June 2010
£12.00

Project Report No. 470

Targeting winter and spring barley disease management

by

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This project commenced on 1st July 2004. The work was funded by a contract for £32,694 from the HGCA (Project RD-2004-3017). The project was sponsored by ADAS Consulting Ltd, Scottish Agricultural College, UAP Ltd, CSC Crop Protection Ltd, Masstock Arable (UK) Ltd, Agrovista UK Ltd, BASF plc, and Bayer Crop Science through the Sustainable Arable LINK programme (£833,052).

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1. ABSTRACT

Field experiments tested the mechanisms by which fungicide treatment affects the growth of barley, so that sprays might be better targeted. Experiments were conducted on winter and spring barley at two 'core' research sites: ADAS Rosemaund (Herefordshire) and SAC Aberdeen. Two designs of experiments were undertaken on winter barley. In the first design, different seed rates and N regimes (amount and timing) were imposed to cause variation in the source-sink balance of the crop similar to that which normally occurs between crops due to differences in plant establishment and N availability (here 'source' is defined as the dry matter available for grain filling and 'sink' is the capacity of grains to store the dry matter). The effect of source:sink balance on response to fungicide treatment was then tested. In the other design, fungicide timing was varied to determine the effects of disease control during specific crop developmental phases on canopy growth, light interception, and yield formation. In the spring barley experiments, seed rate, N regime and fungicide timing treatments were combined. Forty seven trials were conducted by industry partners to test the effects of fungicide timing on yield and validate the findings from the core sites across a wide range of varieties and sites.

The effects of fungicide treatment on canopy growth, deposition of stem-storage reserves, and yield components was similar in spring and winter barley and for two and six-row varieties, indicating that the key growth stages for disease management in barley are common – albeit that those growth stages occur at different calendar dates according to sowing date. Protection should start during tillering to maximise grain numbers by protecting production and survival of tillers and spikelets. Early tillering, stem-extension (T1) and, to a lesser extent, booting/ear emerged (T2) applications increased grain numbers per m^2. T1 and T2 treatments increased average grain weight – an effect which appeared to be the result of an increase in grain storage capacity (sink) rather than increased supply of dry matter for grain filling (source). At industry sites where there was little or no visible disease, the average yield response to fungicide was similar to those sites where there was disease. Effects on grain numbers were not simply the result of protection of green area, but may have involved direct effects of fungicides on grain site formation or prevention of deleterious effects from symptomless infections. The findings from this project have been incorporated in the HGCA Barley Disease Management Guide.

2. SUMMARY

2.1 Background and aims

For barley production to be economically sustainable, high yields of quality grain need to be obtained consistently, without prohibitive input costs. The splash-borne diseases of barley are particularly damaging and can require substantial fungicide inputs for effective control. However, there can be considerable variation in yield response to fungicide between crops. Hence, fungicide treatments need to be well targeted. Rational treatment decisions are also needed to avoid unnecessary exposure of pathogen populations to fungicides and the consequent increased selective pressure for fungicide insensitivity.

Management decisions about the control of foliar disease are usually based on a consideration of the amount of visible disease present in the crop and the risk of further epidemic development. At present, understanding of the mechanisms by which disease impacts on yield and the agronomic and environmental factors that influence this relationship is not developed enough to allow fungicide treatments to be tailored according to the state of the crop and its likely yield response.

Yield formation in cereals is often considered in terms of the interplay between source and sink. Here, 'source' is defined as the dry matter available for grain filling from post-anthesis photosynthesis and remobilisation of pre-anthesis storage reserves and 'sink' is the number and capacity of grains to store assimilate. Current evidence indicates that the yield of UK barley tends to be sink-limited (limited by the storage capacity of grains rather than supply of assimilate to fill them) and that the extent of the source-sink imbalance varies between sites and years. This may explain field observations that report variable responses to late season disease control, as strongly sink-limited crops are likely to be unresponsive to late treatments which only increase post-anthesis source.

Pathogens may reduce crop growth by affecting radiation interception, radiation use efficiency (RUE, the amount of dry matter produced per unit of light energy intercepted) or the partitioning of assimilates. RUE is used in this report to describe photosynthetically active radiation (PAR) -use efficiency, i.e., RUE is biomass production per unit PAR intercepted by healthy tissue. The impact of these effects on yield is likely to depend on the timing of the disease epidemic and the relative source-sink balance of the crop. If the epidemic occurs early, these effects could restrict the

development of sink capacity by limiting the number of ears produced and the potential number of grains per ear. Later epidemics, on the other hand, around the time of flowering, could restrict sink capacity by limiting grain development and potential grain size. At the same time, disease may restrict the supply of assimilate for grain filling by constraining the expansion of the canopy green area before flowering, canopy duration after flowering, or the pre-anthesis accumulation and post-anthesis remobilisation of storage reserves.

Rationale use of fungicides dictates that treatments are applied only to those crops likely to respond with an increase in grain yield or quality. The first step in targeting disease management is to understand how disease affects yield and to identify which developmental phases are most important to protect. Thus the aims of the project were to improve understanding of the physiological mechanisms underlying the response of barley to disease to help target fungicide treatment by:

quantifying the effect of source-sink balance on the response to disease control,

quantifying the effects of disease during different developmental phases on growth, resource capture and yield formation.

The project was structured to reconcile the focus required for mechanistic studies with the need for broad applicability of the findings. Experiments at ADAS Rosemaund and SAC Aberdeen undertook a detailed investigation of the physiological responses of winter and spring barley crops to fungicide. Forty seven experiments conducted by industry partners over a range of sites determined the yield response to a common set of fungicide treatments in a selection of winter and spring cultivars grown with contrasting drilling dates, agronomic regimes and disease pressures.

2.2 Methods and Results

2.2.1. The effect of source-sink balance on the response of winter barley to fungicides

Methods

A two-row winter barley variety (*Hordeum vulgare* L. cv Haka) was sown between 23rd September and 1st October 2005 and 2006 at ADAS Rosemaund and SAC Aberdeen. Treatments consisted of a factorial combination of four nitrogen levels, two seed rates and two fungicide levels. The nitrogen treatments were made up of different combinations of application rate and timing (Table I). The full recommended N fertilizer

requirement was calculated on the basis of previous cropping and an analysis of soil mineral N. Treatments then ranged from 0.66 to 1.33 times the full recommended rate and were applied as a split dressing of ammonium nitrate. The first split was targeted at mid-tillering (GS 23/24) to promote tiller production and the second split 4-6 weeks later after the start of stem extension (GS 32/39) to promote retention of green leaf area post-anthesis. The time between the split was intended to generate a large contrast in canopy growth and duration. The seed rates were 150 and 300 seeds m^{-2} at ADAS Rosemaund and 150 and 350 seeds m^{-2} at SAC Aberdeen, for the 'low' and the 'high' seed rates, respectively.

Table I. N treatments at Aberdeen and Rosemaund

Treatment label	Fraction of full recommended rate	
	1st Split, GS 23/24	2nd Split, GS 37/39
*HH	2/3	2/3
HL	2/3	1/3
LH	1/3	2/3
LL	1/3	1/3

Where: * H = high rate, L = low rate; HH is high followed by high, HL is high followed by low, etc.

The disease of interest was *Rhynchosporium secalis*. There were two fungicide treatment programmes: [1] untreated and [2] full fungicide treatment comprising three applications; autumn, GS 31/32 and GS 59. At each of the three times the same fungicide mixture was used with the aim of giving complete control of rhynchosporium: epoxiconazole + boscalid (half rate, 0.75 l ha^{-1} Tracker, BASF) plus prothioconazole + fluoxastrobin (half rate, 0.625 l ha^{-1} Fandango, Bayer) plus fenpropimorph (0.3 or 0.4 of full rate, 0.3-0.4 l ha^{-1} Corbel, BASF). All plots were treated with quinoxyfen (full manufacturers recommended rate; 0.3 l ha^{-1} Fortress, Dow) at GS 30 to prevent powdery mildew infection without affecting *R. secalis*. Measurements were made of % disease and % green leaf area, absolute leaf area, biomass, stem water soluble carbohydrates, canopy light interception, grain yield and yield components.

Disease, source-sink balance and yield

The combination of seed rate and N treatments was successful in achieving variation in the growth and duration of source structures relative to sink. For example, low seed rate reduced the amount of soluble carbohydrate reserves at flowering relative to grain numbers and low N reduced healthy area relative to grain numbers. There was significant Rhynchosporium leaf scald (>5% surface covered by lesions averaged over top 3 leaves) at both sites and in both years, but disease severity was greater at Aberdeen.

Fungicide treatment increased post-anthesis PAR interception and the healthy canopy area (HAI) and quantity of stem soluble carbohydrates at flowering. These effects were associated with an increase in yield (average 1.4 t ha^{-1} at 100% DM) and its components: ears m^{-2}, grains m^{-2} and mean grain weight (MGW). Significantly, the yield response to fungicide was influenced by seed rate and N regime. In a cross site-year analysis, fungicide application resulted in an additional 0.3 t ha^{-1} at high seed rate compared to low and with high (HH) and late N (LH) compared to early (HL). Further, the additional yield response was almost entirely associated with an increase in grain number m^{-2}. The effects of seed rate and N treatments on the response to fungicide were not explained by changes in disease severity with these treatments.

Effect of fungicides on grain numbers

Grain number m^{-2} was linearly related to PAR interception by healthy tissue across sites and years when the period from crop emergence to ear emergence was considered. At low seed rate the increase in grain number m^{-2} with fungicide treatment was explained in terms of an increase in healthy area PAR interception pre-anthesis. At high seed rate, the increase in grain number with fungicide was not associated with an equivalent increase in PAR interception. Instead, there was 30% increase in the number of grains produced per unit of PAR intercepted (increase in slope, Table II).

Table II. Slope of the relationship between pre-anthesis PAR interception (x axis) and grain number (y axis) for winter barley cv Haka at different seed rates with and without a full fungicide programme. ** = significant (P<0.001).

Seed rate/m^2	Fungicide	Slope (b)		Intercept (a)	
150	Untreated	35.15		-1227	
150	Treated	38		-1888	
300/350	Untreated	36.07		-1796	
300/350	Treated	47.92	**	-6265	**

By contrast, the interaction between N regime and fungicides on grain number was not associated with effects on pre-anthesis PAR interception or changes in the sensitivity of grain site formation to intercepted PAR. It was associated with a greater increase in PAR interception post-anthesis following fungicide application in the HH and LH regimes compared to early N (HL). It is possible that fungicide treatment increased fertilization and grain set or the survival of late developing tillers more when availability of N late in the season was high.

Effect of fungicides on mean grain weight

No significant interaction between seed rate or N regime and fungicide treatment on mean grain weight (MGW) was found, implying that MGW was equally responsive to fungicide under all these treatments. The potential supply of assimilate for grain filling was calculated from the post-anthesis PAR interception multiplied by the RUE plus the pre-anthesis storage reserves. The seed rate and N treatments gave a large variation in potential supply, but fungicide increased MGW over the same broad range of potential supply found in untreated plots (Fig. I). These results suggest that fungicides increase MGW by influencing grain development and the capacity for storage of dry matter, and not by increasing the supply of assimilate for grain filling. Fungicides might have a direct growth regulatory effect on grain development, or they might be controlling symptomless pathogen infection. If pathogen infection restricts grain development without, or before, visible symptom expression, control of the pathogen

could lead to an increase in grain storage capacity without an associated increase in estimated healthy area PAR interception and assimilate supply.

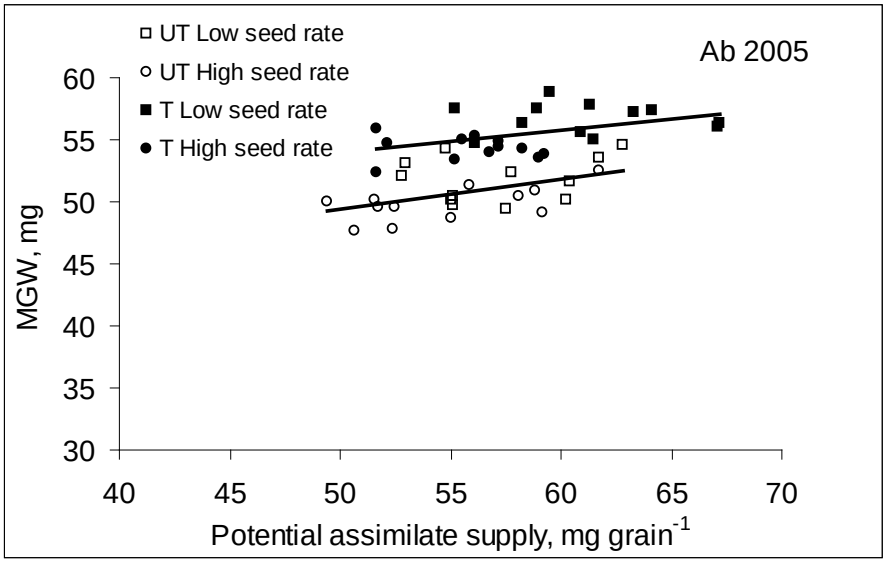


Figure I. Relationship between potential assimilate supply per unit grain number post-anthesis and mean grain weight at 100 % DM for fungicide treated (T) and untreated (UT) crops. Within each fungicide treatment, data for seed rate and N treatments have been pooled. Results are for the Aberdeen (Ab) site in 2005.

2.2.2 The effect of fungicide timing on the growth, photosynthetically active radiation (PAR) interception and yield of winter barley

Methods for Experiment 1

Fungicide timing at a single seed rate and N regime. Barley cv Haka was grown at a standard seed rate (300 seeds m^{-2} Rosemaund, 350 seeds m^{-2} Aberdeen) and N regime (split application of full recommended rate). Fungicide treatments consisted of a factorial combination of spray timings: autumn (GS21/22 Aberdeen and GS 22/24 Rosemaund), spring (GS 31/32; T1) and summer (GS 49/59; T2) The 8 fungicide treatment timings were: [1] autumn, T1, T2 (+++), [2] autumn and T1 (++-), [3] autumn and T2 (+-+), [4] T1 and T2 (-++), [5] autumn only (+--), [6] T1 only (-+-), [7] T2 only (--+), [8] untreated (---). At each application timing the same fungicide mixture was used (active ingredients as described above). Disease and crop growth

assessmnts, PAR interception and final harvest yield and grain quality measurements were also as described above.

Methods for Experiment 2

Fungicide timing in combination with seed rate and N treatments. Barley cv Haka was sown at 150 and 450 seeds m^{-2}. The experiment was a factorial design with 2 N timings x 2 seed rates x 6 fungicide regimes, as a split-split plot design with N treatments as main plots, seed rate as sub-plots and fungicide treatments as sub-sub plots. The N treatments were early (GS 23/24) or late (GS 31) application of the full recommended rate for feed barley. There were 6 fungicide treatments: [1] autumn, T1 and T2 (+++), [2] autumn and T1 (++-), [3] autumn only (+--), [4] T1 only (-+-), [5] T2 only (--+), [6] untreated (---). Fungicide mixtures and rates at each timing were the same as described above. Disease and % green leaf area were assessed at key growth stages, but no growth analysis was undertaken. Harvest and grain quality data were recorded as described above.

Results for Experiment 1: Disease, Yield and yield components

The main disease at each site and in each year was rhynchosporium. Disease severity was generally much lower in 2006 than 2005 at each of the sites. There were significant yield responses to fungicide application in each site-year combination except Aberdeen 2006 (Table III). The most consistent yield responses came from autumn and GS 31 (T1) applications. In 2005 there were also significant responses to applications at GS 49/59 (T2). There was no significant response to T2 application at Rosemaund in 2006 when the application was unavoidably delayed until GS 65 (Table III).

Table III. Grain yield response to different fungicide timings. Values are main effect means after ANOVA. Responses highlighted in bold are significant at P <0.05. (3 site mean determined by ANOVA for unbalanced designs after removing Ab 2006).

Year	Site	Fungicide	Yield, t ha^{-1} @100% DM		
			Autumn	T1	T2
			GS 21/24	GS 31/32	GS 49/59
2005	Ab	untreated	5.42	5.21	5.17
		treated	5.70	5.91	5.95
		response	0.28	0.70	0.79
2005	Rm	untreated	6.17	6.11	6.04
		treated	6.87	6.92	6.99
		response	0.70	0.81	0.95
2006	Ab	untreated	5.76	5.79	5.80
		treated	5.84	5.81	5.80
		response	0.09	0.02	0.00
2006	Rm	untreated	7.77	7.62	7.95
		treated	8.28	8.44	8.10
		response	0.50	0.82	0.15
3 site mean		untreated	6.50	6.31	6.44
		treated	6.94	7.14	7.01
		response	0.44	0.83	0.57
df 50		SED	0.074	0.074	0.074

Where Ab is Aberdeen and Rm is Rosemaud

Analysis showed significant effects of autumn, T1 and T2 fungicide applications on yield, but no significant interaction between the different timings. This implies that the response to a particular application timing was not affected by applications at other timings, i.e., the effects of each timing on yield appeared to be additive.
Grain number per m^2 was increased by autumn, T1 and to a lesser extent T2 applications (in some site-years). Mean grain weight, on the other hand, was increased by T1 and to a greater extent T2 applications. Where there was a response of MGW to fungicide, it appeared to arise from an increase in storage capacity of the grain rather

the supply of assimilate for grain filling. This was the case for both T1 and T2 applications.

A comparison of the effects of each fungicide timing on PAR interception over particular growth stages with the increase in grain number elicited suggests that the T1 application may have particularly strong effects compared with other timings. The increase in grain number per unit of additional PAR intercepted was twice that observed with autumn and T2 treatments.

Results for Experiment 2

Fungicide treatment had a significant effect on yield, grains per m^2 and MGW at each site and there was a significant interaction between seed rate and fungicide on yield at Rosemaund, and near significant (P=0.055) interaction at Aberdeen. At high seed rate there was a significant increase in yield with all fungicide timings. At low seed rate, however, there was a significant increase only when autumn treatment was combined with T1 and T1 plus T2, not with single applications. There was no significant interaction between N regime and fungicide treatment.

2.2.3. The effects of source-sink balance on the response to fungicides in spring barley

Methods

Spring barley (*Hordeum vulgare* L. cv Cocktail) was sown on 23 March in 2006 and 2007 at ADAS Rosemaund and 5 April 2007 and 20 April 2008 at SAC Aberdeen. Experimental treatments consisted of two nitrogen timings, two seed rates and four fungicide timings. The N was applied either at crop emergence (early N) to promote tiller production, or after the start of stem extension (late N) to promote retention of green leaf area post-anthesis. Seed rates were 100 and 600 seeds per m^2 representing low and high seed rate extremes respectively. The fungicide treatment programmes were: [1] untreated (- -), [2] application at T1 only (GS 31) (-+), [3] application at T2 only (GS 45-59) (-+), [4] application at T1 and T2 (++). The same fungicide active ingredients and products were used as described previously. At Rosemaund, all plots were treated with metrafenone to prevent powdery mildew infection and allow rhynchosporium to develop. At Aberdeen, as there was no sign of rhynchosporium, powdery mildew was allowed to develop instead.

Disease, green leaf area and source-sink balance

At Aberdeen in both 2007 and 2008 significant mildew epidemics occurred (average 7-8% for the top three leaves at anthesis). At Rosemaund in 2006, significant leaf scald developed late in the season with the greatest severity occurring after ear emergence (average of 4% on the top three leaves). There was little disease at Rosemaund in 2007. The seed rate and N regimes altered the relative source-sink balance of the crop at flowering. Thus, when averaged over fungicide treatments, late N significantly reduced the amount of storage reserves relative to eventual grain numbers and low seed rate increased the healthy canopy area relative to grain numbers when compared with the early N and high seed rate treatments respectively. Furthermore, in fungicide treated plots, low seed rate also led to a greater retention of green leaf area during the grain filling period.

Yield and yield components

There was a significant yield response to fungicide (0.5-1.6 t per ha) in each site-year, including Rosemaund in 2007 when the amount of visible disease was negligible. Both T1 and T2 applications increased yield relative to untreated controls. However, the combined T1 plus T2 application was more effective than either treatment on its own at Aberdeen, but not Rosemaund. An increase in grain numbers accounted for around 68-75% of the yield response observed with each fungicide treatment. The T1 application was the most consistent at increasing grain numbers, although T2 on its own was effective in some site-years. Effects of fungicide treatments on MGW also differed between site-years with the most consistent response coming from the T1 +T2 treatment.

In general, interactions between fungicide timing and either seed rate or N regime on yield were small and inconsistent and were not found in a cross-site-year analysis. No interaction was found between fungicide and either seed rate or N regime on MGW even though these treatments altered the potential supply of post-anthesis assimilate relative to the number of grains.

Effects of fungicide on PAR interception, grain numbers and MGW

When seed rate was used to vary the plant population and canopy area, there was a positive linear relationship between the amount of PAR intercepted by healthy tissue pre-anthesis and the number of grains produced per m^2. Separate lines were needed to describe the relationship for early and late N regimes. However, in each case fungicide treatment increased grain numbers over a range of healthy area PAR interception similar to that observed for untreated plots. The results indicate that the effects of fungicide treatment on grain numbers cannot be explained simply in terms of the protection of leaf area and increased PAR interception before flowering. As was observed with winter barley, analysis of the relationship between the potential assimilate supply post anthesis and MGW suggests that fungicides increased MGW through effects on grain storage capacity and not just the amount of assimilate available for grain filling. Effects of fungicide on canopy growth, storage reserve deposition and sink capacity in spring barley were comparable to those found in winter barley (Table IV).

Table IV. Summary of effects of fungicide treatment on eventual grain number and canopy components at GS 59 in winter and spring barley. Data are summarised from cross site-analyses on winter and spring barley experiments (effects of fungicide averaged across seed rate and N treatments).

Component	Winter	Spring
HAI	+ ***	+ ***
Above-ground biomass	+ ***	+ *
Fertile shoot number m^{-2}	+ ***	ns
Eventual grain number m^{-2}	+ ***	+ ***
WSC (% DM)	ns	ns
WSC (g m^{-2})	+ ***	+**
HA per grain	+ ***	ns
WSC per grain	ns	ns

Where: + = significant increase (*, $p<0.05$; **$p<0.01$; *$p<0.001$) with fungicide, ns = indicates no significant effect of fungicide. HAI is healthy area index of the canopy; WSC, stem water soluble carbohydrates.

2.2.4. Yield response of winter and spring barley to fungicide timing across sites and varieties

The aim of work in this part of the project was to provide independent data from a range of varieties and locations to validate findings from the experiments at the Rosemaund and Aberdeen sites described previously. Six industry partners, Agrovista UK Ltd., BASF plc, Bayer Crop Science Ltd., CSC Crop Protection Ltd., Masstock Arable (UK) Ltd. and UAP Ltd., provided results from experiments on a selection of winter and spring barley varieties, from a range of locations.

Methods

A total of 24 winter and 23 spring barley sites were contributed by the UK industry partners during the project. The fungicide treatments for winter barley were a factorial combination of autumn (GS 21/24), T1 (GS 31/32) and T2 (GS 49/59) applications, as described above for the ADAS and SAC winter barley experiments. For spring barley,

T1 and T2 spray times were used, and a T0 spray applied prior to GS 30 for some forward crops. For each site, fungicides were selected from the following: epoxiconazole + boscalid (Tracker, BASF), prothioconazole + fluoxastrobin (Fandango, Bayer), fenpropimorph (Corbel, BASF), quinoxyfen (Fortress, Dow) and spiroxamine (Torch Extra, Bayer). Products were allowed to be changed between spray times, but had to be the same within each spray time. Dose rates were selected to give good disease control at each application. In all three years, the majority of sites used epoxiconazole + boscalid, half rate (0.75 l ha^{-1}) with prothioconazole + fluoxastrobin, half rate (0.625 l ha^{-1}), and fenpropimorph, half rate (0.5 l ha^{-1}), at each of the three application times. In 2005 only, the Bayer winter barley sites did not have an autumn treatment. Assessments were made of disease severity at GS 12-14, 30-39 and 71, green leaf area at GS 59, final ear number per m^2, grain yield, MGW and specific weight. Because many of the data provided were site averages, no statistical analysis has been conducted.

Winter barley

Rhynchosporium, net Blotch, powdery mildew and brown rust were the main diseases recorded in winter barley over three years, with large variation in incidence between sites, varieties and years. Yields from plots receiving the full fungicide programme ranged from 3.1 to 11.8 t per ha, with yield responses to fungicide from 0.12 to 3.56 t per ha. The early disease recorded at GS 30-39, although low, was as good an indicator of yield response as the late disease. Yield response tended to be larger for sites with disease at GS 30/39 than without, although in general the yield response was poorly related to disease severity.

On average, across all sites and years, the contribution to yield from the three fungicide timings was not additive. Thus, the response was greatest when the fungicide at a particular timing was the sole treatment rather than in combination with other timings (Table V). The implication is that if a treatment is missed or gives inadequate disease control the damage to yield potential might be recoverable, at least in part, with a later fungicide application.

Table V. Winter barley yield responses (t per ha), industry partner sites 2005-2007, 24 UK sites in total.

Fungicide timing	Response comparison	Yield response, t/ha
Autumn (GS 21/24)	Autumn - UT	0.54
	Fully treated – (T1 + T2)	0.12
	Mean	0.25
T1 (GS 31/32)	T1 – UT	1.18
	Fully treated – (autumn + T2)	0.37
	Mean	0.69
T2 (GS 39/49)	T2 - UT	1.04
	Fully treated – (autumn + T1)	0.35
	Mean	0.61

The autumn spray resulted in an increase in yield and grain number, but the T1 and T2 timings gave larger responses. The T2 spray gave as large a response as the T1 spray, unlike the core research sites where it was the T1 spray that gave the largest grain number response. The Industry sites tended to have earlier T2 timings than the core sites (GS 39-49 cf 59), which could explain the larger grain number response with T2 that occurred at the industry sites.

When data for varieties with a common ear type were averaged, the 6-row varieties produced a greater yield and number of grains per m^2 than the 2-row varieties, but the difference was not large. The yield response to the full fungicide programme was comparable for 6- and 2-row varieties (approx. 1.8 t per ha at 85% DM).

The sites were assigned to three main UK regions as follows: [1] North (Scotland, Yorkshire), [2] West (Cheshire, Herefordshire, Hampshire, Dorset, Wiltshire) and [3] S / SE (Leicestershire, Essex, Kent, Norfolk, Hertfordshire, Suffolk). There were few winter barley sites in the north. There were no clear effects of region on the yield response to fungicide, nor the response of particular yield components.

The sites were categorised into those with high (> 600) and low (<400) ear numbers per m^2 in untreated plots. For >600 ears per m^2, the varieties were Carat, Haka and Pearl, and for <400 ears per m^2, the varieties were Saffron, Haka, Pearl and Siberia. The yield response with the T1 spray was larger for the thick crops. For the T2 spray however the yield response was larger for the thin crops. There was no large difference in yield response between the thick or thin crops with the autumn treatment.

Spring barley

Rhynchosporium was the predominant disease in spring barley, noted in 13 out of 20 sites assessed compared to 7 out of 23 sites for winter barley. Net blotch was much less frequent than in winter barley, but ramularia was noted at several sites whereas winter barley had none. Yields from fully treated plots ranged from 4.4 to 9.5 t per ha at 85% DM, with yield responses to fungicide (treated minus untreated) from 0 to 1.8 t per ha. The yield response at sites with disease at both GS 30-39 and later growth stages was similar to the yield response at sites which had no disease.
Overall the T2 treatment gave a larger yield response than the T1 treatment and, as with winter barley, the treatment timings were not additive (Table VI).

Table VI. Spring barley yield responses, industry partner sites 2005-2008, 23 UK sites in total.

Fungicide timing	Response comparison	Yield response, t/ha
T1 (GS 31/32)	T1 - untreated	0.28
	Fully treated – T2	0.06
	Average	0.17
T2 (GS 39/49)	T2 - untreated	0.52
	Fully treated – T1	0.31
	Average	0.42

The yield response to both T1 and T2 was via an increase in grains per m^2. There was some treatment effect on MGW at individual sites, but not when data were averaged across sites. The yield response in thinner crops tended to be larger than for thick crops, for both the T1 and T2 treatments.

2.3. Conclusions & Implications

The effects of fungicide on canopy growth, deposition of storage reserves, yield and yield components was broadly similar in spring and winter barley (Table IV) and for two and six-row winter barley varieties, indicating that a common approach to disease management in barley is appropriate.

Protection of the canopy should start early in the season and continue after flowering to maximise grain numbers and potential grain size (i.e. grain storage capacity). In winter barley significant yield responses were found with autumn, T1 and T2 treatments at the core research sites on variety Haka and the effect at all three timings was additive. Over the wider range of varieties and locations covered by industry partners, the response to autumn fungicide was smaller and less consistent and the effects of the different timings were not additive. Nevertheless, the greatest yield responses were observed with a three (autumn + T1 +T2) or two spray programme (T1 + T2). Similarly in spring barley the greatest yield response was often obtained with the combined T1 and T2 treatment.

T1 and T2 applications coincide with the phase of tiller and spikelet mortality, a major determinant of the number of potential grain sites. However, effects of fungicides on grain numbers cannot be explained just by the control of visible disease symptoms and consequent increase in healthy leaf area and PAR interception. Thus, in both winter and spring barley, grain numbers were increased where there was little change in pre-anthesis PAR interception. Alternative explanations for the observations are 1) fungicides control low levels of visible disease or symptomless pathogen infection, leading to an increase in grain numbers via effects on RUE or partitioning of assimilates without increasing PAR interception; 2) fungicides have direct effects on plant development and grain numbers in the absence of disease.

Yield response to fungicide varied with the physiological state of the crop. This was most pronounced in winter barley where the increase in grain number in response to fungicide was greater in crops at a standard seed rate (referred to as ‘high’ in this study) rather than low. The same trend was also found across the locations and varieties used at the industry sites. Effects of seed rate and N regime on the response to fungicide in spring barley were less consistent. The implications of these findings are that fungicide treatments should not be relaxed on relatively dense winter barley crops on the false assumption that they have tillers and potential grain numbers to spare.

The inconsistent effect of seed rate on the response to fungicide in spring barley could be associated with the shorter periods of tiller production and mortality in spring varieties.

Fungicides appeared to increase MGW by influencing grain development and the storage capacity of grain rather than simply the supply of dry matter for grain filling. In both winter and spring barley, MGW was increased over a wide range of potential assimilate supply. Grain storage capacity is believed to be determined by developmental events shortly before and after anthesis. This suggests that so long as this phase of grain development is protected, it may not be important for T2 fungicide treatments to ensure retention of green leaf area late in the grain filling period. However, this hypothesis needs to be tested directly.

There was no evidence, in either winter or spring barley, that the response to a T2 fungicide was influenced by agronomic treatments designed to vary the source-sink balance. Thus, the suggestion that fungicide treatment could be relaxed in crops where the supply of assimilate for grain filling (source) significantly exceeds the storage capacity of the grain (sink) was not supported by the results of this study. This is probably because fungicides themselves somehow increase the storage capacity of the grain. Thus, there appears to be little scope for tailoring T2 applications according to in-field assessments of canopy size relative to potential ear or grain number.

The need for fungicide cannot be predicted from an assessment of the risk or presence of visible disease in the crop on its own. Yield response to fungicide was poorly related to the amount of visible disease in the crop. This was especially the case for spring barley where the average yield response at industry sites with little or no disease was comparable to that for sites with disease. The poor relationship probably results from significant effects of low levels of visible disease, or symptomless infection, on plant growth and yield formation, or the possible direct effects of fungicides on plant development described above. Possible growth regulatory effects of fungicides have been reported previously (e.g. delayed leaf senescence). However, contrary to these reports, results from the current study indicate that any direct effect of fungicide must occur early in crop development when grain numbers are being determined and are not associated with an increase in PAR interception. Further research is needed to identify the mechanisms by which fungicides elicit their effects on grain numbers and MGW in order to determine whether treatments can be targeted more effectively to those crops most likely to respond.

3. TECHNICAL DETAIL

3.1 The effect of source-sink balance on the response of winter barley to fungicides

3.1.1 Introduction

Management decisions about the control of foliar disease in cereals are usually based on a consideration of the amount of visible disease present in the crop, the risk of further epidemic development and the likely response of grain yield and quality to the application of fungicide. Although it has long been recognised that the yield response to disease control can differ widely between crops expressing comparable levels of visible disease symptoms (Schafer, 1971; Kramer et al. 1980), current understanding of the mechanisms underlying such variation is not developed enough to allow fungicide treatments to be tailored according to the likely response.

Pathogens may reduce crop growth by affecting PAR interception, PAR use efficiency (RUE) or the partitioning of assimilates (Boote et al., 1983; Johnson, 1987; Gaunt, 1995). RUE is used in this report to describe photosynthetically active radiation (PAR)_use efficiency, i.e., RUE is biomass production per unit PAR intercepted by healthy tissue. The impact these effects have on grain yield will depend to a large extent on the source-sink relations of the crop and the developmental stage at which disease occurs. Pathogens may reduce yield by restricting the formation of each of the major yield components (Gaunt, 1995; Madden & Nutter, 1995). Early epidemics of foliar pathogens which develop during canopy expansion, may reduce the number of ears produced and the potential number of grains per ear because disease infection coincides with the period of tiller and spikelet production and survival (Brooks, 1972; Lim & Gaunt, 1986; Conry & Dunne, 1993). By contrast, late epidemics may restrict the later formed yield components, namely the final number of grains per ear and the mean grain weight. It follows that the smaller yield loss of some crops to disease (greater apparent tolerance) compared to others could arise through the possession of traits that maintain PAR interception, RUE and dry matter partitioning to the developing ears and grains in spite of disease (Bingham et al., 2009).

The control of grain filling is often considered in terms of source- or sink-limitation. Here 'source' refers to the potential supply of dry matter from post-anthesis photosynthesis and remobilisation of pre-anthesis storage reserves, whilst 'sink' refers to the number and storage capacity of grains. There is evidence that even under the low light conditions of NW Europe grain filling of barley is predominantly sink limited as the potential supply of assimilate exceeds the total storage capacity of grains (Bingham et al., 2007ab). The extent of the source-sink imbalance has been found to vary widely when the same barley genotype was grown at different sites and in different years (Bingham et al., 2007a). The implications of these observations for disease management in barley are two-fold. Firstly, protection is required early in crop development to maximise the formation of potential grain sites and hence sink capacity. This is consistent with the results of fungicide timing experiments where significant yield responses are often found to applications made during early stem extension (Carver and Griffiths, 1982; Lim and Gaunt 1986; Conry and Dunne 1993). Secondly, agronomic treatments and environmental factors that alter the growth of the canopy relative to the number of developing ears and grains would be predicted to influence the yield response to fungicide. For example, it has been suggested that crops with large canopies and/or large storage reserves relative to grain numbers would be more tolerant of post-anthesis loss of leaf function to disease, and hence less responsive to fungicide, than those whose source and sink capacities are more closely balanced (Gaunt & Wright, 1992; Bingham et al., 2009). This is because in the former case there could be sufficient assimilate available to meet the demands for grain filling in spite of the loss of functional leaf area.

The objectives of experiments reported in this section were to investigate whether altering the structure and growth of the crop canopy using combinations of seed rate and N timing treatments influences its response to fungicide, and to determine whether these effects are associated with changes in the amount of visible disease or the physiological state of the crop. The combination of treatments was designed to vary the relationship between canopy size, canopy duration and grain number production within the range found in commercial practice.

3.1.2 Materials and Methods

Sites and experiment design

A two-row winter barley variety (*Hordeum vulgare* L. cv Haka) was sown in 2004 and 2005 at each of two sites, ADAS Rosemaund and SAC Aberdeen to give crops harvested in 2005 and 2006 respectively. The fields occupied a rotational position that was representative for barley production in the region, and were selected to give a high risk of *Rhynchosporium secalis.* Sowing dates were 23 and 26 September at Rosemaund and 27 September and 1 October at Aberdeen in 2005 and 2006 respectively. The variety Haka was selected for its relatively high susceptibility to *R. secalis* but good resistance to powdery mildew. The same source of seed was used for both sites. Fertilizer P and K were applied in autumn according to soil mineral analysis and anticipated crop demand. Micronutrients, molluscicides, herbicides, insecticides and growth regulators were applied to all plots, as per standard farm practice.

