SESSION 3B

UNDERSTANDING LONG TERM, FIELD-SCALE EFFECTS OF PESTICIDES IN TERRESTRIAL ECOSYSTEMS

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INVITED PAPERS 3B-1 to 3B-5

THE EFFECTS OF PESTICIDES ON TERRESTRIAL NON-TARGET ARTHROPODS IN BROAD-ACRE CROPS – A STRATEGY FOR DATA COLLECTION

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ABSTRACT

Recent studies with pyrethroid insecticides and non target arthropods in laboratory and field experiments are analysed and used to suggest a strategy for assessing the effects of pesticides on 'natural enemies' in broad-acre crops. Three levels/tiers of testing are proposed:

i) laboratory tests to identify the possibility of effects on non-target arthropods.
ii) replicated field studies to produce quantitative data on short term effects.
iii) large scale unreplicated studies to identify possible long term effects.

INTRODUCTION

During recent years much attention has been given to the possible effects of pesticides in broad-acre crops on non-target arthropods. This paper focusses on 'natural enemies' and uses as an example experience from experiments concerned with the use of pyrethroid insecticides to control summer populations of aphids in UK cereal crops. Different approaches have been used to assess these effects ranging from laboratory tests to large scale multi-year monitoring studies. The relative merit of each approach is discussed with a view to proposing a strategy for data generation which it is hoped will be applicable not only to the use of pyrethroids in summer cereals but also to the use of pesticides in broad-acre crops in general.

In the UK attention has centred on the use of pyrethroid insecticides to control summer populations of aphids (*Sitobion avenae* and *Metopolophium dirhodum*) in cereal crops. Pyrethroids have, until recently, been excluded from such use because of concern about possible adverse effects on non-target arthropods. Coats (1986) expressed the view that longer term field studies were required to determine whether arthropod populations remained resilient over years of exposure to pyrethroids. This concern was re-iterated, specifically in relation to cereal aphid control, by Carter *et al*. (1989). They observed that a summer application of deltamethrin gave good control of cereal aphids but cautioned that since pyrethroids are broad-spectrum insecticides the possible long term consequences for 'natural enemy' populations should be assessed before they can be recommended for summer use in cereals.

The principal arthropod groups in question are those natural enemies which may limit aphid outbreaks in some years (Edwards *et al*, 1979; Chambers & Sunderland, 1983; Powell, 1983). These consist of polyphagous predators (carabid beetles, staphylinid beetles and spiders) which are field resident, and aphid specific predators (coccinellid beetles, syrphid flies and lacewings) which, together with the parasites, arrive in the crop as the aphid population grows.

Other arthropods that do not attack aphids but may be affected by summer aphicides include pollinators such as honey bees (which may forage on aphid honeydew) and phytophagous groups such as sawfly larvae and chrysomelid beetles which may serve as food for gamebird chicks.

Polyphagous predators can be found in most U.K. cereal fields, although there are regional differences in species composition. There can also be large differences in the composition and size of populations in adjacent fields. In contrast, since the aphid specific predators and the parasites tend to arrive in the crop in response to an aphid outbreak they cannot be relied upon to appear in reasonable numbers in any given year. The gamebird chickfood species tend to be associated with weeds and occur more commonly close to field margins.

LABORATORY TESTS

Initial laboratory screening tests for efficacy by manufacturers give the first indication as to whether a herbicide or fungicide has the potential for affecting arthropods. (Typically all compounds synthesised will be screened for fungicidal, herbicidal and insecticidal activity.) Similarly, for insecticides early performance evaluation reveals whether there is a broad spectrum of activity against a wide range of groups or whether there is likely to be selectivity.

If it is considered that a product is unlikely to have effects on non-target arthropods then laboratory tests are probably the most effective way of demonstrating lack of activity; the IOBC/WPRS working groups have published many laboratory methods for testing non-target arthropods (Hassan, 1989). An example of the use of laboratory tests demonstrating lack of activity is given by Brown *et al.*, (1983) who showed that pirimicarb residues on glass plates were non-toxic to adults of the carabid *Pterostichus melanarius*, a result borne out by subsequent field studies (Brown, 1983; Powell *et al.*, 1985). Laboratory studies can also indicate groups which may be more likely to be affected than others. For example, Brown *et al.* (1983) showed that in glass plate exposure experiments *Coccinella septempunctata, Erigone* spp. and *Syrphus* spp. were more susceptible to cypermethrin than the carabids *Nebria brevicollis* and *P. melanarius*. In many cases however, insecticides which have appeared harmful in the laboratory have, when tested in the field, been found to be less hazardous than anticipated, or to be innocuous. Perhaps the most striking and best documented example is that of the pyrethroid insecticide alphacypermethrin (FASTAC*) and honey bees, where, despite high toxicity in laboratory tests (Murray, 1985) an extensive series of field experiments did not reveal any hazard in use (Shires *et al.*, 1984a; Shires *et al.*, 1984b).

From the above it is clear that when laboratory tests with non-target arthropods indicate the potential for effects in the field the next step is to proceed to field testing where exposure will be more realistic. For those products where early efficacy data have indicated a likelihood of activity against non-target groups it may often be more informative to move directly to field testing rather than conduct an initial series of laboratory tests with non-target arthropods. However, for some groups (e.g. adult parasitic Hymenoptera), for which field data are difficult to obtain, laboratory studies may be the only available method for obtaining an early indication of possible effects in the field.

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REPLICATED FIELD STUDIES

These studies are intended to provide the quantitative assessment of short term (within season) effects of pesticide applications on the non-target arthropod fauna. There are a number of requirements for these studies. The site for the study should be a field with the crop grown under typical commercial conditions. Sampling of the non-target arthropods in a number of fields prior to selection of the trial site will help to ensure selection of a good site. Applications of the pesticide should follow normal agricultural practice with the test chemical being applied at the highest recommended rate. The inclusion of untreated control plots and a positive control (toxic standard) is necessary to allow interpretation of the data and the use of a negative control (soft/selective standard) can also be of value. All treatments should have a minimum of 4 replicates. A wide variety of sampling methods for non-target arthropods are available (pitfall traps, D-vac, sweep net, visual counts etc.) and selection should be made on the basis of the crop under study and the fauna potentially at hazard. Two approaches to replicated plot studies are available, and each is outlined below.

Small plot replicated studies

A replicated small plot study has often been the first step into the field, particularly in broad-acre crops such as cereals. Plot sizes vary from a few square metres to approximately 100 m^2 (Shires, 1985a; Matcham & Hawkes, 1985). Because the polyphagous predators are active and can quickly recolonise small treated plots from untreated surrounding areas such designs have tended to use buried polythene or metal barriers to contain the surface active invertebrates within treated plots. Obviously these barriers also act to exclude further colonisation of the plots, so the timing of their erection is critical. For example, to evaluate a summer aphicide it is important to establish barriers after the second generation of carabid beetles have emerged. Studies of this type tend to be effective for carabid and staphylinid beetles and for linyphiid spiders (although if movement by ballooning occurs data for this last group may not be valid). Figs. 1 and 2 show the effects of pirimicarb and cypermethrin on Carabidae and Linyphiidae following their application to small barriered plots in summer cereals (Brown, 1983). The lack of effect of both compounds on Carabidae and the short term effects of cypermethrin on the Linyphiidae can be compared with the laboratory results obtained by Brown *et al.* (1983). These laboratory experiments indicated both compounds to be toxic to *Erigone* spp. and neither to be toxic to *P. melanarius*. In small plot replicated studies effects on developing parasites can also be ascertained by examining emergence from mummies. However, they do not generally provide information on adult parasites or aphid specific predators.

Small plot studies have a number of advantages that are not immediately obvious. Because the whole experiment can be conducted in a relatively small area it is possible, by pre-trial sampling of the fauna, to select sites with naturally-occurring high populations of predatory invertebrates. The small area utilized also tends to minimise between-plot variance in populations of non-target arthropods. Together these factors serve to increase the resolving power of the experiment, allowing effects due to treatment to be detected when they occur. The presence of barriers in these experiments prevents/limits the re-invasion of many epigeal predators and the acute effects can therefore be representative of events following applications to very large areas. With these small plot experiments it is often practical to have many treatments in a fully replicated experiment design.

There are, of course, disadvantages of small plot studies with barriers. Most notably it may be possible to 'trap-out' predators within the plots and the presence of the barrier may alter predator behaviour. Studies with barriered plots will be unlikely to generate data for aphid specific predators or chick food species. However, data for aphid specific predators and parasites have been generated from small plot studies when mesh cages, constructed over the crop, have been used (Langley, 1985; Inglesfield pers. comm.).

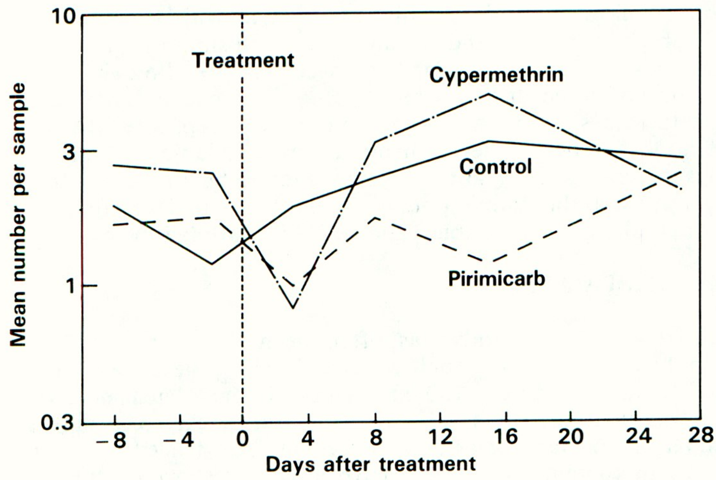

FIG 1. Effects of insecticides on Carabidae caught in pitfall traps in small barriered plots (After Brown, 1983)

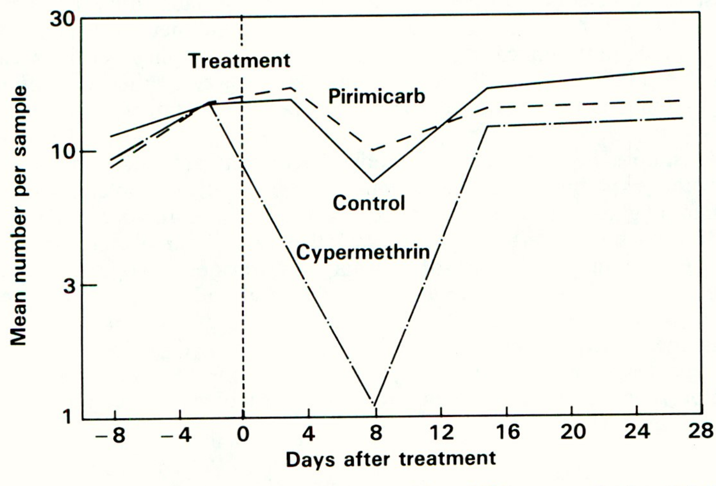

FIG 2. Effects of insecticides on Linyphiidae caught in pitfall traps in small barriered plots (After Brown, 1983)

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Large plot replicated studies

In these experiments the plots are not barriered and plot sizes typically range from 1 to 3 ha with sampling normally restricted to the central area of each plot. With four replicates required even the 1 ha plot study, allowing a field margin of 50 m and 15 m discards between plots, will need a field of 30 ha. Because such studies need this large area of land it is often not possible to find sufficient suitable sites to allow site selection to be based on pre-trial sampling. If a large and uniform population of non-target fauna can be found across a trial site then this design is clearly superior to the small barriered plot approach. It more closely approximates to commercial practice in terms of scale; there is no possibility of oversampling and it may yield data on a wider range of taxa. However, in practice, the distribution and abundance of non-target arthropods within very large fields are often extremely patchy. This patchiness serves to decrease the resolving power of the experiment, reducing its ability to detect significant effects should they occur. Finally, even with 1 – 3 ha plots re-colonisation, particularly by the more mobile species, can occur quite rapidly.

LARGE PLOT UNREPLICATED FIELD STUDIES

The results from these studies can only be interpreted qualitatively but because of their scale they do have a high degree of realism. With these large plots, often whole fields, the number of treatments that can be incorporated into an experiment declines and rarely have more than 2 treatments been included. Application of the pesticide(s) under study should follow recommended commercial practice with regard to timing and rate. Non-target arthropods can be sampled using the methods suggested for the replicated plot studies. One advantage of this design is that they can fit into standard agricultural practice and can therefore last for more than one season.

Shires (1985b) treated large (~10 ha) adjacent fields of winter wheat with cypermethrin and demeton-s-methyl by aerial application (a third untreated field served as a control). He found that neither compound produced serious long-term effects on carabids or linyphiids, though both had temporary effects on the numbers of linyphiids, with a more marked effect resulting from the cypermethrin treatment (Figs. 3 and 4). In general, the results of this study were in line with the findings of earlier small barriered-plot experiments with cypermethrin (Brown, 1983; Shires, 1985a).

