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SELECTIVE ACTION OF FUNGICIDES AND DEVELOPMENT OF RESISTANCE IN FUNGI TO FUNGICIDES

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Summary Selective action with respect to the combination of host and parasite is a prerequisite for systemic fungicides. Most of these fungicides appear to act selectively on fungi. This may cause a shift in the microbiological balance in the soil or on the plant surface by elimination of sensitive organisms or by a change in the antagonistic properties of tolerant ones. It appears that resistance to fungicides, which interfere rather selectively with the metabolism of fungi, may develop more readily than to non selective toxicants. Resistance mechanisms are discussed in relation to the sites of action of selective fungicides: chitin synthesis in cell wall, cell membrane, energy production in mitochondria, protein synthesis on ribosomes, and nucleic acid metabolism.

Emergence of resistance in the laboratory does not necessarily mean that resistance problems will be encountered under field conditions. Moreover it should be possible to develop systemic fungicides, to which resistance does not emerge easily. Provided that resistance problems can be kept within bounds and application occurs judiciously, systemic fungicides may become of great value for plant disease control in the future.

INTRODUCTION

As epidemics of fungal plant diseases may cause severe fluctuations in yield or even annihilate a crop in a certain area, measures of various kinds have to be taken to insure against hazards. Among the most important ones are the breeding of resistant varieties and the use of fungicides. Although the use of resistant varieties seems most attractive to the grower, since in that case special control measures may be omitted, breeding for resistance can not solve all phytopathological problems. As the selection of a new variety and the multiplication of seed for release to the grower usually takes 10 years or more, it usually cannot provide a short term answer to a disease problem. Moreover, if no genes for resistance are available in a certain crop, incorporation of resistance genes from other sources may require many back crosses to obtain a variety with commercially acceptable qualities, and in some cases lack of sources of resistance even may preclude breeding for resistance.

When resistant varieties are not or not yet available, the use of fungicides may provide immediate relief. Control by fungicides is usually obtained by prevention, the fungicide being applied as a cover on plant parts or seeds. The use of these classic types of fungicide, however, does have some shortcomings. As a rule, they do not exert a curative action, once the fungus has penetrated into the plant tissue, and are ineffective against deep seated infections of seeds and vascular pathogens. Moreover they have to be applied rather frequently because of washing off by rain and the appearance of new, unprotected growth.

This situation has prompted a search for fungicides which are absorbed by the plant and subsequently transported in the system of the plant, and protect also plant parts which have not been hit by the fungicide. This has resulted in the discovery of various chemicals with systemic action against fungal diseases, so called "systemic fungicides" or "systemics". In recent years some of these have reached the phase of practical application.

As systemic fungicides penetrate in the cells both of parasite and hostplant, a highly selective action is required. The fungus should be killed or its growth inhibited without injury to the plant. It appears that most systemic fungicides act rather selectively also with respect to fungi. Some fungi or groups of fungi are highly sensitive, others rather tolerant to a particular systemic.

The consequences of this selective action with respect to the microbiological balance, and its relation to the phenomenon of acquired resistance to fungicides will be discussed.

SELECTIVE ACTION

The systemic fungicides which are in practical use act rather selectively on fungi. With respect to benomyl, Bollen and Fuchs (1970) observed a remarkable correlation between sensitivity to benomyl and taxonomic position, especially with respect to sporogenesis. Completely or relatively resistant fungi belong to the Oomycetes and Zygomycetes and to those groups of the Moniliales, which are characterised by producing porospores or annellospores. Sensitive fungi belong to the Ascomycetes, the Sphaeropsidales and to those groups of the Moniliales, which are characterised by forming blastospores, phialospores and aleuriospores. A similar relation exists for thiophanate-methyl, which is not surprising since this compound is converted to 2-benzimidazole carbamic acid methyl ester (BCM), which is also the fungitoxic principle of benomyl (Selling et al., 1971). The oxathiin compounds carboxin and oxycarboxin are only active against fungi belonging to the Basidiomycetes, while the pyrimidine derivatives dimethirimol and ethirimol have a spectrum of activity which is even more narrow; only powdery mildew fungi are sensitive. The antibiotic kasugamycin is so specific in its action that it inhibits only one fungus, Piricularia oryzae on rice.

This selective action on fungi may have consequences for the microbial balance, when this type of fungicides is applied to the soil or to plants. It is well known that the activity of plant pathogens may be influenced by the presence of other microorganisms, which compete for food and space, or produce antibiotic substances. When a selective fungicide eliminates rhizosphere fungi antagonistic to a plant pathogen, the latter, when relatively insensitive to the fungicide, may be favoured. Such a shift in the microbiological balance may result in increased damage to the plant. Evidence that such a phenomenon may occur, has been obtained at our laboratory by van den Berg and Bollen (1971). The incidence of wilting of china aster, Callistephus chinensis, caused by Phytophthora cryptogea, was studied after mixing Benlate W.P. 50%, at a concentration of 5 ppm active ingredient, through the soil. One to two months after inoculation wilting of asters was much higher in the benomyl treated soil than in the control. The difference in incidence of wilting did, however, decrease with time, so that three months after inoculation no difference between treated and untreated soils was found. Although benomyl did not affect the final level of infestation of the soil by P. cryptogea, this level was attained much earlier in the presence of benomyl. Analysis of the soil microflora before and after application of benomyl showed a shift in the fungal population in favour of benomyl tolerant fungi. The rapid increase of wilting in the benomyl treated soil therefore might be due to the fact that benomyl sensitive antagonists of P. cryptogea are eliminated, while this fungus itself is rather insensitive to benomyl.

In a field experiment with rye, van der Hoeven and Bollen (1972) observed increased incidence of sharp eye spot, caused by Rhizoctonia solani after benomyl treatment.

It is therefore of importance to realize that other dangerous soil pathogens which are not sensitive (Pythium species) or are relatively insensitive to benomyl, may increase as a result of treatment with this fungicide. This may, of course, also apply for other selective fungicides.

That similar phenomena may take place also on above ground parts of plants, will be discussed later on.

ACQUIRED RESISTANCE

It is a well known fact that the effect of a biocide against noxious organisms may decrease after it has been used for some time. Known examples of this phenomenon are the acquired resistance of bacteria to antibiotics, and of insects to insecticides. This has caused many problems in the control of bacterial diseases of humans and insect pests of plants. In the past the control of fungal diseases with fungicides has scarcely been hampered by comparable problems. Exceptions are some fungicides belonging to the "aromatic hydrocarbon group", such as diphenyl, which is used against Penicillium fruit rot in citrus, and hexachlorobenzene, a seed disinfectant of wheat against bunt. The state of affairs with respect to fungicide resistance has been described in an excellent review by Georgopoulos and Zaracovitis (1967), who conclude that development of tolerance to fungicides by plant pathogenic fungi is reported surprisingly rarely.

Shortly after the introduction of systemic fungicides, however, various cases of acquired resistance have been reported. Dimethirimol, which provided excellent control of powdery mildew in Dutch cucumber greenhouses in 1968 and early 1969, failed in some greenhouses later on. Experiments, carried out in Wageningen, revealed the presence of tolerant strains in this fungus, which prohibited the further use of this fungicide.

Another example of fungicide resistance has been encountered in Dutch cyclamen culture, where heart rot, caused by Botrytis cinerea, was successfully treated with benomyl. From a greenhouse where the benomyl treatment failed, strains of B. cinerea were isolated, which tolerated 1000 ppm of benomyl in an agar medium, while the wild type fungus was eliminated at 0.5 ppm of this fungicide (Bollen and Scholten, 1971). The tolerant strain showed cross resistance to the benomyl-related compounds thiabendazol and fuberidazol, and further also to thiophanate-methyl. Scholten, at Aalsmeer, made the remarkable observation that in benomyl treated greenhouses, cyclamen plants infected with the benomyl-resistant strain of B. cinerea, were more severely diseased than plants in non treated greenhouses. Experiments showed that this could not be attributed to an increased virulence of the resistant strain. It was further noticed that after prolonged benomyl treatment disease incidence was reduced again to the original level. From these plants Bollen (1971) isolated two strains of Penicillium brevicompactum, which also had acquired resistance to benomyl. Both strains tolerated a concentration of 1000 ppm of benomyl in an agar medium, while growth of a wild type strain was completely inhibited at a concentration of 0.5 ppm. When tested on malt agar, a striking antagonism between benomyl resistant strains of B. cinerea and P. brevicompactum was obtained. This antagonism, showing a growth free zone between the two fungi, was even stronger in the presence of benomyl than without this fungicide in the medium. This indicates that the use of selective fungicides on the above ground plant parts also, might interfere with the relation between microorganisms, pathogenic as well as non-pathogenic.

Several other cases of acquired resistance to benomyl in practice have been reported. Schroeder and Provvidenti (1969) observed a decreasing effect of benomyl against cucumber powdery mildew, which they attributed to acquired resistance. Netzer and Dishon (1970) found resistance to benomyl in powdery mildew on muskmelon. In laboratory experiments resistance in fungi to many selective fungicides has been obtained, as will be discussed below.

MECHANISMS OF RESISTANCE

As the genetic aspects of fungicide resistance will be considered in the next lecture, I shall not discuss these, but ask your attention for the way in which genetically determined resistance may come to expression. It is obvious that an explanation of the mechanism of resistance in fungi will only be possible if we know the mechanism of action of a fungicide. It has to be admitted that at this moment, little detailed information is available about the mechanism of action of the new systemic fungicides. Therefore also experimental antifungal compounds with selective action, such as antibiotics and antimetabolites will be included in this discussion.

Fungicides may interfere with the metabolism of the fungal cell at various sites. Some examples of fungicidal action and acquired resistance are given below.

site:	interference with:	fungicide:	acquired resistance in:
cell wall	chitin synthesis	kitazin	Piricularia oryzae
protoplast-membrane	permeability	pimaricin amphotericin B dodine	Botrytis cinerea Candida sp. Hypomyces solani f. cucurbitae
mitochondria	energy production	antimycin A carboxin	Venturia inaequalis Ustilago maydis,
ribosomes	protein synthesis	cycloheximide kasugamycin	Saccharomyces cerevisiae Piricularia oryzae
nucleus	nucleic acid metabolism	6-azauracil benomyl	Cladosporium cucumerinum various fungi

Resistance of fungi to a fungicide may be brought about in various ways (Dekker, 1969). The fungal cell may be changed in such a way that the fungicide does not reach the site of action. This may be due to a decreased permeability of the protoplast membrane, or an increased capability of the fungus to detoxify the fungicide. When the fungicide does reach the site of action, the following possibilities may be discerned:

a. a decreased affinity to the fungicide at the site of action.

b. changes in the fungal metabolism resulting in a compensation for the inhibiting effect or a circumvention of the blocked site.

These mechanisms will be discussed in relation to the sites of action of fungicides.

Chitin synthesis. Recently two fungicides have been introduced which inhibit the synthesis of chitin, namely the antibiotic polyoxin D and kitazin. At our laboratory evidence was obtained by de Waard, that also hinosan interferes with chitin synthesis. Polyoxin D was found to interfere with penetration of N-acetyl glucosamine through the protoplast membrane and with its polymerisation into chitin (Sasaki et al., 1968). A kitazin-tolerant strain of Piricularia oryzae was obtained by de Waard, but the mechanism, responsible for resistance, has not yet been studied.

Permeability. A group of antifungal antibiotics, so called polyene macrolides, are known to act upon the protoplast membrane of the fungal cell. They interfere with ergosterols in this membrane, which results in leakage and ultimate death of the cell. Fungi which lack ergosterol in their membrane, such as Pythium and Phytophthora species are insensitive to these antibiotics (Schloesser, 1965). Although this example concerns an existing difference in sensitivity between various classes of fungi, acquired resistance has also been reported. At our laboratory strains of Botrytis cinerea were obtained, which are tolerant to pimaricin. Winner and Athar (1971) found strains of Candida species with increased tolerance to amphotericin B and nystatin, which showed a decreased sterol content. Resistance in this case might be brought about by a decreased affinity at the site of action, due to absence of sterols in the membrane.

Dodine acts on the permeability of the cell wall. A decreased effect of this fungicide against apple scab has been observed by Szkolnik and Gilpatrick (1969). The genetic basis of radiation-induced resistance to dodine in Hypomyces solani f. cucurbitae was studied by Kappas and Georgopoulos (1970), who found that four unlinked loci may contribute to resistance. The physiological-biochemical background of this type of resistance has not yet been elucidated.

Energy production. The antibiotic antimycin A acts upon electron transport in the respiratory chain between cytochrome b and c. After irradiation of conidia of Venturia inaequalis with ultraviolet rays Leben (1955) obtained strains which tolerated 5000 times as much antimycin A as the wild strain. He proved that the resistance was due to gene mutation. The resistance mechanism has not been investigated in this case.

Tolerance of Ustilago maydis to antimycin A could be attributed to a shift in the electron transport, at a site preceding cytochrome c, to an alternate terminal oxidase (Georgopoulos and Sisler, 1970). This alternate pathway is apparently of considerable value to U. maydis for growth in the presence of the antibiotic, which is demonstrated by the fact that a mutant which lacked this alternate pathway, was sensitive to antimycin A. This mutant was tolerant to the oxathiins carboxin and oxycarboxin. Although it is not yet clear how oxathiin resistance is controlled by the gene which eliminates the antimycin A tolerance, it is assumed that the action of the two compounds is related and that the oxathiins must also affect respiration (Georgopoulos, 1971).

Protein synthesis. Various antibiotics and probably also oxathiins and botran interfere with protein synthesis. Resistance in Saccharomyces cerevisiae to cycloheximide appeared to be located in the ribosomes, and might be due to a modification of the sensitive site on the ribosome (Cooper et al., 1967).

Ohmori (1967) obtained strains of Piricularia oryzae resistant to the antibiotic kasugamycin. The mechanism of kasugamycin resistance has been studied in Escherichia coli mutants by Helser et al. (1971). Also in this case the resistance appeared to be located in the ribosomes, namely in the 16 S RNA unit of the ribosome. The oligonucleotides of the 16 S RNA unit contain dimethyladenine in the sensitive bacteria, but unaltered adenine in the resistant mutant. It seems that mutation confers resistance to kasugamycin by preventing cells from methylating this adenine residue to yield dimethyladenine. Apparently the affinity to the fungicide has been reduced or lost by this change, without loss of the capacity to synthesize proteins.