The experimental design was a randomised complete block with three replicate plots per treatment. Treatments consisted of a factorial combination of four nitrogen levels, two seed rates and two fungicide levels. The nitrogen treatments were made up of different combinations of application rate and timing (Table 1). The full recommended N fertilizer requirement was calculated on the basis of previous cropping and an analysis of soil mineral N (to 60-90 cm depth depending on soil conditions) made between December and February according to local practice. N treatments then ranged from 2/3 to 1 1/3 times the full recommended rate and were applied as a split dressing of ammonium nitrate. The first split was targeted at mid-tillering (GS 23/24) to promote tiller production and the second split 4-6 weeks later after the start of stem extension (GS 31/32) to promote retention of green leaf area post-anthesis. The long time period between the first and second split was intended to generate a large contrast in canopy growth and duration. At Aberdeen in both 2005 and 2006 the crop progressed rapidly from GS 32 to GS 39 and thus the second split was a little later than intended based on growth stage.

The seed rates were 150 and 300 seeds m^{-2} at ADAS Rosemaund and 150 and 350 seeds m^{-2} at SAC Aberdeen, for the ‘low’ and the ‘high’ seed rates, respectively. The ‘high’ seed rate is in fact the standard rate used for commercial

barley production in the region, but is referred to here as 'high' to distinguish it from the lower seed rate. A greater 'high' seed rate was used in Aberdeen compared to Rosemaund to offset the potentially lower tiller production and greater overwinter plant losses in the north.

Table 1. N treatments, application dates and growth stages (GS) at Aberdeen (Ab) and Rosemaund (Rm)

	Fraction of full recommended rate				Full rate, kg N
Treatment	1st Split		2nd Split		
HH	2/3		2/3		
HL	2/3		1/3		
LH	1/3		2/3		
LL	1/3		1/3		
	Date	GS	Date	GS	
Ab 2005	23-Mar	23/24	12-May	37	165
Ab 2006	05-Apr	24	16-May	37/39	165
Rm 2005	10-Mar	24	9-Apr	32	150
Rm 2006	20-Mar	25/30	28-Apr	37/39	120

The disease of interest was rhynchosporium. There were two fungicide treatment programmes: [1] untreated and [2] full fungicide treatment comprising three applications; autumn, GS 31/32 and GS 59. At each timing, the same fungicide mixture was used with the aim of giving good control of *R. secalis*: epoxiconazole + boscalid (half rate, 0.75 l ha^{-1} Tracker, BASF) plus prothioconazole + fluoxastrobin (half rate, 0.625 l ha^{-1} Fandango, Bayer) plus fenpropimorph (0.3 or 0.4 of full rate, 0.3-0.4 l ha^{-1} Corbel, BASF). All applications were made in 225 l ha^{-1} water by hand operated gas-pressured sprayer. In the case of GS 31/32 and GS 59 treatments, applications were made after the crop had been sampled for growth analysis and disease assessments. All plots were treated with quinoxyfen (full manufacturers recommended rate; 0.3 l ha^{-1} Fortress, Dow) at GS 30 to prevent powdery mildew infection, without affecting *R. secalis*.

Sampling and measurements

Disease and green area assessments

In 2005, disease was assessed at the 2-4 leaf stage, and GS 31, 59 and 71. In 2006 additional assessments were made, one at GS 39/45 and one during grain filling, GS 71 + 2 weeks. Ten plants (or ten shoots after GS39) were sampled at random from along the entire length of plots. Disease was assessed on one side of each fully emerged mainstem leaf by estimating visually the % area occupied by sporulating disease lesions, excluding the area of associated chlorosis. The latter was accounted for in a separate assessment of the % green area that considered both natural and disease-induced chlorosis and necrosis. Disease was not assessed on leaves with advanced senescence, as identification becomes unreliable, however, such leaves were recorded as being present and scored for % green area. Any disease on the stem was also recorded.

Canopy senescence assessments were begun after the final disease assessment and when the flag leaf had begun to yellow. In-field assessments of the % green area of the flag leaf and the stem were made two to three times a week at two locations per plot until complete senescence of the canopy had occurred in all plots. At each location the percentage green area was estimated visually as an average of 5-10 plants.

Photosynthetically active radiation (PAR) interception

In 2005, interception of PAR was determined at GS 31 and GS 59, between 10.00 and 14.00 hours, using a Sunscan Canopy Analysis System (Delta T Devices, Cambridge, UK). Simultaneous measurements of PAR were made above, and at ground level below the canopy. The measurements of transmitted PAR were made at 8-10 randomly selected locations along the length of the plot at an angle of approximately 45^{o} to the crop rows. In 2006, additional measurements were made at GS 39/45. Where possible measurements were made on the same day plants were sampled for disease assessment, but always within one or two days of the sampling.

Biomass and leaf area

Approximately a third to a half the plot length was designated for taking destructive quadrat samples for biomass and leaf area determination, the rest was reserved for combining. The end of the plot designated for sampling was randomised from block to block. Destructive samples were taken at GS 31 and GS 59. In 2005, plants were sampled from two 'quadrats' (0.5 m x 6 rows) located at opposite ends of the designated sampling area which were bulked and processed as one. In 2006, three smaller 'quadrats' (0.5 m x 4 rows) were sampled and pooled to better account for the spatial variability in crop growth. The 'quadrats' were positioned at least 0.5 m from the edge the plot and previous sampling areas, and more than 1.0 m from tramlines and the ends of the plot.

Plants were pulled up with their basal roots still attached, and placed into plastic bags to prevent moisture loss and taken to the laboratory for analysis. Samples were processed immediately or stored in sealed plastic bags in the dark at 4 °C to await analysis. All growth analysis was completed within five days of sampling. After washing soil from the base of shoots the tissue was gently blotted dry and weighed to the nearest 0.01 g. Plants were counted and divided into two subsamples by weight; subsample one (SS1) and subsample two (SS2). At GS 31, SS1 was 20% of the total and SS2 the remainder; at GS 59, SS1 was 10% of the total and SS2 20%. Each subsample was weighed fresh before severing the roots and reweighing. The roots were discarded and the number of potentially viable, dead and dying shoots counted. A dead or dying shoot was classed as one with no green material, or where its newest expanded leaf had begun to senesce. The potentially fertile shoots and dead and dying shoots in SS2 were weighed fresh, dried in a forced draft oven at 80^{o}C for 48 h and reweighed. The N concentration of the combined fertile and dead and dying shoots of SS2 was determined by Dumas combustion.

In 2005, the potentially fertile shoots of the SS1 subsample were divided into leaf laminae, stem plus leaf sheath and at GS59 ear (including awns) fractions and the projected area of each fraction measured using an automated leaf area meter (Delta T Devices, Cambridge, UK). Any leaf tissue that had completely senesced was measured and recorded separately. Each tissue fraction was then dried at 80^{o}C for 48 h and weighed. The area and dry weight of dead and dying

shoots was determined separately. In 2006, SS1 at GS 31 was processed as described above. At GS 59, the potentially fertile shoots were separated into zones representing each of the upper five culm leaf layers within the canopy. Thus zone 1 included the flag leaf (leaf 1) and the stem material from the base of the flag leaf up to the collar of the ear; zone 2 included leaf 2 and the stem material from the base of leaf 2 up to the base of the flag leaf etc. The bottom zone comprised leaf 5, the stem section and any senescent basal leaves below leaf 5, and the stem between leaf 5 and leaf 4. Ears plus awns were taken as a separate fraction. The projected area of stem and laminae in each zone was measured separately. In zones 1-4 the area measured included diseased, senescent and healthy tissue. In zone 5 basal senescent leaves were measured separately from leaf 5. This ensured that the stratification of the leaf area measurements corresponded with the disease and % green leaf area assessments described above. After determining the area, each fraction was dried at 80^{o}C for 48 h and weighed. The area and dry weight of dead and dying shoots was determined separately without stratification by leaf layer.

Stem water soluble carbohydrates

The concentration of water-soluble carbohydrates in stem tissue was assessed after >90% of shoots had reached GS 59. Ten shoots were sampled at random from each plot (including both designated sampling and harvest areas) between 10.00 and 13.00 hours after all other field measurements had been completed. Shoots were sealed in polythene bags and placed in a pre-chilled cool box for transport to the laboratory. Immediately on arrival the sample was processed. Leaf laminae were excised at the ligule, along with emerging leaves and ears. The remaining fraction (stem plus leaf sheaths) was then weighed fresh, placed in a gauze tray and dried rapidly at 100^{o}C in a pre-heated fan-assisted oven for at least two hours followed by a further 24 h at 80^{o}C before reweighing. The WSC concentration of the stem plus attached leaf sheath fraction was quantified on water extracts of milled tissue after hydrolysis by sulphuric acid and colourimetric determination using the anthrone reagent. The residual moisture content of the tissue was also determined on a separate sample of the tissue and used to express the WSC concentrations on a 100% dry matter basis.

Pre-harvest assessments

Ear numbers m^{-2} were determined shortly before final harvest by counting the number of ears along a 0.5 m cane placed between two rows of plants. Counts were made in the row on each side of the cane at six locations selected at random along the entire length of each plot. The outer two rows were avoided to minimise edge effects.

Grain yield and quality

At crop maturity, the area of each plot designated for yield determination was harvested using a small plot combine. A sample of grains was taken for determination of mean grain weight and moisture content.

Meteorological records

Weather data were recorded at, or within a km of, the site. Measurements were made of daily rainfall, daily maximum, minimum and mean temperature and total incident solar PAR. Incident photosynthetically active radiation was estimated as 0.5 x incident solar radiation.

Calculations and data analysis

Crop growth and yield components

Above ground biomass and canopy area index (CAI, total tissue projected area/unit ground area) were calculated from the SS2 dry weight and SS1 projected area measurements after adjusting for the sub-sampling using the subsample/total sample fresh weight ratio. CAI includes both diseased and healthy area of laminae, stem and ears and is thus distinct from the green area index (GAI) which considers only green tissue and the leaf area index (LAI) which considers only leaf laminae. The healthy area index (HAI) was calculated from the CAI for stem plus leaf sheath, leaf laminae and ear (after GS 59) adjusted by the % green area of each these fractions. In 2005 an average % green area for the whole canopy was used based on scores for the stem and top five leaves. In 2006 at GS 59, values of CAI for ears, stem and leaves in individual zones down the canopy were adjusted by their own % green area

score and summed to give the canopy HAI. The yield component grains m^{-2} was estimated as the grain yield/mean grain weight.

At GS 31 and during the latter half of grain filling some of the lower leaves had senesced completely making reliable assessment of disease difficult. Therefore, for consistency of presentation, disease severity has been summarised by averaging over the top 3 leaves at each growth stage and % green area over the top four leaves.

PAR interception

An equipment malfunction meant that fractional PAR interception data were not available for Rosemaund 2005. The analysis of PAR interception and related properties was therefore based on data from the remaining three site-years, which were: Aberdeen 2005, Aberdeen 2006 and Rosemaund 2006. At GS 31 and GS 59 a canopy light extinction coefficient (k) was calculated for each plot from measured values of fractional PAR transmission and CAI as

$$k = ln\ (I/I_0)/CAI \qquad (1)$$

where I_0 is the incident PAR and I is the PAR transmitted to the base of the canopy. The fraction of PAR intercepted (F) by healthy tissue was then estimated from measurements of HAI and k using the Beers law analogy

$$F = 1\text{-}exp\ (-k*HAI) \qquad (2)$$

Healthy area PAR interception (HAint) was then estimated in daily time steps over defined developmental periods as:

$$HAint = F*I_0 \qquad (3)$$

Interpolation pre-anthesis

Daily values of HAI were estimated from the date of 50% crop emergence to GS 31 using accumulated thermal time (base temperature 0^oC) over the period and measured values of HAI at GS 31. HAint was then calculated using the value of k at GS 31, daily HAI and incident PAR. In 2005, HAint from GS 31-59 was

calculated from *k* and HAI estimated by linear interpolation over the period. In 2006, separate estimates for GS 31-39 and GS 39–59 were made. It was assumed that lamina CAI at GS 39 was the same as that at GS 59. The stem area at GS 39 was assumed to be 60% of that at GS 59 (confirmed by measurements of stem area on plants used for disease assessments) and no contribution from ears and awns. HAI was then estimated from the stratified CAI measured at GS 59 and % green leaf area scores at GS 39. HAint was estimated assuming that *k* was the same as at GS 59.

To check the validity of the assumptions made in estimating CAI at GS 39, fractional PAR interception was calculated from the estimated values of CAI and compared with measured values at GS 39. The estimated and measured values agreed well (slope of regression 0.998, r^2 0.6; data not shown).

Interpolation between assessment dates post-anthesis

CAI and *k* were assumed to remain constant after GS 59, whilst % green area declined as a result of natural and disease-induced senescence. Independent measurements have shown this to be a reasonable assumption until late in the grain filling period when the grain approaches physiological maturity (Bingham, unpublished data). Thus HAI during grain filling was determined from disease and % green area scores at mid and late grain filling and CAI and *k* at GS 59. Linear interpolation was used to estimate daily values of HAI between individual assessment dates and the date of final canopy senescence for calculation of daily HAint. Daily values of HAint were summed to give the total for the post-anthesis period.

Radiation use efficiency and potential post-anthesis assimilate supply

Pre-anthesis radiation use efficiency (RUE) was determined for each plot as the ratio of biomass gain between GS 31 and 59 and the PAR intercepted by healthy tissue. Potential assimilate supply (PAS) for grain filling per unit grain number was given as:

$$PAS = (HAint\ per\ grain * RUE) + WSC\ per\ grain + initial\ grain\ weight \quad (4)$$

The initial grain weight was assumed to be 3.0 mg (Scott et al. 1983; Bingham et al. 2007a) and represents the biomass of the husk forming tissues at flowering. Equation 4 assumes that post-anthesis RUE is the same as that pre-anthesis. Whilst this is clearly a simplification, when averaged over sites and years, pre- and post-anthesis RUE of winter barley cv. Pearl did not differ significantly (Bingham et al. 2007a).

Statistical analysis

Statistical analysis of the effects of seed rate, N regime and fungicide treatment, and the interactions between them, on components of source and sink was conducted using GenStat 11.1 (VSN International Ltd, Hemel Hempstead, UK). Data were analysed by ANOVA incorporating site-years as a factor. Only main effects and first order interactions between fungicide and site-year, seed rate and N regime are presented. Data were checked for normality and homoscedasticity and transformed prior to analysis as required. For ease of interpretation back-transformed mean values for treatments are presented, but transformed values of SEDs. Simple linear regression and regression with groups were used to analyse relationships between source and sink components and the significance of differences between slopes and intercepts.

3.1.3 Results

Disease

The main disease at each site was rhynchosporium leaf scald (*Rhynchosporium secalis*). Only negligible levels of powdery mildew (*Bulmeria graminis* f. sp. *hordei*) and brown spotting were observed. As the severity of spotting was so low, its underlying cause (physiological spotting or symptoms of infection by *Ramularia collo-cygni*) was not diagnosed. There were significant differences ($P<0.001$) between sites and years in the severity of rhynchosporium found on the top three leaves. Severity tended to be greater in 2005 than 2006, especially at ear emergence (GS 59) and during grain filling (GS 71) and greater at Aberdeen than Rosemaund (Fig. 1). The greater disease severity in 2005 compared to 2006 was associated with a larger reduction in % green leaf area

(GLA) on the top four leaves (reduction of 40-50 % in relative GLA at GS 71 in 2005 and only 20-30 % in 2006). The relative GLA is that of non-fungicide treated plots expressed relative to that of fungicide treated plots after the latter has been normalized to 100%. Expressing the data on a relative basis allows the effects of disease (or more correctly lack of fungicide treatment) on green leaf area to be separated from those of natural senescence observed in fungicide-treated control leaves. The relative % GLA was comparable between sites in spite of a lower disease severity at GS 71 in Rosemaund in 2005 compared with Aberdeen.

Seed rate had relatively little effect on the severity of rhynchosporium. At GS 31, when averaged across site-years and N treatments disease severity was significantly greater (21%) at high seed rate compared to low, but there was no significant difference at the later growth stages (Table 2). Nor was there any interaction between seed rate and site-year on disease severity. By contrast, N treatment had a highly significant effect ($P<0.001$) on the severity of rhynchosporium at ear emergence and during grain filling. Disease severity tended to be greater in the high early N treatments (HH and HL) and was least at low N (treatment LL, Table 2). The effect of N treatment on disease severity was partly dependent on the site, as shown by the significant site-year x N interaction at GS 59. At this growth stage, the effects of high early N were only observed at Rosemaund and not Aberdeen. Effects of N treatment on disease severity were more consistent across sites (no significant SY x N interaction) when disease was measured during grain filling.

Yield and yield components

When averaged across N and seed rate treatments, the yield of fungicide-treated crops differed widely between sites and years (Table 3). Yields were consistently greater at Rosemaund compared to Aberdeen. At Aberdeen yields were greater in 2005 than 2006, whereas at Rosemaund the reverse was the case. The greater yield at Rosemaund compared to Aberdeen was the result of the production of a much larger (39-92% depending on the year) number of grains m^{-2}, offset to some extent by a smaller (8-15%) mean grain weight (MGW). Application of fungicide gave a significant yield response in each site-year and was associated with increases in both grain number m^{-2} and MGW (18 and 8%

respectively, averaged over site-years). The magnitude of the response of yield and its components to fungicide differed between site-years as indicated by a significant site-year x fungicide interaction (Table 3).

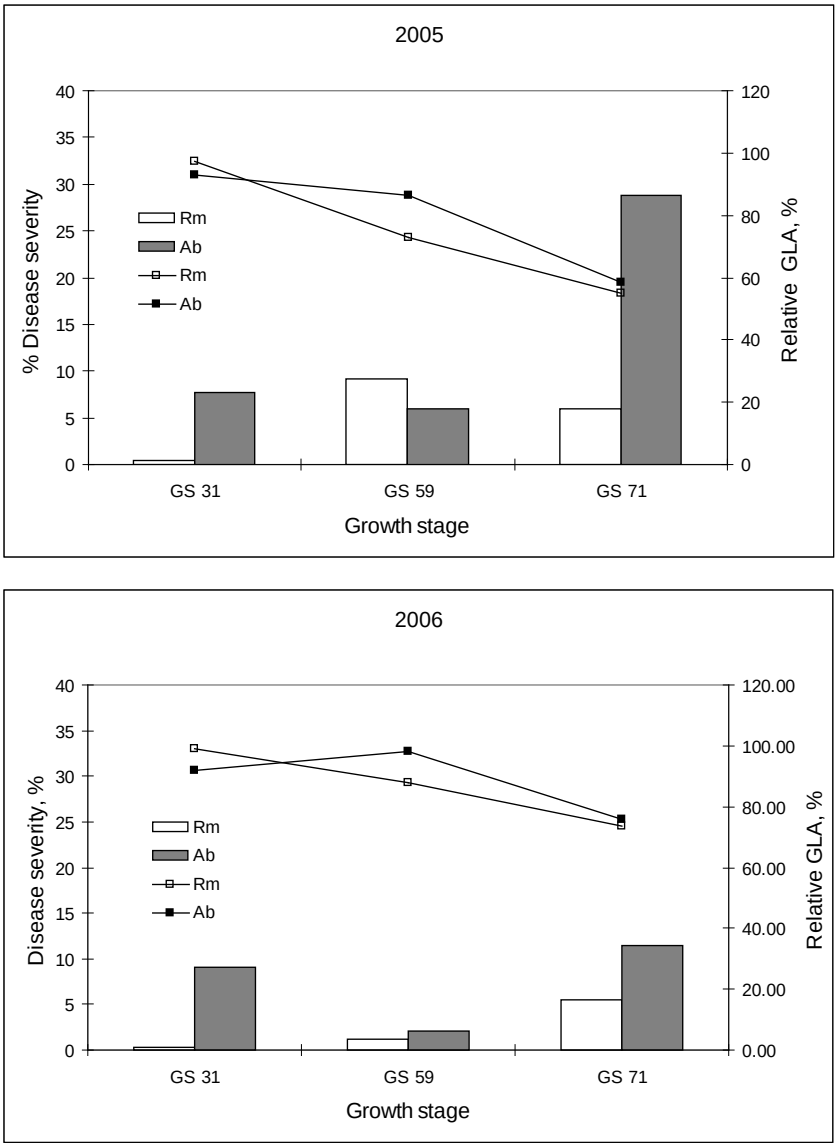


Figure 1. Severity of rhynchosporium infection and relative % green leaf area (GLA) of non-fungicide treated plots in 2005 and 2006 averaged across seed rate and N treatments. Rm = ADAS Rosemaund, Ab = SAC Aberdeen.

Disease severity (bars) is expressed as the % surface area covered by lesions averaged for the top three leaves. Relative % GLA (symbols and lines) is the average % green leaf area for the top 4 leaves of untreated plots expressed relative to that of fungicide treated plots (values for fungicide treated plots normalized to 100%). Raw % data were converted to fractions and arcsine

transformed prior to analysis. Values shown are back-transformed main effect means for each site-year combination. SEDs (transformed values, 8 df) for disease severity are: GS 31, 0.0124; GS 59, 0.0246; GS 71, 0.0340.

Table 2. Severity of rhynchosporium infection (averaged for top 3 leaves) in non-fungicide treated plots as influenced by agronomic treatments. Percentage data were converted to fractions and arcsine transformed prior to analysis. Values shown are back-transformed main effect means for seed rate and N and means for the site-year (SY) x N interaction at GS 59. SED values are the transformed values. Significance levels are ***, P<0.001; ** P<0.01; ns, P>0.05. SEDs and degrees of freedom (df) for interactions involving SY are for comparison of N or SR treatments within the same level of SY. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

		Growth stage		
Factor	Treatment	GS 31	GS 59	GS 71
Seed rate	Low	2.8	3.9	12.1
	High	3.4	4.1	11.3
N	HH	2.9	5.4	14.2
	HL	3.5	4.5	13.8
	LH	2.8	3.4	10.4
	LL	3.2	3.0	8.7
	df	SED; significance		
Seed rate (SR)	56	0.0060; **	0.0068; ns	0.0156; ns
Nitrogen (N)	56	0.0085; ns	0.0095; ***	0.0221; ***
SY*SR	56	0.0120; ns	0.0135; ns	0.0313; ns
SY*N	56	0.0169; ns	0.0191; ***	0.0442; ns
SR*N	56	0.0120; ns	0.0135; ns	0.0313; ns
	GS 59			
SY*N	HH	HL	LH	LL
Ab 2005	6.5	5.7	5.1	6.3
Ab 2006	2.4	2.0	1.8	2.0
Rm 2005	13.1	11.0	8.6	5.1
Rm 2006	2.4	1.7	0.6	0.6

Table 3. Yield and yield components of fungicide treated and untreated plots. Values are main factor means and their interaction with fungicide. Response refers to the increase in yield or its components resulting from fungicide treatment. Significance levels; *** (P<0.001); ** (P<0.01); * (P<0.05); ns (P>0.05). Ab = SAC Aberdeen, Rm = ADAS Rosemaund..

		Yield, t ha^{-1} @ 100% DM			Grains m^{-2}			MGW, mg @100% DM		
		Untreated	Treated	Response	Untreated	Treated	Response	Untreated	Treated	Response
Site-year (SY)	Ab 2005	4.7	6.2	1.6	9251	11265	2014	50.8	55.5	4.7
	Ab 2006	3.9	4.9	0.9	7745	9172	1427	50.7	52.8	2.1
	Rm 2005	5.4	7.4	2.0	12851	15678	2827	41.7	47.1	5.4
	Rm 2006	7.3	8.6	1.3	15812	17633	1821	46.4	48.8	2.5
Seed rate (SR)	Low	5.3	6.6	1.3	11166	12893	1727	48.0	51.5	3.5
	High	5.4	7.0	1.6	11663	13981	2318	46.8	50.6	3.8
Nitrogen (N)	HH	5.5	7.1	1.6	11732	14101	2369	47.5	51.2	3.7
	HL	5.5	6.8	1.3	11830	13274	1444	47.4	52.0	4.6
	LH	5.2	6.8	1.6	11142	13596	2454	47.1	50.4	3.4
	LL	5.1	6.4	1.3	10954	12777	1823	47.5	50.6	3.1
Fungicide (F)		5.3	6.8	1.4	11415	13437	2022	47.4	51.1	3.7
		df	SED	signif	df	SED	signif	df	SED	signif
Main effects	SY	8	0.28	***	8	531	***	8	0.58	***
	SR	118	0.06	***	118	122	***	120	0.22	***
	N	118	0.08	***	118	172	***	120	0.30	*
	F	118	0.06	***	118	122	***	120	0.22	***
Interactions	SY*F	118	0.12	***	118	243	***	120	0.43	***
	SR*F	118	0.08	*	118	172	*	120	0.30	ns
	N*F	118	0.12	p = 0.056	118	243	*	120	0.43	ns

Table 4. Pre-harvest ear numbers in fungicide- treated and untreated plots. Values are main factor means and their interaction with fungicide. Significance levels; *** (P<0.001); ** (P<0.01); * (P<0.05); ns (P>0.05). Ab = SAC Aberdeen, Rm = ADAS Rosemaund..

		Final ear no. m^{-2}	
		Untreated	Treated
Site-year (SY)	Ab 2005	636	704
	Ab 2006	425	490
	Rm 2005	961	1043
	Rm 2006	780	839
Seed rate (SR)	Low	679	718
	High	722	820
Nitrogen (N)	HH	728	810
	HL	692	737
	LH	692	774
	LL	692	755
Fungicide (F)		701	769

		df	SED	signif
Main effects	SY	8	38	***
	SR	118	13	***
	N	118	18	*
	F	118	13	***
Interactions	SY*F	118	26	ns
	SR*F	118	18	*
	N*F	118	26	ns

Low seed rate significantly reduced yield and grain number m^{-2}, but increased the MGW relative to the high seed rate. Nitrogen rate and timing also had a significant effect on yield and its components. Importantly, there were significant seed rate x fungicide and N x fungicide interactions indicating that when averaged over site-years the response to fungicide was modified by seed rate and N treatment. The yield response was increased by a further 0.3 t ha^{-1} at high seed rate compared to low, and by the same margin with late N (HH and LH) compared to early or low N (HL and LL). The additional yield was associated entirely with an increase in grain number m^{-2}. There was no significant seed rate x fungicide or N x fungicide interaction on MGW (Table 3). The interaction between seed rate and N with fungicide on yield appears to be consistent between sites and years, because the 3-way interaction between these treatment combinations and site-year was not statistically significant.

Final ear population densities were determined just prior to harvest. Ear numbers were increased by fungicide treatment at each site and in each seed rate and N treatment (Table 4). There was a significant interaction between seed rate and fungicide, with the increase in ear number with fungicide treatment at high seed rate more than double that at low seed rate.

Pre-anthesis growth of source and sink components

Pre-anthesis canopy growth differed markedly between sites. Above-ground biomass at GS 59 at Rosemaund was almost double that at Aberdeen in 2005 and 2006 (Table 5). This was associated with a greater healthy canopy area (HAI) and larger fertile shoot number at Rosemaund compared with Aberdeen. Seed rate and N treatments had contrasting effects on canopy growth. Thus, when averaged across site-years and other treatments, there was a significantly larger biomass at high seed rate compared to low, but no difference in HAI. By contrast, N regime had no significant effect on canopy biomass, but HAI was significantly greater at high N (HH) compared to low (LL).

Biomass and HAI were increased by fungicide application. The increase in HAI was the result of both an increase in shoot number and healthy area per shoot (Table 5). There was a significant interaction between seed rate and fungicide on biomass, but none of the other components. However, as with biomass the

response of HAI and fertile shoot number to fungicide was consistently greater at high seed rate compared to low.

There was no interaction between N regime and fungicide for any of the canopy components. The concentration of water soluble carbohydrates in stem tissue differed widely (> 2 fold range) between sites and years and this was accompanied by large differences in the amount of WSC per m^{-2} of ground area and WSC per final grain number (Table 6). By contrast the effects of seed rate and N treatments on WSC were small. High seed rate increased the concentration of WSC in stem tissue by 10%, the amount m^{-2} by 16% and the amount per unit grain number by 20% compared to low seed rate. The concentration of WSC in tissue was reduced by high fertilizer N application (HH) compared to low (LL), but the total quantity and amount per grain number were unchanged (Table 6). Fungicide application had no significant overall effect on the concentration of WSC in tissue, although in one site-year (Aberdeen in 2005) the concentration was increased significantly. However, fungicide consistently increased the total quantity of WSC as a result of its effects on shoot numbers, but had no effect on the amount of WSC available per grain. There was no seed rate or N x fungicide interaction indicating that the response of WSCs to fungicide was not modified by seed rate or N regime.

The HA per unit grain number provides an index of the healthy surface area available for photosynthesis at the start of the grain filling period relative to the number of grains that need to be supplied with photosynthate. The ratio varied significantly between site-years and with N regime. The ratio was greatest with high N (HH) compared to low N (LL). Fungicide application increased the ratio by 19% ($P<0.001$) indicating that the effects of fungicide on healthy canopy area exceeded those on the formation of grain numbers.

N concentration in above-ground tissue (stem leaf sheaths and laminae combined) at ear emergence was influenced most strongly by seed rate, N regime and fungicide application. The concentration was greatest at low seed rate compared to high, and by high (HH) and late (LH) N applications compared to low (LL) and early (HL) applications (Table 7). The concentration was also reduced by fungicide treatment. However, when total N offtake was considered (the product of N concentration and above-ground biomass), only the effects of

N regime were significant. Thus the effects of fungicide and seed rate on tissue N concentrations were offset by those on biomass.

Relationship between pre-anthesis light interception and grain number formation

The relationship between final grain number and PAR interception by healthy tissue was not consistent between sites and years if PAR interception was just considered between crop emergence and GS 31 or GS 31 and GS 59 (data not shown). Only when the entire period from crop emergence to GS 59 was included in the analysis was the relationship consistent over sites and years; here a single regression model accounted for 87% of the variation in grain number (Fig. 2). This suggests that processes before and after GS 31 are important in determining final grain number and that the relative importance of each of these periods differs between years and sites. The entire period from crop emergence to GS 59 was, therefore, included in the analysis of the effects of fungicide application on the relationship between PAR interception and grain numbers.

When data for the three site-years, seed rate and N treatments were pooled and separate lines fitted by linear regression to data for fungicide-treated and untreated crops, the slopes were found to differ significantly ($P<0.05$), but not the intercepts (Fig. 3). The effect of seed rate on the response to fungicide was analysed by fitting separate models to different combinations of seed rate and fungicide treatment. The slope and intercepts did not differ between high and low seed rate in the absence of fungicide nor between fungicide-treated and untreated crops at low seed rate. Only at high seed rate did fungicide treatment increase the slope and reduce the intercept significantly compared to the other treatment combinations (Table 8). A similar analysis on the effects of N regime on the response to fungicide revealed no significant difference in slope or intercept between the different fungicide-N regime combinations.

Table 5. Canopy components at ear emergence (GS 59). Values are main factor means and their interaction with fungicide. Significance levels; *** (P<0.001); ** (P<0.01); * (P<0.05); ns (P>0.05). Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

			Biomass, g DW m^{-2}		[i]HAI		Fertile shoots m^{-2}		[ii]HA, cm^2 $shoot^{-1}$	
			Untreated	Treated	Untreated	Treated	Untreated	Treated	Untreated	Treated
Site-year (SY)	Ab 2005		524	619	3.0	3.8	560	617	52.8	61.5
	Ab 2006		571	684	2.1	2.7	451	497	45.7	53.3
	Rm 2005		1111	1267	6.2	9.1	920	978	68.0	94.5
	Rm 2006		931	1019	5.6	7.1	842	894	66.2	80.0
Seed rate (SR)	Low		771	838	4.2	5.5	646	682	61.4	76.5
	High		796	957	4.2	5.8	740	811	54.9	68.2
Nitrogen (N)	HH		816	942	4.7	6.3	722	784	62.8	77.8
	HL		773	862	4.3	5.5	658	719	62.1	73.7
	LH		799	869	4.1	5.5	725	757	54.6	69.7
	LL		748	916	3.7	5.3	668	725	53.1	68.1
Fungicide (F)			784	897	4.2	5.7	693	746	58.2	72.3
		df	SED	signif	SED	signif	SED	signif	SED	signif
Main effects	SY	8	32	***	0.45	***	34	***	4.2	***
	SR	119	18	***	0.14	ns	14	***	1.1	***
	N	119	26	ns	0.2	***	20	**	1.6	***
	F	119	18	***	0.14	***	14	***	1.1	***
Interactions	SY*F	119	41	ns	0.28	***	29	ns	2.5	***
	SR*F	119	26	*	0.2	ns	20	ns	1.6	ns
	N*F	119	36	ns	0.28	ns	29	ns	2.5	ns

[i]HAI = healthy area index [ii]HA = healthy area

Table 6. Healthy area (HA) per unit grain number and stem water soluble carbohydrates (WSC) at GS 59 expressed as a % of stem DW, g m^{-2} ground area and mg per unit eventual grain number. Significance levels; *** (P<0.001); ** (P<0.01); * (P<0.05); ns (P>0.05). $ HA data were transformed (log_{10} x + 1) prior to analysis; mean values shown are after back transformation, SEDs are the transformed values. Ab = SAC Aberdeen, Rm = ADAS Rosemaund..

			WSC, % DW		WSC, g m^{-2}		WSC, mg $grain^{-1}$		$HA, cm^2 $grain^{-1}$	
			Untreated	Treated	Untreated	Treated	Untreated	Treated	Untreated	Treated
Site-year (SY)	Ab 2005		27.7	31.6	129	175	14.1	15.9	3.16	3.29
	Ab 2006		35.6	36.1	169	202	22.1	22.5	2.65	2.91
	Rm 2005		25.2	23.3	187	192	13.6	13.5	4.29	6.27
	Rm 2006		13.9	14.5	111	136	7.1	7.7	3.44	3.98
Seed rate										
(SR)	Low		24.3	25.2	137	163	13.2	13.3	3.32	3.82
	High		26.9	27.5	161	189	15.3	16.5	3.37	4.12
Nitrogen (N)	HH		23.9	24.3	152	171	15.3	14.7	3.88	4.54
	HL		25.9	27.0	151	176	13.4	14.5	3.25	3.88
	LH		24.9	26.0	144	174	13.6	14.9	3.24	3.81
	LL		27.6	28.2	150	182	14.6	15.5	3.04	3.68
Fungicide (F)			25.6	26.4	149	176	14.2	14.9	3.34	3.97
		df	SED	signif	SED	signif	SED	signif	SED	signif
Main effects	SY	8	2.5	***	16	***	1.6	***	0.0289	***
	SR	119	0.5	***	5	***	0.5	***	0.0095	ns
	N	119	0.7	***	7	ns	0.7	ns	0.0134	***
	F	119	0.5	ns	5	***	0.5	ns	0.0095	***
Interactions	SY*F	119	1.0	***	9	*	1.0	ns	0.0189	***
	SR*F	119	0.7	ns	7	ns	0.7	ns	0.0134	ns
	N*F	119	1.0	ns	9	ns	1.0	ns	0.0189	ns

Table 7. Concentration of N in above-ground biomass and total canopy N offtake at ear emergence (GS 59). Significance levels; *** (P<0.001); ** (P<0.01); * (P<0.05); ns (P>0.05). Degrees of freedom (df) and SED for the SY x F interaction are for comparison of means within the same level of SY. Ab = SAC Aberdeen, Rm = ADAS Rosemaund..

