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Cereals for bioethanol: quantifying the alcohol yield of UK hard wheats and the grain yields and N requirements of triticale in the second cereal position

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ABBREVIATIONS USED

ANOVA	Analysis of Variance	RL	HGCA Recommended Lists
AY	Alcohol yield	RV	Residue viscosity
CEL	Crop Evaluation Ltd	SED	Standard Error of Difference
DM	Dry matter	SWRI	Scotch Whisky Research
ha	Hectare		Institute
hL	Hectolitre	t	Metric tonne
L	Litre	SF	Suffolk (field trial site)
LSD	Least Significant Difference	Spwt	Specific weight
Ν	Nitrogen	то	Towthorpe (field trial site)
NUE	Nitrogen Use Efficiency	TT	Terrington (field trial site)
PGR	Plant Growth Regulator		

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CONTENTS

1.	ABSTR	ACT6
2.	SUMMA	ARY7
	2.1. Ir	ntroduction7
	2.1.1.	Objectives7
	2.1.2.	Background7
	2.1.3.	Summary9
	2.2. M	aterials and methods9
	2.2.1.	Hard wheat samples for alcohol yield testing9
	2.2.2.	Wheat-triticale N response experiments10
	2.3. R	esults10
	2.3.1.	Alcohol yield of hard wheats10
	2.3.2.	Grain yields and N requirements of wheat and triticale11
	2.4. Di	iscussion/Conclusions and implications15
3.	TECHN	ICAL DETAIL17
	3.1. Ir	ntroduction17
	3.1.1.	Overall aim17
	3.1.2.	Specific objectives
	3.1.3.	Background
	3.1.4.	Benefits of Group 3 and 4 wheats as biofuel feedstocks in the first
		cereal position
	3.1.5.	Advantages of triticale19
	3.1.6.	Summary
	3.2. M	aterials and methods22
	3.2.1.	Hard wheat samples for alcohol yield testing
	3.2.2.	Alcohol yield determination 22
	3.2.3.	Field Experiments to compare performance of wheat and triticale 24
	3.2.4.	Grain protein analyses25
	3.2.5.	Data analysis25

3.3.	Results27
3.3.1.	Alcohol yield of hard wheats from RL 200927
3.3.2.	Wheat and triticale 2010: Grain yield, specific weight and optimum N rate
3.3.3.	Wheat and triticale 2010: Protein concentration and alcohol yield 34
3.3.4.	Wheat and triticale 2010: Supplementary study, Suffolk species trial 36
3.4.	Discussion40
3.4.1.	Alcohol yields of hard wheats40
3.4.2.	Agronomic performance of wheat and triticale and predicted alcohol yields
3.4.3.	Observations on grain quality44
3.4.4.	Conclusions and Recommendations45
3.5.	References

1. ABSTRACT

This project is the first to consider the performance of hard wheat varieties for bioethanol production in the UK. It is also the first to describe a series of experiments in which modern winter wheat and triticale varieties have been studied under comparable conditions of nitrogen (N) nutrition and crop management. This latter work was carried out in the context of identifying the best cereals for bioethanol production, particularly in the second position in the rotation.

In the first part of the study, 10 wheat varieties were taken from six HGCA Recommended Lists sites in 2009 (56 samples in total) and analysed for alcohol yield (AY) and residue viscosity (RV) using a method previously applied to distilling wheats. There were significant differences between hard wheat varieties; Conqueror and Oakley had particularly high AY and Ketchum had low AY. Glasgow as a soft wheat reference variety demonstrated superior AY, outperforming all the hard wheats. There were no differences in RV between hard wheat varieties, indicating that they are equally amenable for bioethanol processing. The higher AY of Conqueror and Oakley was principally due to their lower grain proteins, which probably reflect a yield dilution effect, rather than underlying genetic differences in grain composition. The combination of high AY and high grain yield meant that Conqueror and Oakley had the highest yield of alcohol per hectare.

With regards to alternative feedstocks for bioethanol, the results point to a substantial opportunity for the use of triticale to displace wheat. In five out of six trials carried out between 2007 and 2010, triticale out-yielded wheat when studied in the first or second cereal position on high yield potential 'wheat land'. At the sixth site, triticale matched but did not out-yield wheat, only because of post-maturity lodging at the higher N rates. Within these experiments, where full N response trials were carried out, triticale had a lower N optimum than wheat in one experiment, and the same optima as wheat in another two. In a fourth experiment, there were two triticale varieties with lower optima and two with similar optima to wheat. N optima for triticale appear to be higher than stated in the Defra Fertiliser Manual. Given the higher grain yield with the same and/or less N, and higher straw yields, these results clearly indicate that triticale has higher N use efficiency than wheat. The report makes recommendations for further work on wheat and triticale for bioethanol production, and to identify more N efficient cereal species.

2. SUMMARY

2.1. Introduction

2.1.1. Objectives

 To determine alcohol yields of current Group 3 and Group 4 hard wheat varieties
To evaluate the grain yield, alcohol yield and N requirements of triticale in relation to wheat

2.1.2. Background

With the opening of the Ensus bioethanol plant on Teeside in spring 2010 and the Vivergo plant on Humberside scheduled to open in 2011, growing grain for bioethanol is now a reality, with up to 2 million tonnes of grain expected to be used from the 2011 harvest. Both variety and agronomy can affect the value of grain to the processor, mainly through the alcohol yield achieved per tonne of grain. In addition, through Carbon Reporting in the Renewable Energy Directive the Greenhouse Gas (GHG) savings of the biofuel are also important to the processor. Given that over 67% of the GHG costs of bioethanol are due to crop production, reducing these GHG costs will ultimately be important, and will be part of specifications to the grower.

The keys to maximising the GHG savings associated with bioethanol production include maximising grain yields, and minimising Nitrogen (N) inputs. Nitrogen fertilisation constitutes over 70% of the GHG and 20% of the economic cost of production of wheat.

In a first wheat position, to maximise both profitability and GHG savings, variety choice will be driven primarily by a high grain yield (as reported in the HGCA Recommended Lists), but processing performance and alcohol yield will still be important to the industry. In a second wheat position the situation is different because yield tends to be lower due to build-up of diseases such as take-all, because N applications are generally higher, and because grain protein is generally higher (hence reducing alcohol yields). All of these factors (especially low yield and higher N inputs) reduce the profitability of the crop and the potential GHG savings of the biofuel. It is possible that alternative cereals may have similar or higher yields than wheat in the

second or later rotational positions, and have a lower N requirement. Of the likely candidates, barley and oats are unlikely to be of use due to their husked grain and hence relatively low starch content, and rye has low yields. This leaves triticale, a hybrid of rye and wheat.

Varieties in HGCA Recommended List trials are tested for their alcohol yields by the Scotch Whisky Research Institute (SWRI) and there are some indicators of the best varietal types suitable for distilling (see HGCA Information Sheet 11, 2010). These tests are currently restricted to soft wheats, however, with the existing distilling industry experiencing problems in processing hard wheats. In contrast, the new bioethanol plants inevitably take in a range of wheat varieties including hard endosperm types. However, little is known about the performance of hard wheat varieties for alcohol production in the UK. Clearly some hard wheat varieties may be useful for bioethanol production, particularly those with high yield potential and lower grain proteins than milling wheat varieties (e.g. Oakley). However, further work is needed to determine the AY and processing characteristics of the Group 3 and 4 hard wheats to guide variety choice in the developing bioethanol industry.

Carbon reporting for the RED now applies at farm level and a GHG incentivisation scheme is likely. Early work has demonstrated that the ideal wheat variety for bioethanol is high-yielding with low protein content and low N fertiliser requirements. However, wheat varieties showing big improvements in N use efficiency are not currently available within elite germplasm. Initial investigations have suggested that triticale could deliver comparable yields at a lower N input, and moreover that this species was being used in Sweden for bioethanol production.

Agronomy trials carried out around 5-10 years ago indicated that modern triticale varieties could yield up to 9 t/ha as a first cereal and 8 t/ha as a second or third cereal when grown on sites with high yield potential and managed according to best agronomic practice for wheat (e.g. good disease and lodging control).

Given the potential displayed by triticale in a 'look-see' study in 2007, a dedicated trial funded by breeders was subsequently designed to compare wheat and triticale in a second cereal position on good wheat land in 2009. Importantly, the design incorporated an N response trial, with identical N rates for each variety, to allow curve fitting and determination of economic N optima. Based on the fitted optima, triticale

yielded approximately 10% more than wheat, with *ca*. 20% lower N optima. Even with a discount applied to the value of triticale grain (relative to wheat grain), triticale still showed a greater financial margin over N inputs than did wheat.

Clearly, these are very important results, both in the context of improving profitability of cereal production, but also in terms of reducing GHG emissions, assuming that modern triticale varieties could find a ready market in the bioethanol industry today. Based on this one N-response trial, there are insufficient data to take a conclusive message to the industry, and further work is needed to validate the potential of triticale to wheat in the second or third cereal position. Therefore the work outlined in the current project aimed to repeat the study described above at two sites in 2009/10. The results of an additional breeder-funded study are included, to bring the number of N response data sets from 2010 harvest to three.

2.1.3. Summary

The aims of this project were to: (i) provide information on the suitability of hard Group 3 and 4 winter wheat varieties for alcohol production, and (ii) investigate the performance of winter triticale in the second cereal position as a potential low-input, high yield feedstock for alcohol production. An additional commercial trial harvested in 2010 is also reported for wheat and triticale in the first cereal position.

2.2. Materials and methods

2.2.1. Hard wheat samples for alcohol yield testing

Fifty-six wheat grain samples were provided by Crop Evaluation Ltd (CEL) from the 2009 winter wheat Recommended List trial series. The ten varieties selected were Glasgow, Warrior, Oakley, Duxford, JB Diego, Ketchum, Panorama, Grafton, Conqueror and KWS Sterling. Alcohol yield and residue viscosity was tested in the laboratory based on the 'wheat-cook' method of the Scotch Whisky Research Institute, using samples of soft wheats Glasgow and Warrior as reference varieties.