In a five-year unreplicated experiment (Inglesfield, 1989) alphacypermethrin was applied annually to a 4 ha plot and the effects on non-target arthropods compared with those resulting from the application of other commercially appropriate insecticides applied to an adjacent 4 ha plot. The results indicated no long-term adverse effects of alphacypermethrin on the groups studied, although, as in earlier studies with cypermethrin, there were short term effects on linyphiid spiders. The study allowed Inglesfield to conclude that any differences between the effects of alphacypermethrin and the non-pyrethroid products on entomophagous arthropods were short lived, with no evidence that any of the treatments had long term adverse effects on any of the taxa studied. This, along with other published data on pyrethroids, indicated that their use in European arable ecosystems is unlikely to have long term effects on the structure and integrity of the arthropod communities.

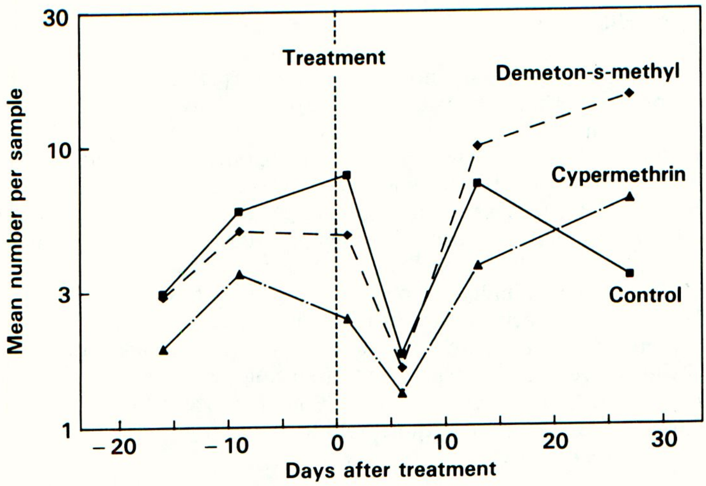

FIG 3. Effects of insecticides on Carabidae caught in pitfall traps in an unreplicated whole field experiment (After Shires, 1985b)

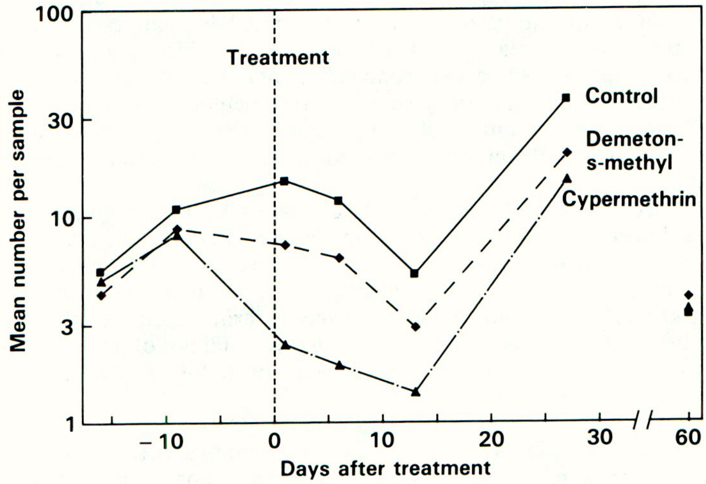

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FIG 4. Effects of insecticides on Lynphiidae caught in pitfall traps in an unreplicated whole field experiment (After Shires, 1985b)

COMMERCIAL MONITORING

The monitoring of non-target arthropods following commercial use of a product appears at first to be more relevant than the procedures so far described. However, since replication under conditions of commercial use is unlikely to be possible the results from such monitoring will tend to be qualitative. Also, commercially managed crops are likely to receive applications of many products in addition to the one in question. Any reduction in numbers of non-target arthropods occurring during monitoring of commercial practices will therefore be difficult to assign to any particular treatment. In contrast to all the previous approaches, which tend to be confined to a single locality, commercial monitoring could be conducted simultaneously in different regions and would, if required, provide the opportunity to confirm the long term acceptability of the product in use.

DISCUSSION AND STRATEGY

Three groups of non-target arthropods which serve as 'natural-enemies' in broad acre crops were identified: polyphagous predators, pest specific predators and parasitoids. Drawing specifically on recent experience with pyrethroid insecticides in cereals, a series of studies, ranging from simple laboratory tests through to commercial monitoring, has been outlined. A broad indication as to which type of study is capable of generating useful information on the effects of pesticides on each of the three groups of natural enemies is given in Table 1. Table 2 provides a summary of the main characteristics of each of the field designs and highlights the relative strengths of the different methods. A consideration of these strengths indicates that in general, small and large replicated plot studies have important features in common as do large unreplicated studies and commercial monitoring.

TABLE 1. Suitability of laboratory and field experiments for generating data on three groups of 'natural enemies'.

	Laboratory	Field Small plot Replicated	Field Large plot Replicated	Field Large plot Unreplicated	Commercial Monitoring
Polyphagous Predators	+	+ + +	+ + +	+ + +	+
Aphid Associated Predators	+ + +	–	–	+	+
Parasitoids					
adults	+ + +	+ (cages)	–	+ + +	–
mummies	+ + +	+ + +	+ + +	+ + +	+ + +

+ + + very suitable
+ acceptable
– unlikely to produce useful data.

TABLE 2. Outline summary of the major features of the different field experiment designs.

	Small plot replicated	Large plot replicated	Large unreplicated	Commercial monitoring
Quantitative	Yes	Yes	No	No
Number of treatments	≥4	3 – 4	1 – 2	1
Suitable for manipulation	Yes	No	No	No
Realism	Low	Some	Good	Excellent
Risk of trapping out	Yes	No	No	No

Common important strengths are highlighted

A consideration of the information in Tables 1 and 2 leads us to conclude that broadly three levels (tiers) of study are present.

i) Laboratory tests – primarily used to identify the possibility of effects on non-target arthropods or to produce data on specific organisms for which data were not available following field studies.
ii) Replicated field studies – used to produce quantitative data on short term (within season) effects and allowing comparison with reference compounds.
iii) Large scale unreplicated studies including commercial monitoring – these studies provide qualitative data under 'realistic' conditions, the number of different treatment regimes will be small, but such studies can provide data on possible long term effects, through multi-year studies.

It is suggested that using the approach outlined here, the possible effects of pesticides on non-target arthropods can be investigated effectively. It is not envisaged that all, or indeed many, products would require testing at all levels.

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AN INTEGRATED APPROACH TO ASSESSING EFFECTS OF SOME PESTICIDES IN GRASSLAND

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ABSTRACT

A combination of field surveys, whole field experiments and microplot experiments has been used to study the effects on non-target invertebrates of a range of pesticides used in grassland. Field surveys of ground beetles and spiders in northern England & southern Scotland showed that the community is determined primarily by factors such as soil moisture and density (ground beetles), or altitude and pasture management (spiders). Repeated use of chlorpyrifos against *Tipula* larvae had a noticeable effect, although management factors other than pesticide application were more important in the short term. Whole-field experimental studies on newly-sown fields in southern England showed that both chlorpyrifos and fonofos affect the activity of the common ground beetle species; the effect seems to result from direct toxicity rather than shortage of invertebrate prey, and was short-lived in the most actively re-colonising species. Microplot experiments confirmed that reseeding operations are more devastating to soil fauna than subsequent pesticide usage. The impact of pesticides on the carabid *Pterostichus melanarius* was least if the beetles were already present in the plot before spraying, rather than being introduced afterwards, and if the soil was relatively un-disturbed, as would be true in many upland pastures.

INTRODUCTION

Grassland occupies over 60 per cent of the agricultural land in the United Kingdom (Hopkins, 1988); some 350,000 ha of grass are sown each year. However, despite the large area that this crop covers it receives little insecticide - less than 5 per cent of that applied to cereals (Wilkins, 1985). In recent years, however, farmers' awareness of pest problems in newly-sown grass has increased. Consequently there is a greater likelihood of insecticides being used. However, the impact of these chemicals on non-target and beneficial fauna, e.g. carabid beetles (Carabidae), has not been widely studied in field scale experiments for newly-sown grassland. This paper outlines three approaches that have been used to assess the effects on predatory non-target invertebrates of some pesticides used in grassland. These were:

i) Field surveys of the ground beetles and spiders of lowland reseeds and upland improved pastures treated with chlorpyrifos to control Tipula larvae.

ii) Whole field experiments on the impact on carabids of chlorpyrifos and fonofos applied to newly-sown fields in southern England.

iii) Small plot and microplot experiments, to investigate; (a) the effects of a range of pesticides on soil Collembola and mites, and on frit fly and its parasitoids; (b) rapid screening of the effects of pesticides on carabid beetles.

It was envisaged that these three investigations would complement each other. Microplot experimentation is already widely used for screening pesticides for their effects on non-target invertebrates in cereals (Hassan, 1985), but suffers from the potential problems associated with un-natural environmental conditions. Whole field experiments enable the relevance of such small-scale trials to be assessed, but the cost and effort needed (vide the Boxworth experiment using cereals, see Grieg-Smith, 1989) severely limit the numbers of replicates and treatments that can be tested in practice. Field surveys on farms enable a wider range of treatments and conditions to be evaluated, but without the ability to carry out conventional statistical analyses of variance. However, multivariate analyses offer the opportunity to assess the main sources of variation in the data (Rushton & Luff, 1988), and to generate hypotheses that can be tested experimentally on a more limited scale as in the other sections of this work. Detailed results of all three approaches will be decribed in more detail elsewhere: this paper merely outlines each and indicates how they complement one another.

FIELD SURVEYS

Methods

The field work was divided equally between two years. In 1987 ten upland pasture sites covering a range of pesticide treatment histories were selected at the M.A.F.F. Experimental Husbandry Farm at Redesdale, Northumberland. These were sampled using an existing sampling protocol, of nine pitfall traps per site, filled with ethylene glycol and emptied at monthly intervals from April to October in each year. Taken in conjunction with existing data from 1985 and 1986 (Luff & Rushton, 1989), this provided spider and ground beetle community data for a total of 19 sites on this farm differing in pesticide usage and management history. Between-year variation was known to be generally less than management effects (Luff & Rushton, 1989).

During 1988, 10 lowland pasture sites in south-west Scotland were also sampled using the same protocol. Together with existing data on lowland sites (Eyre et al., 1989, 1990) this provided spider community information for a total of 22 pastures and ground beetle community information for 37 pastures.

The effects of pesticide usage on the upland sites alone and in combination with the lowland sites, were investigated using Classification, Ordination and Discriminant Analysis procedures in combination.

Classification, using two-way indicator species analysis (TWINSPAN, Hill, 1979a) divides the sites into groups (by repeated binary division of the data) and identifies the characteristic 'indicator' species at each level of the classification.

Ordination, using detrended correspondence analysis (DECORANA, Hill, 1979b) sumarises differences between sites and represents variation within the data on axes which are easier to comprehend than the original data matrix. Canonical correspondence analysis (CANOCO, Ter Braak, 1987) enables the effects of measured environmental variables to be included in the ordination procedure.

Stepwise linear discriminant Analysis (James, 1985) was also used to assess which environmental factors differed most between groups of sites previously identified using Twinspan.

Results

The upland sites (both improved and unimproved) were classified and ordinated on the basis of their species lists of spiders and ground beetles. For both the spiders and the ground beetles three groups of sites were identified by classification; one unimproved and two improved site-groups. For both invertebrate groups the discriminant analyses suggested that the most important factor influencing the classification was whether or not the pasture had been improved. The unimproved sites formed a discrete class in each case. The species composition of unimproved grassland sites was more diverse than that of the improved sites. For the ground beetles the major distinction between the two improved site-groups was the intensity of pesticide use. It was concluded that repeated pesticide use is more damaging to larger beetle species because these tend to have longer life-cycles and hence are more likely to receive lethal doses of chlorpyrifos over the course of their lives than are smaller species with shorter life-cycles. For the spiders pesticide usage was less important in determining the separation between the two improved site-groups than was the year of sampling. It was concluded that the spider fauna of improved sites reflects colonisation from, and hence the species composition of, the aeroplankton in each year and this depends on climatic factors such as prevailing wind direction (Rushton et al., 1989).

Further analyses were carried out on the improved (i.e. more intensively managed) sites from both upland and lowland sites together. The relative importance of pesticide use, soil characteristics (water content, organic matter and bulk density) and site altitude as factors determining the structure of the ground beetle communities present on each site is summarised in Fig. 1. Substrate conditions, especially soil water and soil density appeared to be important in influencing community distribution, whilst pesticide application appeared to have less influence on community structure (Eyre et al., 1990).

