Effects of certain herbicides on the *in vitro* hatch of *Globodera rostochiensis* and *Heterodera schachtii*

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SUMMARY

Low concentrations of the active ingredient of various herbicides did not elicit any hatch from cysts of *Globodera rostochiensis*, or have any marked effect on subsequent hatch in host root diffusates (RD). Placing cysts into solutions of aldicarb plus herbicides before transfer to RD reduced the rate of hatch but not the total hatch. Exposing cysts of *G. rostochiensis* and *Heterodera schachtii* to the formulated herbicide compounds at medium field application rates before transfer to RD adversely affected hatch : two herbicides gave only slight effects on subsequent hatch but the thiocarbamate herbicides prevented any hatch in RD.

RÉSUMÉ

Influence de certains herbicides sur l'éclosion in vitro *de* Globodera rostochiensis *et* Heterodera schachtii

A faibles concentrations, la matière active de divers herbicides ne provoque aucune éclosion hors des kystes de *Globodera rostochiensis* et n'a aucun effet marqué sur l'éclosion ultérieure dans le diffusat radiculaire (ou DR) de la plante-hôte. Le trempage des kystes dans une solution d'alticarbe additionnée d'herbicide, avant leur transfert dans le DR, réduit le taux d'éclosion mais non sa valeur totale. L'exposition des kystes de *G. rostochiensis* et *Heterodera schachtii* à des herbicides en formule composée, aux taux moyens d'utilisation en champs et avant transfert dans le DR, a une influence variable sur l'éclosion : deux herbicides n'ont qu'un effet léger sur l'éclosion ultérieure, mais les herbicides contenant du thiocarbamate suppriment toute éclosion dans le DR.

An initial step in the hatching sequence of some cyst nematodes is a change in permeability of the lipid layer of the eggshell which is likely to be a Ca^{2+} - mediated structural alteration induced by hatching agents (Perry, 1987). Substances which alter the permeability of the lipid layer may initiate hatching and result in the death of juveniles in the absence of host plants. Alternatively, such substances may render the unhatched juvenile susceptible to environmental extremes by altering the protection afforded by the eggshell and the trehalose in the eggfluid (Perry, 1983).

Thiocarbamate herbicides are known to cause membrane disintegration and altered permeability in plant cells. Kraus and Sikora (1981, 1983) found that di-allate, the active ingredient (a.i.) of the herbicide Avadex® (Monsanto plc), significantly increased hatch of *Heterodera schachtii* perhaps because of an effect on the lipid layer of the eggshell, the presence of which has recently been demonstrated (Perry & Trett, 1986). The current a.i. of Avadex BW® is tri-allate, and we examined the effects of tri-allate and the formulated compound, together with other herbicides, on the *in vitro* hatch from cysts of *Globodera rostochiensis* and *H. schachtii.*

Materials and methods

Cysts of both species were from single generations extracted from pot cultures using standard techniques (Southey, 1986). *G. rostochiensis* Ro 1, grown on potato cv. Désirée, were stored dry at 5° for at least six months after extraction. *H. schachtii,* grown on cabbage cv. Hispi, were stored in moist soil at 2° for six months and extracted immediately before experimentation.

Root diffusate was collected (Fenwick, 1949) from 10 wk-old potato plants (PRD) or 6-7 wk-old sugar beet plants (SBRD) grown in sterile loam pot cultures. Diffusate was stored in polythene bottles at 2° until required when SBRD was used undiluted and PRD was diluted with glass distilled water (GDW) 1 in 4 by volume. Soil leachate (SL) was obtained (Fenwick, 1949) from unplanted pots of sterilised loam.

Before hatching tests, cysts of *G. rostochiensis* and *H. schachtii* were pretreated by soaking for 7 d and 1 d respectively in GDW at 20°. For each test solution, hatching tests were done on four batches of 25 soaked cysts, each batch held at 20° in a covered excavated glass block containing approximately 2 ml of solution. Counts of hatched juveniles were made at weekly intervals when cysts were rinsed in GDW and fresh test solution added. At the end of each test, the cysts were broken open and the number of viable, unhatched juveniles were counted in order to determine the percentage hatch. Batches of cysts in GDW, PRD and SBRD were routinely used as controls. Results for each experiment were subjected to

two way analysis of variance after arcsin transformation of percentages.

