HGCA

PROJECT REPORT No. 80

MONITORING SENSITIVITY TO THE DMI GROUP OF FUNGICIDES IN NET BLOTCH (*Pyrenophora teres*) OF WINTER BARLEY

AUGUST 1993

PRICE £3.00

HGCA PROJECT REPORT No. 80

MONITORING SENSITIVITY TO THE DMI GROUP OF FUNGICIDES IN NET BLOTCH (*Pyrenophora teres*) OF WINTER BARLEY

by

M.J. HIMS
MAFF Central Science Laboratory, Hatching Green,
Harpenden, Hertfordshire AL5 2BD

Final report of a three year project at the Central Science Laboratory. The work commenced in August 1988 and was funded by a grant of £25,069 from the Home-Grown Cereals Authority (HGCA Project No. 0096/3/87).

Abstract

During the four years 1988-91 net blotch, caused by the fungus *Pyrenophora teres*, was the most damaging of the splash-borne diseases recorded in the CSL/ADAS National Surveys of Winter Barley Diseases in England and Wales. Despite the fact that many crops were treated with one or more fungicides, estimated losses of up to £7m are likely to have occurred.

Chemicals in the 'DMI' group (eg cyproconazole, flusilazole, flutriafol, propiconazole, prochloraz & triadimenol), either singly or as components of formulated mixtures, dominate fungicide use on winter barley but particularly so after flag leaf emergence; during the period 1987-91 up to 82% of crops surveyed received an application of at least one 'DMI' fungicide. More than 50% received an application after flag leaf tip emergence (GS37), almost invariably propiconazole. Most of the fungicides offering effective control of net blotch belong to the 'DMI' group and base-line data on current sensitivity is required in order to detect rapidly changes which may lead to resistance problems.

Leaves showing symptoms of net blotch from the 1988, 1989 and 1991 CSL/ADAS National Survey of Winter Barley Diseases were used to establish isolates of the fungus from all the cereal growing areas of England and Wales. In all, 264 samples provided 1485 isolates which were tested for *in-vitro* sensitivity to the DMI fungicides prochloraz and propiconazole. Concentrations of 0.1 and 1.0 µg ai (0.5 and 5.0 µg ai in 1989) prochloraz and 0.5 and 5.0 µg ai (0.1 and 1.0 µg ai in 1989) propiconazole per ml agar were used. Sensitivity varied greatly between samples and isolates, but showed no reduction in 1988 and 1989 below that found in preliminary studies at Long Ashton Research Station on isolates made in 1979 and 1980.

However, there appears to have been a slight reduction in sensitivity of the population obtained during 1991 compared to that obtained during 1988. In general, reductions in sensitivity were associated with previous use of a DMI during the growing season, and sensitivity to both DMIs was well correlated in all three years (0.46, 0.39 & 0.81 1988-91 respectively $P < 0.0001$). The significance of this apparent reduction needs to be assessed on the basis of *in-vivo* and field crop sensitivity tests but it does nor currently appear to cause disease control problems in the field. The results of this work indicate that it is important to continue to monitor for possible further shifts towards insensitivity to DMI fungicides in populations of *Pyrenophora teres* at appropriate intervals.

Objectives

To monitor populations of *Pyrenophora teres* for sensitivity to DMI fungicides. Samples were collected from winter barley crops for a 3-year period (1988, 1989, 1990) as part of the CSL/ADAS National Surveys of Winter Barley Diseases. In reality the 1990 testing was postponed until 1991 because of a low incidence of net blotch in winter barley during the winter and early spring period of 1989-90.

Introduction

Net blotch, caused by the fungus *Pyrenophora teres* (Drechs.), is a relatively common disease of barley throughout the temperate regions of the world (Shipton *et al.*, 1973) and is particularly prevalent in areas of high rainfall and humidity. In the UK, the disease showed a dramatic increase in winter barley in the late 1970's, probably favoured by a number of changes in agronomic practice, including the widespread cultivation of the highly susceptible cultivar Sonja (Table 1). The epidemic of 1979 was particularly severe (Jordan, 1981) and surveys in the early 1980's showed net blotch to be the most, second most and fourth most damaging disease in 1981, 1982 and 1983 respectively (Table 2). Following a lull during the drier summers of the mid-1980's, net blotch was again the most severe foliar disease in 1987, and was more severe in 1988 than 1986, despite the widespread introduction of cultivars with high levels of field resistance (Table 1).

Fungicides are widely used for disease control in winter barley. During the period 1988-91, 88-94% of all crops surveyed by CSL and ADAS (Polley, 1988-1991) received at least one fungicidal spray, almost invariably a DMI fungicide. In addition over 50% were sprayed after flag leaf tip emergence (GS37), specifically for foliar disease control. More than half of these crops received a spray of propiconazole or another fungicide in the DMI group ('azoles' eg. cyproconazole, flusilazole, flutriafol, propiconazole, prochloraz & triadimenol), some members of which are regarded as giving the only effective control of net blotch.

However, widespread use of a few chemicals with a similar mode of action, with little effective control afforded by fungicides with differing modes of action, may lead to the development of insensitivity and cross-resistance within all members of the azole group. In New Zealand, resistance to triadimenol seed dressings has become widespread since it was first detected in 1981, and cross-resistance to nuarimol is also common (Sheridan *et al.*, 1987). In Sweden, variations in sensitivity to DMI's between sub-populations of *P. teres* have already been found that are sufficiently large to be classed as resistance according to the definition of Fuchs *et al.* (1984) (Olvang, 1988). In Yugoslavia triadimefon failed to inhibit the growth of *P. teres* in *in-vitro* tests whereas propiconazole, prochloraz and tebuconazole gave high levels of growth inhibition (Milevoj & Groznik, 1989).

Sensitivity to a wide range of antifungal compounds was investigated at Long Ashton Research Station (LARS) in the early 1980's but no loss in sensitivity was found in isolates from plots which had received sprays of propiconazole through the season (Jordan *et al.*, 1981, Kendall *et al.*, 1982). Though failure to control the disease in the field has yet to be established, base-line data on sensitivity to fungicides in current populations of *P. teres* is required in order to detect any future drift towards insensitivity. Should this occur, alternative control strategies may then be developed before the loss of control presents serious problems.

The annual National Surveys of Winter Barley Diseases, carried out by CSL and ADAS, afforded an ideal opportunity to monitor the pathogen as over 300 crops from throughout England and Wales are assessed for all foliar diseases at the watery ripe stage (GS71). Sensitivity to the two most commonly used azoles for net blotch control, propiconazole and prochloraz, was examined in isolates established from crops grown in the 1988, 1989 and 1991 harvest years.

Materials and methods

Sampling

Samples sent in mid-June to early July as part of the CSL/ADAS National Survey of Winter Barley Diseases were assessed for net blotch. Flag and second leaves showing clear symptoms were wrapped individually in dry filter paper and stored overnight in a sealed plastic vessel containing silica gel. After drying, samples were labelled and stored at -18°C until required for testing. Up to 10 leaves per sample were preserved.

Isolation

As required, leaves were removed from storage and placed into Petri-dish damp chambers then incubated at 17°C under continuous near ultra-violet light for 24-48 hrs. Spores produced within areas of netting were carefully picked off using a sterile scalpel blade, and transferred, 3-5 spores each, to plates containing potato dextrose agar (PDA - Oxoid). Germination and growth was monitored daily over the next 2-3 days to ensure no contaminants were present, then the colony was allowed to grow under near ultra-violet light at 17°C until required.