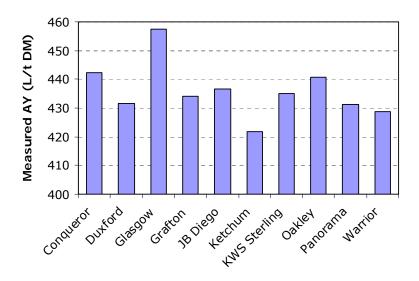
2.2.2. Wheat-triticale N response experiments

Two wheat-triticale N response experiments were carried out in the 2009/10 field season in the second cereal position. The first was at Towthorpe, Near Malton, East Yorkshire and the second at Terrington St Clement, King's Lynn, Norfolk. At each site, two winter wheat (JB Diego and Viscount) and two winter triticale (Benetto and Grenado) varieties were each tested at five nitrogen (N) rates. A third wheat-triticale experiment funded by breeders was carried out in the 2009/10 growing season within a larger species trial comparing winter wheat, triticale, barley, oats and rye. This experiment was located at Cransford in Suffolk on a clay loam soil, also in the second cereal position. Grain yield was determined at harvest, and grain protein and specific weight determined in all experiments, and the economic optimum N rate determined by curve fitting. In individual trials, assessments were made of lodging, incidence of take-all, and pre-harvest grab samples taken for determination of total biomass.

2.3. Results

2.3.1. Alcohol yield of hard wheats

The wheat samples selected were all feed types, with relatively low grain protein contents. There were significant variety differences in protein content, with Conqueror, Glasgow, Duxford and Oakley having lower protein contents than Grafton, Ketchum, Panorama and Warrior. Measured alcohol yield (AY) for the hard wheats, compared to the reference varieties of soft wheat; Glasgow (high AY) and Warrior (low AY) averaged across sites are shown in Summary Figure 1. Variety had a significant effect on AY whereby Glasgow was confirmed as the superior wheat, having significantly higher AY than the other varieties. However, Conqueror and Oakley also had significantly higher AY than the worst varieties Ketchum and Warrior.

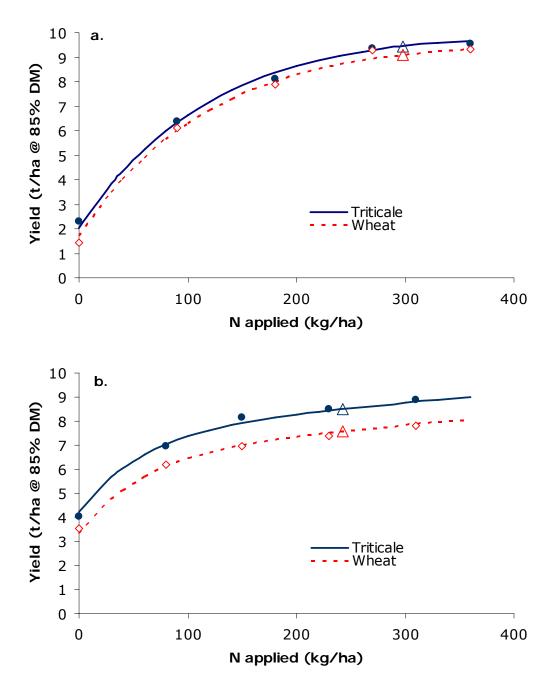


Summary Figure 1. Alcohol yield of ten wheat varieties from the 2009 Recommended List.

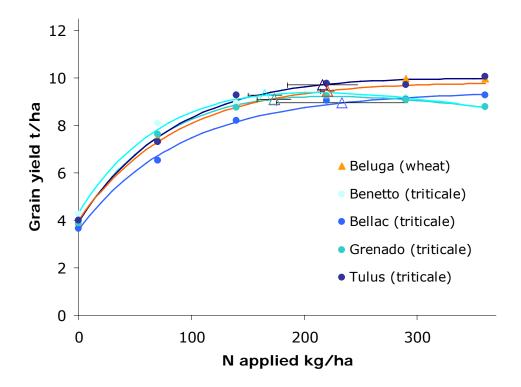
The good performance of Conqueror and Oakley was in part due to their lower protein contents. When the protein effect was removed by standardising AY at 11.5% protein, only Glasgow was shown to be significantly different to the rest. The hard wheats were very similar in AY at a fixed protein content. When combined with grain yields to estimate alcohol yields per hectare, Conqueror and Oakley were seen to be the best performing hard wheats. Residue viscosity was significantly influenced by variety, with Warrior having the highest residue viscosity (indicative of problems during processing). However no other hard wheat variety was significantly different to the others in terms of residue viscosity.

2.3.2. Grain yields and N requirements of wheat and triticale

In both the Towthorpe and Terrington N response experiments, the triticale varieties significantly out-yielded the wheat varieties (Summary Figure 2). A third N x species experiment in 2010, where four triticale varieties were compared to a number of wheat varieties, showed that triticale matched, but did not exceed, the yield of winter wheat (Summary Figure 3).



Summary Figure 2. Effect of N on yield of triticale and wheat (data points and fitted curves), including yields at optimum N rates (triangles) at a) Towthorpe, and b) Terrington in 2010.



Summary Figure 3. Effect of N on yield of triticale and wheat (data points and fitted curves), at a second cereal site in Suffolk in 2010, including yields at optimum N rates (triangles).

Over two field seasons (2009 and 2010) where full N response experiments were carried out, we have therefore shown that on high yield potential land in the second cereal position, triticale has out-yielded wheat on three occasions and matched wheat in the fourth. In 2009 triticale had significantly lower N optima, but in 2010 it had the same N optima as wheat at two sites (Terrington and Towthorpe), and an N optima that varied between varieties at the third site (Suffolk); two triticale varieties having lower optima than wheat varieties (albeit associated with greater lodging at higher N rates), and two others having N optima similar to wheat. The N optima in these experiments (Suffolk, 199; Terrington, 243; Towthorpe, 298 kgN/ha) were higher than those that would be recommended in the Fertiliser Manual where the highest recommendation is currently 150 kgN/ha for triticale.

Overall, grain protein contents and likely alcohol yields of triticale can be expected to be similar to wheat, with variation between cultivars, as is the case for wheat.

Some measures of take-all were made on the Suffolk trial and its incidence was shown to be significantly lower in triticale than in the wheat varieties tested (8% and 19% respectively). Further work over a wider range of seasons is required to quantify the true take-all resistance of modern triticale varieties, and to distinguish this from traits such as a faster rate of root expansion, which could enable the crop to overcome pathogen attack and give increased ability to capture N.

Better nitrogen use efficiency (NUE) is also an important trait in triticale. In 2009, better grain and straw yields (i.e. greater total biomass) than wheat were observed, with less applied N. The resources were not available in 2010 to examine N partitioning and total biomass in all these experiments, but it was confirmed that triticale in the Suffolk trial had greater straw biomass (particularly for the variety Benetto). Nevertheless, with the Towthorpe and Terrington crops, it is clear that triticale produced more grain with the same amount of N applied, i.e. better NUE. However, the basis for this better performance remains unknown. It seems to be due to a combination of greater recovery of soil N (i.e. higher nil-N yields) and greater recovery/utilisation of fertiliser N; analysis of dry matter and nitrogen harvest indices is required to understand the better NUE of triticale.

One disadvantage of triticale in the 2010 experiments at two sites was lodging at the highest N rates, although the variety Tulus was fairly resistant to lodging and gave the highest yield of both species. The results suggest that if lodging could be better controlled in more of the triticale varieties, yields could be even higher. This warrants further work, both on plant growth regulators, and in understanding inherent lodging risk and how triticale relates to wheat in terms of root plate spread, stem strength etc. It should be noted, however, that there was no lodging at the Towthorpe site in 2010.

The ADAS work to date has focussed on triticale in the second cereal position as it is believed this is where the main advantage of triticale will lie, by making better use of its inherent take-all resistance and NUE when roots are compromised. In the first cereal position, we may not expect triticale to outperform wheat, which has had more sustained breeding effort in the UK. Despite this, a 0.5 t/ha yield advantage of triticale was seen in first wheat experiment in 2007 compared to 40 wheat varieties. Also in 2010, Agrovista carried out a trial at Eryholme, Nr Darlington, where wheat and triticale followed oilseed rape, where the two triticale varieties out-yielded the wheat average by 1.83 t/ha, and out-yielded the top yielding wheat (Robigus) by 0.47 t/ha.

These results point to a substantial opportunity from the use of triticale to displace wheat for animal feed as well as bioethanol use; in five of the six trials we have studied, which have compared wheat with triticale over the past four years, triticale has significantly and substantially out-yielded wheat, whether in first or second cereal positions. At the other site, triticale did not out-yield wheat only because of postmaturity lodging at the higher N rates.

2.4. Discussion/Conclusions and implications

This report is the first to publish actual alcohol yields and residue viscosities of hard wheat varieties from UK Recommended List trials, and to compare them to reference varieties of soft wheat using a laboratory method. It is also the first to describe a series of N response experiments in which wheat and triticale have been studied under truly comparable conditions with respect to fertiliser N, with assessments made of both grain yield and quality.

With respect to the alcohol yield of hard wheat varieties grown on a number of Recommended List sites in 2009, the conclusions are as follows:

- 1. There are significant differences in AY between hard wheat varieties, with Conqueror and Oakley having particularly high AY, and Ketchum a particularly low AY.
- 2. Glasgow as a soft wheat reference demonstrates superior AY, outperforming the hard wheat varieties.
- 3. There were no significant differences in residue viscosity between hard wheat varieties, and none with the undesirable character of high residue viscosity, as demonstrated by the soft wheat variety Warrior.
- 4. The higher AY demonstrated by Conqueror and Oakley were most likely due to their lower grain protein contents (compared to the other hard wheat varieties grown at the same sites) and hence is more likely to reflect a yield (protein dilution) effect, rather a solely genetic effect.
- 5. The combination of high grain yield and high alcohol yield meant that Oakley and Conqueror had the highest alcohol productivity per unit area, indicating their value for maximising GHG savings.