The spider community data from the same managed upland and lowland sites were similarly analysed. Spider communities on upland sites were more species rich than those of lowland sites. The results of the Canonical Correspondence Analysis suggested that the most important factors influencing the communities were altitude and the pasture utilisation strategy. The main effects of altitude appeared to be related to the surrounding areas of unmanaged grasslands in upland, which provided a larger pool of dispersing or

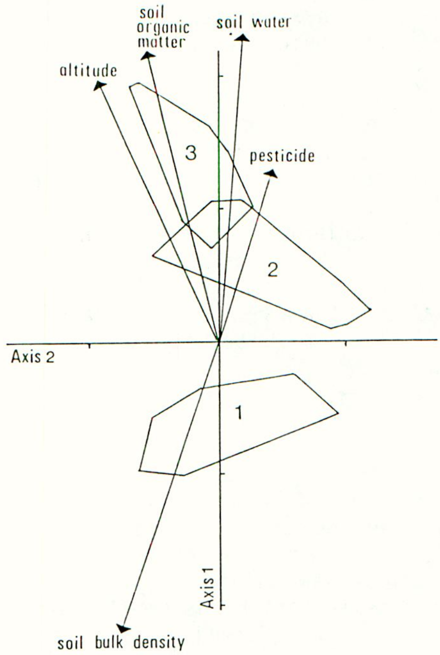

Figure 1

CANOCO biplot of ground beetle communities on managed grassland sites; polygons enclose each TWINSPAN group of sites. The amount of variation due to each environmental variable is shown by the relative length of the arrows.

colonising species than in the more intensively farmed lowland areas. Management practices such as silage production appeared to influence lowland grassland communities particularly. Pesticide application did not appear to be as important in determining the spider community structure as these other variables.

WHOLE FIELD EXPERIMENTS

Methods

We studied the impact on carabids of the insecticides chlorpyrifos and fonofos, applied to 12 newly-sown fields near Hurley. The 12 fields chosen were as similar as possible in terms of previous cropping history, soil type and soil moisture content. Four fields were left untreated, four were sown with fonofos treated seed (at between 0.11 and 0.18 kg a.i./ha) in early September 1988 and four were sprayed with chlorpyrifos (0.72 kg a.i./ha) in mid-September 1988.

The carabid population was sampled in each field from September 1988-December 1989 and February-June 1990, using pitfall traps (Clements et al., 1988). After collection, carabid larvae and adults were extracted, washed and identified to species.

In order to evaluate the effects of the pesticides on growth of the larval stages, measurements were made of the elytral and pronotal widths of the subsequent generation of adults. Measurements were made of *Nebria brevicollis* adults trapped in October 1988, which would have been active as larvae prior to the start of the experiment. These were compared with *N. brevicollis* adults trapped in May 1989, which would have been active as larvae at or shortly before the time of pesticide treatment. To assess effects on fecundity, adult female beetles from each field were dissected to determine the number and maturity of the eggs they contained.

The data was found not to be normally-distributed, so the mean pitfall catches for each treatment were analysed by a Kruskal-Wallis non-parametric test over a range of time intervals. This analysis produces a mean rank for each treatment, reflecting the number of beetles caught.

Results

From the mean ranks for carabid adults caught, the rank for the chlorpyrifos treated fields is much lower than for the other fields ($P < 0.01$). This test was carried out for all pitfall catches, from the first sample after the chlorpyrifos was sprayed, to the end of the experiment, and shows that the effects of chlorpyrifos persisted for more than 18 months after spraying (Table 1). Similarly, the mean ranks for their larvae show that chlorpyrifos consistently reduced the numbers caught ($P < 0.01$).

More detailed data, not presented here, showed that the numbers of various common species of carabids were apparently affected differently by the chlorpyrifos treatment. For example, numbers of adults and larvae of _Nebria brevicollis_ were greatly reduced for 18 months, whereas _Trechus quadristriatus_ was affected for only nine months after treatment.

TABLE 1. Results of Kruskal-Wallis test on mean ranks of Carabidae caught in pitfall traps, based on four fields per treatment. Sampling period October 1988 - June 1990 (except for _N. brevicollis_ larvae, - June 1989). Differences between mean ranks followed by a different letter (horizontal comparisons only) are significant at $P < 0.05$ (*) or $P < 0.01$ (**)

Carabid species	Mean rank Control	Chlorpyrifos	Fonofos	H-value (2 d.f.)
All adults	56.4 ^{a}	34.3 ^{b}	54.8 ^{a}	12.45 **
All larvae	58.6 ^{a}	28.9 ^{b}	57.9 ^{a}	23.72 **
N. brevicollis adults	53.2 ^{a}	37.1 ^{b}	55.2 ^{a}	8.13 *
N. brevicollis larvae	34.4 ^{a}	16.3 ^{b}	36.3 ^{a}	16.77 **
Notiophilus species	69.5 ^{a}	32.6 ^{b}	43.5 ^{c}	29.65 **
T. quadristriatus	57.5 ^{a}	33.6 ^{b}	54.4 ^{a}	13.92 **

No significant differences were found between treatments in elytral and pronotal widths or in the number of mature eggs per female _N. brevicollis_. The inference from this is that the larvae developing in the treated fields after insecticide application were not deprived of prey. Therefore the initial effect of chlorpyrifos on adults and larvae must be due to direct toxicity. The reduced trap catches more than a year after spraying must also have resulted from the initial direct toxic effect.

MICROPLOT EXPERIMENTS

Soil microfauna, frit fly and parasitoids

Methods

To study soil microfauna, and frit fly and its parasites, field trials were established in 1987-88 and 1988-89 with various pesticides applied in a randomized block design with five blocks and one replicate of each treatment per block; plots measured 7 x 3m. A range of chemicals (listed below) likely to be used on either established ('permanent') or newly-sown Italian ryegrass were applied at the times and rates used in normal agricultural practice.

Chemicals used were: chlorpyrifos, methiocarb and an untreated control (on both permanent and reseeded pastures); propiconazole, MCPA/2,4 DB/Benazolin, triazophos, gamma HCH and triclopyr (on the permanent pasture only); fonofos seed treatment, benomyl/captan, carbosulfan, drazoxolon, omethoate and mecaprop (on the reseeded pasture only).

To assess the microfauna, 5cm turf cores were taken from each plot before the application of the pesticides and at intervals of 2,4, 16 and 32 weeks after treatment. Microfauna were extracted by a tullgren funnel system and identified to major groups.

In order to sample frit fly and its parasitoids, 5cm turf cores were taken from each plot in both the permanent and newly-sown grass. The number of plants and tillers in each core were counted and then each tiller dissected to locate the frit larva within. After identification, each larva was dissected and inspected for internal parasites.

Results

From the soil microfauna results obtained in 1987-88, it is clear that the destruction of an existing grass sward and the associated seed bed preparations for reseeding are far more detrimental to soil microfauna than any of the pesticides used. For instance, the total microfauna population in September 1987 in untreated permanent pasture was 35,000/m^2 compared to 19,000/m^2 in untreated plots of the reseeded grass. The equivalent figures for September 1988 (two weeks after treatment), although higher, follow the same trend: 80,000/m^2 for permanent pasture and 25,000/m^2 for the reseeded grass. This reduction in numbers after cultivation resulted mainly (>85% in both years) from a very high kill of Collembola. Soil mites and other microfauna were relatively unaffected. Possible changes in species composition within major groups were not studied.

Two weeks after treatment (when the effects of the pesticides might be expected to be most obvious) there was no significant reduction of any microfauna group for any pesticide treatment when compared to the controls in either the permanent or reseeded pasture.

In both years the microfauna populations increased from September to November, particularly in the reseeded areas, stabilized through winter, and then increased markedly by April and May. By then the populations were higher in the reseeded pasture in all three treatments which were common to both the reseeded and permanent grass (control, methiocarb and chlorpyrifos).

From these results it appears that (i) the most commonly used grassland pesticides have few, if any, detrimental effects on soil microfauna, and (ii) soil disturbance is a greater problem, although populations return to normal within 6-12 months.

Results of the frit fly experiments were hampered by low numbers of the pest. In both 1987-88 and 1988-89, despite early drilling to optimize the level of infestation, larval densities in the late autumn were low, with 6% of tillers infested in the normally susceptible new-sown Italian ryegrass in 1987 and only 2-3% in 1988. In both years the infestation in the permanent pasture was 1-3%.

While some caution is necessary in interpreting the effects of pesticides at these low densities, some points of interest arise. First, given that the host (frit fly larvae) densities were low, the levels of parasitism in 1987-88 in the control plots (20% in the reseeded grass and 40% in the permanent pasture) suggest that the parasites are effective in searching out suitable hosts. Second, as with the soil microfauna, soil cultivations are more damaging to the parasitoids (the main one of which cannot fly and is therefore likely to be adversely affected by soil disturbance) than pesticides. Third, omethoate appears to be more effective in controlling frit fly than either chlorpyrifos spray or fonofos seed treatment.

In both the 1987-88 and 1988-89 experiments there were no significant differences in yields in the first grass harvests taken in the late autumn (2 December 1987 and 11 November 1988), in any pesticide treatment compared to the controls. Since the main pest attacking grass at this time is frit fly and the densities were known to be low, large differences in yield were unlikely. However, based on the treatments applied to the permanent pasture in the autumn, there was an order of decreasing yield in 1987 from propiconazole (2.78 t/ha) to the control (2.59 t/ha) which was significantly correlated (rank correlation) with a decreasing % of cryptostigmatid mites in the total microfauna, and also with the ratio of cryptostigmatid to mesostigmatid mites. In autumn 1988, propiconazole was again the highest yielding treatment in the permanent pasture with 4.1 t/ha, and although the order of decreasing yields was different to autumn 1987, with MCPA the lowest (3.14 t/ha), there was still a highly significant correlation with the order of decreasing % cryptostigmatid mites and the ratio of the Cryptostigmata to Mesostigmata. The role of the different chemicals on the predatory mesostigmatid mites, and/or effects of amounts of decaying weed material on the detritivore Cryptostigmata warrants further attention. This should involve analysis through an annual cycle in both types of pasture.

Pesticides and carabid beetles

Methods

Microplots were constructed from large barrels, cut lengthways and dug into holes in the ground. Each barrel was filled with soil, 'cultivated', and sown with grass seed or left fallow. Carabid beetles (*Pterostichus melanarius*) caught in pitfall traps in nearby fields were introduced into each microplot before or after the application of pesticides. Activity was recorded with pitfall traps in each microplot, and mortality assessed by the total recovery of dead or living beetles after 10 days.

Results

In the first experiment 10 P. melanarius were introduced into each microplot after spraying or sowing treated or untreated grass seeds into a fine tilth. There was 100% mortality in the chlorpyrifos-sprayed plots but only 10% in plots treated with fonofos granules or sown with fonofos-treated seed.

In a second experiment, 10 beetles were introduced into the microplots 5,10 and 24 hours after chlorpyrifos had been sprayed onto a fine tilth. Again there was 100% mortality in all these treatments, but only 20% dead or missing in the control.

In a third experiment, the soil was left in rough clods or prepared as a fine tilth, and the beetles were introduced into microplots 48 hours before spraying. This gave them the opportunity to shelter beneath the larger clods in the rough soil treatment. There was 80% mortality in the sprayed fine tilth, 24% in the treated 'rough soil' and 16% in the control.

DISCUSSION

All three approaches used in this work have shown that pesticides used in grassland can affect the non-target invertebrates that were studied. However, at all levels of scale from microplot experiments to surveys of whole farms, insecticide application was clearly not the dominant factor influencing the potentially beneficial invertebrates. Whether on lowland reseeded pastures, or upland improved grasslands, the cultivation process itself is seriously damaging to the invertebrate fauna present. This was especially evident from analysis of the field survey data, in which the application of pesticides was never the major source of variation.

The whole field experiments also used normally managed commercial farms, and gave a realistic evaluation of the consequences of using insecticides on carabid beetles. Fonofos seed treatment had some minor effects, but the numbers of many common carabid species e.g. N.brevicollis and T. quadristriatus were greatly reduced in the swards treated with chlorpyrifos.

N. brevicollis is strongly polyphagous, feeding on flies, springtails, mites, spiders and earthworms (Hengeveld, 1980). Having such catholic tastes, it is unlikely to have suffered from shortage of food. Further, the beetles were the same size in all fields and contained similar numbers of mature eggs. The inference of this is that the larvae developing in the treated fields after insecticide application were not deprived of prey. Therefore the effect of chlorpyrifos, which persisted more than a year after spraying, was probably due to initial direct toxicity on the adult beetles.

The initial effect of chlorpyrifos on T. quadristriatus was very similar to that on N. brevicollis. However, numbers of the former recovered in 1989, probably because T. quadristriatus is an active flyer, and has little difficulty in recolonizing a pasture.