Sensitivity testing

Prior to testing samples from the 1988 survey twenty isolates were tested against a range of fungicide concentrations in PDA (0.05 µg ai/ml to 50 µg ai/ml) to determine the two optimum concentrations for sensitivity monitoring (Figures 1 & 2). Thereafter, all isolates were tested against two concentrations only for each fungicide as follows: propiconazole 0.5 & 5 µg ai/ml, prochloraz 0.1 & 1 µg/ml. In 1989 the concentrations of the two fungicides were inadvertently reversed so that prochoraz was tested at 0.5 & 5.0 µg/ml and propiconazole was tested at 0.1 & 1.0 µg/ml. Pairs of isolates were tested concurrently against each fungicide by transferring 5mm plugs of agar from the colony edge of the original isolation plates to Petri dishes containing PDA amended with fungicide to the appropriate concentration. Each pair was also plated onto unamended PDA as a control. All plates were incubated at 18°C and measured after 7 and 14 days. Growth was expressed as a percentage of the control growth for each isolate.

Results

Figures 1 and 2 show the average dosage-response curves to prochloraz and propiconazole found in the preliminary sensitivity testing. The variation in response between isolates at each concentration is illustrated by frequency curves of the inhibition. Clearly, the range of variability

in response is greater at lower concentrations. Additionally, isolates varied more in their response to propiconazole than to prochloraz. The EC_{50} value for propiconazole appears to be around 0.087µg/ml (Figure 2) while that for prochloraz, by extrapolation of Figure 1, may be around 0.0032µg/ml. Complete inhibition of all isolates occurred at 10 µg/ml for both fungicides giving a minimum inhibitory concentration (MIC) somewhere between 5 and 10 µg ml^{-1}.

Figures 3 and 4 show the average dosage-response curves to prochloraz and propiconazole at the original concentrations (1988) and the reversed concentrations (1989) found in the populations tested during 1988 and 1989. The variation in response between isolates at each concentration is illustrated by frequency curves of the inhibition. Clearly, the range of variability in response is greater at lower concentrations. Additionally, isolates varied more in their response to propiconazole than to prochloraz.

A comparison of the 1988 and 1991 data indicates that there may have been a slight reduction in the sensitivity of the population sampled in 1991 compared to that sampled in 1998. This is particularly evident at the low concentrations (Figure 5) of the two fungicides at which a small number of isolates exhibited no growth inhibition compared to the unamended controls. At the high concentration more isolates exhibited less than 50% inhibition of growth in 1991 compared to 1988 (Figure 6). See also Figures 7 and 8.

During the three years of testing *P. teres* was successfully established from: 110 samples giving a total of 743 isolates (1988), 69 samples giving a total of 290 isolates (1989) and 85 samples giving a total of 452 isolates in 1991. For the three years, the growth rate in the control plates varied considerably between isolates with a mean: for 1988 of 50.9 mm (range 13-85 mm), for 1989 of 56.8 mm (range 23-74 mm) and for 1991 of 74.5 mm (range 26-80 mm) after 7 days. Mean growth, as a percentage of control growth within each population, for each chemical is given in Tables 3.1 and 3.2. There was a significant correlation within isolates (r = 0.46('88), 0.36('89), 0.81('91) $P < 0.0001$) between the degree of growth in the presence of propiconazole and that found in the presence of prochloraz.

Isolates varied in sensitivity regionally; in 1988 the lowest mean percentage growth rates for both chemicals was found in the south-west (Bristol) and the highest in the south-east (Wye). Isolates from the south-east (Wye) behaved similarly in 1991 although those from the south-west were conversely amongst isolates with the highest mean percentage growth rates (Table 4.1). For 1989 isolates from the south-east (Wye) gave the lowest percentage growth rates for the high rate of propiconazole which was only double that of the low rate of propiconazole used in 1988. Results for the low concentration of prochloraz and the high concentration of propiconazole (more comparable to the low concentrations of both fungicides used in 1988) are given in Table 4.2.

During 1988 only four isolates were found to grow in excess of 40% of the unamended control rate at 5.0 µg/ml propiconazole, each from different fields. For the higher rate of prochloraz (1.0 µg/ml), three isolates from two different fields grew in excess of 40% of control rates. The fungicides applied to these crops are listed in Table 5.1.

During 1989 nine isolates were found to grow in excess of 60% of the unamended control rate at 1.0 µg/ml propiconazole, three from the same field, the remainder each from different fields. For the higher rate of prochloraz (5.0 µg/ml), no isolates grew in excess of 60% of control rates. The fungicides applied to these crops are listed in Table 5.2.

During 1991 seven isolates were found to grow in excess of 40% of the unamended control rate at one or other of the high concentrations of the two test fungicides. The fungicides applied to these crops are listed in Table 5.3.

Tables 4.1 and 4.3 indicate an apparent association between the extent of growth on fungicide amended agar and current season use of DMI fungicides. This relationship is even more readily apparent from Table 6 which distinguishes between those isolates (and their associated current season, field-applied fungicides) that grew the most and those that grew least on the high rates of the two test fungicides in agar. The national means are particularly striking with greater differences between 1988 and 1991 for the isolates that grew the most.

Discussion

Despite the reversal in the concentrations of prochloraz and propiconazole used in 1989 compared to those used in 1988 the dosage-response curves in Figures 3 and 4 indicate that the data generated by the two years work is perfectly acceptable in terms of detecting shifts within populations of *P. teres* for insensitivity to DMI fungicides. The two graphs indicate that there was no significant change in the mean sensitivity of *P. teres* populations to DMI fungicides nor was there a significant change in the distribution of sensitive and less sensitive isolates about the mean from 1988 to 1989. However for a direct comparison only the data from 1988 and 1991 may be used. Figures 5 and 6 illustrate the growth distribution curves for 1988 and 1991 for both fungicides. There is a clear shift towards reduced sensitivity in the population surveyed in 1991 compared to that in 1988. Not only is there a greater 'spread' in the growth distribution curve (Figures 5-8) but the mean percentage growth for the population has increased by 1.43-5.0% at the high concentrations and by 4.97-13.73% at the low concentrations (Table 4.1). For individual regions these differences were considerably greater but also smaller in some instances (Table 4.1).

In the regional context it was very useful to depict the apparent 'reducing sensitivity' in populations of *P. teres* on maps of England and Wales. Figures 9 and 10 compare regional data from 1988 and 1991 for the sensitivity of isolates as measured by the percentage of isolates that grew to a degree greater than 25% of their respective controls. There appears to be some fairly

obvious differences between the two years as well as between regions for both propiconazole and prochloraz. Clearly, for the future, it may be sensible to concentrate on those areas where the greatest apparent shift has occurred to date.

The overall results of this project indicate that isolates of *P. teres* vary in their *in-vitro* sensitivity to the two fungicides tested, most noticeably at low concentrations, and particularly when considering propiconazole. However, the bimodal distribution of sensitivity found in Sweden by Olvang (1988) was not apparent here. Olvang found that isolates could be classified into two groups with EC_{50} values for the more sensitive group at least one sixth that of the less sensitive isolates. This survey has found no evidence that resistance to propiconazole or prochloraz, as defined by Fuchs *et al.* (1984), exists within the population at present in England and Wales. Furthermore, mean ED_{50} values estimated in this study are lower than those established by workers at LARS on a limited number of isolates made in 1979 and 1980 (Kendall *et al.*, 1982). Nonetheless, although some isolates were completely inhibited by 5.0 µg/ml propiconazole or 1.0 µg/ml prochloraz, the majority of isolates were capable of growth around 10-20% of that found on unamended agar. Additionally, some isolates were capable of growth in excess of 40%(1988) and far more were so in 1991. Moreover, some isolates were capable of growth in excess of 60% of control growth in 1991. While most of these isolates were from fields where DMI's had been used at least once on the crop, it is interesting to note that one isolate in each year was from a field where only Missile had been used, a fungicide completely unrelated to the DMI group. Thus, variation in sensitivity between isolates exists in the field in the absence of selection pressure from the use of DMI's.