Taking into account the three wheat and triticale N response experiments carried out in 2010, together with a previous experiment carried out in 2009, the conclusions are as follows:

- 6. Triticale out-yielded wheat on three occasions and matched wheat yield in the fourth when grown in the second cereal position and with similar N applications.
- Relative grain protein contents and predicted alcohol yield between triticale and wheat are broadly similar, but differences are inconsistent between sites and protein measurement methods.
- 8. Triticale had a lower N optimum for yield than wheat in one experiment, had the same N optima as wheat in two experiments, and in one experiment there were two triticale varieties with lower N optima than wheat, and two with similar N optima to wheat.
- 9. N optima for triticale appear to be higher than stated in the Defra Fertiliser Manual, however this is the first series of experiments to study the performance of triticale on 'typical wheat' land of high yield potential.
- 10. Given the higher yield with the same and/or less N, these results clearly indicate that triticale can have higher nitrogen use efficiency than wheat.
- 11. These performance benefits of triticale could be greater in a year with a higher incidence of take-all.
- 12. Triticale also appeared to produce more straw and hence total biomass then wheat, which could be particularly valuable in the context of burning biomass for energy.
- 13. Significant lodging in triticale was seen in two trials, although if the crops had been harvested earlier, it is likely that triticale would have outperformed wheat to an even greater extent.
- 14. The results confirm that triticale tends to have lower specific weights than wheat, even in the second cereal position, although this may not be important for bioethanol production.
- 15. In an ADAS trial in 2007 triticale out-yielded wheat by 0.59 t/ha, and an independent commercial trial in 2010 showed that triticale out-yielded wheat by 1.83 t/ha, both trials being carried out in the first cereal position.

Recommendations for further work are described in the scientific report.

3. TECHNICAL DETAIL

3.1. Introduction

3.1.1. Overall aim

To maximise profits and GHG savings from bioethanol production, by identifying best variety choice including possible use of triticale in second cereal positions.

3.1.2. Specific objectives

- 1. To determine alcohol yields of current Group 3 and 4 hard wheat varieties
- 2. To evaluate the grain yield, alcohol yield and N requirements of triticale in relation to wheat

3.1.3. Background

With the opening of the Ensus bioethanol plant on Teeside in spring 2010 and the Vivergo plant on Humberside scheduled to open in 2011, growing grain for bioethanol is now a reality, with up to 2 million tonnes of grain expected to be used from the 2011 harvest. Both variety and agronomy can affect the value of grain to the processor, mainly through the alcohol yield achieved per tonne of grain (Smith *et al.*, 2006). In addition, through Carbon Reporting in the Renewable Energy Directive the Greenhouse Gas (GHG) savings of the biofuel are also important to the processor. Given that over 67% of the GHG costs of bioethanol are due to crop production (Biofuels GHG Calculator, HGCA), reducing these GHG costs will ultimately be important, and will be part of specifications to the grower.

The keys to maximising the GHG savings associated with bioethanol production include maximising grain yields, and minimising Nitrogen (N) inputs (Kindred *et al.*, 2008a). Nitrogen fertilisation constitutes over 70% of the GHG and 20% of the economic cost of production of wheat.

In a first wheat position, to maximise both profitability and GHG savings, variety choice will be driven primarily by a high grain yield (as reported in the HGCA Recommended Lists), but processing performance and alcohol yield will still be

important to the industry. Consideration of the utility of Group 3 and 4 wheats for bioethanol is given below (3.1.4).

In a second wheat position the situation is different because yield tends to be lower due to build-up of diseases such as take-all, because N applications are generally higher, and because grain protein is generally higher (hence reducing alcohol yields). All of these factors (especially low yield and higher N inputs) reduce the profitability of the crop and the potential GHG savings of the biofuel. It is possible that alternative cereals may have similar or higher yields than wheat in the second or later rotational positions, and have a lower N requirement. Of the likely candidates, barley and oats are unlikely to be of use due to their husked grain and hence relatively low starch content, and rye has low yields. This leaves triticale (a hybrid of rye and wheat), which is considered further below (3.1.5).

3.1.4. Benefits of Group 3 and 4 wheats as biofuel feedstocks in the first cereal position

Varieties in HGCA Recommended List trials are tested for AY by SWRI, and there are some indicators of the best varietal types suitable for distilling (see HGCA Information Sheet 11, 2010). However these tests are currently restricted to soft wheats, with the existing distilling industry experiencing problems in processing hard wheats.

In contrast the new bioethanol plants inevitably take in a range of wheat varieties including hard endosperm types. However, little is known about the performance of hard wheat varieties for alcohol production in the UK, other than an assessment via lab screening of 30 feed wheat samples carried out by Davis-Knight *et al.* (2010). Until the biofuel industry has had some months experience of utilising current varieties, and in the absence of an industry mechanism for testing the performance of new varieties (in the way that the potable alcohol industry does), there exists a substantial gap in our knowledge.

Initial work in the GREEN Grain LINK project (Sylvester-Bradley *et al.*, 2010) using a comparison of one hard (Option) and one soft wheat (Riband) showed that the AY of the hard wheat responded in a similar way to applied N, as did a soft wheat (Kindred *et al.*, 2008b). Clearly some hard wheat varieties may well be advantageous for bioethanol production, particularly those with high yield potential and lower grain

proteins than milling wheat varieties (e.g. Oakley). However, further work is needed to determine the AY and processing characteristics of the Group 3 and 4 hard wheats to guide variety choice in the developing bioethanol industry.

3.1.5. Advantages of triticale

Carbon reporting for the RED now applies at farm level and a GHG incentivisation scheme is likely. Our early work demonstrated that the ideal wheat variety for bioethanol is high-yielding with low protein content and low N fertiliser requirements. We know from GREEN grain trials that wheat varieties with big reductions in N requirements are not currently available. However, an analysis by Sylvester-Bradley and Kindred (2009) showed triticale to have the highest nitrogen use efficiency (NUE) of cereal species in the UK. Other investigations suggested that triticale could deliver comparable yields at a lower N input, and moreover that this species was being used in Sweden for bioethanol production. In the UK, triticale is a cereal which is assumed to require 30% less N than wheat (RB209, 8th edition; Anon 2010), and therefore presents significant opportunities for reductions in growing costs and GHG emissions. Following these initial observations, laboratory work funded by HGCA confirmed that triticale samples harvested from Descriptive List trials gave comparable alcohol yields to a control wheat variety, and showed similar grain characteristics in terms of size, shape and protein content as wheat (Davis-Knight and Weightman, 2008).

Savings in GHG emissions reported for fuel alcohol production vary widely, dependent on assumptions made for yields and levels of inputs used in the assessments. By modelling the use of triticale for bioethanol production with the HGCA Carbon Calculator, and entering typical yields and input scenarios, Weightman and Davis-Knight (2008) quantified the typical GHG savings, and demonstrated a potential benefit of triticale over wheat, particularly in second and third wheat situations. However, there has been no good information on actual performance of triticale in the UK, in fair comparisons with wheat (e.g. on the same soil types, with similar crop protection and nutrition regimes).

Agronomy trials carried out *ca*. 5-10 years ago indicated that modern triticale varieties could yield up to 9 t/ha as a first cereal (unpublished data, ADAS Rosemaund) and 8 t/ha as a second or third cereal (Overthrow and Carver, 2003; HGCA project report 306) when grown on sites with high yield potential, and managed

according to best agronomic practice for wheat (e.g. good disease and lodging control).

The historic N-response data on which the fertiliser recommendations were based (e.g. Aquilina, 1987) are no longer available. Moreover, these studies were performed on old varieties originally released in the 1980s (published information reviewed by Davis-Knight and Weightman, 2008), often on low yield potential sites or on light soils (e.g. Cleal, 1993) triticale being seen as a 'low input' crop (Gutteridge *et al.*, 1993). Moreover, where comparisons of wheat and triticale have been made at a single N level, it is never clear from the results whether the wheat has been under-fertilized, or the triticale over-fertilized (e.g. Overthrow and Carver; HGCA project report 306).

Hence it was clear that some proof of concept was required to compare the relative yield responses of wheat and triticale to N, in high yield potential situations. As an initial 'look-see' triticale was included in a wheat variety experiment carried out at ADAS Terrington in 2007 in a first cereal position at nil-N and with 140 kg/ha (50 kg/ha less N than recommended for the wheat); the results indicated a significantly higher yield for triticale than any of the wheat varieties at either N level (Table 1).

Table 1. Grain yields (t/ha @85% DM) for wheat (Robigus) and triticale (Benetto) grown at ADAS Terrington in 2007 in a first cereal position.

	Yield (t/ha)		
N rate (kg/ha):	0	140	
Variety (species):			
Robigus (wheat)	6.55	9.09	
Benetto (triticale)	7.13	9.68	

Given the potential displayed by triticale in 2007, a dedicated trial funded by breeders was subsequently designed to compare wheat and triticale in a second cereal position on good wheat land, where the greater take-all resistance of triticale would be expected to enhance any yield advantage (2009 harvest season). Additionally, the design incorporated an N response trial, with identical N rates for each variety, to allow curve fitting and determination of economic optima. Grain yields are shown in Figure 1 (work supported by Senova and Syngenta Seeds).

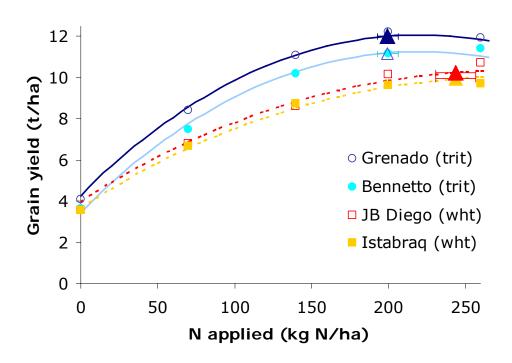


Figure 1. Grain yields (t/ha @85% DM) for two varieties of wheat (JB Diego, Istabraq) and triticale (Grenado, Benetto) grown on a clay loam soil in Suffolk in 2009, in a second cereal position (triangles represent economic N optima).

Based on the fitted optima, triticale yielded approximately 10% more than wheat, with *ca*. 20% lower N optima. Even with a £15/t discount applied to the value of triticale grain (relative to wheat grain), triticale still showed a greater financial margin over N inputs than did wheat (Kindred *et al.*, 2010a,b).