			N, % DW		N offtake, g m^{-2}	
			No Fungicide	Fungicide Treated	No Fungicide	Fungicide Treated
Site-year (SY)	Ab 2005		2.20	1.89	11.5	11.7
	Ab 2006		1.92	1.81	12.3	11.0
	Rm 2006		1.96	1.84	18.4	18.6
Seed rate (SR)	Low		2.11	1.89	13.9	13.1
	High		1.94	1.80	14.2	14.4
Nitrogen (N)	HH		2.38	2.12	17.2	15.7
	HL		1.96	1.80	12.8	12.5
	LH		2.10	1.94	15.3	14.8
	LL		1.68	1.51	11.2	12.0
Fungicide (F)			2.03	1.85	14.1	13.8
		df	SED	signif	SED	signif
Main effects	SY	6	0.053	*	0.68	***
	SR	90	0.029	***	0.47	ns
	N	90	0.041	***	0.67	***
	F	90	0.029	***	0.47	ns
Interactions	SY*F	90	0.050	**	0.82	ns
	SR*F	90	0.041	ns	0.67	ns
	N*F	90	0.058	ns	0.94	ns

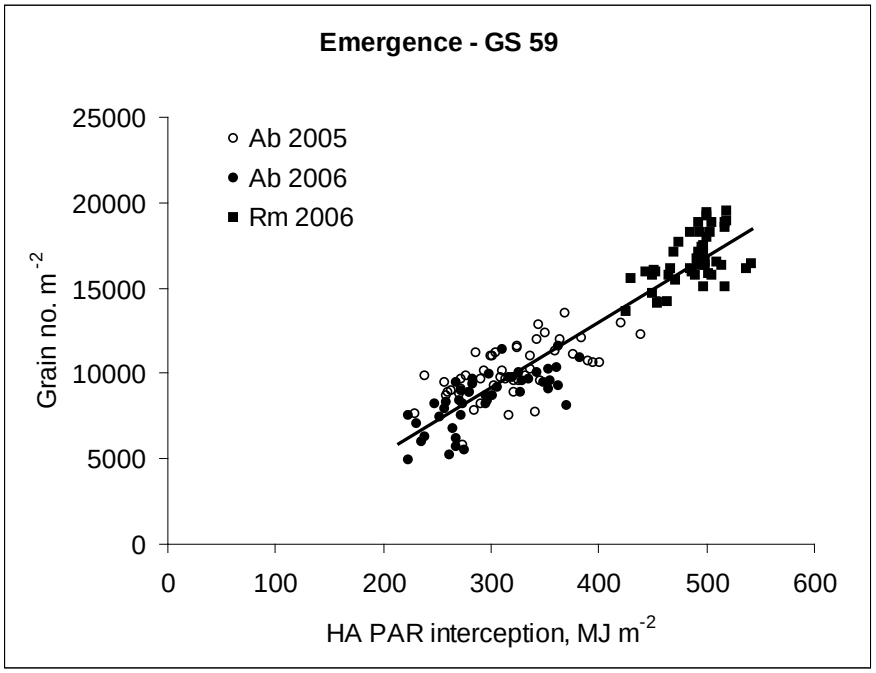


Figure 2. Relationship between PAR interception by healthy tissue between crop emergence and GS 59 and final grain number for three site-years where PAR data were available. Line fitted by regression ($y = -2252 + 38.16x$, $r^2 = 0.88$, $P<0.001$). Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

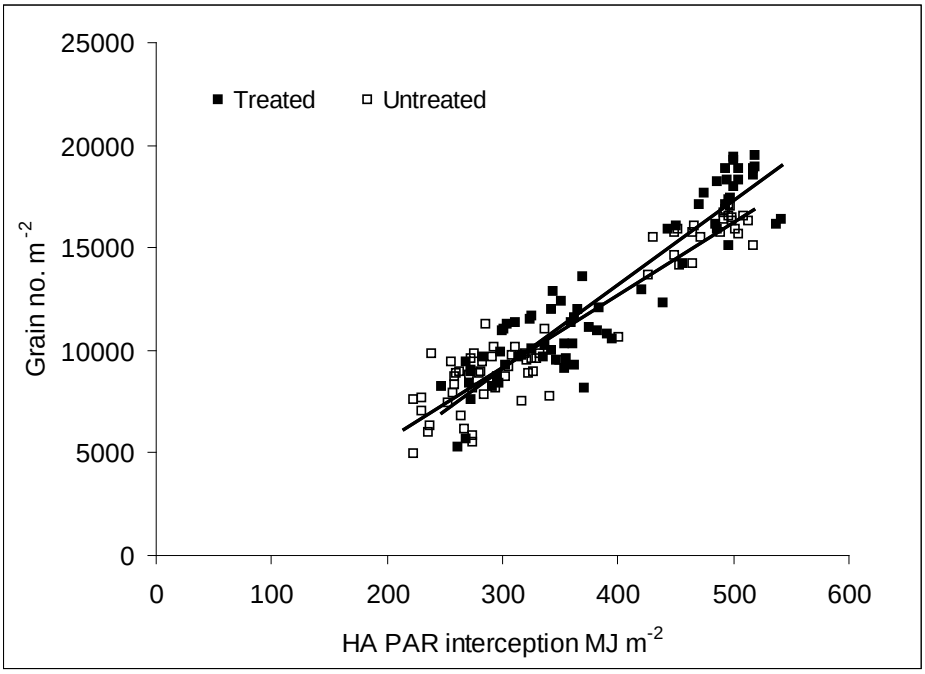


Figure 3. Relationship between PAR interception by healthy tissue from crop emergence to GS 59 and final grain number. Data for site-years, seed

rate and N treatments have been pooled. Separate lines fitted by linear regression for fungicide treated ($y = –3187 + 40.95x$, $r^2 = 0.86$) and untreated ($y = –1415 + 35.36x$, $r^2 = 0.90$) crops. Slopes differed significantly at $P<0.05$, but not intercepts.

Table 8. Values of slope and intercept for linear regression models (y = a + bx) relating final grain number m^{-2} (y) to pre-anthesis healthy area PAR interception (x). Separate models were fitted to data for seed rate and fungicide treatment combinations and the slopes and intercepts compared to the reference treatment Low Untreated. Significance ** (P<0.01); ns (P>0.05).

Seed rate	Fungicide	Slope (b)		Intercept (a)	
Low	Untreated	35.15		-1227	
Low	Treated	38.00		-1888	
High	Untreated	36.07		-1796	
High	Treated	47.92	**	-6265	**

Post-anthesis PAR interception and potential assimilate supply for grain filling

Post-anthesis PAR interception by healthy tissue was greater at Rosemaund 2006 than Aberdeen in 2005 and 2006 (Table 9), as a result of the greater canopy size and incident radiation at Rosemaund. Seed rate had no significant effect on PAR interception by healthy tissue, but interception was increased by high N (HH) compared to lower N (LH, HL and LL) due to differences in the size of the healthy canopy and its duration post-anthesis. Fungicide treatment also increased PAR interception by increasing healthy canopy area at GS 59 and prolonging green area duration post anthesis. However, the effects of fungicide on grain number were greater than those on post-anthesis PAR interception, hence when PAR interception by healthy tissue is expressed per unit grain number, interception was greater in untreated compared to treated plots (data not shown).

Figure 1.4 presents a theoretical analysis of the effects of the source-sink balance of the crop on the relationship between assimilate availability for grain filling and mean grain weight (MGW). If grain filling of a diseased crop is source-

limited (limited by the availability of assimilate) then an increase in assimilate supply in response to fungicide treatment should lead to an equivalent increase in MGW (line A-C). If however, the increase in supply exceeds the spare capacity for storage, the MGW will increase initially until the storage capacity is met (line A-B) and then remain constant as the crop moves from a state of source-limitation to one of sink-limitation (line B-E). If, on the other hand, grain filling of the diseased crop is sink-limited (i.e. line D-E; potential assimilate supply exceeds storage capacity) an increase in MGW with fungicide treatment would indicate an increase in storage capacity (sink capacity) per grain (broken line, F-G).

To investigate the physiological basis for the effects of fungicide treatment on MGW, the potential assimilate supply per grain during the post-anthesis period was estimated for each seed rate-N regime combination and plotted against the actual MGW. The potential supply was calculated according to equation 4 above. Estimates of RUE varied widely between individual plots because they were based on just two biomass assessments, one at GS 31 and the other at GS 59. A cross site-year anova indicated no significant effect of seed rate and N regime on PAR use efficiency, nor any interaction of these treatments with site-year. There was a significant fungicide effect ($P = 0.04$), with PAR use efficiency being increased by fungicide treatment. Thus, for estimation of potential assimilate supply, average values of PAR use efficiency for fungicide treated and untreated plots in each site-year were used.

The seed rate and N regime treatments resulted in a range of potential assimilate supply per grain in each of the site-years (Fig. 5). The exceptionally high values in some plots at Aberdeen 2006 were the result of unusually low grain numbers in these treatments. In general, estimated potential assimilate supply exceeded measured grain weight at Aberdeen in 2005 and 2006. Although there was a significant relationship between potential assimilate supply and MGW in Aberdeen 2005 ($P < 0.05$) the amount of variation in MGW accounted for was small ($r^2 < 0.25$). At Aberdeen in 2006 there was no significant relationship. Importantly in both years, fungicide treatment increased MGW over a wide range of potential supply (Fig. 5), which is consistent with fungicides increasing the storage capacity of grain (i.e. the potential grain size).

Interpretation of the effects of fungicide on MGW at Rosemaund in 2006 is more

difficult. Parallel regression analysis indicated no significant difference in slope and intercept when separate lines were fitted to data from fungicide-treated and untreated plots. However, when a common slope was used, there was a trend for intercepts to differ (P = 0.07; data not shown).

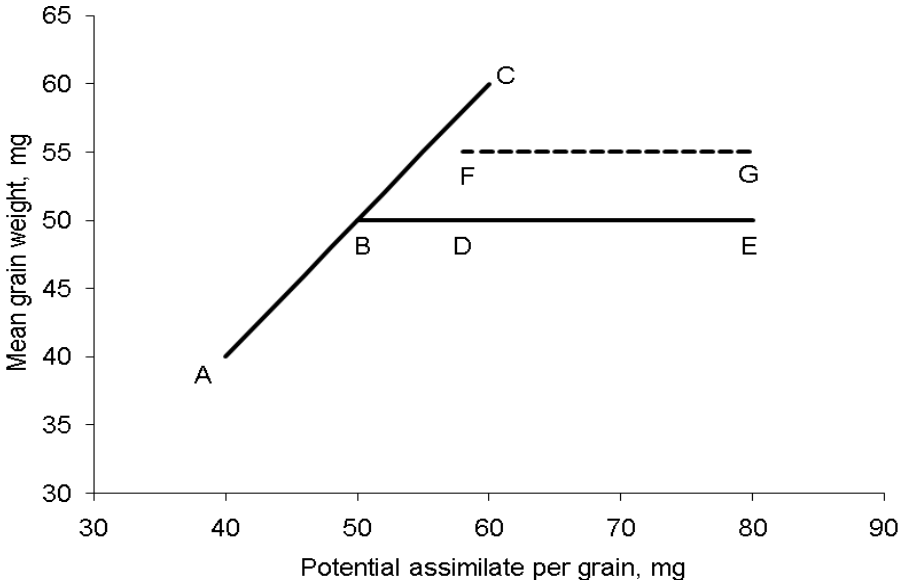


Figure 4. Theoretical relationships between potential assimilate supply per grain post-anthesis and mean grain weight as affected by the source-sink balance of the crop. See text (page 42) for a full explanation.

Table 9. Post-anthesis PAR interception by healthy tissue and pre-anthesis RUE

		PAR interception		RUE, g DW MJ^{-1} PAR	
		Untreated	Treated	Untreated	Treated
Site-year (SY)	Ab 2005	211	248	1.81	1.91
	Ab 2006	179	197	2.24	2.33
	Rm 2006	267	299	2.01	2.27
Seed rate (SR)	Low	219	249	2.03	2.14
	High	220	247	2.01	2.20
Nitrogen (N)	HH	237	276	2.09	2.16
	HL	225	247	1.95	2.17
	LH	215	252	2.03	2.08
	LL	200	218	2.01	2.25
Fungicide (F)		219	248	2.02	2.17

		df	SED	signif	SED	signif
Main effects	SY	6	9.3	***	0.20	ns
	SR	90	1.9	ns	0.07	ns
	N	90	2.7	***	0.10	ns
	F	90	1.9	***	0.07	*
Interactions	SY*F	90	3.3	***	0.12	ns
	SR*F	90	2.7	ns	0.10	ns
	N*F	90	3.8	***	0.14	ns

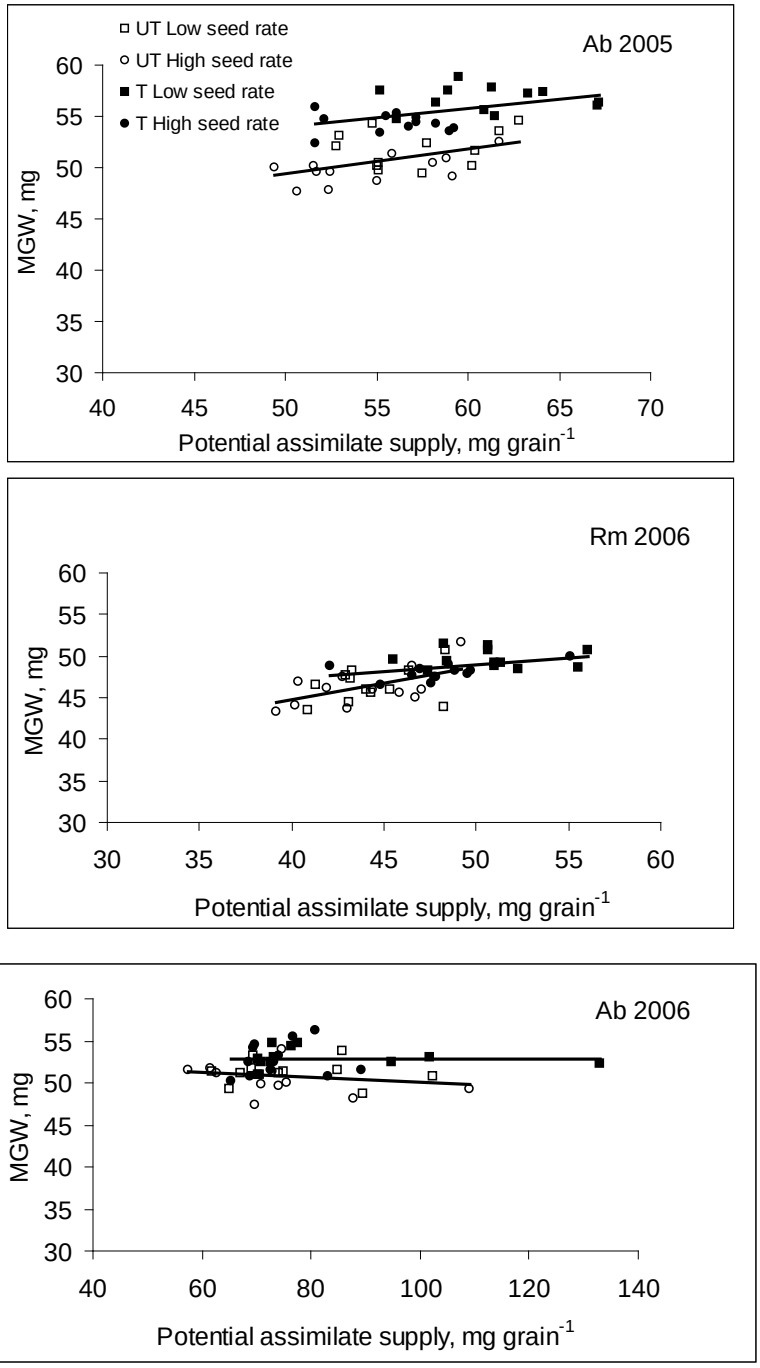


Figure 5. Relationship between estimated potential assimilate supply per grain and actual mean grain weight (MGW) for three site-years. Legend for all site-years is given in the top, Ab 2005 graph. Each point represents an individual plot. Lines fitted by linear regression for fungicide treated (T) and untreated (UT) plots: Ab 2005 (UT, r^2 0.24, P = 0.016; T, r^2 0.20 P = 0.027); Ab 2006 (UT, r^2 0.05, P = 0.322; T, r^2 <0.01, P = 0.995); Rm 2006 (UT, r^2 0.26, P = 0.011; T, r^2 0.16, P = 0.05). Note different scales used on x axis.

3.1.4 Discussion

The combination of seed rate and N treatments was designed to vary the growth and duration of source structures relative to the growth of the yield bearing structures in order to investigate their effects on yield response to fungicide. A wide range of measures demonstrates that this variation in growth was achieved. The fungicide programme used in these experiments also influenced the growth of both source and sink structures. In the absence of fungicide, the healthy canopy area (HAI), the quantity of stem WSCs at flowering, and post-anthesis PAR interception were reduced by disease. These effects were associated with a reduction in yield and its components; ears m^{-2}, grains m^{-2} and MGW. The effects of fungicide observed here are consistent with previous reports of the impact of early and persistent disease epidemics on the growth of the canopy, deposition of storage reserves, formation of grain numbers, post-anthesis canopy duration and MGW of barley (Carver and Griffiths, 1982; Lim and Gaunt 1986; Gaunt and Wright 1992).

However, the current experiments show that the yield response to fungicide is influenced by seed rate and N regime, with fungicide resulting in an additional 0.3 t ha^{-1} at high seed rate compared to low and with high and late N compared to early or low N. Further, the additional yield response was almost entirely associated with an increase in grain number m^{-2}.

To what extent can the effects of these agronomic treatments on the response of grain numbers to fungicide be explained by changes in disease severity or variation in disease tolerance with agronomy? Higher rates of fertiliser N are known to increase the severity of a range of foliar diseases and in some cases this may be related to increases in leaf N concentrations (Leitch and Jenkins 1995; Neumann et al., 2004; Walters and Bingham, 2007). In the current work, high N applications (HH) resulted in relatively high concentrations of N in the tissue and the greatest severity of rhynchosporium in the upper canopy at flowering and during grain filling. This was associated with a large grain number response to fungicide. However, whilst early N (HL) resulted in significantly more disease than late N (LH), of the two it was late N that gave the greater grain number and yield response to fungicide. Thus, the interaction between N and fungicide treatments on grain numbers cannot be explained entirely by effects of

the N regime on disease severity. A similar argument applies to the effects of seed rate. The grain number response to fungicide was greater at high seed rate compared to low, yet seed rate had little effect on the severity of visible disease after GS 31. Even at GS 31, the differences in severity were small.

Fungicides and grain number m^{-2}

Final grain number is a function of the numbers of tillers and spikelets that are produced and survive to form ears and fertile florets respectively, and the number of florets that are successfully fertilized. Shading experiments have shown that both ear number and the number of grains per ear of barley are sensitive to variations in PAR interception by the canopy, but that the scale of the effect and yield component most affected depends on the timing of shading (Grasshoff and dAntuono, 1997; Arisnabarreta and Miralles, 2008). Shading just 10 days before ear emergence had the greatest effect on grain numbers per ear, whilst shading at the end of tiller production and early stem extension was as effective in reducing ear number as shading during booting (Arisnabarreta and Miralles, 2008). There is also evidence that shading from ear emergence to the end of grain filling can reduce final ear numbers (Grasshoff and dAntuono, 1997).

In the current study, grain number m^{-2} was linearly related to PAR interception by healthy tissue across sites and years when the period from crop emergence to ear emergence was considered. When just the pre-stem extension phase, or the period from the start of stem extension to ear emergence was considered, a single relationship did not explain the variation between sites and years. This suggests that developmental events influencing grain number both before GS 31 (i.e. tiller production and spikelet initiation) and after GS 31 (tiller and spikelet survival) were sensitive to variation in PAR interception in this study.

At low seed rate the increase in grain number m^{-2} with fungicide treatment can be explained in terms of an increase in healthy area PAR interception pre-anthesis, since there was no significant difference in the regression models fitted to data for untreated and treated plots. At high seed rate, the increase in grain number with fungicide was not associated with an equivalent increase in PAR interception. Rather, there was an increase in the apparent sensitivity of grain number formation to PAR interception, with a 30% increase in the number of

grains produced per MJ intercepted. The interaction of seed rate and fungicide found on grain numbers was also observed with final ear number suggesting that fungicide treatment leads to a relatively greater production and/or survival of tillers at high seed rate compared to low. It is conceivable that fungicide treatment increased assimilate partitioning to young tillers favouring their survival and that survival was more pronounced at high seed rate than low. This could arise if there is greater competition between shoots for light in a dense canopy leading to a greater dependence of young tillers on the main shoot for assimilates. The increase in shoot number with fungicide treatment at high seed rate did not lead to a proportional increase in PAR interception because the healthy area per shoot was smaller and because the relationship between HAI of the canopy and PAR interception is non-linear (Hirose 2005).

In contrast to the effects of seed rate on the crop's response to fungicide, the interaction between N regime and fungicide on grain number could not be explained through changes in the sensitivity of grain site formation to intercepted PAR. Nor was it associated with effects of N on total pre-anthesis PAR interception (N and fungicide interaction P = 0.77, data not shown). There was, however, a significant N and fungicide interaction on post-anthesis PAR interception by healthy area in which the greatest increase in interception with fungicide application occurred in the high and late N treatments (HH and LH). Moreover, there was no significant difference between regression models fitted to data for post-anthesis PAR interception and grains m^{-2} implying that the differences in grain number between N regimes was associated with variation in their post-anthesis PAR interception. However, we cannot determine from the current data whether the relationship is causal or not. It is possible that fungicide treatment increased fertilisation and grain set or the survival of late developing tillers was greater under the HH and LH regimes. This might be a direct result of the greater post-anthesis PAR interception and an increased assimilate supply to the ears, but we cannot rule out the possibility of some independent mechanism. It has been suggested that grain number per ear is influenced by the N concentration in the ear at flowering (Abatte et al., 1995; Sinclair and Jamieson, 2006). Although the high and late N treatments increased total N concentration in the canopy compared to the other regimes, fungicide reduced the concentration to the same extent in each case. Unless fungicide altered the relative partitioning of N between canopy and ear differentially under

each N regime, variation in N concentration of the ear at flowering is unlikely to explain the observed N x fungicide interaction on grain number.

It is also worth highlighting again that early N (HL) resulted in significantly more disease than late N (LH), yet fungicide increased post-anthesis PAR interception more in the late N regime. It would appear that late N applications and higher canopy N concentrations at flowering increase the effects of fungicides on green area retention post-anthesis. Delayed leaf senescence and increased canopy duration has been associated with the strobilurin group of fungicides (Grossmann and Retzlaff, 1997; Pepler et al., 2005), such as fluoxastrobin used here.

Fungicides and MGW

We hypothesised that crops with a large potential supply of assimilate for grain filling relative to the number of grains to fill would be more tolerant of post-anthesis disease than those where the source and sink are more closely balanced. Treatments used to vary canopy size also tended to vary grain numbers in the same direction, and thus the range of variation in healthy area per unit grain number at flowering was relatively small. Nevertheless, when canopy duration, post-anthesis PAR interception and stem WSC reserves were taken into account, there was a sufficiently large range of potential assimilate supply per unit grain number to test the hypothesis. No significant interaction between seed rate or N regime and fungicide treatment on MGW was found implying that crops were equally responsive to fungicide under all these treatments.

In untreated crops at Aberdeen in 2005 and Rosemaund in 2006 there was a significant linear relationship between potential assimilate supply and MGW, but the slope of the relationship was small (0.24 and 0.40 for Aberdeen and Rosemaund respectively). If MGW was simply limited by the amount of dry matter available for grain filling, an increase in supply should lead to an equivalent increase in MGW. A slope well below unity, as observed here, is consistent with a co-limitation of MGW by both source and sink (Boras et al. 2004; Bingham et al., 2007ab). There is evidence that the grain storage capacity is governed by the size of the carpel established shortly before anthesis and the number of endosperm cells produced early post-anthesis, the latter being

influenced by irradiance and assimilate supply to the grain (Brocklehurst, 1977; Cochrane and Duffus, 1983; Scott et al., 1983; Singh and Jenner, 1984). A greater assimilate supply during early grain development could therefore lead to a larger potential grain size and simultaneously provide the additional dry matter for grain filling. At Aberdeen in 2006, the potential supply was far in excess of the actual MGW, so perhaps here grain development was saturated with assimilate and hence there was no change in storage capacity as potential supply increased.

At Aberdeen in 2005 and 2006, fungicide treatment increased MGW over a wide range of potential supply. These results suggest that fungicides increase MGW by influencing grain development and increasing the capacity for storage of dry matter, but not by increasing assimilate availability. Even at Rosemaund, where the displacement of potential supply for fungicide-treated and untreated crops was greater, parallel regression analysis indicated a significant difference in intercept, but not slope, consistent with the view that fungicide increased the storage capacity of grain.

Our data provide no evidence as to the mechanisms involved. Fungicides might increase partitioning of the available assimilate towards the grain so that it has a greater impact on grain development. Alternatively, fungicides might have a direct growth regulatory effect on grain development. Several groups of fungicides including the triazoles and strobilurins have been reported to possess growth regulatory activity (Grossmann and Retzlaff, 1997; Rademacher, 2000). Another possible explanation is that the fungicides are controlling symptomless pathogen infection. If pathogen infection restricts grain development without, or before, visible symptom expression, control of the pathogen could lead to an increase in grain storage capacity without an associated increase in estimated healthy area PAR interception and assimilate supply. Recent evidence has demonstrated that *R. secalis* and *Ramularia collo-cygni* can be present in the plant without producing symptoms (Havis et al., 2006; Fountaine et al., 2007; Walters et al., 2008). However, to date the effects of asymptomatic pathogen growth on host physiology have not been determined.

3.1.5 Conclusions and implications

Good disease management must be achieved, even in crops with high plant densities, to ensure shoot survival and final grain numbers are maximised. The sensitivity of grain number formation to intercepted PAR both before and after the start of stem extension implies that disease management must start early during tillering and be sustained through stem extension for maximum effect on grain numbers.

Fungicide-treatment increased MGW to the same extent irrespective of the potential assimilate supply, probably through effects on grain development and the capacity for grain filling. Since the development of potential grain size is determined shortly before and after ear emergence, these results emphasize the importance of disease management during this period. This study investigated the response to fungicide of a single variety. If similar results are also found for other varieties, it would call into question the current focus of late-season disease management to sustain canopy lifespan for as long as possible after flowering. Although an increase in potential grain size will lead to a greater dry matter requirement for grain filling, in the current study this was in the order of 3-4 mg per grain. Even in the absence of fungicide, potential assimilate supply often exceeded the measured grain weight by more than this suggesting that there would be sufficient assimilate to meet the increase in storage capacity of the grain without requiring a longer canopy duration. This hypothesis remains to be tested.

3.2 The effect of fungicide timing on the growth, radiation interception and yield of winter barley

3.2.1 Introduction

The yield of UK grown winter barley is considered to be largely sink limited. When the same variety was compared across sites and years, differences in the number of grains produced m^{-2} accounted for the majority of the variation in yield (Blake et al., 2006). Although MGW can also vary significantly (Bingham et al., 2007b), compared with grain number, it tends to be more stable. Results of experiments in section 3.1 demonstrated that grain number m^{-2} is related to

interception of photosynthetically active radiation (PAR) before GS 31 and during stem extension (GS 31-59). An association between post-anthesis PAR interception and final grain number was also found when the fertilizer N regime was used to modify post-anthesis green area duration. These results suggest that protection of the canopy from disease may be necessary throughout crop growth in order to maximise grain number production and hence the potential sink capacity and yield of the crop. However, the disease management treatments in Section 3.1 consisted of a full three-spray programme (including quinoxyfen) to give maximum control of all foliar disease, and a control that received just quinoxyfen to prevent mildew infection. Although such a three spray programme is typical for commercial crops of winter barley where rhynchosporium is the major disease threat, the treatment provides no information on the relative contribution of each application timing to the formation of yield. Nor does it provide any information on the effects of disease during specific developmental stages on the growth of the individual source and sink structures that contribute to yield. Such information is necessary in order to identify opportunities for rationalising fungicide inputs.

The results of section 3.1 also suggest that fungicides increase mean grain weight (MGW) through effects on potential grain size (i.e. the capacity of grains for storing dry matter) rather than the supply of assimilate for grain filling. Fungicides increased MGW over a large range of potential supply, when seed rate and N treatments were imposed to vary the latter. However, the effect of these treatments on the response of the crop specifically to late season (post-anthesis) disease could not be tested, because fungicide timing was not used to vary the progress of the disease epidemic. Thus, we cannot determine whether the requirement for ear emergence fungicides is modified by other agronomic treatments such as seed rate and N rate and timing.

Two experiments are reported in this section. The aim of experiment 1 was to investigate the effects of disease infection during different developmental phases on the growth, resource capture and yield of winter barley. This was achieved using different fungicide application timings and frequencies to vary the disease epidemic. The aim of experiment 2 was to investigate the effects of seed rate and N regime on the yield response to fungicide timing. If the tolerance of late season disease depends on the relative source-sink balance after flowering we

would expect to find a significant interaction between fungicide applications made at ear emergence and either seed rate or N regime.

3.2.2 Materials and Methods

Experiment 1

A two-row winter barley variety (*Hordeum vulgare* L. cv Haka) was sown in 2004 and 2005 at two sites, ADAS Rosemaund and SAC Aberdeen to give crops harvested in 2005 and 2006 respectively. At each site the experiment was located in the same fields used in section 3.1. Site selection criteria, general husbandry and sowing date were as described in section 3.1. The seed rates were 300 seeds m^{-2} at Rosemaund and 350 seeds m^{-2} at Aberdeen to reflect local commercial practice and plots were 24 m long. Fertilizer N was applied as a split dose of ammonium nitrate. Half the full recommended rate was applied at Zadoks growth stage (GS) 23/24 during tillering and the second half applied at the start of stem extension (GS 31). The recommended rate was determined from soil analysis, previous cropping and predicted requirement for feed barley crops according to standard fertilizer practice for the area (RB209 at Rosemaund; SAC Technical Note T516, 2002 at Aberdeen).

The experiment was laid out as a complete randomised block with three replicate plots per fungicide treatment. Fungicide treatments consisted of a factorial combination of autumn (GS21/22 Aberdeen and GS 22/24 Rosemaund), spring (GS 31/32; T1) and summer (GS 49/59; T2) applications (Table 1). At Rosemaund in 2006, the T2 application was delayed until GS 65 by poor weather conditions. At each application timing the same fungicide mixture was used with the aim of giving good control of *R. secalis*: epoxiconazole + boscalid (half rate, 0.75 l ha^{-1} Tracker, BASF) plus prothioconazole + fluoxastrobin (half rate, 0.625 l ha^{-1} Fandango, Bayer) plus fenpropimorph (0.3 or 0.4 of full rate, 0.3-0.4 l ha^{-1} Corbel, BASF). The low rate of fenpropimorph was selected to control established mildew but without scorching leaves. All applications were made in 225 l ha^{-1} water by hand operated gas-pressured sprayer. In the case of GS 31/32 treatments, applications were made after the crop had been sampled for growth analysis and disease assessments. All plots were treated with quinoxyfen (full manufacturers recommended rate; 0.3 l ha^{-1} Fortress, Dow) at GS 30 to prevent

powdery mildew (*Blumeria graminis*) infection without affecting *R. secalis*. Disease and crop growth assessments, PAR interception and final harvest yield and grain quality measurements were as described in section 3.1, including the year and site variations. In 2005, treatments 3 and 6 were not sampled for growth analysis, but were sampled for disease and taken to final grain yield.

Table 10. Fungicide application combinations. Application made (+), no application (-)

Treatment	Autumn GS 21/24	T1 GS 31/32	T2 GS 49/59
1	+	+	+
2	+	+	-
3	+	-	+
4	-	+	+
5	+	-	-
6	-	+	-
7	-	-	+
8	-	-	-

Growth analysis, healthy area PAR interception and potential post-anthesis assimilate supply were calculated as described in section 3.1, including the year and site variations. At the GS 31 sampling, treatment 1 was identical to treatments 2, 3 and 5, and treatment 8 was identical to treatments 4, 6 and 7 as sampling occurred prior to the spring fungicide application. Thus, only treatments 1 and 8 were sampled and values for individual blocks used for the corresponding treatments in calculations of light extinction coefficient and biomass production from GS 31 to 59. At Rosemaund in 2005, treatments 3 and 6 were not combined. Thus, yield and yield component data were analysed using analysis of variance for unbalanced designs in Genstat 11.1 (VSN International Ltd, Hemel Hempstead, UK).

Experiment 2

In 2006 winter barley cv Haka was sown at two sites, SAC Aberdeen and ADAS Rosemaund. Fields were selected using the criteria described in section 3.1.

Sowing dates were 18 September in Aberdeen and 21 September at Rosemaund. The experiment was a factorial design with 2 N timings x 2 seed rates x 6 fungicide regimes. It was laid out as a split-split plot with N treatments as main plots, seed rate as sub-plots and fungicide treatments as sub-sub plots. Plots were 24 m long. The N treatments were early (GS 23/24) or late (GS 31) application of the full recommended rate for feed barley. N was applied as ammonium nitrate with the recommended rate determined from soil mineral N analysis, previous cropping and predicted crop requirement as described for experiment 1. The seed rates used were 150 and 450 seeds m^{-2} for the low and high rates, respectively. There were 6 fungicide treatments: [1] autumn, GS 30/31 and GS 49/59 (+++), [2] autumn and GS 30/31 (++-), [3] autumn only (+--), [4] GS 30/31 only (-+-), [5] GS 49/59 only (--+), [6] untreated (---). The GS 30/31 application is referred to as the T1 and the GS 49/59 as the T2 timings. Fungicide mixtures and rates of application at each timing were the same as those used in experiment 1. All plots were treated with quinoxyfen (Fortress, Dow) to prevent powdery mildew (*Blumeria graminis*) infection without affecting *R. secalis*. In Aberdeen two half rate applications (0.15 l ha^{-1}) were made at GS 22 and GS 31. All other husbandry including P, K and Mn fertilizer, growth regulator and herbicide applications were as standard farm practice for the area.

Disease and % green leaf area were assessed at GS 31, 39, 59, 71 and further assessment of % green area made at GS 71 + 2 weeks. PAR interception by the canopy was determined at GS 31 and GS 69/71 and final ear number m^{-2} counted prior to harvesting. At harvest, grain yield was recorded on the combine and samples taken for determination of grain moisture and mean grain weight (MGW). All assessments were conducted as described in section 3.1. Data were analysed using analysis of variance routines in in Genstat 11.1 (VSN International Ltd, Hemel Hempstead, UK).

3.2.3 Results

Experiment 1

Disease and % green leaf area

As in section 3.1, the main disease at each site and in each year was rhynchosporium leaf scald (*Rhynchosporium secalis*). Only negligible levels of powdery mildew and brown spotting were observed. In 2005 disease severity in Aberdeen was greater than that at Rosemaund at GS 31 and 71, but comparable at GS 59 (Fig. 6). At Rosemaund post-anthesis leaf senescence progressed more rapidly than at Aberdeen, especially in the untreated plots and thus the relative green leaf area at the final assessment was similar at the two sites in spite of the much greater disease severity at Aberdeen.

Disease severity was generally much lower in 2006 than 2005 at each of the sites. In Aberdeen there was an appreciable amount of rhynchosporium present at GS 31, but thereafter the % severity declined as new leaf layers were produced that remained relatively free of disease (Fig. 6). The much greater disease severity at Aberdeen at GS 31 did not result in an appreciable reduction in relative GLA because treated plots (those that received autumn fungicide) showed similar levels of infection (9 and 11 % for treated and untreated plots respectively). As observed in 2005, post-anthesis senescence of untreated leaves progressed more rapidly at Rosemaund than Aberdeen, thus giving rise to a more pronounced decline in relative GLA in spite of the comparable disease severity at the two sites.

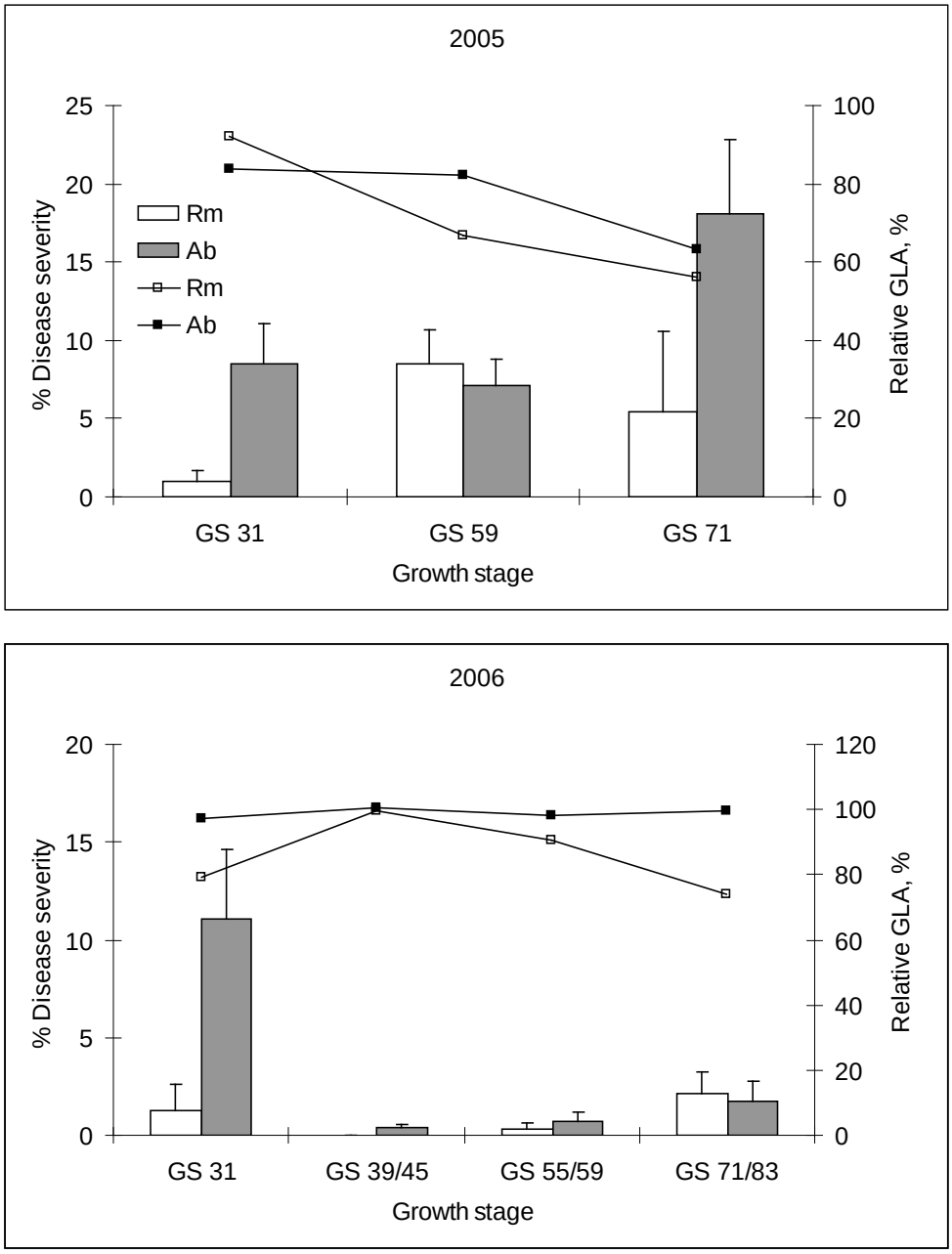


Figure 6. Severity of rhynchosporium infection and relative % green leaf area on non-fungicide treated plots in 2005 and 2006. Disease severity (columns) is expressed as the % surface area covered by lesions averaged for the top three fully expanded leaves. Relative % GLA (symbols and lines) is the average % green leaf area for the top four fully expanded leaves of untreated plots expressed relative to that of fungicide treated plots (values for fungicide treated plots normalized to 100%). Values are means of three replicate plots; bars on columns are SD. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Yield and yield components

There were significant yield responses to fungicide application in each site-year combination except Aberdeen 2006 (Table 11). The yield values presented are main effect means for the different fungicide timings. For example the autumn 'treated' value is the mean of all fungicide combinations containing the autumn application (Table 10). The most consistent yield responses came from autumn and GS 31 (T1) applications. In 2005 there were also significant responses to applications at GS 49/59 (T2). Interestingly, there was no significant response to T2 application at Rosemaund in 2006 when the application was unavoidably delayed until GS 65.