Nucleic acid metabolism. Benzimidazole carbamic acid methylester (BCM), the fungitoxic principle of benomyl and thiophanate-methyl, interferes with DNA synthesis or with some post DNA synthesis aspect of the cell replication process in Ustilago maydis and Neurospora crassa (Sisler, 1971). Of the following fungi benomyl resistant strains were obtained at our laboratory: Botrytis cinerea (Bollen and Scholten, 1971), Penicillium brevicompactum and P. corymbiferum (Bollen, 1971) and Cladosporium cucumerinum. Acquired resistance to benomyl has further been reported in various fungi, o.a. in Aspergillus nidulans and Neurospora crassa (Hastie and Georgopoulos, 1971), Fusarium oxysporum f. sp. melonis (Bartels-Schooley and MacNeil, 1971) and muskmelon powdery mildew (Netzer and Dishon, 1970). As the exact mechanism of action of benomyl is not yet known, so the mechanism of resistance still has to be elucidated. It has been suggested that, to be fungitoxic, BCM should be metabolized to its nucleotide, a situation analogous to the one described for 6-azauracil by Dekker (1968).

It has been shown that 6-azauracil (AzU), an antimetabolite of uracil, interferes with nucleic acid synthesis. This compound is active as a systemic fungicide against powdery mildews and various other diseases, for example cucumber scab. The fungus Cladosporium cucumerinum has been used to study the fungitoxic action. It appeared that AzU, after conversion by the fungus into its nucleotide, 6-azauridine monophosphate (AzUMP), interferes with the activity of one enzyme in the pyrimidine biosynthesis. After irradiation of conidia of C. cucumerinum with ultraviolet rays, strains were obtained which tolerated much higher concentrations of AzU than the wild strain (Dekker, 1967). Different types of resistant strain could be discerned, and at least three mechanisms of resistance appeared to be operative. The first one is a decreased absorption of AzU by the fungal cell (Dekker, 1971) and the second one a decreased conversion of AzU into its nucleotide (Dekker, 1968; Dekhuijzen and Dekker, 1971). The resistance of a third type of strain could not be explained by one of these mechanisms. To study this type of resistance the fungus was grown for one or two weeks on a modified Czapek Dox medium in Roux flasks. The mycelium was then harvested, washed three times in a phosphate buffer solution, ground in a mortar with sand and extracted with the mentioned buffer solution. The decarboxylation of carboxyl-^{14}C labeled orotidine monophosphate by the medium in which the fungus has grown, by the supernatants obtained after washing and centrifugation and by the fungal extracts was then determined, according to a method described earlier (Dekker, 1968).

It appeared that the activity of OMP decarboxylase produced by the resistant strain was at least three times as high as that in the wild strain. It seems plausible that an increase of activity of the inhibited enzyme will require more of the fungicide to obtain a certain degree of inhibition. This could explain the observed tolerance to 6-azauracil in this strain.

PROSPECTS

As has been mentioned above, no serious resistance problems have been encountered in practice with the classic type of superficially acting fungicides, such as copper fungicides, dithiocarbamates etc. The assumption, that fungi would be less apt to develop tolerance to biocides than bacteria or insects has nevertheless proved to be wrong. Especially after the introduction of systemic fungicides various cases of acquired resistance in practice have been reported.

When discussing the problem of acquired resistance to biocides, not only the nature of the organisms concerned, but also the type of the biocides should be taken into consideration. The antibiotics and insecticides to which resistance emerged in bacteria and insects, interfere rather selectively with the metabolism of these organisms. Most of the classic fungicides, however, are non-selective plasma toxicants, which interfere with the metabolism of the living cell at numerous sites. In spite of this these toxicants still can be applied to plants, thanks to the fact that they hardly penetrate at all into the plant tissue. It will be clear that the effects of these multi-site inhibitors cannot be reduced easily by single gene mutations.

In contrast to this, systemic fungicides do act selectively on the plant-parasite combination thanks to physiological-biochemical differences between the cells of host and parasite. As has been indicated by laboratory experiments with experimental fungicides, such as antibiotics and metabolites, these fungicides probably act at one or only a few sites in the fungal metabolism. It seems plausible that fungi may adjust more readily to selective than to non-selective fungicides by gene mutation.

As has been outlined above fungi possess numerous opportunities to acquire resistance to fungicides which interfere selectively with their metabolism. Laboratory experiments may provide an estimate of the frequency with which resistant mutants may emerge under certain conditions. Bartels-Schooley and MacNeil (1971) found that, after treatment of conidia of Fusarium oxysporum f. sp. melonis with ultraviolet rays, one out of $4.6x10^5$ of these conidia give rise to a benomyl-resistant colony, and without u.v. treatment one out of $8.6x10^7$. Considering the large numbers of spores which may be produced by fungi, the development of resistant strains in cases like this is likely to be a common phenomenon.

In view of this the question may arise whether resistant strains will ultimately develop to all systemic fungicides after a certain period of practical use. This of course would seriously endanger the future of these fungicides in the control of plant diseases.

Without trying to give a definite answer to this question, I should like to make a few remarks.

a. The frequency with which resistant mutants arise, may vary greatly for various systemic fungicides. This depends on the number of mutations required to reach a certain level of resistance and on the mutability of the genes at the loci concerned. A generalisation therefore does not seem to be justified. Chemicals which act at more than one site in the metabolism of the fungal cell should be preferred to those which act at only one site.

b. It has been shown that systemic compounds without direct fungicidal activity may provide disease control by increasing the resistance of the host plant. An example is procaine hydrochloride, a local anesthetic without fungicidal activity in vitro. It protects cucumber plants against powdery mildew (Niemann and Dekker, 1966), without being inhibitory to spore germination or elongation of germ tubes of Sphaerotheca fuliginea in vitro. This induced resistance might possibly be compared with the natural "horizontal" resistance, which is not easily broken down by fungi.

c. Evidence has been obtained that the emergence of fungicide resistance under laboratory conditions does not necessarily mean that resistance problems will be encountered also under field conditions. As the survival of resistant mutants in practice will be discussed in the next lecture, I shall confine myself to an interesting observation made by Ohmori (1967) while studying the control of Piricularia oryzae on rice by the antibiotic kasugamycin. In laboratory experiments isolates of this fungus were obtained, which grew on a medium containing 100 μg/ml of the antibiotic, while the wild strain was inhibited at a concentration of 1 μg/ml. Although kasugamycin has been used against the Piricularia disease of rice during a number of years, no resistance problems have arisen in the field. It was observed that the kasugamycin-resistant strains obtained in the laboratory were less infectious and that the formation of spores, in spite of the tolerance of mycelium growth to kasugamycin, was still inhibited by the antibiotic.

d. The emergence of a resistance problem in practice will also depend on the type of disease. For a fast spreading disease, caused by a heavily sporulating pathogen, the situation may differ from that for a disease, which spreads slowly. Moreover the frequency of emergence of resistant mutants may be different for various fungi.

In the foregoing we have discussed two possible disadvantages of selective action by systemic fungicides, namely a disturbance of the microbiological balance and an increased chance for fungi to develop resistance. These, however, do not alter the fact that systemic fungicides may have distinct advantages above classic types of fungicides, as mentioned in the introduction. In addition the possibility remains open that systemic fungicides interfere in a beneficial way with the metabolism of the host plant. Benomyl, for example, may delay senescence, which under certain circumstances and in certain crops might be desirable (Sisler) 1971.

Provided that resistance problems can be kept within bounds, and application occurs judiciously, it may be expected that systemic fungicides will be of great value for plant disease control in the future.

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FUNGICIDES AND THE FUNGUS POPULATION PROBLEM

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Summary The increasing numbers of reports of fungal mutants tolerant of fungicides should not be unexpected since populations of plant pathogens tend to be large, so that the total number of mutants, which continually occur, is also relatively large. Instances of tolerant mutants causing serious practical difficulties are, however, not as common, so that it is important to determine the factors which cause tolerant mutants to become more or less frequent in pathogen populations. It is also important to consider how the integration of the use of fungicides with breeding for disease resistance may help to maintain both fungicide sensitivity of the pathogen and host resistance.

Note on terminology: Simultaneous consideration of the interaction between pathogen, fungicide and host may lead to confusion in use of the terms resistance and tolerance. In the interaction between host and pathogen the host resists attack or is susceptible to it. After it is attacked, the host is then either tolerant or sensitive to the presence of the pathogen. Following this usage, it is convenient to consider the interaction of pathogen and fungicide only in terms of tolerance and sensitivity.

INTRODUCTION

In considering the importance of fungicide tolerance, there are four basic questions which must be answered:

i. Can the mechanism necessary for tolerance of a fungicide be accommodated within the limits of fungal metabolism?

ii. Is tolerance heritable and if so in what way?

iii. Does the occurrence of tolerance have any consequence of economic significance in the pathogen population?

iv. Can we reduce the effects of tolerance by alterations in fungicide use, or by manipulation of host interactions?

In this paper some facets of these questions are discussed.

OCCURRENCE OF FUNGICIDE TOLERANCE

Two factors indicate that the frequency of fungicide tolerant mutants may be expected to be high. The first is that the fungi are known to possess the potential for an extremely wide range of metabolic variation. The potential frequently exists, therefore, for initially sensitive systems to change to higher levels of tolerance. The second factor is that where mutation rates towards tolerance have been measured (Bartels-Schooley and MacNeill, 1971, Georgopoulos and Panopoulos, 1966, Shaw and Khaki 1971, and Wilkie and Lee 1965) they have been found to be high relative to the size of the fungus population.

Not surprisingly therefore, since the last reviews of fungicide tolerance by Georgopoulos and Zaracovitis (1967) for organic fungicides and Ashida (1965) for metal toxicants, there has been a considerable increase in the number of tolerant mutants described, a range of which is given in Table 1. The increase has been attributed to the greater specificity of some materials more recently introduced, but may also be due simply to greater usage of the wider range of fungicides now available.

From the examples given in the Table it can be seen that the proportion which is of practical importance is small. This may be because the occurrence of tolerant mutants in the laboratory, either naturally or by induction, does not give any immediate indication of the potential importance of mutants in the population. Also, the frequency of detection of mutations determined in the laboratory, where selection can be controlled, may be expected to be greater than the frequency of mutant detection in the field. Indeed, because of the difficulty of detection of mutations occurring after commercial applications of fungicides, tolerance may in fact, be of greater significance than is at present realised.

The situation is similar to that found in breeding for disease resistance or from the use of other pesticides. For any high degree of pathogen or pest control exerted by host resistance, pesticide or fungicide, mutations for virulence or tolerance in the pathogen or pest may occur, sooner or later. The important questions concern therefore, the factors determining the frequency of the mutants and their subsequent fate in the pathogen population.

INHERITANCE OF TOLERANCE

The frequency of occurrence of tolerance will be determined initially by the equilibrium value of forward and back mutation rates for tolerance, by the number of loci with this potential, and by the interactions between them.

i. Intra-allelic variation

First it is necessary to consider the size of effect of the mutation. MacKenzie *et al*. (1971) found that tolerance to thiram in *Cochliobolus carbonum* was inherited quantitatively so that the effect of each tolerance allele was small. On the other hand, there are numerous cases of single tolerance alleles exerting easily measurable effects, for example, in tolerance of chlorinated nitrobenzenes and dodine in *Nectria haematococca* (Georgopoulos and Panopoulos 1966, Kappas and Georgopoulos, 1970) and in tolerance of cadminate and actidione in *Cochliobolus carbonum* (MacKenzie *et al*., 1971).

Where mutations occur at more than one locus in the genome, then the alleles involved may condition similar degrees of tolerance, as in *Nectria haematococca*, (Georgopoulos and Panopoulos, 1966) or different degrees as in the tolerance of yeast to actidione (Wilkie and Lee, 1965).

Table 1

Some recent examples of fungicide tolerance
*denotes examples considered to be of practical importance

a. Tolerance of organic fungicides

Benzimidazoles	*Botrytis cinerea	Bollen & Scholten (1971)
	Fusarium oxysporum melonis	Bartels-Schooley & MacNeill (1971)
	Oidiopsis taurica	Netzer et al. (1970)
	Sclerotinia sclerotiorum	Paster & Dinoor+
	*Sphaerotheca fuliginea	Schroeder & Provvidenti (1969), Dinoor+
	Ustilago hordei	Ben Yefet & Dinoor+
Captan	Aspergillus sp.	Dinoor+
	Puccinia graminis	Polyakov & Mende (1965)
	Venturia inaequalis	Golyshin (1964)
Dicloran	Botrytis cinerea	Webster et al. (1970)
	Rhizopus stolonifer	Webster et al. (1968)
	*Sclerotium cepivorum	Locke (1968)
Dithiocarbamates	Cochliobolus carbonum	MacKenzie et al. (1971)
Dodine	Nectria haematococca	Kappas & Georgopoulos (1970)
	*Venturia inaequalis	Szkolnik & Gilpatrick (1969)
Ethirimols	Erysiphe graminis	Wolfe (unpubl.)
	*Sphaerotheca fuliginea	Bent et al. (1971)
Oxathiins	Ustilago hordei	Ben Yefet & Dinoor+
	Ustilago maydis	Georgopoulos (1971)
Triarimol	Sclerotinia sclerotiorum	Paster & Dinoor+

b. Tolerance of metal toxicants

Boron	Piricularia oryzae	Yamasaki et al. (1964)
Cadmium	Cochliobolus carbonum	MacKenzie et al. (1971)
	*Sclerotinia homoeocarpa	Cole et al. (1968), Massie et al. (1968)
Copper	Piricularia oryzae	Yamasaki et al. (1964)
Mercury	*Pyrenophora avenae	Noble et al. (1966), Bainbridge (1969) Malone (1968)

+ personal communication

The number of loci found to be involved in fungicide tolerance will depend essentially on the metabolic mechanisms required and on the size of the population analysed. In most genetic analyses which have been reported, more than one locus conditioning tolerance has been found, and sometimes up to six (Wilkie and Lee, 1965). Georgopoulus and Panopoulos (1966) determined five loci conditioning tolerance of chlorinated nitrobenzene in Nectria haematococca with apparently widely different mutation rates, and they pointed out that surveys of a larger population may well have revealed further loci with mutation rates not detectable in the size of population which they used.

