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COMPUTATIONS ON SOIL FUMIGATION WITH METAM-SODIUM

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Summary Soil fumigation with metam-sodium was simulated with computer models. The influence of rate of conversion of metam-sodium on the dose pattern for methyl isothiocyanate was checked. Equations and solutions are given for simultaneous vapour diffusion and leaching. In a series of simulations, the effect on dose pattern was traced of a period of rainfall soon after fumigation.

Résumé La désinfection du sol avec du métam-sodium a été simulée à l'aide des modèles mathématiques. L'effet de la vitesse de transformation du métam-sodium sur les doses de methyl isothiocyanate obtenues a été vérifié. Des équations et des solutions adequates décrivant le transport du composé par diffusion dans la phase gaseuze et par convection avec la phase liquide ont été données. Dans une série de calculs par ordinateur, l'effet de précipitation quelque jours après la fumigation a été simulé.

INTRODUCTION

In late years there is an increasing interest in metam-sodium, particularly for possible use in large-scale fumigation in arable farming against plant-parasitic nematodes. The desired characteristics of such applications are: effectiveness at low rates, low cost of the chemical, inexpensive techniques of application with the usual injection apparatus, only a short period with risk of after-effects and minimum consequences for the environment. In the past, results with metam-sodium were very variable and occasionally there were prolonged after-effects. This indicated that understanding of soil behaviour of the active compound was inadequate so that the selected soil conditions and application techniques were not optimum.

A detailed study on the behaviour of metam-sodium and the active conversion product methyl isothiocyanate was thus necessary. Data on the conversion rates and on the distribution of methyl isothiocyanate over the phases in soil were determined in the laboratory (Smelt and Leistra, 1973). Computation models were developed starting from those made for the behaviour of the isomers of 1,3-dichloropropene in soil (Leistra, 1971, 1972). Several modifications and extensions were made and assumptions had to be checked for the particular metam-sodium situation. The computation models were checked with detailed field experiments including measurement of soil physical characteristics and gas chromatography of methyl isothiocyanate concentrations (Leistra et al, 1973; Smelt et al, 1973). After testing, the combination of computation models and

basic data can be used to simulate soil fumigations. Suitable tools are then obtained to make quantitative predictions on effectiveness and after-effects under a wide variety of conditions (Leistra and Smelt, 1973; Leistra, 1973b).

On this occasion attention is concentrated on the lay-out of the computation models as adapted and extended for metam-sodium. Illustrations are given for some typical situations.

The standard computation model

By far the majority of farmers and contractors apply fumigant with horizontal-blade and plough injectors, so the initial distribution is in a plane. Injection depth is usually about 18 cm. The area density of methyl isothiocyanate formed (M_{init}, $\mu g\ cm^{-2}$) is calculated from the volume of metam-sodium solution per area (recommended rate 400 l./ha), the concentration of metam-sodium (usually 0.38 kg/l.), the fraction that is converted to methyl isothiocyanate (about 0.92), and the molecular weights of precursor and fumigant. On the basis of measured soil physical characteristics, the profile is divided into, for example, 5 layers: 3 above and 2 below the injection level. The soil profile is further divided into computation compartments of, for example, 2 cm thickness and concentrations are computed for points in the middle of these compartments. Compartment thickness, Δx, is selected in such a way that a suitable arrangement over the layers is obtained, with which transitions between layers coincide with computation points. However, another selection of compartment arrangement is possible (Leistra, 1972).

Values are assigned to temperature-dependent quantities like the diffusion coefficient of methyl isothiocyanate in air, D_a, the water/gas phase distribution ratio, $K_{w/g}$, the decomposition rate constant, k_r, and for each of the soil layers the value of the soil/gas phase distribution ratio, $K_{s/g}$ (Smelt and Leistra, 1973). Next the values of the soil physical characteristics for each layer are introduced including bulk density, ρ_b, volume fraction of the water phase, ε_w, volume fraction of the gas phase, ε_g, and the estimated quotient D_p/D_a, where D_p is the coefficient for diffusion of methyl isothiocyanate through the gas-filled pore system in soil. These estimates of D_p/D_a are obtained from literature data and from experience with dichloropropene (Leistra, 1972). A convenient quantity is the fumigant capacity factor, ϕ, for the concentration in the gas phase, C_g, defined as $\phi = \varepsilon_g + \varepsilon_w K_{w/g} + \rho_b K_{s/g}$. This capacity factor connects the concentration of fumigant in the whole soil, Q, with C_g: $Q = \phi\ C_g$.

The differential equation for fumigant vapour diffusion in heterogeneous soil systems (Leistra, 1971) is

$$\partial(\phi\ C_g)/\partial t = \partial(D_p\ \partial C_g/\partial x)/\partial x \qquad (1)$$

The change in soil conditions with position and time is expressed in the position and time dependence of the coefficients ϕ and D_p.

To get a suitable distribution of methyl isothiocyanate concentrations over the grid points at the start of the numerical computations, an analytical solution for diffusion from an instantaneous plane source (Leistra, 1971) can be used, provided the rate of formation is not restrictive. The time, t_{anal}, for which this distribution is calculated is confined to the period in which only the two layers around the injection depth are important for the diffusion and is usually about a day. Because decomposition rates of methyl isothiocyanate in soils are rather high, the amount, M_{anal}, for which this distribution is calculated is obtained with: $M_{anal} = M_{init} \exp(-k_r\ t_{anal})$. The comparatively low resistance to upward diffusion, for example after injection with a blade injector results in a fraction of 2/3 to 3/4 that diffuses upwards in the initial period.

The numerical solution for Equation 1 is obtained by conversion into an explicit difference equation. The first-order forward difference in time is combined with the second-order central difference in space (Smith, 1969). For heterogeneous systems the following difference equation is obtained:

$$C_g(i,j+1) = r_{dm}(i)\ C_g(i-1,j) + r_d(i)\ C_g(i,j) + r_{dp}(i)\ C_g(i+1,j) \qquad (2)$$

In this, the computation factors for diffusion alone, r_d, are:

$$r_{dm}(i) = D_{pm}(i)\ \Delta t/(\phi(i)\ (\Delta x)^2)$$

$$r_{dp}(i) = D_{pp}(i)\ \Delta t/(\phi(i)\ (\Delta x)^2)$$

$$r_d(i) = 1 - r_{dm}(i) - r_{dp}(i)$$

The subscript m refers to the range between x(i - 1) and x(i) while p as second subscript refers to the range between x(i) and x(i + 1).

The numerical computations start with the assessment of the time step, Δt, that can be used. This time step is limited because the highest value for the quotient $D_p(i)\ \Delta t/[(\phi(i)\ (\Delta x)^2]$ in a soil profile should be smaller than 0.5. The value of Δt is strongly influenced by the prevailing soil conditions and usually a value in the range 0.1-0.5 day can be taken. The computed diffusion and capacity characteristics for the layers are arranged over the compartments. At the transitions between layers, average capacity factors are taken. For each period with constant conditions, the computation factors have to be computed only once, so the computations in the dynamic section are as simple as possible. The concentration at the soil surface is set at zero, the methyl isothiocyanate being immediately removed by wind. The lower boundary is mostly taken at about 50 cm depth and the nature of the condition here depends on the local situation. The ultimate value of $C_g(i,j+1)$ at time $t(j+1)$ is obtained by taking account of decomposition during Δt by multiplication by the factor $\exp(-k_r\ \Delta t)$.

One of the objects of the computations is to obtain the cumulative concentration-time product [dose, ct(i,j)] for methyl isothiocyanate in the water phase at various depths. For this integration the trapezium rule can be used

$$ct(i,j+1) = ct(i,j) + 0.5\ \Delta t\ K_{w/g}\ [C_g(i,j) + C_g(i,j+1)] \qquad (3)$$

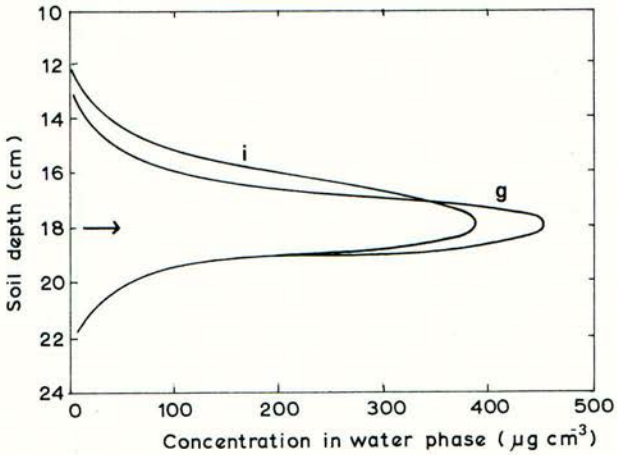
The computation of concentrations and doses are repeated until the time arranged for output, for example for comparison with measured concentrations of methyl isothiocyanate. If conditions are constant, computations can be continued with the same computation factors till concentrations are very low.

Changes in the conditions with time are introduced according to need and as far as available basic data permits. First the fumigant concentrations in whole soil are computed for each of the compartments. With changes in temperature, new values have to be set for a number of quantities: D_a, $K_{w/g}$, $K_{s/g}$ and k_r. If soil moisture or structure change, new values should be set for ε_w, ε_g, and the quotient D_p/D_a. New capacity factors, $\phi(i)^{new}$ are calculated and if necessary a new time step taken. The computation factors, r_d, per point have to be calculated again. The concentrations with which the new period starts are: $C(i,j)^{new} = Q(i,j)/\phi(i)^{new}$.

Effect of rate of formation of methyl isothiocyanate

Conversion of metam-sodium to methyl isothiocyanate takes some time and this may have some effect on diffusion patterns. In a modified computer model, the analytical starting solution was replaced by a section describing gradual formation of methyl isothiocyanate. A first-order rate equation was used: $dM_p/dt = -k_{rp}M_p$. The area density of precursor, M_p, is expressed in methyl isothiocyanate equivalent ($\mu g\ cm^{-2}$) and k_{rp} is the first-order conversion rate constant. It was assumed that the value of k_{rp} was 3.0 day^{-1}, which means that 95% of the metam-sodium applied was converted after one day, when the total amount of methyl isothiocyanate was formed. In the computations, the first day

Fig. 1 Computed distribution of methyl isothiocyanate in the profile one day after injection of metam-sodium into humic sand soil.
i = instantaneous plane source;
g = gradual formation.



was divided into ten time steps. The area density of methyl isothiocyanate, ΔM, formed per time step, Δt, is: $\Delta M = [1-\exp(k_{rp}\,\Delta t)]\,M_p$, where M_p is the area density of precursor at the beginning of the time step. At each time step ΔM was introduced in a computation compartment of 2 cm thickness around the injection depth.

The concentrations after one day computed for a moist humic sand soil (pF 2.0) at 15 °C are given in Fig. 1. As could be expected, concentrations near the injection depth are higher with the gradual conversion than with the instantaneous source. Continuation of the computations with the two starting distributions showed that the position of the dose line (e.g. Fig. 2) near the soil surface was hardly 0.2 cm lower after the gradual formation. Because the conversion rate in the example was lower than found for most soils and conditions (Smelt and Leistra, 1973), both approaches for the initial period may be used. The introduction of gradual conversion might be more relevant with precursors like dazomet, with a slow conversion under some conditions.

Fig. 1 shows that after one day distribution in the profile of methyl isothiocyanate is still restricted. This illustrates the slow diffusion in the humic sand soil if at the wet side of the favourable range.

Diffusion and leaching

With many soils, more than half the amount of methyl isothiocyanate is present in the water phase (Smelt and Leistra, 1973). With rainfall, leaching may thus be an important process. With some extension and modification, leaching may be built into the computation model described.

A differential equation that can be used for the description of simultaneous diffusion and leaching of fumigants in soil is

$$\partial Q/\partial t = \partial(D_p\ \partial C_g/\partial x)/\partial x + \partial(D_{disp}\ \partial C_w/\partial x)/\partial x - v\ \partial C_w/\partial x \qquad (4)$$

There is a convective transport caused by a water flux, v, resulting from the difference between precipitation and evaporation at the soil surface. An important associated spreading phenomenon is hydrodynamic dispersion represented by the coefficient D_{disp}. The equation is simplified by changing over to one dependent variable, C_g, and by combination of the coefficients for the spreading processes into one coefficient D_{spr}. With the relations $Q = \emptyset C_g$ and $C_w = K_w/g C_g$, the equation becomes

$$\partial(\emptyset C_g)/\partial t = \partial(D_{spr} \partial C_g/\partial x)/\partial x - v K_{w/g} \partial C_g/\partial x \qquad (5)$$

To approach the value for the coefficient of hydrodynamic dispersion for the fumigant in the water phase, the following relation can be used: $D_{disp} = DISP\ v$. The factor DISP is the dispersion distance which assumes a value of, for example, 1 cm for soils under leaching conditions (Frissel et al, 1970). The expression of the spreading coefficient in the considered system is: $D_{spr} = D_p + DISP\ v\ K_w/g$.

Conversion of Equation 5 into an explicit difference equation gives

$$C_g(i,j+1) = r_{dlm}(i)\ C_g(i-1,j) + r_{dl}(i)\ C_g(i,j) + r_{dlp}(i)\ C_g(i+1,j) \qquad (6)$$

The computation factors for diffusion plus leaching, r_{dl}, are

$$r_{dlm}(i) = D_{sprm}(i)\ \Delta t/[\emptyset(i)\ (\Delta x)^2] + v\ K_{w/g}\ \Delta t/(\emptyset(i)\ \Delta x)$$

$$r_{dlp}(i) = D_{sprp}(i)\ \Delta t/[\emptyset(i)\ (\Delta x)^2]$$

$$r_{dl}(i) = 1 - r_{dlm}(i) - r_{dlp}(i)$$

With compartments in computations on convective transport, a pseudo-spreading is introduced. The value of the pseudo-spreading coefficient, D_{spr}^{pseudo}, with respect to the concentration in the water phase can be approached with: $D_{spr}^{pseudo} = v\ \Delta x/2$ (Goudriaan, 1973; Leistra, 1973a). In the computations, only a net spreading coefficient has to be added: $D_{spr}^{net} = D_p + DISP\ v\ K_w/g - v\ K_{w/g}\ \Delta x/2$.

Effect of rainfall on dose pattern

One of the possible applications of the computation model involving vapour diffusion and leaching is to simulate the effect of rainfall on the dose pattern. In a series of computations, injection of 400 l. of metam-sodium solution (0.38 kg/l.) was simulated for a humic sandy soil. Soil moisture was on the wet side (pF about 2.0) of the favourable range. Details are given in Table 1. In the standard case, conditions were kept constant throughout fumigation. The computed dose pattern for methyl isothiocyanate in the water phase is given in Fig. 2. At 50 $\mu g\ cm^{-3}$ day, nematode mortality is presumably high.

In the next run, injection under the same conditions was simulated but there was a total rainfall surplus of 20 mm on the second and third day. The situation was simulated by taking in this period a constant percolation rate and a higher moisture content in the layers 0-28 cm (pF about 1.5). After the third day, moisture content in the layers 0-28 cm was lowered to the original values and that of the layer 28-60 cm was set higher for three days. From the sixth day onwards, moisture condition in the entire profile corresponded again with pF 2.0. The two most significant phenomena, percolation and temporary increase in moisture content, were thus allowed for, though in a simplified way. In a third run, rainfall on the fifth and sixth day after injection was simulated in the same way.

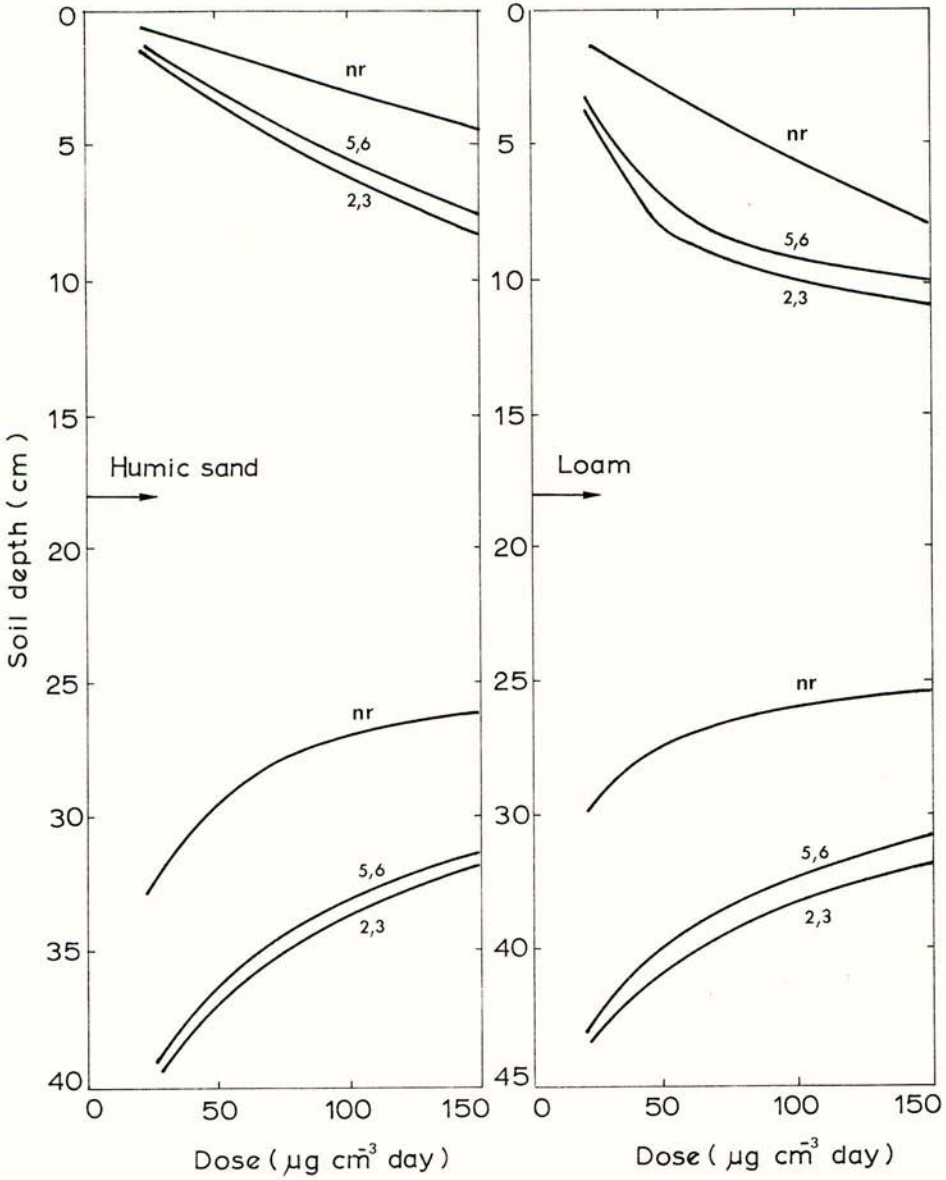
In Fig. 2, the dose patterns for the different situations are given. As a result of the rainfall, the dose patterns are clearly displaced downwards. The displacement shows most clearly at the lower end and amounts to about 7 cm for the most important range of concentration. At the top of the profile, displacement is somewhat less because the soil surface acts as a sort of turning-point

Table 1

Soil physical properties with diffusion and distribution characteristics for methyl isothiocyanate

Soil type, temperature, pF	Decomp. rate constant (day^{-1})	Layer (cm)	Organic matter content ($g\ g^{-1}$)	Bulk density ($g\ cm^{-3}$)	Water phase ($cm^3 cm^{-3}$)	Gas phase ($cm^3 cm^{-3}$)	D_p/D_a approx. range	D_p/D_a estim. value	K_s/g	Capacity factor ($cm^3 cm^{-3}$)
Humic sand,	0.06	0- 8	0.050	1.25	0.31	0.20	0.02-0.06	0.040	42	120
15°C,		8-18	0.050	1.30	0.33	0.16	0.01-0.04	0.025	42	126
pF 2.0		18-28	0.040	1.45	0.33	0.10	0.01	0.007	34	121
		28-60	0.020	1.55	0.26	0.14	<0.03	0.015	17	83
Humic sand,	0.06	0- 8	0.050	1.25	0.36	0.15	0.01-0.03	0.020	42	130
15°C,		8-18	0.050	1.30	0.38	0.11	0.01	0.010	42	137
pF 1.5		18-28	0.040	1.45	0.38	0.06	<0.01	0.004	34	131
		28-60	0.020	1.55	0.31	0.09	0.01	0.008	17	93
Loam,	0.11	0- 8	0.023	1.10	0.30	0.25	0.04-0.10	0.070	9.5	76
15°C,		8-18	0.023	1.25	0.34	0.14	<0.03	0.020	9.5	85
pF 2.0		18-28	0.021	1.35	0.35	0.09	0.01	0.007	8.7	87
		28-60	0.021	1.30	0.34	0.12	<0.02	0.015	8.7	85
Loam,	0.11	0- 8	0.023	1.10	0.33	0.21	0.02-0.07	0.045	9.5	82
15°C,		8-18	0.023	1.25	0.38	0.10	0.01	0.015	9.5	94
pF 1.5		18-28	0.021	1.35	0.39	0.05	<0.01	0.005	8.7	96
		28-60	0.021	1.30	0.38	0.08	0.01	0.010	8.7	93

Fig. 2 Dose pattern computed for methyl isothiocyanate in the water phase after injection of metam-sodium into humic sand soil and loam soil, respectively.
nr = no rainfall; 2,3 = rainfall on second and third day;
5,6 = rainfall on fifth and sixth day after injection.



for the dose pattern. The spreading of the concentration pattern after one day is rather limited, so leaching in the earlier period caused a large proportion of the fumigant to be leached into the denser layer below the level of injection.

The same series of simulations was carried out for a loam soil, with a lower adsorption and a higher decomposition rate (Smelt and Leistra, 1973). Details of soil properties and interaction characteristics are given in Table 1. The computed dose patterns are represented in Fig. 2. In the standard case, the top centimetres of the profile were badly disinfected and activity at 30 cm depth was very poor. Fumigation under somewhat drier conditions would have resulted in a much better dose pattern (Leistra and Smelt, 1973). With this soil type too, rainfall caused a substantial displacement of the dose pattern. The effect is even stronger with the loam soil than with the humic sand soil. As a result of the lower adsorption and the higher decomposition rate, the dose pattern at the top is even more unfavourable. The greater extent of leaching is partly offset at some decimetres depth by the greater part of the fumigant being decomposed near the injection depth.

GENERAL DISCUSSION

A further step has been taken to describe fumigant behaviour in soil quantitatively with the development of computation models. Although a number of simplifying assumptions are made, the importance of various aspects of methyl isothiocyanate behaviour in soil can be traced. Differences in the rate of conversion of metam-sodium to methyl isothiocyanate in the practical range are not very important.

Under rather wet conditions, methyl isothiocyanate is easily leached over several centimetres and this will reduce its effectiveness. In practice, there are several other complicating factors. Under rather wet conditions, horizontal distribution of fumigant will be very irregular, so its effectiveness is less than the average dose line would indicate. Further, low concentrations presumably contribute little to the effective dose so that the flatter dose lines are even more unfavourable. A limited amount of rain after injection into a drier soil will be less unfavourable for the effect in the top layer, particularly if methyl isothiocyanate is spread to a considerable extent. A main effect is then the increase in moisture content in the top layer accompanied by a decrease in diffusion rate (Smelt et al, 1973).

The same computation model can be used to estimate leaching of fumigant residues in the winter season. Residues of methyl isothiocyanate can be leached over several decimetres and contamination of ground and surface water is thus possible. Only with application under somewhat drier conditions in the period up to early autumn is the risk of leaching out of the profile very small. The rather low diffusion rate and the high decomposition rate then favour elimination in the top of the profile.

References

FRISSEL, M.J., POELSTRA, P. and REINIGER, P. (1970) Chromatographic transport through soils. III. A simulation model for the evaluation of the apparent diffusion coefficient in undisturbed soils with tritiated water. Plant and Soil, 33, 161-176.

GOUDRIAAN, J. (1973) Dispersion in simulation models of population growth and salt movement in the soil. Netherlands Journal of Agricultural Science, 21, (in press).

LEISTRA, M. (1971) Diffusion of 1,3-dichloropropene from a plane source in soil. Pesticide Science, 2, 75-79.

LEISTRA, M. (1972) Diffusion and adsorption of the nematicide 1,3-dichloropropene in soil. Pudoc, Wageningen, Netherlands. Agricultural Research Reports, 769, 105 pp.

LEISTRA, M. (1973a) Computation models for the transport of pesticides in soil. Residue Reviews, 49 (in press).

LEISTRA, M. (1973b) Predictions on effectivity and after-effects by simulating soil fumigations with metam-sodium. (submitted).

LEISTRA, M. and SMELT, J.H. (1973) Optimum soil conditions for fumigation with metam-sodium. In: Proceedings of the International Symposium on Soil Disinfestation, Heverlee-Louvain, Belgium, September 24-28, 1973. (submitted).

LEISTRA, M., SMELT, J.H. and NOLLEN, H.M. (1973) Concentration-time relation for methyl isothiocyanate in soil after injection of metam-sodium. (submitted).

SMELT, J.H. and LEISTRA, M. (1973) Conversion of metam-sodium to methyl isothiocyanate and basic data for the behaviour of methyl isothiocyanate in soil. (submitted).

SMELT, J.H., LEISTRA, M., SPRONG, M.C. and NOLLEN, H.M. (1973) Soil fumigation with dichloropropene and metam-sodium: effect of soil cultivations on dose pattern. (submitted).

SMITH, G.D. (1969) Numerical solution of partial differential equations. Oxford University Press, London.

NOTES

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FUNGICIDAL CONTROL OF WINTER WHEAT FOLIAGE DISEASES IN FRANCE

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Summary Trials started in 1970 showed that it is possible to control foliage diseases on winter wheat with fungicides such as benomyl, benomyl + mancozeb or methylthiophanate + maneb. The best results were obtained after treatments during stem extension and flowering or, more often, at both stages. The experimentation in progress will allow us to measure the risk for wheat in each region in order to know precisely the conditions in which fungicide application will be profitable.

Résumé L'expérimentation réalisée sur le blé tendre d'hiver depuis 1970 a mis en évidence la possibilité de limiter les dégâts provoqués sur cette culture par les maladies fongiques grâce à l'utilisation de produits fongicides tels que le bénomyl et les mélanges mancozèbe + bénomyl ou manèbe + méthylthiophanate. Les résultats les plus intéressants ont été observés à la suite d'applications réalisées, soit courant montaison soit en fin épiaison, soit, le plus souvent, successivement à ces deux époques. L'expérimentation en cours doit permettre d'étudier localement les risques encourus par la céréale, afin de préciser les conditions dans lesquelles il est opportun d'intervenir compte tenu du coût des traitements.

INTRODUCTION

Fungicidal control of cereal foliage diseases has been possible for a long time only with seed-dressing. This seems to be an excellent treatment against seedborne diseases such as damping-off, bunt and loose smut but it does not protect wheat from diseases which infect after seed germination. New systemic fungicides which can be sprayed onto the foliage appeared on the market in 1972 and made it possible for farmers to protect the plant during the whole growing season. These new chemicals can also be used as seed-dressings, but the yield advantage has appeared to be best following spray application.

The aim of the trials done by the "Institut Technique des Céréales et des Fourrages" since 1970 is : (a) to measure the importance of damage caused by foliage diseases on yield and on grain quality, (b) to compare the efficiency of treatments performed at different growth stages of wheat.

Treatment was carried out against many fungus diseases and not against one in particular. Many different diseases were generally found in the same plot and it was difficult to foresee which would be dominant in each case.

METHODS AND MATERIALS

From 1970 to 1972, 103 trials have been carried out on winter wheat with the following chemicals (doses kg/ha a.i.) benomyl (0.3) ; benomyl (0.3) + mancozeb (2) ; methylthiophanate (1) + maneb (2) ; mancozeb (2). The materials were applied at the following stages (Feekes-Large scale) : erect (stage 5) ; stem extension, last leaf just visible (stage 8) ; flowering (stage 10.5) ; repeated applications in same plots at stages 5 and 8 ; 5 and 10.5 ; 8 and 10.5.

The main varieties used were : Capitole (27 trials) ; Hardi (13 trials) ; Champlein (12 trials) ; Joss (7 trials) ; 26-10 (3 trials). The trials were situated in the main region of wheat production in France.

The diseases observed were :
- Foot rot : eyespot (Cercosporella herpotricoides) ; brown foot rot (Fusarium spp.)
- On leaves and heads : leaf spot (Septoria spp.) ; ear blight (Fusarium spp.) ; mildew (Erysiphe graminis) ; yellow and brown rust (Puccinia spp.).

