LONG TERM CONTROL OF ALOPECURUS MYOSUROIDES IN WINTER WHEAT

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Summary Pre-emergence and post-emergence herbicides were compared over five years in a continuous wheat rotation. The herbicides were applied to the same area each year or in alternate years. Large increases in yield were obtained where herbicides were applied each year, but the annual residual weed population was sufficient to warrant continuing control measures. Herbicides merely improved yields and eased harvest difficulties without eliminating the weed problem.

INTRODUCTION

<u>Alopecurus myosuroides is a serious weed on heavy alkaline soils and because</u> of its winter annual habit is greatly encouraged by a high proportion of winter sown cereals. Much seed is shed before the cereal is harvested and germinates after a short period of primary dormancy, if conditions are suitable. Cultivation does not appear to influence germination but burying seed, especially by ploughing, induces secondary dormancy which encourages survival to the following year (Wellington and Hitchings, 1965). Secondary dormancy accounts largely for low spring germination and only in early sown spring cereals does the weed occasionally present a problem.

Seedlings of <u>A</u>. myosuroides do not emerge from depths greater than $2 \cdot 5$ cm and where ploughing is undertaken for seed-bed preparation most plants arise from seed shed in previous years whose dormancy is broken when brought to the soil surface (Naylor 1970).

The ability of seed to survive more than one year as a result of secondary dormancy is an important factor in the maintenance of <u>A</u>. myosuroides populations where autumn sown cereals are the main enterprise. In this situation annual or biennial herbicide treatment is necessary to ensure profitable crop yield.

METHODS AND MATERIALS

The trial was sited on a silty loam of Upper Greensand over Gault in Buckinghamshire. The first year of the experiment was situated in the sixth winter wheat crop.

The following treatments were applied to the same areas each year during 1966-71.

1. Pre-sowing

24 oz a.i./ac triallate incorporated by harrows.

2. Post-sowing pre-emergence

32 oz a.i./ac terbutryne (1966-68)

45 oz a.i./ac methabenzthiazuron (1970-71)

3. Post-emergence of crop and weeds

5 oz a.i./ac barban (A. myosuroides 1-2 leaves) or

16 20z a.i./ac simazine + methoprotryne (A. myosuroides 3-4 leaves)

4. Control, no spray

The experimental design was a randomised block with four replications. Two blocks received treatments each year and two in alternate years only. Each plot was 15 ft x 132 ft in size and sprays were applied across the cereal drills. Combine yields of grain were obtained from areas of 10 ft x 120 ft in each plot. Each year seedbeds were prepared by ploughing and cultivation and the trial area received the same fertilizer treatment as the farm crop. Spring wheat was sown in 1969 because the wet autumn of 1968 precluded winter wheat.

		Table 1 Site Details			
	1966/67	1967-68	1968-69	1969-70	1970-71
Crop Date sown and	W.Wheat	W.Wheat	S.Wheat	W.Wheat	W.Wheat
pre-emergence application	12.11.66	13.10.67	28.3.69	20.11.69	20.10.70
Seed rate 1b/ac	168	160	162	160	150
Variety	Cappelle Desprez	Cappelle Desprez	Janus	Cappelle Desprez	Cappelle Desprez
Post-emergence application & stage of growth	14.2.67	29.2.68	22.4.69	25.4.70	31.3.71
Crop	2-3 leaves	3 ¹ / ₂ leaves	5 leaves	5 leaves	5 leaves
Alopecurus myosuroides	$2-2\frac{1}{2}$ leaves	31-4 leaves	1-3 leaves	4-5 leaves	4-5 leaves
Date of harvest	21.8.67	22.8.68	6.9.69	12.8.70	13.9.71

RESULTS

Counts were made of <u>A</u>. myosuroides plants prior to post-emergence treatment and assessment of control by counting inflorescences per unit area prior to harvest. The yield of wheat was assessed as cwt/ac clean grain at 85% dry matter. Results are shown in Table 2.

	1966	67	. 1	967/68		1	968/69	
Year Treatment	% Control	Yield	Blackgrass ft ² (22.11.67)	% Control	Yield	Blackgrass ft ² (7.5.69)	% Control	Yield
Annual Application								
No Spray	0	27.1 cwt/ac	15.7	0	11.0 cwt/ac	25.8	0	29.5
Pre-emergence 1	52	115	16.5	73	165	8.1	76	121
2	37	114	16-8	55	115	13.0	10	111
Post-emergence 3	61	122	16.4	60	143	25.0	18	110
SE Means (%)	10.5	5.5		14.1	8.7		10.9	1.7
Alternate Year Application			Resi	dual Yea	ar	S	prayed	
No Spray			18.5	0	10-4 cwt/ac	27.3	0	26.9
Pre-emergence 1			17.0	10	97	10.3	79	126
2			20.3	3	100	20-8	46	113
Post-emergence 3			19.1	30	107	18.4	35	111
SE Means (%)				13.7	10.4		7.9	5.7

Table 2 Control of Alopecurus myosuroides and grain yields 1966-71

1966-67 results mean of all plots

Blackgrass ft² = number of plants emerging with crop and date counted. % control = % reduction in inflorescences relative to control plots. Yield = percentage of control yield.

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Table 2 Continued

	1	969/70		1	1970/71			
Year Treatment	Blackgrass ft ² (24.2.70)	g Control	Yield	Blackgrass ft (3.3.71)	% Control	Yield		
Annual Application								
No Spray	24.3	0	25.9 cwt/ac	91 • 4	0	9.3 cwt/ac		
Pre-emergence 1	10-1	51	140	30-8	66	250		
2	12.6	70	136	32-3	65	139		
Post-emergence 3	22.9	18	124	69.7	24	166		
SE Means (%)		8.5	4.9			15.9		
Alternate Year Application	Resi	dual Year		S	prayed			
No Spray	32 • 1	0	24.9 cwt/ac	108.0	0	9.1 cwt/ac		
Pre-emergence 1	26.4	16	119	48.1	56	288		
2	23.7	14	119	61.7	43	157		
Post-emergence 3	25.7	8	117	74.4	31	182		
SE Means (%)		7.7	5.1	12.2		16.2		

Treatment Annual application Pre-emergence 1 2		Average increase in grain yield per annum over control (1966-71) cwt/ac 85% dry matter
Pre-emergence	1	8.9
	2	4.9
Post-emergence	3	5.6
Alternate year	application	
Pre-emergence	1	6.1
	2	3-1
Post-emergence	3	3.8

Table 3 Mean Yield Increase Per Annum Over Five Years

DISCUSSION

Counts of A. myosurcides plants soon after emergence show the effect of current and residual pre-emergence treatments but only the residual effect of postemergence treatment from previous years. Counts in 1967 were done too early as plants were still dying on the pre-emergence herbicide plots. Blocks treated in alternate years showed a residual effect of previous herbicide treatments but this tended to be small relative to the control plot populations.

The level of control of <u>A</u>. myosuroides was poor but the average response to herbicides in grain yield over the five years was large (Table 3). Yields on the control plots were very low and responses to herbicides very large when early sowing encouraged a high <u>A</u>. myosuroides population (1968 and 1971). Triallate gave the most consistent results. The responses in yield from the post-emergence herbicide were greater than expected from the degree of control as measured by inflorescence counts, this has been noted in other trials in the area.

As A. myosuroides counts were undertaken before the post-emergence herbicide was applied a comparison could be made of annual applications with spraying in alternate years on the weed population. Differences are still small after five years of treatment (Table 2). In a continuous wheat rotation, herbicide effects on the <u>A. myosuroides</u> population will tend to be delayed until seed shed in previous years has germinated and been eliminated. In addition the incomplete control by herbicides allows a considerable return of seed to the soil reservoir. It is hoped to continue this trial as a long-term project.

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COMPARISON OF SYSTEMS ON CONTROL OF AVENA FATUA IN SPRING BARLEY

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<u>Summary</u> A long term study was undertaken on the behaviour of A.fatua under seven systems of control applied to the same areas each year in commercial crops of spring barley. Late sowing to kill germinating <u>A.fatua</u> was compared with pre-emergence triallate e.c. and barban post-emergence applied each year, in alternate years only and the two herbicides alternately. An area was left untreated as control and samples were taken at intervals to measure the number of seeds of <u>A.fatua</u> in the soil. Plant populations of <u>A.fatua</u> varied considerably between years and this was reflected in variable yield responses to treatments. At the end of four years there was evidence that control measures were effectively reducing the population of <u>A.fatua</u> to roguable levels and the value of the extra yield of barley covered the cost of treatment.

INTRODUCTION

Under commercial conditions populations of wild oats vary considerably from year to year in known infested fields. Factors of importance are:-

a) the soil "reservoir" of seeds and their age

b) the number of seeds germinating and establishing plants

c) the date of sowing and competitive ability of the cereal crop

d) the relative dates of emergence of wild oat and cereal plants

e) control measures adopted in the past and effect on the soil "reservoir" of seed.

Thurston (1963) considered that germination of <u>A.fatua</u> depended on properties of the seed more than weather conditions or depth and time of cultivation. More seedlings of <u>A.fatua</u> appeared in the second rather than the first spring after shedding. Thurston showed that time of emergence also depended on the depth of the seed. Each one inch depth delaying germination by one day down to nine inches but with older seeds taking progressively longer to emergence. <u>A.fatua</u> emerging later than the cereal crop also produces less seed, a 14 to 18 day delay reducing seed production by 40% and 63% respectively. Early sowing of the spring cereal gives the crop and advantage over the main flush of <u>A.fatua</u> but fewer seedlings would be killed by seedbed cultivations.

Selman (1970) examined the population dynamics of <u>A.fatua</u> in spring barley under various treatments. In the untreated crop the annual rate of increase of <u>A.fatua</u> varied from a multiplication factor of 1.3 to nearly 6 with a mean of 3.05 over 6 years. With late sowing and triallate e.c. treatment, there was variation in the annual rate of decrease around a mean of three. Selman concluded that it was not possible to predict populations of $\underline{A.fatua}$ but discussed situations where treatment would be profitable and where savings could be made in control measures.

The trial described in this paper is a study of the behaviour of <u>A.fatua</u> populations in cereals under treatments which vary in efficiency of control. In this way it may be possible to show the effect on the weed population in the soil, whether the most effective treatments can eliminate wild oats or whether it is more profitable to maintain populations at a level not affecting cereal yield by cheaper control methods.

METHOD AND MATERIALS

The trial was laid down on a calcareous boulder clay (Hanslope series) at Hardmead, Bucks where a minimum of autumn cultivation followed by winter ploughing is the normal practice between cropping. Size of plot was 20 yd by 80 yd to allow combine yield determinations. There was no replication except in the untreated control plots. Treatments were as follows:-

1. Late sowing of the crop

Seedbed prepared 3 weeks before sowing and seedling wild oats killed by two spring time cultivations

2. Triallate e.c. at 20 oz a.i./ac pre-sowing incorporated

Applied each year Applied alternate years only Alternate years with barban

3. Barban at 5 oz a.i./ac post-emergence

Applied each year Applied alternate years only Alternate years with triallate e.c.

Two control plots were included. Spraying was carried out across the cereal drills and plots were large enough for two combine yield cuts of 10ft x 200ft per treatment. Details of the site are shown in Table 1.

Table 1

Details of Site

	1968	1969	1970	1972
Sowing date				
Normal	28.3.68.	9.4.69.	6.5.70.	17.4.71.
Late	22.4.68.	2.5.69.	6.5.70.	4.5.71.
Cultivar	Deba Abed	Deba Abed	Sultan	Zephyr
Seed Rate 1b/ac	140	140	140	150
Application of triallate	22.3.68.	9.4.69.	6.5.70.	15.4.71.
Application of barban and	2.5.68.	22.5.69.	1.6.70.	19.5.71.
stage of growth of A.fatua	13-2 leaves	2-23 leaves	1-23 leaves	2 leaves

The 1968 crop followed three spring barley crops which received commercial applications of triallate or barban overall. In 1970 a wet spring delayed sowing of the whole area until early May.

RESULTS

The effect of treatments on panicle number and yield of clean grain in cwt/scis given in tables 2 a and b, panicles were categorised for size and sub-sampled for spikelet number but are not presented here. Where possible counts of wild oat seedlings were undertaken prior to seedbed cultivation and post-emergence of the crop at the 2 leaf stage. Soil samples were taken at intervals to assess the number of seeds of <u>A.fatua</u>. In 1968 the herbicide plots were sprayed overall and the alternate treatments started in 1969.

Table 2a

Seed in soil and emerging plants of A.fatua

					TRIALLAT	E		BARBAN	
	Cor	ntrol	Late Sown	Annual	Alter- nate only	Alter- nate Barban	Annual	Alter- nate only	Alter- nate Trial- late
A.fatua see	ds								
10 kg soil	Feb 6	3 10	12		9			11	
	Sept 6	3 107	-					-	
	Sept 6	9 123	-		+			-	
	Sept 72	2 52		1	7	2	58	52	4
A.fatua 2	1968	-	70.2	-	-	4	4	4	14
PRE DRILL	1969	26.1	26.1	22.5	25.2	26.1	24.3	29.7	22.5
THE DICINE	1970	105.3	10.8	-	-	-	-	-	-
	1971	70.7	18.0	2.7	16.2	3.6	16.2	47.7	3.6
A.fatua 2	1968	26.5	-		- 2.2			25.2	
plants yd POST DRILL	1969	154.8	8.1	8.1	32.4	28.8	74.7	40.6	9.0
	1970	49.0	4.5		- 2.7			- 20.7	
	1971	56.7	4.5	0.9	9.0	9.9	15.3	29.7	7.2

Table 2b

A.fatua panicles and yield of barley

					TRIALLA	TE		BARBAN		
	Cont	rol	Late Sown	Annual	Alter- nate only	Alter- nate Barban	Annual	Alter- nate only	Alter- nate Trial- late	
Panicles of	1968	140.0	11.7		6.7			7.6 -		
A.fatua yd ²	1969	248.9	23.4	9.9	77.4	25.2	66.6	78.3	12.6	
	1970	9.9	0.6	0*	2.0	0.8	8.0	8.4	0.8	
	1971	34.0	1.6	0*	10.4	0.4	13.2	13.6	0*	
Yield of	1968	17.4	26.3		- 29.7			- 29.6 -		
Barley cwt/ac clean grain	1969	16.3	31.9	33.5	23.6	31.4	26.2	23.8	33.9	
	1970	25.1	27.4	24.8	25.7	25.4	23.5	25.5	27.4	
	1971	33.8	25.1	33.6	34.0	34.0	34.2	32.1	32.3	

* No panicles within quadrats but few visible within plots

Table 3

		TRIALLATE BAR						
	Control	Late Sown	Annual	Alter- nate only	Alter- nate Barban	Annual	Alter- nate only	Alter- nate Trial- late
% Control of Panicle numbers		89	95	81	93	69	67	83
Average Yield cwt/ac annum	23	28	30	28	30	28	27	30
Total extra Yield over Control cwt/ac		18	29	20	28	21	18	30

Summary of four years control systems

DISCUSSION

In the first year of the study 25 A.fatua plants per yd² emerged with the crop and both herbicides gave good results. Yields were severely reduced on the unsprayed plots and production of panicles was little restricted by crop competition. Late sowing gave good control of A.fatua but grain yield was 3 cwt/ac less than with early sowing plus herbicides. Seeds of A.fatua in the soil of the control plots built up by a factor of 10 with one year's free seeding. The crop was drilled later in the second year and there was a large germination of wild oats before and after drilling, slightly more germinated with the crop on the barban block. The number of panicles left after treatment with triallate were similar to the previous year on the continuous and alternate herbicide treatments. Barban was not as effective in reducing panicle numbers as in the previous year. The residual benefit of previous herbicide use was large with both herbicides, i.e. large in terms of crop yield and panicle numbers. Late sowing gave similar results to the previous year. <u>A.fatua</u> was very competitive on the control plots and produced many panicles but the increase in seed population in the soil was not as large as in the previous year.

In the third year all plots were late sown because of weather conditions and fewer wild oats emerged with the crop. This factor combined with overall spraying obscured residual effects of previous treatment although fewer panicles matured on the barban - triallate - barban treatment. Twice as many <u>A.fatua</u> plants emerged on the control area compared to the first year but were not competitive, produced few panicles and had little effect on crop yield.

A.fatua was again not competitive in the fourth year, few panicles were produced relative to the initial population emerging and yields were similar on the control and herbicide plots. The residual value of triallate was greater than barban if wild oats germinating in the spring are taken as an indication of the reservoir of seeds in the soil.

The trial provides evidence of a decline in the population of <u>A.fatua</u> where triallate is used annually. The importance of crop competition in suppressing panicle and seed production is shown on the control plots in 1970 and 1971 where 50 wild oats per yd emerged with the crop but had little effect on the yield. The results indicate that after 3 or 4 years of effective herbicide use quite high initial populations of <u>A.fatua</u> can be reduced to levels that are hand roguable and the financial inducements in terms of increased crop yield (Table 3) warrant the adoption of such techniques.

It is hoped to continue this trial for further years under cereal cropping in particular to monitor the changes of wild oat seed population in the soil.

Acknowledgements

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THE USE OF PLANT GROWTH REGULATORS IN THE CONTROL OF AGROPYRON REPENS

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Summary Eight plant growth regulators, including MH as a standard, were applied at 2. 4 and 8 kg/ha to the foliage or soil of pot grown Agropyron repens at the 5-6 leaf, 1-2 tiller stage. Shoot number and weight. rhizome weight and node numbers, both total and sprouted, were recorded six weeks after treatment. None of the compounds were more effective than MH as a herbicide, but the higher doses of chlorflurecol were almost as phytotoxic. The ability of 2-chloroethylphosphonic acid and chlorflurecol to break dormancy and alter the ratio of shoots to rhizomes indicated potential as adjuvants to existing control measures. The performances of paraquat applied to foliage and pronamide applied to the soil were not enhanced by 2-chloroethylphosphonic acid and it was concluded that although buds were released from dormancy, they invariably developed into rhizomes increasing the plants' survival potential. Low rates of chlorflurecol which induces buds to develop into leafy shoots enhanced the performance of dalapon and paraquat under certain conditions, the relative times of application being important with dalapon and temperature with paraquat. Although herbicide activity can be enhanced with chlorflurecol, more reliable growth regulators are required for successful field application.

INTRODUCTION

Flant growth regulators may be considered as adjuvants to existing control measures or as weed killers in their own right. It is well established that the major difficulty of controlling <u>A. repens</u> by cultivations and/or herbicides is due mainly to regeneration of weed from dormant buds on the underground rhizomes and it is in relation to this aspect of the biology of <u>A. repens</u> that growth regulators might have particular potential.

Growth regulatory responses which could improve existing weed control techniques include:

1) Release of buds from dormancy resulting in utilization of carbohydrates and increased underground surface area. This would render the plant more vulnerable to cultivations, the sprouting buds may provide a sink for herbicides and more surface area for contact with soil-applied compounds;

2) Alteration of ratio of shoots to rhizomes resulting in the development of shoots at the expense of rhizomes, thus increasing the target for foliage-applied herbicides. The opposite effect i.e. increased rhizome production, would only have control potential if no new photosynthetic tissue developed and the buds on the rhizomes continued to sprout until the carbohydrate reserve was depleted;

3) Development of rhizomes above ground so that the rhizome would be a ready target for herbicides, surface cultivations, and desiccation;

4) Modification of cuticle condition and stomatal opening so that penetration of foliage-applied herbicides is improved. Concurrently transpiration would be

increased favouring the uptake of soil-applied herbicide and opportunity for desiccation of tissues would be facilitated;

5) General toxicity both to buds on the underground rhizome and to the shoot, thereby allowing the growth regulator to be used alone for the control of <u>A. repens</u>.

