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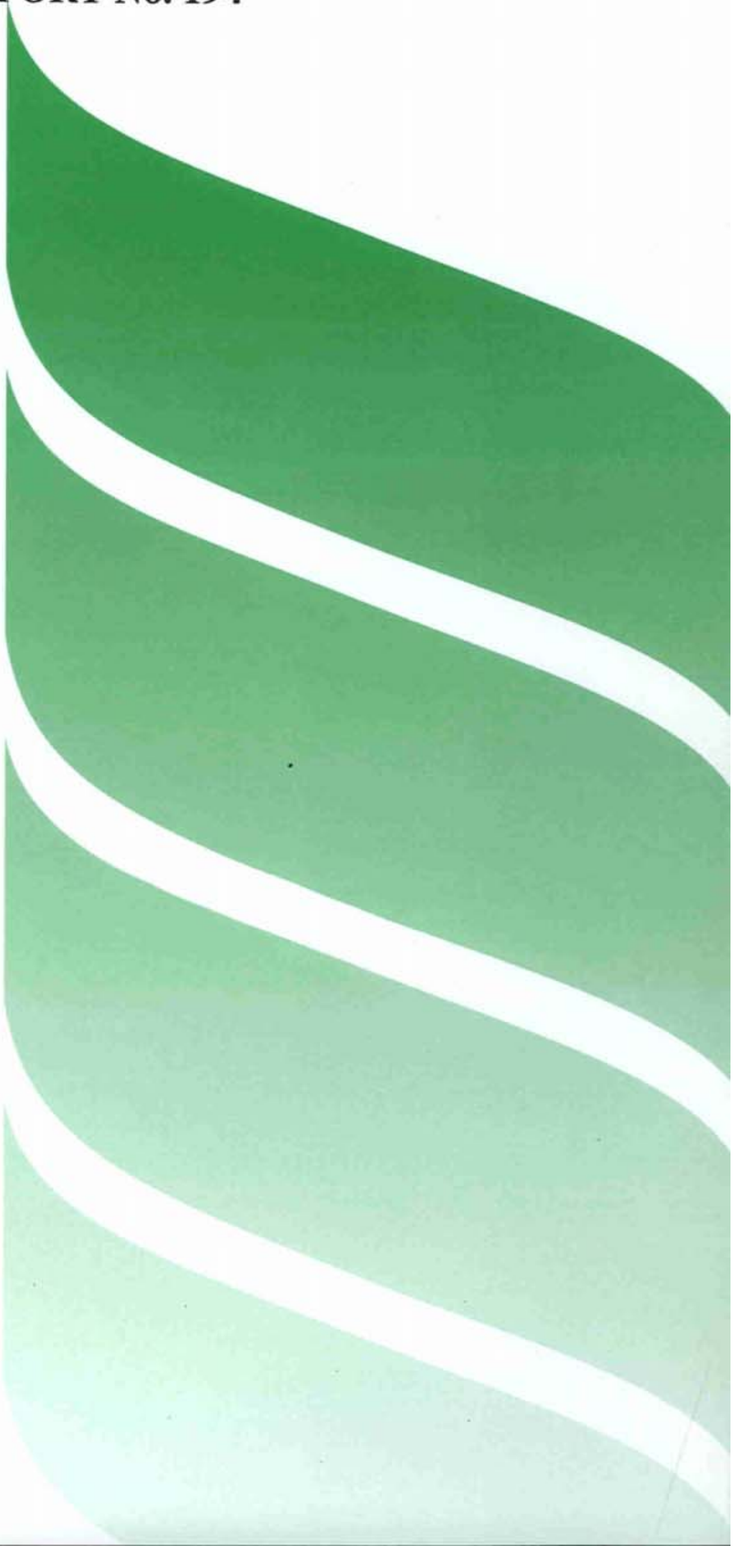

SOURCING ADDED VALUE FOOD INGREDIENTS FROM HOME-GROWN OATS

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SOURCING ADDED VALUE FOOD INGREDIENTS FROM HOME-GROWN OATS

'Innovation'
by

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ABSTRACT

A study was carried out to examine the potential for the fractionation of oat grains and the use of the resultant components as food ingredients. The study included the following elements:

- Assessment of agronomic and environmental effects on grain composition
- Development of a dry milling process for oats
- Analysis of the properties of oat components following dry milling
- Evaluation of the properties of oat flour as a thickener in a béchemal sauce mixture
- A literature review of oat fractionation technology

Grain quality is likely to be an important aspect for oat fractionation and guidelines for oat growers are necessary in order to meet the demands of this market - quality criteria will be more stringent than in other areas where oats are traded. Grain protein content can be largely controlled by the grower through application of appropriate amounts of nitrogen fertiliser as ammonium nitrate or foliar urea but β-glucan and oil contents are more difficult to control and mostly vary due to variety. **β-glucan content was increased in the study through application of foliar urea though further studies are needed to understand the mechanism for this effect. The effect of foliar urea on β-glucan content has significant implications for the growing of oats and provides a means by which the value of the crop can be increased in many markets.**

It was demonstrated that dry milling followed by sieving can be used to prepare starch-rich and bran-rich flours streams. The starch-rich stream contained typically 60% starch, 9% protein and 1% β-glucan while the bran-rich stream contained 48% starch, 15% protein and 5% β-glucan. It was necessary to dehull oats prior to milling but otherwise there were no problems using this technique. Adjustment of mill parameters was necessary to optimise the extraction of components into the flour streams and maximum separation was achieved using a three pass process in which the oversieve material at each stage was passed through the mill again. Further refinement of the milling than that in the study may yield greater separation of components and there was evidence that aspects of grain quality (as yet unidentified) may impact on the efficiency of the process.

Studies of isolated components were concentrated on starch, β-glucan, antioxidants and polar lipids with emulsifier properties. Starch properties were shown to be similar to those published in the literature and did not appear to be affected by agronomy or environment. **An important finding was the presence of significant amounts of α-amylase activity in some samples. There were no significant differences in β-glucan properties which related to agronomic or environmental treatments.** There was evidence that extraction efficiency of β-glucan can be affected by variety and foliar urea application and if proven this could have crop management implications. **Significant antioxidant activity was found in the flour samples.** This was not affected by agronomy but there was evidence that it may be affected by soil water availability. There was also some evidence that milling may be a critical stage in the recovery of antioxidant activity. The glycolipid emulsifier was identified in milled flours but the level did not appear to be affected by any of the factors in the study, in contrast to earlier studies which indicated large differences due to site.

The properties of oat flour in béchemel sauce preparations was compared with chemically modified maize starch and wheat flour. **The oat flour was found to be acceptable for this purpose and sensory properties of the sauce were comparable to sauces prepared with the other thickeners.**

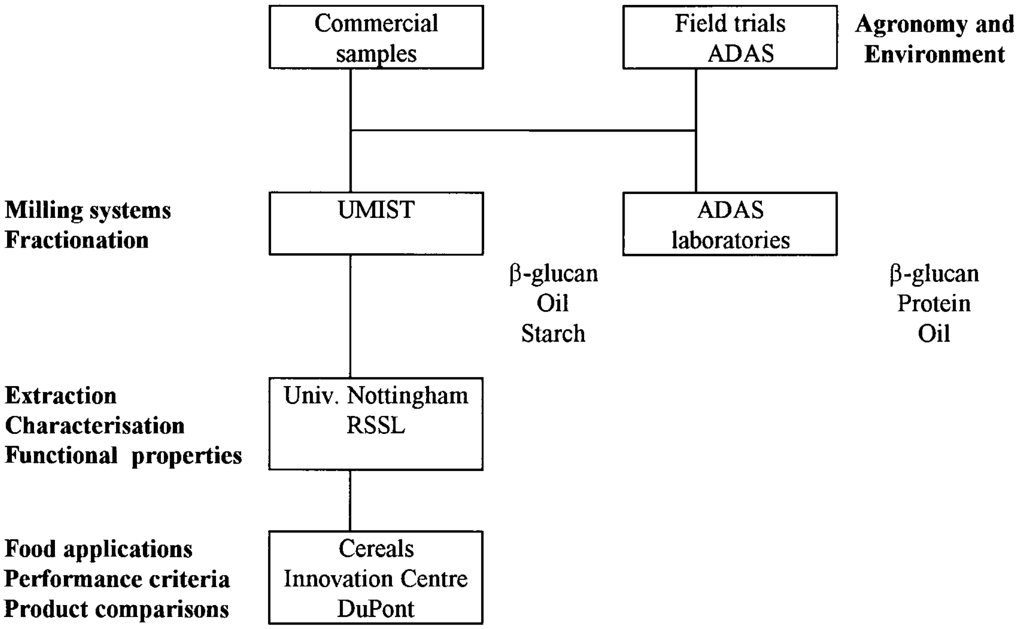
The project demonstrated that considerable potential exists to source food ingredients from oats with dry milling as a preliminary separation stage. **A review of processes available for fractionation of oats revealed that patents surrounding existing processes are not likely to be a barrier to commercialisation of a process.** Further impetus is likely to come from examination of the other oat components which were beyond the scope of this study and also markets for components in other industry sectors. Some of these additional studies are currently being examined by the Objective 5B funded Oatec project.

INTRODUCTION

This reports details the findings of a study aimed at evaluating the potential for the fractionation of oat grains and the use of the resultant components as food ingredients. The study involved research groups at ADAS, the Satake Centre for Grain Process Engineering (UMIST), University of Nottingham, DuPont (UK) Ltd. and Reading Scientific Services Ltd. (RSSL). A number of aspects were covered by the project as follows:

1. The effects of agronomy and environment on grain quality.
2. Enrichment of components into flour fractions by milling and sieving.
3. Extraction and analysis of antioxidants, emulsifiers, starch and β-glucan.
4. Evaluation of oat starch as a thickener in sauce preparations.
5. Review of current oat fractionation technology.

These elements were linked as shown below:



The study addressed a number of key areas for the development of a fractionation process for oats in the UK. The study was intended to complement the Oatec project which is being funded by a consortium of public bodies and private companies including MAFF, the Countryside Agency, the EU Structural Fund, Semundo, The Superioat Co., Ceapro, ConAgra, and Boots plc. The Oatec project began in 1997 and runs until 2001 and is a feasibility study to demonstrate the viability of a UK based oat fractionation plant. The Oatec project is considering mainly non-food uses of oat fractions and hence the current study is complementary by considering food uses. The overall goal of both projects is to provide sufficient incentives for investment in an oat fractionation plant in the UK. In order to provide identity the current project has been named 'Innovation'.

EXECUTIVE SUMMARY

This reports details the findings of a study aimed at evaluating the potential for the fractionation of oat grains and the use of the resultant components as food ingredients.

An important prerequisite for the utilisation of oats for fractionation is the supply of grain of the desired quality. Grain quality will determine the processing efficiency and hence permit high levels of recovery of specific components of interest such as β-glucan and antioxidants. From the growers' perspective this will be important for two reasons: Firstly, the sale of oats into this market will be likely to depend upon strict quality criteria with the necessity for production of oats of this quality being consistent across regions and years. Secondly, the successful manipulation of components of interest may enable premiums to be established for oats with specific and measurable quality criteria. At this stage the optimal design of the fractionation process is not clear, nor are the identity of the components of most interest; hence the area of the study examined gross composition. The effects of agronomy and environment on protein, oil and β-glucan were studied since these components were most likely to be specified for overall grain quality. In this respect the effects of plant nitrogen nutrition and soil water availability were of specific interest since these factors had been shown in previous studies to be of importance for the components examined. A comparison of different oat varieties was also undertaken and the principal factors studied were examined at two contrasting sites over two crop years.

Dry milling was investigated as a preliminary stage in a fractionation process since it was believed to be both economical and also suitable for initial enrichment of bran and starch components. Previous studies had shown dry milling to be effective though this was using oats of lower oil content than those generally found in the UK. For these reasons it was initially felt that low temperature milling may be required. In practice this was not found to be necessary with the oats being successfully milled at ambient temperature. The study also showed that enrichment of components was possible by sieving. The study thus concentrated on milling at ambient temperature and separation of bran-rich and starch-rich streams by sieving.

Following the studies on dry milling more detailed analysis of grain components was undertaken at the University of Nottingham and RSSL; these studies were targeted to areas which were judged to have most current interest in terms of utilisation by the food industry. The areas examined were starch, β-glucan and polar lipid constituents which showed antioxidant and emulsification properties. Other aspects which had originally been proposed for study (oat protein) were deemed to be of lesser importance and were thus not covered by the work.

The studies at DuPont (UK) Ltd. concentrated specifically on the use of a starch enriched flour fraction in sauce preparations; this was chosen for study since it was felt that this is an area where commercial opportunities exist at present. Other areas were felt to be of commercial significance but were beyond the scope of the current project.

Agronomy and environment studies on grain quality

The aim of the agronomy and environment studies was to examine the factors affecting grain quality, which would be of significance for the production of oats for fractionation. Winter oats were grown under a range of conditions, which allowed examination of the effects of site, season, variety, and nitrogen fertilisation. Efforts were also made to examine the effects of soil moisture supply but the relatively high rainfall experienced during the two years of the study meant that only minor effects of modifying soil moisture levels were noted in grain quality. In particular, the high rainfall around anthesis in both years meant that low soil moisture conditions were not possible at either site. The factor targeted for examination in the irrigation experiments was drought, since previous reports had indicated that this can have a significant effect on β-glucan content. Since these two years were unusually wet, compared to previous seasons, it cannot yet be ruled out that drought may affect grain β-glucan levels.

In addition to the development of husbandry guidelines, oats were sourced from this part of the study for use in the milling and component characterisation studies, performed later in the project. This was to allow examination of the effects of agronomy on milling performance and the properties of the grain components.

Three grain components were examined in the field studies – β-glucan, protein and oil. These are discussed in turn below.

β-glucan

β-glucan was of particular interest since it has a high market value. This is estimated at up to £5 per kg for low purity high volume uses and up to £1,500/kg for very high purity low volume uses. The results of the study showed that there were two possible means to manipulate grain β-glucan content – firstly through selection of the variety highest in this component and secondly by the application of foliar urea at anthesis. Careful consideration of variety will be necessary since there was no correlation between grain yield and β-glucan content. Selection of a lower yielding variety may be offset by the premium paid for grain of higher β-glucan content. It is important to note that there were no differences in β-glucan content between naked and conventional varieties when compared on a groat for groat basis. Application of foliar urea at anthesis gave a clear increase in β-glucan content on a percentage basis and this was found at both sites in both years. Interactions between variety and foliar urea were not examined in detail but there was some evidence that variety Image gave a greater response to application of foliar urea than Gerald. In addition to increased β-glucan, foliar urea also gave increases in grain yield and protein content, though had no effect on grain oil content. The overall benefit of applying foliar urea can be seen by considering that application of foliar urea to variety Image at Rosemaund in Year 1 resulted in an additional 40 kg β-glucan per hectare. If only a small proportion of this were paid as a premium to the grower it would make a considerable difference to the economics of oat production. However, the effectiveness of increasing β-glucan using foliar urea would require testing on a wider range of varieties due to the possibility of an interaction between the two factors. The physiological assessments showed that the foliar urea affected crop growth by prolonging green area and thus maintaining the potential for photosynthesis for longer than normal. Other means may be possible to achieve this, which could also be worth examining. These results did support the current theory that much β-glucan is deposited late in development due to thickening of cell walls, which were initiated at an earlier growth stage. The physiological basis for the increase in β-glucan content using foliar urea is not clear and further studies are necessary to establish the basis of the effects seen.

Oil

Oil components could also be of much value as antioxidants and emulsifiers. Trends in the food ingredients industry indicate that the value could be considerable where advantageous properties are identified such as improved functionality compared with existing raw materials. Oil is believed to be mainly determined by genotype although effects of season have also been reported. There was evidence that oil content may be lowered by application of foliar urea, though this did not occur in every situation studied. Further studies are necessary to confirm the effects seen. Efforts by breeders are continuing to develop varieties with higher oil content. This is particularly true with naked oats and estimates from the current study showed that naked varieties were higher in oil on a groat-for-groat basis, even though protein and β-glucan were similar between naked and conventional types.

Protein

Although of less importance in this study, oat protein has potential for use in food systems for its texturising and emulsifying properties. Oat protein content was successfully manipulated through applications of ammonium nitrate or foliar urea, both of which resulted in significant increases. The increases in grain protein content through ammonium nitrate application were independent of grain oil or β-glucan content which showed no response. Other factors affecting grain protein content were site, season and variety.

Grain physical quality

Significant effects on grain physical quality (specific weight and thousand grain weight) were noted with ammonium nitrate application although these may be of lesser importance with respect to grain fractionation. The current measure of quality using specific weight would not necessarily reflect the quality of grain for fractionation and it is not recommended that this should be used for trading purposes where specific grain chemical components are of interest. More accurate measures of quality would be obtained by measuring β-glucan content directly though the cost and time taken for this analysis may preclude its use for trading purposes. The rapid technique using Near Infra-Red Spectroscopy developed at ADAS Laboratories shows promise in this respect but requires further development and would need frequent calibration.

Grain yield

Grain yield was maximised by applying both ammonium nitrate and foliar urea. The optimal amount of ammonium nitrate was site dependent with higher applications at one of the sites studied (Rosemaund) resulting in significant lodging which depressed grain yield. In the absence of a significant premium many growers may seek to maximise grain yield while maintaining quality and the results of the study showed that in most cases this would be straightforward. A complicating situation would exist if 'functional food' products were to be prepared from the milled grain. In this case a specific minimum amount of a component may be required in order to satisfy regulatory requirements (e.g. the cholesterol lowering effect of oat β-glucan). In this case the amount of the component present may be close to the regulatory limit; hence to meet the specification set by food processors it may be necessary for growers to forego yield increases in favour of meeting specifications. Premiums would be required to compensate for the loss of yield and thus grain quality would be of considerably more importance than at present.

Milling studies

There were two aims of the milling studies. Firstly, to develop a dry milling procedure which could be used as a preliminary stage in a fractionation process and secondly to produce milled samples for use in subsequent areas of the project. The mill used for the studies was a Satake STR 100 experimental roller mill. All mill parameters were adjustable and allowed complete optimisation for the oat samples. Careful setting of the mill operating parameters enabled the oats to be milled without the problems of clogging of roll flutes which had been anticipated. Preliminary examination of the milling process utilised commercial samples of oats, varieties Gerald and Kynon but later studies utilised the samples grown in the ADAS experiments.

Groat stabilisation

First stages for consideration in the milling process were stabilisation and dehulling. Stabilisation is necessary to inactivate lipases which are present at high levels in oat groats, particularly after dehulling during which damage occurs to the pericarp layers of the grain. Lipase activity can produce free fatty acids (FFA) which can give rise to rancidity and off-flavours. In the context of fractionation, damage to the groats could also stimulate the production of enzymes which able to degrade the components of interest such as β-glucan and starch. In order to prevent deterioration of these components all samples were dehulled, milled and analysed within as short a time frame as possible. Arrangements were made to transfer milled flours from UMIST to Nottingham within 24 hr, all samples were vacuum packed in a nitrogen atmosphere for transit and they were then analysed immediately upon receipt. This was the only realistic option in the proposed dry milling process since it was likely that steaming would modify the milling properties of the oats and also change the functionality of some of the components. Some studies were carried out at an early stage to confirm that the flour could be stored safely within the timescale proposed. The results showed that although both FFA production and flour acidity began to increase after milling, the flour would not deteriorate to a significant degree within the timescale proposed between milling and extraction. Heat treatment was shown to retard the deterioration of the flour but it was not possible to completely inactivate lipase activity. Commercialisation of the dry milling procedure will require further studies of the storage properties of the whole oats and milled flour since these aspects may be more important in a larger scale process and will be further affected by subsequent extraction steps which have yet to be determined.

Dehulling

Dehulling was a particularly onerous procedure due to the limitations of the laboratory dehuller. No pilot scale dehuller was available for the study although some assistance was granted by Quaker Oats Ltd., later in the study, for which the project is extremely grateful. All samples were dehulled prior to milling since separation of hulls following milling was only partially successful and also generated hazardous dust.

Separation of flour by sieving

Following milling the resultant flour was size fractionated using a series of sieves. Initial studies used a stack of seven sieves to produce eight fractions for analysis. These fractions varied in the proportions of starch, protein and β-glucan but limitations of sample size necessitated combining fractions for the further studies envisaged in the project. An interesting fraction accumulated in the 600 µm sieve which was high in ash and lipid components and it is postulated that this may contain embryo fragments. If so, this could prove to be particularly valuable for the isolation of certain components and warrants further investigation. The 420 µm sieve proved the most appropriate for separation of starch-rich and bran-rich material (see below) so this was chosen as the cut-off for all flours produced from the oats grown in the field studies.

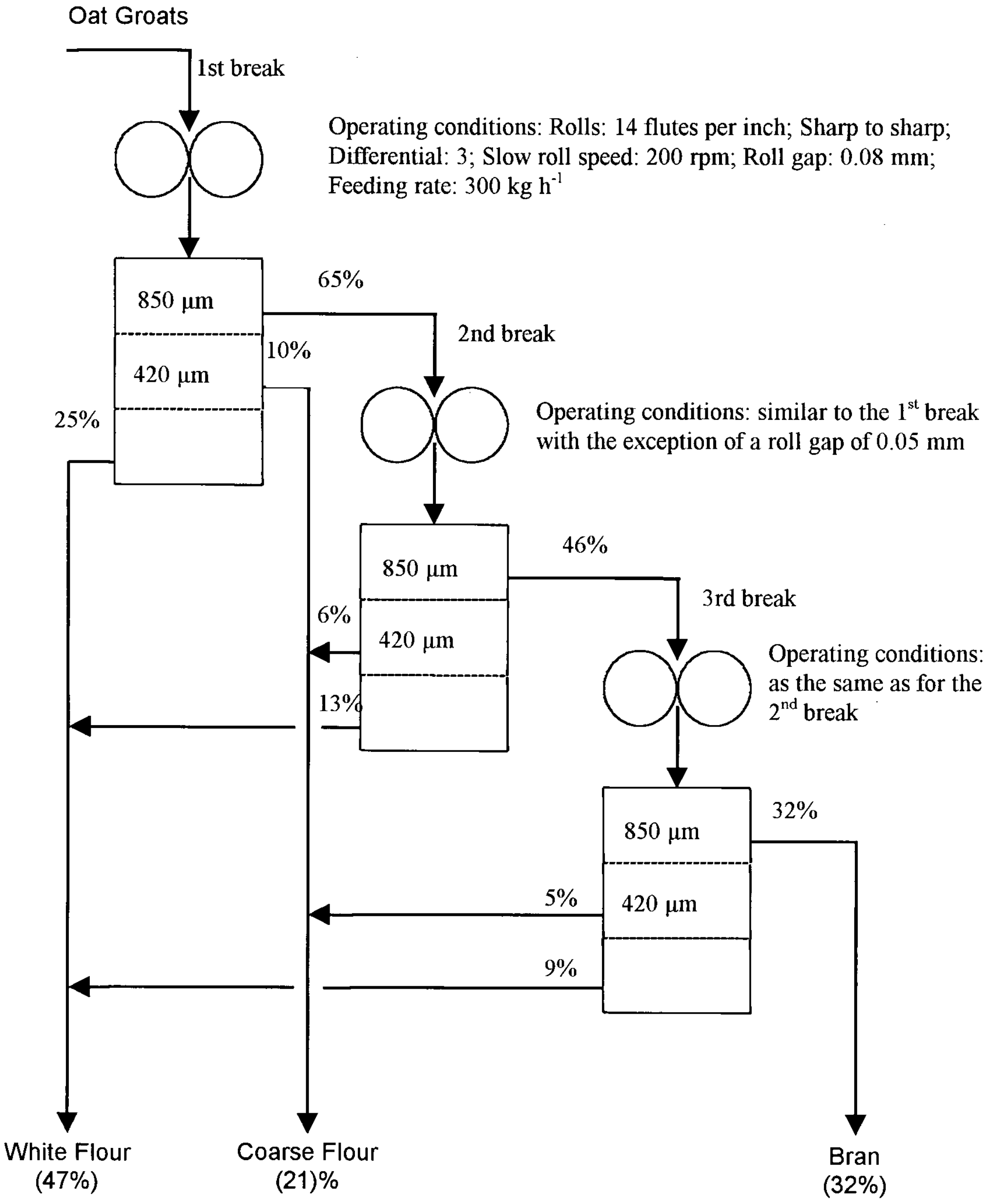
Milling process

An early objective was to develop a milling process which could be used for routine fractionation of flours into starch-rich and bran-rich streams. Mill variables examined were roll speed and differential, roll gap, roll configuration, and the use of successive mill breaks. Optimum slow roll speed was set at 200 rpm with a roll speed differential of 3. Higher speed differentials resulted in more white flour production but could not be justified in terms of increased power consumption and safety considerations. Smaller roll gaps also produced more white flour; hence a gap of 0.08 mm was chosen. This produced flours which when separated by the 420 µm sieve gave very different ash contents and hence indicated good separation of starch and bran components. Ash content was high in bran and low in starch fractions so was shown to be a convenient means to monitor the migration of components during milling and sieving. Roll configuration was also shown to be an important milling parameter with greater numbers of flutes per roll enhancing the production of both sieve fractions. Hence 14 flutes per inch was chosen as the standard roll configuration. Optimal separation was achieved using three successive mill breaks and three sieve fractions. While suitable for a large scale process it was not possible to mill all samples in this way in the study since it was too time consuming. For routine screening, samples were milled using a single break and the flour separated using a 420 µm sieve. Combined with the monitoring of the purity of samples using ash content this provided a convenient and reproducible procedure which could be carried out in the timescale required and also avoided deterioration of samples due to time delays. The optimised milling procedure is depicted below.

Under the milling and sieving conditions selected for the samples in the study two flour fractions were produced which were termed 'starch-rich' and 'bran-rich'. The coarse fraction (>420 μm, 'bran-rich') contained approximately 15% protein, 48% starch and 5% β-glucan while the fine fraction (<420 μm, 'starch-rich') contained approximately 9% protein, 60% starch and 1% β-glucan. Examination of the two flour streams showed that the fine fraction (starch-rich) was largely derived from the endosperm whereas the coarse fraction (bran-rich) was from the exterior layers of the grain (pericarp, testa, aleurone and sub-aleurone). Results showed that lipids were distributed roughly evenly between the two flour fractions so there appears to be no benefit in terms of lipid composition in fractionating the flour in this way if crude oat oil is the main fraction sought.

Samples were selected from those grown by ADAS for examination of milling performance and subsequent extraction and analysis of components at Nottingham University, DuPont and RSSL. Time taken for dehulling, milling, extraction of components and their analysis did not permit assessment of all samples from the field experiments. Samples were selected for milling on the basis of comparisons of the most interesting agronomic and environmental variables identified in the earlier stages of the project (see below). The rationale for selection of the samples milled for analysis at DuPont was based on differences identified in the pasting properties of the starch-rich fraction. The amounts of starch-rich and bran-rich flours produced on milling the samples were recorded. Although differences were noted they did not appear to relate to any of the agronomic variables examined. It was thus not possible to determine the influence of agronomic treatment on flour yield.

The milling process developed in the study provided a convenient means for producing crude fractions from whole oats enriched in either starch or β-glucan. It is possible that these flours could be used with further processing, such as was demonstrated later in the project, for example in sauce preparations. Other potential uses for the flour fractions exist e.g. the inclusion of the bran rich fraction in functional food products, associated with the cholesterol lowering health claim for β-glucan. However, it is also likely that further processing would be required to enrich or extract specific components and further studies are needed. A particular area of interest is the extraction of starch; removal of the contaminating bran from the starch using a combination of dry milling and sieving procedures may enhance the recovery of starch. This could influence its properties during subsequent processing and it is recommended that this area be investigated further. The dry milling process thus provided a convenient first step for commercial oat fractionation.



Prototype milling process for dry milling of oats.

Samples selected for dry milling at UMIST

Sample code	Location	Variety	Soil moisture control (irrigation)	Ammonium nitrate kg N/ha	Foliar urea Kg N/ha
G1	Rosemaund	Gerald	-	0	0
G2			-	0	60
G3			-	40	0
G4			-	40	60
G5			-	100	0
G6			-	100	60
I1		Image	-	0	0
I2			-	0	60
I3			-	40	0
I5			-	100	0
I6			-	100	60
N1	Gleadthorpe	Gerald	-	40	0
N2			-	100	0
N3			-	140	0
IR1		Gerald	Nil	100	0
IR2			Early	100	0
IR3			Late	100	0
V1	Rosemaund	Aintree	-	80	0
V2		Chamois	-	80	0
V4		Emperor	-	80	0
V5		Gerald	-	80	0
V7		Image	-	80	0
V9		Krypton	-	80	0
V10		Kynon	-	80	0
V11		Lexicon	-	80	0
V12		Solva	-	80	0

Extraction and analysis of antioxidants, emulsifiers, starch and β-glucan

It was decided that the studies at Nottingham University would concentrate on starch, β-glucan, antioxidants and the glycolipid emulsifier. A number of oat samples were selected for analysis from those grown by ADAS (see section above). The samples were dehulled and milled at UMIST and then sent for immediate extraction of components and analysis at Nottingham.

Starch

The starch component was important since it represents a large proportion of the total grain weight; the absence of a suitable market for the starch would mean that large amounts of waste material would be likely to be produced. Favourable economics for a fractionation process would thus be likely to be dependent on there being a suitable market for this component. It is unlikely that oat starch could compete in commodity markets and thus specific unique properties of oat starch would need to be identified for its successful commercialisation.

The gelatinisation and pasting properties of oat starches were examined using differential scanning calorimetry (DSC) and a Rapid Visco Analyser (RVA™). All starches showed peak gelatinisation temperatures (Gt) close to 60°C and similar endotherm (δH) values. It was only possible to examine a limited number of samples so it is not known whether Gt or δH were affected by variety or agronomy. The gelatinisation temperatures were close to recorded values for wheat and barley but lower than those for maize and potato starches. Pasting properties were examined for all samples milled in the project. It was noted that in general the oat starches examined showed a high pasting temperature and low peak viscosity. Only rice starch showed a lower peak viscosity than oat starch. Many samples were also shown to contain α-amylase activity, as evidenced by differences in pasting properties when silver nitrate (amylase inhibitor) was included in the analysis. The reason for the presence of α-amylase is not known since the grain was harvested at low moisture content under good field conditions. One possibility was that the samples varied in the degree of maturity at harvest since it was noted that samples to which foliar urea was applied showed a greater difference between samples pasted with and without addition of silver nitrate. It was also interesting to note that the fraction examined was that from the starch-rich stream in which it would be expected that α-amylase activity would be low (since it develops in the aleurone layer of the grain). This could indicate that the α-amylase activity of the bran-rich fraction may be even higher. Small differences between peak and final viscosities were found in RVA results between samples but did not appear to be linked to varietal or agronomic factors.

β-glucan

Oat β-glucan was included in the study since there has been much recent interest in its cholesterol-lowering properties, as well as the potential for use as a thickener in food systems. A number of methods were possible for extraction of β-glucan from the samples but that reported by Dawkins and Nnanna (1993) was chosen since it was relatively straightforward while producing β-glucan of high purity. All samples were defatted with isopropanol prior to β-glucan extraction since this was found to give a considerable increase in recovery. Essentially the method for extraction involved stirring in aqueous suspension at pH 9.2 and centrifuging to remove the starch, adjusting to pH 4.5 and centrifuging to remove the protein, then adjusting to pH 7.0 and making up to 50% isopropanol to precipitate the β-glucan which was then dried. Despite the rigorous nature of the procedure subsequent analysis revealed that some β-glucan precipitates were contaminated with starch and created problems with subsequent chromatographic analyses. This was found with samples extracted from both starch-rich and bran-rich milling streams. It was interesting to note that all bran-rich samples derived from the ADAS variety trial were free of starch contamination and that the starch-rich flours from these samples were also shown to be free of traces of α-amylase. The detection of zero α-amylase activity was thus shown to be an important quality criterion for industrial oat fractionation where starch and β-glucan are to be extracted.

Recoveries of β-glucan from the starch-rich stream were 1.5 to 3.9% and from the bran-rich stream were 4.3 to 11.0% and there did not appear to be any firm correlation between the agronomic treatments and the recovery. Although there were obvious differences between varieties it was not possible to determine whether these were real or artefactual though it was noted that Chamois, which had the highest grain β-glucan content, also gave the highest recovery in the extraction. There was also slight evidence that application of foliar urea to the crop was associated with higher recovery of β-glucan. These are aspects which should be explored further.

Analysis of β-glucan extracts by SEC/MALS gave reproducible estimates of molecular weight (MW) for the bran-rich but not the starch-rich stream (due to the presence of contaminating starch). The values for MW obtained were consistent with those reported in the literature, lying in between the range reported. It must be noted though that there was some evidence of the presence of β-glucanase in some of the samples which may have degraded the β-glucan thus giving an underestimation of the MW. Due to the possible presence of starch and β-glucanase in some samples it could not be determined whether true differences in the MW of β-glucan existed between the two milling streams. There were also no discernible differences between samples from different agronomic treatments. It was particularly noticeable that all bran-rich samples from the variety trial were free of contaminating starch. All samples from the variety trial were of significantly higher MW than those from other experiments which may have thus contained some β-glucanase activity. The absence of both α-amylase and β-glucanase in samples may thus be key parameters to be considered for the quality of oats for industrial fractionation, even in fractionation schemes in which high temperature solvent extraction is used as a first step (as in this study). Both enzymes are produced in the scutellar and aleurone regions of the grain and would be expected to be concentrated in the bran-rich milling stream. Further studies should be carried out to determine the conditions required to stabilise the grain and prevent deterioration of starch and β-glucan by these enzymes.

Antioxidants and glycolipid emulsifier

There has been much interest in obtaining natural antioxidant compounds for use in the food industry in addition to the interest in antioxidants for their health benefits per se. Oat polar lipid extracts had previously been shown to contain high levels of phenolic antioxidants. There has also been much interest in the glycolipid emulsifier from oats and its potential for use in chocolate. Both the antioxidant components and glycolipid emulsifier were present within the polar lipid fraction of the oat oil.

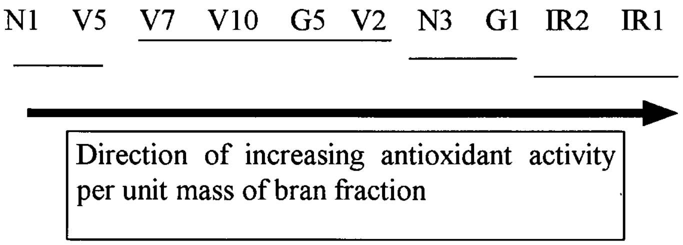
An initial aim in the study was to determine the optimal extraction conditions for the polar lipid components. A method was developed which used isopropanol (IPA) at 70°C for 2 hr. This was shown to extract high levels of polar lipids as well as minimising the amount of solvent required which was likely to be important in a commercial fractionation process. It was also possible to extract both antioxidant and emulsifier components using this procedure. The recovery of lipids using this procedure was similar to that quoted for other methods in the literature (5-6% w/w of whole grain) and the reproducibility of the method was confirmed from analysis of replicate extraction from the same material. The stability of the extracts was also demonstrated in that they were shown to retain antioxidant activity when stored in the dark at -80°C for 6 months.

A novel method for assessment of antioxidant activity was developed in the project which was based on a chemiluminescent reaction. This showed much higher sensitivity than the commonly used β-carotene bleaching method. The chemiluminescent method showed that the antioxidant activity was close to that reported in previous studies in which oats were examined.

Similar content of total phenolic components was found between the oat varieties examined. Although some significant differences were noted these were small in real terms. There was a close correlation between antioxidant activity and total phenolic content. Nitrogen fertilisation appeared to have no influence on total phenolic content or antioxidant activity at either site and there were no differences between the sites despite quite large differences in other aspects of grain quality. There did however appear to be a significant effect of soil water availability on total phenolic content (though not antioxidant activity) at the Gleadthorpe site; samples from the Rosemaund site were not examined at this stage of the study. Irrigation at pre-anthesis ('early') and post-anthesis ('late') stages resulted in a stepwise decrease in the level of total phenolics. This may indicate that drought conditions can result in higher levels of phenolic components.

All the above observations were made on whole milled oat samples. A comparison was also made between the starch-rich and bran-rich milling streams. Total phenolic components were shown to be enriched in the bran-rich stream to between 52-196% in all samples except those grown in the variety trial - these showed the reverse pattern with higher total phenolics in the starch-rich stream. This may indicate the sensitivity of the milling and sieving steps in the process since it is possible that the size class of the flour particles containing the phenolics is very close to the 420 μm sieve size which was selected as the cut-off point for the separation. There was indeed some evidence in the early assessment of the milling process that specific fractions which were combined with others in the final scheme were particularly high in lipids. These results indicated there is considerable further potential to refine the dry-milling process to produce flours with much greater enrichment of certain phenolic components. However, as was shown in this study, grain composition as influenced by agronomic and environmental factors is likely also to be important and should be considered in relation to any process proposed. Further studies should include examination of a wider range of flour particle size fractions and the effect of grain composition on the distribution of components between these fractions.

Calculation of 'phenolic equivalents' revealed that the bran-rich fraction contained a more potent mixture of phenolic components in terms of antioxidant activity than the starch-rich fraction. This is despite the fact that in general the total phenolic content was mirrored by the antioxidant activity. This showed that it was possible to enrich the antioxidant activity of the flour without changing the overall levels of total phenolics. The relative rankings of samples in terms of antioxidant activity were calculated and showed that there were significant differences between samples.



Although some discrepancies in the ranking order of samples were found on comparing antioxidant activity results between bran-rich fractions and whole flour, this result showed that selective extraction of antioxidant compounds into the bran-rich milling stream was taking place. However, due to the limited number of samples examined it was not possible to determine whether this was affected by the agronomic conditions under which the grain samples were grown. It can be speculated from the results that selective separation at the milling stage of certain phenolics may cause synergistic or inhibitory effects which could lead to increases or decreases in antioxidant activity in the milled sample streams.

From previous work on the additions of oat phenolics to plant oils (to enhance oxidative stability), commercially viable effects were observed at 0.01% (w/w) of total phenolics. Given this value, and the concentration of phenolics in the extracts obtained in this study, one can estimate that 1 kg of oats would have to be processed to yield enough phenolics to be added to 1 kg of oil.

The glycolipid emulsifier component was separated from the other components in the polar lipid extracts by thin-layer chromatography and quantified by image analysis. Absolute measurement of this component was not possible since no standard material was available but the relative amount was compared with polar lipid material extracted from a standard sample. The presence of the glycolipid in the samples was confirmed but there were no differences in the relative amounts between oat varieties or agronomic treatments. There were also no differences in amounts between the starch-rich and bran-rich milling streams. These results contradicted those from previous studies at RSSL which indicated that large variations existed in the amounts of the glycolipid between different oat samples. However, the result was important since it showed that samples for fractionation could be selected on the basis of other oat components without affecting the amounts of emulsifier extracted.

Evaluation of oat starch as a thickener in béchamel sauce preparations

The use of oat flour as a thickener for sauce preparations was chosen as a suitable application to test the samples produced in the study. Chemically modified waxy maize starches for example Colflo 67 a product of National Starch & Chemical Ltd are currently used for this purpose due to their superior processing properties. There is, however, much interest in using natural (non-chemically treated) starch sources in such applications, to enable 'clean labelling' declarations to be listed on food packaging. Commercial wheat flours are available which could be considered for use in such applications. Viceroy flour, a product of RHM Ltd, was included in the study, as a reference material, against which the oat flour could be compared.. The starch rich oat flour fraction was chosen for the sauce evaluation. The bran rich fraction was too coarse in texture and showed a high grey/brown colour which would not have been suitable in a sauce application.

A preliminary study was carried out to examine the behaviour of the oat flour in a sauce system and to determine the optimal process for preparing the sauce. During this initial study, sensory panellists were trained to recognise the textural and organoleptic characteristics of the sauces, in preparation for the larger study.

In the larger study, oat samples were selected such as to enable comparisons to be made between varieties and to examine the effects of nitrogen application. The varieties were selected based on their contrasting rheological properties (RVA^{TM} pasting profiles) as identified in studies at Nottingham University. The samples were dehulled, milled, heat-treated in a laboratory scale cooker, and sauces prepared for sensory profiling. The oat samples were found to be very similar to the sample prepared with wheat flour, for many of the attributes examined. There were some differences, most notably the oat samples were thinner, darker and some were less buttery. The sauce made with the naked oat sample Kynon was also found to be similar to the wheat flour sample in terms of the degree of 'setting' when allowed to go cold. The oat samples (and wheat flour) were all very different to the modified maize starch in terms of texture and appearance although there were some similarities in aroma, flavour and aftertaste. There were no large differences in organoleptic properties between the oat samples analysed. The differences which were noted, relate to the presence of dark specks (arising from incomplete dehulling during flour preparation) lightness and the extent of setting when left to go cold. No differences could be discerned due to the application of nitrogen during crop growth. The differences observed between varieties, which were mainly in appearance, may have been due more to the processing of the samples than due to inherent differences in grain composition.

The results of this part of the study indicated that the oat flour had considerable potential for substitution in place of wheat flour in such applications. It was also shown that the quality of oats was not critical in this particular application since the range of samples examined had quite different physical and chemical characteristics. It was particularly important to note that no off-flavours were noted considering that there was a significant time delay (1-2 weeks) between dehulling the preparation of the sauces. Rancidity problems due to lipase activity might have been expected.

Industrial fractionation technology

A review was undertaken to examine possible fractionation technology available at the current time. There is little commercial fractionation of oats taking place world-wide though a number of initiatives have been attempted in recent years, though there is no large scale processing at present. This creates an opportunity for commercial developments in this area in the UK. The review showed that there is no barrier in terms of methods for fractionating oat components. Although many patents exist this is not likely to prevent the development of a suitable process. The current project demonstrated that dry-milling oats may be a logical first step in any process and this would fit well with many of the methods published.

Significance of the study for commercial opportunities

The quality of UK grown winter oats was shown to be suitable for many of the proposed applications for oat fractions in both food and non-food markets. The project has contributed to the understanding of factors affecting these quality attributes and has demonstrated practical means by which the grower can enhance the value of the crop where specific components such as β-glucan are required. This will assist in achieving quality premiums for growers.

The project has demonstrated the viability of dry-milling as a primary extraction step; this has much significance for commercialisation of the fractionation process since dry-milling is well established in the wheat milling industry and could be readily modified to process oats. Unlike many other processes for fractionation of oats dry-milling is not restricted by being covered by patents. The process is also extremely flexible, allowing secondary processes to be added as required. For lower value applications the basic dry-milling process may produce oat flour of sufficient quality in an economic fashion for many purposes. This may open possibilities for using oats in food ingredient applications which would otherwise not be possible. It was particularly important to note that the oat flour examined was robust in nature with no rancid flavours developing during its processing and examination. This was despite no heat treatment of the original oats and the storage over a two week period. A heat treatment step was used in the flour samples evaluated in the sauce application. This was required not only to inactivate lipases but also to inactivate any amylase enzymes present in the flours.

In addition to sourcing relatively crude oat fractions by dry-milling alone, the results of the studies at Nottingham University showed that potential exists to source novel and high value components from oats. High antioxidant properties were demonstrated in the polar lipid extracts and it was shown that there was scope for manipulation of these components by agronomic and milling techniques. The glycolipid emulsifier was also shown to be present in these extracts thus permitting the isolation of both components in a single step. Other components (β-glucan, starch) were shown to have potential in this respect although they may require further development both in terms of extraction and identification of markets.

The project has contributed significantly to the ultimate aim of attracting investment in a UK based oat fractionation plant. The commercial opportunities for using oats in non-food and food applications will continue to be examined by the Oatec project which is generating considerable interest world-wide.

Recommendations for further studies

These are included within each separate section of the report.

Sourcing added value food ingredients from home grown oats *'Innovation'*

Part 1

Agronomy and environment effects on grain composition

by

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Summary

The effects of agronomic and environmental variables on grain yield and quality were studied in field experiments. These were part of a larger study examining the potential for sourcing novel food ingredients from oats. This part of the study was carried out by ADAS and was aimed at producing crop management guidelines for oats of high quality for industrial fractionation, as well as grain samples for use in other areas of the study.

Variables examined were water supply and nitrogen availability. The experimental sites were chosen to represent differences in soil type, with Rosemaund having a medium soil with high water holding capacity, and Gleadthorpe a light soil with low water-holding capacity and propensity for summer droughts. Soil moisture deficit was monitored throughout the study using 'Irriguide', and in combination with irrigation systems set up within the experiments was used to control the availability of water to the crops. This resulted in a number of treatments aimed at optimising crop growth either 'early' (irrigated during Growth Stages 31-61) or 'late' (irrigated during Growth Stages 61-87). These experiments were carried out at both sites but at Rosemaund the control of water availability was examined in combination with the nitrogen treatments. Nitrogen supply to the crops was varied by applying different amounts of ammonium nitrate as prills, and urea was applied as a foliar spray at flowering. These treatments were carried out at both sites and the experiments were repeated over two years.

Addition of ammonium nitrate resulted in higher shoot numbers through increased tiller survival, increased crop height, and increased Green Area Index. There were also increases in the amounts of stem carbohydrate reserves and an increased crop dry mass, reflecting the changes in shoot numbers and Green Area Index. These changes with ammonium nitrate enabled the crop to intercept more solar radiation and to maintain photosynthetic capacity, increasing the yield potential. The effects were found mainly where additional ammonium nitrate had increased canopy size to between GAI 5 and 6 since canopies larger than this showed negligible increases in photosynthetically active radiation interception, biomass or yield. Application of foliar urea was also shown to have significant effects on the physiological status of the crop, resulting in delaying senescence compared to plots which received no foliar urea. This extended the grain filling period by maintaining light interception for longer than normal and was reflected in higher grain yield. The impact of irrigation was not as great as had been expected due to the high rainfall at critical points during the season.

All factors examined within the study (variety, site, season, ammonium nitrate, foliar urea and water supply) were shown to influence grain yield and many of these also affected grain quality. The most significant differences were due to variety and site, though large seasonal effects were noted at both sites. Large differences in yield, grain physical characteristics and composition were noted between varieties and the experiment provided a basis on which a variety could be ranked according to grain yield and quality. Varieties producing the highest yield were generally those producing the greatest amounts of protein, oil and β-glucan. A comparison of conventional and naked varieties on a groat-for-groat basis (assuming the husk contributes 25% to the total weight of the grain) showed that there was no difference

in the amounts of protein and β-glucan produced but that naked oats were richer in oil.

Application of ammonium nitrate increased grain yield but this reached a maximum at one of the sites in the study (Rosemaund) in which yield declined due to the effects of lodging. Effects were also noted in grain physical characteristics with decreases in both specific weight and TGW at higher ammonium nitrate levels. Increased yield also resulted from application of nitrogen as foliar urea in most cases. Both ammonium nitrate and foliar urea influenced grain composition. Ammonium nitrate application produced a stepwise increase in grain protein content but had no effect on oil or β-glucan content. Application of foliar urea resulted in increased protein levels and also β-glucan. Concurrent with these changes were decreases in grain oil. **This in combination with the increase in yield seen is a significant development since it is the first recorded means for increasing β-glucan content in field crops.** No explanation can be given at this stage for the effect of foliar urea on β-glucan but there was some evidence of a varietal influence on the ability to manipulate β-glucan in this way. Recommendations are made for further research in this area.

The application of irrigation at the 'early' phase resulted in increased grain yield at Gleadthorpe in Year 1 but not Year 2. There were no other significant effects of soil water availability at either site and no effects on any of the aspects of grain quality examined. The lack of response to irrigation was due to the unusually wet weather during the study which meant that the crops did not experience drought stress. It is recommended that further studies be carried out using automated crop shelters to create true drought conditions.

The present study has shown that potential exists to manipulate the quality of oats by using appropriate ammonium nitrate and foliar urea programmes. **The increase in grain β-glucan content with foliar urea application is of particular importance for manipulating the quality of grain for industrial fractionation.**

Recommendations for further studies

- Examination of the effects of foliar urea on a wider range of winter oat varieties since significant interactions between variety and foliar urea may exist. This is of importance for both industrial fractionation and also in relation to the manufacture of 'functional foods' based on the FDA health claim for oat soluble fibre.
- Examination of the effects of other factors which can increase grain filling and may thus increase grain β-glucan content. This should include the use of strobilurin fungicides, which have been shown to lead to increases in grain filling in wheat.
- Further studies examining the effects of drought on grain composition. These should be carried out using automated crop shelters to guarantee drought conditions are experienced. This will also allow examination of drought at specific growth stages.
- More detailed studies of crop growth and development for winter oats. Although the current study provided some information, much greater detail is required for full understanding of the basis for grain yield and quality.

Introduction

A key factor in the establishment of a commercial oat fractionation plant will be the ability to source oats of suitable quality for processing. It is important that oat growers can produce grain with components of interest at high concentration and in an easily extractable form. The aim of this part of the study is to identify the key agronomic and environmental factors affecting grain quality. The research was intended to result in crop management guidelines for the oat crop destined for fractionation. This will enable growers to produce grain of the desired quality for the new markets identified in other areas of the project. It will also provide end-users with information regarding specifications required for optimal processing and component functionality. This will enable quality criteria to be identified which will assist the establishment of grower premiums.

Selection of the correct variety is likely to remain the major consideration for grain yield and quality in oats and significant effort is being directed towards breeding new varieties adapted to specific markets (Valentine, 1996). However, numerous reports have also demonstrated the importance of correct agronomic practice and favourable environmental conditions for meeting specific quality criteria within a given variety. A recently completed MAFF study showed that there was considerable variation in composition as a result of sowing date, nitrogen fertilisation, site and season, although the relative importance of these factors was not clear (Laverick, 1997; Davies and Givens, 1998).

There is much current interest in oat β-glucan, both in terms of its viscometric properties and also for its positive health benefits. The reports above were the first detailed investigations of variation in β-glucan content in winter oats and build on a number of other reports concerning spring oats (Brunner and Freed, 1994; Humphreys *et al.*, 1994; Saastamoinen *et al.*, 1992; Saastamoinen, 1995). The variations in β-glucan content in these reports appeared to be more due to environmental than agronomic effects but with an underlying genetic factor. It has been reported that β-glucan can be affected largely by soil moisture (Schuster *et al.*, 1967; Bendelow, 1975; Smart, 1976), though the literature in this area is confusing since some conflicting reports also exist which indicate that actual evapotranspiration is more important than soil moisture per se. Many of these studies have been carried out under artificial conditions and have concentrated on barley rather than oats. One of the aims of the current field studies is to determine the extent to which the amount of β-glucan can be manipulated. Utilising field studies will enable these findings to be readily exploited for the benefit of the cereal grower.

In addition to β-glucan, protein and oil are also of much interest for novel food and industrial applications. Protein content in oats is known to be largely determined by the fertility of the soil and the amount of nitrogen fertiliser applied to the crop (Laverick, 1997; Chalmers *et al.*, 1998) but less information is available regarding factors affecting oil content. It is generally believed that this is genetically determined and is less affected by agronomic practice or environment than protein or β-glucan. It is important to understand the effects of agronomic and environmental factors on these components in order to produce crop management guidelines. Of particular interest are the interactions between these components and how the amount and their extractability and their functionality can be maximised.

A key factor underlying the manipulation of grain yield and quality is the 'canopy management' principle which has been developed for wheat (HGCA, 1997). Nitrogen fertilisation is a key husbandry factor which must be considered for optimal grain yield and also quality and is a key element of the concept of 'canopy management'. Nitrogen is essential for production of plant proteins including those associated with photosynthesis and is assimilated from the soil as ammonium or nitrate ions. Although soil reserves of nitrogen exist from mineralisation during the season most crops benefit from application of additional nitrogen as fertiliser during the spring. In most cases the amount of nitrogen applied has a significant effect on the capacity of the crop to intercept sunlight and therefore to accumulate dry matter and enhance yield. This occurs due to a number of changes, including increasing the number of fertile shoots through encouraging tiller survival, a factor shown to account for a large proportion of variation in grain yield in wheat (HGCA, 1997). This is the basis of canopy management since it has been shown that husbandry effects on leaf number and leaf size are small. Essentially, the larger the canopy the greater the capacity to intercept sunlight and hence higher yield. Further applications of nitrogen are also possible later in development as foliar sprays; these have no effects on overall canopy size but can enhance grain yield by delaying senescence, enabling the crop to photosynthesise for longer and produce additional assimilate for grain filling.

In the current study nitrogen fertiliser was applied in the spring as solid ammonium nitrate prills. The levels applied in the study were as follows:

- 'Low' - none applied at Rosemaund, 40 kg N/ha at Gleadthorpe - very low levels aimed at producing grain with much lower than normal protein content.
- 'Medium' - 40 kg N/ha at Rosemaund, 100 kg N/ha at Gleadthorpe - a sub-optimal amount aimed at producing grain with slightly lower than normal protein content.
- 'High' - 100 kg/ha at Rosemaund, 140 kg/ha at Gleadthorpe - an amount slightly above that normally recommended to produce grain with elevated protein content.

Grain protein content was further modified by applying foliar urea in combination with the ammonium nitrate treatments.

Since the study was replicated at two sites with differing soil fertility it was expected that a wide range in grain composition would result, with both low and high grain protein samples in each year of the study. The rationale behind the nitrogen treatments on the structure of the crop canopy are shown in Table 1.

Table 1. Anticipated effects of canopy manipulation through applied nitrogen

Nitrogen level	Ammonium nitrate applied at GS 30-32 kg N/ha	Foliar urea applied at GS 61 kg N/ha	Canopy size	Canopy retention
'Low'	0 (40)	0	Small	Poor
	0 (40)	60		Good
'Medium'	40 (100)	0	Medium	Poor
	40 (100)	60		Good
'High'	100 (140)	0	Large	Poor
	100 (140)	60		Good

( ) Amount applied at Gleadthorpe – higher than that at Rosemaund due to lower soil nitrogen supply at this site

Adequate soil water supply is a further factor expected to influence grain yield and quality. It is essential at all stages of plant growth and is determined by two principle factors in field crops - rainfall and soil water holding capacity. Drought conditions are more likely in light soil types even with adequate rainfall since water drains rapidly from the soil in addition to the loss from the transpiring canopy. The experiments in this study took place at two sites - Rosemaund in Herefordshire, which is a silty clay loam with high water holding capacity, and Gleadthorpe in Nottinghamshire, which is a sandy loam with low water-holding capacity and propensity for summer droughts. Despite the differences in location the rainfall was similar at each site, differences in soil moisture levels arising as a result of the contrasting soil water holding capacity. Irrigation facilities were established at both sites using 'trickle' systems in Year 1 (1996/7) with 'trickle' being used at Rosemaund again in Year 2 (1997/8) though overhead sprays were used at Gleadthorpe in this year. The aim of these experiments was to manipulate crop growth by maintaining water supply through irrigation at specific developmental phases whilst unirrigated plots acted as controls. It was anticipated based on previous experience that drought conditions were possible in the control plots, particularly at Gleadthorpe. The irrigation was further split into 'early' and 'late' phases, optimising tiller retention and stem carbohydrate reserves in the 'early' phase (GS 31-61) and grain production and grain filling in the 'late' phase (GS 61-87). An additional treatment was established at Gleadthorpe in which all phases were optimised by applying irrigation throughout the whole growth period ('thoughout'). It was not intended that the irrigation treatments would be advocated to growers; this was merely used as a tool for investigating the effects of soil water supply on grain quality. A summary of the irrigation treatments is shown in Table 2.

Table 2. Anticipated effects of manipulating crop growth and biomass partitioning through water supply

Timing of irrigation (GS)	Crop growth parameters optimised
31-61 ('Early')	Tiller retention Soluble carbohydrate reserves
61-87 ('Late')	Grain production (no.) Grain filling

At Rosemaund the irrigation treatments were combined with the nitrogen treatments in order to investigate possible interactive effects.

The influence of variety on grain quality was also examined by sourcing grain from a variety trial at the same site in Year 1.. The inclusion of two varieties (Gerald, Image) in the experiments examining nitrogen fertilisation also allowed the comparison of the effects of nitrogen between two varieties with contrasting degrees of tillering.

In addition to investigating the effects of the field treatments grain samples were sourced from the experiments for the dry milling studies at UMIST. It was not possible to mill all grain samples so a sub-selection was made of those samples which showed the greatest interest in terms of the effects on grain quality.

The study will provide important information relevant to the establishment of an oat fractionation plant in the UK. This study will be complemented by the Oatec project which is being funded by a consortium of public bodies and private companies including MAFF, the Countryside Agency, the EU Structural Fund, Semundo, The Superioat Co., Ceapro, ConAgra, and Boots plc. The project began in 1997 and runs until 2001 and the aim is to undertake a feasibility study to demonstrate the viability of a UK based oat fractionation plant. The Oatec project is considering mainly non-food uses of oat fractions and hence the current study is highly complementary by considering food uses. It is hoped that the combination of Oatec and Innovation will attract suitable investment for the establishment of an oat fractionation plant in the UK.

Objectives

1. To investigate agronomic and environmental factors affecting grain composition in UK winter oats.

2. To provide a range of samples for milling and fractionation studies in which grain composition has been altered due to the effects of nitrogen, soil water supply and site.

Materials and Methods

Winter oats were grown in experimental field plots in which the effects of variety, site, ammonium nitrate, foliar urea and soil water supply were examined. These experiments were carried out over a two year period in order to examine the effects of season. Details are given below regarding the treatments and assessments carried out in the study. Further detail, if required, can be obtained upon request from ADAS.