RESULTS

1. Effect of the treatments on yield and quality of wheat

Table 1

Overall yield increases obtained after two fungicide treatments growth stages 8 and 10.5 with benomyl + mancozeb

Year	No. of trials	Yield of control plot t/ha	Yield increase t/ha	% increase
1970	21	4.870	0.350	7
1971	42	4.800	0.510	11
1972	40	5.750	0.580	10

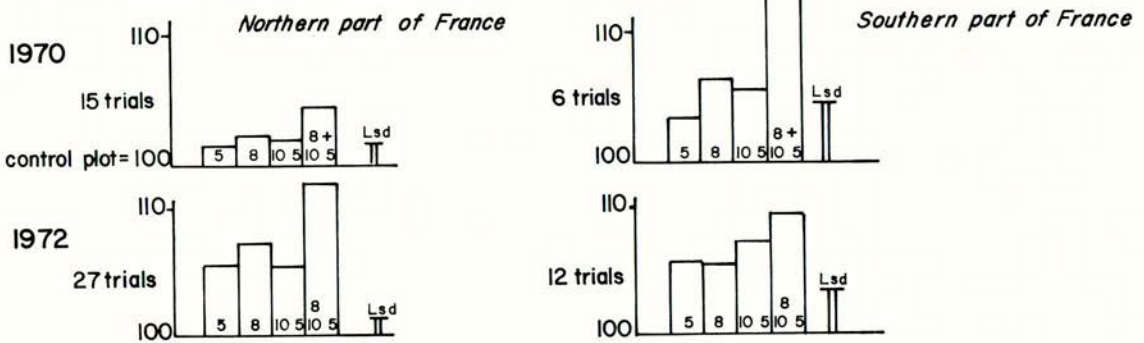
The overall yield increases shown in Table 1 following treatment with benomyl and mancozeb at growth stages 8 and 10.5, were due mainly to the increase of 1000 grains weight and, sometimes, in the number of heads. They show the favourable effect of fungicides on migration of dry matter in the host and give an idea of the minimum damage caused by the diseases.

These trials also made it possible to compare different kinds of fungicides. Benomyl used alone at flowering (stage 10.5) gave a yield increase of 180 kg/ha whereas the mixture benomyl + mancozeb gave 320

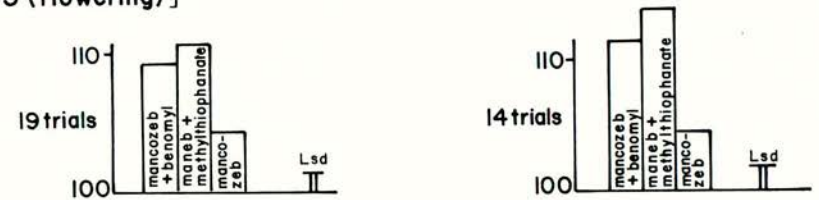
Fig. I

Winter wheat : effect on yield of different foliage treatments performed in 1970, 1971 and 1972 (% of control plots)

1) Comparison between benomyl + mancozeb applications at different wheat growth stages in 1970 and 1972 (Feekes-Large scale)



2) Comparison between chemicals [1971 applications at stages 8 (stem extension) and 10-5 (flowering)]



kg/ha increase (average of 40 trials in 1972). Mancozeb usually gave a lower yield gain than that obtained with systemic fungicides (Fig. 1-2°). The activity of systemic fungicides increases when they are mixed with chemicals such as mancozeb.

Comparison between treatments given at different stages of plant growth is shown in Fig. 1-1°. Two periods in particular seem to be favourable for treatment : (a) during stem extension, from the beginning of stem extension till the boot stage ; this treatment is effective against foot-rot and diseases of the first leaves ; (b) at flowering ; this treatment is successful against foliage and head diseases. Plant protection is well ensured with repeated applications at these two growth stages. Nevertheless it is possible to obtain partial protection using one of these treatments.

2. Profitability of treatments

An example of the cost of treatments is shown in Table 2

Table 2

Costs of fungicide treatments

		1 treatment	2 treatments
Loss due to wheelings		25 F	25 F
Cost of chemicals		109 F	218 F
Cost of treatment		20 F	40 F
Total cost	Francs	154 F	283 F
	Kg (100 kg = 50 F)	310 kg	570 kg

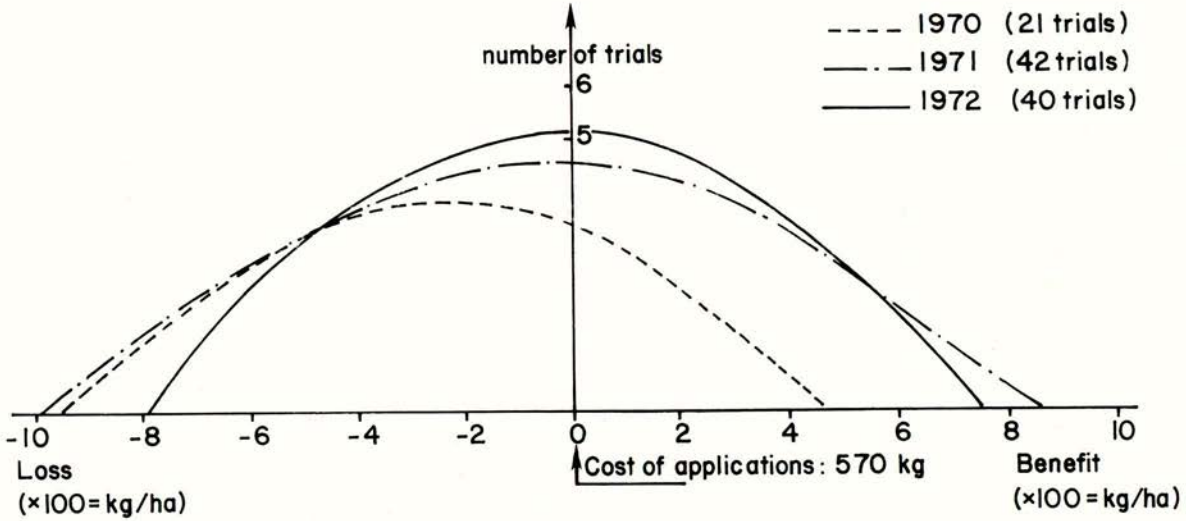
Fig. 2 shows that the yield increase may be important but that it is also variable. For the last three years the yield increase in many cases has not repaid the cost of treatment because of the present high cost of systemic fungicides. It is therefore necessary to specify the risk before deciding on the most suitable treatment. At present it is not possible to prove any precise correlation between yield loss and disease level. The risk can only be determined through investigation of the different factors which are favourable to the diseases.

a) Influence of site and year

Fig. 1 shows that the yield increase after treatment differs according to site and climate. The benefit of treatment did not differ from year to year in the south part of France whereas, in the north, it was

Fig. 2

Profitability of treatments on winter wheat (benomyl + mancozeb) applied during stem extension (8) and at flowering (10-5)



ineffective in 1970 but beneficial in 1971 and 1972. The losses caused by disease in 1971 may be explained by the very dry and warm period in the beginning of July. In contrast, in 1972, the early attack of eyespot and brown foot-rot was favoured by a mild winter which seemed to be of great importance.

b) Influence of soil type

Table 3 shows that in 1971 and particularly in 1972, the yield increase was higher on light than on heavy soils.

Table 3

Yield increase on heavy and light soils following benomyl + mancozeb treatment at growth stages 8 and 10.5

Yield increase (kg/ha)

Clay content	1971	1972
> 12.5 %	470 (16 trials)	530 (16 trials)
< 12.5 %	530 (16 trials)	660 (18 trials)

c) Influence of previous crop

In the same trials the yield increase was higher when the previous crop was wheat or maize than when it was not a cereal, eg. potatoes, beet or rape (Table 4).

Table 4

Influence of previous crop on yield increase following benomyl + mancozeb treatment at growth stages 8 and 10.5

Yield increase (kg/ha)

Previous crop	1971	1972
Potatoes, beet, rape	450 (15 trials)	470 (11 trials)
Wheat or maize	580 (17 trials)	610 (26 trials)

d) Influence of varieties

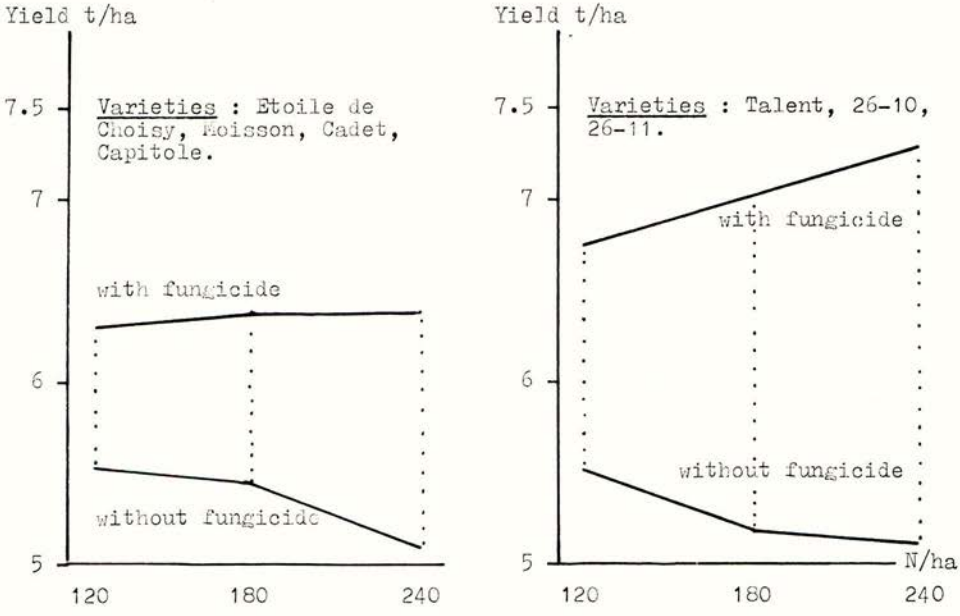
Trials on winter wheat in 1972 showed that certain varieties such as Joss, Talent, 26-10 and 26-11 gave a yield increase of 950 kg/ha to 1140 kg/ha, whereas varieties such as Hardi, Capitole and Champlein gave an increase of only 730 kg/ha to 860 kg/ha.

e) Influence of nitrogen

The incidence of fungicide treatments was studied in 1972 on 7 varieties with 3 different rates of nitrogen : 120, 180 and 240 units (Fig. 3). In these trials, the yield of each variety declined as the rate of nitrogen increased. In the presence of the fungicide, this phenomenon was not observed and it was possible to distinguish two groups of varieties : those which did not gain in yield with an increasing nitrogen rate (Etoile de Choisy, Moisson, Capitole, Cadet) and those which did increase in yield with an increasing nitrogen rate (Talent, 26-10 and 26-11).

Fig. 3

Influence of fungicide treatments with variable nitrogen rate on different varieties of winter wheat (average of 4 trials situated in the southern part of France)



DISCUSSION

Fungicide treatments on wheat foliage promote yield increases and also have a favourable influence on grain quality. However, in the current economical situation such treatment cannot be advised as a matter of course and it is necessary to study the different factors which influence the gravity of diseases before deciding on treatment. The conditions most likely to cause a yield decrease seem to be the following : a dry climate at the end of the vegetative period (southern part of France and on light soils in the northern part) - poorly drained soils - mild winter (favourable for foot-rot spread) - when the previous crop is a cereal - when the variety is sensitive to the diseases - when nitrogen supply is at a high level - when minimum tillage is practised, leaving crop debris on the surface.

According to the risk, the grower has to choose between the four following possibilities : (a) no treatment ; (b) one treatment during stem extension which is mainly effective against foot-rot diseases ; (c) one treatment at flowering which protects the late leaves and the heads ; (d) two treatments, during stem extension and at flowering. These two treatments give the most regular and largest yield increases but, even when the risk is at its highest, one treatment only is more economical in many situations.

Fungicide treatment of wheat foliage should not be systematic but there are still many problems concerning specific recommendations in each situation (for example, the behaviour of each variety, the economical dosage of chemicals). Another problem is deciding whether to treat or not. At present it is difficult to foresee the benefit of treatment in each single case but the experimentation now in progress should allow us to draw up a map of the losses caused by disease.

If it does become possible to protect cereals against disease by the use of fungicides, this could raise many questions concerning crop rotation, cultivations, choice of varieties, sowing dates and fertilizer rates. Indeed, following the use of fungicides, it is not certain that current agricultural techniques will remain the most satisfactory, both from a technical as well as an economic point of view.

Reference

I.T.C.F. (1973) Proceedings of the Conference "La lutte contre les maladies des Céréales".

Proceedings 7th British Insecticide and Fungicide Conference (1973)

THE EFFECT OF TRIDEMORPH ALONE AND IN MIXTURES WITH OTHER FUNGICIDES ON DISEASES OTHER THAN ERYSIPHE GRAMINIS

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Summary Tridemorph, in addition to controlling cereal powdery mildew, Erysiphe graminis, has a marked effect against certain other cereal pathogens. Tridemorph applied either alone or in mixture with metiram, has given good control of Puccinia striiformis (yellow rust) on wheat and barley. Treatment of wheat with a mixture of tridemorph and metiram reduced levels of Septoria nodorum (glume blotch).

Tridemorph applied to barley reduced levels of Rhyncosporium secalis (leaf blotch), while an improved effect was obtained with a mixture of tridemorph and carbendazim.

Résumé Les essais au champ ont montré qu'en plus du contrôle de l'oidium des céreales, Erysiphe graminis, le tridemorphe avait un effet marqué contre certains autres agents pathogènes des céreales. Le tridemorphe appliqué soit seul, soit en mélange avec le metiram, a donné un bon contrôle de Puccinia striiformis (rouille jaune) sur blè et sur orge. Le traitement du blè avec un mélange de tridemorphe et de metiram diminuait le taux de Septoria nodorum (septoriose sur glume).

Le tridemorphe appliqué sur orge diminuait le taux de Rhyncosporium secalis (rhyncosporiose sur feuille) et par ailleurs un meilleur effet a été obtenu avec le mélange de tridemorphe et de carbendazim.

INTRODUCTION

Since the detection of the fungicidal activity of tridemorph and other N-alkyl substituted 2,6-dimethylmorpholines, numerous experiments have been reported showing the effectiveness of tridemorph against cereal powdery mildew. Jung and Bedford (1971) and Evans and Hawkins (1971) have described work carried out in England with tridemorph against cereal powdery mildew.

During the course of some of these investigations, it was observed that tridemorph had an effect against diseases other than Erysiphe graminis i.e. Puccinia striiformis, Puccinia recondita, Septoria nodorum, Rhyncosporium secalis.

In 1972 and 1973 some of these effects have been investigated further.

METHODS AND MATERIALS

In all trials a randomised block design with four replicates was used. Plots measured 4m x 12.5m or 15m. Treatments were applied with a Van der Weij knapsack sprayer fitted with cone nozzles. Tridemorph was applied at 0.52 kg a.i./ha. Carbendazim was applied at the rates stated. Metiram (Zineb-polyethylene thiuram-disulphide complex) was applied at 1.6 kg a.i./ha. All sprays were applied in 250 l/ha water.

Formulations

Tridemorph - 75% a.i. w/v emulsifiable concentrate
Carbendazim - 50% wettable powder (BAS 3460F)
Metiram - 80% wettable powder

Disease assessments were made using the appropriate key from the Guide for the Assessment of Cereal Diseases devised by the Plant Pathology Laboratories, Harpenden. Cereal growth stages are expressed using the Feekes-Large scale (Large, 1954).

Trials were harvested using a "Hege 125" small plot combine or Claas Compact 25 combine.

RESULTS

Wheat

Puccinia striiformis (yellow rust)

Results from a trial carried out in 1971 indicated that tridemorph had a marked effect against yellow rust (P. striiformis). The figures for disease control and the effect on grain weight are presented. (Table 1)

Table 1

The effect of tridemorph applied to winter wheat (cv. Joss Cambier) on levels of P. striiformis and grain weight

Treatment	% disease level on flag leaf at GS 10.1	at GS 10.5	Thousand grain wt. (g)
Untreated	3.60	20.4	45.5
Tridemorph	0.42	5.8	47.4

Tridemorph was applied at GS 9 when infection was present on the lower leaves, but not on the flag leaf.

Observations indicated that the effect of tridemorph was mainly curative; pustules appeared to dry up within 5 days of application. In trials carried out in 1972 the effect of tridemorph was again recorded and yields following tridemorph application were significantly higher than in untreated plots (Table 2).

Table 2

The effect of tridemorph applied to winter wheat (cv. Joss Cambier) on levels of P. striiformis and yield at two sites (A and B)

Treatment	% disease level on flag leaf at GS 10.5		Yield kg/ha	
	A	B	A	B
Untreated	12.5	68	4167	3534
Tridemorph	2.5	47	5071	4067
Standard error ±			96	408

In Trial A, application was made at G.S. 10 where the flag leaves were free of yellow rust, and in Trial B, application was at G.S. 10 with 10-15% infection on the flag leaf.

Septoria nodorum (glume blotch) and Puccinia recondita (brown rust)

In further trials carried out in 1973 with a mixture of tridemorph and metiram, effects against S. nodorum (glume blotch) and P. recondita (brown rust) were observed. (Tables 3 and 4)

Table 3

The effect of an application of tridemorph plus metiram on foliar and ear diseases and yield of winter wheat (cv. Maris Nimrod) at GS. 9 (25/5/73)

Assessment (% infection at GS. 10.5 - 26/6/73)		Untreated	Treated	s.e.±
Septoria nodorum	- ear	22.5	17.5	
Septoria nodorum	- flag leaf	21.0	11.0	
Puccinia striiformis	- flag leaf	5.0	1.75	
Puccinia recondita	- flag leaf	2.5	1.0	
Green tissue	- flag leaf	26.0	40.0	
Senescence *		45.5	46.25	
Yield (kg/ha)		5034	5687	209

* Senescence which could not be directly related to any of the diseases present.

Table 4

The effect of an application of tridemorph plus metiram to winter wheat (cv. Maris Ranger) at GS 10.3 (8/6/73)

Assessment (% infection at GS 11.1 - 12/7/73	Untreated	Treated
Erysiphe graminis - ear	10.6	2.5
Erysiphe graminis - flag leaf	1.25	0.5
Erysiphe graminis - 2nd leaf	16.25	5.0
Septoria nodorum - flag leaf	4.37	1.5
Septoria nodorum - 2nd leaf	46.25	18.75

Barley

Puccinia striiformis (yellow rust)

In 1973, severe infections of yellow rust were recorded in some winter barley crops. Applications of tridemorph plus metiram gave marked reductions in disease levels. (Table 5)

Table 5

The effect of tridemorph plus metiram applied at two growth stages to winter barley (cv. Astrix) on levels of P. striiformis

Date of assessment	15/5		16/6		19/6	
GS at assessment	8		10.5		11.1	
GS and date at application	% infection on leaf *		% infection on leaf		% green tissue on leaf	
	3	4	1	2	1	2
Untreated	2.0	14.0	14.4	33.7	20	20
GS 6 - 28/4	2.0	3.5	5.0	20.8	25	30
GS 8 - 14/5	-	-	4.7	11.8	40	55
GS 6 + GS 8	-	-	2.0	7.5	50	60

* Leaves 1 and 2 free of infection at GS 8

Treated plots showed marked decreases in disease levels for a considerable period after application.

Rhyncosporium secalis (leaf blotch)

In 1973 trials were laid down to evaluate the effect of tridemorph, alone and in mixture with carbendazim, on infections of R. secalis on spring barley. One trial was laid down on the cultivar Deba Abed (seed treated with ethirimol)

and the second on plots of Maris Mink drilled at two different dates within the same field. In both trials no mildew was recorded.

Table 6

The effect of tridemorph, alone and in mixture with carbendazim, applied to spring barley (cv. Deba Abed) at GS 8-9 on levels of R. secalis and yield

Treatment	% disease level at GS 10.5 Leaf 1	Leaf 2	Yield kg/ha	Relative yield
Untreated	42.0	86.6	3565	100
Tridemorph	18.1	65.7	3816	107
Tridemorph + carbendazim 0.25 kg	5.7	45.0	4318	121
Tridemorph + carbendazim 0.5 kg	3.5	27.4	4268	120
Standard error ±			184	

Disease levels at GS 8-9: Flag leaf - trace, leaf 2 - 1%, leaf 3 - 7.5%

Tridemorph alone gave a marked reduction in disease level, while mixtures with carbendazim gave a greater degree of control.

Table 7

The effect of tridemorph, alone and in mixture with carbendazim, applied to spring barley (cv. Maris Mink) sown at two dates, on levels of R. secalis and yield

Treatment	Crop drilled 12/3: % disease level (at GS 10.5) on leaf 1	2	3	Yield kg/ha	Crop drilled 28/3: % disease level (at GS 10) on leaf 1	2	3	Yield kg/ha
Untreated	21.2	40.1	44.5	4872	7.6	16.3	28.5	4961
(E) Tridemorph	21.6	36.6	39.6	5014	8.9	12.2	19.7	5468
(E) Tridemorph + carbendazim	4.3	14.8	15.8	5486	0.7	2.4	4.6	5468
(L) Tridemorph	11.5	20.6	23.1	5312	5.2	8.8	14.0	5260
(L) Tridemorph + carbendazim	3.4	9.2	16.8	5618	1.9	3.5	12.3	5590
Standard error ±				299				206

(E) = application at GS 4-5 on late drilled crop and GS 5-6 on early drilled crop (24th May)

(L) = application at GS 8-9 on late drilled crop and GS 9-10 on early drilled crop (14th June)

Observations on grain surface microflora (Hill, personal communication) have shown that populations of Alternaria spp., Penicillium spp. and Aspergillus spp. were eliminated, and populations of Cladosporium spp. and Sporobolomyces reduced, by foliar applications of tridemorph.

DISCUSSION

The results presented indicate that tridemorph has a marked curative effect against P. striiformis on wheat, resulting in significant yield increases (Table 2). Further trials with this mixture in 1973 again showed an effect against P. striiformis and in addition effects against S. nodorum. These results (Table 3) indicated that treatment gave an increase in yield, but it is not possible to relate this effect to individual disease levels. It would appear that an application of tridemorph plus metiram, after flag leaf emergence, markedly reduces levels of the important late season diseases of wheat.

The results presented on barley show that tridemorph plus metiram also has a marked effect against P. striiformis on this crop (Table 5). In addition, applications of tridemorph to barley gave considerable reductions in levels of R. secalis. The extent of this effect varied with time of application and levels of disease present in the crop, but in each instance yield was increased. The degree of the control and effect on yield was increased when a mixture of tridemorph and carbendazim was used (Tables 6 and 7). Foliar applications of tridemorph have also been shown to reduce levels of certain fungi in the grain microflora.

Previous work (Jung and Bedford, 1971 and Evans and Hawkins, 1971) has shown that tridemorph is an effective treatment for control of powdery mildew (E.graminis) on barley. Although mildew is the most widespread disease of barley (King, 1972) other diseases can also cause considerable yield loss (James, Jenkins and Jemmett, 1968). The effects of tridemorph on certain of these diseases, in particular P. striiformis and R. secalis may represent an added benefit from the use of this material for the routine treatment of mildew in barley.

Acknowledgements

The authors wish to thank Mr. R. A. Hill of Rothamsted Experimental Station, for permission to quote his observations on the effect of tridemorph on grain microflora. Thanks are also due to their colleagues of BASF United Kingdom Ltd., and to the growers who kindly co-operated with the trials.

References

EVANS, E.J. and HAWKINS, J.H. (1971) Timing of Fungicidal Sprays for Control of Mildew in Spring Barley. Proceedings 6th British Insecticide Conference 1971, 33-41.

JUNG, K.U. and BEDFORD, J.L. (1971) The evaluation of application time of tridemorph for the control of Mildew in Barley and Oats in the United Kingdom. Proceedings 6th British Insecticide Conference 1971, 75-81

KING, J.E. (1972) Surveys of foliar diseases of spring barley in England and Wales 1967-70. Plant Pathology, 21, 23-35

LARGE, E.C. (1954) Growth Stages in Cereals. Plant Pathology, 3, 128-9

JAMES, W.C. JENKINS, J.E.E. and JEMMETT, J.L. (1968) The relationship between leaf blotch caused by Rhyncosporium secalis and losses in grain yield of spring barley. Ann. Appl. Biol. 62, 273-288.

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THE TIMING OF APPLICATION OF BENODANIL (BAS 3170F) FOR THE CONTROL OF CEREAL RUST DISEASES

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Summary BAS 3170F was tested against Puccinia striiformis on wheat and barley and against Puccinia hordei on barley. Good control of both diseases was obtained. For optimum yield benefit, however, timing of application was found to be important. For the control of P. striiformis in wheat and barley greatest yield benefit resulted when the flag leaf was protected from infection. Under conditions of early, continuous infection, two applications may be required.

For the control of P. hordei, best yield responses were obtained with applications made at or before the onset of the rapid phase of rust development. Under conditions of early and prolonged infections again two sprays may be necessary for maximum yield benefit.

Resume BAS 3170F a été essayé contre Puccinia striiformis sur bleet sur orge ainsi que contre Puccinia hordei sur orge. Un bon contrôle des deux maladies a été obtenu. Cependant pour obtenir un gain de rendement optimum, il est apparu que l'époque d'application était importante. Sur blè et sur orge dans la lutte contre Puccinia striiformis les plus gros gains de rendement ont été obtenus quand l'etendart était protégé de l'infestation. Deux applications peuvent être nécessaires dans des conditions d'infestation précoce et continue. Pour la lutte contre Puccinia hordei les meilleures responses quant au rendement ont été obtenues avec des applications effectuées au moment où avant le debut de la phase rapide de développement de la rouille. De même, dans des conditions d'infestation précoce et prolongée, deux pulverisations peuvent être nécessaires pour obtenir un gain maximum de rendement.

INTRODUCTION

Benodanil - the proposed common chemical name for 2 iodo-benzanilide - is a related chemical to mebenil (Pommer and Kradel, 1969). Preliminary results showed benodanil to be more active than mebenil (Pommer and Zwick, 1971), and development of mebenil was discontinued. During the period 1971-73, benodanil has been evaluated for control of Puccinia striiformis Westend on wheat and barley, and Puccinia hordei Otth. on barley. At an early stage in the development of benodanil, it became apparent that timing of application is of prime importance. The trials presented are intended to show the optimal timing of a single application of benodanil for maximum yield benefit.

METHOD AND MATERIALS

Benodanil was used as a wettable powder formulation containing 50% a.i. and is referred to by the code number BAS 3170F.

In the trials for control of Puccinia striiformis on wheat and Puccinia hordei on barley, BAS 3170F was applied at a rate of 1.5 kg/ha a.i. at various stages of growth of the crop. The modified Feekes Scale (Large, 1954) was used to assess the growth stages. In the trials for control of P. striiformis on barley, BAS 3170F was applied at 1.0 kg/ha a.i. Volume of application was 250 l/ha in all trials.

In the trial described in Table 1, the two varieties were included as split plots within a randomised block design. All trials were replicated four times. Plot size was 12.5 or 15 metres x 4 metres.

Leaf disease assessments were made by assessing percentage of leaf area infected, using a logarithmic scoring system (1-9) in the 1972 trials, the figures being converted to percentages for presentation. In the 1973 trial, percent infection of leaf area was scored directly. Where ear infection occurred, it was measured by counting the number of infected glumes on ten ears per plot, and expressing this figure as a percentage.

Trials were harvested using a Hege 125 combine or 'Claas Compact' combine harvester.

RESULTS

Results are presented on the effect of timing of application on disease level and yield.

Puccinia striiformis on Winter Wheat

Table 1

The effect of application of BAS 3170F to two cultivars at various growth stages on disease levels and yield

Growth stage and date at application	% disease level at application[1]		% disease level at GS 10.5.3[2]		Relative yield	
	A	B	A	B	A	B
Untreated	-	-	29.0	45.0	100	100
G.S 5 - 26.4.72	< 2.5	-	17.5	25.0	145*	145*
GS 6-7 - 15.5.72	< 5	< 2.5	16.0	21.0	146*	175*
GS 8-9 - 31.5.72	5	5	11.0	13.0	165*	183*
GS 9-10 - 15.6.72	15	20	17.5	19.0	146*	141*
GS 10.3 - 23.6.72	25	30	27.0	32.5	128*	138*
GS 6-7,9-10,10.3	-	-	7.5	10.0	183*	187*
Yield of untreated (kg/ha)					3,228	2,724
L.S.D. (P = 0.05)					36.8	29.0

Yellow rust was first seen in the crop at the end tillering.
Cultivar A - Maris Beacon
B - Joss Cambier

1 - on whole plant
2 - on flag leaf

In each case, an application at G.S. 8 gave the lowest disease levels on the flag leaf at G.S. 10.5.3 and resulted in the highest yield.

Table 2

Comparison of applications of BAS 3170F at growth stages 8 and 10 to cv. Joss Cambier in two trials (A and B)

Growth stage at application	% disease level at GS 10.5.1[1]		% ear infection		Relative yield	
	A	B	A	B	A	B
Untreated	14.0	40.0	19.0	29.0	100	100
8	2.5	15.0	13.5	19.7	138***	127***
10	2.5	27.0	10.2	16.0	126***	115
Yield of untreated (kg/ha)					4,068	3,516
L.S.D. (P = 0.001)					19.8	13.2

Yellow rust first appeared in Trial A at G.S. 7, and in Trial B at G.S. 5-6.

1 = on flag leaf

In both trials, application at G.S. 8 gave a 12% higher yield response than did application at G.S. 10.

Table 3

The effect of application of BAS 3170F applied at three growth stages to plots of cv Joss Cambier in which yellow rust first appeared at early tillering

Growth stage at application	% disease level at application [1]	% disease level at GS 10.5.1[2]		Relative yield	Thousand grain wt (g)
Untreated	-	40	(85)	100	35.8
5	20	30	(51)	114***	34.3
7	16	30	(24)	115***	37.2
9	26	7.5	(51)	122***	37.8*
Yield of untreated (kg/ha)				2,916	
L.S.D. (P = 0.05)				-	5.6
(P = 0.001)				11.8	-

1 - mean of top three leaves
2 - flag leaf, with 2nd leaf in brackets

Application at each growth stage gave an increase in yield, but only the later applications gave an increase in grain size.