The objective of the experiments described here was to evaluate the suitability of a number of growth regulators as adjuvants and/or herbicides for <u>A. repens</u> control following application to the foliage and soil. In addition some of the factors which affect performance such as temperature and time of application of an adjuvant in relation to the herbicide are considered.

METHOD AND MATERIALS

Single node 2.5 cm rhizome fragments of <u>Agropyron repens</u> L. (Beauv) clone 31 (Headington clone) were grown in Levington compost in 9 cm pots in a glasshouse until they reached the 5-6 leaf stage with one to two tillers and 1-4 cm of rhizome.

The glasshouse controls were set to give 16° C⁺ 5. The temperature rarely fell below this but in warm weather often exceeded 21° C. Humidity ranged between 40-80% R.H. The plants were grown in the glasshouse after herbicide application except in the experiment reported in Table 2 when plants were kept in controlled environment cabinets set up as follows:

Tempe day,	erature /night	% Relativ day/	night	Water Vapo deficit (day,	our Pressure (leaf/air) /night
20	15	92	89	1.38	1.41
10	5	85	85	1.40	1.00

The growth regulators and herbicides, all with 0.2% v/v Tergitol NFX wetter, were applied to the foliage with a laboratory pot sprayer set to deliver 352 1/hm at 2.11 kg/cm pressure. Soil applications were made as a high volume drench of 10 ml/pot. To ensure that only the foliage was treated with herbicide in shootapplied treatments the compost surface was covered with peat before spraying and the latter removed subsequently. When growth regulators and herbicides were applied 'simultaneously' the compounds were not mixed in the spray tank, but in each case the herbicide was applied first followed after a few minutes by the growth regulator. Generally the plants were kept in the glasshouse or growth cabinets for approximately 6 weeks and then harvested. Following assessments 2 to 6 below, the rhizomes were replanted and grown in the glasshouse for a further 6 weeks. The plants in each experiment and environment were set up in two randomised blocks each containing two replicate pots per treatment.

The following assessments were made: 1) type of symptoms; 2) number of shoots; 3) fresh weight of shoots; 4) fresh weight of rhizomes; 5) total number of nodes; 6) number of sprouted nodes; 7) regrowth shoot number.

RESULTS

Fig. 1 summarises two experiments and in order to make a direct comparison between all the compounds the results have been expressed as percent of controls. In general the chemicals were most active as foliage-applied sprays. The effects of B-995 (succinic acid-2,2-dimethyl hydrazide), Du 17623 (7-chloro-4-ethoxycarbonylmethoxy-5-methyl-2,1,3-benzothiodiazole) and NIA 10637 (ethyl hydrogen 1propylphonate) treatments on rhizome weight and total node number were not of



Fig. 1. Effect of eight growth regulators on the development of Agropyron repens

Percent of control

Horizontal bars are means of 4 replicates each of 2, 4 and 8 kg/ha (except TIBA, used only at 4 kg/ha) applied to foliage (F) and soil (S) harvested 6 weeks after treatment

sufficient magnitude to justify further investigation. The cytokinin 6-benzylamiropurine, which induces sprouting in <u>Cyperus rotundus</u> (Parker and Dear, 1972), was the least effective of the compounds tested even when used as a 200 ppm foliar dip.

MH applied to the foliege fulfilled the requirements of a growth regulator/weed killer as shoots, rhizomes and nodes were all reduced and at 4 and 6 kg/ha the plants were killed by the time of the regrowth assessment. At 2 kg/ha MH reduced the shoots in size but the number was increased at six weeks and regrowth after 12 weeks was more prolific than the controls. TIBA and chlorflurecol are similar in the morphological resuonse they induce but chlorflurecol is more active. Both compounds suppressed apical dominance and released bucs from dormancy with subsequent development of leafy shoots with a 'cork-screw' appearance and lacking normal geophototropic responses. While chlorflurecol- and TIBA-treated plants had more shoots their weight and size were reduced and this is a reflection of the substantially reduced rhizome system. At 6 weeks after application of chlorflurecol the number of nodes was greatly reduced at 4 and 8 kg/ha and the regrowth 6 weeks after replanting the rhizomes consisted of 10 and 8 small 'cork-screw' shoots respectively compared with 38 normal shoots on the control. Chlorflurecol at 2 kg/ha increased the number of shoots over 2-fold initially but by the time of the regrowth assessment the plants were as controls. Thus chlorflurecol showed herbicidal effects especially at the higher rates and was more effective following soil application than MH. In marked contrast to chlorflurecol, 2-chloroethylphosphonic acid (G 996) had little effect. but was most effective at releasing buds which invariably had rhizome like characteristics even when growing above the ground (Caseley 1970). The number of shoots including 'aerial rhizomes' was doubled with 2 kg/ha and 4 kg/ha and trebled with 8 kg/ha and the latter rate increased rhizome fresh weights 34% over the controls.

The data in Table 1 was recorded 10 weeks after application of the chemicals in July compared with six weeks for that in Fig. 1 and after this period the influence of G 996 on shoot number had disappeared. Furthermore the high temperatures in the

Treatment	kg/ha	Shoot No.	Shoot Wt. g.	Rhizome Wt. g.	Total Number of Nodes	Number of Sprouted Nodes
Control		26.4 (1.6)	18.4 (0.7)	12.4 (1.1)	65.8 (6.3)	24.5 (3.7)
Chlorflurecol	0.5	27.3 (2.3)	18.8 (0.4)	12.2 (1.4)	68.5 (7.6)	23.5 (1.5)
G 996	2.0	24.3 (3.6)	22.8 (3.9)	14.1 (1.1)	101.8 (12.5)	28.5 (6.3)
Paraquat	0.13	12.8 (4.9)	4.6 (1.8)	1.1 (0.3)	5.0 (2.6)	2.0 (2.0)
Paraquat + Chlorflurecol	0.13	0.3	1.4 (0.4)	1.2 (0.1)	4.8 (1.8)	2.5 (1.0)
Paraquat + G 996	0.13	15.8 (1.1)	8.9 (1.7)	3.3 (1.1)	20.8 (9.0)	10.8 (6.2)

Table 1 Effect of chlorflurecol and G 996 on paraquat toxicity to Agropyron repens

Each mean is of 4 replicates for treatments and 12 replicates for controls and the standard errors are shown in brackets. glasshouse (21° mean weekly, max. 29°C) during the experiment resulted in rapid manifestation of G 996 symptoms and a subsequent shortened period for their disappearance. This is in agreement with unpublished results obtained in controlled environment experiment at WRC where temperature was a variable. While the shoot number of G 996 treated plants was as control, the total number of nodes was significantly increased and combination with paraguat resulted in a decreased phytotoxicity. Chlorflurecol at 0.5 kg/ha induced slight loss of geo/phototropic response in the weeks immediately following application, but had no prolonged significant effect. However, this chemical enhanced the performance of paraquat and 3 out of 4 plants were killed compared with one plant for paraquat alone. In an experiment started in May when the glasshouse was cooler chlorflurecol enhanced the paraquat activity less and data from an experiment in controlled environment cabinets where temperature was the only variable are shown in Table 2. These results indicate that chlorflurecol enhanced paraquat performance under warm conditions, but reduced paraguat activity at low temperatures.

1.8	DT	e	2	
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Treatment	kg/ha	Shoo 10 ⁰ C	t No. 20 ⁰ C	Rhizon 10 ⁰ C	wt.g. 20 [°] C	Total N of No 10°C	des 20°C	Numbe Sproute 10°C	er of ed Nodes 20°C
Paraquat	0.13	3.3 (3.3)	46.6 (7.4)	2.5 (1.8)	16.9 (5.8)	0	21.4 (9.1)	0	30.8 (13.2)
Chlorflurecol	1.0	169.1 (13.0)	141.8 (9.4)	84.0 (29.5)	49.4 (6.6)	95.2 (33.4)	33.3 (6.9)	121.7 (33.3)	143.6 (30.1)
Paraquat + Chlorflurecol	0.13	6.5 (3.8)	10.6 (6.4)	4.2 (1.7)	3.1 (1.4)	3.2 (3.2)	2.8 (2.1)	8.7 (8.7)	15.4 (15.4)

Means are expressed as percent of control for each temperature and standard errors are shown in brackets

The data in Table 3 illustrate the effect of the timing of application of dalapon in relation to chlorflurecol. For this combination of chemicals the application of the herbicide first followed by the growth regulator resulted in the greatest activity. Similar experiments with paraquat and chlorflurecol did not show any advantage with sequential applications.

DISCUSSION

Compounds with growth regulatory activity are screened regularly at WRO for the ability to release dormant buds from apical dominance in isolated rhizome systems (Chancellor and Leakey, 1972). Some representative compounds which showed low or no activity in this test were included in the whole plant studies as it was found with G 996 that foliage attached to the rhizome was necessary for activity (Chancellor, 1970). However, in the case of B 995, 6-benzylaminopurine, Du 17623 and NIA 10637, the results of the whole plant studies (Fig. 1) are in general agreement with Chancellor's findings and none of the responses were of a magnitude to justify further studies.

MH was included as a reference growth regulator herbicide as it was first suggested as a herbicide for <u>A. repens</u> in 1950 (Crafts <u>et al.</u>) and has been approved

Treatment	kg/ha	Shoot No.	Shoot Wt. g.	Rhizome Wt. g.	Total Number of Nodes	Number of Sprouted Nodes
Control	0	11.1 (1.0)	18.3 (1.6)	7.0 (1.0)	30.5 (5.3)	3.5 (0.9)
Chlorflurecol	0.5	24.5 (1.4)	11.0 (0.7)	6.9 (0.5)	16.3 (2.0)	(5.3 (1.1)
Dalapon	4.0	23.8 (3.3)	5.1 (1.1)	5.0 (0.7)	21.0 (6.9)	5.3 (2.3)
Dalapon simultaneous + Chlorflurecol	4.0 + 0.5	11.8 (2.1)	3.1 (0.6)	4.4 (1.1)	20.0 (4.3)	4.8 (2.8)
Dalapon 48 h <u>after</u> Chlorflurecol	4.0 + 0.5	12.3 (0.8)	4.5 (0.4)	4.7 (0.8)	16.3 (6.3)	3.5 (1.0)
Dalapon 48 h <u>before</u> Chlorflurecol	4.0 + 0.5	6.3 (0.5)	1.6 (0.2)	1.3 (0.1)	1.0 (0.0)	0.8 (0.3)

Table 3

Effect of timing of application of dalapon in relation to chlorflurecol

Each mean is of 4 replicates for treatments and 12 replicates for controls and the standard errors are shown in brackets.

as a post-harvest treatment in Finland since 1966 (Rantanen and Lallukka, 1968). The only compounds which showed herbicidal activity approaching that of MH were chlorflurecol and TIBA. Although toxic to <u>A. repens</u> at the higher rates chlorflurecol was ultimately less effective than MH. However chlorflurecol performed well in field experiments at WRO, autumn application suppressing growth through the spring (Holrcyd 1972).

TIBA induced the same morphological effects as chlorflurecol, but its activity was lower and this is in agreement with results reported for other species (Schneider, 1970). Chlorflurecol has more potential as an additive to other herbicides, than as a phytotoxic agent in its own right. It releases buds from apical dominance and increases the number of shoots in relation to rhizomes and may induce other responses in the plant which are conducive to control. G 996 had a low phytotoxicity at the doses employed (Fig. 1), but had considerable potential for releasing buds from apical dominance.

Experiments were conducted to exploit the dormancy breaking ability of chlorflurecol and G 996 (Table 1). The failure of G 996 to enhance paraquat toxicity while chlorflurecol achieved some success may be partly attributed to the nature of the growth from the bud following release from dormancy. In the case of G 996 this was always a rhizome which bore more buds and as these developed above and below ground the photosynthetic capacity was only partially reduced. Chlorflurecol treatment, on the other hand, always resulted in the production of deformed 'spiralled' leaves. The relative regenerative capacity is reflected in the almost complete absence of nodes following chlorflurecol treatment and the threefold increase with G 996 (Fig. 1). It was envisaged that prolific rhizome production with G 996 might improve control of <u>A. repens</u> used in conjunction with a soil-acting herbicide. In an experiment where well established <u>A. repens</u> plants, with several coils of rhizome around the exterior of the soil mass, were transferred to larger pots containing pronamide (4 kg/ha) treated soil, G 996 at 4 kg/ha had no effect on the performance of the pronamide. The results reported here and field experiments with aminotriazole, asulam, dalapon, sodium 2,2,3,3-tetrafluoropropionate and paraquat in combination with G 996 indicate that the herbicide without G 996 generally gave better control (Blair, 1972). These results suggest that dormancy breaking per se does not necessarily enhance control. It would appear that the growing buds do not become a sink for the soil or foliage-applied herbicides and the nature of the subsequent growth seems to be of major importance. Alternatively it is possible that G 996 interferes with the transport and/or activity of the herbicide.

Chlorflurecol releases buds from apical dominance and these tend to develop into leafy shoots and in an outside pot experiment chlorflurecol improved the control of <u>A. repens</u> by aminotrizole and dalapon (Parker and Richardson, 1972). However herbicide performance is not always enhanced and temperature (Table 2) and other factors may be of considerable importance. Environmental factors are known to affect endogenous growth regulator levels e.g. moisture stress increases abscisic acid (Wright and Hiron, 1969), and since chlorflurecol acts via the endogenous regulator system of the plant (Schneider, 1970) it is not surprising that the performance of this chemical is influenced by aerial and soil conditions. The importance of the timing of herbicide versus the growth regulator application may be related to uptake and transport and the endogenous regulators system may also be involved.

The results presented here indicate that growth regulators can enhance herbicide performance, but at present not enough is known of the factors that control the effectiveness of the operation. Either these must be elucidated or new compounds more tolerant of variation in environmental factors introduced to allow the successful field exploitation of this type of technique.

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THE EFFECT OF SOME PLANT GROWTH REGULATORS ON THE SPROUTING OF CYPERUS ROTUNDUS AND ITS RESPONSE TO HERBICIDES

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<u>Summary</u> Thirty-eight plant growth regulators were tested for their ability to increase sprouting of <u>Cyperus rotundus</u> tubers. The cytokinins were very active in promoting extra sprouting, while chlorflurecol, naptalam, TIBA and a few other compounds were active to a lesser degree. Attempts to increase the susceptibility of <u>C. rotundus</u> to herbicides by the use of 6-benzylaminopurine (BA) and chlorflurecol have been generally disappointing but indications of some useful interaction between BA and terbacil were observed. Pre-treatment with gibberellins A₂ and A₄₊₇ caused tubers of <u>C. rotundus</u> to the soil surface.

INTRODUCTION

The programme of work described, has been based on the assumption that problem perennial weeds such as <u>Cyperus rotundus</u> owe much of their success to the abundance of dormant buds on their underground systems which enable them to recover rapidly after chemical or mechanical damage. It was postulated that if the majority of these buds could be brought into active growth, then the plant would be more susceptible to control measures, especially by herbicides.

The underground system of <u>C. rotundus</u> consists of chains of tubers linked by rhizomes 10-20 cm long. The tubers each have up to 10 viable buds most of which remain dormant. One or two lateral buds, usually on the lower side, grow out to form continuation rhizome, but all other lateral buds stay dormant in the intact system. The apical bud may or may not grow out to form a leafy shoot and the term 'dormant tuber' is applied to those in which the apical bud has remained dormant. In the intact system, therefore, at least 75% of the tuber buds remain dormant. When the tuber system is broken up, most tubers immediately sprout and several buds on each tuber may begin to grow, but as in seven-node rhizome sections of <u>Agropyron repens</u> (Chancellor, in preparation) a dominance system is soon re-established and only one or two continue to develop to the point of emergence from the soil.

As an index of the ability of a compound to interfere with this 'dormancy' system, we have measured the number of sprouts which continue to grow on each tuber following treatment with the chemical. Where high activity was indicated the compound was tested further to assess its capacity to overcome the dormancy of apical or lateral buds of the tubers in the intact plant system. Subsequently, the most active compounds were used in an attempt to achieve our ultimate objective of increasing the susceptibility of the weed to herbicide.

One further parameter was studied, namely depth of basal bulb formation. When an isolated tuber is planted, the emerging shoot or shoots form a swelling or 'basal bulb' very approximately half way between the parent tuber and the soil surface. If the tuber sprouts from a relatively great depth, the basal bulb and hence the apical meristem and the origin of the primary rhizomes will be placed several cm deep in the soil and may not be so readily affected by a herbicide applied to the soil surface or only shallowly incorporated. Therefore, any compounds which would cause the basal bulb to be formed nearer the soil surface were of interest.

METHODS AND MATERIALS

The stocks of <u>C. rotundus</u> used as a source of tubers for these experiments were originally obtained from Rhodesia. Plants were grown in the glasshouse in plain loam soil at temperatures ranging from $18-27^{\circ}$ C. Supplementary light from fluorescent tubes was provided in the winter and plants were fed regularly with a liquid fertilizer. For work on isolated tubers, dormant tubers were separated from stock plants. 9-15 months old, and washed in tap water before treatment.

The tubers were treated and incubated in one of three different ways:

(a) immersion for 24 h in solutions of growth regulator (30 tubers per 150 ml solution) in stoppered conical flasks in the dark at 26 ± 1^{9} C. followed by washing in de-ionised water and placement in petri dishes. Five tubers were placed in each dish on four layers of Whatman No. 1 filter paper moistened with 0 ml distilled water and the dishes were enclosed in polyethylene bags to reduce moisture loss and were incubated in the dark at 26° C. There were usually three replicate dishes per treatment and observations were made twice weekly for up to three weeks on the number of tubers sprouted and the number of actively growing sprouts per tuber. The main assessment was at 10 days, when about 90° of tubers had sprouted in controls and each had 2-3 active sprouts.

(b) unsoaked tubers placed directly into petri dishes in which the filter papers were moistened with the growth regulator solution instead of distilled water. This is referred to as 'continuous exposure'. Assessments were as for method (a).

(c) (not used in the earliest experiments) tubers soaked for 24 h as (a) but then planted directly into loam soil in 8.5 cm diameter pots and grown under normal glasshouse conditions. Tuber numbers and replication were as for method (a). Numbers of emerged shoots were counted weekly up to 5-7 weeks when the plants were harvested. The pattern of rhizome growth was then assessed for any abnormality and the foliage, roots and tubers plus rhizome were dried and weighed separately. Tubers were normally planted 0 cm deep and the depth of the basal bulb was measured at the time of harvest but for some compounds information was obtained from special experiments in which a similar procedure was used.

All compounds were tested at three concentrations as thought appropriate but the special experiments on depth of basal bulb usually included only one dose of each compound. Formulated compounds were used when available but BA, kinetin. IAA, GA₃, salicylic acid and p-coumaric acid required dissolving in 5% HCl or 5% NaC₂C₃ followed by appropriate neutralisation. Acetone was used to help dissolve² SD 8350 and methanol for GA₄₊₇, while the DU compounds were dissolved in xylene and emulsified using Triton B 1956.

For the further experiments on interactions with herbicides, the growth regulator pre-treatment of isolated tubers was done by method (c), usually using a solution of BA at 50 mg/l for 24 h.

Application to established plants was done by dipping, drenching or spraying. For the dipping treatments, the plant system plus soil mass were removed from the pots and lowered into 200 ml of solution. The plant was held in such a way that the solution wetted all the rhizomes and tubers on the outside of the soil mass and after about 20 seconds it was removed. having absorbed up to 100 ml of solution. Drenching consisted of flooding a solution on to the soil surface at 20 ml per 8.5 cm diameter pot.