The three isolates showing the least sensitivity to prochloraz in 1988 were from fields where the only DMI used had been flutriafol. Similarly, in all three years not all isolates with reduced sensitivity to propiconazole were from fields that had received propiconazole alone or in mixtures with other non-DMI fungicides. This, coupled with the significant correlation between the degree of insensitivity within isolates to propiconazole and prochloraz, suggests that any resistance developing in the future may be across many members of the DMI group.

Along with several other groups of fungicides, the DMI's interfere with sterol synthesis and affect fungi by altering cell membrane functions. All members of the DMI group, however, act specifically by preventing demethylation at the C-14 position, and therefore use a common pathway. For this reason it is not surprising that cross-resistance is likely and has already been reported in New Zealand in *P. teres* for nuarimol and triadimenol (Sheridan *et al.*, 1987). The DMI's, and more particularly propiconazole and prochloraz, are used almost exclusively for net blotch control and widely within winter barley for disease control generally. It is likely that such continued intensive use places strong selection pressure on the pathogen for resistance. A decline in the level of control of wheat mildew in the Netherlands has been attributed to decreased

sensitivity to DMI's in the pathogen which correlates with an increase in their use (De Waard *et al.*, 1986).

Finally, there is the question of how this project should be pursued in the future. Clearly the apparent slight shift towards insensitivity detected in 1991 appears unlikely to lead to field disease control problems in the immediate future. However, because this apparent shift may be the first indication of an incipient resistance problem, monitoring for further shifts should continue, perhaps on an *ad-hoc* basis, associated with the CSL/ADAS National Winter Barley Disease Survey of England and Wales or the National UK Cereal Pathogen Virulence Survey. Surveys of England and Wales should also take diseased leaf samples from Scotland and Northern Ireland. This type of monitoring need not be expensive especially if it were to concentrate, at least in the first instance, on areas of greater potential for fungicide insensitivity eg. south-east and south-west England plus any apparent disease control failures in field crops. Such a concentration of effort would allow for field testing on 'cereal tussocks' similar to that achieved at CSL for *Septoria tritici* in 1992. It is even more important to maintain a monitoring programme whilst some relatively net blotch susceptible varieties continue to be introduced into the UK and may gain a major share of the market (Table 1).

References

Fuchs, A., Vries, F.W. de & Waard, M.A. de (1984) Simultaneous resistance in fungi to ergosterol biosynthesis inhibitors and dicarboximides. *Netherlands Journal of Plant Pathology* **90,** 3-11.

Jordan, V.W.L. (1981) Aetiology of barley net blotch caused by *Pyrenophora teres* and some effects on yield. *Plant Pathology* **30,** 77-87.

Jordan, V.W.L., Best, G.R. & Allen, E. (1981) Fungicide tests in the glasshouse. *Report Long Ashton Research Station* **1981,** 88.

Kendall, S.J., Carter, G.A. & Court, L. (1982) Sensitivity of *Pyrenophora teres* to ergosterol biosynthesis inhibitors. *Report Long Ashton Research Station* **1982,** 91-92.

Milejvoj, L. & Groznik, K. (1989) Influence of some fungicides on *Pyrenophora graminea* (Died.) Ito et Kurib. and *Pyrenophora teres* (Died.) Drechsler. *Zastita Bilja* **40,** 201-209.

Olvang, H. (1988) Sensitivity of *Drechslera teres* and *Septoria nodorum* to sterol-biosynthesis inhibitors. *Netherlands Journal of Plant Pathology* **94,** 57-68.

Polley, R. W. (1988-1991) Winter Barley Survey Reports. *Ministry of Agriculture, Fisheries & Food, Harpenden Laboratory.*

Sheridan, J.E., Grbavac, N., Sheridan, Mary H. & Soteros, J.J. (1987) Further studies on triadimenol resistance in the barley net blotch pathogen *Pyrenophora teres*. *New Zealand Journal of Agricultural Research* **30,** 101-105.

Shipton, W.A., Khan, T.N. & Boyd, W.J.R. (1973) Net blotch of barley. *Review of Plant Pathology* **52,** 270-290.

Waard, M.A. de, Kipp, E.M.C., Horn, M.N. & Nistelrooy, J.G.M. van (1986) Variation in sensitivity to fungicides which inhibit ergosterol biosynthesis in wheat powdery mildew. *Netherlands Journal of Plant Pathology* **92,** 21-23.

Table 1. Mean severity of net blotch (second leaf) at GS71-73 on the range of varieties of winter barley encountered in the survey during 1988-91 (number of samples in each year).

Variety	Survey Year				NIAB net blotch
	1988	1989	1990	1991	resistance rating#
Blanche	0.00(0)	0.00(0)	0.00(0)	1.76(1)	?
Concert	3.88(5)	2.04(1)	0.00(1)	0.00(0)	3
Finesse	0.00(0)	0.00(1)	0.00(4)	0.00(1)	7
Frolic	0.00(0)	0.00(1)	0.01(15)	0.18(28)	8
Gaulois	0.00(0)	0.00(2)	4.00(2)	10.29(6)	6
Gerbel	0.72(1)	0.00(0)	0.00(0)	0.00(0)	6
Gypsy	0.00(0)	0.00(0)	0.00(0)	0.39(3)	8
Halcyon	0.27(13)	0.01(12)	0.06(21)	0.05(17)	8
Igri*	**3.91(133)**	**0.94(82)**	**0.35(45)**	**3.23(29)**	**5**
Kira	0.00(0)	0.00(2)	0.04(15)	0.06(25)	8
Magie	0.22(37)	0.08(62)	0.08(43)	0.47(35)	8
Manitou	0.00(0)	0.00(0)	0.00(0)	7.68(3)	7
Marinka	0.19(28)	0.03(62)	0.00(124)	0.36(113)	9
Maris Otter*	**0.03(11)**	**0.15(13)**	**0.00(9)**	**0.04(5)**	**7**
Melusine	0.00(0)	0.00(1)	0.00(3)	0.00(0)	7
Mimosa	0.00(0)	0.00(0)	0.00(0)	0.00(1)	9
Nevada	0.00(3)	0.00(1)	0.16(1)	0.00(0)	7
Opera	0.00(0)	0.00(1)	0.00(0)	0.00(0)	7
Panda	2.85(73)	1.96(35)	0.01(10)	2.85(8)	6
Pastoral	0.00(0)	0.00(0)	0.00(3)	0.80(25)	6
Pipkin	0.05(14)	0.02(10)	0.00(24)	0.18(33)	8
Pirate	11.20(1)	0.44(1)	0.00(0)	0.00(0)	7
Plaisant	2.83(18)	1.13(29)	0.35(22)	5.17(9)	?
Posaune	0.00(0)	0.00(0)	0.00(0)	0.76(1)	5
Puffin	0.00(0)	0.00(0)	0.00(0)	1.35(33)	4
Sonja*	**0.04(2)**	**0.97(3)**	**0.06(2)**	**0.04(1)**	**3**
Tipper	2.00(1)	1.12(1)	0.00(1)	0.00(0)	7
Torrent	1.43(10)	0.21(24)	0.17(27)	0.83(16)	7
Vixen	0.00(0)	0.00(1)	0.00(0)	0.00(0)	9

*In 1981 these three varieties constituted 96% of the winter barley area surveyed compared to 38% in 1988.