Puccinia striiformis on Winter Barley

Table 4

The effect of BAS 3170F at various growth stages on the disease development and yield of cv. Astrix

Growth stage and date of application	% disease at application on leaves 1 & 2	3 & 4	% disease on flag leaves 6.6.	19.6.	Relative yield
Untreated	-	-	9.0	70.0	100
GS 6 - 28.4.	1	45	5.5	77.5	105
GS 6-7 - 7.5.	0	50	4.0	60.0	115*
GS 8 - 14.5.	0	40	1.5	32.5	116*
GS 10.1 - 22.5.	0	X	2.0	8.75	119*
GS 10.5 - 31.5.	8	X	3.5	15.0	110
GS 6,6-7,8,10.1,10.5			0	0	129*
Yield of untreated (kg/ha)					3659

X - leaves not assessed due to senescence

L.S.D. (P = 0.05) 13.4

Significant yield responses were obtained with treatments applied before infection of leaves 1 and 2, except for the first application, in which early re-infection occurred.

Table 5

The effect of application of BAS 3170F at various growth stages on brown rust levels and yield of cv. Proctor

Date of application	Growth stage at application	% brown rust at application		% brown rust Date of assessment 3.7.72		Relative yield	% grain >2.2 mm
		Top 3 leaves	Lower leaves	Flag leaf	2nd leaf		
Untreated				10	68	100	48.1
19.5.72 + 15.6.72 + 5.7.72	5)) 9)) 10)			0	1	186***	80.4
19.5.72	5	0	2.5	6	18	144**	40.2
1.6.72	6	2.5	5	3	9	147***	52.6
12.6.72	8	7.5	10	4	12	150***	67.6
22.6.72	10 - 10.5	35	60	3	13	131**	75.4
5.7.72	10.5	67	67			123**	57.0
Yield of untreated (kg/ha)						2716	
L.S.D. (P = 0.01)						23.7	
(P = 0.001)						33.2	

Good control of brown rust was obtained with all treatments, single treatments giving a control lasting approximately 5-6 weeks.

Best yield response was obtained with 3 applications of BAS 3170F, the best single treatment was that applied at G.S. 8 shortly before the onset of the rapid phase of rust development.

Table 6

Comparison of two timings of BAS 3170F against P. hordei in spring barley

Location and cultivar	Treatment	Growth stage at application	% brown rust at application		% brown rust Assessed 20.7.1972		Relative yield	% grain >2.2 mm
			Top 3 leaves	Lower leaves	Flag leaf	2nd leaf		
A. Creeting St. Mary	Untreated						100	88.9
	3170F	10.5	2	3			113	94.8
Proctor	3170F	10.5.4	5	8			110	95.1
B. Foxearth	Untreated				80	70	100	71.1
Midas	3170F	9 - 10	1	2.5	8	12	113**	84.0
	3170F	10.5	20	-	80	70	103	81.4
C. Foxearth	Untreated				15	70	100	92.7
Sultan	3170F	10.1	1	3	3	3	109**	95.2
	3170F	10.5	2	4	5	35	106	94.6

Yield of untreated: A - 3740.8 kg/ha; B - 3740.8 kg/ha; C - 4619.5 kg/ha

L.S.D. B - (P = 0.01) 10.1

L.S.D. C - (P = 0.01) 7.9

DISCUSSION

A. Puccinia striiformis on wheat

The results in Table 1 show that under the conditions experienced, i.e. a susceptible crop with rust first appearing immediately after tillering, the optimal timing of application for BAS 3170F was at, or soon after, emergence of the flag leaf. Where applications were made later, the effect on disease levels and yield were reduced. The results in Table 2 support these observations, and also show that treatment of the crop before ear emergence gives a reduction in disease levels on the ear.

Where applications were made to a crop infected before tillering, (Table 3) the disease control and yield increases were smaller. There is an indication that the earliest treatment was increasing grain number and becoming severely reinfected, whereas later applications gave an increase in grain size. These figures therefore suggest that under conditions of early and continuous infection, it may be necessary to make two applications for optimal benfit.

B. Puccinia striiformis on barley

Good control of P. striiformis was obtained with BAS 3170F applied at all growth stages. The period of control lasted 3-4 weeks in this cultivar.

Yield increases would seem to be directly related to the degree of rust control obtained, and where applications have been made shortly before the flag and second leaf became infected.

The applications made at G.S. 6 and G.S. 10.5 did not increase yields significantly, probably because with the first application early reinfection occurred, and with the late application premature leaf secescence on lower leaves plus infection on the upper leaves was not prevented.

Results presented in Table 4 show that applications made after flag leaf emergence, before infection on the upper leaves takes place, gave the highest yield benefit from a single application.

Two applications may be worthwhile in situations of early and prolonged attack.

C. Puccinia hordei on barley

BAS 3170F gave good control of brown rust in barley when applied before, or at the onset of, rust infection. Applications of BAS 3170F on established rust infection appeared to have little effect in controlling the disease (see Trial B, Table 6). This was confirmed in the yield results, the greatest responses resulting from applications of BAS 3170F made at, or just prior to, the phase of rapid development within the crop. Applications at this stage also resulted in substantial grain quality improvement (see Table 5).

In situations of early (G.S. 5) and severe brown rust infection, as reported in Table 5, it would appear necessary to treat more than once for maximum yield increase.

The action of BAS 3170F against brown rust in barley seems to be protectant with little or no eradicant activity.

References

LARGE, E.C. (1954) Growth Stages in Cereals. Plant Pathology, 3, 128-129.
POMMER, E.H. and KRADEL, J. (1969) Mebenil (BAS 3050F) - a new compound with specific action against some Basidiomycedes. Proceedings 5th British Insecticide and Fungicide Conference, 2, 563-569.
POMMER and ZWICK, (1971) Personal Communication.

Proceedings 7th British Insecticide and Fungicide Conference (1973)

CONTROL OF CERCOSPORELLA HERPOTRICHOIDES IN WINTER WHEAT IN GERMANY

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Summary Eyespot disease in cereals (Cercosporella herpotrichoides), is controlled efficiently by one application of benomyl, thiophanate methyl, carbendazin and other systemic fungicides in late spring. The effectiveness of spraying depends largely on correct timing. Earlier investigations by the authors had shown that the extent of infection is largely determined by the dependence of sporulation and infection on temperature, relative humidity and duration of periods with favourable temperature and relative humidity. The infection probability can be calculated for a given period based on these meteorological parameters. Fungicidal treatment should not take place before the end of a 40 days' period with high infection probability. The method makes use of the chemotherapeutic properties of the systemic fungicides and the long incubation period of the disease. The use of this method for a forecasting system in Germany is being investigated on a broad scale.

Résumé Le piétin-verse des céréales (Cercosporella herpotrichoides), peut être combattu efficacement par une application de benomyl, thiophanat M, bavistin ou autre fongicide systémique. L'efficacité d'une application dépend en particulier du moment où elle a lieu. Des précédantes recherches ont montré que la sporulation et l'etendue de l'infection dépendent en premier lieu de trois paramêtres meteorologiques (temperature, humidité relative de l'air ainsi que la durée des optima des deux précédants paramêtres). Une programmation numérique fut developpée pour permettre de calculer la probabilité de l'infection pour une période donnée; pour le calcul il est seulement tenu comptes des facteurs meteorologiques susmentionnés. Le traitement fongicide ne doit pas être appliqué avant la fin d'une période de 40 jours, qui accuse une grande porbabilité d'infection, afin de profiter de l'effet chimiothérapeutique du fongicide systemique et du grand temps d'incubation de la maladie. Actuellement, cette méthode est à l'essai pour développer un système d'alarme applicable à l'Allemagne.

INTRODUCTION

The fungus Cercosporella herpotrichoides causes eyespot disease of cereals, which gives rise to considerable losses in winter wheat and winter barley in Germany. Until several years ago, it was not possible to control the pathogen effectively by means of chemical plant protection. The main reason for the current importance of the disease is that many farmers have increased the percentage of wheat and barley in the rotation. Application of chlormequat (Cycocel) prevents plants from lodging, but the physiological damage caused by the fungus is not abolished by this procedure. The new systemic fungicides, based on benzimidazole and thiophanate, have stepped into this breach.

MATERIALS AND METHODS

Experimental methods and mathematical procedures were described earlier (Fehrmann 1970, Fehrmann and Schrodter 1971, 1972, Schrodter and Fehrmann 1971 a, b).

RESULTS

Table 1 demonstrates the average results from four field experiments in 1970, in which benomyl (Du Pont Benlate, 50% a.i.) was applied to winter wheat at four different dates. The fungicide controls the pathogen effectively, depending largely on the correct time of spraying. So far, in most cases, the best results were obtained after application of a systemic fungicide from stem extension up to the first node stage (end of stage 5 until stage 6 according to Feekes 1941 and Large 1954; stage H - I according to Keller and Baggiolini 1954). The economic injury level of the disease proved to be advantageous: a rate of 480 g/ha of fungicide at the most favourable spraying date lowered the percentage of diseased plants considerably, but yield was increased only to a minor extent when compared with the effect of half the amount of the substance (240 g/ha). In the same way, combinations of four sprayings, each at 240 g/ha, between the beginning of April and the beginning of June decreased disease incidence considerably, but yield was increased only slightly. The effects of chlormequat and benomyl on yield were apparently independent of each other. Applications of benlate in the autumn were much less effective than those in spring.

From these and other data we concluded that for practical purposes

- one spring application of a systemic fungicide is sufficient;
- the correct date of spraying is of major importance;
- the amount of fungicide should be sufficient to lower the incidence to the economic injury level: eradication of the fungus is not necessary;
- use of chlormequat does not exclude application of a fungicide, and vice versa.

As mentioned above, until now farmers in Germany have been recommended to spray a systemic fungicide at the beginning of stem extension (25 - 30 cm height) of the host plant. However, from the results of our ecological investigations on the epidemiology of Cercosporella herpotrichoides (Fehrmann and Schrodter 1971, 1972;

Table 1

Eyespot incidence and yield in winter wheat: dependence on timing of fungicidal treatment (Giessen 1970)

Spraying date	Benomyl g/ha	Chlormequat l/ha	Incidence (0 - 100)	Yield dz/ha	Yield %
-	-	-	75.0	54.0	100.0
9/4	240	-	73.0	53.3	98.7
28/4	240	-	63.3	55.3	102.4
14/5	240	-	47.9	59.0	109.3
2/6	240	-	62.7	55.9	103.5
14/5	480	-	29.5	59.9	110.9
all dates	240	-	17.0	59.9	110.9
14/5	-	1.5	74.0	61.2	113.3
14/5	240	1.5	48.0	65.5	121.3

(average from four experiments, four replicates each)

Schrodter and Fehrmann 1971 a, b) a method is being developed to predict the most favourable date for spraying. In this research it has been found that under field conditions, the processes of sporulation and infection are determined mainly by temperature, air humidity, and by the minimum length of periods with optimal conditions of the two parameters. It is possible to calculate the infection probability within a given period of time with high significance from these meteorological data.

Figure 1 demonstrates such curves for infection probability at two locations in the Frankfurt area in spring 1970. They are valid for the results of the fields experiments listed in Table 1. There we had found that May 14th was the best time for the application of the fungicide. Benomyl is able to control all infections which occured at least 40 days before spraying, but only those which occured five to ten days after application (Fehrmann 1970). Hence, it is possible to make use of the chemotherapeutic properties of systemic fungicides for predicting the optimum spraying date. In this connexion it is advantageous that the incubation time of eyespot is long, about 6 to 8 weeks. At the end of a period of 30 to 40 days with high infection probability, farmers are recommended to spray.

At present, this system is being tested on a broad scale in Germany. In 1972, ten field experiments with different spraying dates were carried out in various parts of the country. On average, application of benomyl at the right time increased yeild by 15.7% (untreated control: 38.9 dz/ha).

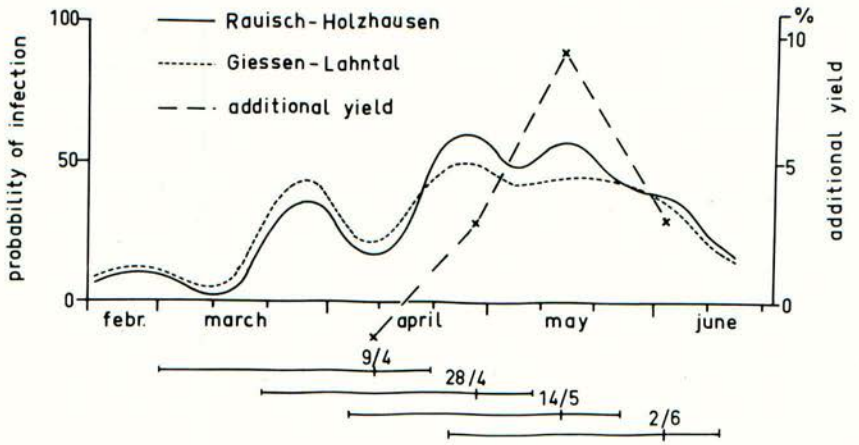

Figure 1 Left ordinate: Probability of infection of winter wheat by Cercosporella herpotrichoides at two different locations in Germany 1970 (Giessen-Lahntal, Rauisch-Holzhausen);

Right ordinate: Increase of yield after application of benomyl at different dates (data from Table 1);

Below: Approximate periods of effectiveness of benomyl against Cercosporella infections

In Figure 2, the curves for infection probability in spring 1970 at four different locations in Germany are plotted; each site of meteorological recording was in the area of one or several of the field experiments (Wulfshagen/Schleswig-Holstein: 2 experiments; Braunschweig: 1, Münster: 1, Weihenstephan/Bavaria: 5). Straight lines demonstrate the length of efficiency of the fungicide against infections (arrow = date of spraying). There is a good coincidence between those spraying dates which proved experimentally to be the most favourable, and the calculated optimum dates.

This result becomes even more clear from Figure 3. Here, the increase of yield (DM/ha, costs for fungicide deducted) at different spraying dates was plotted against time. It is very striking, that

- correct timing of a fungicidal treatment is most important for its economic effectiveness;
- calculated and experimentally determined optimum dates for treatment are close to each other.

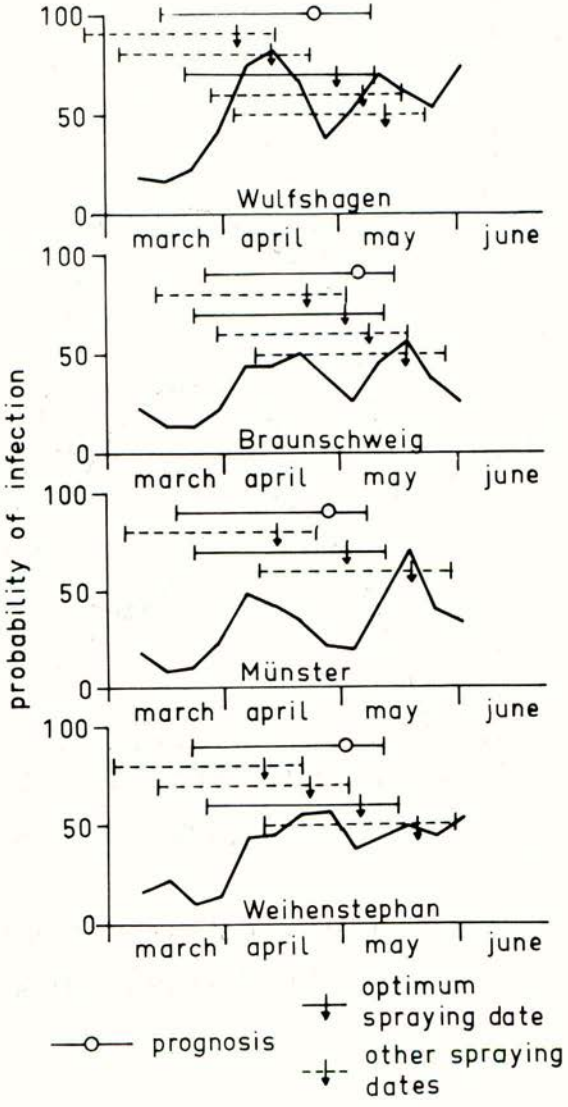

Figure 2 Curves for infection probability at four locations, Germany 1972. Experimentally and mathematically (prognosis) determined optimum dates for fungicidal treatment against Cercosporella herpotrichoides.

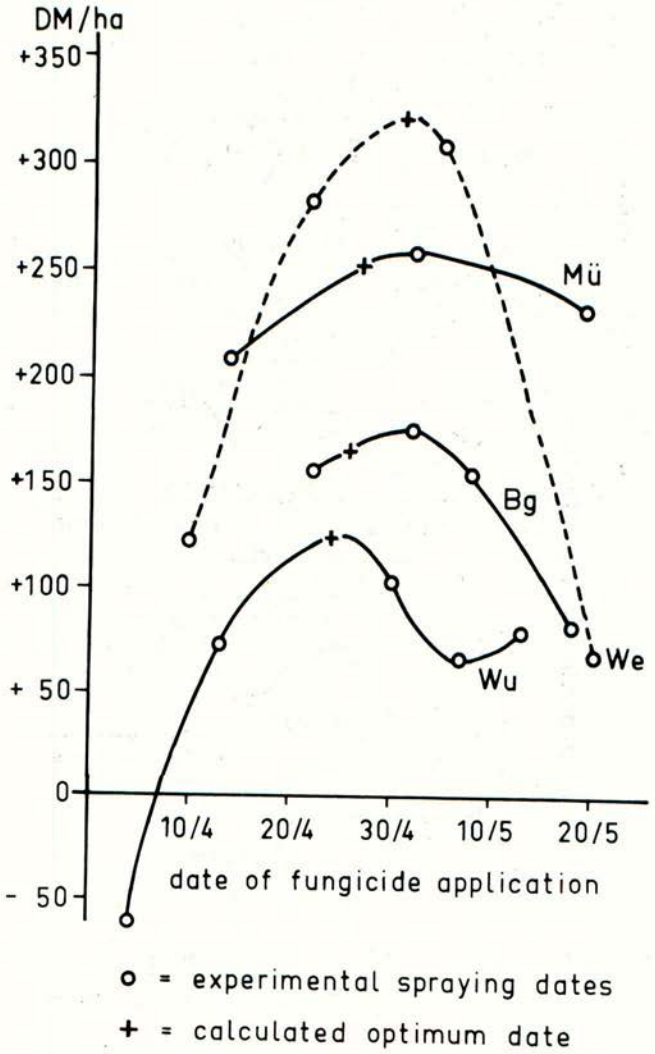

Figure 3 Increase of yield (DM/ha, costs for fungicide deducted) after application of benomyl at different dates (total of ten experiments).

The results justify the expectation, that by cooperation between the Plant Protection Service and the Meteorological Service, a forecasting system for the control of Cercosporella herpotrichoides in winter wheat will be established in Germany within a few years time.

DISCUSSION

At present, only a small part of the wheat area (1 - 3%, Hanf, 1973) is sprayed with fungicides. In the near future we expect a drastic increase in the consumption of systemic fungicides, especially for the northern part of the country. Here, the chemical control of Cercosporella herpotrichoides, Septoria spp., powdery mildew and to a certain extent rusts, may become a widely used routine procedure in wheat.

As has been pointed out, the effectiveness of spraying against the eyespot fungus largely depends on correct timing. For this reason, in most cases a combined application of chlormequat and fungicides against Cercosporella herpotrichoides is not possible: chlormequat is sprayed when the crop is about 10 to 15 cm high. The most effective spraying against glume-blotch takes place after heading. Where mildew appears together with Septoria nodorum on the heads, both fungi may be controlled by thiophanate methyl (Löcher and Hampel, 1973).

As far as the control of Cercosporella herpotrichoides is concerned, there is still a problem. Due to the long incubation period, wheat growers are often unable to evaluate whether or not the application of a fungicide is economically justified. This is especially true when, after a period of high infection probability in March and April, no symptoms can be seen at the time of stem extension. Absence of symptoms, however, does not always mean absence of the fungus in the plants. Spraying may be recommended in all cases, where cereals are dominant in the crop rotation and where the farmer knows that Cercosporella herpotrichoides is a potential problem in his field. The menace of the eyespot fungus is greater on heavier soils than on lighter, sandy ones, so that the economic risk of fungicidal treatment is less in fields with soil optimal for wheat.

Acknowledgements

Thanks are due to members of the German Plant Protection Service for carrying out field experiments.

References

FEEKES, W. (1941) De tarwe en haar milieu. Verslag XVII. Technische Tarwe Commissie, Groningen, p. 560-561.

FEHRMANN, H. (1970) Bekämpfung der Halmbruchkrankheit des Weizens mit Benomyl. Nachrichtenblatt Deutscher Pflanzenschutzdienst (Braunschweig), 22, 136-139.

FEHRMANN, H., and SCHRÖDTER, H. (1971) Ökologische Untersuchungen zur Epidemiologie von Cercosporella herpotrichoides. I. Jahreszeitliche Abhängigkeit von Infektionen im Freiland. Phytopathologische Zeitschrift, 71, 66-82.

FEHRMANN, H. and SCHRÖDTER H. (1972) Ökologische Untersuchungen zur Epidemiologie von Cercosporella herpotrichoides. IV. Erarbeitung eines praxisnahen Verfahrens zur Bekämpfung der Halmbruchkrankheit des Weizens mit systemischen Fungiziden. Phytopathologische Zeitschrift, 74, 161-174.

HANF, M. (1973) Derzeitiger Stand der Pflanzenschutzmittel-Anwendung im Getreidebau. BASF-Mitteilungen für den Landbau, 3/73, 122-130.

KELLER, C. and BAGGIOLINI, M. (1954) Les stades repères dans la végétation du blé. Revue Romande de Agriculture, Viticulture et Arboriculture (Lausanne), 10, 17-20.

LARGE, E.C. (1954) Growth stages in cereals. Illustration of the Feekes scale. Plant Pathology, 3, 127-128.

LÖCHER, F. and HAMPEL, M. (1973) Neue Fungizide zur Bekämpfung von Getreidekrankheiten. BASF-Mitteilungen für den Landbau, 3/73, 146-165.

SCHRÖDTER, H. and FEHRMANN, H. (1971 a) Ökologische Untersuchungen zur Epidemiologie von Cercosporella herpotrichoides. II. Die Abhängigkeit des Infektionserfolges von einzelnen meteorologischen Faktoren. Phytopathologische Zeitschrift, 71, 97-112.

SCHRÖDTER, H. and FEHRMANN, H. (1971 b) Ökologische Untersuchungen zur Epidemiologie von Cercosporella herpotrichoides. III. Die relative Bedeutung der meteorologischen Parameter und die komplexe Wirkung ihrer Konstellationen auf den Infektionserfolg. Phytopathologische Zeitschrift, 71, 203-222.

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CONTROL OF CEREAL DISEASES WITH CARBENDAZIM

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Summary In several trials carbendazim (methyl-2-benzimidazole carbamate) was effective against eyespot (Cercosporella herpotrichoides) and leaf and ear diseases (Septoria nodorum, Erysiphe graminis) in wheat. The most favourable period for the control of Cercosporella was between growth stages 5/6, i.e. end of tillering and first nodul stage. The treatment with the fungicide was also worthwhile when the infestation with Cercosporella did not lead to lodging. The application of carbendazim for the control of glume blotch and mildew during the period shortly before and after heading resulted in high yield increases; the fungicidal treatment was most effective after the wheat had formed ears. It was worthwhile applying several treatments when eyespot and ear diseases appeared in succession.

INTRODUCTION

Since the successful introduction of fungicides for the control of powdery mildew at the end of the sixties, cereal diseases have been receiving increasing attention. In the meantime numerous tests have shown that foot, leaf and ear diseases of cereals are important factors that can reduce yield. The development of broad-spectrum fungicides of the benzimidazole and thiophanate groups opened new possibilities for controlling foot and ear diseases, which up till then could only be controlled by plant hygiene. This paper reports on trial results obtained so far with carbendazim, a benzimidazole derivative, when used as a spray for the control of foot and ear diseases of cereals.

METHOD AND MATERIALS

Carbendazim contains 50% methyl-2-benzimidazole carbamate. The following results were obtained during field trials in 1972. Infestation by Cercosporella was assessed according to instructions given by the Biologische Bundesanstalt für Land- und Forstwirtschaft (Anon., 1971), the German equivalent of ADAS. Unless noted otherwise, the assessment was carried out when the cereal was milky ripe to hard (Feekes Scale 11.1 - 11.3). Lodging was expressed in figures on a scale of 1-9 (1 = no lodging, 9 = total lodging). The assessments of infestation with glume blotch (Septoria nodorum) and mildew (Erysiphe graminis) were carried out during milky to hard ripeness. The infestation was evaluated on a scale 1-9 (1 = not infested, 9 = totally infested). Details on application times are based on the growth stage of the cereal (according to Large).

RESULTS

1. The fungicidal efficacy of carbendazim against eyespot

Infections of eyespot in winter cereals can occur from autumn to late spring. Trials by various authors (Bruehl and Cunfer 1972, Huber and Mulanax 1972) have shown that treatments with benomyl in autumn were in general less effective than applications in spring. These reports conform with tests carried out by Fehrmann and Schrödter (1970) in which eyespot was effectively controlled by spraying approx. 40 days after the pathogen was inoculated into wheat. Spraying before the onset of infestation was less successful. For this reason we examined the influence of carbendazim on eyespot by applying in spring when the disease first appeared.

The trial results in Table 1 show that control measures can be taken over a relatively long period, i.e. when tillers form to the time when the last leaf is just visible. The most favourable time to apply carbendazim for controlling eyespot is during the growth stages 5/6, i.e. end of tillering to formation of the first node. This is rather later than the application date for the growth regulator chlormequat (® Cycocel).

Table 1

Control of eyespot in winter wheat with carbendazim

(25 trials)

	g a.i./ha	Date of application	Cercosporella % infestation	Lodging (1-9)	Yield kg/ha	Yield rel.
Control	-	-	92.2	5.1	4040	100
Chlormequat*)		3/4	90.0	4.2	4090	101
Carbendazim	125	3/4	83.5	4.1	4320	107
Chlormequat*) + carbendazim	125	3/4	83.1	3.4	4350	108
Chlormequat*) + carbendazim	125	5/6	71.9	3.1	4480	111

*) application rates depending on locations and/or varieties

Considerable improvements in yield were achieved after the application of carbendazim, although the eyespot infestation was apparently only slightly reduced. It must, however, be taken into account that the assessments of infestation shown in Table 1 took place relatively late, i.e. when the wheat was hard ripe.

Table 2 demonstrates how the eyespot infection progressed during the vegetative period. Immediately after application with carbendazim, the infection was suppressed to a large degree. However, as the vegetative period progressed the efficacy gradually diminished. The main reason for the notable increase in yield is that following treatment, the cereal remains healthy for a longer period than the control so that eyespot infection cannot occur until later and thus will not cause so much damage. The lower scores recorded towards the end of the vegetative period (growth stage 11.1 - 11.3) do not represent a real reduction in eyespot infestation, but are brought about by the fact that symptoms of infection are more difficult to recognise when the crop is fully ripe, and they could no longer be fully recorded in the assessments.

Table 2

The course of infection by eyespot in winter wheat during the vegetative period

(2 trials)

	% infestation of eyespot during growth stage 3/4	7/8	10.1/10.5	11.1/11.3
Control	28	58	78	66
Carbendazim		17	52	40
Relative to control		29	67	61

Time of application: 5/6

The growth regulator chlormequat is first and foremost known to reduce the physiological lodging of cereals. When chlormequat and carbendazim are applied as a combination, both physiological and parasitic lodging can be checked. Table 3 shows how wheat responds to treatment with chlormequat or the combination chlormequat and carbendazim. Whilst application with chlormequat was only worthwhile when lodging was severe, carbendazim brought improvements in yield irrespective of the intensity of lodging. These trials indicate that eyespot causes reductions in yield even if parasitic lodging does not occur and that fungicidal treatment is justifiable regardless of the intensity of lodging.

Table 3

Control of eyespot, degree of lodging and its influence on the yield of winter wheat

	slight lodging*)			more severe lodging**)		
	Lodging (1-9)	Cercosporella % infestation	Yield	Lodging (1-9)	Cercosporella % infestation	Yield
Control	1.7	91.6	100 (4660 kg/ha)	6.6	91.3	100 (3790 kg/ha)
Chlormequat	1.3	88.4	100	5.1	86.5	106
Chlormequat + carbendazim	1.1	69.9	106	4.4	71.3	113
No. of trials		11			16	

*) Trials with an evaluation score of 1 - 3.9 in Control
**) Trials with an evaluation score of 4 - 9 in Control

Up to now literature on the subject has supported the view that barley is more resistant to eyespot than wheat, and for this reason suffers only small yield losses. Our own tests on the extent of infestation by eyespot in winter barley do not confirm this view. Application of the fungicide to winter barley (Table 4) resulted in yield increases similar to those in winter wheat.

Table 4

Control of eyespot in winter barley

(5 trials)

	g a.i./ha	eyespot % infestation	Yield kg/ha	rel.
Control	-	93	5910	100
Carbendazim	125	84.8	6370	108

Time of application: 5/6

Similar observations have also been made in rye.

2. The fungicidal efficacy of carbendazim against ear diseases (glume bloth, powdery mildew) in wheat

Melville and Jemmett (1971) and Douchet et al. (1972) have reported on the efficacy of benomyl and methyl thiophanate against glume blotch (Septoria nodorum). In 1972 there was a severe outbreak of ear mildew (Erysiphe graminis) as well as glume blotch in West Germany. There was a perceptible reduction of infection in the case of both pathogens after treatment with carbendazim (Table 5). Although the outbreak of ear diseases could not be checked completely, treatment with the fungicide resulted in increases in yield of about 10%. Test results show that control measures taken even before the heading stage bring about useful improvements in yield, although the fungicide is doubtless best applied after heading.