Sites

Both sites in the study were at ADAS Research Centres, details of which are shown in Table 3. These sites were chosen to represent the extremes of soil type in terms of water-holding properties. Previous experience had shown that the soil at Rosemaund had a high capacity for water retention, while that at Gleadthorpe was prone to drought due to the high drainage properties of the light sandy soil at this site.

Table 3. Site details

Site	Location	Soil type
Rosemaund	Preston Wynne, Herefordshire	Silty clay loam
Gleadthorpe	Mansfield, Nottinghamshire	Loamy medium sand

Agronomic treatments

Agronomic factors examined in the field experiments are shown in Table 4. Four replicates of each treatment in a full factorial design were grown at each site. All farm operations were as standard practice for winter oats.

Table 4. Experimental treatments examined in field experiments

Expt. no.	Site	Variety	Irrigation	Ammonium nitrate	Foliar urea
1	Rosemaund	Gerald	None GS 31-61 GS 61-87	0 kg/ha 40 kg/ha 100 kg/ha	0 kg/ha 60 kg/ha
2	Rosemaund	Image	None	0 kg/ha 40 kg/ha 100 kg/ha	0 kg/ha 60 kg/ha
3	Rosemaund	Various	None	80 kg/ha	None
4	Gleadthorpe	Gerald	None	40 kg/ha 100 kg/ha 140 kg/ha	0 kg/ha 60 kg/ha
5	Gleadthorpe	Gerald	None GS 31-61 GS 61-87	100 kg/ha	None

Ammonium nitrate
Ammonium nitrate was applied by hand at both sites; it was applied as a split application with 40 kg/ha at GS 31 and the remainder at GS 32.

Foliar urea
Foliar urea was applied as a spray at GS 61. This was applied in the evening or in overcast weather (to prevent leaf damage) as a single application in a total volume of 300 litres in water.

Control of soil water availability
At Rosemaund irrigation was applied using the 'trickle' tape system in both years. This was also used at Gleadthorpe in Year 1 but overhead irrigation from a 'linear move' boom was used in Year 2. Soil moisture deficit (SMD) was monitored using the ADAS Irriguide model for each site (Bailey and Spackman, 1996; Bailey *et al.*, 1996). Guideline targets for application of irrigation were followed as closely as possible within the practical limits of the experiments. The target SMD above which irrigation was applied was 50 mm and the target amount applied to the plots was 25 mm per application.

Experimental design

This was as shown in Table 5. Due to the practicalities involved in operating the irrigation system it was necessary to 'block' the irrigation treatments as main plots within Experiment 1. Replicates were arranged in four blocks in the field.

Table 5. Experimental designs used within the field experiments

Expt.	Design
1	A split plot design with irrigation as a main plot treatment and then a fully randomised 3 x 2 factorial within the main plots for the nitrogen treatments.
2	A 3 x 2 factorial design with the treatments fully randomised
3	Fully randomised
4	A 3 x 2 factorial design with the treatments fully randomised
5	Fully randomised

Assessments and data collected

Meteorological data were collected from the ADAS meteorological stations at each site. Details of solar radiation (kW m^{-2}), maximum, minimum and mean temperature (°C), rainfall (mm), wind speed (m s^{-1}) and relative humidity were measured. This information was used in conjunction with details of the crop to calculate SMD values for scheduling the irrigation treatments. Incident, reflected and absorbed photosynthetically active radiation were also measured in the crop at Rosemaund in Experiments 1 and 2 using a Ceptometer.

Soil mineral nitrogen was measured in the spring at each site. One sample was taken per replicate of six soil cores bulked from 0-30, 30-60, and 60-90 cm horizons. Plant manganese was measured in the spring to check for possible manganese deficiency.

Growth was assessed using the decimal code of Tottman and Broad (1987). This was monitored to standardise the timing of treatments at the two sites. A standard protocol was used for crop sampling to ensure that a representative sample was taken from the crop. This consisted of taking a rectangular quadrat 1.0 m in length from the central six rows of the plot. This was then sub-sampled for analysis in the laboratory. Where necessary a further sub-sample was immediately dried in a forced-air oven.

Crop growth was monitored as shoot numbers, Green Area Index, and biomass at GS 61+75°C days (hereafter approximated and referred to as GS 65) and at mid-senescence. Stem carbohydrate reserves were measured in dried sub-samples at both of these growth stages.

Crop height was measured across plots and lodging was assessed as a percentage of the crop lodged compared to the area of the whole plot.

Grain chemical composition (β-glucan, protein and oil) was analysed at ADAS Laboratories. β-glucan was measured using a modification of the McLeary enzymic assay (McCleary and Mugford, 1997) in which oil was extracted and the defatted sample ball-milled prior to enzymic hydrolysis. Protein was estimated as N x 5.83, with nitrogen content measured using the Dumas procedure. Oil was measured gravimetrically following soxhlet extraction with petroleum ether. Full details of all analytical methods are available from ADAS Laboratories.

Results

Full tables of results are shown in the Appendix. Each individual data entry in these tables is the mean of four replicate field plots with each plot being analysed as an independent sample and in random order. Statistical values are also shown in the Appendix and in the following discussion of the results, treatments are referred to as 'different' only when the effect is significant at the 5% level.

In the following discussion ammonium nitrate treatments are referred to as 'low', 'medium' and 'high', representing the three levels applied in the study at each site (Rosemaund - 0, 40, 100 kg N/ha; Gleadthorpe - 40, 100, 140 kg N/ha). These differences were necessary due to the differential fertility of the soil at each site.

Weather

Graphs of the monthly rainfall, sunshine hours and air temperature are shown in Figures 1 to 4. Only rainfall is presented from the Gleadthorpe site since other data were not available. The weather during the study showed an abnormal pattern compared to the long term trends and the data in Figures 1 to 4 reveal a number of differences between the years of the study. The Rosemaund site in Year 1 was characterised by a relatively dry spring (March/April) followed by high June rainfall and then dry July (Figure 1). This created low soil moisture levels prior to grain filling but then the significant rainfall at the onset of grain filling ensured that sufficient water was available for normal crop growth throughout the rest of the season. A similar pattern was also found at Gleadthorpe in this year (Figure 4). As a result of the rainfall pattern much irrigation was applied in the 'early' phases, with less in the 'late' phase, particularly at Gleadthorpe. The consequence of this was that the most significant effects were noted with the 'early' irrigation treatment. The pattern of weather was again unusual in Year 2, with low rainfall in February followed by very wet April, dry May, wet June and then dry July and August. Again, the pattern of weather was similar at the two sites except that July and August were wetter at Gleadthorpe than at Rosemaund (though the rainfall was still below the long term mean). This pattern of weather was considerably different from the long term means and made scheduling of the irrigation treatments difficult. The consequence was that increasing soil water availability at Rosemaund had little or no impact on the growth of the crop, grain yield or quality. At Gleadthorpe, greater impact of soil water availability on crop growth was noted, due to the light soil type, particularly irrigation during the 'early' phase in Year1. One of the consequences of the high rainfall at Rosemaund in Year 2 was that the crop was heavily infected with Crown Rust, with the consequent reduction in grain yield and quality. There was no Crown Rust infection at Gleadthorpe.

Monthly mean temperatures (Figure 2) at Rosemaund were close to normal but there were unusual patterns of monthly sunshine (Figure 3). The most significant difference from the long term mean was that very low sunshine hours were recorded for June in both years and they were also very low in July in Year 2. Low levels of sunlight would be expected to result in delayed harvest due to slower grain maturation with lower grain yield than normal and both of these effects were noted in the study.

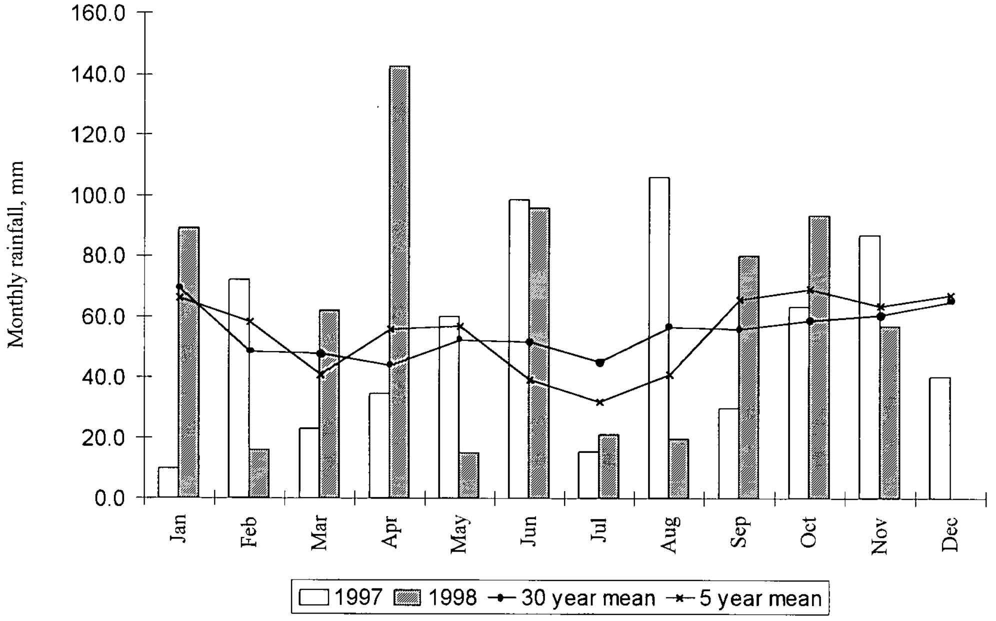

Figure 1. Monthly rainfall at Rosemaund in 1997 (Year 1) and 1998 (Year 2)

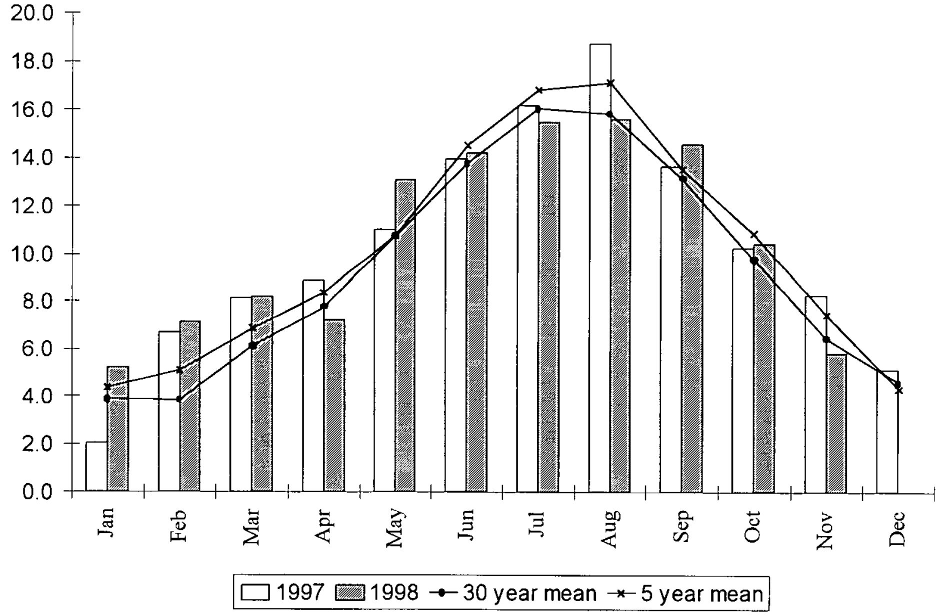

Figure 2. Mean monthly temperature at Rosemaund in 1997 (Year 1) and 1998 (Year 2)

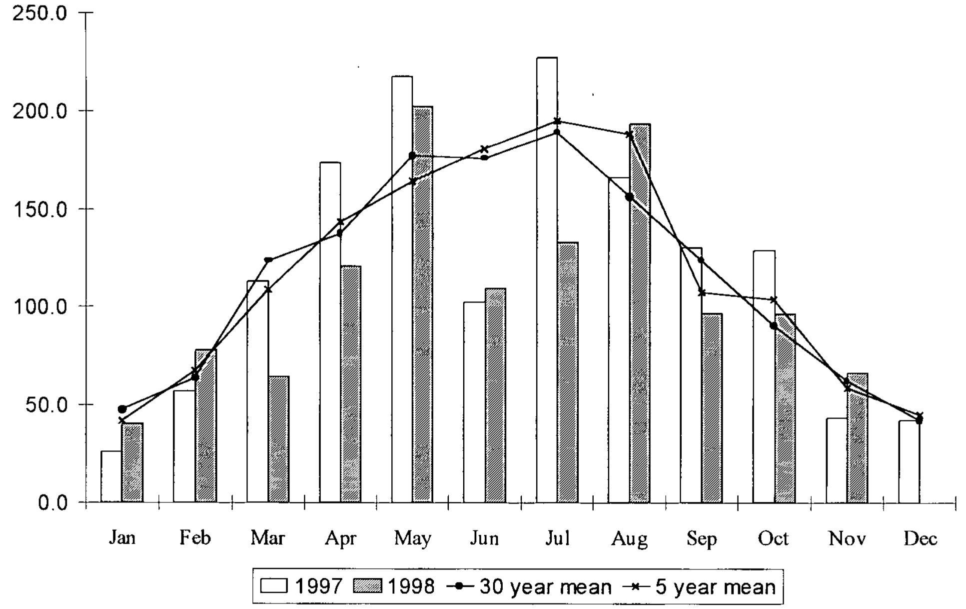

Figure 3. Monthly sunshine hours at Rosemaund in 1997 (Year 1) and 1998 (Year 2)

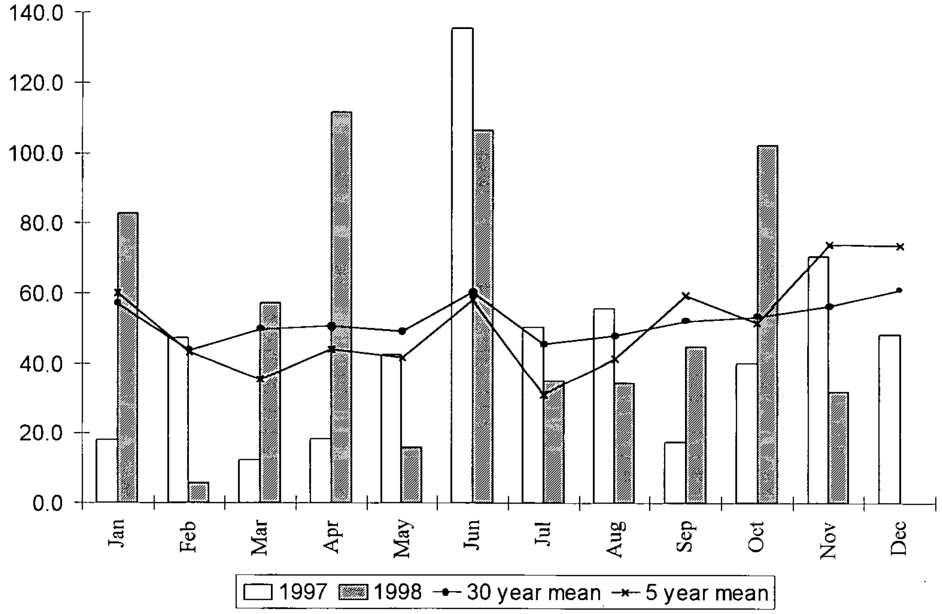

Figure 4. Monthly rainfall at Gleadthorpe in 1997 (Year 1) and 1998 (Year 2)

Control of soil water availability

The weather data were used in combination with crop growth parameters to monitor soil moisture deficits throughout the season via the Irriguide model. This enabled scheduling of the frequency of irrigation applied in the experiments. Irrigation applied at each site is shown in Figures 5 and 6 and the effects of the applied irrigation on soil moisture deficits are shown in Figures 7 to 10. These graphs show that most irrigation was applied at Rosemaund in Year 1. Although less irrigation was applied during the 'early' phase at Gleadthorpe in Year 1 it did have a large impact on grain yield due to the rapid loss of water from the control plots. This clearly demonstrated the need to consider the interaction between water-retaining properties of the soil and the irrigation applied. For example, despite applying large amounts of irrigation to the plots receiving the 'late' treatment at Rosemaund, little difference was noted between these plots and the control plots which received no irrigation. This was due to the high water retaining potential of the soil at this site and hence lack of crop stress. It should be noted that a soil moisture deficit of 104 mm is required for crop stress at Rosemaund whereas the corresponding figure for Gleadthorpe is 67 mm (defined as 50% AWC). No 'throughout' treatment is shown at Gleadthorpe in Year 1 since the amounts of irrigation applied were the same as those for the 'late' treatment. In Year 2 much irrigation was applied to the 'late' treatments at both sites. However, due to the carryover effect of high April and June rainfall the timing of this irrigation was too late in the season to significantly affect crop growth. Despite these conditions the soil moisture deficits were maintained broadly in line with the original objectives.

Overall, the impact of soil water supply, manipulated using field irrigation, was small in terms of the effects on grain yield and quality due to the lack of crop stress experienced during the course of the study. In future studies automated crop shelters would ensure that crop water stress is achieved, even in seasons in which high rainfall occurs.

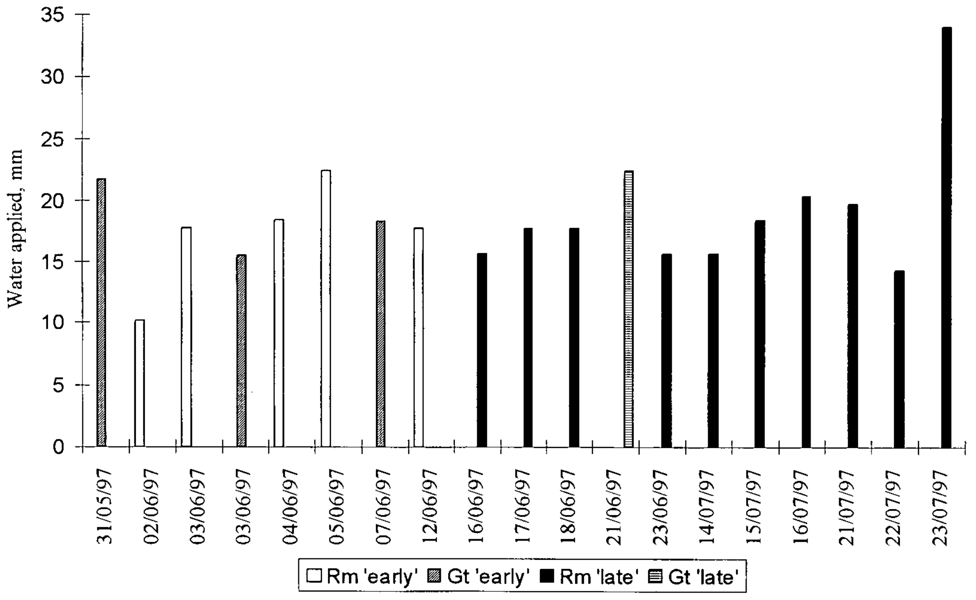

Figure 5. Irrigation applied in 1997 (Year 1)

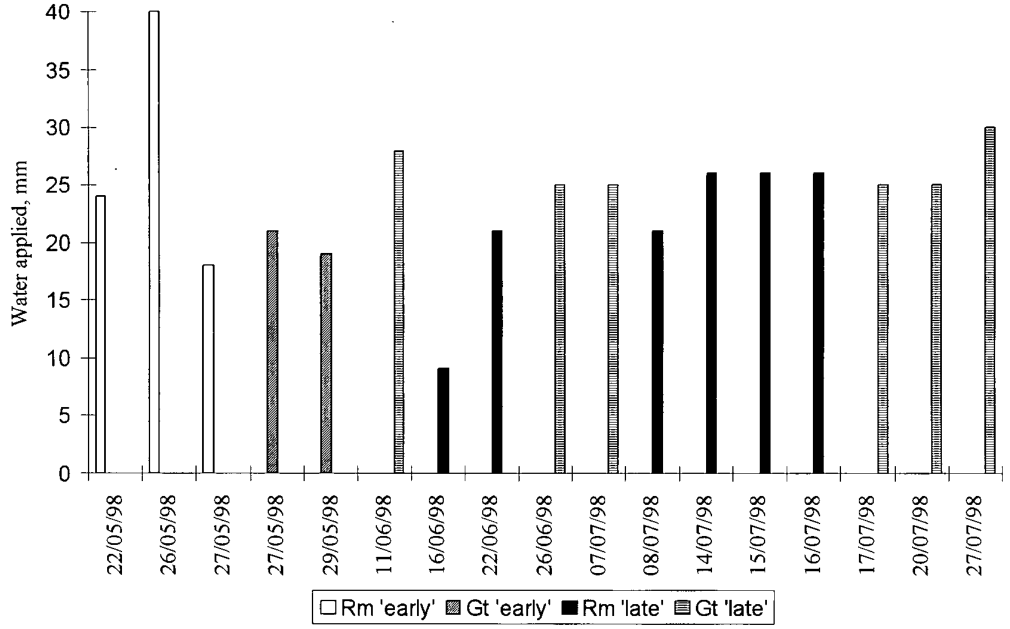

Figure 6. Irrigation applied in 1998 (Year 2)

Soil mineral nitrogen

Values for soil mineral nitrogen for each horizon are shown in Table 6. These data showed that more nitrogen was available to the crop in Gleadthorpe in Year 2 than Year 1, though at Rosemaund similar amounts were available each year.

Table 6. Soil mineral nitrogen values.

Site	Depth cm	Year 1	Year 2
Rosemaund	0 - 30	37.6	37.2
	30 - 60	15.6	28.8
	60 - 90	18.8	10.4
	Total	72.0	76.4
Gleadthorpe	0 - 30	22.0	27.5
	30 - 60	8.0	18.7
	60 - 90	7.0	30.1
	Total	37.0	76.3

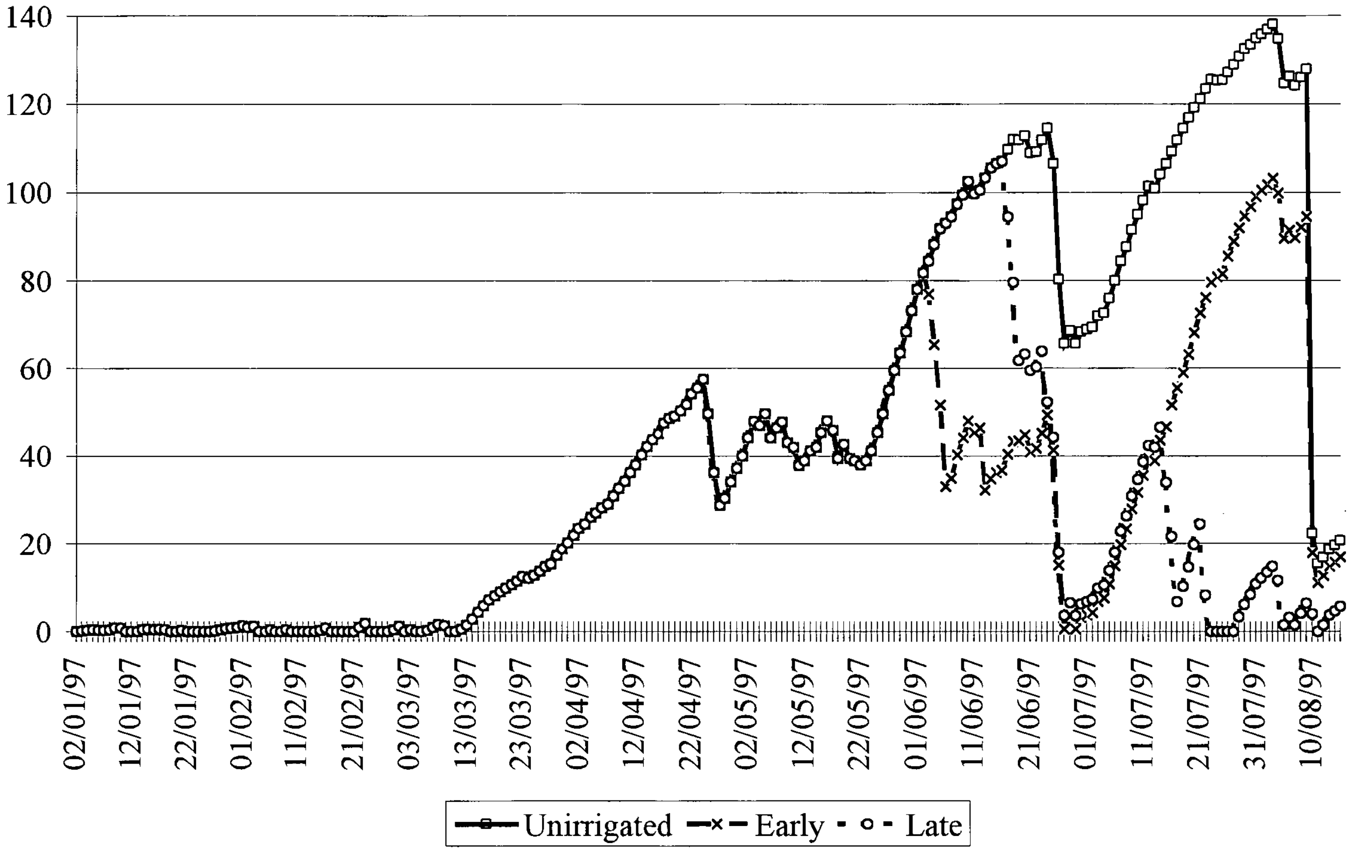

Figure 7. Soil moisture deficits (mm) at Rosemaund in 1997 (Year 1)

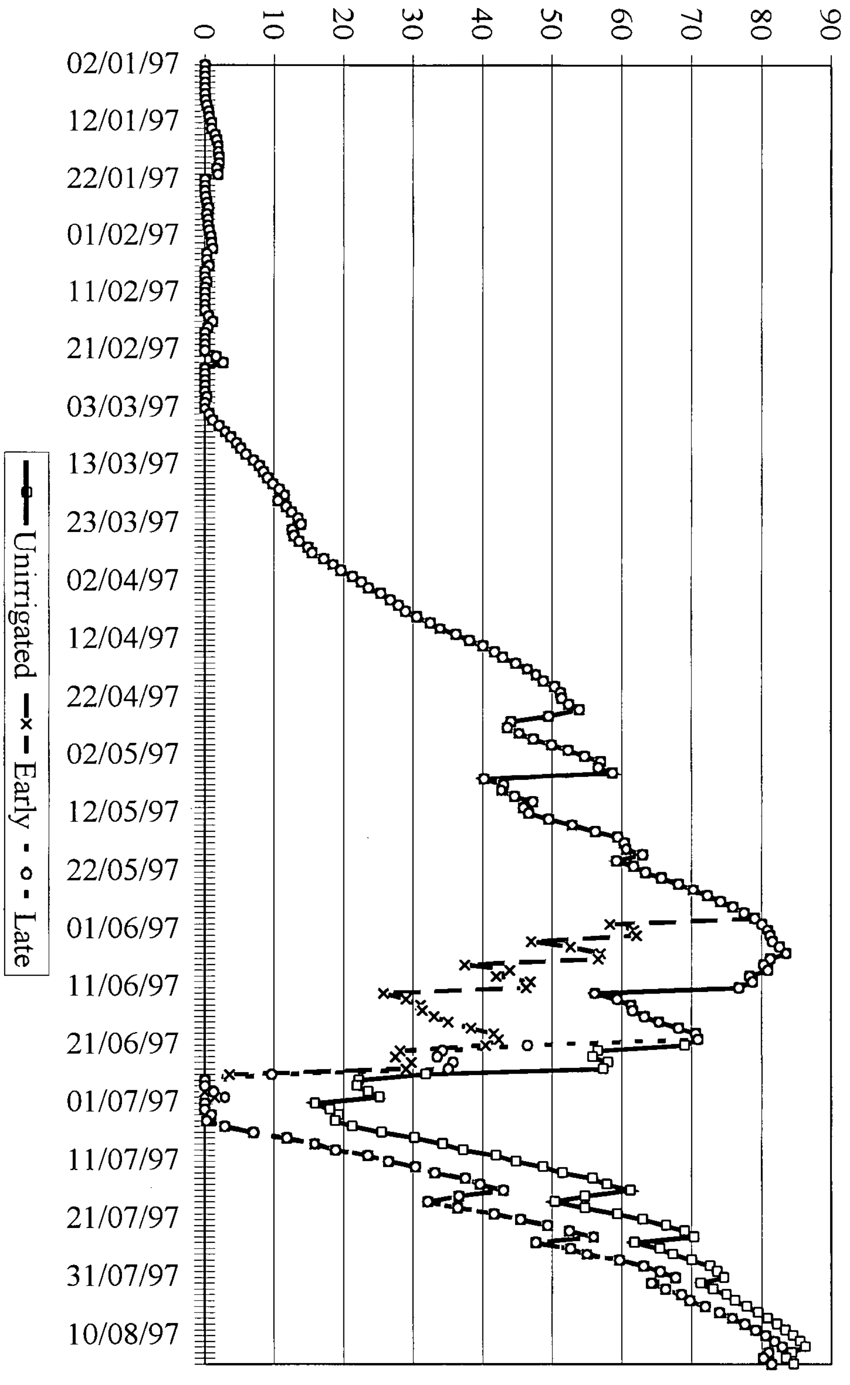

Figure 8. Soil moisture deficits (mm) at Gleadthorpe in 1997 (Year 1)

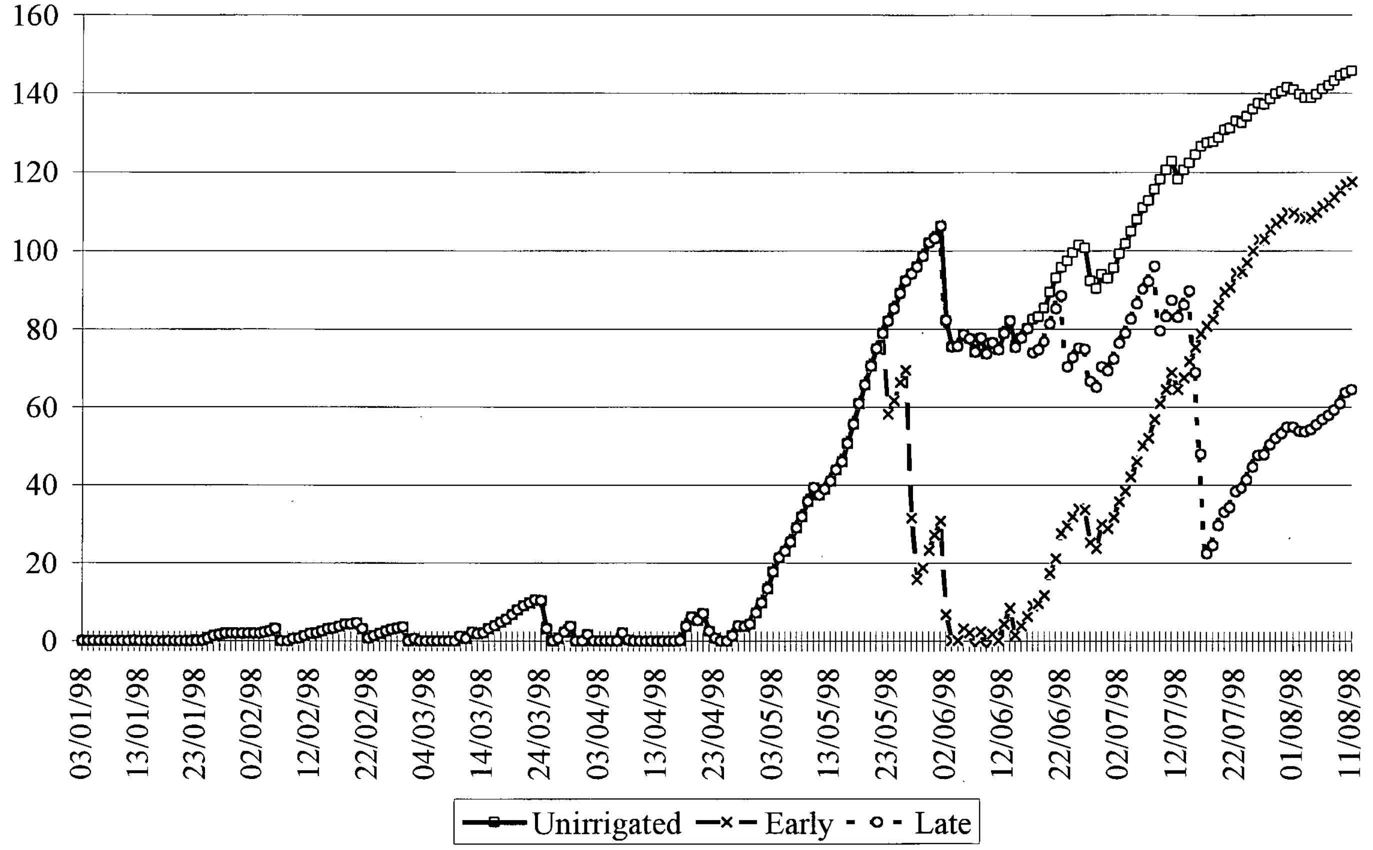

Figure 9. Soil moisture deficits (mm) at Rosemaund in 1998 (Year 2)

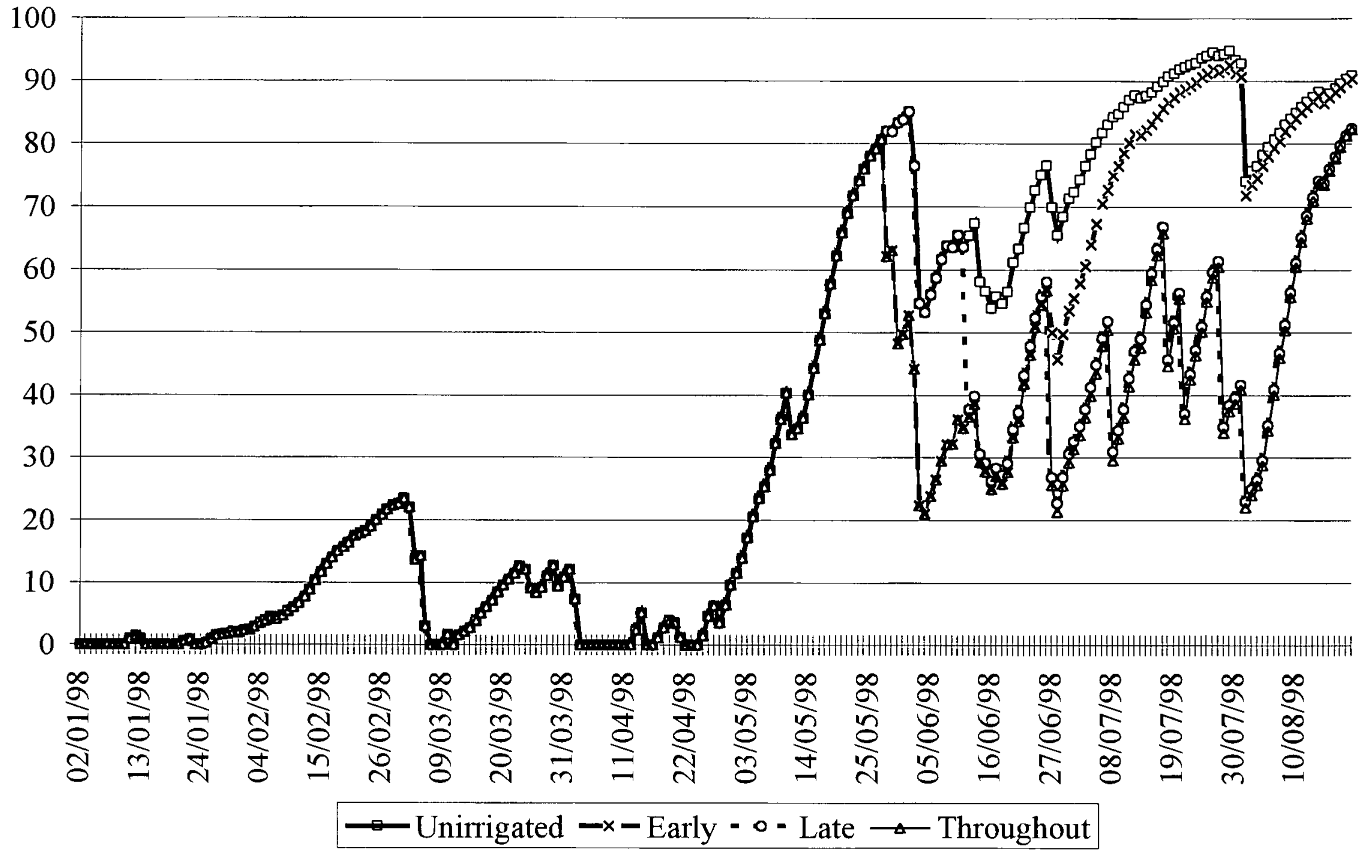

Figure 10. Soil moisture deficits at Gleadthorpe in 1998 (Year 2)

Nitrogen uptake

The amounts of nitrogen taken up by the crop each year are summarised in Figures 11 to 13. Full data are given in Tables A1 to A4. The data showed that considerably more nitrogen was taken up than would have been expected based on the amounts of mineral nitrogen available in the soil measured early in the season (Table 6). It is widely believed that oats are efficient at scavenging nitrogen from the soil and there are a number of explanations for this:

- Further mineralisation of organic nitrogen throughout the season.
- Uptake of nitrogen from deeper regions of the soil.
- Some nitrogen was present in the crop (which was at approximately GS 30) at the time when SMN was measured.

Further indication of the efficiency with which the crop was able to take up nitrogen was that uptake increased with applied ammonium nitrate, even though high levels of nitrogen had already been taken up by the crop. Efficiency of nitrogen uptake was also high when applied as foliar urea, and was on a par with the efficiency of uptake when applied as ammonium nitrate. However, no effect was noted of water supply on nitrogen uptake.

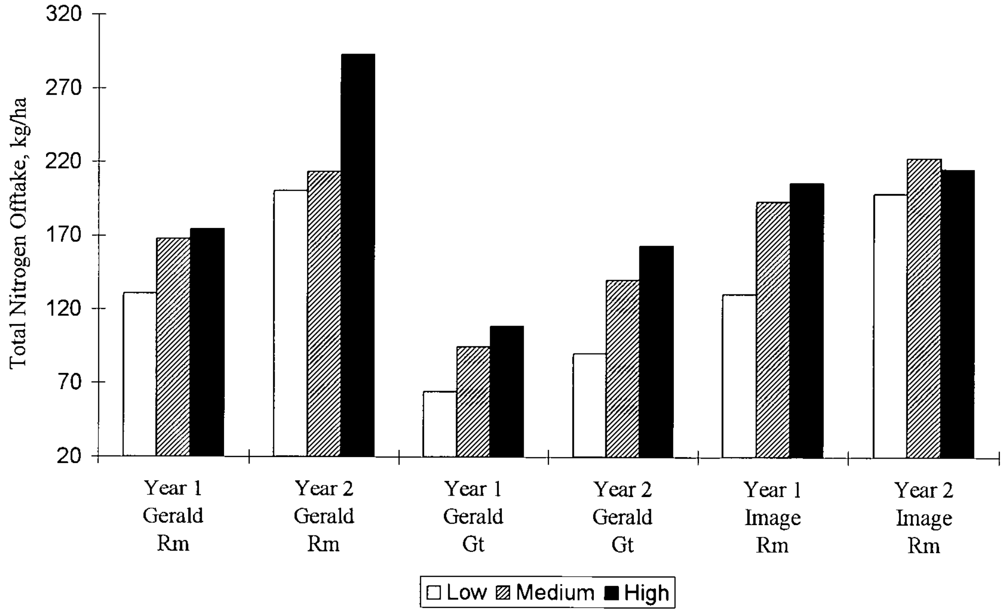

Figure 11. Effects of ammonium nitrate (Low, Medium, High) on Total Nitrogen Offtake.

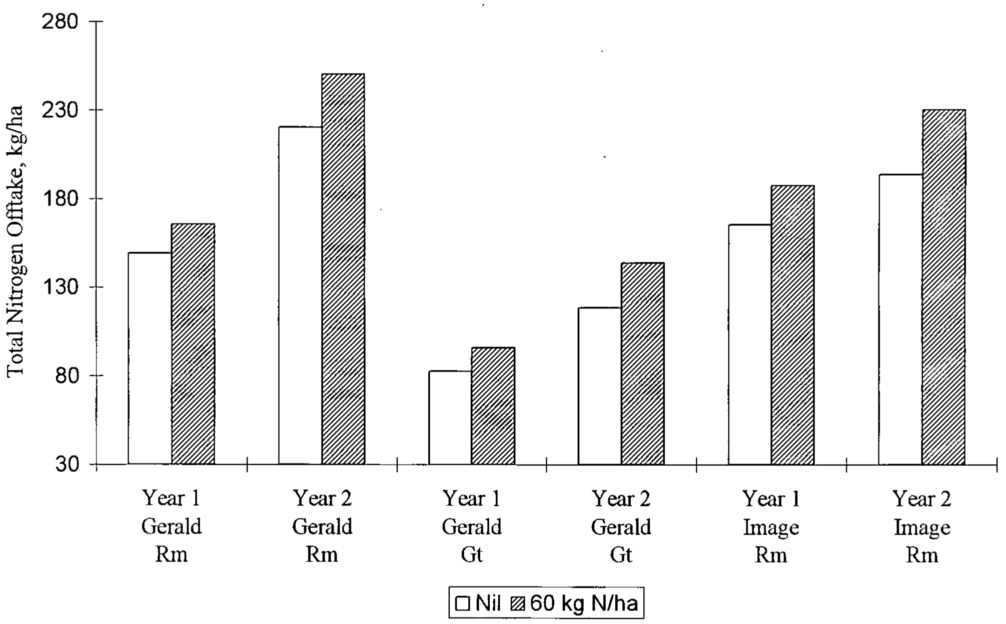

Figure 12. Effects of foliar urea on Total Nitrogen Offtake.

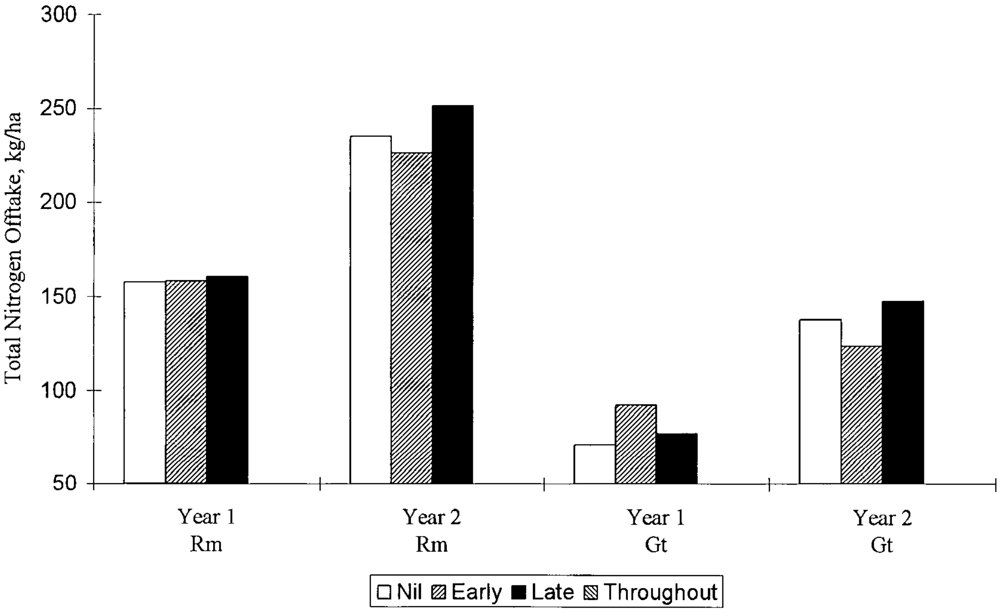

Figure 13. Effects of soil water supply (Nil, Early and Late irrigation) on Total Nitrogen Offtake.

Crop assessments

In order to understand how the agronomic inputs had affected grain yield and quality the study undertook to examine the growth and development of the crop. The rationale follows the recent HGCA funded projects carried out on wheat in which changes in crop growth are monitored through measurable aspects of crop structure relating to photosynthetic capacity and yield potential. Considerable research has been carried out in this area on wheat so comparisons with wheat are made where this is relevant. The underlying hypothesis concerning canopy development and yield is that increased nitrogen uptake leads to greater numbers of tillers which produces higher Green Area Index; this allows more interception of solar radiation which produces more biomass and hence higher yield. However, this relationship is not linear, with the response decreasing as canopy size increases; in wheat, little or no increase in yield is seen above a GAI of 6. The effects of late nitrogen such as foliar urea would be expected to influence yield by increasing the duration of canopy survival since the leaves would contain higher levels of nitrogen and are thus able to maintain photosynthesis for longer, since the developing grain removes nitrogen from the leaves. The use of irrigation to increase soil moisture supply would prolong grain filling by delaying senescence brought about by drought.

Assessments were carried out at two stages during development, GS 61+75°days (hereafter approximated and referred to as GS 65) and at mid-senescence. The rationale for sampling at these stages is that GS 65 is the stage at which Green Area Index (GAI) and stem carbohydrate reserves are close to maximal. At mid-senescence the crop is still able to capture photosynthetically active radiation though the potential for this is decreasing rapidly; prolonging green area at this stage increases potential yield through increasing the rate and duration of grain filling. Measuring GAI at these developmental stages gives an indication of the effectiveness of the agronomic treatments at maximising yield potential through increasing 'source'. Measurements of shoot number give an indication of potential for higher yield through increased 'sink'. A further aspect of importance for yield is the establishment of an efficient canopy supported by stems and roots sufficiently strong to resist lodging. This was measured by monitoring the area of crop which lodged through the season.

Full results of the crop assessments and statistical analysis of the effects of the treatments are given in the Appendix (Tables A1 to A30).

Shoot numbers

Tillering is one of the most important processes affecting canopy formation and potential grain yield. Maximum tiller number is reached in the spring but frequently more tillers are produced than are able to survive to form panicles, due to competition for resources. Application of ammonium nitrate in the spring encourages the survival of tillers resulting in more shoots per unit area with a larger canopy thus enhancing yield. It has been shown in wheat that tiller number accounts for a large proportion of variation in canopy size. However, the survival of too large a number of tillers can also have negative effects since high shoot numbers competing for available reserves can give rise to weak stems which are prone to lodging. A balance must therefore be achieved between increase in canopy size and lodging risk. Increased tillering has also been shown to be beneficial in buffering against unfavourable environmental conditions in spring oats (Lawes, 1977). Soil nitrogen supply has been shown to be one of the most important factors affecting tillering in oats and where there are low nitrogen reserves tillering stops earlier in the season (Chandler, 1969).

Full results for assessment of shoot numbers are given in Tables A5 to A8 (Appendix). Large effects of ammonium nitrate were noted in fertile shoot numbers at GS 65 and at mid-senescence at both sites although there were differences between the two years of the study (Figures 14 and 15). In Year 1 an increase in ammonium nitrate from 'low' to 'medium' resulted in increased numbers of fertile shoots at both sites with no additional change in fertile shoot numbers when ammonium nitrate was increased further to the 'high' level. The results in Year 2 were different in that no overall effect of ammonium nitrate on fertile shoot numbers was seen at Rosemaund; this contrasted sharply with the result at Gleadthorpe where a stepwise increase in shoot numbers was found with increasing ammonium nitrate. It was clear that shoot numbers were influenced by available soil nitrogen in both years. The difference between the two years of study at Gleadthorpe was likely to be due to the lower soil moisture levels prior to anthesis in Year 1 which limited the survival of tillers at this stage. The total nitrogen uptake in 'Low' nitrogen plots at Rosemaund in Year 2 (Figure 11) was on a par with levels in other sites and years where high nitrogen was applied; thus there was sufficient nitrogen available to the crop and this was the reason for the lack of response seen to applied ammonium nitrate in the experiments.

Large differences in fertile shoot numbers at GS 65 and mid-senescence were noted between both sites and seasons (Figures 14 to 16). Variation in fertile shoot number was not adequately accounted for by mineral nitrogen reserves and indicated that they were influenced by a number of factors present. This was particularly noticeable in variety Image grown at Rosemaund; much lower shoot numbers resulted in Year 2 at 'medium' and 'high' ammonium nitrate levels compared to 1997. The reasons for these differences are not clear from the data presented. A noticeable feature of variety Image was the high numbers of fertile shoots produced compared to Gerald, which was possibly one reason for the greater susceptibility of Image to lodging. Some differences between sites were not easy to explain. For example, higher shoot numbers were recorded at Gleadthorpe than at Rosemaund in Gerald in Year 1, despite the lower soil moisture availability at the Gleadthorpe site in this year.

Higher numbers of fertile shoots may have been expected to result in a larger number of smaller grains through an increase in the 'sink' capacity of the crop. This was evidenced by a reduction in specific weight and thousand grain weight with increased ammonium nitrate levels. It may be expected that this would affect grain composition through different proportions of grain components. However, there was no evidence for this within the study since other than protein content there was no correlation between shoot number and grain quality in terms of β-glucan or oil content.

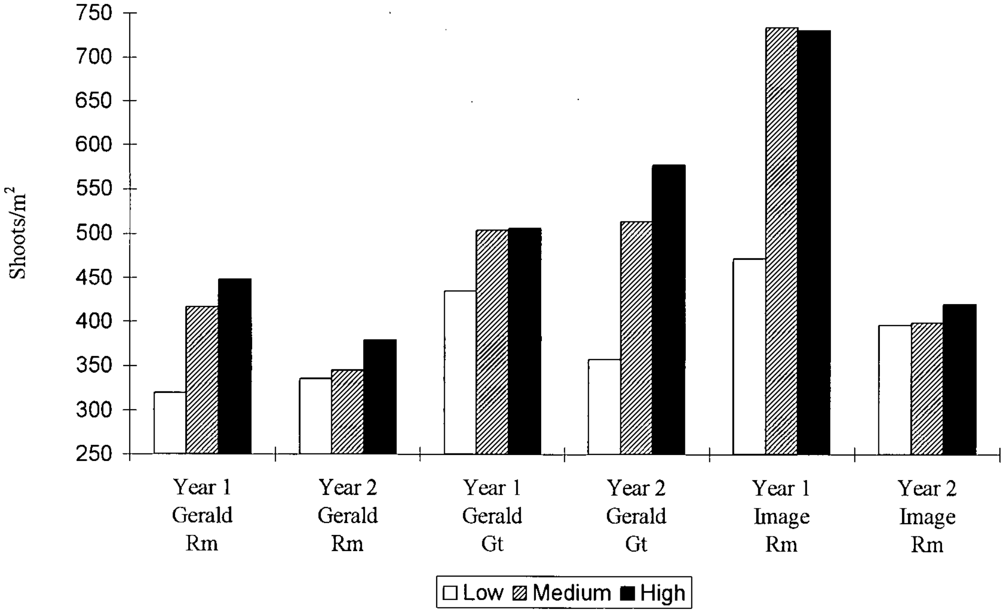

Figure 14. Effects of ammonium nitrate (Low, Medium, High) on fertile shoot numbers at GS 65.

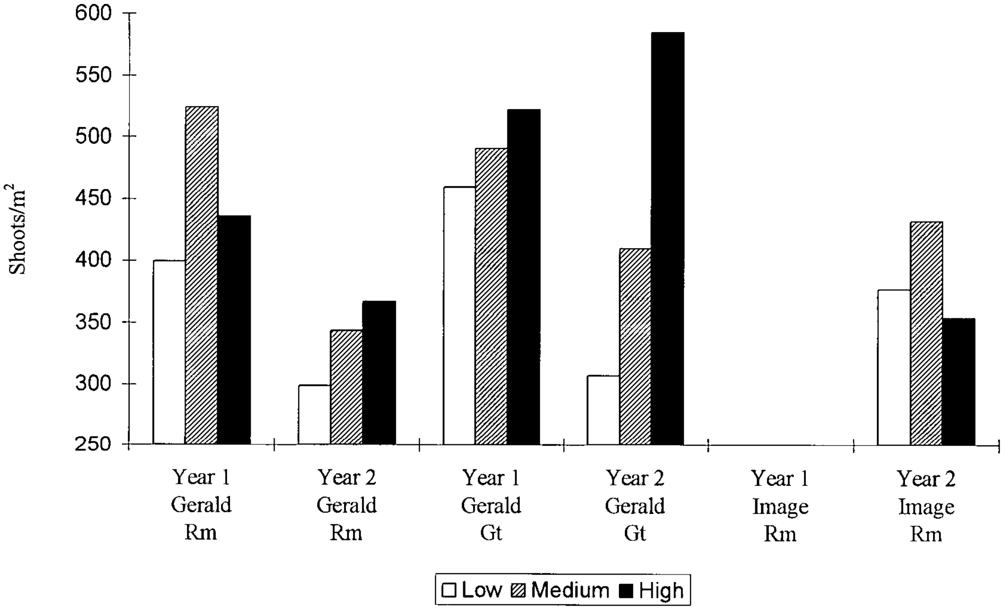

Figure 15. Effects of ammonium nitrate (Low, Medium, High) on fertile shoot numbers at mid-senescence.

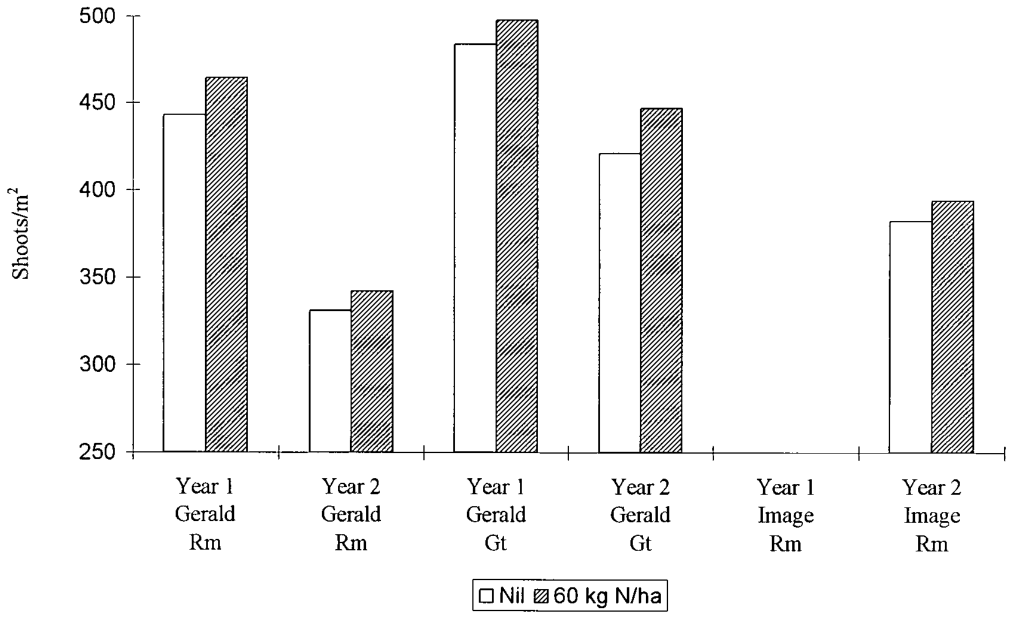

Figure 16. Effects of foliar urea on fertile shoot numbers at mid-senescence.

Canopy size

Canopy size measured as Green Area Index (GAI – the ratio of projected planar area of green material to ground area) is a key determinant of yield potential. Size of the canopy indicates the ability of the crop to intercept incident radiation. Increased GAI results from increases in fertile shoot numbers and can be manipulated by applying larger amounts of nitrogen as ammonium nitrate. In addition to canopy size, survival of the canopy is also important. An attempt was made in the study to manipulate this by modifying soil water supply and applying additional nitrogen as foliar urea during the period following anthesis.

GAI was measured separately for leaves, stems and ears since all three crop components contribute to grain filling. Total GAI is thus the sum of these components and represents the overall capacity of the crop to trap sunlight for photosynthesis. This is important since a significant amount of photosynthesis occurs in the flag leaf and the ear late in grain development. Full results of the assessments of the effects on GAI are given in Tables A9 to A12.

Increases in ammonium nitrate resulted in higher Total GAI at GS 65, mainly as a result of greater shoot numbers at higher ammonium nitrate levels, giving increases in leaf and stem green areas (Figure 17). Higher GAI would be expected to result in higher yield through greater capacity for photosynthesis, though little effect would be likely above a GAI of 6 due to the effects of shading (assuming a parallel with wheat). An increase in yield is also possible through greater duration of green area at higher nitrogen levels. This effect was noted at both sites (Figure 18) though not in Year 2 at Rosemaund due to the effects of Crown Rust infection.

There was strong evidence of interactions between the effects of ammonium nitrate and other factors on GAI. In Year 1 at Rosemaund there was a much greater increase in GAI from 'low' to 'medium' than from 'medium' to 'high' ammonium nitrate whereas in Year 2 there were similar increases at both levels. In Year 1 at Gleadthorpe the magnitude of the increases in GAI with applied ammonium nitrate was similar at all levels but in Year 2 a greater increase was seen at the lower level. A possible reason for this is that the availability of the applied ammonium nitrate and its uptake by the crop were affected by environmental factors. There was considerably higher Total GAI at Gleadthorpe in Year 2 compared to Year 1, which may have resulted from higher rainfall during the canopy expansion phase or possibly higher soil mineral nitrogen.

