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Improving resource use efficiency in barley, through protecting sink capacity.

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1. Abstract

The aim of this project was to identify how fungicides increase the resource use efficiency and yield of spring barley so that fungicide strategies can be tailored more effectively to account for the disease resistance of the variety and the potential physiological response of the crop. Specifically, three broad research questions were addressed which have scientific and commercial relevance. 1) What duration of protection of canopy light interception is required post-anthesis to maximise yield? 2) How do fungicides increase yield where visible disease severity is low or absent? 3) How should fungicides be timed in low disease risk situations?

Detailed physiological measurements were made in experiments conducted by research partners at two main sites, ADAS in Herefordshire and SRUC in Edinburgh. Experiments by industry partners tested the validity of the findings over a wider range of varieties and sites differing in disease pressure. Results showed that light interception by the canopy must be protected for approximately the first 75% of grain filling in order to maximise yield; a period of 3–5 weeks from 50% ear emergence depending on the site and year. After that period yield is insensitive to major reductions in light interception, probably because grain filling can be completed using dry matter from storage reserves. Treatment of disease-susceptible varieties with prothioconazole plus pyraclostrobin (products Proline and Comet 200, respectively) at the start of stem extension gave adequate protection of the canopy over the critical first 75% of grain filling when disease pressure was low. Under higher disease pressure an additional treatment during booting was needed. Although later applications after ear emergence protected the canopy for longer, they had no effect on yield because the additional protection occurred late in, or after, the critical period.

Field experiments over a wide range of varieties and sites showed that, on average, yield responses in the order of 0.3–0.4 t ha^{-1} were obtained from treatment with Proline and Comet in the absence of visible disease. The yield increases were largely the result of an increase in the number of grains produced m^{-2}. A comparison of the effects of Proline and Comet with that of chlorothalonil (product Bravo 500) indicated that the grain number response was not the result of the control of visible disease, the control of symptomless pathogen infection and leaf saprophytes, or a delay in leaf senescence. It appeared to result from a direct effect on plant metabolism which occurred before flowering. A single application during booting was sufficient to elicit the response. The results have implications for fungicide treatments in low disease risk situations, e.g. where resistant varieties are grown or where the disease pressure is low. If there is no disease present at the start of stem extension, fungicide treatment can be withheld. However, an application of prothioconazole plus pyraclostrobin at booting can be justified economically as it will provide insurance against late season disease and will result in yield enhancement, even if disease fails to develop. It can be further justified in terms of improvements in N use efficiency and reduced greenhouse gas emissions per tonne of grain yield.

2. Introduction

Spring barley is a valuable component of cropping systems. It provides specific conservation and wildlife benefits over winter crops. For example, overwinter stubble affords more foraging and nesting opportunities for farmland birds. Barley production in general also aids effective land use as it can be grown at marginal sites less suitable for the production of other arable crops and is an important component of good rotational practice. However, for barley production to be economically and environmentally sustainable, high yields of quality grain need to be obtained consistently with the minimum of inputs.

Foliar disease reduces the efficiency with which crops use water and energy. Disease can affect plant water relations through effects on root growth, the integrity of the leaf cuticle and stomatal regulation (Ayres, 1981; Walters, 1985; Prats *et al.,* 2006; Grimmer *et al*., 2012). Disease decreases energy efficiency by reducing the dry matter produced per unit of energy expended in crop husbandry (Berry *et al.*, 2008). As fertilizer nitrogen (N) accounts for approximately half the total energy input into arable production, management of disease to maximise yield per unit of fertilizer N applied is essential to maximise the energy efficiency of barley production. However, control of disease with fungicides needs to be targeted only at those crops likely to give a significant response in terms of increased grain yield or quality, in order to minimise any environmental impacts and selective pressure for fungicide resistance (Bingham *et al.,* 2012a) and for economic reasons. Identifying potentially responsive crops requires an understanding of how disease influences the yield forming process and the effects of fungicides on both the pathogen and plant.

