodes, with the recommendation that the product is suspended in water and applied to a total area of 20 m^2 of moist soil, using a watering can fitted with a rose. Research on this nematode is continuing in a project involving the Agricultural Genetics Company Ltd and IACR Long Ashton Research Station, under the MAFF LINK Technologies for Sustainable Agriculture Programme. The aims of this project include: establishing the principles for effective, economic inundative use of this nematode as a biocontrol agent against slugs in arable crops; determining the feasibility of encouraging natural epizootics of the nematode in slug populations in arable crops; investigations of the effects of the nematode on non-target organisms, including slugs and snails in field margins and hedgerows adjacent to sprayed areas; and compatibility with other forms of slug control and agrochemicals used in arable crops.

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Session 6B

**Fate and Effects of
Pesticides - Assessing
Environmental Impact and
Managing Risk**

Chairman

Dr P W Greig Smith

Session Organiser

Dr R Allan

Papers

6B-1 to 6B-5



AQUATIC ECOLOGICAL RISK ASSESSMENT IN THE USA

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ABSTRACT

Risk assessment is implicit in the pesticide regulatory process in the USA. For aquatic systems, the basic principles of risk assessment were formalized in 1986 around a deterministic "quotient" approach linking simple estimates of exposure with toxicity values. Regulatory action was triggered if the quotient exceeded a predetermined "level of concern". More recently there have been many advances in aquatic risk assessment which have resulted in the development of probabilistic models that operate levels of scale from toxicodynamics through organisms and metapopulations to entire populations at a "landscape scale". In the pesticide regulatory arena there are currently three different approaches to aquatic risk assessment. Of these, that being proposed by the Aquatic Risk Assessment and Mitigation Dialogue Group (ARAMDG) appears the most comprehensive.

HISTORY

Since 1970, the federal regulatory authority and basis for pesticide registration in the USA has rested with the Environmental Protection Agency (EPA) under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA). FIFRA, as amended, [Section 3 (c) (5) (D)] requires that the Administrator shall register a pesticide if they determine that "when used in accordance with widespread and commonly used practice it will not generally cause unreasonable adverse effects on the environment". Risk assessment is implicit under FIFRA and is conducted by the science branches of the Office of Pesticide Programs (OPP). Determining whether or not a risk is unreasonable is a risk management function, risk managers being the senior managers within OPP.

Up to 1992, formal risk assessment was in practice used largely as a framework for triggering increasingly complex studies, culminating in field studies, though there were intentions to formalize and expand single species estimates to communities and ecosystems (Urban & Cook, 1986). The objective of these field studies was to rebut or confirm a presumption of risk made from simple risk assessment. For aquatic systems, these field studies were initially conducted in pre-existing water bodies, usually ponds. However, the difficulty of achieving manageable replication and controls was considered too great to adequately perform this task. This led to the development of the "mesocosm" studies. Between 1986 and 1992, about 15 mesocosms were conducted for pesticide regulatory

purposes, at a cost to industry of *circa* \$75M, not including the cost of review within the EPA. None of these studies were considered by the EPA to rebut the presumption of risk, whereas many were peer reviewed and published in the open literature drawing the opposite conclusion.

The differing interpretations were due to three basic reasons. (1) a multitude of endpoints were measured with widely varying power, (2) there was no *a priori* agreement as to what constituted a biologically significant effect in the studies, (3) out of concern for environmental protection, the EPA departed from the usual statistical procedure and sought to constrain not only α (the false positive rate) but also β (the false negative rate). In other words, this meant that it was not possible to accept the null hypothesis unless it had been rejected. In a complex and fluctuating system of low statistical power, this is impossible.

The expenditure, delays in review and lack of closure resulting from this approach was troublesome to both EPA managers and to industry as it blocked the system of registration and re-registration. In order to by-pass this problem, the EPA issued a memo on 29 October 1992, known as "The New Paradigm" (Campt, 1992). This laid out a new strategy for resolving these problems that had arisen with aquatic, terrestrial and groundwater field studies. For aquatic systems, a simple risk assessment was to be conducted on the basic toxicity data and assumptions about exposure (Expected Environmental Concentration - EEC) derived from simple product chemistry, application methods and rate. If a predetermined "Level of Concern" (LOC) was exceeded a detailed risk assessment was conducted, (largely a refinement of the exposure term using PRZM and EXAMS programs). If at this point the LOC was still exceeded, the registrant was required to make mitigations to the product label and to monitor for residues and/or biological effects to demonstrate the effectiveness of the mitigation. The aquatic levels of concern are shown in Table 1.

TABLE 1. Aquatic Levels of Concern as Currently Quoted by EPA.