At Rosemaund in Year 2 a significant reduction in Total GAI resulted from a Crown Rust infection; this was noted for leaf but not stem or ear GAI. The result was that Total GAI for Year 2 was lower than that for Year 1 at this site.

A varietal comparison between Gerald and Image was possible at Rosemaund and indicated that there were differences in Total GAI at GS 65 in both years (Figure 17). In Year 1 similar Total GAI was found in both varieties at 0 kg N/ha but Image had higher GAI at the 40 and 100 kg N/ha levels. This was largely as a result of greater tillering in Image compared to Gerald, though would not be expected to have a large influence on yield since Total GAI was high at both sites. No comparison was possible in Year 2 since the greater susceptibility of Image to Crown Rust resulted in lower Total GAI at all levels of ammonium nitrate addition.

Effects of soil water supply on GAI at GS 65 are shown in Figure 19. No significant effect of soil water supply was noted in GAI at Rosemaund in Year 1 and only small effects in Year 2. At Gleadthorpe 'early' irrigation resulted in increased GAI (all components) in Year 1 but not Year 2, due to the higher rainfall in the latter year.

GAI was measured at mid-senescence in order to determine the extent to which the treatments had delayed senescence and thus maintained the capacity of the crop to photosynthesise. No measurements of GAI were made in variety Image at Rosemaund in Year 1 due to severe lodging within these plots before the assessments were due to be carried out. A significant increase in Total GAI at mid-senescence was found with higher ammonium nitrate application in both years at Gleadthorpe and in Year 1 at Rosemaund in variety Gerald (Figure 18). In Year 2 at Rosemaund this effect was not found, possibly due to the Crown Rust infection. Senescence at Gleadthorpe was more rapid in Year 2 than Year 1 due to the drier weather during the main grain-filling period whereas heavy rainfall was noted in Year 1 during early grain-filling. At this stage the relative contributions of leaf, stem and ear to radiation interception had changed, with the ear contributing significantly more than the stem or leaves.

There was significantly higher Total GAI at mid-senescence at Rosemaund than Gleadthorpe in both years, despite the Crown Rust infection at the Rosemaund site. No significant effect of soil water supply was found in Year 2 at Rosemaund but there were highly significant differences at Gleadthorpe in Year 2 (Figure 20). The effects were most noticeable in the 'late' and 'throughout' treatments and since there had been no effect of 'early' irrigation on GAI at GS 65, these effects showed that the treatments successfully delayed senescence in this experiment.

Effects of foliar urea on canopy duration were noted (Figure 21). If it is assumed that shoots are dying at this stage due to removal of nitrogen by the developing grain then the application of foliar urea would be expected to delay senescence. The end result would be to prolong grain filling by maintaining the capacity for photosynthesis. In several instances application of foliar urea resulted higher in Green Area Index at mid-senescence, thus potentially prolonging grain-filling; this was noted in all three components, leaf, stem and ear but was statistically significant only at Gleadthorpe in Year 1 and at Rosemaund (Gerald only) in Year 2. The effect was greater in cases where more green matter remained at the time foliar urea was applied, such as at higher ammonium nitrate levels. This was possibly the reason why no effect was seen at Rosemaund in Year 2 since the Crown Rust infection had caused significant green material to senesce prior to application of the urea. Decisions concerning the use of foliar urea should thus consider the status of the crop at the time at which it is to be applied.

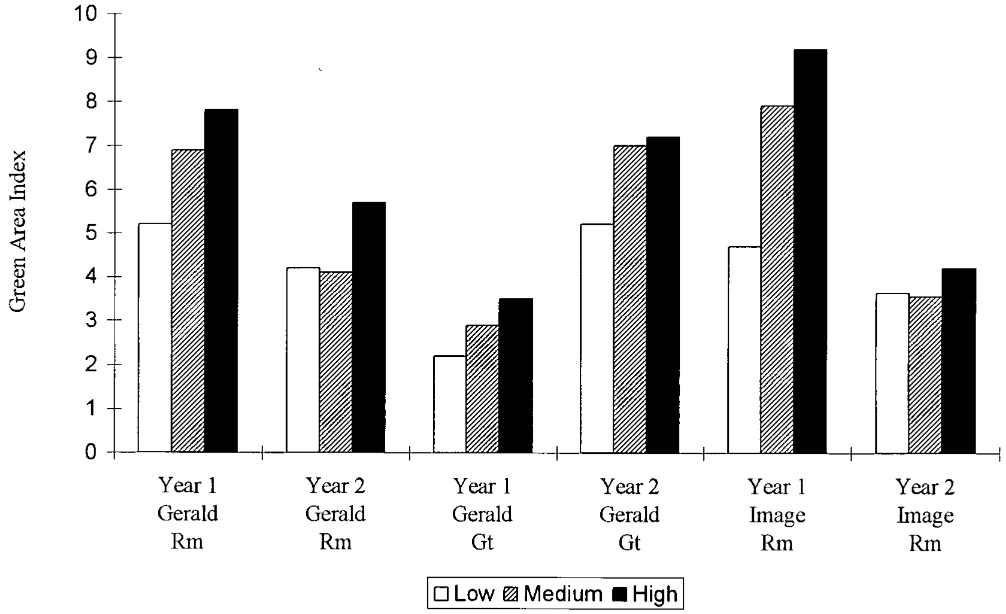

Figure 17. Effects of ammonium nitrate (Low, Medium, High) on Green Area Index at GS 65.

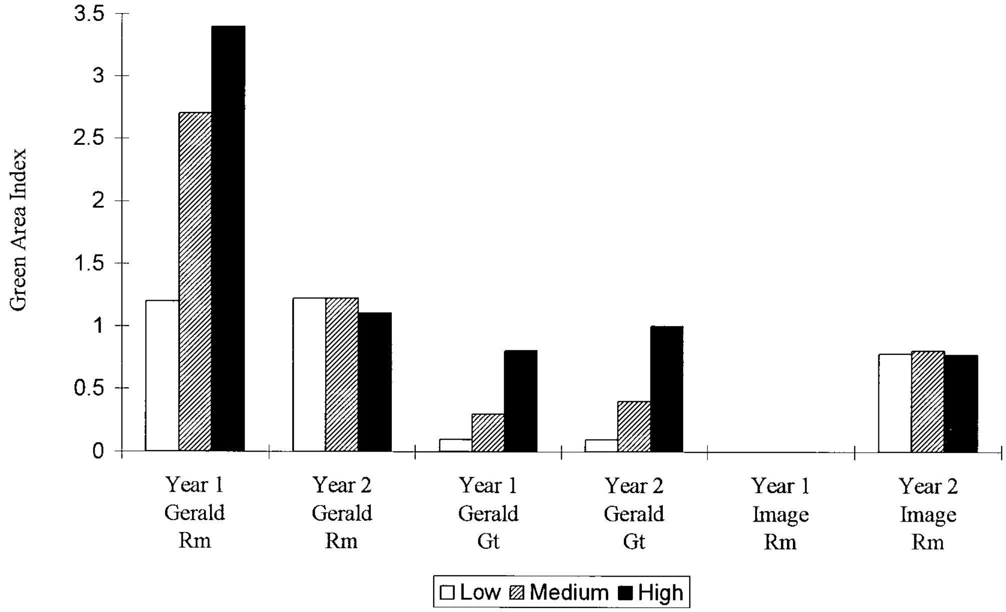

Figure 18. Effects of ammonium nitrate (Low, medium, High) on Green Area Index At mid-senescence.

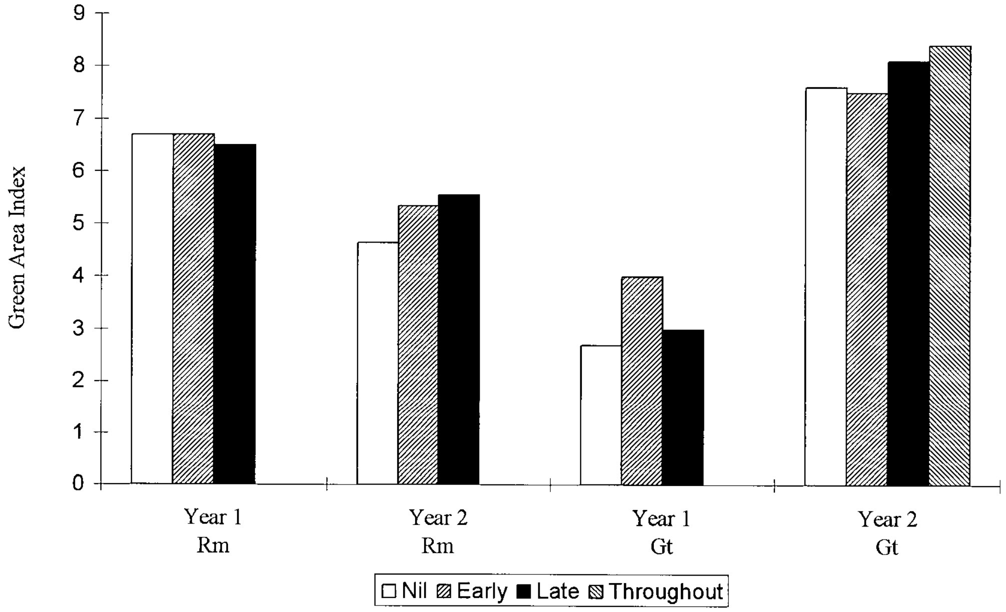

Figure 19. Effects of soil water supply (Nil, Early, late and Throughout irrigation) on Green Area Index at GS 65.

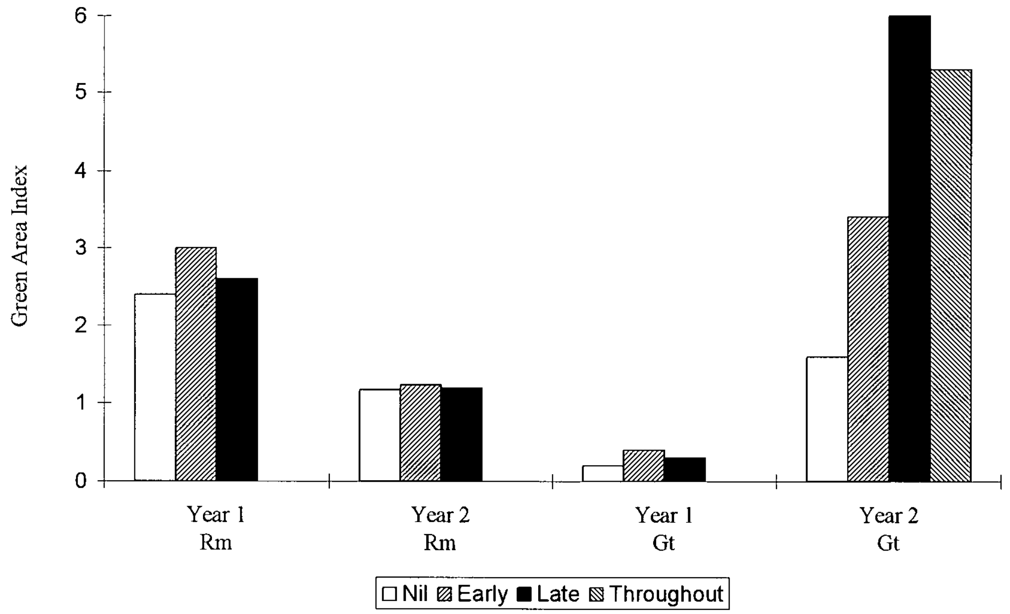

Figure 20. Effects of soil water supply (Nil, Early, Late and Throughout irrigation) on Green Area Index at mid-senescence.

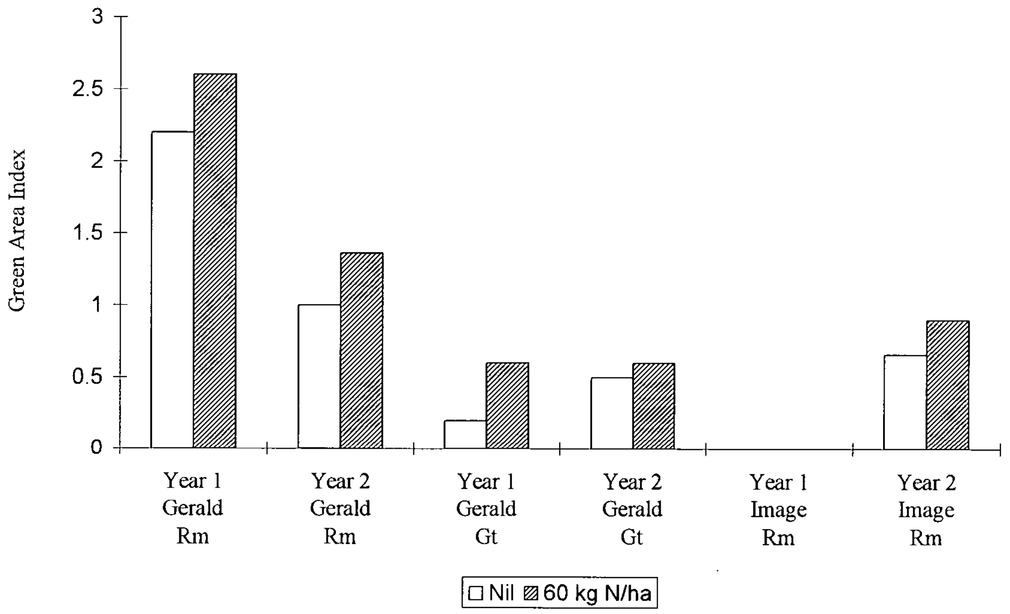

Figure 21. Effects of foliar urea on Green Area Index at mid-senescence.

Light interception

In order to determine the effect of changes in canopy size on the proportion of incident photosynthetically active radiation (PAR) intercepted by the crop, Ceptometer measurements were made at Rosemaund. The results in terms of radiation abosorbed by the crop are given in Figures 22 and 23 with full results given in Tables A13 to A16 (Appendix). Higher ammonium nitrate levels resulted in increases in the amount of PAR intercepted, although the magnitude of the effect was relatively small in Year 1 (Figures 22), as would have been expected by comparison with wheat since GAI was above 5 in all treatments. In Year 2 the differences were larger. The reduction in PAR interception resulting from the smaller canopy, particularly in the absence of applied nitrogen, would have been expected to result in reduced yield. The effects noted were a direct result of increases in GAI as a result of ammonium nitrate application. The largest effects of ammonium nitrate were found in July at both sites, the stage at which the canopy was beginning to senesce. A small, but significant increase in PAR interception was also noted with application of foliar urea, indicating the potential of this input to increase yield, particularly if nitrogen deficiency is expected to curtail canopy survival.

The results of the two years taken together tend to confirm that the optimum canopy size for PAR interception is close to that for wheat, with a canopy of greater than GAI 5 intercepting >90% of PAR in both years.

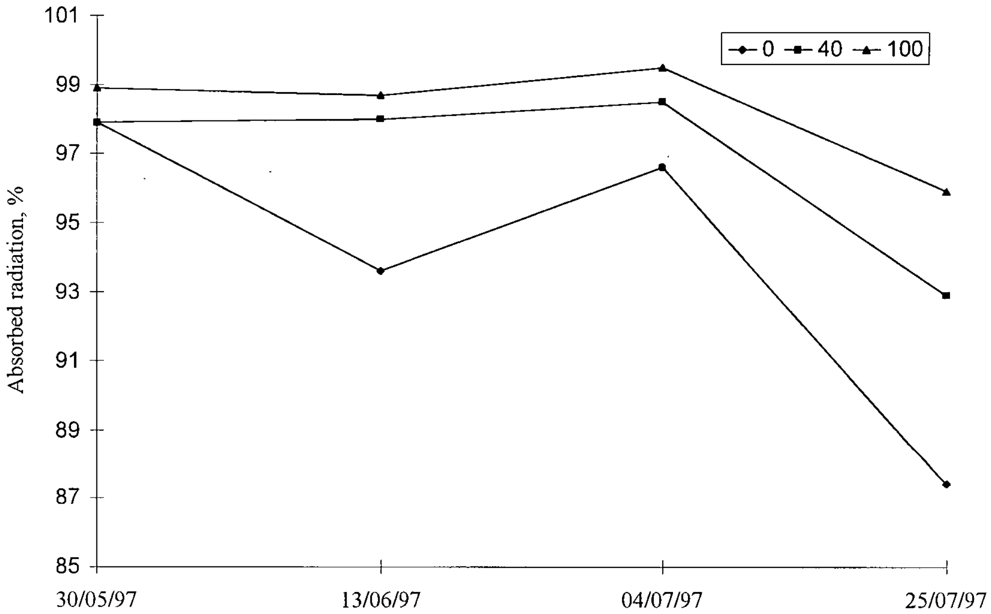

Figure 22. Effects of ammonium nitrate on absorbed radiation at Rosemaund in Year 1.

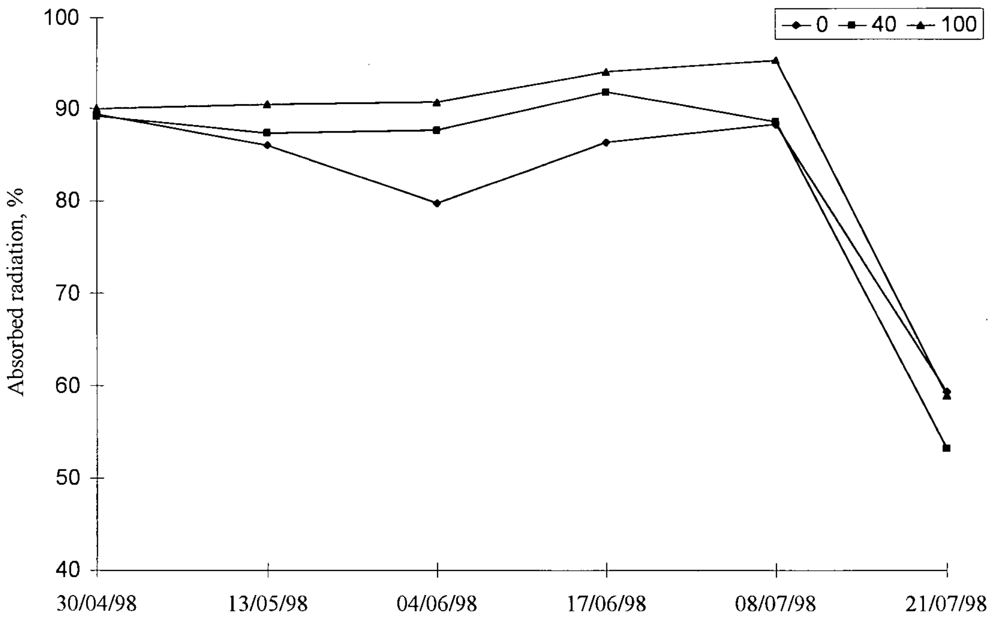

Figure 23. Effects of ammonium nitrate on absorbed radiation at Rosemaund in Year 2.

Crop dry matter

Full results of the assessments of accumulated dry weight (biomass) of the various crop components are shown in the Appendix (Tables A17 to A24). Soil water supply did not significantly affect any of the biomass components at Rosemaund in either year of the study. Effects noted such as increases in leaf GAI would have been expected to have arisen as a result of increased leaf biomass. However, any such increases would have been relatively small in comparison to overall crop mass and little effect is noted in the result tables. 'Early' irrigation at Gleadthorpe in Year 1 resulted in higher leaf, stem and ear biomass, in line with increases in Green Area Index for these components but in Year 2 there was no effect of irrigation at either stage.

Increases in biomass were noted in some experiments as a result of higher ammonium nitrate levels or foliar urea application. These increases were small relative to the overall crop mass and were not consistent throughout all experiments. A large difference in biomass was seen between the two years of the study at Gleadthorpe where much higher biomass was produced in Year 2, possibly as a result of higher rainfall in that year.

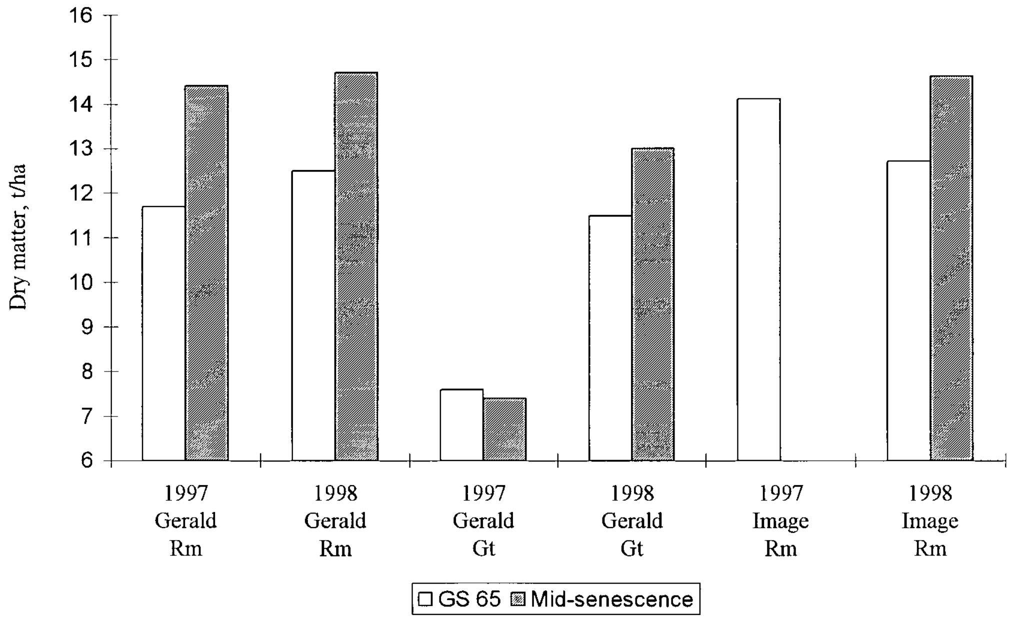

Figure 24. Total crop dry matter at GS 65 and mid-senescence.

Crop height

Crop height was measured in Year 1 at Rosemaund and at both sites in Year 2. Full results are given in Tables A25, A26, A33 and A34 (Appendix). The height of the crop at Rosemaund was significantly increased by application of ammonium nitrate at all levels (Figure 25). Although crop height was not measured at Gleadthorpe in Year 1 it was generally observed that the crop was considerably shorter than in Year 2, possibly as a result of dry conditions during the period of stem elongation. At Gleadthorpe increases in height were only seen from 40 to 100 and not from 100 to 140 kg/ha N. Variety Image was considerably taller than Gerald at Rosemaund in Year 2 (also observed though not measured in Year 1 due to lodging), a factor contributing to the greater propensity to lodging in this variety. The higher fertility and water holding capacity of the soil at Rosemaund compared to Gleadthorpe resulted in higher overall crop height at this site.

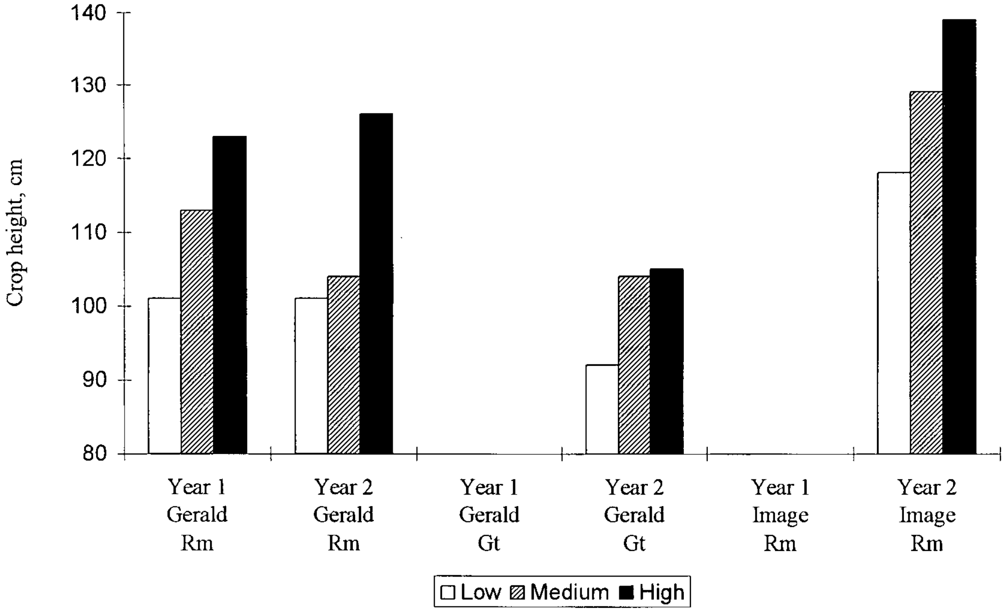

Figure 25. Effects of ammonium nitrate (Low, Medium, High) on final crop height.

Lodging

No lodging was observed at Gleadthorpe in either year of the study even at the highest levels of ammonium nitrate applied. In contrast, significant lodging occurred at Rosemaund in both years of the study. Full results are given in Tables A25 and A26. Lodging was more severe in Image than Gerald, particularly in Year 1 when Image lodged early in the season. This resulted in growth of secondary tillers and a high proportion of small grains in the final samples. No secondary tiller growth was found with Image in Year 2 probably due to the lodging occurring much later in the season. The severity of lodging was greatly influenced by the amount of ammonium nitrate applied (Figure 26) with the severity much greater at higher ammonium nitrate levels in all cases. Nil or very low levels of lodging was found in the 0 and 40 kg N/ha plots of variety Gerald at Rosemaund in either year. The effects of lodging were also exacerbated by 'early' irrigation in Year 1 though no similar effect was seen in Year 2.

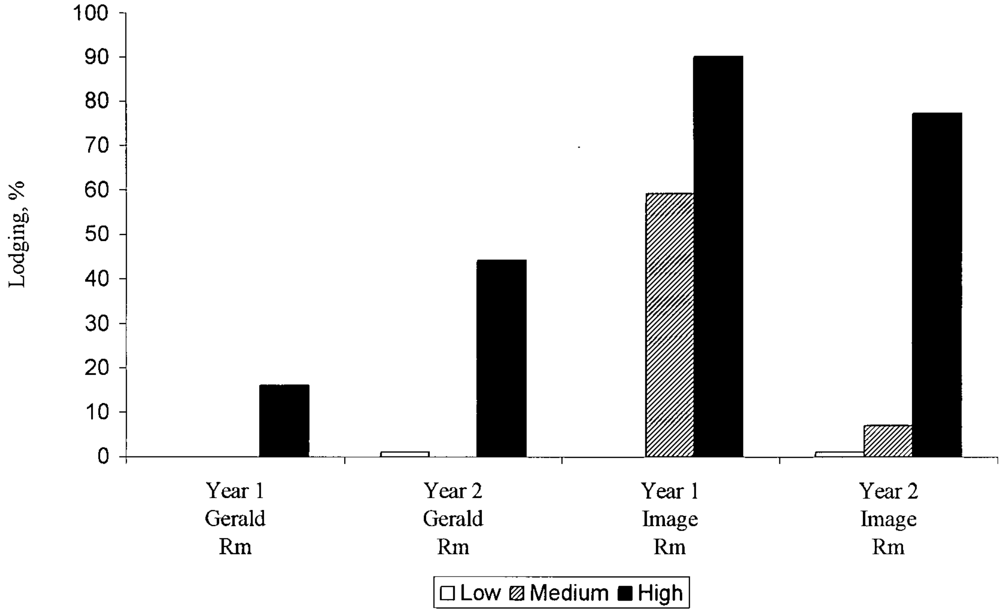

Figure 26. Effects of ammonium nitrate (Low, Medium, High) on crop lodging.

Stem carbohydrate reserves

Stem carbohydrate reserves were measured in order to determine the ability of the crop to maintain yield in adverse conditions such as drought. Soluble sugars accumulate in the stems and contribute significantly to grain yield during normal crop growth. These reserves have been shown to contribute from 20 to 50% of grain yield in wheat (HGCA, 1997) and accumulation of high levels of stem carbohydrate reserves has thus been associated with high yield potential. They have also been shown to buffer against adverse conditions such as drought where they can contribute significantly to grain filling by re-distribution from the stems; hence lower final levels would be expected under unfavourable conditions. These sugars have been termed stem carbohydrate reserves and were measured in the study at GS 65 and mid-senescence. Stem carbohydrate reserves in wheat are produced mainly during stem extension and are maximal at flowering; a similar pattern of deposition has been assumed for oats, hence their measurement at GS 65. From this point onwards they may be redistributed to the grain sites; in stressed plants they will be largely utilised by harvest but in unstressed plants significant amounts may still remain. Maintaining Green Area Index such as by applying irrigation or foliar urea would be expected to result in increased amounts of stem carbohydrate reserves at mid-senescence, hence their measurement at this stage of the study since it gave an indication of the success of the treatments and potential for increased yield through prolonged grain filling.

Full results of the analysis of stem carbohydrate reserves are given in Tables A27 to A30 (Appendix). There was generally a reduction in 'maximum' stem carbohydrate reserves with increased levels of ammonium nitrate. This may have been due to increased demand for structural carbohydrates, due to increased shoot numbers/m^2. The only exception to this was Gerald grown at Rosemaund in Year 2. There was no consistent effect of foliar urea on stem carbohydrates at mid-senescence, though this was mainly due to large variations between replicate plots. Larger amounts of stem carbohydrates were found at Gleadthorpe than Rosemaund in both years and at both growth stages. A large increase in stem carbohydrate reserves was noted at Gleadthorpe with application of foliar urea in year 2; this was due to higher GAI providing greater current photosynthesis and hence less demand for these reserves.

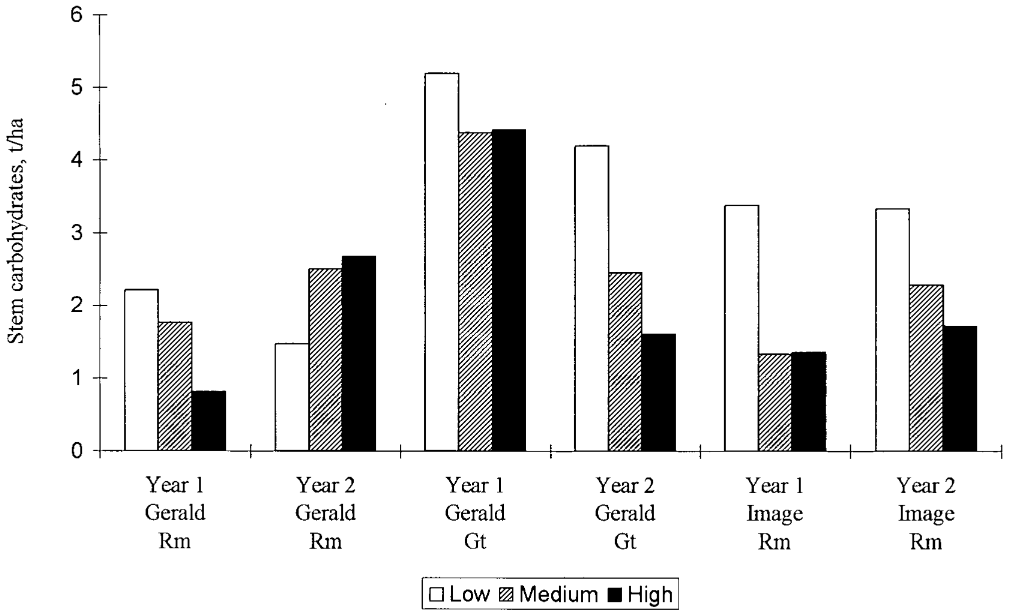

Figure 27. Effects of ammonium nitrate (Low, Medium, High) on stem carbohydrates reserves at GS 65.

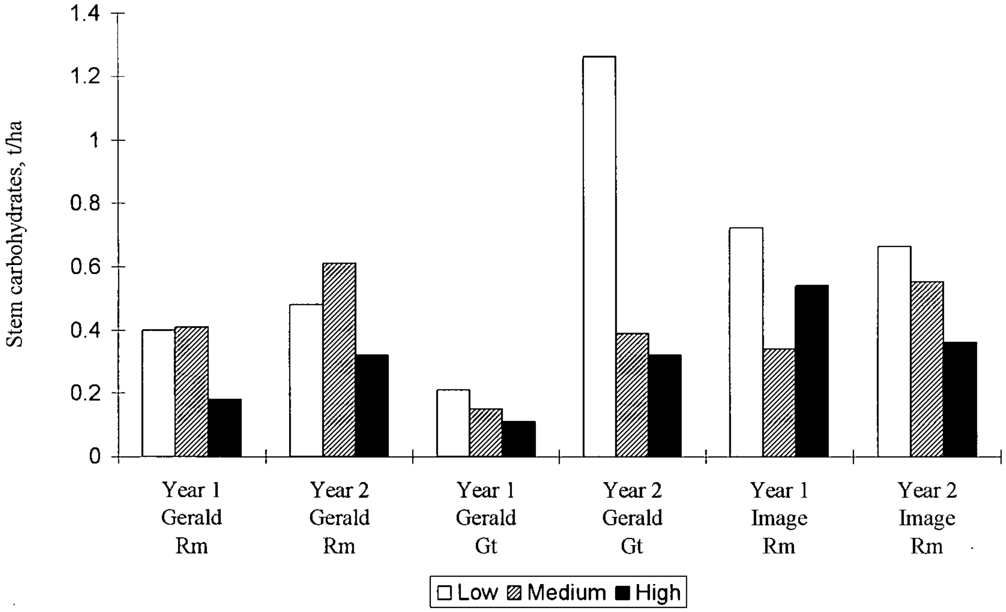

Figure 28. Effects of ammonium nitrate (Low, Medium, High) on stem carbohydrate reserves at mid-senescence.

Harvest indices

Harvest index (HI) is a measure of the partitioning of dry matter to the grain and is measured as the ratio of grain to total crop dry weight. An associated measure of nitrogen partitioning is Nitrogen Harvest Index (NHI) and is an indication of the efficiency of the plant to mobilise nitrogen from the vegetative tissues and translocate it to the grain. These values are shown in Tables A1 to A4.

Applications of both ammonium nitrate and foliar urea resulted in significant increases in total nitrogen offtake (TNO) but lower NHI (Figures 11, 12, 29 and 30) at both sites. Differences in HI as a result of applied nitrogen were not significant in Image but were in Gerald at Rosemaund in both years and at Gleadthorpe in year 2. No effects of control of water supply were found in HI, NHI or TNO at Rosemaund and there were no interactions between ammonium nitrate and irrigation. At Gleadthorpe higher TNO was found with 'early' irrigation in Year 1 but no effect was noted with 'late' irrigation (Figure 13). There was an indication (though not statistically significant) that NHI was lower when 'early' irrigation was applied. This was most likely due to improvement in vegetative growth with early irrigation but a restriction on grain growth later due to water supply limiting the use of stored nitrogen. Significant differences in NHI between years were noted at Rosemaund in both varieties. The results indicated that although greater amounts of nitrogen were taken up by the crop in Year 2 much less was assimilated by the grain than in Year 1. This was possibly a result of the Crown Rust infection at this site limiting grain growth.

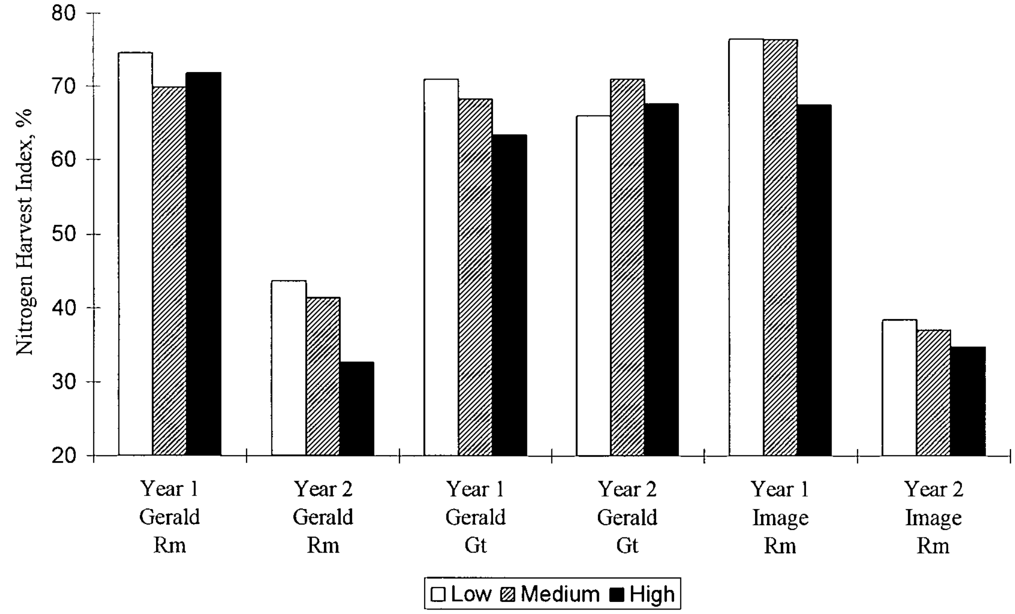

Figure 29. Effects of ammonium nitrate (Low, Medium, high) on Nitrogen Harvest Index.

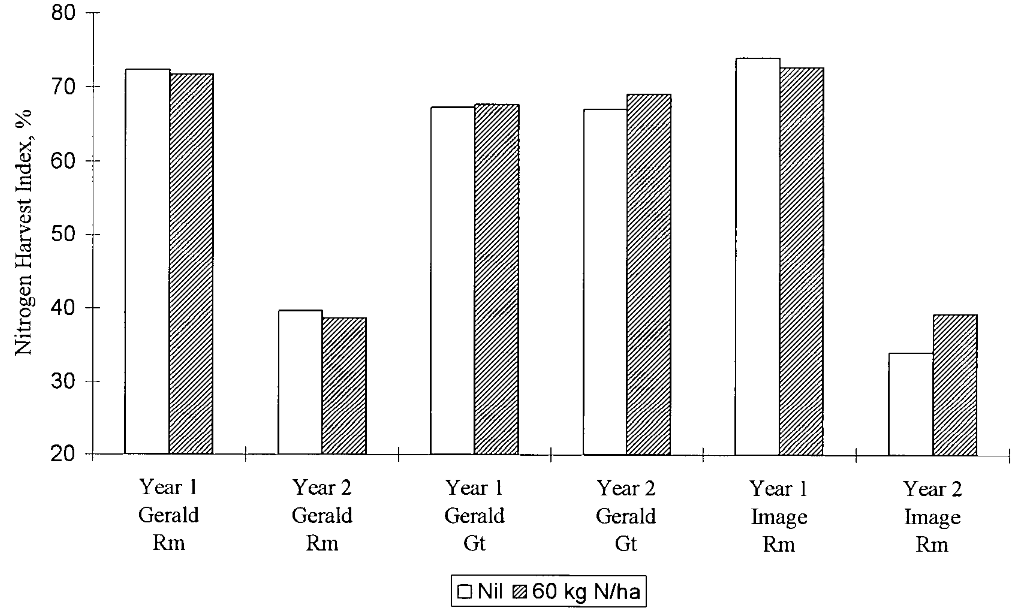

Figure 30. Effects of foliar urea on Nitrogen Harvest Index.

Grain yield

Full results for grain yield are given in the Appendix (Tables A31 to A34). Addition of ammonium nitrate at the 'medium' level resulted in significant increases in grain yield (compared to 'low') at both sites in both years (Figure 31). Differences were evident between the two years when Gerald was compared at each site; in Year 1 the yield increase was greater at Rosemaund but this contrasted with Year 2 where it was much higher at Gleadthorpe. There were two possible reasons for these differences; firstly, dry conditions at Gleadthorpe in Year 1 compared to Year 2 which limited the uptake of nitrogen and subsequent vegetative growth, and secondly there was sufficient nitrogen for crop growth at Rosemaund in Year 2 even in the absence of additional nitrogen applied. In both years higher overall yield was recorded at Rosemaund and was apparently due to the greater water-holding capacity of the soil at this site. In no instances did ammonium nitrate applied at the 'high' level result in greater yield than when it was applied compared to the 'medium' level. This was to be expected since the additional GAI produced by high nitrogen resulted in a negligible increase in PAR interception. In fact, at Rosemaund in Year 2 a significant decrease in yield was noted in Gerald plots receiving 'high' amounts of ammonium nitrate since significant lodging had occurred, disrupting photosynthesis and hence grain filling. This was noted in both years with variety Image due to higher levels of lodging than Gerald (Figure 26). Even in the absence of lodging and in low fertility soil such as at Gleadthorpe there was no benefit in terms of yield by applying ammonium nitrate over the 'medium' level, although the possibility does exist that higher levels of ammonium nitrate at Gleadthorpe may have resulted in higher yield if water supply had not been limiting.

Application of foliar urea resulted in significant increases in grain yield in most cases (Figure 32). There was no evidence of an interaction between foliar urea and the amount of ammonium nitrate at either site.

There was no effect of soil water supply on yield at Rosemaund in either year of the study, due to the presence of adequate soil moisture for growth during the course of the experiment. Some effects of irrigation were noted at Gleadthorpe though they differed between the two years. Gleadthorpe experienced particularly dry conditions during the 'early' growth phase (GS 31-61 approx.) in Year 1; hence the application of irrigation during this period resulted in a significant increase in yield (Figure 33). A smaller increase in yield was noted with irrigation during the 'late' growth phase. The 1998 season was by contrast much wetter than that in 1997 and that there were no significant effects of any irrigation treatments on yield in Year 2. Although rainfall in this year was below average during May, July and August (Figure 4) there was a significant 'carryover' effect of the high rainfall during April and June which meant that adequate soil moisture was available throughout the season.

Comparison of grain yield of a range of winter oat varieties was made in Year 1. Results for conventional and naked varieties are compared separately in Tables 7 and 8. There were large differences in the severity of lodging which must be taken into account when the results for yield are compared. For example, Gerald and Jalna which gave the greatest yield were the varieties most resistant to lodging. Grain yield varied considerably between the conventional varieties, from 6.9 t/ha in Chamois to 9.1 t/ha in Jalna. Smaller (though statistically significant) differences in yield were found between the naked varieties. If it assumed that the husk contributed 25% to the final grain weight in the conventional varieties then compared on a groat-for-groat basis similar yields were obtained between the naked and conventional types. There was no evidence of differences in the severity of lodging between naked and conventional types.

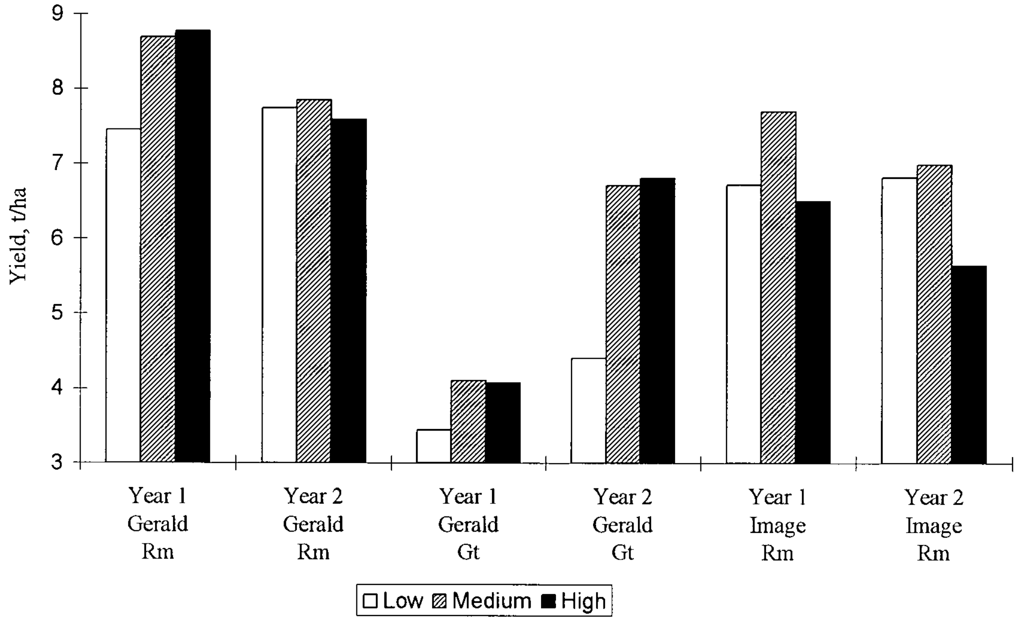

Figure 31. Effect of ammonium nitrate (Low, Medium, High) on grain yield.

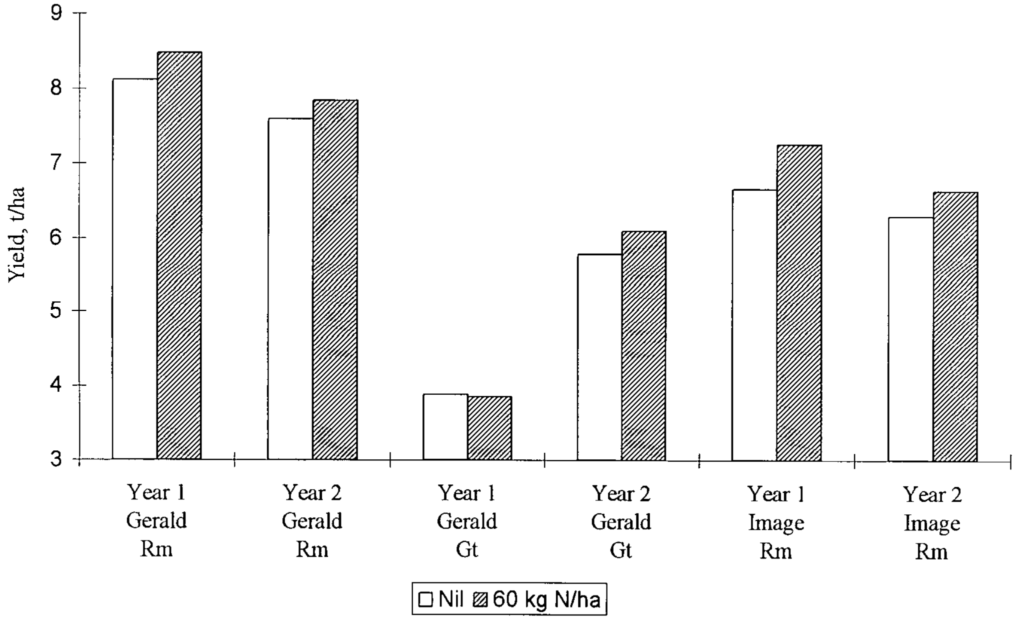

Figure 32. Effect of foliar urea on grain yield.

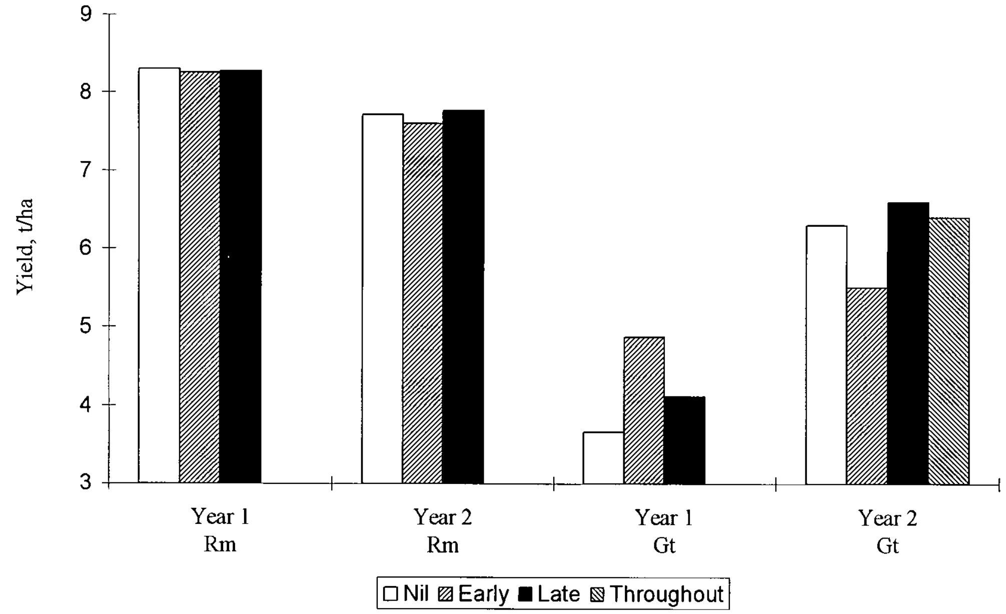

Figure 33. Effect of soil water supply (Nil, Early, Late and Throughout irrigation) on grain yield.

Table 7. Yield and grain quality of conventional (husked) winter oat cultivars grown at Rosemaund in Year 1.

Variety	Lodging %	Yield	Specific Weight kg/hl	TGW g	β-glucan %	Protein %	Oil %
Aintree	56	7.4	48.8	25.2	3.3	10.6	8.0
Chamois	79	6.9	47.3	27.8	3.8	11.5	7.5
Craig	60	7.9	46.5	27.5	3.6	11.3	7.0
Emperor	82	8.0	44.1	30.1	3.2	10.9	6.9
Gerald	19	8.8	48.8	31.2	2.6	9.7	7.1
Image	76	7.1	44.6	25.4	3.0	11.4	7.7
Jalna	23	9.1	49.6	32.0	3.5	10.6	7.0
Solva	72	7.7	46.9	28.3	2.8	10.8	7.3
Sovereign	56	8.0	44.6	30.3	2.8	10.4	7.4
Grand mean	58	7.9	46.8	28.6	3.2	10.8	7.3
Sig. effects(F)	0.001	0.001	<0.001	<0.001	0.002	<0.001	0.028
SED	15.3	0.466	1.268	1.309	0.283	0.343	0.318

Table 8. Yield and grain quality of naked winter oat cultivars grown at Rosemaund in Year 1.

Variety	Lodging %	Yield	Specific Weight kg/hl	TGW g	β-glucan %	Protein %	Oil %
Harpoon	79	5.5	57.6	24.9	3.9	11.8	8.9
Krypton	87	5.2	53.6	21.1	3.5	11.4	10.0
Kynon	51	5.2	58.6	22.1	3.7	12.1	9.1
Lexicon	53	5.9	53.6	22.2	3.5	11.1	9.9
Grand mean	68	5.5	55.9	22.6	3.6	11.6	9.4
Sig. effects(F)	0.241	0.044	0.173	0.053	0.156	0.232	0.361
SED	20.0	0.235	2.590	1.187	0.178	0.475	0.714

Grain quality

Full results for analysis of grain quality are given in the Appendix (Tables A31 to A34) and in Tables 7 and 8.

Application of higher levels of ammonium nitrate resulted in decreases in grain specific weight and TGW (Figure 34 and 35). In most cases this was found at both 'medium' and 'high' levels of addition. The decreases in specific weight and TGW are most likely to have resulted from increases in both fertile shoot numbers (Figure 14) and lodging. The effects of lodging on specific weight are particularly evident in variety Image at Rosemaund in which severe lodging occurred. The effects of nitrogen application on grain physical characteristics were complicated by a site interaction. Grain samples were of lower specific weight at Gleadthorpe than Rosemaund in Year 1, though they were similar at both sites in Year 2. Thousand grain weights were similar at each site in Year 1 but were much higher at Gleadthorpe in Year 2. These differences were mainly due to the higher rainfall at Gleadthorpe during the grain filling period in Year 2.

In general, effects of foliar urea on specific weight and thousand grain weight were small and not statistically significant. There were some exceptions to this at Gleadthorpe where applied foliar urea resulted in a decrease in specific weight in Year 1 and an increase in thousand grain weight in Year 2.

Little effect of soil water supply was noted in grain quality at either site, although a small decrease in specific weight with 'early' irrigation was found at Rosemaund in Year 1. The only effect of irrigation on grain composition was that protein content was slightly higher in the control (no irrigation) treatment at Rosemaund compared to the irrigated plots.

A significant varietal effect was noted in all physical and chemical grain quality parameters for the varieties examined (Tables 7 and 8). Differences between conventional and naked varieties were explained mainly by the contribution of the husk, with similar composition when compared on a groat-for-groat basis. There is a strong likelihood that significant interactions exist in the response of different varieties to applied ammonium nitrate and foliar urea for the various aspects of grain quality considered. This is likely to be due to differences in standing power of the varieties. However, examination of these effects was beyond the scope of the present study. It is recommended that this aspect be considered further in future work.

A key area of the study concerned the effects of the agronomic treatments on grain β-glucan levels. There was no consistent effect of ammonium nitrate on β-glucan; application of ammonium nitrate in Year 2 resulted in small increases at Gleadthorpe and decreases at Rosemaund. The effects of foliar urea application were interesting in that they resulted in significant increases in grain β-glucan content in a number of cases (Figure 36). The magnitude of the effect appeared to be variety dependent and was greatest in Image at Rosemaund where it was found in both years of the study. It was also seen in Gerald though only in Year 2 (at both sites), there being no response in Year 1. The response to foliar urea did not appear to be affected by the level of ammonium nitrate applied or any irrigation applied (only examined at the Rosemaund site). The combination of increases in both yield and β-glucan in response to foliar urea application resulted in a significant amount of additional β-glucan produced by the crop per unit area. Only two oat varieties were examined in these experiments, but a much wider range of winter oat varieties were analysed for β-glucan content in a further experiment. The results of these analyses (Tables 7 and 8) showed that significant differences between varieties in β-glucan content existed. Possible site and seasonal differences in β-glucan content were also noted although these were not consistent throughout the study (Figure 36). In both years higher β-glucan content was found at Gleadthorpe compared to Rosemaund though this was most likely a dilution effect of increased yield at the Rosemaund site. It is interesting to note that no difference in β-glucan content was found between Gerald grown at Gleadthorpe in Year 1 compared to Year 2 in the 'nil' foliar urea treatments; however, a significant difference was found in the samples which had received foliar urea. There was evidently an interaction between seasonal factors and the application of foliar urea.

Other grain components responded in a predictable manner to the experimental treatments. Applications of ammonium nitrate or foliar urea resulted in significant increases in grain protein in all experiments (Figures 37 and 38). The effect was stepwise and indicated that further additions of ammonium nitrate may have resulted in even higher grain protein contents than those seen. There was generally a greater effect of foliar urea on grain protein at lower levels of ammonium nitrate, where nitrogen was likely to be more limiting in terms of supply for grain filling; an exception though was noted with Image at Rosemaund in Year 2 where much greater increases in protein were found at 'medium' ammonium nitrate level. The reason for this was possibly due to more severe lodging in these plots compared to those which received no ammonium nitrate. This would have restricted carbohydrate accumulation in the grain to a greater extent than nitrogen uptake.

There was evidence that grain oil content decreased as a result of foliar urea application, but there was no effect of ammonium nitrate noted. There were also some small differences between varieties (Tables 7 and 8). This indicates that there may be some scope to manipulate oil content through selection of variety and by agronomic techniques. No effect of soil water supply was found in oil content but an effect of season was noted (Figure 39) at both sites. The seasonal effect on grain oil content has been noted before, but the underlying cause of the differences is not known.

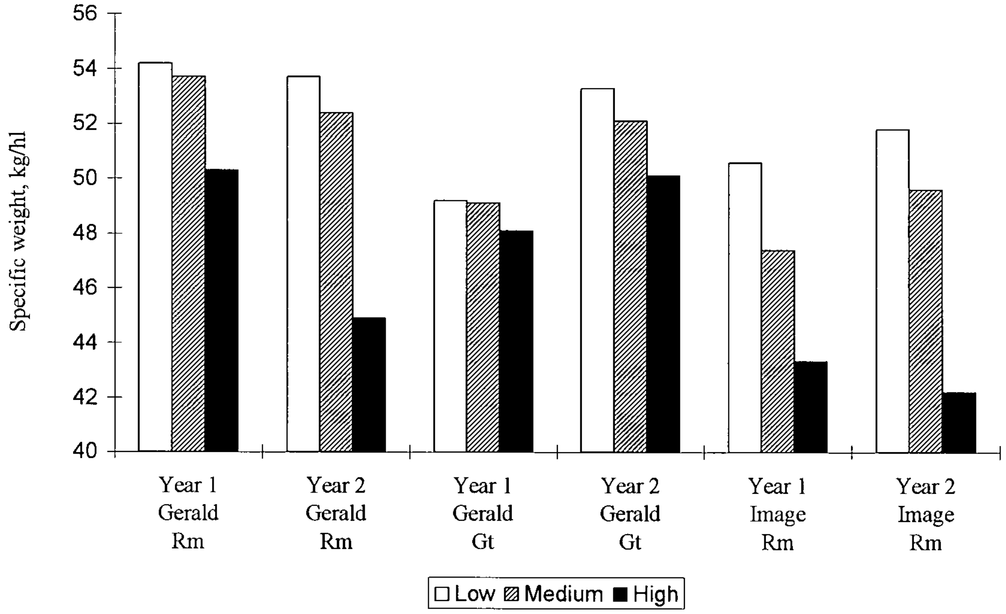

Figure 34. Effect of ammonium nitrate (Low, Medium, High) on specific weight.