Clearly, these are very important results, both in the context of improving profitability of cereal production, but also in terms of reducing GHG emissions, assuming that modern triticale varieties could find a ready market in the bioethanol industry today.

A single N-response experiment is clearly insufficient to deliver a conclusive message to the industry, so further evaluation was needed to validate the potential of triticale to wheat in the second or third cereal position. This project therefore aimed to repeat the study described above at two sites in 2009/10. The results of an additional breeder-funded study were included, to bring the number of N response data sets from 2010 harvest to three.

3.1.6. Summary

To support UK growers supplying the bioethanol industry, there is a need to: (i) provide information on the suitability of hard Group 3 and 4 wheats for alcohol production, and (ii) investigate the potential for improving profitability in the second cereal position, and maximising biofuel yields and GHG savings by growing triticale as a feedstock for alcohol production.

3.2. Materials and methods

3.2.1. Hard wheat samples for alcohol yield testing

Fifty-six wheat grain samples were provided by Crop Evaluation Ltd from the 2009 Recommended List trial series. The ten varieties selected were Glasgow, Warrior, Oakley, Duxford, JB Diego, Ketchum, Panorama, Grafton, Conqueror and KWS Sterling, each represented across six sites (with the exception of Oakley, Duxford, JB Diego and Panorama from the Lincolnshire site which were not tested). The site details and average yields and protein levels are shown in Table 2.

Table 2. Site details and average grain yield and protein at each of six RecommendedList sites in 2009 used to provide wheat samples for alcohol yield testing.

Site code	9KW120T	9NA114T	9ES104T	9AN112T	9AN108T	9HH110T
Site	Framling.	Wolfertn	Humbie	Welbourn	Croft	Rudston
Location	Suffolk	Norfolk	E. Lothian	Lincs	N. Yorks	E. Yorks
Previous crop	W-rape	F-peas	W-rape	W-rape	W-bean	W-rape
Soil	D-clay	D-silt	Medium	Shallow	Medium	Shallow
G. yield (t/ha)*	12.96	12.99	10.80	13.01	11.45	9.79
Protein (%DM)	10.68	11.74	9.54	11.30	10.51	7.67

*Grain yield at 85% DM

3.2.2. Alcohol yield determination

Alcohol yield (AY) and residue viscosity (RV) were determined in duplicate using an ADAS method adapted from that of the Scotch Whisky Research Institute (SWRI; Agu *et al.*, 2006). Wheat grain was milled using a Glen Creston hammer mill fitted with a 2mm screen, and the moisture content of the flour determined on a subsample by

drying overnight at 100°C. Wholemeal flour (15 g fresh weight basis) was placed in a stainless steel beaker with 40.5 mL of water and 250 µL of a thermostable alphaamylase (added in excess) to rapidly break down starch to oligosaccharides (Termamyl 120L, Novozyme). The slurry was then heated in a waterbath to 85°C with frequent stirring, before being autoclaved at 126°C for 11 min. The sample was returned to the waterbath and further 250 μ L of the amylase was added when the slurry returned to 85°C, to minimise retrogradation. The cooked slurry was then reduced in temperature and mashed at 65°C for an hour with inclusion of barley malt that contains a relatively high a and β amylase content and also supplies modified starch and free amino nitrogen to the yeast (20% malt to 80% wheat on a dry weight basis). The slurry was pitched with distillers yeast (0.4% w/w) and fermented at 30°C for 68 hours before being distilled and the distillate measured for alcohol content using an Anton Paar density meter. The residue after distillation was adjusted to 125 mL with water before being centrifuged and the supernatant filtered twice through GF/A filter papers. Viscosity of the supernatant was determined at 20 oC using a Utube viscometer (PSL-BS/U B, Poulten Selfe and Lee, Essex, UK).

Glasgow (a soft wheat, consistently rated as a good quality distilling wheat) and Warrior (no distilling rating) were used as examples of high and low AY wheats respectively, sourced from three different RL sites (9ES104T, 9AN108T and 9HH110T). Duplicate grain samples of each were screened in parallel at the Scotch Whisky Research Institute (SWRI) and at ADAS, to confirm that the AY testing methodology was comparable between labs (Table 3). It should be noted that the SDs represent variation between sites rather than analytical variation. All further AY testing on the hard wheat samples were carried out by ADAS.

Alcohol yield (L/t DM)				
Lab	SWRI	ADAS		
Variety	SWI			
Glasgow	460	459		
	(4.8)	(7.5)		
Warrior	445	432		
	(7.1)	(1.0)		

Table 3. Average alcohol yield (SD, n=3) for contrasting wheat varieties tested at two laboratories (grain samples from 2009 RL sites).

3.2.3. Field Experiments to compare performance of wheat and triticale

Wheat-triticale N response experiments were carried out in the 2009/10 field season in the second cereal position at two sites. The first was at Towthorpe (TO), Near Malton, East Yorkshire and the second at Terrington St Clement (TT), King's Lynn, Norfolk. At each site, two winter wheat (JB Diego and Viscount) and two triticale varieties (Benetto and Grenado) were each tested at five nitrogen (N) rates. At TO, where the optimum N rate was predicted to be the higher of the two sites, N treatments were 0, 90, 180, 270 and 360 kg N/ha, and at TT they were 0, 80, 150, 230, 310 kg N/ha. At both sites, N was applied as per the timings recommended in the Fertiliser Manual (Anon., 2010) and each treatment combination was replicated three times.

A third wheat-triticale experiment was studied in the 2009/10 growing season within a larger species trial comparing wheat, triticale, barley, oats and rye: The experiment was located at Cransford in Suffolk (SF; grid reference TM 328 645) on a clay loam soil (Ragdale series) following winter wheat. The experiment design used a split-split-plot with species (5 treatments, wheat and triticale shared a main plot but triticale plots were separated from wheat plots by discards) as the main plot, N (6 treatments) as a split plot and variety within a species (8 varieties for wheat, 3 for barley, triticale and oats, 1 for rye) as a split-split plot giving 102 treatments replicated 3 times (306 plots in total). Nil-N guard plots separated species and N treatments. Species main plots were arranged to allow separate management and harvesting if necessary. The trial was surrounded by winter barley to allow access for harvesting barley plots. Plots were 2 m wide x 12 m long and drilled at 350 seeds m⁻² using an Øjyord plot drill.

Nitrogen application rates were determined following measurement of soil mineral nitrogen (SMN) in autumn, which was 50 kgN/ha. This gave a soil nitrogen supply (SNS) of index 1, for which the N rate recommended by RB209 for wheat on a clay soil was 220 kgN/ha. Rates used were 0, 70, 140, 220, 290 and 360 kgN/ha. The same N rates were used for all species. Nitrogen was applied by hand as ammonium nitrate prills. Where possible common herbicide, fungicide and PGR applications were made across all species to achieve effective weed, disease and lodging control.

Agronomic management at the three sites was robust to avoid pest, disease and weed control problems and was the same for triticale as for wheat to avoid confounding effects. At TT and SF, for each plot, the proportion of plot area that was leaning (0-45° from vertical) or lodging (45 - 90° from vertical) was recorded at harvest. Both wheat-triticale trials were harvested in August 2010, the N x species trial harvested in early September 2010 and yields (t/ha @ 85% dry matter) and specific weights were determined. Grain samples were sent for N content determination using both near-infrared reflectance (NIR) and oxidative combustion (using a LECO instrument) methods.

3.2.4. Grain protein analyses

Grain protein was determined by Near Infra Red (NIR) reflectance spectroscopy using an Infratec instrument (FOSS UK Ltd) with appropriate calibrations for wheat and triticale. In addition, for the TT and TO sites, grain were also analysed by the Dumas combustion reference method, and protein estimated as Nx5.7. It was therefore possible to compare the robustness of NIR for predicting grain protein by NIR.

3.2.5. Data analysis

All data were analysed by using Genstat v. 12 (VSN International Ltd.).

Alcohol yield of hard wheats

Alcohol yield (L/t DM) from the RL 2009 samples were combined with RL yields (source CEL, RL Plus) adjusted to 100% DM to give an alcohol yield per unit area (L/ha).

Alcohol yields (L/t DM and L/ha) from the RL2009 samples were subjected to REML analysis, to account for the 4 missing values at the Lincolnshire site. This analysis gave predicted alcohol yield and residue viscosity means for each variety over all sites and significance as determined by Wald tests.

ADAS Wheat vs triticale experiments

For the 2010 TT and TO sites, alcohol yields (AY) were not determined directly on samples using the SWRI laboratory method, but were estimated from a predictive equation as follows:

AY (L/t, DM basis) = $-7.31 \times \text{protein} + 519$ as reported by Davis-Knight and Weightman (2008).

Yield, specific weight, protein and AY data from HM were analysed by ANOVA. Due to technical difficulties at TT, the wrong N amounts were applied to some plots. This meant that the data had to be analysed using an unbalanced ANOVA, giving predicted, rather than arithmetic, treatment means.

The response of yield to N was estimated for each experiment using the linear plus exponential function (LEXP; George, 1984).

$$y = a + b.r^{N} + c.N$$

where y is yield in t/ha at 85%DM, N is total fertiliser N applied in kg/ha, and a, b, c and r are parameters determined by statistical fitting.

Optimum N rates (Nopt) were then derived from the fitted LEXP parameters using: Nopt = [ln(k-c)-ln(b(ln(r)))]/ln(r)

where k is the breakeven price ratio between fertiliser N (\pounds/kg) and grain ($\pounds/tonne$). The breakeven ratios used in this study was 5.

A grain N (%) response curve was then fitted to the data from each experiment. The data were fitted with a Normal Type curve with Depletion. The function for the normal with depletion curve is:

$$N\% = d + c.\exp(-\exp(-a.(N - b)))$$

where *a*, *b*, *c* and *d* are parameters determined by fitting, and N is applied N (kg/ha).

Grain N% estimates were then derived at each Nopt estimate.