Action	Acute LOC	Chronic LOC
Endangered Spp : no action	EEC < 0.1 LC ₁₀ or EEC < 0.05 LC ₅₀	EEC < LOEC
Endangered Spp : consultation with Office of Endangered Species	EEC ≥ 0.1 LC ₁₀ or EEC ≥ 0.05 LC ₅₀	EEC > LOEC
Aquatic Effects : no action	EEC < 0.1 LC ₅₀	EEC < LOEC
Aquatic effects : consider action	EEC ≥ 0.1 LC ₅₀ EEC < 0.5 LC ₅₀	
Aquatic Effects : restricted use	EEC ≥ 0.5 LC ₅₀	EEC > LOEC

The "New Paradigm" put much more emphasis on risk assessment and the existing simple, deterministic system outlined earlier (Urban & Cook, 1986) became stretched beyond its original intentions. This resulted in a need to re-assess the approach to risk assessment. The following describes approaches to aquatic ecotoxicology being used in the USA and the proposals for aquatic risk assessment for pesticides.

NEW METHODS

There is considerable academic interest in risk assessment and this is influencing the development of regulatory guidelines in the area of ecotoxicology. There are two issues that are surfacing as foci for risk assessment models. These are probabilistic models as opposed to the current deterministic ones and questions of scale. Barnhouse (1992) has proposed that pesticide ecological assessment at the population and ecosystem levels could benefit from adopting the existing techniques used to assess ecological effects from other types of environmental stressors.

Probabilistic v Deterministic Models

Existing risk assessment models will concentrate on picking representative species and representative exposures. These can be mid-point values (such as EC_{50} s). The attraction and the problem with these deterministic models is that they give a single answer to the issue of whether or not something is a risk. As attractive as this is from a policy level, it is a poor model of the world which is necessarily much more complex. Additionally, if mid point values are used, the assumption is that a 50% effect is acceptable. To avoid this from happening, safety factors are applied to the deterministic model. For example Urban & Cook (1986) give the LOC for aquatic species to be when the estimated environmental concentration (EEC) exceeds $1/2$ the acute EC_{50} or for endangered species $1/20$ the acute EC_{50} EEC or $1/10$ the acute EC_{10} . The problem here is that the safety factors are arbitrary and may be grossly over protective or under protective so that though the answer seems clear, there is no way of knowing the confidence in that answer.

Probabilistic models show the probability of various outcomes of the risk assessment. This makes the task of risk management more difficult, in that more judgement is required than in the "yes/no" deterministic model, but ultimately more powerful and representative of the real world. Probabilities can be introduced to models either by looking at the "exceedence probabilities" or by introducing distributions of numbers in simulation models (Monte Carlo methods). Exceedence probabilities can either be calculated on a single species basis (Figure 1) or on a multi-species basis (Figure 2).

Figure 1. The calculation of exceedence probabilities from probability of exposure and single-species toxicity data.

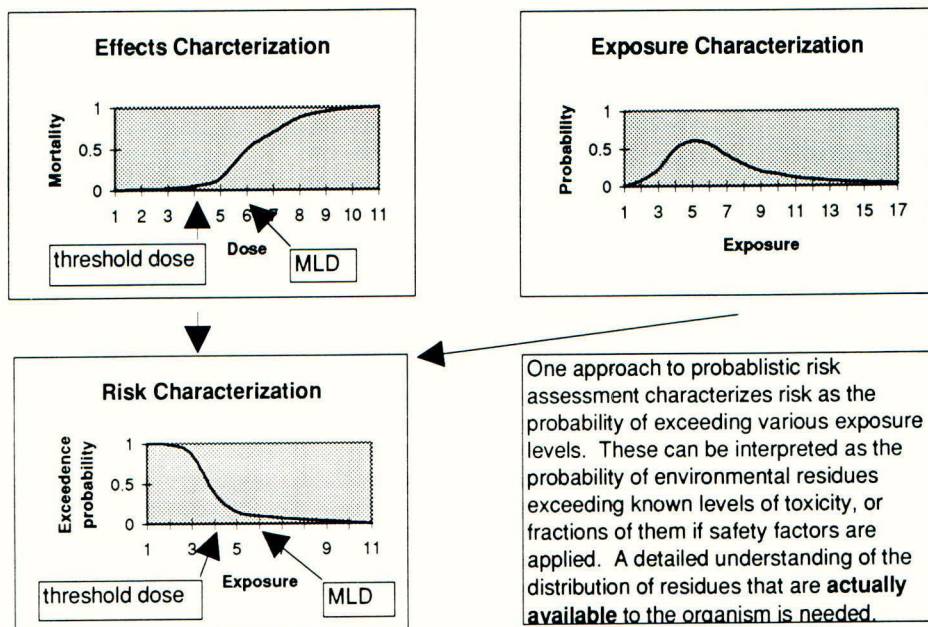
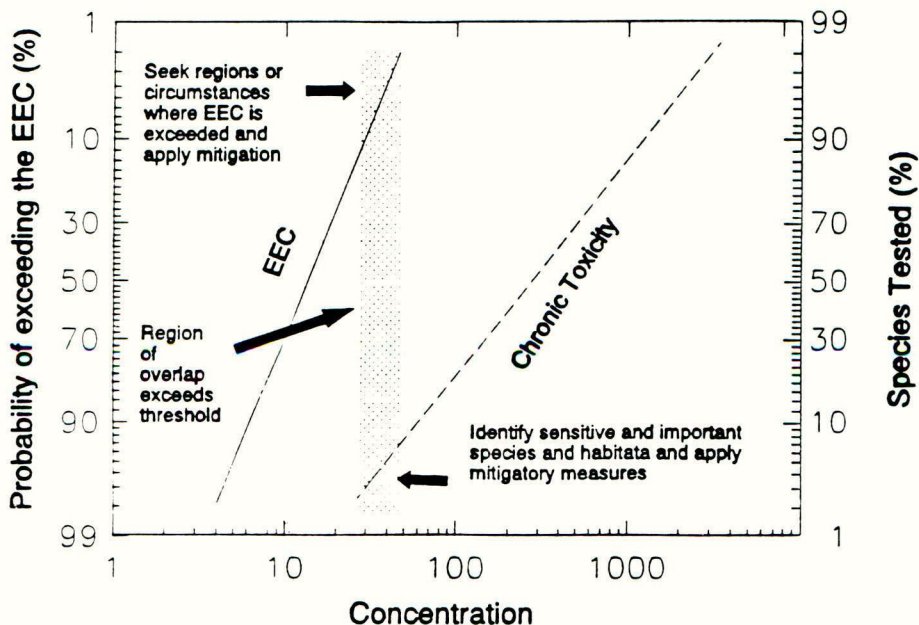


