

EFFECTS OF TIMING AND DOSE ON RESIDUES OF CHLORMEQUAT IN WHEAT, BARLEY AND OATS

APRIL 2004

Price £4.50

PROJECT REPORT No 334

EFFECTS OF TIMING AND DOSE ON RESIDUES OF CHLORMEQUAT IN WHEAT, BARLEY AND OATS

by

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This is the final report of a 15-month project that started in October 2002. This work was funded by the HGCA with a grant of $\pounds 69,998$ (Project 2737).

The Home-Grown Cereals Authority (HGCA) has provided funding for this project but has not conducted the research or written this report. While the authors have worked on the best information available to them, neither HGCA nor the authors shall in any event be liable for any loss, damage or injury howsoever suffered directly or indirectly in relation to the report or the research on which it is based.

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ABSTRACT

Chlormequat is a very effective lodging control agent in winter wheat. It has been shown to reduce lodging risk by the same amount as improving varietal standing power by 1-2 units, delaying sowing by 2-4 weeks and establishing about 100 plants m^{-2} fewer. In the UK, plant growth regulator products containing chlormequat are applied to about 65% of the winter wheat area and almost 50% of the winter barley and oat areas.

Previous surveys have recorded chlormequat residues in the harvested grain of most cereal crops that have been treated with chlormequat. Whilst the residues found in cereal grains are almost always below the maximum residue limit (MRL), pressure may be exerted in the future to further reduce residues due to the Food Standard Agency's aim to minimise pesticide residues in food irrespective of the MRL. Published literature has shown that the level of chlormequat residues may be determined by the time of PGR application and dose rate, and possibly by the crop's growing conditions. This study aimed to quantify the impact of management practices and crop conditions that affect chlormequat residues in wheat, barley and oat grain, whilst maintaining the ability of chlormequat to reduce lodging.

The experiments in this study were carried out at three UK sites during the 2002-03 growing season. Chlormequat residues were found in the grain of all of the wheat, barley and oat crops that were treated with a range of timings and rates of chlormequat. Residues ranged from 0.01 to 0.34 mg kg⁻¹ for wheat, 0.01 to 0.46 mg kg⁻¹ for barley and 0.01 to 0.77 mg kg⁻¹ for oats. None of the residues exceeded the MRL set for chlormequat in wheat and barley of 2 mg kg⁻¹, or oats of 5 mg kg⁻¹. In winter wheat, the most effective method of reducing chlormequat residues, without significantly reducing lodging control, was achieved by applying chlormequat earlier in the plant's life-cycle. The effect of changing the application timing from GS31 to tillering was to reduce the chlormequat residues in the grain by 60% and only cause a small and non-significant reduction in efficacy. In oats, the trials in this single season study indicated that chlormequat residues could not be reduced by changing the time of application or reducing the dose rate without greatly reducing the efficacy of lodging control. In winter barley, chlormequat did not reduce lodging or crop height, therefore the effectiveness of chlormequat as a lodging control agent for this species must be considered carefully. Applying chlormequat at late tillering reduced residues by 33% compared with applications at GS30. Reducing the dose rate to ¼ only reduced the residues by 36%.

SUMMARY

INTRODUCTION

In the UK, plant growth regulators (PGRs) are applied to 84% of the winter wheat area, 73% of winter barley and 65% of winter oats. About 79% of the PGRs applied to winter wheat and about 65% of the PGRs applied to winter barley and oats contain chlormequat. By far the most common chlormequat containing PGR product is 3C chlormequat. Other chlormequat containing products include New 5C Cycocel, Meteor, Stronghold and Upgrade. Chlormequat regulates growth by inhibiting the biosynthesis of gibberellic acid. This growth regulating property is used primarily to reduce lodging in cereals. Chlormequat is a very effective lodging control agent in winter wheat and has been shown to reduce lodging risk by the same amount as improving varietal standing power by 1-2 units, delaying sowing by 2-4 weeks and establishing about 100 plants m⁻² fewer.

A review of literature has shown that chlormequat applications usually result in residues of chlormequat in cereal grains. In 2002, a HGCA survey of chlormequat residues in UK-produced wheat grain showed that 44 of the 48 samples contained chlormequat at levels of between 0.02 and 0.50 mg kg⁻¹. These levels are significantly below the maximum residue limits (MRL) of 2 mg of chlormequat per kg of wheat or barley. The MRL for oats is 5 mg kg⁻¹. Whilst this survey and other European surveys have shown that chlormequat residues in cereal grains are almost always below the MRL, there may be pressure to further reduce them due to the Food Standard Agency's aim to minimise pesticide residues in food irrespective of the MRL. Published literature indicates that chlormequat residues of wheat and oats are reduced when chlormequat is applied in smaller amounts or earlier on in the plant's life-cycle. The duration of the interval between applying the PGR and harvesting the grain has been shown to be negatively related to the size of chlormequat residues. There is also some evidence that chlormequat residues are increased when the supply of water to the plant is limiting.

This study investigated the scope for minimising chlormequat residues in cereal grains, whilst maintaining the ability of chlormequat to reduce lodging, through the use of earlier and smaller applications of chlormequat. Sites with different rainfall and soil types were used to investigate the effect of water supply on chlormequat residues. A further experiment investigated the scope for minimising lodging in oats without recourse to PGRs through the manipulation of seed rate and nitrogen supply.

MATERIALS & METHODS

Experiments. The experiments were carried out at three UK sites during the 2002-03 season: ADAS Rosemaund has a silt clay loam, ADAS Gleadthorpe has a loamy medium sand and Queens University

Belfast has a loamy sand. A winter wheat and a winter barley trial were carried out at both ADAS Rosemaund and ADAS Gleadthorpe. Winter oat trials were carried out at ADAS Rosemaund and Queens University Belfast. The winter wheat trials used cv. Option and cv. Equinox, the winter barley trials used cv. Sumo and cv. Pearl and the winter oat trials used cv. Millenium and cv. Gerald. Within each experiment, the chlormequat treatments were applied to plots with an area of at least 30 m², and the plots were arranged in a randomised block design with three replicates per treatment. Chlormequat was applied as New 5C Cycocel (645 g l⁻¹ chlormequat chloride). The New 5C Cycocel treatments for each of the cereal species are described in Table 1.

Table 1. New 5C Cycocel treatments

Winter wheat and winter barley	Winter oats.
Nil	Nil
2.5 l ha ⁻¹ at GS24	2.5 l ha ⁻¹ at GS30
2.5 l ha ⁻¹ at GS30	2.5 l ha ⁻¹ at GS31
2.5 l ha ⁻¹ at GS31	2.5 l ha ⁻¹ at GS32
2.5 l ha ⁻¹ at GS32	2.5 l ha ⁻¹ at GS33
2.5 l ha ⁻¹ at GS37	1.25 l ha ⁻¹ at GS30 *
0.625 l ha ⁻¹ at GS31	1.25 l ha ⁻¹ at GS31 *
1.2511 ha ⁻¹ at GS31	1.25 l ha ⁻¹ at GS32
1.875 l ha ⁻¹ at GS31	1.25 l ha ⁻¹ at GS33 *
	1.25 l ha ⁻¹ at GS30 & GS32 *

* Queens University Belfast only.

An additional experiment was set up at ADAS Rosemaund to investigate the influence of crop management on the lodging risk and yield of winter oats. The experimental treatments included variety (Buffalo and Gerald), 250 and 400 seeds m⁻² and six nitrogen treatments with different timings and amounts of nitrogen fertiliser.

Measurements. Field measurements included plot yield and specific weight, and the incidence of lodging was recorded throughout the season. Additionally, during grain filling a large number of lodging associated plant measurements were measured on both wheat trials and the oat trial at ADAS Rosemaund. These measurements included height, natural frequency, height at centre of gravity, number and ear area of the shoots; the spread and depth of the root plate; together with the length, diameter, wall width and breaking strength of the bottom two internodes. These were used to calculate the leverage of the shoots, the strength of the stem base, the strength of the anchorage system and the overall lodging resistance in terms of the wind speed required to cause lodging. Chlormequat residues were estimated from a 500 g sub-sample that was representative of the whole plot. Chlormequat residue analysis was carried out by the Central Science Laboratory and had a reporting limit for chlormequat residues of 0.01 mg kg⁻¹.

RESULTS & DISCUSSION

Chlormequat residues were detected in all of the cereal crops that were treated with New 5C Cycocel (range 0.01 to 0.77 mg per kg of grain). None of the residues exceeded the MRL set for chlormequat in wheat and barley of 2 mg kg⁻¹ or oats of 5 mg kg⁻¹.

Large differences in the level of chlormequat were found between the cereal species and sites (Figure 1). Across the treatments applied at GS30, 31 and 32, winter wheat had the lowest residues at 0.10 mg kg⁻¹, followed by winter barley (0.27 mg kg⁻¹) and winter oats (0.28 mg kg⁻¹). For each species, large differences were observed between the sites, e.g. winter wheat residues averaged 0.05 mg kg⁻¹at ADAS Rosemaund and 0.15 mg kg⁻¹at ADAS Gleadthorpe (Figure 1). None of the factors measured in this study explain the large differences between sites and species. The site and species differences for the interval between applying the chlormequat and harvest appear to be too small to explain the variation in residues. Similarly the yield differences are not large enough for the residue differences to be caused by a dilution effect. It is difficult to draw conclusions about the effect of water availability because the site with the drought prone soil type experienced greater rainfall than the site with the water retentive soil type between the dates of treatment application and harvest. Other possible reasons for the site differences include the use of different cultivars and different levels of nitrogen fertiliser. Some existing literature shows that cultivar affects chlormequat residues and high nitrogen fertiliser can increase chlormequat residues in straw. However, other literature contradicts these findings. A much more detailed study taking into account chlormequat uptake, climatic conditions at the time of treatment application, cultivar, fertiliser use and late season drought would be required to fully explain site differences in the absolute residues.



Figure 1. Mean chlormequat residues for the 2.5 l ha⁻¹ of New 5C Cycocel treatments at GS30, GS31 and GS32 combined. ADAS Rosemaund (closed columns), ADAS Gleadthorpe (hatched) and Belfast (open). S.E.D. = 0.050, 11 df).

Each cereal species had a similar pattern of New 5C Cycocel treatment effects on the level of chlormequat residues across both sites at which they were tested. This was despite the large variation in chlormequat residues between sites described above. Statistically significant interactions occurred between the site and New 5C Cycocel treatments for wheat and oats, but these interactions were usually due to changes in the size of the differences rather than a change in ranking of the chlormequat treatments between the sites. Also the level of statistical significance for the interaction was always smaller than the level of significance for the treatment effects. For these reasons the mean chlormequat residues across sites are presented for each cereal species. The following sections describe how the chlormequat treatments affect the level of chlormequat residues in the grain and lodging resistance for winter wheat, winter oats, and winter barley.