Herbicides were applied by a laboratory sprayer applying 352 1/ha and postemergence sprays always included a wetting agent. Pre-planting treatments were sprayed on to an 8 cm deep layer of soil, in tins, which was then thoroughly mixed before being placed in 8.5 or 12.5 cm diameter pots and the tubers planted. The soil in all cases was a sandy loam from one of the fields at Begbroke Hill. After postemergence spraying, 24 h elapsed before a thorough overhead watering of the foliage. Fresh weights of shoot systems were measured 6-12 weeks after post-emergence sprays. Underground systems were not assessed in detail but were generally observed to be affected to about the same extent as the foliage.

RESULTS

1. Initial 'screening' on isolated tubers

The cytokinins kinetin, BA and SD &339 were particularly effective in promoting extra sprouting of isolated tubers of <u>C. rotundus</u> (Table 1), these results being in close agreement with those independently obtained by Teo <u>et al</u>. (1971). Not only were more sprouts observed on tubers kept in petri dishes, but when pre-soaked tubers were planted into soil a significantly increased number of shoots emerged. Table 2 shows the results for two of the compcunds in more detail. SD &339 was the most active compound tested. In further work, pre-treatment with BA at 50 mg/l consistently increased the number of emerged shoots, even when treatment was delayed for five days when sprouting had begun, in comparison with control tubers which were either unsoaked or soaked only in water.

Chlorflurecol also increased sprouting considerably but in contrast to the cytokinin action other effects such as stunting and loss of geotropic response were observed. Naptalam and TIBA also induced moderate extra sprouting but were much less active than chlorflurecol. A number of other compounds, in particular the herbicidal ether compounds, MO 338, nitrofen and fluorodifen, caused some extra sprouting in petri dishes but this was not always apparent when tubers were planted in soil. 'Continuous exposure' usually resulted in slightly less response than the soaking treatments, perhaps because only part of the tuber was in contact with the solution.

The depth of basal bulb formation in <u>Cyperus rotundus</u> was affected by the gibberellins only. GA_3 caused very marked lengthening of the stem tissue below the basal bulb and a pre-treatment with 50 mg/l before planting tubers at 7 cm depth resulted in the basal bulb forming at 1 cm depth as against 3 cm in untreated plants. The mixture GA_{4+7} was less active.

Table 3 summarises the inactive compounds tested.

2. Effects on the intact plant system

Table 4 shows the effects of some of the more active compounds on mature intact <u>C. rotundus</u> plants with numerous 'dormant' tubers. Plants grown in 12 cm diameter plastic pots for four months were either sprayed in a conventional manner or were dipped into solutions of the growth regulators.

Before treatment a number of mature dormant tubers (about six per pot) were marked and one week later the state of these tubers was observed. In controls, 20%had sprouted, perhaps partly due to the manipulations and disturbance, while BA treatment at 50 mg/l caused over 80% to sprout and many of the tubers showed multiple sprouting. In addition to the apical buds, lateral buds on the upper side of the tuber also developed. These are never seen to sprout in the intact plant system. Chlorflurecol-methyl had a distinct effect, both as a din and as a toliage spray. 2-Chloroethylphosphonic acid was active as a dip at high concentrations but had no effect as a foliage spray even at the high dose of 8 kg/ha. 2,4-D and sodium cacedy-late had no positive effect, whilst GA caused an apparent intensification of dormancy at both 10 and 10C mg/l. This is consistent with results from petri dish tests where reduced numbers of sprouts per tuber were observed. Some similar indications were also obtained by Teo et al. (1971).

Table 1

Compounds having some activity on the sprouting or the depth of basal bulb formation in C. rotundus

Response to growth regulator at doses indicated in brackets (mg/1)

C. rotundus					
Sprouting		Depth	of basal bulb		
++ (10-	1(()	C	(50)		
+ (10) ++ (50-100)	Ċ.	(50)		
+ (100)	C	(50)		
+ (1)		tox	ic at dose sted (5)		
+ (1-1	ec)	C	(50)		
+ (100)	C.	(1-100)		
+ (100)	0	(1-100)		
+ (100)	c	(1-100)		
+ (100)	c	(1-100)		
+ (100)	0	(1-100)		
+ (500)	С	(250)		
+ (500)	O	(250)		
+ (500)	0	(250)		
0 (1-1	(0)	++	(50)		
C (1-1	00)	•	(50-100)		
	Sprout ++ (10- + (10) ++ (+ (10) + (1-1) + (1-1) + (100 + (100 + (100 + (100 + (100 + (100 + (500 + (500 + (500 0 (1-1) 0 (1-1)	$\frac{C. FG}{Sprouting}$ ++ (10-100) + (10) ++ (50-100) + (100) + (100) + (1-100) + (1-100) + (100) + (100) + (100) + (100) + (100) + (100) + (500) + (500) + (500) 0 (1-100) 0 (1-100)	$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$		

Notes: + = moderate response ++ = pronounced response

0 = no positive affect

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SD 8339	=	6-benzylamino-9-(tetrahydro-2-pyranyl)-9H-purine
BA	=	6-benzylaminopurine
kinetin	=	6-furfurylaminopurine
TIBA	=	2,3,5-triiodobenzoic acid
DU 13594	=	5,7-dichloro-4-ethoxycarbonylmethoxy-2,1,3-benzothiadiazole
DU 16333	=	5-chloro-6-ethoxycarbonylmethoxy-2,1,3-benzothiadiazole
PRB &	=	2-(B-chloro-B-cyanoethyl)-b-chlorotoluene
MBR 6033	=	3-trifluoromethylsulfonamido-p-acetotoluidide
MO 338	=	p-nitrophenyl-2,4,6-trichlorophenyl ether
GA, A 7	=	gibberellins

		Sprouts/tub	er on <u>C. rotundus</u> af	ter 10 days
Compound	Dose mg/1	Soak + dish	Soak + soil	Continuous
SD 8339	1	3.57	1.87	n.t.
	10	5.00	3.60	3.60
	100	5.21	4.53	n.t.
MO 338	5	3.47	1,73	n.t.
	50	3.27	1.80	3.29
	500	4.27	2.00	n.t.
control	0	2.40	1.67	2.33
S.E.		± 0.32	<u>+</u> 0.34	± 0.32

Table 2

n.t. = not treated

Table 3

Compounds	having no	positive	activity	in the	systems te	sted

Compound	Chemical name	Doses tested (mg/1)
AMO 1618	2-isopropy1-4-dimethy1amino-5-methy1pheny1-1- piperidine carboxy1ate	1-100
B 995	N-dimethylaminosuccinamic acid	1-100
2-chloroethanol		1,000-
		25,000
chlormequat	2-chloroethyltrimethylammonium chloride	1-100
chlorphonium chloride	tributy1-2,4-dichlorobenzy1phosphonium chloride	1-100
coumarin		1-100
p-coumaric acid		1-100
CP 41845	N,N-bis(phosphonomethy1)glycine	1-100
DU 17623	7-chloro-4-ethoxycarbonylmethoxy-2,1,3-benzo- thiadiazole	1-100
glycocholic acid	(as Na salt)	1-100
glyphosate	N-phosphonomethyl glycine	0.1-100
G 996	2-chloroethylphosphonic acid	1-100
IAA	indole-3-acetic acid	1-100
maleic hydrazide	1,2-dihydropyridazine-3,6-dione	10-1.000
NIA 10637	ethyl hydrogen-l-propylphosphonate	1-100
NC 9565	<pre>([3-(p-chloropheny1)-1,2,4-oxadiazo1-5-y1]thio) acetic acid</pre>	1-100
NC 9634	[(3-pheny1-1,2,4-thiadiazo1-5-y1)thio]acetic acid	1-100
salicylic acid		1-100
thiourea		1-100
U 27658	3,4,5-tribromopyrazole-1-acetic acid	0.1-10
U 29449	ethyl 2-(dimethylamino-2-[(4-chloro-o-tolyl) oxy]propionate	0.1-10
U 29722	$3, 4, 5$ -tribromo- α -methylpyrazole-1-acetic acid	1
WL 17731	ethyl 2-(N-benzoy1-3,4-dichloroaniline)propionate	1-100

Т	a	b	1	e	4
_	_	_	_	_	_

	% sprouting of marked dormant tubers 7 days after treatment					
Compound	dip mg/1	73	spray kg/ha	78		
ВА	10 50	10.5 83.3	n.t. n.t.	1		
2-chloroethylphosphonic acid	250 1000	5.8 55.0	8	15.7		
chlorflurecol methyl	10	52.6	2	61.0		
naptalam	10	37.5	4	29.4		
2,4-D amine	10	16.6	4	29.4		
sodium cacodylate	10	23.6	4	5.5		
GA ₃	10 100	0	n.t. n.t.	Ċ.		
Control	С	20.0				
S.E.		± 14.3		± 14.3		

Effects of several growth regulators on the sprouting of dormant tubers on mature intact C. rotundus plants

n.t. = not treated

3. Combinations with herbicides

Since BA was most effective in promoting extra sprouting, experiments were conducted to assess interactions of this chemical with herbicides. In the first two tests tubers of <u>C. rotundus</u> were soaked for 24 h in BA at 50 mg/l and either planted into herbicide treated soil (five per pot) or grown on in soil for treatment with foliage-applied herbicides after 2-3 weeks.

The first experiment showed the intended effects of BA, sprouts per pot after 20 days being more than trebled from 6 to 21, but the responses to EPTC and sodium cacodylate were unaffected and the activity of paraquat appeared to be substantially less on the BA pre-treated plants.

In a second experiment, partially presented in Table 5, there was less protective effect of BA pre-treatment against subsequent paraguat spraying but there was again no indication that the pre-treatment and resultant extra sprouting made the weed any more susceptible to EPTC, metflurazone or chlorthiamid as soil-incorporated treatments or to sodium cacodylate or MSMA as post-emergence sprays. There was a modest increase in susceptibility to 2,4-D and a striking increase in susceptibility to terbacil at 0.5 kg/ha.

In a further experiment, summarised in Table 0, there was again some sign of synergism between terbacil and a BA pre-treatment but it was considerably less striking. There was no useful interaction between terbacil pre-planting and chlorflurecol. either as a pre-planting or post-emergence application.

The post-emergence treatments with BA and chlorflurecol both induced extra sprouting of the tubers but did not increase the susceptibility of the plants to post-emergence applications of terbacil. In this experiment, 25 day-old plants were treated but in another test, much older plants treated with similar combinations also failed to respond.

Herbicide	kg/ha	Fresh wt. of s No BA	with BA pre-treatment
Decemerance			
terbacil	0.25 0.5 1.0	87.1 105.0 13.1	94.2 18.1 1.3
EPTC	0.25 0.5	81.7 83.4	99.2 75.7
Post-emergence			
2,4-D	1.0	83.9 62.8	64.9 39.8
paraquat	0.2	57.5 15.1	42.8 24.8
Control	C	100 (6.9)*	100.1 (20.3)*
S.E.		<u>+</u> 8.2	± 8.2

Table 5

Response of C. rotundus to four herbicides, with and without pre-treatment with b-benzylaminopurine at 50 mg/1

* Shoot numbers/pot at 14 days

Table 6 indicates that there were antagonistic rather than useful interactions between glyphosate and both BA and chlorflurecol.

In one further experiment a dip treatment of BA at 50 mg/l was used to cause extra sprouting of 9 week-old <u>C. rotundus</u> plants, but there was no increased susceptibility to metflurazone or sodium cacodylate.

Table 6

The interaction of two growth regulators with two herbicides on C. rotundus

Fresh wt. of <u>C. rotundus</u> foliage as % of untreated control

			+ BA	+ BA	+ chlor	flurecol post-emergence	
Herbicide	kg/ha	alone	50 mg/1	100 mg/1	0.4 kg/ha	0.8 kg/ha	
terbacil (pre-planting)	0.4	115.5 40.5	60.6 22.3	n.t. n.t.	88.5 46.9	94.1 44.1	
glyphosate (post-emergence)	0.5	100.8	126.1 45.5	94.0 4.2	n.t. n.t.	72.6 36.6	
Control	0	100.0	100.7	113.4	82.7	81.4	
S.E.		+ 7.4	<u>+</u> 7.4	± 7.4	<u>+</u> 7.4	± 7.4	

n.t. = not treated

DISCUSSION

This series of experiments has illustrated the ability of the cytokinins and to a lesser extent the morphactins and some other compounds to overcome correlative inhibition of bud growth within the Cyperus rotundus tuber system. At present, the cytokinins are too expensive to be considered for use in the field and the soaking pre-treatment used in many of the experiments is a completely artificial and impractical type of treatment, but the aim has been to use them as tools to establish whether such dormancy breaking effects could lead to improved control of difficult perennial weeds. If there were great advantages then the search could continue for more inexpensive treatments. It had been envisaged that with the aid of a dormancy breaker the susceptibility of the weed should be increased in three different ways -(a) the active buds would be more effective sinks for herbicide translocation and would be readily killed leaving less viable dormant buds from which the plant could recover; (b) the extra sprout growth would deplete stored food reserves in the tuber more rapidly and hence reduce the ability of the plant to recover after herbicide treatment and (c) extra shoots would provide extra leaf area and allow greater absorption of translocated herbicides.

Unfortunately our experiments have shown very little evidence of increased susceptibility of <u>C. rotundus</u> to herbicide following increased sprouting of either young or well-established plants. The one possible exception has baen with terbacil where pre-planting treatments may have been more effective on BA pre-treated tubers. There has not, however, been any sign of useful interaction on well-established plants for which improved control is particularly badly needed, and where it was hoped a dormancy breaking chemical would substitute for the dormancy breaking effect of cultivation.

The results so far are disappointing, but the interaction with terbacil and other photosynthetic inhibitor herbicides and the physiological implications of the activity of the cytokinins are being explored further.

The activity of the gibberellins in causing more shallow formation of the basal bulb is potentially interesting and further relevant investigations are in progress.

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PLANT GROWTH REGULATORS AS A PRETREATMENT FOR SUGAR BEET SEEDS

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<u>Summary</u> Sugar beet fruits were pretreated by steeping in water or aqueous solutions of the growth regulators gibberellic acid, kinetin or 6-benzylaminopurine to investigate the possibilities of improving field emergence and accelerating early growth. Steeping in water for 24 hours increased the emergence percentages, shortened the interval between sowing and emergence and increased average seedling weights. Additional responses due to the inclusion of growth regulators in the steep were usually smaller than the initial response to steeping in water, with gibberellic acid at 100 ppm, kinetin at 50-100 ppm and 6-benzylaminopurine at l'-10 ppm the most promising treatments. Pretreatment was particularly effective on the underdeveloped seeds in a sample and improved the performance of early harvested fruits. The greatest responses were obtained in fruits of a large size grade.

INTRODUCTION

The sugar beet 'seed' of commerce is a fruit consisting of a small embryo with some perispermic reserves, located within a corky pericarp. Fruits, as harvested from the mother plant, weigh from 2-30 mg and contain seeds from 0.5-6.0 mg. Starting with such limited growing 'capital' seedlings are slow to emerge and develop and are therefore very susceptible to weed competition. Moreover, the proportion of fruits which produce seedlings varies considerably depending on seed lot and soil conditions so that gappy, and therefore potentially weedy, plant stands occur. Radiography (Longden, et al, 1971) has shown that at least three categories of fruits can be distinguished: seedless fruits, fruits with seeds which appear shrivelled or underdeveloped and those which appear fully developed. Seedless fruits must be eliminated as far as possible during physical processing, but many partially, and even fully, developed seeds also fail to produce seedlings. The objective of the present work was to pretreat fruit stocks to accelerate early growth, thereby minimising gappiness within the crop, and creating an early leaf cover to smother weeds and increase yield.

A number of growth regulators have been used to pretreat seeds, particularly those of vegetable crops (Table 1). Auxins and gibberellins are known to stimulate cell expansion, while the cytokinins may regulate cell division. Gibberellins can stimulate stem elongation in some dwarf or rosette plants, promote a-amylase activity in barley grains and break dormancy in some light sensitive seeds such as lettuce. Both kinetin (6-furfurylaminopurine) and 6-benzylaminopurine (BA) will break dormancy in lettuce seeds by possibly acting as antagonists of germination inhibitors but they may inhibit auxin activity at high concentrations. Wren (unpublished data) has shown that sugar beet seedlings respond to mixtures of indolyl

Workers	Crop	Treatment	Response
Singh and Dohare (1964)	Radish	NAA. 10-80 ppm	10-20 ppm increased emergence % 20-80 ppm increased root size
Sinha (1969)	Rice	NAA. 50, 75, 100 ppm 24 hour soak IAA.	75 ppm increased dry matter production
Saar (1968)	Radish	GA. 10 ppm	Increased dry matter production; small seed benefited most
Teare, Law and Wilson (1970)	Peas	GA. 4g/45 kg peas	Faster emergence at low temperatures
Peterson (1958)	Sugar beet	GA. 1, 10, 100 ppm spray	No effect
Doxtator (1958)	Sugar beet	GA. 100, 1000 ppm dust or 4 hour soak	No effect
Snyder (1959)	Sugar beet	GA. 250 ppm 1½ or 3 hour soak 125-250 ppm 18 hour soak	No effect
Jung and El-Fouly (1967)	Beta	GA. 18 hour soak	Increased emergence %
Vlasyuk <u>et al</u> (1967)	Maize	Kinetin 0.1-0.5 ppm 48 hour soak	Increased germination % and dry matter production
Makino <u>et al</u> (1969)	Radish	BA. 1 ppm	Increased dry matter production

Table 1

The response of seeds to pretreatment with growth regulating chemicals

acetic acid, gibberellic acid and kinetin in culture media, but no successful growthregulator pretreatment for seeds has been reported.

EXPERIMENTAL

1. Preliminary studies

Concentration response curves were determined for gibberellic acid (GA), as GA₃, kinetin (K) and 6-benzylaminopurine (BA). Twenty-five fruits were placed on two filter papers in a Petri dish, with 5 ml of aqueous growth regulator solution or distilled water, and germinated at 20°C in the dark. The optimum concentrations were found to be GA 100 ppm, kinetin 50 ppm and BA 10 ppm (Fig. 1).

A series of experiments followed in which pretreated fruits were sown in controlled environment rooms (20°C and 16 hour photoperiod) and the field. Fruits were steeped in aqueous solutions of the growth regulators (1:20 w/v) for 24 hours



at 20°C; Snyder (1959) and others had failed to find promotion of germination of sugar beet using shorter periods. The fruits were air-dried at 20°C for 36 hours before sowing. Two 'control' treatments were used, viz. completely untreated and steeped in water only. Table 2 shows typical data from a field experiment. The level of emergence was low; the fruit bulk contained 20 per cent seedless fruits and no fungicide was used. The most effective pretreatments nearly doubled the seedling weights.

Pretreatment effects in these preliminary experiments may be summarised as follows :-

Water Steeping the 'seed' in water usually accelerated emergence and increased the emergence percentage and average seedling dry weights. The origin of the improvement may be either in the diffusion and elimination from the pericarp of water soluble substances which inhibit germination or in the initial 'priming' processes which occur during the imbibition period and result in rapid germination and early growth.