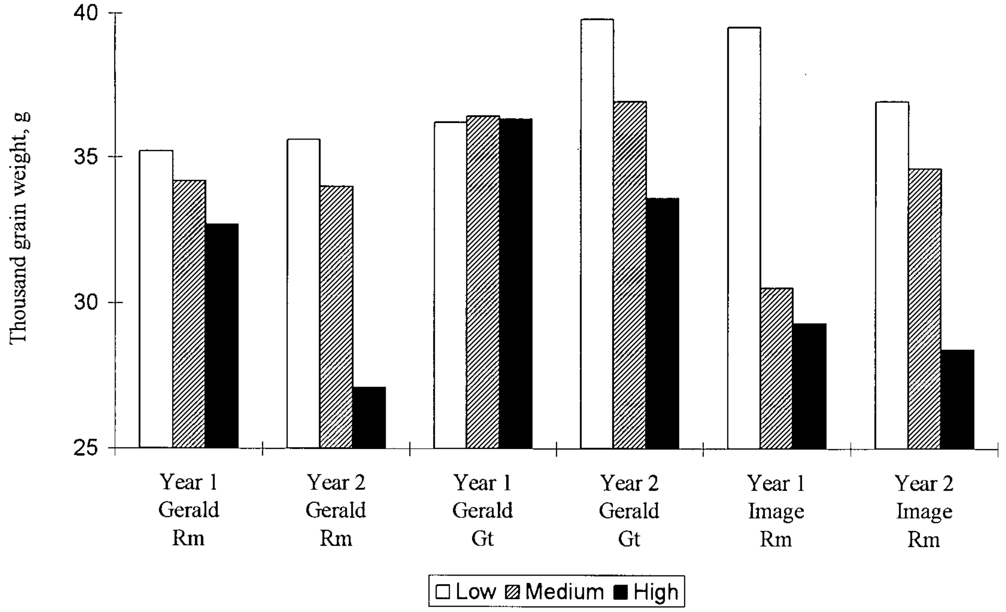

Figure 35. Effect of ammonium nitrate (Low, Medium, High) on thousand grain weight.

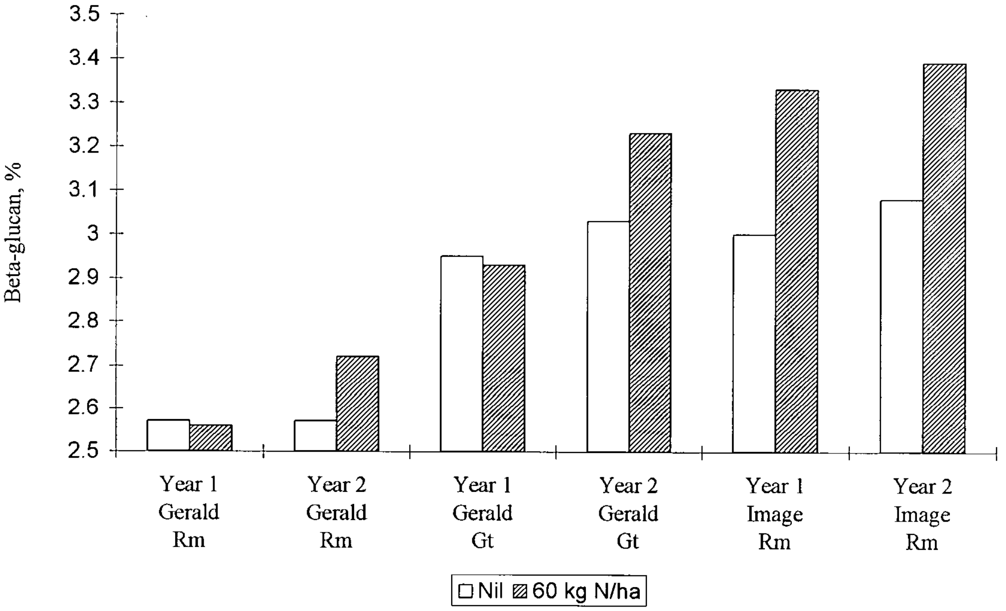

Figure 36. Effect of foliar urea on grain β-glucan.

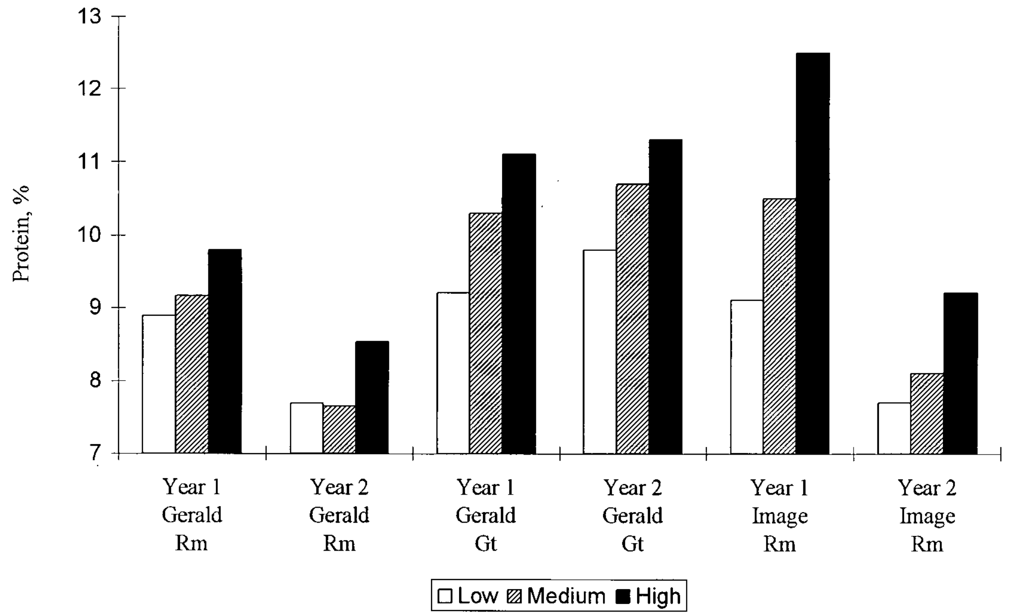

Figure 37. Effect of ammonium nitrate (Low, Medium, High) on grain protein.

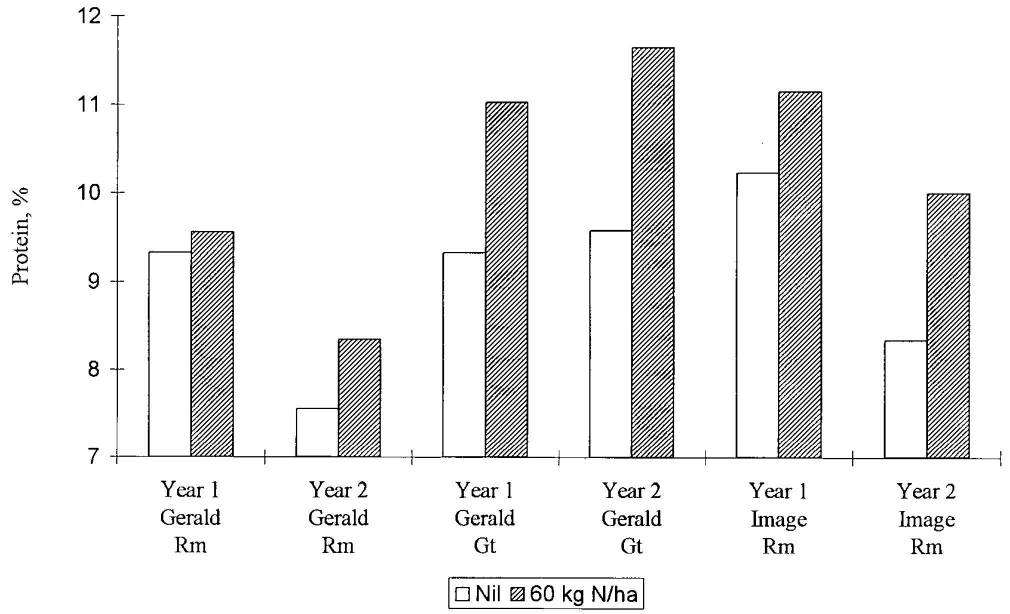

Figure 38. Effect of foliar urea on grain protein.

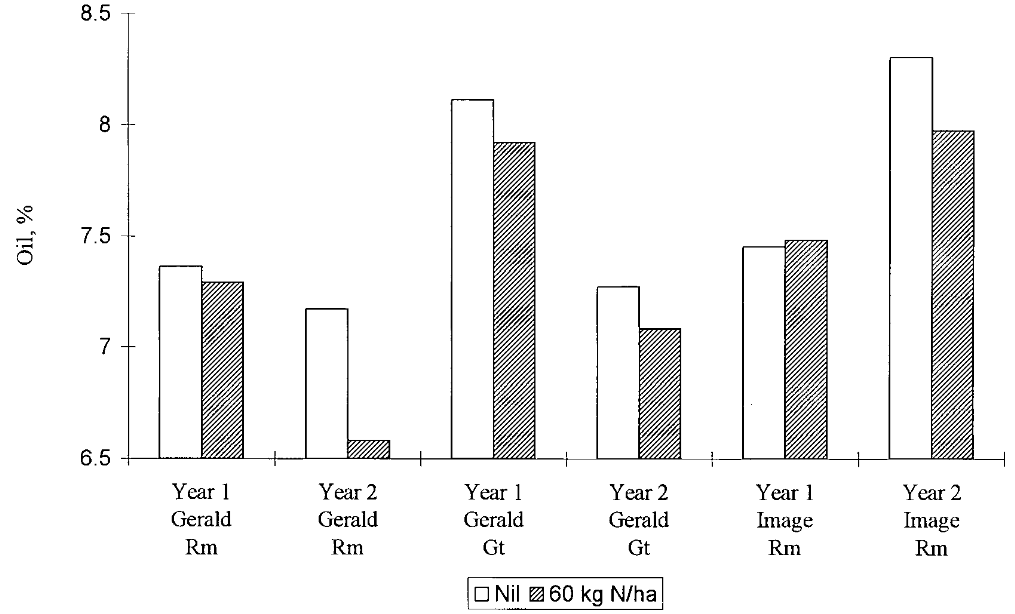

Figure 39. Effect of foliar urea on grain oil content.

Discussion

The study carried out by ADAS was intended to produce crop management guidelines for growing grain of suitable quality for industrial fractionation. Important recommendations can be made at this stage but other aspects of grain quality need further research. This two year research project has identified some aspects of management practices which are not yet fully understood and require further testing.

The effects of the experimental treatments in the study were broadly in line with the results of similar studies involving wheat. Addition of ammonium nitrate resulted in the following effects:

- Higher shoot numbers through increased tiller survival
- Increased crop height
- Increased Green Area Index via increases in all the major components of Green Area, leaf, stem and ear
- Increases in the amounts of stem carbohydrate reserves
- Increased crop dry mass
- Increased yield
- Increased grain protein

These changes with increased ammonium nitrate enabled the crop to intercept more solar radiation and also to maintain photosynthetic capacity for longer, thus increasing the yield potential. This response did however appear to tail off once an optimum canopy size comparable to that in wheat (GAI 5-6) had been achieved. Negative effects of increased ammonium nitrate above these levels were noted at the

Rosemaund site, principally due to lodging. In Year 1 where lodging occurred early in the season this was reflected in lower yield, specific weight, and TGW from plots which had lodged severely. Application of foliar urea also had significant effects on the physiological status of the crop, resulting in higher Green Area Index at mid-senescence compared to plots which received no foliar urea. This is thought to have extended the grain filling period by maintaining the green canopy for longer than normal and was reflected in higher grain yield. The impact of controlling soil water supply was not as great as had been expected, due to the high rainfall at critical points during the season, with only minor effects noted in crop growth.

All factors examined in the study (variety, site, season, ammonium nitrate, foliar urea and water supply) were shown to influence grain yield and many of these also affected grain quality. The most significant differences were due to variety and site, though large seasonal effects were noted at Rosemaund and at Gleadthorpe due to the different rainfall pattern between the two years. The factors of most importance for consideration by the grower are selection of the correct variety and nitrogen fertilisation (in addition to disease control).

Large differences in yield and composition were noted between varieties. Differences were also noted in grain physical characteristics (specific weight, TGW) and these will be of consequence in traditional markets for oats. Previous reports had indicated that varietal differences existed in composition (Hutchinson and Martin, 1955a,b; Welch and Yong, 1980; Welch and Lloyd, 1989; Peterson, 1991; Saastamoinen *et al.*, 1992; Bhatty, 1992; Zhou *et al.*, 1998) but little data exists concerning currently grown winter sown varieties in the UK. Many of these earlier reports also did not define closely the growing conditions of the crops from which the samples were taken. The results of the current study thus serve as a guide to varietal selection. Further information regarding the characteristics of newer varieties is available from the NIAB Recommended List. Many of the newer varieties from the IGER breeding programme have specific characteristics which could be of importance in new markets for oats. The grain from the new naked variety Icon is particularly valuable since it is high in β-glucan, oil and protein. This variety is also resistant to lodging being of short height and stiff-strawed. More detailed information will be available about Icon and other new varieties from the Oatec project in due course. Of particular note will be comparisons of varieties across years to determine in more detail the variation in composition due to season.

Varieties producing the highest yield were generally those producing the greatest amounts of the components of interest. The production in kg/ha of each component based on both grain yield and percentage composition is shown in Table 9. However, this will only be of benefit in cases where a premium is paid for the component of interest. A number of other factors require consideration in selecting a variety from Table 9. The comparison is of varieties grown at only one high yielding site - different results might be obtained at other sites, particularly in low yield situations. Agronomic characteristics of the varieties should be considered alongside the particular site factors (e.g. drought, disease, soil fertility, lodging risk). For example, where high protein content is required, necessitating the use of high amounts of nitrogen fertiliser, the resistance of a variety to lodging should be considered at the chosen site, in addition to its protein yield in the current study since the results may be quite different in a lodged crop compared to one which does not lodge.

Table 9. Production of protein, oil and beta-glucan (kg/ha at 15% moisture) in field studies at Rosemaund and Gleadthorpe

Site	Variety	Ammonium nitrate kg N/ha	Foliar urea kgN/ha	Protein kg/ha		Oil kg/ha		Beta-glucan kg/ha	
				Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
Rosemaund	Gerald	0	0	511	486	461	459	157	165
			60	616	526	478	441	169	185
		40	0	642	458	512	466	180	166
			60	712	563	555	485	186	190
		100	0	719	523	549	464	196	167
			60	741	575	554	391	199	168
Rosemaund	Image	0	0	455	378	418	479	166	180
			60	589	514	459	492	194	208
		40	0	614	367	461	489	191	164
			60	756	606	522	533	239	214
		100	0	664	405	388	371	153	150
			60	713	473	404	336	185	153
Gleadthorpe	Gerald	40	0	243	303	246	266	84	104
			60	292	443	230	276	84	121
		100	0	320	552	275	395	103	169
			60	400	673	273	421	104	185
		140	0	369	586	283	414	106	179
			60	394	713	272	412	100	201
Rosemaund (variety comparison)	*Conventional*								
	Aintree			667	-	503	-	208	-
	Chamois			674	-	440	-	223	-
	Craig			759	-	470	-	242	-
	Emperor			741	-	469	-	218	-
	Gerald			726	-	531	-	194	-
	Image			688	-	465	-	181	-
	Jalna			820	-	541	-	271	-
	Solva			707	-	478	-	183	-
	Sovereign			707	-	503	-	190	-
	Mean			725	-	490	-	215	-
	Naked								
	Harpoon			552	-	416	-	182	-
	Krypton			504	-	442	-	155	-
	Kynon			535	-	402	-	164	-
	Lexicon			557	-	496	-	176	-
	Mean			542	-	439	-	168	-

It was interesting to note that a comparison of conventional and naked varieties on a groat-for-groat basis (assuming the husk contributes approx. 25% to the total weight of the grain) showed that there was no difference in the amounts of protein and β-glucan produced on a kg/ha basis. The situation is different for oil, where naked oats produced on average an additional 71 kg/ha, nearly 20% more than the conventional varieties. Large variations between individual varieties were found and should be considered when selecting a variety.

Use of nitrogen fertiliser both as ammonium nitrate and as foliar urea was shown to be a key tool for manipulating both the yield and quality of grain produced. Application of ammonium nitrate resulted generally in increased grain yield but this reached a maximum at Rosemaund, above which yield declined due to the effects of lodging. Interactions were thus apparent between the resistance to lodging and the benefit from applied ammonium nitrate. A maximum was also reached at the 'medium' ammonium nitrate level at Gleadthorpe but no decline in yield was found at the 'high' level since there was no lodging at this site. Effects were also noted in grain physical characteristics with decreases in both specific weight and TGW at higher ammonium nitrate levels. A further factor shown to be of importance was the rainfall pattern following ammonium nitrate application. In Year 2 of the study a dry period at Rosemaund and plenty of nitrogen already in the soil inhibited the uptake of applied nitrogen by the crop thus limiting the yield response. This was also seen in other experiments at this site in this year. These factors must therefore be considered in relation to ammonium nitrate application and husbandry practices must be flexible to enable modification of both the amount and the timing of ammonium nitrate in response to prevailing weather conditions in a given season. Increased yield also resulted from application of nitrogen as foliar urea in addition to increases in grain quality.

In addition to yield there were considerable benefits of nitrogen fertilisation in terms of grain quality and the combination of the effects on both yield and quality would have been of significant benefit to the grower of oats for industrial fractionation. The increase in grain protein as a result of application of fertiliser nitrogen has been well documented (Peterson, 1976; Eppendorfer, 1977; Chalmers *et al.*, 1998). This was found in the current study at both sites in both years and unlike yield there appeared to be a response at all levels of ammonium nitrate application. This indicated that even where no further yield increase is likely, higher levels of ammonium nitrate might be of benefit if grain protein is a key component in an industrial fractionation scheme. Some previous reports had indicated an inverse relationship between grain protein and oil content (Dent, 1957; Brown *et al.*, 1966; Nik-Khah *et al.*, 1972) although others have found no relationship (Youngs and Forsberg, 1979; Gullord, 1980; Saastamoinen, 1987). In accord with these latter reports, no overall relationship between protein and oil was found in this study.

Of particular interest were the effects of both nitrogen and soil water supply on grain β-glucan levels since β-glucan is potentially the most valuable component for oat fractionation. Current estimates of the value of β-glucan in commercial markets are up to £5 per kg for low purity high volume uses and up to £1,500/kg for very high purity low volume uses. Soil water supply has been cited in previous reports as an important factor in barley, with drought conditions leading to elevated levels of glucan (Schuster *et al.*, 1967; Bendelow, 1975; Smart, 1976). However, no effect of soil water supply was noted in grain β-glucan content in the present study. This may have been due to the fact that during the period of study the crops were not sufficiently drought stressed to produce the response in β-glucan. Previous reports were mainly of studies which had taken place in glasshouse or controlled environments and were able to produce severe drought. Under these conditions changes in β-glucan levels are believed to have occurred as a plant survival mechanism in response to extreme environmental stress (Stone and Clarke, 1992) and if so it is unlikely that these conditions can be replicated in field studies in the UK climate, even in a dry year. The difficulty of scheduling the irrigation treatments clearly demonstrated the problems in carrying out this type of experiment in unpredictable weather conditions such as in the two years of this study. As a result of this it is recommended that future studies should be carried out using automated crop shelters in order to guarantee drought conditions.

The effects of applying nitrogen as late foliar urea were particularly interesting. Application of foliar urea at all ammonium nitrate levels resulted in increases in yield, protein, and β-glucan. There was also a decrease in grain oil content with foliar urea application. The changes in β-glucan and oil were unexpected and have not been recorded before. **In combination with increased yield this represents a practical means to enhance the value of a crop if β-glucan is the main component of interest.** Further work is necessary to investigate whether significant differences between varieties exist in the response to foliar urea application. This could have implications for selection of variety and subsequent husbandry decisions when growing oats for extraction of β-glucan. Although only two varieties were examined in detail in this study, there were indications that β-glucan levels responded to foliar urea more consistently and to a greater extent in Image than in Gerald.

The results for the use of foliar urea are important in terms of the management of winter oats when grown for fractionation since no other reports exist which demonstrate practical measures for enhancing grain β-glucan content. It is important in any further studies to know whether varieties which contain higher levels of β-glucan show an equivalent response to application of foliar urea. In addition, the amount and timing of the foliar urea may be of importance and should be studied further. The overall benefit of applying foliar urea can be seen by considering that application of foliar urea to variety Image at Rosemaund in Year 1 resulted in an additional 40 kg β-glucan per hectare. This would be worth £200 per hectare for low value and as much as £60,000 for high value uses.

The present study has shown that potential exists to manipulate the quality of oats by using appropriate ammonium nitrate and foliar urea programmes. The increase in grain β-glucan content with foliar urea application is of particular importance for manipulating the quality of grain for industrial fractionation. It is clear that considerable interactions between the various factors exist and in many cases the selection of an appropriate variety and a management regime which is aimed at the highest grain yield will also satisfy the requirements in terms of grain quality. Based on the results of this study, Table 10 summarises the possible strategies that can be employed in this respect.

Table 10. Summary of crop management decisions to be considered in producing grain for fractionation

Component	Site	Variety	Ammonium nitrate	Foliar urea
Starch	High fertility and rainfall	High yielding type	Optimal for yield	No
Protein	Not important	Select high protein type	High	Yes
β-glucan	Not important	Select high β-glucan type	Optimal for yield	Yes
Oil	Not important	Select high oil type	Optimal for yield	No

Recommendations for further studies

Further studies are required as follows:

- Examination of the effects of foliar urea on a wider range of winter oat varieties since significant interactions between variety and foliar urea may exist. This could confer significant advantage to the UK grower since it represents a practical means of manipulating grain β-glucan content once a particular variety has been sown. This is of importance for both industrial fractionation and also in relation to the manufacture of 'functional foods' based on the FDA health claim for oat soluble fibre.
- Examination of the effects of other factors which can increase grain filling and may thus increase grain β-glucan content. This should include the use of strobilurin fungicides which have been shown to lead to increases in grain filling in wheat.
- Further studies examining the effects of drought on grain composition. These should be carried out using automated crop shelters to guarantee drought conditions are experienced. This will also allow examination of drought at specific growth stages.
- More detailed studies of crop growth and development for winter oats. Although the current study provided some information, much greater detail is required for full understanding of the basis for grain yield and quality.

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Appendix

Table	Details
A1	HI, NHI, TNO
A2	"
A3	"
A4	"
A5	Shoot numbers
A6	"
A7	"
A8	"
A9	Green Area Indices
A10	"
A11	"
A12	"
A13	Intercepted solar radiation
A14	"
A15	"
A16	"
A17	Crop dry matter
A18	"
A19	"
A20	"
A21	"
A22	"
A23	"
A24	"
A25	Crop height and lodging
A26	"
A27	Stem carbohydrate reserves
A28	"
A29	"
A30	"
A31	Grain yield and quality
A32	"
A33	"
A34	"

Table A1. Effects of ammonium nitrate, foliar urea and soil moisture* on havest index, NHI and total nitrogen offtake for Gerald grown at Rosemaund.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Irrigation	Dry matter harvest index, %		Nitrogen harvest index, %		Total nitrogen offtake, kg/ha	
			Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
0	0	Nil	54.2	45.2	75.7	44.5	115.9	188.8
		Early	53.1	46.2	73.0	44.3	127.2	167.0
		Late	55.8	45.9	75.9	43.6	126.6	187.2
0	60	Nil	52.4	44.2	73.3	42.9	145.2	211.7
		Early	53.4	44.7	72.2	43.9	143.8	219.7
		Late	53.5	46.5	75.8	46.7	140.9	193.5
40	0	Nil	51.7	44.4	68.1	43.3	162.9	180.4
		Early	52.5	42.3	73.9	39.4	150.8	210.2
		Late	48.1	41.8	67.1	36.8	166.0	222.6
40	60	Nil	52.7	41.3	71.4	39.4	172.5	246.6
		Early	51.3	43.6	73.8	39.7	170.3	243.6
		Late	52.3	42.6	75.7	41.6	165.8	245.5
100	0	Nil	53.1	38.0	73.2	31.3	168.6	292.4
		Early	54.7	38.8	69.5	35.6	166.5	236.7
		Late	52.5	38.7	71.0	27.6	180.2	290.3
100	60	Nil	52.0	39.6	70.3	33.8	180.0	293.3
		Early	51.0	38.6	66.3	32.2	191.8	281.4
		Late	52.4	39.0	70.8	26.0	182.8	371.6
Means								
Grand mean			52.6	42.3	72.1	38.5	158.8	237.9
Ammonium nitrate		0	53.7	45.4	74.3	44.3	133.3	194.7
		40	51.4	42.7	71.7	40.0	164.7	224.8
		100	52.6	38.8	70.2	31.1	178.3	294.3
Foliar urea		0	52.8	42.4	71.9	38.5	151.6	219.5
		60	52.3	42.2	72.2	38.5	165.9	256.3
Irrigation		Nil	52.7	42.1	72.0	39.2	157.5	235.5
		Early	52.7	42.4	71.5	39.18	158.4	226.5
		Late	52.4	42.4	72.7	37.040	160.4	251.800
Significance of effects (F)								
Ammonium nitrate (AN)			0.018	<0.001	0.021	<0.001	<0.001	<0.001
Foliar urea (FU)			0.410	0.861	0.846	0.979	<0.001	0.001
Irrigation (I)			0.974	0.968	0.730	0.478	0.903	0.031
AN x FU			0.123	0.778	0.091	0.850	0.455	0.822
AN x I			0.431	0.884	0.846	0.065	0.725	0.146
FU x I			0.405	0.742	0.193	0.361	0.192	0.875
AN x FU x I			0.279	0.596	0.855	0.31	0.715	0.31
Standard errors (SED)								
AN			0.77	0.89	1.44	1.27	4.20	13.14
FU			0.63	0.73	1.18	1.04	3.43	10.73
I			1.24	1.25	1.53	1.91	6.46	7.08
AN x FU			1.09	1.26	2.04	1.80	5.94	18.58
AN x I			1.65	1.77	2.55	2.62	8.78	19.88
FU x I			1.46	1.53	2.10	2.3	7.71	14.9
AN x FU x I			2.13	2.35	3.57	3.4	11.40	30.2

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A2. Effects of ammonium nitrate and foliar urea on havest index, NHI and total nitrogen offtake for cultivar Image grown at Rosemaund.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Dry matter harvest index, %		Nitrogen harvest index, %		Total nitrogen offtake, kg/ha	
		Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
0	0	46.2	38.8	73.4	37.0	117.6	176.9
	60	49.5	40.0	79.4	39.9	142.6	220.1
40	0	47.9	38.2	76.6	32.2	184.0	196.0
	60	50.2	41.3	76.0	41.7	202.9	249.1
100	0	52.3	38.4	72.0	33.1	194.6	208.2
	60	42.6	38.8	62.7	36.2	217.3	221.7
Means							
Grand mean		48.1	39.3	73.4	36.7	176.5	212.0
Ammonium	0	47.9	39.4	76.4	38.4	130.1	198.5
nitrate	40	49.0	39.8	76.3	37.0	193.4	222.5
	100	47.4	38.6	67.4	34.7	206.0	215.0
Foliar urea	0	48.8	38.5	74.0	34.1	165.4	193.7
	60	47.4	40.0	72.7	39.3	187.6	230.3
Significance of effects							
Ammonium nitrate (AN)		0.857	0.600	0.003	0.258	0.003	0.321
Foliar urea (FU)		0.570	0.123	0.524	0.013	0.185	0.012
AN x FU		0.075	0.531	0.023	0.268	0.988	0.441
Standard errors (SED)							
AN		2.92	1.17	2.45	2.22	19.60	15.69
FU		2.39	0.96	2.00	1.81	16.00	12.81
AN x FU		4.13	1.66	3.47	3.13	27.72	22.19

Data are means of four replicate plots

Table A3. Effects of ammonium nitrate and foliar urea on havest index, NHI and total nitrogen offtake for cultivar Gerald grown at Gleadthorpe.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Dry matter harvest index, %		Nitrogen harvest index, %		Total nitrogen offtake, kg/ha	
		Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
0	0	47.8	42.2	69.4	63.6	59.5	80.3
	60	50.4	42.3	72.5	68.1	70.0	100.1
40	0	47.2	49.1	66.6	69.9	89.6	125.3
	60	49.6	50.6	69.9	72.0	99.8	154.8
100	0	48.6	48.7	65.8	67.6	99.4	150.5
	60	46.5	49.8	60.7	67.3	118.0	176.0
Means							
Grand mean		48.3	47.1	67.5	68.1	89.4	131.2
Ammonium nitrate	0	49.1	42.2	70.9	65.9	64.7	90.2
	40	48.4	49.9	68.2	70.9	94.7	140.1
	100	47.6	49.3	63.3	67.5	108.7	163.2
Foliar urea	0	47.9	46.7	67.3	67.0	82.8	118.7
	60	48.8	47.6	67.7	69.1	95.9	143.6
Significance of effects							
Ammonium nitrate (AN)		0.617	<0.001	0.026	0.076	<0.001	<0.001
Foliar urea (FU)		0.433	0.329	0.842	0.236	0.009	<0.001
AN x FU		0.244	0.795	0.199	0.540	0.682	0.720
Standard errors (SED)							
AN		1.50	1.108	2.54	2.08	5.34	5.92
FU		1.22	0.905	2.07	1.70	4.36	4.83
AN x FU		2.12	1.568	3.59	2.95	7.56	8.37

Data are means of four replicate plots

Table A4. Effects of soil moisture* on harvest index, NHI and total nitrogen offtake for cultivar Gerald grown at Gleadthorpe.

Irrigation	Dry matter harvest index, %		Nitrogen harvest index, %		Total nitrogen offtake, kg/ha	
	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
Nil	54.6	48.1	75.2	69.5	71.7	138.0
Early	51.3	46.3	71.4	65.8	92.8	124.2
Late	54.3	47.7	74.5	64.4	77.7	148.1
Throughout	-	47.2	-	65.6	-	142.6
Grand mean	53.4	47.3	73.7	66.3	80.7	138.2
Sig. effects (F)	0.084	0.381	0.261	0.162	0.022	0.212
SED	1.30	1.04	2.19	2.12	5.52	10.69

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A5. Effects of ammonium nitrate, foliar urea and soil moisture* on shoot numbers in cultivar Gerald grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	Irrigation	GS 65				Mid-senescence			
			Fertile		D & D		Fertile		D & D	
			Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
0	0	Nil	320	335	83	90	383	264	135	20
		Early	322	324	78	105	356	275	129	47
		Late	288	397	114	59	353	288	45	93
0	60	Nil	-	-	-	-	415	334	101	57
		Early	-	-	-	-	361	334	94	68
		Late	-	-	-	-	411	334	37	32
40	0	Nil	417	345	59	57	532	387	53	70
		Early	440	345	81	69	455	309	95	41
		Late	447	332	54	59	515	357	113	22
40	60	Nil	-	-	-	-	516	301	130	48
		Early	-	-	-	-	542	407	52	30
		Late	-	-	-	-	460	397	65	35
100	0	Nil	448	379	80	25	413	343	48	26
		Early	428	378	32	68	573	359	64	26
		Late	458	406	75	24	440	327	13	85
100	60	Nil	-	-	-	-	460	391	32	22
		Early	-	-	-	-	524	386	47	14
		Late	-	-	-	-	543	442	65	22
Means										
Grand mean			396	360	73	62	459	346	73	41
Ammonium nitrate		0	310	352	92	84	380	305	90	50
		40	435	340	65	62	503	360	84	41
		100	445	388	62	39	492	375	45	32
Foliar urea		0	-	-	-	-	447	323	77	46
		60	-	-	-	-	470	369	69	37
Irrigation		Nil	395	353	74	57	453	337	83	38
		Early	397	349	64	81	468	345	80	38
		Late	398	378	81	47	454	357	56	48
Significance of effects (F)										
Ammonium nitrate (AN)			<0.001	0.341	0.380	0.086	<0.001	<0.001	0.628	0.411
Foliar urea (FU)			-	-	-	-	0.266	0.003	0.628	0.401
Irrigation (I)			0.997	0.833	0.359	0.215	0.857	0.657	0.174	0.819
AN x FU			-	-	-	-	0.833	0.368	0.703	0.488
AN x I			0.847	0.839	0.606	0.860	0.143	0.992	0.325	0.199
FU x I			-	-	-	-	0.916	0.231	0.562	0.175
AN x FU x I			-	-	-	-	0.239	0.090	0.703	0.206
Standard errors (SED)										
AN			28.2	32.5	22.7	18.9	26.0	17.8	19.4	13.0
FU			-	-	-	-	21.2	14.5	15.8	10.6
I			33.2	52.3	11.3	17.2	30.6	21.9	13.5	19.1
AN x FU			-	-	-	-	36.7	25.1	27.4	18.3
AN x I			51.9	69.6	34.1	31.7	47.8	33.4	30.5	26.5
FU x I			-	-	-	-	40.1	28.2	23.6	23.1
AN x FU x I			-	-	-	-	65.6	45.4	45.3	34.7

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A6. Effects of ammonium nitrate and foliar urea on shoot numbers in cultivar Image grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	GS 65				Mid-senescence			
		Fertile		D & D		Fertile		D & D	
		Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
0	0	472	396	146	18	-	378	-	34
	60	-	-	-	-	-	377	-	52
40	0	734	399	228	28	-	412	-	12
	60	-	-	-	-	-	451	-	9
100	0	730	420	168	24	-	354	-	74
	60	-	-	-	-	-	354	-	37
Means		-		-		-		-	
Grand mean		645	405	181	24	-	388	-	36
Ammonium	0	472	396	146	18	-	377	-	43
nitrate	40	734	399	228	28	-	432	-	11
	100	730	420	168	24	-	354	-	55
Foliar urea	0	-	-	-	-	-	382	-	40
	60	-	-	-	-	-	394	-	33
Significance of effects						-		-	
Ammonium nitrate (AN)		0.037	0.880	0.322	0.793	-	0.089	-	0.197
Foliar urea (FU)		-	-	-	-	-	0.657	-	0.730
AN x FU		-	-	-	-	-	0.791	-	0.525
Standard errors (SED)									
AN		86.4	50.7	51.0	14.8	-	33.3	-	24.2
FU		-	-	-	-	-	27.2	-	19.8
AN x FU		-	-	-	-	-	47.0	-	34.3

Data are means of four replicate plots

Table A7. Effects of ammonium nitrate and foliar urea on shoot numbers in cultivar Gerald grown at Gleadthorpe

Ammonium nitrate kg/ha	Foliar urea kg/ha	GS 65						Mid-senescence					
		Fertile		Dead		Dying		Fertile		Dead		Dying	
		Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
40	0	435	357	24	276	6	19	443	320	16	300	2	0
	60	-	-	-	-	-	-	477	293	14	256	19	2
100	0	504	513	18	239	29	57	487	415	48	271	2	8
	60	-	-	-	-	-	-	496	404	16	255	28	2
140	0	506	577	36	290	52	61	523	527	41	163	6	7
	60	-	-	-	-	-	-	521	644	34	222	110	18
<u>Means</u>													
Grand mean		482	482	26	268	29	46	491	434	28	245	28	6
Ammonium nitrate	40	435	357	24	276	6	19	460	307	15	278	11	1
	100	504	513	18	239	29	57	491	410	32	263	15	5
	140	506	577	36	290	52	61	522	585	37	193	58	12
Foliar urea	0	-	-	-	-	-	-	484	421	35	245	3	5
	60	-	-	-	-	-	-	498	447	21	245	52	7
<u>Significance of effects (F)</u>													
Ammonium nitrate (AN)		0.033	0.023	0.523	0.359	0.035	0.274	0.155	<0.001	0.026	0.249	<0.001	0.349
Foliar urea (FU)		-	-	-	-	-	-	0.579	0.372	0.046	0.997	<0.001	0.737
AN x FU		-	-	-	-	-	-	0.831	0.113	0.168	0.610	0.001	0.529
<u>Standard errors (SED)</u>													
AN		22.8	58.4	15.6	33.2	13.0	25.5	30.2	35.1	7.7	52.3	10.2	7.6
FU		-	-	-	-	-	-	24.6	28.7	6.3	42.7	8.3	6.2
AN x FU		-	-	-	-	-	-	42.6	49.7	11.0	73.9	14.4	10.8

Data are means of four replicate plots

Table A8. Effects of soil moisture* on shoot numbers in cultivar Gerald grown at Gleadthorpe

Irrigation	GS 65						Mid-senescence					
	Fertile		Dead		Dying		Fertile		Dead		Dying	
	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
Nil	505	564	14	27	34	155	523	699	35	173	10	7
Early	598	655	5	26	21	128	576	864	15	118	6	0
Late	615	602	0	0	30	215	506	864	37	114	5	8
Throughout	-	643	-	47	-	269	-	760	-	65	-	2
Grand mean	573	616	6	25	29	192	535	797	29	118	7	4
Sig. effects (F)	0.281	0.440	0.193	0.059	0.700	0.100	0.241	0.047	0.361	0.256	0.822	0.187
SED	66.5	59.2	6.5	14.3	15.9	53.2	38.6	57.8	15.9	49.5	8.1	3.930

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A9. Effects of ammonium nitrate, foliar urea and soil moisture* on Green Area Indices of cultivar Gerald grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	Irrigation	Total Green Area Index				Green Leaf Area Index				Green Stem Area Index				Green Ear Area Index			
			GS 65		Mid-senescence		GS 65		Mid-senescence		GS 65		Mid-senescence		GS 65		Mid-senescence	
			Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
0	0	Nil	5.2	4.16	1.1	0.91	3.5	2.42	0.4	0.25	1.2	1.04	0.5	0.36	0.5	0.70	0.1	0.30
		Early	5.1	3.59	1.1	0.71	3.5	2.25	0.5	0.05	1.1	0.84	0.5	0.36	0.5	0.50	0.1	0.30
		Late	4.4	5.48	1.0	1.14	2.9	3.32	0.4	0.33	1.1	1.46	0.5	0.47	0.4	0.70	0.1	0.35
0	60	Nil	-	-	1.2	1.53	-	-	0.5	0.74	-	-	0.6	0.44	-	-	0.1	0.35
		Early	-	-	1.7	1.86	-	-	1.0	0.93	-	-	0.6	0.57	-	-	0.1	0.36
		Late	-	-	2.1	1.24	-	-	1.3	0.47	-	-	0.6	0.46	-	-	0.1	0.31
40	0	Nil	6.9	4.08	2.3	1.09	4.7	1.99	1.0	0.21	1.6	1.24	0.9	0.46	0.6	0.85	0.3	0.42
		Early	7.1	5.25	3.7	0.92	4.9	2.99	2.3	0.08	1.7	1.39	1.0	0.47	0.5	0.88	0.4	0.36
		Late	7.3	5.62	3.2	1.00	5.0	3.86	1.8	0.14	1.7	1.27	1.1	0.48	0.6	0.49	0.3	0.39
40	60	Nil	-	-	3.1	1.35	-	-	1.9	0.48	-	-	1.0	0.52	-	-	0.2	0.35
		Early	-	-	3.7	1.62	-	-	2.1	0.45	-	-	1.3	0.74	-	-	0.3	0.43
		Late	-	-	2.3	1.59	-	-	1.4	0.48	-	-	0.7	0.65	-	-	0.1	0.47
100	0	Nil	7.8	5.71	3.2	0.99	5.3	3.84	1.8	0.09	1.9	1.32	1.0	0.48	0.6	0.55	0.4	0.43
		Early	7.8	7.22	3.8	1.10	5.2	4.90	2.4	0.18	1.9	1.77	1.1	0.52	0.7	0.56	0.6	0.40
		Late	7.9	5.56	2.9	1.01	5.2	3.14	1.6	0.01	2.0	1.79	0.9	0.78	0.7	0.63	0.5	0.49
100	60	Nil	-	-	3.6	1.20	-	-	2.1	0.16	-	-	1.2	0.59	-	-	0.4	0.45
		Early	-	-	4.5	1.24	-	-	2.2	0.15	-	-	1.4	0.68	-	-	0.6	0.42
		Late	-	-	4.1	1.22	-	-	2.2	0.04	-	-	1.2	0.69	-	-	0.6	0.49
Means																		
Grand mean			6.6	5.19	2.7	1.21	4.5	3.19	1.5	0.29	1.6	1.35	0.9	0.54	0.6	0.65	0.3	0.39
Ammonium nitrate		0	4.9	4.41	1.3	1.23	3.3	2.66	0.7	0.46	1.1	1.11	0.6	0.45	0.5	0.63	0.1	0.33
		40	7.1	4.99	3.1	1.26	4.9	2.95	1.8	0.31	1.7	1.3	1.0	0.55	0.6	0.74	0.3	0.41
		100	7.8	6.16	3.6	1.13	5.3	3.96	2.0	0.1	1.9	1.63	1.1	0.62	0.7	0.58	0.5	0.45
Foliar urea		0	-	-	2.5	0.99	-	-	1.4	0.15	-	-	0.9	0.49	-	-	0.3	0.38
		60	-	-	2.9	1.43	-	-	1.6	0.43	-	-	1.0	0.59	-	-	0.3	0.41
Irrigation		Nil	6.7	4.65	2.4	1.18	4.5	2.75	1.3	0.32	1.6	1.2	0.9	0.48	0.6	0.7	0.2	0.38
		Early	6.7	5.35	3.0	1.24	4.6	3.38	1.7	0.31	1.6	1.33	1.0	0.56	0.6	0.65	0.3	0.38
		Late	6.5	5.55	2.6	1.20	4.4	3.44	1.5	0.24	1.6	1.51	0.8	0.59	0.6	0.61	0.3	0.42
Significance of effects (F)																		
Ammonium nitrate (AN)			<0.001	0.009	<0.001	0.393	<0.001	0.004	<0.001	<0.001	<0.001	0.023	<0.001	0.005	<0.001	0.531	<0.001	<0.001
Foliar urea (FU)			-	-	0.066	<0.001	-	-	0.097	<0.001	-	-	0.092	0.018	-	-	0.983	0.269
Irrigation (I)			0.911	0.299	0.053	0.868	0.728	0.182	0.077	0.697	0.913	0.472	0.117	0.114	0.810	0.924	0.008	0.448
AN x FU			-	-	0.369	0.090	-	-	0.643	0.005	-	-	0.166	0.592	-	-	0.093	0.957
AN x I			0.833	0.096	0.558	0.988	0.802	0.013	0.637	0.864	0.895	0.329	0.663	0.617	0.311	0.492	0.095	0.783
FU x I			-	-	0.961	0.163	-	-	0.553	0.260	-	-	0.421	0.197	-	-	0.810	0.564
AN x FU x I			-	-	0.234	0.145	-	-	0.227	0.169	-	-	0.178	0.786	-	-	0.943	0.327
Standard errors (SED)																		
AN			0.455	0.497	0.264	0.101	0.323	0.34	0.200	0.070	0.131	0.168	0.072	0.523	0.037	0.144	0.047	0.024
FU			-	-	0.216	0.082	-	-	0.163	0.057	-	-	0.059	0.043	-	-	0.379	0.020
I			0.330	0.549	0.208	0.118	0.240	0.354	0.162	0.093	0.103	0.237	0.065	0.046	0.020	0.239	0.019	0.030
AN x FU			-	-	0.373	0.142	-	-	0.283	0.099	-	-	0.102	0.074	-	-	0.066	0.034
AN x I			0.723	0.893	0.427	0.185	0.516	0.597	0.326	0.135	0.212	0.336	0.122	0.087	0.055	0.314	0.068	0.046
FU x I			-	-	0.336	0.155	-	-	0.257	0.116	-	-	0.098	0.070	-	-	0.050	0.039
AN x FU x I			-	-	0.626	0.254	-	-	0.476	0.1816	-	-	0.175	0.126	-	-	0.106	0.062

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A10. Effects of ammonium nitrate and foliar urea on Green Area Indices of cultivar Image grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	Total Green Area Index			Green Leaf Area Index			Green Stem Area Index			Green Ear Area Index		
		GS 65		Mid-sen.	GS 65		Mid-sen.	GS 65		Mid-sen.	GS 65		Mid-sen.
		Year 1	Year 2	Year 2	Year 1	Year 2	Year 2	Year 1	Year 2	Year 2	Year 1	Year 2	Year 2
0	0	4.7	3.6	0.5	2.9	2.0	0.0	1.3	1.2	0.2	0.5	0.5	0.2
	60	-	-	1.1	-	-	0.1	-	-	0.6	-	-	0.4
40	0	7.9	3.6	0.7	5.0	1.8	0.0	2.1	1.2	0.3	0.8	0.6	0.3
	60	-	-	0.9	-	-	0.0	-	-	0.5	-	-	0.4
100	0	9.2	4.2	0.9	6.0	2.1	0.0	2.3	1.4	0.5	0.9	0.7	0.4
	60	-	-	0.7	-	-	0.0	-	-	0.3	-	-	0.4
<u>Means</u>					-			-			-		
Grand mean		7.3	3.8	0.8	4.6	2.0	0.0	1.9	1.3	0.4	0.8	0.6	0.4
Ammonium	0	4.7	3.6	0.8	2.9	2.0	0.0	1.3	1.2	0.4	0.5	0.5	0.3
nitrate	40	7.9	3.6	0.8	5.0	1.8	0.0	2.1	1.2	0.4	0.8	0.6	0.4
	100	9.2	4.2	0.8	6.0	2.1	0.0	2.3	1.4	0.4	0.9	0.7	0.4
Foliar urea	0	-	-	0.7	-	-	0.0	-	-	0.3	-	-	0.3
	60	-	-	0.9	-	-	0.0	-	-	0.5	-	-	0.4
<u>Significance of effects</u>													
Ammonium nitrate (AN)		<0.001	0.682	0.988	<0.001	0.801	0.398	<0.001	0.650	0.913	0.005	0.191	0.118
Foliar urea (FU)		-	-	0.089	-	-	0.346	-	-	0.180	-	-	0.033
AN x FU		-	-	0.073	-	-	0.376	-	-	0.127	-	-	0.074
<u>Standard errors (SED)</u>													
AN		0.265	0.769	0.161	0.235	0.471	0.032	0.119	0.292	0.126	0.076	0.071	0.037
FU		-	-	0.131	-	-	0.026	-	-	0.103	-	-	0.031
AN x FU		-	-	0.227	-	-	0.045	-	-	0.178	-	-	0.053

Data are means of four replicate plots

Table A11. Effects of ammonium nitrate and foliar urea on Green Area Indices of cultivar Gerald grown at Gleadthorpe

Ammonium nitrate kg/ha	Foliar urea kg/ha	Total Green Area Index				Green Leaf Area Index				Green Stem Area Index				Green Ear Area Index			
		GS 65		Mid-senescence		GS 65		Mid-senescence		GS 65		Mid-senescence		GS 65		Mid-senescence	
		Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
40	0	2.2	5.2	0.0	0.1	0.9	3.2	0.0	0.0	0.8	1.3	0.0	0.0	0.6	0.8	0.0	0.0
	60	-	-	0.2	0.2	-	-	0.1	0.1	-	-	0.1	0.1	-	-	0.0	0.0
100	0	2.9	7.0	0.1	0.5	1.3	4.1	0.0	0.1	1.0	1.9	0.1	0.3	0.7	1.0	0.0	0.0
	60	-	-	0.5	0.4	-	-	0.2	0.2	-	-	0.3	0.2	-	-	0.0	0.1
140	0	3.5	7.2	0.4	0.9	1.6	4.2	0.1	0.4	1.1	2.0	0.2	0.3	0.7	1.0	0.1	0.1
	60	-	-	1.1	1.2	-	-	0.4	0.3	-	-	0.6	0.7	-	-	0.2	0.2
Means																	
Grand mean		2.9	6.5	0.4	0.5	1.3	3.8	0.1	0.2	1.0	1.7	0.2	0.3	0.6	1.0	0.1	0.1
Ammonium	40	2.2	5.2	0.1	0.1	0.9	3.2	0.0	0.1	0.8	1.3	0.1	0.0	0.6	0.8	0.0	0.0
nitrate	100	2.9	7.0	0.3	0.4	1.3	4.1	0.1	0.1	1.0	1.9	0.2	0.2	0.7	1.0	0.0	0.0
	140	3.5	7.2	0.8	1.0	1.6	4.2	0.2	0.3	1.1	2.0	0.4	0.5	0.7	1.0	0.1	0.2
Foliar urea	0	-	-	0.2	0.5	-	-	0.0	0.2	-	-	0.1	0.2	-	-	0.0	0.0
	60	-	-	0.6	0.6	-	-	0.2	0.2	-	-	0.3	0.3	-	-	0.1	0.1
Significance of effects (F)																	
Ammonium nitrate (AN)		0.009	0.015	<0.001	<0.001	0.005	0.024	0.002	0.068	0.057	0.008	<0.001	<0.001	0.029	0.027	0.001	0.004
Foliar urea (FU)		-	-	<0.001	0.376	-	-	<0.001	0.928	-	-	<0.001	0.056	-	-	0.008	0.170
AN x FU		-	-	0.105	0.661	-	-	0.167	0.478	-	-	0.209	0.015	-	-	0.032	0.325
Standard errors (SED)																	
AN		0.254	0.501	0.117	0.190	0.142	0.307	0.049	0.111	0.089	0.140	0.059	0.069	0.046	0.070	0.025	0.043
FU		-	-	0.096	0.155	-	-	0.040	0.090	-	-	0.048	0.056	-	-	0.021	0.035
AN x FU		-	-	0.165	0.269	-	-	0.069	0.156	-	-	0.083	0.098	-	-	0.036	0.060

Data are means of four replicate plots

Table A12. Effects of soil moisture* on Green Area Indices of cultivar Gerald grown at Gleadthorpe

Irrigation	Total Green Area Index				Green Leaf Area Index				Green Stem Area Index				Green Ear Area Index			
	GS 65		Mid-senescence		GS 65		Mid-senescence		GS 65		Mid-senescence		GS 65		Mid-senescence	
	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
Nil	2.7	7.6	0.2	1.6	1.1	4.7	0.0	0.4	0.9	1.9	0.1	0.7	0.6	1.0	0.0	0.5
Early	4.0	7.5	0.4	3.4	1.8	4.7	0.1	1.2	1.3	2.0	0.3	1.4	0.9	0.9	0.1	0.8
Late	3.0	8.1	0.3	6.0	1.1	5.1	0.1	3.1	1.1	2.0	0.2	1.9	0.7	1.0	0.1	0.9
Throughout	-	8.4	-	5.3	-	5.3	-	2.7	-	2.1	-	1.6	-	1.0	-	1.0
Grand mean	3.2	7.9	0.3	4.1	1.4	4.9	0.1	1.9	1.1	2.0	0.2	1.4	0.8	0.9	0.1	0.8
Sig. effects (F)	0.010	0.138	0.248	<0.001	0.004	0.166	0.924	<0.001	0.039	0.339	0.038	<0.001	0.011	0.337	0.329	0.041
SED	0.285	0.388	0.096	0.493	0.133	0.265	0.038	0.288	0.111	0.118	0.044	0.172	0.051	0.054	0.026	0.168

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A13. Effects of ammonium nitrate, foliar urea and soil moisture* on intercepted, relected and absorbed solar radiation for cultivar Gerald at Rosemaund in Year 1.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Irrigation	Intercepted radiation, %				Reflected radiation, %				Absorbed radiation, %			
			30/05/97	13/06/97	04/07/97	25/07/97	30/05/97	13/06/97	04/07/97	25/07/97	30/05/97	13/06/97	04/07/97	25/07/97
0	0	Nil	97.9	93.6	96.3	88.6	3.0	3.8	3.0	6.7	94.8	89.9	93.2	82.0
		Early	-	92.7	95.3	83.9	-	3.5	3.1	6.4	-	89.2	92.2	77.4
		Late	-	-	93.6	80.7	-	-	3.4	6.7	-	-	90.2	74.0
0	60	Nil	-	-	96.9	86.1	-	-	2.7	7.0	-	-	94.2	79.1
		Early	-	-	95.4	87.5	-	-	2.6	6.4	-	-	92.8	81.1
		Late	-	-	96.0	87.5	-	-	2.9	6.5	-	-	93.1	81.0
40	0	Nil	97.9	98.0	98.5	92.3	3.0	3.1	2.9	7.1	94.9	94.8	95.6	85.2
		Early	-	97.4	99.0	95.3	-	3.3	2.5	6.8	-	94.2	96.5	88.6
		Late	-	-	96.5	88.4	-	-	2.9	6.7	-	-	93.5	81.7
40	60	Nil	-	-	98.4	93.4	-	-	2.4	7.0	-	-	96.0	86.3
		Early	-	-	98.5	95.5	-	-	2.8	7.1	-	-	95.7	88.4
		Late	-	-	97.9	93.9	-	-	2.9	6.5	-	-	95.0	87.4
100	0	Nil	98.9	98.7	99.3	95.3	2.7	3.3	2.3	6.8	96.2	95.4	97.0	88.5
		Early	-	97.3	99.4	96.7	-	3.1	3.1	6.5	-	94.2	96.3	90.2
		Late	-	-	99.4	96.5	-	-	2.6	6.5	-	-	96.8	90.0
100	60	Nil	-	-	99.7	96.5	-	-	2.3	6.7	-	-	97.4	89.8
		Early	-	-	99.5	96.2	-	-	2.4	6.0	-	-	97.1	90.3
		Late	-	-	99.7	96.3	-	-	2.6	5.8	-	-	97.1	90.5
Means														
Grand mean			98.23	96.28	97.7	91.7	2.9	3.3	2.8	6.6	95.3	92.9	95.0	85.1
Ammonium nitrate		0	97.87	93.16	95.6	85.7	3.0	3.6	3.0	6.6	94.8	89.5	92.6	79.1
		40	97.92	97.7	98.1	93.1	3.0	3.2	2.8	6.9	94.9	94.5	95.4	86.3
		100	98.9	97.99	99.5	96.3	2.7	3.2	2.5	6.4	96.2	94.8	97.0	89.9
Foliar urea		0	-	-	97.5	90.9	-	-	2.9	6.7	-	-	94.6	84.2
		60	-	-	98.0	92.6	-	-	2.6	6.6	-	-	95.4	86.0
Irrigation		Nil	-	96.76	98.2	92.0	-	3.4	2.6	6.9	-	93.4	95.6	85.2
		Early	-	95.8	97.9	92.5	-	3.3	2.8	6.5	-	92.5	95.1	86.0
		Late	-	-	97.2	90.6	-	-	2.9	6.5	-	-	94.3	84.1
Significance of effects (F)														
Ammonium nitrate (AN)			0.314	<0.001	<0.001	<0.001	0.093	0.008	0.001	0.007	0.246	<0.001	<0.001	<0.001
Foliar urea (FU)			-	-	0.083	0.020	-	-	0.007	0.341	-	-	0.010	0.013
Irrigation (I)			-	0.344	0.151	0.135	-	0.741	0.490	0.030	-	0.421	0.091	0.197
AN x FU			-	-	0.476	0.319	-	-	0.299	0.172	-	-	0.254	0.490
AN x I			-	0.823	0.128	0.133	-	0.270	0.387	0.564	-	0.893	0.092	0.143
FU x I			-	-	0.140	0.057	-	-	0.903	0.443	-	-	0.169	0.033
AN x FU x I			-	-	0.700	0.115	-	-	0.094	0.815	-	-	0.470	0.093
Standard errors (SED)														
AN			0.69	0.702	0.363	0.866	0.154	0.128	0.111	0.152	0.833	0.711	0.358	0.859
FU			-	-	0.296	0.707	-	-	0.090	0.124	-	-	0.293	0.701
I			-	0.86	0.442	0.856	-	0.292	0.215	0.125	-	0.921	0.473	0.921
AN x FU			-	-	0.513	1.224	-	-	0.156	0.215	-	-	0.507	1.215
AN x I			-	1.182	0.677	1.494	-	0.327	0.266	0.249	-	1.234	0.693	1.525
FU x I			-	-	0.572	1.218	-	-	0.242	0.197	-	-	0.593	1.260
AN x FU x I			-	-	0.923	2.117	-	-	0.328	0.363	-	-	0.930	2.130