Wheat

Chlormequat was shown to reduce lodging risk by increasing natural frequency and reducing height. These effects can be summarised in terms of the effect on shoot leverage, with a smaller shoot leverage equating to a smaller lodging risk. The results show that 2.5 l ha⁻¹ New 5C Cycocel significantly reduced shoot leverage when applied during tillering, GS30, GS31, GS32 and GS37, with the largest reduction at GS31 (Figure 2). Applying a ³/₄ rate at GS31 also significantly reduced shoot leverage (Figure 3). Earlier applications and lower rates of chlormequat also reduced the chlormequat residues in wheat grain (Figure 2 & 3). Changing the time of application from GS31 to tillering reduced the chlormequat residues by 60% and only caused a small reduction in the treatment's ability to control lodging. These results indicate that chlormequat residues can be reduced, whilst maintaining efficacy of lodging, by applying chlormequat earlier in the plant's lifecycle. The dose rate had to be reduced to ¹/₄ to achieve a statistically significant reduction in chlormequat residues of reducing the dose rate to minimise chlormequat residues appears to be limited because lodging control is greatly reduced when dose rates below ³/₄ are used (Figure 3).



Figure 2. Effect of application timing of New 5C Cycocel (@ 2.5 l ha⁻¹) on shoot leverage (columns) (s.e.d. = 5.77, 32 df) and chlormequat grain residues (----) (s.e.d. = 0.034, 32 df).



Figure 3. Effect of New 5C Cycocel rate (@ GS31) on shoot leverage (columns) (s.e.d. = 5.77, 32 df) and chlormequat grain residues (----) (s.e.d. = 0.034, 32 df).

Oats

Across both sites, chlormequat significantly reduced height when applied at GS32 or GS33 (Figure 4). Calculations of shoot leverage at ADAS Rosemaund showed that the application at GS32 reduced shoot leverage the most, but this effect was not statistically significant. These results indicate that the best lodging control occur when chlormequat is applied at around GS32, in agreement with recommendations for applying chlormequat to oats. Chlormequat residues increased significantly from 0.14 mg kg⁻¹ to 0.44 mg kg⁻¹ as the time of application was delayed from GS30 to GS32 (Figure 4). These results suggest that there is limited scope for minimising chlormequat residues in oats by applying chlormequat at earlier growth stages without reducing lodging control. There also seems to be little scope for minimising residues by reducing dose rate without reducing efficacy since crop height was not significantly reduced by a half dose of chlormequat applied at GS32. Of high importance to the grower were the yield reductions of about 0.5 t ha⁻¹ that resulted from chlormequat applications at GS30, 31 and 32 compared with the nil treatment. Similar results have been observed in some other studies and they may be more common in modern short varieties. Therefore, further studies must investigate the effect of chlormequat on the yield and lodging risk of modern short oat varieties.

In the experiment to investigate lodging control in the absence of PGRs, cv. Gerald experienced moderate lodging, whereas negligible lodging was recorded in the Buffalo plots. The amount of lodging was related to the quantity of N fertilizer. Applying 100 or 120 kg N ha⁻¹ resulted in 10% or less of the plot area lodged. Applications of 140 kg N ha⁻¹ resulted in 38% area lodged and 160 kg N ha⁻¹ resulted in 51-56% area lodged. The amount of lodging was not affected by either the time of nitrogen application or seed rate and yield was not affected by any of the treatments.



Figure 4. Effect of application timing of New 5C Cycocel (@ 2.5 l ha⁻¹) on crop height (columns) (s.e.d. = 23.4, 19 df) and chlormequat grain residues (---) (s.e.d. = 0.057, 19 df).

Barley

This study showed that, in agreement with several previous studies, chlormequat neither reduced plant height, nor reduced brackling. In fact, chlormequat applied during late tillering significantly increased brackling (P<0.05). This small effect that chlormequat has on lodging and height of barley appears to be caused by less efficient translocation of chlormequat to its biochemical targets (Alcock and Morgan, 1968; Lord and Wheeler, 1981), and possibly less uptake into the leaves compared with other cereal species (Hunt and Baker, 1982). Across the two sites, applying a full rate of New 5C Cycocel at GS30, 31 or 32 resulted in the greatest residues of about 0.27 mg kg⁻¹. Applying at either tillering or GS37, or reducing the application rate to ¹/₄, significantly reduced the level of residues to between 0.17 and 0.19 mg kg⁻¹. Halving the dose rate reduced the residues from 0.27 to 0.20 mg kg⁻¹, but this reduction was not statistically significant. Previous studies have not investigated the effect of chlormequat timing on residues in barley, so it is impossible to comment on whether the absence of a trend for increased residues in response to delaying applications is unusual.

Results from this study and published literature indicate that chlormequat is not as effective at reducing lodging in barley as it is in wheat and oats. Nonetheless, it is still applied to almost 50% of the winter barley grown in the UK. This may be due to the possibility that it can increase yield in the absence of lodging. This study observed a non-significant yield increase of about 0.25 t ha⁻¹ (4%) in response to most of the chlormequat treatments in the absence of lodging. Previous studies have shown that chlormequat can improve yield in the absence of lodging by 10-26%. Many of these studies also observed an increase in ear number in response to chlormequat. However, many studies also report no yield improvement and some negative effects have been reported, such as a reduction in grain size. Also, it has been shown that the increase in ear number can be caused by improving tiller survival rather than greater tillering, which is often desired by farmers with a poor crop. It therefore appears that small yield improvements in response to chlormequat are possible in the absence of lodging, but the response is very inconsistent.

Chlormequat is recommended for barley prior to GS31. If we assume that applications during tillering or GS30 are equally likely to produce positive effects in the absence of lodging then there appears to be some scope for minimising residues in barley by applying before GS30. This project showed that applications during late tillering resulted in residues that were 33% less than from applications at GS30-32. There seems to be limited scope for minimising residues by reducing the dose rate since the dose had to be reduced to a quarter to reduce residues by only 36%, and it seems unlikely that the treatment will be as effective at such low rates.



Figure 5. Effect of different treatments of New 5C Cycocel on the level of chlormequat residues in barley grain (s.e.d. = 0.034, 29 df).

CONCLUSIONS

- A recent survey showed that 44 out of 48 wheat samples contained chlormequat with a range of 0.01 to 0.50 mg kg⁻¹. In this study, experiments were carried out at three UK sites during the 2002-03 growing season. Chlormequat residues were found in the grain of all of the cereals treated with New 5C Cycocel. Residues ranged from 0.01 to 0.34 mg kg⁻¹ for wheat, 0.01 to 0.46 mg kg⁻¹ for barley and 0.01 to 0.77 mg kg⁻¹ for oats. None of the residues exceeded the MRL set for chlormequat in wheat and barley of 2 mg kg⁻¹, or oats of 5 mg kg⁻¹.
- In winter wheat, the most effective method of reducing chlormequat residues, without significantly reducing lodging control, would be to apply chlormequat earlier. In this study, changing the application timing from GS31 to late tillering reduced the chlormequat residues by 60% and only caused a small and non-significant reduction in efficacy. There did not appear to be any scope for minimising residues by reducing the dose rate without reducing lodging control.
- In winter oats, the trials in this single season study indicated that chlormequat residues could not be reduced by changing the time of application or reducing the dose rate without greatly reducing the efficacy of lodging control.
- In winter barley, chlormequat did not reduce lodging or crop height, therefore the effectiveness of chlormequat as a lodging control agent for this species must be considered carefully. Applying chlormequat at late tillering reduced residues by 33% compared with applications at GS30. Reducing the dose rate to ¹/₄ only reduced the residues by 36%.

TECHNICAL DETAIL

APPENDIX 1

MINIMISING CHLORMEQUAT RESIDUES IN HARVESTED GRAIN

INTRODUCTION

Chlormequat use in the UK

Chlormequat (2-chloroethyltrimethylammonium) is a plant growth regulator typically applied as the chloride salt (chlormequat chloride). Chlormequat chloride is often referred to as chlorocholine chloride or CCC. It was discovered by N.E. Tolbert of Michigan State University in the United States and its plant growth regulating properties were first reported by Tolbert (1960). Chlormequat regulates plant growth by blocking the biosynthesis of gibberellic acid (Rademacher, 2000). This growth regulating property is used to reduce cereal lodging throughout North West Europe. In the UK, about ³/₄ of the plant growth regulator (PGR) products used on cereals contain chlormequat (Table 1.1). By far the most widely used chlormequat product is 3C chlormequat, which makes up 55% of all PGR usage on cereals. On average, winter wheat receives 1.2 spray rounds containing a chlormequat product, winter barley receives 0.8 spray rounds and oats receive 0.7 spray rounds.

Chemical	Chlormequat	Winter	Winter	oats	Other
	content (g l^{-1})	wheat	barley		cereals
Chlormequat	460 - 700	1,713,934	336,555	56,767	31,394
Chlormequat / choline chloride	640 - 645	218,578	40,710	18,577	2,216
Chlormequat / imazaquin	368	306,889	2,093	-	497
Chlormequat / 2-chloroethylphosphonic acid	305 - 360	49,784	26,393		4,103
Chlormequat chloride / mepiquat chloride	345	93,149	11,502	-	-
Chlormequat chloride / 2-chloroethyl-	230	27,549	23,380	-	3,790
phophonic acid / mepiquat chloride					
Total chlormequat containing PGRs		2,409,883	440,633	75,344	48,000
Other PGRs		622,947	244,191	37,126	18,851
All PGRs		3,032,830	684,824	112,470	66,851

 Table 1.1. Usage of plant growth regulators in Great Britain 2000 (Spray hectares) (Garthwaite and Thomas,

 2000)

Role of chlormequat

Most studies show that chlormequat is very effective at reducing lodging in winter wheat. For example, a full rate application of chlormequat has been shown to reduce lodging risk by the same amount as improving standing power by 1 to 2 units, delaying drilling by 2 to 4 weeks and establishing 100 plants m⁻² fewer (Spink et al., 2003). Evidence indicates that chlormequat is able to control lodging in oats, but not as consistently as in wheat (Kust, 1986; Leitch and Hayes, 1990; White, 2003). The majority of previous studies show that chlormequat is not very effective at controlling lodging in barley (Humphries, 1968; Matthews and Thompson, 1983). The most likely mechanism by which lodging is reduced is by shortening plants. Chlormequat is usually effective at reducing crop height in wheat and oats, for which height reductions of 12 to 24% have been observed in winter wheat (Gill et al., 1974, Page, 1973) and reductions of 10 to 30% have been observed in winter oats (Leitch and Hayes, 1990; White et al., 2003). Chlormequat shortens wheat when applied during tillering or stem extension (Bragg et al., 1984; Berry et al., 1998) and shortens winter oats most when applied at GS32 (Leitch and Hayes, 1990). However, there are some reports of chlormequat having a negligible effect on the height of wheat crops (Matthews and Caldicott, 1981). Chlormequat is much less effective at reducing height in barley, with reductions of 2-3% observed by Green et al. (1985) and Koranteng and Matthews (1982). It has been observed that chlormequat causes early shortening of barley, but final heights are not decreased (Kust, 1986).