Table 5

Efficacy of carbendazim against ear diseases in wheat

(12 trials)

	g a.i./ha	Time of application	Septoria (1-9)	Mildew (1-9)	Yield kg/ha	rel.
Control	-	-	4.2	4.4	4320	100
Carbendazim	125	8/9	3.8	4.0	4590	106
Carbendazim	125	10.1/10.5	3.6	3.4	4860	113

In order to be able to register the extent and intensity of infestation by both pathogens, glume blotch and ear mildew were assessed separately. A selection of trials (Table 6), in which only mildew appeared to any great extent, showed more clearly than the results given in Table 5 that carbendazim reduced infections of ear mildew too, thus promoting increases in yield.

Table 6

Efficacy of carbendazim against ear mildew in wheat

(8 trials)

	Ear mildew	Septoria	Yield kg/ha	rel.
Control	4.7	1.5	5300	100
Carbendazim	3.8	1.4	5670	107

Time of application: growth stage 10.1/10.5

Mildew lesions on the ears of wheat make the plant more susceptible to later infections by glume blotch. Wheat crops whose leaves are quite heavily infested with mildew before heading, and in which a later infection of the ears can therefore be anticipated, should be treated with fungicide as early as possible, i.e. immediately after shooting of the ear up to the beginning of the flowering stage. The main effect of carbendazim on cereal mildew is prophylactic.

In areas or varieties with glume blotch only, control measures can still be taken at a later date, i.e. until the end of the flowering stage. Infestations of the ear by glume blotch usually occur later than mildew infections. Staggered application of fungicides in wheat inoculated with glume blotch revealed that optimum control was achieved when treatment took place immediately before and after infection. Even so, economically important yield increases were obtained when fungicide was applied 16 days after infection.

3. Combined applications of carbendazim and maneb

There was a close correlation between the results of various tests showing that a combined application of carbendazim and maneb resulted in a higher increase in yield than an application of benzimidazole derivatives alone. The superior effect of the combination of carbendazim and maneb is probably due to the following factors:

a) The fungicidal efficacy of carbendazim against the pathogens of ear diseases is intensified by the addition of maneb. This applies to both glume blotch and powdery mildew, although maneb alone is not effective against mildew.

b) The fungicidal spectrum of the mixture is greater than that of the components. Dithiocarbamates control black mould, e.g. Alternaria spp. which attack the ears and they are also effective to a certain extent against leaf diseases breaking out at a later stage, e.g. cereal rusts.

c) In soils with a low manganese content, the nutrient value of the trace elements in maneb may be utilised, thus resulting in yield increases.

d) Lastly it is well-known that the use of maneb in fruit-growing promotes an especially healthy and green foliage. Cereals also remain green longer after treatment with maneb, and are thus capable of assimilating over a longer period which can make additional yield increases possible. It is still not certain however whether the optical improvement achieved by maneb is a result of its fungicidal qualities or the effects of its trace elements.

DISCUSSION

Table 7

Control of foot and ear diseases of wheat with carbendazim

(11 trials)

	g a.i./ha	Time of application	Yield kg/ha	rel.
Control	-	-	4560	100
Carbendazim	125	3/4	4850	106
Carbendazim	125 + 125	3/4 + 10.1/10.5	5140	113

Chlormequat was applied in these trials

There is no doubt that various single measures for the control of crop diseases do achieve success, i.e. yield increases. In agricultural practice, however, it is important to know to what extent repeated applications of fungicides for the control of various crop diseases can increase successive yields, or whether the yield potential of the plant is confined within strict genetic limits. It can be seen from Table 7, taking the foot and ear diseases as an example, that individual measures can have a cumulative effect. The trial results show that a single application of carbendazim for the control of foot diseases can no longer influence or make less serious a subsequent attack of ear diseases. It is, therefore, perfectly justifiable and of economic interest to apply a number of treatments of the fungicide when several cereal diseases appear in succession.

References

ANON. (1971) Vorläufige Richtlinien für die Prüfung von Fungiziden gegen Cercosporella herpotrichoides Fron. (Halmbruchkrankheit) an Getreide. Biologische Bundesanstalt für Land- und Forstwirtschaft, Braunschweig.

BRUEHL, G.W. and B. CUNFER (1972) Control of Cercosporella root rot of wheat by benomyl. Plant Dis. Reptr., 56, 20-23.

DOUCHET, J.P., G. QUERE and J. LHOSTE (1971) Action fongicide de l'association methylthiophanate + manèbe dans la lutte contre les maladies des organes aériens des céréales. Phytiatrie - Phytopharmacie, N° Spécial, 37-50.

FEHRMANN, H. and H. SCHRÖDTER (1972) Ökologische Untersuchungen zur Epidemiologie von Cercosporella herpotrichoides IV. Erarbeitung eines praxisnahen Verfahrens zur Bekämpfung der Halmbruchkrankheit des Weizens mit systemischen Fungiziden, Phytopath. Z., 74, 161-174.

HUBER, D.M. and M.W. MULANAX (1972) Benomyl rates and application times for wheat foot rot control. Plant Dis. Reptr., 56, 342-344.

MELVILLE, S.C. and J.L. JEMMETT (1971) The effect of glume blotch on the yield of winter wheat. Pl. Path., 20, 14-17.

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THE EFFECT OF SEED CONDITION AND FUNGICIDAL DRESSINGS ON THE FIELD EMERGENCE OF BARLEY

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Summary The emergence of seed lots of Golden Promise barley was found to be inversely related to the water-sensitivity of the lots in germination tests and directly related to the rates of oxygen uptake of the lots after 24 h in excess moisture in the laboratory. Organomercurial seed dressings reduced water-sensitivity and improved field emergence. Antibiotics in the germination media and seed dressings reduced the rate of oxygen uptake but did not eliminate differences between seed lots. The relevance of the seed microflora and of physiological differences between seed lots to the laboratory performance and field emergence of barley is discussed.

INTRODUCTION

Emergence has seldom been regarded as a problem in barley, possibly because of the ability of barley crops to compensate for low plant numbers by an increase in grain weight per plant (Kirby, 1969). However, rising seed costs may well lead to a reduction in the sowing densities in general use, and emergence in barley might be examined more critically since reduced densities coupled with poor emergence could diminish yields.

The present study was undertaken to compare the emergence levels of barley seed lots commercially available in Scotland, both in the dressed and untreated condition, and to evaluate the predictive value of the laboratory germination test and of germination in excess water, which has been shown by maltsters to be highly variable in barley grain samples (Essery et al, 1954). Differences between seed lots and chemical seed treatments were related to differences in the oxygen uptake of the seeds.

METHODS AND MATERIALS

Samples of seed lots of barley (variety Golden Promise) produced in Scotland and obtained in an untreated condition from a seed merchant were used, seven seed lots from the 1970 harvest in 1972 and ten from the 1972 harvest in 1973. The term seed is used throughout in the agronomic sense.

Germination was tested in 9 cm diameter Petri dishes containing 70 g of dry dry-sterilised sand to which 9 or 18 ml of distilled water were added. Dishes were placed in deep trays and covered with moisture-proof wrapping and incubated at 20°C for 7 days. The dish weights were checked after 2 days and water was added if necessary; no more than 0.5 ml was ever needed. Normally 20 seeds

were sown in a dish at a depth of 0.5 cm ventral side uppermost and seeds were counted as germinated when the coleoptile emerged.

In some experiments a solution containing two antibacterial antibiotics, streptomycin sulphate and benzyl penicillin, each at 800 ppm, and an antifungal antibiotic, mycostatin, at 400 ppm, was added to the sand instead of water. The pH of the solution was adjusted to 7 before use. Dressing of small quantities of seed was done with a commercially available fungicide containing 1.6% a.i. of organomercurial compounds (ethyl mercury chloride and phenyl mercury acetate) at a rate of 1 to 336 by weight by rotating in a sealed glass jar for 5 minutes.

Seed oxygen uptake was measured at 20°C in a Gilson Differential Respirometer. After 24 h in Petri dishes of moistened sand five samples of 20 seeds were placed in separate 17 ml reaction vessels with 0.5 ml of distilled water. Two flasks containing only 0.5 ml of water were included in each tank as thermoblanks (Carver and Gloyne, 1971). Each sample was dried for 24 h at 105°C and weighed. Eventually oxygen uptake was expressed as microlitres at standard temperature and pressure per hour per g dry weight.

Emergence in the field was determined in a medium loam soil. Seeds were sown by hand, 100 seeds per row, at a depth of 3.8 cm in 152 cm rows set 15.2 cm apart. In January 1972 seven seed lots were sown in the dressed and untreated condition in five replicate blocks. In 1973 ten lots in the dressed and untreated condition were sown on eight dates (Table 1) in five replicate blocks. In a second field experiment in 1973 five lots were sown at two depths (3.8 cm and 10.2 cm) on two dates in a dressed and untreated condition. In this experiment only 50 seeds were sown of each individual treatment in 76 cm rows in each of five replicate blocks. Emergence counts of all experiments were made at regular intervals until constant. The total rainfall for the period 2 days before and 7 days after each sowing in the first experiment in 1973 (Table 1) and hours below 10°C (at 5 cm depth in soil) for the day of sowing and 7 days after, were recorded at a station within 200 m of the sowing site.

RESULTS

The germination of all seed lots was tested in sand at two moisture levels which were produced by the addition of 9 and 18 ml of distilled water to Petri dishes of sand. Germination at the lower moisture level was taken as the laboratory germination. The effect of the higher moisture level on the germination of the seed lots was expressed as follows:

$$\frac{\%\text{ germination with 9 ml} - \%\text{ germination with 18 ml}}{\%\text{ germination with 9 ml}} \times 100$$

and was called % water-sensitivity. This term was first used by Essery et al (1954) who noted differences in water-sensitivity between barley grain samples.

The mean percentage laboratory germination and water-sensitivity of each of the ten lots used in 1973 were determined in the untreated condition for five replicates of twenty seeds. No significant differences were found in laboratory germination which ranged from 93 to 98% but several significant differences in water-sensitivity were found between seed lots, which ranged from 44 to 87%.

The mean field emergences were determined for each seed lot of five replicates of 100 seeds on eight sowing dates, that is, a total of 4,000 seeds per lot. Seed

lots in the untreated condition showed a range of emergences from 70 to 84% and some significant differences between lots. Dressed seed showed a smaller range from 73 to 79% and no significant differences between lots. Over the whole experiment dressing produced a small but significant ($p < 0.01$) increase in emergence from 75 to 77% and the overall emergence of seven out of the ten lots was improved by dressing.

Considerable differences in mean emergence were found between sowing dates (Table 1) which ranged from 50 to 83% for untreated seed and from 49 to 85% for dressed seed. Laboratory germination appeared to be a poor indicator of the field emergence of the seed lots since it was never significantly correlated with emergence. However, water-sensitivity was negatively and significantly correlated with field emergence on the four sowing dates when the mean emergences of the untreated seed were at their lowest (Table 1). These sowing dates occurred either during a time of low soil temperatures (23rd March and 3rd April) or during a period of high rainfall (3rd May and 20th May). In the 20th May sowing when water-sensitivity was highly correlated with emergence, seed dressing produced the greatest improvement in emergence (Table 1). The mean emergences of the ten seed lots over all the sowings were inversely and significantly related to water-sensitivity (Fig. 1a).

Table 1

Mean percentage field emergence of ten seed lots of barley at each of eight sowing dates in 1973 and correlations between emergence and laboratory measures of performance

Sowing Date	Untreated	Dressed	Correlation coeff. with field emergence: Lab germination	Water-Sensitivity	Total rain(mm)	Hours below 10°C
23 Mar	50a	49a	.4774	- .6533*	10.4	192
3 Apr	74bc	76b	.4629	- .6497*	22.6	192
12 Apr	82e	83e	- .1893	- .5944	0	152
20 Apr	83e	85f	- .3688	- .4116	3.5	129
25 Apr	79d	81cde	.1291	.1568	18.0	132
3 May	78cd	79bc	- .2160	- .8391**	41.2	138
20 May	74b	80cd	.0655	- .9080***	23.5	91
28 May	79d	82de	- .3030	- .3189	16.9	29

Significance levels: *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.
Means in columns with a letter in common are not significantly different ($p = 0.05$).

Increasing the depth of sowing considerably reduced emergence of all the five seed lots tested at both sowing dates (Table 2). In all but two comparisons between dressed and untreated seeds sown deeply there was an improvement in emergence with dressing and in three instances this improvement was a significant one. No such significant improvements were seen in the shallower sowing. Seed lots showed some significant overall differences in emergence; seed lot 10 which gave the lowest emergence (52%) had also shown the greatest water-sensitivity (87%).

Table 2

Mean percentage emergence of five seed lots of barley sown on two dates at two depths in a dressed (+) and untreated (-) condition

	21 May				30 May				
	Shallow		Deep		Shallow		Deep		
Seed lot	+	-	+	-	+	-	+	-	Mean
10	86	80	15	16	78	78	42	26	52a
5	94	90	27	9	84	82	86	62	67b
1	95	93	28	24	86	83	80	77	71b
9	94	85	30	21	90	90	81	65	70b
3	89	89	31	19	90	92	76	76	69b

Pairs of means underlined are significantly different ($p = 0.05$).

The effect of seed dressing on emergence was also examined in an early sowing (Jan 26th) in 1972. The mean emergence of seven seed lots, each of which had a laboratory germination greater than 95%, was significantly ($p < 0.001$) improved by dressing from 43 to 51% and the mean water-sensitivity of the same lots was significantly ($p < 0.01$) reduced from 30 to 17% by dressing.

The rate of oxygen uptake of seeds of all ten lots sown in 1973 was determined after 24 h in sand moistened with 9 and 18 ml of distilled water and the same amounts of antibiotic solution. Increasing the amount of moistening liquid significantly ($p < 0.001$) reduced the mean oxygen uptake of five replicates of the ten lots, measured after the removal of the seeds from the sand, from 110.9 to 65.5$\mu l h^{-1} g^{-1}$ dry weight in the case of water and from 102.1 to 46.3$\mu l h^{-1} g^{-1}$ dry weight in the case of antibiotic solution. Antibiotic solution significantly ($p < 0.001$) reduced the oxygen uptake compared with water at the higher moisture level but did not adversely affect germination at either 9 ml or 18 ml, in fact, at the higher moisture level the mean germination of five replicates of ten seed lots was increased from 25 to 31% but not significantly. Dressing the seed with fungicide reduced, but not significantly, the mean rate of oxygen uptake of five replicates of four seed lots after 24 h in sand moistened with 18 ml of water from 74.5 to 63.9$\mu l h^{-1} g^{-1}$ dry weight but did not eliminate differences between lots.

Although the overall effect of increasing the moisture level of the sand was to reduce oxygen uptake not all the ten seed lots were affected similarly. The percentage reduction of oxygen uptake with increasing sand moisture ranged from 3 to 61%. Thus if the oxygen uptake measurements are indicative of the uptake in the sand before removal then seed lots differ in their ability to maintain their oxygen uptake in high moisture conditions.

The mean oxygen uptake of the seed lots after 24 h in high moisture sand was inversely and significantly ($p < 0.05$) related to water-sensitivity (Fig 1b) and directly and significantly ($p < 0.01$) related to field emergence (Fig 1b). Antibiotic solution did not eliminate differences in the rate of oxygen uptake between seed lots after 24 h in high moisture sand, but uptake was no longer significantly related to either water-sensitivity or emergence.

Examinations of five of the untreated seed lots used in 1973, which had water-

sensitivities ranging from 44 to 84% revealed low levels of incidence of Fusarium nivale (2 - 10.5%) and little Septoria nodorum (< 1.5%) and no infections of loose smut (Ustilago nuda) were found in embryo tests on the same lots.

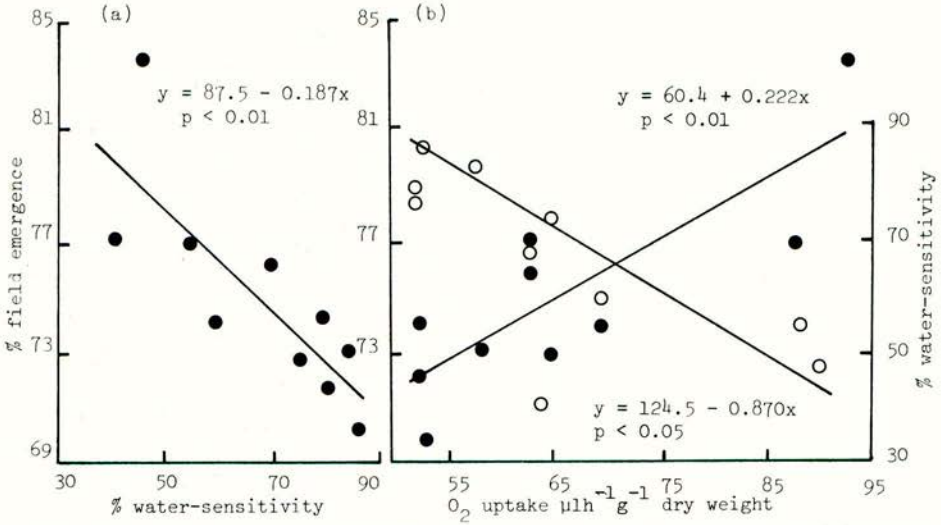

Fig. 1(a) Relation between water-sensitivity and field emergence and (b) relation between oxygen uptake and field emergence (●) and between oxygen uptake and water-sensitivity (O) for ten seed lots of barley

DISCUSSION

The field emergence of barley observed in this work was generally high, usually more than 70% of the seeds sown emerged. However, lower emergences and differences in emergence between seed lots were seen in early sowings, sowings made during periods of high rainfall and in abnormally deep sowings.

Seed lots that emerged well under these relatively adverse conditions were also able to germinate in the laboratory under conditions of excess moisture. Differences in so-called water-sensitivity have been reported for barley before and explained in terms of differing abilities among seed lots to germinate under conditions where oxygen availability is limited (Crabb and Kirsop, 1969).

Gaber and Roberts (1969) overcame the water-sensitivity of barley seeds by the addition of a combination of antifungal and antibacterial antibiotics to the germination medium and suggested that water-sensitivity resulted from competition for oxygen from high populations of microorganisms in conditions where oxygen availability was limited. In the present work antibiotics did not significantly reduce water-sensitivity but when added to germination media they did reduce the rate of oxygen uptake following removal from conditions of excess moisture.

However, antibiotics in the germination media did not eliminate differences between seed lots suggesting that there were physiological differences between seed lots not associated with microorganisms. Nevertheless, high populations of seed-borne microorganisms may sometimes contribute to reduced germination in excess moisture in the laboratory.

The rates of oxygen uptake measured after 24 h in excessively moist conditions were related to water-sensitivity and field emergence when antibiotics were not used in the germination media, low oxygen uptake being a feature of lots that were water-sensitive and emerged less well in the field. If the uptakes measured are indicative of the oxygen uptake of the seeds in the germination media then the inability of some seed lots to maintain aerobic respiration in conditions of limited oxygen availability may be a cause of water-sensitivity and relatively poor emergence under adverse conditions in the field.

The relevance of the seed-borne microflora to field emergence is difficult to evaluate. Organomercurial dressing improved both the field emergence of seed lots, on which few specific pathogens were observed, and their germination in conditions of excess moisture. Dressings also reduced the oxygen uptake of seeds measured after their removal from excessively moist germination conditions. Organomercurial dressings are not only effective against seed-borne pathogens of cereals but their non-selective action has also been found to reduce the saprophytic fungal flora of barley seed (Mills and Wallace, 1970). This general disinfection may have a beneficial effect in emergence situations where oxygen availability is limited by high soil moisture.

Differences exist between seed lots of barley which affect their emergence ability. These differences can be readily observed in germination tests in conditions of excess moisture which may thus provide a method for detecting seed lots of relatively low field emergence potential. These differences should also be kept in mind when chemicals are being tested for their effects on the field emergence of barley.

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References

CARVER, M. F. F. and GLOYNE, A. R. (1971) The effect of variations in ambient temperature on the Gilson differential respirometer. Laboratory Practice, 20,433.

CRABB, D. and KIRSOP, B. H. (1969) Water-sensitivity in barley 1. Respiration studies and the influence of oxygen availability. Journal of the Institute of Brewing, 75, 254-259.

ESSERY, R. E., KIRSOP, B. H. and POLLOCK, J. R. A. (1954) Studies in barley and malt 1. Effects of water on germination tests. Journal of the Institute of Brewing, 60, 473-481.

GABER, S. D. and ROBERTS, E. H. (1969) Water-sensitivity in barley seeds 2. Association with micro-organism activity. Journal of the Institute of

Brewing, 75, 303-314.

KIRBY, E. J. M. (1969) The effect of sowing date and plant density on barley. Annals of Applied Biology, 63, 513-521.

MILLS, J. T. and WALLACE, H. A. H. (1970) Differential action of fungicides upon fungi occurring on barley seed. Canadian Journal of Plant Science, 50, 129-136

NOTES

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THE EFFECTIVENESS OF BIS (8-GUANIDINO-OCTYL) AMINE* AS A SEED DRESSING, ALONE OR IN MIXTURE WITH OTHER FUNGICIDES, AGAINST DISEASES OF CEREALS

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Summary Guazatine was first reported at the 1st Congress of Plant Pathology as having high activity against the spectrum of cereal seed-borne diseases normally controlled by mercury-based seed-dressings, particularly Leptosphaeria nodorum, Calonectria nivalis, Pyrenophora avenae, Tilletia caries and Ustilago avenae. Guazatine is active against these diseases at 0.8 g/kg seed.

More recent work has confirmed this activity but has also made apparent a weakness against barley leaf stripe at high infection levels. A guazatine/carboxin mixture was found to give good control of barley leaf stripe and to widen the activity spectrum to include loose smut of barley. Where loose smut control is not required, guazatine/maneb combined powder seed dressing provides a satisfactory candidate material for disease control.

Résumé La Guazatine a fait l'objet d'une communication au premier Congrès de Pathologie Végétale en 1968 signalant une haute activité à l'egard des maladies des semences de céréales normalement contrôlées par le mercure en traitement des semences, notamment Leptosphaeria nodorum, Calonectria nivalis, Pyrenophora avenae, Tilletia caries et Ustilago avenae. La Guazatine est efficace contre ces maladies à 0.8 g par kilo de semence.

Des recherches plus récentes onto confirmé cette activité mais elles ont aussi mis en évidence une faiblesse contre l'helminthosporiose de l'orge pour une contamination élevée. L'addition de carboxine améliore le contrôle de l'helminthosporiose de l'orge et permet de lutter contre le charbon nu. Là où le contrôle du charbon nu de l'orge n'est pas requis, un mélange sous forme de poudres à base de quazatine et de manèbe en traitement des semences constitue une solution satisfaisante pour le contrôle des autres maladies.

* Common name (ISO proposed name): Guazatine
(Also known as MC 25, MC 200 and EM 379)

INTRODUCTION

The many seed-borne diseases which attack cereals have been adequately controlled with only one or two exceptions, by mercury-based materials applied as seed dressings (Hewitt, 1967). The current trend towards the use of relatively non-toxic pesticides, coupled with the resistance of Pyrenophora avenae to mercury-based treatments, (Greenaway and Cowan, 1970) has led to a restriction in use of these materials in several countries (Granhall, 1971) and to a search for safer alternative fungicides.

It is believed that guazatine is capable of forming the basis of such an alternative (Catling W.S. and Cook I.K., 1968, pers.comm.); it is relatively non-toxic to humans and wildlife, and extensive laboratory and field tests showed it to have promising activity against a wide range of seed-borne pathogens.

Further testing, however, showed that the activity of guazatine was not always comparable with that of mercury treatments, particularly against barley leaf stripe (Unpub. data, 1972). More recent work has therefore been directed towards testing guazatine in mixture with other fungicides in order to extend the activity spectrum; preliminary laboratory results with guazatine/carboxin and guazatine/maneb mixtures were very promising, and results of subsequent field trials with these mixtures compared with guazatine alone are reported here.

METHOD AND MATERIALS

Formulation used Bis (8-guanidino-octyl) amine (guazatine) was used as the water soluble acetate or insoluble sesquisulphate salt (40% a.i. w/v or 40% a.i. w/w respectively). Other formulations mentioned in the paper are detailed below. The powder formulations contain the sesquisulphate salt and the slurry formulations the acetate salt. (% a.i. w/w for powders, % a.i. w/v for slurries).

30% guazatine powder	(30 G pow.)
30%/50% guazatine/carboxin powder	(30/50 G/C pow.)
26%/25% guazatine/carboxin slurry	(26/25 G/C slur.)
20%/25% guazatine/carboxin slurry	(20/25 G/C slur.)
30%/10% guazatine/carboxin slurry	(30/10 G/C slur.)
30%/30% guazatine/maneb powder	(30/30 G/M pow.)
30%/20% guazatine/maneb powder	(30/20 G/M pow.)

The above were compared with standards of commercially available formulations of organomercury, organomercury/carboxin or carboxin/thiram seed dressings.

Rates of application in terms of weight or volume of formulation per kg seed are given in the relevant tables.

Seed and seed treatment Where possible seed carrying natural infections of the pathogens were used in the trials. Only seed from the harvest year immediately prior to that of the trial was used.

In the cases of Bunt and Oat Loose Smut it was not possible to obtain naturally infected seed; fresh spores of the pathogens were artificially applied to the seed by dusting at the rate of 3.75 g/kg seed for Tilletia caries or by vacuum impregnating oat seed with a 0.2% w/v aqueous suspension of Ustilago avenae spores (A.P.S. 1944).

Powder seed dressings were applied by mixing the dry formulation with the seed for a standard time in a glass bottle, polythene bag or metal 'butter-churn' dresser depending on the scale of the trial.

True liquid solution formulations (guazatine acetate, organomercury) were applied to a thin layer of seed contained in a shallow tray by a laboratory compressed gas-powered sprayer. The spraying operation was done quickly and the seed shaken immediately to obtain a good distribution.

Slurry formulations were applied by shaking the chemical and seed together in a large glass jar.

Trial layout Small plot randomised block arrangements were used throughout these trials. Plot size was 1.1m x 5 or 10m or 3m x 30m (specified in each table) and there were 3 or 4 replicates. The trials were drilled with an Øyjord tractor-mounted small plot drill at a sowing rate of 165 kg/ha for spring varieties and 200 kg/ha for winter varieties.

Assessments For most trials a count of emerged plants was made at GS 1-2 (Feekes scale as modified by Large, 1954). Diseased plants in oat leaf stripe trials were also counted at this stage. In Calonectria nivalis and Leptosphaeria nodorum trials at least 100 plants/plot were randomly sampled and examined for disease symptoms on the shoot base or roots.

In loose smut trials the number of diseased ears per plot was counted and for bunt a random sample of at least 100 ears was taken and the percentage disease determined.

Yields are presented only for the one large plot trial as the other small plots are intended primarily for disease incidence assessment and do not lend themselves to accurate yield analyses.

RESULTS

Data from selected trials are given below to illustrate typical levels of disease control given by guazatine mixtures; the extent to which the disease incidence has been decreased in treated plots below the level in untreated plots is given in most trials as 'percentage control'.

a) Glume Blotch (Leptosphaeria nodorum)

It is mainly the seedling stage of this disease which has been studied in our trials; if chemical treatment controls this seed-borne phase then seedling establishment will be improved and also inoculum potential from this source for the later spread of the leaf and ear phase will be decreased.

Results from one trial are given in Table 1. By means of careful sampling at an early growth stage an assessment of pre-emergence blight (PEB) could be made.

Table 1

Disease control in winter wheat (cv. Cappelle Desprez) infected with L. nodorum

Treatment	Rate per kg	Total emerged plants/m	PEB %incidence	PEB %control	Emerged diseased %incidence	Emerged diseased %control
40 G liq.	2ml	69.3	2.5	59.0	0.8	94.0
26/25 G/C slur.	3ml	64.1	2.1	65.6	0.5	96.3
30/10 G/C slur.	2ml	66.3	1.0	83.6	0.6	95.5
30/30 G/M pow.	2g	64.5	1.3	78.7	0.2	98.5
2% organomercury liq.	1ml	65.2	1.6	73.8	1.6	88.1
Untreated	-	61.5	6.1	-	13.4	-
LSD (P=5%)	-	-	-	-	2.1	-

Plot size 1.1m x 10m, Sowing date 24.11.72 Assessed GS 2-3

All treatments gave very good control of the seedling stage of this disease; the guazatine and guazatine mixtures gave a lower incidence of emerged diseased plants than did mercury.

b) Brown Foot Rot (mostly Calonectria nivalis)

This disease can be severe, depending on local soil conditions, and affects establishment and stand; it is difficult to obtain complete control of this pathogen because of its active soil-borne phase (Moore and Moore, 1961). Table 2 shows results of two trials using the same seed stock of spring wheat; 40% guazatine proved almost as effective as mercury here and the guazatine/carboxin mixture was not significantly better than guazatine alone.

Similar results were obtained from spring barley trials (Table 3); here disease incidence was higher and control by liquid organomercury less good.