Within the locus it has often been found that tolerance is dominant or partially dominant to sensitivity (Shaw and Elliott 1968, Wilkie and Lee, 1965) so that in diploid pathogens, mutations for tolerance will be immediately expressed. The expression of recessive mutations for tolerance may be delayed until a sufficiently large selection pressure is applied to the population. Such delays would not occur, of course, in haploid pathogens such as the powdery mildews.

ii. Inter-allelic variation

Inter-allelic interaction depends on the chromosomal distribution of the loci conditioning tolerance, and the system of recombination in the organism. Studies of recombination have not so far revealed linkage between tolerance loci although linkage with other loci has been demonstrated. The most important of these studies concern linkage between loci conditioning fungicide tolerance and those conditioning pathogenicity* on the host. Georgopoulos (1963a) found that in a series of isolates of Nectria haematococca tolerant of chlorinated nitrobenzenes, some were as pathogenic as the wild type whilst others were less so. In one of the latter, linkage was detected between fungicide tolerance and a locus controlling part of the pathogenicity character. In studying dodine tolerance in the same organism, Kappas and Georgopoulos (1971) found that in one tolerant line which was non-pathogenic, the gene for pathogenicity was independent of the gene for tolerance. Thus the occurrence of reduced pathogenicity in a newly formed tolerant isolate is not necessarily indicative of either a functional association or linkage. Most of the recorded examples of tolerance do not seem to be associated with changes in pathogenicity, assessed in laboratory studies.

Where tolerance alleles are found at different loci in the same organism, it is necessary to determine whether they are duplicate in action, or whether there is an interaction between them. In an example noted above (Georgopoulos 1963a, b), tolerance at three loci was found to be duplicate. On the other hand, Kappas and Georgopoulos (1970) investigating dodine tolerance, also in Nectria haematococca, and Wilkie and Lee (1965) analysing actidione tolerance in yeast, both obtained evidence of positive interactions in recombinants, i.e. the recombinants exhibited higher levels of tolerance than either parent. In the yeast the tolerance was further enhanced by the presence of modifying genes.

The phenomenon of interaction is further complicated by cross-tolerance which frequently occurs for related fungicides (Bartels-Schooley and MacNeill 1971, Edgington et al., 1971, Esuruoso and Wood, 1971, Georgopoulos, 1963b, and Priest and Wood, 1961). Bollen and Scholten (1971) were able to demonstrate cross-tolerance in Botrytis cinerea both for related benzimidazole derivatives and for methyl thiophanate. On the other hand, Parry and Wood (1959)

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*The term pathogenicity is used here to cover virulence and aggressiveness, since the distinction is not clearly made in the relevant references, although aggressiveness is the characteristic most likely concerned.

found that a ferbam-tolerant strain of the same fungus was not tolerant of other closely related fungicides. Further, Georgopoulos and Vomvoyanni (1965) obtained evidence for increased sensitivity to diphenyl in some chlorinated-nitrobenzene tolerant strains.

So far, consideration of inheritance has been restricted mainly to simple Mendelian examples. The phenomena of heterokaryosis and extra-chromosomal inheritance are also common in the fungi and have been implicated in the inheritance of fungicide tolerance. Inheritance of tolerance of dicloran in Rhizopus stolonifer (Webster et al., 1968) and in Botrytis cinerea (Webster et al., 1970, Esuruoso and Wood, 1971) was considered to be due to heterokaryotic effects. Webster et al., (1968) pointed out that in the normal heterokaryotic population, fungicide tolerance may persist at a low level, that is, in a small proportion of the heterokaryotic nuclei. When the selection pressure increases, due to the use of the fungicide, then the proportion of nuclei conditioning tolerance could increase appropriately. On removal of the selection pressure, the nuclear distribution would return to the original proportions, which conditioned maximum fitness in the untreated population.

MacKenzie et al., (1971) in a study of thiram-tolerance inherited quantitatively in Cochliobolus carbonum discussed the difficulty of distinguishing between heterokaryotic effects and cytoplasmic inheritance. Their conclusion for one cross was that the inheritance analysed was cytoplasmic although it was impossible to determine whether or not this was nuclear-dependent.

Although the rate of occurrence of tolerance in heterokaryotic and cytoplasmic systems of inheritance is dependent on mutation rate, these systems do provide further means of variation in the fungal population.

TOLERANCE IN THE FUNGAL POPULATION

In order to cause an economic problem, tolerant forms in the population must increase beyond a critical frequency so that they are both measurable in effect and self-sustaining at that level. The attainment of this critical frequency is dependent on inheritance in the individual, discussed above, competition within the fungus population, factors affecting presentation of the fungicide to the fungus and the influence of pathogen resistance in the host.

i. Competition in the fungus population. Studies within a species, discussed in the previous section, indicate that, in the laboratory, tolerant forms are usually as pathogenic as normal forms: if not, then this may be due to chance recombination between tolerance alleles and alleles conditioning low pathogenicity. Alternatively, a number of loci conditioning tolerance may be determined, some of which may be associated with decreased pathogenicity, whilst others are not. The conclusion from these studies would be that forms could easily evolve which possess the combinations of tolerance and pathogenicity necessary for successful competition with sensitive forms of untreated host material.

This situation may not arise in practise however, because laboratory comparisons of pathogenicity cannot include all factors involved in fitness in the field population. The theory of genetic homoeostasis indicates that, although particular individuals with a characteristic such as fungicide tolerance may be selected, the whole population does not easily shift in this direction since there are many characteristics in the population which are held in a complex balance in the existing environmental situation. Thus, if the new selection pressure is relaxed, or incomplete, the population may tend to shift back to its previous state of balanced adaptation. It is only when the tolerant strains have a large and continuous selective advantage that the population gradually becomes fully adapted.

Where a particular species is eliminated by fungicide, the competitive pressure on other, unaffected species, may be lifted so that a new problem arises. A situation of this kind was described by Farley and Lockwood (1969) and Katan and Lockwood (1970), in which Fusarium spp. and other pathogens, tolerant of pentachloronitrobenzene, were found to increase in soils treated with this fungicide. Other examples are noted by Prof. Dekker elsewhere in this volume.

ii. Presentation of the fungicide. The concentration of the fungicide and the timing of application are generally determined in relation to the degree of disease control and yield increase obtained. Both of these factors will however influence the emergence of tolerant forms. Generally, it might be expected that low concentrations will lead to the appearance of a relatively large number of tolerant forms, although the average degree of tolerance may not be great.

With regard to spray-timing of systemic fungicides, the question is often asked as to whether seed treatment may be more or less conducive to the emergence of tolerant strains than foliar sprays. There is no general answer. Mutants tolerant of the seed-dressing may arise earlier in the season than those tolerant of foliar sprays. If however they are less competitive than the wild type, then as the fungicide concentration decreases, competition increases and the frequency of tolerance declines.

With foliar sprays, on the other hand, there are fewer pathogen generations between the time of application of fungicide and the occurrence of sexual or resting stages, so that the relatively high frequency of tolerance alleles present at the time of genetic reassortment would allow the possible emergence of tolerant forms which were well adapted for further survival.

Considering the duration of the fungicidal or fungistatic effect, older fungicides tended to have a severe but short-lived effect. If mutants tolerant of these compounds did arise, then they would require a strong selective advantage in order to compete successfully in the natural population, following the disappearance of the fungicide effect. The systemic fungicides however, subject the pathogen to a longer-lasting but diminishing effect which favours the selection of mutants at different levels of tolerance. At low concentrations, the appearance of a large number of tolerant forms increases the possibility of association of tolerance with normal pathogenicity and environmental fitness in the population.

Decreasing concentration of systemic fungicides in the host after application is complicated by uneven distribution. Peterson and Edgington (1971) showed that benomyl tends to move upwards in the plant and accumulate in leaf tips and margins so that central areas of leaves would tend to become less inhospitable to the pathogen earlier than the more distal regions. Indeed, tolerance could conceivably arise in such cases from fungicide avoidance rather than direct metabolic tolerance as forms might be selected which were better able to grow on those host organs which had been the first to be depleted of fungicide.

iii. The influence of host resistance. Generally, there is no obvious economic advantage to be gained in applying fungicides to disease-resistant varieties, except where the fungicide may be found to have a direct beneficial effect on the host. However, in such cases, the effects of host resistance and fungicide control interact so that the possibility for production of host-virulent and fungicide-tolerant pathogen forms is reduced to a minimum. With a particularly outstanding host variety, protection in this way, of both host-resistance and fungicide, may not be beyond the bounds of economic feasibility in certain cash-intensive crops.

At the other extreme, the application of fungicides to host varieties highly susceptible to disease may have a dubious economic advantage. Fungicide trials in disease intensive conditions show that, even with high levels of disease control, susceptible varieties do not realise their maximum yield potential. Varieties of barley, highly susceptible to powdery mildew, have been shown to support higher levels of the disease towards the end of the season than untreated controls, since protection in the early part of the season allows the host to develop vigorously and thus become an ideal medium for subsequent pathogen development as the fungicide concentration falls (Wolfe, unpubl.). Under these conditions, development of fungicide tolerant strains may be enhanced.

The most suitable hosts for fungicide use therefore seem to be those with moderate or intermediate resistance in which re-infection is delayed sufficiently for the late season epidemic not to develop to any great extent. Such combinations offer a direct economic advantage, together with the indirect advantage of protection of both the host resistance and the fungicide effect.

In the event of tolerant mutants occurring, Fig. 1, a. and b. shows diagrammatically the course of events which might occur on moderately disease resistant and on disease susceptible varieties both in the situation where the tolerant lines are less competitive than the sensitive and in the situation where they are equally competitive. In Fig. 1a, disease levels are drawn higher at the end of the season than at the fungicide application date, whereas in Fig. 1b, they are drawn lower, illustrating the cross-protection of fungicide and host resistance.

AVOIDANCE OF FUNGICIDE TOLERANCE

A number of measures may be suggested to minimise the emergence and increase of fungicide tolerance.

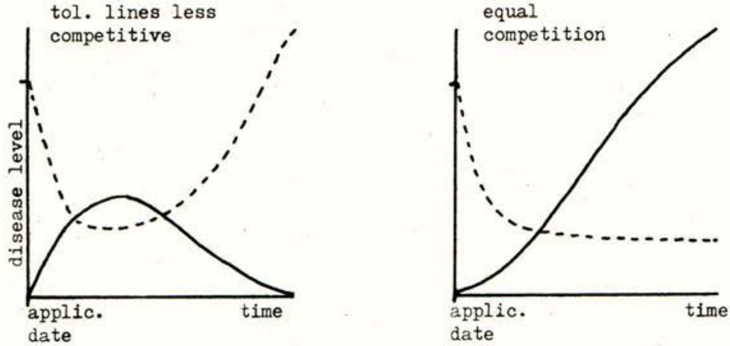
i. Fungicide dose. This should be at the highest level possible, consistent with economic feasibility and avoidance of phytotoxicity. Low levels may be conducive to the development of tolerance.

ii. Area of fungicide application. This should be kept to a minimum consistent with disease control and economic return, both nationally and on the individual farm. There are two reasons for this, the first being simply that tolerance does not develop where fungicides are not used. Secondly the maintenance of a small reservoir of wild type population may help to reduce the frequency of tolerant forms because of competition subsequent to depletion of the fungicide effect.

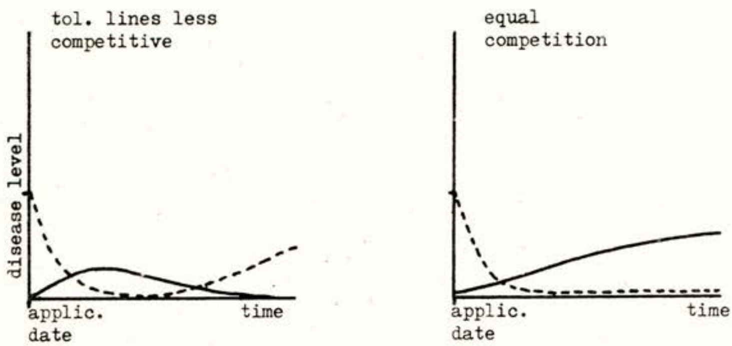
iii. Spray programmes. Where more than one fungicide application is considered economically feasible for disease control, then different fungicides should be used even though there may be a slight initial economic or practical disadvantage to the grower. This will have the effect of minimising the development of tolerance since there will be a requirement for the incorporation of two or more tolerance mechanisms in one pathogen strain. Repeated use of one fungicide however, will create a continuous selection pressure for a high level of tolerance involving only one mechanism. The comparison is illustrated diagrammatically in Fig. 1c.

Fig. 1. Population frequency of fungicide tolerant lines in different hypothetical situations. —— = tolerant lines, - - - = sensitive lines

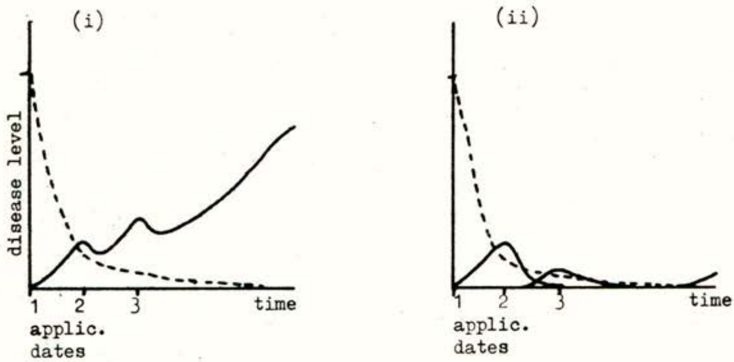
a. On disease susceptible host



b. On disease resistant host



c. same fungicide repeated (i) or different fungicides used (ii), in equal competition situations



iv. Choice of host variety. Host varieties should be selected which have at least a moderate degree of resistance in order both to exploit the interaction of host resistance and fungicide control, and to insure against failure of host resistance and of the fungicide (see Fig. 1a and b.).