Figure 2. The calculation exceedence probabilities from probability of exposure and multi-species toxicity data.



The Question of Scale

Barnhouse (1992) reviewed modeling approaches developed in the last decade and their relevance to ecological risk assessment as tools for prediction, explanation and extrapolation. He discussed models in two categories. First, individual-based where the emphasis is on population characteristics and second, regional models which incorporate large-scale spatial patterns. For these "landscape" scale models, detailed information can be obtained from satellite imagery and numerous data bases can be analyzed and graphed using Geographical Information System (GIS). These methods enable the prediction of ecological risk at a landscape level. This is of particular significance given some of the more recent issues in pesticide regulation for example, risk to endangered species.

The pesticide regulatory fraternity is most familiar with mechanistic models which handle fate; these have been used in the regulatory process to understand fate and transport of pesticides. In contrast, individual-models are mechanistic models for effects which have not been used in pesticide regulation. These models are capable of simulating effects of stress on biota at all levels of organization. For example, Rose *et al* (1988) describe a community-level model developed from microcosm data. Individual-based models have been used extensively in fisheries research (e.g., van Winkle & Rose, 1993, DeAngelis *et al*, 1991) and have been applied in toxicological studies (e.g., Barnhouse *et al*, 1990, Bartell, 1990, Madenjian & Carpenter, 1993).

The individual-based approach provides a framework within which natural processes are conceptualized and empirical studies can be combined with modelling in a synergistic manner (Van Winkle & Rose, 1993). Unlike a classical population model (e.g., Leslie matrix) where variables are limited to size or age stages within which there is no individual distinction, individual based models can include any characterized process (Barnhouse, 1990). They also offer several advantages from an ecological risk assessment perspective (Bartell *et al*, 1992). A large amount of research has been conducted into these systems which have successfully modelled aquatic production dynamics at different levels of biological organization. The equations which describe the fundamental processes such as mortality, feeding, growth and reproduction can be readily converted to include sublethal physiological effects of contaminants. It is expected that the large amount of data available for pesticides is amenable to providing the necessary toxicity and toxicokinetic information for these models.

At an organism-level, individual toxicodynamic models can relate risk of death to the uptake and internal concentration of the contaminant. For example, Bartell *et al* (1988) express dose as a 48-h integrated body burden for various organisms. In this particular model, both fate and effects are integrated in order to assess the effects of the biota on the fate of the contaminant.

The Standard Water Column Model (SWACOM) described by Bartell *et al* (1992) is an example of how previous research experience with aquatic food web models can be developed into a risk-assessment tool to detect higher order effects. SWACOM models populations of organisms at various trophic levels (phytoplankton, zooplankton, forage fish and piscivores) and can account for the sublethal effects of toxicants on physiological

processes and ecological interactions.