Table 2

Effects of	pretreatment	on emer	gence in	the f	ield and	seed1:	ing dry	weights	
Treatment (ppm)	Untreated	Water	GA 100	GA 500	к 40	к 80	BA 10	BA 40	S.E.
Emergence %	25	25	29	29	33	25	27	18	±1.5
Dry weight/ seedling (mg)	685	679	1145	963	1147	1173	810	506	±131

<u>Gibberellins</u> The inclusion of gibberellic acid in the steep produced larger responses when fruits were sown in the field in spring than at 20°C in the controlled environment. It may be that GA stimulates growth more effectively at lower temperatures and this possibility is being investigated. No distinct optimum concentration was found. Seedlings were pale green with elongated hypocotyls and cotyledons.

<u>Cytokinins</u> Emergence percentages were improved in several cases with kinetin and again response was greater in the field. Optimum concentrations for kinetin were in the range 50-100 ppm, but for BA were in the range 1-10 ppm. At higher concentrations emergence was delayed and seedlings were stunted, although there was little effect on the overall emergence percentages.

<u>Fungicide</u> Ethyl mercury phosphate (E.M.P.) at 40 ppm for 20 minutes is a standard steep treatment for sugar beet fruits to control the seed-borne pathogen <u>Phoma betae</u> (Byford, 1963). In our experiments this treatment alone usually improved emergence percentages, without affecting seedling weights. However, when done after growth regulator pretreatment, although emergence percentage was improved the beneficial effects of pretreatment on seedling growth were diminished. It is possible that the concentration range of E.M.P. tolerated after pretreatment was much narrower.

The following experiments investigated the benefits of pretreatment of fruit lots known to differ in performance.

2. Pretreatment of small and large fruits

Radiography showed that small fruits (3.6-4.0 mm) contained smaller seeds than large fruits of the same stock (4.8-5.2 mm) and more of them were underdeveloped (35 and 27 per cent respectively). Six replicates, each of 100 fruits were pretreated and sown 2 cm deep in compost in seed trays which were arranged in controlled environment rooms (20°C and 16 hour photoperiod).

The main promotory effects were obtained by steeping in water alone (Table 3). The large fruits were responsive to a wider range of pretreatments and showed a higher magnitude of response than the small fruits, e.g. the most effective pretreatment increased average seedling weights from the large and small fruits by 43 and 28 per cent respectively. Fruits of this stock were radiographed after pretreatment then sown individually in predetermined positions to monitor their

Table 3

Pretreatment effects on fruits from two size grades

Size grade (mm)	Untreated	Water	GA 100	GA 1000	К 50	К 100	BA 10	BA 40	EMP	Water + EMP
3.6-4.0	44.8	54.5	53.5	55.2	54.2	56.2	59.8	52.2	50.1	62.3
4.8-5.2	70.5	84.0	78.3	84.0	82.2 (S.E.	86.3 ±2.93)	81.7	85.0	79.0	91.8
Mean	57.7	69.3	65.7	69.6	68.2 (S.E.	71.3 ±2.07)	70.8	68.6	64.5	77.1
Size grade (mm)	Untreated	Water	GA 100	GA 1000	к 50	К 100	BA 10	BA 40	EMP	Water + EMP
3.6-4.0	26.6	34.1	32.9	30.2	34.0	32.2	33.6	27.3	26.6	27.6
4.8-5.2	32.5	43.5	44.1	40.8	43.6 (s.e.	46.4 ±2.11)	41.9	37.2	32.1	32.7
Mean	29.6	38.8	38.5	35.5	38.8 (S.E.	39.3 ±1.49)	37.8	32.2	29.3	30.2

(a) Percentage emergence

Table 4

Radiography of pretreated fruits : percentage emergence from normal and underdeveloped seeds

	Untreated	Water	GA 100
Normal	52.4	84.8	70.0
Underdeveloped	39.1	77.4	71.6

performance. Pretreatment was particularly effective on seeds which appeared underdeveloped (Table 4). The lower level of response of the small fruits compared with that of the large (Table 3) suggests that more of the underdeveloped seeds within the large fruits were capable of responding, whereas many of the structures classified as underdeveloped seeds within the small fruits were simply incapable of developing whether pretreated or not. The proportion of fruits which failed to develop seedlings was not simply accounted for by those which appeared seedless; only 10-15 per cent of the small fruits and 4-5 per cent of the large fruits appeared seedless. An E.M.P. steep super-imposed upon a water steep-drying routine was the most effective treatment combination in improving emergence, the level of emergence approaching the maximum possible as indicated by radiography. The failure of the small fruits to attain this potential tends to reinforce the suggestion that many of the underdeveloped seeds were too small to develop or emerge from this depth.

3. Pretreatment of early and late harvested fruits

In a cool, wet summer seed crops ripen later and an unusually high proportion of the true seeds are not fully developed at harvest, so that low emergence percentages and seedling weights may result. This effect is most marked if the crop is harvested early and it is with the smaller fruits that such immaturity is likely to be most critical.

A preliminary experiment was carried out using fruits produced in the cool, wet 1968 season. Fruits from two harvest dates (5 and 19 September) and of two size grades (2.8-3.35 and 4.0-4.75 mm) were examined. The emergence percentages and seedling weights were improved by delaying harvesting until 19 September, particularly with the small fruits. Similar effects were not found in 1970 when crops ripened quickly. A radiographic comparison of the two fruit lots to determine the proportion of underdeveloped seeds has not yet been carried out.

Table 5

The effects of fruit size and harvest date in 1968 on the performance of seed

Fruit size	Emerg	ence %	Seedling dry weight (mg)		
	2.8-3.35	4.0-4.75	2.8-3.35	4.0-4.75	
Harvest date					
5 September	28.7	57.5	18.5	25.8	
19 September	35.5	56.3	25.4	33.1	
	(S.E.	±2.31)	(S.E.	±2.02)	

The 1968 fruits were pretreated and four replicates of fifty fruits were sown in compost in seed trays, which were arranged in controlled environment rooms (20°C and 16 hour photoperiod). The quantity of fruits in the grade 4.0-4.75 mm proved to be insufficient to allow completion of this experiment and the grade 3.35-4.00 mm was substituted.

Comparisons of the data in Table 5 and that for untreated fruits in Table 6 show that the 'large' grades used in the two experiments differed in performance, in that fruits used in the second experiment appeared more immature at the early harvest, giving lower emergence percentages and seedling dry weights.

In this experiment not all of the benefit from pretreatment was obtained by steeping in water alone. Growth regulator treatment produced additional responses and with the exception of BA 10 ppm, the degree of response to the different growth regulators was strikingly similar. It is possible that a combination of gibberellins and cytokinins might be more effective. Again large fruits were more responsive to pretreatment, particularly when harvested early.

		Pretreatme	nt of large	and smal	1 fruit	s from	early a	nd late		
			h	arvests	in 1968					
(a) Percentage emergence										
Fruit size		Harvest date	Untreated	Water	GA 100	GA 1000	BA l	BA 10	к 50	K 100
G	5	September	16.5	20.0	26.0	23.0	20.0	22.5	17.0	25.0
Small	19	September	29.5	33.0	31.0	29.0	38.5	31.0	25.5	38.5
					(S	.E. ±3.	76)			
-	5	September	25.0	31.0	44.5	41.5	45.0	33.5	44.5	34.0
Large	19	September	51.0	58.0	54.5	65.0	64.0	51.0	63.0	55.0
Mean			30.5	35.5	39.0	39.6	41.9	34.5	37.5	38.1
					(S	.E. ±1.	88)			_
a			(b) See	dling dr	y weigh	t (mg)				
Fruit size		Harvest date	Untreated	Water	GA 100	GA 1000	BA 1	BA 10	К 50	К 100
	5	September	10.8	14.8	16.7	14.5	15.0	14.1	15.2	16.0
Small	19	September	17.5	15.7	14.2	14.5	19.6	15.4	14.8	21.9
					(S	.E. ±1.	85)			
	5	September	15.8	19.8	21.3	20.1	19.2	14.7	22.0	18.2
Large	19	September	16.3	17.8	23.1	18.9	24.0	19.9	20.6	17.3
Mean			15.1	17.0	18.8	17.0	19.5	16.0	18.2	18.4
					(S	.E. ±0.	92)			

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As a first step towards understanding the mechanisms involved the prediction of responses, changes in the natural growth regular content of fruits are being determined as they age and increase in size.

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STIMULATION OF CELERY SEED GERMINATION WITH PLANT GROWTH REGULATORS

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Summary In laboratory tests, a mixture of the gibberellins A_4 and A_7 (GA₄/7) with some other plant growth regulators stimulated the germination of celery seed incubated at high temperature in the dark. The most satisfactory treatment, namely GA₄/7 and N-dimethylaminosuccinamic acid (Alar), was used in further experiments in the glasshouse and field. Germination and seedling uniformity were improved when seeds were soaked with dichloromethane solutions of this mixture and then dried before sowing.

INTRODUCTION

The seeds of some umbelliferous crops such as celery germinate more readily in the light particularly at temperatures above 20°C, and this factor may contribute to the difficulties encountered in both glasshouse- and field-sown crops (Robinson, 1954). Celery is not normally direct-sown since field emergence is erratic and seedling growth slow resulting in variable crop stands. Weed control can be difficult because post-emergence herbicide treatments are not recommended until seedlings reach the two-leaf stage, by which time the weed population may be well established. Palewitch <u>et al.</u> (1971) found that the light requirement of celery seeds could be eliminated by germinating them in solutions of a mixture of the gibberellins A_{\perp} and A_7 ($GA_{\perp}/7$) in combination with other growth regulators. The objective of the current work was to determine the most suitable treatment for stimulating the germination of dark-incubated celery seeds, and to investigate whether treating seeds with these growth regulators prior to sowing would improve seedling establishment in the field and thereby increase crop uniformity.

MATERIALS AND METHODS

Pure grade chemicals were used in all experiments with the exception of N-dimethylaminosuccinamic acid (Alar) which was obtained as a 98% a.i. wettable powder. The celery seeds cv. Lathom blanching were purchased in bulk from Asmer Seeds Ltd. In the laboratory, seeds were sown on two layers of filter paper in transparent, polystyrene boxes containing 1.6 ml of test solution made up in M/75 phosphate buffer at pH 6.3. They were kept at 22°C either in the dark or under 400 watt, mercury-fluorescent lamps. Germination was defined as emergence of the radicle through the testa and was assessed over a period of 30 days. Each treatment was replicated twice on 50-seed samples and all experiments were repeated on a number

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of occasions. In glasshouse and field tests, seeds were soaked for 42-48 hours in dichloromethane solutions of the growth regulators (Meyer and Mayer, 1971), airdried and stored at 5°C until required. The treated seeds were sown at uniform depth either in 10.5 cm pots containing J.I. Compost No. 1 in an unheated glasshouse at NVRS, in unprotected Dutch-light frames at NVRS or in a mixture of compost and soil in polystyrene flats in an open greenhouse at Bet Dagan in Israel. The percentage germination was recorded over a period of up to 30 days and samples of the resulting seedlings subsequently measured.

RESULTS

a) Laboratory tests

A series of preliminary experiments was used to determine the most promising chemical combinations and to calculate the active concentration ranges of these treatments. The results of these experiments are not presented. In the final fully-randomised, replicated experiment the most effective treatments were applied and the results are shown in Fig. 1.

Fig. 1

Germination of celery cv. Lathom blanching seeds in the dark at 22°C


Seeds incubated in the light germinated rapidly after 5 days but dark-incubated seeds did not germinate over the 30-day test period unless treated with growth-regulator solutions. $GA_{4/7}$ induced some seeds to germinate in the dark but the greatest response was obtained when the cytokinins N⁶-benzyladenine (BA) and 4-pyridylphenylurea (NC5392) or the growth retardant N-dimethylaminosuccinamic acid (Alar) were used in combination with the gibberellins. Although treatments with either cytokinin resulted in a higher final percentage germination than Alar treatment when used with GA_{4/7}, they were not employed in further experiments because subsequent development of the seedling radicles was inhibited by the cytokinins. Treatment with the mixture of Alar and GA_{4/7} resulted in seedlings with larger cotyledons and sturdier roots than those from untreated seeds, so in all further experiments only these chemicals were used.

Glasshouse tests

The results of the first experiment in which seeds pretreated with dichloromethane (DCM) solutions of growth regulators were sown 1 cm deep in pots in an unheated glasshouse are presented in Fig. 2.

Fig. 2

Germination of celery cv. Lathom blanching in glasshouse



Although some increase in germination rate, final germination percentage and seedling size as determined by fresh weight measurements was obtained by $GA_{4/7}$ treatment, the mixture of Alar and $GA_{4/7}$ was even more effective. Subsequently a further experiment was carried out with this treatment, in which seeds were sown either 13 or 26 mm deep in pots, the results of which are presented in Table 1.

Table 1

Sowing depth (mm)	Treatment	% ge	day no	on at	Averag	e seedling measures after 35 days	nents
		13	15	21	F. wt. (mg)	Hypocotyl length (mm)	Leaf no.
13	Untreated Alar + GA4/7	5 36	51 42	1	77 253	4.5 6.8	2.8
26	Untreated Alar + $GA_4/7$	2	5 35	37 36	50 106	2.8 3.7	2.7
L.S.D	. 5% level	-	-	-	44	2.5	-

Germination and seedling development of celery cv. Lathom blanching in the glasshouse

Seedling emergence was delayed but the stimulatory effect of chemical treatment was not affected by the increased sowing depth. At both depths, the final percentage germination of untreated and chemically-treated seeds was similar, but the spread of emergence of treated seeds was less. This resulted in increased uniformity, advanced development and a higher weight of the seedlings from treated seeds at sampling time. Seedlings produced from treatments involving the use of gibberellins appeared normal with no apparent excessive hypocotyl extension.

Field tests

An experiment in which seeds were sown 20 mm deep in Dutch-light frames yielded similar results (Table 2). Treatment with Alar, $GA_{4/7}$ or a mixture of these compounds increased the germination rate and improved seedling development. Although treatment with either $GA_{4/7}$ alone or the mixture of $GA_{4/7}$ and Alar resulted in similar germination, seedlings from the latter treatment were the more vigorous as is indicated by the cotyledon length measurements.

Table 2

Germination and seedling development of celery cv, Lathom blanching in Dutch-light frames at NVRS

_	% germination at day no.		Average seedling measurements after 22 days			
Treatment	14	16	F. wt. (mg)	Seedling length (mm)	Cotyledon length (mm)	
GA4/7 + Alar	21	39	15	8.3	9.8	
GA4/7	22	42	16	8.3	8.6	
Alar	11	27	13	5.8	8.3	
Untreated	8	28	7	4.5	4.8	
L.S.D. at 5% le	vel 6	5	7	2.3	2.9	

In the final experiment, either untreated seeds or seeds treated with $GA_{4/7}$ and Alar were sown in Dutch-light frames at NVRS (max. temp. $21^{\circ}C$) or in polystyrene flats in Israel (max. temp. $27^{\circ}C$). The percentage emergence was assessed over a period of 29 days and the results are presented in Fig. 3.



Fig. 3

The emergence rate of treated seeds was higher than that of untreated seeds at both locations, but although the final germination percentage was similar at NVRS, few untreated seeds emerged under the higher temperature conditions in Israel.

DISCUSSION

The results obtained from these experiments show that the high temperatureinduced light-requirement of celery seeds can be eliminated by treatment with some growth regulators. Although previous experiments had shown that gibberellic acid (GA3) was not effective in stimulating the germination of celery seeds (Palevitch et al, 1971), the most effective treatments were those involving the use of $GA_4/7$ in combination with the cytokinins BA and NC5392 or Alar. The cytokinins were more effective than Alar in promoting germination but inhibited subsequent radicle growth, confirming the observations of Knypl and Chylinska (1972) with other species. The mixture of GA4/7 and Alar, when applied as a dichloromethane seed soak, also stimulated field emergence and subsequent seedling development. The chemical effect on germination percentage was more apparent under the high temperature conditions in Israel, but enhancement of germination rate and seedling development was obtained in all glasshouse and field experiments. The use of organic solvents to introduce these chemicals into seeds may be advantageous in that they evaporate rapidly when the seeds are air-dried. It seems possible that other chemicals such as systemic fungicides could also be introduced in this way. It is envisaged that chemical treatment may assist in obtaining even seedling emergence and subsequent uniform crop stands. The increase in emergence rate induced by these treatments may also be of value in weed control by advancing the time of herbicide application and decreasing the effect of weed competition.

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THE EFFECT OF CHLORFLURECOL ON THE GROWTH OF TURIONS OF MYRIOPHYLLUM VERTICILLATUM L.

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Summary Preliminary experiments were carried out with chlorflurecol on turions of the submerged weed <u>Myriophyllum verticillatum</u> L. in the greenhouse. Results were variable but successful growth inhibition was achieved with concentrations of between 0.001 and 0.1 mg/l. There was also evidence that root initiation was inhibited by concentrations as low as 10⁻⁵ mg/l.

INTRODUCTION

In many freshwater situations where submerged weed growth must be controlled, the complete elimination of plants is undesirable because they oxygenate the water and provide shelter for fish fry and fish-food organisms. The use of herbicides is aimed at the elimination of most vascular plants for as long as possible. When they are used after growth has started they put the fish life at risk through the possibility of deoxygenation. In an attempt to reach a compromise which would reduce but not destroy weed growth initial studies were carried out on the effect of growth retardant chlorflurecol on the submerged weed <u>Myriophyllum verticillatum</u> L.

The effects of chlorflurecol on terrestrial weeds have been described by Berker et al (1968) and Scheider and Mohr (1970) and indicate activity over a wide range of plants. The mode of action claimed for chlorflurecol (Berker et al, 1968) is a dwarfing effect attributed to a reduced rate of cell division. This effect would seem well suited to the needs of submerged weed control by producing shorter compact growth and thus less resistance to water flow while retaining active photosynthesis and thus oxygen evolution, and adequate cover for small animals.

This paper describes a series of laboratory and greenhouse experiments on turions (winter buds) of <u>M. verticillatum</u> aimed at an initial assessment of the action of chlorflurecol in freshwater.

METHODS AND MATERIALS

The object of the initial experiments with chlorflurecol was to find the most suitable concentrations, and the evaluation technique using turions (winter buds) of <u>M. verticillatum</u> described by Robson (1970) was used. In all these experiments the culture medium was 10% Hoaglands solution in deionised water. The beakers (either 1000 ml or 500 ml) were covered with perforated zinc sheets to reduce light and were kept in a greenhouse with temperatures varying diurnally between 15° and 20° C and day lengths of 16 or more hours. The experiments were designed as randomised blocks with 4-6 replications.

In the first experiments the turions were germinated and allowed to grow for 7-8 days before treatment with chlorflurecol. Subsequently ungerminated turions were introduced into the test solutions at intervals of time after the start of the experiments to measure residual activity. The concentrations tested in this series of experiments were from 10 mg/l. to 10^{-7} mg/l. (0.0000001 mg/l.) made up in 10% Hoaglands solution (Hoagland and Arnon, 1950).

RESULTS

The results obtained in two experiments with pre-germinated turions are presented in table 1.

Table 1

Length increment	(mm) of Myr:	iophyllum	verticillatum	turions				
following treatment with chlorflurecol								
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	Exp	ot. I		Expt. II	
Chlorflurecol (mg/l.)	7 days	15 days	8 days	13 days	21 days
1.0	61.5	73.5	-	-	
0.1	67.5	77.5	10.7	13.3	16.5
0.01	65.3	73.0	11.2	11.1	15.3
0.001	62.8	78.0	19.8	22.2	25.7
0.0001	-	-	19.8	23.7	27.0
0.00001		-	22.7	27.0	29.0
Control	108.3	1 35.0	21.3	24.7	28.8
S.E.	6.46	7.87	3.0	4.85	4.72

The growth inhibition obtained in the first experiment (I) at all concentrations was of the same order and amounted to almost a 50% reduction in growth over the 2 week period. The effect did not appear to fall off at the lowest concentration of 0.001 mg/l. and this indicated that chlorflurecol may be active at lower concentrations. The second experiment (II) was designed to investigate this, but no growth inhibition was obtained at concentrations lower than 0.01 mg/l.