#A high figure indicates that the variety shows the character to a high degree (rating is the lowest published on the recommended list from 1981 onwards).

Table 2. *Incidence of net blotch in samples of winter barley in ADAS Surveys and mean severity of infection on the flag and second leaves at GS71-73.*

Year	Incidence in samples (%)	Severity on top two leaves	
		Flag	Second
1981	90	6.7	14.2
1982	60	0.2	0.8
1983	36	0.4	1.1
1986	63	0.5	1.9
1987	82	1.1	3.7
1988	59	1.1	2.2
1989	43	0.3	0.6
1990	20	<0.1	0.1
1991	36	0.4	1.1

Table 3.1. Growth of 1988 & 1991 isolates of P. teres *in the presence of differing concentrations of prochloraz and propiconazole (growth is expressed as a percentage of control growth for each isolate).*

Chemical	Concentration (µg/ml)	% growth (after 7 and 14 days)	
		7 (range)	14
1988			
Propiconazole	0.5	31.14 (0-95)	35.15
Propiconazole	5.0	6.44 (0-47)	12.27
Prochloraz	0.1	30.38 (0-81)	34.38
Prochloraz	1.0	13.01 (0-54)	15.93
1991			
Propiconazole	0.5	38.03 (9-156)	44.88
Propiconazole	5.0	14.37 (0-76)	17.27
Prochloraz	0.1	34.05 (7-123)	39.35
Prochloraz	1.0	15.43 (0-62)	17.36

Table 3.2. Growth of 1989 isolates of P. teres *in the presence of differing concentrations of prochloraz and propiconazole (growth is expressed as a percentage of control growth for each isolate).*

Chemical	Concentration (µg ml/1)	% growth (after 7 and 14 days) 7	14
Propiconazole	0.1	46.35	58.86
Propiconazole	1.0	22.53	28.89
Prochloraz	0.5	19.89	21.38
Prochloraz	5.0	12.89	13.17

Table 4.1. 1991 compared to (1988) regional variation in growth of isolates of P.teres *in the presence of low and high concentrations of prochloraz and propiconazole after 14 days (growth is expressed as a percentage of the control growth for each isolate).*

Region	Chemical			
	Prochloraz		Propiconazole	
	1.0 µg/ml	0.1 µg/ml	5.0 µg/ml	0.5 µg/ml
Wales	15.04 (13.57)	25.31 (30.53)	7.78(11.02)	28.80(34.20)
North (Newcastle)	11.37 (13.43)	29.88 (36.50)	5.86(8.69)	34.58(30.76)
North (Leeds)	14.85 (16.68)	38.67 (35.26)	21.12(13.84)	39.76(36.39)
Midlands & Western	15.73 (14.36)	32.51 (32.25)	15.04(12.95)	40.01(30.98)
South-west (Bristol)	19.04 (12.77)	37.48 (27.25)	18.08(10.32)	42.10(30.12)
South-west (Starcross)	18.11 (18.09)	38.44 (38.08)	15.51(12.99)	39.67(37.84)
East	18.14 (15.39)	42.45 (34.26)	19.31(11.51)	48.24(34.56)
South-east (Reading)	16.90 (17.83)	42.49 (36.54)	16.80(12.41)	48.25(36.84)
South-east (Wye)	17.32 (18.88)	45.54 (39.37)	19.94(14.69)	55.55(40.70)
Average (all regions)	17.36 (15.93)	39.35 (34.38)	17.27(12.27)	44.88(35.15)

Table 4.2. Regional variation in growth of isolates of P. teres *in the presence of low concentrations of prochloraz and propiconazole after 14 days (growth is expressed as a percentage of control growth for each isolate).*

Region	Chemical	
	Propiconazole	Prochloraz
Wales	24.40	18.46
North(Newcastle)	29.72	23.53
North(Leeds)	35.18	23.05
Midlands & Western	26.44	20.35
South-west (Bristol)	27.72	20.09
South-west (Starcross)	20.84	26.53
East	29.07	23.88
South-east (Reading)	30.81	22.52
South-east (Wye)	22.17	23.13
Average (all regions)	27.38	22.11

Table 5.1. Fungicidal treatments applied to crops where isolates were found that grew in excess of 40% of control rates at the higher concentrations of propiconazole and prochloraz.

Sample number	Fungicides applied	
	First spray	Second spray
240	Tilt	-
313	Sportak Alpha	Corbel
435	Missile	-
453	Sportak	Radar
252	Tank mix[1]	Tank mix[1]
347	Tank mix[2]	

Tank mix[1] = fenpropidin + flutriafol + maneb

Tank mix[2] = fenpropimorph + flutriafol

Table 5.2. Fungicidal treatments applied to crops where isolates were found that grew in excess of 60% of control rates at the higher concentration of propiconazole.

Sample number	Fungicides applied First spray	Second spray
72(3 isolates)	Tilt Turbo	Tilt Turbo
125	Tank mix[1]	Tilt Turbo
215	Early Impact	-
241	Missile	-
278	Tank mix[2]	Tilt Turbo
383	Tank mix[3]	Tank mix[3]
407	Sportak Alpha	Radar

Tank mix[1] = carbendazim + flutriafol

Tank mix[2] = prochloraz + fenpropidin

Tank mix[3]= propiconazole + fenpropidin

Table 5.3. Fungicidal treatments applied to crops whose isolates showed the greatest growth on 1.0 µg/ml prochloraz and 5.0 µg/ml propiconazole, compared to their corresponding control growths.

Sample number	Fungicides applied First spray	Second spray
69	Tilt 250 EC	DMI/non MBC mix
122	Early Impact	-
223	Glint	Tilt turbo 375 EC
324	Sportak alpha	MBC/DMI mix
363	MBC/DMI mix	MBC/DMI mix
431	Sanction	Radar
444	Early impact	-

Figure 1. Dosage-response curve for prochloraz (mean of 20 isolates of *Pyrenophora teres*)