The reliance on laboratory studies has resulted from the problems associated with conducting ecosystem-level studies. However, assessments on organism-level toxicity studies have not been without criticism (e.g., Kimball & Levin, 1985). Given this, models appear to be an appropriate bridge, as they are directed at predicting effects at population and ecosystem levels where test data are scarce or information is missing. Modelling could be a very useful and effective tool in pesticide risk characterization with knowledge of limitations and uncertainties. Model "validation" has been a topic of much discussion, however, no model can truly be called valid. Models are inherently inaccurate because they only simulate reality and therefore differ from actual reality. The quality and quantity of data will limit the validity of the model. This is apparent in the model validation procedure described by Bartell *et al* (1986) which uses repeated comparisons of model outputs to the data and assesses the relative contributions of model and data variance. In general, uncertainty associated with the model can be quantified by predictive outputs checked by experimental testing, peer review by technical specialists and use in regulatory practice (Suter *et al*, 1993).

APPROACHES IN REGULATION OF AGROCHEMICALS

Currently there is much activity in risk assessment at USEPA, as well as within state EPAs such as Florida, California and New York, and in other federal agencies such as Department of Agriculture (USDA) and the Fish and Wildlife Service (USFWS). Below we will consider only the USEPA methods, but even here there are now three separate approaches, the Aquatic Risk Assessment and Mitigation Dialogue Group (ARAMDG), the Corn Cluster Analysis and the Ecological Effects Branch (EEB) LOC Project.

ARAMDG

The Aquatic Risk Assessment and Mitigation Dialogue Group (ARAMDG) was set up in 1993 to address issues of risk assessment and risk mitigation in aquatic ecosystems arising from the publication of the "New Paradigm" document. The group met under the auspices of the Society for Ecotoxicology and Chemistry (SETAC) and was comprised of representatives from the USEPA (regulators and researchers), agrochemical companies, academia, environmental and agricultural interest groups. This group is probably the most important in its recommendations as it represents a consensus of opinion between the main stake-holders.

The recommendations of the group are now at an advanced stage but are as yet unpublished. Full disclosure of the results would necessarily be premature, however, indications of the approaches that will be proposed are now well known.

The approach to risk assessment is tiered. The first tier is a deterministic screen, based on a single-event, worst case assumption of a direct overspray of a 2 m deep, 1 ha pond or 1-10% run-off (depending on solubility) from a 10 ha field. The acute and chronic toxicity values come from three species of animal and one plant (if relevant). The trigger for Tier 2 is that the EEC exceeds the acute LC₅ of the most sensitive species or lowest chronic

NOEC.

Tier 2 is probabilistic. Modelled EECs come from the upper 10th percentile of the run-off from a small number of climate/soil scenarios for each proposed pesticide market and toxicity values from an additional two species from the most sensitive group represented in Tier 1. Triggers are for markets which the upper 10th percentile EECs exceed the lower 10th percentile of the LC₅s or NOECs. Tier 3 is similar to Tier 2, only with more detailed modelling of the EECs to include geographical as well as a more inclusive range of soil/climate factors. Tier 4 has more detailed geographical modelling and additional toxicity data as appropriate. Spray-drift deposition estimates based on the model currently being developed by USEPA and the Spray Drift Task Force (SDTF) will be added where appropriate.

This approach differs from the existing system by formally including probabilistic approaches to exposure and toxicity and approaching an estimation of the potential of effects on non-target populations by accounting for geographical factors.

Corn Cluster Analysis

This is an analysis of the risks to human health, avian and aquatic wildlife of 25 products (13 herbicides and 12 insecticides) frequently used in corn in the USA. Phase I of the analysis addresses the models for risk assessment of the individual products and Phase II examines the effects on cost-effectiveness and overall predicted risk of various regulatory strategies. The analysis was conducted by Abt Associates Inc. of Bethesda, MD, for the USEPA Office of Policy Analysis.

With respect to aquatic species, the corn cluster analysis uses a simple deterministic "quotient" approach similar to the water quality criteria (USEPA, 1985) to assess the Acute Toxicity Threshold (ATT) and assumes that this is indicative of effects to communities in the field. These thresholds were set at ½ the concentration giving a predicted EC₅. In the absence of chronic toxicity data, the Chronic Toxicity Thresholds (CTTs) were predicted by applying a constant safety factor to the acute data. This factor is known as an "acute to chronic ratio" or ACR and is somewhat anachronistic and not well regarded by risk assessment scientists these days due to its inaccuracy.

Estimates are made of the potential risk in two scenarios, a farm pond and a reservoir, on the basis that a farm pond is likely to be more highly exposed than a reservoir but of less ecological significance. Exposure in the reservoir was estimated via the SWRBQ model (Arnold *et al*, 1991) and in the farm pond via PRZM-2 (transport to the pond) and the fate of the chemical in the pond by a model called AQUATOX.