However, the rate of growth over the whole of the second experiment was poor and the untreated control plants increased by only 2.7 mm/day in the first week, 0.6 mm/day over the 2nd and 0.7 mm/day over the 3rd week while those of the first experiment grew at 15.4 mm/day in the first week and 4.0 mm/day in the second. These differences occurred, at least in part, because the turions in the second experiment were from a different batch and collected at a different time. The lack of diurnal variation in temperature and the quality of the light source may also have contributed, but records are inadequate to substantiate this.

In both experiments a concentration of 0.01 mg/l. chlorflurecol or above resulted in a 40-50% inhibition of growth during the first week after treatment and this was maintained for the duration of the experiments.

The effect of low concentrations was further studied in experiment III. A succession of 3 turions were introduced at weekly intervals to determine whether chlorflurecol activity decreased with time as well as to confirm the findings in the previous experiments. The chlorflurecol concentrations were chosen over the very

wide range 10 mg/l. to 10^{-7} mg/l. (0.0000001 mg/l.) and the results are given in table 2.

Ta	ble	2
-		_

		Turion 1		Turion 2 (introduced day 7)			Turion 3 (introduced day 14)	
conce of ch	ntration lorflurecol	5 days	13 days	6 days	8 days	15 days	8 days	
10	mg/1.	0.5	0 (all dead)	7.3	8.3	9.0	8.3	
10-1	mg/1.	26.5	34.3	22.2	31.0	49.0	31.3	
10-3	mg/1.	31.7	41.3	28.0	39.7	52.2	33.1	
10-5	mg/1.	41.0	48.5	31.7	45.8	58.8	36.1	
10-7	mg/1.	36.3	45.3	32.0	44.7	65.2	31.3	
Contr	ol	36.2	49.3	28.3	42.3	59.3	38.0	
	S.E.	3.7	7.0	3.0	2.0	4.1	4.1	

Experiment III. Mean length increments (mm) of 3 turions introduced at weekly intervals

Only at the highest dose of 10 mg/l. did chlorflurecol significantly affect the plants and then it killed them. The survival and reduction in growth of turions 2 and 3 at this dose indicated that while there was probably a decrease in the chlorflurecol concentration sufficient chemical remained to cause a 70-75% decrease in growth.

The reduction in root initiation was much more pronounced than the reduction in shoot growth and the results are presented in table 3.

Table 3

Experiment III. The effect of chlorflurecol on the number of roots developed by turions of M. verticillatum

	Mean number of roots				
Concentration of chlorflurecol 4 mg/l.	Turion 2 8 days	Turion 3 8 days			
10.0	0.3	0			
10 ⁻¹	2.3	0.5			
10-3	4.2	3.3			
10-5	8.3	7.3			
10-7	15.3	5.5			
Control	14	11.3			
S.E.	1.5	1.4			

Turion 1 had germinated and produced roots a week before the experiment started and since experience has shown that most of the roots are initiated within the first week, they were not counted. Root initiation was reduced significantly on turion 2 at 10^{-5} mg/l. and higher concentrations while turion 3 showed the first significant effect at 10^{-3} mg/l. Since it is indicated by the shoot data that chlorflurecol activity dropped during the experiment the discrepancy between turions 2 and 3 may be explained in this way. However, it is clear that very low concentrations of chlorflurecol affect the initiation of new roots.

Experiment IV was designed to study the effect of chlorflurecol at 10, 1.0 and 0.1 mg/l. on <u>M. verticillatum</u> turions planted in separate small pots (3 inch diameter) which were filled with pond mud and immersed in tanks containing 30 l. of de-ionized water. Three turions were placed in each tank and the treatments were replicated four times in a latin square design.

At weekly intervals whole plants were harvested at random from each tank and their length measured. The results are presented in table 4 as a percentage of the untreated controls to allow a comparison to be made between weeks.

Table 4

Experiment IV. <u>Mean shoot length (main stem plus laterals) of M. verticillatum</u> plants expressed as a percentage of the control

Concentration of chlorflurecol	7 days length %	14 days length %	21 days length %
10.0 mg/1.	72.5	12.6	7.1
1.0 mg/1.	94.5	24.8	17.1
0.1 mg/1.	96.5	38.4	29.4
Control	100.0	100.0	100.0
S.E.	3.9	2.9	3.1
Control Mean length	50 mm	331 mm	597 mm

Very little growth took place in the first week and this accounts for the apparent lack of effect at all levels of chlorflurecol. However, by the second week all concentrations had inhibited growth and the reduced rate of elongation continued through the third week as indicated by the lower percentage shoot length after 21 days. In a similar experiment (V) the effect of chlorflurecol at 2.0, 1.0 and 0.5 mg/l. was investigated and the results are presented below (Table 5).

In experiment V the difference between the rate of growth of the treated plants and that of the controls was not as great as in experiment IV. This is primarily because the untreated plants in experiment V did not produce as much lateral branching as did those in experiment IV and this is reflected in the difference in mean length of the control plants after three weeks - experiment IV, 597 mm and experiment V, 144.5 mm. Much of the shoot length in experiment IV was due to the

ncentration of	Days	after treatme	nt
lorflurecol mg/1.	7 days %	14 days %	21 days %
2.0	90.6	60.9	46.7
1.0	81.2	62.2	49.7
0.5	80.6	46.1	52.2
Control	100.0	100	1.00
S.E.	8.4	4.6	5.8
Control mean length	40.5 mm	108.75 mm	144.5 mm

Experiment V. Mean shoot length at weekly intervals after treatment expressed as a percentage of untreated control

long lateral branches which started developing in the second week of the experiment. The mean numbers of laterals per plant are compared in table 6 and in addition to showing a difference between experiments they indicate that chlorflurecol may also inhibit the initiation of branches.

Table 6

Mean number of branches de	veloped on the plants in experiments IV and V	IV
Concentration of chlorflurecol	No. of branches	v
		-
10 mg/1.	0 –	
20 mg/1.	- 0	
1.0 mg/1.	0.5 0	
0.5 mg/1.	- 0	
0.1 mg/1.	2 -	
Control	7 2	

and

DISCUSSION

Chlorflurecol retards the elongation of the turions of M. verticillatum but the extent to which it does so is variable. In experiment I a 50% inhibition was obtained with 0.001 mg/l. was required and in experiment III a concentration of 0.1 mg/1. was almost ineffective. The difference between experiments I and II may be due to the difference in growth rate (9.0 mm/day and 1.9 mm/day over the first 2 weeks respectively) but this does not explain the poor response to chlorflurecol in experiment III when the growth rate was intermediate (3.8 mm/day over 2 weeks).

In experiments IV and V different plants were measured at each sampling date so individual rates of growth could not be assessed and it was necessary to relate the growth of the treated plants to that of the controls. However, these data do illustrate the need for caution in referring to percentage inhibition because of the variations in the inherent rate of growth of untreated plants.

In all the experiments where successful growth retardation was achieved it was evident that the inhibition continued for at least two to three weeks and experiments IV and V show how the difference in size between the treated and untreated plants increased with time.

The possible use of chlorflurecol on a field scale will depend upon results from further studies to clarify the conditions responsible for the variability in activity encountered in these preliminary experiments. Further work will be necessary also to determine its activity on other species of submerged weed and on plants at a more advanced stage of growth with the object of establishing dose rates. Once this stage is reached the toxicity of chlorflurecol to fish and other forms of freshwater life must also be considered.

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THE EFFECTS OF PLANT GROWTH-REGULATORY CHEMICALS ON SEED GERMINATION

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Summary Thirteen plant growth-regulatory chemicals were tested at various concentrations for their effects upon the germination of 12 weed species. The greatest stimulation obtained was from 100 ppm GA on <u>Thlaspi arvense</u>. None of the chemicals, however, showed any stimulatory or inhibitory effects common to all the test species. The results indicate that, although species vary in their dormancy and their dormancy-breaking requirements, chemicals may well be found that will in some measure supply these requirements.

INTRODUCTION

One of the great difficulties in reducing the number of viable weed seeds in the soil by cultivation is that many have innate dormancy, which prevents them from germinating even when the conditions are favourable. This dormancy is lost through various agencies in nature; by leaching out of inhibitors; by decay of the seed coat; by chilling over-winter etc.; the method apparently depending upon the type of dormancy present. These dormancy systems enable the seed to survive prolonged periods unfavourable to the plant. The rate of decline in numbers of a seed population is therefore slow, being only about 45% per year (Roberts, 1962). It has been suggested (Povilaitis, 1956) that the best way of ridding the soil of seeds is to stimulate them to germinate. A search was initiated in 1970 at the Weed Research Organization for chemicals that would stimulate the germination of dormant seeds. An initial success was obtained with 2-chloroethylphosphonic acid (Chancellor <u>et al</u>, 1971) and results of further investigations with other chemicals are presented in this paper.

METHOD AND MATERIALS

In a series of dormancy-breaking experiments batches of seed of the following species were used:- Matricaria matricarioides (Less.) Porter, Onopordum acanthium L., Avena fatua L., Chenopodium album L., Datura stramonium L., Thlaspi arvense L., Rumex obtusifolius L., Urtica urens L., Plantago lanceolata L. and Smyrnium olusatrum L. All seed was freshly-collected during autumn 1970 in the Oxford area and stored dry in the laboratory until used. These species were selected because their seeds were dormant and available in quantity and because they included a range of important weed species from nine plant families and so represented various types of dormancy.

One hundred seeds of each of the 10 species were placed in 9 cm petri dishes lined with 4 sheets of Whatman No. 1 filter paper moistened with 6 ml of either distilled water or a chemical solution. These were unreplicated. The dishes were kept at 23 °C in a dark room, receiving short periods of light every 2-3 days when all germinated seeds were counted and removed. The dishes were kept for at least 20 days before being discarded and if still germinating at 20 days for a full week after the last seed germinated. In a second test series, seeds of <u>Striga hermonthica</u> Benth. (6 year old seed) and <u>Orobanche aegyptiaca</u> Pers. were prepared and tested as described by Kasasian and Parker (1971); but without using a sorghum root exudate other than in controls. Each treatment was replicated three times, each with its own control. In tests with <u>S. hermonthica</u> and <u>O. aegyptiaca</u>, because of the impossibility of counting out the seeds, the system of using filter paper discs scattered over with seed gave a range of seed numbers. For <u>S. hermonthica</u> the range was 9-112 seeds/disc, the mean 32, with the majority between 20 and 45. For <u>O. aegyptiaca</u> the range was 9-59, with a mean of 24 and a majority between 15 and 40 seeds/disc.

The 13 chemicals tested during the period October 1970 to March 1971 were $2-(\beta-chloro-\beta-cyanoethyl)-6-chloro-toluene, (PRB-8);$ ethyl hydrogen 1-propylphosphonate, (NIA 10637); 6-benzylaminopurine, (BA); 6-furfurylaminopurine, (kinetin); indole-3-acetic acid, (IAA); gibberellic acid, (GA); 4-hydroxy-5-isopropyl-2-methyl-phenyltrimethyl-ammonium chloride, 1-piperidine carboxylate, (AMO 1618); N-dimethylaminosuccinamic acid, (B-995); tributyl-2,4-dichlorobenzylphosphonium chloride, (Chlorphonium chloride); 5,7-dichloro-4-ethoxycarbonylmethoxy-2,1,3-benzothiadiazole, (Du 13594); 5-chloro-6-ethoxycarbonylmethoxy-2,1,3-benzothiadiazole, (Du 17623); and salicylic acid.

Chlorphonium chloride and NIA 10637 were water miscible liquids, PRB-8 was an emulsifiable concentrate, while AMO 1618 and B-995 were directly water soluble. IAA and GA were dissolved in a few ml of 0.1 N NaOH and salicylic acid in 5% Na $^{CO}_3$ and all were neutralised with 5% v/v HCl during dilution. Kinetin and BA were 3 dissolved in 5% v/v HCl and neutralised with Na $^{CO}_3$ during dilution. Du compounds 13594, 16333 and 17623 were all dissolved in xylene with Triton B 1956 added as emulsifier.

The 10 species in the first series were also tested in February and March with standard dormancy-breaking methods to gauge the relative effectiveness of the growth-regulatory chemicals. The standard methods were pricking the seeds, soaking in 0.2% w/v KNO₃ and chilling at 4° C for 28 days prior to incubation at 23°C.

RESULTS

In the first test series with PRB-8 and NIA 10637 not all 10 species were included as some had not then been collected. <u>S. hermonthica</u> and <u>O. aegyptiaca</u> were also not available for some tests.

The results of both test series are given in Table 1 expressed as the percentage germination of the seeds tested. For <u>S. hermonthica</u> and <u>O. aegyptiaca</u> they are the means of the three replicates.

Controls with a sorghum-root exudate were set up for <u>S. hermonthica</u>, which gave 29-57% germination with a mean of 44%, and for <u>O. aegyptiaca</u>, which gave 13-80% germination in the various tests.

The high germination percentages of <u>M. matricarioides</u> obtained in the test with IMA and GA are presumably due to some environmental influence occurring before or during the testing, which did not recur in other experiments.

The results of the standard dormancy-breaking techniques for the 10 species in the first test series are given in Table 2. The comparisons of pricked and unpricked seeds and 0.2% KNO₃ and water-treated seeds were made in February, i.e. between the AMO 1618 test group and the group of Du compounds in Table 1, which is in chronological order. The comparison of chilled and unchilled seed was made in March between the Du compound group and the salicylic acid test.

Т	a	b	Le	1
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Chemical	ppm	Matricaria matricarioides	Onopord um acanthium	Avena fatua	Chenopodium album	Datura stramonium	Thlaspi arvense
Controls	-			6	0	0	0
PRB-8	0.5			8	1	0	0
	5			3	1	0	0
	50			3	2	0	0
NTA 10637	0.5			7	1	0	0
	5			12	0	0	0
	50			8	1	0	0
Controls		7	50	5	6	0	0
BA	1	7	53	7	0	0	1
	10	51	36	8	3	0	2
	100	65	29	11	1	0	8
Kinetin	1	4	59	16	1	0	0
	10	8	54	13	3	0	1
	100	62	37	18	1	0	10
Controls		54	49	37	12	0	0
IAA	1	33	48	33	0	0	0
	10	55	42	10	3	0	1
	100	64	57	17	1	0	1
GA	1	46	45	18	0	0	1
	10	40	40	22	0	0	0
	100	50	58	22	4	4	93
Controls		14	27	13	2	0	0
AMO 1618	1	12	34	21	0	0	1
	10	7	46	14	0	0	0
	100	12	37	18	0	0	0
B-995	1	11	38	18	1	0	0
	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	0					
	100	14	42	15	2	1	1
Chlorphonium	1	16	35	15	0	0	0
chloride	10	8	24	13	4	0	0
	100	8 .	33	15	0	0	0
Controls		17	20	21	1	0	0
Du 13594	1	5	37	27	2	0	0
	10	5	54	35	2	0	0
	100	7	34	25	3	0	0
Du 16333	1	15	49	15	6	0	1
	10	21	28	21	1	0	0
	100	6	25	12	1	0	1
Du 17623	1	2	36	24	4	0	4
	10	0	12	22	3	0	0
and the second	100	0	20	19	1	0	0
Controls		15	69	25	3	0	2
Salicylic	1	7	72	25	5	0	0
acid	10	11	74	33	2	0	1
	100	3	74	19	3	0	0

The effects of plant growth-regulatory chemicals on the percentage germination of twelve species

Table 1 (continued)

Chemical	ppm	Rumex obtusifolius	Urtica urens	Plantago lanceolata	Smyrnium olusatrum	<u>Striga</u> hermonthica	Orobanche aegyptiaca
Controls						0	
PRB-8	0.5					0	
	5					0	
	50					0	
NIA 10637	0.5					0	
	5					0	
	50					0	
Controls	-	0	2	58	0	0	0
BA	1	0	7	46	0	1.8	0
	10	0	5	50	0	7.8	0
	100	0	8	1	0	0	0
Kinetin	1	0	4	43	1	0	0
IAA	10	0	6	57	0	2.5	0
	100	1	3	21	0	0.4	0
Controls		8	4	58	0	0	0
	1	8	8	38	0	1.2	0
	10	22	16	59	0	0.4	0
	100	30	6	51	0	0	0.5
GA	1	35	8	49	0	0.8	0.5
	10	9	7	80	0	0.7	0
	100	78	8	79	2	0.5	0
Controls		21	20	43	0	0.3	
AMO 1618	1	42	17	57	0	0	
	10	28	22	43	0	0	
	100	16	17	19	0	0	
B-995	1	28	10	36	0	0	
	10	26	4	43	0	0	
	100	23	21	53	0	0	
Chlorphonium	1	20	41	54	0	0	
chloride	10	30	14	62	0	0	
	100	35	1	17	0	0	
Controls		19	14	44	0	0	0
Du 13594	1	22	8	49	0	0	0
	10	5	23	10	0	0	0
	100	9	9	5	0	0	0
Du 16333	1	16	30	31	0	0	0
	10	10	5	16	0	0	0
	100	7	10	3	0	0	2.3
Du 17623	1	19	31	29	0	0	2.4
	10	8	23	13	0	0	0.8
La maria	100	18	13	11	0	0	0.7
Controls	-	6	8	53	0		
Salicylic	1	10	19	40	0		
acid	10	2	20	45	0		
	100	5	7	21	0		

The effects of plant growth-regulatory chemicals on the percentage germination of twelve species

Species	Seeds pricked	Seeds unpricked	Seeds with KNO ₃	Seeds without KNO ₃	Seeds chilled	Seeds unchilled
M. matricarioides	55	18	56	14	3	36
O. acanthium	43	59	62	34	24	49
A. fatua	28	31	21	7	22	19
C. album	69	6	37	3	1	7
D. stramonium	0	0	0	0	0	0
T. arvense	55	0	3	0	4	0
R. obtusifolius	100	72	90	44	91	69
U. urens	54	16	7	3	3	8
P. lanceolata	60	57	33	44	13	62
S. olusatrum	0	0	0	0	3	0

The effects of standard dormancy-breaking techniques on the seeds of ten species (% germination)

Table 2

DISCUSSION

Some of the chemicals (Table 1) such as PRB-8, NIA 10637, B-995 and salicylic acid had little or no effect on the germination of any of the species but others appeared to be stimulatory. BA at 10 and 100 ppm and kinetin at 100 ppm increased greatly the germination of M. matricarioides and to a limited extent T. arvense and S. hermonthica. Kinetin also increased A. fatua germination slightly. IAA possibly decreased the germination of A. fatua and increased that of R. obtusifolius and U. urens, and S. hermonthica slightly. However, large variations were obtained in the germination of untreated seeds of R. obtusifolius (Tables 1 and 2) so that the results with this species may be unreliable. In view of current theories on germination (Carlson, 1972), GA was expected to have general stimulatory effects, especially on A. fatua. However, although there have been many reports of positive results with this species (Simpson, 1965; Green and Helgeson, 1957 etc) GA had no effect in our tests, perhaps because the concentrations were too low (Corns, 1960). GA stimulated the germination of other species e.g. T. arvense showed 93% with 100 ppm; which confirms the finding elsewhere. (H.A. Roberts - personal communication). Some germination of GA-treated S. hermonthica seed was evident and that of R. obtusifolius may have been improved, but more noticeable was the low but obvious germination of S. olusatrum and D. stramonium, both of which had responded to virtually no other treatment. O. aegyptiaca showed some slight stimulation by two of the Du compounds.