Table 2

Disease control in spring wheat (cv. Kloka) infected with Calonectria nivalis at two sites A and B

Treatment	Rate per kg	% infected plants			Ears/m			Relative yield		
		A	B	Mean	A	B	Mean	A	B	Mean
40 G pow.	2g	2.0	5.3	3.6	61.7	91.2	74.6	120	110	115
30 G pow.	2g	2.0	9.8	5.9	58.4	69.5	64.0	101	106	103.5
30/50 G/C pow.	2g	3.75	4.8	4.3	56.1	91.2	73.6	90	105	97.5
1% organomercury pow.	2g	1.25	3.3	2.3	60.0	81.3	70.6	113	112	112.5
Untreated		20.0	25.3	22.6	60.3	71.8	66.0	100	100	100
								(kg/ha = 2234	2208	2221)

Plot size 3m x 30m, Sowing date 29.3.68
Assessed GS 4

Table 3

Brown root rot control in spring barley (unknown cultivar)

Treatment	Rate per kg	Plants/m	Emerged diseased plants	
			% incidence	% control
40 G liq.	2ml	42.8	15.5	82.0
20/25 G/C slur.	3ml	41.6	15.5	82.0
30/10 G/C slur.	2ml	40.1	26.2	69.0
30/30 G/M pow.	2g	41.8	14.1	83.6
2% organomercury liq.	1ml	44.3	37.3	56.6
Untreated	-	41.1	86.0	-
LSD (P = 5%)		-	20.4	-

Plot size 1.1m x 10m, Sowing date 17.4.73
Assessed GS 4-5

c) Bunt (Tilletia caries)

Bunt was formerly a very serious and widespread disease but is now adequately controlled by mercury (Moore and Moore, 1961). Data from two trials is shown in tables 4 and 5; guazatine gives slightly lower control than mercury, particularly in the face of a very high infection level. Guazatine/carboxin and guazatine/maneb mixtures were both better than guazatine alone (Table 5), the guazatine/carboxin mixtures also being better than mercury in this trial.

Table 4

Bunt control in spring wheat (cv. Kloka)

Treatment	Rate per kg	Diseased ears/plot	% control
40 G pow.	2g	10.5	95.4
1% organomercury pow.	2g	1.5	99.3
Untreated	-	228.5	-
LSD (P = 5%)		46.8	

Plot size 1.5m x 5m, Sowing date 21.3.67 Assessed 30.8.67

Table 5

Bunt control in spring wheat (cv. Kolibri)

Treatment	Rate per kg	% diseased ears	% control
40 G pow.	2g	7.6	87.9
30/50 G/C pow.	2g	0.8	98.75
30/10 G/C pow.	2g	1.6	97.4
30/30 G/M pow.	2g	3.8	93.9
1% organomercury pow.	2g	3.3	94.7
Untreated	-	62.5	-
LSD (P = 5%)	-	10.6	-

Plot size 1.1m x 5m, Sowing date 12.4.73 Assessed 7.8.73

d) Barley leaf stripe (Pyrenophora graminea)

This can be a severe disease, an infected plant yielding virtually no useful grain (Moore and Moore, 1961). Trials have been carried out on several varieties with different disease levels; Table 6 gives data from a trial with a high level and Table 7 from a trial with a lower infection level. Guazatine alone is not sufficiently active against this disease, particularly on seed stocks with infection levels of over 2%, but mixtures with carboxin and maneb were more satisfactory. The 30/30 guazatine/maneb mixture performed very well in trials on spring barley cv. Tern and was only slightly inferior to mercury (Table 6).

Table 6

Disease control in spring barley (cv. Tern) carrying a 7% infection of Pyrenophora graminea; data from two sites A and B

Treatment	Rate per kg	Diseased plants/plot			% control		
		A	B	Mean	A	B	Mean
40 G liq.	2ml	65.3	73.0	69.1	65.9	72.3	69.1
20/25 G/C slur.	3ml	18.7	30.0	24.3	90.3	89.8	90.0
30/10 G/C slur.	2ml	32.3	55.6	43.9	83.1	81.1	82.1
30/30 G/M pow.	2g	11.0	19.3	15.1	94.0	93.5	93.75
carboxin/thiram pow.	2g	18.0	28.0	23.0	90.6	90.5	90.55
2% organomercury liq.	1ml	4.0	6.6	5.3	97.9	97.8	97.85
Untreated	-	191.3	295.0	243.1	-	-	-
LSD (P = 5%)		46.5	-	-			

Plot size 1.1m x 10m, Sowing date 6.4.73(A) 17.4.73(B) Assessed GS 10-10.1

Table 7

Disease control in spring barley (cv. Sultan) carrying a 1.2% infection of P. graminea

Treatment	Rate per kg	Diseased plants/plot	% control
40 G liq.	2ml	3.3	92.4
20/25 G/C slur.	3ml	0.0	100
30/10 G/C slur.	2ml	1.3	97.0
30/30 G/M pow.	2g	0.3	99.3
carboxin/thiram pow.	2g	0.7	98.4
2% organomercury liq.	1ml	0.3	99.3
Untreated	-	44.3	-

Plot size 1.1m x 10m, Sowing date 6.4.73 Assessed GS 10

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e) Oat leaf spot (Pyrenophora avenae)

This disease is now only locally severe, mainly in Scotland, but it is interesting because of the widespread occurrence of isolates of the pathogen which are resistant to mercury dressings. Table 8 shows results from two trials carried out this year; symptoms assessed were the striping and marked spotting visible on the first leaves in the seedling stage. Guazatine alone and in mixture gave good control of the disease, although none were completely effective, they were far superior to mercury.

Table 8

Oat leaf spot control in spring oats (cv. Astor) at two sites A and B

		Emerged plants/m			% diseased plants			% control		
Treatment	Rate/kg	A	B	Mean	A	B	Mean	A	B	Mean
40 G liq.	3ml	39.9	43.2	41.5	2.2	0.9	1.55	74.1	87.3	80.7
20/25 G/C slur.	4.5ml	44.6	46.2	45.4	1.6	0.9	1.25	81.2	87.3	84.25
30/10 G/C slur.	3ml	41.6	48.9	45.25	2.7	1.7	2.2	68.2	76.1	72.15
30/30 G/M pow.	3g	41.1	44.8	42.9	2.7	1.9	2.3	68.2	73.3	70.75
carboxin/thiram pow.	3g	39.9	43.4	41.6	2.8	3.9	3.3	67.1	45.1	56.1
2% organomercury liq.	1.5ml	37.8	45.6	41.7	9.5	5.6	7.55	0	21.1	10.55
Untreated	-	36.4	45.4	40.9	8.5	7.1	7.75	-	-	-
LSD (P = 5%)		-	-	-	1.8	-	-			

Plot size 1.1m x 10m, Sowing date 6.4.73 (A) 17.4.73 (B) Assessed GS 2

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f) Oat Loose Smut (Ustilago avenae)

Table 9 shows results from two trials; guazatine was inferior to mercury which gave only moderate control.

Table 9

Loose smut control in artificially inoculated spring oats (cv. Ayr Line)

		Emerged plants/m			Diseased ears/plot			% control		
Treatment	Rate/kg	A	B	Mean	A	B	Mean	A	B	Mean
40 G liq.	3.0ml	38.6	20.7	29.6	102	57.3	79.6	39.6	42.1	40.8
2% organomercury liq.	1.5ml	38.9	25.1	32.0	97	28.5	62.7	42.6	71.2	56.9
Untreated	-	36.4	24.9	30.6	169	99.0	134	-	-	-
LSD (P = 5%)		-	-	-	55.0	49.3	-			

Plot size 1.1m x 5m, Sowing date 18.4.72 (A) 28.4.72 (B) Assessed GS 10.5

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g) Net blotch and foot rot of barley (Pyrenophora teres and Cochliobolus sativus)

Only limited work has been carried out against these diseases in the UK, but unpublished data from collaborator trials overseas indicates that guazatine gives promising control of both. Neither disease is very important in this country at present.

h) Loose smut of barley (Ustilago nuda)

Although guazatine itself shows no activity against this pathogen, trials have shown that the 30/50 G/C powder and 20/25 G/C slurry gave similar levels of control to the mercury/carboxin standard. The 30/10 G/C mixture gave less and more variable control (Table 10)

Table 10

Control of Ustilago nuda in spring barley (cv. Sultan) at two sites A and B

Treatment	Rate per kg	Mean smutted ears/plot A	B	Mean	% control A	B	Mean
30/50 G/C pow.	2g	0.7	2.7	1.7	99.4	98.7	99.0
30/10 G/C pow.	2g	10.0	122.7	66.3	90.9	39.5	65.2
1% mercury/55% carboxin pow.	2g	0	1.0	0.5	100	99.5	99.7
Untreated	-	110.3	203.0	156.6	-	-	-

Plot size 1.1m x 10m, Sowing dates 6.4.73 (A) 17.4.73 (B) Assessed GS 10.5

DISCUSSION

The results of many trials confirm that guazatine, either as a powder or liquid formulation, shows activity of the same order as organomercury dressings against L. nodorum and C. nivalis and slightly less activity against T. caries and U. avenae. In the trial shown in Table 3 the incidence of C. nivalis was higher than the 30% seed-borne infection detected by an agar test (Hewitt, P.D. pers.comm.), which would suggest that the pathogen had spread by secondary growth through the soil. This situation would perhaps explain the poor control given by mercury in this trial. The control of U. avenae by both guazatine and mercury was poor (Table 9), a factor which could be attributable to the method of inoculation (Leukel, 1937).

Guazatine is more active than mercury against P. avenae but less active against P. graminea. This situation with P. graminea has become more evident since seed stocks carrying high infection levels have been available for trials. However, as the barley cv. Sultan in Table 7 carried a 51% infection as determined by agar tests (Hewitt, P.D. pers.comm.) and was adequately controlled, it is possible that the actual depth of infection also affects the result.

These trials show, therefore, that guazatine on its own could be a suitable replacement for mercury-based dressings on wheat but not spring barley.

The guazatine/carboxin mixtures formulated to give a high carboxin loading on the seed (30/50 G/C pow. at 2g/kg or 20/25 G/C slur at 3g/kg) give good control of P. graminea and T. caries and also have a wider activity spectrum which includes U. nuda. These mixtures would be suitable replacements for organomercury/carboxin (Murganic) formulations for the stock seed trade. The 30/10 G/C mixture does not give a high enough improvement of P. graminea activity.

The 30/30 guazatine/maneb powder shows great promise against the whole range of seed-borne organisms so far tested except U. nuda, and is a candidate for the general seed market on both wheat and barley.

The crop safety of guazatine and guazatine mixtures has been well proven in laboratory germination tests (Cook, I.K., unpub. data) and in emergence assessments of the trials reported here. Large plot trials are to be carried out to provide confirmatory yield data.

Trials in Sweden (Dahlberg, S, pers.comm.) confirm the biological activity of guazatine. Farmer-scale handling trials are planned in Sweden this year with guazatine liquid, and distribution and loading data following commercial application of guazatine are at present being obtained.

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References

AMERICAN PHYTOPATHOLOGICAL SOCIETY (1944) Greenhouse method for testing dust seed treatments to control certain cereal smuts. Phytopathology, 34, 401-404

GRANHALL, I (1971) Environmental aspects of using persistent pesticides. Proceedings 6th British Insecticide and Fungicide Conference, 3, 735-740

GREENAWAY, W. and COWAN, J.W. (1970) The stability of mercury resistance in Pyrenophora avenae. Transactions of the British Mycological Society, 54, 127-138

HEWITT, P.D. (1967) Chemical treatment for cereal seed. Agriculture, 74, 555-559

LARGE, E.C. (1954) Growth stages of cereals. Plant Pathology, 3, 128-129

LEUKEL, R.W. (1936) Seed treatment experiments with oats naturally and artificially inoculated with smuts. United States Department of Agriculture Technical Bulletin 568

MOORE, W.C. and MOORE, F.J. (1961) Cereal diseases, Ministry of Agriculture and Fisheries Bulletin No. 129, 3rd Edition.

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1% mercury/55% carboxin pow.	2g	0	1.0	0.5	100	99.5	99.7
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MOORE, W.C. and MOORE, F.J. (1961) Cereal diseases, Ministry of Agriculture and Fisheries Bulletin No. 129, 3rd Edition.

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PATHWAYS OF UPTAKE AND TRANSLOCATION OF SOME SOIL-APPLIED PESTICIDES

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Summary The uptake and translocation by roots of intact barley plants of several pesticides with contrasting physico-chemical properties have been compared in solution culture. Of the seven compounds investigated, six (four triazines, diuron and ethirimol) appeared to move passively with the transpiration stream. The results suggested that whereas in their passage across roots lipophobic compounds were largely confined to the free space, lipophilic compounds could penetrate the protoplasts of the cortical cells, leading to higher rates of translocation.

Although in some circumstances movement of 2,4-dichlorophenoxyacetic acid was clearly associated with metabolism, in common with the other substances the concentration in the transpiration stream seemed to be related to the concentration of the most readily diffusible component in the roots.

There is evidence that totally different mechanisms can operate in uptake by the base of shoot systems of barley and radish, which may be important in some field situations.

Résumé On a comparé en solution nutritive l'absorption et le transport par les racines intactes de plantes d'orge, de plusieurs pesticides aux propriétés physicochimiques differents. Des sept substances qui ont été examinées, six (quatre triazines, le diuron et l'ethirimol) semblent être transportées dans les plantes passivement avec le courant de transpiration. Les resultats suggèrent que lors de leur passage à travers les racines, les substances lipophobiques étaient largement limitées dans le "free space" des racines, les substances lipophiliques pouvaient pénétrer les protoplastes des cellules corticales, resultant en une augmentation de transport.

Bien que dans certaines circonstances l'absorption de l'acide 2,4-dichlorophénoxyacétique soit evidamment liée avec le metabolisme, comme pour les autres substances, la concentration dans le courant de transpiration paraît être en rapport avec la concentration de la fraction la plus diffusible dans les racines.

L'absorption par les zones basales des pousses s'effectue par des mechanismes différents, un processus qui peut être important en agronomie.

INTRODUCTION

In this report an attempt is made to summarize some recent work on the absorption and translocation of five herbicides (2,4-D, diuron, and three triazines), a related non-phytotoxic triazine 2-hydroxy-4-ethylamino-6-isopropylamino -1,3,5-triazine (hydroxyatrazine) and the systemic fungicide ethirimol. Individual aspects of the work will be reported and discussed in greater detail elsewhere. The compounds studied contrast greatly both in their physico-chemical properties and in the extent to which they are absorbed and translocated in plants and it was the aim of the present work to establish whether any general relationships existed between these characteristics.

Many organic compounds are taken up by roots and appear to move passively with water in the transpiration stream (Minshall, 1954; Sheets, 1961; van Oorschot, 1970; Shone and Wood, 1972; Walker, 1972; Crowdy, 1972). The relationship between transport of a solute to the shoots and uptake of water is conveniently expressed as a Transpiration Stream Concentration Factor (TSCF) (Russell and Shorrocks, 1959) defined as:-

$$TSCF = \frac{\text{Quantity of solute in shoots per ml water transpired}}{\text{Quantity of solute per ml ambient solution}}$$

For many nutrient ions at concentrations comparable with those in soil solutions the TSCF may greatly exceed unity, implying the dependence of uptake on metabolism. Values less than unity are characteristic of some herbicides, and are consistent with passive movement with water in the transpiration stream. However, the present study indicates that transport of 2,4-D is different in that it is strongly influenced by metabolism.

It has been suggested that retention in the root, or impermeability of the endodermis, may restrict movement to the shoots to varying extents for different compounds (Crowdy and Rudd Jones, 1956; Crafts, 1964; Crowdy, 1972). Retention in the root is here expressed as a Root Concentration Factor (RCF) where:-

$$RCF = \frac{\text{Quantity of solute in roots per g fresh wt}}{\text{Quantity of solute per ml ambient solution}}$$

Earlier work (Collander, 1959; Crowdy, Grove and McCloskey, 1959) suggested that absorption and retention of organic molecules, and their ability to penetrate cell membranes, might be related to their partition coefficients in oil/water systems; this was accordingly investigated for the compounds considered here.

Recent work (Mercer et al, 1973) has shown that simazine can also enter, and be translocated from, the basal zones of barley shoots and hypocotyls of radish seedlings. Consideration to the relevance of these findings is given in the Discussion.

METHOD AND MATERIALS

The methods used for growing barley seedlings (Hordeum vulgare, cv. Proctor) for measuring transpiration rates and for assaying radioactivity in leaves, roots and xylem sap have been described previously (Shone and Wood, 1972). In brief, ^{14}C-labelled pesticides were incorporated in the uptake solution at a level of 0.1 or 0.2 ppm; except with 2,4-D, altering the concentration of the uptake solution over the range 0.1-2.0 ppm had no significant effect on the TSCF and little on the RCF. pH was controlled to within 0.3 units by incorporating $10^{-3}M$ potassium dihydrogen phosphate, and at pH4, $10^{-4}M$ ammonium sulphate. Thin-layer chromatography of xylem

exudate showed that for representative compounds (simazine, 2,4-D, diuron and ethirimol) there was no appreciable breakdown in the transpiration stream.

Partition coefficients were measured by shaking aqueous solutions of the pesticides with an equal volume of n-dodecane or olive oil, and counting aliquots of the oil and water phases.

Uptake of simazine or ethirimol by the basal zones of barley shoots and roots, and by radish hypocotyls, was measured concurrently with that of water in glass micropotometers.

RESULTS

Physicochemical parameters

Table 1 lists the pK's and partition coefficients in the dodecane/water and olive oil/water systems. The patterns of variation in partition coefficient in the two systems are broadly comparable; atraton and ethirimol, which protonate as the pH is lowered are less lipophilic at pH4 than at pH6. The partition coefficients for hydroxyatrazine were too low for this effect to be clearly demonstrated. On the basis of a pK for 2,4-D of 3.3, about 20% of this compound will be in the undissociated form at pH4; this is reflected in the higher values of the partition coefficient at this hydrion concentration.

Table 1

Physico-chemical properties of compounds investigated

Compound	pK	Partition Coefficient			
		n-dodecane/water		Olive oil/water	
		pH4	pH6	pH4	pH6
Atrazine	1.68[a]	2.6	2.6	44	50
Diuron	-1 to -2[b]	0.70	0.76	22	26
Simazine	1.65[a]	0.44	0.52	3.6	3.6
Atraton	4.20[a]	0.21	1.0	6.1	11
Ethirimol	4.8[c]	0.01	0.04	0.42	4.9
2,4-D	3.3[d]	0.01	<0.01	2.0	0.05
Hydroxyatrazine	5.2[e]	<0.01	<0.01	<0.05	<0.05

a, Weber (1967); b, Bailey et al (1968); c, Riley, personal communication; d, Freed (1964); e, based on value for bisisopropyl compound (Weber, 1967)

Absorption by roots

Table 2 shows results of experiments in which roots of intact plants were placed in labelled solutions at pH4 of some pesticides and tritiated water (THO) at either 20°C or 1°C. After 2 and 5 minutes the roots were removed from the solutions, blotted and counted. The quantities of compounds taken up are expressed as RCF's.

Table 2

Uptake after 2 and 5 min of pesticides and tritiated water (THO) at pH4 by barley roots at 20°C and 1°C

Compound	Temperature 20°C		1°C	
	2 min	5 min	2 min	5 min
2,4-D	1.7	3.0	0.50	0.65
Diuron	1.8	2.1	1.5	2.0
Simazine	0.75	0.95	0.65	0.77
THO	0.82	0.94	0.57	0.79
Ethirimol	0.45	0.55	0.45	0.50

There was little effect of temperature, except for 2,4-D at pH4; the very marked reduction in the rate of uptake at the lower temperature suggests that the absorption of this compound was influenced by metabolism. Results obtained at 20°C for all the other compounds listed in Table 1, at both pH4 and pH6, showed that these could be divided into two groups: those which were absorbed at a rate greater than or comparable with THO (diuron, atrazine, simazine and atraton), and those for which the rate of absorption was less (hydroxyatrazine and ethirimol). With the exception of ethirimol at pH6 in the olive oil/water system, the partition coefficients of the second group were consistently lower than those of the first (Table 1).

The RCF for THO cannot exceed unity, which will be approached when water in the cell vacuoles has equilibrated with the external medium. The results suggest that the lipophilic compounds diuron, atrazine, simazine and atraton resembled tritiated water in diffusing across cell membranes and into the vacuoles, although the observation that the RCF for these compounds rapidly exceeded unity (as shown for diuron in Table 2) was also consistent with adsorption by tissues of the root. Further evidence is given below that lipophobic compounds diffuse across the cortex largely in the free space, whereas lipophilic compounds can permeate the cortical cells.

Transport to shoots and effects of pH and calcium ion concentration

The concentration in the transpiration stream relative to that in the ambient medium (TSCF) contrasted widely between different substances. For 2,4-D at pH4 the TSCF was greater than 3, suggesting the influence of metabolism on transport of this substance to shoots; in the presence of 10^{-3}M sodium azide the TSCF was reduced to unity. For all the other substances the TSCF was less than 1, ranging from 0.09 (ethirimol) to 0.90 (simazine), which was consistent with passive movement in the transpiration stream. Table 3 shows that there was no effect of varying the pH on the TSCF of atrazine, which has a low pK, but pH and changes in calcium ion concentration did affect transport of those compounds with pK's above 4.0.

Table 3

Effects of pH and calcium chloride concentration on TSCF values

Standard errors of means are given

Compound	TSCF measured after uptake for 24 h		
	pH6.5	pH4.0	
	10^{-4}M $CaCl_2$	10^{-4}M $CaCl_2$	10^{-2}M $CaCl_2$
Atrazine	0.75 ± 0.04	0.76 ± 0.03	0.73 ± 0.01
Atraton	0.78 ± 0.02	0.47 ± 0.02	0.51 ± 0.02
Hydroxyatrazine	0.25 ± 0.03	0.56 ± 0.03	0.20 ± 0.04
Ethirimol	0.09 ± 0.02	0.18 ± 0.02	0.13 ± 0.02

For atraton, lowering the pH reduced the TSCF which however was not further decreased when the concentration of calcium was raised from 10^{-4} to 10^{-2}M. By contrast, the TSCF for hydroxyatrazine was increased on lowering the pH to 4, at which hydrogen ion concentration 10^{-2}M calcium reduced the TSCF by a factor of nearly three, presumably because of competition for negative sites in the free space or apoplast of the root. Ethirimol behaved in a similar manner to hydroxyatrazine. Earlier work (Shone et al, 1973) suggests that by contrast with some monovalent ions, calcium largely crosses the cortex in the free space rather than from cell to cell in the symplast. The observation that calcium reduced transport of hydroxyatrazine and ethirimol therefore suggested that these compounds were largely located in the free space, whereas the lack of any apparent competitive interaction between calcium and atraton was evidence that the latter compound had entered the protoplasts of the cortical cells.

Relationships between absorption by roots and transport to shoots

When detached barley roots were placed in solutions of labelled simazine, the concentration in the exudate relative to that in the ambient medium increased with time until at 24 h it approached the value of the TSCF (Shone and Wood, 1972). With 2,4-D at pH4 retention in the root was more marked; at no time over 24 hours exudation did the concentration in the exudate exceed 6% of that in the ambient medium. These results suggest that when transpiration is eliminated, retention in the root may limit transport to the shoot. However, with intact plants, increasing

the period of uptake of simazine and ethirimol from 24 to 48 h did not result in any increase of the TSCF.

In further experiments, the roots of intact plants were placed in labelled solutions of 2,4-D, diuron, simazine or ethirimol for 24 h. One group of plants was then sampled, and a second group transferred either to solutions containing no compound, or to solutions containing the compounds in the unlabelled form for a further period of 24 h. In no treatment was there any significant increase in the quantity of compound in the shoots in the second period of 24 h, although there was considerable net loss to the external solution. Thus a large proportion of the material absorbed by the roots was not readily translocated to the shoots.

Previous work (Shone et al, 1973) had shown that when roots of intact plants which had absorbed certain triazine herbicides for 24 h were eluted with water, the quantity of labelled compound lost by the roots could be assigned to three components, characterized by widely differing half-times of elution. The procedure for achieving this separation has been described in detail earlier (MacRobbie and Dainty, 1958; Dainty and Hope, 1959) and was applied to all the compounds listed in Table 1; for atratone, hydroxyatrazine and ethirimol, the pH and calcium concentration were also varied.

Fig. 1 shows that although there was no clear relationship between the TSCF and the full RCF measured after uptake for 24 h (RCF_f), there appeared to be a correlation between the TSCF and the RCF of the component which diffused most rapidly out of the roots (RCF_r) with half-times ranging from 0.25 to 1.5 min. The behaviour of diuron (not shown on the figure) was however anomalous; the value of RCF_r (1.41) was greatly in excess of the TSCF (0.81). Reasons for this are discussed later. Omitting this compound and 2,4-D at pH4, for which the TSCF and RCF_r were greater than 3 but in close numerical agreement, the correlation coefficient between the TSCF and RCF_r was $r = 0.853$, significant at $P = 0.001$.

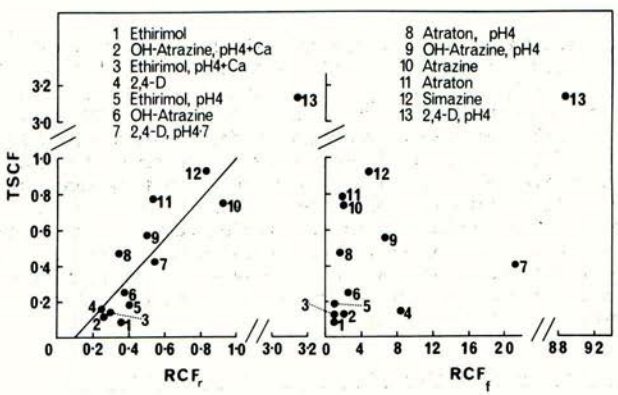

Fig. 1. Comparison of values of TSCF with RCF of readily diffusible component in roots (RCF_r) and of total amount in roots (RCF_f)

Values were measured after 24 h uptake. Uptake solution was at pH6.5 and contained 10^{-4}M calcium chloride unless otherwise stated. + Ca refers to 10^{-2}M calcium chloride.

Uptake of pesticides by the basal zones of roots and shoots

By using micropotometers to measure concurrently the uptake of simazine or ethirimol and water by the basal (proximal) 1.5 cm of young barley roots, it was found that the TSCF calculated from these measurements agreed well with values obtained when the whole root system was immersed in solution containing the pesticides. However, both simazine and ethirimol can also enter, and be translocated from, the basal region of barley shoots, provided that this is not protected by the coleoptile tissue. The quantity of simazine transported to the shoots of radish seedlings when applied to the hypocotyls was approximately the same per unit length of tissue as that transported by the roots (Mercer et al, 1973). By contrast with roots, absorption of the pesticides by basal portions of barley shoots or radish hypocotyls was not accompanied by uptake of water.

In further experiments, 1-2 μl of a solution of atrazine or simazine was quantitatively applied to radish hypocotyls; it was assumed that rapid evaporation of the water took place. Nevertheless, after a period of 4 hours, 70 to 80% of the applied herbicide was recovered in the cotyledons and young leaves. Wetting agents such as Lissapol, and differences in humidity and light intensity seemed to have little effect on the movement of the herbicide. By contrast, only about 5% of 2, 4-D applied at pH4 was translocated to the cotyledons and leaves.

DISCUSSION

In common with ions, the uptake by roots of organic molecules is characterised by an initial, rapid phase followed by a slower absorption by the tissues of the root. With the exception of 2,4-D, however, this initial process is probably largely passive for the substances considered here. The concentration attained in the root in the initial uptake varies greatly between different compounds; the contrasting behaviour of lipophilic and lipophobic compounds in their absorption by roots, in their transport to shoots and in the effects of hydrogen and calcium ions suggest that the former may diffuse into the vacuoles of the cortical cells at a rate comparable with, or greater than that of water, whereas the lipophobic compounds are largely confined to the free space of the cell walls. The possibility that the lipophilic herbicide monuron may diffuse across the protoplasts of cells has been suggested earlier by Crafts and Yamaguchi (1960).

For a solute which is at the same concentration in the cell walls of the root as that in the uptake solution, the rapidly diffusible component RCF_r can provide an estimate, in ml per g fresh weight, of the apparent volume of the free space. The lowest values obtained for RCF_r - hydroxyatrazine and ethirimol at pH4 in the presence of 10^{-2}M calcium chloride (0.26 and 0.27 respectively) and 2,4-D at pH6.5 (0.25) - are reasonably close to estimates previously made of the free space associated with the cell walls in barley roots, 0.23 and 0.17 ml/g fresh weight (Epstein, 1955; Shone, 1964 respectively). As has recently been discussed in detail by Tanton and Crowdy (1972), these values will all overestimate the true volume of the free space because of unavoidable surface contamination and this will be relatively more serious for low values of RCF_r. At pH4 in the absence of a high concentration of calcium, RCF_r is greater for both hydroxyatrazine and ethirimol, presumably reflecting reversible absorption onto negative sites, and hence a higher concentration of these substances in the free space than in the original uptake solution. For the non-polar compounds atrazine and simazine, RCF_r approaches unity, suggesting that the exodiffusion of these compounds takes place not only from the free space but also from the cells of the cortex. The data shown in Fig. 1 imply that it is this readily diffusible fraction which reaches the transpiration stream, although it is quite possible that the endodermis presents a barrier to movement of some substances. The high value of RCF_r for diuron, by comparison with its TSCF, probably arises from the extremely rapid rate at which this compound diffuses into

(Table 2) and out of roots, leading to imprecision in the resolution of the elution curve into three components. The close agreement between RCF_r and the TSCF for 2,4-D at pH4 suggests that the influence of metabolism on the absorption of this compound contrasts with that for ions. Whereas for 2,4-D a proportion of the quantity absorbed by roots, including the readily diffusible fraction which appears to reach the transpiration stream, can readily be removed from the roots by washing with water, for many nutrient ions the active uptake process seems at least partly to depend on the ability of plant membranes to restrain exodiffusion.