The microplot results indicate that (i) carabid beetles are highly active when first introduced into the new environment of a microplot, and likely to acquire a lethal dose from any toxic sprayed chemical, especially from a well-prepared fine tilth, and (ii) if the beetles are introduced before spraying and given time to habituate to the new environment (as would occur in nature), and if the soil is relatively underprepared (as would be true of many upland pastures) then the hazard of chlorpyrifos is greatly reduced. The mortality levels at around 20% were then similar to those recorded in the whole field experiments.

The same microplots show that the soil microfauna may recover from physical disturbance within 6-12 months, as also shown on reclaimed spoil heaps (Hutson & Luff, 1979), and is relatively unaffected by grassland pesticides. This gives further support to the conclusion that any changes in surface fauna such as carabid beetles are unlikely to result from a shortage of their food.

Changes in the soil and vegetation structure of upland grasslands following pasture improvement result, howwever, in a permanently impaired invertebrate fauna on the soil surface (Rushton et al., 1989). This fauna may itself consist of only those species which are pre-adapted to disturbed conditions, and able rapidly to colonise new sites. In both upland and lowland pastures, the ability of natural enemies to recover from insecticide application depends on their mobility, and on the availability of adjacent, unsprayed habitats.

Each of the three sections in this work has both merits and drawbacks, as outlined in the Introduction to this paper. Integrating all three has shown that some features, such as the over-riding importance of physical disturbance to natural enemies, are common to all approaches. It seems reasonable that whole field experiments under real farm conditions are the ideal way of testing the effects of pesticides; such experiments combine realism with conventional statistically analysable data. The combined project has expanded this 'core' approach in two ways. The larger scale survey data has provided more generality to support conclusions from the whole field experiments. They have also yielded data sets large enough to analyse whole invertebrate communities rather than individual species. In the other direction, the microplot approach has enabled more chemicals to be tested with greater replication than on a whole field scale. The microplot data has suggested mechanisms for the survival of some natural enemies when sprayed, as well as supporting the hypotheses about availability of prey to surface-active invertebrates. The three approaches together suggest that presence of potentially beneficial natural enemies in grassland may depend more on other cultural practices than on insecticide usage, as long as pesticide amounts and frequency of application remain at their present levels.

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DEVELOPMENT OF AN EXPERIMENTAL PROGRAMME TO PURSUE THE RESULTS OF THE BOXWORTH PROJECT

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ABSTRACT

The results and experience from the Boxworth Project form the basis for further research, funded by the Ministry of Agriculture, Fisheries and Food (MAFF), to extend knowledge of the environmental side-effects of modern farming methods and examine the consequences of adopting lower pesticide and fertiliser inputs. Two closely linked series of new trials, SCARAB (Seeking Confirmation About Results At Boxworth) and TALISMAN (Towards A Lower Input System Minimizing Agrochemicals and Nitrogen), have been started. They involve four MAFF Experimental Husbandry Farms and include a range of arable crop rotations. SCARAB will focus on assessment of the ecological impact of two contrasting pesticide regimes on populations of invertebrates in collaboration with Southampton University. TALISMAN will concentrate on the agronomic effects of reducing inputs and introducing cropping systems which are intended to be more environmentally benign than present practices.

INTRODUCTION

In 1981, the Ministry of Agriculture, Fisheries and Food (MAFF) began a major long-term, multi-disciplinary project to investigate the environmental effects of pesticide use in intensive cereal production. Because of its location on the Boxworth Experimental Husbandry Farm (EHF), near Cambridge, this study became known as the Boxworth Project. At that time in the UK, routine pesticide use in cereals, particularly in winter wheat, had become widespread (Sly, 1986). However, apart from the Game Conservancy monitoring studies of the insect fauna of cereal fields (Potts & Vickerman, 1974), no large-scale and long-term investigation of the whole cereal ecosystem and pesticide use had been undertaken.

Boxworth EHF provided a dedicated farm area for an experimental study of the ecological and economic effects of crop protection strategies, over a relatively long period. Staff from the Agricultural Development and Advisory Service (ADAS), Research Institutes and Universities collaborated in the Project. After two years of baseline flora and fauna monitoring, three different treatment strategies were applied to matched areas of the farm for a five year experimental phase to compare reduced pesticide treatment regimes with a prophylactic approach. The three pesticide regimes comprised a pre-planned Full Insurance insecticide, fungicide and herbicide routine, a Supervised programme in which pesticide decisions were based on crop monitoring and thresholds and an Integrated treatment intended to reduce pesticide use further by additionally exploiting changes in crop husbandry. Site details and the monitoring studies were described in detail by Hardy (1986) and in the Boxworth Project Annual Reports (ADAS, 1983-89). A final report, in book form, summarizes the results from all the research studies associated with this seven-year study at Boxworth (Greig-Smith *et al,* 1990).

EXPERIENCE GAINED FROM THE BOXWORTH DESIGN

Boxworth was primarily an ecological study, with economic inputs and outputs monitored incidentally. The principal ecological effects identified concerned invertebrate populations, particularly those of predators. Reductions, frequently short-term, in polyphagous beetle and spider populations have been described from studies of effects of single pesticide applications on insect predator/prey relationships, (Vickerman & Sunderland, 1977; Edwards *et al*, 1979; Cole *et al,* 1986; Sotherton *et al,* 1987; Purvis *et al*, 1988). The Boxworth Project extended this knowledge by identifying several categories of insect predators (Burn, 1988; Vickerman, 1988) that were affected to varying degrees, depending on their dispersive abilities and life-histories, by the combination of pesticide applications in the high input regime.

The Project also demonstrated the validity of the whole-farm approach for research on the long-term effects of pesticides and highlighted the scale of crop monitoring necessary and the practical difficulties of operating a supervised system. The broad findings are complemented by results from farm system studies in Europe (El Titi, 1986; Booij & Noorlander, 1988).

Inevitably the Boxworth design was a compromise between the conditions required for long-term study of different aspects (Table 1). A major criticism of the experimental design was the lack of standard plot replication. However, the study involved detailed long-term monitoring of a wide range of non-target fauna and flora, including mobile vertebrate and invertebrate populations. Consequently, a sufficiently large experimental area to demonstrate severity and duration of effects on these very active species was paramount. On this scale, it is extremely difficult to build in the desired degree of replication, for practical reasons and through financial limitations. Therefore treated areas were not replicated in the Boxworth Project although a small plot replicated trial was established to provide corroborative data.

Other criticisms of the Project arose from the inherent inflexibility of the design as the study developed (Table 1). No modifications were made to follow current trends in pesticide use or husbandry. Latterly, the inputs used in the Full Insurance programme were higher than had become the norm for cereal production. Nevertheless, this pre-planned high input did provide a consistent reference for comparative purposes, throughout the experiment. In addition, the continuous wheat cropping pattern itself was no longer typical of current farming. Also, the restriction of the Boxworth Project to a single crop rotation on one farm limited its immediate extrapolation for prediction of effects of pesticides on a wider scale. However, the knowledge gained and the lessons learned from the Boxworth design formed a sound basis for future research aimed at understanding and ameliorating any undesirable effects from current farming practices.

TABLE 1. Values and constraints of the Boxworth design.

Boxworth Design	Reason for Selection	Compromise Accepted
Large experimental areas (whole fields and boundaries)	Essential for monitoring mobile species, minimizing edge-effects. Reflect scale of commercial applications	Replication not feasible for practical and financial reasons
Tight control over whole farming system	Distinguish pesticide effects from other changes in farm practice	Practical difficulties in operating research studies alongside realistic farming
Observations in continuous wheat	Pesticide inputs generally high; suitable system for initial study; monitoring techniques available	No crop rotation. Results not necessarily applicable more widely
Focus on pesticides	Minimize confounding effects from other variables	Exclusion of other environmentally important practices eg fertilisers
Long-term comparison of contrasting pesticide regimes	Stronger interpretation possible than from simple before/after contrasts of individual pesticides at one site	Regimes chosen primarily for experimental reasons. (Integrated not true integrated farming system)
Rigid Full Insurance pesticide regime	Representative of high input farming to identify major effects; provide standard contrast to Supervised/Integrated	Input levels not modified to reflect changing farm practices over experimental period
Broad range of ecological groups monitored	Opportunity to identify interactions and indirect effects	Treatment areas not equally suitable for all groups
Baseline monitoring for 2 years	Identify level of similarity between experiment areas	Differences still developing could affect results
5 year experimental phase	Reveal effects that emerge relatively slowly	Longer term effects not detected
Economic appraisal of inputs and outputs	Complement the environmental results	These two approaches not readily compatible within same design

STRATEGY FOR FUTURE STUDIES

In 1988, MAFF held a Topic Review of the Boxworth Project to consider the priorities for subsequent studies and how they should be designed. A wide range of interests in agricultural research and farmland ecology was represented and this meeting confirmed support for continuing research to extend the findings from Boxworth, taking into account its strengths and limitations. Prolonging full monitoring under this rigid design (Table 1), although possibly a means of identifying any as yet undetected longer-term effects, would not allow broader extrapolation. It was agreed that resources might be more usefully devoted to new projects. However, a limited continuation at Boxworth, to monitor the extent to which populations recovered after a relaxation of the high input regime, was proposed and subsequently implemented.

There was an obvious priority for further work to confirm the generality of the effects on invertebrate populations under a wider range of conditions, soil types and crop rotations and with input levels more closely reflecting current practice. At the same time, there was also an urgent need to examine in detail the agronomy and economics of reduced nitrogen and pesticide use to support the formulation of policies on land use and agricultural practice. The environmental and economic aspects, although complementary, had proved difficult to investigate simultaneously. Therefore, for the future follow-up work, there was merit in separating these components into parallel studies. Experience at Boxworth indicated that some further studies, in particular of the ecology of farmland birds, mammals and flora could be most appropriately investigated in specific, separate trials. In the Boxworth Project, the focus was on pesticide effects on wildlife. Other important environmental effects, such as contamination of soil water with nitrates or pesticides were considered, but proved better suited to specific trials. Similarly, it was concluded that comparisons of organic and conventional systems could not be directly combined with the follow-up studies but might be investigated separately.

After an extensive planning phase two closely linked series of trials, known by the acronyms SCARAB and TALISMAN, have been started. They were designed to focus on the environmental effects of pesticide use or on the economic and agronomic implications of reducing inputs, respectively.

SCARAB: AIMS, DESIGN AND TREATMENTS

SCARAB, which stands for Seeking Confirmation About Results At Boxworth, directly derives from the Boxworth Project. It will extend current knowledge to a wider range of crops and soil types. Thus, using plots large enough to monitor mobile invertebrates, it is designed to establish the broad ecological consequences of applying two different pesticide regimes at three locations (MAFF EHFs) in representative six-course rotations that include cereals and common break crops. At High Mowthorpe, in North Yorkshire, other combinable arable crops (winter oilseed rape, spring beans) form the break crops (Table 2). The rotation at Gleadthorpe, in Nottinghamshire, includes sugarbeet and potatoes or beans. Grassland has been the subject of several studies on pesticide effects on invertebrate populations (Luff & Rushton, 1989) and at Drayton, in Warwickshire, a ley/two year cereals rotation has been included.

TABLE 2. Crop rotations for SCARAB.

Harvest year	HIGH MOWTHORPE (3 sets of paired plots)		GLEADTHORPE (3 sets of paired plots)			DRAYTON (2 sets of paired plots)	
	1 (Bugdale)	2 & 3 (Old Type)	1 (South Field)	2 (N. Kingston)	3 (Balk Field)	1 (Field 5)	2 (Field 1)
1990 (baseline)	winter barley	winter barley	winter barley	sugarbeet	winter barley	ley 4	ley 5
1991	oilseed rape	spring beans	potatoes	spring barley	sugarbeet	ley 5	winter wheat 1
1992	winter wheat	winter wheat	spring wheat	winter barley	spring wheat	winter wheat 1	winter wheat 2
1993	spring barley	winter barley	winter barley	spring beans	winter barley	winter wheat 2	ley 1
1994	spring beans	oilseed rape	sugarbeet	winter wheat	potatoes	ley 1	ley 2
1995	winter wheat	winter wheat	spring wheat	winter barley	spring wheat	ley 2	ley 3
1996	winter barley	spring barley	winter barley	sugarbeet	winter barley	ley 3	ley 4

A limited economic appraisal of input costs and yields will be made in SCARAB but this experiment is primarily environmental. Regular crop monitoring and recording of weed distribution and crop development will be done by EHF staff, with support for pest, disease and weed assessment provided by ADAS Regional Specialists. Routine invertebrate monitoring by pitfall traps and D-vac suction net will be carried out through collaboration of EHF staff, for sampling, and Southampton University, under MAFF Open Contract funding, for identification, analysis and interpretation.