Only a few studies have investigated whether chlormequat reduces lodging by manipulating traits other than height. In general, these have shown that chlormequat does not consistently affect the stem strength and anchorage properties of cereals. For example, no consistent PGR effects have been found on the diameter and wall width of the stem of oats or wheat (Gendy and Hofner, 1989; Crook and Ennos, 1995; Berry *et al.*, 2000). Berry *et al.* (2000) showed that chlormequat actually reduced the material strength of the stem slightly. Crook and Ennos (1995) observed an increase in crown root number after chlormequat was applied at the beginning of stem extension. However, neither Crook and Ennos (1995) nor Berry *et al.* (2000) observed any effects of this treatment on the spread of the root plate and rigidity of the surface roots. These root observations were supported by Easson *et al.* (1995) who found no effect on the breaking load, cross sectional area or Young's modulus of the top few centimetres of individual winter wheat roots following separate applications of chlormequat at GS30 and ethephon with mepiquat chloride at GS32.

Several studies report that chlormequat can increase yield in the absence of lodging. In a review of literature Berry *et al.* (2004) found that chlormequat has been shown to increase yield in 10 studies, decrease yield in 2 studies and have no effect in 23 studies. The reports of yield increases exist for almost all of the major cereal species and can occur in response to PGR applications before or after the onset of stem elongation. Winter

barley had the greatest proportion of studies showing a yield increase, with four out of seven. Three of these studies also reported an increase in ear number.

Chlormequat residues in grain

Almost all studies show that the application of chlormequat to wheat, oats and barley, as a foliar spray, results in chlormequat residues within the grain and other plant parts (Teittinen, 1975; Mach-Hansmann and Rexilius, 1991; Zmrahl and Machackova, 1981; Kuhbauch and Amberger, 1971; El-Fouly and Fawzi, 1972; Gans *et al.*, 2000). Only two of these studies found residues above the maximum residue limits (MRL) of 2 mg of chlormequat per kg of wheat or barley or 5 mg kg⁻¹ for oats. Teittinen (1975) found 4 out of 31 spring wheat grain samples contained more than 2 mg kg⁻¹, two of which resulted from using greater than recommended rates of chlormequat. Zmrahl and Machackova (1981) observed 1 out of 16 wheat samples to exceed the MRL, which appeared to be associated with a very late application of chlormequat. Surveys of chlormequat residues in commercially grown wheat grain (Table 1.2) show that the majority of grain samples contain chlormequat residues, but all at levels below the MRL. A survey of pesticide residues in cereal products between 2000 and 2002 showed that 71% of flour and 41% of bread products sampled contained chlormequat at levels below the MRL (Table 1.2). All but 2% of these samples were known to be produced in the UK.

Country	Product	Number of samples analysed / with chlormequat	Residue range (mg kg ⁻¹)	Reference
UK	grain	n=48/44	0.02-0.50	Griffiths and Mason (2002)
Denmark	grain	n=50/42	0.004-0.62	Granby and Vahl (2001)
Norway	grain	n=39/16	0.05-0.33	Varran et al. (2000)
Germany	grain	n=285/187	0.05-1.14	Bruggemann and Ocker (1986)
UK	flour	n=72/51	0.05-0.30	www.pesticides.gov.uk (2000-02)
UK	bread	n=499/206	0.05-0.20	www.pesticides.gov.uk (2000-02)

Table 1.2. Chlormequat residue surveys for commercially grown wheat grain.

The most important factors that determine the level of the residues are the amount of chlormequat applied and the timing of application. Increasing the amount of chlormequat applied was shown to increase the residue levels in the grain (Jung, 1964; El-Fouly and Fawzi, 1972; Gans *et al.*, 2000; Zmrhal and Machackova, 1981). The residues did not always increase linearly with the amount of chlormequat applied.

Gans *et al.* (2000) discovered that doubling the rate of chlormequat only increased the residues from 1.7 to 2.0 mg kg⁻¹. Teittinen (1975) observed that decreasing the pre-harvest interval (PHI) for applying 2.5 kg ha⁻¹ chlormequat from 98 to 65 days increased the residues in wheat grain from 0.16 to 3.2 mg kg⁻¹. Zmrhal and Machackova (1981) and Jung (1964) also found that later applications to wheat resulted in larger residues. In oats, delaying the application of 1.38 kg ha⁻¹ of chlormequat from GS31/32 to GS45 increased the residues from 0.23-0.33 to 2 mg kg⁻¹ (Gans *et al.*, 2000). Jung, (1968; 1969) found greater chlormequat residues in oats than wheat and attributed these differences to the later application for oats. No studies have directly compared the chlormequat residues of barley with other cereal species, nor can we find any studies that have investigated the effect of chlormequat timing and dose on the residues in barley grain. The level of residues in straw have consistently been shown to be greater than those in the grain (Zmrhal and Machackova, 1981; Jung and El-Fouly, 1969; Bohring, 1982).

The majority of studies have shown that the rate at which chlormequat is metabolised in higher plants is negligible (Blinn, 1967; Schilling and Bergman, 1971; Faust and Bier, 1967; Jung and E-Fouly, 1969; Keller, 1990; Bohring, 1982; Muller and Schuphan, 1975) and that chlormequat residues are stable during storage (Bohring, 1982; Muller and Schuphan, 1975). In contrast to this, two studies have indicated that chlormequat may be metabolized in wheat (Dekhuijzen and Vonk, 1974; Stephan and Schutte, 1970), and Jung and El-Fouly (1969) observed chlormequat levels in grain to decline during 12 months of storage. The balance of the evidence indicates that chlormequat is relatively stable in cereal plants (only 0-10% metablolized) and this would explain why later applications result in large residues in the grain. Chlormequat behaves very differently in the soil, where it was deactivated within four weeks at 20 °C (Jung, 1965).

High rainfall or irrigation during cereal growth have been associated with lower chlormequat residues (Jung and El-Fouly; Kuhbauch and Amberger, 1971; Gans *et al.*, 2000). This may be caused by the dilution effect that results from more dry matter accumulating when the supply of water is adequate. Alternatively, Gans *et al.* (2000) postulated that increased water supplies may affect the transportation of chlormequat around the plant. No other factors have been shown to consistently affect the level of chlormequat residues. Teittinen (1975) observed differences between spring wheat varieties, but Gans *et al.* (2000) did not observe differences between three oat varieties. Zmrhal and Machackova observed greater residues in wheat straw after larger applications of nitrogen fertilizer, but Teittinen (1975) observed no effect of nitrogen fertilization.

The current literature shows that chlormequat residues in winter wheat, oats and barley grain are common, but they seldom exceed the MRL. It is also clear that chlormequat residues in wheat and oats can be reduced by reducing the dose and making earlier applications. We hypothesize that there is scope to further reduce chlormequat residues in cereal grains, whilst maintaining good lodging control, by optimising the dose and application time. However, quantitative information about the effect of dose and application timing on both

the residue levels and the reduction in lodging risk is not available. This project aimed to test the hypothesis by investigating the effect of chlormequat dose rate and timing on 1) the level of residues in the grain and 2) the change in lodging resistance for winter wheat, winter barley and winter oats. Soil types that varied in their water retentiveness were included in the study to investigate the effect of water availability on chlormequat residues. A model of the lodging process in wheat (Berry *et al.*, 2003) was used to quantify the effect of chlormequat on the lodging resistance of wheat and of oats at one site. Observations of lodging and crop height were used to estimate changes in lodging resistance for barley and oats grown at the second site.

MATERIALS AND METHODS

Field experiments

The experiments were carried out at three UK sites during the 2002-03 season: ADAS Rosemaund (52.1°N, 2.5°W) has a silt clay loam (Bromyard series), ADAS Gleadthorpe (55.1°N, 1.6°W) has a loamy medium sand (Cuckney series) and Queens University Belfast (54.7 °N, 6.0 °W) has a loamy sand (surface water gley over limestone till). Winter wheat and winter barley trials were carried out at ADAS Rosemaund and ADAS Gleadthorpe. Winter oat trials were carried out at ADAS Rosemaund and Queens University Belfast. All crops were managed following standard farm practice, such that fungicides were applied to minimise disease impact on grain yield, weeds were controlled to remove any competition with the crop and micronutrients, molluscicides and insecticides were applied where deemed necessary. Table 1.3 describes the cultivar used, its date of drilling, seed rate, the amount of nitrogen applied and the harvest date for each experiment.

Within each experiment, the chlormequat treatments were applied to plots with an area of at least 30 m², and the plots were arranged in a randomised block design with three replicates per treatment. Chlormequat was applied as New 5C Cycocel (645 g l⁻¹ chlormequat chloride). The New 5C Cycocel treatments for each of the cereal species are described in Table 1.4. Treatments were applied using a Knapsack Sprayer in 225 litres of water and with a medium spray quality. The appropriate nozzle to achieve this spray quality was used. At the time of application, the wind speed at boom height was between 2 and 6 mph (3.2 - 9.6 kph) (Force 1-3 Beaufort Scale at a height of 10 m) and the crop foliage was either dry or only slightly damp. The date of applying each PGR treatment is described in Table 1.5.

	ADAS Rosemaund		ADAS Gleadthorpe		Belfast	
Species	W wheat	W barley	W oats	W wheat	W barley	W oats
Cultivar	Option	Sumo	Millenium	Equinox	Pearl	Gerald
1999-2000 crop	W wheat	W wheat	W wheat	carrots	W wheat	W oats
2000-01 crop	W barley	W OSR	W oats	W wheat	onions	S barley
2001-02 crop	W OSR	W wheat	W wheat	W OSR	W wheat	peas
Date of sowing	2/10/02	2/10/02	18/10/02	5/11/02	20/9/02	7/10/02
Seed rate	350 s m ⁻²	250 s m ⁻²	300 s m ⁻²	250 kg ha ⁻¹	156 kg ha ⁻¹	155 kg ha ⁻¹
N applied (kg ha ⁻¹)	118	110	127	205	118	*81
Harvest date	14/8/03	21/7/03	6/8/03	7/8/03	21/7/03	15/8/03

Table 1.3. Description of each experimental crop.

* Broiler manure also applied pre-sowing at 50t ha⁻¹.