EXPERIMENT 1

Solutions of tri-allate (1, 2, 3 und 4 μgn^{-1}) in SL and (4 $\mu g\ ml^{-1}$) in PRD were used in hatching tests with *G. rostochiensis* (1 $\mu g\ ml^{-1}$ is equivalent to 1 ppm.). After 4 wk in the solutions, cysts from all treatment were rinsed in GDW and placed in PRD to determine hatch over a further 3 wk.

EXPERIMENT 2

Cysts of *G. rostochiensis* were set to hatch in a range of thiocarbamate herbicides (cycloate, pebulate, vernolate, chloridazon, metribuzin and tri-allate) each used as the a.i. diluted in GDW to give 5 and 50 $\mu g\ ml^{-1}$. After 4 wk, cysts were transferred to PRD for a further 3 wk.

EXPERIMENT 3

To test the combination of herbicide and aldicarb, cysts of *G. rostochiensis* were placed for 4 wk in solutions (5 and 50 $\mu g\ ml^{-1}$ a.i.) of the above herbicides, plus lenacil, with an additional series of treatments using a solution of each of the seven herbicides (5 $\mu g\ ml^{-1}$) mixed with aldicarb (10 $\mu g\ ml^{-1}$) and aldicarb (10 $\mu g\ ml^{-1}$) alone. After 4 wk, cysts previously exposed to herbicide/aldicarb solutions were transferred to PRD for a further 3 wk. An additional control was used for experiments 2 and 3, with cysts given a 4 wk presoak in GDW before transfer to PRD for 3 wk.

EXPERIMENT 4

Cysts of *G. rostochiensis* and *H. schachtii* were placed in solutions of the formulated compounds of the above seven herbicides used at field application rate (FAR), FAR $\times\ 10^{-2}$ and FAR $\times\ 10^{-3}$. The FAR, based on the manufacturers recommended medium rates, were : Ro-Neet® (a.i. : cycloate; Stauffer Chemical Co.), 3.6 kg a.i. ha^{-1} (equivalent to $15 \times 10^3\ \mu g\ ml^{-1}$); Tillam® (pebulate; Stauffer Chemical Co.), 5.0 kg a.i. ha^{-1} ($21 \times 10^3\ \mu g\ ml^{-1}$); Vernam® (vernolate; Stauffer Chemical Co.), 2.8 kg a.i. ha^{-1} ($12 \times 10^3 \mu g\ ml^{-1}$); Pyramin® (chloridazon; BASF UK Ltd), 2.5 l a.i. ha^{-1} ($10.4 \times 10^3\ \mu g\ ml^{-1}$); Sencorex® (metribuzin; Bayer UK Ltd), 0.6 kg a.i. ha^{-1} ($2.5 \times 10^3\ \mu g\ ml^{-1}$); Avadex BW® (tri-allate; Monsanto plc), 1.7 kg a.i. ha^{-1} ($7 \times 10^3\ \mu g\ ml^{-1}$); Venzar® (lenacil; Du Pont UK Ltd), 1.2 kg a.i. ha^{-1} ($5 \times 10^3\ \mu g\ ml^{-1}$). Cysts were kept in the test solutions for 4 wk before transfer to PRD or SBRD for a further 3 wk.

EXPERIMENT 5

Four of the above compounds were chosen for further study on the possible antagonistic effects of the formulated compound with root diffusates. Avadex BW, Pyramin, Sencorex and Vernam were made up to FAR as above) in root diffusate and in GDW. Cysts of *G. rostochiensis* and *H. schachtii* were kept in the herbicide solutions for 4 wk before transfer to PRD or SBRD for a further 5 wk.

Results

EXPERIMENT 1

The addition of tri-allate to SL did not increase hatch of *G. rostochiensis* over SL controls (Tab. 1). Subsequent hatch after transfer to PRD for 3 wk gave a similar hatch to cysts in PRD controls (80.5 %). At these concentrations, tri-allate appears to have no stimulatory or inhibitory effect on hatch. However, there is an indication that tri-allate may interfere with the action of PRD, for the hatch from cysts exposed to PRD plus 4 $\mu g\ ml^{-1}$ was significantly less ($P < 0.01$) than in PRD controls and remained significantly less ($P < 0.01$) after 3 wk in PRD alone.

Table 1

Total percentage hatch from cysts of *Globodera rostochiensis* after exposure to soil leachate (SL)/tri-allate (1, 2, 3 and 4 $\mu g\ ml^{-1}$) and potato root diffusate (PRD)/tri-allate (4 $\mu g\ ml^{-1}$) solutions for 4 wk and subsequently in PRD alone for a further 3 wk.