Table 11. Grain yield response to different fungicide timings. Values are main effect means after analysis of variance. Responses highlighted in bold are significant at P <0.05. Three site mean determined by anova for unbalanced designs after removing Ab 2006. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Year	Site	Fungicide	Yield, t ha^{-1} @100% DM		
			Autumn	T1	T2
			GS 21/24	GS 31/32	GS 49/59
2005	Ab	untreated	5.42	5.21	5.17
		treated	5.70	5.91	5.95
		response	**0.28**	**0.70**	**0.79**
2005	Rm	untreated	6.17	6.11	6.04
		treated	6.87	6.92	6.99
		response	**0.70**	**0.81**	**0.95**
2006	Ab	untreated	5.76	5.79	5.80
		treated	5.84	5.81	5.80
		response	0.09	0.02	0.00
2006	Rm	untreated	7.77	7.62	7.95
		treated	8.28	8.44	8.10
		response	**0.50**	**0.82**	0.15
3 site mean		untreated	6.50	6.31	6.44
		treated	6.94	7.14	7.01
		response	**0.44**	**0.83**	**0.57**
df 50		SED	0.074	0.074	0.074

As there was very little disease and no significant yield response to fungicide application at Aberdeen in 2006, this site-year has been excluded from

subsequent analyses. A combined analysis of the remaining site-years with site-year as a blocking factor indicated significant effects of autumn, T1 and T2 fungicide applications on yield, but no significant interaction between the different timings. This implies that the response to a particular application timing was not affected by applications at other timings. In other words, the effects of each timing on yield appeared to be additive.

There was a consistent effect of autumn and T1 fungicide applications on grains m^{-2} for the three sites showing an overall yield response to fungicide (Table 12). T1 treatments resulted in a greater increase than autumn treatments in all site-years. When averaged across site–years, the increase in grain number in response to autumn treatment was associated with a significant increase in ear number m^{-2}. There was a comparable, though not statistically significant, increase in ear number in response to T1 treatment. Fungicide application at GS 49/59 resulted in a smaller increase in grain number m^{-2}, but no significant effect on ear number, implying that the increase in grain number was solely the result of an increase in the number of grains ear^{-1}. By contrast, MGW was increased by T1 and T2 treatments, with no significant effect of autumn application. As found with yield, there were no significant interactions between any of the fungicide timings on the individual yield components.

Pre-anthesis canopy growth

When averaged over three site years, autumn fungicide led to a small, but significant increase in healthy canopy area (healthy area index, HAI) at flowering (Table 13). Spring treatment increased HAI to almost twice the extent (0.9 units of HAI compared to 0.5 for autumn treatment). There was no significant effect of either fungicide timing on the number of potentially fertile shoots present at GS 59 in this experiment (but a trend towards an increase with spring application, $P = 0.068$), thus the increase in healthy canopy area was associated for the most part with an increase in healthy area per shoot. There was also no significant effect of fungicide on the concentration of stem water soluble carbohydrate (WSC) reserves. The quantity of WSC per m^{-2} was increased to a small but significant extent by autumn fungicide treatment.

Table 12. Yield components. Values are main effect means after analysis of variance. Responses highlighted in bold are significant at P <0.05. Three site mean determined by anova for unbalanced designs. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Year	Site	Fungicide	Grains m^{-2}			MGW, mg @100% DM			Ears m^{-2}		
			Autumn	T1	T2	Autumn	T1	T2	Autumn	T1	T2
2005	Ab	untreated	10338	10028	10182	52.29	51.87	50.69	685	674	678
		treated	10750	11060	10907	52.93	53.35	54.54	686	697	693
		response	**412**	**1032**	**725**	0.64	**1.48**	**3.85**	1	23	15
2005	Rm	untreated	15123	15011	15324	40.67	40.61	39.36	851	874	903
		treated	16359	16471	16157	41.86	41.93	43.17	944	924	898
		response	**1236**	**1460**	**833**	1.19	**1.32**	**3.81**	**93**	51	-4
2006	Rm	untreated	16314	16261	16725	47.61	46.82	47.49	801	811	809
		treated	17386	17439	16974	47.6	48.39	47.71	834	825	827
		response	**1072**	**1178**	249	-0.01	**1.57**	0.22	33	14	18
3 site mean		untreated	13825	13634	13977	47.54	46.87	46.53	769	774	786
		treated	14699	14890	14547	47.90	48.57	48.91	809	804	793
		response	**874**	**1256**	**570**	0.36	**1.70**	**2.38**	**39**	31	7
		P	<0.001	<0.001	<0.001	0.316	<0.001	<0.001	0.024	0.086	0.717
		SED; df	128.3; 50			0.325; 50			17.78; 49		

Healthy area PAR interception and grain number m^{-2}

Autumn fungicide application increased PAR interception by healthy tissue before GS 31 by 27 MJ m^{-2} (Table 14). The increase was close to significance (P = 0.068). Autumn application also increased PAR interception between GS 31 and 59 and to a lesser extent after ear emergence (GS 59 – maturity). The values represent the difference in average PAR interception between all those treatment combinations containing an autumn application and all those without one. Since there was no significant interaction between fungicide timing on PAR interception in this experiment, the applications appear to operate independently of each other. T1 application also increased PAR interception between GS 31 and 59 and GS 59 – maturity. The increase before ear emergence was comparable to that found with autumn fungicide, whilst after ear emergence it was more than double. T2 application had no significant effect on PAR interception before ear emergence, but increased it significantly afterwards.

Table 13. Canopy components at GS 59 and their response to fungicide treatment in autumn or spring. Values are main effect means for fungicide timing in an analysis across three site-years; residual degrees of freedom, 38. Levels of significance; * P<0.05, *** P<0.001, ns P >0.05.

Variate	Timing	Fungicide		SED	Signif
		Untreated	Treated		
HAI	Autumn	6.6	7.0	0.258	*
	T1	6.3	7.3	0.258	***
	T2	7.0	6.5	0.294	ns
Fertile shoots m^{-2}	Autumn	983	980	28	ns
	T1	952	1011	28	P = 0.068
	T2	992	965	32	ns
HA, cm^2 $shoot^{-1}$	Autumn	64.8	69.4	1.58	***
	T1	63.5	70.8	1.58	***
	T2	67.9	66.0	1.8	ns
WSC % DM	Autumn	21.2	21.4	0.48	ns
	T1	21.2	21.4	0.48	ns
	T2	21.2	21.4	0.55	ns
WSC g m^{-2}	Autumn	155	168	6.38	*
	T1	159	164	6.38	ns
	T2	163	159	7.28	ns

Table 14. Increase in PAR interception by healthy area during specific developmental phases resulting from fungicide application timing. Grain no. MJ^{-1} is the increase in grain number (Table 12) for the fungicide timing expressed per unit of additional total PAR interception. Values are main effect means for a 2 site analysis (SAC Aberdeen 2005 & ADAS Rosemaund 2006). Levels of significance; * P<0.05, *** P<0.001; # P= 0.068.

Fungicide timing	Increase in HA PAR interception, MJ				Grain no. MJ^{-1}
	Emergence - GS 31	GS 31-59	GS 59 - maturity	Total	
Autumn	26.6 #	13.5 ***	9.9 *	50.0	14.5
T1		12.5 ***	26.7 ***	39.2	28.6
T2		4.1 ns	27.9 ***	32.0	14.7
df	5	29	29		
SED	11.5	2.44	4.20		

The increase in grain number m^{-2} with T2 treatment was small compared with T1 treatment (Ab 2005 and Rm 2006 Table 12), yet the gain in PAR interception was comparable (Table 14). Thus, the increase in grain number per MJ of additional PAR intercepted following the T1 treatment was twice that following the T2 application. These results suggest that the physiological effect of fungicide on grain numbers occurs predominantly before ear emergence.
By contrast autumn fungicide led to a similar increase in PAR interception during stem extension (GS 31-59) but a larger total pre-anthesis interception (emergence – GS 59) than T1 application, yet had a smaller effect on grain number formation. The increase in grain number per MJ of additional PAR intercepted was about half that found with T1 fungicide. These results suggest that the effect of T1 fungicide on grain number formation is disproportionate to its effect on PAR interception. This is supported by Fig. 7. When data for Ab 2005 and Rm 2006 were pooled, the slope of the relationship between healthy area PAR interception and grain number m^{-2} was greater with T1 fungicide than without it (but not statistically significant, P = 0.092). When the data from each

site are considered separately, it is apparent that at both Aberdeen and Rosemaund T1 fungicide increased grain numbers over a range of PAR interception comparable to that observed in the absence of T1. This again supports the conclusion that the effect of T1 fungicide on grain numbers is disproportionate to its effect on PAR interception.

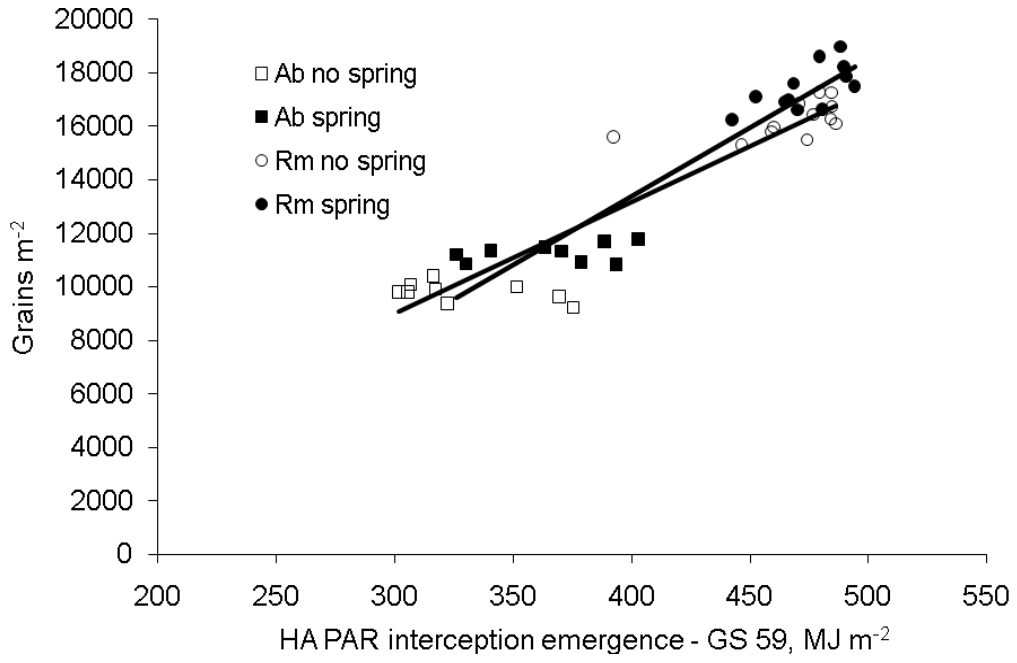
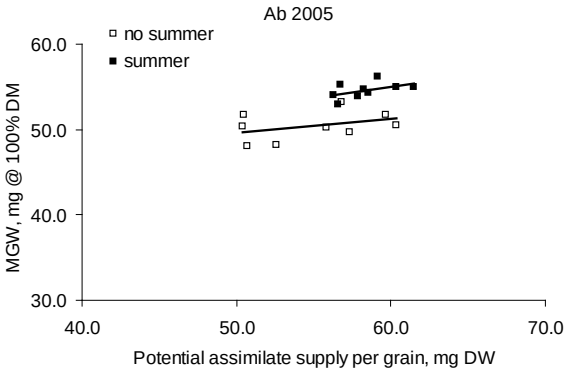
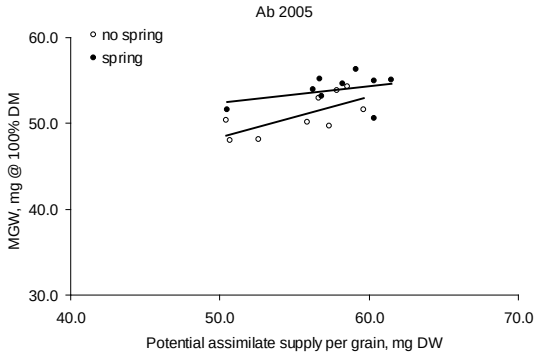
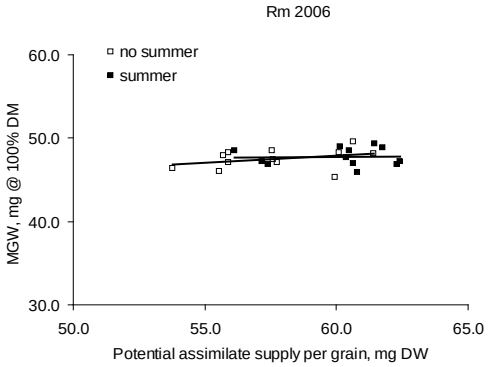


Figure 7. Relationship between PAR interception by healthy area pre-anthesis and grain number m^{-2} for fungicide treatment combinations with and without spring application. Data are pooled from Ab 2005 (squares) and Rm 2006 (circles). Without spring fungicide y = 41.7x – 3516 r^2 = 0.87; with fungicide y = 51.4x - 7160.3 r^2 = 0.89. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.







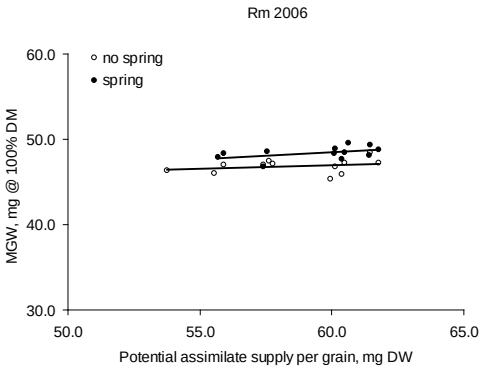


Figure 8. Relationship between potential assimilate supply post-anthesis per unit grain number and mean grain weight (MGW) for fungicide treatment combinations with and without T1 (spring) and T2 (summer) applications. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Mean grain weight

At Aberdeen in 2005 T2 fungicide increased both the potential post-anthesis assimilate supply per grain and the MGW (Fig. 8). However, MGW was increased even where the range of potential assimilate supply for fungicide-treated and untreated crops overlapped. By contrast there was no overall effect of T2 fungicide on MGW (Table 12 & Fig. 8) at Rosemaund in 2006 even though analysis of variance indicated there was a significant increase ($P = 0.02$) in potential assimilate supply per grain (data not shown). It should be remembered that this application was delayed until GS 65. T1 fungicide, on the other hand, increased MGW at Rosemaund, as well as Aberdeen, over the whole range of potential post-anthesis assimilate supply.

Experiment 2

Disease severity

The main diseases and disorders at Aberdeen were rhynchosporium and brown spotting (Fig. 9). Levels were low (<1.3 %) during early stem extension (GS 31 – 39). The severity of spotting increased between GS 39 and 59 and that of rhynchosporium between GS 59 and 77. Significant mildew infection also occurred after ear emergence. In spite of the low apparent disease severities at GS 31, the % green leaf area (GLA) averaged over the top four leaves of untreated plots was low (65%). However, this was largely the result of natural rather than disease-induced senescence of lower-most leaves since the GLA of fungicide-treated plots was equally low giving a relative GLA (GLA of untreated relative to fungicide-treated plots) of 100%.

At Rosemaund the major disease was rhynchosporium. Some net blotch was found, but the average severity on the top three leaves never exceeded 0.6% (Fig. 9). Trace levels of brown rust and powdery mildew were observed in some plots. During stem extension, as the rate of new leaf production exceeded the rate of epidemic development, the severity of rhynchosporium declined from 3% observed at GS 31 to 1% at GS 59. The severity then rose to nearly 8% after ear emergence.

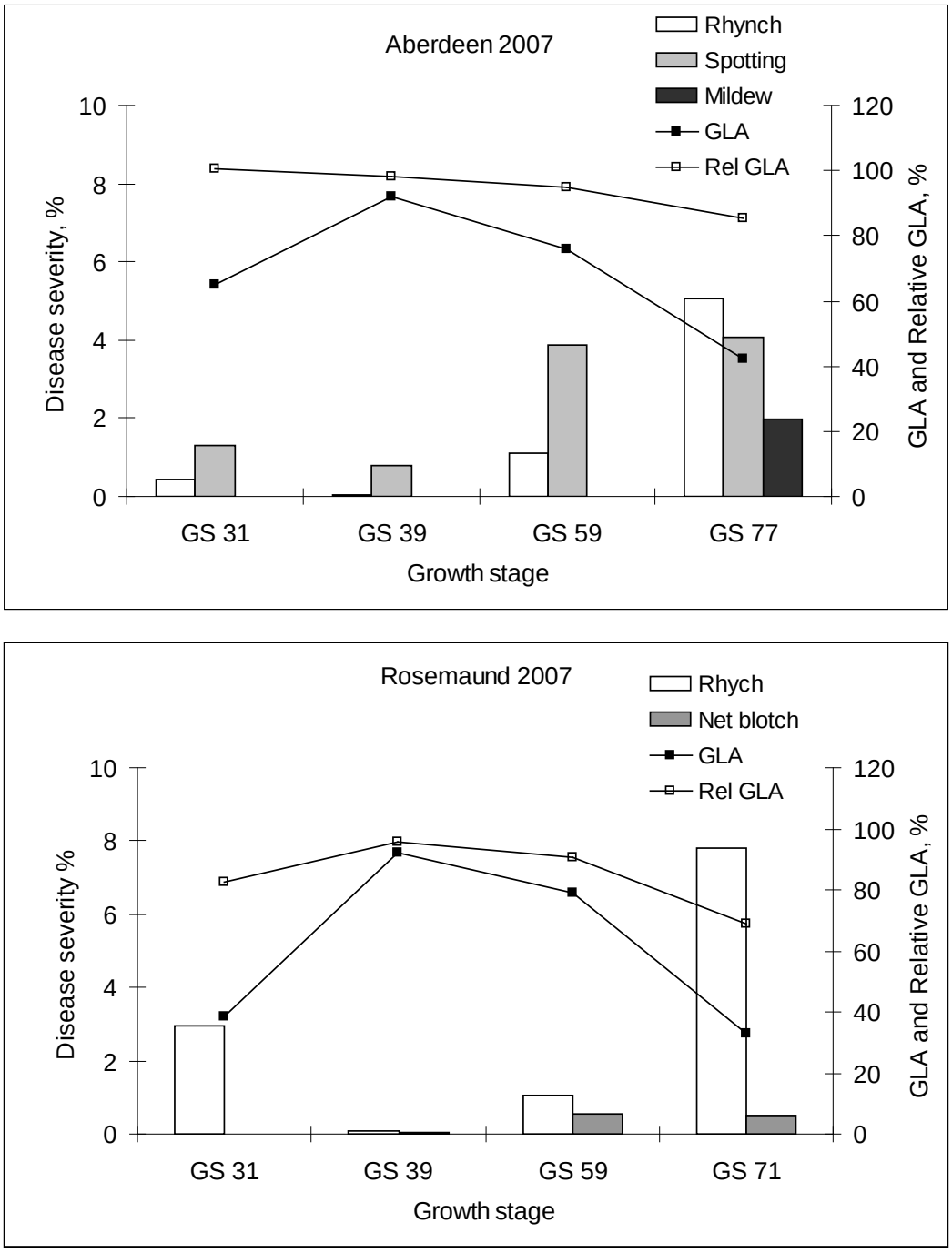


Figure 9. Severity of the main diseases and % green leaf area present in untreated plots in experiment 2. Disease severity (columns) is expressed as the % surface area covered by lesions averaged for the top three fully expanded leaves. GLA is the % green leaf area averaged for the top four fully expanded leaves of untreated plots. Relative GLA is the GLA % of untreated plots expressed relative to that of fungicide treated plots (values for fungicide treated plots normalized to 100%).

At Rosemaund early N applications resulted in significantly greater rhynchosporium infection of non-fungicide treated plots compared with late N at GS 31, and GS 59 (Table 15). There was no significant effect of seed rate on disease severity until after flowering when more disease was found at high seed rate. Further, there was no significant interaction between N regime and seed rate on disease severity at any growth stage.

At Aberdeen there was, in general, no effect of either N regime or seed rate on the severity of rhynchosporium (Table 16). At GS 39 a statistically significant increase was observed with early N compared to late, but in each treatment disease severities were negligible (0.1% or less) at this time. Brown spotting tended to be more severe with late N compared to early N and at high seed rate compared to low from GS 39 onwards.

Table 15. Effects of N regime and seed rate on the severity of rhynchosporium infection averaged over the top three fully expanded leaves of non-fungicide treated plots at Rosemaund 2007. Values are non-transformed main effect means. SEDs are arcsine transformed values (in radians) after analysis of variance. Significance; * P <0.05, ns P > 0.05.

		Rhynchosporium, %			
Factor	Treatment	GS 31	GS 39	GS 59	GS 71
N	Early	2.3	0.2	1.9	10.4
	Late	0.9	0.0	0.2	5.2
Seed rate	Low	1.8	0.0	0.5	4.4
	High	1.4	0.2	1.7	11.3
	df	SED, significance			
N	2	0.012 *	0.017 ns	0.014 *	0.036 ns
SR	4	0.024 ns	0.017 ns	0.028 ns	0.049 *
N*SR	4	0.026 ns	0.024 ns	0.032 ns	0.061 ns

Table 16. Effects of N regime and seed rate on the severity of rhynchosporium infection and brown spotting averaged over the top three fully expanded leaves of non-fungicide treated plots at Aberdeen 2007. Values are non-transformed main effect means. SEDs are arcsine transformed values (in radians) after analysis of variance. Significance; *** P<0.001, * P <0.05, ns P > 0.05.

		Rhynchosporium, %				Spotting, %			
Factor	Treatment	GS 31	GS 39	GS 59	GS 77	GS 31	GS 39	GS 59	GS 77
N	Early	0.6	0.1	1.7	4.1	1.4	0.2	1.7	2.7
	Late	0.3	0.0	0.5	6.1	1.1	1.3	6.0	5.4
Seed rate	Low	0.4	0.1	1.2	4.8	0.8	0.4	2.5	2.1
	High	0.5	0.0	1.0	5.4	1.7	1.2	5.2	6.0
	df	SED, significance				SED, significance			
N	2	0.018 ns	0.0006 ***	0.022 ns	0.015 ns	0.017 ns	0.008 *	0.003 ***	0.036 ns
SR	4	0.010 ns	0.005 ns	0.024 ns	0.020 ns	0.025 ns	0.009 *	0.019 *	0.037 *
N*SR	4	0.021 ns	0.007 ns	0.034 ns	0.029 *	0.030 ns	0.013 *	0.026 *	0.052 ns

Table 17. Main effects of agronomic treatments on yield and yield components in 2007. Yields and mean grain weight (MGW) are expressed at 100% dry matter. Significance; *** P <0.001, ** P <0.01, * P <0.05, ns P >0.05. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

		Yield, t ha^{-1}		Grains m^{-2}		MGW, mg	
Factor	Treatment	Ab	Rm	Ab	Rm	Ab	Rm
N	Early	6.27	7.27	13218	15678	47.50	46.38
	Late	3.53	7.20	8363	15066	42.20	47.84
Seed rate (SR)	Low	4.93	7.30	10449	15232	46.60	47.95
	High	4.86	7.18	11132	15512	43.10	46.27
Fungicide (F)	Untreated	4.48	6.73	10133	14776	43.75	45.54
	Autumn (Aut)	4.75	7.00	10899	14983	43.18	46.81
	T1	4.74	7.26	10296	15430	45.43	47.13
	T2	4.71	7.30	10390	15246	44.83	47.92
	Aut+T1	5.36	7.27	11423	15617	46.23	46.69
	Aut+T1+T2	5.34	7.86	11602	16180	45.68	48.58
	df	SED, significance					
N	2	0.523 *	0.087 ns	1068 *	400 ns	1.628 ns	0.674 ns
SR	4	0.211 ns	0.037 *	563 ns	125 ns	0.672 **	0.425 *
F	40	0.134 ***	0.121 ***	375 ***	326 **	1.049 *	0.654 ***
N*F	40	ns	ns	ns	ns	ns	ns
SR*F	40	P = 0.055 *		ns	ns	ns	ns

Yield and yield components

When averaged over the other the other treatments, late N application almost halved the yield at Aberdeen compared to early application (Table 17). This was associated with both a smaller grain number m^{-2} and smaller MGW. However, the latter was not statistically significant in this split-split plot analysis because the degrees of freedom for the N main plots was low. N regime had no significant effect on yield or its components at Rosemaund. High seed rate led to a small reduction in yield compared to low, an effect associated with a significant reduction in MGW at each site. Fungicide treatment had a significant effect on yield, grains per m^2 and MGW at each site and there was a significant interaction between seed rate and fungicide on yield at Rosemaund, and near significant (P=0.055) interaction at Aberdeen.

A cross site analysis of the seed rate x fungicide interaction is given in Table 18. Comparison of the yield response to fungicide with the seed rate x fungicide LSD indicates that at high seed rate there was a significant increase in yield with all fungicide timings. At low seed rate, however, there was a significant increase only when autumn treatment was combined with T1 and T1 plus T2, not with single applications. Thus, there was a significant difference in yield response between high and low seed rate following autumn, T1 and autumn plus T1 and T2 combinations (difference in yield response exceeded LSD for seed rate x fungicide interaction). Although overall the interaction between seed rate and fungicide on grains m^{-2} was not statistically significant, the trend in grain numbers followed that of yield. Thus, at high seed rate there was a large increase in the response of grain numbers to single autumn and T1 treatments (exceeding the LSD for interaction), but not at low seed rate. MGW did not follow the same trend. Thus an appreciable difference in response between seed rates was observed only when autumn treatment was combined with T1 or T1 plus T2. Examination of the main effect of fungicide on MGW (average of high and low seed rate), indicates that it was the T1 and T2 treatments alone or in combination that gave the significant increase in MGW.

Table. 18. Cross site analysis of the response of yield and yield components to fungicide timing at high and low seed rate. The response is the difference between the yield or yield component at a given timing and the untreated control. Absolute value for the untreated control is given in italics. Significance, df, SEDs and LSDs are given for the main effect of fungicide and the seed rate x fungicide interaction. Values for yield and MGW are expressed at 100% DM. Significance; *** P <0.001, * P <0.05, ns P >0.05.

		Yield response, t ha^{-1}				Grains m^{-2} response				MGW response, mg		
Fungicide		Low	High	Difference		Low	High	Difference		Low	High	Difference
Autumn (Aut)		-0.01	0.55	0.56		-12	986	998		-0.09	0.79	0.88
T1		0.20	0.59	0.39		-27	843	870		1.74	1.53	-0.21
T2		0.39	0.41	0.02		429	298	-131		1.42	2.04	0.62
Aut+T1		0.49	0.94	0.45		823	1308	485		0.85	2.78	1.93
Aut+T1+T2		0.81	1.18	0.38		1243	1629	386		1.61	3.35	1.74
Untreated		*5.80*	*5.41*			*12431*	*12478*			*46.35*	*42.94*	
		SED	df	LSD		SED	df	LSD		SED	df	LSD
F	***	0.098	100	0.19	***	246	100	488	***	0.618	100	1.23
SR*F	*	0.159	57	0.32	ns	414	49	832	ns	0.927	82	1.84

Table 19. Effects of agronomic treatments on canopy size (CAI) and % PAR interception at GS 69/71 Aberdeen and Rosemaund 2007. Significance; *** P <0.001, ** P< 0.01, * P <0.05, ns P >0.05.

		Aberdeen			Rosemaund		
Factor	Treatment	CAI	% PAR interception	% interception per 1000 grain	CAI	% PAR interception	% interception per 1000 grain
N	Early	4.35	92.5	7.1	7.0	98.4	6.3
	Late	2.42	76.3	9.4	5.3	95.7	6.4
Seed rate (SR)	Low	3.22	82.8	8.3	6.1	96.9	6.4
	High	3.55	86.0	8.2	6.2	97.2	6.3
Fungicide (F)	Untreated	3.29	83.8	8.6	5.5	96.0	6.5
	Autumn (Aut)	3.31	83.9	8.1	6.0	96.9	6.5
	T1	3.35	84.2	8.6	6.2	97.1	6.3
	T2	3.38	84.3	8.6	6.2	97.2	6.4
	Aut+T1	3.46	84.8	7.8	6.3	97.1	6.3
	Aut+T1+T2	3.52	85.4	7.7	6.6	97.8	6.0
	df	SED, significance			SED, significance		
N	2	0.174 **	1.90 *	0.82 ns	0.18 *	0.1 **	0.16 ns
SR	4	0.074 *	0.64 **	0.38 ns	0.10 ns	0.28 ns	0.04 ns
F	40	0.060 **	0.44 **	0.29 **	0.13 ***	0.24 ***	0.13 **
N*F		ns	ns	ns	ns	ns	ns
SR*F		ns	ns	ns	ns	ns	ns

Canopy size and PAR interception

Detailed measurements of canopy growth were not made in this experiment, but canopy area index (CAI; diseased and healthy leaf laminae plus stem and leaf sheaths) early post-flowering (GS 69/71) was estimated from measurements of % PAR interception and an assumed light extinction coefficient of 0.6. Fertilizer N regime had a significant effect on % PAR interception, and by inference canopy size with late N reducing CAI by 45% at Aberdeen and 25% at Rosemaund compared to early N (Table 19). Seed rate, by comparison had a smaller effect on % PAR interception and estimated canopy size. They were increased significantly by high seed rate at Aberdeen, but not Rosemaund. The effects of early N regime and high seed rate on canopy size were accompanied by increases in grain number m^{-2}, such that the % PAR interception per unit grain number was not significantly altered by the treatments. Fungicide application increased % PAR interception and estimated CAI and reduced % interception per grain. However, it should be emphasized that the values presented are for CAI and light interception by the healthy plus diseased parts of the canopy. As such the decline in % PAR interception per unit grain number with fungicide treatment would tend to be reduced or even reversed if PAR by healthy tissue alone were considered.

3.2.4 Discussion

The objective of experiments reported in this section was to determine the relative contribution of the individual components of a three-spray fungicide programme to yield formation in barley, and to provide insights into the physiological basis of their effects. As was found in section 3.1, the full fungicide programme resulted in a significant increase in yield and was associated with increases in each of the major yield components, MGW, grain number m^{-2}, and its sub-components ears m^{-2} and by inference grains ear^{-1}. The autumn and T1 applications resulted in the largest increase in grain numbers, although the T2 application also increased grain numbers to a small extent.

These effects are consistent with the phasing of the developmental processes controlling grain site formation. Tiller production and spikelet initiation occur before GS 31. The survival of the initiated tillers and spikelets is then

determined for the most part from GS 31 to GS 59 and coincides with the phase of rapid of stem extension. Autumn applications of fungicide will therefore provide protection of the canopy during the periods of both initiation and survival of tillers and spikelets. T1 applications, on the other hand, provide protection during the survival phase. T2 applications made during booting provide protection during late floret development; a period that has been shown to be critical for the determination of the number of grains per ear (Arisnabarreta and Miralles, 2008).

The production and survival of both tillers and spikelets are sensitive to variations in light interception. The main evidence for this has come from shading treatments designed to reduce the amount of solar radiation incident upon the canopy. The results of shading experiments are usually interpreted in terms of the availability of photosynthates for metabolism or regulation of growth, although it should be noted that changes in availability are generally inferred and supporting measurements of carbohydrate concentrations in the tissue are rarely made (Jenner, 1980). Shading of spring barley during stem extension resulted in a greater reduction in grain number than shading from crop establishment to the end of ear initiation (Willey and Holliday, 1971) suggesting that the mortality of tillers and spikelets may be more sensitive to variation in light interception and assimilate availability than tiller and spikelet production. This may account in part for the greater response of grain numbers to T1 applications compared to autumn fungicide in winter barley.

However, our results suggest that the response to fungicide may be more complex than is generally appreciated and that other factors may underlie the difference in response to autumn and T1 fungicides. Autumn and T1 applications increased PAR interception by healthy area between GS 31 and 59 to a similar extent, yet T1 applications had the greatest effect on grain numbers. Thus, the greater response to T1 application cannot be explained simply in terms of improved light interception during the sensitive period of tiller and spikelet mortality. There may be additional effects of fungicide, possibly via an increase in PAR use efficiency or partitioning of assimilates to the developing tillers and ears, that result from either control of the pathogen or direct effects of fungicide on host metabolism. Moreover, to account for the difference in response of grain numbers to autumn and T1 fungicide, these additional effects must be

dependent on the time of application with application at GS 31 being more effective than in the autumn (GS 21/24).

Interaction between fungicide and seed rate provides further evidence of the complexity in fungicide action. In section 3.1 plots grown at a standard commercial seed rate showed a greater yield and grain number response to fungicide than plots at low seed rate. The greater response could not be explained in terms of the control of a greater disease severity and improved light interception. A similar interaction was found in experiment 2 above at both Aberdeen and Rosemaund. At Aberdeen, seed rate had no significant effect on disease severity, but the yield response to fungicide was greater at high seed rate than low. Although the severity of brown spotting was increased by high seed rate, spotting was not affected by fungicide treatment and, therefore, cannot account for the greater yield response to fungicide at high seed rate. At Rosemaund, seed rate had no effect on disease severity before ear emergence. In the 2007 experiment, the interaction between seed rate and fungicide appeared to be associated with early fungicide applications. When averaged across both sites, there was a significant yield response to a single application made in autumn and T1, at high seed rate but not at low seed rate. The response was associated with an increase in grain number. At low seed rate, increases in grain number only occurred with combinations of autumn, T1 and T2 applications, not single applications. It is conceivable that effects of fungicides on grain number formation are dose dependent and that at low seed rate higher doses, as provided by a sequence of applications, are required to elicit the response than at high seed rates.

In experiment 1 MGW was increased by both T1 and T2 fungicide, but not by autumn applications. The analysis of MGW determination presented in section 3.1 suggested that fungicide increased MGW by influencing grain development and the capacity for storing dry matter (referred to as potential grain size), rather than the availability of dry matter for grain filling. This was based on a comparison of untreated plots with those receiving a full three spray programme. The current experiments enable the same type of analysis to be conducted to compare the effects of T1 and T2 fungicide applications. At Aberdeen in 2005 both T1 and T2 applications increased MGW over a range of potential assimilate supplies, suggesting that in each case, the treatment

increased MGW through an increase in potential grain size. At Rosemaund 2006, T1 application increased MGW in the same manner over a range of assimilate availability, whereas T2 did not, even though the potential assimilate availability was increased. It may be significant that in Aberdeen, where a response of MGW to T2 was found, the T2 was applied during late booting, whereas at Rosemaund T2 was delayed until GS 65. It is tempting to speculate that the fungicide-responsive physiological events that regulate MGW occur prior to fertilization, possibly operating via effects on carpel size. However, the response of MGW to fungicide timing either side of anthesis would need to be determined at a larger number of sites in order to test this hypothesis more rigorously.
No significant interaction was found in experiment 2 between fungicide treatment and either seed rate or N regime on MGW. This implies that the need for a T2 fungicide to protect grain size was not modified by these particular agronomic treatments. This is in spite of the fact that disease severity in non-fungicide treated plots was altered by agronomy (Rosemaund 2007).