Several chemicals decreased germination, especially at the highest concentration, presumably through phytotoxic effects. Both BA and possibly AMO 1618 affected <u>P. lanceolata</u> and chlorphonium chloride affected <u>U. urens</u>; but the most consistent effects were by the three Du compounds, which all decreased the germination of <u>R. obtusifolius</u>, <u>M. matricariodes</u> and <u>P. lanceolata</u>. The susceptibility of <u>P. lanceolata</u> to several chemicals may be associated with the mucilaginous envelope that develops around the seed. This may affect uptake because the chemical solution may be incorporated into the mucilage, thereby bringing the seed into greater contact with the chemical.

The results presented here show that although the species vary both in type of dormancy and consequently in dormancy-breaking requirements, chemicals can in some measure supply these requirements. There are no obvious practical uses for the chemicals used in these tests but it is hoped that by further testing more effective ones may be discovered.

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THE EFFECT OF PLANT GROWTH REGULATORS ON DOMINANCE IN AGROPYRON REPENS (L.) BEAUV. RHIZOMES

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Summary Nine of a number of plant growth regulators tested, delayed to some extent the onset of a new dominance system in decapitated 7-node rhizome fragments grown in vitro. These were 6-benzylaminopurine, kinetin, TIBA, glyphosate, fluorodifen, nitrofen, SD 8339, WL 17731 and NC 9565. One chemical, MBR 6033 affected dominance in some primary lateral shoots so that secondary laterals grew out from them. A new dominance system was asserted sooner with GA, 1AA and U 29449. Polarity was affected by fluorodifen and to a lesser extent by SD 8339.

INTRODUCTION

Most rhizomatous weeds have many dormant buds on their rhizomes which can regrow rapidly if the plant is cut up by cultivation. However, only relatively few buds continue to grow and many more remain as dormant buds or short re-inhibited shoots (Chancellor, 1968) as reserves against future disturbance. This system of dormancy is normally only susceptible to a prolonged series of carefully-timed cultivations (Fail, 1956) or effective herbicide treatments. In order to improve the effectiveness of control measures, a search has been started at the Weed Research Organization to find chemicals which prevent or delay the onset of, or interfere with the new dominance system in rhizome fragments.

Both 2-chloroethylphosphonic acid and chlorflurecol-methyl have been shown to affect growth and dominance in rhizome fragments to a considerable extent (Chancellor, 1970). These results have proved that interference is feasible and the search has continued for further compounds which will achieve the same or other effects. This paper describes preliminary results obtained with a number of compounds.

METHODS AND MATERIALS

In all the tests rhizomes of <u>Agropyron repens</u> (L.) Beauv. clone 31 (Headington clone) of the Weed Research Organization's couch-grass collection were used. In each experiment seven-node fragments were cut from freshly-dug, young rhizomes, then washed and stripped of their roots and scale leaves. The fragments were then attached to strips of Whatman's 3MM chromatographic paper, moistened with water or chemical solution and incubated at 23°C in the dark in glass jars as described by Blair <u>et al.</u> (1970). At least three concentrations of each chemical were employed. A minimum of four rhizomes per treatment were used with sometimes larger numbers.

The chemicals tested were indole-3-acetic acid, (1AA); 6-furfurylaminopurine, (kinetin); gibberellic acid, (GA₃); 6-benzylaminopurine, (BA); 2,3,5-triiodobenzoic acid, (TIBA); 2-chloroethyltrimethylammonium chloride, (chlormequat); N-dimethylamino succinamic acid, (B-995); 4-hydroxy-5-isopropyl-2-methylphenyltrimethylammonium chloride, 1-piperidine carboxylate, (AMO 1618); tributyl 2,4-dichlorobenzyl phosphonium chloride, (chlorphonium chloride); 2-(β-chloro-β-cyanoethyl)-6-chlorotoluene, (PRB-8); ethyl hydrogen 1-propylphosphonate, (NIA 10637); 5,7-dichloro-4ethoxycarbonyl-methoxy-2,1,3-benzothiadiazole, (Du 13594); 5-chloro-6-ethoxycarbonylmethoxy-2,1,3-benzothiadiazole, (Du 16333); 7-chloro-4-ethoxycarbonylmethoxy-5methyl-2,1,3-benzothiadiazole, (Du 17623); salicylic acid; 1,2-dihydropyridazine-3,6-dione (MH); N-(phosphonomethyl)glycine, (glyphosate); sodium glycocholate; 3trifluoro-methyl-sulphonamido-p-acetotoluidide, (MER 6033); 2,4-dinitro-4-trifluoromethyldiphenyl ether, (fluorodifen); 2,4-dichlorophenyl-4-nitrophenyl ether, (nitrofen); 4-nitro-2,4,6-trichlorodiphenyl ether (Mo 338); ethyl-2-(N-benzoyl-3,4dichloroanilino) propionate, (WL 17731); N,N-bis(phosphonomethyl)glycine, (CP 41845) 6-benzylamino-9-(tetrshydropyran-2-yl)-9H-purine, (SD 8339); 3,4,5-tribromo- αmethylpyrasole-1-acetic acid, (U 27658); ethanol-2-(dimethylamino)-2-[(4-chloro-otolyl)oxy] propionate, (WL 29449); ([3-(p-chlorophenyl)-1,2,4-oxadiazol-5-yl]thio) acetic acid, (NC 9565); [(3-phenyl-1,2,4-thiadiazol-5-yl)thio] acetic acid, (NC 9634).

The experiments were continued for 20 days, during which time the length of each shoot was measured to the nearest mm every 2-3 days. Roots were occasionally measured and counted.

RESULTS

Because of the large number of aspects of growth observed, the results are presented in note form. All comparisons are with water-treated controls at 20 days unless otherwise stated.

<u>1AA</u> at 1, 10 and 100 ppm caused a slight reduction in shoot lengths and in the number of shoots growing. The length of roots, measured at 5 days, was also reduced by 10 and 100 ppm.

<u>Kinetin</u> at 1 and 10 ppm had no apparent effects upon growth, but 100 ppm delayed dominance initially and reduced the mean root length by 58% after 6 days.

GA, at 1, 10 and 100 ppm hastened the onset of dominance by about 4-5 days and the dominant shoots were longer (83% longer with 100 ppm). The mean root length was increased with 1 and 10 ppm.

<u>BA</u> at 10 and 100 ppm delayed shoot re-inhibition over the first 11 days but thereafter it was similar to controls. There was no effect on shoot or root length at 10 ppm but 100 ppm reduced the mean shoot length by 30% at 20 days and root length by 59% at 6 days. The same concentrations were repeated in a later experiment, again with no effects on dominance. Root number and length was reduced at 10 ppm while 100 ppm almost completely inhibited root production, besides reducing shoot length by 36%.

<u>TIBA</u> at 1 and 10 ppm reduced shoot length but there was no effect on the dominance system. There were slightly fewer roots with 10 ppm. At 100 ppm there was a delay of about 8 days in the onset of dominance, the shoots grew in circles or spirals and were virtually rootless - only 1 short root was present after 20 days.

Chlormequat, B-995, AMO 1618 and Chlorphonium chloride all had no apparent effects at 1, 10 or 100 ppm.

<u>PRB-8</u> at 0.05 ppm had no effect on dominance but shoots were 28% shorter than in controls. At 5 ppm the number of growing shoots was halved and their length was 72% shorter than controls; roots were thickened but were no shorter.

NIA 10637 at 50 ppm had no obvious effect but at 500 ppm there were fewer growing shoots (about two-thirds fewer) and these were shorter on average by

65%; roots were also reduced in length by two-thirds.

<u>Du 13594</u> was **in**effective at 1 and 10 ppm but 100 ppm delayed the onset of growth by 1 week. This was later found to be due to the xylene in which the chemical was dissolved. At 1 ppm one rhizome had fisciated roots throughout, but this is not thought to be due to the treatment.

 $\underline{\text{Du 16333}}$ effects were similar to those of Du 13594 except that the solvent killed all six rhizomes.

Du 17623 effects were the same as those of Du 13594.

<u>MH</u> at 10 ppm had no effect on dominance, but the shoots were 22% longer than controls. At day 8 roots were unaffected. At 100 ppm shoots had all ceased growth by day 15 the longest being 164 mm (the longest control shoot was 292 mm at day 15); roots too were noticeably shorter. At 1000 ppm all shoots had ceased growth by day 6, the longest being 16 mm as compared with 70 mm for the longest control. Root initiation was completely suppressed at this concentration.

<u>Glyphosate</u> was ineffective at 0.1 and 1 ppm but reduced root length and not root number at 10 ppm. Although there was no effect on dominance shoot length was also decreased. The tests were repeated using 10 and 100 ppm on 12 rhizomes in each instance. At the lower concentration growth was erratic, although dominance was undoubtedly delayed; the roots were short and stumpy and leaf blades on the shoots were shortened. At 100 ppm the chemical was toxic, for buds grew a maximum of 3 mm in 20 days and no roots were produced.

<u>MBR 6033</u> at 1 ppm reduced shoot lengths and increased the number of roots. At 10 ppm more roots were produced, the shoots were very short with broader leaf blades and some of them developed a single secondary lateral shoot from the base. At 100 ppm shoots were little more than long, flattened buds. The roots were increased in number by 35% at day 8, but were stunted. There were few shoots growing at any time on rhizomes treated with the 10 and 100 ppm doses.

<u>Fluorodifen</u> at 5 ppm had no apparent effects. At 50 ppm there was possibly some effect on polarity for the longer shoots occurred more towards the middle or base of the rhizomes. The 500 ppm treatment delayed the onset of growth by about a week with a consequent delay in the onset of dominance. Some effect on polarity was evident and fewer roots were formed (there were none until day 8).

<u>Nitrofen</u> at 5 ppm had no effects on growth. At 50 ppm there was possibly a short delay in the onset of dominance, more shoots were growing and there were more longer ones. All rhizomes were killed rapidly by 500 ppm.

MC 338 at 5 ppm had little effect. At 50 ppm the longest shoots were less rigid than controls and roots fewer. Rhizomes were all killed by 500 ppm.

<u>WL 17731</u> at 1 ppm delayed dominance by about a week and shoots were 24% longer than controls. At 10 ppm there was no effect on dominance and the shoots were 37% shorter. At 100 ppm shoots ceased growth earlier than in the controls; at day 20 none at all were growing and the mean shoot length was then 56% less than that of the controls.

<u>CP 41845</u> at 1, 10 and 100 ppm had no effect on dominance. At 100 ppm leaf blades were shorter and the mean shoot length was reduced by 35%.

SD 8339 at 1 ppm had no effect on dominance but roots and shoots were shorter. At 10 ppm there were more shoots growing throughout, especially near the base, and dominance was delayed. The roots were extremely short. At 100 ppm both shoots and roots made little growth. <u>U 27658</u> at 0.1, 1 and 10 ppm had no effect on dominance. The 10 ppm treatment increased the mean shoot length by 42% in 20 days and the lower concentrations by lesser amounts.

<u>U 29449</u> at 0.1 had no effects but at 1 and 10 ppm there were fewer roots, all of which were very short and deformed at the higher concentration. Shoot growth was strongly inhibited and absolute dominance was imposed very early i.e. before 4 days in 4 of the 8 rhizomes treated.

<u>NC 9565</u> at 1 and 100 ppm had no effect on dominance, but at 10 ppm dominance was delayed by about a week and the mean shoot length was 61% longer than controls.

<u>NC 9634</u> at 1, 10 and 100 ppm had no effect on dominance. The shoots were slightly longer than controls at the two lower concentrations.

Salicylic acid at 10 ppm had no apparent effects.

Sodium glycocholate at 1, 10 and 100 ppm had no effects.

DISCUSSION

The main purpose of these tests was to find chemicals which prevent or delay the onset of a new dominance system in freshly decapitated 7-node rhizome fragments. It is envisaged that effective chemicals may be of considerable value in controlling this weed. Several of the chemicals tested were found to affect dominance in various ways.

BA at 10 and 100 ppm delayed the onset of dominance, in that up to 20% more shoots grew on the treated rhizomes, but this effect was lost by day 13. In contrast, Caseley (1972) found a reduction in the number of aerial shoots when it was applied through the soil to whole plants. Kinetin at 100 ppm was also found to delay dominance, but this was lost by day 18. It is possible that more persistent effects may have been obtained with both these chemicals in the presence of exogenous nutrients. The possibility of adding nutrients to all solutions in these tests is being considered.

TIBA also delayed dominance as was expected. At 100 ppm the number of buds growing indicated a delay on average of about 8 days in the onset of new dominance systems. Meyer and Buchholtz (1963) found that TIBA applied to the centre of an intact rhizome interfered with the dominance of its apex and allowed three buds to the basal side of the treated area to grow out. Caseley (1972) found an increase of 7% in the number of aerial shoots when TIBA was applied to the foliage of intact plants. These results suggest that TIBA blocks auxin movement basipetally as in other plants (Panigrahi & Audus, 1966). Shoots on rhizomes treated with 100 ppm grew in circles or spirals and were virtually rootless, a condition similar to that produced by chlorflurecol (Chancellor, 1970) although no secondary laterals were produced with TIBA.

In a preliminary test with glyphosate at 0.1, 1 and 10 ppm (4 rhizomes/treatment) no effects on dominance were observed, but further tests at 10 and 100 ppm on 12 rhizomes each showed that although the lower concentration had no effect initially, after 14 days sustained yet erratic growth continued at a higher level than controls. The 100 ppm treatment proved toxic. Thus, if this chemical is applied as a herbicide and only a small dosage is received by the plant, the number of shoots could be increased.

Fluorodifen at 50 ppm and nitrofen at 50 ppm also appeared to have a delaying effect on dominance. WL 17731 at 1 ppm and NC 9565 at 10 ppm delayed it by about a week and SD 8339 at 10 ppm by 7-10 days. As was expected, most rhizomes with delayed dominance had a greater mean shoot length than those in controls. With NC 9656 treatment this was by as much as 61% after 20 days. However, with SD 8339 the mean shoot length was similar to that of the controls, which may suggest that its mode of action is different.

All the chemicals considered so far affected the dominance system within the rhizome itself. However, MBR 6033 appeared to break dominance within the lateral shoots, thereby allowing secondary laterals to grow out from their bases. At 10 ppm, four out of 28 shoots were affected in this way and they were on 3 out of the 4 rhizomes. Only chlorflurecol is known to have a similar action (Chancellor, 1970) but unlike chlorflurecol there was an increase in the number of roots with MER 6033.

Equally interesting although possibly less important from the control point of view, some chemicals enhanced the dominance system, so that the overall number of growing buds was reduced. Some treatments were undoubtedly directly toxic to the rhizomes, but three chemicals actually appeared to strengthen dominance itself.

All rhizomes treated with GA, had dominance systems imposed about 5 days earlier than controls. This result is in agreement with other findings (Phillips, 1969) as is the increased length of the dominant shoots, which were as much as 81% longer with 100 ppm. It is suggested by Ruddat and Pharis (1966) that GA3 participates with auxin in maintaining lateral buds in an inhibited state. Certainly 1AA too had a slight enhancing effect upon dominance in the rhizomes at all three concentrations used but no mixtures of GA with 1AA have so far been tested on rhizomes. The third chemical which appeared to enhance dominance was U 29449. At 1 and 10 ppm the number of buds growing was consistently low but the rate of growth of the growing shoots was often greater than that of the controls, indicating that the chemical was not toxic.

Two chemicals seemed to affect polarity, in that the long potential or actual dominant shoots were more often in the middle or towards the base of the rhizomes than near the apical end as in untreated rhizomes. These were fluorodifen and to a lesser extent SD 8339 both at 50 ppm. Dominance was also delayed and this possible effect on polarity may be indicative of a mode of action different from that of the other chemicals delaying dominance.

The results reported here indicate that a number of chemicals are capable of regulating bud behaviour in rhizomes of <u>A. repens</u>. Further testing is necessary to assess their practical value in controlling this weed.

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A CONTINUOUS FLOW METHOD FOR STUDYING ADSORPTION AND DESORPTION OF PESTICIDES IN SOILS AND IN SOIL COLLOID SYSTEMS

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<u>Summary</u> A continuous flow method was used to study some adsorption and desorption characteristics of dimefox [bis(dimethylamino)fluorophosphine oxide], paraquat (1,1'dimethyl-4,4'-bipyridylium dichloride), and prometone (2,4-bisisopropylamino-6-methoxy-1,3,5-triazine) by soils, and by clay and humic acid preparations. Although some practical details must still be resolved it is suggested that the technique can be of value for predicting field application rates of pesticides.

INTRODUCTION

Osgerby (1970) has outlined a number of barriers which must be overcome by a pesticide when proceeding from the soil environment to the site of action in the plant. He has emphasized that adsorption by soil can be a major impediment to the presence of a biocidal concentration of chemical in the soil solution.

It is recognized that laboratory determined soil adsorption data can give erroneous indications about the performances of herbicides in the same soils under field conditions, where climatic and soil physical conditions can have important influences (see, e.g. Hance et al., 1968). Neverless, the adsorption and desorption characteristics of a chemical in any particular soil should influence field application rates, and this paper will describe our attempts to develop a method which will remove much of the tedium involved in determining these characteristics. The principles of this technique have already been outlined by Grice et al. (1972) when describing the adsorption of prometone by a humic acid preparation. Use of the technique is extended here to a study of adsorption of paraquat and dimefox by soils and by clay and humic acid preparations, and the difficulties which are encountered in such studies are emphasized. It is hoped that it will be possible to introduce a high degree of automation into the procedure in the future.

Operation of the Continuous-Flow Cell

Grice et al. (1972) have described minor alterations which were made when adapting an Amicon Model 12 Ultrafiltration Cell (from Amicon Ltd., 57 Queens Road, High Wycombe, Bucks) for studying the adsorption of prometone by a humic acid material. Briefly, the instrumentation consists of a reservoir to contain a known concentration of adsorbate solution, a cylindrical cell containing a magnetic stirrer and adsorbent suspended in water overlying a membrane, and a fraction collector. A constant pressure (nitrogen gas) is applied to the reservoir to force the adsorbate into the cylindrical cell system, and known volumes of the effluent from this cell are collected by means of the fraction collector and analysed by appropriate techniques.

In some instances soils were mixed with glass fibre and separated from the magnet and membrane by means of a perforated stainless steel disc.

Three types of membranes were used in the various adsorption studies. Two of these, the UM-2 (from Amicon) and the Pellicon SPAC [from Millipore (U.K.) Ltd., Millipore House, Abbey Road, Park Royal, London, NW10 7SP] had molecular weight exclusion values of <u>ca</u>. 1000, and the PM-10 membrane (from Millipore) had a similar quoted value of <u>ca</u>.10000.

Fig. 1 shows an example of wash-in and wash-out curves which are obtained when adsorbate is passed through a cell containing water and an aqueous suspension of adsorbent.



Fig. 1. Specimen 'wash-in' and 'wash-out' curves in the absence (I) and presence (II) of adsorbent

Methods for constructing adsorption isotherms from data contained in wash-in curves have been described in detail by Grice et al. (1972). The procedure can be readily understood by reference to Fig. 1. Consider any point X shown in the wash-in curve. At this point the following conditions must apply:

(i), the vol. of adsorbate solution that has entered the cell is equal to the eluate vol. V. Thus the amount of adsorbate which has passed into the cellⁿwill be $(c_{max} \cdot V_n)$;

(ii) The total amount of adsorbate that has left the cell will be given by the area under the curve AX (i.e. area AXB).