Laboratory studies on the uptake of pesticides by the complete root system may help towards an understanding of how these substances reach the transpiration stream from the ambient medium. Conclusions based on these studies may however be misleading when applied to some field situations. It has been shown (Mercer et al, 1973) that even after 4 months under normal rainfall, over 80% of the simazine which had been applied to the surface of the soil remained in the top 1 cm. Uptake by the basal portions of roots and shoots may therefore be important for some pesticides. Preliminary investigations suggest that this pathway of uptake and translocation is associated with mechanisms totally different from those postulated for roots.

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References

BAILEY, G.W., WHITE, J.L. and ROTHBERG, T. (1968) Adsorption of organic herbicides by montmorillonite: Role of pH and chemical character of adsorbate. Soil Science Society of America Proceedings, 32, 222-234.

COLLANDER, R. (1959) Cell membranes: Their resistance to penetration and their capacity for transport. In Plant Physiology, Ed. Steward, F.C. Vol. II Academic Press, London, 3-102.

CRAFTS, A.S. (1964) Herbicide behaviour in the plant. In The Physiology and Biochemistry of Herbicides, Ed. Audus, L.J. Academic Press, London, 75-110.

CRAFTS, A.S. and YAMAGUCHI, S. (1960) Absorption of herbicides by roots. American Journal of Botany, 47, 248-255.

CROWDY, S.H. (1972) Translocation. In Systemic Fungicides, Ed. Marsh, R.W. Longman, London, 92-115.

CROWDY, S.H., GROVE, J.R. and McCLOSKEY, P. (1959) The translocation of antibiotics in higher plants. 4. Systemic fungicidal activity and chemical structure in griseofulvin relatives. Biochemical Journal, 72, 241-249.

CROWDY, S.H. and RUDD JONES, D. (1956) The translocation of sulphonamides in higher plants. I. Uptake and translocation in broad beans. Journal of Experimental Botany, 7, 335-346.

DAINTY, J. and HOPE, A.B. (1959) Ionic relations of cells of Chara Australis. I. Ion exchange in the cell wall. Australian Journal of Biological Sciences, 12, 395-411.

EPSTEIN, E. (1955) Passive penetration and active transport of ions in plant roots. Plant Physiology (Lancaster), 30, 529-535.

FREED, V.H. (1964) Determination of herbicides and plant growth regulators. In The Physiology and Biochemistry of Herbicides, Ed. Audus, L.J. Academic Press, London, 39-71.

MacROBBIE, E.A.C. and DAINTY, J. (1958) Ion transport in Nitellopsis obtusa. Journal of General Physiology, 42, 335-353.

MERCER, E.R., SHONE, M.G.T., HILL, D. and WOOD, A.V. (1973) Agricultural Research Council Letcombe Laboratory Report 1972, 44-55.

MINSHALL, W.H. (1954) Translocation path and phase of action of 3-(4-chlorophenyl)-1:1-dimethyl urea in bean and tomato. Canadian Journal of Botany, 32, 795-798.

OORSCHOT, J.L.P. van (1970) Effect of transpiration rate of bean plants on inhibition of photosynthesis by some root applied herbicides. Weed Research, 10, 230-242.

RUSSELL, R.S. and SHORROCKS, V.M. (1959) The relationship between transpiration and the absorption of inorganic ions by intact plants. Journal of Experimental Botany, 10, 301-316.

SHEETS, T.J. (1961) Uptake and distribution of simazine by oat and cotton seedlings. Weeds, 9, 1-13.

SHONE, M.G.T. (1964) Initial uptake of silica by excised barley roots. Nature, 202, 314-315.

SHONE, M.G.T., CLARKSON, D.T., SANDERSON, J. and WOOD, A.V. (1973) A comparison of the uptake and translocation of some organic molecules and ions in higher plants. In Ion Transport in Plants, Ed. Anderson, W.P. Academic Press, London, 571-582.

SHONE, M.G.T. and WOOD, A.V. (1972) Factors affecting the absorption and translocation of simazine by barley. Journal of Experimental Botany, 23, 141-151.

TANTON, T.W. and CROWDY, S.H. (1972) Water pathways in higher plants. II. Water pathways in roots. Journal of Experimental Botany, 23, 600-618.

WALKER, A. (1972) Availability of atrazine to plants in different soils. Pesticide Science, 3, 139-148.

WEBER, J.B. (1967) Spectrophotometrically determined ionisation constants of 13 alkylamino-s-triazines and the relationships of molecular structure and basicity. Spectrochimica Acta, 23A, 458-461.

NOTES

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UPTAKE AND REDISTRIBUTION OF N-METHYL PYRIDINIUM CHLORIDE, A MODEL SYSTEMIC COMPOUND, BY WHEAT PLANTS

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Summary Radiolabelled N-methyl pyridinium chloride was applied to the second oldest leaf of wheat plants at the 3-4 leaf stage, and the plants analysed at intervals to discover how the compound was distributed. Uptake was complete by the first analysis, 4h after treatment, at which time most of the absorbed solute was located within the cells of the treated part of the leaf. During the subsequent 8 days the compound moved out of the cells and was transported away from the treated area by the phloem and xylem. The distribution of N-methyl pyridinium chloride was compared to that of the more highly phloem-mobile α amino isobutyric acid, which showed very little accumulation at the treated part of the leaf, or movement in the xylem. The way in which solutes in general are able to move in the phloem or xylem is discussed, with particular reference to the two experimental compounds.

Résumé Du chlorure de N-méthyl pyridinium marqué au carbone radio-actif a été appliqué à la deuxième des premières feuilles de plantules de blé dont le stage de croissance avait atteint 3 à 4 feuilles, et les plantules ont été analysées à des intervalles afin de découvrir comment le composé était distribué. L'absorption de la surface de la feuille était complète à la première analyse, 4 heures après le traitement et à ce moment la plus grande partie du soluté absorbé a été trouvée dans les cellules de la partie traitée de la feuille. Au cours des 8 jours qui suivirent, le composé s'est déplacé des cellules et a été transporté au delà de la zone traitée, par le phloème et le xylème. La distribution du chlorure de N-méthyl pyridinium a été comparée à celle de l'acide α amino butyrique - qui se déplace bien plus facilement dans le phloème - lequel a presenté très peu d'accumulation à l'endroit traité de la feuille, ou de mouvement dans le xylème. La manière par laquelle les solutés en général sont capables de se déplacer dans le phloème ou le xylème est examinée, en particulier avec les deux composés à l'essai.

INTRODUCTION

The technique of whole plant autoradiography has established that different phloem-mobile compounds can produce very different overall patterns of distribution within the same species (Crafts 1967). Such observations have generally been interpreted as relative movement within the apoplast which is basically the water in or on cell walls and in the xylem lumen, or symplast which consists of the membrane bounded protoplasm present in all living cells including the phloem and the cytoplasmic strands (plasmodesmata) that connect the protoplasm of adjacent cells. The concept of apoplast and symplast (Münche 1930), while providing a useful tool for the interpretation of these observations, has tended to

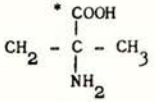
compartmentalise the two systems and to tie the solute-accumulating characteristics of all living plant cells very closely to that of the phloem. The integrity of the symplast and its supposed insulation from the apoplast has been questioned during the past five years following work in Russia by Kursanov and Brovchenko (1969, 1970) who showed that newly synthesised sugars were accumulated in the free space of the leaf and were transferred to conducting cells without prior passage through the mesophyll symplast. At the same time, Pate and Gunning (1969) were developing the concept of transfer cells, which are identified by wall ingrowths and increased area of plasmalemma and by their proposed role of facilitating interchange between the xylem and phloem. Some of our own recent electron microscope studies of the wheat leaf have supported these observations; the cell wall appeared to provide the only feasible route across the leaf, and even if symplast movement is inherently efficient the value of its contribution seems doubtful because of the low density of plasmodesmata observed between mesophyll cells. On the basis of this evidence a model system for the transport of leaf-applied solutes, is described which combines apoplastic movement to the vascular system and membrane-mediated interchange between xylem, phloem and other cells. Some of the first attempts to test this model have been made with N-methyl pyridinium chloride (MPC) which has the advantage of being accumulated by cells as well as moving in both the phloem and the xylem. Data on ∝ Aminoisobutyric acid (AIBA) has been included for comparison because in our system it is highly phloem-mobile and does not remain in the treated part of the leaf or move in the xylem.

METHOD AND MATERIALS

Wheat plants var. Kolibri were grown to the 3-4 leaf stage in controlled environment cabinets giving 85% relative humidity. Solutions containing ^{14}C-labelled N-methyl pyridinium chloride and a 0·01% C-12,4 ethylene oxide wetter were applied as five, 2 µl droplets to the surface of the second oldest leaf. The plants were maintained under the same conditions for up to 8 days, when the surface residues of the applied chemical were removed by painting and stripping with nuloidin prior to analysis of chemical distribution within the tissues. For most distribution studies the treated segment was cut out and extracted three times in 0·05% Tris buffer pH 7·2 and then frozen and thawed twice, in order to disrupt membranes, and extracted again three times. For convenience the first three extractions are taken to be the 'free space' component, and the second three the 'membrane-enclosed' component. The segment was finally combusted in a Tricarb sample oxidiser to give the radioactivity of the unextracted residues. To indicate xylem movement the area of leaf extending from the treated segment to the tip was either extracted by the above procedures or combusted intact to give total radioactivity. As a measure of phloem movement the remainder of the plant was divided into total root and total leaf and combusted. All radioactive samples were measured in a Packard 3380 scintillation counter fitted with the Absolute Activity Analyser (AAA). Autoradiographs were prepared of the whole plants to provide additional information, particularly with regard to the location of the phloem translocated solute.

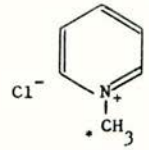
∝ amino isobutyric acid

AIBA



N-methyl pyridinium chloride

MPC



^{14}C-MPC was synthesised by quaternisation with ^{14}C-methyl iodide and converted to the chloride by ion exchange chromatography. ^{14}C-AIBA was obtained from The Radiochemical Centre, Amersham. Extracts from all parts of the plants were analysed by paper chromatography to reveal the extent of metabolic breakdown of the applied chemical. No significant breakdown was observed during the experimental period and no visual evidence of phytotoxicity was evident at concentrations higher than those used in these experiments. The results are expressed either as p moles of solute or as percentage distribution of isotope, and are derived from the mean of four replicates each containing two plants. The method is highly reproducible, the data from Table 1 were derived from experiments performed on two occasions and the medium concentration result of Table 2 agrees well with the 48h sample of Table 1, at the same concentration. The major inaccuracy lies in the amount applied to the leaf surface and this is minimised by expressing the results as the percentage of solute which has penetrated the plant.

RESULTS

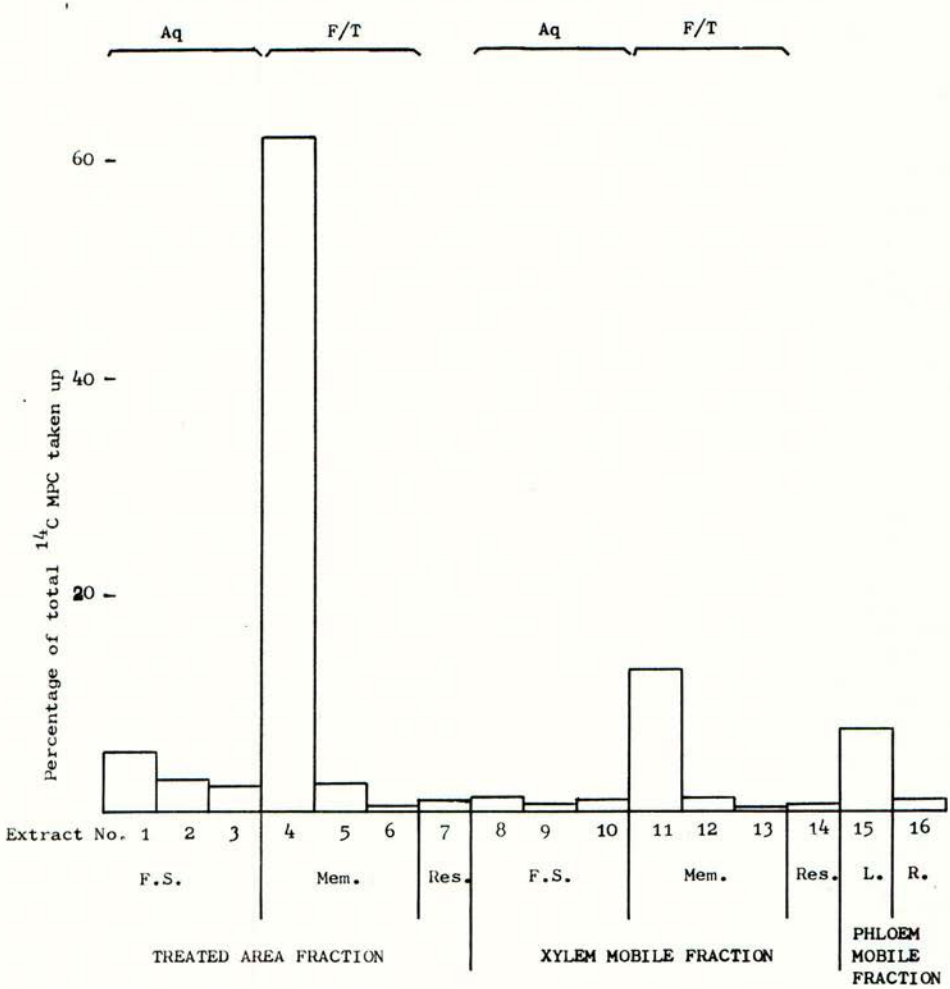
The data presented in Fig. 1 illustrate the distribution of N-methyl pyridinium chloride 4h after treatment with a 3 mM solution. Extracts 1, 2 and 3 were released from the treated area by successive aqueous extraction and were probably removed from the free space and other non-membrane-enclosed parts of the tissue but the fact that there is no major fall off at the third extraction suggests some leakage from behind the membranes. The next three extractions (4-6) which were released after the tissue had been frozen and thawed, illustrate the importance of membrane integrity for the retention of this solute. These extractions are paralleled in the xylem-mobilised fraction (extractions 8-10 and 11-13) suggesting that solute removed from the treated area in the xylem also equilibrated with cells to give identifiable free space (and free xylem) components and a membrane-enclosed (within cell) component. The phloem-mobilised fraction was found in the leaves but did not penetrate to the roots to any extent, and this characteristic was maintained over a period of several days. By 4h 88·0% of the applied solute (22·9 n moles) had penetrated the plant and there was no significant increase during a further 8 days. This means that any subsequent increase in the phloem and xylem fractions could only come about by intra-plant redistribution.

The pattern of redistribution after successive periods of time is shown in Table 1. The free space component of the treated area fell initially and then remained constant, but the membrane-enclosed component fell throughout the experimental period. The xylem-mobile fraction showed reverse behaviour, the free space component increasing initially before remaining constant while the membrane-enclosed component increased up to 4 days and then began to fall. The phloem-mobile fraction increased throughout the experimental period. If the percentage distribution or quantity of MPC in each fraction are plotted against time, on a log scale, all three fractions approximate to a straight line (Fig. 2). The slope of the xylem-mobile fraction is very similar to that of the phloem but when they are projected back the xylem cuts the x axis at 0·5h while the phloem does so at 1·5h. The treated area contains 100% of the solute at 0·5h and when projected forward it cuts the x axis at approximately 33 days. It is not certain how accurate these projections are nor to what extent they are dependent on local variables such as the physiological state of the plants.

The above data were obtained by applying an arbitrary concentration of 3·0 mM. The effect of changing the concentration is shown in Table 2. Increasing the concentration increased the percentage uptake so that at high concentration only 2·4% remained on the surface. Relative accumulation in the cells of the treated area was reduced as concentration increased, with corresponding increases in the

Fig. 1

The percentage distribution of radioactivity in wheat 4h after treatment with ^{14}C-MPC



Abbreviations used in this figure:

Ag	-	Cold aqueous buffer extraction of whole tissue segments
F/T	-	Tissue frozen and thawed then extracted with buffer solution
F.S.	-	Free space component
Mem.	-	Membrane-enclosed component
Res.	-	Residue
L	-	Leaves
R	-	Roots

Table 1

Redistribution of ^{14}C-MPC in wheat

	Time (h) 4	12	24	48	96	192
Treated area fraction						
Free space component	8·6	6·7	5·9	7·2	3·9	3·4
Membrane enclosed component	64·9	43·0	41·4	27·9	24·5	17·1
Residue	0·8	0·9	1·1	0·7	1·3	1·5
Total	74·4	50·5	48·4	35·9	29·7	22·0
Xylem-mobile fraction						
Free space component	2·8	7·6	5·9	6·7	6·3	7·7
Membrane enclosed component	14·1	22·2	23·1	27·9	38·3	32·1
Residue	0·3	0·6	0·9	0·8	1·8	2·3
Total	17·2	30·3	29·9	35·4	40·1	42·1
Phloem-mobile fraction						
Leaf	7·4	18·1	19·0	25·6	27·9	33·5
Root	1·0	1·1	2·7	3·1	2·4	*2·4
Total	8·4	19·2	21·7	28·7	30·3	35·9
Total amount in plant as % of amount applied	88	86	-	90	87	89
Amount applied to the plant (n moles)	22·9	31·2	-	26·5	29·9	29·1

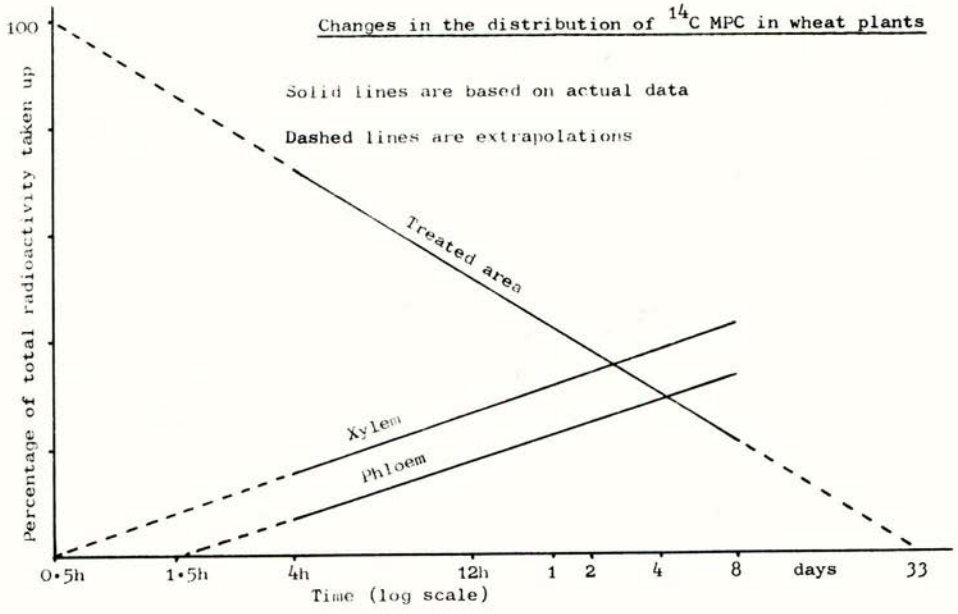
All figures except those of the two bottom lines are percentages of the ^{14}C-MPC which was taken up into the plant.

Concentration of applied MPC = 3·0 mM.

* Roots too large and contaminated for accurate analysis.

Fig. 2

Changes in the distribution of ^{14}C MPC in wheat plants



xylem and phloem fractions. The root component of the phloem fraction was generally 7-8% of the total phloem-translocated solute but at the highest concentration this increased to 20%. The data for α-amino-isobutyric acid which is included for comparison in Table 2 reveal a different pattern of distribution with very little cell retention in the treated area but considerable phloem mobility, including root accumulation.

Table 2

Influence of concentration of applied ^{14}C-MPC on its distribution in wheat after two days and comparison with AIBA distribution

	Concentration of MPC 0·22 mM	3·0 mM	35·5 mM	AIBA conc. 0·9 mM
Treated fraction				
Free space component	6·8	5·9	5·2	1·3
Membrane enclosed component	38·8	39·0	22·7	3·9
Residue	2·0	1·4	0·4	2·5
Total	47·6	37·3	28·3	7·7
Xylem mobile fraction				
Free space component	4·8	7·3	8·5	
Membrane enclosed component	25·5	28·9	26·7	
Residue	1·3	1·3	0·7	
Total	31·6	37·5	35·9	3·2
Phloem mobile fraction				
Leaf	19·1	23·0	28·7	67·0
Root	1·6	2·2	7·0	22·4
Total	20·7	25·2	35·7	89·4
Total amount in plant as % of amount applied	82	93	98	46
Amount applied to the plant (n moles)	2·3	20·9	329·4	8·5

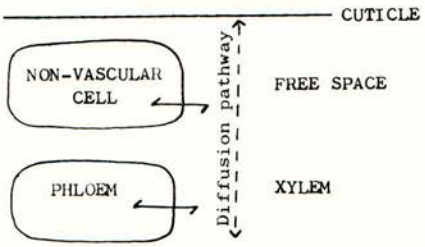
All figures except those of the two bottom lines are percentages of the ^{14}C-MPC which was taken up into the plant.

DISCUSSION

A solute which has penetrated the cuticle to enter the free space of the leaf tissue has three possible modes of distribution. It can move with the transpiration stream in the xylem, in the opposite direction in the phloem, or remain immobilised in the tissue at the point of entry. More than 80% of MPC immobilised within the treated segment of leaf is held behind membranes (the cell component of Tables 1 and 2) and much of the remainder has probably leaked into the free space from cells during the extraction procedure. This means that the treated area fraction is essentially a cell-accumulated fraction and that subsequent movement is controlled by the rate of movement out of the cells and into the mobile phases of the xylem and phloem. The xylem, phloem and cells of the treated area can be modeled as compartments into which solute will penetrate (Fig. 3).

Fig. 3

Model of solute redistribution in wheat leaves



The cell compartment is heterogenous, comprising a variety of cell types, usually aggregated into tissues and all with various inclusions such as vacuoles and membrane-bounded organelles. It is possible that the membrane characteristics here attributed to the cell are primarily those of the vacuole, but so far there is no information regarding the cytological location of MPC. Solute within this compartment is not mobilised or distributed through the plant. The phloem compartment is also heterogeneous and includes the companion cells and possibly the phloem parenchyma as well as the sieve tubes, and is characterised by its ability to accumulate sugars against a concentration gradient and to distribute solutes against the transpiration flow. Although the phloem parenchyma and companion cells are closely associated with the phloem and may be involved in the selection and accumulation of solutes prior to transport in the sieve tubes, they have no major translocating function of their own and may retain MPC in the same way that other leaf cells do. The phloem tissue exists in close proximity to the xylem and at the end of a diffusion gradient extending through the free space to the cuticle. The xylem of the wheat leaf has no membrane around it and can be regarded as an extension of free space with internal diffusion controlled by the structure of the surrounding cell walls. Any solute which enters the free space is potentially capable of systemic movement in the xylem.

Initially the three compartments are not equally accessible to a surface-applied solute because it must pass through the non-vascular tissue of the treated area to reach the phloem and xylem. At the first analysis of ^{14}C-MPC, four hours after treatment, 74·4% was retained in the treated area, including 64·9% within the cells (Table 1) but during the following 8 days this falls to 22% (17·1% in the cells). The rate of efflux from the non-vascular tissue is at least 67 times slower than the net influx which achieved the 74·4% figure from zero within four hours. Another comparison of flux rates is derived from the extrapolations in Fig. 2 which suggest that influx may have been complete within 0·5h and efflux potentially complete by 33 days, a ratio of influx to efflux of 1600:1. There are too many unknown factors such as the rate of trans-cuticular flux, the concentration adjacent to the membrane and the rate of diffusion past the membrane, to permit any calculation of the membrane permeability coefficient, but the fact that the percentage retained in the cell after two days is reduced by increasing the concentration, indicates that forces other than simple diffusion are involved. The phloem has long been known to accumulate sugars against a concentration gradient and has recently been shown to similarly accumulate herbicides such as paraquat and 2,4-dichlorophenoxyacetic acid, and plant growth regulators such as maleic hydrazide. (Peel 1972). Tobacco leaf cells have also been shown to accumulate a number of organic compounds including MPC (Morrod 1973). MPC is capable of entering and leaving cells at a rate dependent on the relative concentration inside and outside the membrane and a similar situation exists in the

phloem, with assimilates entering in regions of high concentration and leaving at 'sinks' where a low external concentration is maintained by metabolism. This means that there must be a ratio of external to internal concentration at which no net flux across the membrane will occur, the equilibrium distribution ratio. Deviation from this equilibrium ratio will result in a net influx or efflux. The cells of the treated area never achieve a stable situation because the free space containing the external component is in contact with the xylem and phloem systems which are removing solute away from the immediate vicinity of the cells. Solute removed in the xylem is brought into contact with cells in the terminal part of the leaf where the distribution ratio favours uptake, at least until the cells at the treated part of the leaf become depleted. It is possible that movement from xylem to phloem is important at the tip of the leaf where the concentration in the phloem is relatively low.

FREE SPACE → CELLS OF TREATED AREA → XYLEM → CELL OF LEAF TIP → PHLOEM ?

The xylem and phloem are situated close to each other and so it is surprising that there should be the 1h delay between mobilisation in the xylem and mobilisation in the phloem (Fig. 2). The explanation may lie in the multicellular structure of phloem, with the companion cells and parenchyma coming to equilibrium with the solute before its symplastic transfer to the sieve tubes. This non-mobile phloem fraction would be analysed as part of the cell component of the treated area fraction. Changes in the mobilisation of solute in the phloem relative to the xylem are shown by the phloem affinity ratio, defined as:

$$\frac{\text{Solute translocated in the phloem}}{\text{Solute translocated in the phloem + xylem}}$$

. At 4h the ratio is 0·33 but by 24h it has increased to over 0·4, reaching 0·5 by 8 days. Increasing the concentration of MPC also increased the ratio from 0·37 at low concentration to 0·40 at medium, to 0·50 at high concentration after two days (figures derived from Tables 1 and 2), but it never reached the 0·97 characteristic of AIBA. It is likely that at high concentration and after long time periods, the change in phloem affinity is influenced by movement from the xylem to the phloem at the tip of the leaf. The phloem affinity ratio also reflects some physiological aspects of the leaf, for instance, increasing the humidity to over 95% can give a value of 0·73 (unpublished data) presumably by reducing the rate of removal by the xylem and so increasing the concentration near the phloem. It is possible that all the additional solute entering the phloem between 4h and 8 days was derived from the membrane-enclosed component of the xylem-mobile and treated area fractions, but AIBA is able to enter the phloem without accumulating in cells of either fraction to the same extent as MPC suggesting that the mesophyll symplast is not an essential requirement for phloem loading and translocation.

The three-compartment model is obviously a considerable simplication but most of the complexities lie within the compartments and are important only when they influence compartmental capacity, or permeability. Equilibrium between the compartments is prevented by the constant removal of solute by the xylem and phloem which over a period of days can lead to the removal of compounds whose equilibrium distribution ratio is strongly in favour of cell retention. When mass flow is more rapid than equilibration local solute concentration will occur in xylem and phloem, leading to simultaneous net influx and efflux at different parts of the same compartment. For example MPC enters the xylem at the treated part of the leaf but is leaving it near the tip and entering the cells and possibly the phloem. This may result in the same compound travelling in either system depending on the direction and rate of movement; AIBA has been reported to move from the roots to leaves in the xylem (Joy,1962) but also clearly moves from the leaves to the roots via the phloem. The mode of distribution and ultimate location of any solute including pesticides will be determined by its route into the plant, its phloem membrane permeability and its affinity for the various binding and retention sites with which it comes into contact.

Acknowledgements

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References

Crafts, A.S. (1967) Ann.New York Acad. Sci. 144 : 357-36.

Kursanov, A.L., Brovchenko, M.I. (1969) Soviet Plant Physiology 16 : 801-807.

Kursanov, A.L., Brovchenko, M.I. (1970) Can. J. Bot. 48 : 1243-1250.

Joy, K.W. (1962) Nature 195 : 618-619.

Morrod, R.S. (1973) J. Exp. Bot. (in press).

Münche, E. (1930) Die Stoffbewegungen in der Pflanze. Jena : Gustav Fisher.

Pate, J.S., Gunning, B.E.S. (1972) Ann. Rev. Plant Physiol. 23 : 173-196.

Peel, A.J. (1972) Pestic. Sci. 3 : 631-641.

NOTES

Proceedings 7th British Insecticide and Fungicide Conference (1973)

EFFECTS OF APHIDS ON THE DISTRIBUTION OF PESTICIDE MOLECULES AND INORGANIC IONS IN BEAN PLANTS

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Summary Aphids were caged on foliage at one node on young broad bean plants, and radio-labelled solutes were added to the nutrient solution or applied to non-infested leaves.

When ^{32}P-phosphate was added to the nutrient solution, aphid colonies caged 1-3 days before this treatment removed more radio-label than aphids caged 1 day after. ^{35}S-sulphate was affected similarly, both in cut and rooted plants. The amounts of labelled phosphate or sulphate removed from the plants by an established aphid sink were greater than those present in the leaf at the feeding site.

Solvent-soluble radio-label derived from ^{14}C-phorate and ^{14}C-ethirimol added to the nutrient solution was less strongly attracted to established aphid sinks, and the aphids did not significantly increase movement of phorate applied to an adjacent leaflet. Water soluble-phorate metabolites, however were strongly attracted to and removed from the feeding sites by the aphids.

Résumé Les pucerons ont été encagés sur le feuillage de jeunes plantes de fève au niveau d'un noeud et on a, ou ajouté des isotopes radioactifs marqués aux solutes du milieu nutritif, ou on les a appliqués aux feuilles non infectées.