3.2.5 Conclusions and implications

The results indicate that protection of the canopy with fungicide is required throughout the growth of winter barley crops in order to maximise yield. Significant yield responses were found with autumn, T1 and T2 application timings, although the greatest response was associated with the T1 application. There is evidence that the yield response to T1 is not simply the result of the protection of green area and an increase in PAR interception by healthy tissue, but that other mechanisms may be at work. The response of grain numbers to autumn and T1 applications may be modified by other agronomic treatments such as seed rate; the response being less at low seed rate. Effects of late season fungicides (during booting) on MGW of winter barley may be associated with increases in the potential size (storage capacity) of grains. There is no evidence to suggest that the need for a T2 fungicide was modified by variations in the source-sink balance of the crop.

3.3 The effects of source-sink balance on the response to fungicides in spring barley

3.3.1 Introduction

Spring and winter-sown varieties of barley follow the same pattern of development through the season. After seed germination, both go through a phase of vegetative development in which the stem apex initiates leaves and tillers, until reproductive development begins. After switching to reproductive development the stem apex forms the ear via the initiation and subsequent development of spikelets and florets. Grain formation is the last major phase of development to occur and commences with fertilization of ovules during, or shortly after, ear emergence. Although both spring and winter barley follow the same developmental sequence, they do so at different rates as governed by their contrasting responses to temperature and daylength. Spring varieties have no vernalization requirement and develop faster than winter varieties.

The main constraints to yield are also similar in spring and winter varieties. Variation in yield across sites and years is strongly associated with variation in the number of grains produced per m^2 (Blake et al. 2006). By contrast mean grain weight tends to be more stable leading to the view that the yield of both winter and spring barley is limited primarily by the number of grains produced and their capacity for storing dry matter (sink-limitation), rather than the supply of assimilate available for grain filling (source-limitation) (Bingham *et al*. 2007ab). A consequence of the above similarities between winter and spring crops is that the aim of disease management in each is also broadly the same. Thus, the aim is to maximise grain number formation by protecting the canopy during the period of tiller and spikelet production and survival, and then to protect grain development and filling.

Results reported in the previous two sections highlight the importance of early season protection in winter barley, as significant yield responses were found with autumn and T1 applications of fungicide. However, the results also indicate that the yield response to fungicide can be complex and is not related simply to the size and timing of the disease epidemic, but that it can vary with the physiological and developmental state of the crop. Thus, yield responses were greater in crops at high population densities compared to low (section 3.1).

Moreover, the timing of fungicide application in relation to developmental stage may be important. T1 applications resulted in a larger increase in grain numbers compared to autumn applications, even though their effect on light interception by healthy tissue during stem extension was comparable (section 3.2).

Whilst the broad aims of disease management are the same for winter and spring crops, the faster rate of development and shorter growing season for spring varieties could modify their response to fungicide compared to winter types. For example, the faster rate of development could mean there is less time for grain numbers to increase in response to disease control. The relative source-sink balance could be different, reflecting the different duration of pre-anthesis to post-anthesis growth in winter and spring crops. Furthermore, in commercial practice, the timing of fungicides for spring and winter crops tends to differ with the stem extension application (T1) occurring earlier (GS 25-30) in spring compared to winter (GS 30-32) crops. This could be significant if fungicides have direct effects on crop development that impact on grain number formation, rather than operating simply through protection of green leaf area.

The objective of experiments reported in this section was, therefore, to determine the effects of altering the source-sink balance of spring barley, by varying seed rate and N timing, on its response to fungicide. By varying seed rate, N regime and fungicide timing in the same experiment, we test whether the response to a T2 fungicide (GS 41-49) is influenced by the source-sink balance of the crop. This will identify whether there is any scope for modifying the T2 application based on an assessment of the risk of disease, the state of the crop and its likely response to fungicide. Evidence was presented in section 3.1 and 3.2 suggesting that fungicides increase mean grain weight through effects on the storage capacity of grain rather than how well the grain fills. The analysis is based on the assumption that pre-anthesis radiation use efficiency (RUE) reflects the potential RUE post-anthesis. In the current experiments measurements of both pre-anthesis and post-anthesis light interception and biomass gain were made to test the validity of this assumption for spring barley.

3.3.2 Materials and Methods

Sites and general husbandry

Spring barley (*Hordeum vulgare* L. cv Cocktail) was sown in 2006 and 2007 at ADAS Rosemaund and in 2007 and 2008 at SAC Aberdeen in plots of 24 x 1.5 m. At each site, the fields occupied a rotational position that was representative for barley production in the region. Sowing dates were 23 March in 2006 and 2007 at Rosemaund and 5 April 2007 and 20 April 2008 at Aberdeen. The variety Cocktail was selected for its relatively high susceptibility to *Rhynchosporium secalis*, initially the main disease of interest. The same source of seed was used for both sites. Fertilizer P and K were applied to the seedbed according to soil mineral analysis and anticipated crop demand. Micronutrients, molluscicides, herbicides, insecticides were applied to all plots, as per standard farm practice. A plant growth regulator (chlormequat 1.25 l ha^{-1} at GS 31) was applied to plots at Rosemaund, but not Aberdeen reflecting local practice in the region.

In order to try and generate some splash dispersal of *R.secalis* inoculum plots at Aberdeen in 2008 were irrigated with an overhead irrigation system from GS 30 to GS 59. Six mm of water was applied Monday and Thursday of each week if the rainfall during the interval between scheduled days was less than 6 mm. If rainfall exceeded 6 mm over the previous interval, no irrigation was applied on the scheduled irrigation day.

Treatments and experimental design

Experimental treatments consisted of two nitrogen timings, two seed rates and four fungicide timings. The full recommended N fertilizer requirement was calculated on the basis of previous cropping (Aberdeen) and an analysis of soil mineral N (to 90 cm) made in the first week of February (Rosemaund). The N was then applied either at crop emergence (early N) to promote tiller production, or at the start of stem extension (late N) to promote retention of green leaf area post-anthesis. Seed rates were 100 and 600 seeds per m^2 representing low and high seed rate extremes respectively.

The fungicide treatment programmes were: [1] untreated, [2] application at T1 only (GS 31), [3] application at T2 only (GS 45-59), [4] application at T1 and

T2. At each timing, the same fungicide mixture was used with the aim of giving good control of *R. secalis*: epoxiconazole + boscalid (half manufacturers recommended rate, 0.75 l ha^{-1} Tracker, BASF) plus prothioconazole + fluoxastrobin (half rate, 0.625 l ha^{-1} Fandango, Bayer) plus fenpropimorph (0.3 or 0.4 of full rate, 0.3-0.4 l ha^{-1} Corbel, BASF). The low rate of fenpropimorph was selected to control established mildew but without scorching leaves. All applications were made in 225 l ha^{-1} water by hand operated gas-pressured sprayer. As the disease of primary interest was *R. secalis* all plots at Rosemaund were treated with metrafenone (full manufacturers recommended rate; 0.5 l ha^{-1} Flexity, BASF) at GS 25/30 to prevent powdery mildew infection without affecting *R. secalis*. However, as there was no evidence of any *R. secalis* infection at Aberdeen by GS 30 in both 2007 and 2008, plots were not treated with metrafenone and powdery mildew epidemics were allowed to develop.

In 2006 at Rosemaund the experiment was laid out in a split-plot design with N as the main plot and seed rate plus fungicide treatments fully randomised within sub-plots. At Rosemaund in 2007 and at Aberdeen in 2007 and 2008 the design was a split-split plot with N as whole plots, seed rate as sub plots and fungicide treatments as sub-sub plots. In all years at each site there were 4 replicate blocks.

Sampling and measurements

Disease and green area assessments

Disease was assessed at approximately two week intervals commencing at GS 31 and coinciding broadly with GS 39/45, 59/65, and 71/75. Ten plants (or ten shoots after GS 39) were sampled at random from along the entire length of plots. Disease was assessed on one side of each fully unfolded mainstem leaf by estimating visually the % area occupied by sporulating disease lesions, excluding the area of associated chlorosis. The latter was accounted for in a separate assessment of the % green area that considered both natural and disease-induced chlorosis and necrosis. Disease was not assessed on leaves with advanced senescence, as identification becomes unreliable, however, leaves were recorded as being present and scored for % green area. Any disease on the stem was also recorded.

Canopy senescence assessments were begun after the final disease assessment and when the flag leaf had begun to yellow. In field assessments of the % green area of the flag leaf and the stem were made two to three times a week at two locations per plot until complete senescence of the canopy had occurred in all plots. The date when the leaves and then stem had lost green colour was recorded.

Radiation interception

Interception of photosynthetically active radiation (PAR) was determined within a day or two of the disease and % green area assessments outlined above. Simultaneous measurements of PAR were made above the canopy, and at ground level below the canopy, between 10.00 and 14.00 hours, using a Sunscan Canopy Analysis System (Delta T Devices, Cambridge, UK). The measurements of transmitted PAR were made at 8-10 randomly selected locations along the length of the plot at an angle of approximately 45^{o} to the crop rows.

Biomass, leaf area and water soluble carbohydrates

Approximately a third to a half the plot length was designated for taking destructive quadrat samples for biomass and leaf area determination, the rest was reserved for combining. The end of the plot designated for sampling was randomised from block to block. Destructive samples were taken at GS 31, GS 39/45 and GS 59/65 (post anthesis sampling is described in a separate section below). Plants were sampled from 3 ‘quadrats’ (1 m x 2 rows) located along the length of the designated sampling area which were bulked and processed as one. The ‘quadrats’ were positioned at least 0.5 m from the edge the plot and previous sampling areas, and more than 1.0 m from tramlines and the ends of the plot.

Plants were pulled up with their basal roots still attached, and placed into plastic bags to prevent moisture loss and taken to the laboratory for analysis. Samples were processed immediately or stored in sealed plastic bags in the dark at 4 °C to await analysis. All growth analysis was completed within 5 days of sampling. After washing soil from the base of shoots the tissue was gently blotted dry and weighed to the nearest 0.01 g. Plants were counted and divided into two subsamples by weight; subsample 1 (SS1) and subsample 2 (SS2). At GS 31,

SS1 was 20% of the total and SS2 the remainder; at GS 59, SS1 was 10% of the total and SS2 20%. Each subsample was weighed fresh before excising the roots and reweighing. The roots were discarded and the number of potentially viable and dead and dying shoots counted. A dead or dying shoot was classed as one with no green material, or where its newest expanded leaf had begun to senesce. The potentially fertile shoots and dead and dying shoots in SS2 were weighed fresh, dried in a forced draft oven at 80^{o}C for 48 h and reweighed. At GS 31 the potentially fertile shoots of the SS1 subsample were divided into leaf laminae, stem plus leaf sheath fractions and the projected area of each fraction measured using an automated leaf area meter (Delta T Devices, Cambridge, UK). Any leaf tissue that had completely senesced was measured and recorded separately. Each tissue fraction was then dried at 80^{o}C for 48 h and weighed. The area and dry weight of dead and dying shoots was determined separately. At later growth stages, the potentially fertile shoots were separated into zones representing each of the upper 5 culm leaf layers within the canopy. Thus zone 1 included the flag leaf (leaf 1) and the stem material from the base of the flag leaf up to the collar of the ear; zone 2 included leaf 2 and the stem material from the base of leaf 2 up to the base of the flag leaf etc. The bottom zone comprised leaf 5, the stem section and any senescent basal leaves below leaf 5, and the stem between leaf 5 and leaf 4. At GS 59/65 ears plus awns were taken as a separate fraction. The projected area of stem and laminae in each zone was measured separately. In zones 1-4 the area measured included diseased, senescent and healthy tissue. In zone 5 basal senescent leaves were measured separately from leaf 5. This ensured that the stratification of the leaf area measurements corresponded with the disease and % green leaf area assessments described above. After determining its area, each fraction was dried at 80^{o}C for 48 h and weighed. The area and dry weight of dead and dying shoots was determined separately without stratification by leaf layer. Water soluble carbohydrates were determined on a random sample of ten shoots per plot taken at GS 59 as described in section 3.1.

Post anthesis shoot biomass (Aberdeen 2007, 2008)

At weekly intervals commencing at GS 59, ten (2007) or twenty (2008) shoots were sampled at random from along the length of the plot. Shoots were divided into ear and stem plus leaf fractions and each fraction dried at 80^{o}C for 48 h before weighing.

Pre-harvest assessments

Ear numbers m^{-2} were determined shortly before final harvest by counting the number of ears along a 0.5 m cane placed between two rows of plants. Counts were made in the row on each side of the cane, and at six locations selected at random along the entire length of each plot. The outer two rows were avoided to minimise edge effects.

Grain yield and quality

At crop maturity, the area of each plot designated for yield determination was harvested using a small plot combine. A sample of grains was taken for determination of mean grain weight and moisture content. Data are expressed on a 100% dry matter basis.

Meteorological records

Weather data were recorded at, or within a km of, the site. Measurements were made of daily rainfall, daily maximum, minimum and mean temperature and total incident solar radiation. Incident photosynthetically active radiation (PAR) was estimated as 0.5 x incident solar radiation.

Calculations and data analysis

Crop growth and yield components

Above ground biomass and canopy area index (CAI, tissue projected area/unit ground area, includes diseased and healthy leaf lamina, stem plus leaf sheaths and ear plus awns when present) were calculated from the SS2 dry weight and SS1 projected area measurements after adjusting for the sub-sampling using the subsample/total sample fresh weight ratio. The healthy leaf area index (HAI)

was calculated by adjusting the CAI by the % green area of the canopy. At GS 31 an average % green area for the whole canopy was used based on scores for the stem and top 5 leaves and the total canopy CAI. At GS 39/45 and GS 59/65, values of CAI for ears (when present), stem and leaves in individual zones down the canopy were adjusted by their % green area score and summed to give the canopy HAI. The yield component grains m^{-2} was estimated as the grain yield/mean grain weight.

At GS 31 and during the latter half of grain filling some of the lower leaves had senesced completely making reliable assessment of disease difficult. Therefore, for consistency of presentation, disease severity has been summarised by averaging over the top 3 leaves at each growth stage unless stated otherwise, and % green area over the top 4 leaves.

Post-anthesis above-ground biomass was calculated from the weekly shoot samples as:

$$\textit{biomass shoot}^{-1} \textit{ x no. fertile shoots } m^{-2} \qquad (1)$$

In order to achieve comparability of biomass estimates pre and post anthesis, data for biomass calculated using equation 1 were normalised to those estimated from quadrat samples using values of total biomass and biomass per shoot taken at the same sampling time (i.e. GS 59/65). Post-anthesis sampling was of potentially fertile shoots, whereas quadrat sampling of biomass included potentially fertile and dead plus dying shoots. However the dead and dying shoots accounted for only 0.3% of the total biomass and the proportion did not differ between seed rate, N and fungicide treatments. Thus the type of shoot sampled was not an important source of error in comparing biomass estimates pre and post-anthesis.

PAR interception

An equipment malfunction meant that fractional PAR interception data were not available for Rosemaund in 2006. Long spells of rainfall also meant that PAR interception measurements were possible only for some, but not all, sample occasions at Rosemaund in 2007.

At GS 31, GS 39/45 and GS 59/65 a canopy light extinction coefficient (*k*) was calculated for each plot from measured values of fractional PAR transmission and CAI as

$$k = ln\ (I/I_0)/CAI \qquad (2)$$

where I_0 is the incident PAR and *I* is the PAR transmitted to the base of the canopy. The fraction of PAR intercepted (*F*) by healthy tissue was then estimated from measurements of HAI and *k* using the Beers law analogy

$$F = 1\text{-}exp\ (-k*HAI) \qquad (3)$$

Healthy area PAR interception (HAint) was then estimated in daily time steps over defined developmental periods as:

$$HAint = F*I_0 \qquad (4)$$

Interpolation pre-anthesis

Daily values of HAI were estimated from the date of 50% crop emergence to GS 31 using accumulated thermal time (base temperature 0^oC) over the period and measured values of HAI at GS 31. HAint was then calculated using the value of *k* at GS 31, daily HAI and incident PAR. Estimates of HAint were made for the intervals GS 31 – GS 39/45 and GS 39/45 – GS 59/65 by linear interpolation of HAI and *k* from measured values at GS 31, GS 39/45 and GS 59/65.

Interpolation between assessment dates post-anthesis

Values of *k* were calculated from measured values of PAR interception during grain filling and the CAI measured at GS 59/65. CAI was assumed to remain constant after GS 59, whilst % green area declined as a result of natural and disease-induced senescence. Thus HAI during grain filling was determined from disease and % green area scores at mid and late grain filling and CAI at GS 59. Linear interpolation was used to estimate daily values of HAI and *k* between individual assessment dates and the date of final canopy senescence for

calculation of daily HAint. Daily values of HAint were summed to give the total for the post-anthesis period.

Radiation use efficiency and potential post-anthesis assimilate supply

Radiation use efficiency (RUE) was determined for each treatment as the slope of the relationship between biomass gain and the PAR intercepted by healthy tissue between GS 31 and GS 85/87. Data from GS 85/87 onward were excluded from the analysis because of the possible shedding of senesced leaves, and consequent loss of biomass after this time. Potential assimilate supply (PAS) for grain filling per unit grain number was calculated as described in section 3.1. Because a complete data set for post-anthesis PAR interception at Rosemaund was not available, analysis of RUE and potential assimilate supply was conducted for the Aberdeen site only.

Statistical analysis

Statistical analysis of the effects of seed rate, N timing and fungicide treatment, and the interactions between them, on components of source and sink was conducted using GenStat 11.1 (VSN International Ltd, Hemel Hempstead, UK). Data were analysed by ANOVA, initially for individual site-years. A cross site-year analysis was then conducted using treatment means from individual site-year combinations. This sequential approach was taken rather than using individual plot data in a cross site analysis because a different experimental design was used at Rosemaund in 2006 (split plot) compared to the other site-years (split-split plot). Data were checked for normality and homoscedasticity and transformed prior to analysis as required. For ease of interpretation back-transformed mean values for treatments are presented. Simple linear regression and regression with groups has been used to analyse relationships between source and sink components and the significance of differences between slopes and intercepts. In regression with groups, models with a common slope and intercept were fitted first, followed by models with separate intercepts but a common slope, and finally using separate intercepts and slopes. The P value reported is for the improvement in % variance accounted for by each step in the process.

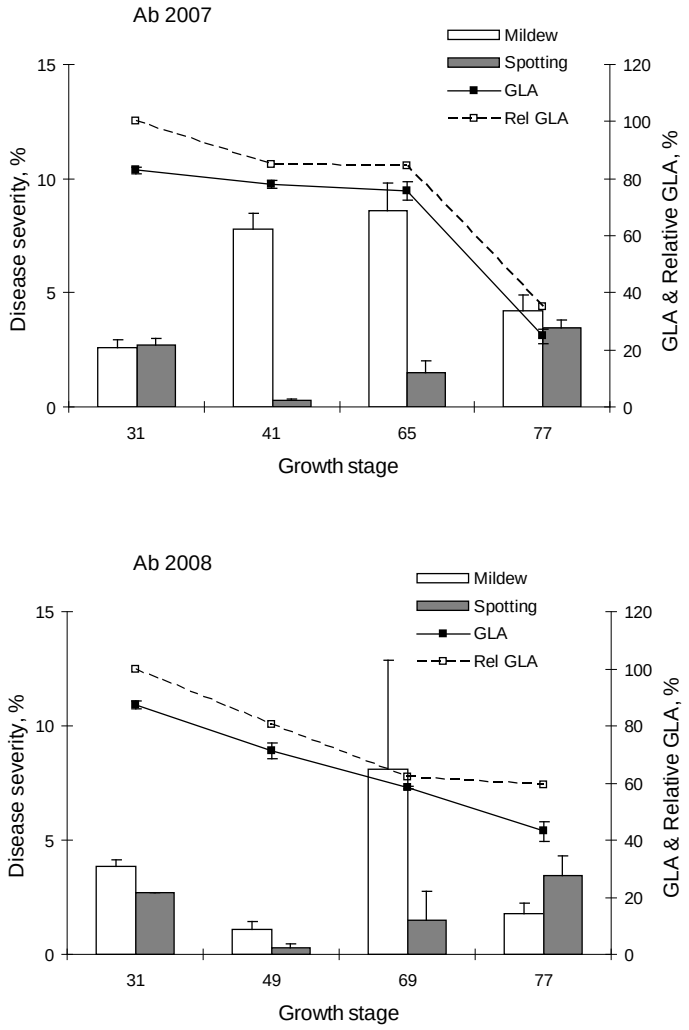


Figure 10. Disease severity (columns) and % green leaf (% GLA, symbols and lines) in non-fungicide treated plots at Aberdeen. Disease severity is the % surface area covered by lesions averaged over the top 3 fully unfolded leaves (except GS 77 where it is top 2 leaves only). % GLA is the % green leaf area averaged for the top 4 leaves. Relative % GLA is the GLA of non-fungicide treated plots relative to that of plots given a T1 plus T2 fungicide. Values are means across seed rate and N regime; vertical bars are SE. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

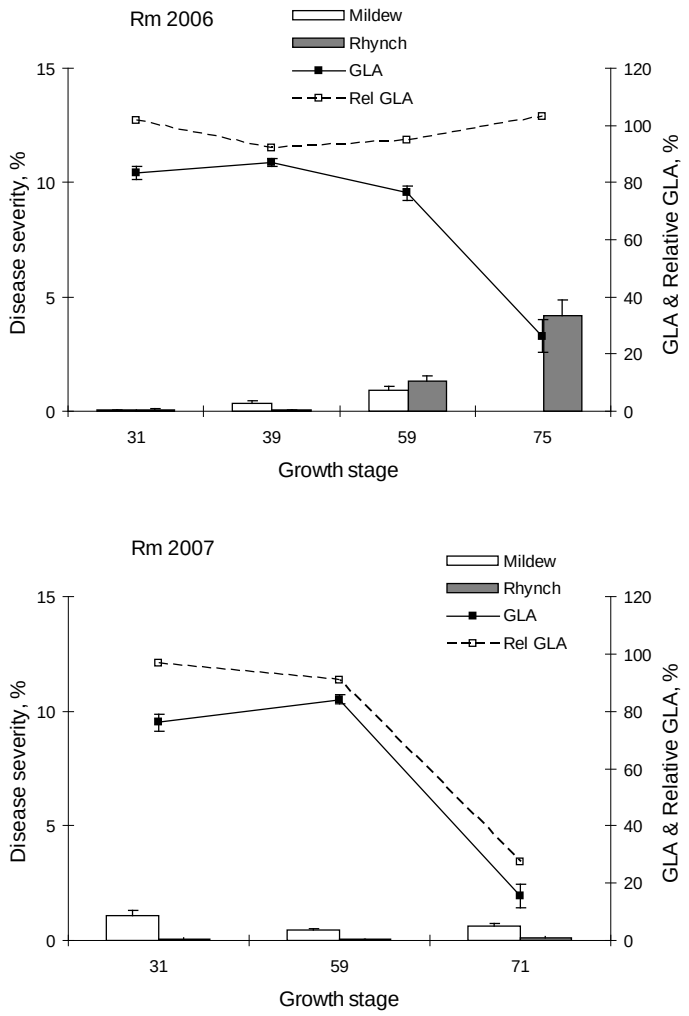


Figure 11. Disease severity (columns) and % green leaf (% GLA, symbols and lines) in non-fungicide treated plots at Rosemaund. Disease severity is the % surface area covered by lesions averaged over the top 3 fully unfolded leaves (except GS 75 in 2006 where it is the top 2 leaves). % GLA is the average % green leaf area averaged for the top 4 leaves. Relative % GLA is the GLA of non-fungicide treated plots relative to that of plots given a T1 plus T2 fungicide. Values are means across seed rate and N regime; vertical bars are SE. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

3.3.3 Results

Disease and % green leaf area

At Aberdeen in both 2007 and 2008, no rhynchosporium leaf scald had developed in the crop by GS 30. Consequently the application of metrafenone (Flexity) was omitted and powdery mildew epidemics were allowed to develop. The severity of mildew was high in untreated plots (Fig. 10) reaching close to 10% averaged over the top 3 leaves, by flowering in each year. The epidemic persisted into the grain filling period with severities of 4 and 2% in 2007 and 2008 respectively for the top 2 leaves. There was also a significant amount of necrotic spotting on leaves in each year. The cause of the spotting was not diagnosed, but is likely to include physiological spotting, old mildew lesions and ramularia (*Ramularia collo-cygni*) depending on the crop growth stage. The % green leaf area (GLA) for the top 4 unfolded leaves was around 80% prior to flowering in 2007 and then fell to 25% by mid grain fill. In 2008 there was a steady decline in % GLA from GS 31 onwards. In each year the decline in % GLA was associated primarily with disease-induced chlorosis and necrosis since there was a parallel decline when the data were expressed relative to the GLA of fungicide treated plots (Fig. 10).

At Rosemaund there were traces of rhynchosporium in the crop at GS 30 and thus the crop was treated with metrafenone (Flexity) to control mildew in the expectation that a rhynchosporium epidemic would develop. In 2006, significant leaf scald did develop, but only late in the season with the greatest severities occurring after ear emergence (GS 59) (Fig. 11). In 2007, a rhynchosporium epidemic failed to develop.

The % GLA at Rosemaund in 2006 was between 76 and 87% between GS 31 and 59, thereafter it fell to 25% (Fig. 11). The decline after ear emergence did not appear to be associated principally with the rhychosporium infection, because there was a comparable decline in % GLA of fungicide-treated plants as shown by the high values of relative % GLA at GS 75 (Fig. 11). By contrast, in 2007, the decline in % GLA after ear emergence was considerably greater in untreated plots than treated ones, as shown by the large decline in relative % GLA between GS 59 and 71. The reason for this rapid fall in % GLA of untreated plants in the

Table 20. Effects of fungicide treatments on the severity of the main disease (% area covered by lesions, averaged for top 3 leaves at GS 59/69 & top 2 leaves at GS 71/77) and % green leaf area (GLA, averaged for the top 4 leaves). Values are main effect means averaged over seed rate and N regimes. Significance of fungicide interactions with N and seed rate (SR) are shown. Significance levels are ***, P<0.001; ** P<0.01; * P<0.05; ns P>0.05. LSDs for fungicide treatments are at P = 0.05. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Growth stage	Treatment	Ab 2007		Ab 2008		Rm 2006		Rm 2007
		Mil	GLA	Mil	GLA	Rhynch	GLA	GLA
GS 59/69	None	7.7	75.9	7.5	58.8	1.3	77.2	84.1
	T1	1.1	88.5	1.5	90.8	0.9	86.3	85.6
	T2	6.6	76.7	4.4	69.1	1.4	73.1	72.4
	T1 + T2	0.8	89.9	0.6	94.3	0.8	81.3	80.5
	lsd	*2.9*	*3.1*	*2.0*	*3.9*	*0.5*	*4.0*	*3.3*
GS 71/77	None	4.2	24.8	1.8	43.0	4.5	26.2	15.5
	T1	1.2	51.4	2.3	51.1	4.7	36.9	27.5
	T2	2.4	45.9	1.8	54.2	3.4	31.5	48.4
	T1 + T2	0.6	70.9	0.6	72.3	1.6	25.4	56.2
	lsd	*1.2*	*4.2*	*1.0*	*8.1*	*1.8*	*7.0*	*7.0*
GS 59/69	F	***	***	***	***	*	***	***
	N*F	ns	ns	ns	ns	ns	**	ns
	SR*F	ns	ns	ns	**	ns	ns	ns
GS 71/77	F	***	***	*	***	**	**	***
	N*F	ns	ns	ns	ns	ns	ns	ns
	SR*F	ns	ns	ns	ns	ns	**	***

absence of significant disease is not known with certainty. It could be associated with mildew infection that went unrecorded if mildew pustules were washed off leaves by heavy rain prior to assessment.

In general there was little effect of N or seed rate on disease severity in non-fungicide treated plants (data not shown). Fungicide significantly reduced disease severity at all sites except Rosemaund in 2007 where there was little disease present (Table 20). At the GS 59/69 assessment, the greatest reduction was found with the T1 application either alone, or in combination with T2. By contrast, T2 had little effect on disease severity probably because of the relatively short time interval between application and assessment. The T1 application gave a corresponding increase in % GLA compared to untreated plants at each site except Rosemaund in 2007. Here there was a significant reduction in % GLA with T2 and T1+T2 compared to untreated plants suggesting some possible temporary leaf scorch from the T2 application.

At the GS 71/77 assessment, the greatest effect of fungicide on disease severity on the top two leaves was found with a combination of T1 and T2. Single applications at T1 and T2 either had no significant effect on disease severity compared to untreated controls (Ab 2008 and Rm 2006) or the reduction was less than that with T1 + T2 (Ab 2007). Similarly T1 + T2 in general resulted in a greater % GLA than either application alone. The exception was at Rosemaund in 2006 where there was an unusually low % GLA in the T1 + T2 treatment. There was no significant interaction between fungicide and either N regime or seed rate on disease severity (Table 20). Some significant interactions were found between fungicide and seed rate on % GLA, but they were inconsistent and varied between site-years and crop growth stage. However, in each case, the interaction resulted from a greater increase in % GLA with fungicide (relative to untreated controls) at high seed rate compared to low seed rate.

Of the agronomic treatments, seed rate and N regime, seed rate had the greater effect on % GLA. In fungicide treated plots, low seed rate resulted in a greater post anthesis % GLA compared to high seed rate in 3 of the 4 site-years (Fig. 12).

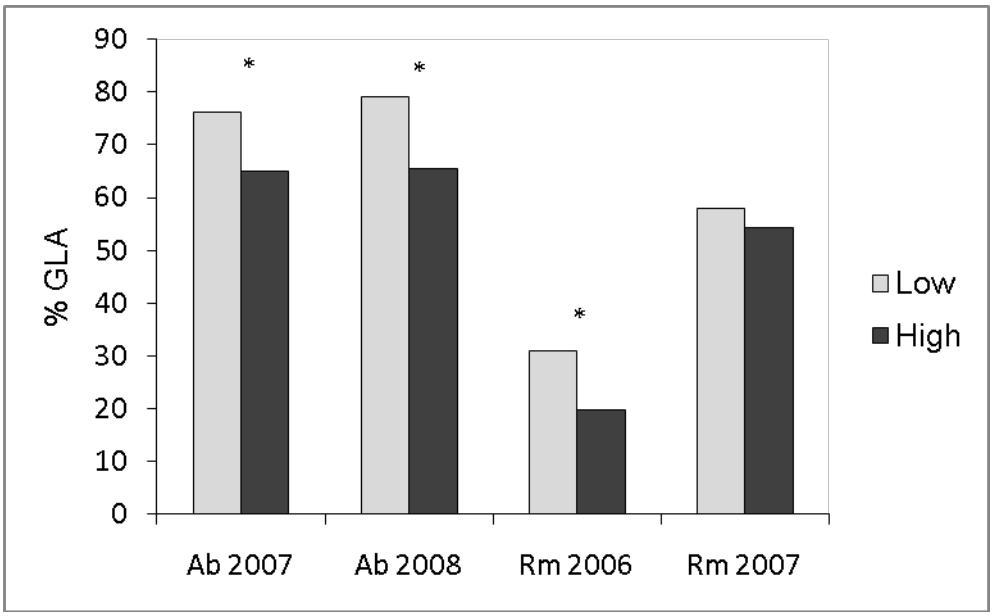


Figure 12. Effects of seed rate on % GLA averaged over the top 4 leaves at GS 71/77. Values are for plots given a T1 + T2 fungicide application. * indicates a significant difference between seed rates at P = 0.05. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Yield and yield components

When averaged across N regime and seed rates, fungicide application significantly increased yield relative to non-fungicide treated controls in each site-year (Table 21). At Aberdeen in both 2007 and 2008, there was a significant yield response to a single application made at either T1 or T2, but in each year the greatest response (0.9-1.6 t ha^{-1}) was to the T1 + T2 treatment. At Rosemaund in 2006 there was a significant yield response to T1, but not T2. In 2007 both T1 and T2 applications increased yield relative to untreated controls, but the combined T1 plus T2 application was no more effective than either treatment on its own.

Table 3.2. Grain yield (t ha^{-1} @100% DM). Values are main effect means for N regime, seed rate and fungicide treatments averaged across the other treatments. Significance levels are ***, P<0.001; ** P<0.01; * P<0.05; ns P>0.05. LSDs for fungicide treatments are at P = 0.05. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Treatment	Level	Ab		Rm	
		2007	2008	2006	2007
N	Early	5.7	4.8	6.0	5.0
	Late	5.8	4.4	5.5	5.0
Seed rate	Low	5.3	4.2	5.2	4.4
(SR)	High	6.3	5.0	6.3	5.6
Fung	None	4.9	4.1	5.5	4.1
(F)	T1	5.9	4.4	6.0	5.1
	T2	5.7	4.7	5.6	5.3
	T1 + T2	6.5	5.0	6.0	5.5
	df	Significance (LSD, P = 0.05)			
N	3	ns	**	ns	ns
SR	6	***	***	***	***
F	36	*** (0.14)	*** (0.27)	*** (0.21)	*** (0.49)
N*F	36	ns	***	ns	ns
SR*F	36	***	ns	ns	ns

Table 22. Grain numbers per m^2. Values are main effect means for N regime, seed rate and fungicide treatments averaged across the other treatments. Significance levels are ***, P<0.001; ** P<0.01; * P<0.05; ns P>0.05. LSDs for fungicide treatments are at P = 0.05. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Treatment	Level	Ab		Rm	
		2007	2008	2006	2007
N	Early	16146	12475	15031	12542
	Late	16905	13222	14487	12816
Seed rate	Low	14196	11080	12116	11008
(SR)	High	18856	14616	17406	14350
Fung	None	15451	11855	14409	10527
(F)	T1	17079	12770	15047	13107
	T2	15725	13095	14502	13450
	T1 + T2	17848	13673	15079	13632
	df	Significance (LSD, P = 0.05)			
N	3	ns	ns	ns	ns
SR	6	***	***	***	***
F	36	*** (936)	*** (523)	*** (419)	*** (1133)
N*F	36	ns	*	*	ns
SR*F	36	**	ns	ns	ns

Table 23. Mean grain weight (mg @100% DM). Values are main effect means for N regime, seed rate and fungicide treatments averaged across the other treatments. Significance levels are ***, P<0.001; ** P<0.01; * P<0.05; ns P>0.05. LSDs for fungicide treatments are at P = 0.05. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Treatment	Level	Ab		Rm	
		2007	2008	2006	2007
N	Early	35.95	38.46	40.28	39.98
	Late	34.89	33.46	39.11	38.89
Seed rate	Low	37.39	37.87	43.13	39.93
(SR)	High	33.46	34.05	36.26	38.94
Fung	None	32.58	35.29	38.61	38.94
(F)	T1	35.27	35.04	40.43	39.12
	T2	36.49	36.58	39.02	39.58
	T1 + T2	37.45	36.94	40.73	40.09
	df	Significance (LSD, P = 0.05)			
N	3	*	**	ns	ns
SR	6	***	**	***	ns
F	36	*** (1.93)	* (1.32)	*** (0.90)	ns
N*F	36	ns	ns	ns	ns
SR*F	36	ns	ns	ns	ns

Table 24. Cross site analysis of treatment effects on yield and yield components. Values are main effect means for N regime, seed rate and fungicide treatments. Significance levels are ***, P<0.001; ** P<0.01; * P<0.05; ns P>0.05. LSDs for fungicide treatments are at P = 0.05.

Treatment	Level	Yield t ha^{-1}, 100% DM	Grains m^{-2}	MGW mg, 100% DM
N	Early	5.4	14048	38.67
	Late	5.2	14358	36.59
Seed rate	Low	4.8	12100	39.58
(SR)	High	5.8	16306	35.68
Fung	None	4.7	13060	36.36
(F)	T1	5.4	14501	37.46
	T2	5.3	14193	37.92
	T1 + T2	5.7	15058	38.78
	df	Significance (LSD, P = 0.05)		
N	45	*	ns	***
SR	45	***	***	***
F	45	*** (0.25)	*** (746)	*** (1.43)
N*F	45	ns	ns	ns
SR*F	45	ns	ns	ns

The relative contribution of the yield components grains m^{-2} and MGW to the overall yield response to fungicide differed between sites and years, although the cross site analysis (Table 24) indicated that effects on grain numbers accounted for around 68-75% of the yield response observed with each fungicide treatment. At Aberdeen in 2007 and Rosemaund in 2006, T1 applications increased grain numbers significantly, whereas T2 on its own had no effect. By contrast, at Aberdeen in 2008 and Rosemaund in 2007, T1 *and* T2 as single applications increased grain numbers.