The Phase I analysis is set out to be a conservative screen, which indeed it is, and is as good as the data used as inputs. However, because of the conservative nature of this model and inconsistencies in the selection of data the comparative risk assessments can be misleading and its use in Phase II to assess the overall risk, highly inaccurate.

The Ecological Effects Branch LOC Project

This is an internal and as yet unfinished exercise with in USEPA Ecological Effects Branch (EEB) which attempts to rank 58 registered pesticides for their acute and chronic LOCs for birds, fish and aquatic invertebrates. The LOCs for aquatic species are shown in Table 2.

TABLE 2. Quotients and LOCs in the EEB LOC Project.

Risk	Quotient	LOC
Acute Fish (stream)	EEC/LC ₅₀	0.5
Acute Fish (pond)	EEC/LC ₅₀	0.5
Chronic Fish (stream)	EEC/LEL ¹	1.0
Acute Aquatic Invertebrate (stream)	EEC/LC ₅₀	0.5
Acute Aquatic Invertebrate (pond)	EEC/LC ₅₀	0.5
Chronic Aquatic Invertebrate (stream)	EEC/LEL	1.0

¹ LEL is an EPA term meaning least effect level. Officially, it is a point between the no observable effect level (NOEL) and the lowest observable effect level (LOEL). Conventionally this would be the maximum acceptable tolerable dose (MATC), the geometric mean of the NOEL and LOEL; however, in this analysis and in all most EPA risk assessments this is taken to be the more conservative NOEL.

Three scenarios are being modelled, a farm pond receiving run-off; a farm pond receiving run-off and 5% aerial spray-drift and a stream. The final results of this analysis are as yet unknown. The toxicity values from the most sensitive species in EPA data bases were used and compared in the quotient assessments in Table (2) with modelled EECs from SWRB (for streams) and STREAM (for streams). This analysis is as yet unfinished.

CONCLUSIONS

Of these approaches, the "Corn Cluster" and "LOC project" are the most similar in that they are deterministic quotients. However, they are different as the utilize different toxicity data sets and endpoints, different scenarios with different assumptions about chemical movement via run-off and/or drift and different models for calculating aquatic EECs. Though, assuming that the underlying data is consistent, these approaches would provide a

comparative ranking mechanism; it is unlikely that they will give an overall absolute risk assessment that is meaningful.

The ARAMDG approach is preferable to industry as it is probabilistic, has known safety factors for the representative nature of toxicity and exposure estimates, is capable of making some estimates of absolute risk in the field and represents consensus of most stakeholders in aquatic risk assessment in the USA. It is important for these different approaches to be rationalized for future clarity in pesticide regulation.

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GIS, REMOTE SENSING AND THE NEW EPA PARADIGM

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ABSTRACT

The adoption by the USEPA of their "New Ecological Risk Paradigm" has changed the way in which manufacturers approach registration and re-registration of pesticides in the United States. The movement of the EPA away from field studies and towards regulatory decisions based on hazard data have prompted many manufacturers to re-examine their options. One of these options includes the collection of exposure data based on the physical relationship between crops and sensitive habitats using remotely sensed data (satellite and airborne imagery) incorporated into a geographic information system (GIS). The data generated by quantifying these spatial relationships can become a valuable tool for justifying exposure scenarios based on environmental measurements rather than on assumed environmental conditions. In addition, the data developed from the use of remote sensing and GIS can be incorporated into exposure models which can then be executed in a probabilistic manner to represent the range and frequency of characteristics found in the environment.

INTRODUCTION

Recent years have seen the steady growth of both satellite and aerial remote sensing and geographic information system (GIS) technologies. Computer hardware and software technology has progressed to the point where satellite images that previously required expensive super computers for processing can now be analyzed in a timely manner using PC technology. In addition, the increased quality of current digital imaging systems coupled with more rapid data capture rates has given the scientific community a powerful tool with which to map and monitor land cover characteristics. As this technology continues to mature, new applications utilizing satellite and airborne imagery will be developed. One of these developing applications involves the use of satellite and airborne digital imagery, in conjunction with GIS, to assist in the quantification of environmental characteristics that influence pesticide risk analyses.