Barley yield in the UK is predominantly sink-limited. Yield is determined by the number of grains produced and their capacity to store assimilates, rather than the crop's ability to provide assimilates to fill potential storage (Bingham *et al*., 2007a & b). The number of grains is determined before flowering by the production and survival of tillers and spikelets; processes that are sensitive to variation in light interception (Arisnabarreta and Miralles, 2008). The potential size (storage capacity) of grains is believed to be set by the development of the ovary pre-flowering and the grain endosperm early post-flowering (Bingham *et al.,* 2007b; Hasan *et al.,* 2011).

Pathogens may reduce crop growth by reducing radiation interception, radiation use efficiency (biomass production per unit of radiation interception by healthy green tissue) and the partitioning of assimilates (Boote *et al*., 1983; Johnson, 1987; Gaunt, 1995). This, in turn, may reduce yield by restricting the formation of each of the major yield components (Gaunt, 1995). Early epidemics of foliar pathogens which develop during canopy expansion may reduce the number of ears produced and the number of grains per ear because disease infection coincides with the period of tiller and spikelet production and survival (Brooks, 1972; Lim and Gaunt, 1986; Conry and Dunne, 1993). A key fungicide timing in winter barley is at the start of stem extension as this maximises tiller and spikelet survival and hence the formation of grain sites. Later applications just prior to ear emergence (i.e. during booting) often give smaller additional increases in yield through an increase in average grain weight (Bingham *et al*., 2010). In spring barley, the stem extension and booting application timings generally result in more comparable yield responses depending on the nature of the disease epidemic (Bingham *et al.,* 2010).

A widely held view within the industry is that late season (post-anthesis) disease reduces average grain weight through effects on the availability of assimilate for grain filling and hence disease management should seek to maximise the duration of canopy green area post-flowering (as in wheat). However, recent evidence suggests that fungicides applied just prior to ear emergence increase average grain weight predominantly by increasing potential grain size rather than assimilate availability for grain filling. Average grain weight was largely unaffected by agronomic treatments designed to vary the amount of post-anthesis assimilate per unit grain number, but was increased by fungicide in both winter and spring barley (Bingham *et al.,* 2010). Since potential grain size is determined over a relatively short period of time either side of flowering, the practical implication of these findings is that protecting green leaf area late into the grain filling period may be unnecessary. However, this hypothesis required testing, as the point at which green area light interception can be reduced without affecting yield was not known.

Thus, in contrast to wheat where the objective of disease management is to protect the post-flowering production of assimilate for grain filling, the primary aim of disease management in barley is to protect the development of sink capacity (Bingham *et al*., 2010; HGCA, 2013a). However, it has also been demonstrated that yield responses to fungicide in barley are variable and do not relate well to the amount of visible disease present, which suggests that fungicides may influence the development of sink capacity in ways other than through the control of visible disease (Bingham *et al.*, 2010; Bingham *et al*., 2012a). Treatment of low, or sometimes apparently nil, disease can result in substantial increases in grains per m^2, and hence in sink capacity and yield (Bingham *et al.,* 2012a). Visual assessment of disease is subjective, but the extent of the discrepancy between disease severity and yield is too large to be attributed to assessment error. This has important implications for the rational use of fungicides, because it means that the requirement for fungicide treatment, expressed in terms of likely improvement in yield or quality, cannot be predicted just from an assessment of the amount of visible disease present in the crop, or the risk of a disease epidemic developing.