3.3. Results

3.3.1. Alcohol yield of hard wheats from RL 2009

The wheat samples selected were all feed types, with relatively low grain protein contents. There were significant variety differences (P<0.001) in protein content with Conqueror, Glasgow, Duxford and Oakley having lower protein contents than Grafton, Ketchum, Panorama and Warrior (Figure 2). Measured AY for the hard wheats, compared to the reference varieties of soft wheat; Glasgow (high AY) and Warrior (low AY) averaged across sites are shown in Figure 3a. Variety had a significant effect on AY (P<0.001) whereby Glasgow was confirmed as the superior wheat, having significantly higher AY than the other varieties. However Conqueror and Oakley also had significantly higher AY than the worst varieties Ketchum and Warrior (based on the LSD, 95%).

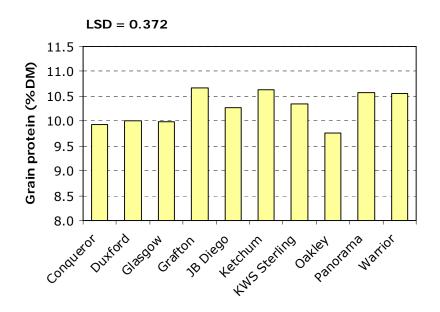
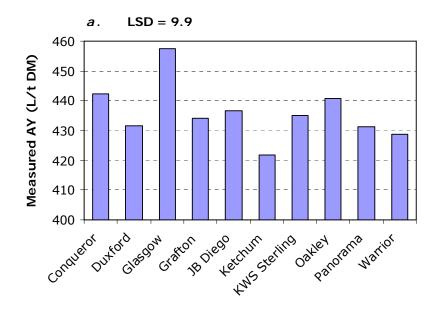


Figure 2. Grain protein content of 10 wheat varieties from the 2009 Recommended List (data supplied by CEL).

Since AY is predominantly a reflection of the starch content of each sample (and inversely related to its protein content), the AY were adjusted to a standard protein content in order to look for non-protein related genetic effects. Protein was standardised at 11.5% DM by reducing or increasing the AY by 6.65 L for each % protein above or below 11.5% protein respectively. These adjusted AY (adjAY) standardised to 11.5% protein, were then subjected to ANOVA in the same way as the

measured AY (Figure 3b). Again variety had a highly significant effect on adjAY (P<0.001), but now only Glasgow was shown to be significantly different to the rest based on the LSD. The remaining hard wheats were very similar in AY at a fixed protein content.



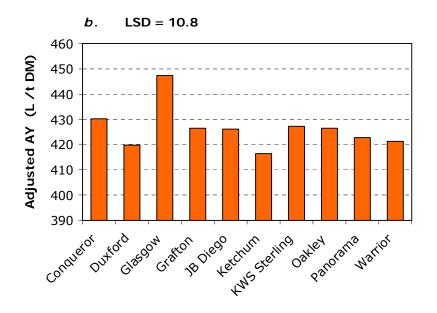


Figure 3. Alcohol yield of 10 wheat varieties from the 2009 RL either a) as measured in the laboratory or b) adjusted to 11.5% protein.

Residue viscosity was significantly influenced by variety (P>0.001), with Warrior having the highest residue viscosity, significantly higher than all other varieties (Figure 4). However no other variety was significantly different to the others based on the LSD (95%).

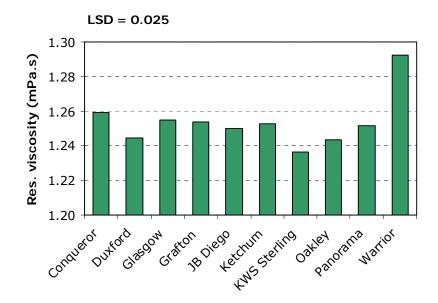


Figure 4. Residue viscosity of 10 wheat varieties from the 2009 RL following determination of AY.

3.3.2. Wheat and triticale 2010: Grain yield, specific weight and optimum N rate

At TO, grain yields significantly differed with species grown (P = 0.003) and N rate (P<0.001; Table 4). When averaged over all N rates, triticale gave 0.42 t/ha (@ 85% DM) higher yields than wheat. When varieties were compared, both triticale varieties gave higher yields than both wheat varieties (Table 5). The highest-yielding triticale variety tested was Benetto and the highest-yielding wheat variety tested was JB Diego (Table 5). There was no significant interaction between species and N rate with yields of both species significantly (P<0.001) increasing at each N level up to 270 kg N/ha (Table 4).

Species	N rate	Yield	Specific
	(kg N/ha)	(t/ha @ 85% DM)	weight
			(kg/hL)
Triticale	(Averaged across	7.14	70.1
Wheat	N treatments)	6.72	71.9
(Averaged across	0	1.87	67.4
species and	90	6.25	69.1
varieties)	180	8.00	72.3
	270	9.33	73.4
	360	9.43	73.0
Triticale	0	2.31	68.9
	90	6.37	68.1
	180	8.13	70.8
	270	9.36	70.6
	360	9.54	72.0
Wheat	0	1.42	65.9
	90	6.13	70.1
	180	7.87	73.7
	270	9.29	76.7
	360	9.32	74.0
Species	P-value	0.003	0.139
	SED	0.112	1.30
N rate	P-value	<0.001	0.015
	SED	0.1771	2.05
Sp x N	P-value	0.204	0.318
	SED	0.2505	2.90

Table 4. The effect N applied on the grain yield and specific weight of triticale and wheat grown at Towthorpe in 2010.

Species	Variety	Yield (t/ha @	Specific
		85% DM)	weight
			(kg/hl)
Triticale	Benetto	7.25	68.7
	Grenado	7.03	71.5
Wheat	JB Diego	6.85	72.2
	Viscount	6.58	71.6
Variety	P-value	0.022	0.25
	SED	0.1662	1.86

Table 5. The effect of variety of triticale or wheat grown at Towthorpe in 2010, on yield and specific weight.

When N response curves were fitted to the yields, there was statistical justification for fitting a different curve for each species (Figure 5a). Since only the intercepts of the curves significantly differed, the species shared the same optimum N rate (Nopt; 298 kg N/ha). The fitted yields at the optimum N rate were: 9.45 t/ha for the triticale and 9.10 t/ha for the wheat (Figure 5a).

At TT, as at TO, the triticale gave significantly (P<0.001) higher yields than the wheat (Table 6), but here the overall difference was larger than at Towthorpe (0.92 t/ha). At TT there was again no interaction between the species and N rate (Table 6), and when N response curves were fitted, again only the intercept of the curves significantly differed (Figure 5b). At Terrington, the optimum N rate of both the wheat and triticale was 243 kg N/ha, and the fitted yield at the optimum N rate was 7.57 t/ha for the wheat and 8.50 t/ha for the triticale.

The area of each plot that was either leaning or lodging was significantly (P<0.001) lower for wheat than triticale (Table 6) when measured at TT. Of the triticale varieties, Benetto appeared to suffer more from leaning than Grenado (data not shown). There were significant interactions between the species grown and N rate in both leaning (P= 0.014) and lodging (P= 0.007), whereby they increase by more at the higher N rates in the triticale than the wheat (Table 6).

At both sites, wheat gave, on average, a higher Spwt than triticale (Table 4, 6), but the effect was only significant (P < 0.001) at TT (wheat was 4.4 kg/hL greater than

triticale; Table 6). Conversely, higher N rates led to significantly (P = 0.015) higher Spwt at TO (Table 4) but not at TT (Table 6).

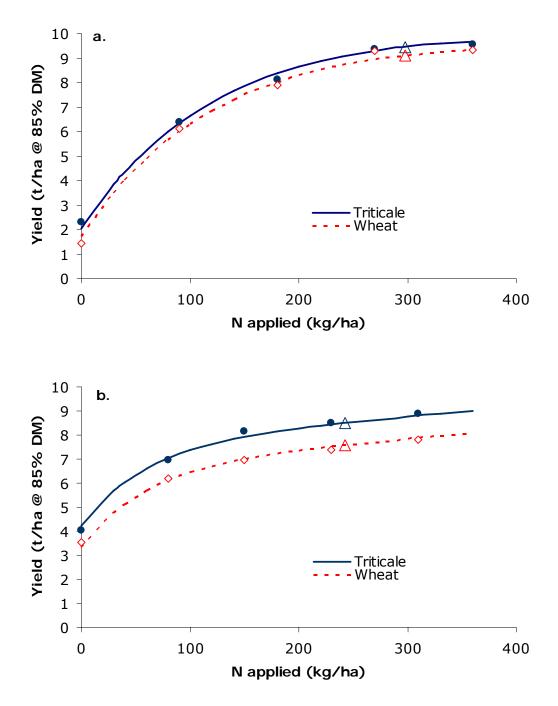


Figure 5. Effect of N on yield of triticale and wheat (data points and fitted curves), including yields at optimum N rates (triangles) at a) Towthorpe, and b) Terrington in 2010.

Species	N rate	Yield	Leaning	Lodging	Specific
		(t/ha @	(% of	(% of	weight
		85% DM)	plot)	plot)	(kg/hL)
Triticale	(Avge across	7.33	30.59	7.06	67.2
Wheat	N treats)	6.41	4.77	0.25	71.6
(Averaged across	0	3.81	-0.02	-0.01	68.2
species and	80	6.58	9.12	0.75	68.3
Varieties)	150	7.56	28.62	8.85	70.3
	230	7.95	18.09	0.03	70.9
	310	8.35	34.03	9.50	69.2
Triticale	0	4.05	-0.07	0.00	68.3
	80	6.95	17.88	1.39	65.2
	150	8.14	52.19	17.39	68.1
	230	8.51	31.30	0.06	68.7
	310	8.88	52.71	17.58	65.9
Wheat	0	3.56	0.03	-0.02	68.0
	80	6.20	0.06	0.09	71.6
	150	6.95	4.20	0.01	72.7
	230	7.38	4.40	-0.01	73.3
	310	7.80	14.69	1.14	72.7
Species	P-value	<0.001	<0.001	<0.001	<0.001
	SED*	0.1147	4.389	1.968	0.80
N rate	P-value	<0.001	<0.001	<0.001	0.084
	SED*	0.1818	6.956	3.119	1.27
Sp x N	P-value	0.26	0.014	0.007	0.059
	SED*	0.2585	9.888	4.433	1.81

Table 6. The effect N applied on the grain yield, leaning, lodging and specific weight of triticale and wheat grown at Terrington in 2010 (means displayed are predicted means from unbalanced ANOVA).