Table 1.4. New 5C Cycocel treatments

Winter wheat and winter barley	Winter oats.
Nil	Nil
2.5 l ha ⁻¹ at GS24	2.5 l ha ⁻¹ at GS30
2.5 l ha ⁻¹ at GS30	2.5 l ha ⁻¹ at GS31
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS31	2.5 l ha ⁻¹ at GS32
2.5 l ha ⁻¹ at GS32	2.5 l ha ⁻¹ at GS33
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS37	1.25 l ha ⁻¹ at GS30 *
0.625 l ha ⁻¹ at GS31	1.25 l ha ⁻¹ at GS31 *
1.251 l ha ⁻¹ at GS31	1.25 l ha ⁻¹ at GS32
1.875 l ha ⁻¹ at GS31	1.25 l ha ⁻¹ at GS33 *
	1.25 l ha ⁻¹ at GS30 & GS32 *

* Queens University Belfast only.

Table 1.5. Date of growth stages that correspond to the New 5C Cycocel application dates

	ADAS Rosemaund			ADAS Gleadthorpe		Belfast
	wheat	barley	oats	wheat	barley	oats
GS24	18 Mar	18 Mar	-	18 Mar	-	-
GS30	8 Apr	8 Apr	8 Apr	31 Mar	18 Mar	30 Apr
GS31	24 Apr	16 Apr	24 Apr	16 Apr	26 Mar	7 May
GS32	30 Apr	23 Apr	30 Apr	24 Apr	10 Apr	14 May
GS33	-	-	14 May	-	-	30 May
GS37	14 May	29 Apr	-	9 May	24 Apr	-

In addition to the experiments to investigate the residues and efficacy of chlormequat, an experiment was set up at ADAS Rosemaund to investigate the influence of crop management on the lodging risk and yield of winter oats. The experiment was a split plot design with three replicates in which the cultivars Buffalo and Gerald formed the main plots and seed rate and nitrogen treatments formed the sub-plots. The seed rate treatments were 250 and 400 seeds m⁻². Six nitrogen treatments were used to investigate the effects of timing and amount of nitrogen fertiliser (Table 1.6). Each plot measured 2m by 24m.

Treatment	March	GS30/31	GS37-39	Total
1	40	40	20	100
2	40	40	40	120
3	40	40	60	140
4	40	40	80	160
5	40	60	60	160
6	40	0	60	100

Table 1.6. Nitrogen treatments (kg ha⁻¹) for the non PGR winter oat experiment at ADAS Rosemaund

Measurements

Chlormequat residues

A single sub-sample of each grain sample (20 g of milled cereal) was extracted with methanol : water (1 : 1, v/v), filtered and analysed by HPLC-MS/MS with electrospray ionisation. Collisionally induced dissociated product ions at m/z 122>58 and 124>58 were monitored. For the purposes of recovery, chlormequat was added to samples of 'chlormequat free' cereal at 0.1 mg kg⁻¹. Any residues were quantified using five point bracketed standard calibration curves, prepared in 'chlormequat free' cereal extract. Matrix-matched calibration solutions in the range of 0.002 μ g ml⁻¹ and 0.2 μ g ml⁻¹ (equating to 0.01 mg kg⁻¹ to 1.0 mg kg⁻¹) were used for quantification of residues. The reporting limit for chlormequat residues was 0.01 mg kg⁻¹. For further details of the method of analysis see Startin *et al.* (1999).

Lodging

A visual assessment of the percentage area of crop that was lodged at 5° to 45° (from the vertical), lodged at 45° to 85° and lodged flat at 85° to 90° was made within the unsampled half of each plot ($10m \times 2m$), including its edges. During lodging assessments, the dominant mechanism and point of failure was identified i.e. whether by stem failure or anchorage failure. Assessments were done after each rain event and preharvest.

Lodging associated plant characters

Plant height (to the ear tip) was measured for all experiments by measuring five plants per plot just prior to harvest. In addition, plant characters that have been associated with lodging by a recently developed model of wheat lodging (Berry *et al.*, 2003) were measured for both wheat experiments and the oat experiment at

ADAS Rosemaund. These measurements were made during grain filling (GS71 to GS79). Ten plants were selected randomly from one half of each plot, avoiding the outer three rows, and the natural frequency was measured on each main shoot before the plants were excavated with a hand fork to a depth of about 100mm. The intention at sampling was to ensure that the structural crown roots were completely recovered. Laboratory measurements included the spread and depth of the root plate; the number of shoots per plant; the height at centre of gravity and ear area of each main shoot; together with the length, diameter, wall width and breaking strength of the bottom two internodes (internodes 1 and 2). The methods for these measurements are described in detail by Berry *et al.* (2000).

<u>Grain</u>

A plot combine harvester was used to measure the grain yield on at least 20 m² of each plot. A minimum of 1 kg of grain was sampled from each plot using 'a little and often' technique to ensure the sample was representative. After cleaning, part of this sample was used for determination of the moisture content and specific weight using a Dickey John grain analysis meter. The remainder of the sample (at least 500g) was analysed for chlormequat residues.

Calculations

The failure yield stress of the stem wall (σ) was calculated for internodes 1 and 2 using from the breaking strength of the internode (F_s), its length (h), radius (a) and wall width (t).

$$\sigma = \frac{F_s ha}{\pi \left(a^4 - \left(a - t\right)^4\right)} \tag{1.1}$$

The stem failure moment (B_S) is calculated from:

$$B_s = \frac{F_s h}{4} \tag{1.2}$$

The shoot base bending moment (B) was obtained from the following expression (Baker et al. 1998):

$$B = \frac{1}{2} \rho A C_d X V_g^2 \left(1 + \frac{g}{(2\pi n)^2 X} \right) \left(1 + e^{-\pi \xi} \frac{\sin(\pi/4)}{\pi/4} \right)$$
(1.3)

where ρ is the density of air (1.2 kg m⁻²), *A* is the projected ear area, *X* is the shoot's height at centre of gravity, *Vg* is the gust speed (ms⁻¹), *n* is the shoot's natural frequency, *g* is the acceleration due to gravity (9.81 ms⁻²), ξ is the shoot's damping ratio (0.08), *C_d* is the drag coefficient of the ear (1.0) and the remaining symbols take their usual meanings.

The anchorage failure moment (B_R) is calculated from:

$$B_R = k_3 s d^3 \tag{1.4}$$

where k_3 is taken as 0.43, *s* is the soil shear strength and *d* is the root cone diameter. Soil shear strength was calculated using equation 1.5, in which *i* is the daily rainfall, *l* is the structural rooting depth, *f* is the soil moisture content at field capacity, *w* is the soil moisture content at permanent wilting point, ρ_s is the density of soil and ρ_w is the density of water. S_D and S_W are values for soil shear strength at permanent wilting point and field capacity for which methods of calculation are described in Baker *et al.* (1998).

$$s = s_D - \frac{i}{\frac{\rho_s}{\rho_w} (f - w) l} (s_D - s_w)$$
(1.5)

The wind speeds required to buckle internodes 1 (V_{gSI}) and 2 (V_{gS2}) and cause anchorage failure (V_{gR}) were calculated by combining and rearranging equations (1.2) and (1.4), with equation (1.3) (Berry *et al.* 2000):

$$V_{gS1} = \sqrt{\frac{2B_{S1}}{(\rho A C_D X) \left(1 + \frac{g}{(2\pi n)^2 X}\right) \left(1 + e^{-\pi \delta} \frac{\sin(\pi/4)}{\pi/4}\right)}}$$
(1.6)

$$V_{gS2} = \sqrt{\frac{2B_{S2}}{\left(\frac{X - h_1}{X}\right) (\rho A C_D X) \left(1 + \frac{g}{(2\pi n)^2 X}\right) \left(1 + e^{-\pi \delta} \frac{\sin(\pi/4)}{\pi/4}\right)}}$$
(1.7)

$$V_{gR} = \sqrt{\frac{2B_R}{N (\rho A C_D X) \left(1 + \frac{g}{(2\pi n)^2 X}\right) \left(1 + e^{-\pi \delta} \frac{\sin(\pi/4)}{\pi/4}\right)}}$$
(1.8)

where B_{SI} and B_{S2} represent the failure moments of internodes 1 and 2 respectively, h_I represents the length of internode 1 and N represents the number of shoots per plant.

Statistical analysis

Analysis of variance procedures within Genstat 6 (Payne 2002) for fully randomised and split plot designs were used to test for differences among treatments and calculate standard errors of differences between means. Each cereal species was analysed separately with site treated as a main plot and the chlormequat treatments treated as sub-plots.

RESULTS

Weather

ADAS Rosemaund and ADAS Gleadthorpe experienced a similar amount of spring rainfall, which was below the long-term average for both sites (Figure 1.1). The soil type at ADAS Gleadthorpe is much less water retentive than the soil at ADAS Rosemaund, which indicates that the wheat and barley crops at ADAS Gleadthorpe were under greater water stress than at ADAS Rosemaund during the spring months. ADAS Gleadthorpe experienced heavy rain (94 mm) between the 22 and 30 June, which is likely to have provided an adequate water supply to the crops at this site for the rest of the growing season, whilst the crops at ADAS Rosemaund are likely to have continued under water stress.



Figure 1.1. Site rainfall between October 2002 and August 2003 for ADAS Gleadthorpe (black columns) and ADAS Rosemaund (open columns).

Chlormequat residues

Chlormequat residues were detected in all of the cereal crops that were treated with New 5C Cycocel (range 0.01 to 0.77 mg per kg of grain). None of the residues exceeded the MRL set for chlormequat in barley and

wheat of 2 mg kg⁻¹ and 5 mg kg⁻¹ for oats. Four of the untreated grain samples had chlormequat residues of between 0.02 and 0.05 mg kg⁻¹. These small residues may have arisen from several sources such as chlormequat residues persisting from the previous crop or grain contamination within the combine.

In winter wheat, chlormequat residues were reduced by applying New 5C Cycocel earlier (Table 1.7). At ADAS Gleadthorpe, applying 2.5 l ha⁻¹ of New 5C Cycocel during tillering (March) resulted in residues of 0.072 mg kg⁻¹. Residues increased to 0.173 mg kg⁻¹ after applications at GS31 (P<0.01) and to 0.264 mg kg⁻¹ after applications at GS37 (P<0.001). At ADAS Rosemaund, the same dose at tillering, GS31 and GS37 produced residues of 0.019, 0.045 and 0.101 mg kg⁻¹ respectively. Chlormequat residues were also reduced by applying lower rates of New 5C Cycocel (Table 1.7). Reducing the rate from 2.5 l ha⁻¹ to 0.625 l ha⁻¹ reduced the chlormequat residues from 0.173 to 0.047 mg kg⁻¹ at ADAS Gleadthorpe and from 0.045 to 0.019 mg kg⁻¹ at ADAS Gleadthorpe and Rosemaund respectively, but this was not a statistically significant reduction. Large differences in the size of the chlormequat residues were observed between the sites, with ADAS Gleadthorpe averaging 0.125 mg kg⁻¹ and ADAS Rosemaund averaging 0.043 mg kg⁻¹. Reasons for this difference are considered in the discussion.