Si on ajoutait du phosphate 32-P, au milieu nutritif, les colonies de pucerons encagés un jour plus tard. Le sulfate 35-S a été affecte de la même monière dans les plantes coupées comme dans les plantes enracinées. Les quantités de phosphate ou de sulfate retirées des plantes par le système d'écoulement d'un puceron établi étaient plus élevées que celles présentes dans la feuille à l endroit où il se nourrissait.

Un isotope marqué soluble dans le solvant, derivé du phorate 14-C et de l'ethirimol -14C ajouté au milieu nutritif était moins fortement attiré par le systeme d'écoulement du puceron, et les pucerons n'augmentaient pas de façon significative le mouvement du phorate appliqué au foliole adjacent. Cependant les métabolites aquasolubles du phorate, etaient fortement attirés et evacués des endroits où les pucerons se nourrissaient.

INTRODUCTION

All plant systemic insecticides in general use move predominantly in the xylem and tend to accumulate in areas of the plant where evaporative water loss occurs. Crisp (1972) applying the Munch concept of plant transport mechanism, has concluded that the pattern of movement of insecticides is predominantly apoplastic and that unless an acidic group is attached to the molecule these compounds are normally unable to penetrate and to be transported by the symplast. Although the symplast/apoplast boundary appears to be a considerable barrier to these compounds, aphids feeding predominantly in the phloem pick up significant amounts of toxicant. Thus a degree of leakage must occur between apoplast and symplast. This leak may be induced or accentuated by the nutrient sink developed by feeding aphids; in the transpiration process only water loss occurs, whereas at an aphid sink both water and solutes are taken from the plant. This loss of sap is replaced in an individual sieve element by longitudinal movement down the pierced element and by lateral movement from surrounding elements. The length of the pierced element affected in this manner has been termed the contributory length and may extend for a considerable distance on either side of the point of stylet penetration (Weatherley et al., 1958; Peel and Weatherley, 1962; Ford and Peel, 1966). Larger aphid colonies increase the contributory length accordingly (Peel and Ho, 1970).

Sap lost from the aphid-infested area must be replaced from neighbouring regions The water may be obtained by an increased flow in the ascending apoplast, but a large proportion of the osmotica must be synthesised by the plant, though storage material may be used temporarily. In this context an aphid infestation on a brussels sprout plant leaf has been shown to increase assimilation rate in adjacent leaves (Way and Cammell, 1970).

Thus the sink may affect both symplastic and apoplastic compounds to a variable extent depending on the magnitude of the sink in relation to the size and physiological state of the plant.

This hypothesis, coupled with an observation that aphids already present on the plant appeared to be more rapidly affected by a root application of a systemic insecticide than aphids caged subsequently, prompted the investigations described in this report.

METHODS AND MATERIALS

Plants

Broad bean plants (Vicia faba L., var Sutton) were pot-grown in a 3: 1 peat: sand mix in a glasshouse. Plants with at least three fully expanded leaves were used in the experiments and were either cut just above the cotyledons or taken complete from the pots. The compost was was carefully removed from the roots under running water. Both cut shoots and rooted plants were maintained in a nutrient solution (Long Ashton solution, trace elements omitted) in 100 mm x 36 mm specimen tubes. The experiments were carried out in a constant temperature room at $20^{\circ}C \pm 1^{\circ}C$ with a 16 h daylength provided by four 80 W fluorescent lamps mounted 60 cm above the bench. The relative humidity was not controlled and fluctuated from about 60% RH during the daylight period to about 85% RH at night.

Cut plants were treated with ^{32}P-phosphate and ^{35}S-sulphate by standing the shoot in about 0.5 ml of nutrient solution to which the radio-isotope had been added. This volume was absorbed in 1-3h after which a further 0.5 ml of 'cold' nutrient solution was added. When this too was absorbed the nutrient solution

was restored to its original volume (50 ml) and maintained at that level for the remainder of the experiment.

This technique could not be followed with the rooted plants and in these experiments the radiolabelled compounds were added directly to the nutrient solution surrounding the roots.

Aphids

Black bean aphids (Aphis fabae L.) were used to produce the aphid sinks. The aphids were cultured on bean plants brought into the constant temperature room at the two-leaf stage.

For the sulphate and phosphate experiments 50-60 fourth instar and adult aphids were selected and when caging times exceeded two days, young aphids were removed periodically to prevent a greater number of aphids producing a correspondingly greater sink.

In the pesticide experiments about 20 alatae were caged on one leaf and allowed to produce young for 24 h after which time the adults were removed and the larvae reduced to about 200. The cage was retained for the phorate metabolite experiments but not for the other experiments. Aphids seldom leave a leaf except when overcrowded. The pesticides were applied five days later, before adults had begun to appear in the colonies, and the experiments terminated for analysis after a further four days.

Honeydew collection

In the phosphate, sulphate and phorate metabolite experiments, honeydew was collected on untreated filter-paper linings to the aphid cages (Galley, 1974). In the other two experiments, with labelled phorate or ethirimol, the honeydew was collected on pieces of aluminium foil 70 mm x 50 mm supported under infested leaves.

Radio-isotopes

^{35}S-sulphate and ^{32}P-phosphate were obtained from the Radiochemical Centre, Amersham, the phosphate in phosphate buffer (initially 1mCi/mg P) and the sulphate carrier free in isotonic saline(5mCi/µgS).

The ^{14}C-phorate was labelled in the ethoxy-group with a specific activity of 42.5 µCi/mg and the ^{14}C-ethirimol was ring-labelled with a specific activity of 22 µCi/mg.

Isotope extraction and counting

Autoradiographs of the leaves at the end of these experiments showed that the distribution of radio-activity in the leaf following the ^{32}P-phosphate and ^{35}S-sulphate treatments was relatively uniform. The ^{14}C-pesticide distribution showed a pattern more typical of apoplastic movement with accumulation of label beginning to occur round the leaf margins.

The concentrations of label in the phosphate and sulphate experiments were determined by punching 10 mm diameter disks from the centre of each leaflet coincident also with the caged area in infested leaves, and counting these for 10,000 counts with a Geiger-Muller detector and Timer/Scaler. Disks were stored at $0^{o}C$ in polythene film when long counting times caused delay between samples.

In order to correct the counting efficiencies of the disks to that of the aphid and honeydew counts some disks were also ground in a glass pestle and mortar with 0.5 g sand and 5 ml water containing 0.05% w/v laboratory detergent and 1mM of both sulphate and phosphate ions (sodium salts). The homogenate was filtered and washed with a further 2 x 2.5 ml of the water solution and a 1 ml aliquot of the filtrate was counted in 10 ml Bray's scintillant.

Aphids were homogenized in an all glass hand-homogenizer with a little of the sulphate, phosphate, detergent solution. The homogenate was made up to a known volume and an aliquot counted in Bray's scintillant.

The honeydew was extraced from the filter papers by shaking them in vials with 2 x 2 ml of the same aqueous solution followed by washing down with a further 1 ml. A 2 ml aliquot was counted in Bray's scintillant. These procedures gave better than 95% recoveries from samples to which known amounts of the isotopes had been added.

For ^{14}C extractions, whole leaves were ground as described above but the solvent used was chloroform: methanol (9:1). Leaves were ground and washed with 3 x 10 ml aliquots of solvent into a conical flask containing 1.5 g finely divided activated charcoal and 2 g anhydrous sodium sulphate. After swirling for two to three minutes, the solvent was filtered through a 5 cm buchner funnel and evaporated almost to dryness in a rotary evaporator. The residue was transferred to a counting vial with three aliquots of toluene totalling 10 ml and containing 8 g/l butyl-PBD. In the phorate metabolite experiments this residue was taken up in benzene and transferred to the top of a silicic acid column 8 mm internal diameter and containing 6 g silicic acid ('Silicar', Malinkrodt CC7). Phorate, its sulphone and sulphoxide were eluted with 40 ml benzene with 5% v/v acetone and 40 ml acetone respectively. This procedure was a modification of the method developed by Bowman et al. (1969) for phorate and metabolites. The column effluents were concentrated nearly to dryness and transferred to counting vials as before. The water soluble fraction was extracted with distilled water from the charcoal/ homogenate residues and an aliquot from a know volume counted in Bray's scintillant.

Aphids were extracted similarly by using the hand homogenizer with the chloroform/methanol solvent system.

Aluminium foils with honeydew samples were carefully rolled into a cylinder and inserted into 10 cm B24 stoppered test tubes. About 5 ml of methanol: water (3:1) were added and the tubes rolled for thirty min. The foils were carefully withdrawn and washed down with 1 ml methanol into the tube. This extract was shaken in the tube with 3 x 5 ml of the chloroform/methanol solvent and the chloroform fractions concentrated in the rotary evaporator and transferred to counting vials with the toluene-based scintillant.

Recoveries from spiked samples with these procedures were good with the exception of ^{14}C-phorate in honeydew. Counts were lost from 'honeydew' samples of ^{14}C-phorate dissolved in a 10% sucrose / 50% aqueous acetone 'honeydew' applied to foils and filter papers, when the samples were dried. This loss probably arose from evaporation of the pesticide during the drying process and may extend also to the toxic metabolutes. Though up to 50% of the phorate and possibly less of the sulphoxide and sulphone may have been lost from the honeydew samples, the water soluble fraction would not be so affected. Similarly, ethirimol has a low vapour pressure, and breakdown is less rapid in the plant; thus the organic solvent fraction would have removed the greater proportion of radio-label at the end of these relatively short experiments.

RESULTS

Phosphate and sulphate in cut plants

The amounts of radio-activity present in the leaves varied considerably between individual plants though the same trends were found among the replicates for each set of treatment conditions. The results are expressed as means of the proportions found in each of 3 plants. Fig.1 shows the distribution of ^{32}P at each leaf with aphids caged at the third leaf for various times before and after the application of 2μCi of ^{32}P-phosphate to the cut shoot. The levels of activity are expressed as a percentage of the total counts per min per cm^2 with aphid and honeydew counts related to the caged area (1.95 cm^2). It can be seen from Fig. 1 that the proportions of isotope removed by the aphids were greatest when the aphid colony was established before isotope application, the greater effect occurring with the longer established colony.

A similar effect was found when 5μCi ^{35}S-sulphate was added to the nutrient, though in this case the distribution pattern in the plant was markedly different with more sulphate accumulating in the younger leaves. (Fig. 2a) When an established aphid colony was present on the third leaf the proportion of label entering the younger leaves was reduced.

Sulphate in rooted plants

5μCi ^{35}S-sulphate added to rooted plants showed a distribution pattern similar to that in the cut plants and aphids caged four days beforehand again attracted more label than those caged on the plant after the isotope application (Fig. 2b)

Autoradiographs were prepared of the leaves on which the aphid colonies were sited from both cut and rooted plants. The position of the cage site was never clearly marked with either isotope, indicating that the radio-label was being transported into the region at the same rate at which it was removed by the aphids. Experiments were also conducted to detect if there was any overshoot of labelled material into the caged area once the aphids were removed. Leaves autoradiographed thirty min after aphid removal occasionally showed slight evidence of the site of the aphid cage but with the general variability of the plant material the results were inconclusive.

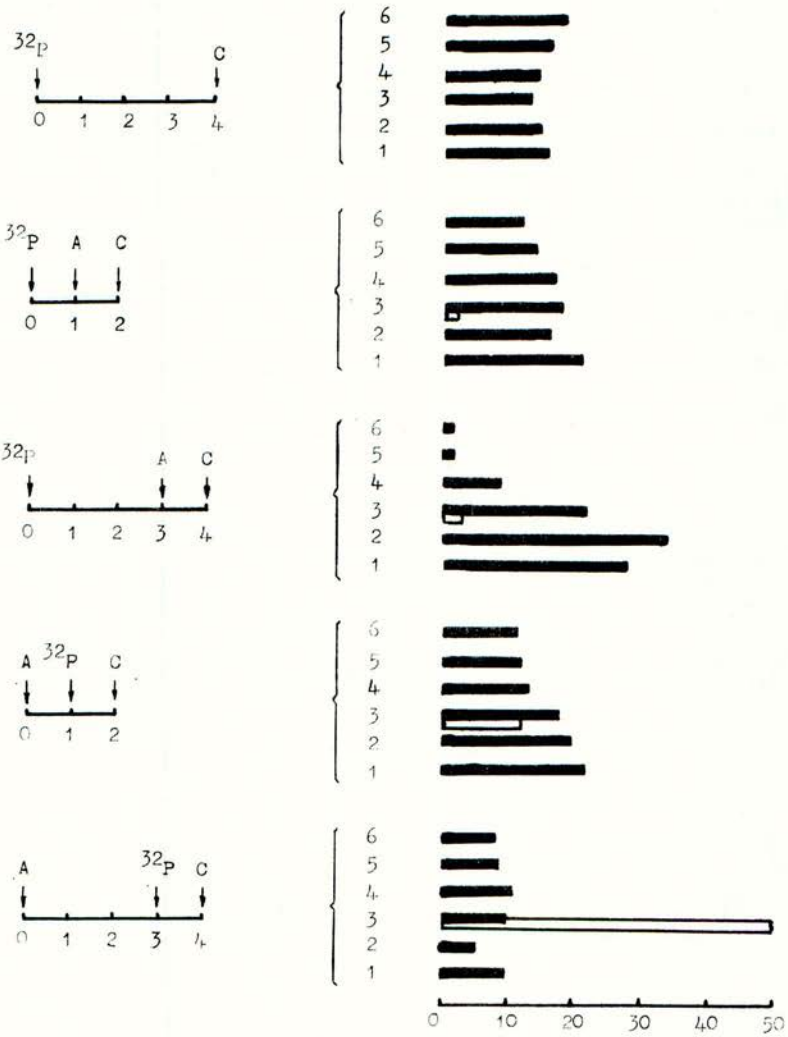
Ethirimol in rooted plants

The radio-activity found in the leaves, aphids and honeydew four days after adding 64 μg ^{14}C-ethirimol to the nutrient of each of two plants are given in Table 1. The control plants with no aphid colonies are included for comparison. Distribution in the plants tended to be of the sulphate pattern with the younger leaves receiving more label and, as in the phosphate and sulphate experiments, the aphids removed a proportion of the radio-activity from the leaflet.

Phorate in rooted plants

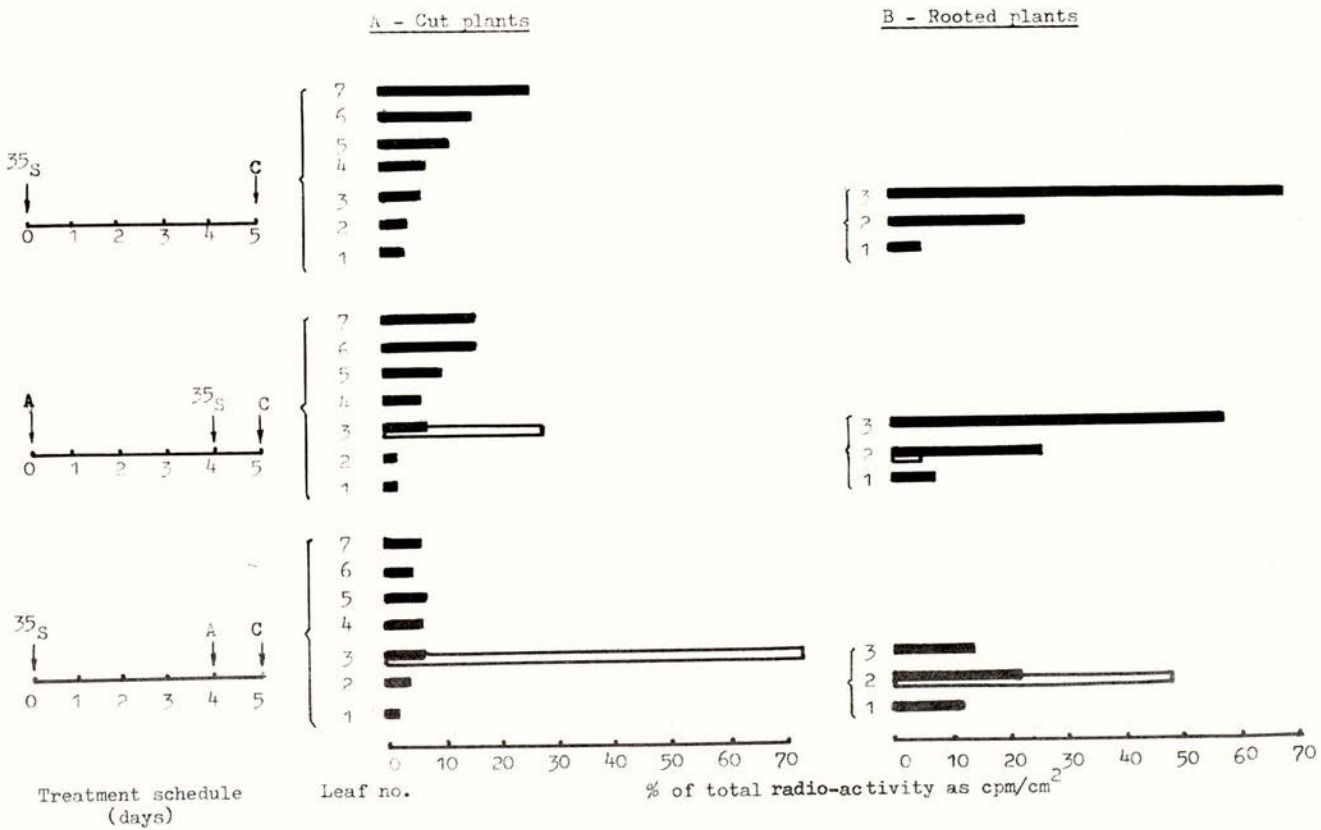
The distribution of label following the application of a sublethal quantity (8.8 μg per plant) of ^{14}C-phorate to the nutrient solution, 5 days after infestation, is shown in Table 2. A similar trend was found in this experiment but the aphids, again on the second leaf took up a little less from the infested leaf (1/20 and 1/10 of the amount of phorate present) compared with ½ and 1/6 for ethirimol. The reduction is probably attributable to the response of the aphids to the insecticide, even though the dose was sub-lethal for the duration of the experiment. Two days

Fig 1. Distribution of ^{32}P in cut plants and in an aphid sink on the third leaf



^{32}P = addition to nutrient solution; A = aphids caged on plant; C = radio-activity counts; 1 = oldest, 6 = youngest leaves: open areas on histograms are combined aphid and honeydew counts.

Fig 2. Distribution of ^{35}S in the plants and the aphid sink



Explanations given under Fig 1.

after application, the feeding rate fell and many individuals left the leaf in search of more suitable feeding sites. These aphids were taken from the plant and kept at -20°C until homogenized with the remaining insects. In both root uptake experiments the amounts of phorate and ethirimol taken up varied considerably from plant to plant. The quantities found in the aphids and honeydew did not contribute significantly to the amounts present in the infested leaves nor was there any discernible effect on the general pattern of distribution.

Table 1

Radio-label distribution following the addition of ^{14}C-ethirimol to the mutrient solution

	Leaf no.	Leaves of control plant	Leaves of infested plant	Aphids	Honeydew
Replicate A	3	430	686	-	-
	2	310	376	43.2	130
	1	208	251	-	-
Replicate B	3	464	170	-	-
	2	379	128	5.7±3%	14.3±2%
	1	395	85	-	-

Values given are ng equivalents of ethirimol on both leaflets. 2σ counting errors were better than ±1% except where indicated.

Table 2

Solvent-soluble radio-label distribution following the application of ^{14}C-phorate to the nutrient solution

	Leaf no.	Leaves of uninfested plant	Leaves of infested plant	Aphids	Honeydew
Replicate A	4	110	26.0	-	-
	3	94	115	-	-
	2	82	67.6	1.3±4%	2.5±3%
	1	95	97.7	-	-
Replicate B	4	108	43.4	-	-
	3	150	51.6	-	-
	2	121	46.6	1.8±4%	3.1±3%
	1	100	64.8	-	-

Values are ng equivalents of phorate. Counting errors as in Table 1.

Phorate applied to an adjacent leaflet

A sub-lethal dose (3.5 µg) of ^{14}C-phorate was applied in 5x5 µl droplets of acetone to the axial surface of one leaflet of the second leaf. Table 3 gives the relative quantities of label as phorate, phorate sulphoxide and phorate sulphone found six days after application. Though toxic compounds did move from leaflet to leaflet, what movement there was seems to have been depressed by an aphid sink established on an adjacent leaflet 5 days before application of phorate.

Table 3

Distribution of radioactive phorate and toxic metabolites in treated and adjacent leaflets

Plant	Leaflet	Phorate	Phorate sulphoxide	Phorate sulphone
Control A	treated	129	144	200
	adjacent aphid-free	3.1±3%	4.2±3%	2.4±3%
Control B	treated	176	140	504
	adjacent aphid-free	2.8±3%	1.7±4%	1.6±4%
Infested A	treated	131	146	235
	adjacent aphid-infested	1.2±4%	0.45±7%	0.34±8%
	aphids	0.17±11%	0.15±12%	0.14±12%
	honeydew	0.32±8%	2.2±3%	0.99±5%
Infested B	treated	217	117	123
	aphid-infested	0.52±7%	0.55±6%	0.46±7%
	aphids	0.50±7%	0.36±8%	0.31±9%
	honeydew	0.20±10%	0.40±8%	1.1±5%

Units and counting errors as in Table 2.

Table 4

Distribution of radioactive phorate and metabolites in the third leaf following application of 7µg ^{14}C-phorate to the first leaf

	Phorate	Phorate sulphoxide	Phorate sulphone	Water soluble fraction
Leaf	1.1±0.9	1.1±0.7	0.78±0.6	2.8±1.1
Aphids	1.5±0.4	1.2±1.1	1.2±1.1	10.4±3.2
Honeydew	0.15±0.1	1.2±0.9	0.72±0.7	41±21

Values given are mean ng equivalents of phorate with standard deviations.

Phorate applied to a lower leaf

In this experiment 200 aphids were reared on each leaflet of the third leaf and 3.5 µg ^{14}C-phorate applied in acetone to each leaflet of the first (oldest) leaf. Table 4 gives the activity appearing in the different metabolite fractions present in the third leaf and in the aphid sink after 4 days. The figures are the means from three plants, all of which showed the same general pattern but differed considerably in the levels of activity present. The most striking aspect of this experiment is the amount of radioactivity found in the water-soluble fraction from the honeydew and to a lesser extent in the aphids, very much more than in the leaf from which it originated. The water soluble phorate metabolites were not further identified but since the compound was labelled in the ethoxy-group the fraction probably contained a mixture of diethyl phosphoric acid and analogues.

DISCUSSION

The phosphate and sulphate experiments showed that the aphid sink had a greater effect on these isotopes ascending the plant than when they were distributed in the leaves. Some of this effect could perhaps be attributed to a slower feeding rate at the beginning of the caging period. It takes 12-16 h

for A.fabae to achieve a normal feeding rate (Banks and Nixon, 1959). The aphids caged 1 day before the isotope application also removed less from the plant than those caged 3 days beforehand, indicating that even when the aphids were feeding at their optimum rate at the time of the isotope application the full sink effect was not established until sometime later.

A factor which may apparently enhance movement to the sink from the ascending stream is the additional transpiration caused by the increased assimilation rate that would be expected from the observations of Way and Cammell (1971). This transpiration flow will bring with it a proportion of label which, combined with other solutes, would be used by the plant to contribute to the osmotic potential in the sink system.

The distribution of radio-label in the leaves of the two plants given the root application of phorate was atypical. Generally a pattern more like that found in the ethirimol treatment might be expected with higher concentrations of insecticide in the younger leaves. However this experiment did demonstrate the appearance of compounds which were solvent-soluble and therefore probably toxic in the aphid honeydew, and this was confirmed in the subsequent experiments by the separation of phorate and its sulphoxide and sulphone. The separation technique used did not distinguish the phorate metabolites from phoratoxon and its sulphoxide and sulphone, but preliminary studies using TLC techniques failed to show any appreciable quantities of these compounds in the plants. Any traces that were present would also be eluted from the column with the phorate metabolites.

The movement from xylem to phloem may occur over a considerable distance from a large aphid colony since the contributory length for an individual aphid extends for many centimetres along the pierced element on either side of the aphid stylets (Weatherley et al, 1959). When very many of the sieve elements are affected it is likely that this effect is extended further both longitudinally and laterally. Sulphate and phosphate readily move laterally in phloem tissue, more readily even than water (Peel, 1970) though in general lateral movement in vascular tissue is poor (Canny, 1971). Pesticide molecules are larger and less readily transported, except in a passive manner, and appeared from these experiments to be less affected by the sink than the mobile ions. Water soluble phorate metabolites however were drawn by the sink and the amounts present there suggest that it may have entered the leaf in that form, perhaps in the transpiration stream, rather than have been produced there. Some hydrolysis would be expected in the sink leaf and in the aphids and honeydew while water was present, but the export of toxic compounds from treated leaves was low and the levels of these compounds in untreated leaves were also low. Thus it seems likely that much of the water soluble fraction in the sink may have been formed in the treated leaf and then exported to appear in the sink as did the ^{32}P-orthophosphate in the earlier experiment.

It may be concluded that an established aphid colony had a negligable influence on the movement of pesticide molecules in the bean plants but had a much greater effect on smaller anionic species. The attraction to the feeding site was greater when the anions were ascending the plant than once they were dispersed in the leaf.

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References

BANKS, C.J. and NIXON, H.L. (1959) The feeding and excretion rates of Aphis fabae Scop. on Vicia faba, L. Entomologia Experimentalis et Applicata, 2, 77-81.

BOWMAN, M.C., BEROZA, M., and HARDING, J.A. (1969) Determination of phorate and five of its metabolites in corn, Journal of Agricultural and Food Chemistry 17, 138-142.

CANNY, M.J. (1971) Translocation: Mechanisms and Kinetics. Annual Review of Plant Physiology, 22, 237-260.

CRISP, C.E. (1972) The molecular design of systemic insecticides and organic functional groups in translocation. Proceedings of the Second International IUPAC Congress of Pesticide Chemistry, Vol. 1 (Insecticides) 211-264.

FORD, J. and PEEL, A.J. (1966) The contributory length of sieve tubes in isolated segments of willow, and the effect on it of low temperatures. Journal of Experimental Botany, 17, 522-533.

GALLEY, D.J. (1974) Relative feeding rates of B.brassicae and M.persicae on brussels sprout plants in relation to their susceptibility to systemic insecticides. Annals of Applied Biology, 76, (in the press).

PEEL, A.J. (1970) Further evidence for the relative immobility of water in sieve tubes of willow. Physiologia Plantarum, 23, 667-672.

PEEL, A.J. and HO, L.C. (1970) Colony size of Tuberolachnus salignus (Gmelin) in relation to mass transport of ^{14}C-labelled assimilates from the leaves of willow. Physiologia Plantarum, 23, 1033-1038.

PEEL, A.J. and WEATHERLEY, P.E. (1962) Studies in sieve tube exudation through aphid mouthparts: The effects of light and girdling. Annals of Botany (N S), 26, 633-646.

WAY, M.J. and CAMMELL, M. (1970) Aggregation behaviour in relation to food utilization by aphids. In 'Animal Populations in Relation to their Food Resources', Ed. Watson, A., British Ecological Society Symposium No. 10, 229-247.

WEATHERLEY, P.E., PEEL, A.J. and HILL, G.P. (1959) The Physiology of the sieve tube. Preliminary experiments using aphid mouthparts. Journal of Experimental Botany, 10, 1-16.

NOTES

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UPTAKE OF RADIOACTIVE PHORATE BY MAIZE IN RELATION TO GRANULE PLACEMENT AND TO CONTROL OF FRIT FLY

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Summary In order to account for differences in frit fly control in maize after the use of different methods of application of insecticidal granules, trials were carried out in the field and glasshouse on the uptake of ^{32}P- phorate from impregnated fuller's earth granules applied in different ways to silage maize. Control of frit fly was greater after a broadcast top dressing over plants than after a furrow treatment under the seeds or after a soil surface dressing at the side of plants. Counts of radioactivity of plants from the trials showed levels of uptake up to 17 ppmw fresh weight of maize for a limited period after the broadcast treatment, which coincided with the peak frit attack in June. Furrow treatment gave less uptake, up to 12.8 ppmw, but levels were maintained over a longer period and activity per plant increased up to 9 weeks after application. Examination of autoradiographs of plants confirmed this behaviour, and showed greater activity at leaf tips, in veins and at growing points.

Resumé Pour expliquer les différences en lutte contre l'Oscinelle chez Maïs suivant des différentes méthodes d'application des insecticides granulés, des essais sur maïs variété INRA 270 ont mis à éxecution aux champs et en serre sur l'absorption de l'insecticide phorate, marqué au p^{32}, impregné sur des granules de fuller's earth, et appliqués par des routes différentes. La lutte était plus efficace après application audessus des plantes que sous les semences ou à côté des plantes sur le sol. La radioactivité dans les plantes a montré une période d'activité plus courte mais plus forte, 17 ppm, après application audessus qui coincide avec la période d'attaque de frit en juin. Traitment dans le sillon a produit moins activité, 12,8 ppm, mais pour une période plus longue. L'activité par plante s'est augmentée jusqu'à neuf semaines après l'application des granules. L'examen des autoradiographies a confirmé une plus forte activité aux extremités des feuilles, dans les veines et les pointes de croissance.

INTRODUCTION

Granules are of increasing importance as a method of

application of insecticide because of ease and safety of application, accurate placement and the way toxicant release is controlled. This is particularly so in cereals, when they can be drilled under the seed, applied to the soil surface, or broadcast as a top dressing over the crop when some of the granules are retained by the plants (Walker 1961).