In designing SCARAB, considerations similar to those encountered for Boxworth had to be addressed (Table 3). As before, plot size and degree of replication required a compromise solution. Optimum plot size for studies on polyphagous invertebrates is difficult to determine and differs for species with different behaviour and mobility (Sotherton *et al* 1988). Ideally, areas of a few hectares are advisable; Sotherton *et al,* (1987) compared gross effects between pesticides in 2-3 ha plots. Plots of a similar size, or slightly smaller, were used for work on autumn pyrethroids (Purvis *et al,* 1988) and in similar subsequent ADAS studies. The Boxworth experience emphasized the importance of field margin habitats and hedgerows as spring sources of invertebrate predators (Sotherton 1985). Hence for SCARAB, comparable plots extending about 150 m into the field from a common boundary were required.

After due consideration it was concluded that suitable paired plots, each of approximately 100 m width with buffer zones between field boundaries and each other, could be accommodated in candidate fields at High Mowthorpe and Gleadthorpe. Both these EHFs have three sets of paired plots, with two of the sets at High Mowthorpe located at north and south ends of a single large field (Table 2). At Drayton (two sets) the smaller fields available reduced the plot widths to 70 m.

Over the whole multi-site experiment, repetition (8 sets of paired plots) makes it feasible for valid comparisons of effects to be made by non-parametric statistical methods. For true replication at each site to be a practical option plot sizes well below the minimum considered necessary for field-scale realism or for assessment of a broad range of species would have to be used.

There are two treatments (contrasting pesticide regimes, Table 3):

Current Farm Practice (CFP) will be based on current practice, at the particular EHF, to represent pesticide use by a technically competent, financially aware farmer in comparable farming situations. Previous experience on the farm and an assessment of risk related to each intended crop will provide a framework of crop protection requirements for the crop year. Subsequent monitoring of pest, disease and weed problems will ensure that treatments are made at the optimum time and that routine treatments give adequate control. During the course of the experiment, some recommendations, rates or active ingredients may be superseded and the treatment components of CFP will evolve to reflect these changes.

TABLE 3. **Comparison of SCARAB, TALISMAN and Boxworth designs.**

	SCARAB	TALISMAN	BOXWORTH
Dominant observations	Monitoring of mobile invertebrates	Economic and agronomic aspects	Wide range of wildlife monitoring
Experimental areas	Large paired plots 150 m x 100 m; common boundary	24 m x 24 m plots; replicated trial	Whole fields; matched farm areas
Replication of sites	8 sets (3 High Mowthorpe; 3 Gleadthorpe; 2 Drayton)	4 trials (Boxworth; Drayton; Gleadthorpe; High Mowthorpe)	-
Experimental treatments	Pesticides: 1. Current Farm Practice - evolve during experiment 2. Reduced Input Approach - no insecticides Other treatments: Kept same across paired plots	Pesticide and Nitrogen inputs: 1. Standard Rotation I 100% 2. Standard Rotation I 50% 3. Standard Rotation II 100% 4. Standard Rotation II 50% 5. 'Green' Rotation 100% 6. 'Green' Rotation 50% (D and HM - extra 'Green' pair)	Pesticides: 1. Full Insurance - rigid 2. Supervised - regular crop monitoring 3. Integrated - additional husbandry modifications Other inputs: As required for particular field
Rotations	6-course rotation Cereals with combinable break crops (HM) Cereals with roots/beans (G) Ley with 2 year cereals (D)	6-course rotation (Standard rotations: HM and G similar to SCARAB; B and D cereals with combinable break crops)	Intensive wheat; some fields wheat with winter oilseed rape break on five-year rotation
Timescale	1990 baseline monitoring - check similarity of CFP and RIA plots 6 year experimental period	1990 baseline monitoring - check uniformity of trial 6 year experimental period	2 baseline years - check similarity of fields/area 5 year experimental period

The Reduced Input Approach (RIA) is intended to contrast with CFP in its severity on non-target invertebrate populations. It will consist of minimal (necessary) usage of fungicides and herbicides determined by regular crop monitoring and the use of managed disease control guidelines and thresholds. As a rule no insecticides will be used. A limited loss in yield from omitting insecticides is acceptable but severe reductions in plant or shoot density are particularly undesirable because the habitat and hence the abundance or activity of insects might then differ between the paired plots. Extreme crop loss might also jeopardise the credibility of the experiment. Insecticide use would be considered only where a severe threat is evident; specific insecticides, placement methods or lower rates known to have less impact on non-target species would then be used.

TALISMAN

TALISMAN (Towards A Low Input System Minimizing Agrochemicals and Nitrogen) will measure the longer term agronomic and economic effects of using lower levels of agrochemicals and nitrogen than currently recommended. The experiment follows a more conventional trials design than SCARAB (Table 3): randomised block layout; plots 24m x 24m; four replicates at Drayton, Gleadthorpe and High Mowthorpe and three at Boxworth. There are three pairs of core treatments at each site (Table 3). Two of the pairs utilize the standard six-course rotation at each site (Table 4(a)) but a different phase of this rotation is represented by each pair. Both sequences start, in 1990/91, with break crops; after six years, two full sets of data will have been obtained for each crop.

TABLE 4(a). Standard crop rotations for TALISMAN.

Year	Boxworth	Drayton	Gleadthorpe	High Mowthorpe
1	W beans	W beans	Sugarbeet	W beans
2	W wheat	W wheat	S wheat	W wheat
3	W wheat	W wheat	W barley	W barley
4	W oilseed rape	W oilseed rape	Potatoes	W oilseed rape
5	W wheat	W wheat	W wheat	W wheat
6	W wheat	W wheat	W barley	W wheat

The third pair of treatments consists of a modified rotation (Table 4(b)) requiring lower pesticide and nitrogen inputs, hence potentially more environmentally benign ('green'), than the standard rotation. An optional fourth pair of treatments (Drayton and High Mowthorpe) use a different phase of this cropping pattern.

Within each pair, one treatment will receive full pesticide and nitrogen inputs in line with current farm practice for the respective crop at each site. Nitrogen levels for each crop will be selected through 'Fertiplan', the ADAS fertiliser planning service. The other treatment will be a fifty per cent reduction in both pesticide and nitrogen treatments. This reduction will be achieved primarily by omitting whole applications but will be supplemented, where appropriate, by use of partial doses.

TABLE 4(b). Modified, lower input rotations for TALISMAN.

Year	Boxworth	Drayton	Gleadthorpe	High Mowthorpe
1	S beans	S beans	S beans	S beans
2	W wheat	Triticale	S wheat	W wheat
3	S wheat	Triticale	S barley	S barley
4	Linseed	S oats	Peas	Linseed
5	W wheat	Triticale	S wheat	W wheat
6	S wheat	Triticale	S barley	S barley

The replicated, small-plot design for TALISMAN is necessary for reliable yield measurement. A supplementary evaluation of the effect on yields of the reduction of fungicide, herbicide or insecticide inputs, in specific combinations, is planned on three blocks of the basic trial. Environmental implications from TALISMAN will be assessed through experience in SCARAB backed by a limited monitoring of the invertebrate fauna, by pitfall trapping and D-vac, in the block adjacent to the field boundary. Invertebrate populations are generally highest near field margins and only large, short-term effects on the more mobile species are likely to be distinguished under the TALISMAN design. Identification of invertebrates in TALISMAN is being undertaken by ADAS Regional Entomology Departments.

DISCUSSION

SCARAB and TALISMAN were specifically designed to pursue the results and hypotheses developed from the Boxworth Project, concentrating on the environmental effects on invertebrate species or on the economic aspects of reducing inputs (Table 3). In many respects, Boxworth was a pilot study to test possible interactions between agriculture and the environment, envisaged at the time of planning. The Topic Review and wide interest in the study, from research workers, the agrochemical and farming industries generally and policy-makers, indicated many directions that subsequent studies could justifiably take. Only a limited number of these ideas can be accommodated in the direct follow-up work outlined here. In addition, several suggestions were considered worthy of detailed study in their own right, either as specific experiments or within other wider research programmes, and may be expected to provide linking or supportive results.

The present wide interest in the interaction of farming practices and the environment highlight the necessity for a scientific basis for the rational assessment of the subject. Building on the full range of research experience in this area, these new MAFF collaborative studies should indeed help to confirm results from Boxworth (SCARAB) and lead towards lower inputs (TALISMAN) and more environmentally benign farming generally.

ACKNOWLEDGEMENT

The role of the SCARAB and TALISMAN working parties in designing the trials and in helping to summarize their development is gratefully acknowledged.

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LONG-TERM CHANGES IN NUMBERS OF CEREAL INVERTEBRATES ASSESSED BY MONITORING

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ABSTRACT

For the past twenty years, The Game Conservancy has monitored abundances of cereal invertebrates in over 100 fields in Sussex in late June. The total number of invertebrates (excluding Acari, Collembola and Thysanoptera) recorded per sample has dropped by almost half in the course of the study, corresponding to a quarter of what was present in pre-pesticide times. This overall change was the result of widespread declines in Araneae and Opiliones, Lepidoptera, Aphididae (Hemiptera), Parasitica and Symphyta (Hymenoptera), Staphylinidae, Cryptophagidae and Lathridiidae (all Coleoptera) and Lonchopteridae (Diptera); these groups constituted 72%, on average, of the total by number. Taking Staphylinidae as an example, the decline occurred equally across all farms, in *Tachyporus* and non-*Tachyporus* spp. alike and, within *Tachyporus*, across age classes and species. The staphylinid decline could not be attributed to weather, to hedgerow removal or to changes in proportions of spring and winter cereals. A possible cause was a drop in the availability of fungal food, as the level of mildew and rusts infecting crops dropped in line with the increasing use of foliar fungicides.

INTRODUCTION

Agricultural practices in Britain have changed dramatically over the past few decades. In many areas the traditional approach of rotational ley farming of small fields with mixed grazing has been largely abandoned in favour of a much more intensive approach involving the loss of grass and livestock, hedgerow removal, greater mechanisation, the use of more productive - and demanding - cereal varieties and a shift from spring to winter cereals (Jenkins, 1984). Equally dramatic has been the rise in inputs - pesticides, fertilisers, growth regulators - applied to the crops (Rands *et al*., 1988). Intuitively, these major changes in arable farming are bound to have had some impact upon the animals and plants living in and around the crops - the questions are what kind and how much of an impact, and have they had any long-term effects upon the cereal ecosystem?

By their very nature, long-term effects require long-term studies in order to be detected. This paper summarises the results from a twenty-year monitoring study of invertebrates in cereal crops on 62 km² of the Sussex Downs. This period, 1970-1989, has seen major changes in farming practices in the area, in line with the rest of the country (Potts, 1986). It started too late to monitor the impact of the introduction of herbicides (mainly late 1950s), but it spans the introduction of foliar fungicides, the loss of ley farming, the increase in insecticides and the move towards monoculture (O'Connor & Shrubb, 1986; Potts, 1986).

METHODS

The study area covers 62 km² of the South Downs, Sussex. Each year from 1970 onwards, invertebrates in cereal crops were sampled by vacuum suction trapping (Dietrick 1961) in the third week of June. Approximately 100 fields were sampled annually, and each sample, comprising 5 subsamples, corresponded to an area of 0.46 m². At the same time as the invertebrates were sampled, crop type and levels of disease present in the crop (on an ordinal scale of 0=no disease to 6=entire crop infected) were noted. Further details of the area and methodology are given in Potts & Vickerman (1974), Potts (1986) and Aebischer (in press).

The results presented here derive from a 28 km² core area of five farms whose cereal fields were sampled systematically throughout the study period. Analysis was carried out using annual mean numbers of invertebrates from each farm, transformed to logarithms (base 10) and weighted by sample size (Aebischer, in press). Long-term increases or decreases were detected by linear regression of the logarithmically transformed means against time, after adjusting for between-farm differences (Kendall, 1976). The annual rate of change was obtained as 10^r-1, where r was the regression coefficient.

Continuous series of daily temperature and rainfall figures from 1969 to 1989 measured at Worthing Meteorological Station, about 5 km south of the centre of the study area, were used to calculate mean temperatures and total rainfall by calendar month and for the month before sampling, as well as sums of day-degrees above 0 °C, 1 °C, etc. to 10 °C from 1 January to the median sampling date.

RESULTS

Summary of trends

The overall trend in numbers of cereal invertebrates, excluding mites (Acari), springtails (Collembola) and thrips (Thysanoptera), which were not counted in all years of the study, was significantly downward (Fig. 1). The mean number of invertebrates recorded per sample during the first five years of the study (1078/m²) was almost twice as high as during the last five years (563/m²); the rate of the decline averaged 5.3% per annum.