New 5C Cycocel treatment	ADAS	ADAS	Mean
	Gleadthorpe	Rosemaund	
Nil	0.010	0.010	0.010
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS24	0.072	0.019	0.045
2.5 l ha ⁻¹ at GS30	0.119	0.048	0.083
2.5 l ha ⁻¹ at GS31	0.173	0.045	0.109
2.5 l ha ⁻¹ at GS32	0.171	0.058	0.115
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS37	0.264	0.101	0.183
0.625 l ha ⁻¹ at GS31	0.047	0.019	0.033
1.25 l ha ⁻¹ at GS31	0.134	0.029	0.081
1.875 l ha ⁻¹ at GS31	0.140	0.057	0.098
Mean	0.125	0.043	0.084
Site P-Value	0.0	01	
Site s.e.d. (4 df)	0.00	072	
Treatment P-Value	<0.0	001	
Treatment s.e.d. (32 df)	0.0	187	
Site * Treatment P-value	0.0	06	
Site * Treatment s.e.d (32)	0.02	259	

Table 1.7. Winter wheat chlormequat residues (mg kg⁻¹)

In winter barley, the greatest residues of about 0.196 mg kg⁻¹ at ADAS Gleadthorpe and 0.352 mg kg⁻¹at ADAS Rosemaund resulted from applications of 2.5 l ha⁻¹ at GS30, 31 or 32 (Table 1.8). These were greater (P<0.05) than the residues after applications during tillering or GS37, which averaged 0.100 mg kg⁻¹ at ADAS Gleadthorpe and 0.263 mg kg⁻¹at ADAS Rosemaund. Reducing the rate of application at GS31 from

2.5 l ha⁻¹ to 0.625 l ha⁻¹ reduced the chlormequat residues from an average over both sites of 0.272 mg kg⁻¹ to 0.175 mg kg⁻¹ (P<0.05). Halving the application rate of chlormequat reduced the residues from 0.272 to 0.204 mg kg⁻¹, but this reduction was not statistically significant.

New 5C Cycocel treatment	ADAS	ADAS	Mean
	Gleadthorpe	Rosemaund	
Nil	0.011	0.013	0.012
2.5 l ha ⁻¹ at GS24	-	0.247	-
2.5 l ha ⁻¹ at GS30	0.218	0.321	0.269
2.5 l ha ⁻¹ at GS31	0.191	0.353	0.272
2.5 l ha ⁻¹ at GS32	0.178	0.381	0.280
2.5 l ha ⁻¹ at GS37	0.100	0.279	0.190
0.625 l ha ⁻¹ at GS31	0.103	0.247	0.175
1.25 l ha ⁻¹ at GS31	0.154	0.254	0.204
1.875 l ha ⁻¹ at GS31	0.144	0.351	0.248
Mean	0.134	0.272	0.203
Site P-Value	0.	001	
Site s.e.d. (4 df)	0.0	0172	
Treatment P-Value	<0	.001	
Treatment s.e.d. (29 df)	0.0	338	
Site * Treatment P-value	N	NS	
Site * Treatment s.e.d (29 df)	0.0	0482	

Table 1.8. Winter barley chlormequat residues (mg kg $^{-1}$)

In winter oats, the chlormequat residues were reduced by applying New 5C Cycocel earlier (Table 1.9). At ADAS Rosemaund, applications of 2.5 l ha⁻¹ of chlormequat at GS30 produced a residue of 0.151 mg kg⁻¹ and this increased to 0.246 mg kg⁻¹at GS32. At Belfast, the same treatment at GS30 and GS32 caused the residues to increase from 0.133 to 0.623 mg kg⁻¹. The site x treatment interaction appears to be mainly caused by the magnitude of the effects produced by the half dose treatment. The rankings of the other treatments were similar at both sites. Table 1.10 shows the effect of a greater range of chlormequat rates and timings that were carried out at Belfast. This shows that halving the rate of New 5C Cycocel reduced the chlormequat residues at all the timings that were investigated (GS30, 31, 32 or 33) to on average 64% of the full rate residue levels. This also demonstrates that the effect of two half rate applications at different timings on the amount of residue are additive.

Wheat, barley and oats were grown at the ADAS Rosemaund site and each species had common treatments of 2.5 l ha⁻¹ of New 5C Cycocel at GS30, 31 and 32. This enabled the effect of species to be analysed at this site. This showed that barley had significantly greater residues at GS30, 31 and 32 than oats or wheat (P<0.05). On average, barley accumulated 0.351 mg kg⁻¹, oats accumulated 0.188 mg kg⁻¹and wheat

accumulated 0.050 mg kg⁻¹. However, at ADAS Gleadthorpe the residues for wheat and barley were not significantly different at 0.150 and 0.155 mg kg⁻¹ for wheat and barley respectively.

New 5C Cycocel treatment	Belfast	ADAS	Mean
-		Rosemaund	
Nil	0.030	0.039	0.034
2.5 l ha ⁻¹ at GS30	0.133	0.151	0.142
2.5 l ha ⁻¹ at GS31	0.357	0.144	0.250
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS32	0.623	0.246	0.435
2.5 l ha ⁻¹ at GS33	0.640	0.425	0.533
1.25 l ha ⁻¹ at GS32	0.373	0.085	0.229
Maan	0.250	0.192	0.270
Mean	0.359	0.182	0.270
Site P-Value		0.017	
Site s.e.d. (4 df)	C	0.0454	
Treatment P-Value	< 0.001		
Treatment s.e.d. (19 df)	0.0574		
Site * Treatment P-value	0.014		
Site * Treatment s.e.d (19 df)	0	0.0870	

Table 1.9. Winter oat chlormequat residues (mg kg⁻¹)

Table 1.10. Winter oat chlormequat residues (mg kg⁻¹) at Belfast

New 5C Cycocel treatment	Belfast
Nil	0.030
2.5 l ha ⁻¹ at GS30	0.133
2.5 l ha ⁻¹ at GS31	0.357
2.5 l ha ⁻¹ at GS32	0.623
2.5 l ha ⁻¹ at GS33	0.640
1.25 l ha ⁻¹ at GS30	0.082
1.25 l ha ⁻¹ at GS31	0.260
1.25 l ha ⁻¹ at GS32	0.373
1.25 l ha ⁻¹ at GS33	0.413
1.25 l ha ⁻¹ at GS30 & GS32	0.443
Mean	0.336
Treatment P-Value	< 0.001
Treatment s.e.d. (18 df)	0.0479

Wheat

In wheat, the rate at which shoots oscillate, known as the natural frequency, has been identified as a key determinant of lodging risk (Berry *et al.*, 2003). A higher natural frequency results in a smaller leverage force being exerted by the shoot on its base and consequently a lower risk to both stem and root lodging. This study showed that the natural frequency of the shoots was significantly increased by 2.5 l ha⁻¹ of New 5C Cycocel applied at GS30, 31 and 32, and by 1.875 l ha⁻¹ applied at GS31 (Table 1.11). New 5C Cycocel applied during tillering, GS37 or at half or quarter rates at GS31 did not significantly increase natural frequency. Height at centre of gravity is also an important determinant of lodging risk. New 5C Cycocel applied at GS30, 31 and 37 reduced the height at centre of gravity the most, but this was not statistically significant due to an unusually large SED for this parameter. Crop height was significantly reduced by the same treatments that significantly affected natural frequency (Table 1.11), with 2.5 l ha⁻¹ New 5C Cycocel applied at GS31 reducing height the most with a 6 cm reduction.

New 5C Cycocel did not significantly affect any of the other plant characters that were measured including; shoot number per plant, ear area, internode length, stem diameter, stem wall width, stem material strength, failure moment of the bottom two internodes, root number per plant, root plate spread and the structural rooting depth (see Appendix 2 for this data). The lack of statistically significant effects on the length of the bottom two internodes was a surprise. In fact, $2.5 \ 1 \ ha^{-1}$ New 5C Cycocel at GS31 reduced the length of internode 1 from about 54mm to 44mm at ADAS Gleadthorpe. At GS32, $2.5 \ 1 \ ha^{-1}$ New 5C Cycocel reduced the length of internode 2 from 88 to 81 mm at ADAS Gleadthorpe and from 94 to 85 mm at ADAS Rosemaund. A large SED for the lengths of internodes 1 and 2 meant that these differences were not statistically significant at the 5% level. The lack of effects on the plant characters that determine the strength of the stem base and anchorage system is almost entirely consistent with other chlormequat studies on winter wheat (Crook and Ennos, 1995; Easson *et al.*, 1995; Berry *et al.*, 2000). The only difference being that Crook and Ennos (1995) observed an increase in crown root number from 9 to 11 per plant after chlormequat was applied during early stem extension.

Table 1.11 Effect of New 5C Cycocel on the characters that determine shoot leverage in wheat

New 5C Cycocel	Natural frequency		Height at centre of			Crop height				
application	1 (000	(Hz)			gravity (mm)			(mm)		
	GT	RM	mean	GT	RM	mean	GT	RM	Mean	
Nil	0.92	0.73	0.83	428	420	424	781	641	711	
2.50l/ha @GS24	1.01	0.80	0.90	411	418	415	763	628	695	
2.50l/ha @GS30	1.04	0.77	0.91	403	412	408	737	608	673	
2.50l/ha @GS31	1.04	0.80	0.92	407	398	403	718	582	650	
2.50l/ha @GS32	1.03	0.78	0.91	414	411	413	733	627	680	

2.50l/ha @GS37	1.00	0.80	0.90	421	389	405	746	635	690
0.625l/ha @GS31	0.95	0.73	0.84	426	423	425	785	656	721
1.25l/ha @GS31	1.00	0.72	0.86	425	414	419	759	632	696
1.875l/ha@GS31	1.04	0.83	0.93	410	425	418	731	620	676
Mean	1.00	0.77	0.89	416	412	414	750	626	688
Site D value		0.008			NS			0.017	
		0.008						0.017	
Site SED (4 df)		0.0465			9.2			31.7	
Treatment P_Value		0.030			NS			0.001	
		0.037			10.0			0.001	
Treatment SED (32 df)		0.0332			10.2			14.2	
Interaction P-value		NS			NS			NS	
					10			110	
Interaction SED (32 df)		0.0642			16.4			36.9	

Barley

The efficacy of the New 5C Cycocel treatments for reducing lodging risk was assessed in terms of differences in the percentage area lodged and crop height. At ADAS Rosemaund, New 5C Cycocel applied during tillering increased the percentage area brackled at harvest from 38% to 77% (P<0.05). New 5C Cycocel applied at GS30, 31 and 32 also increased the amount of brackling to between 48 and 57%, but these increases were not statistically significant. There was no lodging or brackling at ADAS Gleadthorpe, crop height was therefore used to indicate any effects of treatment on lodging risk. However, New 5C Cycocel did not shorten crops significantly (Table 1.12).