Recent developments within the regulatory community in the United States have also opened new doors for the acceptance of alternative methodologies for data generation and analysis. In October of 1992 the United States Environmental Protection Agency (USEPA) published their "New Paradigm" for ecological risk assessment. In an effort to streamline the ecological risk assessment process (USEPA, 1986) (and to make it possible to meet their deadlines as set forth in the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) of 1988), the USEPA all but eliminated the requirement for avian and aquatic field testing. The time required to conduct and review such studies had become prohibitive, and these studies were

not providing risk managers with data that significantly improved their ability to make regulatory decisions. As a result, the USEPA shifted from a reliance on large-scale field studies to laboratory testing, incident data, and "other information" as the basis for their regulatory decisions. Although this was intended to speed up the regulatory process, it did not bode well for those registrants that had products demonstrating a high degree of toxicity (avian or aquatic) in the laboratory. In these cases the issue became one of exposure. Registrants faced with this concern are now left with the formidable task of presenting the USEPA with exposure data that not only defends their product, but also provides the USEPA with data that can be used meaningfully within the context of an ecological risk assessment. The "New Paradigm" clearly leaves room for the registrant to pursue avenues of data generation other than those traditionally pursued:

"The Assistant Administrator strongly supports efforts to move toward more realistic exposure estimates based on the most realistic and sophisticated EEC calculations using our most qualified personnel and employing the best available data and modeling techniques." (USEPA, 1992)

REMOTE SENSING

In the technical sense, remote sensing is the acquisition of data about an object or phenomenon without physically coming in contact with that object or phenomenon (Jensen, 1986). In terms of the application of remote sensing for ecological risk assessment applications, digital satellite and airborne imagery are generally used.

Although ecological risk assessment is a relatively new application for remote sensing, the technology has been used for environmental characterization and assessment for over 20 years (Hoffer et al., 1979; NASA, 1974). Typical uses of remotely sensed data include land-cover / land-use mapping, forest type mapping, geological exploration, soils mapping, agricultural monitoring, water quality assessment, and wetlands delineation. Because the sensors are located on satellites, their imagery covers very large areas on the ground. It is the ability of remote sensing to analyze such large areas in their entirety that has gained the attention of scientists involved in exposure analysis. Additionally, satellite and airborne imagery provides the user with digital data that can easily be manipulated with a computer for crop area measurements, average field size analysis, crop adjacency analysis, crop buffer composition analysis, etc.

Although the use of remote sensing data for environmental characterization is well documented (Colwell et al., 1983), several factors have aided in its recent maturity enabling the agrochemical industry to consider it for use in risk assessments. These factors include the advancement in sensor technology and number of available satellites, the advances in computer processing technology, the maturation of remote sensing software and the increased knowledge base of the end user. Probably the most important of these factors is the advancement in sensor technology and number of commercially accessible satellites. First, recent advances in sensor technology, have provided low cost airborne sensors capable of collecting high resolution multispectral digital data (Pearson et al., 1992). These data lend themselves to rapid, accurate manipulation and computation for environmental measurements. Secondly, the growing number

of commercial satellites has increased the probability of data acquisition during critical vegetation phases resulting in increased accuracy of land-cover analysis. Additionally, the number of satellites and their highly repetitive orbits provide an opportunity to conduct analyses on recently acquired data (within 2 weeks) while at the same time providing total coverage of study areas consisting of thousands of acres. These developments, in combination with the recent developments in probabilistic modeling, the large aerial coverage and current data provided by the remote sensing systems will soon become an invaluable base for quantifying exposure within an ecological risk assessment.

GEOGRAPHIC INFORMATION SYSTEMS (GIS)

A GIS is a computer system designed to store and analyze spatial data (Burrough, 1986). Whereas all of the popular spreadsheets and databases are capable of relating common data columns, the GIS uses geographic coordinates as the common denominator between spatial datasets. Using this geographic commonality, the GIS can begin to query and analyze many different layers of spatial data such as soils, elevation data, meteorologic data, hydrology, and land-cover types. Some of the operations that can be performed using the GIS include proximity analyses, buffering, statistical analyses, and modeling (Aronoff, 1989; ESRI, 1992). The remotely sensed data provides one or more of the spatial layers generally analyzed within the GIS, therefore the utility of both of these tools is improved when they are used in conjunction with each other. This is especially true for ecological risk assessments where current data about vegetation and cropping must be combined with other environmental factors such as soil types.

One of the most important areas that will rely heavily on GIS is that of probabilistic modeling. Once constructed, the GIS can be used to calculate the frequency of occurrence of parameters such as soil type, slope, field size, field perimeter, and adjacent land-cover. By using the frequency distribution, data from the GIS can be processed iteratively through different models to produce probability distributions of results.

TIERED TESTING, REMOTE SENSING, AND GIS

Under the "New Paradigm" the tiered testing for avian and aquatic organisms as defined in Subdivision E is still in effect (EPA, 1982). Figure 1 is an expansion of the tiered testing flowchart for aquatic organisms. The expansion is intended to include the additional parameters necessary for a complete evaluation of risk due to the labeled uses (or intended labeled uses) of the compound. Within this tiered testing flowchart, the "New Paradigm" effects those studies conducted under tier 4 which includes "simulated/actual field studies" and "special tests". Within tier 4 the "New Paradigm" switches the emphasis away from the previously required "simulated/actual field studies" to the "special tests" which gives the registrant more flexibility in determining the methodologies for developing product specific data. It is at this point in the tiered testing flowchart that the "New Paradigm" functions as a gate through which the GIS and remote sensing technologies can be incorporated into the tiered approach to risk analysis. An example of how GIS and remote sensing can be used to generate environmental data relevant to an ecological risk assessment is presented in Figure 2.