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A14. Effects of ammonium nitrate, foliar urea and soil moisture* on intercepted, relected and absorbed solar radiation for cultivar Gerald at Rosemaund in Year 2.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Irrigation	Intercepted radiation, %						Reflected radiation, %						Absorbed radiation, %					
			30/04/98	13/05/98	04/06/98	17/06/98	08/07/98	21/07/98	30/04/98	13/05/98	04/06/98	17/06/98	08/07/98	21/07/98	30/04/98	13/05/98	04/06/98	17/06/98	08/07/98	21/07/98
0	0	Nil	89.3	86.0	79.7	86.3	87.5	61.6	4.7	4.7	4.1	4.4	4.4	7.3	84.6	81.3	75.6	81.9	83.1	54.3
		Early	-	-	86.2	90.9	84.3	53.9	-	-	4.1	4.5	4.8	7.0	-	-	82.1	86.4	79.5	46.9
		Late	-	-	-	-	83.6	54.3	-	-	-	-	4.4	6.7	-	-	-	-	79.2	47.6
0	60	Nil	-	-	-	-	88.9	57.0	-	-	-	-	3.8	7.2	-	-	-	-	85.1	49.9
		Early	-	-	-	-	83.1	54.6	-	-	-	-	3.9	6.7	-	-	-	-	79.1	47.9
		Late	-	-	-	-	85.3	52.7	-	-	-	-	4.3	6.5	-	-	-	-	81.1	46.2
40	0	Nil	89.1	87.3	87.6	91.7	86.5	51.8	4.0	4.4	3.6	4.2	4.3	6.2	85.2	82.9	84.1	87.5	82.2	45.6
		Early	-	-	86.9	85.1	89.7	61.8	-	-	3.6	4.4	4.2	7.0	-	-	83.3	80.6	85.4	54.8
		Late	-	-	-	-	89.1	58.1	-	-	-	-	4.9	6.7	-	-	-	-	84.2	51.4
40	60	Nil	-	-	-	-	90.5	54.6	-	-	-	-	3.4	6.8	-	-	-	-	87.0	47.9
		Early	-	-	-	-	88.7	61.2	-	-	-	-	3.8	6.8	-	-	-	-	85.0	54.5
		Late	-	-	-	-	90.4	58.4	-	-	-	-	3.7	6.7	-	-	-	-	86.7	51.9
100	0	Nil	89.9	90.4	90.6	94.0	95.0	58.6	4.9	3.8	3.4	3.9	3.9	6.7	85.0	86.6	87.3	90.1	81.0	52.0
		Early	-	-	93.3	91.9	95.9	59.0	-	-	3.3	5.2	3.9	6.5	-	-	90.0	86.7	92.0	52.5
		Late	-	-	-	-	94.5	55.8	-	-	-	-	3.8	7.5	-	-	-	-	90.7	48.3
100	60	Nil	-	-	-	-	95.5	59.2	-	-	-	-	3.6	6.9	-	-	-	-	97.9	52.3
		Early	-	-	-	-	95.0	59.7	-	-	-	-	3.7	6.5	-	-	-	-	91.3	53.2
		Late	-	-	-	-	95.0	58.4	-	-	-	-	3.6	6.7	-	-	-	-	91.4	51.6
Means																				
Grand mean			89.4	87.9	87.4	90.0	89.9	57.3	4.5	4.3	3.7	4.4	4.0	6.8	84.9	83.6	83.7	85.5	85.9	50.5
Ammonium nitrate		0	89.3	86.0	82.9	88.6	85.5	55.7	4.7	4.7	4.1	4.5	4.3	6.9	84.6	81.3	78.9	84.2	81.2	48.8
		40	89.1	87.3	87.3	88.4	89.1	57.7	4.0	4.4	3.6	4.3	4.1	6.7	85.2	82.9	83.7	84.1	85.1	51.0
		100	89.9	90.4	92.0	92.9	95.1	58.5	4.9	3.8	3.3	4.6	3.8	6.8	85.0	86.6	88.6	88.4	91.4	51.6
Foliar urea		0	-	-	-	-	89.6	57.2	-	-	-	-	4.3	6.8	-	-	-	-	85.3	50.4
		60	-	-	-	-	90.3	57.3	-	-	-	-	3.8	6.8	-	-	-	-	86.5	50.6
Irrigation		Nil	-	-	86.0	90.7	90.6	57.1	-	-	3.7	4.2	3.9	6.8	-	-	82.3	86.5	86.7	50.3
		Early	-	-	88.8	89.3	89.5	58.4	-	-	6.7	4.7	4.1	6.8	-	-	85.2	84.6	85.4	51.6
		Late	-	-	-	-	89.6	56.3	-	-	-	-	4.1	6.8	-	-	-	-	85.5	49.5
Significance of effects (F)																				
Ammonium nitrate (AN)			0.956	0.021	<0.001	0.103	<0.001	0.514	0.538	0.005	0.005	0.753	<0.001	0.397	0.983	0.009	<0.001	0.113	<0.001	0.463
Foliar urea (FU)			-	-	-	-	0.301	0.955	-	-	-	-	<0.001	0.520	-	-	-	-	0.071	0.919
Irrigation (I)			-	-	0.521	0.273	0.687	0.750	-	-	0.762	0.293	0.230	0.949	-	-	0.513	0.075	0.636	0.732
AN x FU			-	-	-	-	0.704	0.789	-	-	-	-	0.005	0.543	-	-	-	-	0.487	0.795
AN x I			-	-	0.143	0.071	0.060	0.237	-	-	0.902	0.161	0.114	0.025	-	-	0.140	0.058	0.053	0.267
FU x I			-	-	-	-	0.185	0.984	-	-	-	-	0.911	0.198	-	-	-	-	0.175	0.954
AN x FU x I			-	-	-	-	0.904	0.960	-	-	-	-	0.024	0.558	-	-	-	-	0.901	0.960
Standard errors (SED)																				
AN			2.81	1.13	1.67	2.17	0.83	2.44	0.85	0.17	0.19	0.33	0.08	0.16	3.30	1.13	1.69	2.13	0.82	2.37
FU			-	-	-	-	0.68	1.99	-	-	-	-	0.07	0.13	-	-	-	-	0.67	1.94
I			-	-	3.89	1.03	1.42	2.69	-	-	0.10	0.43	0.11	0.27	-	-	3.86	0.72	1.49	2.67
AN x FU			-	-	-	-	1.17	3.44	-	-	-	-	0.12	0.22	-	-	-	-	1.16	3.35
AN x I			-	-	4.34	1.46	1.84	4.37	-	-	2.39	0.57	0.16	0.35	-	-	4.32	2.57	1.89	4.29
FU x I			-	-	-	-	1.64	3.63	-	-	-	-	0.14	0.32	-	-	-	-	1.70	3.57
AN x FU x I			-	-	-	-	2.34	6.07	-	-	-	-	0.22	0.45	-	-	-	-	2.36	5.94

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A15. Effects of ammonium nitrate and foliar urea on intercepted, relected and absorbed solar radiation for cultivar Image at Rosemaund in Year 1.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Intercepted radiation, % 30/05/97	Intercepted radiation, % 13/06/97	Reflected radiation, % 30/05/97	Reflected radiation, % 13/06/97	Absorbed radiation, % 30/05/97	Absorbed radiation, % 13/06/97
0	0	97.4	96.3	3.6	3.5	94.0	92.9
	60	-	-	-	-	-	-
40	0	98.7	98.5	2.8	2.9	95.9	95.6
	60	-	-	-	-	-	-
100	0	98.8	99.1	2.8	2.6	96.1	96.5
	60	-	-	-	-	-	-
Means							
Grand mean		98.3	98.0	3.1	3.0	95.3	95.0
Ammonium	0	97.4	96.3	3.6	3.5	94.0	92.9
nitrate	40	98.7	98.5	2.8	2.9	95.9	95.6
	100	98.8	99.1	2.8	2.6	96.1	96.5
Significance of effects							
Ammonium nitrate (AN)		0.075	0.008	<0.001	0.003	0.011	0.001
Foliar urea (FU)		-	-	-	-	-	-
AN x FU		-	-	-	-	-	-
Standard errors (SED)							
AN		0.57	0.60	0.05	0.14	0.55	0.52
FU		-	-	-	-	-	-
AN x FU		-	-	-	-	-	-

Data are means of four replicate plots

Table A16. Effects of ammonium nitrate and foliar urea on intercepted, relected and absorbed solar radiation for cultivar Image grown at Rosemaund in Year 2.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Intercepted radiation, %						Reflected radiation, %						Absorbed radiation, %					
		30/04/98	13/05/98	04/06/98	17/06/98	10/07/98	22/07/98	30/04/98	13/05/98	04/06/98	17/06/98	10/07/98	22/07/98	30/04/98	13/05/98	04/06/98	17/06/98	10/07/98	22/07/98
0	0	91.8	90.5	85.5	78.8	54.3	91.5	4.2	5.3	4.3	4.5	6.3	8.1	87.5	85.2	81.2	74.3	48.0	83.4
	60	-	-	-	-	60.5	91.0	-	-	-	-	5.2	7.5	-	-	-	-	55.3	83.5
40	0	91.4	92.9	83.6	84.7	64.1	90.3	3.8	5.1	3.9	4.3	5.7	8.0	87.6	87.8	79.7	80.4	58.4	82.3
	60	-	-	-	-	67.1	92.7	-	-	-	-	5.7	7.2	-	-	-	-	61.5	85.5
100	0	94.6	94.9	90.4	86.7	71.7	89.3	3.5	4.6	3.1	3.9	5.2	8.2	91.1	90.3	87.3	82.8	66.6	81.1
	60	-	-	-	-	70.0	90.3	-	-	-	-	4.8	7.5	-	-	-	-	65.2	82.8
Means																			
Grand mean		92.6	92.8	86.5	83.4	64.6	90.8	3.9	5.0	3.7	4.2	5.5	7.8	88.8	87.7	82.7	79.2	59.2	83.1
Ammonium	0	91.8	80.5	85.5	78.8	57.4	91.2	4.2	5.3	4.3	4.5	5.8	7.8	87.5	85.2	81.2	74.3	51.7	83.5
nitrate	40	91.4	92.9	83.6	84.7	65.6	91.5	3.8	5.1	3.9	4.3	5.7	7.6	87.6	87.8	79.7	80.4	59.9	83.9
	100	94.6	94.9	90.4	86.7	70.9	89.8	3.5	4.6	3.1	3.9	5.0	7.9	91.1	90.3	87.3	82.8	65.9	81.9
Foliar urea	0	-	-	-	-	63.4	90.4	-	-	-	-	5.7	8.1	-	-	-	-	57.7	82.3
	60	-	-	-	-	65.9	91.3	-	-	-	-	5.2	7.4	-	-	-	-	60.7	83.9
Significance of effects																			
Ammonium nitrate (AN)		0.498	0.069	0.316	0.214	0.017	0.449	0.001	0.039	<0.001	0.016	0.601	0.867	0.413	0.060	0.236	0.182	0.012	0.306
Foliar urea (FU)		-	-	-	-	0.467	0.419	-	-	-	-	0.463	0.121	-	-	-	-	0.375	0.137
AN x FU		-	-	-	-	0.626	0.301	-	-	-	-	0.801	0.943	-	-	-	-	0.569	0.242
Standard errors (SED)																			
AN		2.79	1.50	4.08	3.98	4.03	1.37	0.10	0.20	0.12	0.14	0.85	0.50	2.83	1.65	4.04	3.96	3.99	1.24
FU		-	-	-	-	3.29	1.12	-	-	-	-	0.70	0.41	-	-	-	-	3.26	1.01
AN x FU		-	-	-	-	5.70	1.94	-	-	-	-	1.21	0.70	-	-	-	-	5.65	1.75

Data are means of four replicate plots
SED (AN x FU)

Table A17. Effects of ammonium nitrate, foliar urea and soil moisture* in crop dry matters in cultivar Gerald grown at Rosemaund in Year 1.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Irrigation	Total d.m, t/ha		Straw d.m, t/ha		Ear d.m, t/ha	
			GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.
0	0	Nil	11.4	13.3	8.6	7.1	2.8	6.3
		Early	9.9	13.1	7.5	7.0	2.5	6.1
		Late	10.2	11.4	7.7	6.8	2.5	4.6
0	60	Nil	-	14.3	-	7.5	-	6.8
		Early	-	11.4	-	5.9	-	5.5
		Late	-	12.5	-	6.5	-	6.0
40	0	Nil	11.6	14.6	8.7	7.6	2.9	7.1
		Early	12.2	14.5	9.1	7.8	3.0	6.7
		Late	12.8	14.9	9.6	7.9	3.2	7.0
40	60	Nil	-	14.6	-	7.7	-	6.9
		Early	-	16.8	-	8.9	-	8.0
		Late	-	14.0	-	7.2	-	6.8
100	0	Nil	12.3	14.3	9.1	7.5	3.3	6.8
		Early	11.9	14.7	8.7	7.5	3.2	7.2
		Late	12.9	13.5	9.4	7.2	3.5	6.4
100	60	Nil	-	15.2	-	7.6	-	7.7
		Early	-	15.6	-	8.4	-	7.2
		Late	-	16.3	-	8.5	-	7.8
Means								
Grand mean			11.7	14.2	8.7	7.5	3.0	6.7
Ammonium nitrate		0	10.5	12.7	7.9	6.8	2.6	5.9
		40	12.2	14.9	9.1	7.9	3.0	7.1
		100	12.4	14.9	9.0	7.8	3.3	7.2
Foliar urea		0	-	13.8	-	7.4	-	6.5
		60	-	14.5	-	7.6	-	7.0
Irrigation		Nil	11.8	14.4	8.8	7.5	3.0	6.9
		Early	11.3	14.4	8.4	7.6	2.9	6.8
		Late	12.0	13.8	8.9	7.4	3.1	6.4
Significance of effects (F)								
Ammonium nitrate (AN)			0.025	<0.001	0.055	<0.001	0.002	<.001
Foliar urea (FU)			-	0.094	-	0.325	-	0.054
Irrigation (I)			0.574	0.382	0.616	0.647	0.491	0.202
AN x FU			-	0.362	-	0.106	-	0.756
AN x I			0.604	0.244	0.619	0.121	0.617	0.475
FU x I			-	0.888	-	0.954	-	0.587
AN x FU x I			-	0.110	-	0.080	-	0.167
Standard errors (SED)								
AN			0.67	0.51	0.52	0.25	0.17	0.31
FU			-	0.42	-	0.21	-	0.26
I			0.60	0.45	0.49	0.24	0.12	0.24
AN x FU			-	0.72	-	0.36	-	0.44
AN x I			1.12	0.85	0.88	0.43	0.27	0.50
FU x I			-	0.68	-	0.35	-	0.39
AN x FU x I			-	1.23	-	0.62	-	0.74

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A18. Effects of ammonium nitrate, foliar urea and soil moisture* in crop dry matters in cultivar Gerald grown at Rosemaund in Year 2.

Ammoniu nitrate kg/ha	Foliar urea kg/ha	Irrigation	Total d.m, t/ha		Straw d.m, t/ha		Ear d.m, t/ha	
			GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.
0	0	Nil	12.3	11.5	9.3	6.2	3.1	5.5
		Early	12.0	13.3	9.0	7.1	3.1	6.2
		Late	13.2	15.0	9.9	8.2	3.3	6.9
0	60	Nil	-	14.0	-	7.3	-	6.7
		Early	-	13.6	-	7.0	-	6.6
		Late	-	12.7	-	6.6	-	6.1
40	0	Nil	11.8	14.9	8.7	7.9	3.0	6.5
		Early	12.7	14.6	9.4	8.1	3.4	6.5
		Late	12.1	15.2	9.0	8.2	3.1	7.0
40	60	Nil	-	14.9	-	8.0	-	6.9
		Early	-	16.5	-	8.6	-	7.9
		Late	-	15.3	-	8.1	-	7.2
100	0	Nil	13.4	16.4	9.9	9.6	3.5	6.8
		Early	15.4	15.5	11.2	8.7	4.2	6.7
		Late	13.9	16.5	10.2	9.6	3.7	6.9
100	60	Nil	-	16.2	-	9.2	-	7.1
		Early	-	15.3	-	8.8	-	6.6
		Late	-	17.0	-	9.5	-	7.5
Means								
Grand mean			13.0	14.9	9.6	8.2	3.4	6.8
Ammonium nitrate		0	12.5	13.3	9.4	7.1	3.2	6.3
		40	12.2	15.1	9.1	8.1	3.2	7.0
		100	14.2	16.2	10.4	9.2	3.8	6.9
Foliar urea		0	-	14.7	-	8.2	-	6.6
		60	-	15.0	-	8.1	-	6.9
Irrigation		Nil	12.5	14.6	9.3	8.0	3.2	6.6
		Early	13.4	14.8	9.9	8.1	3.5	6.8
		Late	13.1	15.3	9.7	8.4	3.4	6.9
Significance of effects (F)								
Ammonium nitrate (AN)			0.036	<0.001	0.079	<0.001	0.003	0.033
Foliar urea (FU)			-	0.409	-	0.773	-	0.089
Irrigation (I)			0.323	0.614	0.485	0.687	0.121	0.626
AN x FU			-	0.772	-	0.839	-	0.686
AN x I			0.641	0.641	0.685	0.624	0.520	0.765
FU x I			-	0.373	-	0.364	-	0.497
AN x FU x I			-	0.328	-	0.483	-	0.273
Standard errors (SED)								
AN			0.77	0.58	0.60	0.33	0.18	0.27
FU			-	0.47	-	0.27	-	0.22
I			0.68	0.71	0.45	0.41	0.13	0.34
AN x FU			-	0.82	-	0.47	-	0.38
AN x I			1.28	1.10	1.00	0.63	0.29	0.51
FU x I			-	0.92	-	0.53	-	0.44
AN x FU x I			-	1.48	-	0.85	-	0.67

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A19. Effects of ammonium nitrate and foliar urea on crop dry matters of cultivar Image grown at Rosemaund in Year 1 (no measurements were made at mid-senescence).

Ammonium nitrate kg/ha	Foliar urea kg/ha	Total d.m t/ha	Straw d.m., t/ha	Ear d.m., t/ha
0	0	13.9	10.7	3.1
	60	-	-	-
40	0	13.9	9.8	4.1
	60	-	-	-
100	0	14.6	10.3	4.3
	60	-	-	-
Means				
Grand mean		14.1	10.3	3.8
Ammonium nitrate	0	13.9	10.7	3.1
	40	13.9	9.8	4.1
	100	14.6	10.3	4.3
Foliar urea	0	-	-	-
	60	-	-	-
Significance of effects				
Ammonium nitrate (AN)		0.456	0.278	<0.001
Foliar urea (FU)		-	-	-
AN x FU		-	-	-
Standard errors (SED)				
AN		0.57	0.52	0.13
FU		-	-	-
AN x FU		-	-	-

Data are means of four replicate plots

Table A20. Effects of ammonium nitrate and foliar urea on crop dry matters of cultivar Image grown at Rosemaund in Year 2

Ammonium nitrate kg/ha	Foliar urea kg/ha	Total d.m, t/ha		Straw d.m, t/ha		Ear d.m, t/ha	
		GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.
0	0	12.5	12.6	9.3	7.2	3.2	5.4
	60	-	14.7	-	8.4	-	6.2
40	0	12.6	14.8	9.2	8.6	3.5	6.1
	60	-	15.7	-	9.0	-	6.7
100	0	13.0	16.6	9.2	10.2	3.8	6.4
	60	-	13.0	-	7.7	-	5.3
Means							
Grand mean		12.7	14.6	9.2	8.5	3.5	6.0
Ammonium nitrate	0	12.5	13.6	9.3	7.8	3.2	5.8
	40	12.6	15.2	9.2	8.8	3.5	6.4
	100	13.0	14.8	9.2	8.9	3.8	5.9
Foliar urea	0	-	14.7	-	8.7	-	6.0
	60	-	14.5	-	8.4	-	6.1
Significance of effects							
Ammonium nitrate (AN)		0.954	0.268	0.994	0.102	0.413	0.414
Foliar urea (FU)		-	0.801	-	0.476	-	0.783
AN x FU		-	0.024	-	0.007	-	0.11
Standard errors (SED)							
AN		1.70	0.97	1.30	0.52	0.59	0.47
FU		-	0.79	-	0.43	-	0.38
AN x FU		-	1.37	-	0.74	-	0.66

Data are means of four replicate plots

Table A21. Effects of ammonium nitrate and foliar urea on crop dry matters of cultivar Gerald grown at Gleadthorpe in Year 1.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Total, t/ha		Leaves, t/ha		Stem, t/ha		Ears, t/ha		Dead leaves, t/ha		Dead stems, t/ha		D & D, t/ha	
		GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.
40	0	7.1	7.0	0.6	0.0	3.6	0.0	2.3	4.0	0.7	0.7	-	2.0	0.0	0.0
	60	-	6.3	-	0.0	-	0.2	-	3.7	-	0.7	-	1.5	-	0.1
100	0	7.8	7.4	0.7	0.0	3.7	0.1	2.8	4.3	0.7	0.8	-	1.9	0.1	0.1
	60	-	7.7	-	0.1	-	0.6	-	4.4	-	0.8	-	1.6	-	0.0
140	0	7.8	8.4	0.8	0.0	3.5	0.5	2.6	4.9	0.6	1.0	-	1.8	0.2	0.1
	60	-	7.4	-	0.1	-	1.0	-	3.8	-	0.8	-	1.0	-	0.2
Means															
Grand mean		7.6	7.4	0.7	0.1	3.6	0.4	2.6	4.2	0.7	0.8	-	1.6	0.1	0.1
Ammonium nitrate	40	7.1	6.6	0.6	0.0	3.6	0.1	2.3	3.8	0.7	0.7	-	1.8	0.0	0.0
	100	7.8	7.5	0.7	0.1	3.7	0.4	2.8	4.4	0.7	0.8	-	1.7	0.1	0.1
	140	7.8	7.9	0.8	0.1	3.5	0.8	2.6	4.4	0.6	0.9	-	1.4	0.2	0.1
Foliar urea	0	-	7.6	-	0.0	-	0.2	-	4.4	-	0.8	-	1.9	-	0.1
	60	-	7.1	-	0.1	-	0.6	-	4.0	-	0.8	-	1.4	-	0.1
Significance of effects (F)															
Ammonium nitrate (AN)		0.285	0.005	0.013	0.126	0.580	<0.001	0.130	0.037	0.211	0.005	-	0.049	0.004	0.015
Foliar urea (FU)		-	0.083	-	0.005	-	<0.001	-	0.021	-	0.245	-	<0.001	-	0.211
AN x FU		-	0.129	-	0.317	-	0.153	-	0.034	-	0.350	-	0.295	-	0.085
Standard errors (SED)															
AN		0.43	0.33	0.06	0.03	0.18	0.09	0.19	0.22	0.09	0.04	-	0.14	0.03	0.03
FU		-	0.27	-	0.03	-	0.07	-	0.18	-	0.04	-	0.11	-	0.02
AN x FU		-	0.46	-	0.05	-	0.13	-	0.31	-	0.06	-	0.19	-	0.04

Data are means of four replicate plots

Table A22. Effects of ammonium nitrate and foliar urea on crop dry matters of cultivar Gerald grown at Gleadthorpe in Year 2.

Ammonium nitrate kg/ha	Foliar urea kg/ha	Total, t/ha		Leaves, t/ha		Stem, t/ha		Ears, t/ha		Dead leaves, t/ha		Dead stems, t/ha		D & D, t/ha	
		GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.
40	0	11.1	10.8	1.4	0.0	5.4	0.0	2.5	5.0	0.7	1.5	1.0	3.9	0.2	0.2
	60	-	11.1	-	0.0	-	0.1	-	5.2	-	1.4	-	4.1	-	0.2
100	0	12.0	13.8	1.7	0.0	5.8	0.4	3.4	7.5	0.4	1.5	0.4	3.9	0.2	0.2
	60	-	14.0	-	0.1	-	0.3	-	8.1	-	1.5	-	4.2	-	0.2
140	0	11.5	13.1	1.7	0.0	5.6	0.4	3.3	7.6	0.4	1.5	0.4	4.0	0.2	0.1
	60	-	15.1	-	0.1	-	0.9	-	8.5	-	1.7	-	4.2	-	0.2
Means															
Grand mean		11.5	13.0	1.6	0.0	5.6	0.4	3.1	7.0	0.5	1.5	0.6	4.0	0.2	0.2
Ammonium nitrate	40	11.1	11.0	1.4	0.0	5.4	0.0	2.5	5.1	0.7	1.5	1.0	4.0	0.2	0.2
	100	12.0	13.9	1.7	0.0	5.8	0.4	3.4	7.8	0.4	1.5	0.4	4.0	0.2	0.2
	140	11.5	14.1	1.7	0.1	5.6	0.7	3.3	8.1	0.4	1.6	0.4	4.1	0.2	0.2
Foliar urea	0	-	12.6	-	0.0	-	0.3	-	6.7	-	1.5	-	3.9	-	0.2
	60	-	13.4	-	0.1	-	0.4	-	7.3	-	1.5	-	4.2	-	0.2
Significance of effects (F)															
Ammonium nitrate (AN)		0.285	<0.001	0.014	0.031	0.460	<0.001	0.003	<0.001	0.010	0.387	0.05	0.882	0.376	0.164
Foliar urea (FU)		-	0.195	-	0.042	-	0.222	-	0.108	-	0.572	-	0.285	-	0.445
AN x FU		-	0.441	-	0.463	-	0.039	-	0.661	-	0.133	-	0.942	-	0.791
Standard errors (SED)															
AN		0.51	0.73	0.09	0.02	0.31	0.11	0.16	0.39	0.06	0.09	0.22	0.25	0.04	0.04
FU		-	0.59	-	0.01	-	0.09	-	0.32	-	0.08	-	0.21	-	0.03
AN x FU		-	1.03	-	0.02	-	0.16	-	0.55	-	0.13	-	0.36	-	0.05

Data are means of four replicate plots

Table A23. Effects of soil moisture* on crop dry matters of cultivar Gerald grown at Gleadthorpe in Year 1.

Irrigation	Total, t/ha		Leaves, t/ha		Stem, t/ha		Ears, t/ha		Dead leaves, t/ha		Dead stems, t/ha		D & D, t/ha	
	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.
Nil	6.6	6.6	0.6	0.0	3.1	0.3	2.3	3.9	0.4	0.7	-	1.6	0.2	0.1
Early	8.2	9.0	0.8	0.0	4.2	0.5	2.8	5.1	0.4	0.8	-	2.5	0.1	0.0
Late	7.3	7.4	0.5	0.0	3.4	0.3	2.7	4.3	0.5	0.8	-	1.7	0.2	0.1
Grand mean	7.4	7.7	0.6	0.0	3.5	0.4	2.6	4.5	0.4	0.8	-	1.9	0.1	0.1
Sig. effects (F)	0.018	0.012	0.017	0.959	0.017	0.084	0.038	0.014	0.203	0.328	-	<0.001	0.550	0.064
SED	0.41	0.56	0.06	0.01	0.40	0.08	0.16	0.29	0.08	0.07	-	0.14	0.07	0.02

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A24. Effects of soil moisture* on crop dry matters of cultivar Gerald grown at Gleadthorpe in Year 2.

Irrigation	Total, t/ha		Leaves, t/ha		Stem, t/ha		Ears, t/ha		Dead leaves, t/ha		Dead stems, t/ha		D & D, t/ha	
	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.	GS 65	Mid-sen.
Nil	11.2	15.3	2.0	0.1	5.5	0.9	2.9	7.7	0.5	1.8	0.4	4.1	0.3	0.2
Early	10.0	16.8	1.8	0.3	5.0	1.9	2.6	8.4	0.4	1.8	0.4	4.2	0.2	0.1
Late	11.2	17.6	2.0	0.9	5.2	3.7	2.8	8.4	0.4	1.4	0.4	2.7	0.3	0.1
Throughout	10.9	16.0	2.0	0.7	5.2	3.0	2.7	8.1	0.5	1.4	0.5	3.0	0.5	0.1
Grand mean	10.8	16.4	1.9	0.5	5.3	2.4	2.8	8.1	0.5	1.6	0.4	3.5	0.3	0.1
Sig. effects (F)	0.177	0.391	0.463	<0.001	0.599	<0.001	0.205	0.733	0.220	0.163	0.91	0.019	0.157	0.230
SED	0.57	1.30	0.10	0.08	0.35	0.23	0.15	0.67	0.06	0.22	0.10	0.45	0.11	0.04

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A25. Effects of ammonium nitrate, foliar urea and soil moisture* on crop height and lodging of cultivar Gerald grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	Irrigation	Crop height cm		Lodging, %		
			Year 1	Year 2	Year 1 08/08/97	Year 2 27/07/98	07/08/98
0	0	Nil	101	101	0	0	0
		Early	107	99	0	0	0
		Late	99	100	0	0	0
0	60	Nil	100	100	0	1	0
		Early	105	94	0	0	0
		Late	101	97	0	0	0
40	0	Nil	110	101	0	0	23
		Early	118	106	0	0	0
		Late	115	102	0	0	1
40	60	Nil	115	106	0	0	30
		Early	119	102	0	0	0
		Late	114	100	0	0	21
100	0	Nil	123	124	19	27	98
		Early	123	113	28	18	98
		Late	121	118	21	39	97
100	60	Nil	122	127	13	60	99
		Early	126	119	36	64	99
		Late	120	125	26	52	99
Means							
Grand mean			113	107	8	14	37
Ammonium nitrate		0	102	98	0	0	0
		40	115	103	0	0	13
		100	122	121	24	43	98
Foliar urea		0	113	107	8	9	35
		60	113	108	8	20	38
Irrigation		Nil	112	110	5	15	41
		Early	116	105	11	14	33
		Late	112	107	8	15	36
Significance of effects (F)							
Ammonium nitrate (AN)			<0.001	<0.001	-	-	-
Foliar urea (FU)			0.598	0.655	-	-	-
Irrigation (I)			0.028	0.461	-	-	-
AN x FU			0.529	0.048	-	-	-
AN x I			0.556	0.599	-	-	-
FU x I			0.974	0.599	-	-	-
AN x FU x I			0.329	0.584	-	-	-
Standard errors (SED)							
AN			1.09	1.60	-	-	-
FU			0.89	1.31	-	-	-
I			1.40	3.58	-	-	-
AN x FU			1.54	2.26	-	-	-
AN x I			2.09	4.24	-	-	-
FU x I			1.78	3.92	-	-	-
AN x FU x I			2.81	5.06	-	-	-

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A26. Effects of ammonium nitrate and foliar urea on crop height and lodging of cultivar Image grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	Crop height cm		Lodging, %				
				Year 1	Year 2			
		Year 1	Year 2	08/08/97	23/06/98	09/07/98	27/07/98	07/08/98
0	0	-	116	0	0	0	0	0
	60	-	120	0	1	2	1	34
40	0	-	128	63	9	8	6	49
	60	-	130	56	0	0	9	98
100	0	-	140	93	49	49	70	91
	60	-	138	87	45	44	85	96
Means								
Grand mean		-	129	50	17	17	28	61
Ammonium	0	-	118	0	0	1	1	17
nitrate	40	-	129	59	4	4	7	73
	100	-	139	90	47	46	77	94
Foliar urea	0	-	128	52	19	19	25	47
	60	-	129	48	15	15	32	76
Significance of effects								
Ammonium nitrate (AN)		-	<0.001	-	-	-	-	-
Foliar urea (FU)		-	0.212	-	-	-	-	-
AN x FU		-	0.136	-	-	-	-	-
Standard errors (SED)								
AN		-	1.3	-	-	-	-	-
FU		-	1.1	-	-	-	-	-
AN x FU		-	1.8	-	-	-	-	-

Data are means of four replicate plots

Table A27. Effects of ammonium nitrate, foliar urea and soil moisture* on stem carbohydrate reserves in cultivar Gerald grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	Irrigation	Stem carbohydrate reserves, t/ha			
			GS 65		Mid-senescence	
			Year 1	Year 2	Year 1	Year 2
0	0	Nil	2.21	1.47	0.29	0.46
		Early	1.92	2.85	0.32	0.43
		Late	2.04	3.07	0.37	0.40
0	60	Nil	-	-	0.50	0.49
		Early	-	-	0.55	0.17
		Late	-	-	0.60	0.39
40	0	Nil	1.77	2.50	0.33	0.87
		Early	1.71	2.34	0.29	0.54
		Late	1.58	2.61	0.56	0.32
40	60	Nil	-	-	0.48	0.35
		Early	-	-	0.50	0.58
		Late	-	-	0.67	0.59
100	0	Nil	0.81	2.68	0.12	0.31
		Early	0.80	2.17	0.33	0.57
		Late	0.96	2.01	0.22	0.36
100	60	Nil	-	-	0.24	0.33
		Early	-	-	0.38	0.54
		Late	-	-	0.38	0.26
Means						
Grand mean			1.55	2.41	0.39	0.44
Ammonium nitrate		0	2.06	2.46	0.44	0.39
		40	1.69	2.48	0.47	0.54
		100	0.86	2.29	0.28	0.39
Foliar urea		0	-	-	0.31	0.47
		60	-	-	0.48	0.41
Irrigation		Nil	1.60	2.22	0.32	0.47
		Early	1.48	2.45	0.40	0.47
		Late	1.53	2.56	0.47	0.39
Significance of effects (F)						
Ammonium nitrate (AN)			<.001	0.882	0.002	0.051
Foliar urea (FU)			-	-	<.001	0.265
Irrigation (I)			0.741	0.674	0.016	0.772
AN x FU			-	-	0.585	0.948
AN x I			0.850	0.263	0.318	0.128
FU x I			-	-	0.996	0.301
AN x FU x I			-	-	0.954	0.034
Standard errors (SED)						
AN			0.167	0.428	0.538	0.067
FU			-	-	0.044	0.055
I			0.154	0.382	0.033	0.129
AN x FU			-	-	0.076	0.095
AN x I			0.282	0.742	0.083	0.175
FU x I			-	-	0.063	0.146
AN x FU x I			-	-	0.125	0.198

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A28. Effects of ammonium nitrate and foliar urea on stem carbohydrate reserves in cultivar Image grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	Stem carbohydrate reserves, t/ha GS 65 Year 1	GS 65 Year 2	Mid-senescence Year 1	Mid-senescence Year 2
0	0	3.37	3.32	0.64	0.65
	60	-	-	0.80	0.66
40	0	1.33	2.28	0.34	0.55
	60	-	-	0.33	0.55
100	0	1.35	1.72	0.52	0.30
	60	-	-	0.56	0.42
Means					
Grand mean		2.02	2.44	0.53	0.52
Ammonium nitrate	0	3.37	3.32	0.72	0.66
	40	1.33	2.28	0.34	0.55
	100	1.35	1.72	0.54	0.36
Foliar urea	0	-	-	0.50	0.50
	60	-	-	0.56	0.54
Significance of effects					
Ammonium nitrate (AN)		0.065	0.003	0.016	0.231
Foliar urea (FU)		-	-	0.507	0.732
AN x FU		-	-	0.761	0.917
Standard errors (SED)					
AN		0.789	0.455	0.116	0.1664
FU		-	-	0.094	0.1359
AN x FU		-	-		0.2354

Data are means of four replicate plots

Table A29. Effects of ammonium nitrate and foliar urea on stem carbohydrate reserves in cultivar Gerald grown at Gleadthorpe

Ammonium nitrate kg/ha	Foliar urea kg/ha	Stem carbohydrate reserves, t/ha			
		GS 65		Mid-senescence	
		Year 1	Year 2	Year 1	Year 2
0	0	5.19	4.80	0.28	0.53
	60	-	-	0.15	1.28
40	0	4.37	2.75	0.15	0.40
	60	-	-	0.15	0.33
100	0	4.41	1.80	0.13	0.50
	60	-	-	0.10	0.20
Means					
Grand mean		4.66	3.12	0.16	0.54
Ammonium nitrate	0	5.19	4.80	0.21	0.90
	40	4.37	2.75	0.15	0.36
	100	4.41	1.80	0.11	0.35
Foliar urea	0	-	-	0.18	0.48
	60	-	-	0.13	0.60
Significance of effects					
Ammonium nitrate (AN)		0.466	0.016	0.002	<0.001
Foliar urea (FU)			-	0.011	0.24
AN x FU			-	0.024	<0.001
Standard errors (SED)					
AN		0.701	0.724	0.022	0.115
FU		-	-	0.018	0.094
AN x FU		-	-	0.03136	0.163

Data are means of four replicate plots

Table A30. Effects of soil moisture* on stem carbohydrate reserves in cultivar Gerald grown at Gleadthorpe

Ammonium nitrate kg/ha	Stem carbohydrate reserves, t/ha			
	GS 65		Mid-senescence	
	Year 1	Year 2	Year 1	Year 2
Nil	2.07	2.10	0.06	0.30
Early	2.49	2.10	0.08	0.60
Late	2.56	2.50	0.04	0.70
Throughout	-	2.10	-	0.80
Grand mean	2.37	2.20	0.06	0.60
Sig. effects (F	0.458	0.663	0.079	0.001
SED	0.392	0.348	0.013	0.078

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A31. Effects of ammonium nitrate, foliar urea and soil moisture* on grain yield and quality of cultivar Gerald grown at Rosemaund

Ammonium nitrate kg N/ha	Foliar urea kg N/ha	Irrigation	Yield t/ha		Specific Weight kg/hl		TGW g		Beta-glucan %		Protein %		Oil %	
			Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
0	0	Nil	7.21	7.66	53.9	53.3	34.6	34.2	2.56	2.54	8.34	7.46	7.53	7.05
		Early	7.22	7.43	54.6	53.9	34.9	35.7	2.43	2.53	8.82	6.79	6.95	7.00
		Late	7.53	7.51	53.4	53.5	35.8	35.3	2.36	2.79	8.70	7.36	7.35	7.38
0	60	Nil	7.69	7.82	54.5	54.1	35.8	37.0	2.59	2.78	9.43	7.92	7.33	6.64
		Early	7.66	7.87	54.3	52.1	34.9	38.5	2.64	2.87	9.26	8.32	7.05	7.16
		Late	7.56	7.84	54.1	54.1	35.0	36.4	2.75	2.55	9.65	7.90	7.23	7.45
40	0	Nil	8.43	7.49	53.3	52.5	33.1	33.7	2.51	2.61	8.96	7.19	7.15	7.33
		Early	8.53	7.96	53.0	51.7	34.1	33.1	2.72	2.56	8.91	7.11	7.30	6.93
		Late	8.31	8.06	53.8	52.3	35.4	34.2	2.67	2.47	9.05	6.97	7.65	7.53
40	60	Nil	8.95	8.18	54.1	52.2	35.2	34.3	2.45	2.74	9.36	8.10	7.30	6.98
		Early	8.98	8.04	52.3	52.4	33.6	34.3	2.57	2.68	9.55	8.17	7.40	7.00
		Late	8.91	8.18	53.4	52.3	35.7	34.9	2.64	2.69	9.62	8.50	7.28	7.15
100	0	Nil	8.73	7.66	50.2	45.1	31.8	27.4	2.64	2.56	9.69	8.03	7.40	7.13
		Early	8.58	7.18	48.9	46.0	31.8	29.7	2.47	2.48	9.27	8.02	7.08	7.03
		Late	8.77	7.34	49.8	45.0	33.1	28.7	2.87	2.37	10.04	7.38	7.53	7.30
100	60	Nil	8.81	7.51	50.4	44.7	33.5	26.8	2.65	2.64	9.90	9.02	7.40	6.13
		Early	8.58	7.15	48.5	45.5	31.5	28.1	2.79	2.54	10.17	8.66	7.33	6.88
		Late	8.58	7.69	49.3	45.5	32.9	28.2	2.89	2.50	10.35	8.16	7.30	7.30
Means														
Grand mean			8.28	7.70	52.3	50.3	34.0	32.8	2.62	2.61	9.39	7.84	7.31	7.07
Ammonium nitrate		0	7.48	7.69	54.1	53.5	35.2	36.2	2.55	2.68	9.03	7.63	7.24	7.11
		40	8.68	7.98	53.3	52.2	34.5	34.1	2.59	2.62	9.24	7.67	7.35	7.15
		100	8.68	7.42	49.5	45.3	32.4	28.1	2.72	2.52	9.90	8.21	7.34	6.96
Foliar urea		0	8.14	7.59	52.3	50.4	33.8	32.4	2.58	2.55	9.09	7.37	7.33	7.18
		60	8.41	7.81	52.3	50.3	34.2	33.2	2.66	2.67	9.70	8.31	7.29	6.96
Irrigation		Nil	8.30	7.72	52.7	50.3	34.0	32.2	2.57	2.65	9.28	7.96	7.35	6.87
		Early	8.26	7.60	51.9	50.3	33.5	33.2	2.60	2.61	9.33	7.84	7.18	7.00
		Late	8.28	7.77	52.3	50.4	34.7	33.0	2.70	2.56	9.57	7.71	7.39	7.35
Significance of effects (F)														
Ammonium nitrate (AN)			<0.001	0.001	<0.001	<0.001	<0.001	<0.001	0.098	0.002	<0.001	0.003	0.393	0.098
Foliar urea (FU)			0.038	0.061	0.952	0.885	0.271	0.145	0.213	0.002	<0.001	<0.001	0.615	0.005
Irrigation (I)			0.969	0.821	0.026	0.976	0.281	0.826	0.107	0.461	0.712	0.443	0.101	0.116
AN x FU			0.190	0.600	0.411	0.914	0.840	0.037	0.194	0.773	0.248	0.545	0.891	0.215
AN x I			0.924	0.481	0.011	0.481	0.704	0.505	0.483	0.560	0.667	0.316	0.156	0.131
FU x I			0.782	0.935	0.121	0.458	0.046	0.917	0.635	0.277	0.909	0.732	0.091	0.004
AN x FU x I			0.948	0.508	0.729	0.344	0.958	0.874	0.624	0.011	0.143	0.367	0.711	0.272
Standard errors (SED)														
AN			0.154	0.140	0.238	0.383	0.430	0.592	0.079	0.044	0.112	0.180	0.087	0.091
FU			0.125	0.114	0.194	0.313	0.351	0.483	0.064	0.036	0.091	0.146	0.071	0.075
I			0.186	0.266	0.217	0.833	0.672	1.653	0.052	0.065	0.365	0.179	0.083	0.197
AN x FU			0.217	0.198	0.337	0.541	0.608	0.837	0.111	0.062	0.158	0.254	0.123	0.129
AN x I			0.286	0.331	0.401	0.994	1.174	1.853	0.123	0.090	0.398	0.311	0.149	0.236
FU x I			0.241	0.300	0.322	0.917	0.798	1.755	0.095	0.078	0.382	0.254	0.120	0.217
AN x FU x I			0.391	0.410	0.575	1.195	0.907	2.117	0.184	0.117	0.442	0.440	0.212	0.284

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Table A32. Effects of ammonium nitrate and foliar urea on grain yield and quality of cultivar Image grown at Rosemaund

Ammonium nitrate kg/ha	Foliar urea kg/ha	Yield t/ha		Specific Weight kg/hl		TGW g		Beta-glucan %		Protein %		Oil %	
		Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
0	0	6.51	6.64	52.4	52.0	39.7	37.0	3.00	3.19	8.22	6.70	7.55	8.48
	60	6.91	6.98	48.9	51.6	39.3	36.7	3.30	3.51	10.03	8.67	7.82	8.30
40	0	7.16	6.55	46.8	49.3	29.9	33.2	3.14	2.94	10.09	6.60	7.58	8.78
	60	8.19	7.38	48.0	50.0	31.1	36.0	3.43	3.42	10.86	9.66	7.5	8.50
100	0	6.31	5.71	42.8	43.3	28.1	29.8	2.86	3.10	12.38	8.34	7.23	7.65
	60	6.67	5.56	43.9	41.1	30.4	26.9	3.26	3.23	12.58	10.00	7.12	7.10
Means													
Grand mean		6.96	6.47	47.1	47.9	33.1	33.3	3.17	3.23	10.69	8.33	7.47	8.13
Ammonium nitrate	0	6.71	6.81	50.6	51.8	39.5	36.9	3.15	3.35	9.12	7.69	7.69	8.39
	40	7.68	6.97	47.4	49.6	30.5	34.6	3.29	3.18	10.47	8.13	7.54	8.64
	100	6.49	5.64	43.3	42.2	29.3	28.4	3.06	3.16	12.48	9.17	7.18	7.37
Foliar urea	0	6.66	6.30	47.3	48.2	32.6	33.3	3.00	3.08	10.23	7.21	7.45	8.30
	60	7.26	6.64	46.9	47.6	33.6	33.2	3.33	3.39	11.15	9.44	7.48	7.97
Significance of effects													
Ammonium nitrate (AN)		<0.001	0.001	0.001	<0.001	<0.001	<0.001	0.241	0.240	<0.001	0.123	0.255	0.078
Foliar urea (FU)		0.015	0.192	0.768	0.402	0.201	0.915	0.006	0.005	0.002	0.001	0.895	0.463
AN x FU		0.338	0.303	0.267	0.273	0.382	0.098	0.892	0.368	0.055	0.580	0.792	0.938
Standard errors (SED)													
AN		0.264	0.307	1.597	0.864	0.940	1.233	0.1279	0.117	0.306	0.691	0.304	0.542
FU		0.216	0.251	1.304	0.705	0.767	1.007	0.1044	0.096	0.250	0.564	0.249	0.442
AN x FU		0.373	0.434	2.259	1.221	1.329	1.774	0.1809	0.166	0.433	0.977	0.431	0.766

Data are means of four replicate plots

Table A33. Effects of ammonium nitrate and foliar urea on grain yield and quality of cultivar Gerald grown at Gleadthorpe

Ammonium nitrate kg/ha	Foliar urea kg/ha	Crop height cm	Yield t/ha		Specific Weight kg/hl		TGW g		Beta-glucan %		Protein %		Oil %	
		Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
40	0	92	3.52	4.30	50.0	53.0	35.4	38.7	2.80	2.85	8.12	8.30	8.23	7.28
	60	92	3.36	4.60	48.9	53.5	36.9	40.9	2.94	3.10	10.22	11.33	8.05	7.05
100	0	105	3.99	6.50	49.6	51.8	36.9	36.6	3.05	3.05	9.45	9.99	8.13	7.15
	60	104	4.21	6.90	48.6	52.5	35.9	37.3	2.90	3.15	11.19	11.48	7.65	7.18
140	0	106	4.17	6.60	48.5	50.8	36.8	33.8	3.00	3.19	10.42	10.45	7.98	7.38
	60	105	3.98	6.90	47.6	49.3	35.8	33.4	2.96	3.43	11.68	12.15	8.05	7.03
Means														
Grand mean		100	3.87	6.00	48.8	51.8	36.3	36.8	2.94	3.13	10.18	10.62	8.01	7.18
Ammonium nitrate	40	92	3.44	4.40	49.2	53.3	36.2	39.8	2.87	2.97	9.17	9.81	8.14	7.16
	100	104	4.10	6.70	49.1	52.1	36.4	36.9	2.98	3.10	10.32	10.73	7.89	7.16
	140	105	4.07	6.80	48.1	50.1	36.3	33.6	2.98	3.31	11.05	11.30	8.01	7.20
Foliar urea	0	101	3.89	5.80	49.3	51.9	36.4	36.4	2.95	3.03	9.33	9.58	8.11	7.27
	60	100	3.85	6.10	48.2	51.8	36.2	37.2	2.93	3.23	11.03	11.65	7.92	7.08
Significance of effects (F)														
Ammonium nitrate (AN)		<0.001	<0.001	<0.001	0.063	<0.001	0.919	<0.001	0.314	0.028	<0.001	<0.001	0.420	0.948
Foliar urea (FU)		0.465	0.683	0.003	0.012	0.648	0.722	0.051	0.808	0.051	<0.001	<0.001	0.222	0.111
AN x FU		0.971	0.248	0.836	0.811	0.003	0.043	0.041	0.207	0.747	0.247	0.005	0.353	0.378
Standard errors (SED)														
AN		0.129	0.130	0.130	0.487	0.290	0.520	0.470	0.078	0.113	0.242	0.209	0.242	0.132
FU		0.011	0.106	0.100	0.398	0.230	0.425	0.380	0.064	0.093	0.198	0.171	0.198	0.108
AN x FU		0.018	0.184	0.180	0.689	0.400	0.736	0.660	0.111	0.160	0.343	0.296	0.343	0.187

Data are means of four replicate plots

Table A34. Effects of soil moisture* on grain yield and quality of cultivar Gerald grown at Gleadthorpe

Irrigation	Crop height cm	Yield t/ha		Specific Weight kg/hl		TGW g		Beta-glucan %		Protein %		Oil %	
	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
Nil	103	3.66	6.3	49.65	50.6	38.67	33.4	2.85	2.77	10.22	10.85	8.00	7.80
Early	103	4.87	5.5	49.55	50.0	39.38	34.0	2.77	2.89	9.18	10.85	7.85	7.63
Late	102	4.11	6.6	50.40	47.0	38.80	33.7	2.73	2.99	9.52	10.71	7.90	7.45
Throughout	101	-	6.4	-	48.1	-	33.2	-	2.89	-	10.53	-	7.42
Grand mean	102	4.21	6.2	49.87	48.9	38.95	33.6	2.78	2.88	9.64	10.73	7.92	7.58
Sig. effects (F)	0.862	0.016	0.221	0.270	0.131	0.655	0.360	0.592	0.897	0.106	0.241	0.533	0.332
SED	0.023	0.289	0.53	0.513	1.500	0.784	0.46	0.111	0.295	0.409	0.168	0.129	0.216

Data are means of four replicate plots

* Controlled through irrigation at 'early' and 'late' developmental stages

Sourcing added value food ingredients from home grown oats *'Innovation'*

Part 2

Development of a dry milling process for oats

by

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Summary

Dry roller milling technology was evaluated for its applicability to oat milling and fractionation. This was adapted from wheat milling technology and offers the opportunity to produce enriched fractions at low cost. The study was part of the Innovation project, which aimed to establish the basis for the economic production of novel food ingredients from oats.

Oats have a high oil content, compared with wheat, potentially causing difficulties for dry milling. Hence the feasibility of dry roller milling of oats was first investigated. Having demonstrated dry roller milling of oats to be feasible, the process was then investigated in detail, in terms of milling operation, to develop a prototype oat milling process. The effects of roll gap, differential and fluting on particle size distribution and composition were investigated. Based on these analyses, size separation at 420 μm was identified as producing "starch-enriched" and "bran enriched" streams. Multi-pass milling was studied, and a prototype three-pass milling process designed which produced approximately equal proportions of starch- and bran-enriched fractions.

Samples of oat varieties grown under a range of agronomic conditions were milled using single pass milling, and separated into starch-enriched and bran-enriched streams. These streams were sent to Nottingham University for detailed component analysis. Based on these results, six varieties were identified by DuPont as having potential use as food ingredients. These six varieties were milled, and the starch-enriched stream sent to DuPont for sensory evaluation in a sauce product.

The project has demonstrated that dry roller milling technology can be successfully transferred from wheat flour milling to oat milling. It has been shown that oat components are unevenly distributed throughout the milled stocks, and can therefore be enriched through size separation. This work has established the basis for developing a full process for optimal separation of valuable components; detailed process design would depend on the particular components identified, their relative value and the total economics of producing a range of enriched fractions. Dry milling of oats offers the opportunity to extract added-value enriched fractions at relatively low cost.

Introduction

The key to developing economic processes to produce novel food ingredients from oats is to find uses for all the oat fractions. Oats contain several high value components in varying quantities, few of which can be extracted economically without consideration of how to exploit the remaining fractions. For example, oats contain in small quantities a unique patented emulsifier with excellent viscosity-reducing properties, which cannot currently be extracted economically as there is no market for the remaining fraction. The *Innovation* project therefore aimed to investigate potential food ingredient uses for all the major oat fractions and to develop an economic dry milling process to separate the fractions and maximise their functionality and value. This report highlights the results of the application of dry roller milling technology to oats. This part of the study took place at the Satake Centre for Grain Process Engineering, a subsidiary of the Dept. of Chemical Engineering at UMIST. The Centre has wide expertise with dry milling of wheat, technology which it was hoped would be applicable to oats grown within the study.

Oats contain components of high value, where the value is derived firstly by breeding in high levels of these components; secondly by manipulating growing conditions to maximise production of these components; and thirdly by processing to extract the components economically into a range of ingredients of high functionality. Dry milling offers the opportunity to adapt available wheat milling technology to oats, to develop new processes which do not infringe current patents and which produce enriched fractions for a range of food uses.

Oat grains are softer in texture than wheat, partly as a result of the high oil content of the endosperm. This was expected to give problems during dry-milling since there were previous reports of clogging of mill rolls. Because of this it was expected that the study would examine cryogenics as a means to modify the fracture properties of oats before milling. It was evident early in the study that the clogging of mill rolls as had been reported was not likely to cause a problem with the oats examined, even though some of the samples contained high levels of oil. Hence it was not necessary to examine the use of cryogenics and this was dropped from the study. This enabled other aspects of the milling process to be studied in more detail.

Materials and Methods

At the beginning of this project the analytical methods listed in Table 1 were identified and standardised for the determination of oat components. Throughout the project these methods were also applied for the analyses of component distribution in various milling streams in order to study the separation efficiency of the milling process.
Preliminary studies to develop a dry milling procedure for oats were performed on the Satake STR 100 experimental roller mill. Two varieties of oats, Kynon and Gerald, were used as testing materials. Component contents of these two oat samples are listed in Table 2 The Kynon is a naked variety, while Gerald is not; the compositions of whole and dehusked Gerald are shown.

Table 1. Analytical methods used throughout the project.

Component	Method	Condition/Equipment
Groat content	Manual dehulling	10 g oats
Moisture	Dry weight	110°C, 8 hours
Ash	Furnacing	660°C, 6 hours
Protein (N×6.25)	Nessler's spectrophotometry	Digesdahl
Phosphorus	Molybdovanadophosphoric acid	Digesdahl
Starchy materials	Acid hydrolysis	pH 1.3, 130°C, 20 min
Free lipids	Diethyl ether extraction	Soxtec system HT6
Total lipids	AACC methods 02-01A and 02-02A	3 M HCl, 130°C; 25 min
β-glucan	AACC method 32-23	
Flour acidity	pH of flour suspension in distilled water	
Free fatty acid	Titration	1 g L^{-1} standard KOH

Table 2. Quantitative analysis of initial oat samples.

Components	Cultivars		
	KYNON	GERALD	GERALD KERNEL
Groat content (%)	100	100	73.9
Moisture in oats (%)	13.2	13.3	13.3
Moisture in flours (%)	10.9	10.6	10.5
Starchy materials (%, db)	62.2	63.6	45.0
Starch (%, db)	58.0	59.8	41.9
Protein (%, db)	11.5	10.1	6.5
Phosphorus (%, db)	0.4	0.5	0.4
Ash (%, db)	1.6	1.6	2.5
β-glucan (%, db)	4.2	3.9	3.1
Total lipid (%, db)	8.3	8.4	8.2
Free lipid (%, db)	5.8	5.7	5.4

For practical reasons, and because a commercial process to convert oat grain to added-value ingredients could be so designed that no intermediates will be stored for more than 24 hours, lipase inactivation by heat treatment of groats before milling was not intensively investigated. With every parameter fully adjustable on the test mill, a milling procedure was developed by investigating each facet independently. In the search for parameters as indications of milling and separation efficiencies the distributions of oat components, including ash, phosphorus, protein, starchy materials, lipids and β-glucan, in milling streams were analysed. Results indicated that ash and starchy materials were the two most convenient representatives of separation effectiveness. This was not only because of the ease of their analyses, but also because of their natural origins in bran and endosperm, respectively.

Results and Discussion

Based on the following definition of oat bran from AACC in 1989 (below) the development of the milling procedure was targeted to produce two streams of equal weight. The coarse fraction was "oat bran" while the fine portion was referred to as white flour.

Oat bran is the food which is produced by grinding clean oat groats or rolled oats and separating the resulting oat flour by sieving, bolting and /or other suitable means into fractions such that the oat bran fraction is not more than 50% of the starting material, and has a total β-glucan content of at least 5.5% (dry weight basis) and a total dietary fibre content of at least 16.0% (dry weight basis), and such that at least one-third of the total dietary fibre is soluble fibre.

Separation of milled flour by sieving

While milling is essential for adequate "break-down" of the raw materials, sieving is a crucial procedure for component separation or condensation. Figure 1 shows comprehensive comparisons of various component distributions in streams separated by sieve series. This clearly identified 420 μm as an appropriate sieve size for the separation of bran and white flour. Under optimal operating conditions the starting materials were separated into 26.2% white flour and 73.8% bran using a single milling operation. The operating conditions for the mill were: Roll gap: 0.08 mm; feed rate: 304 kg h-1; differential: 3; slow roll speed: 200 rpm; roll configuration: 10.5 flutes per inch.

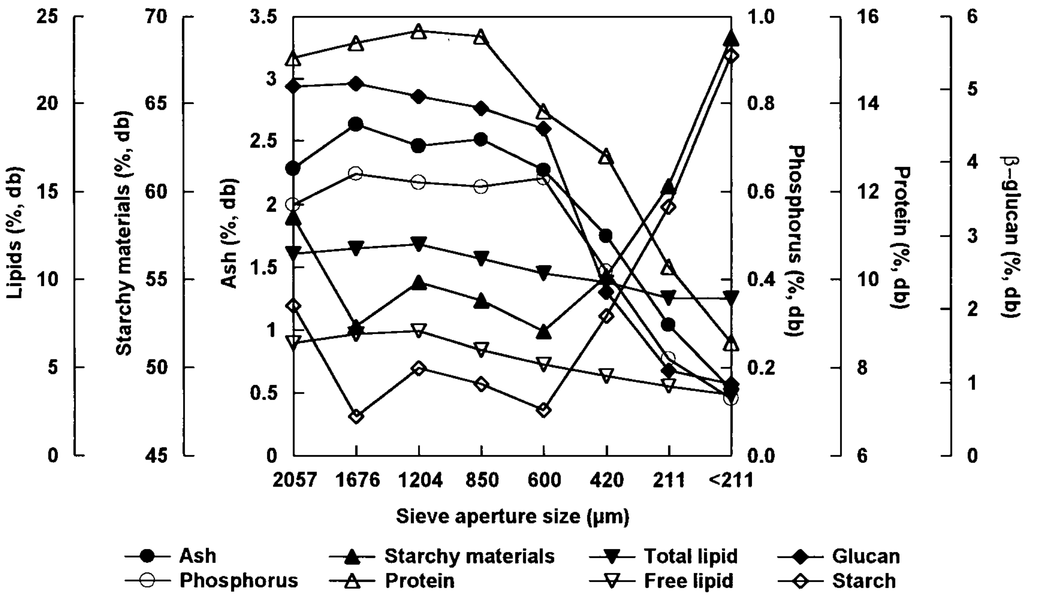

Figure 1. Effects of sieve aperture size on component separation.