Effects of fungicide treatments on MGW also differed between site-years (Table 23). There was no significant effect of any fungicide treatment on MGW at Rosemaund in 2007. MGW was increased by T1 at Rosemaund in 2006, by T2 at Aberdeen in 2008, and by both T1 and T2 applied alone at Aberdeen in 2007.

When averaged over the other treatments, high seed rate resulted in a signficantly greater yield in each site-year compared to low seed rate, whereas the effects of N regime on yield were small and inconsistent (Table 21). High seed rate increased grain numbers m^{-2}, but reduced MGW in 3 of the 4 site-years compared to low seed rate. N regime, on the other hand, had no signficant effect on grain numbers, but early N did result in a significantly greater MGW at Aberdeen compared to late N.

There were few significant interactions between fungicide treatment and either N regime or seed rate on yield (Table 21). Where an interaction was found it tended to be the result of effects of seed rate or N regime on the scale of the response to fungicide rather than its direction (Appendix). For example a significant SR x F interaction was found in Aberdeen 2007 where the response to fungicide, especially the T1 + T2 application, was greater at high seed rate compared to low. At Aberdeen in 2008 there was a significant N x F interaction where the increase in yield with T1 was significant only under the early N regime not the late N. Both these interactions were associated with effects on grain numbers m^{-2} (Table 22). The interactions were not consistent or strong enough to carry through to the cross site analysis (Table 24). Importantly, there was no significant interaction between N or seed rate and fungicide on MGW (Table 23) indicating that the response of MGW to fungicide was not influenced by treatments designed to vary the source-sink balance of the crop.

Table 25. Effects of treatments on the final number of ears m^{-2} at harvest. Values are main effect means for a treatment. Significance levels are ***, P<0.001; ** P<0.01; * P<0.05; ns P>0.05. LSDs for fungicide treatments are at P = 0.05. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Treatment	Level	Ab	Rm		
		2008	2006	2007	
N	Early	932	861	718	
	Late	1252	847	877	
Seed rate	Low	950	710	696	
(SR)	High	1233	999	898	
Fung	None	1089	889	677	
(F)	T1	1121	805	797	
	T2	1034	842	832	
	T1 + T2	1124	881	882	
	df	Significance (LSD, P = 0.05)			
N	3	***	ns	**	
SR	6	***	***	***	
F	36	ns	ns		*** (85.8)
N*F	36	ns	ns	ns	
SR*F	36	*	ns	ns	

Data for final ear number are available for just 3 site-years (Table 25). Both T1 and T2 fungicide treatments increased ear numbers by 18-23% at Rosemaund in 2007. There was no significant effect of fungicide at Rosemaund in 2006 or Aberdeen in 2008. High seed rate resulted in a larger (average 33%) ear number compared to low seed rate. Late N increased ear numbers relative to early N in two of the three site years for which there are data.

Pre-anthesis growth of source and sink components

Results of a cross site analysis of the effects of N regime, seed rate and fungicide treatment on canopy growth and stem water soluble carbohydrate reserves are given in Tables 26 and 27. Above ground biomass and healthy

canopy area (HAI) were greater with early N compared to late N and at high seed rate compared to low. With early N the greater HAI was associated with fewer shoots but larger area per shoot, whereas the greater HAI at high seed rate was the result of a larger number of smaller shoots (Table 26). T1 fungicide (alone and in combination with T2) increased biomass and HAI relative to untreated controls. The greater HAI was the result of a comparable increase in the number of shoots m^{-2} (9-13%) and healthy area per shoot (11-13%), although only the latter was statistically significant ($P<0.05$). When the results were averaged across sites, the concentration (% DW) of water soluble carbohydrates in stem tissue was affected relatively little by any of the treatments, although the concentration was slightly greater (16%) at high seed rate compared to low (Table 27). However, since stem biomass was influenced by N regime, seed rate and fungicide treatments, there were significant effects of each of these treatments on the quantity of water soluble carbohydrates per m^2. The quantity was greatest at high seed rate and with early N and following a T1 application of fungicide.

The quantity of WSC and healthy area expressed per unit eventual grain number represent the amount of soluble carbohydrate reserves and photosynthetically active surface area available for supplying each grain with carbon substrates at the start of the grain filling period. Early N increased the WSC reserves and healthy area per grain relative to late N. Seed rate had no effect on WSC reserves per grain, even though the total quantity of reserves was greater at high seed rate compared to low, because there was an equivalent increase in the number of grains m^{-2} at high seed rate. There was, however, a smaller healthy area per grain at high seed rate. Fungicide treatment had no significant effect on either WSC or healthy area per grain.

Table 26. Effects of treatments on canopy components at anthesis. Values are main effect means averaged over sites and years. Significance levels are ***, P<0.001; ** P<0.01; * P<0.05; ns P>0.05.

Treatment	Level	Biomass g m^{-2}	HAI	Fertile shoots m^{-2}	HA cm^2 $shoot^{-1}$
N	Early	870	4.38	848	54.5
	Late	771	4.05	914	48.4
Seed rate	Low	717	3.77	621	61.2
(SR)	High	924	4.67	1141	41.8
Fungicide	None	776	3.69	827	48.7
(F)	T1	850	4.61	900	55.0
	T2	786	3.83	860	48.2
	T1 + T2	870	4.73	937	54.0
	df	Significance (LSD, P = 0.05))			
N	45	***	*	*	***
SR	45	***	***	***	***
F	45	* (70)	***	ns	** (4.2)
N*F	45	ns	ns	ns	ns
SR*F	45	ns	ns	ns	ns

Table 27. Effects of treatments on stem water soluble carbohydrates (WSC) and healthy canopy area per unit eventual grain number at anthesis. Values are main effect means averaged over sites and years.

Treatment	Level	WSC, % DW	WSC g m^{-2}	WSC, mg $grain^{-1}$	HA cm^2 $grain^{-1}$
N	Early	22.4	115	8.46	3.56
	Late	21.4	93	6.61	3.26
Seed rate (SR)	Low	20.3	89	7.52	3.66
	High	23.5	119	7.55	3.16
Fungicide (F)	None	20.7	98	8.08	3.38
	T1	22.9	111	7.79	3.50
	T2	21.5	95	6.75	3.16
	T1 + T2	22.5	113	7.52	3.61
	df	Significance (LSD, P = 0.05)			
N	45	ns	***	***	*
SR	45	***	***	ns	***
F	45	ns	** (11)	ns	ns
N*F	45	ns	ns	ns	ns
SR*F	45	ns	ns	ns	ns

Relationship between pre-anthesis light interception and grain number formation

When seed rate was used to vary the plant population and canopy area, there was a positive linear relationship between the amount of PAR intercepted by healthy tissue pre-anthesis and the number of grains produced m^{-2}. However, a single relationship did not account for a particularly large proportion of the variation in grain number between site-years, N timing and fungicide treatments. At Aberdeen in 2007, parallel regression analysis indicated that

significantly more variation was accounted for when separate slopes and intercepts were fitted to data for N timing and fungicide treatments (only the untreated and T1+T2 fungicide regimes were considered) (Fig. 13). In 2008, there was a significant improvement when separate intercepts were included in the regression model, but not slopes. The results indicate that the effects of fungicide treatment on grain numbers cannot be explained simply in terms of protection of leaf area and increased PAR interception before anthesis. Thus, fungicide tended to increase grain numbers over a range of healthy area PAR interception similar to that observed for untreated plots. At Aberdeen in 2007 the effect was most pronounced at high seed rate giving rise to the greater slope after fungicide application (with the early N regime). At Aberdeen in 2008 and Rosemaund in 2007, the effect was similar at each seed rate resulting in a significant increase in elevation with fungicide, but not slope.

Late application of N had no significant overall effect on final grain numbers (Table 22), but at Aberdeen in 2008 and Rosemaund 2007 reduced the amount of radiation intercepted by healthy tissue pre-anthesis (Fig. 13). Thus, the regression lines for late N treatment were displaced to the left of those for the early N treatment. At Aberdeen in 2007 there was only one week between early and late N applications and thus, not surprisingly, there was little effect on PAR interception.

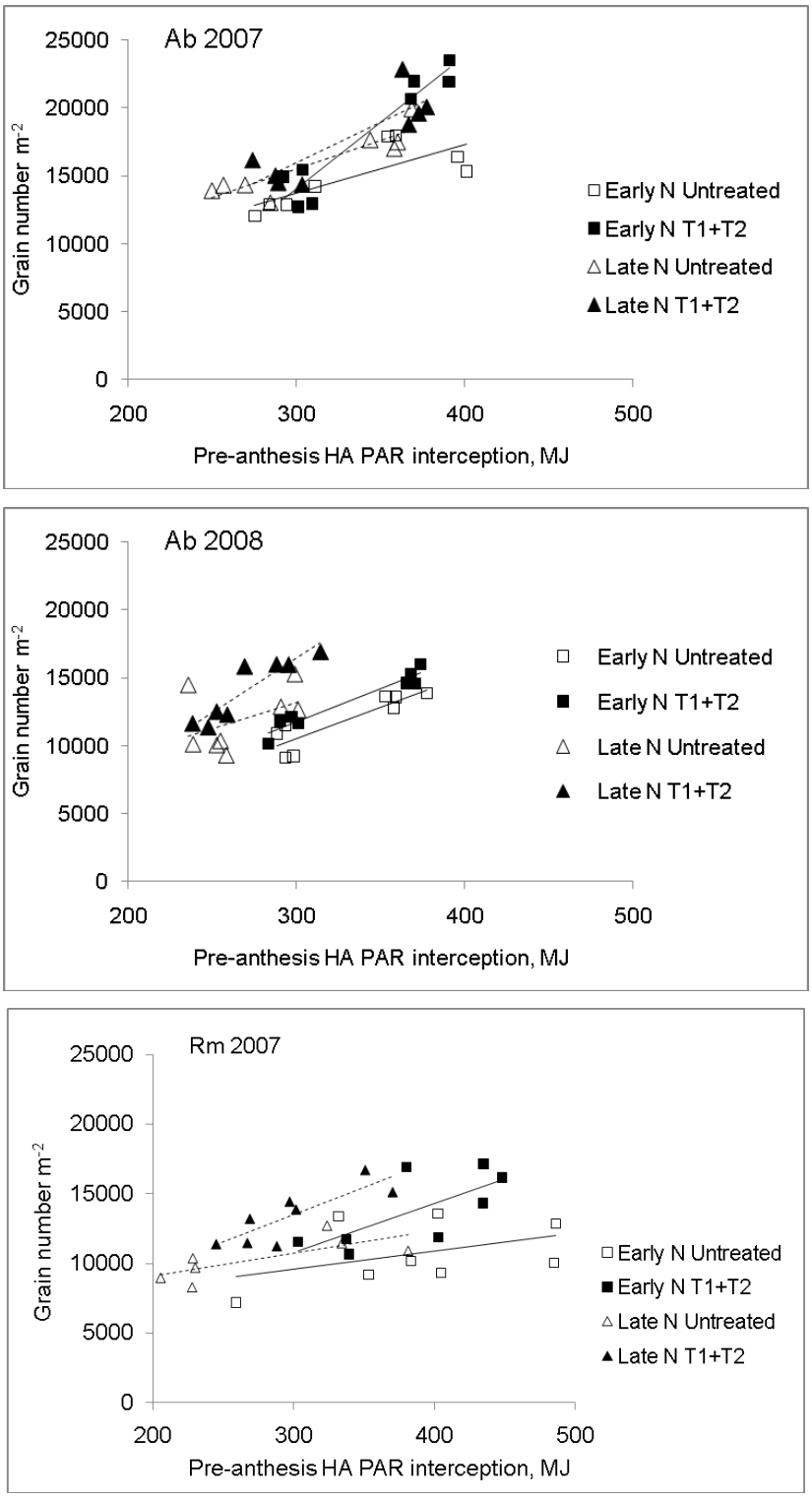


Figure 13. Relationship between PAR interception by healthy tissue from crop emergence to anthesis and eventual grain number. Separate relationships have been plotted for N regime and fungicide treatment combinations. Within a particular combination, points represent individual plot values from the high and low seed rate treatments. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Radiation use efficiency (RUE)

Radiation use efficiency at the Aberdeen site was determined from the slope of plots of above ground biomass against PAR interception by healthy tissue. Data for the period from GS 31 to maximum dry weight were included in the analysis. The latter was reached towards the end of grain filling. Later values were omitted from the analysis because there was a net dry matter loss from the crop during the period of grain ripening. A step-wise polynomial regression approach was used to analyse the data. Data were fitted initially with a first order polynomial and then a second order.

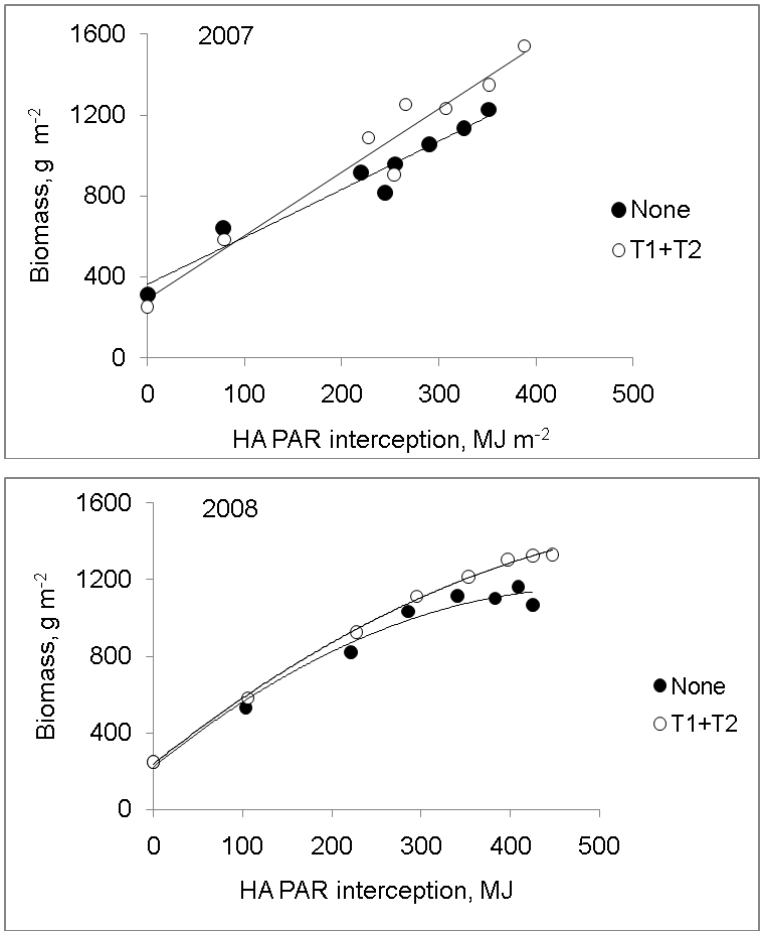


Figure 14. Relationship between accumulated above ground biomass and PAR interception by healthy tissue at Aberdeen for the period GS 31 to late grain filling. Data are for early N at high seed rate (other N and seed rate combinations show a similar pattern); T1 and T2 fungicide treatments are omitted for clarity. Each point is the mean of 4 replicates plots.

Table 28. Effects of fungicide treatments on RUE (g DW MJ^{-1} PAR intercepted) at Aberdeen in 2007. Data were analysed by linear regression with groups. Significance values refer to the improvement in variance accounted for when separate slopes and intercepts were fitted to the different fungicide treatments compared to models with a common slope. Values in italics give the common RUE when there was no significant difference in slope.

N regime	Seed rate	Fungicide	RUE	Significance	% variance
Early	Low	None	2.9	ns	96.3
		T1	2.7	*2.9*	
		T2	3.2		
		T1+T2	3.0		
Early	High	None	2.4	ns	93.8
		T1	2.6	*2.6*	
		T2	2.3		
		T1+T2	3.1		
Late	Low	None	2.6	P = 0.003	97.3
		T1	2.2		
		T2	2.6		
		T1+T2	3.2		
Late	High	None	2.5	P = 0.027	94.4
		T1	2.9		
		T2	1.9		
		T1+T2	2.8		

In 2007, there was no significant improvement in fit, for any of the treatment combinations, when the quadratic term was added to the regression model and a linear model was accepted (Fig. 14 for an example). In 2008, there was a distinct non-linearity to the plots (Fig. 14) and a significant improvement was found when a second order polynomial was fitted. Non-linearity was observed in each combination of seed rate and N regime, and for both fungicide treated and

untreated plots. The non-linearity implies that RUE declined over the measurement period.

Effects of fungicide treatment on RUE were examined for each seed rate-N combination at Aberdeen in 2007. Parallel regression analysis indicated that there was no significant difference between slopes (i.e. RUE) between fungicide treatments with early N at either high or low seed rate (Table 28). There were significant differences with late N, but they did not conform to a consistent and biologically plausible pattern with treatment. Plots given a T1 + T2 application generally had a high RUE, but it was not significantly greater than untreated plots. A comparable analysis of second order polynomials fitted to data from 2008 revealed significant differences between slopes for fungicide treatments only with the early N-low seed rate combination. At other seed rate-N combinations, the best fits were obtained with parallel curves of different intercepts. Thus there is no evidence of a clear and consistent effect of fungicide treatment on RUE.

The value of the quadratic term from 2nd order polynomials can be used as an indicator of the degree of non-linearity in RUE, even where the quadratic term does not represent a significant improvement in fit over a linear regression. When the quadratic term was plotted against the eventual grain number, a significant negative relationship was found, with grain number accounting for 54% and 29% of the variation in the quadratic term at high and low seed rate respectively (Fig. 15).

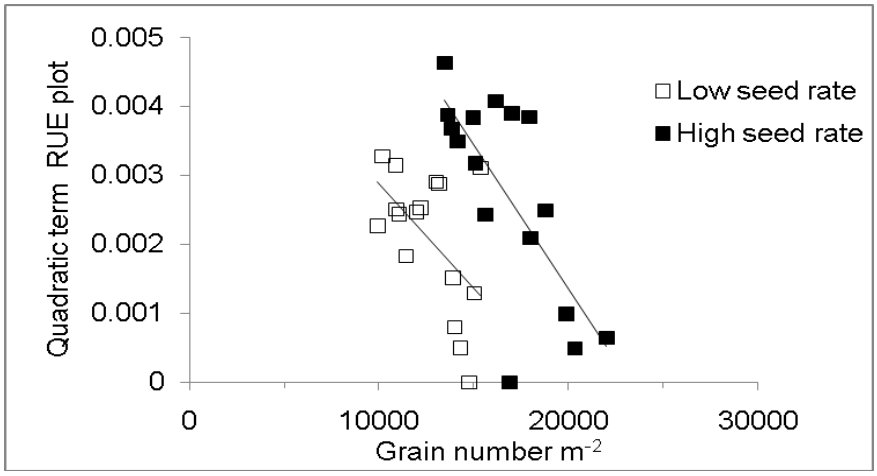


Figure 15. Relationship between value of the quadratic term in plots of biomass gain versus healthy area PAR interception (RUE) and final grain number. For clarity of presentation, the sign of the quadratic term has been ignored. Data pooled from 2007 and 2008. Lines fitted by linear regression; low seed rate $P<0.05$, $r^2 = 0.29$; high seed rate $P<0.05$, $r^2 = 0.54$.

Post anthesis PAR interception and potential assimilate supply

Although fungicide treatments increased the amount of PAR intercepted by healthy tissue post-anthesis in both 2007 and 2008, there was no significant effect when PAR interception was expressed per unit grain number (data not shown). By contrast, low seed rate increased PAR interception per grain by around 40% when compared to high seed rate. When data for seed rate and N regimes were pooled there was a positive linear relationship between MGW and PAR interception post-anthesis (Fig. 16). In 2007, parallel regression analysis indicated a significant difference in intercept between fungicide-treated and untreated plots, but no difference in slope. In 2008, there was no significant difference between treatments in either intercept or slope.

Post anthesis assimilate supply per unit grain number was estimated from the post-anthesis PAR interception, pre-anthesis RUE and the amount of water soluble carbohydrate reserves per grain at anthesis. There was a positive linear relationship between potential assimilate supply per grain and MGW in both

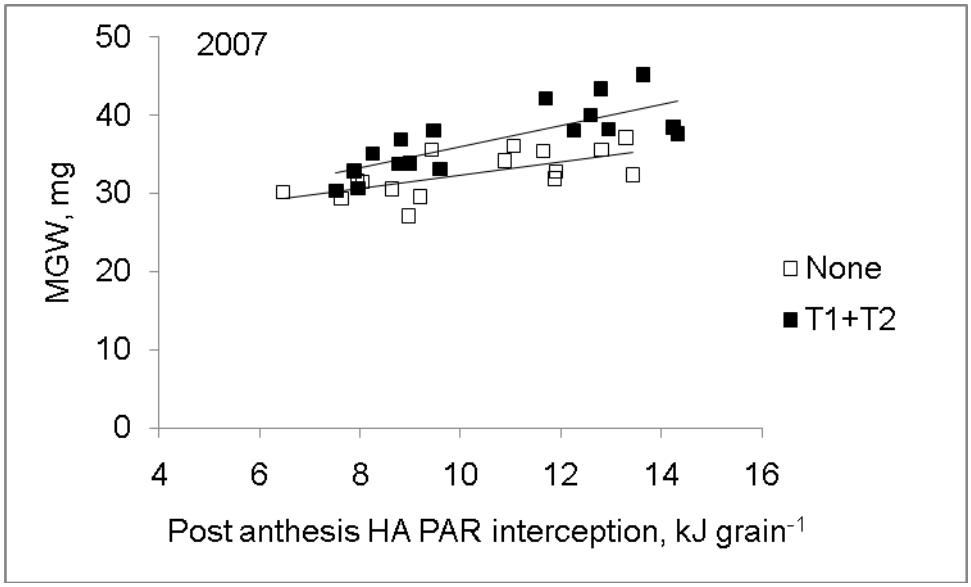
2007 and 2008 (Fig. 17). In 2007 the regression models for fungicide treated plots had a significantly greater intercept than that for untreated plots, but the slopes did not differ significantly. In 2008, a single model with common slope and intercept described well the data for untreated plots and plots given a T1+T2 application of fungicide (Fig. 17).

3.3.4 Discussion

Comparison of spring and winter barley

Although the type and severity of disease tended to differ between the spring barley experiments reported here and the winter barley experiments reported in section 3.1, there were a number of similarities in the growth and yield response to fungicide observed in the spring and winter crops. The yield response to the full fungicide treatment (T1+T2) in spring barley ranged from 0.5 to 1.6 t ha-1 (@100% DM) with an average of 1.1 t ha^{-1}. The response was associated with an increase in grain number m^{-2} in all site-years and an increase in MGW in 3 out of 4 site-years. However, the increases in grain number contributed most to the overall yield responses (50-85%). In winter barley the yield response to the full fungicide programme averaged over 4 site-years was 1.4 t ha^{-1}, of which an increase in grain numbers contributed 69% (section 3.1).

As with winter barley, the yield responses observed in spring barley were only weakly associated with the amount of visible disease present. For example, in 2007 comparable yield responses were found at Aberdeen and Rosemaund, yet disease severity was considerably greater at Aberdeen. At Rosemaund, in 2006 disease severity was in general slightly greater than in 2007, yet the yield response to fungicide in 2006 was only a third of that in 2007. Other similarities in the effects of fungicide treatments on the growth of the canopy, deposition of storage reserves and grain number formation by spring and winter crops are summarised in Table 29.



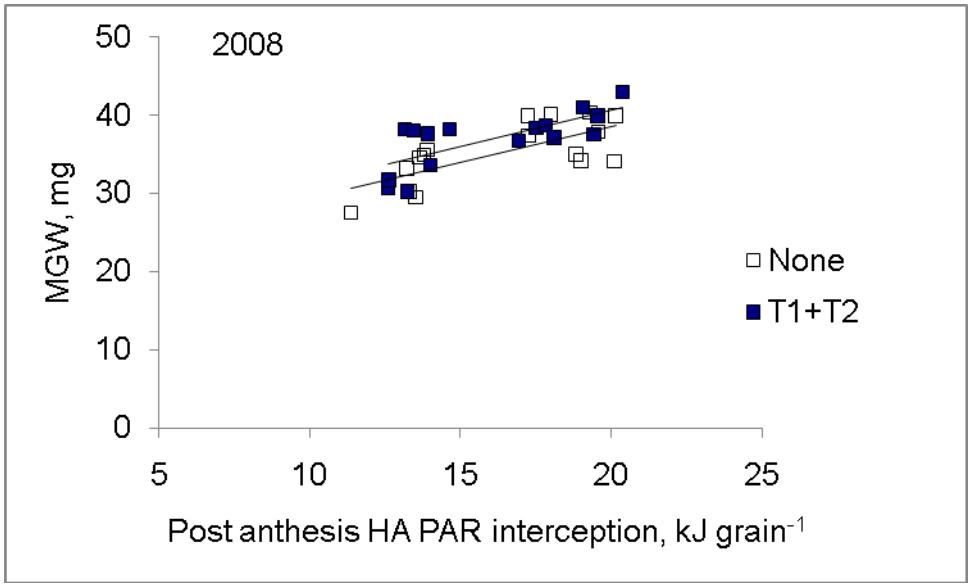
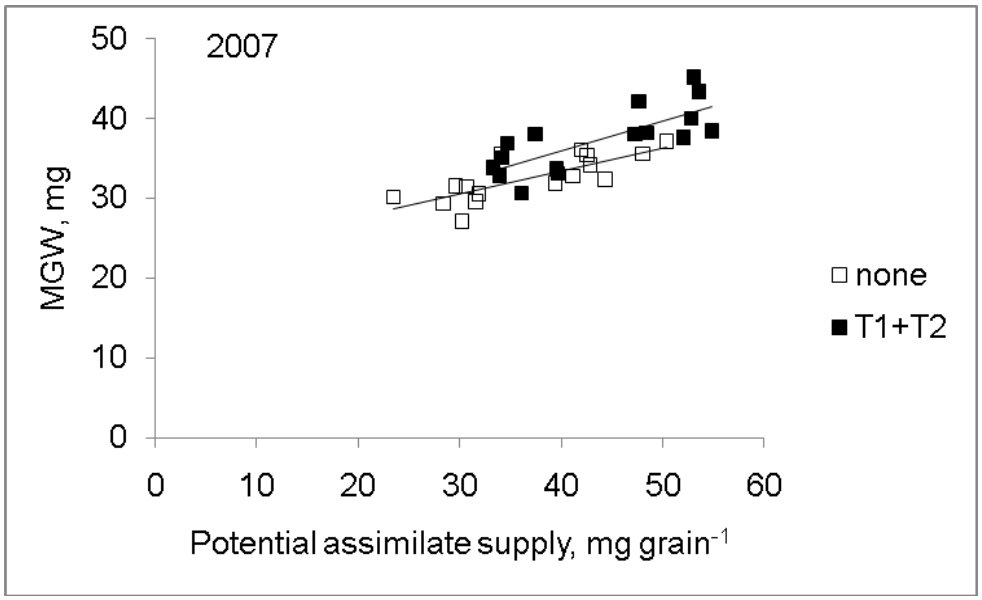


Figure 16. Relationship between post-anthesis PAR interception by healthy tissue, expressed per unit grain number, and MGW. Data for seed rate and N regimes have been pooled for analysis. For clarity only untreated (none) and T1+T2 fungicide treatments are shown.



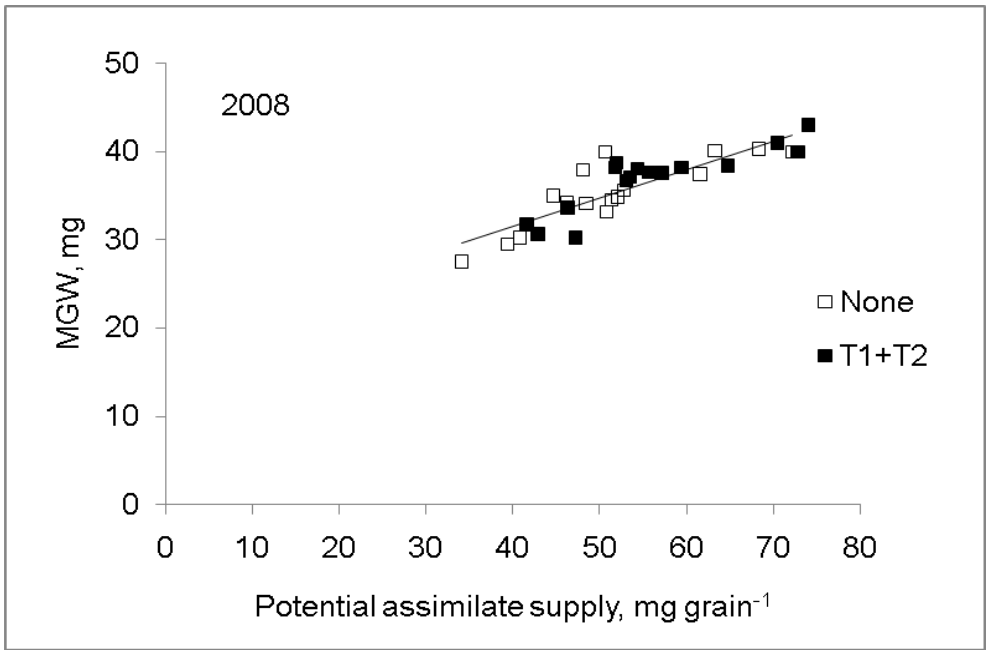


Figure 17. Relationship between potential assimilate supply post-anthesis, expressed per unit grain number, and MGW. Data for seed rate and N regimes have been pooled for analysis. For clarity only untreated (none) and T1+T2 fungicide treatments are shown. Lines fitted by least squares linear regression; 2007 none, y = 0.284x + 22.1, r^2 = 0.61; 2007 T1+T2, y = 0.375x + 21.0, r^2 = 0.59; 2008, y = 0.322x + 18.71, r^2 = 0.72.

Table 29. Summary of effects of fungicide treatment on eventual grain number and canopy components at GS 59 in winter and spring barley. Data are summarised from cross site-analyses in the current section (spring barley) and section 3.1 (winter barley) and are effects of fungicide averaged across seed rate and N treatments. A + indicates an increase with fungicide, ns indicates no significant effect of fungicide.

Component	Winter	Spring
HAI	+	+
Above-ground biomass	+	+
Fertile shoot number m^{-2}	+	ns
Eventual grain number m^{-2}	+	+
WSC (% DM)	ns	ns
WSC (g m^{-2})	+	+
HA per grain	+	ns
WSC per grain	ns	ns

Seed rate and N-fertilizer timing treatments had little effect on disease development in the spring barley experiments, but did have significant effects on crop growth, PAR interception and the deposition of soluble carbohydrate storage reserves. Even though there was little effect of these treatments on disease severity, significant interactions were observed between fungicide treatment and either seed rate or N regime on grain numbers in some site-years. Thus at Aberdeen in 2007, fungicide application had a greater effect on grain numbers at high seed rate than low, and in 2007 fungicide application had a greater effect with late N than early N. These effects are comparable to those found in winter barley (section 3.1), but the interactions were weaker and less consistent over site-years and thus did not lead to statistically significant interactions in a cross site-year analysis (Table 24).

Grain number formation

The number of grains produced per m^2 is governed by the development and survival of tillers and spikelets. As discussed in sections 3.1 and 3.2, evidence from shading experiments suggests that these processes are sensitive to light

availability during the pre-anthesis period. In our experiments when canopy CAI of spring barley was varied by seed rate a linear relationship was found between final grain number and pre-anthesis PAR interception. However, the relationship was not consistent between site-years or N treatments. Late N applications resulted in final grain numbers comparable to early N applications, but with a smaller pre-anthesis PAR interception. The effects of N fertilizer on tillering are well documented and early applications are often recommended to promote tiller production. Our results indicate that delaying N application until the start of stem extension led to the establishment of a larger number of fertile shoots by anthesis, but the shoots were smaller resulting in a smaller above ground biomass, canopy healthy area and HA per shoot (Table 26). Thus tiller production appears to have been delayed rather than restricted by late N application. As a consequence pre-anthesis PAR interception was smaller than with early N applications. At Aberdeen in 2007, the late N treatment was applied earlier than intended, and so there was only two weeks between early and late timings. This may account for the smaller differences in pre-anthesis PAR interception between N treatments at this compared to the other site-years. The results demonstrate that grain number formation is not a simple function of the amount of PAR intercepted pre-anthesis, but that other factors may influence the process.

The N regimes used in the winter barley experiments reported in section 3.1 were different to those used with spring barley, thereby precluding a direct comparison of the tillering response in the two crop types. In the winter barley experiments, both early and late N was applied to each plot, but the amount applied at each timing was varied. When data from the different site-years were pooled, the relationship between PAR interception and grain numbers for the HL and LH nitrogen treatments did not differ.

There is evidence from regression analysis that the increase in grain numbers resulting from fungicide application cannot be explained by protection of green leaf area and increased PAR interception by healthy tissue pre-anthesis. Thus grain numbers were increased by fungicide application either just at high seed rate (e.g. Aberdeen 2007 Early N; Fig. 13) or both seed rates (Aberdeen 2008 and Rosemaund 2007; Fig. 13) with little change in PAR interception pre-anthesis. During stem extension there is a rapid increase in the area of the

canopy as new leaves emerge and unfold. Because the latent period for infection of many foliar pathogens is longer than the phyllochron, development of visible symptoms on the uppermost leaves usually occurs after the final leaf has unfolded (GS 39). As a result visible disease tends to be located mostly in the lower canopy during stem extension, where it has a relatively small impact on canopy PAR interception because the majority of the PAR is intercepted by the upper healthy leaves (Bingham et al. 2009; Bingham and Topp 2009).
So what other mechanisms might be responsible for the effects of fungicides on grain numbers? Grain numbers are largely determined by events pre-flowering through the control of ear and spikelet numbers and by the success of pollination. In some circumstances final ear numbers may be influenced by post-anthesis shoot mortality. However, this does not appear to be a plausible explanation for the effects of fungicides on grain numbers observed in the current experiments. Final ear numbers were broadly comparable with fertile shoot numbers at anthesis for each of the fungicide treatments. There was no indication that shoot or ear numbers declined after anthesis to a greater extent in untreated plots compared to those receiving fungicide, thus the fungicide effects on grain numbers are likely to be exerted before flowering. There are several possible explanations including:

1. Control of disease (visible or symptomless) with fungicide resulting in a greater RUE, biomass production and grain number formation. An increase in RUE could arise through effects of disease control on photosynthetic metabolism (Scholes 1992), although it is unlikely to be important quantitatively given the location of disease in the canopy. Alternatively, it could be associated with changes in assimilate partitioning in favour of developing tillers and ears as a consequence of disease control.
2. Fungicides increase grain numbers independently of disease control by influencing tiller and/or spikelet survival through direct control of apical development or partitioning of assimilates.

Unfortunately it is not possible to distinguish between these mechanisms with the current data. Regardless of the mechanism a greater survival of tillers would, in addition to increasing eventual grain numbers, also contribute to an increase in biomass and RUE. An increase in spikelet survival, on the other hand,

is likely to increase grain numbers with relatively little effect on biomass gain and RUE pre-anthesis. Although not statistically significant, when averaged across site-years around 60% of the increase in grain numbers was associated with an increase in ear number m^{-2} and 40 % with an increase in grains ear^{-1}. This relatively equal contribution may explain why we found no evidence of either a consistent increase in pre-anthesis RUE with fungicide treatment or a significant increase in eventual grain number per unit of above ground biomass at anthesis (data not shown). Small increases in each are likely to have been 'lost' in the overall variability of the data.

Fungicides and MGW

An analysis of the effects of fungicides on MGW in terms of source or sink limitation of grain filling was developed in section 3.1. The realised MGW is compared with the quantity of assimilate potentially available for grain filling. The latter is based on an estimate of post-anthesis PAR interception, RUE and stem WSC reserves deposited pre-anthesis. Determination of post-anthesis PAR interception and stem WSC reserves is relatively straightforward. Determination of post anthesis RUE, on the other hand, is problematic because it can decline during canopy senescence and the physiological mechanisms responsible for the decline are not known with certainty (Bingham et al., 2007a). It has been argued that for fungicide-treated winter barley crops, the decline in RUE is the result of negative feedback on photosynthesis from a limited sink capacity. Evidence comes from a positive relationship between green area per unit grain number at anthesis (an indicator of the source:sink capacity) and the extent of the subsequent decline in RUE (Bingham et al., 2007a). For this reason it was assumed in section 3.1 that pre-anthesis RUE is the same as the potential RUE post-anthesis i.e. RUE in the absence of feedback from a limited sink capacity. But does this assumption also hold for spring barley?