These general figures cover a wide variety of invertebrate taxa, and the next step is to examine whether all declined in the same way, or whether the overall decline was the result of declines in particular taxa or trophic groups. The invertebrates were therefore split into five main groups of relevance to pest management and conservation: aphids (Aphididae), parasitoid wasps (Parasitica - predominantly aphid parasitoids), aphid-specific predators (Coccinellidae, Cantharidae, Neuroptera, Syrphidae), polyphagous predators (Araneae, Carabidae, Staphylinidae, Dermaptera, predatory Diptera) and chick-food items, i.e. taxa upon which partridge and pheasant chicks depend for survival during the first two weeks of life (Symphyta, Lepidoptera, non-aphid Hemiptera, Chrysomelidae, Curculionidae - Potts & Aebischer, 1989). These groups constituted 69% of the total by number. A sixth, miscellaneous, group included the remaining families which were numerically important in the

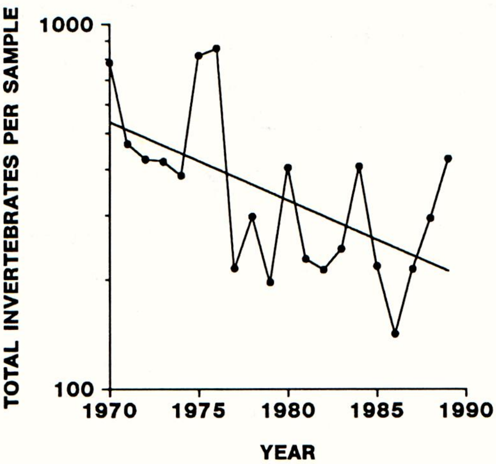

FIGURE 1. Mean numbers (logarithmic scale) of invertebrates, excluding Acari, Collembola and Thysanoptera, recorded per sample in the Sussex study in each of the years 1970 to 1989. Numbers declined significantly in the course of the study, at an average rate of 5.3% ($F_{1,18}$=11.0, P<0.01).

Sussex samples (arbitrarily defined as making up at least 2% by number of the captured invertebrates); it made up a further 21% of the total.

The numerical importance of the different groups and the long-term trends which they exhibit are summarised in Table 1. It is clear that the overall change in invertebrate abundance was caused by wide-ranging declines across the various taxa. Out of the six general groups defined above, declines occurred in the four numerically most important ones (aphids, parasitoids, polyphagous predators, miscellaneous). Among the heterogeneous group of polyphagous predators, the decline can be traced to decreases in numbers of spiders (Araneae) and of rove beetles (Staphylinidae); in the miscellaneous group, whose taxa were selected on the basis of their abundance, three out of five had declined: two families of small beetles (Cryptophagidae, Lathridiidae) and one of flies (Lonchopteridae). Finally, one of the most important component taxa in the group of chick-food items, sawflies (Symphyta) and Lepidoptera (Potts 1986), also showed a significant downward trend. Numerically, the taxa in decline accounted for 72%, on average, of the invertebrates recorded in cereal fields in Sussex. The annual rates of decline varied from 4.1% to 12.7%, with a mean of 6.8%. This corresponded approximately to a halving of abundance over ten years.

As yet, the causes of the declines are unclear in most cases. In the next section, we concentrate on one particular group, the rove beetles (Staphylinidae) to demonstrate some features of their decline which recur in other groups, to illustrate some of the difficulties in interpretation, and to investigate a possible role of pesticides as causative agents.

TABLE 1. Summary of long-term trends among a variety of invertebrate taxa present in the Sussex samples from 1970 to 1989. The taxa were chosen as belonging to five biologically relevant groups, plus a miscellaneous group of numerically important taxa which did not fit into the other groups.

Taxon	% in samples (by number)		Trend		Annual rate of change (%)	
Total (excluding Acari, Collembola, Thysanoptera)	100.0		Down		-5.3	
Aphididae	37.2		Down		-8.4	
Parasitoid wasps (Parasitica)	15.0		Down		-4.7	
Aphid-specific predators	0.5		None		-	
Coccinellidae		0.3		None		-
Others		0.2		None		-
Polyphagous predators	13.6		Down		-3.8	
Carabidae		0.5		None		-
Staphylinidae		6.5		Down		-7.7
Araneae, Opiliones		3.0		Down		-4.1
Dolichopod-,Empid-,Scathophagidae		3.6		None		-
Others		<0.1		None		-
Chick-food items	3.0		None		-	
Symphyta, Lepidoptera		0.5		Down		-4.5
Hemiptera (excl. Aphididae)		2.2		None		-
Chrysomelidae, Curculionidae		0.3		None		-
Miscellaneous	21.1		Down		-6.9	
Cryptophagidae		4.9		Down		-6.7
Lathridiidae		2.4		Down		-12.7
Cecidomyiidae		8.2		None		-
Lonchopteridae		2.4		Down		-5.4
Drosophilidae		3.2		None		-

Staphylinidae

The decline in rove beetles was replicated across all farms (Fig. 2): there was no significant difference between the slopes of the regression lines ($F_{4,90}$=0.25, n.s.), so that, on average, the rate of decline was the same on all farms. There were also similarities in year-to-year variation among the farms: for instance, common peaks occurred in 1972-1973 and 1984-1985, common troughs in 1975, 1983, 1986, 1989. Such features - similar long-term trends and similar patterns of year-to-year variation - were to be found in all the taxa examined in Table 1. Because of the similarities between farms, the data in subsequent analyses were pooled across farms.

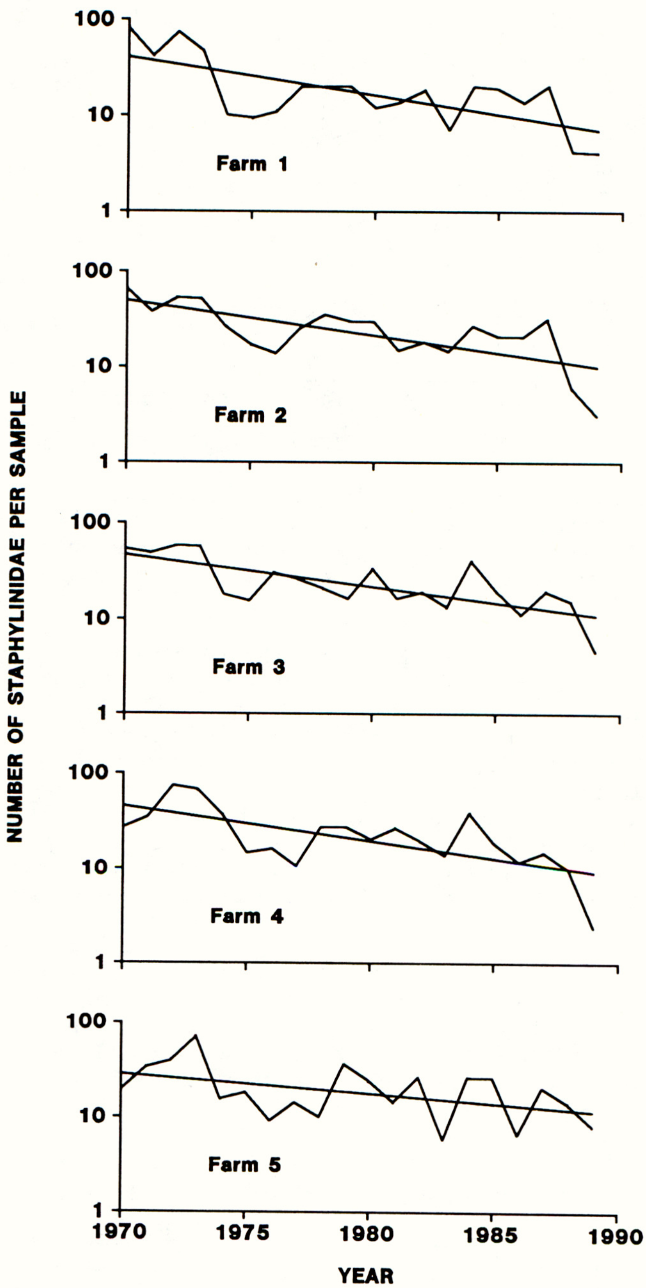

FIGURE 2. Annual mean numbers (logarithmic scale) of Staphylinidae recorded per sample on each of five farms in the Sussex study area from 1970 to 1989. The average rate of decline in the course of the study was 7.7% ($F_{1,94}$=72.9, P<0.001).

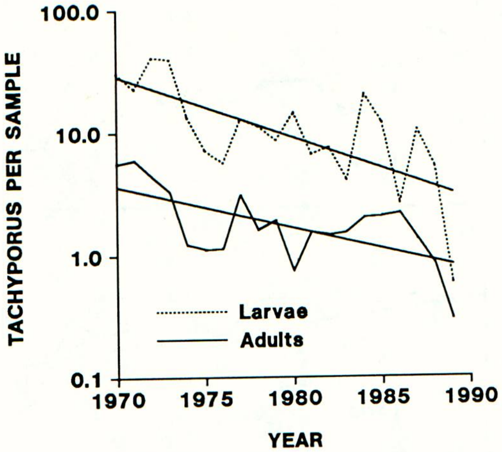

FIGURE 3. Mean numbers (logarithmic scale) of adult and immature *Tachyporus* recorded per sample in the Sussex study in each of the years 1970 to 1989. Abundances in both age classes declined significantly in the course of the study, at an average rate of 9.2% ($F_{1,37}$=32.5, P<0.001).

Within the rove-beetle complex, 65% of individuals belonged to the genus *Tachyporus*, and this genus was examined more closely. Figure 3 shows that the abundance of both adults and larvae was reduced during the period of the study. There was no significant difference in the slopes of the two regression lines ($F_{1,36}$=1.23, n.s.): the ratio of larvae to adults remained approximately constant at about 6:1, implying that changes in breeding rate were not responsible for the decline. According to identification based on adults, the genus was represented by four species in the samples: *T. chrysomelinus*, *T. hypnorum*, *T. nitidulus* and *T. obtusus*. Numbers of adults of all four species declined at similar rates (Fig. 4).

The overall decline in numbers of rove beetles was not, however, produced solely by the decrease in numbers of *Tachyporus*. The remaining, non-*Tachyporus*, rove beetles also showed a significant decrease in abundance ($F_{1,18}$=12.4, P<0.01), at an average annual rate of 5.8%. Although the latter was lower than the rate of decline of *Tachyporus* adults and larvae (9.3%), the two did not differ significantly ($F_{1,55}$=2.46, n.s.).

Possible causes of the staphylinid decline

Is it possible to discover the cause of the decline in rove beetles? The similarity in the effect across the five farms suggests that whatever the cause, it must have acted on a broad geographical scale rather than at the field or even the farm level. Therefore it is likely to be either a climatic effect or else a change in farming practice so widespread that it was effective throughout the study area.

The influence of climate was investigated using the daily weather records from Worthing Meteorological Station. None of the obvious relevant climatic variables, ranging from temperature and rainfall through

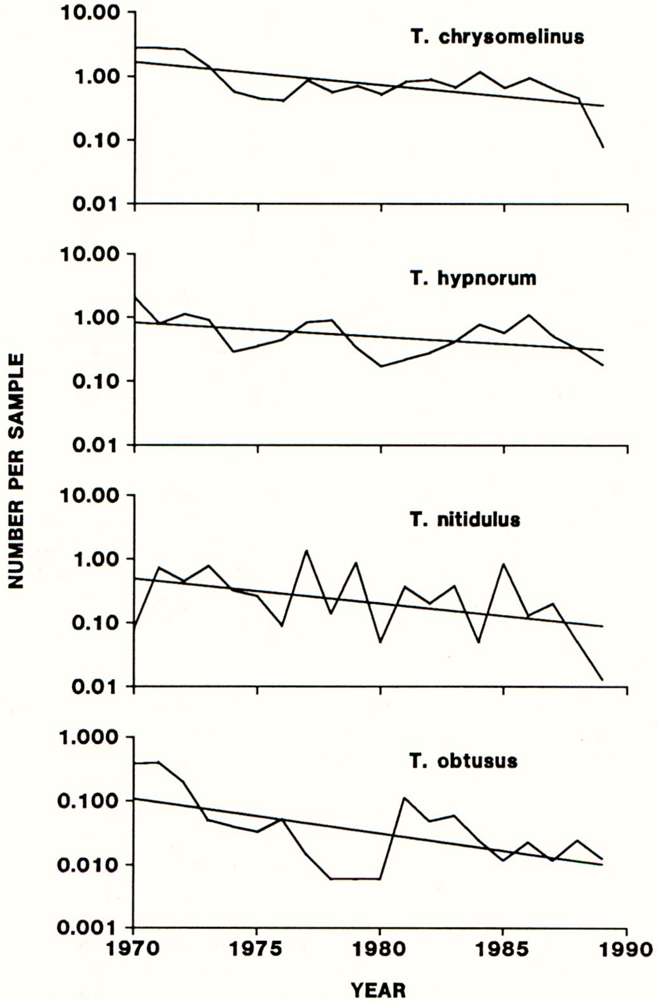

FIGURE 4. Annual mean numbers (logarithmic scale) of each of the four species of *Tachyporus* represented in the Sussex samples from 1970 to 1989. In each case numbers declined in the course of the study, at an average rate of 9.1% ($F_{1,75}$=26.3, P<0.001).

to day-degree sums, were able to account for the observed decline. This focused attention onto the farming methods.