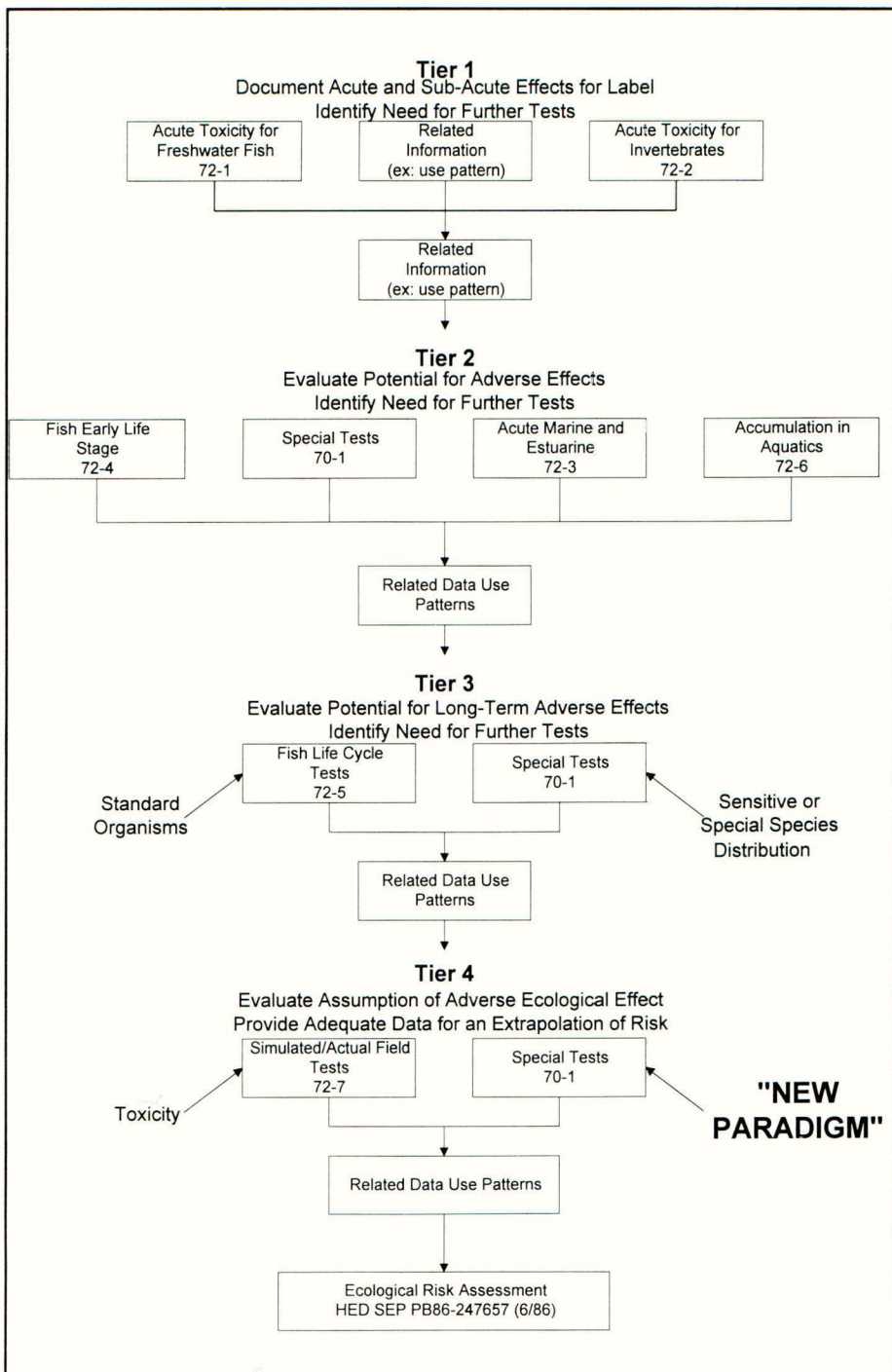


Figure 1. Aquatic organism testing flowchart (Modified from Subdivision E guidelines for aquatic organism hazard evaluation) (USEPA, 1982)

DATA GENERATION AND ANALYSIS

Figure 2 proposes essentially four main stages to data development under Tier 4. The first of these stages is site selection. Although this stage is not listed as falling under the realm of the GIS, in most cases the use of the GIS would significantly improve the site selection process. Data are currently available for the United States that can be used to select areas that represent distinct physiographic and ecological environments. Several of these datasets include the USEPA's Ecoregion designations for the Conterminous U.S. and the Major Land Resource Areas (MLRA) as defined by the U.S. Department of Agriculture Soil Conservation Service (USDA-SCS). These data can be used in conjunction with cropping information, pest pressure data, and/or soil data to identify the number and location of sites across the United States that would be required to generate a statistically representative sample of a products interaction with the environment.

