EFFECT OF FIELD APPLICATIONS OF ISOTHIOCYANATE PRECURSORS ON PESTS AND DISEASES OF OILSEED RAPE

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INTRODUCTION

The glucosinolates are a group of compounds characteristic of the order Capparales and particularly the family Brassicaceae (= Cruciferae), including oilseed rape (Brassica napus L.). When the cells in which they are stored are damaged, they are catabolised to various compounds, including isothiocyanates, which are biologically active against pests and pathogens (Chew 1988). Experiments at Rothamsted have shown that applications of synthetic isothiocyanate-releasing compounds decreased the incidence of several diseases in oilseed rape plots (Rawlinson et al. 1985). Here we present data from field experiments in 1988-89 and 1989-90 involving applications to plots of formulations of precursors designed to release 3-butenyl isothiocyanate and phenylethyl isothiocyanate, compounds which are perceived by certain oilseed rape pests (Blight et al. 1989), and are toxic to certain fungal pathogens in the vapour phase (Dawson et al. in preparation). We monitored the distribution of pests and the incidence and severity of fungal disease on treated plots to determine the ecological effects and possible crop protection benefit of altering the volatile isothiocyanate profile of the crop.

MATERIALS AND METHODS

The compounds used as precursors of 2-phenylethyl isothiocyanate and 3-butenyl isothiocyanate were 3,5-bis-(2-phenylethyl)-1,3,5,-thiadiazine-2-thione (Compound A) and 3,5-bis-(3-butenyl)-1,3,5,-thiadiazine-2-thione (Compound B), respectively. Their synthesis, degradation and other properties will be described elsewhere (Dawson et al. in preparation). They were formulated as emulsifiable concentrates in xylene (36%), acetophenone (12% v/v) and Ethylan BV (3% w/v) in 1988-89 and as solutions in tetrahydrofurfuryl alcohol in 1989-90.

Five treatments (I - V) were applied to field plots (4 x 4m. in 1988-89 and 10 x 3m. in 1989-90) of cv. Cobra on three occasions in each season as shown in Table 1. Standard pesticides were applied at recommended rates using a hydraulic sprayer but formulations of precursors were applied using an electrostatically-charged rotary atomiser sprayer (Arnold and Pye 1980). Treatments I-V were replicated five times in each season; plots were arranged in a quasi-complete Latin square design to compensate for possible unilateral invasion by insects (Freeman 1979). Pest incidence was assessed prior to, and for several days after, each application; disease was assessed prior to, and one month after, each application.

Complementary bioassays (Griffiths et al. 1989; Dawson et al. in preparation) confirmed that isothiocyanates were released from the formulations at concentrations which influenced insect feeding and inhibited fungal development in the laboratory.

Table 1. Design of experiments

		1988-89	1989-90
	Times of Treatments:	9 November 12 April 2 June	6 November 4 April 23 May
I		Control	
II	Insecticides:	Gamma HCH Gamma HCH Triazophos	Gamma HCH Gamma HCH Triazophos
	Fungicides:	Prochloraz Prochloraz Iprodione	Prochloraz Prochloraz Iprodione
III	Compound A:	6.0% w/v ; 252 g. a.i. ha^{-1}	
IV	Compound B:	4.2% w/v 176g. a.i. ha^{-1}	8.4% w/v 352g. a.i. ha^{-1}
V	Mixed formulation:	compound A (3.0%) compound B (2.1%)	compound A (3.0%) compound B (4.2%)

EFFECT ON PEST AND DISEASE INCIDENCE

Precursor applications affected the incidence of *Psylliodes chrysocephala* (cabbage stem flea beetle), but not consistently. The application in November 1988 had no immediate effect but by the following February, there were significantly fewer larvae on plots which had received an application of compound A; there was no effect in 1989-90 (data not shown).

The most significant effects of precursors were on the incidence and severity of infestation by *Meligethes* spp. (pollen beetle) and *Ceutorhyncus assimilis* (seed weevil) after the second and third applications, respectively, in each experiment (Table 2, A., B.). In 1988-89, compound B decreased the incidence of *Meligethes* on primary racemes and also the proportion of pods with *C.assimilis* feeding holes. Consequently, the application rate for compound B was doubled in the 1989-90 experiment. In this season, similar effects on the incidence of *Meligethes* and *C.assimilis* occurred after application of both compound B and the mixed formulation; compound A also had a transient effect on the number of *C.assimilis* feeding holes on pods (Table 2, A., B.). Few *Dasineura brassicae* (brassica pod midge) were present in 1988-89, but in 1989-90 compound B and the mixed formulation significantly decreased the number of eggs and larvae in pods (Table 2, C.).