* SED is average SED from unbalanced ANOVA analysis

3.3.3. Wheat and triticale 2010: Protein concentration and alcohol yield

At the TO and TT sites, the differences in protein concentration between the two species, based on the reference method for N determination (Dumas combustion; LECO N x 5.7) showed different trends: At TO, on average, there was no significant difference in protein concentration between triticale and wheat (Table 7), and this was also the case when the individual varieties were examined (Table 8). In contrast, at TT, wheat gave significantly (P<0.001) higher protein concentrations when averaged over all N rates (10.86 %) than triticale (9.55 %; Table 9). This meant that since there is a linear negative relationship between protein concentration and alcohol yield, the triticale is predicted to give significantly (P<0.001) higher alcohol yields than wheat (11.05 L/t higher; Table 9).

At both sites, there were significant (P<0.001) effects of the amount of N applied on protein concentration (Table 7, 9). The protein concentration response to N applied was fitted using a linear with depletion curve, and the protein concentration at Nopt was determined. This showed that, at TO the fitted protein concentration at Nopt was 11.97 % for triticale and 11.31 % for wheat, equivalent to 432 and 436 L/t alcohol yield, respectively. At TT, the protein concentration at Nopt was 11.247 for wheat, equivalent to 438 and 428 L/t alcohol yield, respectively.

When protein was determined by NIR, results differed from those obtained using the reference method (LECO). The grain protein content of the TO samples were higher using NIR than LECO (Table 7), with the average protein concentration of the triticale measured as 0.78 % higher using NIR than LECO, and wheat 0.32 % higher. When these data were analysed, it showed that triticale had significantly (P<0.001) higher protein concentrations than wheat; a different conclusion to that based on the reference methodology (Table 7).

The different analysis methods also led to different conclusions at TT. Here, grain protein determined using NIR indicated no significant difference between the two species, whereas LECO method had shown that the average protein concentration of wheat was significantly higher than that of triticale (Table 8). At both sites, NIR appeared to over-estimate the protein concentrations for triticale at each N rate (Tables 7, 9).

Table 7. The effect N applied on the protein concentration (measured by NIR or oxidative combustion reference method (LECO) and predicted alcohol yield of triticale and wheat grown at Towthorpe in 2010.

Species	N rate	Protein	Protein	Predicted
	(kg N/ha)	concentration	concentration	alcohol yield
		LECO (% DM)	NIR (% DM)	(L/t DM)*
Triticale	(Avge across	10.35	11.13	443
Wheat	N treats)	10.07	10.39	445
(Averaged across	0	8.51	8.98	457
species and	90	8.55	9.00	456
Varieties)	180	10.85	11.55	440
	270	11.47	12.17	435
	360	11.78	12.26	433
Triticale	0	8.24	8.72	459
	90	8.60	9.18	456
	180	11.03	12.03	438
	270	11.84	12.77	432
	360	12.06	12.93	431
Wheat	0	8.78	9.24	455
	90	8.51	8.82	457
	180	10.67	11.06	441
	270	11.02	11.44	438
	360	11.50	11.59	435
Species	P-value	0.17	<0.001	0.17
	SED	0.1783	0.151	1.303
N rate	P-value	<0.001	<0.001	<0.001
	SED	0.2818	0.2387	2.06
Sp x N	P-value	0.181	0.001	0.181
	SED	0.3986	0.3376	2.914

* Predicted alcohol yield estimated as AY = -7.31 x protein + 519, where protein is based on LECO N x 5.7

Table 8. The effect of variety of triticale or wheat grown at Towthorpe in 2010 on protein concentration (measured by NIR or oxidative combustion (LECO) and predicted alcohol yield (based on LECO-N).

Species	Variety	Protein LECO	Protein NIR (%	Predicted
		(% DM)	DM)	alcohol
				yield (L/t)
Triticale	Benetto	10.30	11.18	444
	Grenado	10.41	11.08	443
Wheat	JB Diego	10.10	10.49	445
	Viscount	10.03	10.29	446
Variety	P-value	0.359	<0.001	0.359
	SED	0.2161	0.182	1.579

3.3.4. Wheat and triticale 2010: Supplementary study, Suffolk species trial

A supplementary study is reported here for wheat and triticale grown in a species experiment also including barley, oats and rye (data reported elsewhere). This work was directly funded by breeders, but was monitored in part through the present project under an HGCA studentship, thereby giving added value. Due to the wet August of 2010 this trial was late harvested and whilst there was no substantial lodging pre-maturity before early August, there was significant post-maturity lodging by time of harvest in early September which was likely to have affected grain yields. There was a significant (P<0.01) difference in the yields among the varieties (Table 10). When averaged over all N rates, the triticale variety Tulus gave the highest yield (8.36 t/ha), followed by the wheat variety Beluga (8.17 t/ha). Unlike the trials at TO and TT reported above, there was no overall difference in yield between the wheat and the triticale varieties. In the Suffolk trial there was a significant (P = 0.03) interaction between the varieties and N rates, whereby the yields of some of the varieties (e.g. Tulus) continued to increase at high N rates whereas those of other varieties (e.g. Benetto) gave lower yields at the highest N rate than at 290 kg N/ha (Table 10). This result appeared to be associated with the differences in lodging of the varieties; Tulus, the highest yielding variety, suffered least from lodging, although generally, there was more lodging in the triticale than the wheat varieties (Table 11). Data from the other 7 wheat varieties/treatments in the trial have been averaged to compare with one wheat variety, Beluga, and the four triticale varieties.

Species	N rate	Protein	Protein	Predicted
		concentration	concentration	alcohol yield
		LECO (% DM)	NIR (% DM)	(L/t) [†]
Triticale	Avge across	9.55	10.63	449
Wheat	N treats	10.86	10.54	440
(Averaged across	0	7.96	8.36	461
species and	80	8.72	9.21	455
Varieties)	150	10.33	10.85	444
	230	11.48	11.96	435
	310	12.33	12.69	429
Triticale	0	7.49	8.38	464
	80	7.99	8.90	461
	150	9.84	10.93	447
	230	10.67	12.13	441
	310	11.71	12.96	433
Wheat	0	8.47	8.33	457
	80	9.51	9.55	450
	150	10.86	10.77	440
	230	12.35	11.77	429
	310	13.00	12.39	424
Species	P-value	<0.001	0.662	<0.001
	SED*	0.1642	0.1476	1.2
N rate	P-value	<0.001	<0.001	<0.001
	SED*	0.2595	0.234	1.9
Sp x N	P-value	0.609	0.133	0.609
	SED*	0.3712	0.3344	2.7

Table 9. The effect N applied on the protein concentration measured by NIR or oxidative combustion (LECO) and predicted alcohol yield (based on LECO-N) of triticale and wheat grown at Terrington.

* SED is average SED from unbalanced ANOVA analysis

⁺Predicted alcohol yield estimated as AY = -7.31 x protein + 519, where protein is based on LECO N x 5.7

Species	Variety	N applied (kg N/ha)				Mean	Opti		
									ma
		0	70	140	220	290	360		
Wheat	Beluga	4.04	7.21	8.87	9.41	9.79	9.72	8.17	221
	Variety								
	average	3.70	7.06	8.82	9.27	9.41	9.58	7.97	209
Triticale	Bellac	3.66	6.52	8.22	9.05	9.11	9.27	7.64	233
	Benetto	4.26	8.09	8.92	9.19	9.19	8.79	8.07	165
	Grenado	3.90	7.62	8.76	9.25	9.08	8.79	7.90	173
	Tulus	3.99	7.32	9.27	9.77	9.72	10.1	8.36	216
Mean		3.93	7.30	8.81	9.32	9.38	9.37	8.02	
Variety	P-value	<0.00	1						
	SED	0.1375	5						
N Rate	P-value	<0.00	1						
	SED	0.1375	5						
Var x N	P-value	0.03							
	SED	0.3368	3						

Table 10. Effects of applied N fertiliser on grain yields (t/ha @ 85% DM) of wheat and triticale varieties grown in Suffolk in 2010.

The interaction between variety and N rate showed through in N response curves fitted to the data, with statistical justification for using separate curves for each variety, with a common R parameter, giving different N optima for each variety (Figure 6). Benetto and Grenado gave very similar yield responses to N, and gave the lowest N optima. The other triticale varieties Bellac and Tulus however gave N optima similar to the Beluga and the other wheat varieties in the trial.

In the SF trial, the grain protein concentrations of the wheat varieties (determined by NIR) were generally lower, and the predicted AY generally higher than in the triticale (Table 11). This is consistent with the results from the TO trial.

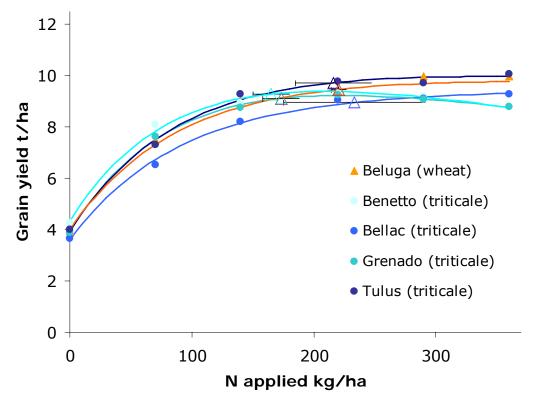


Figure 6. Effect of N on yield (@85%DM) of triticale or wheat varieties (fitted curves) grown in Suffolk in 2010, including optimum N rates (triangles).

Table 11. Effects of wheat and triticale varieties grown in Suffolk in 2010 on grain
protein concentration by NIR, predicted alcohol yield (PAY), specific weight and
leaning and lodging averaged across N rates.