New 5C Cycocel application	ADAS Rosemaund Percentage area brackled on 16 July	ADAS Gleadthorpe Crop height (mm)
Nil	38	773
2.50l/ha @GS24	77	-
2.50l/ha @GS30	57	742
2.50l/ha @GS31	48	765
2.50l/ha @GS32	50	748
2.50l/ha @GS37	37	765
0.625l/ha@GS31	55	733
1.25l/ha @GS31	53	744
1.875l/ha@GS31	57	772
Mean	52	755
Treatment P-Value	0.013	NS
Treatment SED (13 df)	8.7	28.1

Table 1.12. Effect of New 5C Cycocel on brackling at ADAS Rosemaund and crop height at ADAS Gleadthorpe

Oats

At ADAS Rosemaund, the efficacy of the New 5C Cycocel treatments for reducing lodging risk was assessed in terms of the changes made to the plant characters found to determine the lodging risk in wheat and the percentage area lodged. The reductions in height at centre of gravity and overall crop height, together with the increase in natural frequency, indicate that New 5C Cycocel at GS32 reduced lodging risk the most (Tables 1.13 and 1.14). The half rate dose at GS32 did not significantly change the plant characters associated with lodging. These results are supported by the lodging data (Table 1.13). The New 5C Cycocel treatments had no effect on the panicle area or shoot number, stem diameter, stem wall width, material strength of the stem wall, overall stem strength, root number, root plate spread or structural rooting depth (see Appendix 3 for this data). No lodging was observed at the Belfast site, however 2.5 ha⁻¹ New 5C Cycocel applied at GS30, 31, 32 or 33 reduced height by similar amounts (81 to 110 mm) (Table 1.14). Half rates of New 5C Cycocel reduced height by a similar amount to the full rate at GS31, 32 and 33, but did not reduce height at GS30.

Table 1.13 Winter oats at ADAS Rosemaund. Effect of New 5C Cycocel on lodging and plant characters that may determine shoot leverage.

New 5C Cycocel application	Percentage area lodged (30 July)	Shoot height at centre of gravity (mm)	Shoot natural frequency (Hz)
Nil	16.7	650	0.56
2.501/ha @GS30	5.0	611	0.65
2.50l/ha @GS31	8.3	605	0.64
2.501/ha @GS32	10.0	577	0.66
2.501/ha @GS33	18.3	649	0.54
1.25l/ha @GS32	18.3	635	0.58
Mean	12.8	621	0.60
Treatment P-Value	NS	0.061	NS
Treatment SED (10 df)	7.84	23.1	0.059

Table 1.14. Winter oats height (mm)

New 5C Cycocel treatment	Belfast	ADAS	Mean
		Rosemaund	
Nil	1077	1247	1162
2.5 l ha ⁻¹ at GS30	983	1246	1115
2.5 l ha ⁻¹ at GS31	983	1298	1141
2.5 l ha ⁻¹ at GS32	996	1207	1098
2.5 l ha ⁻¹ at GS33	967	1209	1088
1.25 l ha ⁻¹ at GS32	974	1257	1115
Mean	997	1243	1112
Site P-Value		0.001	

Site s.e.d. (4 df)	29.1	
Treatment P-Value	0.049	
Treatment s.e.d. (19 df)	23.4	
Site * Treatment P-value	0.062	
Site * Treatment s.e.d (df 19)	42.0	

Grain yield and specific weight

Large differences were observed between sites for both grain yield and specific weight. On average, the winter wheat at ADAS Rosemaund yielded 2.5 t ha⁻¹ more than at ADAS Gleadthorpe (P<0.010) (Table 1.16) and the specific weight was 2.4 kg hl⁻¹ greater than at ADAS Gleadthorpe (P<0.01). Across similar treatments, the winter barley at ADAS Rosemaund yielded 0.9 t ha⁻¹ more than at ADAS Gleadthorpe (P<0.05) (Table 1.17). However, the winter barley specific weight at ADAS Rosemaund was 1.4 kg hl⁻¹ less (P<0.05). On average, the oat yields at ADAS Rosemaund were 3.7 t ha⁻¹ more than at Belfast (Table 1.18).

Across both sites, the oat yield was reduced by 0.62 t ha^{-1} when New 5C Cycocel was applied at GS30 (P<0.05; Table 1.17). At Belfast, a half rate application at GS30 approximately halved the size of this yield reduction. Later applications (GS31 to GS33) caused smaller yield losses. In general, the yields and specific weight of wheat and barley (Tables 1.15 and 1.16) were not affected by applications of New 5C Cycocel. The only statistically significant effect on yield was found in the barley trial at ADAS Rosemaund where the treatment during tillering reduced yield by 0.53 t ha^{-1} .

New 5C Cycocel treatment	ADAS	ADAS	Mean
	Gleadthorpe	Rosemaund	
Nil	7.27	10.26	8.77
2.5 l ha ⁻¹ at GS24	7.71	10.06	8.89
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS30	7.65	9.98	8.81
2.5 l ha ⁻¹ at GS31	7.82	10.06	8.94
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS32	7.90	10.12	9.01
2.5 l ha ⁻¹ at GS37	7.52	10.12	8.82
0.625 l ha ⁻¹ at GS31	7.04	10.07	8.56
1.25 l ha ⁻¹ at GS31	7.90	10.20	9.05
1.875 l ha ⁻¹ at GS31	7.73	9.92	8.83
Mean	7.62	10.09	8.85
Site P-Value	0.0	10	
Site s.e.d. (4 df)	0.4	27	
Treatment P-Value	Ν	S	
Treatment s.e.d. (32 df)	0.1	78	
Site * Treatment P-value	Ν	S	
Site * Treatment s.e.d (32)	0.4	.89	

Table 1.16. Winter wheat yield (t ha⁻¹)

New 5C Cycocel treatment	ADAS	ADAS	Mean
	Gleadthorpe	Rosemaund	
Nil	7.00	8.11	7.55
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS2	-	7.47	-
2.5 l ha ⁻¹ at GS30	7.27	7.86	7.56
2.5 l ha ⁻¹ at GS31	7.27	8.08	7.68
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS32	7.24	8.17	7.70
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS37	7.12	8.30	7.71
0.625 l ha ⁻¹ at GS31	7.23	8.07	7.65
1.25 l ha ⁻¹ at GS31	7.20	8.12	7.66
1.875 l ha ⁻¹ at GS31	7.47	8.13	7.80
Mean	7.15	8.03	7.59
Site P-Value	<0	0.05	
Site s.e.d. (4 df)	0.2	225	
Treatment P-Value	<0	.01	
Treatment s.e.d. (29 df)	0.1	149	
Site * Treatment P-value	Ν	IS	
Site * Treatment s.e.d (29 df)	0.3	300	

Table 1.17. Winter barley yield (t ha⁻¹)

Table 1.18. Winter oats yield (t ha⁻¹)

New 5C Cycocel treatment	Belfast	ADAS	Mean
-		Rosemaund	
Nil	6.30	9.71	8.00
2.5 l ha ⁻¹ at GS30	5.58	9.18	7.38
2.5 l ha ⁻¹ at GS31	5.78	9.34	7.56
2.5 l ha ⁻¹ at GS32	5.75	9.34	7.55
2.5 l ha ⁻¹ at GS33	5.86	9.62	7.74
1.25 l ha ⁻¹ at GS32	5.90	10.03	7.97
Mean	5.86	9.54	7.70
Site P-Value		< 0.001	
Site s.e.d. (4 df)		0.265	
Treatment P-Value		0.023	
Treatment s.e.d. (20 df)		0.191	
Site * Treatment P-value		NS	
Site * Treatment s.e.d (20 df)		0.363	

Non PGR winter oat experiment

Most of the plots for cv. Gerald experienced some lodging, whereas negligible lodging was recorded in cv. Buffalo plots. For cv. Gerald, the nitrogen treatments significantly affected the amount of lodging (Table 1.19), whereas seed rate had no effect. The amount of lodging was related to the quantity of N fertilizer. Applying 100 or 120 kg N ha⁻¹ resulted in 10% or less of the plot lodged. Applications of 140 kg N ha⁻¹ resulted in 38% area lodged and 160 kg N ha⁻¹ resulted in 51-56% area lodged. The time of application appeared to have little effect on the amount of lodging. Yield was not affected by the variety, nitrogen or

seed rate treatments. The nitrogen treatment did affect the specific weight (P<0.001), with the 40, 40, 20 kg N ha⁻¹ treatment resulting in the greatest specific weight (50.8 kg hl⁻¹) and the 40, 40, 60 kg N ha⁻¹ treatment having the smallest specific weight (49.0 hl⁻¹). Ear number m⁻² appeared to be reduced in cv. Gerald by omitting the GS30-31 N application but was not affected by seed rate.

 Table 1.19. Effect of nitrogen treatments on lodging

Nitrogen	n treatment ($(kg N ha^{-1})$	% area lodged at harvest
			for cv. Gerald
			(mean across seed rates)
March	GS30/31	GS37-39	
40	40	20	7
40	40	40	10
40	40	60	38
40	40	80	51
40	60	60	56
40	0	60	5
P value			< 0.001
SED (6 d	f)		8.7

DISCUSSION

Large differences in the level of chlormequat were found between the cereal species and sites (Figure 1.2). Across the treatments applied at GS30, 31 and 32, winter wheat had the lowest residues at 0.10 mg kg⁻¹, followed by winter barley (0.27 mg kg⁻¹) and winter oats (0.28 mg kg⁻¹). For each species, large differences were observed between the sites, e.g. winter wheat residues averaged 0.05 mg kg⁻¹at ADAS Rosemaund and 0.15 mg kg⁻¹at ADAS Gleadthorpe (Figure 1.2). None of the factors measured in this study explain the large differences between sites and seasons. The site and species differences for the interval between applying the chlormequat and harvest appear to be too small to explain the variation in residues. Similarly the yield differences are not large enough for the residue differences to be caused by a dilution effect. It is difficult to draw conclusions about the effect of water availability because the site with the drought prone soil type experienced greater rainfall than the site with the water retentive soil type between the dates of treatment application and harvest. Other possible reasons for the site differences include the use of different cultivars and different levels of nitrogen fertilizer. Some existing literature shows that cultivar affects chlormequat residues and high nitrogen fertilizer can increase chlormequat residues in straw. However, other literature

contradicts these findings. A much more detailed study taking into account chlormequat uptake, climatic conditions at the time of treatment application, cultivar, fertilizer use and late season drought would be required to fully explain site differences in the absolute residues.