Solutions	*Time (wk)*	
	4	*7*
SL	2.3	77.4
SL + 1 $\mu g\ ml^{-1}$	0.7	77.3
SL + 2 $\mu g\ ml^{-1}$	2.7	79.2
SL + 3 $\mu g\ ml^{-1}$	2.3	80.4
SL + 4 $\mu g\ ml^{-1}$	3.8	81.7
PRD	79.8	80.5
PRD + 4 $\mu g\ ml^{-1}$	35.2	61.4

EXPERIMENT 2

Table 2 shows that exposure of cysts of *G. rostochiensis* to herbicide solutions for 4 wk did not elicit any significant hatch at either concentration; hatches were between 0.5-2.3 %, similar to the hatch in GDW controls (1.2 %). The herbicides did not have any marked effect on subsequent hatch, for transfer to PRD for 3 wk resulted in hatches from all treatments (40.4-56.4 %) commensurate with the hatch from control cysts (4 wk soak/3 wk PRD : 56.9 %). Hatching activity of this batch of PRD was somewhat less than previous batches, but does not detract from the conclusion that the a.i. of the herbicides had no effect on hatching.

Table 2

Total percentage hatch from cysts of *Globodera rostochiensis* after exposure to the active ingredient (5 and 50 µg ml^{-1}) of various herbicides for 4 wk and subsequently in potato root diffusate (PRD) alone for a further 3 wk. PRD and glass distilled water (GDW) as controls.

Solutions		*Time (wk)*	
		4	*7*
PRD		54.9	
GDW		1.2	
Cycloate	5 µg ml^{-1}	0.9	41.5
	50 µg ml^{-1}	0.9	45.9
Pebulate	5 µg ml^{-1}	1.1	42.5
	50 µg ml^{-1}	0.7	44.9
Vernolate	5 µg ml^{-1}	0.9	44.1
	50 µg ml^{-1}	0.8	47.4
Tri-allate	5 µg ml^{-1}	1.3	56.4
	50 µg ml^{-1}	0.5	48.3
Chloridazon	5 µg ml^{-1}	1.1	53.2
	50 µg ml^{-1}	0.6	47.2
Metribuzin	5 µg ml^{-1}	1.9	40.4
	50 µg ml^{-1}	2.3	45.1

Experiment 3

A marginally greater hatch of *G. rostochiensis* in GDW (5.2 %) than above was reflected in similar hatches from herbicide treated cysts (1.5 - 5.3 %) but, as with experiment 2, the herbicides did not enhance hatch or affect subsequent hatch in PRD (Tab. 3). Mixing aldicarb with the herbicides virtually eliminated any hatch (0.1 - 0.7 %) but did not affect the subsequent total hatch when cysts were transferred to PRD for 3 wk (Tab. 3). However, there was a difference in the rate of hatch between the two sets of treatments. Most juveniles hatched from cysts exposed to herbicides 1 wk after transfer to PRD, whereas hatch from cysts exposed to herbicide/aldicarb mixtures was delayed until 2 wk after transfer to PRD (Tab. 4).

Experiment 4

Using the formulated compounds of the herbicides at FAR gave a marked effect by some herbicides on hatch. No increase in hatch of *G. rostochiensis* or *H. schachtii* was observed but subsequent hatch after cysts were transferred to root diffusates was altered (Tab. 5).

The hatch from cysts of *G. rostochiensis* after transfer from FAR × 10^{-2} and FAR × 10^{-3} concentrations of herbicides to PRD were all similar (range : 23.5-39.7 %) and were less than the control hatch in PRD (47.6 %). However, the most marked effects followed exposure to

Table 3

Total percentage hatch from cyst of *Globodera rostochiensis* after exposure to the active ingredient (5 and 50 µg ml^{-1}) of various herbicides and to herbicide (5 µg ml^{-1})/aldicarb (10 µg ml^{-1}) solutions for 4 wk and subsequently in PRD alone for a further 3 wk. Potato root diffusate (PRD) and glass distilled water (GDW) as controls.