Species	Variety	Protein	Predicted	Spwt	Leaning	Lodging
		conc. NIR	AY (L/t)	(kg/ hL)	(% of	(% of
		(% DM)			plot)	plot)
Wheat	Beluga	10.42	443	68.3	1.94	4.44
	Wheat	10.64	441	70.3	2.88	6.98
	var avge					
Triticale	Bellac	11.86	432	61.9	16.11	30.28
	Benetto	11.20	437	66.7	35.83	20.83
	Grenado	10.86	440	66.3	7.22	39.44
	Tulus	11.32	436	64.2	1.39	0.00
Variety	P-value	<0.001	<0.001	<0.001	< 0.001	<0.001
	SED	0.19	1.409	0.69	7.93	8.02

3.4. Discussion

3.4.1. Alcohol yields of hard wheats

Using the laboratory method for AY determination, the results show that there were significant differences in performance between hard wheat varieties, with Conqueror and Oakley having the highest AY, and Ketchum the lowest AY. The results confirmed that the soft wheat Glasgow still outperforms the hard wheat varieties in terms of its AY per tonne of grain.

Residue viscosity is another important measure of processing quality as far as distillers are concerned. The hard wheat varieties behaved similarly, with no variety showing the high viscosity typical of a poor distilling quality wheat, such as Warrior. As the test method for AY is based on the Scotch Whisky method, it is too early to say whether the method is wholly appropriate for the (fuel) bioethanol industry, but the data suggest that no particular hard wheat variety would give cause for concern at this stage. This supports earlier observations with a limited number of hard wheats (Kindred *et al.*, 2008b; Davis-Knight *et al.*, 2010).

Conqueror and Oakley demonstrated relatively high AY, and this was principally due to lower protein contents in their grain, which reflects a response of those varieties to their environment as well as a genetic effect *per se*. When the AY were adjusted to a standard protein content, only the soft wheat Glasgow had significantly higher AY than the other varieties. It could be argued that adjusting to fixed protein content is a crude method of standardisation. However all previous work has shown that statistically, parallel lines can only be justifiably fitted to the relationships between AY and grain protein for different varieties (Smith *et al.*, 2006; Clarke *et al.*, 2008). It is therefore the best method we have at present to examine genetic differences in AY between wheat varieties. Nevertheless, low protein grain is still valuable for alcohol production, whatever the route of its production.

Alcohol productivity (AP) describes the output of alcohol per hectare, which is a critical determinant when quantifying the GHG savings associated with bioethanol production. This was estimated by taking the grain yields from RL Plus, and multiplying these by the AY determined in the laboratory as described above. The AP and corresponding AY data are shown in Figure 7. For the hard wheat data in the present study, only five

40

sites are included: the Lincs site was excluded because it was an incomplete data set, and its high site yield (Table 2) biased the AP assessments for certain varieties. While the distilling industry have no intention of moving to hard wheats, equivalent soft wheat data are shown along side (left hand figure 7) for reference.

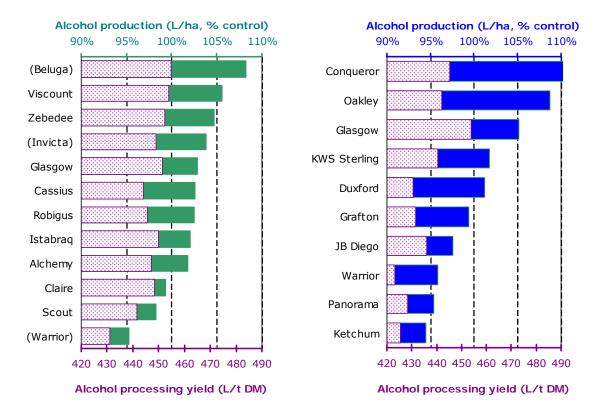


Figure 7. Alcohol production and alcohol processing yield (AP) of (left) soft wheats analysed by SWRI 2006-2009, and (right) hard wheats analysed in the present study from five sites in RL 2009. Reference AP = 4,097 L/ha for soft wheats and 4,407 L/ha for hard wheats. Soft wheat varieties in brackets represent limited number of observations.

It can be seen that the main determinant of AP as discussed elsewhere (Kindred *et al.*, 2008a) is grain yield. To exemplify this, the soft variety Glasgow, although it has a superior AY per tonne of grain, is outperformed in terms of grain yield and hence AP by Conqueror and Oakley. Regarding their potential for maximising GHG savings, these latter two varieties appear to be well suited for bioethanol production, by combining high AY with high grain yield (as also do Beluga and Viscount in the soft wheat class).

3.4.2. Agronomic performance of wheat and triticale and predicted alcohol yields

In both TO and TT experiments in this project, the triticale varieties significantly outyielded the wheat varieties. This is consistent with the experiment in 2009 (see Figure 1) in a 2nd cereal position, which showed that triticale gave 2 t/ha higher yields than wheat and required 50 kg/ha less N fertiliser (Kindred *et al.*, 2010). However at the SF trial in 2010, four triticale varieties were compared to a number of wheat varieties and showed no overall significant difference in yield between the two species.

Over two field seasons we have therefore shown that on high yield potential land in the 2nd cereal position, triticale has out-yielded wheat on three occasions and matched wheat in the fourth. In one season triticale had significantly lower Nopt, but in the second season it had the same Nopt as wheat at 2 sites (TT and TO), and an N optima that varied between varieties at the 3rd site (SF); two triticale varieties having lower optima than wheat varieties (albeit associated with greater lodging at higher N rates), and two others having N optima similar to wheat. The N optima in these experiments (SF, 199; TT, 243; TO, 298 kg/ha) were higher than those that would be recommended in the Fertiliser Manual (Anon., 2010) where the highest recommendation is currently 150 kg N/ha for triticale. However it should be noted that 2010 was not a severe take-all year, and so although grown in the 2nd cereal position, the true benefit of triticale in being able to overcome root damage may not have been seen in the field. Some measures of Take-all were taken on the SF trial and incidence was shown to be significantly lower in triticale than in the wheat varieties tested (8% vs 19% respectively). However no further resources were available for Take-all assessments. Further work over a wider range of seasons is required to quantify the true take-all resistance of modern triticale varieties, and to distinguish this from traits such as a faster rate of root expansion, which could enables the crop to overcome pathogen attack and give an increased an ability to capture N.

Better nitrogen use efficiency (NUE) is also an important trait in triticale: In 2009, better grain and straw yields (i.e. greater total biomass) than wheat were observed, with less N. The resources were not available in 2010 to examine N partitioning and total biomass in all these crops, but work under an HGCA studentship demonstrated that the triticale in the SF trial had greater straw biomass (particularly for the variety Benetto). Nevertheless with the TO and TT crops, it is clear that triticale produced

42

more grain with the same amount of N applied, i.e. better NUE. However the basis for this better performance remains unknown. Further physiological studies are required to assess rooting characteristics in particular. Interestingly, the biggest yield advantage of triticale is not always expressed at zero fertiliser, so it does not seem that the better performance of triticale can be mainly ascribed to the recovery of soil N (in contrast with a crop like oats, which has been the highest yielding species without N in both years of the SF trials). The better performance of triticale seems to be due to a combination of greater recovery of soil N (ie higher nil-N yields) and greater recovery/ utilisation of fertiliser nitrogen than wheat (Kindred *et al.*, 2010a). Analysis of dry matter and nitrogen harvest indices is required on a wider set of trials to understand the better NUE of triticale. It should be noted also that 2010 was a dry year overall, and a greater advantage of triticale might have been observed if the experiments had been carried out on light land.

One disadvantage in triticale in the 2010 experiments at two sites was lodging at the highest N rates, although the variety Tulus at the SF site was fairly resistant to lodging and gave the highest yield of both species. The results suggest that if lodging could be better controlled in more of the triticale varieties, yields could be even higher. This warrants further work, both on PGRs, and in understanding inherent lodging risk and how triticale relates to wheat in terms of root plate spread, stem strength etc.

The ADAS work to date has focussed on triticale in the 2nd cereal position as it is believed this is where the main advantage of triticale will lie, by making better use of its inherent take-all resistance and nitrogen use efficiency when roots are compromised. In the 1st cereal position, we may not expect triticale to outperform wheat, which has had more sustained breeding effort in the UK. Despite this, a 0.5 t/ha yield advantage of triticale was seen in first wheat experiment in 2007 compared to 40 wheat varieties (Kindred *et al.*, 2010b). Also in 2010, Agrovista carried out a trial at Eryholme, Nr Darlington, where wheat and triticale followed oilseed rape, and 220 kg N/ha were applied (in two doses of 110 kg N/ha each). A standard wheat fungicide programme was applied and no lodging was recorded. The two triticale varieties out-yielded the wheat average by 1.83 t/ha, and out-yielded the top yielding wheat (Robigus) by 0.47 t/ha (Table 12).

43

Species	Variety	Grain yield			
-	_	(t/ha @ 85%DM)			
Triticale	Bennetto	11.27			
	Grenado	11.54			
Wheat	Robigus	10.94			
	Grafton	9.89			
	Invicta	9.77			
	Warrior	9.16			
	JB Diego	9.08			
	Scout	9.09			
	CPBT 160	9.54			
	Glasgow	9.11			
	LSD	0.32			

Table 12. Yield of wheat and triticale varieties grown at Eryholme in 2010 in the first cereal position (date supplied by Agrovista).

This points to a substantial opportunity from the use of triticale to displace wheat for animal feed as well as bioethanol use; in five of the six trials we know of that have compared wheat with triticale over the past four years, triticale has significantly and substantially out-yielded wheat, whether in first or second cereal positions. At the other site, triticale did not out-yield wheat due to post-maturity lodging at the higher N rates.

3.4.3. Observations on grain quality

One point which is often levelled against triticale is its lower specific weight (Spwt) compared to wheat. In RL booklets, it is generally not possible to make fair comparisons between the two species, because the wheats are generally grown in 1st place in the rotation, with the triticale in a later cereal position. In the 2009 study (Section 2) triticale had lower Spwt than wheat (71.5 vs 76.8 kg/hL). In the present study, at the TO site there was no significant difference between the two species (70.1 vs 71.9 kg/hL) whereas at the other two sites triticale had lower Spwt then wheat (TT, 67.2 vs 71.6 kg/hl; SF, 65.5 vs 70.2 kg/hL), all grown in the 2nd cereal position.