The interaction of host and fungicide is extremely complex, but the possible existence of relationships between host resistance mechanisms and fungicide mechanisms may offer a fruitful field for future exploitation. Less direct relationships may be found in host characteristics which affect for example, uptake and distribution of the fungicide within the plant.

v. Monitoring of tolerance. Since it can be argued that fungicide tolerance in some form and at some level will almost certainly exist in a pathogen population, monitoring for this should provide an 'early warning' advantage. As soon as tolerance is detected then a range of tolerant isolates may be analysed to gain information on the tolerance level, pathogenicity etc., which will indicate the potential importance of the newly developing strains and the kind of action necessary to deal with the problem. A similar system has been in operation for several years for the detection of pathogen races virulent on disease resistant hosts in cereals and this has had a number of important successes in avoiding exploitation of initially resistant varieties which were potentially at risk. In the field of cereal disease control there exists, therefore, the possibility for a useful collaboration in determining both fungicide tolerance and host virulence in the pathogen population.

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ENVIRONMENTAL ASPECTS OF USING PERSISTENT PESTICIDES

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Summary The background is given to the attitudes and decisions in Scandinavia on the two major groups of persistent chemicals, namely organomercury fungicides and organochlorine insecticides. The current situation in Scandinavian countries is described and it is emphasized that the decisions are strongly influenced by the Scandinavian need or lack of need for pesticides and should not necessarily influence decisions in other countries.

The influence of pesticides on the environment is, to a high degree, linked to their persistence. This is, of course, not the only criterion of importance, but in today's debate it is a dominating one. I have interpreted my task here today to present some views on the relevant situation on these problems in Scandinavia, and especially in my home-country Sweden.

Two groups of persistent pesticides have attracted special interest in our corner of the world - the organomercury fungicides and the organochlorine insecticides. Whilst in the United States and in most European countries the presumed phenomenon of "Silent Spring" was mainly attributed to DDT and its relatives, high mercury contents in seed-eating birds and birds of prey were causing the major alarm in Sweden.

Mercury compounds had been extensively for 30 to 40 years on seeds of autumn and spring sown cereals, sugar beet and other root crops, and many vegetables. Many different types of mercury compounds have been used during this period and the application techniques have also undergone great changes. After some years of wet treatment (steeping), the dry method (dusting) prevailed until the early 1950's when liquid formulations took over. These were mostly methylmercury compounds applied by about 2100 specially equipped seed dressing stations according to the so-called "Panogen" method. The transition from dry to liquid formulations was greatly encouraged for hygiene reasons, the farmers being relieved from the risks of inhaling and absorbing the poisonous dust.

Some years later, from 1958 on, Dr Karl Borg of the wild life survey of the National Veterinary Institute started to publish results of chemical analysis of different wild birds, shot or found dead. From the high mercury contents noted and from feeding experiments conducted with dressed grain it was concluded that mercury treatments of seeds could be held responsible for lethal and sublethal effects on grain-feeding birds and their predators (Borg 1958, Borg et al. 1969, Hansen 1965, Otterlind and Lennerstedt 1964). In some cases increased effects relating to combined aldrin-mercury treatments could be observed. The general public and the ornithologists in particular were alarmed and the discussions culminated at a conference in September 1965, convened by an ad hoc Committee on Nature Resources, acting under the Ministry of Agriculture.

In February 1964 the National Institute for Plant Protection and the State Seed Testing and Certification Institute issued a joint recommendation that the dosage of mercury should be reduced by a half (1,6 g Hg/ha, instead of 3,2 g Hg/ha) and that mercury treatment should be carried out only when the seed analysis had revealed a significant infection of seed-borne diseases (15% or more of fungi attacking the seeds during germination and early growth) (Esbo 1965, Granhall 1965). In October 1965 these recommendations were made compulsory by law as regards mercury treatment of spring-sown cereals. For winter cereals no restrictions have been imposed nor are there restrictions on the use of mercury-free seed dressings.

As from February 1966 the National Poisons and Pesticides Board withdrew the registration of the alkylmercury seed dressings which were found especially persistent. Instead of these, alkoxyalkyl mercury compounds, considered less stable to hydrolysis, were registered to be used in "full dose" (3,2 g Hg/ha). The new compounds have the same liquid formulation as have the old ones and can be applied in the existing equipment.

The regulations have brought about remarkable changes in the seed dressing situation in Sweden. Before then mercury treatment had been established as a routine prophylactic measure. Nearly all winter cereals were treated and 70-80 per cent of the spring sown seeds. In 1968 and the following years, however, the percentage of treated spring seeds was below 10. In 1971 a slight increase has been noted (see table 1).

Table 1 Frequency of seed-borne diseases and mercury treatment in Swedish spring-sown cereals, 1962-71

Year	Percentage above 15% infection level x)	Percentage actually treated
1962	31	)
1963	45	)70-80
1964	32	)
1965	33	)
1966	50	23
1967	33	13
1968	20	10
1969	12	7
1970	6	3
1971	17	15
1972 xx)	30	

x) 1962-65 treatment recommended, 1966-72 treatment permitted

xx) prel. figure

There are two causes of the treatment decrease. Firstly, the seeds have been unusually healthy because of favourable growing and harvesting weather in the years 1967-1970. This means that relatively few seed lots have been over the "permissible level" of 15% infection. The harvest of 1971 has shown a more "normal" degree of infection. Secondly and even more important, as a result of the public discussions on the hazards involved, many farmers have refrained from treatment with mercury even in cases of comparatively high infection.

In the other Nordic countries the mercury debate has not produced such drastic changes. In table 2 the Hg-treated areas have been estimated for the years 1965 and 1968, i.e. before and after the Swedish restrictions (Granhall 1965, Lihnell 1969). In Norway the transition from methyl to alkoxyalkyl mercury went more smoothly. In Finland and Denmark dry treatments with alkoxyalkyl and phenyl compounds have dominated all the time. The acreage of Hg-treatment is very low in Norway and Finland, especially in relation to the total land area. This is not the case in Denmark, but, nevertheless, no widespread bird incidents were reported.

Table 2 Mercury seed dressing in the Nordic countries, 1965 and 1968

Country	Total land area 1000 ha	Arable land 1000 ha	Cereals 1000 ha	Mercury treated area 1965 1000 ha	1968 1000 ha
Sweden	41.100	3.290	1.470	1.180	400
Norway	30.900	850	240	120	90
Finland	30.500	2.680	1.020	300	300
Denmark	4.300	2.820	1.570	1.290	1.250

How has the mercury threat to the Swedish bird populations developed after the decrease in treated area and the shift to less persistent compounds? In 1964 as much as 75.2% of ring-doves shot had shown more than 1 ppm Hg in their liver and 30.5% more than 5 ppm. In 1967 the National Veterinary Institute (Wanntorp et al. 1967) reported only 8.5% of analysed doves having shown more than 1 ppm and none over 5 ppm. In 1968 improvements were also noted in dove-hawks (Borg 1969), 30% of which had over 10 ppm in 1963-64 but none over 3 ppm in 1967-68. Similar results were obtained by other investigators (Westermark et al. 1969), and the decrease was attributed to the low persistence of the new alkoxyalkyl mercury, which has a "50% biological break-down time" of about 1/10 of that of methyl mercury.

These reports, of course, have been noted with satisfaction. The public interest in possible mercury sources has turned away from agriculture and more attention is now being given to industry, especially the paper-pulp mills and chlorine-alkali factories which have been revealed as sources of mercury contamination of a considerably greater magnitude. Contamination of rivers and lakes with their effluents has resulted in a high mercury content in fish. Many waters have been black-listed by the Institute of Public Health because of mercury contents in the fish higher than 1 ppm, and all kinds of fishing has been prohibited in these waters. Measures taken have produced encouraging results and the situation is improving.

In agriculture the weakening of the defences against seed-borne diseases has, however, not gone by without consequences. Several fungi which were well controlled or practically eradicated have come back, in particular *Helminthosporium gramineum* in barley and other species of the same genus in barley and oats, *Tilletia caries* in winter wheat, and recently also *Ustilago hordei* in barley. A troublesome new-comer, *Tilletia controversa*, dwarf bunt in winter wheat, although in principle a soil-borne fungus, has no doubt also profited from the reduced mercury treatments. Due, no doubt, to these experiences the farmers' interest in seed treatment now seems to be reviving. Mercury-free compounds, in particular systemic fungicides, have given promising results in field experiments (Olofsson 1971) and are ready to take over the dressing of winter wheat and rye with the exception of wheat seed lots with high infection of bunt. For the spring--sown cereals, however, the mercury compounds are still unsurpassed for giving protection.

Now to the other category of persistent pesticides. The use of organochlorine insecticides in Sweden and in the other Nordic countries has always been at a comparatively modest level. The four most interesting compounds are aldrin, dieldrin, lindane and DDT.

Aldrin was used during some years in combined seed dressing with methylmercury compounds. Only about 2% of the cereal seeds sown were treated this way as an insurance against wireworm (Elateridae) attacks. This small amount, however, may have been the spark that set alight the whole seed dressing debate. The effect on the seed-eating birds was apparently acute. In 1964 aldrin was prohibited for seed treatment of cereals, and in 1966 the ban was extended to seed and soil treatment in cabbage, onion and carrots. Only against wireworms in potatoes, where no substitute had been found, was the use of aldrin allowed for a further period of years but in practice it was only applied on 2000-3000 ha annually. Dieldrin had only been used in small quantities, mostly in vegetables, and it was finally withdrawn in 1966.

Lindane has a wider application against several insect pests in the field and in storage. Because of its comparatively short persistance lindane has not been implicated to a great extent in the discussions on contamination of the environment. On the other hand its use has been limited because it can produce taint in products for consumption. The annual amount used in recent years in Sweden has been about 30 tons (active substance).

The DDT contamination of environment on the other hand is a problem of world-wide concern. Of the total production of about 100,000 tons annually the Swedish consumption of DDT only amounted to 40-60 tons. Wind transportation from other parts of Europe and, maybe, also from afar, must therefore be taken into consideration to explain the sometimes rather high quantities of DDT and its metabolites found everywhere in different organisms, in agricultural regions as well as in the most northern wilderness, in the inland streams and lakes as well as in the Baltic Sea. To a considerable degree the chemically related industrial products PCB's (polychlorinated biphenols) have complicated and aggravated the situation. Correlated with high contents in wild birds, cases have been reported of low reproduction because of thin-shelled and fragile eggs, and there have been mass bird deaths attributed to increased sensitivity to adverse weather conditions. In general, however, the Swedish experience follows the same pattern as that reported from the United Kingdom (Anon. 1964 and 1969), France (Siriez 1970) and Austria (Beran 1970), indicating occasional disturbances but no significant general reduction of wild life populations by the action of DDT and related substances.

In March 1969 the Swedish Poisons and Pesticides Board convened a DDT Conference in Stockholm to which several well-known international experts were invited. The Board's communication after the final session of the Conference attracted wide attention :

"The Poisons and Pesticides Board has decided today that further use of certain chlorinated hydrocarbon pesticides will not be accepted in Sweden. The decisions imply the following :

1. All use of aldrin and dieldrin will be forbidden from the 1st of January 1970.

2. The use of DDT and lindane in household preparations and similar preparations for home gardening will be forbidden from the same date.

3. Other uses of DDT will be forbidden for a test period of two years, from January 1st 1970. In exceptional instances there will be the possibility of an exemption. The Board assumes that during the test period research will be carried out aiming at elucidating the effect of the restriction".

The communication stated that the decisions were not based on any immediate risks to human health but on the risks shown to the environment regarded as an integrated biological system. The low demand for DDT in Sweden and the local possibilities of replacing it by other compounds or control methods less disturbing to the environment were also referred to. On the other hand it was duly recognized that variations in types of climate and differences in other conditions highly influence the need for and the risks of the organochlorines in different parts of the world. The statement made by the World Health Organization that DDT is for the time being indispensable in the campaigns against malaria and other vector-borne diseases was also quoted.

In the communication the possibility of exemptions from the DDT prohibition is mentioned. Two applications for such exemptions were immediately presented. The farmers' co-operative organizations claimed that continued use of DDT against the blossom beetle (Meligethes aeneus) and other insects in oil-seed rape was essential. After reference to the possibilities of using non-persistent organo-phosphorous compounds and the non-persistent organochlorine substance methoxychlor the request was refused. The other opposition came from the private and governmental forest administrations and concerned the DDT treatment of pine and spruce planting material against the pine weevil (Hylobius abietis). The absence of an efficient substitute and the important economic values at stake made a provisional exemption necessary in this case. On the other hand a proposal for the use of DDT against barkbeetles in stored and transported timber was not accepted by the Board.

The 1969 decision of the Pesticides Board refers to a two year test period for DDT, but nobody believes that the DDT-ban will be cancelled from January 1st 1972 or any later date. It is, however, still too early to look for effects of the restrictions applied. In Denmark, Finland and Norway similar restrictions on the use of DDT have been issued. The Hylobius campaign has also been exempted in Finland and Norway where the situation is analogous to that of Sweden.

An EPPO survey recently published (Mathys edit. 1970) reflects the situation in other parts of Europe where important quantities of DDT are still being used. Partial or total ban is, however, coming into force in most countries. The corresponding reports for lindane are also forecasting restrictions but considerably less rigorous ones.

The general trend all over the world is to restrict or ban the use of persistent pesticides as far and as soon as possible and from the environmental point of view there are no doubt good reasons for this attitude. On the other hand extraordinarily great care must be taken in the selection and marketing of new pesticides. The criteria for an acceptable pesticide are nowadays much more difficult to define than they were 25 years ago when DDT was launched. It might easily happen again that today's blessings are tomorrow's blame.