Milling operations

Roller milling is critically affected by mill operation, including roll differential, roll gap and surface finish (coarse or fine flutes or smooth rolls). The effect of these milling parameters on oat milling was investigated in order to evaluate the suitability of dry roller milling for oats, and to identify optimum settings for effective separation. Multi-pass milling of the oats to increase white flour yield was also investigated.

Roll speed differential

With the slow roll speed set at 200 rpm the effects of roll speed differential were investigated. By increasing differential from 2.5 to 3.5 white flour production ranged from 25% to 30% (Figure 2). Further increases in differential led to higher production of white flour and at the same time resulted in increases in power consumption. Bearing in mind also the safe operation of laboratory equipment a differential of 3 was finally selected as one of the standard operating parameters. With this value a white flour production of 27% of the starting materials could be obtained from the first break.

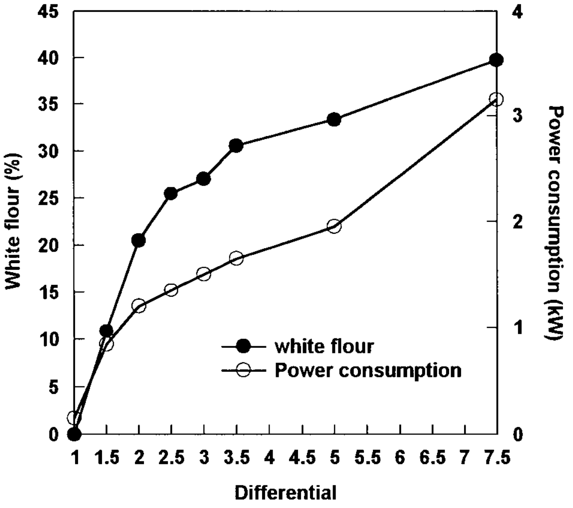

Figure 2. Effects of roll speed differential on flour production and power consumption.

Roll gap

In order to optimise roll gap the differential was set at 3 while all the other parameters were maintained as in the previous milling operation. Milling results clearly demonstrated an inverse relationship between roll gap and white flour production (Figure 3). When roll gap was reduced to 0.06 mm white flour production reached over 30% of the starting materials. Analyses of ash distribution in milling streams from different roll gaps revealed that some particular oat components, probably embryos, were accumulated by the 600 μm sieve (Figure 4). In more detailed milling studies, this stream should not undergo any further milling. The 211 μm sieve separated the 'throughs' from the 420 μm sieve into two streams of very different ash content, especially in the cases of roll gaps of 0.06 mm and 0.08 mm (Figure 4). The combination of these two streams produced white flour with an ash content below 0.8% on a dry basis (Figure 5). For detailed studies of milling and component extraction, however, these two streams should be treated separately. These studies will require large amounts of oats as starting material.

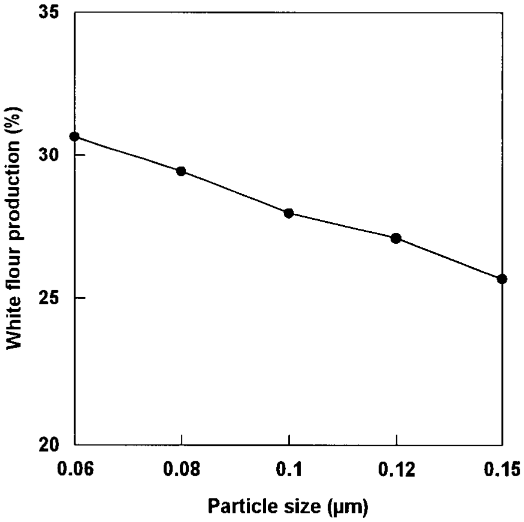

Figure 3. Effects of roll gap on white flour production.

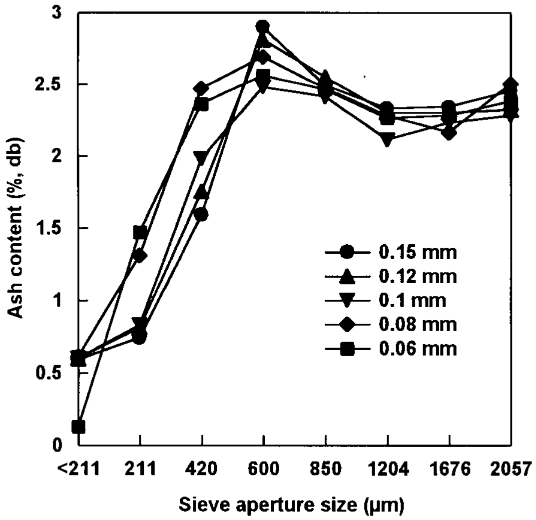

Figure 4. Ash content of milling streams.

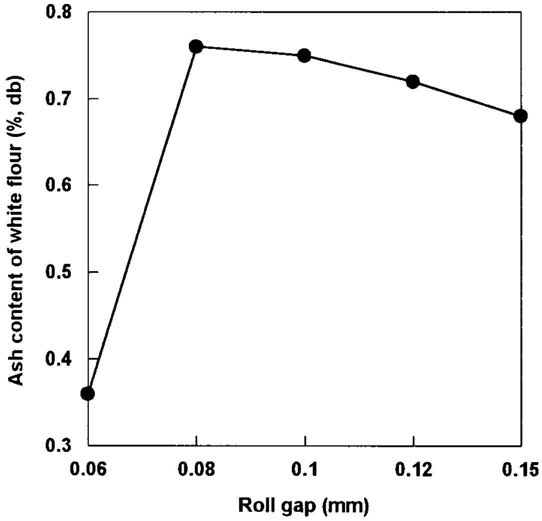

Figure 5. Effect of roll gap on ash content of white flour.

Roll configuration

A comparison was made of the performance of rolls with different flute configurations. Rolls of 14 flutes per inch displayed better milling performance than those of 10.5 flutes per inch and extended the production of white flour and the coarse fraction (Figure 6) while the formation of intermediate particles was slightly reduced. The application of smooth rolls, however, failed to produce any positive effects on the improvement of milling performance (data not included).

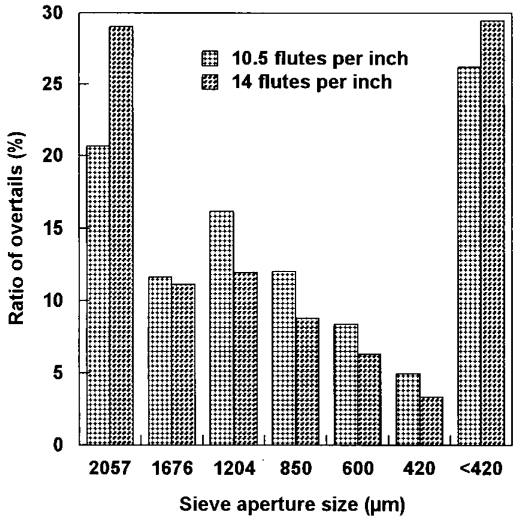

Figure 6. Effects of roll fluting on flour production.

Successive millings

Successive millings were carried out with the same operating parameters optimised for first break with the exception of a smaller roll gap of 0.05 mm. Using heat treated Kynon groat the second break raised white flour (<420 μm) production to 52.6% of the starting material on a dry basis. (Figure 7). In this flour the content of starch materials was enhanced to 71%. Further milling (third break) brought white flour production up to 59.6% with a slight increase in ash content, which indicated bran contamination of the flour. Due to the process of heat pretreatment ash content in the white flour was higher than 1%. Using unheated Kynon the ash content of white flour was frequently below 1% on a dry basis.

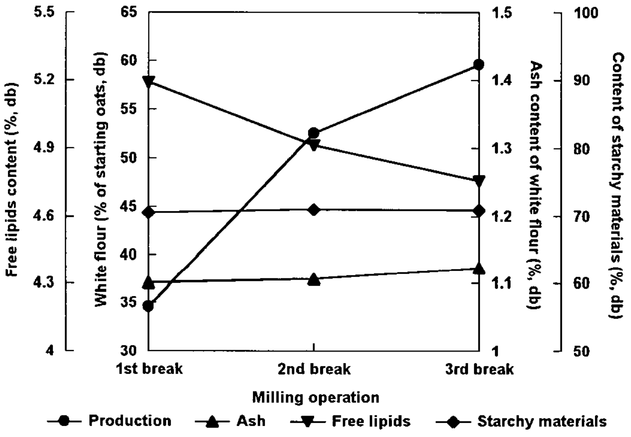

Figure 7. Production and component composition of white flour from successive milling.

Component separation

During the development of the milling process the distributions of various oat components in milling streams were analysed. Based on the results the following conclusions were drawn:

1. White flour contains higher starch and lower protein content while bran contains higher protein and lower starch content.
2. For long-term storage of oat flour, heat treatment of the flour post-milling (rather than the

groats before milling) seems more effective for lipase deactivation.

3. Compared with coarse fractions white flour contains less lipid and β-glucan, but the total lipids and β-glucan that appear in the white flour were not insignificant because white flour represented a large portion of the total groat weight.

Final milling process

According to the results obtained throughout the project a final milling process was decided, as depicted in Figure 8. At the laboratory scale this process was frequently simplified by combining coarse flour into bran, so that only one sieve of 420 μm was required for the sieving separation of milled components.

Samples for analysis at Nottingham University and DuPont

Prior to milling, samples for Nottingham were dehulled using a Streckel & Schrader laboratory dehuller. Samples for Dupont were dehulled by Quaker Oats Limited using a custom built dehuller at the Quaker site. Those dehulled at Quaker contained approximately one third of the original hulls contaminating the samples and due to time constraints no further dehulling was possible. Samples were milled for analysis at Nottingham University using the conditions shown in Figure 8 but using only a single break operation. The resultant flours were separated into two fractions using the 420 μm sieve and designated 'starch-rich' (<420 μm) and 'bran-rich' (>420 μm). Following milling, samples were flushed with nitrogen gas, vacuum packed in plastic pouches and sent to Nottingham by next day courier; the samples were analysed at Nottingham immediately on arrival to limit any deterioration of components. Table 3 lists the samples milled for analysis at Nottingham, and Table 4 those for DuPont. Details of the identity of samples are given in Part 1 of this report.

Table 3. Milling records of samples for Nottingham University:

Sample code	White flour (% of sample size)	Ash content (%; db)	
		>420 µm	<420 µm
G2	61.1	3.0	0.8
I1	47.1		
I2	50.2		
I5	42.2		
I6	49.6		
V1	58.5		
V5	59.5		
V7	61.0		
V2	59.4		
V10	52.6	2.9	1.2
V11	60.7	2.5	1.3
G6	62.0	3.2	1.0
G1	63.6	3.1	0.9
G5	63.8	3.0	0.9
G3	55.2	2.9	1.7
G4	57.1	2.80	0.8
I3	55.3	2.5	0.7
N1	51.6	2.5	0.7
N3	46.8	2.4	0.7
IR1	51.7	2.6	0.8
IR2	53.2	2.5	0.8

Table 4. Milling records of samples for DuPont.

Sample code	<420 µm (%)	>420 µm (%)
G1	50.1	50.0
I1	30.0	70.0
V2	44.7	55.3
V5	34.3	65.7
V7	29.1	70.9
V10	31.9	68.1

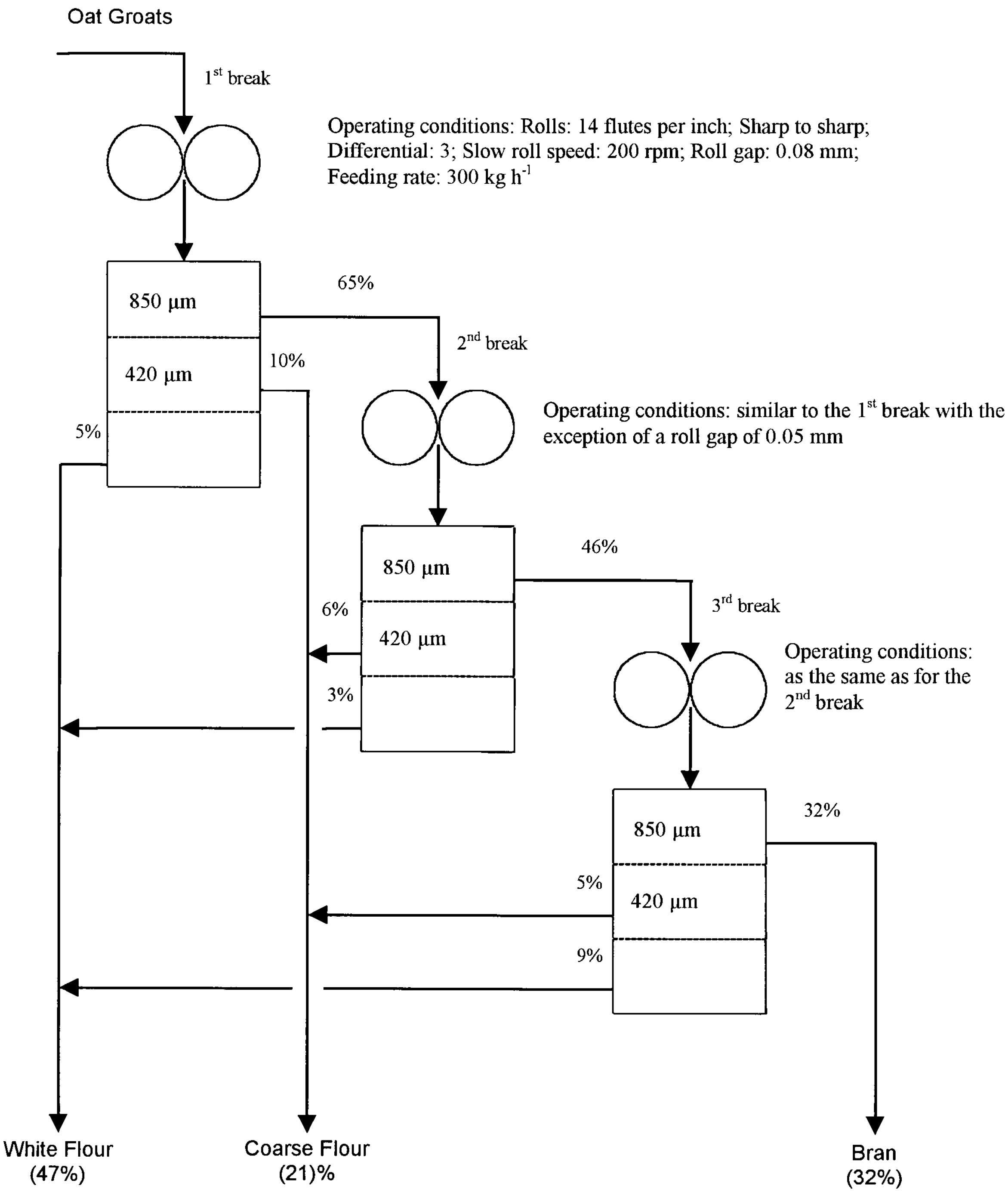

Figure 8. Prototype milling process for dry milling of oats. Samples milled for analysis at Nottingham University and DuPont used only the first break.

Conclusions

Dry roller milling technology has been successfully applied to oat milling. The milled stocks show an uneven distribution of oat components, allowing enrichment of streams by size separation. Multi-pass milling allows more effective and selective separations and enhanced yields. Separation effectiveness depends critically on roller milling conditions. Fluted rolls with 14 flutes/inch, operating at differential of 3 and roll gap of 0.08 mm, produce first break stocks with a natural separation into starch- and bran-enriched fractions at around 420 μm. A prototype three-pass process has been designed, which produces approximately equal fractions of starch- and bran-enriched material. This work has established the basis for detailed process design, which would need to be tailored to the particular added value components sought, under the constraint of the total economics of producing all the fractions. Dry milling technology offers the opportunity to produce enriched ingredient fractions at relatively low cost.

Sourcing added value food ingredients from home grown oats

'Innovation'

Part 3

Analysis of oat constituents

by

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Summary

Studies at Nottingham University consisted of detailed examination of the properties of starch, β-glucan and polar lipid fractions in the oat samples grown by ADAS. The final samples for analysis were chosen in order to investigate the effects of variety, agronomy and environment on properties of the grain components studied. Some analytical techniques were already available; others required further development. Initial development of methods utilised commercial oat samples, but in later studies the samples came from the ADAS field experiments. Samples were investigated either as whole oat grain flour or dehulled and selectively milled (fractionated) samples. The dehulling and fractionation were also part of this project and took place at UMIST. Whole grain flour and fractionated oat flour were compared to determine whether antioxidant activity decreased after dehulling and storage, or during the milling and storage of the milled samples. The results showed no deterioration in antioxidant activity after several months of storage of the dehulled grain. Under the conditions used (transport/storage of less than 48 hr at ambient temperature) there was no change in antioxidant activity in the milled samples. This result gave confidence in the storage properties of both the whole oats and the milled fractions.

The starch granule size and gelatinisation temperatures for the test samples agreed with data previously reported for oat starch. From pasting curve data, "pure" oat starch had a late and low viscosity peak compared to all other starches tested, except for rice starch. The starch-rich fraction from the milling trials appeared to contain other contaminating material. All the milled samples provided by UMIST had similar gelatinisation temperatures, but there were major differences in the maximum paste viscosities, which were shown to be greatly reduced by the presence of silver ions. The likely cause of this is that several samples contained high levels of active amylase.

β-glucan was extracted from both the starch and bran rich fractions of milled oats. Several methods were evaluated in an attempt to obtain reproducible results for the quantities and molecular weights of the β-glucan. Recovery of β-glucan was higher and more reproducible if the samples were defatted prior to extraction. However, there were still difficulties in obtaining the precision necessary to give confidence in the results. The β-glucan samples were also frequently found to be contaminated with starch. As expected, the amounts of β-glucan were higher in the bran-rich fraction compared to the starch. The average molecular weight for the β-glucan was approximately 3.1×10^5, with values ranging from 1.4 to 6.3×10^5. These results fall within the data range previously reported for molecular weights of glucans, but the methodology and extraction may have affected the values obtained. Storage of some samples was also found to cause a marked decrease in the molecular weights of β-glucan. It was thought that this was due to endogenous enzymes rather than secondary contamination.

A particular effort was made to measure the activity of phenolic antioxidants found in oats. The extraction method for the phenolics was based on isopropanol followed by solid phase purification. Typical values for the amount of phenolics recovered were in the region of 100 mg of total phenolics per kg of oats. It was clear that the bran-rich milled fraction contained a higher concentration of phenolics than the equivalent starch-rich stream. In previous work on the additions of oat phenolics to plant oils (to enhance oxidative stability), commercially viable effects were observed at 0.01% (w/w) of total phenolics. Given this value, and the concentration of phenolics in our extracts, one can estimate that 1 kg of oats would have to be processed to yield enough phenolics to be added to 1 kg of oil.

A sensitive and rapid chemiluminescent method for screening antioxidants was developed in the study. The amount of antioxidant activity in the polar lipid extract was calculated as the Inhibition Concentration. This is the equivalent mass of oats required to decrease the chemiluminescence by 50%, IC_{50}. The greater the activity of the antioxidant the lower the IC_{50} value. Results indicate that these values range between 0.3 and 2.5 mg of oats depending on the sample analysed. Generally the antioxidant potency of the polar lipid extracts from the bran-rich fractions is higher than those from the starch-rich fractions. Furthermore, when the IC_{50} values were calculated relative to the total phenolic concentration in that sample, the mass of phenolic compounds required to cause 50% quench was obtained. This value ranged between 27 and 106 ng of total phenolics and was significantly lower in the bran-rich fractions. This provides indirect evidence that distinct populations of phenolic compounds exist in these oat flour fractions, or that an inhibitor or stimulator of antioxidant activity is concentrated in one of the fractions.

The emulsifier component previously identified was examined in collaboration with RSSL. All polar lipid extracts of whole grain flour, bran-rich and starch-rich fractions tested contained emulsifier but the method used to measure the relative concentration of the glycolipid emulsifier in these extracts did not reveal any differences between the samples tested.

One of the main objectives of this section of the study was to determine whether differences exist between oat grain samples as a result of varietal, agronomic or environmental effects. Although there were clear differences between samples examined in this study, it was not possible to precisely define the quantitative effects of the factors examined.

Introduction

Studies at Nottingham University examined the properties of components from oat grains in terms of their potential for utilisation as food ingredients. Early discussions within the management group deemed that the components of most interest were likely to be starch, β-glucan and antioxidants and emulsifiers present within the polar lipid extracts.

The aims of the studies at Nottingham were:

1. To develop and compare methods for the extraction and analysis of these components.
2. To compare the relative amounts and properties of the components in the starch-rich and bran-rich fractions produced by dry-milling at UMIST.
3. To compare the amounts and properties of the components in samples grown under a range of conditions at ADAS.

Starch is a major commodity in the food industry and is the main constituent of oat flour. For oat production and fractionation to be viable a market has to be found for this component. Characterisation of the starch was therefore undertaken to establish if the behaviour was measurably different to that of starches derived from other sources. Identification of novel functionality would allow the targeting of a suitable market.

Much recent interest has been generated in oat β-glucan as a result of numerous studies of its clinical effect and the FDA ruling for a health claim in the functional foods sector (Chen and Anderson 1979; Kirby *et al* 1981; Wood 1986; Klopfenstein and Hoseney 1987; Dawkins and Nnanna 1993). In addition to this β-glucan also offers potential as an agent which can alter the rheological properties of food (Wood 1986).

Field studies on a series of oat cultivars were carried out separately by ADAS; a selection of these samples were investigated by SEC/MALS in order to determine whether growing conditions and/or variety of oats would be factors influencing the molecular weight properties of β-glucan. Preliminary work had been carried out to determine the best fractionation method for β-glucan from milled oats and these studies also showed that it was necessary to defat the oats prior to fractionation, in order to reduce the amount of starch carried through the fractionation and in particular into the β-glucan fraction. Determination of the molecular weight of β-glucan is important because the molecular weight influences rheological properties.

Oats are also a potential source of natural compounds which show strong antioxidant and emulsifier properties. Antioxidants have a high profile because of their ability to extend the shelf-life of many food products and due to their apparent health benefits. These hydroxycinnamic acid conjugate compounds have amphiphilic properties and can therefore be enriched in polar lipid extracts (Forssell *et al* 1992; Duve and White 1991). The use of oat flour to prevent oils becoming rancid was patented as early as the 1930's (Musher, 1936). The relatively high antioxidant properties of oat flour have been attributed to the phenolic compounds, predominantly ferulic and caffeic acid derivatives (Collins, 1986;

Daniels and Martin, 1961; Daniels *et al.,* 1963; Daniels and Martin, 1967; Daniels and Martin, 1968; Durkee and Thivierge, 1977; Pokorny, 1991; White, 1995). Synthetic antioxidants such as butylated hydroxy toluene (BHT), butylated hydroxy anisole (BHA) and propyl gallate (PG) are used in some food products to delay the onset of oxidative rancidity. As there is serious concern about the toxic and carcinogenic effects of these substances (Branen, 1975), there is great interest in the possibility of substituting them with natural antioxidants (Pratt, 1992).

Phenolic compounds in cereals can be classified as free (6%), soluble (17-30%) and bound (66-80%). The distribution of phenolic compounds into these groups is influenced by the methods employed to fractionate the material and the cereal which is being analysed (Dimberg *et al.,* 1993; Hatcher & Kruger, 1997). In general, the majority of phenolic compounds in cereals are bound through a covalent association with cell wall polysaccharides. The removal of these compounds requires extreme conditions. Simple phenolics in the free form are rarely found. This is probably a consequence of their phytotoxicity (Collins, 1986). The soluble fraction is therefore of greatest interest in this work.

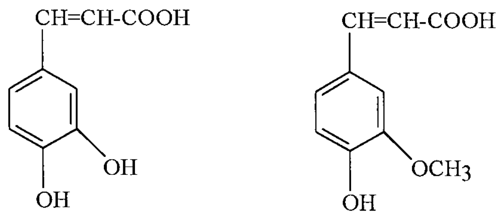
A range of phenolic compounds exist in oats with varying solubilities (see Figure 1). These include ether or ester linked glycosides (Collins, 1986), anthranilic acids and avenanthramides (Dimberg *et al.,* 1993), ester linked glycerol conjugates (Taketa, 1957; Vogel, 1961) and ester linked alkyl conjugates (Daniels and Martin, 1967).

Polar lipid extracts from oats have been credited with containing high levels of antioxidant activity (Collins, 1986; White, 1995). Few workers have analysed the complete chemical structure of the phenolic compounds in a polar lipid extract from oats. One exception is the work by Daniels and Martin (1967). They identified a range of long chain (C_{26} and C_{28}) mono and diesters of caffeic and ferulic acids. The antioxidant capacity of these compounds appears to be mediated through their ability to react with free radicals and so break the chain propagation cycle associated with peroxidation reactions.

More recent work (Duve and White, 1991; Tian and White, 1994; Xing and White, 1997) has highlighted the potential of extracted oat antioxidants as a commercially viable source of natural antioxidants for the food industry. An essential aspect of this would be the selection of a cost-effective process for extracting antioxidants. Workers have used different solvents and extraction procedures to obtain the polar lipid fraction, which is enriched with the phenolic compounds of interest (Forssell *et al.,* 1992; Duve and White, 1991). No researchers, however, have directly compared the activities of such extracts with a view to rationalising an industrial scale process. Commercial considerations will also influence the methods used to measure the antioxidant activity of the oat extracts. For example, continual monitoring of the industrial process would be required, and so a method for measuring the antioxidant activity would be necessary. Some reports suggest that chemiluminescence is a convenient method for measuring antioxidant activity (Ashida *et al.,* 1991; Burkow *et al.,* 1995; Hirayama *et al.,* 1997).

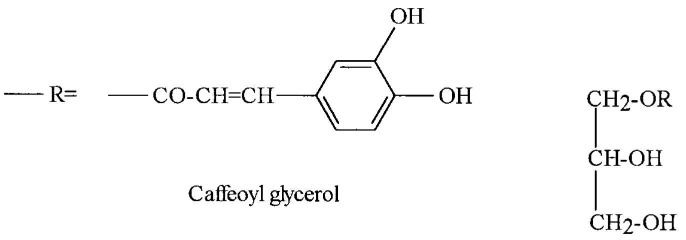
Oat polar lipid extracts have also been shown to contain a novel emulsifier with properties which could be used by the confectionery industry. This was discovered by a team from RSSL in the late 1980s (see Figure 2 for its structure). A patent was published (Evans, 1988) describing the use of this emulsifier, primarily in the manufacture of chocolate. Characteristically, emulsifiers such as lecithin are used to maintain optimum rheological properties with a reduced cocoa butter formulation. Unpublished results from RSSL further showed that the emulsifier performed well in stabilising water in oil emulsions, in bread making as a loaf volume improver, in margarine as both an emulsifier and an anti-spattering agent, and in ice-cream as an emulsifier, giving fine droplet size and minimal fat droplet aggregation. For these reasons the emulsifier is likely to be a valuable element in a proposed fractionation scheme.

Surface active agents which reduce viscosity are normally limited to natural lecithins and the synthetic products YN and polyglycerol polyricinoleate (PGPR). Of all the lecithins, (egg, soya, groundnut, sunflower and rapeseed), only soya is widely used because of considerations of cost, availability and performance. Egg is expensive, groundnut and sunflower difficult to obtain, and rapeseed has lower activity and gives a strong burnt note. The compounds present in soya lecithin are phosphatidyl choline, phosphatidyl ethanolamine, and phosphatidyl inositol (see Figure 3), together with their lyso forms and phosphatidic acid. YN and PGPR are very effective as emulsifiers and are odourless; however they are synthetic, and are not legally permitted for food use in many countries. The components of YN are complex, but are mainly salts of phosphatidic acid monomers and dimers, while PGPR is a complex polymer (see Figure 4). A group of glycolipids was identified as the active components in a polar lipid extract of oats; these glycolipids were responsible for the observed viscosity-reducing effect when added to chocolate. They are derivatives of glycerol, with a digalactosyl residue attached to one of the primary hydroxyls of the glycerol molecule. The remaining two glycerol hydroxyls are esterified with fatty acids. One or more of these fatty acids is an unsaturated hydroxy fatty acid, which is esterified through the hydroxyl group with a further fatty acid. An example of the structure of one of these rheologically active glycolipids is given in Figure 2.

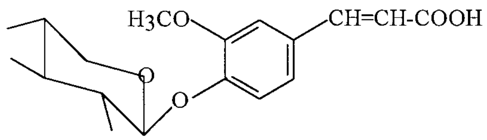


Caffeic acid

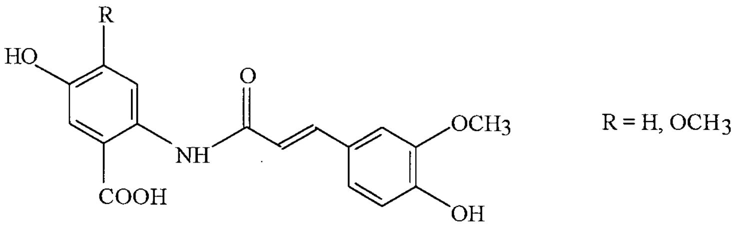
Ferulic acid



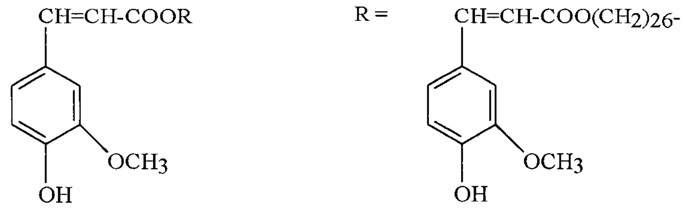
Caffeoyl glycerol



Ferulic acid 4-O- ▶-glucoside



Avenanthramides



R = $C_{26}H_{52}$

1-O-feruloyl-n-hexacosan-1-ol

1,26-di-O-feruloyl-n-hexacosan-1,26-diol

Figure 1. Structures of antioxidants in oats

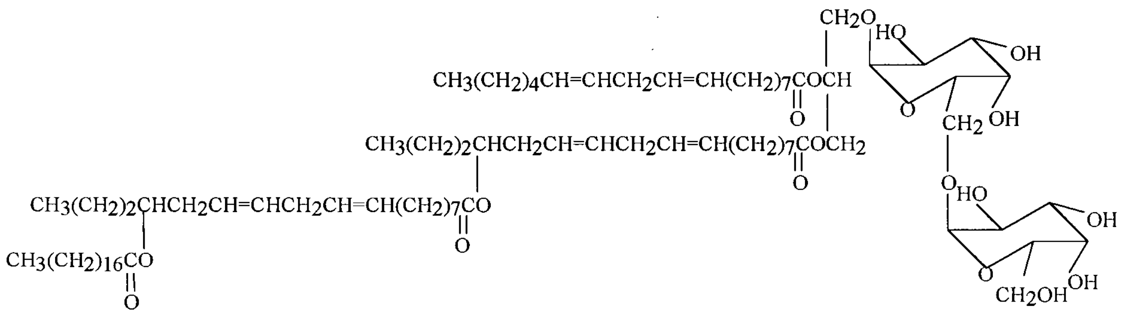
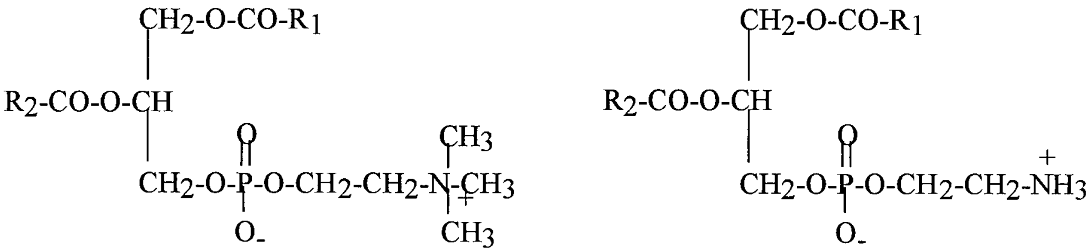
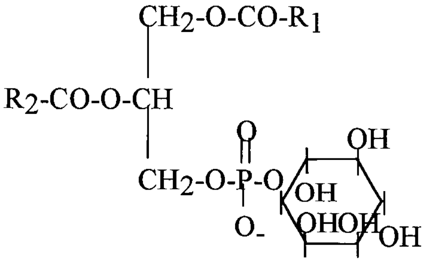


Figure 2. The active component of oat emulsifier



Phosphatidyl choline

Phosphatidyl ethanolamine



Phosphatidy inositol

R_1, R_2 = acyl

Figure 3. The components of lecithin

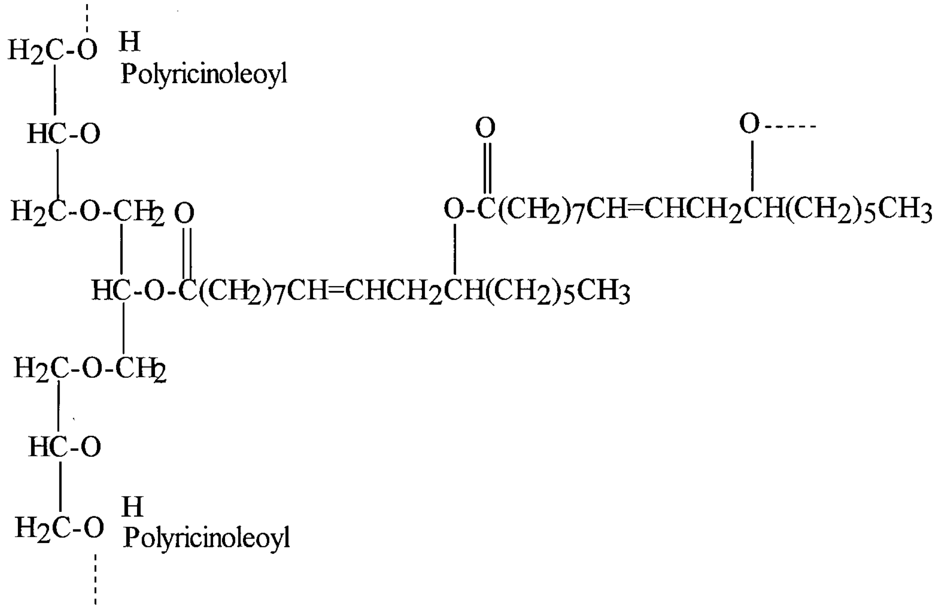


Figure 4. Structure of Polyglycerolpolyricinoleate (PGPR)

Materials and Methods

Starch

In the initial studies three oat samples were acquired.

1) Oat flour (1st break), milled and sieved to <211 µm
2) Oat flour similarly milled and sieved (Mornflake)
3) Purified oat starch (ADAS Rosemaund)

It should be emphasised that only sample 3 is pure starch; samples 1 and 2 are flours, albeit finely sieved.

Differential Scanning Calorimetry

DSC accurately measures the temperatures at which substances take up or give out energy (endo- or exo- thermic processes respectively). Starch granules take up energy when they reach their gelatinisation temperature or, in other words, the temperature at which they become swollen. This temperature is characteristic for starches from different sources and plant species. Additionally, the amount of energy taken up or the enthalpy difference is important.

A 1:3 (w/w) suspension of starch or flour in water was left overnight to fully hydrate. A known amount of the suspension was placed in a DCS pan and the pan placed in the calorimeter. A temperature gradient of 20°C to 100°C was then applied at a rate of 10°C min^{-1}.

RVA viscosity

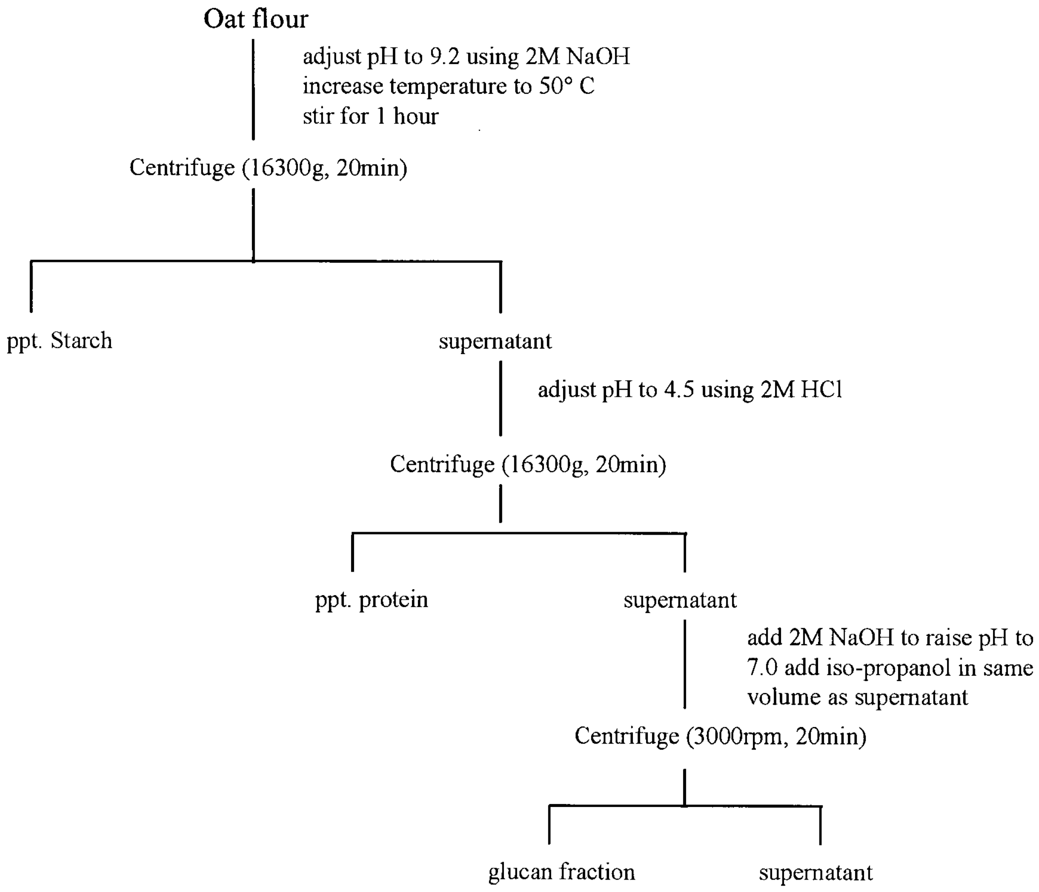
Oat samples for RVA analysis were based on 3.00 g (uncorrected for moisture) of samples in 25 g of distilled water (pH 5.9 at 25°C). Buffering salts were not required as the samples were analysed immediately. A paddle was placed into the canister containing the sample and water. The paddle blade was vigorously jogged through the sample up and down 10 times. The sample was then inserted into the RVA Series 4 (Newport Scientific, NSW, Australia) which was used along with the accompanying software (Thermocline). A standard 1 profile was used and can be described as follows: idle and hold at 50°C; 0-1 minutes = 50°C, 1-4:45 minutes = ramp up to 95°C; 4:45-7:15 minutes = hold at 95°C; 7:15-11 minutes cooling (set at 50°C); hold at 50°C to 13 minutes.

β-glucan

Extraction of β-glucan

Oat samples were defatted using isopropanol as described elsewhere (see section on polar lipids) and milled with a rotary mill through a 250 μm mesh. Fractionation of samples was carried out according to the method by Dawkins and Nnanna (1993). A schematic outline of this procedure is shown below.

The β-glucan fractions were freeze-dried and weighed and an aliquot of each sample was dissolved in 0.1M NaCl in a boiling water bath and left to stir overnight. Samples were tested for the presence of starch using 0.1N KI/I_2 solution - absence of any blue colour was an indication that no starch was present.



Determination of absolute molecular weight and molecular weight distribution by SEC/MALS

Size exclusion chromatography (SEC) separates molecules according to decreasing occupied volume, provided that there are no non-size exclusion mechanisms interfering with the separation. For a homologous series this results in a separation according to decreasing molecular weight. Dual detection with in-line mass and light scattering detectors allows determination of absolute molecular weight according to:

$$\frac{Kc}{R_\theta} = \frac{1}{P(\theta)}\left(\frac{1}{Mw} + 2A_2c\right)$$

where K is the polymer constant for a particular scattering system, c is the sample (fraction) concentration, R_θ is the excess Rayleigh factor, M_w is the weight average molecular weight, P(θ) is the particle scattering function which contains information about particle dimensions (eg. radius of gyration, R_g), and A_2 is the second virial coefficient which is a measure of solvent-solute interactions and to a first approximation can be taken as zero due to the extremely low concentration of the individual sample fractions ($c \approx 1 \times 10^{-5}$).

The light scattering cell is illuminated by a 5 mW He-Ne laser (wavelength 633 nm) and the intensity of the scattered light is measured at 18 angles simultaneously. Weight average molecular weight for each fraction, $M_{w,i}$, is then obtained by extrapolation to zero angle using a plot of $Kc_i/ R_{(\theta),i}$ versus $\sin^2\theta/2$ (Zimm plot). Although the molecular weight at each slice is, according to the above equation, the weight average molecular weight, if the slices are assumed to be approximately monodisperse, then the number and z-averages over the whole distribution can be found from the usual equations:

$$M_n = \frac{\sum c_i}{\sum (c_i / M_i)}$$

$$M_w = \frac{\sum c_i M_i}{\sum c_i}$$

$$M_z = \frac{\sum c_i M_i^2}{\sum c_i M_i}$$

Chromatographic conditions

The columns were eluted with aqueous 0.1M NaCl solution. The SEC/MALS system consisted of a Waters 590 Solvent Delivery module (Waters, Millipore, Watford, UK), a Rheodyne injection valve (Model 7125) fitted with a 100 μl loop (Rheodyne, St Louis, US), a guard column and three analytical columns (TSK G600PW, TSK G500PW, TSK G4000PW, ToSoHaas, Japan). Scattered light intensities were measured using a Dawn F multiangle light scattering photometer (Wyatt, Santa Barbara, US). Concentrations were measured using an Optilab 903 (Wyatt, Santa Barbara, US) interferometric refractometer. The eluent was pumped at a flow rate of 0.8 ml/min at ambient temperature and full loop injections of samples (5 mg/ml) which had previously been filtered through 0.45 mm syringe filters (PVDF, Whatman, Watford, UK) were performed.

Antioxidants

All oat samples for analysis were milled at UMIST as described in Part 2 of the report. Solvents were analytical reagent grade and purchased from Fisher Scientific. All other chemicals were purchased from Sigma-Aldrich Chemical Co. unless otherwise stated; t-butylhydroperoxide (70% aq. solution), $Na_2B_4O_7.10H_2O$, luminol (HPLC grade), cytochrome c (horse heart, 99% pure), silicic acid (100 mesh), tannic acid (code:T/0150/53; batch: 975829347; $C_{76}H_{52}O_{46}$ MWt. 1701.22) were purchased from FisherScientific.

Preparation of oat samples

i) Grinding of whole oats: A batch of approximately 600 g was ground for 5 minutes in a Phillips Blender (400 W, Type HR 2810/A) at full speed.
ii) Fractionation of dehulled oats: these were dry-milled at UMIST as described in Part 2 of this report.

iii) Transport and storage of samples: After milling, the samples were packed into polyethylene bags, flushed with nitrogen and sent within one day to Nottingham. Lipids were extracted from the samples as soon as they arrived at Nottingham.

Lipid extraction

For this method the extraction solvent was isopropanol at 70°C. The extraction was carried out within two hours with continual stirring. Oat groats (100 g) were weighed into an Erlenmeyer flask (2 l), isopropanol (1.2 l) was added. The Erlenmeyer flask was then placed into a water bath at 70°C and the suspension was stirred with a propeller stirrer at 300 rpm for 2 hours. To obtain a clear solution the extract was centrifuged at 1500 rpm. The centrifugate was rotary evaporated and dissolved in 7.5 ml of methanol. Further fractionation by column chromatography was carried out (below). The extracts were stored in the dark at -80°C until required.

Fractionation of the crude lipid extract

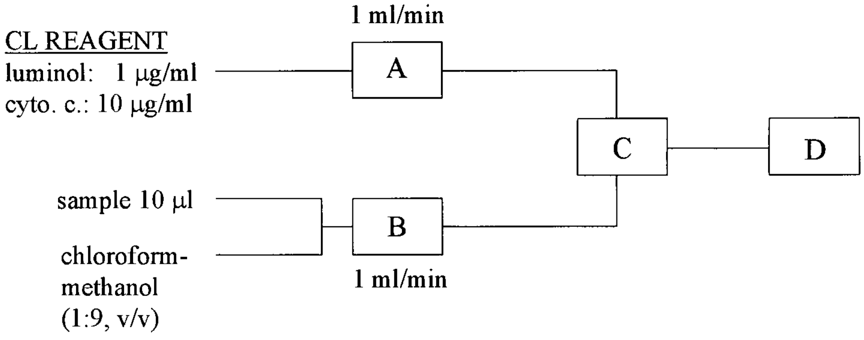
Silicic acid (50 g of 100 mesh) was activated overnight at 105°C. This was then washed three times with methanol (100 ml), then three times with petroleum ether (100 ml). A petroleum ether slurry of the treated silicic acid was then packed into a column (25 mm x 300 mm). The methanolic extract was applied to the top. The first elution was carried out with 300 ml petroleum ether to extract hydrophobic material and the second with 300 ml methanol to extract polar material. The methanol extract was then rotary evaporated, dissolved in 7.5 ml methanol and stored in the dark at -80°C until required.

Determination of total phenolic contents

The total phenolic content was measured by following the AOAC Method (9.110), (1984), with tannic acid as the standard. Samples (0.4 ml of extract) were mixed with Folin-Denis reagent and sodium carbonate at ambient temperature (and at the prescribed concentrations) and the absorbance at 760 nm measured after 30 minutes.

Measurement of Antioxidant Activity

The experiments were carried out as depicted in Figure 5. The "Waters 2690" pump has an automatic probe sampler and an automatic injector with a sample loop of 50 μl. Test samples were injected from this loop into the chloroform-methanol mixture and pumped to the chemiluminescent (CL) detector.

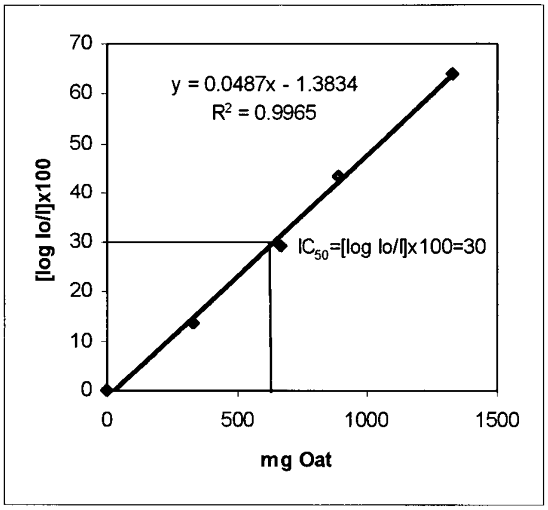


A	peristaltic pump Ismatec sa
B	HPLC pump Waters 2690 with 10 µl injector and sample carousel
C	chemiluminescence detector Camspec
D	integrator Hewlett Packard 3392 A

Figure 5. Flow diagram of the CL method

Calculation of Antioxidant Activity:

A series of dilutions of the extracts in 7.5 ml methanol was made using methanol (1:4000, 1:3000 1:2000; 1:1500; 1:1000 and 1:400); t-butyl hydroperoxide was added to a concentration of 3 µmol/ml. The peak areas of these dilutions were measured and compared to the area of an equivalent solution minus oat extract, which was set to 100 %. The antioxidant activity of each oat extract was calculated as an IC_{50} (inhibition concentration) value, i.e. the concentration of the extract that gives a peak area which is 50 % of the control (Ashida *et al.,* 1991). Unless otherwise stated the IC_{50} values are expressed in terms of the mass of oats equivalent to this extract concentration. Figure 6 shows the effect of oat extract concentration on the % quenching; as concentration increases, so does the % quenching of the chemiluminescence reaction.



[a] Each value represents the average of duplicate measurements which gave a negligible range

[b] Calculation of the IC_{50} value see (Ashida *et al* 1991)

Figure 6. Effect of oat extract concentration on the quenching of the chemiluminescence reaction

Data analysis

All data points were obtained by measuring two separately prepared extracts. For the analysis of the whole flour sample, a single factor ANOVA was carried out and the SED determined to see if there are significant differences between varieties and samples grown under different conditions. Results from the starch and bran analyses were compared using an unpaired t-test or by ANOVA tests.

Emulsifier

For this work the polar lipid extract (see above) was diluted with methanol (1:20, extract:methanol). An aliquot (50 µl) was applied with an automated applicator (CAMAG LINOMAT 3) on chromatographical plates (Silica gel 60, Merck). These were developed in chloroform : ethanol : triethanolamine : water (30:34:28:8 v, v, v, v). The dried plates (5 minutes at 105°C) were sprayed with a 0.2 % solution of 5-methyl resorcinol mono hydrate in 75 % sulfuric acid. After heating the plates for 10 minutes at 105°C the emulsifier appeared as red-violet spots. The intensity of the spots is proportional to the amount of emulsifier. The intensity of the spots of the different samples was measured with the Phoretix 1D gel analysis software Version 4.00 (Phoretix International, Newcastle upon Tyne, UK) and compared against sample N3.

Results and Discussion

Starch

Size of oat starch granules

Oat starch granules are reported (Pomeranz, 1991) to be very small in size (3-10 µm). Table 1 shows sizes of various cereal starches.

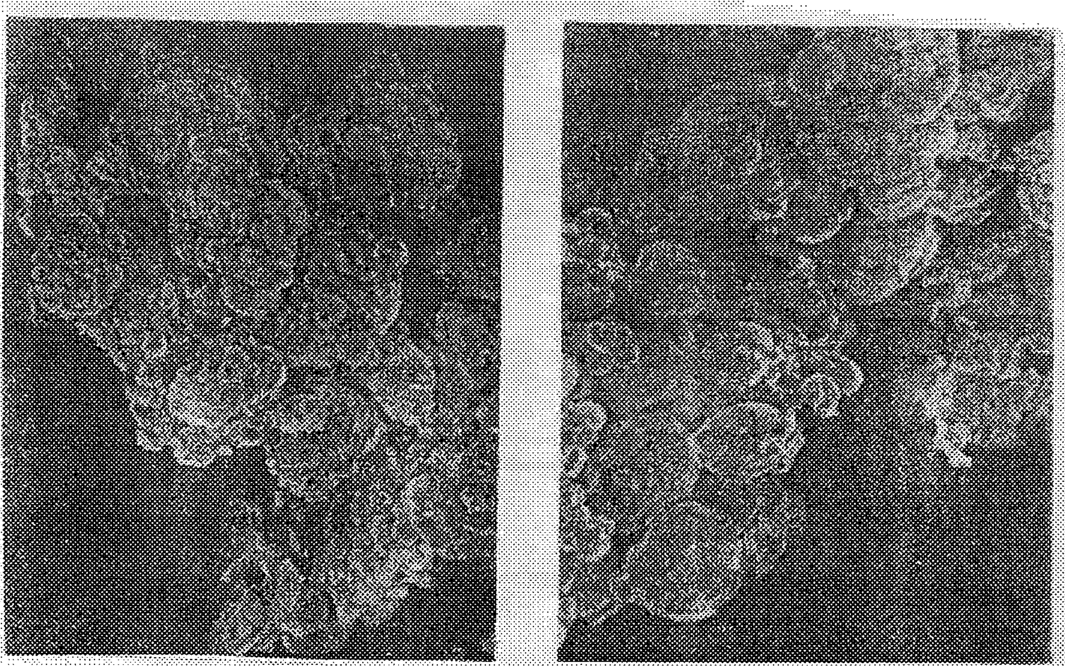
Table 1. Granule size and gelatinisation temperature for a range of starches

Starch	Granule size (µm)	Gelatinization range (°C)
Barley	2-35	56-62
Corn	5-25	62-80
Potato	15-100	56-69
Rice	3-8	61-80
Rye	2-35	57-70
Wheat	2-35	53-72
Oats	2-10	56-62

(from "Functional Properties of Food Components", Y. Pomeranz, Academic Press, 1991)

The fractionated flour samples were examined by Scanning Electron Microscopy (SEM) (Figure 7).

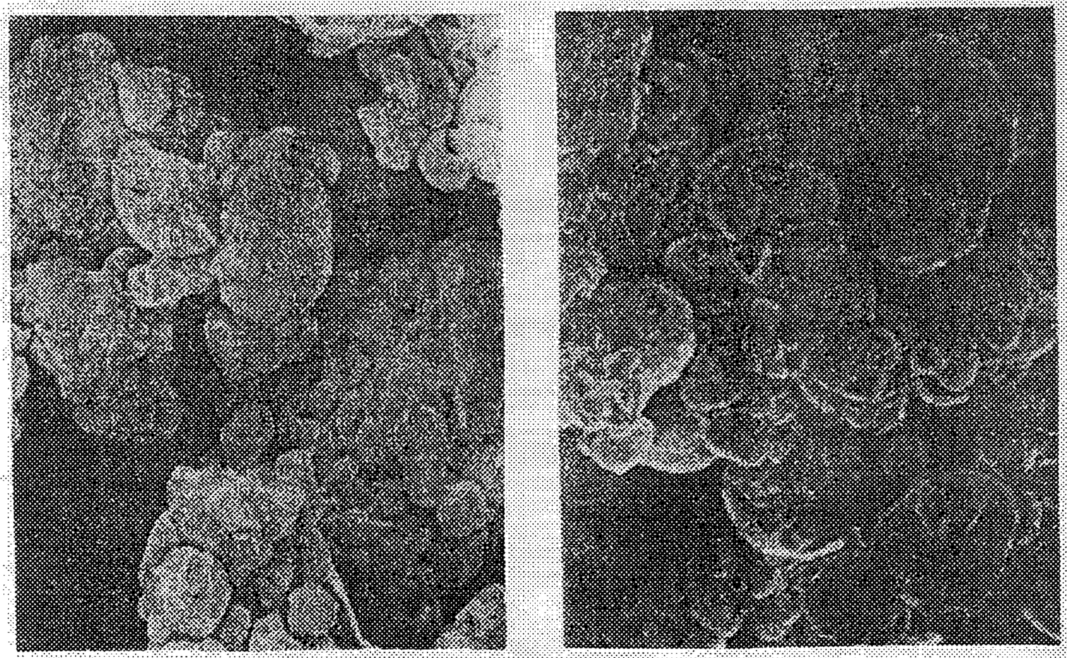
The micrographs indicated aggregated groups of material. There appeared to be two populations of spheroids; one approximately 3x larger than the other. The starches studied by SEM were sent for chemical analysis and the results are given in Table 2.



Flour sample – UMIST, Mag = 575 x

Flour sample – MORNFLAKES

Mag = 575 x



Flour sample – UMIST, Mag = 1,150 x

Flour sample – MORNFLAKES

Mag = 1,150 x

Fig. A1 SEM of UMIST and MORNFLAKE flour

Figure 7. SEM of UMIST and Mornflake oat flour

Table 2. Analysis of oat flour samples

Analysis	UMIST	MORNFLAKE
Nitrogen (%)	1.25	1.38
Protein % (N x 6.25)	7.8	8.6
Ash (%)	0.5	0.7
Moisture (%)	10.5	9.2
Fat (%)	10.1	7.7

Gelatinisation characteristics

The thickening properties of starch depend on the characteristic way in which the granules swell when heated in excess water. The temperature at which gelatinisation occurs or the amount of heat required to melt the crystallites within the starch granule is important. These properties are dependent on the source. These features can be studied by Differential Scanning Calorimetry (DSC). Table 3 gives the results for all three starch samples. The results are similar to those previously reported. The milling, fractionation and other treatments that had been used for the UMIST and Mornflake samples do not seem to have affected these DSC parameters. All three samples showed a peak at very close to 60°C. This indicated that oat starch has a relatively low gelatinisation temperature compared to other starches. In addition, the enthalpy change for each is also very similar with an average value of 7.71 Jg^{-1} of dry material. This corresponds to the same order of magnitude for data on wheat starch samples.

Table 3. Gelatinisation characteristics of oat starches

Starch	Peak Onset T (°C)	Peak T (°C)	Peak End T (°C)	Δ_H (J/g)
UMIST 1	55.7	60.0	65.2	7.3
UMIST 2	55.6	59.8	64.9	6.8
Mornflake 1	57.1	61.3	66.2	6.9
Mornflake 2	50.4	60.5	65.2	8.4
Oat Starch 1	56.5	59.8	63.4	7.9
Oat Starch 2	56.4	59.7	63.8	9.1

Rapid Viscoanalysis (RVA)

In addition to the gelatinisation temperature, the viscosity of the sample during and following pasting is of great importance during the manufacture of foods. The pasting properties can be followed using a Rapid Visco Analyser (Newport Scientific). This instrument measures the viscosity of flour and starch suspensions as they are taken through a programmed heating and stirring profile. The viscosity of the paste is recorded as the starch granules swell and the suspension thickens. A similar procedure is carried out within the milling and baking industry using a Brabender Amylograph to assess flour quality. Typically 3 g of flour or starch (corrected for moisture content) and 25 ml of distilled water were placed in a specially designed container fitted with a plastic vane rotor. The vessel was placed in the RVA and a heating and stirring profile applied.

Figure 8 shows the RVA profiles for a range of starches. Oat starch shows a high pasting temperature and low peak viscosity compared to all the other samples with the exception of rice. Low levels of chemicals within the water used for pasting can also have a significant affect on the RVA results.