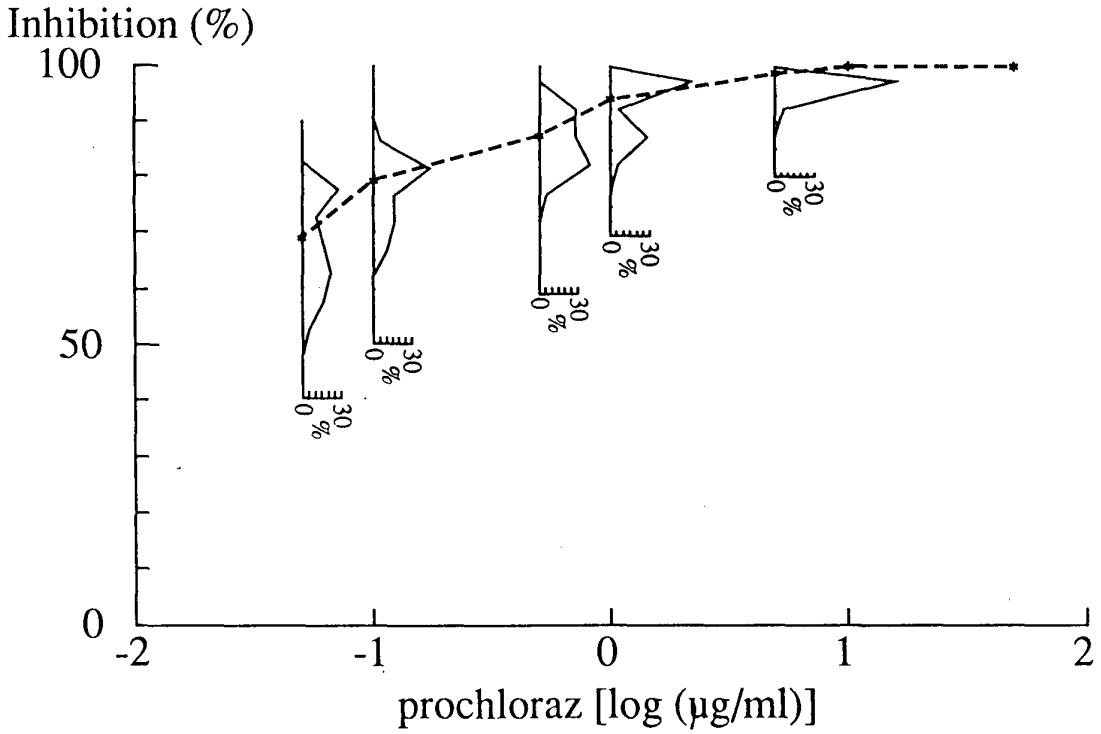

Figure 2. Dosage-response curve for propiconazole (mean of 20 isolates of *Pyrenophora teres*)

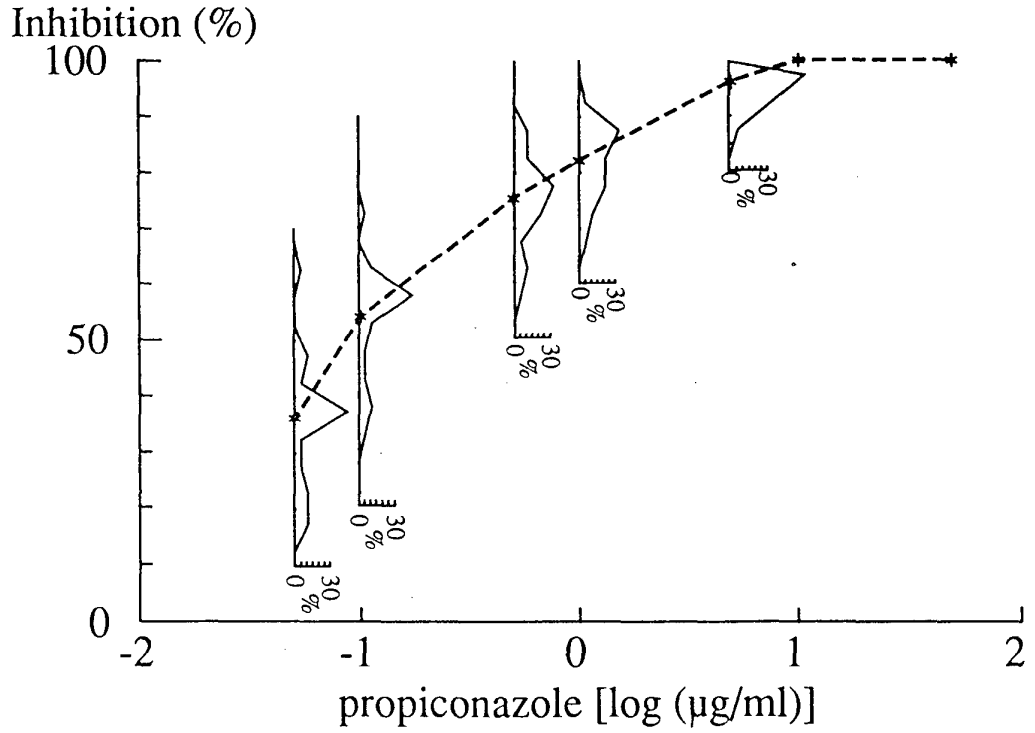

Figure 3. Dosage-response curve for prochloraz
(mean of all isolates for 1988 and 1989)

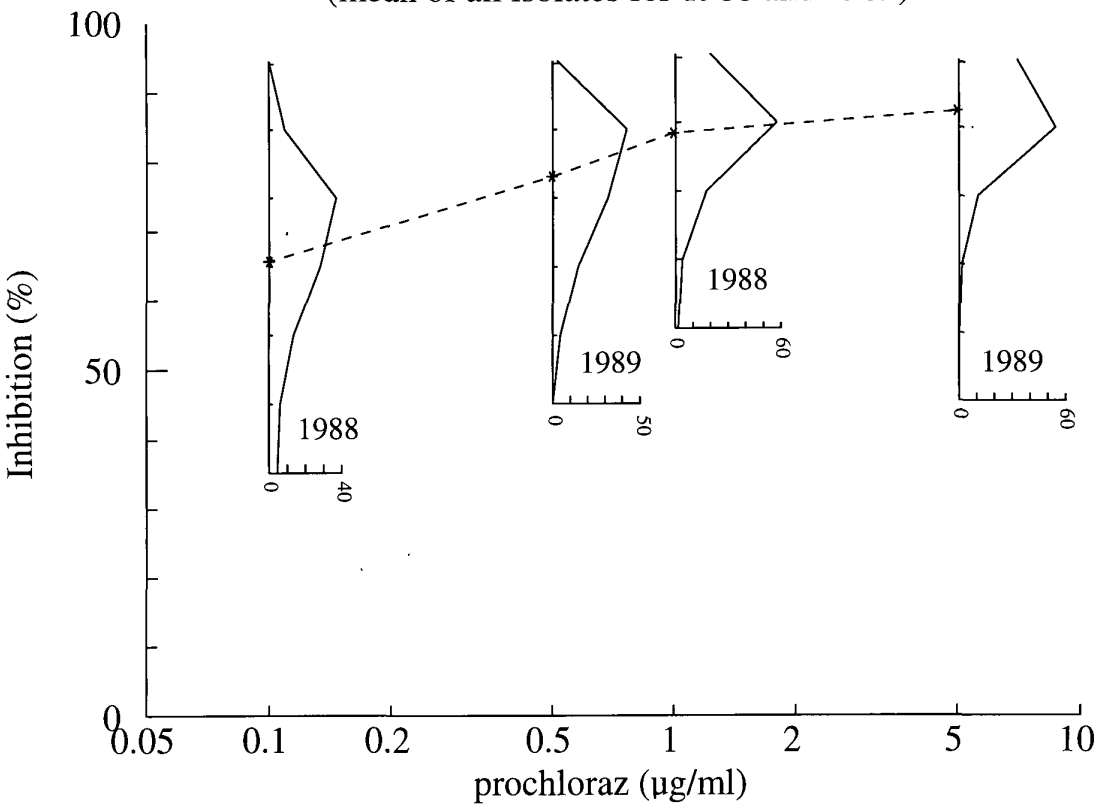

Figure 4. Dosage-response curve for propiconazole
(mean of all isolates for 1988 and 1989)