(iii), The concentration of adsorbate in solution in the ultrafiltration cell (vol. V) is C_n , and hence the amount of adsorbate which is free in solution may be written as V.C_n.

Thus at any point X the amount of adsorbate adsorbed by the adsorbent in the ultrafiltration cell may be expressed by the following relationship:

Adsorption at $C_n = C_{max} V_n - (area AXB + V.C_n)$.

These calculations assume that no adsorption or other interactions take place at the membrane. Where such effects take place a blank run must be carried out and any adsorption obtained must be subtracted out from that recorded when adsorbent is present in the cell.

Desorption may determined by exchanging water for the adsorbate solution in the reservoir. At the beginning of the desorption run the (assuming that the eluate concn is C_{max}) amount of adsorbate contained in the cell will be equal to the total amount (Γ) adsorbed by the adsorbent plus the amount (C_{max} V) which is free in solution in the cell. Consider now the situation which applies at any point Y in the wash-out curve (Fig. 1). The total amount of adsorbate which has left the ultrafiltration cell will be given by the area under the curve DY (i.e. area DYEF). The concentration of adsorbate free in the ultrafiltration cell is given by C_1 and hence the amount of adsorbate free in solution may be written as V.C₁ Thus at the point Y

Adsorption =
$$(\Gamma + V C_{max}) - (area DYEF + V C_{,}).$$

Adsorbent materials

Some characteristics of the four soils used, as supplied by the Rothamsted Experimental Station, are presented in Table 1. Here CEC refers to the cation

Soil	Clay content (%)	Organic carbon content (%)	CEC (meq/100g)	pH value
Adventurers	28	40	170	7.3
Brome Pin	12	1.2	10.7	7.7
Prick Willow	68	13.7	85.4	6.5
YNYS	36	1.0	7.1	4.3

Table 1

exchange capacity in milliequivalents (meq)/100g.

Sodium ion saturated montmorillonite (Na⁺-montmorillonite) was prepared according to the method of Barshad (1969), and the $\langle 0.2 \mu$ fraction, isolated by centrifugation, was used in the adsorption work. It had a CEC of 105 - 108 meq/100g (Pick,1972), and a pH (0.05g in 25 cm³ of distilled water) value of 8.7. Hydrogen ion - saturated humic acid $(H^+ - HA)$ was prepared by extracting a H^+ - ion saturated Fenland soil (80% o.m., washed with IN HC*l*) once with 0.5N NaOH, adjusting the pH of the extract to 1.0 with HC*l*, centrifuging, washing the precipitate with distilled water and then dialysing till chloride could not be detected in the dialysate. Non-dialyzable retentates were diluted with distilled water to the appropriate for adsorption studies. The CEC was 390 meq/100g and the pH value for the suspension was 3.8.

Adsorbate chemicals

Dimefox (99% pure) was supplied by The Murphy Chemical Co. Ltd., Wheathampsted, Herts. Crystalline paraquat was obtained pure from I.C.I., Plant Protection Ltd., Jealotts Hill Research Station, Bracknell, Berks., and prometone (95% purity) was supplied by J.R. Geigy S.A., Basle, Switzerland.

Analytical procedures

A Perkin-Elmer F11 GLC instrument, equipped with a series 900, dual, flame ionisation/halogen - phosphorus specific detector was used for the analysis of aqueous solutions of dimefox. Aliquots $(2 \ 1.)$ were injected into a 5ft. X 1/4 inch glass column packed with silicone elastomer S.E 30 (10% as the liquid phase), on a Universal Support material of 60 - 85 mesh size. Nitrogen (pressure = 31b/in² at a flow rate of 20 cm³/min) was employed as the gas phase. An oven temperature of 110°C and the appropriate (instrument settling $2\frac{1}{2}$) injection block temperature were used. Hydrogen (171b/in²) and air (251b/in²) pressures were as recommended. In order to measure the continual variation in detector response it was necessary to inject a standard sample after every third application of unknown solution.

Aqueous solutions of paraquat were analysed polarographically or spectrophotometrically. For polarography 0.5 cm³ of paraquat solution was added to 5 cm³ of base electrolyte $(0.2\underline{N} \ HC\ell)$. This was degassed and the wave height at -0.97 volts (vs. the mercury pool electrode) was measured using a Davis Differential Cathode Ray Polarograph. Spectrophotometric absorption was measured at 298 n.m. (maximum absorption for paraquat is at 256 n.m.) and a linear calibration curve was obtained at this wavelength for the concentration range used.

Prometone $\binom{14}{C}$ labelled) concentrations were assayed by liquid scintillation counting as described by Grice et al. (1972).

RESULTS AND DISCUSSION

Evaluation of membranes for adsorption studies

The ideal membrane, for use in adsorption studies employing the Continuous-Flow technique, should not interact with the adsorbent or adsorbate, and it should prevent adsorbent materials from being eluted from the cell. Because humic acid is a polydisperse polymeric material, with components ranging in molecular weights from 1.5 X 10³ to greater than 2 X 10⁶, it was necessary to work with the UM-2 and Pellicon SPAC membranes for H⁺- HA systems. These membranes were also used for studies with soils (because of the organic matter present), but it is now realised that the PM-10 membrane, which is satisfactory for studies on clays, can also be used when soils are predominantly saturated with divalent and trivalent exchangeable cations.

There was negligible interaction between the PM-10 membrane and paraquat. However, at a reservoir solution concentration of $2.26 \text{ meq}/\ell$., this chemical was held back by the UM-2 to the extent of $4.15 \times 10^{-3} \text{ meq/cm}^2$ of membrane surface. Under the same experimental conditions interaction with the Pellicon SPAC was appreciably less $(1.2 \times 10^{-3} \text{ meq/cm}^2)$. Paraquat was readily washed from the UM-2 membrane and this could indicate that surface concentration effects and not adsorption was involved. These phenomena are being investigated further.

Pellicon SPAC membranes were used exclusively for adsorption studies with dimefox. No measurable membrane - chemical interactions were detected.

The UM-2 membranes interacted to a small extent with prometone, and it was necessary to subtract out the membrane adsorption from the total adsorption in the cell system. However, maximum membrane adsorption at a reservoir concn of 13 $\mu \underline{M}/\ell$. amounted to only 0.7 $\mu \underline{M}/cm^2$, whereas adsorption by H⁺- HA was 725 $\mu \underline{M}/g$.

Adsorption and desorption isotherms

Dispersed clay was readily held in suspension in the 13 cm^3 capacity cell by the magnetic stirring device used. Aggregation resulted following the adsorption of paraquat, and this caused a deposition of clay to sediment over the membrane surface.



equilibrium concn (meq/l)

Fig. 2 Adsorption of paraquat by H^+- HA and $\text{Na}^+-\text{montmorillonite}$ preparations

Two isotherms are presented in Fig. 2 from data for the adsorption of paraquat by 0.05g of the $\langle 0.2 \text{ size Na}^+$ -montmorillonite clay preparation. Isotherm I was obtained when the concn in the reservoir was 2.26 meq/l. and the flow rate was 8 cm³/h. When the reservoir solution concn and the flow rate were lowered to 1.7 meq/l. and 2.86 cm³/h, respectively, isotherm II was obtained. This isotherm closely agrees with that obtained for the same clay preparation by means of the standard batch equilibration (slurry) technique (Pick, 1972). However, a reservoir concn of 6 meq/l. and the slower flow rate gave an isotherm similar to I. These data suggest that the "hump" in the adsorption isotherm (see curve I, Fig. 2) resulted from the entrapment of paraquat solution in the sedimented clay matrix. This entrapped material did not diffuse sufficiently rapidly into the bulk solution to maintain true adsorption - desorption equilibrium in the cases where the more rapid flow rate and higher reservoir solution concn were used. The isotherm slowly approached the true plateau level (as expressed by curve II) as adsorption was continued.

Fig. 2. also presents an isotherm from continuous-flow data for the adsorption of paraquat (reservoir concn $2\cdot7 \text{ meq}/\ell$.) by H⁺- HA (0.05g in 13 cm⁻³ cell vol.). The stirring device kept the aggregated organic materials suspended in the cell in this instance, and adsorption - desorption equilibrium was readily attained during the course of the experiment. Burns (1970) obtained an isotherm of similar shape by using the slurry technique, but his extent of adsorption was less than that found here. However, he used a H⁺- HA preparation which had a lower CEC (360 meq/g) and which had been freeze-dried.

Isotherms for the desorption of paraquat from montmorillonite (see curve I) and from humic acid are included in Fig. 2. Both of these isotherms indicate that the paraquat which is bound to the respective adsorbents by weak attractive forces can be readily desorbed with water. But most of the adsorbed material (probably bound by an ion-exchange mechanism) could not be desorbed under the experimental conditions used.

There was a tendency for soils to sediment (in the manner described for aggregated clays) during adsorption studies using the



Fig. 3 Isotherms [Continuous-Flow (I); Slurry (II)] for adsorption of paraquat by Prick Willow soil

Continuous-Flow technique. Thus difficulties were experienced in attaining true adsorption - desorption equilibria. In order to avoid sedimentation the Prick Willow soil was mixed with glass fibre in the manner described in the section on Method and Materials. Fig. 3 shows a comparison between the isotherm obtained from data for this modified procedure (curve I, paraquat reservoir solution concn $2 \cdot 5 + M/i$.) and that for the conventional slurry technique (curve II, where the maximum adsorbate concn used was 6 + M/i.). It can be seen that the isotherms are reasonably similar for comparable concn ranges. The general shape of these isotherms suggests that the high organic matter content (Table 1) of the Prick Willow soil influenced adsorption more than did the high clay content of that soil (compare the isotherm shapes of curve II, Fig. 2. with that for H⁺- HA, Fig. 2).

An isotherm for the adsorption of prometone by H^+ -HA from data for the Continuous-Flow method has already been compared with that from slurry technique data (see Grice et al. 1972). These isotherms were shown to be closely similar in every way.

Isotherms for the adsorption of dimefox by H⁺- HA, Na⁺montmorillonite, and YNYS and Prick Willow soils are presented in Fig. 4. All data were obtained from the Continuous-Flow procedure in the absence of a glass fibre filler. Similar experiments were carried out for the Adventurers and Brome Pin soils.



Fig. 4 Adsorption of dimefox

Na⁺-montmorillonite and H⁺- HA were not aggregated by the adsorbed dimefox and thus no difficulties were experienced in obtaining uncomplicated adsorption isotherms. The surface coverage of the clay, calculated from the molecular dimensions of dimefox and the amount adsorbed at the plateau level, amounted to 123 m²/g. This indicates that adsorption took place predominantly on the external surfaces of the clay. It was calculated, by use of the inverse Langmuir equation, that the monolayer coverage by the chemical amounted to 746 m²/g in the case of H⁺- HA.

The isotherm for adsorption on Prick Willow (Fig. 4) shows the characteristic "hump" which has already been attributed to the slow attainment of adsorption - desorption equilibrium in sedimented media. Similar shaped isotherms were obtained for adsorption by Adventurers and Brome Pin soils. Plateaus were characteristically reached (see isotherm for adsorption on Prick Willow) and adsorption of dimefox at these levels amounted to 2.3 and 0.8 g/100 for Adventurers and Brome Pin, respectively. Isotherm "hump" effects were absent in the case of adsorption by the YNYS soil (Fig. 4).

An attempt was made to correlate the adsorption of dimefox with the clay and organic matter contents of the preparations and soils studied. The data in Fig. 5 indicate that adsorption correlated better with clay than with organic matter contents, except in the case of YNYS which did not correlate with either parameter. In Fig. 5 the numbers 1,2,3,4,5, and 6 refer to YNYS, Brome Pin, Prick Willow, Adventurers, Na -montmorillonite and H - HA, respectively, and + refers to data for clays and the circles (o) those for organic matter.





CONCLUSIONS

Data which have been presented indicate that the Continuous-Flow method can provide a technique by which the capacity of a soil to adsorb a particular chemical can readily be determined. The procedure, where membrane - chemical interactions and sedimentation of adsorbent in the cell can be avoided, is theoretically sound. Further investigations should resolve the effects of different flow rates on adsorption - desorption equilibria, as well as the effects of higher adsorbate concentrations and the effects of bigger cell volumes and different adsorbent- adsorbate ratios in the cell.

Preliminary further studies in our laboratories indicate that the technique can be automated where suitable analytical procedures are available. It should also be possible to computerise these data. Comparison of adsorption isotherms with similar isotherms for soils with known herbicide performances could be of considerable value when deciding on field application rates for a particular chemical.

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AN APPROACH TO THE PREDICTION OF THE LEACHING OF HERBICIDES IN SOILS

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<u>Summary</u> The factors affecting the leaching of herbicides in soils are briefly discussed and a method for predicting this leaching is described. This consists of a semi-empirical method based on the equation of continuity and implemented on an analogue computer. The method is illustrated by a consideration of the residue profiles in four European soils of 2,6-dichlorobenzamide, a metabolite of the herbicides chlorthiamid and dichlobenil, following repeated annual applications of chlorthiamid to the soil surface. It is shown that there is good agreement between the residue levels found by the chemical analysis of soil samples and those predicted by the mathematical model.

INTRODUCTION

The rate of transport of a herbicide through the soil increases as the average rate of water flow increases, and this in turn depends upon the moisture characteristics of the soil. The rate of transport is also inversely proportional to the adsorption constant of the herbicide.

In principle it is possible to consider soil as analogous to a chromatographic support and therefore to apply chromatographic theory to calculate the distribution of herbicides in soils during leaching. A number of workers have used this approach. For instance Lindstrom et al (1971) and Oddson et al (1970) have used rate theories of chromatography and King and McCarty (1968) have used the plate theory of chromatography, but in most cases the calculations apply to the transport of herbicides within carefully packed columns of uniform soil through which water flows continuously. Clearly such conditions are not realistic and although their use leads to a better understanding of the principles of the leaching process it does not necessarily lead to a description of the leaching behaviour of herbicides under field conditions. The results of such work are therefore not entirely suitable for predicting either the availability of herbicides to plant roots or their transport and accumulation in soils over a long period of time.

Frissel et al (1970) have shown that it is possible to calculate the distribution of 90Sr in soil as a function of depth and time providing that the history of a particular site is known. These workers used over twenty parameters in a simulation model and their results agreed well with 90Sr distributions determined by the chemical analysis of soil samples. It is possible that their technique could be used to predict future 90Sr levels, although this would presumably involve using estimates of many of their parameters.

The exact prediction of the distribution of a herbicide in soil at times following its application is of course impossible. This would demand not only a knowledge of the history of a particular site but also of the future climatic conditions to which it will be subjected. Rather it is the order of magnitude of the distribution following repeated applications of a herbicide to the soil that is of interest.

This paper describes an attempt to predict the accumulation and distribution of 2,6-dichlorobenzamide (BAM) in four European soils. This compound is a metabolite of the herbicides chlorthiamid (PREFIX, 2,6-dichlorothiobenzamide) and dichlobenil (CASORON, 2,6-dichlorobenzonitrile). It has been shown by Leach et al (1971) to be absorbed by plant roots and to cause leaf margin chlorosis. Both chlorthiamid and dichlobenil are used for weed control in perennial crops and in the situations discussed below repeated annual applications of chlorthiamid were made to vines growing in France and Germany. Beynon and Wright (1968) have shown that chlorthiamid decomposes fairly quickly to dichlobenil in soil and this in turn decomposes more slowly to BAM. BAM is relatively weakly adsorbed by soils and it is leached through the soil much more readily than either of the other two herbicides.

METHOD AND MATERIALS

Long-term field trials established in vineyards near to Avignon, Bordeaux and Versailles in France and at Geisenheim in Germany for the purpose of the biological evaluation of chlorthiamid were used for this work. Chlorthiamid was applied to the soil surface annually in the spring, starting in 1968, as $7^{1}/2^{*}$ (w/w) granules. Plots treated at 12 kg/ha were sampled after harvest, usually in the autumn, using a hydraulic core sampler or by digging a pit. Samples of 10 or 15 cm depth were obtained, down to a total depth which varied between 45 and 60 cm in 1968 to 2 metres in 1971. These samples were analysed for BAM and for total nitrile by gasliquid chromatography using an electron-capture detector (Beynon and Wright, 1968).

The moisture equivalent of the soil samples was measured by the centrifuge method and the wilting point by equilibrating the samples at 15 atmospheres pressure in a pressure-membrane cell. The adsorption of ^{14}C -labelled BAM was determined using the slurry technique.

<u>Mathematical Model</u> The movement of a pesticide in a soil is caused by the movement of the water in the soil and also by the molecular diffusion of the compound in the water or the air. If diffusion in air is neglected the variation of the concentration of the pesticide in the soil water can be described by a combination of Ficks second law, adsorption and mass flow:-

$$\frac{\delta C_W}{\delta t} = D \frac{\delta^2 C_W}{\delta x^2} - \frac{1}{E} \frac{\delta S}{\delta t} - v \frac{\delta C_W}{\delta x} - k C_W$$
(1)

where C_W = concentration of pesticide in soil water ($\mu g/ml$), S = concentration of adsorbed pesticide ($\mu g/g$), E = fractional pore volume, v = velocity of water in the soil pores (cm/week), x = soil depth, positive downwards, D = bulk average diffusion coefficient, k = rate constant for the decomposition of the pesticide in the soil (week⁻¹).

If the adsorption process is represented as a completely reversible reaction with the rate constant for adsorption = k_a and the rate constant for desorption k_d then:

$$\frac{\delta S}{\delta t} = k_a C_W - k_d S$$
(2)

Equations 1 and 2 describe the behaviour of the compound in the soil subject to the assumptions made in their derivation. These are that the adsorption is linear and completely reversible; D is independent of concentration; the velocity of the water in the soil is a constant; the soil does not shrink or swell and so

Soil	depth (cm)	Bordeaux	Versailles	Soil depth (cm)	Avignon
	0-10 10-20 20-30 30-45 45-60	.614 .640 .628 .404 .325	.678 .678 .598 .382 .290	0-10 10-20 20-30 30-40 40-50	.978 .898 .811 .513 .537
		Geisenheim			
	0-15 15-30 30-45	• 337 • 346 • 373			

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14	DIG	1	

Transport	coefficients	calculated	from	soil	and	rainfall	data

Ta	bl	е	2
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Transport coefficients determined from analogue computer

Soil depth (cm)	Avignon	Bordeaux	Soil depth (cm)	Versailles	Geisenheim
0-20	0.600	0.200	0-10	0.068	0.260
20-40	0.331	0.347	10-20	0.100	0.100
40-60	0.700	0.700	20-30	0.120	0.600
60-80	0.800	1.000	30-40	0.120	1.000
80-100	0.600	1.000	40-50	0.120	1.000
100-120	1.000	1.000	50-60	0.120	1.000
120-140	1.000	1.000	60-70	0.120	1.000
140-160	1.000	1.000	70-80	0.120	1.000
160-180	1.000	1.000	80-90	0.120	1.000
180-200	1.000	1.000	90-100	0.120	1.000

Table 3

Predicted equilibrium residue levels (ppm)

Soil depth (cm)	Avignon (1973)	Bordeaux (1972)	Versailles (1978)	Geisenheim (1972)
0-20 20-40 40-60 60-80 80-100 100-120 120-140 140-160	0.16 0.45 0.20 0.27 0.16 0.15 0.15	0.06 0.18 0.09 0.06 0.06 0.06 0.06	0.55 0.32 0.22 0.10 0.03	0.2 0.104 0.03 0.02 0.02
160-180 180-200	0.15	0.06		

E is a constant.