The major diseases were light leaf spot (*Pyrenopeziza brassicae*) and dark leaf and pod spot (*Alternaria brassicae*) on leaves and pods and canker (*Leptosphaeria maculans*) on stems

Table 2. Effect of isothiocyanate precursor applications on pest incidence on oilseed rape plots

A. Pollen Beetle (Number of beetles per primary raceme)

	1988-89		1989-90	
Assessment Date:	13 April	19 April	5 April	9 April
I. Control	8.18	4.36	1.23	1.07
II. Standard	0.56***[1]	0.70***	0.24*	0.07*
III. Compound A	7.40	3.62	1.10	1.15
IV. Compound B	6.28*	4.22	0.85*	0.82
V. Mixed formulation	6.66	3.88	0.88*	1.02
$S.E.D._{.05}$	0.82	0.54	0.11	0.20

B. Adult seed weevil damage to pods

	1988-89 (Percent pods with feeding holes)		1989-90 (Number of feeding holes per pod)[2]	
Assessment Date:	9 June	22 June	31 May	11 June
I. Control	69.8	73.6	1.75	1.92
II. Standard	65.4	54.6*	1.43*	1.69
III. Compound A	60.4	70.0	1.43*	1.82
IV. Compound B	68.0	60.8*	1.23*	1.60*
V. Mixed formulation	74.4	63.8	1.20*	1.56*
$S.E.D._{.05}$	6.85	5.58	0.12	0.13

C. Brassica pod midge (Number of eggs and larvae per pod)[3]

Assessment Date:	31 May 1990	11 June 1990
I. Control	3.14	5.30
II. Standard	1.31	1.66*
III. Compound A	4.88	4.23
IV. Compound B	2.16	2.07*
V. Mixed formulation	1.48	2.43*
$S.E.D._{.05}$	1.44	1.19

Notes:
All figures are mean values for five replications.
[1] - asterisks represent level of significance of difference from control; * - $P<0.05$, *** - $P<0.001$.
[2] - approximately 100% of pods infested in 1989-90.
[3] - very few pod midge present in 1988-89

(Table 3). Precursor-treated plots often had less disease than untreated plots but in neither season were these differences sustained and no precursor formulation affected disease as much as the standard fungicides.

Yields were taken from plots in the 1989-90 experiment, but only plots receiving standard pesticide applications yielded significantly more than control (2.24 t/ha vs. 1.52 control, S.E.D. = 0.12).

Table 3. Effect of isothiocyanate precursor applications on dark leaf and pod spot (DLS), canker (C) and light leaf and pod spot (LLS) incidence in oilseed rape plots.

1988-89

Assessment Date:	(% leaves) 17 April		(stem score[1]) 5 July	(pod infection[1]) 5 July	
	DLS	LLS	C	DLS	LLS
I. Control	8.5	61.4	0.17	2.86	2.75
II. Standard	1.6^{***2}	16.9***	0.04	0.07*	1.08
III. Compound A	8.0	60.0	0.22	1.32	2.62
IV. Compound B	5.5	56.8	0.18	3.60	1.63
V. Mixed formulation	11.3	60.4	0.14	1.66	2.24
S.E.D. $_{.05}$	1.52	5.86	0.078	1.133	0.781

1989-90

Assessment Date:	(% leaves) 18 May		(stem score[1]) 9 July	(pod infection[1]) 9 July	
	DLS	LLS	C	DLS	LLS
I. Control	29.8	58.1	0.35	0.28	0.38
II. Standard	8.7***	24.5***	0.17	0.06***	0.36
III. Compound A	24.6	59.9	0.29	0.28	0.54
IV. Compound B	27.6	62.9	0.33	0.16*	0.47
V. Mixed formulation	17.5	60.2	0.28	0.10	0.54
S.E.D. $_{.05}$	4.54	3.76	0.111	0.045	0.168

Notes:
All figures are mean values for five replications.
1 - severity index derived from Anon (1985)
2 - asterisks represent level of significance of difference from control; * - $P<0.05$, ** - $P<0.01$, *** - $P<0.001$

DISCUSSION

Applications of precursors designed to release specific isothiocyanates have affected pest distribution and to a lesser extent, disease incidence. Studies have shown that specialized pests of oilseed rape are generally attracted to isothiocyanates but this effect appears to be concentration-dependent (Finch 1978); in these experiments, pest incidence was often significantly less on plots treated with precursor formulations, but never significantly greater. Evidence from laboratory studies suggests that the effects of these precursor applications on pests and pathogens are due to the release of isothiocyanates rather than to intermediate breakdown-products or other aspects of formulation; further studies are in progress to confirm that the precursors also release isothiocyanates under field conditions.

Compound B consistently affected pest distribution more than compound A and the effects of the mixed formulation were probably attributable to the presence and concentration of compound B. Compound B degrades faster than compound A in vitro but at this stage we have no further explanation for the ineffectiveness of compound A.

Isothiocyanate-releasing compounds, such as dazomet and metham are effective as soil fumigants against fungal inoculum (Worthing 1983) and isothiocyanate precursors significantly decreased foliar disease in earlier trials (Rawlinson et al. 1985) but they were relatively ineffective in the present experiments. This may be due in part to the application of treatments to coincide as nearly as possible with the arrival of pests rather than with the onset of fungal infection; as a result some diseases were only partially controlled even on plots which received standard fungicides.

Laboratory bioassays are in progress to identify those glucosinolate catabolites which best combine fungitoxicity with a beneficial influence over pest behaviour. These preliminary experiments have demonstrated that the use of synthetic precursors will be useful for testing the efficacy of isothiocyanates in the field. The long-term aim of this work is to reduce reliance on pesticides by identifying compounds or combinations of compounds which could be enhanced in the vegetative parts of oilseed rape cultivars so that they release the most effective protectant compounds during infestation and infection (Dawson et al. 1989).

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