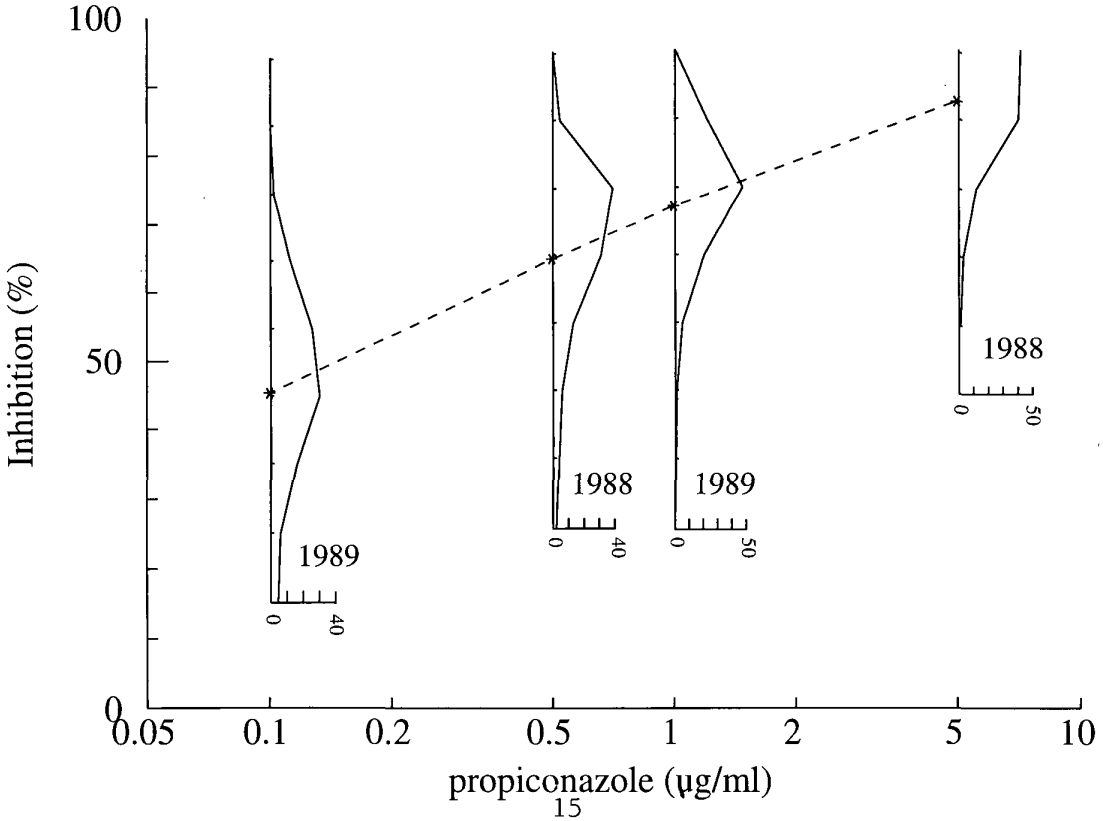

Figure 5. Growth distribution of isolates of *P.teres* after 2 weeks on low concentrations of prochloraz and propiconazole

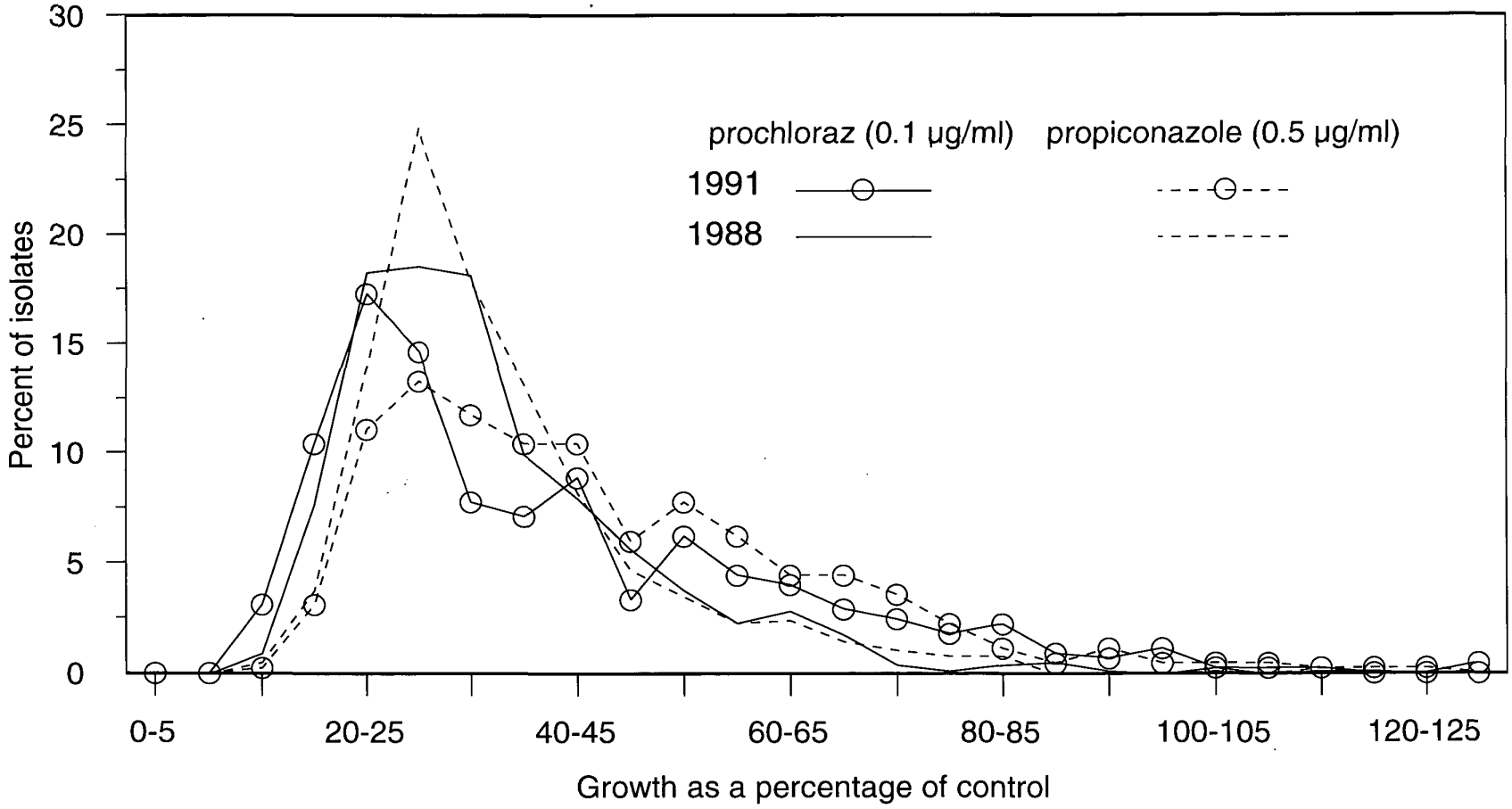

Figure 6. Growth distribution of isolates of *P. teres* after 2 weeks on high concentrations of prochloraz and propiconazole (1988 & 1991 data)