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THE CHANGING PATTERN OF PESTICIDE USAGE IN EUROPE - AN INDUSTRIAL VIEWPOINT

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Summary Changes in the pattern of pesticide usage may arise either from the development of more efficient materials for specific applications, or from the abandonment of chemicals which, although effective, have been found to produce detrimental environmental effects. In the past the former cause has probably been of greater importance, but with the present increase in concern about pollution in general, regulation of pesticide usage in relation to environmental considerations is becoming of greater significance. Regulations or even bans on the use of such materials as organochlorine insecticides and mercury fungicides exist in a number of European countries.

The necessity for these changes is considered in relation to the established knowledge of the possible or probable environmental effects of the materials concerned. The need for information on the possible environmental effects of new pesticides leads to considerable changes in the course of research and development programmes in the pesticide industry.

The last thirty years have covered a period of change from the use of simple inorganic poisons and plant extracts to the use of the diverse armoury of synthetic pesticides produced by a large scientifically based industry. The era of synthetic pesticides is usually considered to start from the recognition of the insecticidal properties of DDT in the early 1940's and from the discovery of the selective herbicidal properties of the synthetic phenoxyacetic acids shortly afterwards. The practical development of these materials was accelerated by the demands of war, so that DDT became widely used in the control of insect-borne tropical disease, and the hormone herbicides were rapidly pressed into service to improve the efficiency of cereal production. With hindsight, it is easy to point out that these developments proceeded with little regard for the possibility of any adverse long-term environmental effects. The intense war-time usage having disclosed no obvious deleterious effects, DDT and other organochlorine insecticides passed into widespread use in agriculture and almost twenty years passed before more detailed research started to disclose the more subtle ecological consequences of widespread and incautious use of powerfully biologically active substances possessing considerable powers of persistence in the environment.

The agricultural use of synthetic pesticides was nowhere more energetically developed than in the U.S.A., and it is not surprising that some of the early doubts as to the wisdom of existing practices arose there. The era of doubt, so far as public opinion is concerned, is usually thought to date from the publication of Rachel Carson's "Silent Spring". The appreciation of the possible hazards arising from pesticides has resulted in a very large volume of scientific investigations of their environmental effects. In practice, the large backlog of work on materials in established use, such as the organochlorine insecticides, has mainly attracted the attention of official and academic research laboratories, while industrial laboratories have largely been devoted to establishing the possible environmental effects of new materials under development. The amount of work now deemed necessary under the broad heading of environmental effects can amount to about one third of the total cost of development of a new pesticide and may take between five and ten years from the point of the initial observation of a potentially useful biological effect to the point of sale to a user.

The pesticide industry was an early target for attack in the period of growing public awareness of the importance of environmental pollution. It has now had some years to set its house in order, and the pressure of public opinion is increasingly directed to a wider range of pollution problems, including motor vehicles, factory chimneys, sewage effluents, oil spillages and jet aircraft.

The pattern of pesticide usage in Europe differs from that of other parts of the world, although it is broadly similar to that of North America, as shown in Table 1. In the developed areas of Western Europe and North America manual labour is becoming increasingly scarce and expensive and the pesticide market is dominated by herbicides. In the underdeveloped tropical countries with plentiful cheap labour, herbicides are less used but the demands for the control of insect borne disease, together with a high incidence of agricultural insect pests, result in insecticides taking top place.

The adverse ecological effects of the organochlorine insecticides are well known. Although much academic discussion is possible in relation to the details of the dynamics of the accumulation of these substances through biological food--chains, it seems prudent to limit the extent to which such persistent materials are allowed to permeate through the environment. The maintenance of present standards of insect-borne disease control in tropical countries is, however, dependent upon the use of DDT, in that no other material at present available can produce a similar effect at an economic cost. It is reasonable, therefore, that the first moves to limit the spread of DDT should take place in agricultural usages in the more developed countries, where alternative, but perhaps more costly, materials may be used.

Table 1

World usage of pesticides by groups of products

Country	% total usage for each area		
	Herbicides	Insecticides	Fungicides
N. America	61	25	7
W. Europe	49	25	23
Far East	25	45	21
Africa	31	42	28
S. America	30	53	16
Australia & N.Z.	45	38	16
C. Amer. & Caribb.	33	51	15

Table 2 lists some of the actions being taken in various European countries on the limitation of organochlorine insecticide usage. In the U.K. action to control the use of organochlorine materials has followed the two Reviews by the Advisory Committee on Pesticides and other Toxic Chemicals. The 1964 review recommended limitations on the use of the more toxic cyclodiene materials, while the second review in 1969 extended these limitations and further recommended the ending of DDT usage on a number of field crops, on bush fruit and for post-blossom applications on top fruit. Accurate figures for quantities of pesticides used on particular crops are not easy to obtain. The best available estimates are those of Strickland[1], whose figures for England and Wales for 1963-64 show a relatively modest annual consumption of DDT of about 250 tons. The restrictions on DDT usage imposed at present appear to affect about 30 tons of DDT annually, so that our action is perhaps more an affirmation of the desirability of control than a major step to reduce the input of DDT to the biosphere. In a local area, however the restrictions on the use of the cyclodiene compounds have produced measurable ecological results in reduced residue levels in predatory birds[2,3]. In some other European countries, the restrictions on the use of organochlorine insecticides affect more substantial applications, as in Spain, where there is estimated to be an annual use of about 1300 metric tons of DDT.

After the organochlorine insecticides, the pesticides which have attracted the greatest attention from those concerned with environmental pollution are the mercury fungicides. In some countries, and notably in Sweden, alkyl mercury compounds have been used in cereal seed-dressings, and there is good evidence from the work of Berg[4], Johnels[5] and others that alkyl mercury compounds derived from agricultural seed dressings accumulated to damaging levels into predatory birds in Sweden. The effect is clearly related to alkyl compounds, as distinct from phenyl or alkoxyalkyl mercury compounds. The latter types of compound, although possessing comparable acute toxicities to those of the alkyl mercury compounds, have a shorter residence time in the animal body and do not accumulate and penetrate to the central nervous system to the same extent as the alkyl compounds.

The ecological significance of agricultural mercury fungicides is complicated by the fact that, unlike organochlorine compounds, mercury compounds occur naturally in the environment. Mercury occurs naturally in the rocks of the earth's crust and is widely distributed by erosion and subsequent transport in water and air. Furthermore, mercury is an important pollutant from its many industrial uses. The use of phenylmercury acetate (PMA) as a fungicide in the paper pulp industry has been shown, again in Sweden, to be of particular importance, since PMA residues in the liquid effluent from paper mills can undergo microbiological methylation in river mud and the resulting methylmercury compounds can then accumulate through aquatic food-chains into fish.

In relation to other uses, the quantity of mercury used in agriculture is small. Data for the U.S.A. show that of a total annual mercury production of about 3000 metric tons, only 104 metric tons is used in agriculture. It is interesting to note that more mercury is used in dentistry (118 metric tons) than in agriculture. The presence of measurable mercury residues in tuna fish has obtained much publicity. One must remember that the tuna is a top predator of the open ocean ecosystem, and that it is not necessary to invoke any manmade pollution to account for small mercury residues in it, concentrated from the small levels of mercury naturally present in the ocean.

Table 2

Controls on organochlorine pesticides in Europe

Country	Chemical	Action
Sweden	Aldrin	Prohibited from 1/1/70
	Dieldrin	Prohibited from 1/1/70
	DDT	Use in home or garden forbidden from 1/1/70 Two year moratorium on agricultural use, but ban rescinded for forest insect control. Tobacco imports from spraying countries to be banned from 1973.
West Germany	Dieldrin	Prohibited
	DDT	Banned in agriculture from 1971, except for certain uses in forestry, which may continue until 1974.
Italy	DDT	To be banned from almost all agricultural uses.
France	DDT	Banned from use on fruit trees, potatoes, rape and vines, from June 1971.
Hungary	DDT	Banned from agricultural use.
U.S.S.R.	DDT	Use stopped. Further manufacture prohibited.
Finland	All OC pesticides	Banned from sale from 13/8/71, except for certain uses in forestry.
Spain	DDT	Banned from April, 1971, except for cotton and forestry, olives after harvest and vines before flowering.
Poland	DDT	Banned or discouraged.
Czechoslovakia	DDT	Banned or discouraged.

Phenylmercury and alkoxyalkylmercury compounds, applied as seed-dressings, are highly efficient fungicides. The normal PMA seed-dressing used on cereals in the U.K. applies 0.06 oz/acre (4.0 g/ha) of mercury to the soil. This small residue is degraded microbiologically in the soil. In experiments on the fate of PMA in soil, about 60% of the applied dose was lost as mercury vapour within 28 days[7]. The natural level of mercury in soil has been found to vary between 0.01 and 0.06 p.p.m. in agricultural land in England, with much higher levels in areas of metalliferous rocks[8,9]. The use of non-accumulative mercury fungicides as seed-dressings thus seems to be a negligible environmental hazard.

From the industrial point of view, the importance of looking at the changing pattern of usage lies in the guidance which this may provide on the types of materials which will be required in the future, and hence on what aims research and development programmes should have. A replacement for DDT, in the form of a material having a broad spectrum of activity yet a low vertebrate toxicity, capable of persisting as a residual deposit for 6-9 months, and yet lacking any properties of accumulation in the biological environment, could clearly be a valid research target. A replacement seed fungicide, to be used where organic mercury compounds are used today, could no doubt be sold, but there is little to suggest that the better of the mercury compounds pose any important environmental problems, so that any research programme on this topic would not warrant any high priority.

The amount of information required to establish the environmental safety of new materials presents some problems to the industry, particularly in relation to the scale of operation which is possible at early stages in the development of a new compound. Initially, only small quantities of the material are available, yet the more complex long-term effects of chemicals in the environment may not become evident until large areas of land have been treated. The manufacturer clearly wishes to know of any important environmental hazards at an early stage, before he is committed to the relatively costly production of large quantities. One approach is to make use of small, well circumscribed natural ecosystems and to study the effects of introducing the chemical into it, making observations on chemical persistence and degradation, effects on microbiological processes, changes in populations or species diversity in the fauna and flora and accumulation of the chemical into organisms. An alternative approach has recently been suggested by Metcalf[10], who has set up artificial simplified ecosystems in the laboratory into which the chemical is introduced and effects are noted.

During the last ten years we have faced the problems of the so-called side effects of pesticides, and I suggest that substantial progress has been made in recognising the difficulties and in replacing, or controlling the use, of the materials which present problems. On new pesticides, a very great deal of work is done to investigate possible environmental effects before they go into commercial use. It is highly unlikely that materials which could produce serious indirect effects, comparable to those of the organochlorine compounds, would go into unrestricted use today. We are perhaps moving, however, to a state of affairs where it is not the ecological implications of side effects which may concern us, but the effect of pesticides imprudently used to do what they were designed to do. Such matters as the unnecessary destruction of hedgerow or roadside verge vegetation by herbicides, or the use of insecticides in such a way that bees and other animal life of value or interest are affected. It is possible that regulatory mechanisms will be less concerned with bans or specific limitations and there is perhaps a need for codes of practice for the proper use of pesticides, to get the maximum benefit from their use in conjunction with cultural and other means of limiting pest damage.

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PRINCIPLES AND METHODS OF THE EUROPEAN COMMUNITIES WITH REGARD TO PESTICIDES AND PESTICIDE RESIDUES

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Summary A brief outline is given of the studies undertaken at the EEC in order to harmonize regulations on pesticides and their residues in foodstuffs in the member countries. These problems are presently a matter of study by various expert committees set up by the Commission to establish tolerance levels for pesticides residues on and in foods and feedingstuffs and to develop the standardized methods for the analysis of both pesticides and pesticides residues. The principles that have been adopted for reaching the required purposes are discussed.

The European Economic Community's establishing instrument, the Treaty of Rome, provides for the framing of common, Community-level legal provisions where differences between national provisions are liable to impede the operation of the Common Market and, in particular, the free movement of goods within the Community.

Among the activities under this heading is the harmonization of legislation on food, animal feedingstuffs and pesticides, with the object of removing technical obstacles to trade in these fields. Several directives on food and feedstuff additives have already been adopted and put into force by the Member States and a number of other proposed enactments, including draft directives and regulations on agricultural pesticides and pesticide residues in food and feedstuffs, are in the pipeline.

Before I go on to the subject of pesticides, it will be as well to give you an outline of Community rulemaking procedure. By the terms of the Treaty, all Community draft enactments have to emanate in the first instance from the Commission, and the Commission is empowered, too, to take implementing action. At the same time, all generally binding and all conspicuously important provisions including those concerning food, feedstuffs and pesticides require the endorsement of the Council of Ministers. Here the Community procedure is in two main stages.

First, the Commission department responsible prepares a preliminary draft and convenes meetings of experts from the member countries' Civil Services to discuss it. At this stage the experts do not actually speak for their Governments but they are able to help the Commission towards arrangements the Governments will regard as sound and practicable. The Commission also consults trade associations and consumer bodies in the Community on drafts in hand. When this preparatory work is completed the proposals embodying the results are submitted, with the Commission's endorsement, to the Council.

Next, the Council has the proposals studied by an ad hoc committee of Government officials, whose work is co-ordinated by the Committee of the Ministers' Permanent Representatives. At the same time it refers them for the opinion of the European Parliament--the European-level political assembly--and of the Economic and Social Committee, an advisory body consisting of representatives of trade and industry, agriculture, organized labour, and so on; the Parliament and the Committee in their turn consult their specialist committees and record their reactions in reports to the Council.

When views have been compared and agreement reached between the six Permanent Representatives and the Commission Representative, the final decision is taken by the Council of Ministers.

The Member States then have to put into effect the provisions enacted according to the requirements of the enactment. This depends on the legal basis given to it. In the case of food and feedingstuffs legislation the enactments normally resorted to are regulations and directives. Regulations are binding in every respect and have direct force of law in the Member States while directives are binding on the States as to the end result but leave the form and means to the discretion of the national authorities.

Community acts are published in the official gazette of the Community and come into force on the date appointed in them.

All the Member States have had regulations for years with regard to the trade and use of agricultural pesticides. To be officially accepted as agricultural pesticides, products have to go through a clearance procedure involving the furnishing of particulars as to their composition, efficacy, use pattern, toxicity, phytotoxicity, residues etc and their marketing too is governed by regulations concerning instructions for use, packing and labelling.