The second stage is the data collection stage. Once the sites are selected, satellite imagery of the area can be acquired. All of the major companies that market satellite imagery have extensive libraries of historical imagery. Therefore, historical data can be selected by the end user or the satellites can be programmed to acquire more current imagery. Airborne imagery may also be acquired during this stage providing a higher level of spatial resolution (1 - 2 meters). This data can be used to address issues related to proximity and landscape mitigation that can not be addressed using the satellite imagery due to its spatial resolution limitations (10 - 30 meters).

Other digital data available from government agencies that can be incorporated into the GIS include soil, elevational, meteorological, hydrological data, and transportation data. In addition, environmental fate and toxicity data for a specific product can be incorporated into the analysis. Although not all product data have a spatial component, they can in some cases be linked to existing spatial data such as soil types and then incorporated into the GIS.

Once the data required to address specific product issues are acquired, the data becomes a part of the GIS as separate layers of information. This includes the satellite data that has been processed into a land-cover map to identify crops or habitats of concern. With all of this data, the GIS can be used to characterize the environment. For example, the hydrology and the satellite data can be used to determine the adjacency of aquatic habitats to a crop; or the soils and satellite data can be used to determine the distribution of soil types and their characteristics for a specific crop; or the aerial imagery can be used to quantify buffers widths and composition around aquatic habitats at high resolution. The combination of remote sensing and GIS provides a multitude of ways in which the environment can be characterized depending on the environmental issues related to a specific product.

As a part of the environmental characterization, the GIS can generate data from which distributions and/or probabilities can be derived. For example, in most cases specific crops are grown on a range of soil types with varying characteristics. The GIS analyzes those soils that are associated with the crop and generates a probability distribution for the association of specific soil types with the crop. Then, when the environmental fate models are implemented

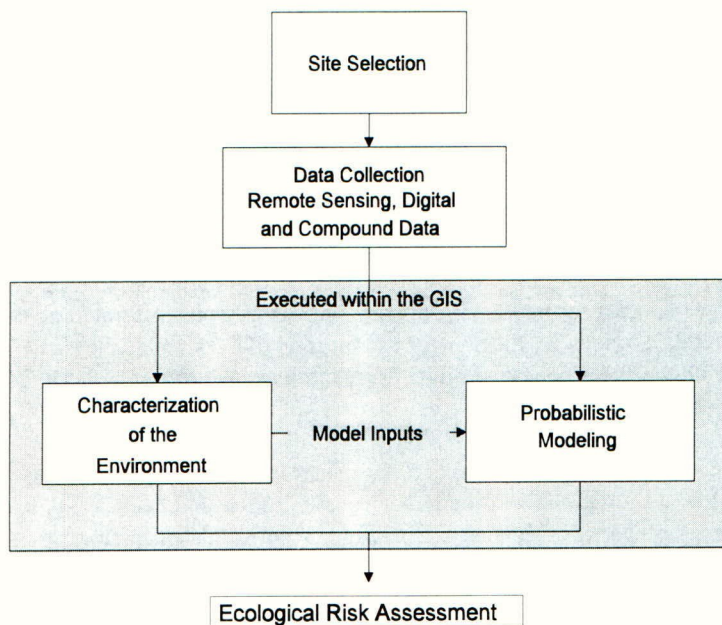


Figure 2. Flowchart detailing the use of remote sensing and GIS for the generation and modeling of data to be used within the context of ecological risk analysis.

using these data, an understanding of the frequency with which this soil type occurs with the crop will be used to generate a probability of occurrence for the modeling result. This same concept can be extrapolated to adjacency of habitats to a crop, buffer widths and compositions, meteorological data, field sizes, and water body sizes, to name a few.

CONCLUSION

The technology related to remote sensing and GIS is well established and ready for new applications and innovative ideas. Applications utilizing this approach are within reach, both technically and financially, and are being evaluated by both the agrochemical industry and the USEPA. Additionally the "New Paradigm" necessitates that the agrochemical industry take the initiative in defending their products (especially for exposure and environmental fate), thereby opening the door for the use of remote sensing and GIS technologies.

USEPA'S RESPONSE

To date this technology has not been through a rigorous regulatory test by the USEPA. There are, however, a number of registrants who are pro-actively pursuing this technology as a

product registration tool. Some of these projects are currently before the USEPA and it is anticipated that many will soon follow. At this point, the USEPA's response to this technology has been favorable.

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