One change which took place across the board on the study area was a gradual switch from spring cereals to winter cereals (Aebischer in press). However, the decline in numbers of rove beetles occurred at the same rate in winter wheat, winter barley and spring wheat, and there was no evidence that numbers were higher in spring barley than in either of the winter cereals. Therefore this change could be discounted as a reason for the decline. The destruction of overwintering habitat (Sotherton, 1984;

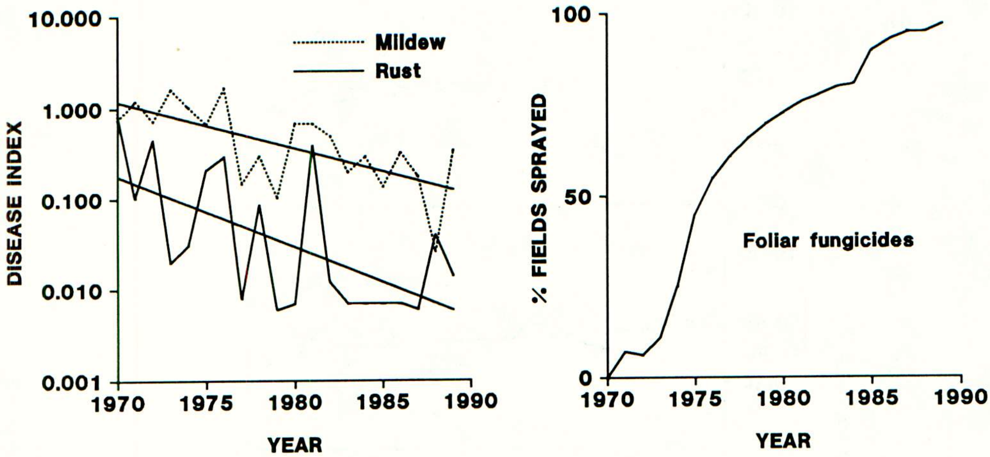

FIGURE 5. Mean index (logarithmic scale) of mildew *(Erysiphe graminis)* and of rusts *(Puccinia* spp.) in cereals on the Sussex study area for each of the years 1970 to 1989 (left), and corresponding increase in the proportion of fields in the study area treated with foliar fungicides (right). Both disease indices declined significantly in the course of the study (mildew: $F_{1,18}=14.3$, $P<0.01$; rusts: $F_{1,18}=9.06$, $P<0.01$).

1985) was also an unlikely cause, as the removal of hedgerows and ploughing up of grass banks occurred on only two of the five farms over the past 20 years (Potts, 1986; Aebischer, in press).

Another possibility was that changes on the farms wrought changes in the food supply of the beetles. As well as being predatory on aphids, the rove beetles, and *Tachyporus* spp. in particular, feed on fungal material, mainly mildews *(Erysiphe* spp.) and rusts *(Puccinia* spp.) (Sunderland, 1975; Dennis *et al*. in press). Figure 5 shows that the incidence of mildew *(E. graminis)* and of rusts has gone down significantly since the study began, while the proportion of fields treated with foliar fungicides increased. The latter were applied on less than 10% of cereal fields until 1974, when the proportion rose rapidly; in recent years, practically all cereal crops were treated as a matter of course. The disease indices suggest that the treatment was effective, as rusts have almost disappeared from the crops, and the level of mildew is much reduced.

Other explanations are possible, such as direct toxic effects of insecticides, increasingly used to control cereal aphids, or a rise in levels of parasitism by parasitoids or nematodes. However, the widespread use of insecticides did not take place until the 1980s on the study area, and we have no information on rates of parasitism (Potts, 1986).

DISCUSSION

Although the Sussex monitoring study did not start until after one of the first big changes in crop management, the use of herbicides, there have been considerable changes in the densities of the invertebrates recorded over the past twenty years. This paper shows that the changes give cause for concern: overall, the abundance of cereal invertebrates (excluding mites, springtails and thrips) dropped by approximately half in the course of the study, and the decrease is the result of declines across a wide range of taxa, from agricultural pests such as aphids, to beneficial arthropods such as spiders and rove beetles. As the effect of herbicide use has been estimated as causing a 50% reduction in invertebrate abundance in cereals (Southwood & Cross, 1969; Potts, 1986), this means that the number of invertebrates in crops now represents only about a quarter of the number present before the pesticide era.

The causes of the declines highlighted by this study are not easy to establish, and need not necessarily be the same for all taxa. Characteristically, the factors responsible for the declines were acting on a broad geographical scale, so that obvious candidates were climatic change on one hand, and widely adopted changes in agricultural practices on the other. Thus Aebischer (1990) found that the decline in sawflies shown in Table 1 was accounted for by changes in summer temperature and rainfall in conjunction with the disappearance of undersowing (cereals used as a nurse for grass). For rove beetles, the only explanation which, so far, seems plausible is that the rapidly increased use of fungicides from 1974 onwards reduced levels of the pathogenic fungi upon which the beetles feed. It is interesting that two other groups of mycophagous beetles, the Cryptophagidae and the Lathridiidae (Potts & Vickerman 1974), also showed sharp declines in abundance since the beginning of the study (Table 1).

The prime role of monitoring is to serve as a warning system. The main advantage of monitoring population densities, as described here, is that it is successful in detecting population change (cf. Table 1) for a relatively small outlay of time and resources. It is, however, a "black-box" approach, in that it records the outcome of internal processes that remain unknown, and that require much additional effort to assess effectively. Assuming that change is taking place, the time taken to detect it will vary from species to species. This depends upon the type of change (step or trend), the magnitude of the change, the accuracy of sampling, the range of natural between-year fluctuations and the confounding effects of different factors, in particular weather. The possible causes of change may be identified through a correlative or regression approach, although from the statistical viewpoint, potential problems of serial correlation between years require the use of time-series modelling, for which the 20-year span of the Sussex data is only just sufficient (Aebischer, 1990). Provided that the monitoring is extensive enough to encompass within-year as well as between-year variation, it should be possible to separate the effects of farming methods varying between farms from climatic effects. Once tentative causes have been identified though correlation, the demonstration of cause and effect must then lie with experimentation.

The Sussex study is currently sounding an alarm over the cereal ecosystem. Because of the vast area of Britain under cultivation (4 million ha), we cannot afford to ignore it. Many members of its invertebrate community are valuable tools for integrated pest management, and many form the basis of a complicated food chain upon which many birds and mammals rely. Amid the growing public concerns about the effects of pesticide and nitrate pollution, the Sussex study adds further cause for urgent research into the ecological implications of modern agriculture.

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EFFECTS OF PESTICIDE APPLICATIONS ON SMALL MAMMALS IN ARABLE FIELDS, AND THE RECOVERY OF THEIR POPULATIONS

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ABSTRACT

A series of experimental field trials was conducted by MAFF between 1977 and 1988 at Bridgets EHF, to evaluate the hazards of pesticide applications for small mammal populations, particularly wood mice *Apodemus sylvaticus*. They covered insecticidal seed treatments, a herbicide spray, and molluscicide pellets. Wood mice were trapped and removed from the site for laboratory investigation, including autopsy examination, histology, biochemical assays and residue analysis. A contrasting, ecological approach was taken in a 7-year study at Boxworth EHF, tracing long-term effects of pesticides on wood mouse populations. Together, these studies demonstrate the value, and potential difficulties of interpretation of studying small mammals in pesticide field trials.

INTRODUCTION

Among the potential environmental consequences of pesticide use, effects on populations of small mammals (mice, voles and shrews) are of concern both for the welfare of these species and for possible secondary poisoning of their predators. The small size and secretive habits of these animals mean that even major impacts on populations might not be noticed by the casual observer, nor be detected through monitoring such as the ADAS Wildlife Incident Investigation Scheme (Greig-Smith 1988). It is therefore all the more important that potential adverse effects should be identified, and taken into account in the course of pre-approval testing of new products or new uses of pesticides.

The MAFF Central Science Laboratory has been involved in a series of field studies to examine the effects on resident small mammals of pesticides applied to arable fields. This has involved a variety of insecticides, molluscicides and herbicides, in various formulations (sprays, seed treatments, pellets). Additional work on molluscicide pellets was carried out independently by Cambridge University, as part of the 'Boxworth Project' (Greig-Smith 1989), a major experimental study of the environmental consequences of high pesticide inputs on cereal crops. Taken together, these studies provide examples of the range of methods available to investigate small mammals and pesticides. They also reveal some of the complicating factors that may limit interpretation of results if these are not derived from a multi-disciplinary approach, combining ecological analysis of populations with toxicological study of individual animals. This paper reviews the principal findings from eight separate MAFF field trials at one location, and a continuous seven-year study at a different site. We address both the scale of initial impacts on small mammal populations, and their recovery.

FACTORS AFFECTING PESTICIDE IMPACTS

The nature and scale of effects on non-target animals depend on the toxicity of the pesticide product used and the animals' exposure to it. Although toxicity data are rarely available for the wild mammal species likely to occur on arable fields, the greatest uncertainty lies in estimates of likely exposure (Somerville and Walker 1990). This is because exposure is highly variable, depending on local features such as: population density; the distribution of refuges of non-crop habitat; the behaviour of individual animals, influencing the amount of time spent on fields; the availability of food supplies; and the time of year at which the application is made. Characteristics of the pesticide are equally important. The likelihood of exposure is affected not only by the application rate, but also by the persistence and pattern of breakdown of the active ingredient. Some formulations may be ingested directly (e.g. palatable pellets or treated seeds) or through grooming after contact with contaminated vegetation, whereas others may cause secondary poisoning through consumption of poisoned invertebrates.

How serious such exposure is depends on the susceptibility of individual animals, which is liable to differ not only between species, but also among age-classes, and between males and females. Impacts may be greater, and recovery slower, at some times of year. If a pesticide is applied in autumn, when the annual cycle of numbers is reaching a peak, and juveniles are dispersing, local population recovery may occur rapidly by immigration. In contrast, an application early in the year, when the population is made up of small numbers of over-wintering adults, may cause a longer-lasting impact because recovery by recruitment is hindered.

As well as these considerations, care must be taken to allow for annual fluctuations in population size, and in the species composition of the small mammal community. There may be major changes even at one site, which limit the confidence that can be placed on the generality of results from isolated, single-season trials.

SPECIES OF SMALL MAMMALS ON ARABLE FIELDS

Most MAFF trials have been conducted at Bridgets Experimental Husbandry Farm (EHF) in Hampshire, using a number of similar arable fields with nearby hedgerows and woods suitable for a wide range of small mammals. The species and numbers trapped at Bridgets EHF in eight trials between 1977 and 1988 are listed in Table 1. This shows that wood mice *Apodemus sylvaticus* were the most common, but yellow-necked mice *Apodemus flavicollis* (of which it can be difficult to distinguish juveniles with certainty from juvenile wood mice) were also frequent in some trials. The relative numbers of these species, and of *Apodemus* compared to bank voles *Clethrionomys glareolus* and common shrews *Sorex araneus*, varied greatly from year to year, in ways that were not obviously related to changes in habitat, nor to the time of year. This prompts a caution about judging the typicality of conditions in a single trial, and emphasises the need to obtain good pre-trial control data (Greig-Smith & Westlake 1988).

TABLE 1. Species of small mammals trapped in field trials at Bridgets EHF. Figures are the numbers of captures of the species as a percentage of total captures.

	Autumn 1977	Autumn 1978	Autumn 1979	Spring 1981	Autumn 1986	Autumn 1987	Autumn 1988
Wood mouse *Apodemus sylvaticus*	51	34	38	56	95	90	33
Yellow-necked mouse *Apodemus flavicollis*	12	17	35		4	6	60
Bank Vole *Clethrionomys glareolus*	36	42	22	9		1	7
House mouse *Mus musculus*					1	3	0.2
Common shrew *Sorex araneus*		6	5	4			
Pygmy shrew *Sorex minutus*		2					
Harvest mouse *Micromys minutus*					0.5		
Field vole *Microtus agrestis*				32			
TOTAL CAPTURES:	**354**	**590**	**321**	**57**	**425**	**105**	**406**

Similarly, at Boxworth EHF in Cambridgeshire, there were major year to year fluctuations in population densities of wood mice (Fig. 1). Although the habitat was similar in many respects to that at Bridgets, yellow-necked mice were not recorded at Boxworth, which is near the limit of the species' range in the UK.

EFFCTS OF PESTICIDES ON WOOD MICE

Partly because of its general abundance, and regularity of occurrence on fields, the wood mouse has been chosen as the most appropriate species for special study (Westlake *et al*. 1980). It may not be an 'indicator' species in the sense of representing the hazards for all small mammals, because exposure and its consequences may be different for entirely herbivorous species such as voles. However, wood mice range widely onto arable fields, and are likely to be exposed to most types of pesticide use. In North America, both deer mice *Peromyscus* spp. and voles *Microtus* are used in a similar way (Jett 1986).