At Aberdeen in 2007, RUE plots were linear indicating that pre- and post-anthesis RUE did not differ. This was found for all seed rate, N and fungicide treatments. In 2008, however, there was an appreciable decline in RUE post-anthesis. When data from the two years were pooled, a significant inverse relationship was found between the number of grains per m^{-2} and the extent of the non-linearity (Fig. 15), suggesting that a small sink capacity may have

contributed to the decline in RUE as postulated for winter barley (Bingham et al., 2007a). Thus there are grounds for using pre-anthesis values of RUE to estimate potential post-anthesis assimilate supply in spring barley.
The increase in MGW with fungicide treatment did not result from the protection of green area and an increase in PAR interception per unit grain number. Moreover, at Aberdeen in 2007 the increase in MGW with fungicide was found over a range of potential assimilate supply suggesting that fungicide treatment in some way increased the storage capacity of the grain. In 2008, the MGW of fungicide-treated and untreated plots was described by a common relationship with potential assimilate supply, which suggests that an increase in either RUE or WSC per grain may have contributed to the increase in MGW. However, even here the increase in MGW after fungicide treatment may not result simply from a greater availability of assimilate for grain filling, but from an effect of fungicide directly, or indirectly via the greater assimilate supply, on grain storage capacity. Given the uncertainties involved in estimating the true potential post-anthesis RUE, we cannot determine unequivocally what the mechanism of the fungicide response is. However, the apparently large surplus of potential assimilate (potential assimilate supply exceeding realised MGW) in 2008 and the slope of the relationship between MGW versus potential supply being considerably less than unity, are consistent with an effect of fungicides on grain storage capacity.

3.3.5 Conclusions and implications

There was a significant yield increase from fungicide treatment in each of the sites and years which would justify, in terms of crop response at least, the decision to spray. T1 applications gave the most consistent response, whilst T2 applications gave a comparable yield increase to the T1 in 3 out of the 4 site-years. Use of a two spray programme (T1 followed by T2) resulted in a significant additional yield increase over a single T1 application only at Aberdeen. Where there was an additional response to T2 in a two spray programme, it was associated with an increase in MGW as well as a small further increase in the number of grains per m^2 (Tables 21,22 and 23). Because of the lack of radiation interception data at Rosemaund for the post anthesis period, it

is not possible to determine the physiological basis for differences in response of MGW to T2 applications observed at Rosemaund and Aberdeen. It could relate to the control of ramularia which is recognised as being more of a problem at northern sites.

Do fungicide decisions need to be adjusted according to seed rate and N fertilizer regime? The seed rates and N regimes adopted in the current experiments on spring barley represent extremes of practice. The response of grain number formation to fungicide was modified by seed rate and N regime in some site-years, but the effects were inconsistent and not likely to be important in the range more typically associated with commercial spring barley production. By contrast, the effects of seed rate on the response of grain numbers to fungicide in winter barley were large enough to be significant in a cross-site analysis. Thus dense crops appear to be more responsive to fungicide than less dense ones and thus it is important to ensure they are adequately protected rather than take the view that they have tillers and potential grain numbers to spare.

It was hypothesised at the outset that crops with a large source (potential assimilate supply) relative to sink (number and storage capacity of grains) would be less responsive to late season fungicides because they would still have sufficient assimilate to fill grains even if some leaf area was diseased. The seed rate and N treatments significantly altered grain numbers and components of potential assimilate supply including leaf area duration, PAR interception, and WSC reserves per grain. However, in no case was a significant interaction found between fungicide treatment and seed rate or N regime on MGW. Thus the need for a T2 treatment was not modified by these extremes of agronomy. The lack of any interaction may in part be because fungicides appear to influence the grain storage capacity rather than simply the availability of assimilate for grain filling.

Given the observed variation in yield responses to fungicide between sites and years there would appear to some scope for adjusting the number and timing of fungicide treatments to spring barley, providing that improvements can be made in our ability to predict in advance the likely response of the crop to treatment. Further research is needed to understand the mechanisms through which fungicides may increase grain numbers and MGW, and the factors that regulate the process, before yield responses to fungicides can be predicted more reliably. Evidence presented in the current study indicates that the response does not

relate simply to the amount of visible disease and the protection of PAR interception by healthy area, either pre or post anthesis.

3.3.6 Appendix

1. Fungicide x N and fungicide x seed rate (SR) interactions on grain yield. Yields are given in t ha^{-1} at 100% DM. LSD values are for comparison of fungicide treatments within the same level of N or seed rate. Ab = SAC Aberdeen.

Treatment	Fung	Ab			
		2007		2008	
N		Early	Late	Early	Late
	None	4.9	5.0	4.3	3.9
	T1	5.9	5.9	4.8	4.1
	T2	5.6	5.8	4.8	4.7
	T1 + T2	6.4	6.6	5.2	4.8
	mean	5.7	5.8	4.8	4.4
	N*F	ns		<0.001	
	lsd	0.19		0.17	
SR		Low	High	Low	High
	None	4.6	5.3	3.8	4.5
	T1	5.4	6.4	4.1	4.8
	T2	5.2	6.1	4.4	5.1
	T1 + T2	5.8	7.2	4.6	5.4
	mean	5.3	6.3	4.2	5.0
	SR * F	<0.001		ns	
	lsd	0.19		0.17	

2. Fungicide x N and fungicide x seed rate (SR) interactions on grain number m^{-2}. LSD values are for comparison of fungicide treatments within the same level of N or seed rate. Ab = SAC Aberdeen, Rm = ADAS Rosemaund.

Treatment	Fung	Ab				Rm	
		2007		2008		2006	
N		Early	Late	Early	Late	Early	Late
	None	14948	15954	11827	11883	14718	14100
	T1	16540	17618	12515	13025	15711	14383
	T2	15094	16355	12284	13906	14539	14465
	T1 + T2	18002	17694	13273	14073	15156	15002
	mean	16146	16905	12475	13222	15031	14488
	N*F	ns		<0.05		<0.05	
	lsd	1323		739		593	
SR		Low	High	Low	High	Low	High
	None	13457	17445	10060	13649	11953	16864
	T1	14831	19326	10984	14557	12272	17822
	T2	13974	17475	11567	14623	12052	16952
	T1 + T2	14520	21177	11708	15637	12186	17972
	mean	14196	18856	11080	14617	12116	17403
	SR * F	<0.01		ns		ns	
	lsd	1323		739		593	

3.4 Yield response of winter and spring barley to fungicide timing across sites and varieties

3.4.1 Introduction

The previous results in the current study were based on two barley varieties and two locations: winter barley, Haka and spring barley, Cocktail, each at two sites, near Hereford (ADAS) and Aberdeen (SAC). Although these sites are good representatives of a north and a south location, there is likely to be a larger range of environmental and agronomic effects on barley grown throughout the UK, with additional variation in drilling date (between autumn and spring), nitrogen nutrition, and disease pressure. Barley is also genetically diverse, with specialised cultivars being grown for malting quality, and a distinction between two- and six-row types. The aim of this work was to provide independent data from a range of varieties and locations to validate findings from the experiments at the Hereford and Aberdeen sites.

Most of the yield variation in barley is due to differences in the number of grains m^{-2} produced by a crop (Blake *et al*., 2006), which in turn is directly related to the amount of photosynthetically active radiation (PAR) intercepted before GS 31 and during stem extension (GS 31 – 59) (Bingham *et al*, 2009; section 3.1). Fungicides act mainly to increase mean grain weight through effects on potential grain size (i.e., capacity of grains to store dry matter) rather than the supply of assimilate. The timing of fungicides is an important determinant of the responses in yield and yield components. The autumn fungicides (GS 21/24) protect the canopy during the initiation and survival of tillers and spikelets. In both winter and spring barley the T1 spray (GS 31/32) protects through the survival phase and the T2 spray (GS 49-59) protects during late floret development which is a critical phase for determination of grain number per ear. In the UK, most winter and spring barley crops receive two sprays, T1 and T2.

The relative contribution of a three-spray fungicide programme for winter barley, or a two-spray programme for spring barley, were investigated in section 3.2 and 3.3. There were significant yield responses for all fungicide timings, but with some greater than others, e.g., for winter barley the T1 sprays gave the largest

increase in yield and grain number. However these results were based on one winter barley variety (Haka) and one spring variety (Cocktail) at two sites. Therefore, it was important to investigate the effects of fungicide timing at a range of locations and varieties, to validate data from one winter and one spring variety at the two research sites, Hereford and Aberdeen.

Six industry partners provided results from experiments on various winter and spring barley varieties, from a range of locations in each year from Scotland to Southern England from 2005-2008, using common treatments and assessments of disease, yield and quality. The partners were: Agrovista UK Ltd., BASF plc, Bayer Crop Science Ltd., CSC Cropcare Ltd., Masstock Arable (UK) Ltd. and UAP Ltd. Variation in crop growth was therefore provided through geographic, seasonal, agronomic and genotypic variation. The objective was to provide data from a sufficient number of barley varieties, sites and seasons to test the extent to which the responses to disease control were affected in particular by fungicide timing and disease severity. The intention was to use fungicide treatment regimes which were commercially appropriate and which optimised economic and environmental sustainability, to be able to apply the findings to farm practice.

3.4.2 Materials and Methods

A total of 24 winter and 23 spring barley sites were contributed by the UK industry partners during the project (Fig. 18, winter barley, and Fig 26, spring barley). The partners were: Agrovista UK Ltd., BASF plc, Bayer Crop Science Ltd., CSC Crop Protection Ltd., Masstock Arable (UK) Ltd. and UAP Ltd. At each site the experimental design was a complete randomised block with at least three replicate plots per fungicide treatment. The fungicide treatments for winter barley were a factorial combination of autumn (GS21/24), T1 (GS 31/32) and T2 (GS 39/49) applications (Table 30). For spring barley, T1 and T2 spray times were used. For very forward spring barley crops, a T0 spray was applied prior to GS 30. All other non-fungicide sprays (herbicide, insecticide, growth regulators, etc.) were applied to plots using standard farm practice. For each site, fungicides were selected from the following: epoxiconazole + boscalid (Tracker, BASF),

prothioconazole + fluoxastrobin (Fandango, Bayer), fenpropimorph (Corbel, BASF), quinoxyfen (Fortress, Dow) and spiroxamine (Torch Extra, Bayer). Products were allowed to be changed between spray times, but had to be the same for all treatments within each spray time. Dose rates were selected to give good disease control at each application. In all three years, at each of the three application times, the majority of sites used epoxiconazole + boscalid, half rate (0.75 l/ha) with prothioconazole + fluoxastrobin, half rate (0.625 l/ha), and fenpropimorph, half rate (0.5 l/ha). In 2005 only, the Bayer winter barley sites did not have an autumn treatment.

Table 30. Barley fungicide treatments for industry partner sites, 2005-2008.

Treatment	Autumn (GS 21/24)	T1 (GS 31/32)	T2 (GS 49/59)
1	+	+	+
2	+	+	-
3	+	-	+
4	+	-	-
5	-	+	+
6	-	+	-
7	-	-	+
8	-	-	-

Emergence counts were made on fully treated and fully untreated plots. Diseases were assessed on a whole plot basis or individual plants following standard practice by the industry partner. For individual plants, disease was assessed on 10 randomly selected plants per plot, as % area diseased by individual leaf layer (current leaf number). Disease was recorded at a range of growth stages for individual sites and therefore was categorised into three phases: GS 30-39, GS 40-61, GS 61+, for the purposes of combining data from all sites. Average disease severity was calculated as a mean of the score from the top four leaves pre-anthesis, and from the top three leaves post-anthesis. Where only one disease was scored the other possible diseases on the same assessment date were given a zero value.

Green leaf area at GS 59 was measured in the fully untreated plots only, on 20 plants per plot. These were sampled and sent on the same day to ADAS Rosemaund, where area was measured within two days using an automated leaf area meter (Delta-t Devices, Cambridge, UK). A final fertile shoot count was made for all plots by counting the ears in three 0.1 m^2 floating quadrats per plot, any time between ear emergence and harvest. Grain moisture content, 1000 grain weight (TGW), specific weight, yield in kg/plot and plot size were recorded. Grain number per ear was calculated at harvest. For many industry partner sites, replicate plot data were not available and therefore statistical analysis of the data was not possible. Yields and TGW were adjusted according to the grain moisture content and are presented at 85% dry matter. The industry partner results were collated without including data from the research sites at ADAS Hereford and SAC Aberdeen (sections 3.1, 3.2 and 3.3), so as to be able to compare data from the industry partner locations with results from the two research sites.

3.4.3 Results

Winter barley

Yield, yield components and fungicide timing

Results were contributed from a range of varieties and locations from 7, 8 and 8 sites in 2005, 2006 and 2007, respectively (Fig. 18). Yields from fully treated plots ranged from 3.1 to 11.8 t/ha, with yield responses from 0.12 to 3.56 t/ha. There was a trend over three years for yield responses to increase with yield (Figure 33), but there was variability year to year, e.g., in 2007 yield response generally increased with yield for winter barley but in 2006 it did not.

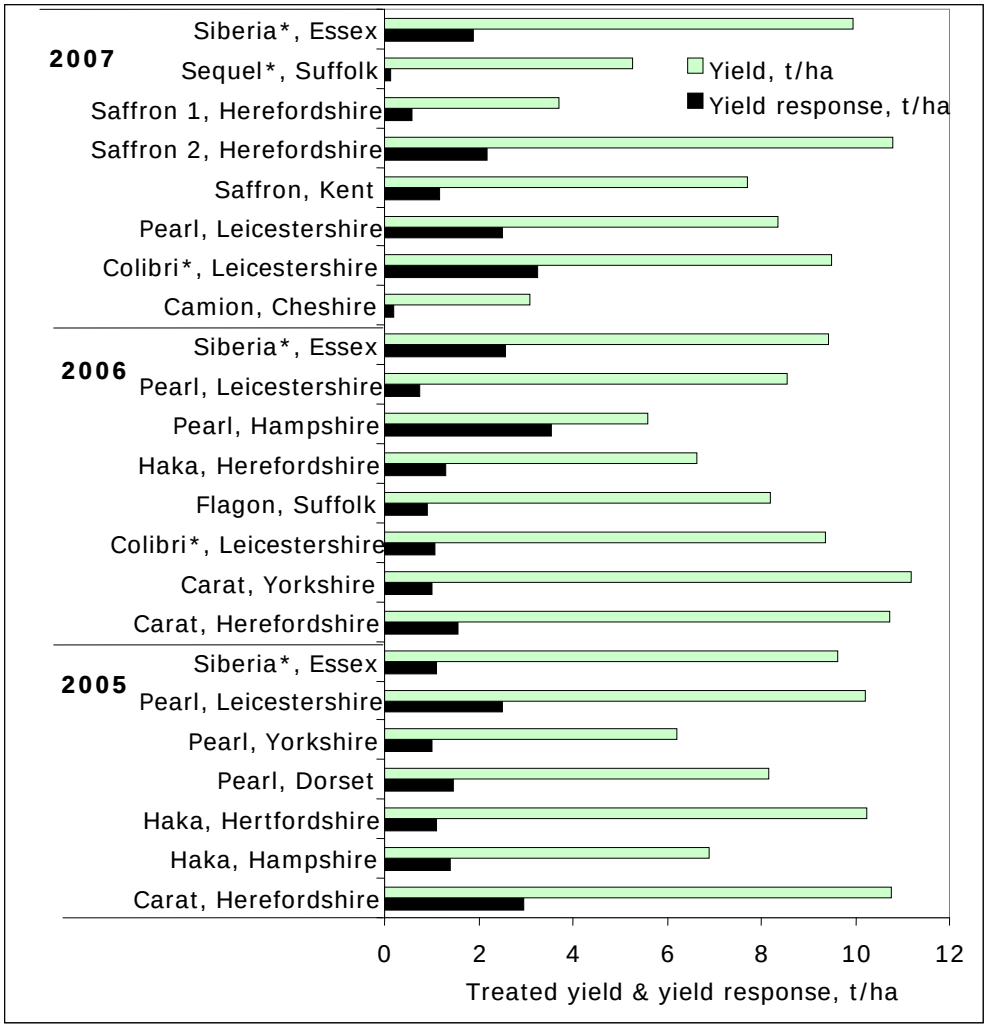


Figure 18. Winter barley fully treated yield and yield responses for industry partner sites, 2005-2007. Fungicides were applied at GS 21-24 (autumn), GS 31/32 (T1) and GS 39-49 (T2). * = 6-row barley.

The largest yield response for each spray timing, autumn, T1 or T2, was when it was used as the only treatment (Table 31). When applied in combination with other application timings, the response to a particular application was smaller. On average, across all sites and years, there was a yield increase when an autumn treatment was included, but this response was variable, e.g., in 2007 there was no yield response on average to the autumn spray when it was applied in addition to the T1 and T2 sprays. Thus, the contribution to yield from the three fungicide timings was not additive for these sites, i.e., if a treatment is missed or gives inadequate disease control the damage to yield potential might be recoverable to some extent.

Table 31. Winter barley yield responses (t/ha), industry partner sites 2005-2007

Fungicide timing	Response comparison	Yield response, t/ha
Autumn (GS 21/24)	Autumn - UT	0.54
	Fully treated – (T1 + T2)	0.12
	Mean	0.25
T1 (GS 31/32)	T1 – UT	1.18
	Fully treated – (autumn + T2)	0.37
	Mean	0.69
T2 (GS 39/49)	T2 - UT	1.04
	Fully treated – (autumn + T1)	0.35
	Mean	0.61

The mean grain number varied by treatment, with the T1+T2 and the three spray programme giving the largest grain/m^2 as expected (Fig 19). The largest response in grain number was with the T1 and T2 sprays (Table 32).

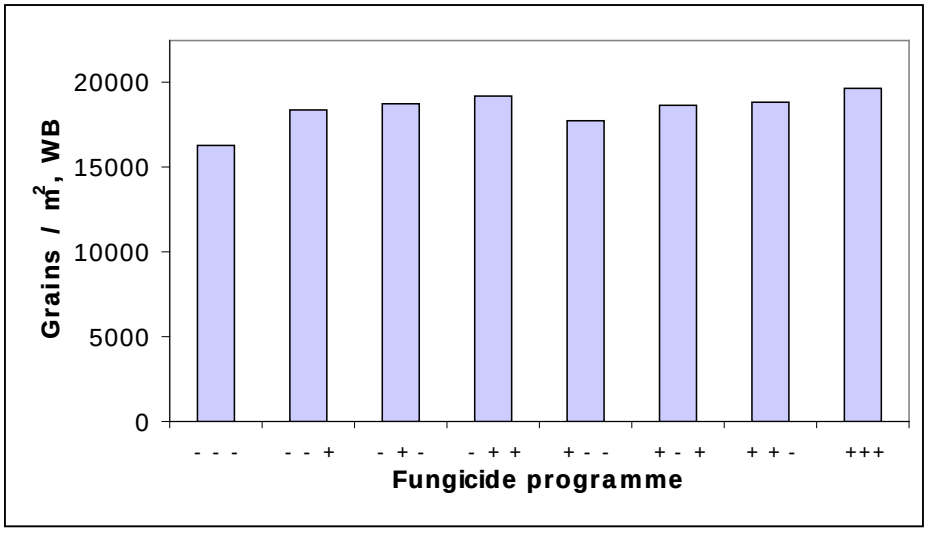


Figure 19. Mean grains/m^2 for fungicide programmes, industry partner winter barley sites 2005 – 2007.

The autumn spray resulted in an increase in grain number, but the T1 and T2 timings gave larger responses (Table 32). The T2 spray gave as big a response as the T1 spray, unlike the core sites where the T1 spray gave the largest grain

number response. The Industry sites tended to have earlier T2 timings than the core sites, which could explain the large grain number response with T2 that occurred at the industry sites but not at the research sites.

Table 32. Mean grains m^{-2} response with fungicide timing, industry partner winter barley sites. Responses are averages of the results from fungicides used at a single timing, and in combination with additional timings.

	Autumn (GS 21-24)	T1 (GS 31/32)	T2 (GS 39/49)
2005	587	1269	1126
2006	678	642	795
2007	387	1361	1458
Mean	551	1090	1126

There was an increase in MGW and yield with fungicide treatment (Fig. 20), for individual sites but not for data combined across sites. In general the sites had similar slopes to each other but with a small range of TGW change with increase in yield. TGW is affected by many factors such as disease, location and variety, which could explain the variability.

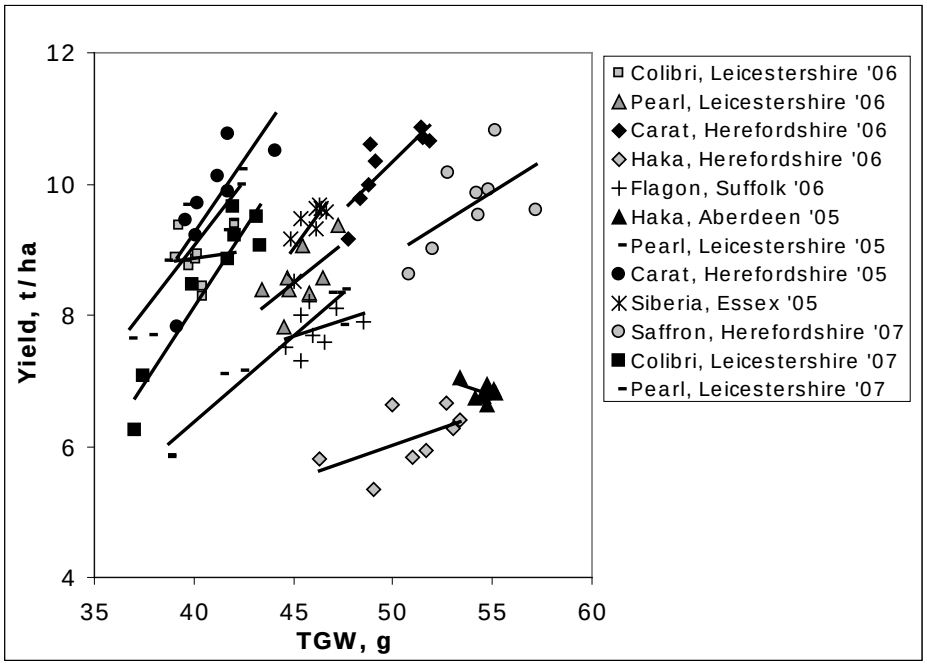


Figure 20. Yield and thousand grain weight (TGW), industry partner winter barley sites.

Disease and yield response

Rhynchosporium, net blotch, mildew and brown rust were the main diseases recorded in winter barley over three years, with large variation in incidence between varieties and years. At GS 30-39, disease was generally low, but by GS61 and later, Rhynchosporium occurred the most frequently (Fig. 21). The early disease recorded at GS30-39, although low, was as good an indicator of yield response as the late disease.

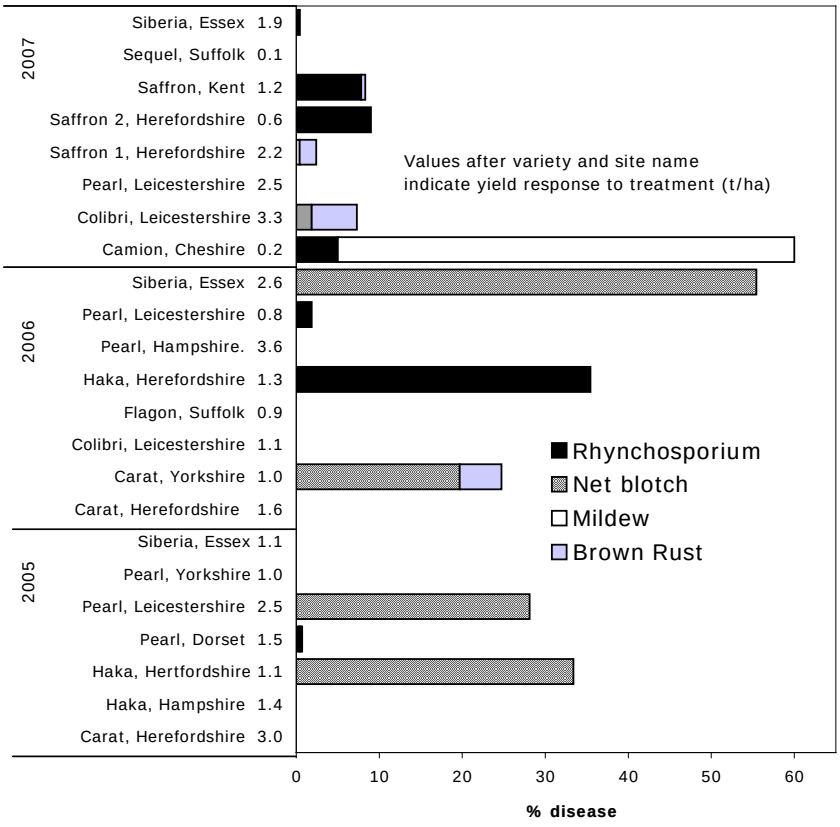


Figure 21. Disease in winter barley at GS 61+, industry partner sites. Disease was calculated from the average severity on the top three leaves.

Yield response tended to be larger for sites with disease at GS 30/39 than without (Table 33). For data categorised by disease or no disease at GS 61+, the yield responses were similar with disease and without. A positive yield response, therefore, occurred in the absence of disease. This could be due to variability in disease assessments or direct effects of fungicides, and needs further investigation. In almost all cases the largest yield response was seen with the fungicide spray as the single spray compared to a no-spray programme.

When Rhynchosporium data for GS 30-39 were examined as the only disease, the yield response was generally larger for sites with Rhynchosporium than without (Table 34). A yield response still occurred without Rhynchosporium, possibly because other diseases caused the loss, or the data were variable, or there were direct effects of fungicides. For sites with Rhynchosporium at GS 61+, there was a yield response but no larger than sites without Rhynchosporium at GS 61+.

Table 33. Mean yield response (t/ha), for winter barley with and without disease in untreated plots at GS 30-39, industry partner sites 2005-2007. Disease = sum of % area of Rhynchosporium, net blotch, mildew and brown rust.

	autumn (GS 21/24)	T1 (GS 31/32)	T2 (GS 39-49)
With disease GS 30/39 (8 sites)	0.23	0.99	0.71
Without disease GS 30/39 (15 sites)	0.29	0.54	0.50

Table 34. Mean yield response (t/ha), for winter barley with and without Rhynchosporium at GS 30-39, industry partner sites 2005-2007.

	autumn (GS 21/24)	T1 (GS 31/32)	T2 (GS 39-49)
With Rhynchosporium, GS 30/39 (4 sites)	0.29	1.25	0.88
Without Rhynchosporium, GS 30/39 (4 sites)	0.39	0.37	0.26

Two-row and six-row barley

As expected, the 6-row varieties had a higher yield and number of grains/m^2 than the 2-row varieties (Fig. 22), but the difference was not large. For both 2-row and 6-row barley, the yield response to fungicide was 19% of the treated yield. The response in grains/m^2 was 14.5% of the treated grain number for 2-row, 15.6% for 6-row.

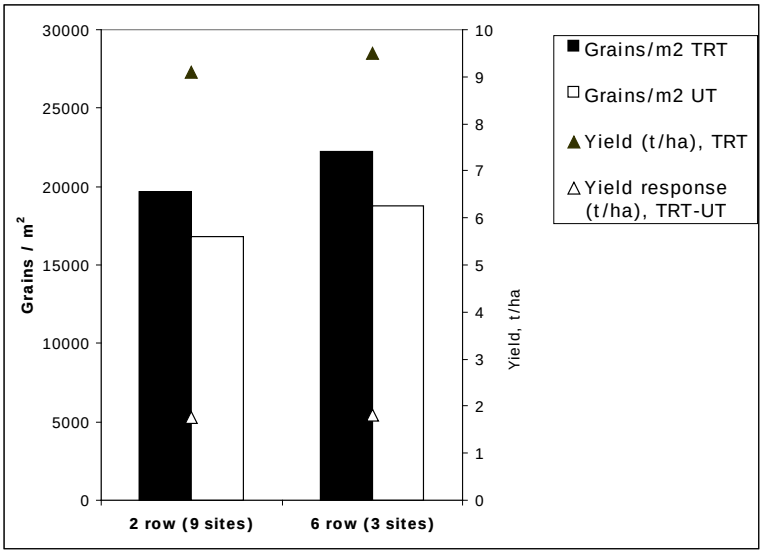


Figure 22. Grains/m^2 and yield for two- and six-row winter barley, industry partner sites 2005-2007. UT = untreated and TRT = full fungicide programme.

The data suggested that the 6-row varieties had a higher average grains m^{-2} count than the 2-row varieties (Fig. 22 and 23; caution: unequal numbers of sites).

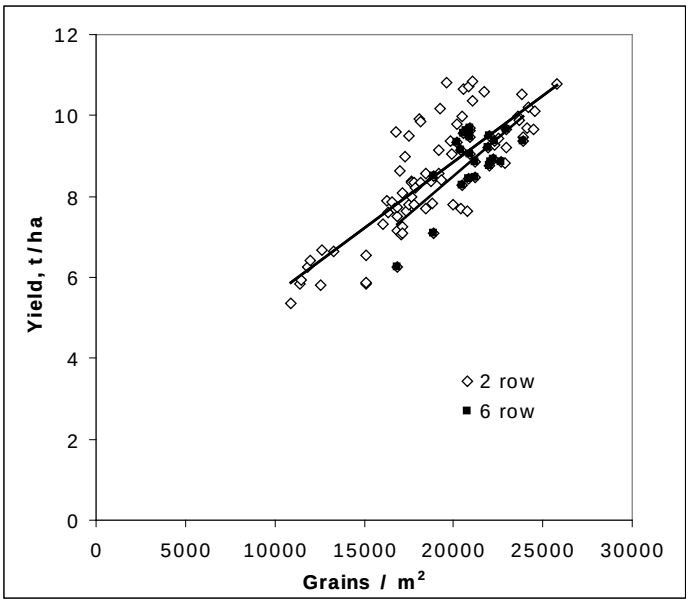


Figure 23. Grains/m^2 and yield for two- and six-row winter barley, industry partner sites 2005-2007. R^2 for 2-row and 6-row barley = 0.65 and 0.46, respectively.

The largest yield response for both 2-row and 6-row barley was with the T1 and T2 sprays (Table 35). The data indicate a larger response for the 6-row barley at GS T1, but the data needs to treated with caution as there were three times as many 2-row sites as 6-row sites. Not all sites could be included because some had missing results from one or more treatments.

Table 35. Yield responses (t/ha) for 2-row and 6-row winter barley, industry partner sites 2005-2007.

	autumn (GS 21/24)	T1 (GS 31/32)	T2 (GS 39-49)
2-row barley (16 sites)	0.25	0.69	0.61
6-row barley (5 sites)	0.36	0.92	0.55

Regional effects

The sites were assigned to three main UK regions as follows: [1] North (Scotland, Yorkshire), [2] West (Cheshire, Herefordshire, Hampshire, Dorset, Wiltshire) and [3] S / SE (Leicestershire, Essex, Kent, Norfolk, Hertfordshire, Suffolk). There were few winter barley sites in the north. There were no clear regional effects of any of the components looked at, e.g., there was little difference in grain number between the west and the S/SE sites (Fig. 24).

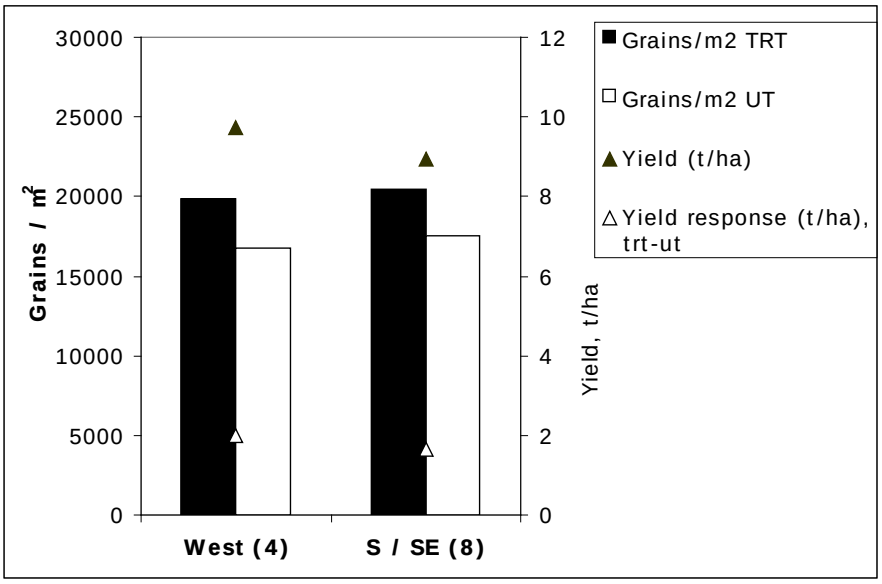


Figure 24. Grains/m^2 and yield by region for winter barley, industry partner sites 2005-2007. UT = untreated and TRT = full fungicide programme. Numbers in parentheses indicate the number of sites in each region.

Final ear counts prior to harvest varied with variety. Ear numbers for variety Haka, used at the research sites at Hereford and Aberdeen, were comparable to other varieties used at he industry partner sites (Fig 25) and therefore tillering habit for Haka was concluded to be representative of most varieties.

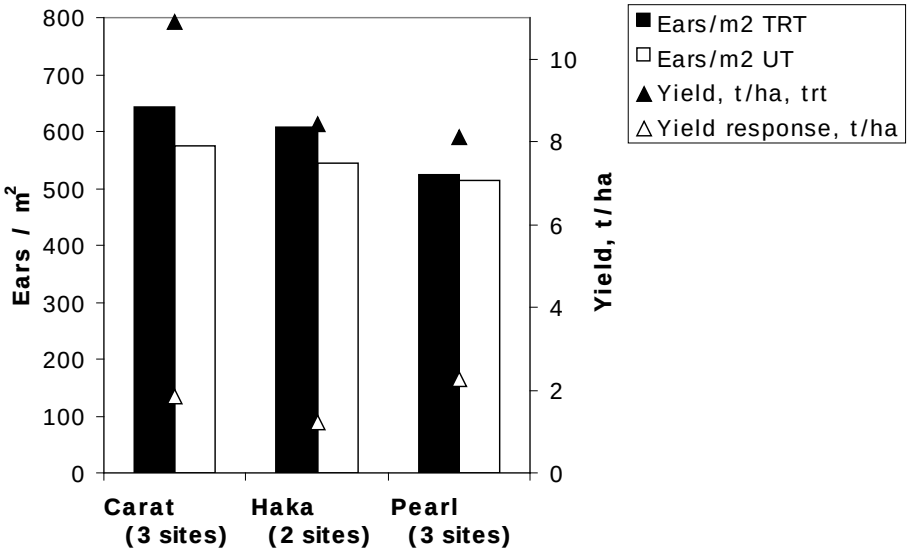


Figure 25. Ears/m^2 and yield for winter barley varieties which had two or more sites, industry partner sites 2005-2007. UT = untreated and TRT = full fungicide programme.

Crop thickness and yield response

The sites were categorised into those with high (> 600) and low (<400) ears populations per m^2 in untreated plots. For >600 ears/m^2, the varieties were Carat, Haka and Pearl, and for <400 ears/m^2, the varieties were Saffron, Haka, Pearl and Siberia. The yield response with the T1 spray was larger for the thick crops (similar to research sites). For the T2 spray however the yield response was larger for the thin crops. There was no large difference in yield response between the thick or thin crops with the autumn treatment.

Table 36. Yield responses (t/ha) for thick and thin winter barley crops, industry partner sites 2005-2007, data for 3 and 4 sites, for crops of > 600 and <400 ears/m^2, respectively.

	autumn (GS 21/24)	T1 (GS 31/32)	T2 (GS 39-49)
> 600 ears / m^2	0.36	1.63	0.38
< 400 ears / m^2	0.29	0.52	0.76

Spring barley

Yield, yield components and fungicide timing

Results were contributed from a range of varieties from 7 sites in 2005, 7 in 2006, 7 in 2007 and 2 in 2008 (Fig. 26), with a high proportion of northern sites as expected. Yields from fully treated plots ranged from 4.4 to 9.5 t/ha, with yield responses from 0 to 1.8 t/ha.