There are several possible mechanisms that might account for yield responses to fungicide where there is little or no visible disease. Firstly, grain number formation may be particularly sensitive to low levels of disease that have a relatively small impact on canopy light interception because of symptom location low in the canopy. Secondly, fungicide treatment may be controlling symptomless pathogen infection. Molecular, microscopic and serological techniques have identified fungal infection in the absence of symptom development in a number of pathosystems including *Rhynchosporium commune* and *Ramularia collo-cygni* of barley (Fountaine et al., 2007; Walters *et al.*, 2008; Sowley *et al.,* 2010; Thirugnanasambandam *et al*., 2011). The fungus may grow systemically within plant tissues before visible symptoms develop, but the impact of this symptomless phase on crop growth and yield formation has not been tested previously (Walters *et al.,* 2008). A third possibility is that fungicides have direct effects on grain sink capacity and yield by modifying plant metabolism or assimilate partitioning. Some triazoles have been reported to impair gibberellin biosynthesis (Rademacher, 2000), which could conceivably increase grain numbers. In wheat, triazole and strobilurin fungicides have been found to delay leaf senescence in the absence of visible disease and the prolonged canopy lifespan is correlated with an increase in yield (Wu and von Tiedemann, 2001; Cromey *et al*., 2004). It has been suggested that the strobilurins may delay senescence by reducing ethylene production and the rate of cytokinin degradation (Grossman *et al.,* 1999), although other lines of evidence suggest that triazoles and strobilurins delay leaf senescence by reducing oxidative stress (Wu and Tiedemann, 2001). In addition, control of saprophytic fungi on the leaf surface has been implicated in the yield response to fungicide in the absence of visible disease (Smedegaard-Petersen and Tolstrup, 1985; Bertelsen *et al.*, 2009). Saprophytes may reduce yield by decreasing leaf lifespan and increasing metabolic costs associated with defence reactions to unsuccessful infection attempts by the fungus (Smedegaard-Petersen and Tolstrup, 1985; Bertelsen *et al*., 2009). Some of the most abundant saprophytes on the leaves of barley, and those responsible for the most frequent penetration attempts, are species of the genus *Cladosporium* (Smedegaard-Petersen and Tolstrup, 1985).

The aim of this project was to identify how fungicides increase the resource use efficiency and yield of spring barley so that fungicide strategies can be tailored more effectively to account for the disease resistance of the variety and the potential physiological response of the crop. Specifically, three broad research questions were addressed which have scientific and commercial relevance:

- What duration of protection of canopy light interception is required post-anthesis to maximise yield?
- How do fungicides increase yield where visible disease severity is low or absent?
- How should fungicides be timed in low disease risk situations?

The specific objectives were to:

1. Quantify the duration of green canopy protection required post-flowering to maximise yield.
2. Measure the importance of disease control during grain filling and the period around flowering when potential grain size is being determined.
3. Identify the mechanisms by which fungicides increase grain numbers and hence improve sink capacity in the absence of visible disease.
4. Test responses of sink components to fungicide treatment at key timings on a range of varieties with high ratings for disease resistance in the Recommended List.
5. Test new understanding across contrasting varieties and environments to develop commercial 'best practice' for disease management and improved yield of high quality grain (industry partner contribution).
6. Calculate the impact of improved disease control on resource use efficiency and greenhouse gas costs of production.

The project was structured such that the detailed physiological measurements needed to answer questions relating to the required duration of post-anthesis protection and the mechanisms of fungicide action were conducted at two research sites, SRUC Edinburgh and ADAS Rosemaund. Industry partners conducted experiments over a range of sites and varieties to test the yield response of spring barley to the same fungicide products under contrasting disease pressure.

Footnote: The above objectives are the same as those stated in the original project proposal, but objectives 1 – 3 in the proposal have been renumbered and reordered in this report to aid presentation of the research findings.

3. Materials & methods

3.1. Duration of canopy protection required post-anthesis (objective 1).

3.1.1. Experimental approach

It would be almost impossible to start and stop a disease epidemic in the field with sufficient precision to determine the duration of canopy protection required. For this reason shading was used to mimic the effects of severe disease on canopy light interception. Commencing at 50% ear emergence (Zadoks growth stage (GS) 55; Tottman, 1987) shade netting was erected at weekly intervals over plots of spring barley and left in place until harvest. Disease was prevented by using a variety (cv. Westminster) with good resistance to foliar disease and the application of a robust fungicide programme. The theoretical relationship between the onset of shading and grain yield is shown in Fig. 1. The netting reduced incident PAR (photosynthetically active radiation) at the top of the canopy by 64% at ADAS and 69% at SRUC. The small differences in extent of shading between sites may be the result of differences in tension applied to the netting. The duration of