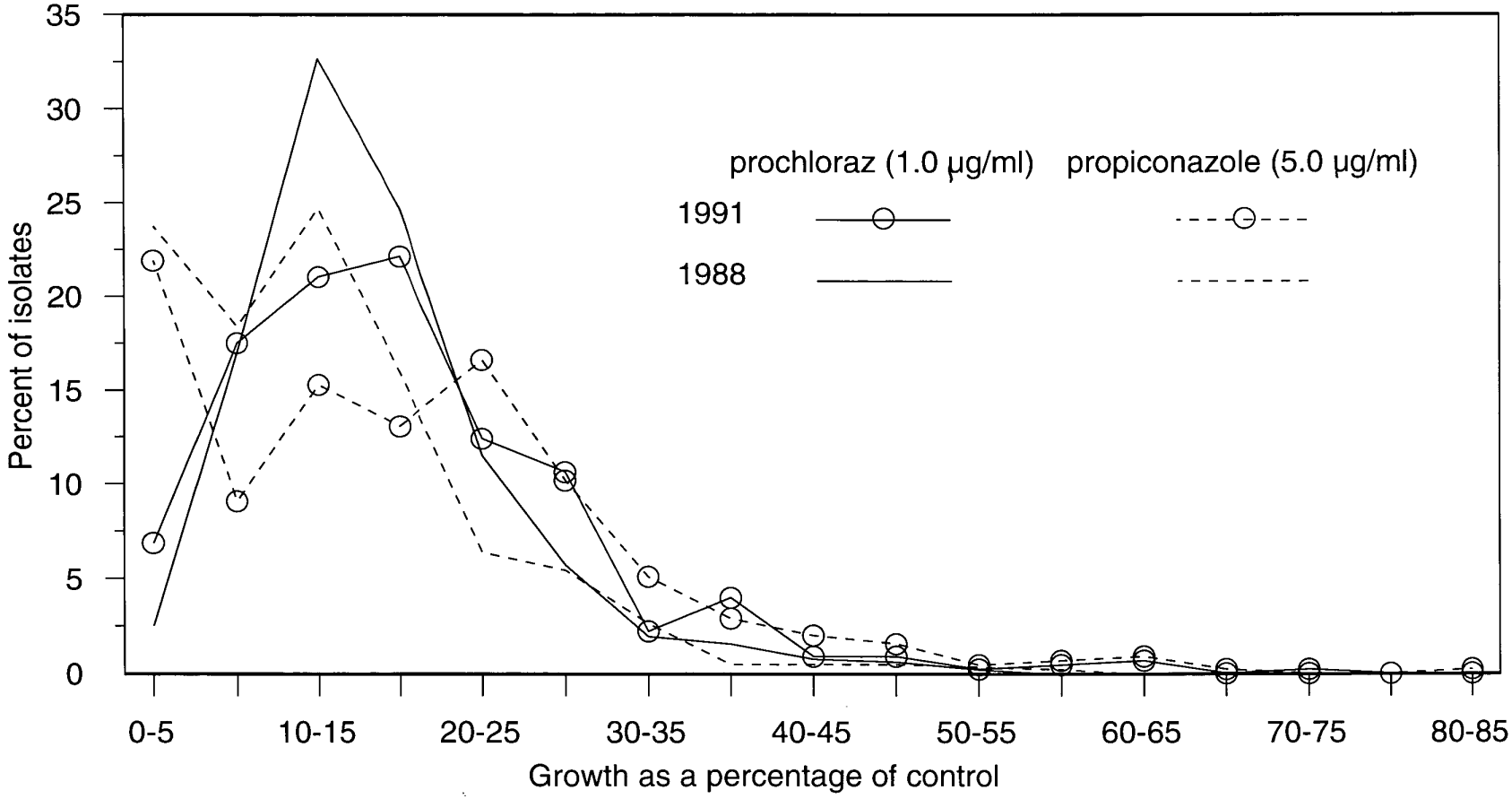

Figure 7. Growth distribution of isolates of *P. teres* after two weeks on high concentrations of prochloraz and propiconazole

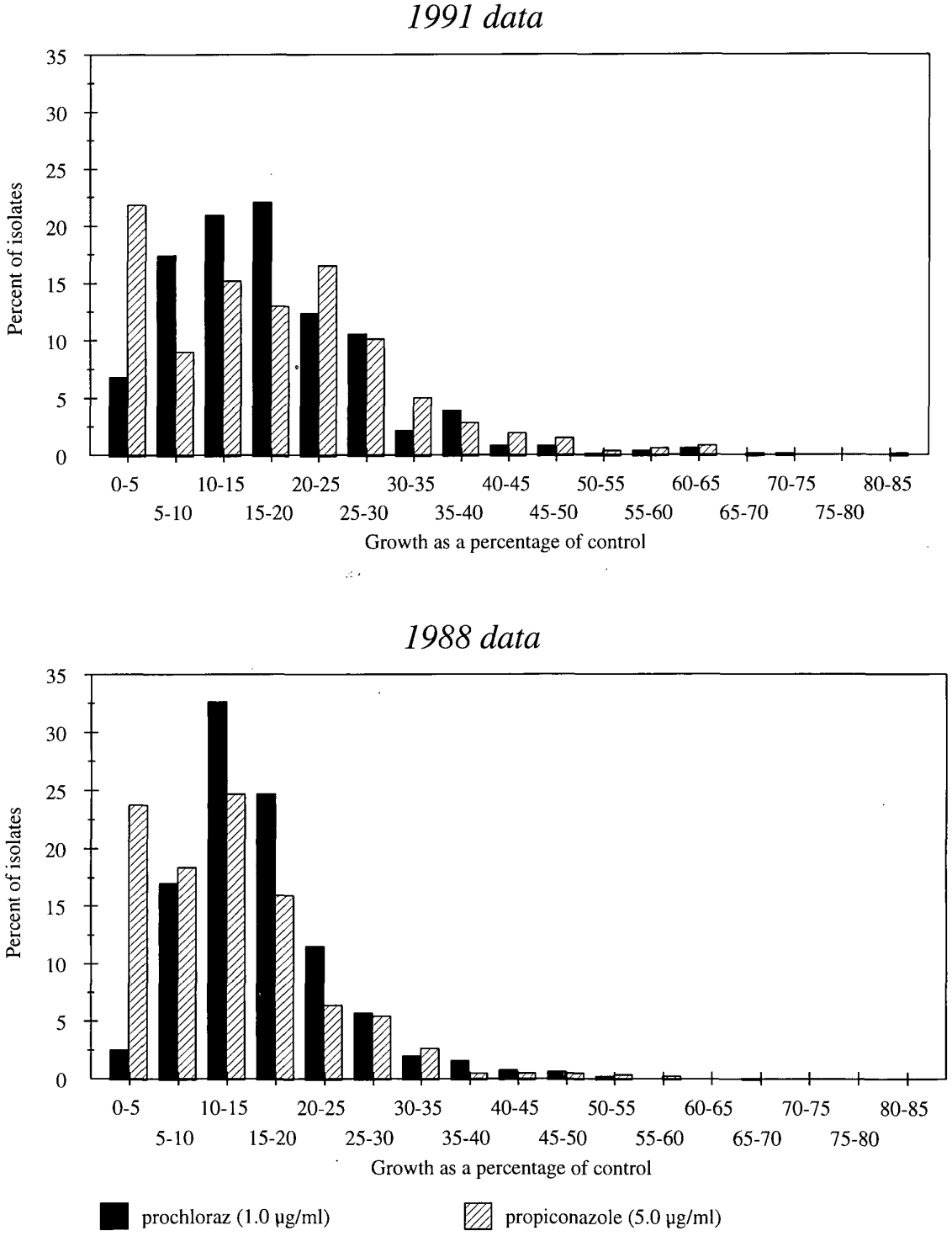

Figure 8. Growth distribution of isolates of *P. teres* after two weeks on low concentrations of prochloraz and propiconazole