Solutions			*Time (wk)*	
			4	*7*
GDW then PRD			4.5	56.9
Aldicarb	10 µg ml^{-1}		0.0	60.3
GDW			5.2	
Cycloate	5 µg ml^{-1}		2.9	59.1
	50 µg ml^{-1}		3.1	55.0
	5 µg ml^{-1}	+ aldicarb 10 µg ml^{-1}	0.3	55.8
Pebulate	5 µg ml^{-1}		3.3	61.4
	50 µg ml^{-1}		4.1	51.2
	5 µg ml^{-1}	+ aldicarb 10 µg ml^{-1}	0.7	53.5
Vernolate	5 µg ml^{-1}		4.5	60.5
	50 µg ml^{-1}		2.7	51.4
	5 µg ml^{-1}	+ aldicarb 10 µg ml^{-1}	0.1	65.6
Tri-allate	5 µg ml^{-1}		5.0	62.4
	50 µg ml^{-1}		1.5	58.0
	5 µg ml^{-1}	+ aldicarb 10 µg ml^{-1}	0.1	53.3
Chloridazon	5 µg ml^{-1}		4.4	58.5
	50 µg ml^{-1}		3.4	57.8
	5 µg ml^{-1}	+ aldicarb 10 µg ml^{-1}	0.2	52.8
Metribuzin	5 µg ml^{-1}		5.1	54.3
	50 µg ml^{-1}		3.8	50.2
	5 µg ml^{-1}	+ aldicarb 10 µg ml^{-1}	0.2	49.5
Lenacil	5 µg ml^{-1}		2.8	47.6
	50 µg ml^{-1}		3.1	56.3
	5 µg ml^{-1}	+ aldicarb 10 µg ml^{-1}	0.1	46.7

herbicides at FAR. After 3 wk in PRD, there was virtually no hatch from cysts previously exposed to cycloate (total hatch : 1.8 %), pebulate (0 %), vernolate (2.6 %) and tri-allate (0.2 %); these four herbicides irreversibly inhibited hatch of *G. rostochiensis*. The hatch from cysts previously exposed to lenacil (27.3 %) was significantly ($P < 0.05$) reduced compared to controls.

The results (Tab. 5) were similar with *H. schachtii*. The reduction in hatch after 4 wk in SBRD from cysts previously exposed to FAR × 10^{-2} and FAR × 10^{-3} was only marginal; all gave over 50 % hatch. The most marked effects resulted from previous exposure to FAR cycloate (total hatch after 3 wk in SBRD : 1.3 %), pebulate (5.2 %), vernolate (6.1 %) and tri-allate (8.3 %) where hatches were significantly ($P < 0.001$) lower than in SBRD control (89.9 %). Hatches in SBRD from cysts previously exposed to FAR metribuzin (46.9 %) and lenacil (61.3 %) were significantly reduced ($P < 0.001$).

Thus, metribuzin and lenacil had only slight effects on subsequent hatch of *G. rostochiensis* and *H. schachtii* whilst prior exposure to FAR concentrations of the thiocarbamate herbicides (cycloate, pebulate, vernolate and tri-allate) abolishes subsequent hatch in root diffusates.

Table 4

Total cumulative percentage hatch from cysts of *Globodera rostochiensis* after 4 wk in herbicide (5 μg ml^{-1}) and herbicide (5 μg ml^{-1})/aldicarb (10 μg ml^{-1}) solutions and then after transfer to potato root diffusate for a further 3 wk.

Solutions			*Time (wk)*			
			4	*5*	*6*	*7*
Cycloate	5 μg ml^{-1}		2.9	57.1	59.0	59.1
Cycloate	5 μg ml^{-1}	+ aldicarb 10 μg ml^{-1}	0.3	16.3	55.6	55.8
Pebulate	5 μg ml^{-1}		3.3	58.1	61.3	61.4
Pebulate	5 μg ml^{-1}	+ aldicarb 10 μg ml^{-1}	0.7	5.2	52.5	53.5
Vernolate	5 μg ml^{-1}		4.5	58.2	60.2	60.3
Vernolate	5 μg ml^{-1}	+ aldicarb 10 μg ml^{-1}	0.1	6.1	64.8	65.6
Chloridazon	5 μg ml^{-1}		4.4	55.8	58.3	58.5
Chloridazon	5 μg ml^{-1}	+ aldicarb 10 μg ml^{-1}	0.2	6.5	51.7	52.8
Metribuzin	5 μg ml^{-1}		5.1	52.0	54.3	54.3
Metribuzin	5 μg ml^{-1}	+ aldicarb 10 μg ml^{-1}	0.2	2.6	48.9	49.5
Tri-allate	5 μg ml^{-1}		5.0	58.5	62.3	62.4
Tri-allate	5 μg ml^{-1}	+ aldicarb 10 μg ml^{-1}	0.1	3.0	52.4	53.3
Lenacil	5 μg ml^{-1}		2.8	43.0	47.4	47.6
Lenacil	5 μg ml^{-1}	+ aldicarb 10 μg ml^{-1}	0.1	1.2	45.7	46.7