FIGURE 1. **Mean density of wood mice caught on three 2 ha trapping grids at Boxworth EHF in July/August 1983-88.**

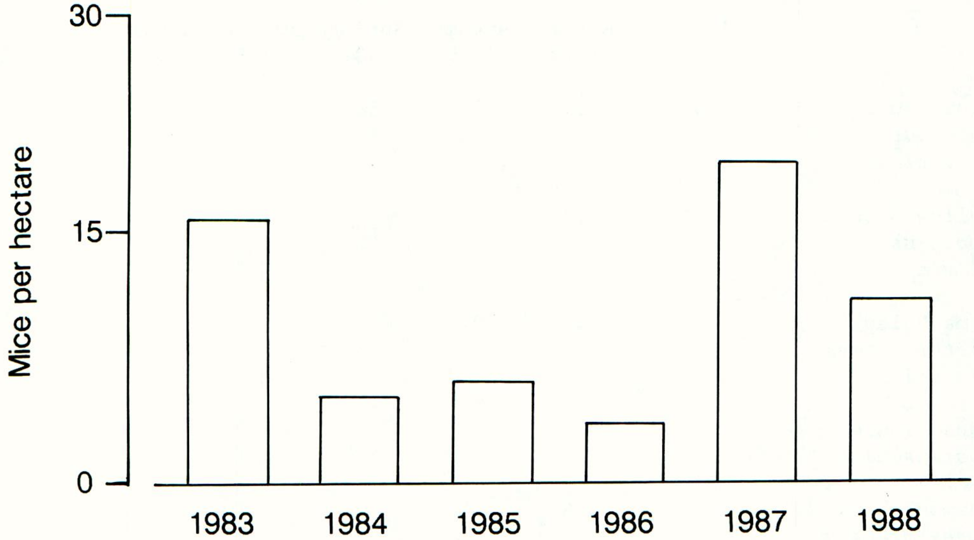

Table 2 summarises the trials at Bridgets EHF, indicating the pesticides involved, and some of the data collected to assess exposure and effects. Small mammals were captured in grids of 'Longworth' or other live traps. In most of these trials, a proportion of the wood mice trapped live was removed for laboratory examination, which included (i) inspection of the body and organs for gross signs of pathological damage, (ii) histological study of tissues, particularly livers, (iii) biochemical assays of brain acetyl cholinesterase (AChE) inhibition, and (iv) chemical analysis for the presence of pesticide residues, in the guts, or tissues. Removal of mice for these purposes caused difficulties for ecological interpretation of the trapping data (Greig-Smith & Westlake 1988), but for some trials it was possible to calculate an index comparing the rate of capture of wood mice just after pesticide application with that just before (see Table 2). More detailed ecological information was obtained at Boxworth EHF, where no wood mice were removed from the population for laboratory studies.

<u>Herbicide Spray</u>

In March 1981, a single application of diclofop-methyl was made as a post-emergence herbicide on a 13 ha field of spring barley. Residues of the chemical in samples of barley declined from 79.8 mg/kg just after spraying (Day 0), to 14.8 mg/kg on Day 5, and 1.1 mg/kg on Day 14; a small residue level was still detectable on Day 28. Wood mice and voles were trapped throughout this period, but no residues were detected in the guts analysed. However, relative liver size (i.e. liver weight/body weight) and plasma enzyme levels were significantly elevated in all the wood mice sampled up to Day 28, and histological evaluation revealed increases in liver cell size and binucleation, which are indicative of a protective response to the presence of a contaminant (Tarrant 1988).

TABLE 2. Summary of pesticide exposure and effects on wood mice in eight experimental field trials at Bridgets EHF.

Date	Active Ingredient+	acute oral LD_{50} to rats (mg/kg)	% wood mice with gut residues	% wood mice with brain AChE inhibition	capture rate index*
a 1977	chlorfenvinphos (I)	10	37%	7%	0.89
a 1978	carbophenothion (I)	6.8	61%	41%	1.22
a 1979	bendiocarb (I)	40	46%	20%	0.56
s 1981	diclofop-methyl (S)	563	0%	-	-
a 1986	XL 79 (M)	c.50	10%	4%	1.50
a 1987	methiocarb (M)	15	0%	0%	1.14
a 1987	metaldehyde (M)	600	0%	-	2.67
a 1988	methiocarb (M)	15	32%	0%	1.00

* Values less than 1 indicate a reduced capture rate after application.

\+ I = insecticide seed treatment; S = herbicide spray;
M = molluscicide pellet

a = autumn; s = spring

Field voles showed transient but significant decreases in plasma enzymes and in brain AChE (which was unaffected in wood mice), as well as similar, though less marked histological changes.

This trial demonstrates exposure of both species to the spray, accompanied by sublethal effects on physiology for about a month after application, while residues of the chemical remained on the field.

<u>Seed Treatments</u>

Three studies concerned the effects of insecticidal seed treatments on autumn-sown cereals. In 1979, bendiocarb was used as a seed dressing on wheat; residues were initially 140 μg per grain, but fell to 11 μg per grain on Day 11. There were no effects on tissue-derived enzymes during the 44 days of the trial. Overall, 46% of the wood mice sampled contained bendiocarb-derived residues, but these were low (0.01 - 0.1 mg/kg) and were probably not high enough to have threatened survival. However, the recapture index value of 0.56 shows that fewer wood mice were captured after application, suggesting the possibility of an effect at the population level.

A trial involving carbophenothion was carried out in the autumn of 1978 (Westlake *et al* 1982). Residues of the compound remained high on the drilled wheat up to Day 159 (100 ppm) and 61% of wood mice contained detectable residues, with 41% having significant depression of brain AChE. Nevertheless, the recapture index was greater than one, indicating a lack of immediate heavy mortality after sowing.

In contrast, residues of a seed treatment containing chlorfenvinphos, applied under similar conditions in 1977, showed a rapid decline to only 5 ppm on Day 30. Residues detected in wood mice reflected this decline over a 35 day period (Westlake *et al* 1980). Only 7% of the mice sampled had depressed levels of brain AChE activity.

These studies demonstrate a high level of exposure to seed treatment chemicals, with variable effects, depending on the toxicity of the active ingredients and on their environmental persistence.

Molluscicide pellets

A trial was conducted in 1986 to evaluate an experimental pellet formulation containing the active ingredient XL 79, which was applied by surface broadcast on three occasions between October and December. Some wood mice showed evidence of having eaten the pellets (blue dye in their stomach contents) and residues of the pesticide were detected in 10% of the animals sampled, but only 4% had significantly depressed levels of brain AChE activity. More casualties were discovered after the December application than in October, perhaps suggesting that the pellets were more attractive when natural food supplies had been depleted.

One trial in the autumn of 1987 was concerned with the use of metaldehyde pellets on a newly-sown field of winter cereals. There was no indication of exposure (none of the wood mice analysed contained detectable residues of metaldehyde) nor of adverse effects on individual wood mice or on the population.

In two further trials, in 1987 and 1988, single surface-broadcast applications of methiocarb-based slug pellets were studied. In 1987, no residues were detected in wood mice, nor were there any substantial effects on brain AChE activity. Also, rates of capture, sex-ratios, and the age-structure of the population were no different on the treated field to a separate 'control' field.

The following year's trial produced different results. Although post-mortem examination did not reveal signs of wood mice having fed on the pellets, 32% contained methiocarb residues. Brain AChE was not significantly depressed, but 8 animals did show spontaneous reactivation of AChE, consistent with exposure to a carbamate compound (see Martin *et al*. 1981). However, the number of wood mice affected by methiocarb was low enough to allow the conclusion that the local population was not endangered. Capture rates were similar before and after application. Together, these two trials show that wood mice may be exposed to surface applications of methiocarb pellets, but effects can differ from year to year, even under apparently similar conditions at the same site.

In contrast to the investigations of methiocarb pellets at Bridgets EHF, directed primarily at measuring exposure and short-term population changes, the study at Boxworth EHF was concerned chiefly with longer term population stability. The experimental regime at Boxworth included some fields which received a surface broadcast application of molluscicide every autumn as a pre-planned 'insurance' treatment, and some which were treated only if slug problems rose above a threshold level (see Greig-Smith 1989). Comparisons between treated and untreated fields in several years revealed severe declines in the numbers of wood mice caught, and in the survival of individual mice marked by fur-clipping before application (Johnson *et al* 1991; Table 3). This strongly suggested that wood mice present on the fields were killed by poisoning after feeding on the pellets, which contain a nutritious cereal base. (The fact that all four wood mice found dead on the fields contained no residues of methiocarb is puzzling, in view of the residues detected in wood mice at Bridgets EHF).

TABLE 3. **Effects of broadcast molluscicide pellets on wood mice at Boxworth EHF, shown by the rates of capture and adult:juvenile ratios before and after applications.**

	Before application	2-4 days after	7-27 days after
rate of capture (mice per 100 traps)	4.33	0	4.58
adult : juv ratio	23:4	-	11:18

TABLE 4. **Comparison of the effects of surface-broadcast and drilling methiocarb pellets with seed, shown by the proportions of individually-marked wood mice that were recaptured after application. Data from Johnson *et al*. 1991.**

	Treated field	Untreated control field
surface broadcast (1988)	5/29 (17%)	29/68 (43%)
drilling with seed (1987)	15/52 (29%)	8/45 (18%)

Although wood mice at Boxworth EHF were apparently killed by methiocarb poisoning, the population level was scarcely affected, because of rapid replacement by immigrants from other habitats. Within two weeks, wood mouse density was restored to its previous level, and there was no long-term difference between the 'Full Insurance' area, where molluscicide was applied annually, and the reduced-input areas, where applications were made only occasionally (Johnson *et al*. 1991).

The problem experienced at Boxworth EHF suggested that the ready availability of molluscicide pellets on the field surface was the cause of the exposure of wood mice to methiocarb. It might, therefore, be possible to eliminate the problem by preventing the mice's access. A comparison of trapping on fields where pellets were broadcast with those where pellets were drilled with the cereal seed (Table 4) confirmed that this was the case; survival of marked mice was no lower than on untreated control fields.

THE VALUE OF LABORATORY TESTS

Alongside these studies in the field, several laboratory experiments have been carried out, to help establish information on toxicity, and the willingness of small mammals to ingest pesticide formulations. Thus, Tarrant & Westlake (1988) demonstrated differences in the palatability, and the toxic effects, of wet and dry slug pellets, by offering pellets to captive wood mice. This kind of complementary study (see also Westlake *et al.* 1982, 1988, Linder & Richmond 1990) can be an important aid to the interpretation of field trial data.

DISCUSSION

In wildlife ecotoxicology, it is generally easier to establish the toxicity of a pesticide (by direct testing, or by extrapolation from a surrogate species) than it is to predict or measure exposure of species whose cryptic habits make observation impractical. The field trials outlined in this paper demonstrate that it is possible to base a variety of diagnostic techniques on simple live-trapping of small mammals on and near treated sites. However, the value of this approach lies in its multi-disciplinary nature; each type of measurement on its own may be open to uncertain interpretation. For example, the range of studies on methiocarb slug pellets shows that the contrasting protocols for ecological and biochemical/residue data collection may produce different conclusions about the hazard to wood mice.

The results also illustrate that effects may differ between years, even at a single site. This is likely to be at least partly due to weather factors and to fluctuations in food supply. However, it may also reflect changes in population density and species composition of the local small rodent community, as a consequence of the annual changes of density in some species (Flowerdew 1985). The implication is that tests carried out in one year cannot be assumed to be generally representative. If it is not possible to study populations for several years, as at Boxworth, the typicality of conditions may be assessed crudely by reference to trends in national monitoring programmes, or by measuring relevant environmental factors (e.g. food availability), provided that the mechanisms of their influence are sufficiently understood.

Particularly because of the variation from place to place and from time to time, it is important to consider not only the initial impact of a pesticide on a small mammal population (e.g. mortality), but also the period of recovery, and the mechanism by which it takes place (recruitment, or immigration). This is heavily dependent on the environmental persistence of the pesticide, which may continue to cause new exposure long after the application, as in the case of carbophenothion. It is also likely to be affected by the availability of refuges from which immigrants can recolonise; if a treated field is close to natural habitat, a large population of small mammals may be potentially at risk, but few of them may spend much time on the field, and recolonisation is likely to be rapid. Clearly, the problems of accurately predicting risks are great.

Despite these difficulties of interpretation, the trials at Bridgets EHF provide a unique opportunity to compare the hazards of different pesticide uses on wood mice at a single locality. Table 2 lists the toxicity of the seven products tested at Bridgets (from Worthing 1987 and Smith 1987) and the exposure experienced by wood mice, shown by the proportion of the animals trapped that contained gut residues. Overall, it appears that wood mice were most likely to be exposed to the most toxic products, but this pattern was due only to their high exposure to seed treatments, presumably as a result of the attractiveness of seeds as food. However, the variability is such that each product must be examined as a separate case.

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