Though, generally speaking, similar in intention, the national regulations do differ in some respects which is why it has been found necessary to prepare Community directives on the subject. A first directive on the classification, packing and labelling of dangerous substances--including the active components of pesticides--was adopted by the Council in June 1967[1]. Two further sets of proposals, one on registration and marketing of pesticides and the other on classification, packing and labelling of dangerous preparations, are in course of elaboration.

Close attention is being paid in this connection to the recommendations of the international organizations which are concerning themselves with the same matters--in particular the Council of Europe's Partial Agreement and the various publications of FAO and WHO, which are standard works of reference for harmonization purposes. The problem of environmental pollution by persistent pesticides is also being carefully studied, and appropriate Community provisions will be enacted as and where necessary.

In recent years tolerances for pesticide residues on and in fruit, vegetables and grain have been introduced in the Netherlands, Germany, Belgium, Luxembourg and Italy. While the tolerances have been fixed with the same basic principles in mind--to safeguard consumers' health and to ensure high-quality agricultural production--there are various differences in regard both to the amounts and nature of the residues permitted and to the agricultural products concerned. In order to dispose of any impediments to trade which might result, the Commission plans to draw up a series of regulations on broad food classes such as fruit and vegetables, grain, dairy products and so forth, setting tolerances for the particular pesticide residues they may contain.

The fruit/vegetable class is being tackled first because many data are available. A draft first regulation, prepared by the working procedure described earlier, was submitted to the Council of Ministers in November 1968,[2] and is still pending, though approved meantime in the main by the Parliament and the Economic and Social Committee. Two further proposals, on fruit and vegetables and on grain, are in preparation.

The proposed first regulation lays down maximum values for some forty pesticide residues on and in fresh fruit and vegetables, listed under the headings of the Common Customs Tariff. The list of residues is not limitative; this means that pesticides not included in it will continue to come under the existing national rules until such time as Community provisions are enacted on them. The proposed tolerances are to apply from the time that the fruit and vegetables become available after harvesting. Exemption may be granted, however, for produce not for immediate consumption and subjected to appropriate controls to ensure that it cannot go to the consumer in its present state. There is to be a procedure for adjusting the tolerances in accordance with advances in technical and scientific knowledge or changes in health and farming requirements.

The tolerance levels set were arrived at by a procedure already in use in many European countries and in particular in most of the Community Member States, taking into account:-

(a) the permissible levels calculated from the acceptable daily intake with a food factor representing the average individual daily intake of the food class in question;

(b) the residue values arising in good agricultural practice, i.e. in the use of pesticides to the extent required by agriculture under normal (and not exceptional) incidences of pest infestation and disease;

(c) the results of check analyses on home and imported produce marketed in Member States.

Within the limits of the permissible toxicological levels, the tolerances are, as a rule, set at the lowest figure compatible with good agricultural practice. It is considered essential to reduce the food contamination hazard even where the permissible toxicological levels for food showed no actual need for this. It is after all possible for some pesticides to enter the human or animal organism otherwise than via the food intake, for example, the widespread use of household insecticides can provide a not inconsiderable source of residues. Moreover, the fact that different pesticide residues and other contaminants may be present together in foodstuffs and in the environment must not be disregarded, even though at present it is not possible to predict any potentiating effect that might occur.

It was not felt necessary to provide for detailed variation in tolerance levels with single food items; with a few special exceptions, the tolerances assigned to a given pesticide cover all the plant species of the same class. This system, using a single food factor representing the average individual daily intake of one food class, has the advantage of being easy to apply in operating controls and of allowing a certain latitude in the range of application of a pesticide without necessitating variation of the tolerance. In addition there is the point that values so calculated should not give rise to complications in international trade in quality products.

The Community proposals on pesticides and pesticide residues provide for harmonized sampling and analytical methods to be worked out for use in official controls on products coming under the enactments. Expert committees have been set up to formulate these and working groups are in action. As the task ahead is such a tremendously large and difficult one it has been decided to make the fullest possible use of methods already accepted by Member States or by national or international bodies. To this end consultations are in progress with IUPAC, CIPAC and the UK Committee on Analytical Methods. We hope very much that these will go forward in close and constructive international scientific co-operation for the greater good of trade within the Common Market and between Community and non--Community countries.

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SOME RECENT WORK ON FUNGUS-TRANSMITTED AND NEMATODE-TRANSMITTED VIRUSES

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Summary Since 1958, soil-inhabiting vectors have been identified for at least fifteen plant viruses. The vectors of some of these viruses are nematodes and of others fungi. Several of the viruses are of considerable economic importance. Where disease-escaping cultivars are not available, the nematode-borne viruses arabis mosaic, raspberry ringspot, tomato black ring and tobacco rattle can be controlled in raspberry, strawberry and potato crops by treating soil with nematicides, especially D-D. This is economically justifiable for high value crops, but barely so for potatoes. The spread of one fungus-transmitted virus, potato mop-top, can also be controlled experimentally by soil treatments. These include decreasing the soil pH to 5, or applying zinc sulphate, but they are not yet recommended for use by farmers on a field scale.

The first proof that some plant viruses are transmitted by soil-inhabiting vectors was obtained more than ten years ago, and these vectors are now known to include dorylaimid nematodes, and chytrid and plasmodiophoromycete fungi. In general however, there is considerable specificity between virus and vector, and any one virus is transmitted only by one or a few closely related species. The diseases caused by these viruses are widespread and include some of major economic importance, such as fanleaf of grapevines, which is common in Continental Europe and also occurs in many other countries, and wheat mosaic, which is mainly found in North America. In Britain, soil-borne viruses cause considerable losses in crops of strawberry, raspberry, potato, pea and lettuce. However most of the viruses are patchily distributed over the country, so that many crops are unaffected though a good proportion of those that are affected have a high incidence of infection.

In this short paper I have no space to discuss all fifteen or more viruses whose soil-inhabiting vectors are known, and I shall deal mainly with recent studies on two of the viruses at the Scottish Horticultural Research Institute, with an emphasis on the possibilities of using soil treatments to prevent their spread.

Nematode-transmitted viruses

Most nematode-transmitted viruses fall into one or other of two groups, called the nepoviruses and the tobraviruses (Harrison et al., 1971). The nepoviruses have isometric particles about 30 nm in diameter, are transmitted by species of Xiphinema or Longidorus, and are exemplified by arabis mosaic virus. The tobraviruses by contrast have straight tubular particles, are transmitted by *Trichodorus* spp and are exemplified by tobacco rattle virus. For some years evidence has accumulated to show that these viruses can persist in their vectors for months or even years. However the nematodes do not retain them through a moult and there is no good evidence that the viruses multiply in their vectors. Our understanding of the transmission process has recently been greatly strengthened by the work of Taylor and Robertson (1969, 1970a, 1970b), who found by examining thin sections of virus-carrying nematodes in the electron microscope, that the virus particles become associated with either the guiding sheath (*Longidorus*) or the oesophageal wall (*Xiphinema* and *Trichodorus*) of the vectors. Thus although many details remain to be established, transmission seems essentially to involve the reversible association of virus particles with specific parts of the surface of the alimentary tract that are bathed with ingested plant material before it reaches the intestine.

The ecology of *Xiphinema* and *Longidorus* spp in relation to that of several of the viruses they transmit in Britain, the role of seed transmission in the survival and spread of these viruses (Murant, 1970), and virus control by nematicide treatment of soil (Harrison, Peachey & Winslow, 1963; Murant & Taylor, 1965) have been well studied, but information on the viruses transmitted by *Trichodorus* spp was more scanty. Recent work has done much to fill this gap. Cooper (1971), in a more sophisticated survey than any done previously, was able to relate the occurrence of tobacco rattle virus and of *Trichodorus* spp to soil type. Using the soil maps made by the Soil Survey of Scotland, he found *Trichodorus* vectors in nearly 90% of fields on freely drained podsols and tobacco rattle virus in almost 70%. Of the vector species, *T. pachydermus* predominated on some soil series, *T. primitivus* on others, the two species occurred together on several soil series and on one series neither virus nor vectors were found. Neither virus nor vectors occurred on heavy, poorly drained soils. Thus the chance of occurrence of tobacco rattle virus can be estimated from a knowledge of the soil series on which a field lies.

Other work indicates that rainfall has an important effect on the spread of tobacco rattle virus, presumably by affecting vector movement, and that weeds are common overwintering reservoirs of the virus (Cooper & Harrison, 1969, 1971). The effects of chemical control of weeds are complicated and interesting. Rigorous weed control during the life of a crop of potatoes, in which tobacco rattle virus causes one type of spraing disease, only slightly decreased the incidence of the disease, but continuing the weed control for another year trebled the amount of spraing that developed in the second potato crop on the same plots, as compared to the incidence in the weed-infested control plots. Presumably this happened because virus-carrying nematodes had no alternatives to potato as hosts in the herbicide-treated plots, whereas in the weed-infested plots the weed plants were preferred to potato by many of the nematodes (Cooper & Harrison, 1971). Weed control therefore seems unlikely to aid the control of spraing disease in the short term and, although we have evidence that the disease is less prevalent when the land is kept free of plants for nine months before potatoes are planted, this would rarely be a practical proposition.

Although a few potato varieties are field immune (Arran Pilot) or resistant (Redskin) to infection with tobacco rattle virus, many of the most popular cultivars are susceptible to infection and several react severely. Cooper and Thomas (1971) therefore tested the ability of several nematicides to control Trichodorus and virus spread. Comparing different plots at one site they found it was necessary to kill 94% of the Trichodorus to decrease the spraing incidence by 90% (Fig. 1). Results obtained with three of the most effective chemicals are shown in Table 1. All substantially controlled spraing in the first crop after treatment, but only D-D gave good control in the second year and none was effective in the third year.

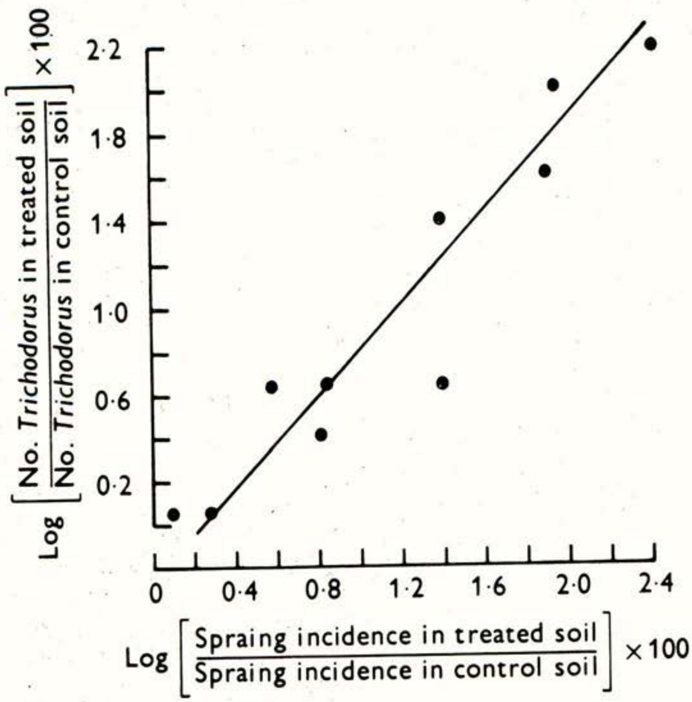

Fig. 1. The relation between number of Trichodorus nematodes in May and incidence of potato spraing in Pentland Dell tubers from plots treated with nematicides (Cooper and Thomas, 1971).

The numbers of Trichodorus at this site were small, but for D-D and dazomet the control of spraing paralleled the decrease in Trichodorus numbers. With methomyl the position was different, control of spraing in the first potato crop being achieved without a corresponding decrease in nematode numbers. This probably happened because methomyl has adverse effects on the nematodes without killing them, as was found with Xiphinema and Longidorus (Taylor and Gordon, 1970). Methomyl is systemically translocated in plants, although its effect in these tests probably mainly resulted from its contact action. Whether systemics without contact action will prove useful remains to be established, but it would be surprising if any act sufficiently rapidly to prevent virus-carrying nematodes inoculating viruses to nematicide-containing cells.

Table 1

Effect of three chemicals on Trichodorus numbers and spraing incidence*

Nematicide+ (kg/ha)		No. Trichodorus per 200 g soil			% tubers with spraing		
		Oct 1969	Aug 1970	Aug 1971	1969	1970	1971
D-D	(224)	0	0.2	3	2	0.8	6
Dazomet	(168)	0	6	11	8	9	9
Methomyl	(9)	2	7	7	2	13	9
None		3	8	8	31	12	8

*Data from Cooper and Thomas, 1971; and J. I. Cooper, unpublished
+Applied April-May 1969

Of the nematicides tested, D-D was therefore the most effective, used at about 200 kg/ha. In earlier work, Harrison, Peachey and Winslow (1963) found that, on a heavy soil in England, late autumn application of D-D was somewhat more effective than spring application for killing Xiphinema diversicaudatum, the vector of arabis mosaic and strawberry latent ringspot viruses. Similarly, on light land in Scotland autumn applications were more effective than spring applications for killing Trichodorus (Table 2) and for controlling potato spraing disease (Table 3).

Table 2

Effect of D-D on Trichodorus numbers*

D-D application (kg/ha)	No. Trichodorus per 200 g soil			
	May 1970	Aug 1970	May 1971	Aug 1971
224, autumn +	0	0.2	0.4	2
112, autumn +	0	1	3	9
224, spring ‡	5	0.2	12	4
112, spring ‡	12	7	61	14
None	204	34	191	22

*Data from Cooper and Thomas, 1971; and J. I. Cooper, unpublished
+Applied October, 1969
‡Applied March, 1970

Table 3

Effect of D-D on spraing incidence*

D-D application (kg/ha)	% tubers with spraing 1970	1971
224, autumn +	0.4	0.3
112, autumn +	0.8	1.8
224, spring ‡	0.2	2.4
112, spring ‡	5.4	3.2
None	21.4	26.6

*Data from Cooper and Thomas, 1971; and J. I. Cooper, unpublished
+Applied October, 1969
‡Applied March, 1970

Presumably the main reason for the superiority of autumn application is that the fumigant was left in the soil for several months longer than was possible after spring application.