EXPERIMENT 5

For this test, the formulated compounds of four herbicides at FAR were selected : chloridazon and metribuzin having only slight effects on hatch, and vernolate and tri-allate preventing hatch. After treatment for 4 wk, cysts were left in PRD or SBRD for 5 wk to ensure that any delay in hatching could be assessed.

Exposing cysts of *G. rostochiensis* to mixtures of the herbicides and PRD completely stopped hatch in all cases; subsequent transfer of the cysts to PRD resulted in hatches of 1.2 % and 0.4 % from cysts previously treated with tri-allate/PRD and vernolate/PRD respectively, and 40.0 % and 40.9 % from cysts with prior exposure to chloridazon/PRD and metribuzin/PRD respectively (Tab. 6). These hatches were all significantly less than the hatch from control cysts in PRD for 9 wk (80.5 %).

Similar results were obtained with *H. schachtii* (Tab. 6). The herbicides interferred with the action of SBRD : virtually no hatch occurred in any treatments and transfer to SBRD resulted in hatches over 5 wk of less than 15 % in each case; all were significantly ($P < 0.001$) lower than the hatch from control cysts in SBRD (51.7 %).

Hatches from cysts of both species exposed to herbicides only before transfer to PRD or SBRD (Tab. 6) confirmed results obtained in experiment 4 (Tab. 5), with the exception of metribuzin where the inhibition of hatch from *G. rostochiensis* cysts is more marked (9.6 %) than in the previous test (30.6 %; Tab. 5).

Discussion

The herbicides used in this work cannot be regarded as hatching agents for *G. rostochiensis* and *H. schachtii*. The putative action of herbicides in affecting the eggshell lipid layer and altering eggshell permeability may not cause hatch; herbicides may fail to mimic the bimodal action of hatching agents (Perry, 1987) where metabolic stimulation of the unhatched juvenile is also important. *H. schachtii* was cultured on cabbage, which results in cysts containing eggs with distinct inner lipoprotein membranes and hatching characteristics similar to *G. rostochiensis* with less than 10 % hatch in water and a need for RD to stimulate hatch; the absence of fungal contamination as a factor in the hatching response has been discussed (Perry & Trett, 1986). Populations of *H. schachtii* from sugar beet may have given different results, as such cysts commonly have a large water hatch and frequently contain fungus surrounding the eggs; exposure to herbicides may remove this fungus resulting in enhanced hatch. Saly and Stanova (1976) and Kraus

Table 5

Total percentage hatch from cysts of *Globodera rostochiensis* and *Heterodera schachtii* after exposure to herbicides at medium field application rate (FAR), FAR × 10^{-2} and FAR × 10^{-3} for 4 wk and subsequently in root diffusate (RD) alone for 3 wk. RD and glass distilled water (GDW) as controls.

Solutions		G. rostochiensis		H. schachtii	
		Time (wk)			
		4	*7*	*4*	*7*
RD		47.6		89.9	
GDW		3.0		6.5	
Cycloate	FAR	0.5	1.8	0.0	1.3
	FAR × 10^{-2}	4.3	29.4	1.7	65.5
	FAR × 10^{-3}	5.0	39.7	4.8	78.9
Pebulate	FAR	0.0	0.0	0.0	5.2
	FAR × 10^{-2}	4.4	25.4	2.3	64.7
	FAR × 10^{-3}	5.0	25.5	5.2	71.3
Vernolate	FAR	1.2	2.6	2.4	6.1
	FAR × 10^{-2}	2.7	27.0	1.6	75.6
	FAR × 10^{-3}	6.9	32.0	6.3	63.1
Tri-allate	FAR	0.1	0.2	0.1	8.3
	FAR × 10^{-2}	3.9	23.5	2.9	79.8
	FAR × 10^{-3}	3.6	28.3	1.3	89.4
Chloridazon	FAR	1.6	44.2	0.0	86.2
	FAR × 10^{-2}	2.0	24.0	3.0	68.8
	FAR × 10^{-3}	2.8	23.5	5.8	77.7
Metribuzin	FAR	2.5	30.6	0.1	46.9
	FAR × 10^{-2}	3.8	27.0	2.9	52.6
	FAR × 10^{-3}	5.7	32.2	4.1	77.9
Lenacil	FAR	1.6	27.3	5.6	61.3
	FAR × 10^{-2}	4.6	30.8	4.2	73.4
	FAR × 10^{-3}	4.0	30.8	7.2	74.6