Granules are of value for fly and moth stem borer control in the tropics, and these trials were intended to give information on the distribution and intensity of insecticide concentrations in maize after granule application to different parts of the plant, and to relate this pattern of uptake to the control of frit fly (Oscinella frit) the larvae of which bore into the base of the maize stem. Granules impregnated with ^{32}P- phorate were used in a field trial (Experiment 1) on frit fly control at Wytham, Oxford, and plants were sampled for radioactivity counts and autoradiography during the trial. Glasshouse experiments were also carried out at Porton (Experiment 2).

MATERIALS AND METHODS

Granules were prepared as follows:

About 15 mc of ^{32}P- phorate (1.4g) were obtained from the Radiochemical Centre at Amersham. This was diluted with 113g. of non-labelled phorate, 38g. of a stabilizer supplied by Cyanamid Ltd. added, and the mixture diluted to a total volume of 250 ml with acetone.

Fuller's earth granules (450g), 22/44 mesh (353-699μ), were placed in an inclined polythene bottle and 100 ml of the phorate solution, diluted with a further 200 ml of acetone, added. The bottle was rotated and a stream of filtered air passed into its open mouth until the solvent had evaporated. The impregnated granules were tumbled for a further 30 min, spread out on grease-proof paper so that final traces of acetone could evaporate and then remixed in the bottle for another hour. The concentration of phorate in the granules was 8.7% w/w.

Experiment 1 Maize var. INRA 270 was sown 21 May 1964. It emerged about 1 June, was thinned to 9 plants per yd^2 (7.52 m^2) on 11 June and harvested 22 September.

Treatments:
1 Granules in seed furrow, applied 21 May.
2 " at side of plants, applied 4 June.
3 " broadcast over plants, applied 4 June.
4 Control (no treatment).

The design was a latin square 4 x 4, with plots each 3.66 m x 4 rows 60 cm apart, making 8.92 m^2 per plot. Seeds were sown double at 15 cm apart to give 200 plants per plot, later thinned to about 100 plants per plot.

Phorate granules were applied at 1.26 kg phorate per ha. In fact 15g. phorate granules were diluted with untreated granules to make 40g. per plot for more accurate application. Treatment (1) was raked in the seed furrow under but mostly not in contact with the seed; (2) was applied to the soil surface in a 5 cm wide band at one side of seedlings when in the three-leaf stage, 5-10 cm high; and (3) was broadcast in a 15 cm band over seedlings at the same stage. Application was made by sprinkling from thick glass boiling-tubes by hand, rubber gloves and boots being worn. Radioactivity on hands and boots was monitored after application.

The weather was very dry in late summer and autumn, and yields were finally low.

Plant samples were taken from the four replicates of each treatment at the times shown in the tables. As treatment 1 was applied before germination, sampling was later after applicatiion than in the other treatments. The number of plants per sample decreased from 30 to 12 as the experiment continued because of the increased plant size and the difficulty of subsequent processing. Each sample was washed free from soil particles, the excess water removed and the sample weighed. The total plant was cut into 5 cm lengths, placed in shallow trays and dried for 2 h at 80°C. in hot-air oven. The drying time was increased in later samples due to the increased amount being processed. After drying the plants were re-weighed and passed through a Cullatti Hammer Mill fitted with a 0.04 in. screen. The resulting fine powder was well mixed by rolling and 1 g. samples pressed into the shape of discs, 2.2 cm diam by 0.5 cm deep, for counting on planchettes with an 'end-window' counter. Standard counting methods were observed. Plants from untreated plots were sampled as controls to determine the level of natural radioactivity; counter background alone was about 13 c.p.m., and plant background about 10 c.p.m. A calibration curve was prepared by counting the activity of 1g discs prepared from a dried powder of untreated plants impregnated with known amounts of labelled phorate. 100 c.p.m. were taken as equivalent to 5 µg phorate in each disc under these conditions and subsequent counts were corrected for background count and decay to this standard.

For autoradiography, young plants from the field were washed, arranged on absorbent paper, dried in an oven at 80°C for 2-10 hours depending on thickness, and mounted with Sellotape strips on fresh paper. They were then autoradiographed by the method of Crafts and Yamaguchi (1960) and Little (1962), with 'Kodirex' X-ray film for the period stated. Packs of mounted plant, film, foam sheet (bathmat), hardboard were pressed together and kept at 0°C. An indication of exposure time to X-ray film after several half-lives is given by Flinn (1964). Plants that could not be mounted immediately were held in deep freeze at -20°C to prevent movement of materials. Films were developed in Kodak D.19B.

Experiment 2 Single maize plants growing in 16 cm diam. pots in John Innes compost in a cool glasshouse were treated with radioactive phorate granules when the plants were about 15 cm high. In one set

of treatments (1), 200 mg of granules were placed in a hole under each plant among the roots, 4 cm below soil level. In a second set (3) 200 mg. granules were placed in the leaf whorl funnels to simulate broadcast application. Plants and soil were kept moist with a mist spray. They were then lifted, washed, dried, mounted, and autoradiographed as above.

RESULTS

Experiment 1 The amount of phorate found, as ppmw phorate-equivalent in fresh maize tissue, is shown in Table 1. Roots were included in all samples except the last.

Table 1

Concentration of phorate-equivalent in plants after the three granule treatments

sample number	weeks after application of treatment (1)	(2)	(3)	concentration as ppmw fresh weight * (1)	(2)	(3)
1	3	1	1	12.8 ± 2.8	3.1 ± 0.5	17.4 ± 0.9
2	5	3	3	9.6 ± 0.4	1.7 ± 0.3	3.7 ± 0.5
3	7	-	-	3.4	-	-
4	9	-	-	2.3	-	-
5	11	-	-	0.08	-	-

* Values shown with standard errors are the means of four determinations. Other values are single determinations from pooled replicates.

From the first time of sampling in each treatment there was a progressive reduction in concentration of phorate-equivalent per unit weight of fresh maize tissue. If the unit plant and its fresh weight is considered, however, the amount of phorate-equivalent per plant is seen to increase with time, particularly in treatment 1 (Table 2).

Table 2

sample number	mean fresh weight of plant (g)	Amount of phorate per plant phorate-equivalent (µg/plant) treatment 1	treatment 2	treatment 3
1	1.6	20.3	5.0	27.8
2	6.6	63.4	11.9	24.4
3	61	202	-	-
4	213	490	-	-
5	323	26	-	-

This clearly illustrates the ability to maintain a prolonged residual effect of insecticide in the plant for nine weeks after treatment (1)(furrow treatment) and for at least three weeks after treatments (2) (side application) and (3)(broadcast). In fact if the dilution of the concentration by growth is allowed for, as in Table 2, absorption of radioactive material by the plant is seen to increase up to nine weeks after application of granules to the soil. In treatment (1) there was a dramatic fall in level after this period.

The intensity of image of plants examined by autoradiography is expressed as * to **** in Table 3. Roots are classified as 'near' or 'far' from granules. These observations are discussed later with the autoradiographs of Experiment 2.

Table 3

Intensity of image of autoradiographs of plants from Experiment 1.

Sample No.	Treatment No.	Days after application	X-ray film exposure (days)	roots		seed	stem	leaves		
				far	near			whorl	blade	tip
X	1	15	16	****	***	***	**	**	**	**
	2	1	16	-	-	-	-	-	-	-
	3	1	16	**	**	**	*	***	****	***
1	1	21	31	***	****	***	***	***	**	***
	2	7	31	**	**	**	**	**	**	**
	3	7	31	**	**	**	***	***	***	****
2	1	35	32	***	***	***	**	**	**	**
	2	21	32	*	*	*	*	*	*	*
	3	21	32	**	**	*	*	**	**	**

- = not autoradiographed.
X = preliminary sample; other sample numbers as in previous tables.

The intensity of image after autoradiography of pot-grown plants, treated with granules under (treatment 1)or over (treatment 3) the plants, is shown in Table 4. Leaves below those receiving granules are indicated as 'leaf below treatment'. Examples of autoradiographs are shown in Fig.1.

Table 4

Intensity of image of autoradiographs of plants from Experiment 2

Treatment No.	days after treatment	exposure (days)	root		stem	leaf			leaf below treatment
			far	near		whorl	blade	tip	
1	1	14	**	***	*	*	*	*	-
3	1	14	0	0	0	***	****	***	0
1	2	16	*	****	*	*	**	**	-
3	2	16	0	0	0	****	****	***	0
1	4	16	*	***	*	*	**	**	-
3	4	16	0	0	*	**	****	****	0
1	8	38	0	**	**	*	**	**	-
3	8	38	0	***	*	***	****	***	0
1	19	52	0	*	**	*	*	*	-
3	19	52	0	0	**	**	*	**	*
1	31	60	*	**	**	*	*	*	-
3	31	60	0	*	**	**	(*)	(*)	0

Symbols are used as in Table 3. (*) denotes very faint image.

Because of variation among the individual plants sampled, differences between treatments sampled at one time were sometimes more apparent than differences between samples at different times. However a pattern of intensity of radioactivity could be seen that agreed well with the autoradiographs and radioactivity counts of plants treated in the field (Experiment 1), and the main features shown in the two experiments will be described together.

Treatment 1: root treatment (field and glasshouse)

(i) One day after application of granules, intense radioactivity was present in the roots nearest to the granules. A little toxicant has been translocated throughout the plant.

(ii) Two days after application the intensity had increased, reaching moderate amounts between four and eight days in the leaf blades and tips. Some activity persisted in the roots and leaves up to thirty-five days after application.

(iii) A high intensity of activity was present in the seed for up to five weeks after application.

Treatment 2: soil surface treatment (field only).

A moderate radioactivity was built up throughout the plant seven days after application. It did not persist as long as or as intensively as after root or leaf treatments.

Treatment 3: leaf treatment (field and glasshouse).

(i) One day after application of granules to the top of the plant intense radio activity was present, greatest at the sites of granule retention, the leaf blades, whorl and leaf axils.

(ii) From two to eight days after application activity was very high in leaf blades and tips, becoming less after eight days from application. It was still as great if not greater than after root application at three weeks after application but never reached such activity in the roots. In the glasshouse, a month after application, activity in all parts except the stem and whorl was less than after root application.

(iii) There was no translocation downwards into leaves not receiving granules after leaf application, as seen in the last column of Table 4. The activity in roots and below the application point was probably due to granules falling on to the soil followed by absorption of toxicant through the roots.

(iv) Some autoradiography show greater radioactivity in the leaf veins and the growing point of the stem.

DISCUSSION

Control of frit fly in 1964, the subject of a paper to be published, as shown by the angular transform of the mean percentage of plants severely attacked by frit on 25th June, was greatest after treatment (3), a top dressing (2.0^{o}); while treatment (1), furrow treatment (14.0^{o}) and treatment (2), soil dressing (15.9^{o}) were less effective. Control (4) value was 41.6^{o} and the standard error $\pm 1.7^{o}$. Top dressing with phorate granules was generally found to be less effective in control of frit fly than furrow application in previous years. Examination of radioactivity in plant samples and autoradiographs here suggests that the explanation is probably the early intense concentration of phorate in the leaves and leaf axils in plants treated from above (treatment 3) in the first week of June. Although root treatment (1) gave a more persistent high concentration of phorate or metabolite from early June to August, it did not appear to reach the same intensity as in leaf treatment (3).

This indicates that for rapid intense insecticidal acivity, a top dressing of granules will give a high concentration in the leaves within a day, but the intensity will decline after a week. For a long lasting activity, particularly against root pests, granules in the furrow beneath the seeds should offer greatest protection. This conclusion might obviously be modified by rain washing off granules and releasing toxicant from them more quickly, or by wind which will shake granules on to the soil (Walker, 1961). Furrow treatment with granules has been found to be more effective than top dressing against some fly pests of cereals, such as Hessian fly (Mayetiola destructor) in Cyprus (Walker, 1971). Furrow treatment was also effective against barley fly (Hylemya arambourgi)in Kenya (Walker, unpublished). These are situations in which a prolonged level of insecticide activity is valuable. The longer persistence of phorate and its metabolites under, rather than on the surface of soil has recently been confirmed by Schulz et al. (1973).

The radioactivity present in the plant is not necessarily insecticidal phorate. Rapid breakdown to metabolites has been shown to occur in cotton and vegetables (Casida, 1962). Oxidation products, sulphoxides and sulphones, are formed first at the thio-ether sulphur and then at the thiono sulphur. These are more potent anti-cholinesterases than phorate and persist in plants for long periods of time. We attempted to identify and follow the changes in metabolites in maize by means of thin layer chromatography but experienced difficulties due to impurities in the radioactive material supplied, and in obtaining pure radio-labelled metabolites as standards. Lichtenstein et al. (1973) have recently reported the replacement of phorate by its sulphoxide and then sulphone in soil.

Information on the uptake of phorate from granules and seed treatments by cotton is given by Hacskaylo, Lindquist and Clark (1961). Hacskaylo, Lindquist, Davich and Morton (1961) found that accumulation of phorate by the roots of the cotton plant was very rapid for the first two days and then declined rapidly. The stem acted mainly for transportation. Accumulation of phorate in the leaves was correlated with transpiration and the factors that favoured it. Lindquist, Hacskaylo and Davich (1961) related the amount absorbed by roots to the volume of root exposed, the most active site being 20-40 mm above the radicle tip.

There appears to be a longer period of absorption of phorate by roots of maize than that reported for cotton (Lindquist, et al. 1961) and Bardner (1964) found a long period of absorption in wheat. The duration of effect of phorate may depend on the rates of growth, and Anderson, et al. (1961) found chrysanthemum plants remained toxic to mites for several months. Paris and Newbould (1964) showed increased absorption of phosphorus from soil by rye and barley at greater growth rates.

The work was continued with radioactive ^{35}S- phorate, which has a longer half-life (to be published later).

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References

ANDERSON, M.A., SCHULZ, J.T. and HIBBS, E.T. (1961) Biological activity and localization of phorate residues in soil-drench treated Chrysanthemums. Journal of economic Entomology, 54, 827-832.

BARDNER, R. (1964) Uptake of phorate applied as a slurry to wheat and mustard seeds. Annals of applied Biology, 53, 445-458.

CASIDA, J.E. (1962) Metabolism of O.P. insecticides in plants; a review in Radioisotopes and Radiation in Entomology p 49. Intern. Atomic Energy Agency, Vienna.

CRAFTS, A.S., and YAMAGUCHI, S. (1960) Gross autoradiography of solute translocation and distribution in plants. Medical and biological Illustration, 10, 103.

FLINN, M. (1964) Calculation of exposure time for successive autoradiographs. Nature, 202, 1036.

HACSKAYLO, J., LINDQUIST, D.A., amd CLARK (1961) Phorate accumulation by cotton plants and recovery from soil. Journal of economic Entomology, 54, 411-3.

HACSKAYLO, J., LINDQUIST, D.A., DAVICH, T.B., and MORTON, H.L. (1961) Accumulation of phorate by cotton plants from solution and sand culture. Botanical Gazette, 123, 46-50.

LICHTENSTEIN, E.P., FUHREMANN, T.W., SCHULZ, K.R. and LIANG, T.T. (1973) Effects of field application methods on the persistence and metabolism of phorate in soils and its translocation into crops. Journal of economic Entomology, 66, 863-866.

LICHTENSTEIN, D.A., HACSKAYLO, J., and DAVICH, T.B. (1961) Absorption and trans-location of phorate by cotton seedlings. Botanical Gazette, 123, 137-140.

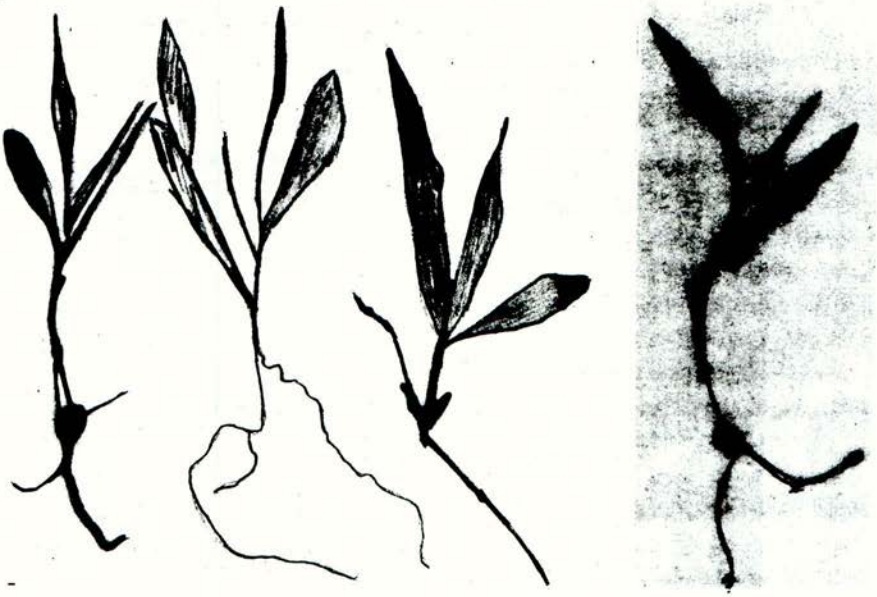
LITTLE, E.C.S. (1962) Improvements in autoradiographic techniques study of movement of C-14 labelled compounds in plants. Medical and biological Illustration, 12, 36-9.

PARIS, P. and NEWBOULD, P. (1964) Ann. Rep. A.R.C. Radiobiol. Lab. 1963-4, p.55. Wantage, Berks.

SCHULZ, K.R., LICHTENSTEIN, E.P., FUHREMANN, T.W. and LIANG, T.T. (1973). Movement and metabolism of phorate under field conditions after granular band applications. Journal of economic Entomology, 66, 873-5.

WALKER, P.T. (1961) Distribution of granules by a hand-operated distributor and their retention on maize plants. Journal of agricultural Engineering Research, 6, 112-8.

WALKER, P.T. (1971) Insecticidal control of Hessian Fly in wheat and barley in Cyprus. Pesticide Science, 2, 267-275.



(1) (2) (3) (3)

Fig. 1 Examples of autoradiographs: treatments (1) in seed furrow, (2) side dressing on soil surface, (3) top dressing on plant.

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CHEMICAL CONTROL OF SEPTORIA LEAF BLOTCH IN WHEAT

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Summary Daconil 2787 and Maneb were found to control economically the damage caused by Septoria tritici in spring wheat. Treatment increased yield by 23% (Daconil 2787) or 17% (Maneb), while the net value of the yield increase was 15% for Daconil 2787 and 13% for Maneb. Distinction is made between biological activity (Daconil 2787 is better than Maneb), reduction of yield loss (Daconil 2787 > Maneb) and economical control (Daconil 2787 = Maneb). It is stressed that evaluation of fungicides should be based not only on their biological activity, but also on their cost : benefit ratio.

Resumé Daconil 2787 et Maneb se sont avérés capable de contrôler économiquement les ravages causés par Septoria tritici chez le blé de printemps. Le traitment augmente le rendement de 23% (Daconil 2787) ou de 17% (Maneb), la valeur nette de l'augmentation du rendement étant de 15% pour le Daconil 2787 et de 13% pour le Maneb. Une distinction est faite entre l'activité biologique (Daconil 2787 s'avère supérieur au Maneb), l'augmentation du rendement (Daconil 2787 > Maneb) et un contrôle à bon marche (Daconil 2787 = Maneb). Le fait que l'évaluation des fongicides ne doit pas se baser seulement sur leur activité biologique mais aussi sur leur rapport coût-bénéfice est souligné.

INTRODUCTION

Septoria leaf blotch (Septoria tritici), the only Septoria disease on wheat in Israel, has become an important factor in wheat production and a cause for much concern among farmers and advisers. Yield loss estimates to date (Eyal, 1972) are few and not applicable for the country as a whole. The wheat farmer, who, under Israeli conditions, invests much money and effort in wheat production, must combat the disease with effective control measures. Since there is no effective host resistance in current commercial varieties, only sanitation and chemical control can be considered.

With chemical control, several questions arise:

1. At what date and at what stage does the disease appear and should chemical control be prophylactic?

2. What is the rate of spread of the disease relative to the growth rate of the wheat plants?

3. What parts of the plant are critical in determining yield loss and efficiency of control?

4. What is the cost: benefit ratio of any chemical control?

Our experiment was designed to follow the disease development of S. tritici and to obtain an estimate of the biological efficiency vs. economical feasibility of two fungicides.

Chemical control in such low income crop as wheat deserves careful consideration for the optimization of economic control measures.

METHOD AND MATERIAL

The field experiment was in randomized blocks with five replicates. Each plot was 40m x 18m. The fungicides were applied in 300 l/ha water from a tractor-driven sprayer with a 9m boom. A susceptible variety Mivhor 1177 (spring wheat) was planted in early December 1971. The fungicides used, the dose rates and dates of application are indicated in Table 1.

Table 1

Details of treatments in the field trial

	Treatment	Dosage	Dates of applications 1972
1.	Maneb	(3.3 kg/ha (2.5 kg/ha	13 Feb. 6 Mar. 25 Mar.
2.	Daconil 2787	(2.4 kg/ha (1.9 kg/ha	13 Feb. 6 Mar. 25 Mar.
3.	Daconil 2787	(2.4 kg/ha (1.9 kg/ha	13 Feb. 12 Mar.
4.	Control	-	-

The spraying dates were determined by consideration of the disease development and the climatic conditions in the field. The first spray was applied when Septoria lesions were found on the third leaf from the bottom (1-10 lesions on 44% of the third leaves). Subsequent sprays were determined by taking into account rainfall and the upward spread of the disease in the crop.

Estimations of disease severity were made every 7-10 days on a sample of 25 plants from each plot and scores were taken on each leaf separately. The scores, as percentages, are according to a scale adapted from James (1971). The number of dry leaves among those of the same age in each sample was also counted.

Yield One strip (4.55m wide, 38m long) was harvested by combine from each plot and yield per hectare was calculated. From each plot, hectoliter weight and thousand kernel weight were determined.

Yield values were calculated according to market prices: one metric ton at hectoliter weight of 76 kg sold at 365 IL/t, but the price decreased proportionately with hectoliter weight to 72 kg, below which grain was sold at 320 IL/t. The cost

of the fungicides and applications have been deducted from the market value of the crop (6 IL/kg for Maneb and 18 IL/kg for Daconil 2787).

RESULTS

Amount of disease. Disease levels on each of the upper four leaves in each of the three treatments were lower than in the control plots. No effect was found on the fifth leaves from the top. The chemical treatments delayed disease development on the upper leaves due probably to the prevention of early spread from the lower leaves. Disease development on the first (flag) leaves, second leaves and third leaves is shown in Fig. 1-3. For each leaf, disease development was postponed by at least 10 days. Daconil 2787, even at the lower dose, controlled the disease better than Maneb.

Another measure of disease severity was the number of dead leaves in each age group. Fourty per cent of the fourth leaves from the top in the treated plots were still green 3 weeks after the fourth leaves in the control plots were dry. Treated flag and upper leaves stayed green till the end of the season, while in the control 15-60% of the leaves were dry 10-20 days earlier.

Yield - Data on yield and yield parameters are presented in Table 2. There are no significant differences between treatments for any of the yield parameters because

Table 2

The effect of chemical control on wheat yields under natural infestation with Septoria leaf blotch (S. tritici)

Treatment		Yield (kg/ha)	Hectoliter weight (kg)	1000 Kernel weight (g)	No. of Kernels per ear	Yield value IL/ha
Maneb	3 sprays	4.6 ab*	73.2	32.6	65.4	1446
Daconil 2787	3 sprays	4.8 a	72.8	32.5	64.6	1477
Daconil 2787	2 sprays	4.2 b	71.9	32.2	67.0	1305
Control		3.9 b	71.9	31.2	61.4	1282

* Different letters indicate significant difference at 1% level

the results were very variable. Despite the variability there were significant differences in total yield. Nevertheless, the trends shown by these results should be considered. It seems that under the conditions in 1972 one of the main effects of S. tritici was the reduction in the number of kernels per ear. The data on yield value shows that chemical control of Septoria leaf blotch can be economical. Under these conditions the net benefit was 13-15%.

DISCUSSION

Control of a disease vs. reduction of disease losses

The biological effect of the fungicides on the disease, throughout the season, was remarkable and graphs cannot illustrate the whole effect seen in the field. However, the expectations for large yield differences did not materialize. One of the reasons was the dry weather towards the end of the season. Treated plants with an appreciable greater amount of green leaves than the untreated plants transpire more and are therefore more subject to water stress. A hot spell during the last days of ripening probably resulted in loss of potential yield expected from the treated crops. In trying to correlate disease control with reduction of damage (Van der Plank, 1963 and others), factors such as the interaction of fungicide, causal organism and host should be taken into consideration as well as the interaction host and environment.

Biological efficiency vs. economical feasibility

Disease control in most crops cannot and for an economic return need not be complete. Control measures should be applied to control the damage economically. Unfortunately this well-agreed principal is not often followed in practice. Fungicides are usually compared and rated according to their biological efficiency rather than their economic value. The effect on disease control is often different from the control of damage, as measured by yield reductions, and the control may be achieved by means that are different in cost. The same fungicide against the same disease on different varieties of the same crop might have a different optimum in economic control (Eshet & Dinoor, 1972). We maintain that efficacy of a fungicide should be measured according to its ability to increase the net profit from a crop. Therefore neither the price of a fungicide nor its biological activity should be considered separately. In our experiment Daconil 2787 is biologically more effective against the disease than Maneb, but with only two sprays it does not increase the yield as much as Maneb. Daconil 2787 is three times more expensive than Maneb but effects better control of yield loss. Economical control of the yield loss can be achieved by increasing the efficiency of a fungicide or reducing its price or both. The reduction in price could be achieved by improving the biological efficiency so that less material is needed or fewer applications are required. In our experiment we attempted to reduce the price of control measures using Daconil 2787 against Septoria, by reducing the dose in each application and by reducing the number of applications, resulting in reduction in the cost of both labour and fungicide. A reduction in the dose made Daconil 2787 as economic as Maneb but reducing both dose and number of applications failed to give an economic return. It seems to us that much more experimentation is needed in order to optimize the economical use of most fungicides in most crops. In such experiments a fixed dosage and a variable price should be compared to a fixed price and a variable dosage. The results will then indicate whether or not economical control of a particular disease by a particular fungicide in a particular crop is feasible.

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References

ESHET, Y. and DINOOR, A. (1972) Chemical control of Oidiopsis powdery mildew on Peppers. Hassadeh, 52 (Hebrew).

EYAL, Z. (1972) Effect of Septoria leaf blotch on the yield of spring wheat in Israel. Plant Disease Reporter, 56, 983-986.

JAMES, C. (1971) A Manual of Assessment Keys for Plant Diseases. Canada Department of Agriculture, Publication No. 1458.

VAN DER PLANK, J.E. (1963) Plant Diseases: Epidemics and Control, 349 pp, New York, Academic Press.

Fig. 1

Percentage disease on flag leaves

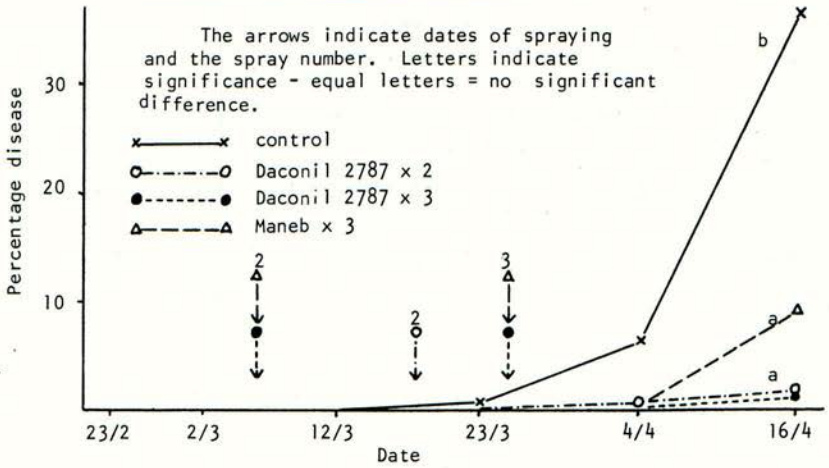

Fig. 2

Percentage disease on second leaves

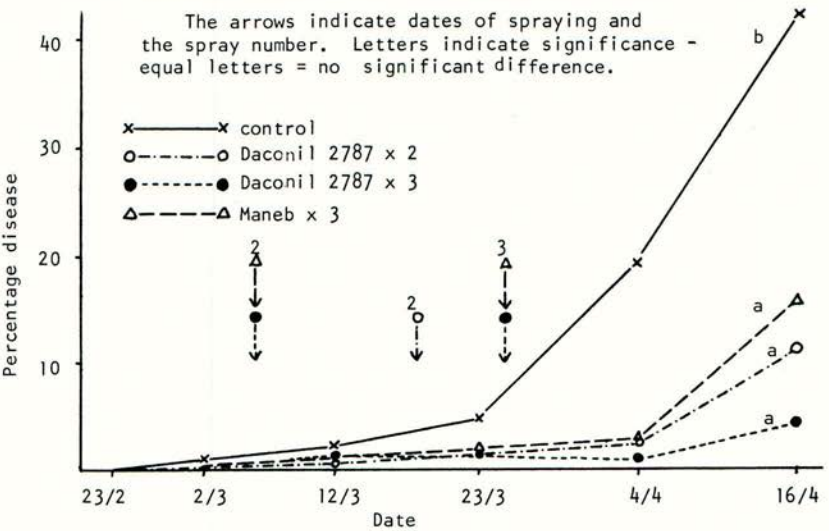

Fig. 3

Percentage disease on third leaves