In Scotland, where seed potatoes are grown only once every six years or so, it is obvious that nematicides will benefit only a single potato crop and the cost of D-D treatment may not be justified. However, spraing caused by tobacco rattle virus is more important to the ware grower than to the seed grower because the virus disfigures the tubers but is usually not passed from mother tuber to progeny plant. Thus D-D application in autumn may be an economic proposition for ware growers who wish to grow crops that are susceptible to tobacco rattle virus or Trichodorus damage more often than one year in three. In other circumstances the emphasis would be changed to finding a material that will merely protect the first crop after treatment. For this purpose substances of the methomyl type have the advantage that they are translocated to the haulms and have an aphicidal action in addition to their effects on nematodes.

A fungus-transmitted virus

Now that seed potatoes are so free of the better known potato viruses, potato mop-top is the commonest virus in many stocks. Potato cultivars differ greatly both in susceptibility to infection and in severity of reaction to the virus, and no immune variety is known. The virus is transmitted by the powdery scab fungus (*Spongospora subterranea*), and although potato is not the only host of the fungus in Britain it is the only species in which the virus is known to occur naturally. The virus survives between potato crops in the resting spores of *S. subterranea*, in which it can be retained for several years (Jones and Harrison, 1969).

Although potato mop-top virus has such a restricted host range, and its control therefore presents a very different problem to that of tobacco rattle virus, again crop rotation is not effective, presumably because of the virus reservoir persisting in the soil in the form of virus-carrying resting spores of *S. subterranea*. However in the absence of new infections from the soil the virus is gradually self-eliminating from potato stocks, because it is only transmitted to about half of the tubers derived from an infected mother tuber. In cultivars that develop obvious symptoms of infection, the elimination process can be much accelerated by careful rogueing out of affected plants, but in other cultivars rogueing is less effective because of the difficulty in identifying infected plants.

Rogueing should if possible be supplemented by a method of preventing infection from the soil, and I will describe two ways of doing this on an experimental scale. In the first, sulphur is applied to the infested soil to lower its pH to about 5. This decreases by 90% or more the incidence of powdery scab, and the spread of potato mop-top virus. The fungus is not killed but infection is inhibited and, when the pH of the treated soil is raised to about 7, indicator plants grown in it again become infected with the virus (Jones and Harrison, 1968). This method has the disadvantage that heavy applications of sulphur would be needed on some soils, and that the sulphur causes scarring on some tubers and decreases tuber weight. It could however be useful for the small plots used to build up Virus-tested stocks from single-plant progenies, where tuber number is more important than tuber size.

In the second method, soil is treated with zinc-containing materials, which kill the zoospores of S. subterranea, and which are used to control crook root of watercress, a disease caused by the watercress form of S. subterranea (Tomlinson, 1958). At first we used a zinc frit, but found that although this prevented virus spread it also caused necrotic leaf spotting and premature senescence of the haulms (Cooper and Harrison, 1971). Later we found that these undesirable effects were caused by boron contained in the frit, and that virus spread can be largely prevented by applying zinc sulphate to the soil. Relatively large applications (300 kg zinc per ha) are however needed to give good control, and the method may not be economic on a large scale. Also, although zinc compounds are relatively non-toxic, the hazards to other crops or to stock using the land subsequently need to be assessed carefully. Again, this method is likely to be of most use for the small plots used in the propagation of Virus-tested stocks.

In this paper, I have tried to provide some illustrations of the kinds of problems that are posed by soil-borne viruses. Plainly the development of control measures is still at an early stage, but the present indications are that the successful outcome of this work may not be without interest for those concerned with crop-protection chemicals.

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PROBLEMS AND PROGRESS IN THE USE OF SYSTEMIC FUNGICIDES

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Summary This review has confirmed the overwhelming success of the systemic fungicides in controlling a large and varied collection of plant diseases. With products representing several major groups of chemicals including the oxathiins, pyrimidines, benzimidazoles, thiophanates and the morpholines it is now possible to control a number of Ascomycetes and Basidiomycetes that attack higher plants. However none of the products currently available are able to control any of the Phycomycetes. Thus their spectrum of activity generally seems to be very specific though not necessarily limited.

The degree of systemicity appears to be largely dependent on (a) the chemical structures of the compounds concerned; (b) the physical and physiological status of the crop; (c) the method of application adopted in practice; (d) the environmental conditions during and after treatment. With these generalisations in mind various systemic products have now been successfully used as seed, soil and foliar (stem) treatments of crop plants and those treatments resulting in maximum distribution of the active ingredients have generally given best results.

Unfortunately in the short time that systemic fungicides have been in use several instances of resistance have already been reported. This upsurge is attributed to the application of "high selection pressure" to a varied wild population of pathogens. By analogy with the field of antibiotics and insecticides various measures are recommended to prevent or delay the onset of resistance to systemic fungicides.

INTRODUCTION

It has been shown (Evans 1968) that dosage response curves for the control of potato blight with non-systemic fungicides such as Maneb, frequently have a ceiling performance under field conditions that is independent of spray frequency. This upper limit is probably due to inadequate distribution of toxicant rather than a lack of activity. Similar reasoning can be applied to the use of other non-systemics as the distribution and persistence of any fungicide on or in the host plant finally limits its performance. The distribution and persistence of fungicide deposits can frequently be modified by formulation or application technology, but on a practical basis there are limits to what can be achieved by these methods, and only by the discovery of truly systemic fungicides have these limitations been overcome.

Such products first became available on an economic basis when the oxathiin, Carboxin, was introduced as a seed dressing for the control of the loose smut pathogen Ustilago nuda on barley (von Schmeling & Kulka, 1966). Today there are at least five major groups of systemic fungicides available in commercial practice.

BIOLOGICAL SPECTRUM OF MODERN SYSTEMIC FUNGICIDES

During the past three years there have been several reviews of the use of systemic fungicides (Byrde 1969, 1970) (Evans 1971) in which the widespread application of these products in both tropical and temperate crops against a wide variety of pathogenic fungi have been noted. Products are now available based on compounds representing the oxathiins, pyrimidines, benzimidazoles, thiophanates and the morpholines.

<u>Oxathiins</u> Represented by the analogues Carboxin and Oxycarboxin the former product being the first truly synthetic systemic fungicide to be used in agriculture as a seed dressing for smuts. Spectrum of activity almost confined to the Basidiomycetes especially the rusts and smuts (Edgington et al 1966).

<u>Pyrimidines</u> Spectrum of activity generally confined to the Ascomycetes in particular the powdery mildews (Erysiphales). The simpler analogues ethirimol and dimethirimol are specific in activity towards the powdery mildews (Elias et al 1968) but Triarimol is now being developed as a multipurpose fungicide due to its activity against several other Ascomycete fungi (Gramlich et al 1969).

<u>Benzimidazoles</u> Compounds with the broadest spectrum of activity noted to date being active against many Ascomycetes and certain Basidiomycetes but specifically inactive towards Phycomycetes. Commonly represented by benomyl (Delp & Klopping 1968) and thiabendazole (Staron and Allard 1964).

<u>Thiophanates</u> Derivatives of thioallophanic acid thiophanate methyl and thiophanate are being commercially developed as direct competitors of the benzimidazoles having similar spectra of activity (Ishii 1970, Aelbers 1970) and possibly a similar mode of action (Selling et al 1970).

<u>Morpholines</u> Dodimorph and Tridimorph are now commercially available in Northern Europe (Pommer & Kradel 1967), the former recommended for the control of rose mildew (Sphaerotheca pannosa) and the latter primarily for the control of barley mildew (Erysiphe graminis).

Systemic control of the rusts, smuts and powdery mildews as well as the ubiquitous Botrytis cinerea on soft fruit is excellent and superior to that of most non-systemics but there is still a need for a systemic product that will control Phycomycetes, a group that includes several of the worlds most devastating plant pathogens.

SYSTEMICITY - ITS PRACTICAL SIGNIFICANCE

Systemic fungicides are defined as compounds which are capable of penetrating and moving within the tissues of a given host plant and there inhibiting the development of a known parasite. They may act as direct toxicants of that pathogen (Snel et al 1970) or else give rise to toxic derivatives (Clemons & Sisler 1969). Such substances would be expected to move considerable distances within the host tissues via the normal transport systems based on the xylem transpiration stream or else in the phloem translocatory system.

Evidence to date suggests that all practical systemics in use today are moved unidirectionally in the transpiration stream being carried upward and outward in the manner shown by Snel & Edgington (1970) to occur in the oxathiins. Bipolar movement in the phloem system is theoretically possible but none of the better known systemic fungicides are capable of mass movement in this fashion.

According to Peterson & Edgington (1970) it is probably more important to distinguish between movement of substances within the living (symplastic) as opposed to the non-living (apoplastic) transport systems in higher plants. In this context movement in the transpiration stream is said to be apoplastic and thereby dependent on the physical factors which affect the process of transpiration. The distribution of functional stomata obviously play an important part in affecting the rate of transpiration and eventually might also affect the final pattern of distribution of the fungicide within the tissues. When substances are constantly supplied to the transpiration stream they eventually tend to accumulate at leaf margins and will induce toxic symptoms at the edges of these leaves if the therapeutic index is not large enough.

As the superiority of systemic fungicides is dependent on their internal distribution it is important to know what factors can affect the rates and patterns of movement within the host plant. From a practical point of view movement can be considered as dependent on,

(a) the molecular configuration of the active ingredient (i.e. the choice of product)

(b) the structure and physiological status of the host (i.e. the agronomic status of the crop)

(c) the method of application to be used (i.e. the placement of active ingredient on the crop)

(d) environmental conditions during and after treatment

Of the four factors listed here only two can be manipulated by the farmer, and much care should always be taken in the correct choice of product and its application in the most desirable fashion.

SYSTEMIC FUNGICIDES AS SEED DRESSINGS

Traditionally seed dressings are used to destroy either superficial seedborne pathogens or to protect seedlings from soilborne pathogens but the introduction of systemic products has added two further possibilities

(a) the control of deep seated pathogens previously inaccessible to chemicals

(b) the control of airborne pathogens of annual crops by using the seed dressing as a reservoir of fungicide during the growth of the crop

The control of deep seated loose smut diseases of wheat and barley by seed dressing has now been clearly established as a practical method and ethirimol is widely used for the control of barley mildew (Erysiphe graminis) (Bent 1969). The dressing process often causes some difficulties as formulation possibilities are already limited by the physico-chemical nature of the active ingredient. This is further complicated by the type of machinery used for dressing the seed, and by the loading capacity of the seed. This is often a limiting factor when attempting to apply high rates of dressing intended to give a prolonged period of fungicidal activity.

As most seed samples are kept dry prior to planting chemical penetration is assumed to take place primarily through the roots at germination. This undoubtedly occurs with the oxathiins (Snel & Edgington 1970) and the pyrimidines (Bent 1969). It can be shown that cotyledons in phaseolus beans are equally effective absorptive organs by removing the testa from beans that have been soaked for 24 hours in distilled water, applying a dressing such as Oxycarboxin to the naked cotyledon and germinating in a moist atmosphere. After 14 days seedlings can be infected with the bean rust pathogen (Uromyces phaseoli) for assay purposes. By this method it can be shown that the testa is an extremely effective barrier to the uptake of the dressing and that cotyledons are very effective absorptive organs as those treatments applied to the seed coat had little effect whilst those applied directly to the cotyledons gave good disease control (Evans unpublished).

SOIL TREATMENTS WITH SYSTEMIC FUNGICIDES

Previously fungicides were only applied to soil for eradication of soilborne pathogenic fungi prior to planting or placed near the germinating seed to prevent attacks by such organisms. The highly selective mobile character of modern systemics means that even perennial crops can be treated in situ and airborne pathogens controlled following root uptake. Thus a benomyl soil drench is recommended for the control of Verticillium wilt of cotton (Hine et al 1968) whilst cucumber mildew is controlled by soil treatment with a variety of products (Aelbers 1970). Spray treatments sometimes allow more rapid control of airborne pathogens such as cucumber mildew (Evans 1971) but may eventually prove to be inferior.

Soil is a notoriously complex medium in which to practice the art of chemical control and systemic chemicals, like all others, are subject to a variety of physical, chemical and biological inactivating forces. Thus the performance of any chemical might differ considerably under various soil conditions as shown by Hock et al (1970) who reported that the uptake of benomyl by roots of elm seedlings was much greater in plants grown in sand culture than in normal potting compost. There was, however, no opinion given as to whether this was due to root structure or to availability differences associated with these substrates.

LEAF AND STEM TREATMENTS WITH SYSTEMICS

The biological activity of any foliar fungicide deposit is ultimately dependent on the inherent fungitoxicity of the product used and the amount of active ingredient which reaches the site of action. The availability of the toxicant is in turn dependent on its distribution and persistence in or on the plant. The initial distribution of any spray deposit, whether systemic or non-systemic, is largely based on the method of application used. The effective distribution for disease control is however the redistributed pattern induced by surface movement of the non-systemic in solution, suspension or vapour (Evans 1968) and by the internal transport of the absorbed toxicant when a systemic is applied. The advantages of systemicity can easily be seen by placing the same quantity of fungicide per leaf either as one or more droplets and then uniformly inoculating the leaves with a known pathogen. Morgan (1952) showed how the activity of a copper deposit could be improved by better distribution and this exercise is even more rewarding with systemic products such as benomyl used on cucumber foliage for the control of mildew. Since systemic fungicide deposits are not subject to excessive weathering like the majority of non-systemics their persistence tends to be better regulated and more dependent on the applied dosage. Even so uptake of systemics is very dependent on environmental conditions during and after application and can be made more effective by the addition of various spray supplements such as glycerol (Gray 1956) and certain surface active agents (Brown & Hall 1970).