Table 6

Total percentage hatch of *Globodera rostochiensis* and *Heterodera schachtii* at 9 wk after exposure for 4 wk to herbicides at medium field application rate alone or mixed with root diffusate (RD), followed by 5 wk in RD

Solutions	G. rostochiensis	H. schachtii
RD	80.5	51.7
Vernolate	1.6	5.1
Vernolate + RD	0.4	8.6
Tri-allate	4.2	7.2
Tri-allate + RD	1.2	9.8
Chloridazon	47.8	49.9
Chloridazon + RD	40.0	12.3
Metribuzin	9.6	13.3
Metribuzin + RD	40.9	12.1

and Sikora (1979) noted that herbicides increased hatch from cysts of *H. schachtii* from sugar beet and Altman and Steele (1982) found increased hatch in 1.25, 2.5 and 5 µg ml^{-1} cycloate. Di-allate at concentrations of 2.5 to 80 µg ml^{-1} stimulated hatch from *H. schachtii* cysts from sugar beet and enhanced invasion (Kraus, 1981; Kraus & Sikora, 1981); by contrast, we found that tri-allate at concentrations up to 50 µg ml^{-1}, did not cause hatch from cysts of *G. rostochiensis.*

Although there are indications that some herbicides increase gall production of *Meloidogyne arenaria* (King, Rodriguez-Kabana & Ingram, 1977) and cyst numbers of *H. schachtii* (Abivardi & Altman, 1978; Kraus & Sikora, 1983) and *H. glycines* (Kraus, Noel & Edwards, 1982) it may be too simplistic to attribute this solely to hatch effects, especially as these species usually have a large water hatch. Other factors may be involved : metribuzin, for example, stimulates movement of hatched juveniles of *G. rostochiensis* and enhances invasion

(Perry, Feil & Beane). Herbicides may also render the plant roots more (unpubl. res.) susceptible to nematode invasion. Exposing cysts to aldicarb/herbicide mixtures only delayed hatch on transfer to PRD. This is unlikely to be due to an effect by the herbicides but probably reflects the time taken for juveniles to recover from the inhibition of acetycholinesterase caused by exposure to aldicarb; certain concentrations of aldicarb reversibly inhibit hatch (Cooke, 1987).

The hatching response of cysts of *G. rostochiensis* and *H. schachtii* to the formulated herbicide compounds confirmed and extended results from experiments with the active ingredients. All herbicides tested at FAR and 10^{-2} and 10^{-3} dilutions elicited no hatch from either species. However, the significant finding of this work relates to the inhibition of hatch. Exposure to FAR concentrations of cycloate, pebulate, vernolate and tri-allate eliminates subsequent hatch of both species in RD. Tests with New Blue R (Shepherd, 1962) showed that tri-allate at FAR killed 90 % of cyst contents.

It is clear from the present work that herbicides cannot be regarded as having a common mode of action relating to hatching. Three of the herbicides tested had slight or no effects on hatch, whilst the four thiocarbamate herbicides at FAR (cycloate, pebulate, vernolate and tri-allate) prevented subsequent hatch in RD; the thiocarbamate group seems to be the important factor in the inhibition. There appears to be no link between the method of formulating the compound and hatch effects : all the thiocarbamates, chloridazon and lenacil were formulated as emulsifiable concentrates, with only metribuzin being formulated as a wettable powder.

The effect of thiocarbamate herbicides on hatch was reduced markedly by dilution so any control strategy based on their use, probably as a pre-emergence treatment, would have to overcome the problem of soil incorporation to ensure that the required concentrations contact the cysts. This aspect and the mode of action are currently being investigated. The interactive effect of herbicides and RD, where hatch is prevented, is also worth further study. Although it may not be feasible to translate this effect into field use as a post-emergence treatment, the chemical basis for the inactivation of RD could be important.

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