The combination of equations (1) and (2) is a third-order partial differential equation, which would be very difficult to solve. This may be simplified by assuming that diffusion is small compared with mass flow (ie. D can be ignored) and also that adsorption equilibrium is attained rapidly (ie. k_d is large). We then have:-

$$\frac{\&C_W}{\&t} = \frac{1}{1 + K/E} \left[-v \frac{\&C_W}{\&x} - kC_W \right]$$
(3)

This may be transformed into a finite difference equation by discretising x:-

$$\frac{d(C_W)_n}{dt} = \frac{v}{1 + K/E} \left[(C_W)_{n-1} - (C_W)_n \right] - \frac{k(C_W)_n}{(1 + K/E)}$$
(4)

Equations essentially similar to (1) and (2) have been solved by Oddson et al (1970) in the form of Bessel functions after introducing the same simplifying assumptions invoked above. These workers and others have shown that this approach leads to a satisfactory description of the leaching of herbicides in columns of soil under laboratory conditions.

The term v/(1 + K/E) may be considered as a transport coefficient. The finite difference equation (4) implies that the soil can be approximated as a series of n bands or compartments of equal depth. Such an equation can be readily solved by means of an analogue computer, each "soil compartment" being represented by a particular computing integrator, each of which is assigned its own potentiometer the setting of which represents the value of the transport coefficient for that particular compartment.

In principle the transport coefficients can be calculated from a knowledge of the terms v, K and E, but these are variables over a period of time in practical situations. An estimate of E was however used based on the assumption that during the period from the application of the herbicide to the time at which soil samples were taken (6-9 months) the volume of water concerned in the leaching process was equal to the difference between the field capacity (or moisture equivalent) of the soil and the wilting point. The velocity of the water through the soil (v) was assumed constant and equal to the total rainfall during the period divided by the length of that period. This is clearly unrealistic and is likely to give a maximum value.

The leaching process was first simulated on the analogue computer using transport coefficients calculated in this way. Later, however, the computer was used to produce curves which most closely matched the BAM distribution curves determined from the chemical analysis of soil samples. This was done by finding the optimum setting of the potentiometers by trial and error. These settings, corresponding to the transport coefficients, were then noted.

RESULTS

Equation 4 was first solved using transport coefficients calculated from soil adsorption data, rainfall records and measurements of the moisture equivalent and wilting point of the various soil samples. These coefficients are shown in Table 1. The solutions were compared with EAM residues found in the autumn of 1968 at the four trial sites following the first application of chlorthiamid in the spring of 1968 and the agreement was found to be fairly good. However, predictions for the residues of EAM that would occur following a further application of chlorthiamid in the autumn of 1969.

Depth (cm)	0-10	10-20	20-30	30-40	40-50	50-60	60-70	70-80	80-90	90-100
Versailles								-		
1968	0.18	0.04	<0.01	-	-	-	-	-		
(Residues found)	(0.09)	(0.03)	-	-	-	-	-	-	-	
1969	0.32	0.10	0.03	<0.01	-	-	-	-	-	
(Residues found)	(0.45)	(0.23)	(0.16)	(0.13)	(0.10)	-	-	-	-	-
1970	0.42	0.17	0.08	0.03	0.01	<0.01	<0.01	-	-	
(Residues found)	(0.34)	(0.08)	(0.06)	(0.06)	(0.06)	(0.03)	(0.01)	(<0.01)		
1971	0.50	0.23	0.11	0.06	0.02	0.01	<0.01	<0.01	-	_
(Residues found)	(0-20 cm	0.12)	(20-40	cm 0.11)	(40-60 cm	n 0.09)	(60-80	cm 0.05)	(80-100	cm 0.03)
Geisenheim										
1968	0.06	0.07	0.01	<0.01	<0.01	-	-	-	-	-
(Residues found)	(0.03)	(0.05)	(0.03)	(0.01)	-	-	-	-	-	
1969	0.09	0.15	0.02	0.01	0.01	<0.01	<0.01	-	-	2
(Residues found)	(0.09)	(0.15)	(0.06)	(0.02)	-	-	-	-	-	_
1970	0.10	0.22	0.03	0.02	0.02	0.01	0.01	<0.01	<0.01	
(Residues found)	(0.24)	(0.11)	(0.03)	(0.02)	(<0.01)	-	-	_	-	
1971	0.10	0.26	0.04	0.02	0.02	0.02	0.02	0.02	0.02	0.01
(Residues found)	(0-20 cm	0.45)	(20-40	cm 0.2)	(40-60 cr	n 0.07)	(60-80	em 0.03)	(80-100	cm 0.03)

Table 4

Calculated residues of BAM in soils (ppm % dry wt.) following application of PREFIX at 12 kg/ha

Depth	(cm)	0-20	20-40	40-60	60-80	80-100	100-120	120-140	140-160	160-180	180-200
Avignon											
1968		0.14	0.29	0.12	0.08	0.06	0.02	0.01	<0.01	-	-
(Residues	found)	(0.04)	(0.20)	(0.06)	-	-	-	-	-	-	-
1969		0.16	0.42	0.20	0.16	0.18	0.10	0.08	0.07	0.05	0.04
(Residues	found)	(0.08)	(0.25)	(0.30)	-	-	-	-	-	-	-
1970		0.16	0.40	0.20	0.15	0.20	0.10	0.09	0.08	0.07	0.06
(Residues	found)	(0.28)	(0.41)	(0.23)	(0.13)	(0.23)	(0.13)	(0.09)	(0.06)	(0.06)	(0.05)
1971		0.16	0.44	0.22	0.19	0.24	0.14	0.13	0.12	0.11	0.11
(Residues	found)	(0.43)	(0.23)	(0.14)	(0.04)	(0.05)	(0.08)	(0.08)	(0.07)	(0.08)	(0.10)
Bordeaux											
1968		0.16	0.07	0.03	0.02	0.01	<0.01	<0.01	<0.01	<0.01	<0.01
(Residues	found)	(0.4)	(0.21)	(0.08)	(<0.01)	-	_	-	-	-	-
1969		0.23	0.13	0.06	0.04	0.03	0.03	0.02	0.02	0.01	0.01
(Residues	found)	(0.29)	(0.21)	(0.14)	(0.07)	-	-	-	_	-	-
1970		0.26	0.16	0.08	0.05	0.05	0.05	0.04	0.04	0.04	0.03
(Residues	found)	(0.26)	(0.16)	(0.06)	(0.03)	(0.04)	(0.03)	(0.03)	(0.02)	(0.02)	(<0.01)
1971		0.26	0.17	0.09	0.06	0.06	0.06	0.05	0.05	0.05	0.05
(Residues	found)	(0.38)	(0.18)	(0.08)	(0.06)	(0.06)	(0.04)	(0.02)	(<0.01)	-	-

Table 4 (contd.)

Calculated residues of BAM in soils (ppm % dry wt.) following application of PREFIX at 12 kg/ha
Better agreement with the BAM residues found in the autumn of 1968 was obtained when the transport coefficients of equation 4 were varied until a best-fitting curve was obtained. It was then found that these same transport coefficients, shown in Table 2, gave reasonably good predictions of the BAM residue levels following repeated applications of chlorthiamid in 1969, 1970 and 1971. These residue levels are shown in Table 4. As would be expected the results show most variation, both from year to year and between calculated and found values in the soil layer nearest to the surface. Within this layer the greatest degree of movement is likely to occur, both downwards due to the leaching action of rain and upwards due to evaporation of water from the soil.

DISCUSSION

It is seen from the results in Table 4 that the method of prediction of EAM residues in soils outlined above adequately describes the long-term distribution of the compound in the four soils used. Although the method is based on sound physical principles in the form of the equation of continuity (equation 1) it is essentially an empirical method. Such a method cannot be implemented until analytical data of the residues in the soil at the end of the first year becomes available. These data are then sufficient to derive the necessary transport coefficients from which the residues of future years can be predicted without alteration. Optimum values of these coefficients could be obtained by updating them as successive annual residue data became available, but clearly one purpose of the method is to eliminate such sampling and analysis. However, since annual variations in soil and climatic conditions could be large occasional soil samples should be analysed to act as a check on the reliability of the predictions.

There is no doubt that the use of this predictive method would be more satisfactory if the transport coefficients could be calculated from soil and climatic data. This may prove to be possible as more experience of the technique is gained. It is clear from Tables 1 and 2 that the term v/[1 + K/E)] already gives values of the same order of magnitude as the more satisfactory coefficients found by the curve-fitting method.

This particular model has been used to predict the probable build-up and distribution of residues of BAM if annual applications of chlorthiamid continue indefinitely. This shows that an equilibrium situation is achieved within four years of continual application at Geisenheim and Bordeaux, within five years at Avignon and within ten years at Versailles. The rather long time required to establish equilibrium at Versailles is probably because a clay pan underlies the soil at this site. These equilibrium levels are shown in Table 3. The model also predicts that if the application of chlorthiamid was stopped once equilibrium had been achieved the residues of BAM would be reduced to low levels within two years at Avignon and Bordeaux and within three to five years at Geisenheim and Versailles.

In principle it is possible to use this predictive model to estimate the uptake of herbicides and their metabolites by plant roots. This involves superimposing a second model for the distribution of perennial plant roots or for the growth and consequent distribution of annual crop roots. This approach has been discussed by Osgerby (1972) but so far little experience has been gained with this technique.

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INFLUENCE OF SOIL FACTORS ON AVAILABILITY OF ATRAZINE AND LINURON TO PLANTS

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<u>Summary</u> The concentrations of atrazine and linuron in the shoots of wheat plants growing in 10 different soils were directly proportional to the concentrations of herbicide in the soil solution estimated from slurry adsorption measurements. There was a discrepancy between the total uptake of each herbicide and the amounts theoretically supplied to the plants by mass flow in response to transpiration. This discrepancy was greater with linuron than with atrazine. The adsorption of atrazine was more nearly reversible than was that of linuron and it is suggested that desorption rates may be of importance in determining herbicide availability to plants in different soils over a period of time.

INTRODUCTION

The results from several investigations into the absorption and translocation of triazine and urea herbicides by plants in culture solution have suggested that uptake is a passive process related to the concentration of herbicide in solution and the amount of water transpired by the plant (Sheets, 1961; Geissbuhler et al., 1963; Wheeler & Hamilton, 1968; Nashed & Ilnicki, 1970; Shone & Wood, 1972). Where uptake from soil is concerned, solution concentrations of herbicide are not easily defined since a large proportion of the total amount of herbicide in the system will be adsorbed by the soil. Adsorption can readily be determined under slurry conditions where a small amount of soil is shaken with an excess of solution but extrapolation from these measurements to adsorption under the soil moisture regimes at which plants grow may not be valid (Grover & Hance, 1970). Continued availability of herbicide to plants in soil will also depend upon the reversibility of the adsorption process. After plant uptake of herbicide and addition of water to the soil to replace that lost through evaporation and transpiration, the initial equilibrium of herbicide between adsorbed and solution phases will be disturbed, and herbicide must be desorbed in order to maintain the initial state. Previous experiments with atrazine have shown that uptake by wheat and turnip seedlings growing in different soils can be predicted with some degree of accuracy when slurry adsorption measurements are used to estimate soil solution concentrations of herbicide (Walker, 1972a). The present experiments were made to compare the uptake of atrazine by wheat with that of linuron, a more strongly adsorbed compound, to determine whether similar predictions of uptake can be made for compounds with different adsorption characteristics.

EXPERIMENTAL METHODS AND RESULTS

The soils used in these experiments were obtained from different fields at the National Vegetable Research Station and from several outside sites. Their properties are shown in Table 1. The herbicides used were carbon-14 ring-labelled atrazine (specific activity, 10.22 µGi/mg) and carbon-14 carbon/1-labelled linuron (specific activity, 0.35 µCi/mg). Heroicide adsorption by the soils was measured by shaking duplicate 5-3 amounts of 2-mm sieved, air-dry soil with 20 ml herbicide solution in 0.02 % calcium chloride. The initial herbicide concentration for both atrazine and linuron was 0.05 μ /ml. After 24 h equilibration, the susbensions were centrifuged and auplicate 2 ml subsamples of the supernatant were transferred to counting vials to which 10 ml scintillation liquid was added (Bray, 1960). The activity of the samples was determined using a Tracerlab Corumatic 200 liquid scintillation counter and quench corrections were determined by the internal standard method. Samples of the original herbicide solutions were counted similarly and the extent of herbicide assorption by the soils calculated from the difference between initial and final count rates. Ausorption was expressed as a distribution coefficient or kd-value for each soil-herbicide combination (Talbert & Fletchall, 1965) and the results are shown in Table 1. To examine the reversibility of the adsorption process, the two herbicides were adsorbed onto the soils as described above, but followin 24 h equilibration, 15 ml of the supernatant was replaced with 15 ml 0.02 % calcium chloride and the suspensions were reshaken for a further 24 h, after which the new distribution of herbicide was determined. The expected concentration of herbicide in solution could be predicted from the initial adsorption measurement and the determined solution concentrations as percentages of those expected are shown in Table 1.

(Soil Site	Carbon (,.)	Clay (%)	Atrazine		Linuron	
			Ads	Des	Ads	Des
NVRS Soils						
Little Cherry	0.70	16	1.7	91	4.2	77
Soakwaters	1.07	27	2.2	37	6.2	77
Townsend	1.32	14	2.4	100	3.0	79
.lum Orchard	1.32	18	3.1	97	6.6	74
Gallas Leys	1.50	34	3.0	35	3.1	70
Others						
Bristol	0.90	19	2.1	35	0.6	72
Esher	0.94	5	1.6	91	9.8	74
kufford	1.01	17	1.9	99	0.7	37
Wokingham	1.30	9	2.9	94	2.7	bd
lamworth	2.02	11	3.1	98	15.8	74

Table 1

Soil properties and herbicide adsorption and desorption

Ads = Adsorption distribution coefficient

Des = Measured equilibrium concentration after 24 h desorption as a percentage of that expected.

To measure uptake of the two herbicides by wheat seedlings, separate amounts

of each soil were treated with atrazine in methanol to give a concentration of 0.005 $\mu_{5/2}$ ary soil, or with linuron in actione to give a concentration of 0.150 $\mu_{5/2}$ dry soil. When the solvents had evaporated, the soils were rewetted to 8 $_{2.5}$ moisture by adding the required amount of water and were thoroughly riddle-mixed. Duplicate pots, containing 250 g soil and 5 pregeninated seeds of wheat, were breaked from each soil-heroicide componition. The surface of the soil was covered with a 1 cm layer of polystyrene granules to reduce surface evaporation of water from the pots. The soil in the pots was moistened to 75 $_{2.6}$ pot capacity by standing in petri-dishes containing the required amounts of water. Pot weights were returned to their original weight by watering from above. After 10 days the plants were cut off at seed level and their herbicide content determined following the extraction and radioassay procedures previously described (Walker, 1972a; 1972b). The roots were also washed free from soil and their nerbicide content determined.

Soil solution concentrations of herbicide were calculated from the following expression which assumes linear adsorption isotherms with slope $K_{\rm d}$:

$$C_{m} = C_{w} (K_{d} + V) \qquad . \qquad . \qquad . \qquad 1.$$

where G_{L} = concentration of herbicide in the soil (µg/g dry soil)

 $C_{uv} = \text{concentration of herbicide in solution } (uv/ul)$

ha = adsorption distribution coefficient

V = soil moisture content (ml/g)

The relationships between the shoot concentrations of atrazine and linuron and these estimated soil solution concentrations are shown in Fig. 1, and it can be seen that these were approximately linear for each herbicide. The amounts of water transpired by the plants in the different treatments were estimated from the differences in weight loss between pots with plants and pots with soil only under the same conditions and in Fig. 2, the determined total uptake of the herbicides by the wheat seedlings is compared with the theoretical supply by mass-flow in response to transpiration (the volume of water transpired multiplied by the soil solution concentration of herbicide). It can be seen that for both atrazine and linuron, the amounts of herbicide taken up by the plants were less than the amounts supplied, with the data for linuron showing a greater difference than those for atrazine.

DISCUSSION

The results in Fig. 1 show that the relative availability of both atrazine and linuron to plants growing in the ten soils is determined by their adsorption by the soils, with linear relationships being obtained between shoot concentrations and soil solution concentrations in each case. The results in Fig. 2, however, show that total uptake of the two herbicides by the seedlings was less than that expected on the basis of supply by mass-flow. The extent of this difference was greater for linuron - the more strongly adsorbed herbicide. Supply to the plants was calculated on the assumptions that firstly, solution concentrations of herbicide could be predicted from slurry adsorption measurements and secondly, that these solution concentrations remained constant throughout the 10 day period of the experiments. Grover & Hance (1970) reported that adsorption of linuron varied with changes in soil : solution ratio; they showed a decrease in Freundlich K value from 12.3 to 2.7 as ratios increased from 1 : 10 to 4 : 1. Similar changes in the extent of adsorption, however, would not explain the apparent discrepancies in the present results - the data in Fig. 2 would suggest an increase in the strength of adsorption at high soil : solution ratios. Such an increase in the





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strength of alsorption was reported by Scott & Lutz (1971) working with realinite suspensions. Using a pressure membrane technique, they showed that the concentration of diuron in the water removed from clay suspensions with applied pressures between 0.3 and 15 atmospheres was only 00 $_{20}$ of that in the solution extracted within the range of applied pressure between 0 and 0.3 atmospheres. In similar experiments with atrazine, the concentrations in the high pressure extracts showed little change from those in the low pressure extract. Green & Obien (1969) also demonstrated that the concentration of atrazine in the water extracted from soils at low soil moisture contents was similar to that predicted from slurry adsorption measurements.

During preparation of the soil samples in the uptake experiments described above, the soil-heroicide mixtures were allowed to dry and were subsequently rewetted before planting of the wheat seedlings. Under these conditions, we are dealing with a desorption rather than an adsorption equilibrium. The results in Table 1 show that neither atrazine nor linuron adsorption is fully reversible within 24 h of vigorous shaking. Graham - Bryce (1967) reported that when soil samples were allowed to dry between adsorption of an insecticide and its subsequent desorption, then desorption did not reach its expected equilibrium. Therefore in addition to possible errors associated with extrapolation from slurry adsorption measurements, the solution concentrations estimated from equation 1 may be overestimates due to incomplete reversibility of the adsorption process. This irreversibility of adsorption may also have important implications concerning the continued availability of the herbicides to the plants during the 10 day experimental period. Although steps were taken to reduce the water losses from the pots, it was necessary to water them at 3-day intervals during the experiment. Since the plants were removing herbicide from the soil and since it was necessary to add more water, the initial equilibrium conditions would be changed and more herbicide must have been desorbed in order to maintain the solution concentrations constant. Incomplete reversibility of adsorption would accentuate the changes in solution concentration and the errors associated with the use of equation 1.

The above results illustrate several factors which may be of consequence in governing herbicide behaviour under field conditions. The data in Fig. 2 provide further evidence that herbicide uptake by plants is a passive process related to the concentration in the soil solution and the amount of water transpired by the plant. With an uneven distribution of herbicide in the soil, uptake of water from the zones in which the herbicide is located will be a prerequisite for effective action. Since under field conditions the herbicide will probably be located close to the soil surface and in soil which is prone to wide fluctuations in moisture content, sufficient moisture in this surface layer will be essential early in the growth of weed seedlings if the herbicide is to be effective. With a sequence of wetting and drying cycles, desorption rates may be important in regulating the amounts of herbicide in the soil solution available for uptake by the plant and it would be interesting to examine the possible importance of moist compared with dry soil surface conditions at the time of application in determining subsequent herbicide activity.

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