Whilst spray or dust treatments may be perfectly adequate for the control of foliar pathogens other techniques are required for the control of systemic pathogens such as Dutch elm disease (Ceratostomella ulmi) and silver leaf disease of stone fruit (Stereum purpureum), both of which could probably be treated by stem injections similar to those used for the correction of mineral deficiencies. Holes are bored tangentially into the trunk and either a dilute solution or a concentrated solid preparation inserted. The former technique is cubersome and the latter can lead to problems of phytotoxicity unless the safety margin of the compound used is very high. Experimentally a whole variety of other techniques have been used for trunk injections and several have been listed by Wain and Carter (1967) in their review of systemic movement.

RESISTANCE TO SYSTEMIC FUNGICIDES

In the short time that has elapsed since the commercial introduction of the first truly synthetic systemic fungicide there have been more reports of resistance to fungicides than ever before in the whole of the history of fungicide usage. Two of the first examples noted were the failure to control cucumber mildew by benomyl (Schroeder & Provvidenti 1969) and dimethirimol (Anon 1969). Since then cross resistance has been recorded within the benzimidazoles (Bartels and McNeill 1971) using mutant forms of Fusarium oxysporium as test organisms. This is a classical pattern of events with resistance of any kind, in particular when dealing with insecticides, and therefore by analogy with this field of experience one might predict that this upsurge of resistance to fungicides is the result of applying a "high selection pressure" to an extremely variable wild population of pathogenic organisms.

The degree of variability and the high rate of reproduction of fungi is well documented and the question therefore arises as to what constitutes a high selection pressure? Experience of systemics in general suggests that this pressure is in some way related to the exceptional efficiency of fungicide treatments in controlling many diseases and that this in turn is dependent on

(a) the high inherent activity of these fungicides

(b) a very efficient distribution of toxicant within host tissues

(c) the specific character of most systemic fungicides

If this reasoning is true then it would seem logical to avoid excessive dependence on any of these three factors by

(a) using the least specific products whenever possible

(b) adopting mixtures or mixed schedules

(c) avoiding sublethal dosages and prolonged contact with the pathogen

(d) making less stringent use of systemicity if possible.

Genetically resistance appears to be a matter of Darwinian selection and according to Hastie (1970) no evidence of mutagenesis in the resistance of certain Aspergillus Nidulans strains to benomyl is apparent although it seems to have induced some genetic instability in the normal population. Since therefore the process of resistance is basically selective, it must be assumed that the resistant strain is normally present in small numbers being under some form of competitive disadvantage, possibly from the susceptible strain or from some other extraneous micro-organisms. If the former is true then it would seem logical to use these highly efficient fungicides in a manner most likely to leave a residue of the susceptible strain to assist in suppressing the upsurge of the resistant organisms.

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ORGANOCHLORINE ALTERNATIVES - A REVIEW OF THE PRESENT POSITION IN THE UK

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Summary For most British crop pests, adequate alternatives are available to replace the persistent organochlorines which have now been largely withdrawn from use. A number of pests, including several soil-dwelling organisms, are not controlled by OC alternatives. Crops at risk from complete withdrawal of organochlorines include vegetables, strawberries and narcissus. General problems are that pest control is becoming more expensive, more hazardous to operators and to plant health, and often less effective than in years when organochlorines were in common use.

INTRODUCTION

Several organochlorine insecticides fall short of the qualities possessed by the ideal pesticide. Residues of aldrin, dieldrin, DDT and heptachlor persist in the environment, have deleterious effects upon beneficial and general wild-life, accumulate within the body tissues of predators and higher vertebrates, and induce resistance problems in some of the pests which they formerly controlled.

These undesirable qualities have led to a substantial decline in the use of such organochlorines - a decline accelerated in many countries by governmental advice or legislation, whereby offending pesticides have either been banned or their use has been severely curtailed. In the United Kingdom, voluntary curtailment of OC usage followed governmental recommendations arising from reviews of the pesticide situation (Anon, 1964; Anon, 1969), so that by 1972 few approved uses will remain for DDT, aldrin and dieldrin. Heptachlor is not available in this country, nor is DDT for home or garden use.

Much interest now centres on methods of control other than chemical, including cultural, physical, biological and integrated measures. On the chemical front the search has intensified for alternative pesticides which give effective control of pests of plants but which are not subject to the limitations of the offending organochlorines.

This paper attempts an assessment of how successfully we have provided chemicals to fill the gaps left by the withdrawal of so many uses of OC pesticides in the United Kingdom. Other speakers in today's session will consider in detail the replacement of organochlorines for use on cereals, oil-seed rape, sugar beet and some vegetable and fruit crops.

THE PEST SITUATION

For present purposes it is convenient to place British crop pests into the following four categories.

Group 1 - pests not controlled by organochlorines or by alternative pesticides.

Group 2 - pests which are effectively controlled by the OCs and by the alternatives (organophosphates, carbamates etc.) which have replaced them.

Group 3 - pests against which the OCs were of little or no use, but which are controlled by alternative chemicals.

Group 4 - pests against which the OCs were effective, but for which we have not as yet developed useful alternatives.

Table 1

Classification of crop pests

Group	Organochlorine effectiveness	OC alternatives effectiveness	Examples
1	-	-	Birds
2	+	+	Lepidoptera
3	-	+	Aphids, slugs
4	+	-	Millepedes

Using this classification, we can now examine the Ministry of Agriculture's List of Approved Products for 1971, in which chemicals officially approved for use in pest control are grouped under various crop headings. We are interested chiefly in Group 2 and Group 4 pests. For Group 2 pests we want to know whether the OC alternatives really are giving as effective a degree of control as the materials they replaced. The presence of Group 4 pests will highlight any gaps in our chemical armoury left by the withdrawal of OC uses.

Table 2

Problem pests in British agriculture

Crop	Group 2 pests (OC's effective, OC alternatives supposedly effective)	Group 4 pests (OCs effective, OC alternatives not effective)
Field brassicas	Flea beetles	Mustard beetle
Cereals	Leatherjackets	Frit fly Wireworm
Grass seed		Leatherjackets
Legumes	Leatherjackets	Seed weevils
Potatoes	Cutworms	Chafer grubs Flea beetles Millepedes
Beet		Chafer grubs Cutworms Nematodes Flea beetles Millepedes Wireworm

Table 2 (continued)

Crop	Group 2 pests	Group 4 pests
Vegetables	Cutworms Cabbage root fly Caterpillars Flea beetles Carrot fly Onion fly	Chafer grubs Millepedes Asparagus beetle Bean seed fly Gall & stem weevils Leek moth Pea thrips
Top fruit	Capsid bugs (apricot, peach) Cherry fruit moth (plum, cherry)	Chafer grubs Pear midge Wasps (nests) Weevils Winter moth (apricot, peach) Nut weevil
Bush fruit	Raspberry moth Blackcurrant gall mite Winter moth	Raspberry cane midge 'Red berry' mite Strawberry rhynchites Weevils Blackcurrant leaf midge
Soft fruit	Cutworms	Xiphinema eelworms Chafer grubs Strawberry mite Strawberry blossom weevil Strawberry rhynchites Stem eelworm
Hops		Flea beetle Hop root weevil Hop strig midge
Flowers and ornamentals		Capsid bugs Earwigs Narcissus bulb fly
Turf	Chafer grubs Earthworms Leatherjackets	
Glasshouse crops	Root knot eelworm Bulb scale mite	Ants Cutworms Earwigs Vine weevil Narcissus bulb fly Rose caterpillars Tarsonemid mite Millepedes

The above list contains among Group 4 pests several soil-dwelling organisms which feed on a wide range of plants, and against which the organochlorines were so effective because of their persistence. Chafer grubs, wireworms, ants and millepedes come into this category, together with general feeding nematodes such as Xiphinema and Ditylenchus dipsaci.

Specific pests in Group 4 include narcissus bulb fly, so adequately controlled by aldrin, and bean seed fly, for which dieldrin seed dressings are still largely effective. The remaining Group 4 pests are at present of minor importance, though changing cultural practices as in sugar beet growing may well bring some of them into prominence.

Turning now to Group 2 pests, we note the frequent mention of cutworms and leatherjackets. A number of insecticides such as trichlorphon, fenitrothion, tetrachlorvinphos and methiocarb give excellent in vitro results against these pests, but field trials are often disappointing and suggest that we are faced with problems of how to apply the alternatives effectively.

Another point of interest is the presence in Group 2 of such important dipterous pests as cabbage root fly, carrot fly and onion fly. Larvae of all three, and of bean seed fly, were controlled by organochlorines until resistance developed in many parts of the country (Coaker, Mowat and Wheatley 1963; Wright and Coaker, 1968; Gostick and Baker, 1966; Gostick, Powell and Slough, 1971). No satisfactory alternatives to aldrin or dieldrin as a root-dip for brassicas has yet been developed, and post-planting spot drench or granule treatments of less persistent alternatives require very careful placement if they are to be successful.

The problem with carrot fly lies not so much in controlling first or second generation larval damage, for which phorate, disulfoton or other materials are adequate, but in controlling the partial third generation larval damage, which is so prevalent on carrots left in the soil during the winter and early spring months.

As far as onion fly is concerned, OC resistance has occurred sporadically in the main onion-growing areas. There are probably several insecticides which give control when applied as granules at sowing, but seed dressing formulations are often phytotoxic to a greater or lesser degree (Wells and Lacy, 1968; Perron, 1968).

Tarsonemid and eriophyid mites also occur in Group 2. Endrin and endosulfan have proved far superior to alternative chemicals, and lime sulphur can hardly be regarded as a satisfactory alternative for use against blackcurrant gall mite.

The Crop Situation

Although pest problems arising from OC withdrawal are to be found in most crops (Table 2), they are of particular concern in vegetables, strawberries and narcissus. The major pests of brassica seed crops attack the plants at the flowering stage. Few alternatives to BHC and DDT are less toxic to bees and none are as effective against pollen beetle and seed weevil as BHC, but several, including azinphos-methyl and malathion, give adequate commercial control.

It is interesting to note that of the six fruit pests listed by Dicker (1965) as needing effective OC alternatives, only two (blackcurrant gall mite, strawberry blossom weevil) remain on my problem list, a number of effective alternatives now being available for the control of pre-blossom apple pests such as winter moth and tortricid larvae, apple sucker, apple blossom weevil and aphids (Vernon and Gould, 1971).

Problems in OC withdrawal

UK governmental agencies recognised that it was not possible to impose a complete ban on persistent organochlorines without leaving certain crops vulnerable to pest attack (see above). The 1971 List of Approved Products therefore retains certain uses for aldrin, dieldrin and DDT, although such uses will no doubt be rapidly phased out as effective alternatives are provided.

The organochlorines were so effective partly because of their persistence, a single pre-planting soil application or one foliar application often giving virtually complete protection for the whole season. For approximately ten years, carrots were grown in the UK with the almost complete protection from carrot fly afforded by pre-sowing dieldrin soil treatment. The degree of control of this and other pests, to which farmers and consumers became accustomed during the aldrin/dieldrin/DDT era of the late 1950s and early 1960s, is not likely to be achieved by the non-persistent chemicals now available.

The cost of pesticide application is becoming much greater as a consequence of OC withdrawal. Not only is it becoming more expensive to develop and market new pesticides, but several applications of the non-persistent alternative may be required compared with only one application of the persistent organochlorine. Pest control now costs approximately three times as much for materials alone as for organochlorines, and in some cases a good deal more. The cost of pest control measures will thus figure more prominently in the variable costs of production for many crops. To give one example, frit fly attacking maize can be controlled by phorate or chlorfenvinphos, but the cost of such treatment can scarcely be justified for maize grown for fodder, as this crop has only a small profit margin.

Many of the problems listed in Table 2 involve minor uses of pesticides, and this raises the question of who is going to test candidate pesticides for crops or crop pests which are of little financial value. The chemical industry justifiably looks to major uses for its products and can hardly be expected to screen chemicals for such unimportant pests as strawberry rhynchites, blackcurrant leaf midge or hop root weevil. Much of the developmental work on minor uses of pesticides is done at present by the Ministry of Agriculture's Advisory Service and by research stations, but whether they will continue to perform this work is uncertain.

One of the best features of the organochlorines was their comparatively low acute toxicity hazard to operators, and the paucity of records of poisoning in this country following the use of DDT, aldrin and dieldrin testifies to this fact. Many of the alternative pesticides are a good deal more poisonous in terms of oral and dermal toxicity and contractors, farmers and operators will need to pay even stricter attention to the safety precautionary measures detailed on the pesticide label.

Another virtue possessed by organochlorines was their comparatively low phytotoxicity. Seed dressings, root dips and foliar treatments of the persistent OCs could be applied to most crop plants without fear of inducing phytotoxic symptoms. Their replacements often lack this virtue. Thus the search for a cheap and efficient alternative to dieldrin seed dressing for onion fly maggot control is hampered by the extreme phytotoxicity of many organophosphates and carbamates when applied as seed treatments to bulb or salad onions.

One of the constant demands of conservationists is that pesticides should be more selective in action, so that ideally only the target pest species is affected by the chemical. Selectivity is obviously important when pesticides are used in integrated control programmes. Apart from new chemicals such as pirimicarb, dicarzol and formetanate which are highly selective against aphids and mites respectively, I see little sign of greater selectivity nowadays than in the days when organochlorines reigned supreme. For example, aldicarb, carbaryl and fenitrothion cover a spectrum of pests as broad as, or even broader than, the organochlorines.

Conclusions

I began by listing certain undesirable features of the persistent organochlorines which led to their severe curtailment in the United Kingdom. Let me end by recalling the virtues of DDT, aldrin, dieldrin and other persistent organochlorines. They were cheap, easy and safe to apply to crops and often gave well-nigh perfect control of many pests.

By withdrawing so many uses for organochlorine insecticides, we move into an era when pest control in this country may be less effective for most crops, more hazardous to those who have to apply the new chemicals, and certainly more expensive. If it is not possible to educate the consumer public into accepting produce with a small amount of pest damage, then we may have to forsake reliance on chemical control methods alone and integrate the new non-persistant pesticides with non-chemical methods to give the degree of pest control to which the British farmer and public have become accustomed. Little progress has so far been made in developing integrated control methods in this country.

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