Figure 9 shows three pasting curves for the samples used for DSC work. Samples used were adjusted so that they all had the same solids content, but not the same starch level. It is therefore not surprising that the purified sample showed a higher peak viscosity than the other two flours. The Mornflake sample shows a very similar pattern compared to the purified starch. However, the UMIST sample was different. The gelatinisation peak occurred 40 seconds later in the profile than for the other two samples. In addition a second peak occurred on cooling the sample. This observation was reproducible. This distinctive RVA profile for the UMIST sample could be due to several factors including the presence of material other than starch. Although many other oat samples indicated the presence of a second peak no other sample from the main body of the work showed the pronounced differences demonstrated by this initial oat sample.

Effects of variety and agronomic conditions

The RVA method was sensitive enough to demonstrate differences in the starch samples. It was therefore used to compare the 18 samples from the ADAS field trials (see Tables 4 and 5). The starch-rich fraction from the UMIST milling experiments was used for these analyses.

The RVA method given in section 3 was carried out but in some cases 0.02 % w/v of $AgNO_3$ was added to the water before pasting the samples. This was necessary to inactivate α-amylase which was shown to be present in some of the samples.

Figure 10 shows some of the pasting profiles obtained. The shapes are all similar, but there is considerable variation in the viscosities obtained. It was found that the presence of $AgNO_3$ made a large difference to the profile. Figure 11 demonstrates the difference in curves in the presence and absence of $AgNO_3$.

It is assumed that the difference in the RVA profile is due to amylase activity which is inhibited in the presence of silver ions. Figure 12 shows the % differences in peak viscosity in the presence and absence of silver ions. It would appear that amylase activity is causing at least a 40% reduction in the peak viscosity of samples G1, G2, N1 and N3. All the Gerald samples seem more affected than the Image samples. However it should be noted that V5, which is also a Gerald sample, shows no amylase activity. It has been reported that oats contain small amounts of α-amylase activity (Meredith and Jenkins, 1973) which may be located in the pericarp tissue (Kruger *et al.*, 1991). It is therefore surprising that significant activity has been found in the starch-rich fraction.

Table 6 lists the peak and final viscosities of the samples. There is a range of values, but it is not possible to link these with any specific variation due to variety or growing conditions. It is notable that the Gerald sample grown in the variety trial showed no amylase activity, in contrast to the irrigation and nitrogen trials. It can be speculated that irrigation/nitrogen application maintained the green leaf area index for longer, making samples more susceptible to sprouting and to amylase activity within the developing grains.

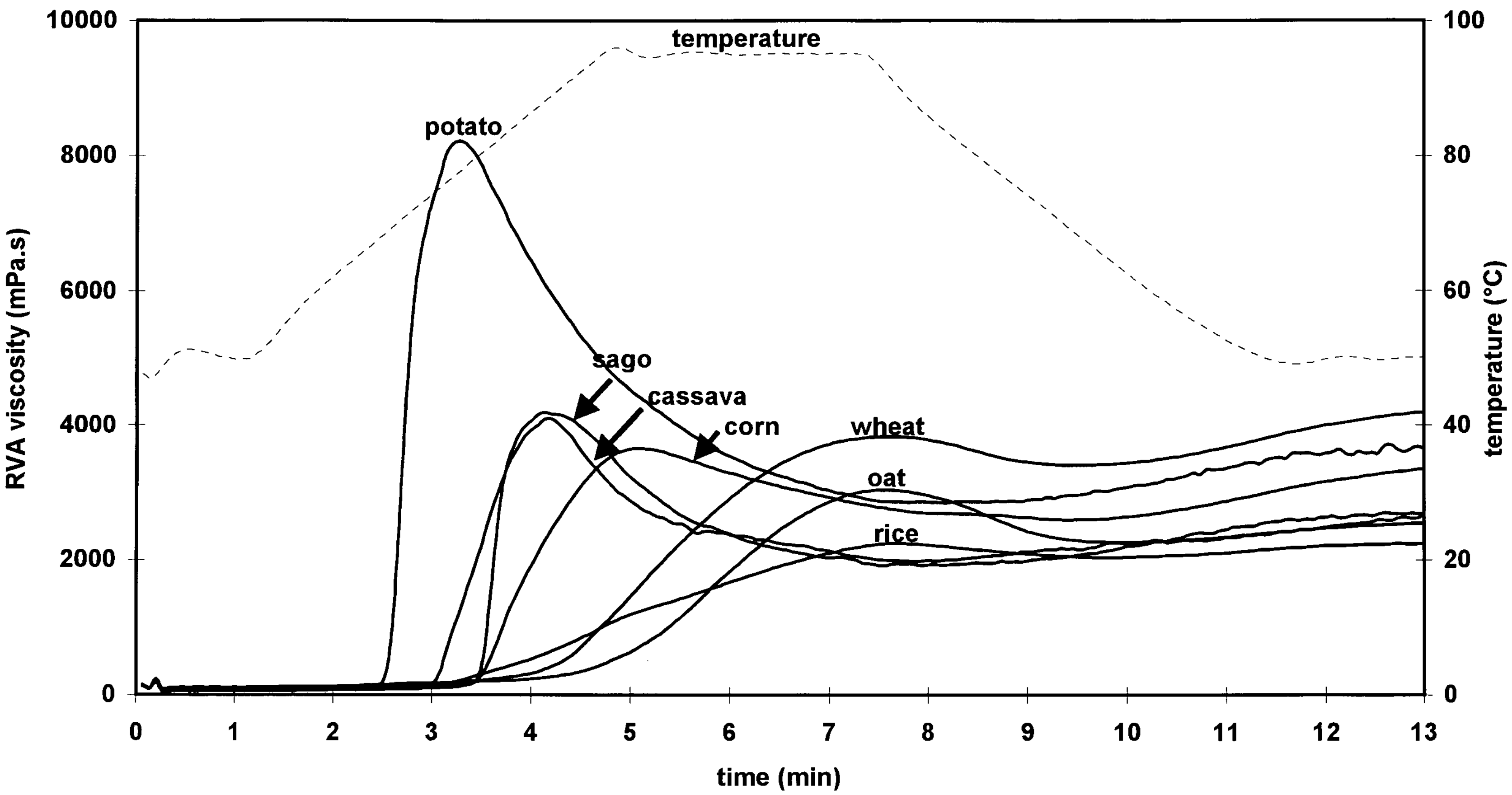


Figure 8. RVA profile for a range of starches from different botanical sources

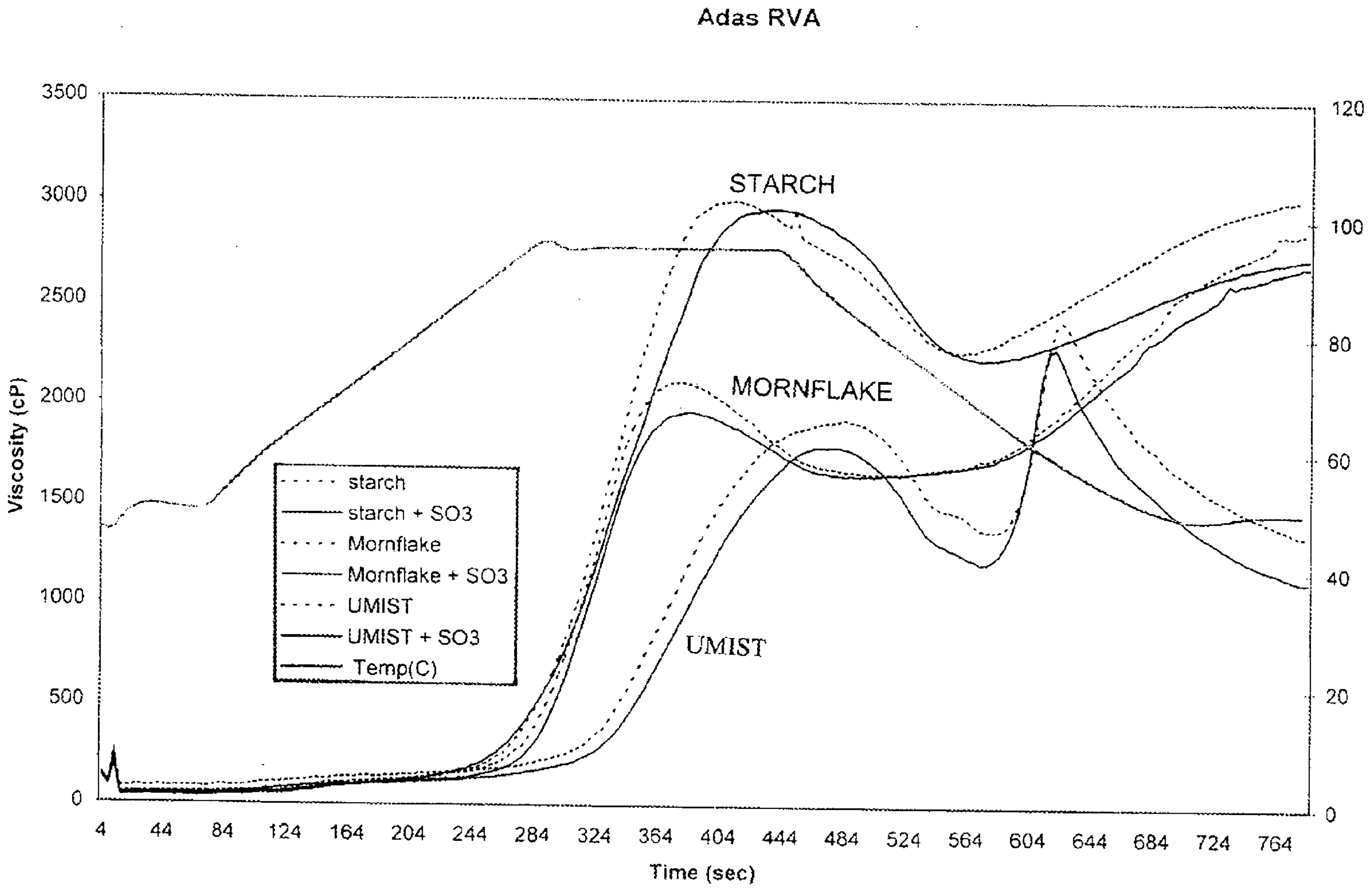


Figure 9. RVA profiles of purified starch, UMIST and Mornflake products

Table 4. Oat samples codes

SAMPLE CODE	LOCATION		VARIETY	IRRIGATION	AN Kg/ha	FU Kg/ha
G1	R		GERALD	NIL	0	0
G2					0	60
G3					40	0
G4					40	60
G5					100	0
G6					100	60
I1	R		IMAGE	NIL	0	0
I2					0	60
I3					40	0
I5					100	0
I6					100	60
N1		G	GERALD	NIL	40	NIL
N2					100	
N3					140	
IR1		G	GERALD	NIL	80	NIL
IR2				EARLY	80	
IR3				LATE	80	
V1	R		AINTREE	NIL	80	NIL
V2			CHAMOIS		80	
V4			EMPEROR		80	
V5			GERALD		80	
V7			IMAGE		80	
V9			KRYPTON		80	
V10			KYNON		80	
V11			LEXICON		80	
V12			SOLVA		80	

Table 5. Analyses carried out at Nottingham

	CODE	ANALYSIS														
		STARCH			β-GLUCAN			TOTAL PHENOLICS			ANTIOXIDANT ACTIVITY			EMULSIFIER		
		WH.	DEHULLED		WH.	DEHULLED		WH.	DEHULLED		WH.	DEHULLED		WH.	DEHULLED	
			S	B		S	B		S	B		S	B		S	B
	G3	▨	▨	▨	▨	▨	▨	✓	▨	▨	✓	▨	▨	✓	▨	▨
	N2	▨	▨	▨	▨	▨	▨	✓	▨	▨	✓	▨	▨	✓	▨	▨
	IR3	▨	▨	▨	▨	▨	▨	✓	▨	▨	✓	▨	▨	✓	▨	▨
	V4	▨	▨	▨	▨	▨	▨	✓	▨	▨	✓	▨	▨	✓	▨	▨
	V9	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	✓	▨	▨
	V12	▨	▨	▨	▨	▨	▨	✓	▨	▨	✓	▨	▨	✓	▨	▨
■	G1	▨	✓	▨	▨	✓	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓
	G5	▨	✓	▨	▨	▨	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓
	N1	▨	✓	▨	▨	✓	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓
	N3	▨	✓	▨	▨	▨	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓
	IR1	▨	✓	▨	▨	▨	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓
	IR2	▨	✓	▨	▨	▨	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓
■	V2	▨	✓	▨	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
■	V5	▨	✓	▨	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
■	V7	▨	✓	▨	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
■	V10	▨	✓	▨	▨	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
	V11	▨	✓	▨	▨	▨	✓	✓	▨	▨	✓	▨	▨	✓	▨	▨
	G2	▨	✓	▨	▨	▨	✓	▨	▨	▨	▨	▨	▨	▨	▨	▨
	G4	▨	▨	▨	▨	▨	▨	▨	✓	✓	▨	✓	✓	▨	✓	✓
	G6	▨	✓	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨
■	I1	▨	✓	▨	▨	✓	✓	▨	✓	✓	▨	✓	✓	▨	✓	✓
	I2	▨	✓	▨	▨	▨	✓	▨	▨	▨	▨	▨	▨	▨	▨	▨
	I3	▨	▨	▨	▨	▨	▨	▨	✓	✓	▨	✓	✓	▨	✓	✓
	I5	▨	✓	▨	▨	▨	✓	▨	▨	▨	▨	▨	▨	▨	▨	▨
	I6	▨	✓	▨	▨	▨	✓	▨	▨	✓	▨	▨	✓	▨	▨	✓
	V1	▨	✓	▨	▨	▨	✓	▨	▨	▨	▨	▨	▨	▨	▨	✓

KEY

✓	Analysis complete
▨	No analysis
■	Varieties selected by Du Pont

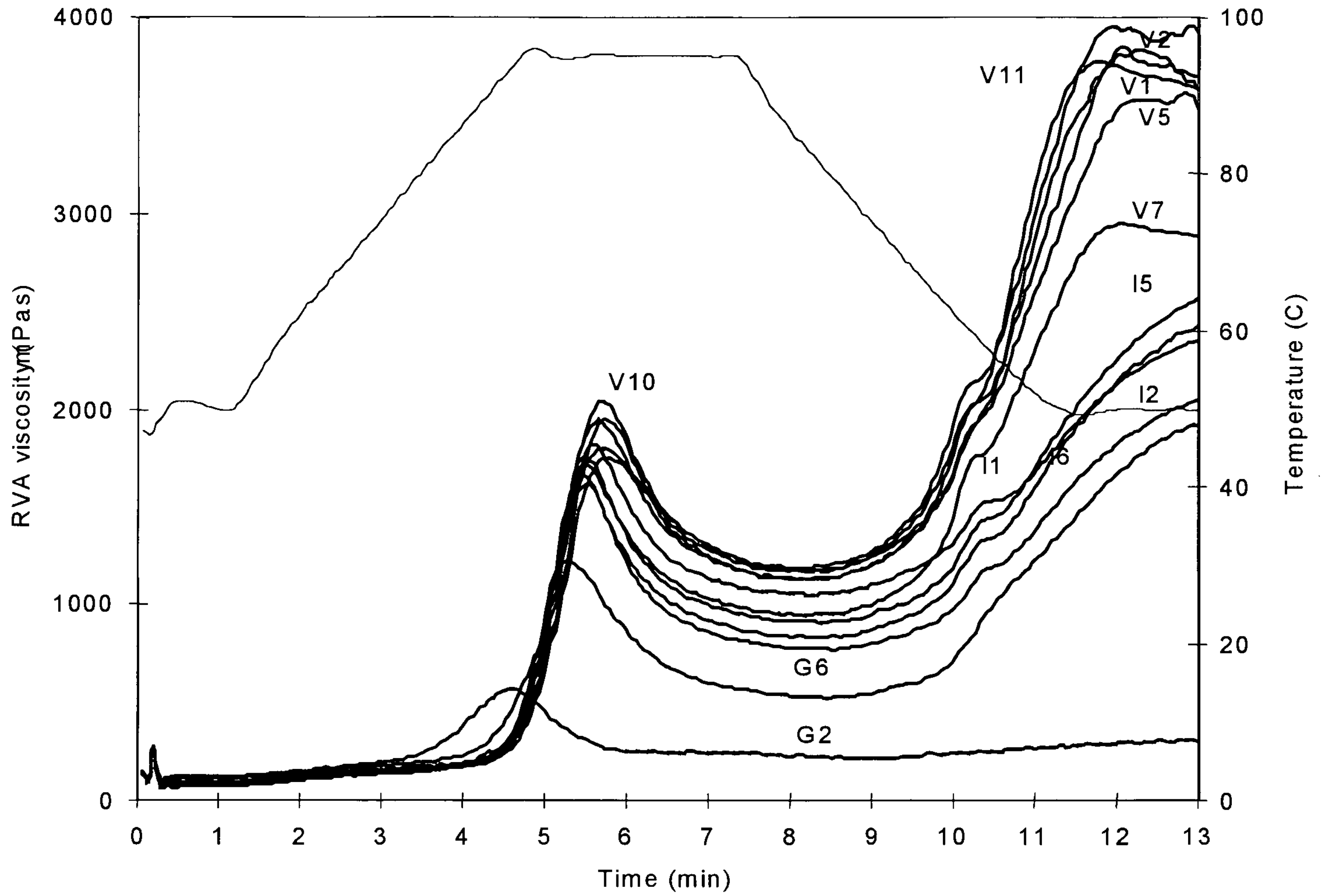


Figure 10. RVA profiles of some oat samples

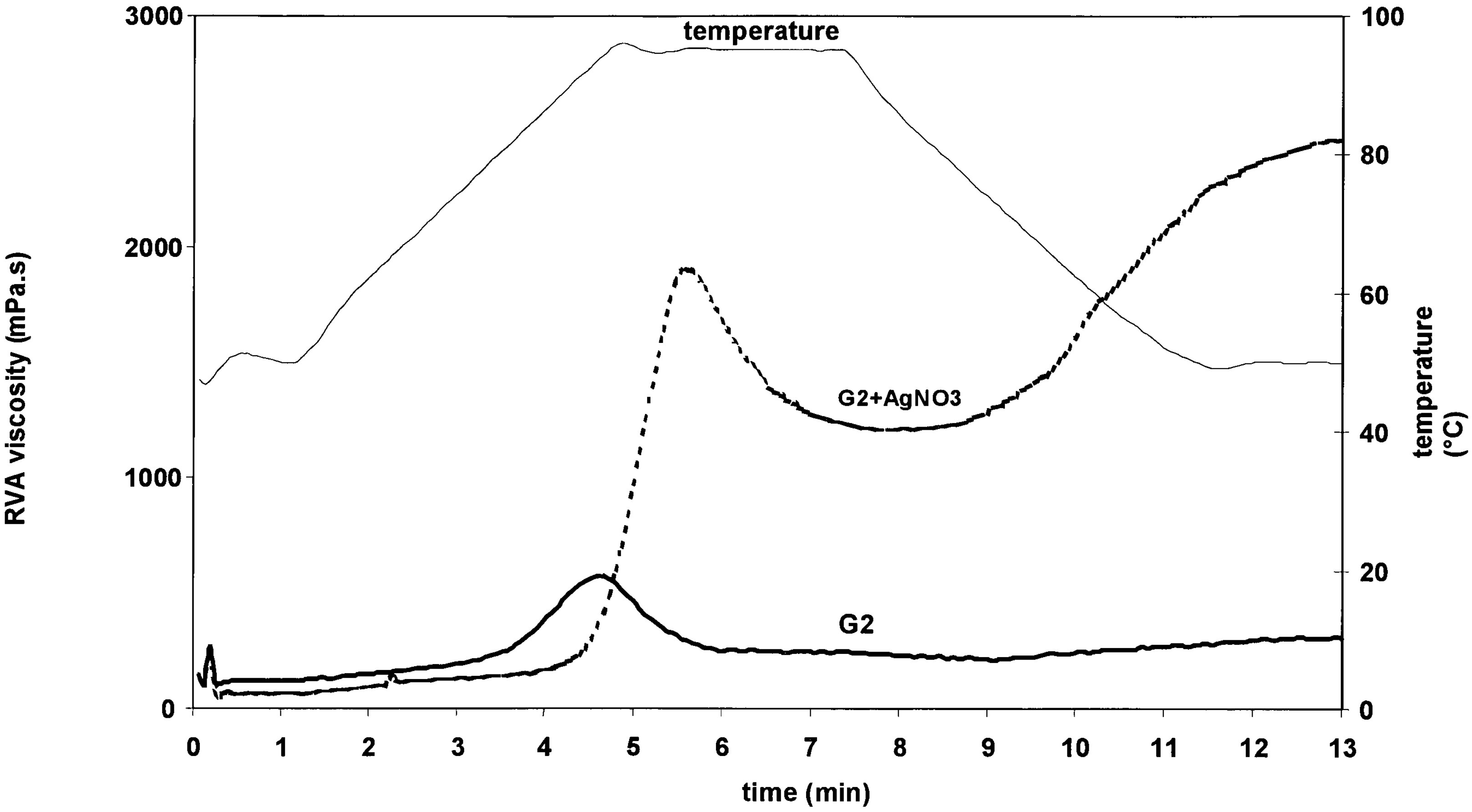


Figure 11. RVA profiles for sample G2 in the presence and absence of $AgNO_3$

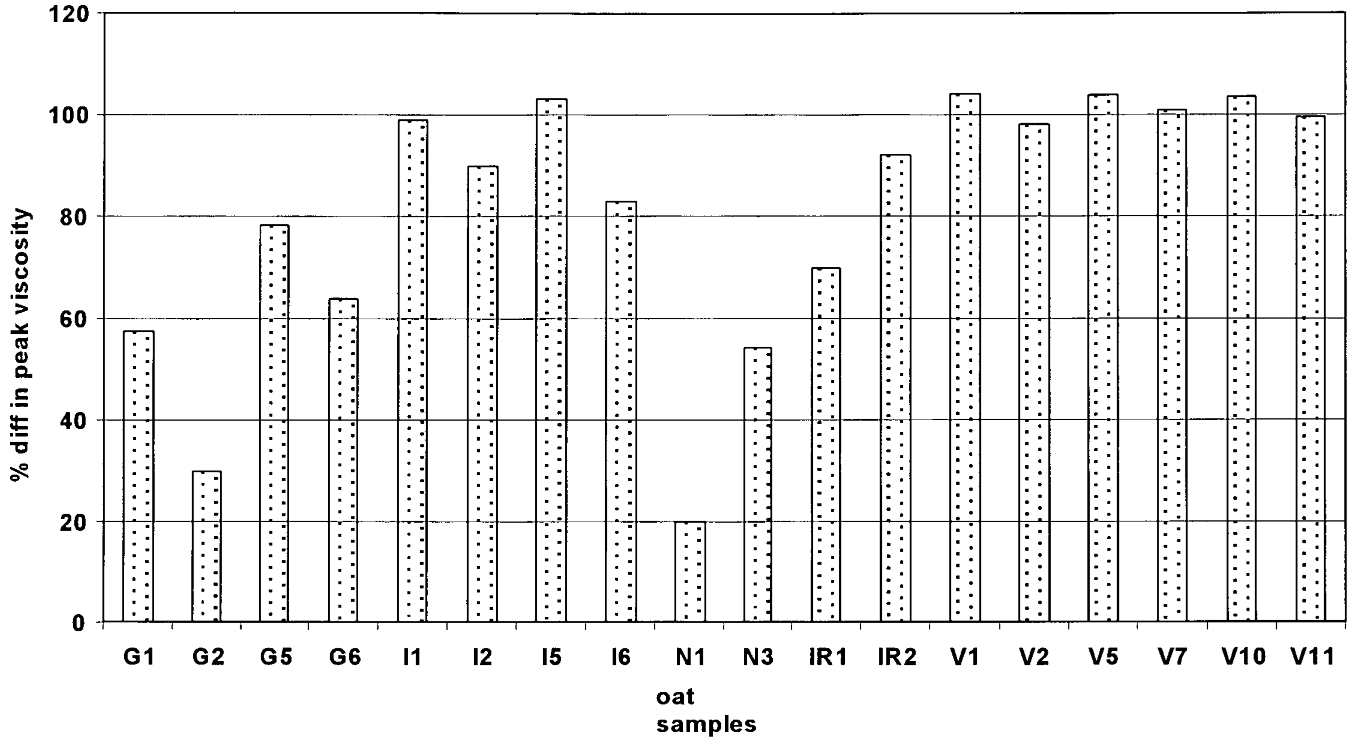


Figure 12. % Differences in RVA peak viscosity in the presence and absence of $AgNO_3$

Table 6 RVA data for oat samples (3g sample+25 g distilled water; pH 5.85 @24.6 degree C) using STD 1 temperature profile

Samples					**RVA viscosity (mPa.s)**								
	peak	difference	hold	difference	breakdown	difference	final	difference	setback	difference	time	temp	
G2	**571**	**1334**	**211**	**996**	**360**	**338**	**303**	**2163**	**92**	**1167**	4.67	50.20	**Lowest**
G2+AgNO3	*1905*		*1207*		*698*		*2466*		*1259*		*5.60*	*49.80*	
G6	1225	**698**	529	**692**	696	**6**	1923	**728**	1394	**36**	5.27	49.90	
G6+AgNO3	*1923*		*1221*		*702*		*2651*		*1430*		*5.53*	*50.25*	
I1	1818	**22**	1052	**147**	766	**-125**	2347	**34**	1295	**-113**	5.60	51.05	
I1+AgNO3	*1840*		*1199*		*641*		*2381*		*1182*		*5.73*	*49.70*	
I2	1619	**180**	772	**295**	847	**-115**	2052	**371**	1284	**72**	5.53	52.05	
I2+AgNO3	*1799*		*1067*		*732*		*2423*		*1356*		*5.60*	*51.10*	
I5	1752	**-49**	908	**142**	844	**-191**	2569	**7**	1661	**-135**	5.47	49.75	
I5+AgNO3	*1703*		*1050*		*653*		*2576*		*1526*		*5.53*	*49.80*	
I6	1672	**341**	833	**402**	839	**-61**	2426	**964**	1593	**562**	5.47	49.75	
I6+AgNO3	*2013*		*1235*		*778*		*3390*		*2155*		*5.73*	*49.80*	
V5	1944	**-71**	1134	**6**	810	**-77**	3537	**-366**	2403	**-372**	5.67	51.05	
V5+AgNO3	*1873*		*1140*		*733*		*3171*		*2031*		*5.60*	*51.10*	
V7	1717	**-15**	945	**23**	772	**-38**	2881	**-58**	1936	**-81**	5.53	49.75	
V7+AgNO3	*1702*		*968*		*734*		*2823*		*1855*		*5.53*	*49.70*	
V1	1811	**-81**	1168	**-81**	643	**0**	3635	**-202**	2467	**-121**	5.73	50.25	
V1+AgNO3	*1730*		*1087*		*643*		*3433*		*2346*		*5.73*	*50.20*	
V2	1749	**32**	1178	**34**	571	**-2**	3701	**-62**	2523	**-96**	5.73	49.70	
V2+AgNO3	*1781*		*1212*		*569*		*3639*		*2427*		*5.80*	*49.80*	
V10	**2042**	**-72**	**1189**	**-19**	**853**	**-53**	**3925**	**-442**	**2736**	**-423**	5.67	50.25	**Highest**
V10+AgNO	*1970*		*1170*		*800*		*3483*		*2313*		*5.73*	*50.25*	
V11	1947	**13**	1131	**26**	816	**-13**	3635	**-298**	2504	**-324**	5.73	49.70	
V11+AgNO	*1960*		*1157*		*803*		*3337*		*2180*		*5.73*	*49.70*	

Note:

peak = peak viscosity (maximum viscosity developed during or soon after the heating portion of the test)
hold = holding strength (minimum viscosity after the peak, normally occurring around the commencement of the sample cooling)
breakdown = peak viscosity minus trough (hold) viscosity
final = viscosity at the end of the test
setback = setback from trough (final viscosity minus trough viscosity)
time = peak time (peak at which the peak viscosity occurred, in minutes)
temp = pasting temperature (temperature where viscosity first increases at least 2 RV over a 20 sec. period using profile STD1, in degree C)
AgNO3 used = 0.02% (water basis)

β-glucan

The amounts of β-glucan recovered from the fractionations are shown in Table 7. There was a large difference in the amounts of β-glucan between the starch-rich stream and the bran-rich stream, i.e., in the starch-rich stream recoveries were between 1.5 and 3.9% and for the bran-rich stream recoveries were generally between 4.3 and 11% with only one sample showing a recovery of less than 4% β-glucan.

Table 7. Recovery of β-glucan (%w/w)

Sample	Bran-rich stream A	Bran-rich stream B	Starch-rich stream A	Starch-rich stream B
G1	n.d.	n.d.	2.2	1.8
G2	6.3	6.1	n.d.	n.d.
I1	4.3	5.0	2.6	2.5
I2	6.8	6.0	n.d.	n.d.
I5	3.5	4.6	n.d.	n.d.
I6	5.4	4.5	n.d.	n.d.
N1	n.d.	n.d.	3.9	2.5
V1	9.4	9.1	n.d.	n.d.
V2	9.6	11.0	2.2	2.0
V5	8.3	8.0	1.6	1.5
V7	7.7	6.5	1.7	1.7
V10	5.8	6.8	2.7	2.1
V11	8.5	8.0	n.d.	n.d.

A and B are replicate extractions

SEC/MALS is a suitable technique for the determination of molecular weights of β-glucan though there is no way of distinguishing between components if they elute together. For this reason a simple qualitative test (iodine test) was used to establish whether starch was still present in any of the β-glucan fractions. The presence of starch would be expected to have a significant effect on the molecular weights determined. The results are shown in Table 8.

Table 8. Presence of starch (iodine test, all samples tested after filtration)

Sample	Bran-rich stream A	Bran-rich stream B	Starch-rich stream A	Starch-rich stream B
G1	n.d.	n.d.	+++	+++
G2	-	-	n.d.	n.d.
I1	+++	+	+++	+++
I2	+	++	n.d.	n.d.
I5	+++	+	n.d.	n.d.
I6	++	+++	n.d.	n.d.
N1	n.d.	n.d.	+	-
V1	-	-	n.d.	n.d.
V2	-	-	++	+++
V5	-	-	++	++
V7	-	+	-	+
V10	-	-	+	+
V11	-	-	n.d.	n.d.

+++ very dark blue colour
++ blue colour
+ hint of blue colour
- no evidence of blue, same colour as blank (i.e., 0.1M NaCl)

In the bran-rich stream starch was found to be present in all β-glucan fractions from variety Image grown in the nitrogen trial at Rosemaund. In contrast, Image grown in the variety trial at Rosemaund contained only small amounts of starch, whilst none of the other varieties showed any evidence of starch. Virtually all samples analysed from the starch-rich stream showed some evidence that starch was still present after fractionation, with the Image variety for the Rosemaund nitrogen trials being the most heavily contaminated.

Weight average molecular weights found for the β-glucan fractions are shown in Table 9. Results from the bran-rich stream appear to be reproducible for duplicate injections of most samples and the difference between the A and B repeats is also (mostly) relatively low. The situation for the starch-rich stream was less satisfactory, in that the results for some repeat injections were reproducible while for others they were not. In addition, some repeats were run after 24 hours and this had a profound effect on molecular weight, mostly causing a decrease in molecular weight except for one sample (I1) where a considerable increase was found.

The lowest molecular weights found in the bran-rich stream were for the Image variety involved in the fertilizer field studies (Rosemaund nitrogen). All other samples (including Image in the varieties trial) showed weight average molecular weights ranging from

300,000 to 500,000 dalton, whereas the samples from the I-series all gave weight average molecular weights below 300,000 dalton.

Having established that starch was present in some of the β-glucan fractions, an attempt was made to correlate this with the molecular weights found. In the case of the fractions from the bran-rich stream it appeared that the presence of starch caused a slight increase in the weight average molecular weight (for example I1: sample A is heavily contaminated with starch, sample B appears to be less contaminated, their molecular weights differ with sample A showing the higher molecular weight). However, this trend could not be established for the starch-rich stream.

Table 9. Weight average molecular weights (M_w) obtained by SEC/MALS

	$M_w(\times 10^5)$			
Sample	Bran-rich stream		Starch-rich stream	
	A	B	A	B
G1	n.d.	n.d.	5.53 6.02	6.30 6.03
G2	3.82 3.84	4.14 3.98	n.d.	n.d.
I1	2.27 2.44	1.39 1.42	3.73 *3.17	2.40 *6.50
I2	1.76 1.45	2.37 2.36	n.d.	n.d.
I5	2.23 2.00	1.59 1.50	n.d.	n.d.
I6	2.20 2.27	2.74 2.99	n.d.	n.d.

Table 9 continued

	$M_w(\times 10^5)$			
Sample	Bran-rich stream		Starch-rich stream	
	A	B	A	B
N1	n.d.	n.d.	2.70	2.57
			2.84	2.17
V1	3.38	3.73	n.d.	n.d.
	3.02	3.41	n.d.	n.d.
V2	4.60	4.92	3.41	3.81
	4.69	4.55	*3.34	4.22
V5	5.12	5.28	1.67	5.18
	4.57	4.90	*0.45	5.41
V7	3.41	4.78	1.39	2.25
	3.51	4.96	0.89	2.08
V10	3.45	4.15	3.24	3.12
	3.48	4.00	2.78	*0.71
V11	3.16	4.00	n.d.	n.d.
	3.41	3.97	n.d.	n.d.

* Sample analysed 24 h after the corresponding sample

Molecular weight values for β-glucan reported by other workers vary from relatively low (27,500 – 63,000 [Acker et al. 1955], 16,000 – 26,800 [Podrazy, 1964]) to several million (2×10^6, Autio et al, 1987, 2.7 and 3×10^6, Wood et al,1991). Such discrepancies are probably related to differences in the origin of the oats, extraction procedures and also solvent conditions and methodology used for molecular weight determination. It is therefore impossible to compare our values with those obtained by other authors. It is also well known (Welch and Lloyd, 1989; Dawkins and Nnanna, 1993) that the presence of starch and endogenous β-glucanases complicates β-glucan determinations and that both of these components are extremely difficult to remove completely. Our results are consistent with these observations. Although it appears that the extraction process used was efficient in removing starch from a number of samples, there were some inconsistencies for duplicate samples, despite the fact that every effort was made to keep the fractionation process as uniform as possible throughout the whole of the sample series. In addition, for the samples where duplicate injections were performed on consecutive days, the molecular weight of samples decreased significantly (except for I1) which may be due to the presence of β-glucanases.

It seems to be quite difficult to draw definite conclusions from these experiments. It appears that the weight average molecular weights measured for the β-glucan from the starch-rich stream are actually lower than those from the bran-rich stream of the same sample. This is surprising as a higher molecular weight might be expected if there is contamination with starch, which seems to be the case for most of the samples. However, the molecular weight of some samples increased when starch was present, so the trend is highly inconsistent.

There were differences in the molecular weights found for the different varieties and growing conditions. However, a difference in molecular weight was also found for Image used in the nitrogen trial and Image from the varieties trial.

Antioxidants

Extraction of polar lipid extracts

Developmental work was carried out to evaluate methods for extracting polar lipids from oats which were enriched in total phenolics (TP) and had significant antioxidant activity. Initial work examined yield and activity of antioxidants extracted from oats by two previously reported extraction regimes. One method (adapted from Xing and White, 1997) was an extraction with methanol over 10 days at room temperature, whereas the other (adapted from Forssell *et al.*, 1992) used isopropanol (IPA) at 70°C for only two hours. Although the isopropanol method yielded slightly lower values of TP (70% of the amount obtained using the methanol extraction) this method was considered to be most suitable for use in the current study. This is due firstly to savings in time and solvent usage (1.4 litres, 2 h for isopropanol compared to 10 litres, 10 days with methanol) and secondly, it was possible to extract the emulsifier with the same procedure.

At an absolute level the quantitative recovery of lipids using this method (5-6% total lipids relative to the total mass of flour) is comparable to amounts quoted in the literature and reported by UMIST during some of their preliminary milling studies. Furthermore, the proportion of this extract which is composed of polar lipids (as determined gravimetrically after the fractionation step) is between 7 – 18 %. This compares well with the values quoted by Forssell (1992) (14 – 23 %), who fractionated crude oat oil into neutral and polar fractions using a supercritical fluid extraction method.

The reliability of the IPA lipid extraction and column fractionation method was checked by measuring the TP values of three different polar lipid extracts obtained from the same oat sample. The following TP values were obtained: 94 mg/kg; 103 mg/kg and 113 mg/kg. These measurements gave an average of 103 mg/kg and a STDEV of 9.5 mg/kg. This recovery of total phenolic compounds in the polar lipid fraction of oats is comparable to values quoted in the literature (171 mg/kg Daniels and Martin, 1967; 30 mg/kg Tian and

White, 1994; 230 mg/kg Xing and White). The method was thus deemed to be suitable for use in the study.

Polar lipid extracts in 7.5 ml methanol were stored in the dark at -80°C and were shown to retain antioxidant activity for at least 6 months.

Measurement of antioxidant activity

The use of chemiluminescence to measure antioxidant activity of polar lipid extracts from oats was evaluated and found to be suitable for this purpose. In summary, antioxidant activity of the oat extracts, as measured by chemiluminescence, was at least comparable with the activity of an oat polar lipid fraction (Xing and White 1997) which was added to soya bean oil in an accelerated storage trial. Chemiluminescence offers the potential to screen oats rapidly for their antioxidant content and to monitor any industrial process designed to recover the active antioxidants from oats. Significantly higher sensitivity (at least two orders of magnitude) was achieved using chemiluminescence compared with using β-carotene bleaching (Marco, 1968). Consequently, large batches of oats (kilograms) are not required for laboratory scale extraction of oat antioxidants; gram or milligram batches of oats/oat flour may be sufficient to extract and measure antioxidant activity.

Effect of oat variety on total phenolic content and antioxidant activity

Initial work involved the analysis of whole oat flour samples which were prepared at Nottingham. The aim was to investigate the effect of oat variety and growing conditions on the antioxidant activity of polar lipid extracts.

The variety experiment utilised oat samples which had been grown at ADAS Rosemaund. These samples were grown in experimental plots but agronomic inputs were as standard commercial rates (80 kg/ha nitrogen as ammonium nitrate). The varieties analysed were as follows:

V2	Chamois
V4	Emperor
V5	Gerald
V7	Image
V10	Kynon
V11	Lexicon
V12	Solva

Total Phenolic Content (TPC) results are shown in Figure 13 and antioxdant activities in Figure 14. All samples appear to have similar concentrations of phenolic compounds in their polar lipid extract. A single factor ANOVA was nevertheless carried out, and revealed some significant differences in TPC between varieties. A SED (standard error of the difference) value of 3 was calculated.

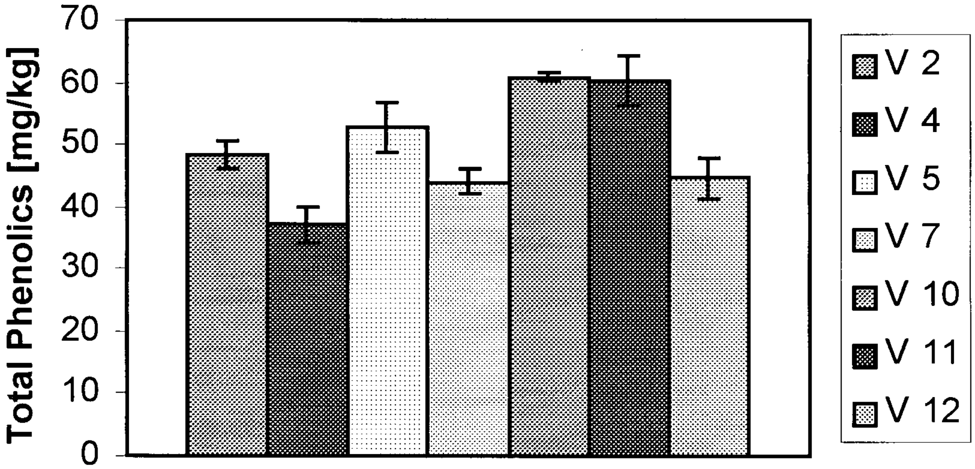


Figure 13. Effect of oat variety on TP content (Rosemaund)

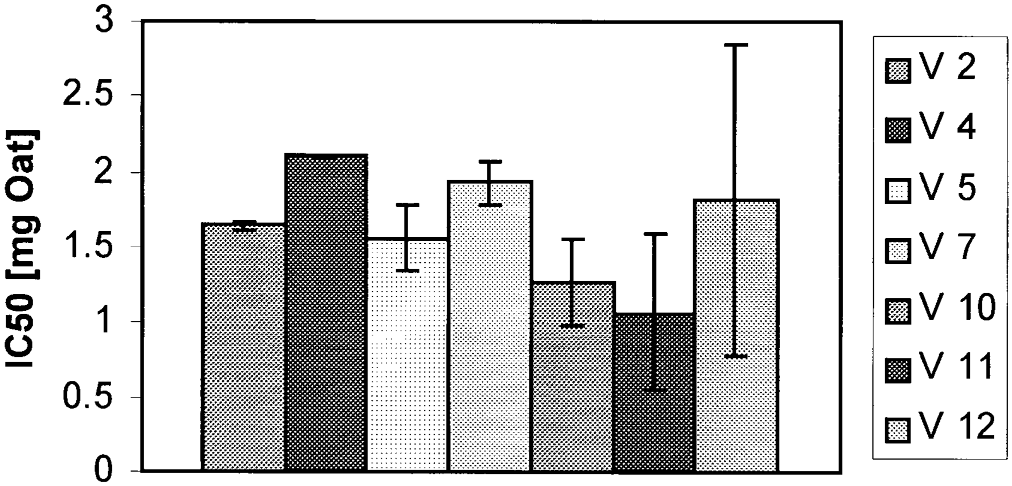


Figure 14. Effect of oat variety on antioxidant activity

In the Newman-Keuls Diagram shown in Figure 15 the average values of TP for the different varieties are compared. Lines connect samples that show no significant differences. For instance the TP content of V4 is significantly lower than that of V7 and V12. The TP contents of V7 and V12, however, are not different. The samples V2 and V5 have mid-range TP contents, whereas V11 and V10 have the highest levels.

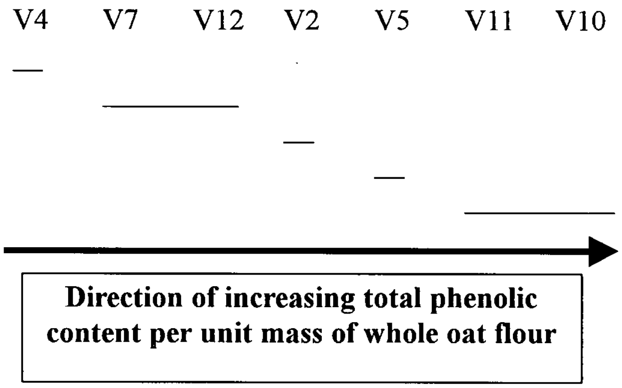


Figure 15. Newman-Keuls diagram for TP content of different oat varieties

The ranking of oat varieties from low antioxidant activity (high IC_{50} value) to high antioxidant activity (low IC_{50} value) matches, almost exactly, the ranking of oat varieties from low TP content values to high ones (see Figure 16), the only difference being that V10 and V11 swap positions. There is therefore a close correlation between the total phenolic content of the polar lipid extracts and their antioxidant activity.

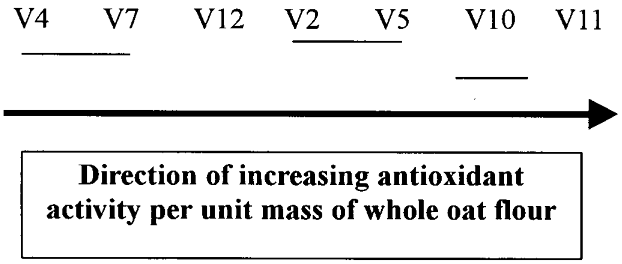


Figure 16. Newman-Keuls diagram for antioxidant activity of different oat varieties

Statistically significant differences in the IC_{50} values between varieties were calculated (V11 and V12 data), which had broad standard deviations, and were removed from the calculation. The connecting lines shown in Figure 16 above, therefore, only refer to samples V2, V4, V5, V7 and V10.

Effect of applied nitrogen on total phenolic content and antioxidant activity

The experiment was carried out on variety Gerald at two ADAS sites, Rosmaund (G) and Gleadthorpe (N). The experiment examined different amounts of nitrogen applied to the plots as ammonium nitrate (AN).

G1	0 kg/ha AN
G3	40 kg/ha AN
G5	100 kg/ha AN
N1	40 kg/ha AN
N2	100 kg/ha AN
N3	140 kg/ha AN

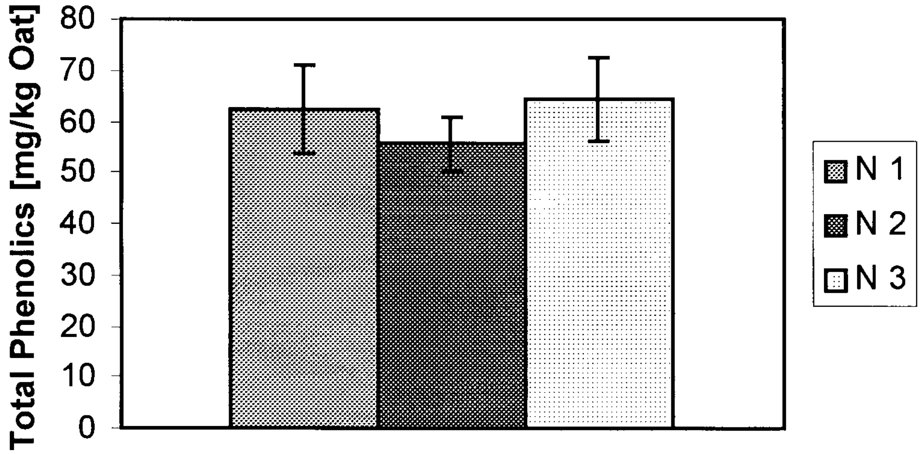


Figure 17. Effect of applied nitrogen on total phenolic content (Gerald, Gleadthorpe)

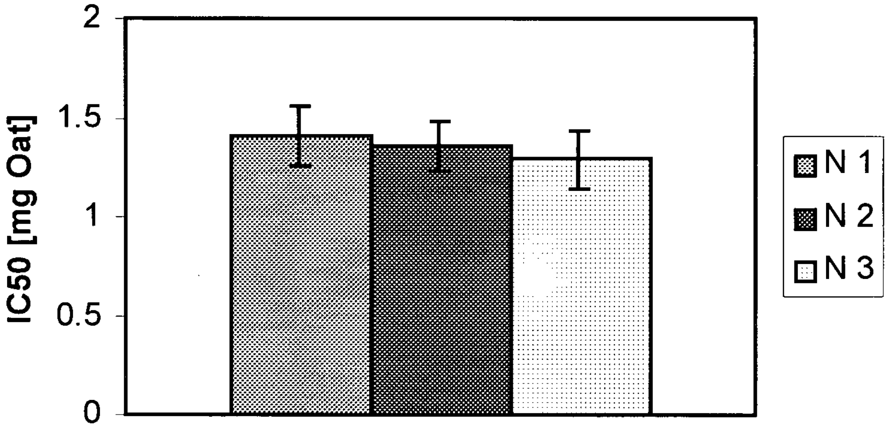


Figure 18. Effect of applied nitrogen on antioxidant activity (Gerald, Gleadthorpe)

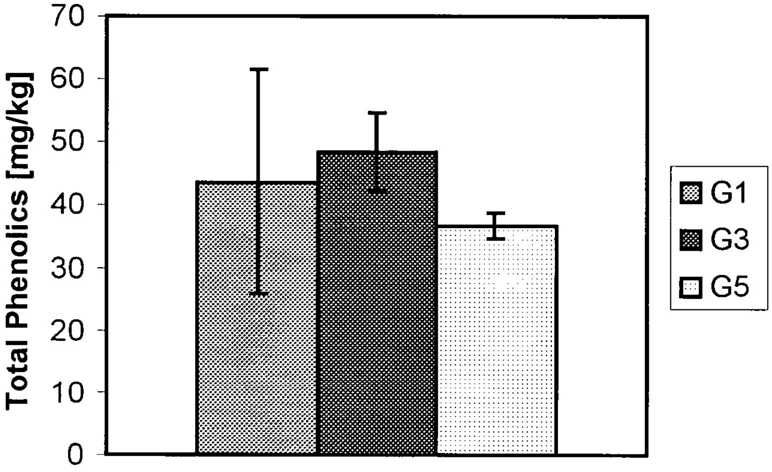


Figure 19. Effect of applied nitrogen on total phenolic content (Gerald, Rosmaund)

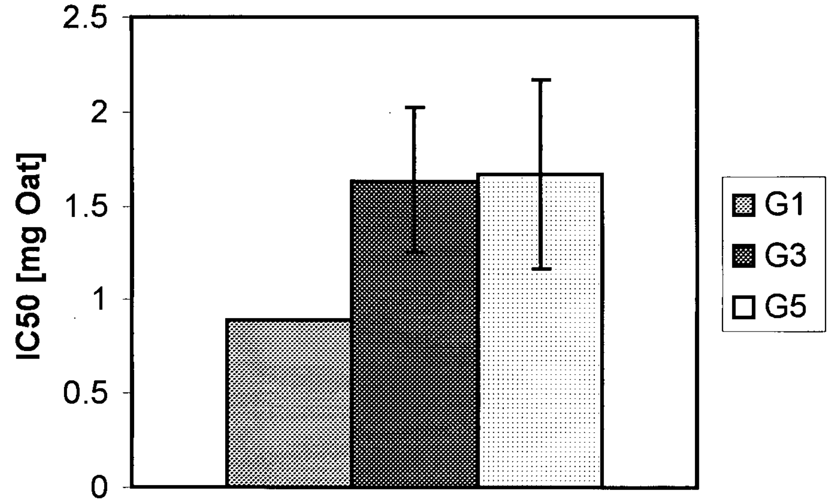


Figure 20. Effect of applied nitrogen on antioxidant activity (Gerald, Rosmaund)

TP contents ranged from 50 - 70 mg/kg, and IC_{50} values ranged from 1 to 1.5 mg oat. From the results of single factor ANOVA tests it was shown that at both sites there are no significant differences in TP content and IC_{50} values between samples grown with a range of applied nitrogen fertiliser treatments. Therefore it can be assumed that application of nitrogen in this study has no effect on the formation of phenolic compounds and their contribution to antioxidant activity.

Effect of growing location on total phenolic content and antioxidant activity

From the above data, samples G3 (from Rosmaund) and N1 (from Gleadthorpe) are broadly comparable since they were derived from the same seed source and grown with the same nitrogen input. Both samples show similar TP values ranging from 50 to 60 mg/kg and similar antioxidant activity of 1.4-1.6 mg oat (see Table 10). A single factor ANOVA test gave no significant differences in TPC and IC_{50} values for samples grown at different locations.

Table 10. Total phenolic concentration and antioxidant activity of oat variety Gerald grown at Rosemaund (GS) and Gleadthorpe (N1)

Sample	(A) [mg/kg] TP	(B) [mg/kg] TP	Average [mg/kg]
G3	53	44	49
N1	68	56	62
Sample	(A) IC_{50} [mg Oat]	(B) IC_{50} [mg Oat]	Average [mg Oat]
G3	1.5	1.8	1.6
N1	1.5	1.3	1.4

Effect of soil water availability on total phenolic content and antioxidant activity

This experiment was carried out on variety Gerald at Gleadthorpe applying nil (IR1), early (IR2), and late (IR3) irrigation in order to modify soil water availability.

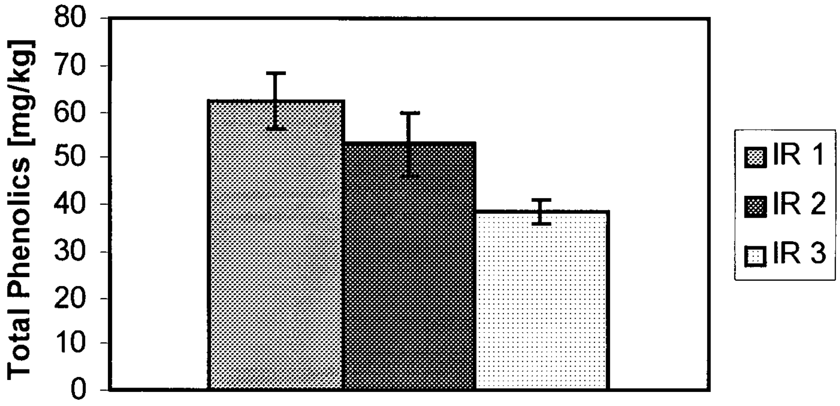


Figure 21. Effect of irrigation on total phenolic content (Gerald, Gleadthorpe)

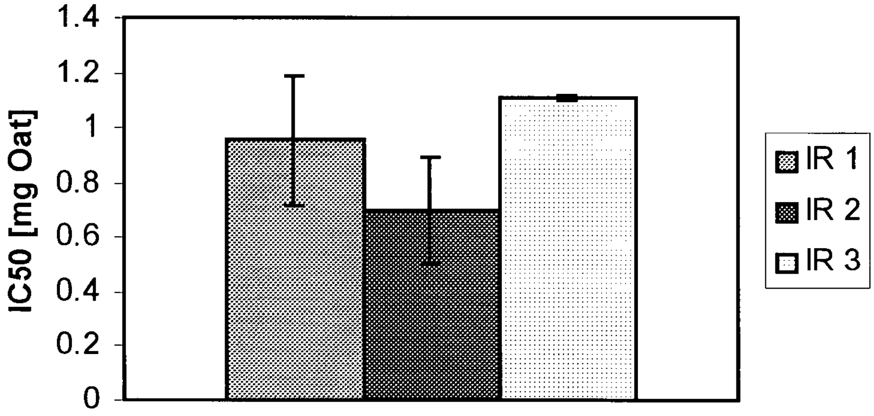


Figure 22. Effect of irrigation on antioxidant activity (Gerald, Gleadthorpe)

There is a decrease in the TP content values in the order of nil to early to late irrigation (see Figure 21). A single factor ANOVA test proved that these differences are significant.

The IC_{50} values all lay in a range between 0.7 and 1.1 mg oat (see Figure 22). A single factor ANOVA test revealed no significant differences between these average values. There is perhaps an indication of potential differences in the IC_{50} values between samples. The ANOVA P value = 0.21 and the SED value=0.18. If this SED value is used as a yardstick it reveals a difference between IR2 and the other samples. It is possible that the high IC_{50} reading for the IR1 sample alone has compromised the statistical impact of the analysis.

Effect of milling on antioxidant activity

An important element to this project is the fractionation of oats through milling to yield starch-rich and bran-rich fractions and it is necessary to ascertain the distribution of the antioxidant activity between these two fractions. The work described below was aimed at comparing the antioxidant activities in the two mill streams and also their activity measured in the equivalent whole oat flour.

The results of this study are shown in Table 11. The relative mass values show that, in general, the starch-rich stream has more mass than the bran-rich stream (approximately 60% and 40% respectively). In terms of total phenolic (TP) content, the concentration is enriched in the bran fraction by between 52% and 196% in oat varieties G1, G5, N1, N3, IR1 and IR2. The relative concentrations of TP content between the two fractions are inverted in the remaining fractions with higher concentrations (enrichment of between 34% and 104%) being found in the starch fractions. Closer analysis of the data using unpaired t-tests revealed that the majority of these differences in TP concentration between the starch and bran-rich fractions are statistically relevant. The two exceptions were IR2 and V7, which showed statistical differences only at the 15% and 23% levels respectively.

Table 11. Comparison of total phenolic content and antioxidant activity values in starch-rich and bran-rich fractions

	% Mass of Dehulled sample		**TP content (mg/kg fraction)**		**IC_{50} value (mg fraction)**		**IC_{50} phenolic equivalent value (ng)**	
	starch	**bran**	**starch**	**bran**	**starch**	**bran**	**starch**	**bran**
G1	63.6	36.4	31	56	1.63	0.51	51	29
G5	63.8	36.2	23	68	2.47	0.70	57	48
N1	51.6	48.4	42	106	1.59	1.00	67	106
N3	46.8	53.2	46	86	2.18	0.58	100	50
IR1	51.6	48.4	62	94	0.53	0.29	33	27
IR2	53.2	46.8	53	84	1.37	0.40	73	34
V2	59.4	40.6	98	48	0.58	0.67	57	32
V5	59.5	40.5	111	55	0.69	0.86	77	47
V7	61.0	39.0	71	53	0.81	0.76	58	40
V10	52.6	47.4	86	52	0.73	0.74	63	38

The distribution of phenolic compounds between the two fractions was on the whole mirrored by the 'distribution' of antioxidant activity (IC_{50} values). If an oat variety showed enrichment of phenolics in the bran fraction (e.g. G1) there was an accompanying enrichment of antioxidant activity (i.e. a relative decrease in the IC_{50} value). As before, an equivalent statistical analysis revealed statistical differences in the IC_{50} values between the two streams in all the varieties except V5, V7 and V10.