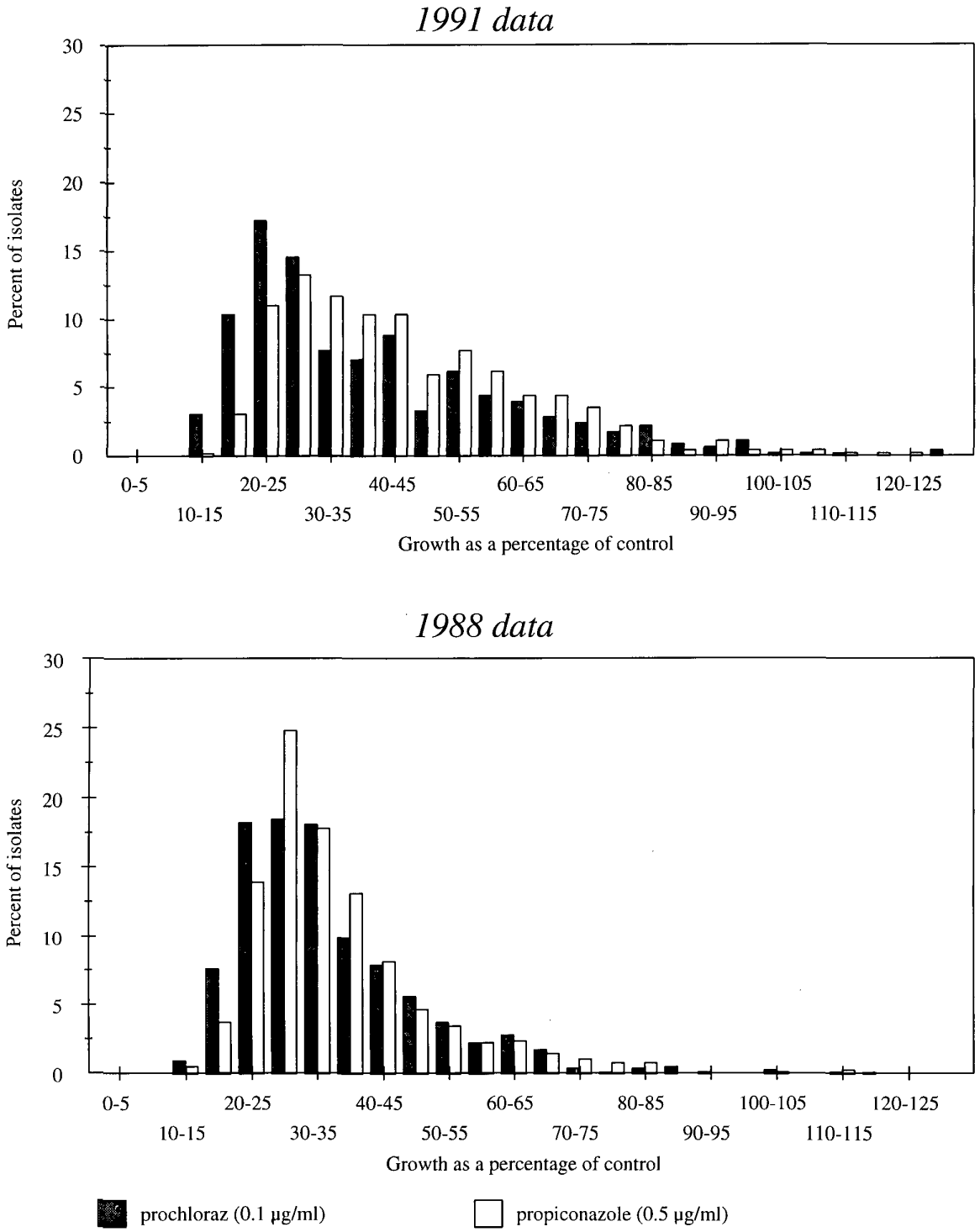

Figure 9. Comparison between the sensitivity of isolates of *P. teres* obtained during 1988 and 1991 to 1.0 µg/ml prochloraz

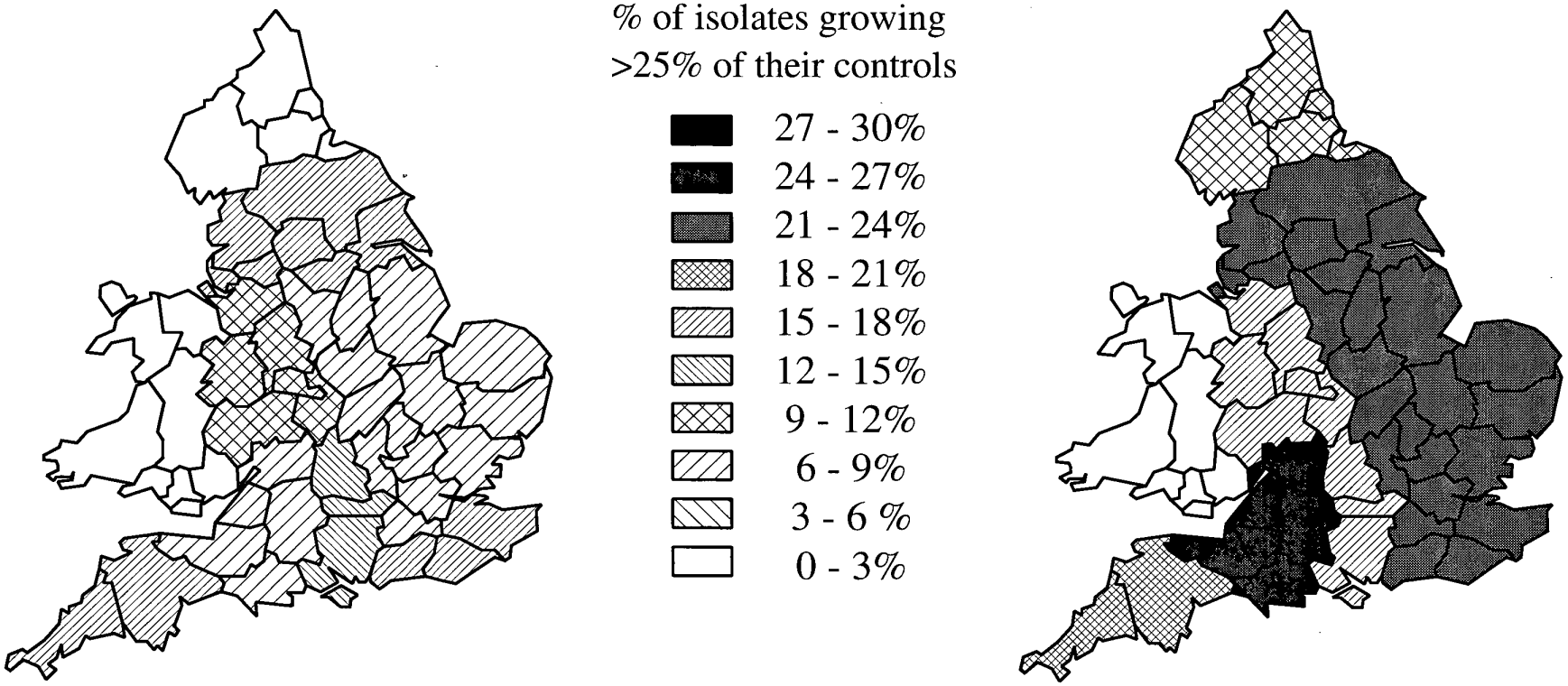

Figure 10. Maps comparing the incidence of reducing sensitivity in isolates of *P. teres* from the ADAS regions of England & Wales to 5.0 µg/ml propiconazole