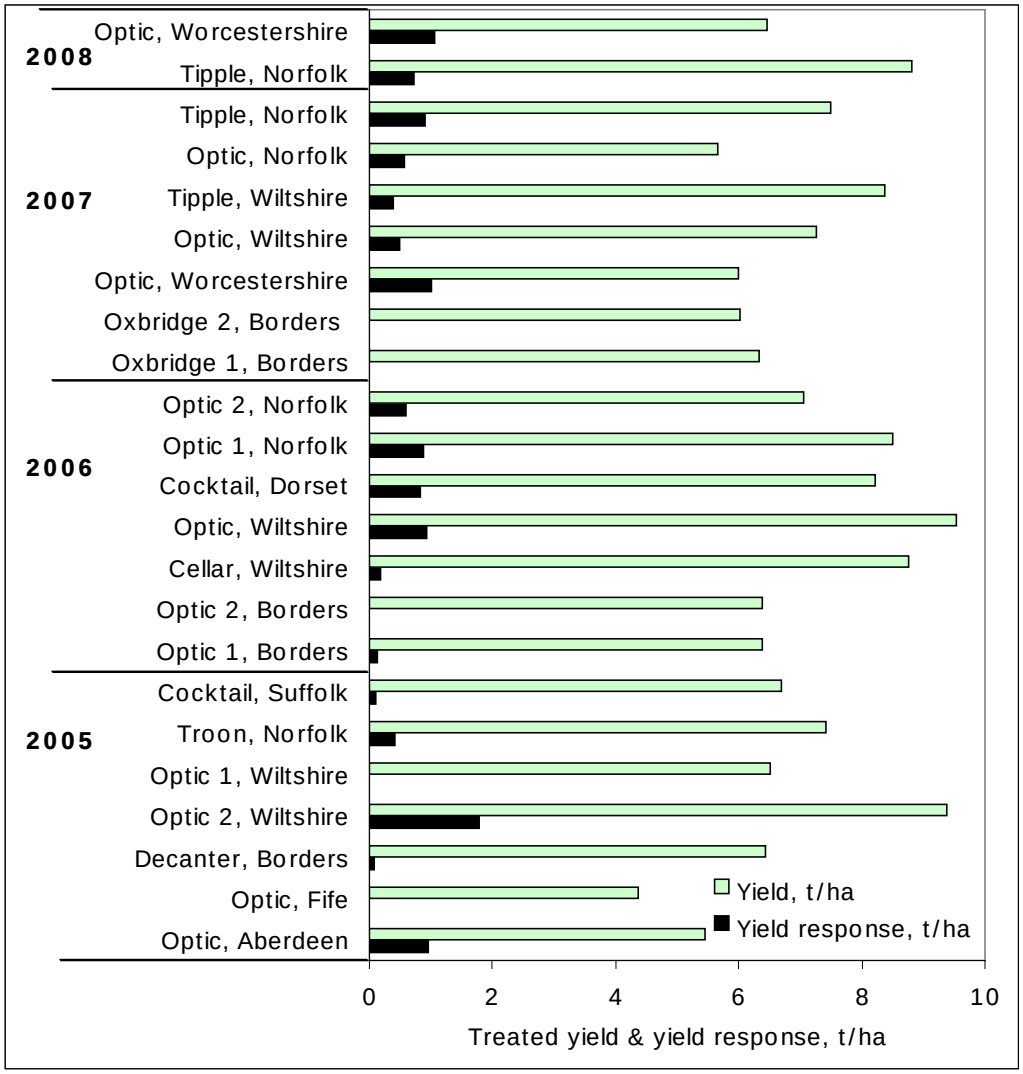


Figure 26. Spring barley fully treated yields and yield responses for industry partner sites, 2005-2007. Fungicides were applied at GS 31/32 (T1) and GS 39-49 (T2).

Overall the T2 treatment gave the largest yield response for spring barley, compared with the T1 treatment (Table 37). As with winter barley, the largest yield response was with a spray used as the only treatment.

Table 37. Spring barley yield responses (t/ha), industry partner sites 2005-2008

Fungicide timing	Response comparison	Yield response, t/ha
T1 (GS 31/32)	T1 - untreated	0.28
	Fully treated – T2	0.06
	Average	0.17
T2 (GS 39/49)	T2 - untreated	0.52
	Fully treated – T1	0.31
	Average	0.42

As with winter barley, grains/m^2 were closely related to yield (Fig. 33, R^2 for all years combined = 0.80, with fully treated plots ranging from 10074 to 21703 grains/m^2 (winter barley fully treated range was 12449 to 25808 grains/m^2).

The T2 only treatment resulted in more grains/m^2 than the T1 only treatment (Fig. 27), comparable to grains/m^2 from the T1 + T2 treatment. The mean number of grains/m^2 was lower with the T1 only fungicide treatment than with the other treatments (Fig. 30).

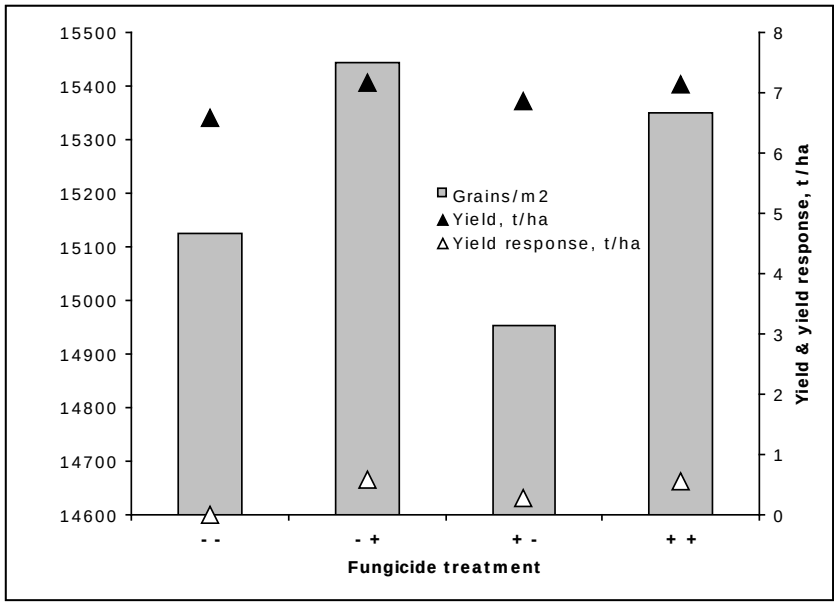


Figure 27. Mean grains/m^2 for fungicide programmes, industry partner spring barley sites 2005 – 2008.

The relatively low grains/m^2 number with the T1 only treatment resulted in a negative grains/m^2 response on average for the T1 treatment (Table 38). The T2 treatment gave a positive grains/m^2 response of 405, but this was low compared to winter barley where the response in grains/m^2 was 1090 for the T1 spray and 1126 for the T2 spray.

Table 38. Mean grains / m^2 response with fungicide timing, industry partner spring barley sites. Responses are averages for all sprays.

	T1 (GS 31/32)	T2 (GS 39/49)
2005 (3 Sites)	7	228
2006 (4 Sites)	-92	456
2007 (2 Sites)	-553	157
2008 (1 Site)	124	779
Mean	-128	405

As with winter barley, there was some treatment effect on TGW (Fig. 28), but for individual sites, not when data were combined.

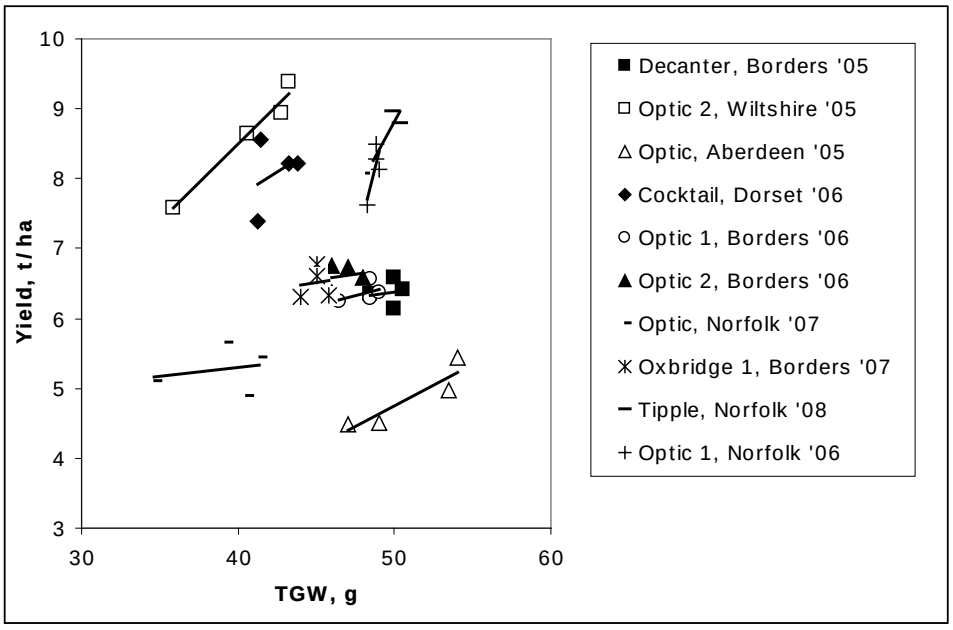


Figure 28. Yield and thousand grain weight (TGW), industry partner spring barley sites 2005-2008.

Disease and yield response

Rhynchosporium was the predominant disease in spring barley (Fig. 29), noted in 13 out of 20 sites assessed. Net blotch was much less frequent than in winter barley, but Ramularia was noted at several sites whereas winter barley had none.

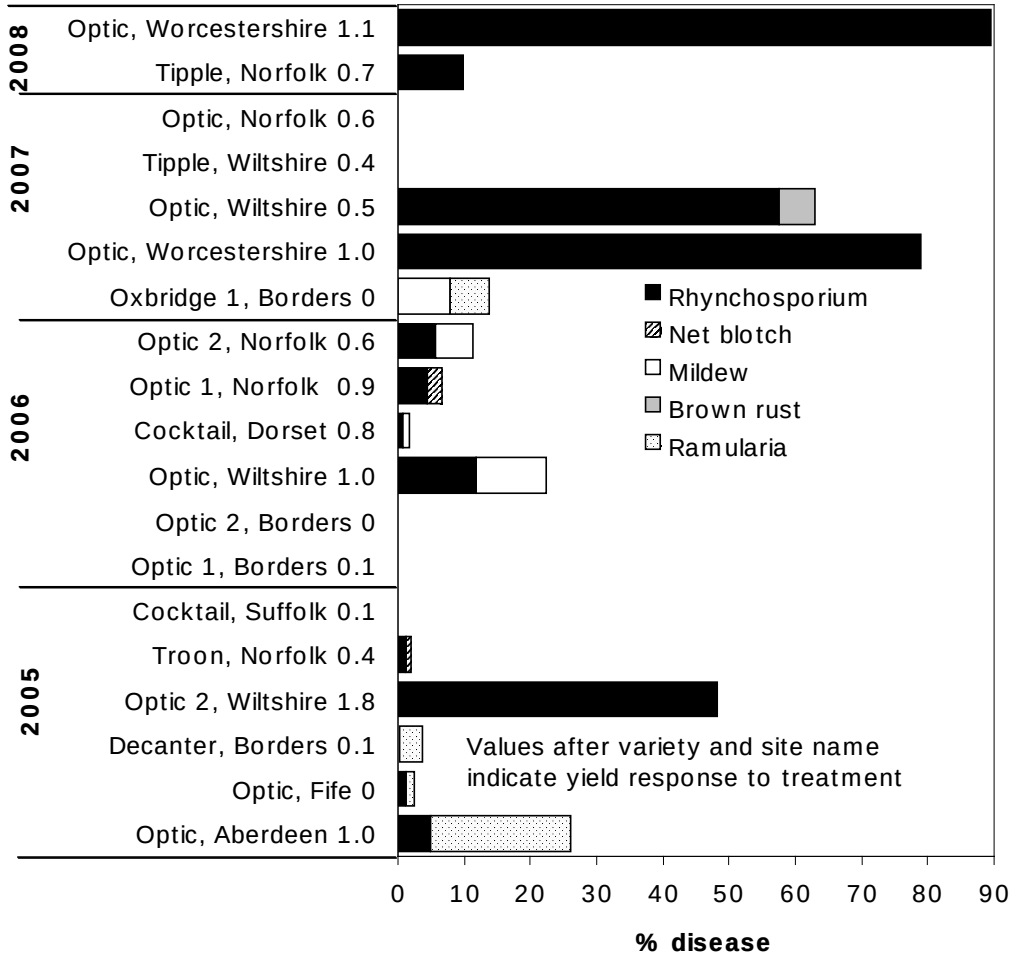


Figure 29. Disease in untreated plots of spring barley at GS 61+, industry partner sites. Disease data missing for Optic 1 Wiltshire 2005, Cellar Wiltshire 2006, Tipple Norfolk 2007 and Oxbridge Berwickshire 2007). Disease was calculated as the average % severity on the top three leaves.

The yield response at the spring barley sites with disease at GS 30-39 was similar to the yield response at sites which had no disease (Table 39), unlike winter barley where there was a higher yield response at sites with disease, particularly early disease at GS 30-39. Categorising spring barley sites by

disease or no disease at later growth stages gave similar yield responses in diseased and no-disease sites. Thus, as with winter barley, there was a yield response in the absence of disease.

Table 39. Mean yield response (t/ha), for spring barley with and without disease in untreated plots at GS 30-39, industry partner sites 2005-2008. Disease = sum of % area of Rhynchosporium, Net Blotch, Mildew, Brown rust and Ramularia.

	T1 (GS 31/32)	T2 (GS 39-49)
With disease GS 30/39 (9 sites)	0.15	0.36
Without disease GS 30/39 (14 sites)	0.11	0.37

As with total disease, there was no obvious difference in yield response between sites with Rhynchosporium at GS 30-39 and sites without (Table 40). As with winter barley, there was a yield response in the absence of Rhynchosporium.

Table 40. Mean yield response (t/ha), for spring barley with and without Rhynchosporium at GS 30-39, industry partner sites 2005-2008.

	T1 (GS 31/32)	T2 (GS 39-49)
With Rhynchosporium, GS 30/39 (5 sites)	0.18	0.33
Without Rhynchosporium, GS 30/39 (6 sites)	0.20	0.53

Regional effects

Yield and grains/m^2 tended to be lower for the northern sites, and yield responses to fungicide were low compared to the West and South/South East sites (Fig. 30). Only Optic was grown at more than one or two sites, and within this variety, yield components were variable across regions as expected, e.g., ears/m^2 varied by location (Fig. 32).

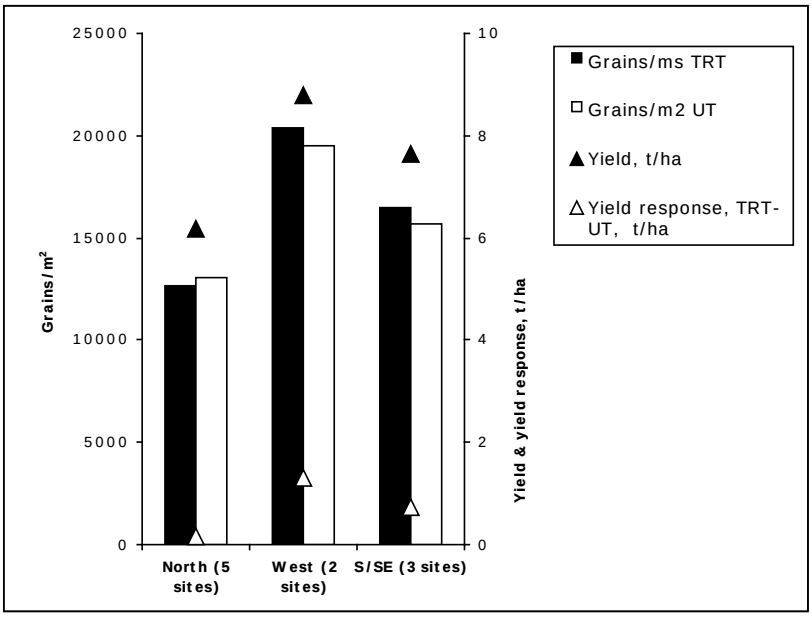


Figure 30. Grains/m^2 and yield by region for spring barley, industry partner sites 2005-2008. UT = untreated and TRT = full fungicide programme.

Crop thickness and yield response

Ears/m^2 varied with spring barley variety, but most varieties were only grown at one or two sites (Fig. 31). The exception was Optic which had 7 sites, across England and Scotland.

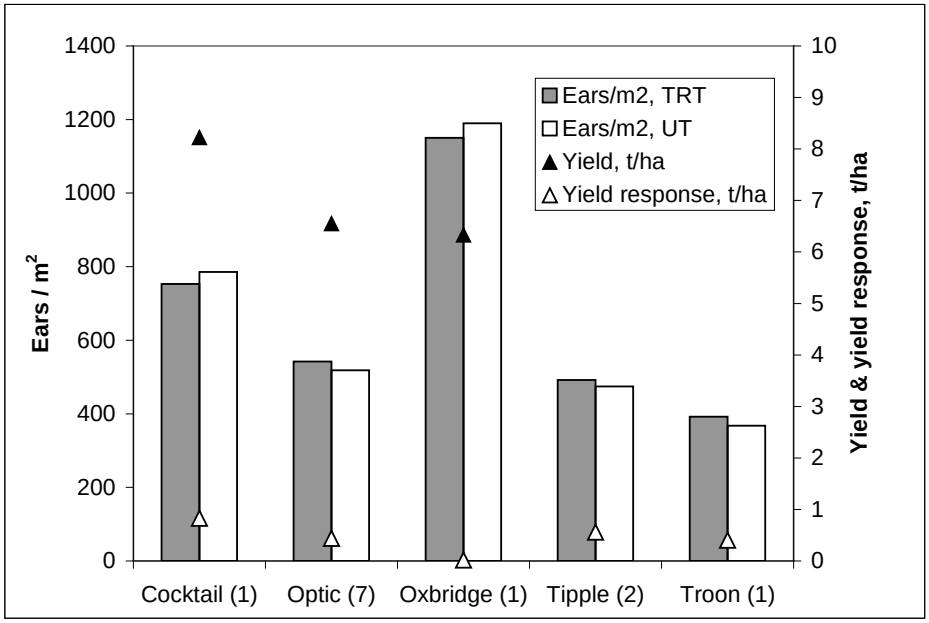


Figure 31. Ears/m^2 and yield for spring barley varieties, industry partner sites 2005-2008. UT = untreated and TRT = full fungicide programme.

The variation in ears/m^2 results for Optic may reflect the performance in ear counts for all varieties across the UK. Ears/m^2 for Optic were high in the north, low in the west and intermediate in the east (Fig. 31). However, ear number did not necessarily reflect yield, e.g., the Worcester and Wiltshire sites yielded as well as the northern sites, with much lower ear counts and with variable yield responses to fungicide (Fig. 32).

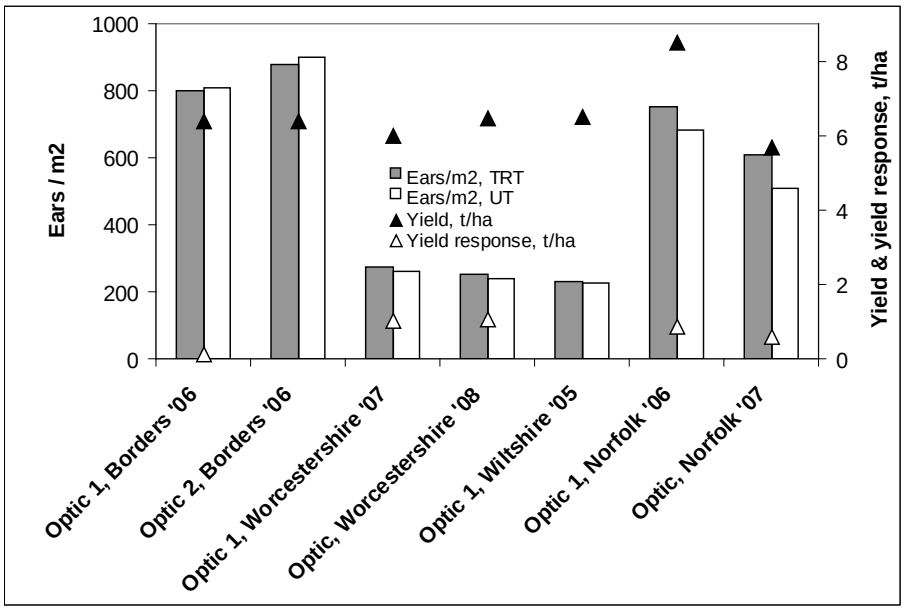


Figure 32. Ears / m^2 and yield for spring barley variety Optic, industry partner sites 2005-2007. UT = untreated and TRT = full fungicide programme.

The yield response in thinner crops tended to be larger than for thick crops (Table 41), for both the T1 and T2 treatments.

Table 41. Yield responses (t/ha) for thick and thin spring barley crops, industry partner sites 2005-2008, for crops of > 600 and <400 ears/m^2, respectively.

	T1 (GS 31/32)	T2 (GS 39-49)
> 600 ears / m^2 (7 Sites)	0.04	0.34
< 400 ears / m^2 (4 Sites)	0.29	0.48

Winter barley and spring barley compared

For all data combined, there was a trend for yield response to increase with potential yield (defined as yield with full fungicide programme) (Fig 33), but in general the relationship was variable. In 2005-6 the results suggested no relationship between yield response and yield, but in 2007 the yield response appeared to increase with yield. Overall, many low yielding crops showed a significant yield response to fungicide, and therefore crops of low yield potential (e.g., at historically low yielding sites) may still be economically worthwhile to treat.

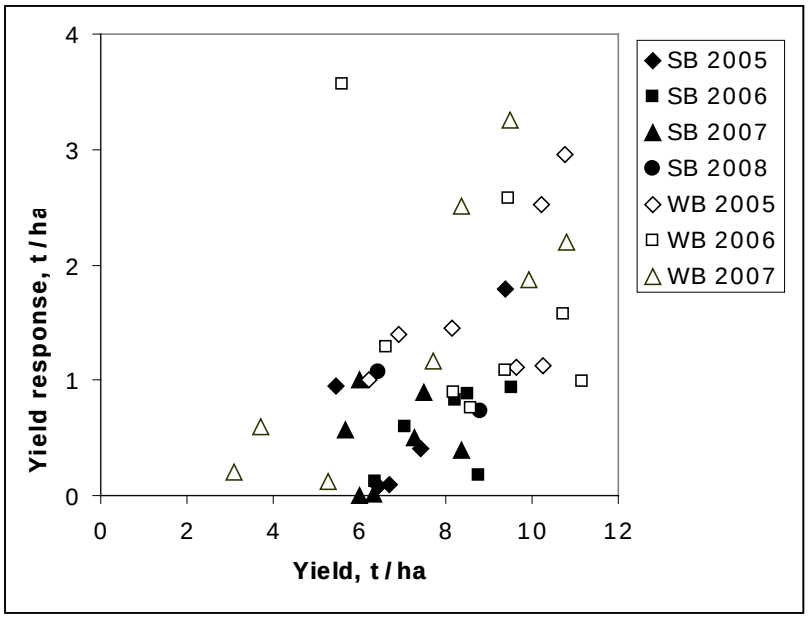


Figure 33. Yield and yield response for winter and spring barley industry sites, 2005-2008. Yield response is yield of fully treated (winter barley 3-spray, spring barley 2-spray) minus that of untreated plots. Yield is that of fully treated plots.

When data from 2005 – 2008 for all the winter (2-row and 6-row varieties) and spring barley sites were combined, grains m^{-2} showed a close relationship with yield (Fig. 34). Spring barley had a smaller range of grains m^{-2} than winter barley (Fig. 32).

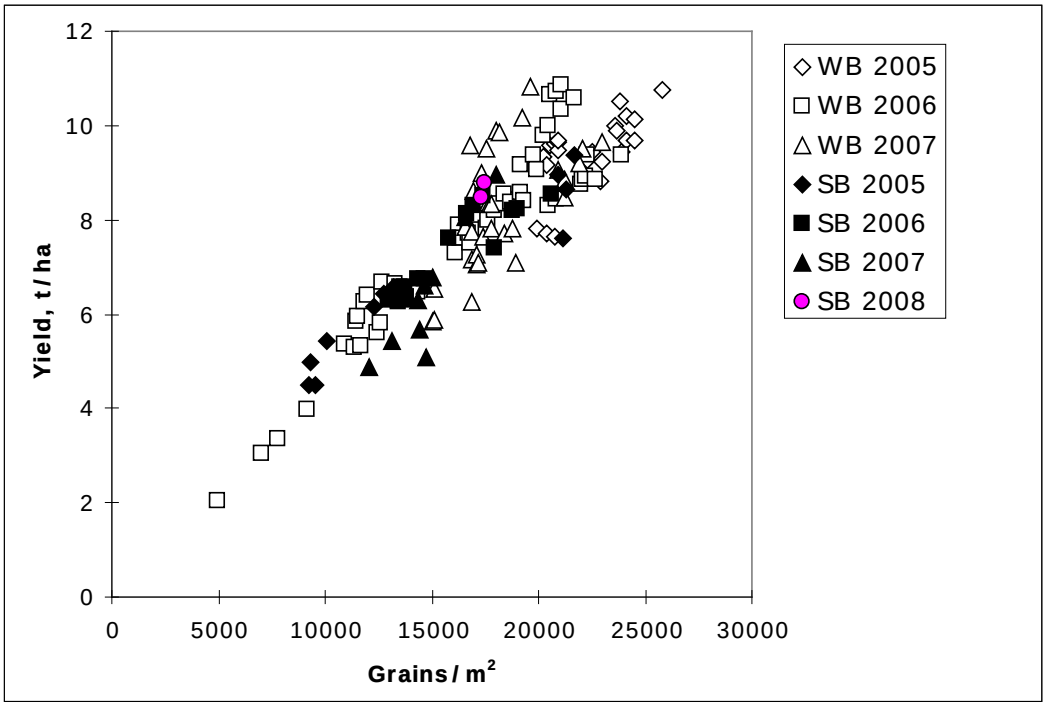


Figure 34. Grains/m^2 and yield for winter and spring barley industry partner sites 2005 – 2008. R^2 = 0.77 for winter barley and spring barley data combined, for all fungicide treatments.

In general, although the data were limited, yield increased with green area index (GAI) up to approximately GAI of 5, with an indication that further GAI increase did not give more yield (Fig. 35), as observed in the Hereford and Aberdeen research sites. The yields and GAI were lower for spring barley compared to winter barley, most probably reflecting the wide range of drilling dates and shorter duration of growth for spring barley. Some winter barley crops gave very high yields of over 10 t/ha with a GAI of only 5.

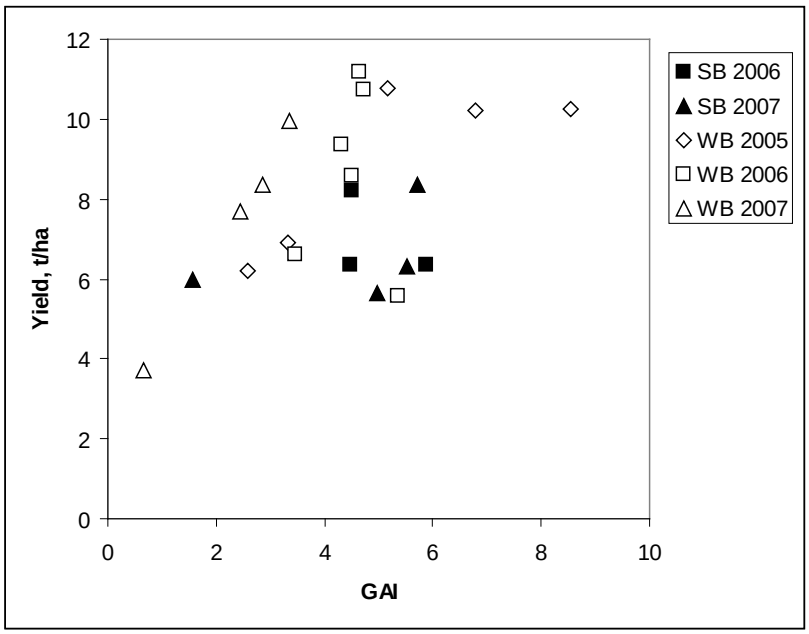


Figure 35. Yield and green area index for winter and spring barley industry partner sites.

Much of the variation in GAI increase was due to the ears/m^2 count (Fig. 36), with a tendency for spring barley to have ear numbers in the upper end of the range. Ears/m^2 had a bigger influence over GAI than did leaf size, which supports the importance of early protection and maintenance of shoots and hence maximising tiller number in barley crops.

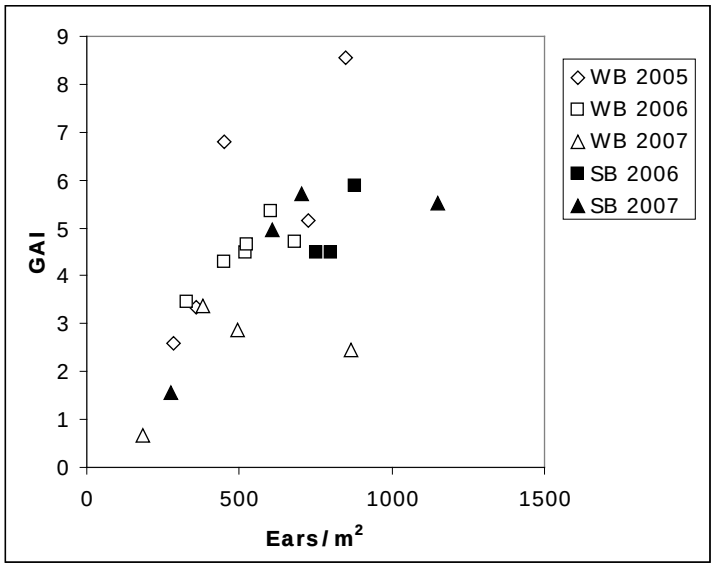


Figure 36. Green area index and ears/m^2 for winter and spring barley industry partner sites.

3.4.4. Discussion

The objectives of this work were to provide independent data from industry partner experiments to test the extent to which the findings from research sites could be generalised to different environments, varieties and disease pressure. Specifically, the industry experiments investigated the relative contribution of individual components of a three-spray fungicide programme to yield in winter and spring barley, and determined the effects of disease during different developmental phases. Similar concurrent experiments on one winter barley variety, Haka, at two research sites located near Hereford (ADAS) and Aberdeen (SAC) showed that a full three-spray fungicide programme gave a significant increase in yield and yield components (mean grain weight, grain number, ear number) The autumn (GS 21/24) and T1 (GS 31/32) sprays gave the largest increases in grain number. The T2 (GS 49/59) spray also increased grain number to a certain extent. In general, most of the conclusions from the industry experiments supported the findings from the research sites. Where industry data did not support the research site findings, this was due to data being inconclusive rather than contradictory. The main limitation was lack of sufficient replicate plot data to allow statistical analysis.

Many potentially low yielding winter and spring barley crops (yield potential here refers to the yield of fully treated crops) showed a large yield response to fungicide. Therefore potentially low-yielding crops may be economically worthwhile to treat. The difficulty is in predicting which crops justify treatment. The relationship between yield response and potential yield was variable, but taking all three years data into account, there was an overall trend for larger yield responses with potentially higher yielding crops, for both winter and spring barley.

For winter barley, autumn spray results were of particular interest. As expected the T1 and T2 sprays gave the largest response for yield and yield components compared to the autumn spray. However the autumn response varied by year, with a significant response in 2005, but little or no response overall in 2007. On average, the autumn spray gave an increase in both yield and ~~a~~ grains/m^2, but

in practice it is difficult to predict in autumn which crops will justify this spray. The standard fungicide application programme for most winter barley crops is two treatments, T1 and T2. It is possible that a treatment prior to T1 at GS23-30, as is often recommended for crops in Scotland, would be more beneficial than an autumn spray, because it could reduce early disease establishment at a critical time for canopy development more effectively than the autumn spray. Further work is needed to investigate whether additional early sprays are justified, and under what conditions. Three years of data from the industry sites suggest that the contribution to yield from three fungicide timings was not additive. In other words, if a treatment is missed or gives inadequate disease control, some of the yield can be recovered by applying the other fungicides. For the research sites, however, the fungicide timings were additive.

At the two research sites using one winter barley variety, Haka, yield was closely related to grains/m^2. For the industry sites, despite a very wide range of locations and varieties, the same result was found, for both winter (two- and six-row) and spring barley. However, the spring barley range of yields and grains/m^2 were less than for winter barley, i.e., winter barley was more variable. The main response of grain number to fungicide for the winter barley industry sites was with the T1 and T2 sprays. These were equal in their effects. This differed from the research sites where the autumn and T1 sprays gave larger grain number responses than the T2 spray. The industry T2 spray for winter barley tended to be earlier than those at the research sites and therefore was more likely to have continued to protect the survival phase of tillers and spikelets, resulting in a larger grain number response with this spray than for the research sites. For spring barley, the response in grain number from the T1 and T2 sprays was very different. On average there was no response from the T1 spray and a positive response from the T2 spray, but this was relatively small compared to the winter barley T2 response. This could reflect both the lower grain numbers on average with spring barley, or high variability across the industry spring barley varieties and sites.

Six-row winter barley tended to have a higher yield and grain number than two-row barley, but not always. The data indicated a larger yield response from six- than two-row barley for the autumn and T1 sprays.

Thousand grain weight (TGW) for both winter and spring barley showed a small but consistent increase with fungicide treatment, but only by individual sites, with more variability in spring barley data than winter barley. Unlike grain number, TGW was not closely related to yield when data for all sites and varieties was included, and therefore TGW data have little predictive value for yield.

Disease types and levels were highly variable across the sites, varieties and years, and therefore it was difficult to generalise the results. Rhynchosporium, net blotch, mildew and brown rust were the most frequently recorded diseases on winter barley, but at varying levels year to year. The same diseases were seen on spring barley but with the addition of ramularia, reflecting the high proportion of spring barley sites in the north. Despite the variability in disease there were some clear effects. Early disease observations at GS 30/31 were as good an indicator of yield response as later disease. For both winter and spring barley, yield responses with fungicides occurred with and without disease, suggesting that fungicides themselves have an effect on yield, which needs further investigation. For winter barley, on average the yield responses were higher in crops with disease than without, for disease recorded at GS 30/31. The results suggest that Rhynchosporium had a larger effect on yield than other diseases, but this would need confirmation by additional experiments designed to investigate specific diseases. Late disease (GS 61+) in winter barley was not related to yields and yield response to fungicide. For spring barley, it was not possible to demonstrate a difference in yield response between crops with or without disease, or specifically, with and without Rhynchosporium. Therefore, as with winter barley, there were clear yield responses with fungicide treatments for spring barley, but the yield responses could not be related to levels of diseases assessed or timing of assessment. This was partly due to variation in the data for disease and yield across the wide range of locations used. Spring barley results mirror those of winter barley in most respects and while the

results for disease at the spring barley industry sites do not support the winter barley results, they do not contradict them, i.e., disease assessments and crop protection should be started early in both crops.

Assigning the sites into groups for North, West and South/South East did not reveal any regional effects for yield or yield components for winter or spring barley, as yield, grain number and yield response to fungicide within a region were variable. The only clear regional difference was the high number of northern spring barley sites at which the yield and grain number tended to be lower than for spring barley at the West and South sites.

The hypothesis that thick crops are more responsive to fungicides as suggested by the results of section 3.1 was not possible to test for the industry sites data. The winter barley sites showed a larger yield response only with the T1 spray, for crops >600 ears/m^2 compared to <400 ears/m^2. The research sites data, based on winter barley variety Haka, showed that thicker crops had higher yield responses, but although the industry site results tend to support this for winter barley, the evidence is not conclusive, most likely because of the limited number of sites and variability of the data. For the spring barley industry sites the converse was true, with the thinner crops tending to have higher yield responses than the thick crops. The interaction between seed rate and fungicide on grain numbers in spring barley was also inconsistent at the research sites. The apparent difference in response of winter and spring crops may be associated with the longer duration of tiller and spikelet production and survival in the winter crop.

The value of the industry site results is they are derived from independent experiments from a wide range of barley varieties, locations and years. Despite the variability in these data, the results in general supported conclusions made from similar experiments at the research sites on one winter and one spring variety. Therefore, the conclusions from experiments at the research sites are broadly applicable to farm practice.

4. Acknowledgments

We thank the DEFRA Sustainable Arable LINK programme and AHDB-HGCA for funding, and Agrovista UK Ltd., BASF plc, Bayer Crop Science Ltd., CSC Cropcare Ltd., Masstock Arable (UK) Ltd. and UAP Ltd. for contributing field experiments and expertise to this project. Statistical advice from Chris Dyer (ADAS) is acknowledged.

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