Figure 1.2. Mean chlormequat residues for the 2.5 l ha⁻¹ of New 5C Cycocel treatments at GS30, GS31 and GS32 combined. ADAS Rosemaund (closed columns), ADAS Gleadthorpe (hatched) and Belfast (open). S.E.D. = 0.050, 11 df).

In agreement with previous literature, this project shows that applications of chlormequat invariably result in chlormequat residues in the grain which are significantly less than the MRL. The primary objective of this project has been to investigate whether dose rate and timing of chlormequat can be optimised for reducing lodging risk and minimising chlormequat residues in the grain. The findings for wheat, oats then barley are considered in the following sections.

Wheat

The chlormequat residues in wheat grain were reduced by smaller rates and earlier applications of chlormequat. These findings are in agreement with published literature (Jung, 1964; El-Fouly and Fawzi, 1972; Gans *et al.*, 2000; Zmrhal and Machackova, 1981; Teittinen, 1975). Chlormequat was shown to reduce lodging risk by increasing natural frequency and reducing height. These effects can be summarised in terms of the effect on shoot leverage, which has been calculated using equation 1.3 with a fixed ear area and wind speed. The results show that 2.5 l ha⁻¹ 5C Cycocel significantly reduced shoot leverage when applied during tillering, GS30, GS31 and GS37, with the largest reduction at GS31 (Figure 1.3). Applying a ³/₄ rate at GS31 also significantly reduced shoot leverage (Figure 1.4). These results indicate that chlormequat residues can

be reduced, whilst maintaining efficacy of lodging control, by applying chlormequat earlier in the plant's lifecycle. Previous work has also shown that chlormequat applications before GS31 can effectively reduce lodging (Bragg *et al.*, 1984). The effectiveness of reducing the dose rate appears to be limited because lodging control is greatly reduced when dose rates below $\frac{3}{4}$ are used.

Oats

In oats, a model of wheat lodging was used to estimate the effects of chlormequat on shoot leverage at ADAS Rosemaund. This data shows that applying chlormequat at GS32 reduced leverage the most, from 741 Nmm to 578 Nmm (Appendix 3.1), with applications at GS30 and GS31 causing smaller reductions and the application at GS33 no reduction. However, none of the treatment differences were significant due to the large variability in height at centre of gravity and natural frequency of the shoots. The calculations for shoot leverage must be interpreted with care because this study assumed that the wheat lodging model of Berry et al. (2003) could be used to estimate the lodging risk of oats. This may not be true due to the different crop structure of oats compared with wheat. Therefore the estimates of how chlormequat affects the lodging risk of oats must be checked with a suitably developed and calibrated model of oat lodging. Changes in crop height have also been used to estimate the effect of chlormequat on lodging risk. Statistically significant effects were observed across both sites and these showed that applications at GS33 reduced height the most, followed by GS32 (Figure 1.5). These height observations, together with the calculations of leverage, indicate that chlormequat effects the greatest reduction in lodging when applied at or soon after GS32. This is in agreement with recommendations for applying chlormequat to oats (Anon., 2003). These results indicate that there is limited scope for minimising chlormequat residues in oats by applying chlormequat at earlier growth stages (Figure 1.5).

Of high importance to the grower were the yield reductions of about 0.5 t ha⁻¹ that resulted from chlormequat applications at GS30, 31 and 32. Yield reductions in oats have also been observed in response to chlormequat by Leitch and Hayes (1989) and in modern short varieties. A recent publication (Anon., 1999) by a commercial breeding company, Semundo Ltd., found that PGRs did not give a positive response on short oat varieties. Further studies must investigate the effect of chlormequat on the yield and lodging risk of modern short oat varieties. This may show that lodging risk in oats can be controlled more profitably through other husbandry methods, such as careful management of nitrogen applications (Table 1.19).

Barley

In the case of barley the first question to answer must be why chlormequat is applied to this crop species? This study showed that chlormequat neither reduced plant height, nor reduced brackling. These results are supported by previous studies (Green *et al.*, 1985; Koranteng and Matthews, 1982). The small effect of chlormequat on lodging and height of barley appears to be caused by less efficient translocation of

chlormequat to its biochemical target(s), and possibly less uptake into the leaves compared with other cereal species. Alcock and Morgan (1968) and Lord and Wheeler (1981) reported that about half as much chlormequat was translocated from barley leaves compared with wheat leaves after similar uptake of chlormequat into leaves for both species. Hunt and Baker (1982) also found that less chlormequat was able to enter the cuticle and leaf tissue of barley compared with wheat.

There is some evidence that chlormequat can increase barley yield in the absence of lodging and can increase tillering in barley. This project observed a non-significant yield increase of 4% in response to chlormequat at the site that experienced no lodging. Several studies have shown a yield improvement of 10-26% (Boothroyd and Nicholson, 1984; Matthews and Thompson, 1983; Matthews *et al.*, 1982). Some of these studies also observed an increase in ear number. This does not necessarily mean that chlormequat improves tillering since Matthews *et al.* (1983) observed that chlormequat increased ear number by reducing the proportion of tillers that died. However, many studies also report no yield improvement in response to chlormequat (e.g. Green *et al.*, 1985; Bragg *et al.*, 1984; Koranteng and Matthews, 1982) and some negative effects have been reported, such as a reduction in grain size (Green *et al.*, 1985). It therefore appears that small yield improvements in response to chlormequat are possible in the absence of lodging, but the response is very inconsistent.

Chlormequat is recommended for barley prior to GS31 (Anon., 2003). If we assume that applications during tillering or GS30 are equally likely to produce positive effects in the absence of lodging then there appears to be some scope for minimising residues in barley by applying before GS30. This project showed that applications during late tillering resulted in residues that were 33% less than from applications at GS30-32. There seems to be limited scope for minimising residues by reducing the dose rate since the dose had to be reduced to a quarter to reduce residues by only 36%, and it seems unlikely that the treatment will be as effective at such low rates. Applying chlormequat after GS30 did not increase residues in the grain and even decreased them when applied as late as GS37. This is in contrast to the observations for wheat and oats, which may indicate that in barley the efficiency of chlormequat timing on residues in barley, so it is impossible to compare these observations with others.



Figure 1.3. Winter wheat. Effect of application timing of New 5C Cycocel (@2.5 l ha⁻¹) on shoot leverage (columns) (s.e.d = 5.77, 32 df) and chlormequat grain residues (———) (s.e.d. = 0.034, 32 df).





Figure 1.5. Winter oats. Effect of application timing of New 5C Cycocel (@2.5 l ha⁻¹) on crop height (columns) (s.e.d. = 23.4, 19 df) and chlormequat grain residues (---) (s.e.d. = 0.057, 19 df).

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APPENDIX 2. Lodging associated measurements on winter wheat

New 5C Cycocel	Ear area			Shoot number			* Shoot leverage			
application		(cm^2)			per plant		(Nmm)			
	GT	RM	Mean	GT	RM	mean	GT	RM	Mean	
Nil	11.0	12.8	11.9	1.23	3.00	2.12	143	198	170	
2.50l/ha @GS2	11.4	12.0	11.7	1.37	3.53	2.45	130	184	157	
2.50l/ha @GS30	11.3	13.0	12.2	1.20	3.40	2.30	126	187	157	
2.50l/ha @GS31	12.1	13.3	12.7	1.33	3.40	2.37	128	177	152	
2.50l/ha @GS32	11.3	12.4	11.9	1.20	3.27	2.23	129	181	155	
2.50l/ha @GS37	12.2	13.5	12.8	1.27	3.30	2.28	134	174	154	
0.625l/ha@GS31	11.9	13.6	12.7	1.50	3.17	2.33	141	200	170	
1.251/ha @GS31	11.4	12.7	12.1	1.27	3.23	2.25	135	200	168	
1.875l/ha@GS31	11.2	13.1	12.2	1.33	3.40	2.37	128	182	155	
Mean	11.6	12.9	12.2	1.30	3.30	2.30	133	187	160	
Site P-value		0.020			< 0.001			0.006		
Site SED (4 df)		0.37			0.186			10.4		
Treatment P-Value		NS			NS			0.008		
Treatment SED (32 df)		0.40			0.218			5.8		
Interaction P-value		NS			NS			NS		
Interaction SED (32 df)		0.65			0.345			13.0		

Appendix 2.1. Effects on ear area, shoot number per plant and shoot leverage

* Shoot leverage calculated using a mean ear area of 12.2 cm^2

New 5C Cycocel	R	oot num	ber	Roo	t plate s	pread	Ro	ot plate d	lepth	Anc	horage st	rength
application		per plan	t		(mm)			(mm)			(Nmm)	
	GT	RM	Mean	GT	RM	Mean	GT	RM	Mean	GT	RM	Mean
Nil	15.8	19.7	17.8	38.5	38.7	38.6	38.6	25.3	32.0	155	154	154
2.50l/ha @GS24	15.4	19.2	17.3	35.4	38.4	36.9	34.9	28.7	31.8	124	153	139
2.50l/ha @GS30	15.6	23.1	19.3	37.2	39.7	38.5	41.0	25.7	33.4	136	165	151
2.50l/ha @GS31	14.7	21.3	18.0	34.7	42.4	38.6	30.8	26.6	28.7	112	199	155
2.50l/ha @GS32	16.3	20.8	18.5	34.0	37.0	35.5	30.5	26.5	28.5	108	133	121
2.50l/ha @GS37	17.1	21.6	19.3	40.8	38.1	39.4	37.1	27.7	32.4	184	145	165
0.6251/ha @GS31	15.9	18.9	17.4	37.3	38.1	37.7	35.7	25.3	30.5	158	145	151
1.251/ha @GS31	14.3	17.9	16.1	36.1	40.9	38.5	30.9	27.2	29.0	124	182	153
1.875l/ha@GS31	15.2	19.2	17.2	35.8	45.6	40.7	34.8	31.1	32.9	121	250	186
Mean	15.6	20.2	17.9	36.6	39.9	38.3	34.9	27.1	31.0	136	170	153
Site P-value		0.044			NS			NS			NS	
Site SED (4 df)		1.58			1.29			2.86			14.6	
Treatment P-Value		NS			NS			NS			NS	
Treatment SED (32 df)		1.56			2.07			2.06			25.2	
Interaction P-value		NS			NS			0.043			NS	
Interaction SED (32 df)		2.61			3.05			3.96			36.7	