It is still not entirely clear whether triticale has on average lower grain protein than wheat, reflecting its typically lower N inputs, or has higher grain protein, as some of the earlier reports suggested (e.g. Naylor, 1987). From the experiments in 2010, there was inconsistency in the predicted protein concentration in triticale relative to wheat, dependent on the analytical method chosen and the site: at the TO site, the reference method (Leco N) suggested no significant difference between the two species, but NIR suggested that triticale had significantly higher protein than wheat; at TT, Leco N showed that triticale had a significantly lower grain protein, whereas NIR suggested that triticale had a higher grain protein than wheat (non-significant). In summary, NIR seems to over-predict the grain protein content of triticale. This is probably due to the fact that NIR calibrations for triticale have been built from far fewer samples than in wheat (mainly older triticale varieties, and not those grown in the UK). More samples are required to be scanned to get a more reliable calibration. In the meantime it is suggested that NIR predictions of protein in triticale grain should be treated with caution. This applies to the results from the SF trial in 2010 (Table 11) where triticale appeared to have higher protein than wheat. Results from the SF trial also shows there is clearly substantial variation in grain protein content between triticale varieties; the variety Grenado gave some of the lowest protein contents in the experiment whilst the variety Bellac gave by far the highest protein contents. Overall it would seem that grain protein content of triticale is likely to be similar to wheat, so the conclusions of Davis-Knight and Weightman (2008) that alcohol yields per t of triticale are likely to be comparable to wheat still stand.

3.4.4. Conclusions and Recommendations

This report is the first to publish actual alcohol yields and residue viscosities of hard wheat varieties from UK RL trials, and to compare them to reference varieties of soft wheat using a laboratory method. It is also the first to describe a series of N response experiments in which wheat and triticale have been studied under truly comparable conditions with respect to fertiliser N, with assessments made of both grain yield and quality. With respect to the alcohol yield of hard wheat varieties grown on six RL sites in 2009, the conclusions are as follows:

- 1. There are significant differences in AY between hard wheat varieties, with Conqueror and Oakley having particularly high AY, and Ketchum a particularly low AY.
- 2. Glasgow as a soft wheat reference demonstrates superior AY, outperforming the hard wheat varieties.
- 3. There were no significant differences in residue viscosity between hard wheat varieties, and none with the undesirable character of high residue viscosity, as demonstrated by the soft wheat variety Warrior.
- 4. The higher AY demonstrated by Conqueror and Oakley were most likely due to their lower grain protein contents (compared to the other hard wheat varieties grown at the same sites) and hence is more likely to reflect a yield (protein dilution) effect, rather a solely genetic effect.
- 5. The combination of high grain yield and high alcohol yield meant that Oakley and Conqueror had the highest alcohol productivity per unit area, indicating their value for maximising GHG savings.

Taking into account the three wheat and triticale N response experiments carried out in 2010, together with a previous experiment carried out in 2009 (Kindred *et al.*, 2010), the conclusions are as follows:

- 6. Triticale out yielded wheat on three occasions and matched wheat yield in the fourth when grown in the 2nd cereal position and with similar N applications.
- Relative grain protein contents and predicted alcohol yield between triticale and wheat are broadly similar, but differences are inconsistent between sites and protein measurement methods.
- 8. Triticale had a lower N optimum for yield than wheat in one experiment, had the same N optima as wheat in two experiments, and in one experiment there were two triticale varieties with lower N optima than wheat, and two with similar N optima to wheat.
- N optima for triticale appear to be higher than stated in the Defra Fertiliser Manual, however this is the first series of experiments to study the performance of triticale on 'typical wheat' land of high yield potential.

- 10. Given the higher yield with the same and/or less N, these results clearly indicate that triticale can have higher nitrogen use efficiency than wheat.
- 11. These performance benefits of triticale could be greater in a year with a higher incidence of take-all.
- 12. Triticale also appeared to produce more straw and hence total biomass then wheat, which could be particularly valuable in the context of burning biomass for energy.
- 13. Significant lodging in triticale was seen in two trials, although if the crops had been harvested earlier, it is likely that triticale would have outperformed wheat to an even greater extent.
- 14. The results confirm that triticale tends to have lower specific weights than wheat, even in the second cereal position, although this may not be important for bioethanol production.
- 15. In an ADAS trial in 2007 triticale out-yielded wheat by 0.59 t/ha, and an independent commercial trial in 2010 showed that triticale out-yielded wheat by 1.83 t/ha, both trials being carried out in the 1st cereal position.

Recommendations for further work are as follows:

- Further screening of alcohol yields in hard and soft wheat varieties should be carried out, ideally using a laboratory method appropriate to a wheat bioethanol refinery. There would be benefit from this being an annual exercise to inform bioethanol processors, growers and breeders of the best varieties for bioethanol use.
- 2. NIR predictions for grain protein in triticale need to be improved as they currently appear to over-predict for triticale compared to wheat.
- 3. Further field research on triticale should be carried out over a wider range of soil types and rotational positions to evaluate and understand its benefits in terms of higher yields, reduced N requirements and take-all resistance/tolerance, to ascertain whether these are achieved through greater uptake of N or more efficient utilisation of N in producing biomass.
- An assessment of rooting characteristics in triticale should be carried out to understand its better yields and lower N requirements, and potentially to transfer such knowledge to wheat.

- 5. A detailed study of the lodging characteristics of triticale are needed to see if the knowledge gained on wheat can be applied to triticale.
- 6. Alcohol yield of triticale should be assessed using the laboratory method, and residue viscosity assessed.

An integrated project with a biofuel producer is required to finally demonstrate the utility of triticale as a bioethanol crop to the market.

3.5. References

Anon. 2010. Fertiliser Manual (RB209). Defra, London 252pp.

- Aquilina M. 1987. The effect of nitrogen on triticale quality. Aspects of Applied Biology, Cereal Quality 15: 293-296.
- Clarke S, Kindred D R, Weightman R M, Dyer C, Sylvester-Bradley R. 2008. Growing Wheat for Alcohol and Bioethanol Production in the North East. An ADAS study commissioned by the North East of England Processing Industry Cluster Ltd (NEPIC). Version 1.8 31-10-2008. Published at: <u>www.adas.co.uk</u>.
- **Cleal R A E. 1993.** Effect of growth regulators on the grain yield and quality of triticale and wheat grown as a second cereal on light soil. Aspects of Applied Biology, Cereal Quality III 36: 281-286.
- **Davis-Knight H, Weightman R M. 2008.** The potential of triticale as a low input cereal for bioethanol production. Project Report No. 434. Home Grown Cereals Authority, Caledonia House, 223 Pentonville Rd, London, N1 9NG, July, 2008.
- Davis-Knight H, Weightman R M, Agu R, Bringhurst T, Brosnan J. 2010. Maximising bioethanol processing yield of UK wheat: effects of non starch polysaccharides in grain. Project Report No. 467. Home Grown Cereals Authority, Stoneleigh Park, Kenilworth, Warwickshire, CV8 2TL, March 2010.
- **George B J. 1984.** Design and interpretation of nitrogen response experiments. In: The nitrogen requirements of cereals. Reference Book 385. UK Ministry of Agriculture Fisheries and Food, MAFF Publications.
- **Gutteridge R**, **Hornby D**, **Hollins T**, **Prew R**. **1993**. Take-all in autumn-sown wheat, barley, triticale and rye grown with high and low inputs. Plant Pathology 42, 425-431.
- Kindred D R, Weightman R M, Clarke S, Agu R, Brosnan J, Sylvester-Bradley R. 2008a. Developing wheat for the biofuels market. Aspects of Applied Biology 90, Biomass and Energy Crops III; 143-152.

- Kindred D R, Verhoeven T M O, Weightman R M, Swanston J S, Agu R C, Brosnan J M, Sylvester-Bradley R. 2008b. Effects of variety and fertiliser nitrogen on alcohol yield, grain yield, starch and protein content, and protein composition of winter wheat. Journal of Cereal Science, 48; 46-57.
- Kindred D R, Roques S, Weightman R M, Sylvester-Bradley R. 2010a. Evaluating triticale as a sustainable, productive and profitable cereal choice; could it displace much of the second wheat feed market ? ADAS report for Defra Crops Hub, Food and Farming Group, London.
- Kindred D R, Weightman R M, Roques S, Sylvester-Bradley R. 2010b. Low Nitrogen input cereals for bioethanol production. Aspects of Applied Biology 101, Non Food Uses of Crops, 37-44.
- **Naylor R E L. 1987.** The weight and nitrogen content of triticale grains as influenced by nitrogen fertiliser and chlormequat. Aspects of Applied Biology, Cereal Quality 15: 195-200.
- **Overthrow R**, **Carver M F**. **2003**. The value of triticale in the 2nd/3rd cereal position in crop sequences. HGCA project report No. 306. HGCA, Caledonia House, 223 Pentonville Rd, London.
- **Sylvester-Bradley R**, **Kindred D**. **2009**. Analysing nitrogen responses of cereals to prioritise routes to the improvement of nitrogen use efficiency. Journal Of Experimental Botany 60, 1939-1951.
- Sylvester-Bradley R, Kindred D R, Weightman, R M, Thomas W, Swanston J S, Thompson D, Feuerhelm D, Creasy T, Argillier O, Melichar J, Brosnan J, Agu R, Bringhurst T, Foulkes J F, Pask A, Cowe I, Hemongway D, Robinson D, Wilcox S. 2010. Genetic reduction of energy use and emissions of nitrogen through cereal production: GREEN grain. HGCA Project report No. 468. Home Grown Cereals Authority, Stoneleigh Park, Kenilworth, Warwickshire, CV8 2TL, UK, June 2010.
- Weightman R M and Davis-Knight H. 2008. Triticale as a low input cereal for alcohol production. II. Potential to reduce greenhouse gas emissions relative to bioethanol from wheat. Aspects of Applied Biology 90, Biomass and Energy Crops III; 165-172.