Appendix 2.2. Effects on plant characters that determine anchorage strength

New 5C Cycocel application		Length	l		Diamete (mm)	r	V	Wall widt (mm)	h	Fail	ure yield (Mpa)	stress	Failur	e moment	(Nmm)
	GT	RM	Mean	GT	RM	mean	GT	RM	mean	GT	RM	mean	GT	RM	Mean
Nil	54.1	55.7	54.9	3.48	4.23	3.85	0.597	0.658	0.627	31.2	26.1	28.6	104	151	127
2.50l/ha @GS24	45.4	57.5	51.5	3.50	4.46	3.98	0.621	0.677	0.649	33.2	18.4	25.8	113	123	118
2.50l/ha @GS30	47.8	57.3	52.5	3.59	4.23	3.91	0.693	0.590	0.642	29.0	25.3	27.1	113	1345	124
2.50l/ha @GS31	43.7	60.6	52.2	3.46	4.28	3.87	0.639	0.619	0.629	40.2	21.8	31.0	137	120	129
2.50l/ha @GS32	46.9	57.5	52.2	3.48	4.17	3.83	0.591	0.620	0.606	33.6	20.9	27.3	113	113	113
2.50l/ha @GS37	51.1	55.2	53.2	3.51	4.45	3.98	0.653	0.685	0.669	36.4	18.7	27.6	130	125	128
0.625l/ha@GS31	41.6	56.1	48.9	3.42	4.26	3.84	0.684	0.628	0.656	36.8	21.7	29.3	125	123	124
1.251/ha @GS31	43.6	60.1	51.9	3.47	4.09	3.78	0.606	0.572	0.589	37.6	23.3	30.5	129	114	121
1.875l/ha@GS31	47.3	66.3	56.8	3.47	4.46	3.96	0.584	0.690	0.637	30.5	18.7	24.6	101	125	113
Mean	46.8	58.5	52.7	3.49	4.29	3.89	0.630	0.638	0.634	34.3	21.7	28.0	118	126	122
Site P-value		0.003			< 0.001			NS			0.006			NS	
Site SED (4 df)		1.81			0.046			0.0163			2.35			8.1	
Treatment P-Value		NS			NS			NS			NS			NS	
Treatment SED (32 df)		3.44			0.070			0.0327			3.03			12.3	
Interaction P-value		NS			NS			NS			NS			NS	
Interaction SED (32 df)		4.93			0.104			0.0466			4.68			18.3	

Appendix 2.3. Effects on plant characters that determine the strength of internode 1

New 5C Cycocel		Length			Diameter	r	V	Wall widt	h	Fail	ure yield	stress	Failur	e moment	(Nmm)
application		(mm)			(mm)			(mm)			(Mpa)				
	GT	RM	Mean	GT	RM	mean	GT	RM	mean	GT	RM	mean	GT	RM	Mean
Nil	87.8	93.6	90.7	3.83	5.04	4.43	0.605	0.712	0.659	21.6	11.2	16.4	91	104	98
2.50l/ha @GS24	79.6	95.4	87.5	4.01	5.20	4.61	0.676	0.762	0.719	21.0	11.7	16.4	107	122	115
2.50l/ha @GS30	81.3	91.6	86.4	4.08	4.79	4.43	0.705	0.651	0.678	18.4	11.2	14.8	100	87	93
2.50l/ha @GS31	81.2	86.6	83.9	3.96	5.17	4.56	0.654	0.694	0.674	25.8	8.8	17.3	125	85	105
2.50l/ha @GS32	80.8	85.0	82.9	3.90	4.89	4.39	0.618	0.708	0.663	22.5	9.9	16.2	100	86	93
2.50l/ha @GS37	89.1	89.0	89.0	3.99	5.18	4.59	0.676	0.702	0.689	22.4	8.9	15.7	112	87	100
0.625l/ha @GS31	83.1	91.6	87.3	3.94	5.11	4.53	0.683	0.723	0.703	22.5	9.3	15.9	109	89	99
1.25l/ha @GS31	76.1	93.0	84.5	3.82	4.95	4.38	0.614	0.714	0.664	24.4	10.5	17.4	106	93	99
1.875l/ha @GS31	80.4	94.2	87.3	3.80	5.28	4.54	0.574	0.744	0.659	22.1	12.1	17.1	89	127	108
Mean	82.2	91.1	86.3	3.92	5.06	4.49	0.645	0.712	0.679	22.3	10.4	16.4	104	98	101
Site P-value		< 0.001			< 0.001			0.033			0.003			NS	
Site SED (4 df)		0.71			0.064			0.0211			1.85			6.2	
Treatment P-Value		NS			NS			NS			NS			NS	
Treatment SED (32 df)		3.11			0.087			0.0399			1.42			9.9	
Interaction P-value		NS			0.019			NS			NS			0.014	
Interaction SED (32 df)		4.21			0.133			0.0572			2.64			14.5	

Appendix 2.4. Effects on plant characters that determine the strength of internode 2

	~	0.11					
New 5C Cycocel	Sten	n failure	wind	Anchorage failure			
application	sp	eed (m s	s ⁻¹)	wind speed (m s^{-1})			
	GT	RM	Mean	GT	RM	Mean	
Nil	11.8	10.4	11.1	11.1	6.65	8.88	
2.50l/ha @GS24	12.4	10.8	11.6	11.7	6.92	9.29	
2.50l/ha @GS30	12.6	10.7	11.6	11.9	6.85	9.35	
2.50l/ha @GS31	12.5	11.0	11.8	11.8	7.02	9.41	
2.50l/ha @GS32	12.4	10.8	11.6	11.7	6.93	9.31	
2.50l/ha @GS37	12.2	11.1	11.7	11.5	7.09	9.30	
0.625l/ha@GS31	11.9	10.4	11.1	11.2	6.62	8.92	
1.251/ha @GS31	12.2	10.3	11.3	11.5	6.61	9.04	
1.875l/ha@GS31	12.5	10.9	11.7	11.8	6.97	9.37	
Mean	12.3	10.7	11.5	11.6	6.85	9.21	
Sita D value		0.000			<0.001		
Sile P-value		0.009			<0.001		
Site SED (4 df)		0.33			0.236		
Treatment P-Value		0.008			0.007		
Treatment SED (32 df)		0.20			0.158		
(52 df)		0.20			0.100		
Interaction P-value		NS			NS		
Interaction SED (32 df)		0.43			0.316		

Appendix 2.5. Effect of changes in shoot natural frequency and Height at centre of gravity on stem failure wind speed and anchorage failure wind speed

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New 5C Cycocel treatment	Ear area	Shoot number	* Shoot leverage
	(cm^2)	per plant	(Nmm)
Nil	26.0	3.87	741
2.5 l ha ⁻¹ at GS30	29.2	3.20	618
2.5 l ha ⁻¹ at GS31	30.7	3.97	614
2.5 l ha ⁻¹ at GS32	24.5	3.67	578
2.5 l ha ⁻¹ at GS33	30.8	3.67	789
1.25 l ha ⁻¹ at GS32	32.9	3.33	687
Mean	29.0	3.62	671
Treatment P-Value	NS	NS	NS
Treatment s.e.d. (10 df)	4.50	0.381	91.2

Appendix 3.1. Effects on ear area, shoot number per plant and shoot leverage

* Shoot leverage calculated using mean panicle area.

New 5C Cycocel treatment	Root number	Root plate spread	Root plate depth	Anchorage strength
	per plant	(mm)	(mm)	(Nmm)
Nil	19.8	35.1	25.2	114
2.5 l ha ⁻¹ at GS30	17.3	30.4	24.7	74
2.5 l ha ⁻¹ at GS31	22.3	31.0	25.8	79
2.5 l ha ⁻¹ at GS32	17.6	32.7	23.4	93
2.5 l ha ⁻¹ at GS33	22.5	32.1	24.7	88
1.25 l ha ⁻¹ at GS32	19.1	30.7	24.8	77
Mean	19.8	32.0	24.8	88
Treatment P-Value	NS	NS	NS	NS
Treatment s.e.d. (10 df)	2.99	1.94	1.02	17.2

Appendix 3.2. Effects on plant characters that deter	mine anchorage strength

New 5C Cycocel treatment	Length (mm)	Diameter (mm)	Wall width (mm)	Failure yield stress (Mpa)	Failure moment (Nmm)
Nil	76	5.07	0.770	19.4	188
2.5 l ha ⁻¹ at GS30	60	4.99	0.761	21.6	203
2.5 l ha ⁻¹ at GS31	70	4.96	0.792	19.5	184
2.5 l ha ⁻¹ at GS32	68	4.58	0.742	25.5	188
2.5 l ha ⁻¹ at GS33	86	5.26	0.817	19.2	220
1.25 l ha ⁻¹ at GS32	71	5.26	0.810	19.1	208
Mean	72	5.02	0.782	20.7	199
Treatment P-Value	NS	NS	NS	NS	NS
Treatment s.e.d. (10 df)	10.7	0.231	0.0422	5.25	55.5

Appendix 3.3. Effects on plant characters that determine the strength of internode 1

Appendix 3.4. Effects on plant characters that determine the strength of internode 2

New 5C Cycocel treatment	Length (mm)	Diameter (mm)	Wall width (mm)	Failure yield stress (Mpa)	Failure moment (Nmm)
Nil	160	5.57	0.630	12.1	132
2.5 l ha ⁻¹ at GS30	134	5.48	0.624	13.7	138
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS31	144	5.44	0.674	10.4	113
$2.5 \mathrm{l}\mathrm{ha}^{-1}$ at GS32	138	5.05	0.618	14.3	119
2.5 l ha ⁻¹ at GS33	159	5.69	0.661	10.1	120
1.25 l ha ⁻¹ at GS32	142	5.66	0.671	10.8	127
Mean	146	5.48	0.646	11.9	125
Treatment P-Value	NS	NS	NS	NS	NS
Treatment s.e.d. (10 df)	15.4	0.209	0.0599	2.86	27.7

New 5C Cycocel treatment	Stem failure	Anchorage
	wind speed	failure wind speed
	$(m s^{-1})$	$(m s^{-1})$
Nil	7.06	6.24
2.5 l ha ⁻¹ at GS30	7.45	6.50
2.5 l ha ⁻¹ at GS31	6.97	5.85
2.5 l ha ⁻¹ at GS32	8.18	6.92
2.5 l ha ⁻¹ at GS33	6.64	5.40
1.25 l ha ⁻¹ at GS32	6.66	5.56
Mean	7.16	6.08
Treatment P-Value	NS	NS
Treatment s.e.d. (10 df)	1.136	1.008

Appendix 3.5. Effect of changes in shoot natural frequency and Height at centre of gravity on stem failure wind speed and anchorage failure wind speed