IC_{50} phenolic equivalent values indicate the mass of phenolic compounds in the sample which cause a 50% quench in the chemiluminscence. This is simply calculated from the IC_{50} and TP data. A low IC_{50} phenolic equivalent value would therefore indicate that a population of active phenolic compounds exists; if a relatively higher value is calculated a population of less active phenolic compounds exists. The latter situation may result from the dilution of active phenolics with a relatively high concentration of phenolic compounds with reduced antioxidant activity, or it could be explained by the loss of synergistic phenolic compounds from the overall population of phenolic compounds. The IC_{50} phenolic equivalent results (see Table 12) show that the bran fraction, from all the varieties tested (with the exception of N1), contains a more potent mixture of phenolic compounds than the starch-rich fraction.

From these findings one can summarise that the bran-rich fraction of the majority of the oat varieties tested is enriched with antioxidant activity compared to the starch-rich fraction. Statistical analysis of the IC_{50} data for the bran-rich fractions would reveal any differences between the varieties tested in terms of their antioxidant activity in this fraction. An ANOVA test showed that differences do exist and the Newman-Keuls representation of the results is shown below.

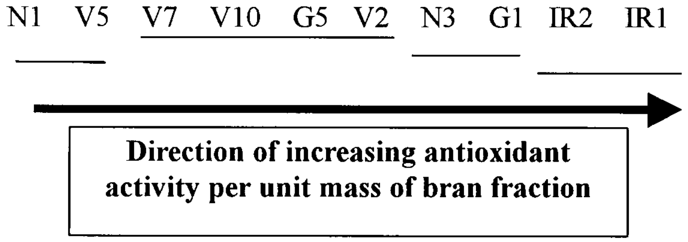


Figure 23. Newman-Keuls diagram of the IC_{50} data in bran fractions from a range of oat varieties

From this analysis of the IC_{50} data, bran fractions from IR1 and IR2 contain the highest antioxidant activity per unit mass of bran fraction compared with the bran fractions from the other oat varieties tested. This is consistent with the whole oat flour data in section 2 where the IR samples were found to have higher IC_{50} values than the other varieties tested. Further comparison of bran-rich antioxidant activity data with that described for whole flour fractions reveals some discrepancies though. For example, although both sets of data (Figures 14 and 23) show variety-dependent differences in the antioxidant activity, the ranking of these differences is altered. Similarly, if one compares the results for N1 and N3 in Figure 23 with those in Figures 17 to 20, an applied nitrogen dependent difference in the antioxidant activity is apparent in the bran-rich fractions which was not manifested in the whole flour samples.

Comparison of antioxidant activity in whole and fractionated oat flour

Results for the total phenolic content and the antioxidant activity of starch and bran-rich oat fractions from section 3.1 above can be combined mathematically to represent the values that may have been obtained if the fractions had been combined in the appropriate relative amounts to form reconstituted whole flour. A comparison of results obtained for this theoretical reconstituted oat flour or 'dehulled flour' with actual whole oat flour data is shown in Table 13 and Figures 24 and 25.

Table 12. Comparison of TP and antioxidant activity in whole flour and in starch-rich and bran-rich fractions

	TP content (mg/kg sample)				**IC_{50} value (mg sample)**				**IC_{50} phenolic equivalent value (ng)**			
	Whole flour	Dehulled flour			Whole flour	Dehulled flour			Whole flour	Dehulled flour		
		starch[a]	bran[a]	SUM[b]		starch[a]	bran[a]	SUM[b]		starch[a]	bran[a]	SUM[b]
G1	43	19.7	20.4	40.1	0.90	1.04	0.18	1.22	39	20	4	24
G5	37	14.7	24.6	39.3	1.67	0.89	0.45	1.34	62	13	12	25
N1	62	21.7	51.3	73.0	1.41	0.82	0.48	1.30	87	18	25	43
N3	64	21.5	45.8	67.3	1.29	1.03	0.31	0.34	83	22	14	36
IR1	62	32.1	45.4	77.5	0.96	0.27	0.14	0.68	60	9	6	15
IR2	53	28.2	38.8	67.0	0.70	0.72	0.19	1.00	37	20	7	27
V2	48	58.1	19.5	77.6	1.65	0.34	0.27	0.56	79	20	5	25
V5	53	66.0	22.3	88.3	1.57	0.41	0.34	0.75	83	27	8	35
V7	44	43.3	20.7	64.0	1.93	0.49	0.30	0.86	85	21	6	27
V10	61	45.2	24.6	69.8	1.28	0.38	0.35	0.62	78	17	9	26

[a] Contribution of this fraction to reconstituted dehulled oat flour (values in table C2 corrected for % mass factor)

[b] Projected values for reconstituted dehulled oat flour

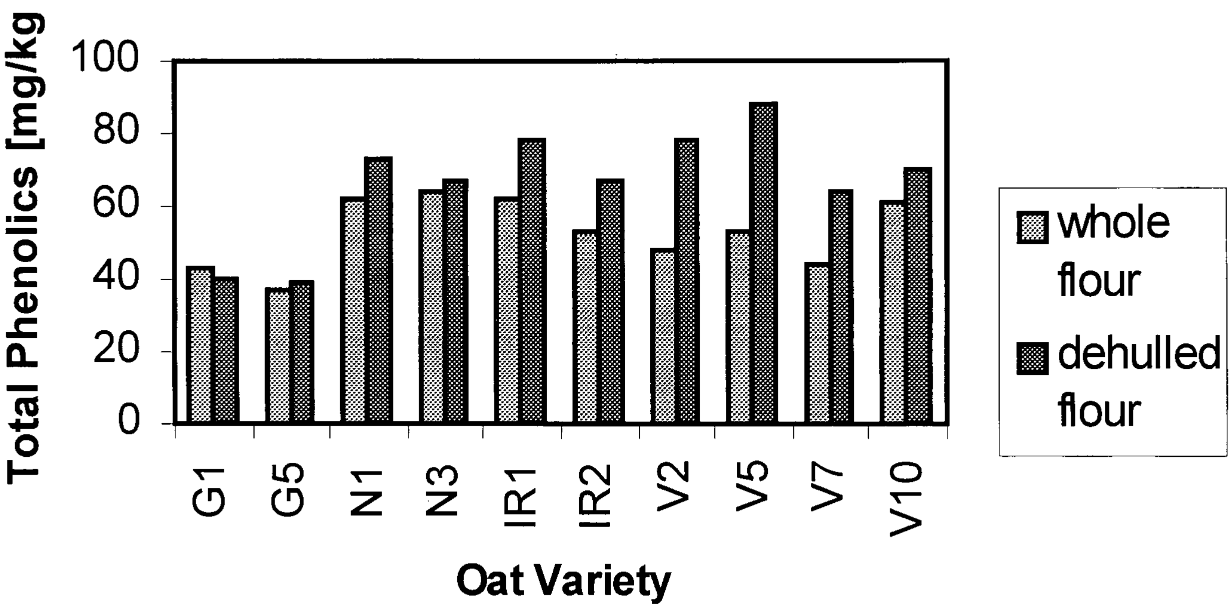


Figure 24. Comparison of TP values between whole and dehulled oat flour

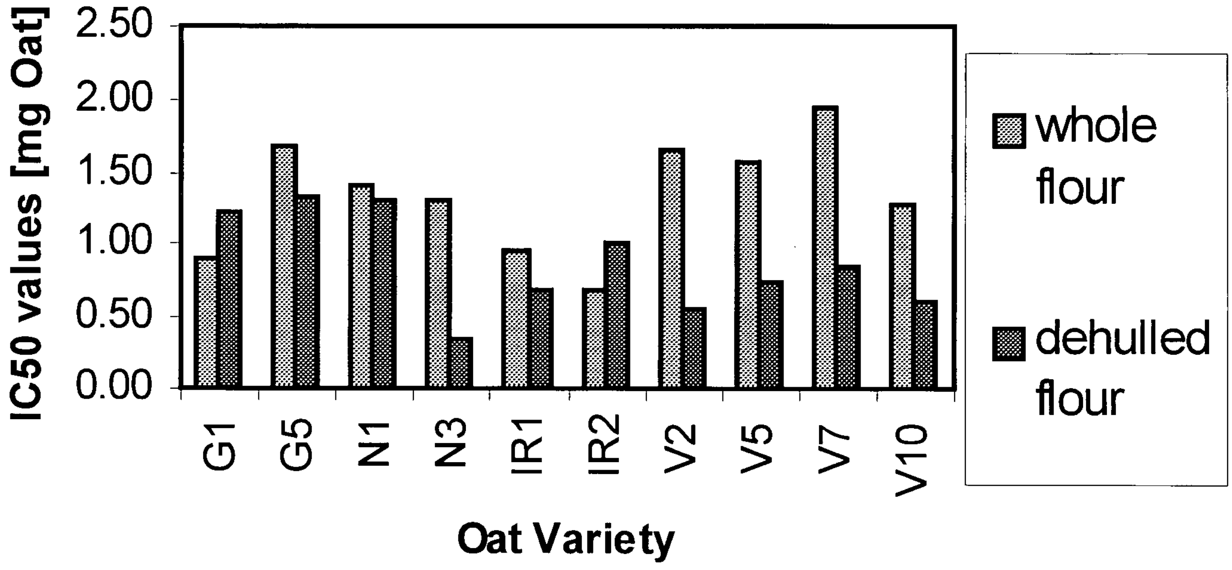


Figure 25. Comparison of IC_{50} values between whole and dehulled oat flour

A brief scan of the data shows clearly that measurable antioxidant activity of a similar order of magnitude was found both in whole flour and in the fractionated flour. Given that the dehulled oats were stored for a period of weeks or months before milling one can state that the process of dehulling and storage did not appear to adversely affect the antioxidant activity within the kernel.

Total phenolic content tends to be higher in the dehulled flour than the whole flour. This could be easily explained by a dilution effect of material from the hull component increasing the mass of the whole flour without contributing to the total phenolic mass.

The differences in the TP values between whole and dehulled oat flour tend to be inverted when one compares the differences in the IC_{50} values. This is consistent with higher TP resulting in higher antioxidant activity (i.e. lower IC_{50} values). In general this inversion is disproportionate and suggests that the increase in antioxidant activity in the theoretical reconstituted oat flour (dehulled flour) can not be fully explained by an increase in the total phenolic content. One must not forget that the dehulled flour IC_{50} values are obtained by extrapolating the starch-rich and bran-rich data. It is possible that actual reconstitution might result in some inhibition effects, leading to IC_{50} values more in line with the whole flour results. One may however speculate that the separation of phenolic compounds by milling could be selective, resulting in distinct populations in the starch and bran-rich fractions. These populations may contain phenolic compounds with enhanced synergistic antioxidant activity, which causes a decrease in the IC_{50} value. This possibility is clearly illustrated if one compares the IC_{50} phenolic equivalent data between the dehulled and whole flour samples.

Emulsifier

An absolute measurement of the glycolipid emulsifier (GE) concentration in the samples was not possible because there was no standard material available. The relative amounts were determined by measuring the intensity of all samples against an arbitrarily chosen sample i.e. polar lipid material extracted from N3 whole flour.

Table C4 shows the results from this investigation. A single factor ANOVA test confirmed that there are no significant differences in the GE content between different oat varieties, and between different agronomic treatments. Values obtained for the starch or bran fractions are slightly higher than those for whole oats. This could be explained by a dilution effect on the GE in the whole flour due to the presence of hull material with a negligible amount of emulsifier present. The concentration of GE in the starch-rich and the bran-rich fractions is similar.

Table 13. Relative concentration of glycolipid emulsifier in polar lipid extracts from oats

		EMULSIFIER CONTENT (Relative to N3 = 1.00)		
		WHOLE	**DEHULLED**	
			S	**B**
	G3	1.10, 0.88		
	N2	0.78, 0.98		
	IR3	1.04, 0.97		
	V4	0.83, 0.88		
	V9	0.99, 1.25, 0.84		
	V12	1.02, 0.98		
▨	**G1**	0.99, 1.06	1.07, 1.11	0.96, 0.88
	G5	1.01, 0.93	1.18, 1.21	1.04
	N1	1.11, 0.94	0.91, 1.12	1.15
	N3	1.00, 0.92	1.03, 1.09	1.06, 0.98
	IR1	1.04, 0.93	1.10, 0.99	1.22, 1.40
	IR2	1.05, 0.81	1.21, 1.44	1.26, 1.07
▨	**V2**	0.85, 1.01	1.30, 1.33	1.23, 1.09
▨	**V5**	1.08, 0.86	1.27, 1.22	1.25, 1.13
▨	**V7**	0.88	1.19, 1.26	1.16, 1.16
▨	**V10**	1.06, 1.02, 1.14	1.02, 1.24	0.95
	V11	1.07		
	G4		1.16, 1.20	1.11
▨	**I1**		0.35	0.94, 0.98
	I3		1.28, 1.28	1.58
	I6			1.32, 1.39
	V1			0.93

Conclusions

Starch

- The starch granule size and gelatinisation temperatures for the test samples fitted with the data previously reported for oat starch.
- From pasting curve data, "pure" oat starch had a late and low viscosity peak compared to all other starches tested. The only sample with a lower profile was rice.
- The starch-rich fraction from the milling trials appeared to contain other material. All the samples had a similar gelatinisation temperature, but there were major differences in the maximum paste viscosities.
- The differences in pasting viscosities could be greatly reduced by the presence of silver ions. It has therefore been assumed that several samples contained high levels of active amylase.
- No significant amylase activity was observed in the starch-rich fractions form the variety trials.
- Gerald and Image samples from the non-variety trials contained amylase activity in their starch-rich fractions.
- Gerald oats from the irrigation and nitrogen trials contained the highest levels of amylase activity.

β-glucan

- Recovery of β-glucan was increased and was more reproducible if the fat was removed from the oat samples prior to extraction.
- The β-glucan samples were frequently contaminated with starch.
- β-glucan was enriched in the bran-rich fraction compared to the starch-rich fraction.
- The average molecular weight for the β-glucan was approximately 3.1×10^5, with values ranging from 1.4 to 6.3×10^5.
- Storage of some samples was also found to cause a marked decrease in their molecular weights. It was thought that this was due to endogenous enzymes, rather than secondary contamination.

Antioxidants

- Heated isopropanol extraction followed by solid phase separation is an effective and reproducible method for preparing a polar lipid extract from oats, which exhibits significant antioxidant activity.
- Flow through chemiluminescene can be used to measure the antioxidant activity in oat extracts. It is particularly well suited to this study where its sensitivity precludes the need to extract lipids from large batches of oats.
- In general, as the total phenolic content of the extract increases so does the antioxidant activity.

- Oat variety influences the antioxidant activity. The ranking of oat varieties according to their antioxidant activity changed after selective milling into bran-rich and starch-rich fractions. Of the varieties which were analysed as whole flour and fractionated flour one may tentatively propose, from an overall comparison of ranking data that, in terms of antioxidant activity, V10 (Kynon) shows a lot of promise, V5 (Gerald) and V2 (Chamois) are good and V7 (Image) is poor.
- No effect of location or fertiliser treatment (ammonium nitrate or foliar urea) on antioxidant activity is evident from the whole flour data. This conclusion is undermined by the fractionated flour results.
- Nil and early irrigation (Gerald, Gleadthorpe) samples performed consistently well, exhibiting the highest antioxidant activity in whole flour and bran-rich fractions. Comparison of the actual rainfall at the two sites may allow further insights into this significant result.
- Dehulling oat samples followed by storage for several weeks had no detrimental effect on the antioxidant activity.
- Selective milling of oats into starch-rich and bran-rich fractions caused a quantitative (IC_{50} results) and qualitative (IC_{50} phenolic equivalent results) enrichment of antioxidant activity into the bran-rich fraction. If one assumes that the lower IC_{50} phenolic equivalent results in the bran-rich fraction are not due to the enrichment of a stimulating compound in this fraction, nor due to the enrichment of an inhibitor in the starch-rich fraction, then we have indirect evidence of a selective partitioning of phenolic compounds between the two fractions.
- Glycolipid emulsifier was present in all samples tested. No influence of oat variety nor of oat flour fractionation on emulsifier concentration or distribution respectively was observed.

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Sourcing added value food ingredients from home grown oats *'Innovation'*

Part 4

Food Applications

by

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Cereals Innovation Centre

Sensory Analysis Report

Title:	**INNOVATION - Final report**
Report to:	Martin Livermore, Charlie Forge, Leo Kelly, Magali Verdes, Sue Lee, Mark Fisher, Christine Walker, Matt Brooker (DuPont CIC) Julian South, Cathy Townsley (ADAS) Colin Webb, Grant Campbell, RuoHang Wang (Satake Centre, UMIST) Sandra Hill, David Gray, Rita Auerbach (Food Science, Nottingham University) Ian Smith, Mike Jee (Reading Scientific Services Ltd.) Paul Meakin (Home-Grown Cereals Authority)
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Project ref.:	Innovation (S002)
Ref.:	**CIC/11/98** *E:\DUPONT.DOC*
Date:	October 1998
Test dates:	Attribute generation: 14, 15, 16, 17 September Profiling on products: 21, 22, 23, 24, 28, 29, 30 September Rating: 5 and 6 October

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Summary

This project represents the DCIC input to a Home-Grown Cereals Authority sponsored project to evaluate new food uses for oats. It provides information on how oat flour selected from different varieties and agriculture practices compares to wheat and modified starch in terms of both functional and sensorial attributes in a particular application. The report also describes determining the right application, the correct process for the production of that application and understanding the oat flour behaviour.

An earlier study was carried out to determine the correct application for testing the oat flour fractions. The thickening functionality of the oat flour was tested in a béchamel sauce. This sauce was chosen as it has a bland taste, and therefore would not mask the characteristics of the oat flour. The sauce process was optimised during this study and the oat flour was found to give an acceptable béchamel sauce. This sauce was compared sensorially with a béchamel sauce prepared with a standard wheat flour (Viceroy), and one with a modified starch (Colflo). The results from this early study indicated that the sample prepared with the oat flour was very similar to the sample made with the wheat flour. The modified starch sample was perceived as being very different from these two samples, it had a stronger aroma and flavour, it was much thicker, lighter in colour and smoother in the mouth than the two other samples.

A second study was set up to investigate the effects variety and agricultural practices have on the thickening functionality of the oat flour. In this study, six samples of oats were selected and compared to sauces made with two common thickeners: standard wheat flour and modified starch. The technique of full sensory profiling was used to describe and quantify the differences and similarities between the eight sauces. However, due to the sample availability, the sauces were only able to be assessed when hot and the original plan of assessing them in the jar, as a consumer may do on purchasing, had to be dropped.

The results from the sensory profiling for this second study indicated that there was no effect from the different agricultural practices, but that there were differences, possibly due to the variety of the oat, in the appearance of the sauces and the amount the sauces set when left to go cold. The Gerald samples contained less dark specks than the other oat samples and were also lighter in colour. The naked oat sample, Kynon, became more set on cooling than most of the other oat samples except for Chamois, which in turn was more set than Image grown under standard crop husbandry.

Background and Objectives

In 1989, a Home-Grown Cereals Authority (HGCA) review of the UK oat crop concluded that there was a lack of research in the UK in comparison to that undertaken in the USA and Canada, on the value of oat fractions for industrial non-food as well as novel food manufacturing uses. In 1996, the HGCA funded a project called Oatec to assess the potential of the oat grain for use in non-food applications. In September 1997, the HGCA also funded the Innovation project, the aim of which is to unlock the potential of the oat grain as a source of added value food ingredients. The key to the project's success is the introduction of new technology for fractionating the oat into its constituent parts. The project objectives are to evaluate the milling and fractionation processes for extraction of components, to investigate the genotypic, husbandry and environmental effects on grain composition, to examine the effects of grain composition on milling and fractionation efficiency, to characterise the structure and properties of components and to assess the functionality of components for food uses. The participants in the project are the Agricultural Development Advisory Service (growth studies), Satake Centre University of Manchester Institute of Science and Technology (milling and extraction studies), Nottingham University (functionality studies) and Reading Scientific Services. Dalgety has been involved in the project since April 1997.

The original objective of the Dalgety (now DuPont) contribution to the project was to assess the sensory properties of the functional components of oat (a fraction rich in starch and a fraction rich in non starch polysaccharide), selected from different varieties, different environments and different agriculture practices, in pasteurised sauce applications and to compare the functionality of oat fractions to a standard wheat flour and a modified starch in this application.

To develop experience in the preparation of sauces using oat flour, an initial study was set up to test the behaviour of the oat flour in a sauce application and to determine the correct process for sauce making. A secondary aim of the first study was to compare the sensory characteristics of a sauce made with oat flour to sauces made with commonly used thickeners such as standard wheat flour and modified starch. The details and results of this study are given in Appendix 1.

The aim of the second study, which is the topic of this report, was to meet the original aim of the project i.e. to investigate the different functionality of oat cereal depending on their varieties and growing conditions and to compare sauces made with these oat flours to sauces made with common thickeners. The six samples of oat selected were: Image and Gerald varieties with two different levels for agriculture practices, Chamois variety interesting for its rheological properties and Kynon variety interesting as it is a naked oat. This design allowed us to compare four different varieties and two different agriculture practices. The samples chosen are all fractions rich in starch and may have different levels of ß-Glucan. The fractions rich in ß-Glucan were very coarse and showed a high grey/brown colour which would not have been suitable in a sauce application.

The project objective was accomplished by preparing eight béchamel sauce samples: six of the samples were made with the various oat flour fractions, one with wheat flour and one with modified starch and using full sensory profiling techniques to identify and quantify similarities and differences in appearance, aroma, flavour, texture and aftertaste of the eight samples.

Experimental Details

Sauce Preparation

Eight sauces were prepared for sensory assessment for this study. The sauces had the same formulation but one was made with a modified starch (Colflo 67), one with a standard wheat flour (Viceroy) and the other six samples with oat flour. The oats had been milled by UMIST, packed in polyethylene, flushed with nitrogen to minimise the oxidative effects following milling and sent to Cambridge by courier and heat-treated. Sample identifications are given in Table 1.

Table 1: Sample Identification

Sample Code	Identification
I1	Rosemaund nitrogen **Image** Nil irrigation, 0 AN, 0 FU
V2	Rosemaund variety **Chamois** (grown under standard crop husbandry)
V5	Rosemaund variety **Gerald** (grown under standard crop husbandry)
V7	Rosemaund variety **Image** (grown under standard crop husbandry)
G1	Rosemaund irrigation/nitrogen **Gerald** Nil irrigation, 0 AN, 0 FU
V10	Rosemaund variety **Kynon** (naked oat - grown under standard crop husbandry)
V	Viceroy (wheat flour)
C	Colflo (modified starch)

Key *AN - ammonium nitrate* *FU - foliar urea*

The sauces were produced by Magali Verdes, Marie Robelin, Mark Fisher and Matthew Brooker in batches of 1kg, at the DuPont Cereals Innovation Centre. The recipes were based on 4.5% of starch (on a dry weight basis) and are shown in Table 2.

The sauces were kept at ambient temperature between preparation and sensory assessment. They were then heated on gas (medium heat) until they reached 85°C and were kept at that temperature, or above, for three minutes. The temperature of the sauces was monitored and was approximately 60°C when the panellists assessed the mouth-feel and flavour. The hot sauces were presented in polystyrene cups and each panellist received a third of a cup.

Table 2: Sauce recipes

	I1	**V2**	**V5**	**V7**	**G1**	**V10**	**Viceroy**	**Colflo**
Ingredients	**g/kg**	**g/kg**	**g/kg**	**g/kg**	**g/kg**	**g/kg**	**g/kg**	**g/kg**
I1	61.8							
V2		62.2						
V5			61.0					
V7				61.5				
G1					61.4			
V10						61.8		
Viceroy							63.1	
Colflo								50.6
Satro	28	28	28	28	28	28	28	28
Salt	6.3	6.3	6.3	6.3	6.3	6.3	6.3	6.3
White pepper	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Butter	47.5	47.5	47.5	47.5	47.5	47.5	47.5	47.5
Double cream	57	57	57	57	57	57	57	57
Lactic acid	2.3	2.3	2.3	2.3	2.3	2.3	2.3	2.3
Shear time (minutes)	2:00	2:00	2:00	2:00	2:00	2:00	2:00	2:00

Sensory Techniques Used

The methodology used for assessment was a full profiling technique. Assessors were presented with each of the products in turn and asked to describe appearance, aroma, texture, flavour and aftertaste. After the individual vocabulary generation there followed panel discussion where a consensus vocabulary was devised for all the attributes. This vocabulary consisted of attributes and descriptors that all assessors understood and found most appropriate to describe the products. This technique involves introducing the panellists to references in order to calibrate the panel on the type of attribute they are assessing. A list of the references may be found in Appendix 2. A score sheet was then devised for the assessors to score each attribute for each product. This was transferred onto the sensory software, FIZZ, when finalised.

Attribute generation

Due to the small quantity of samples of oat flour available, attributes and the protocol for the assessment of sauces, both hot and cold, were generated during the pre-study using commercially available sauces, details of which may be found in Appendix 1. Further attributes were added, where necessary, during the assessment of the eight experimental samples.

Panellists' rating

For the rating each product was presented to the panellists in duplicate using 3-digit codes in a randomised order to avoid positional bias, carryover effects and learning effects. The order of presentation of the samples for the rating is attached in Appendix 3. All attributes were measured on a 10 cm line from "not" to "very", unless otherwise stated, and are listed in Table 3.

Table 3: Questionnaire

Category	Attribute name	Further explanation	Anchors of scale
Aroma	Strength	overall assessment	weak to strong
	Cheesy	Béchamel sauce with vinegar	not to very
	Milky/sweet	Béchamel sauce	not to very
	Roux	Warm butter and cooked flour	not to very
	Vegetable water	Potato and pea water	not to very
	Sour	Sour milk, regurgitated baby milk, citric acid and white sauce	not to very
	Lemon	White sauce and artificial lemon	not to very
	Stewed apples		not to very
	Waxy maize starch	Paste of waxy maize starch and water	not to very
Appearance	Thickness	Tilt the cup very slowly and assess	thin to thick
	Skin coverage		no skin to 100% skin
Texture	Wobbles	Tap cup lightly 2 or 3 times on side of cup	not to very
	Pourability	Tilt the cup in one direction until the sauce reaches 1 cm from cup ridge and assess	does not move to runny
	Coats cup	Return cup to upright and assess	not to very
	Dropping consistency	After stirring, take half a teaspoon of sauce onto the plate and assess while dropping from side of spoon (hold teaspoon at cup height)	not (pour, runny) to very (forms a blob)
	Coats spoon	Quantity of coating	not to very
	Holds its shape	Look at plate	not to very
	Shiny	Overall appearance	not to very
	Refills trough	Do a line with the tip of your spoon in the sample on the plate and assess whether the trough refills or not	not (does not move) to very (filled up quickly)
Flavour	Flavour strength	Overall assessment of flavour	bland to strong
	Tangy	Sensation of cheeks puckering	not to very
	Buttery		not to very
	Milky/creamy	Warm full fat milk	not to very
	Sour milk/sour cheese	Warm curd cheese	not to very
	Vinegar		not to very

Table 3: Questionnaire (cont.)

Category	Attribute name	Further explanation	Anchors of scale
	Starchy	Thickened with starch or corn flour	not to very
	Salty		not to very
	Peppery		not to very
	Sweet		not to very
	Waxy maize starch	Chemical note	not to very
	Lemon	Artificial lemon	not to very
Mouth feel	Slimy mouth feel	uncooked white boiled egg sensation, oyster, overcooked pasta, tapioca pudding	not to very
	Greasy mouth feel		not to very
	Smooth mouth feel		not to very
Measure after swallowing	Starchy mouth feel	floury coating, home-made sauce not cooked enough, sensation	not to very
	Coats mouth	clings to mouth/sticky	not to very
Aftertaste	Vinegary		not to very
	Acidic		not to very
	Lemon		not to very
	Cheesy/creamy		not to very
	Salty		not to very
	Pepper		not to very
	Leaves mouth dry	After swallowing	not to very
Appearance cold	Dark specks	Amount of	not to very
	Shade		light to dark
Texture when cold	Lumpy	overall impression on spoon, plate and in cup	not to very
	Newly formed skin when cold	using the back of the spoon, push the surface of the product and assess the newly formed skin	not to very
	Set when cold	whether the sauce is set when the plate is tilted or whether the sauce moves	not to very (does not move)

Constraints

The largest constraint of the study was the small quantity of oat flour available: this resulted in samples having to be prepared in 1kg batches at a lab scale which generated variability between batches. It also meant that the panellists were not able to assess the samples as many times as is normally necessary for profiling.

Statistical Methods Used

The mean scores for each attribute and each product can be found in Appendix 4. The analyses used included analysis of variance (ANOVA) and the Newman-Keuls test. The ANOVA divided the total variance into six components: assessor, sample, replicate, assessor x sample interaction, assessor x replicate interaction and residual. The first five components were then tested for significance against the residual term.

A significant result indicates the presence of real source effects over and above the base level of random variation. The results from the ANOVA analyses are shown in Appendix 5. To say a result is significant at the 5% level is to say with 95% certainty that there is a real difference between the products. This means that there is a chance of 5 in 100 (or 1 in 20) that there is no real difference between the products.

A significant interaction effect indicates that the relative rating of the samples differs between assessors. Some attributes indicated significant judge-product interaction, but this was mainly due to different rating of samples that were very similar for that attribute, as opposed to completely different sample orders for different panellists. In cases where the sample differences are considerably greater than the interaction effect the interaction effects may be overlooked. This was the case for the majority of the attributes in this profile that showed interactions of this nature. Some attributes, however, indicated that the panellists did not fully understand the protocol for assessment and these attributes have been ignored in the assessment of the results.

The Newman-Keuls test was employed to assess the significance of the differences between individual sample means using a standard error calculated from the residual mean square. It was used to identify groups of means that did not differ significantly at the 5% level. This test allows the statistical differentiation of samples in the groups as opposed to a simple mean comparison by eye.

Results

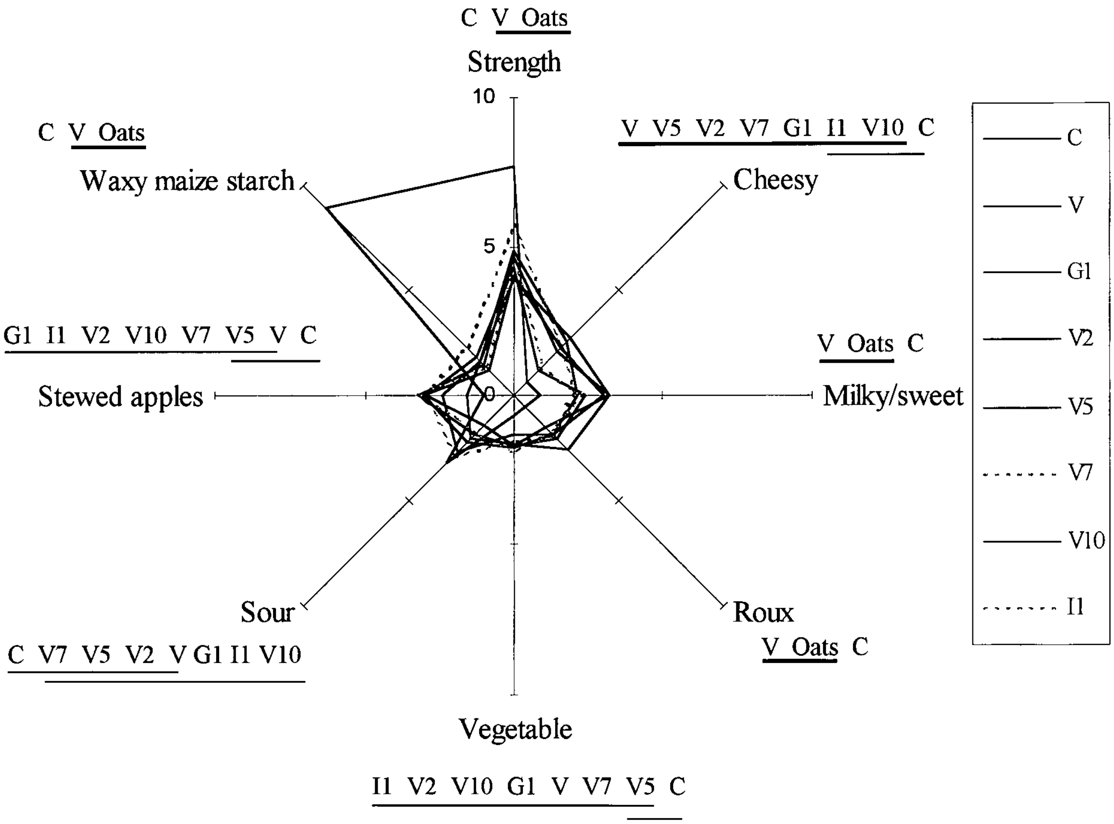
The panellists generated 48 attributes to describe the sauces. Thirty-two of these attributes generated a significant difference between the samples and five attributes were inconclusive, as the statistical analyses performed highlighted problems with the panellists' use of these to differentiate the samples. The panellists had a limited sample awareness of the actual samples due to the limitation of oat flour provided. This could explain the disagreement between the panellists for some of the attributes.

The profiles are shown in Figures 1 to 4. The results from the Newman-Keuls tests is shown in blue type next to the attribute title in order of decreasing means: like samples are underlined. For example (Figure 1) for strength of aroma: oat (samples prepared with oat flour) and sample V (prepared with Viceroy) are underlined indicating that they were not found to be statistically different during the Newman-Keuls analysis. These samples were found to be weaker in aroma than sample C (prepared with Colflo). In the discussion sections after each spider plot, similar samples, such as these, are grouped with an ampersand. There is a full discussion of the results at the end of this section. Samples prepared with Colflo, Viceroy and oat flour will be called "Colflo", "Viceroy" and "Oat" respectively in the results discussion.

There was a significant interaction for the attributes "shiny", "smooth", "starchy/floury", "lumpy" and "newly formed skin", indicating that the panellists were not using the attribute in the same way, therefore no conclusion can be drawn for these attributes.

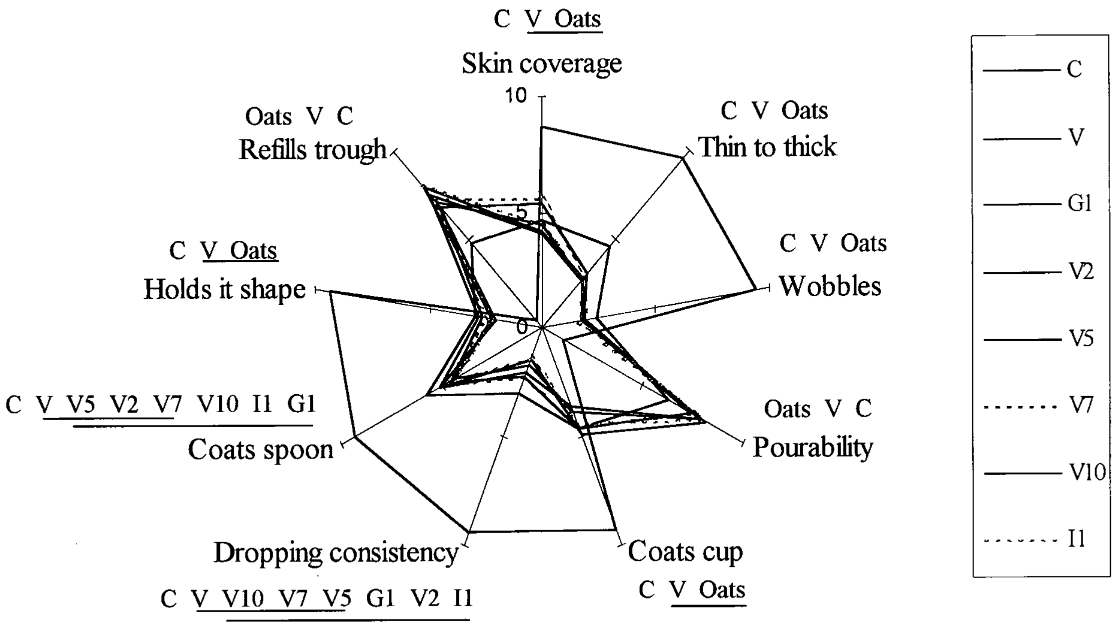
There was no significant differences between the samples for the attributes "lemon aroma", "vinegar flavour", "salty flavour", "peppery flavour", "lemon flavour" and all of the aftertaste attributes.

Figure 1: Aroma Profile



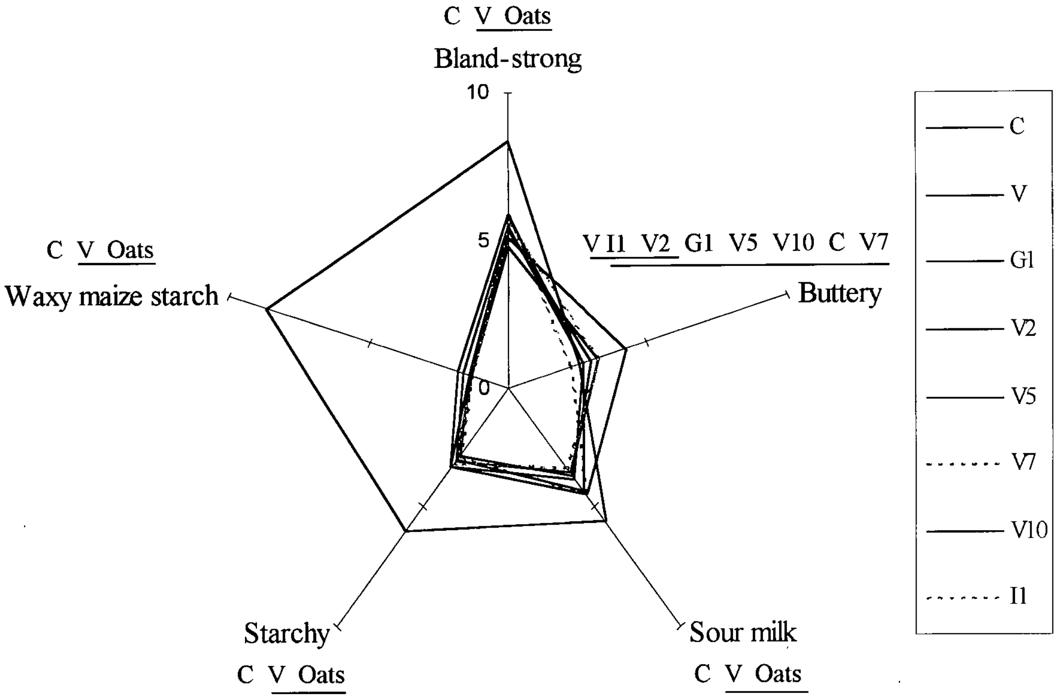
Attribute	Discussion
Strength	Colflo (C) had a stronger aroma than Viceroy (V) & the oat samples, which were not perceived as being different from each other.
Cheesy aroma	The oat samples I1 and V10 had a similar lower cheesy aroma to Colflo. The other oat samples had a similar cheesy aroma to Viceroy.
Milky/sweet	The Viceroy and oat samples had a stronger milky/sweet aroma than Colflo.
Roux	The Viceroy and oat samples had a stronger roux (warm butter and flour) aroma than Colflo.
Vegetable	The oat sample V5 had a similar vegetable aroma to Colflo: the other samples were all found to be similar to Viceroy and were more vegetable-like than Colflo.
Sour	The oat samples V7, V5, V2 & Viceroy were all similar to Colflo in terms of their sour aroma.
Stewed apples	Viceroy & the oat sample V5 had a similar stewed apple aroma to Colflo. The other oat samples were found to have more of this aroma than Colflo and be similar to the Viceroy sample.
Waxy maize starch	The Colflo sample had a stronger aroma of waxy maize starch than either the Viceroy sample or the oat samples, which were not found to be significantly different from each other.

Figure 2: Appearance/Texture Profile



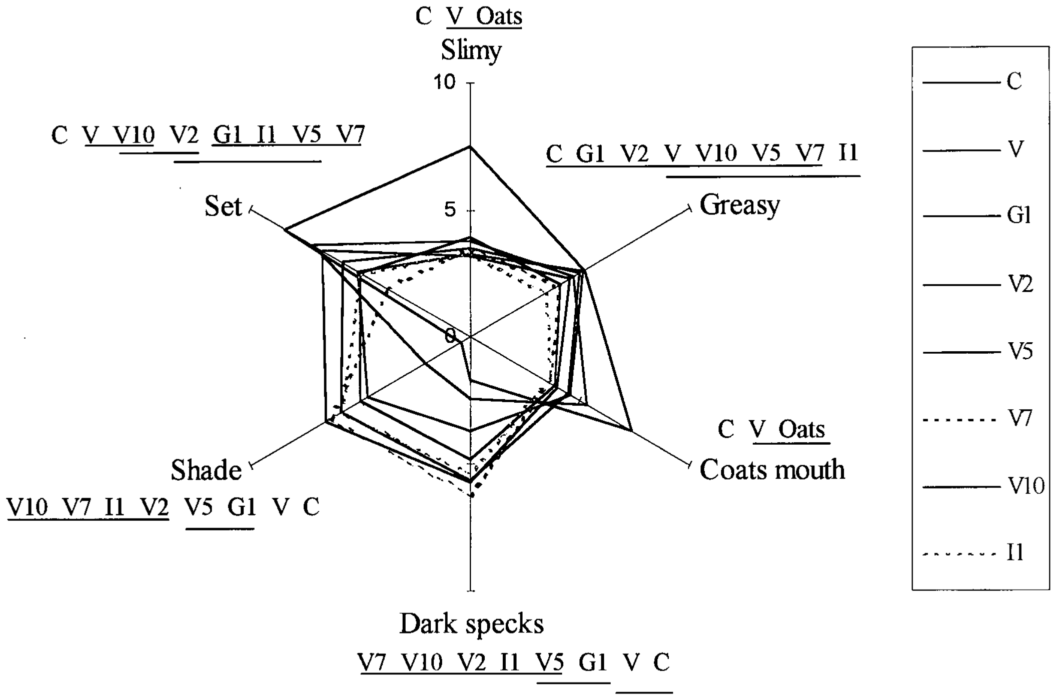
Attribute	Discussion
Skin Coverage	Colflo had more skin on top of the hot sauce in the cup than the Viceroy or oat samples, which were not found to be significantly different.
Thin to thick	Colflo was thicker than Viceroy which was thicker than the oat samples.
Wobbles	Colflo wobbled in the cup more when tapped (gelatinous) than Viceroy which in turn was slightly more wobbly than the oat samples.
Pourability	The oat samples were more runny in the cup than the Viceroy sample which was more runny than the Colflo sample.
Coats cup	The Colflo sample coated the side of the cup after tipping more than the Viceroy or oat samples, which were not found to be significantly different.
Dropping consistency	The Colflo sample formed a set blob on the plate when dropped. The oat samples V10, V7 & V5 were found to be similar to the Viceroy sample and more runny onto the plate than Colflo. The other oat samples were found to be more runny than Viceroy and Colflo.
Coats spoon	The Colflo sample left more quantity of sauce on the spoon than the other samples. The oat samples V5, V2 & V7 were similar to the Viceroy sample which coated the spoon more than the oat samples V10, I1 & G1.
Holds its shape	After dropping on the plate the Colflo sample held its shape more than either the Viceroy sample or the oat samples.
Refills trough	The Colfo sample did not refill the trough on the plate made with a spoon. The Viceroy sample did so slower than the oat samples which were not found to be significantly different.

Figure 3: Flavour Profile



Attribute	Discussion
Bland-strong	The Colflo sample had a stronger flavour than the Viceroy or oat samples.
Buttery	The Viceroy sample was more buttery than Colflo. The oat samples I1 & V2 were found to be similar to the Viceroy sample in butter flavour. The other oat samples were less buttery than the Viceroy sample.
Sour milk	The Colflo sample had more of a sour milk flavour than the other samples.
Starchy	The Colflo sample had a more starchy flavour than the other samples which were not found to be significantly different.
Waxy maize starch	The Colflo sample had the strongest waxy maize starch flavour: the other samples did not taste of waxy maize starch.

Figure 4: Mouth-feel and Appearance when cold



Attribute	Discussion
Slimy	Colflo (C) was perceived as being slimier than the other samples which were not found to be significantly different.
Greasy	The samples were all quite similar for this attribute with most of the oat samples being similar in greasiness to Colflo, except for I1 which was similar to Viceroy and slightly different from V2.
Coats mouth	The Colflo sample again stood out from the other samples and left more of a coating in the mouth after swallowing.
Dark specks	The oat samples had more dark specks than Viceroy & Colflo. Sample G1 had less dark specks than the other oat samples except for V5.
Shade	The Colflo and Viceroy samples were lighter than the oat samples, of which samples V5 & G1 were lighter than the other oat samples.
Set when cold	The Colflo sample became quite set on the plate when cold. The Viceroy sample was similar to the oat sample V10. The other oat samples, except for V2, were less set than V10 when left to go cold.

Results Discussion

The Colflo sample was very different from the other samples in this study. It was stronger in aroma, particularly in the waxy maize starch attribute, was thicker, less pourable and had a stronger flavour, again particularly for the waxy maize starch attribute. The Colflo sample was also perceived as being more slimy and greasy in the mouth and was also the lightest in colour and the most set when left to go cold. Table 4 below, indicates those attributes where similarities between the Colflo and the other samples were found. Many of the flavour attributes and all of the aftertaste attributes indicated no significant differences between any of the samples: these are also shown in Table 4.

Table 4: Similarities between Colflo and the experimental samples

Attribute	**Samples similar to Colflo**
Cheesy aroma	V10 and I1
Vegetable aroma	V5
Sour aroma	V7, V5, V2 and Viceroy
Stewed apple aroma	Viceroy and V5
Tangy, vinegar, salty, peppery, sweet and lemon flavours	No significant difference detected between any of the samples
Buttery flavour	All oat samples
Greasy mouthfeel	All samples except for I1
All aftertaste attributes	No significant difference detected between any of the samples
Dark specks	Viceroy

The oat samples were generally very similar to each other except for the three attributes "dark specks", "shade" and amount of setting when cold. Sample G1 had less dark specks than the other oat samples, except for V5, and samples G1 and V5 were lighter than the other oat samples. Sample V10 was more set when cold than the other oat samples, except for sample V2, which was more set than sample V7. In some cases certain oat samples were similar to the Viceroy or Colflo sample, but these attributes did not indicate actual differences between the samples themselves. For example, for the buttery flavour attribute, oat samples I1 and V2 are similar in butter flavour to the Viceroy sample, but are not more buttery than the other oat samples.

The oat samples were, for most attributes, similar to the sample made with wheat flour (Viceroy) except for those attributes shown in Table 5.

Table 5: Oat samples different to Viceroy

Attribute	**Oat samples different to Viceroy**
Thin to thick	All samples thinner than Viceroy
Wobbles	All samples wobbled less than Viceroy
Pourability	All samples more pourable than Viceroy
Dropping consistency	G1, V2 and I1 more runny than Viceroy
Coats spoon	V10, I1 and G1 coated spoon less than Viceroy
Refills trough	All samples refilled trough more than Viceroy
Buttery flavour	G1, V5, V10 and V7 all less buttery than Viceroy
Dark specks	All samples had more dark specks than Viceroy
Shade	All samples darker than Viceroy
Set when cold	All samples except V10, were less set when allowed to go cold than Viceroy.

The paragraph below gives examples of similarities between the oat samples and the standard products. Sample **V5** (Gerald grown under standard crop husbandry) was similar to Colflo in vegetable and stewed apple aromas and similar to Viceroy in the amount of sample left coating a spoon. Sample **I1** (Image treated with nitrogen) was similar to Colflo in terms of its cheesy aroma and was similar to Viceroy in its buttery flavour. Sample **V10** (Kynon, a naked oat) was similar to Viceroy in its dropping consistency (runniness) and the amount of setting when cold, which was more pronounced than the other oat samples except for V2. This sample was also similar to Colflo in its cheesy aroma. Samples **V2** (Chamois grown under standard crop husbandry) and **V7** (Image grown under standard crop husbandry) were both similar to Viceroy in the amount of sample left coating a spoon. Sample V2 was also similar to Viceroy in its level of buttery flavour. Sample **G1** (Gerald treated with nitrogen) had less dark specks than the other oat samples, except for the other Gerald sample, V5. These two samples (G1 and V5) were also lighter in colour than the other oat samples.

Conclusion

The oat samples were similar to the sample made with **wheat** flour for many of the attributes generated during this sensory study, except in that they were thinner, darker and some were less buttery. Sample V10 (Kynon, a naked oat) was also similar to the Viceroy in the amount of setting when allowed to go cold. The oat samples were all very different to the Colflo sample in texture and appearance but there were some similarities in aroma, flavour and aftertaste.

The six oat samples were all quite similar to each other, the only three differences being in amount of dark specks, lightness and amount of setting when left to go cold. Sample G1 (Gerald treated with nitrogen) had less dark specks than the other oat samples (except for V5) and samples G1 and V5 (Gerald grown under standard crop husbandry) were lighter in colour than the other oat samples. The results therefore indicate that there were some differences detected between the different varieties but mainly in appearance.

In terms of identifying trends in agriculture practices, there appears to be no link between the different growing methods as assessment of all the attributes indicates no specific differences between the grouped samples shown in Table 6.

Table 6: Groups of oat samples for comparison

Groups	**Description**
V7 and I1	Both Image varieties but sample I1 was treated with nitrogen (V7 was grown under standard crop husbandry)
V5 and G1	Both Gerald varieties but sample G1 was treated with nitrogen (V5 was grown under standard crop husbandry)
V7, V5 and V2	All grown under standard crop husbandry

Appendix 1: Preliminary Experiment Details

Cereals Innovation Centre

Sensory Analysis Report

Title:	***INNOVATION - PRE-STUDY***
Report to:	*Martin Livermore, Charlie Forge, Leo Kelly, Magali Verdes, Sue Lee, Mark Fisher, Christine Walker, Matt Brooker*
Report cc:	*Lauren Rogers, Lesley Matthews*
Report from:	*Marie Robelin*
Project ref.:	*Innovation*
Ref.:	***CIC/07/98*** *E:\DUPONT.DOC*
Date:	*28 September 1998*
Test dates:	*Attribute generation: 28, 29, 30 July and 4, 5, 6, 11, 12, 13 August* *Profiling on products: 18, 19, 25, 26 August and 2, 3, 8, 9, 10 September*

Appendix 1: Preliminary Experiment Details (cont.)

Summary

This project represents the DCIC input to a Home-Grown Cereals Authority sponsored project to evaluate new food uses for oats. It will provide information on how oat flour selected from different variety and agriculture practises, compares to wheat and modified starch in terms of both functional and sensorial attributes. The tasks included finding the right application, the right process and understanding the oat flour behaviour in our application. The thickening functionality of the ingredient oat flour was tested in béchamel sauce. This sauce was chosen as it has a bland taste, and therefore would not mask the characteristics of the oat flour. The sauce process was optimised and the oat flour gave an acceptable béchamel sauce. This was then compared sensorially with a béchamel sauce prepared with a standard wheat flour (Viceroy), and one with a modified starch (Colflo). The sensory panellists did a full conventional profiling including the appearance, aroma, flavour, texture and aftertaste of the products.

The sample prepared with the oat flour was very similar to the sample made with the wheat flour, they had similar aroma, flavour and mouth-feel but their appearance and texture varied slightly. The oat sample was darker and greyer in colour, it was slightly thinner and runnier than the wheat sample. The modified starch sample was perceived as being very different from these two samples, it had a stronger aroma and flavour; it was much thicker, lighter in colour and smoother in the mouth than the two other samples.

A larger study is now planned to investigate the effects variety and agricultural practises have on the thickening functionality of the ingredient oat flour. In this study, six samples of oats will be selected and compared to sauces made with common thickeners like standard wheat flour and modified starch.

Appendix 1: Preliminary Experiment Details (cont.)

Background and Objectives

This section was deleted as the information is contained in the main body of the report.

Experimental Details

Sauce making

Three sauces were prepared for sensory assessment for this pre-study. The sauces had the same formulation but one was made with a modified starch (Colflo 67), one with a standard wheat flour (Viceroy) and one with oat flour. The oat flour was milled from a variety of Gerald provided last year by ADAS for UMIST to develop the milling process. Because this oat flour was required for the development of Béchamel processing, the specificity of the material was not very important and the growing conditions are not known. The oat was then milled by UMIST. The flour was packed in polyethylene, flushed with nitrogen to minimise the oxidative effects following milling and it was sent to Cambridge by courier and heat-treated.

The sauces were produced by Magali Verdes and Sue Lee in batches of 1Kg, at Dupont CIC. The recipes were based on 4.5% of starch (on a dry weight basis) and are shown in Table 1.

Table 1: Sauce recipes

	Colflo 67 (C)	*Oat flour (O)*	*Viceroy (V)*
Moisture	*12.5%*	*9.8%*	*12.1%*
Ingredients			
"Starch source"	*50.6*	*61.8*	*63.1*
Satro	*28.0*	*28.0*	*28.0*
Salt	*6.3*	*6.3*	*6.3*
White pepper	*0.4*	*0.4*	*0.4*
Butter	*47.5*	*47.5*	*47.5*
Double cream	*57.0*	*57.0*	*57.0*
Lactic acid	*2.3*	*2.3*	*2.3*
Water	*807.9*	*796.7*	*795.4*
Shear *(Silverson, 6000 rpm)*	*2 minutes*	*2 minutes*	*2 minutes*

The sauces were kept at ambient temperature between preparation and sensory assessment. They were then heated on gas (medium heat) until they reached 85°C and were kept at that temperature or above for three minutes. The temperature of the sauces was monitored and was approximately 60°C when the panellists assessed the mouth-feel and flavour. The hot sauces were presented in polystyrene cups and each panellists received a third of a cup.

Appendix 1: Preliminary Experiment Details (cont.)

Sensory techniques Used

The methodology used for assessment was a full profiling technique. Assessors were presented with each of the products in turn and asked to describe appearance, aroma, texture, flavour and aftertaste. After the individual vocabulary generation there followed panel discussion where a consensus vocabulary was devised for all the attributes. This vocabulary consisted of attributes and descriptors that all assessors understood and found most appropriate to describe the products. This technique involves introducing the panellists to references in order to calibrate the panel on the type of attribute they are assessing. A score sheet was then devised for the assessors to score each attribute for each product. This was transferred onto the sensory software, FIZZ, when finalised.

Attribute generation

Due to the small quantity of samples of oat flour available, the panellists had to be trained first in assessing commercial sauces. The planning of the sessions and the products assessed can be found in Appendices 1, 2 and 3.

Panellists' rating

For the rating, each product was presented to the panellists in duplicate using 3-digit codes in a randomised order to avoid positional bias, carryover effects and learning effects. The order of presentation of the samples for the rating is attached in Appendix 4. All attributes were measured on a 10 cm line from "not" to "very", unless otherwise stated, and are listed in Table 2.

Table 2: Final questionnaire

Category	*Attribute name*	*Further explanation*	*Anchors of scale*
Aroma	*Strength*	*overall assessment -*	*weak to strong*
	Cheesy		*not to very*
	Melted butter		*not to very*
	Dairy/milky		*not to very*
	Starchy	*flour smell from cooked flour and fat together*	*not to very*
	Vegetable water	*cooked peas*	*not to very*
	Sour	*sour milk, regurgitated baby milk*	*not to very*
	Vinegar		*not to very*
	Lemon		*not to very*
	Rancid	*butter left out on a warm day*	*not to very*
	Sweet		*not to very*
	Waxy maize starch	*paste of waxy maize starch and water*	*not to very*
Appearance *tilting the cup very slowly*	*Greasy looking*		*not to very*
	Thickness		*thin to thick*
	Skin coverage	*no skin to total coverage*	*no skin to 100% skin*
	Fissures in skin	*presence of cracks in the skin*	*not to very*

Appendix 1: Preliminary Experiment Details (cont.)

Table 2: Final questionnaire (cont.)

Category	*Attribute name*	*Further explanation*	*Anchors of scale*
Texture	*Wobbles*		*not to very*
	Pourability	*whether the sauce is runny when tilting the cup*	*does not move to runny*
	Skin breaks up	*whether the skin breaks up when tilting the cup or not*	*not to very*
	Coats cup	*amount of sauce left on the side when cup returned to upright*	*not to very*
	Shiny	*overall appearance*	*not to very*
	Dropping consistency	*whether the sauce pours when dropped from spoon or forms blobs*	*not to very*
	Bungee effect	*retractability of sample after dropping from spoon*	*not to very*
	Greasy	*on the back of the spoon*	*not to very*
	Coats spoon		*not to very*
	Holds its shape		*not to very*
	Refills trough	*do a line with your spoon in the sample on the plate and assess whether the trough refills or not*	*not (does not move) to very (filled up quickly)*
	How fast it moves	*when plate tilted*	*slow to fast*
Flavour	*Flavour strength*	*overall assessment of flavour*	*bland to strong*
	Tangy/sharp		*not to very*
	Buttery		*not to very*
	Milky/creamy		*not to very*
	Sour milk	*sour cheese flavour*	*not to very*
	Rancid butter		*not to very*
	Vinegar		*not to very*
	Starchy	*corn flour*	*not to very*
	Salty		*not to very*
	Peppery		*not to very*
	Bitter		*not to very*
	Sweet		*not to very*
	Waxy maize starch		*not to very*
Mouth feel	*Slimy mouth feel*	*uncooked white boiled egg sensation, oyster, overcooked pasta*	*not to very*
	Greasy mouth feel		*not to very*
	Smooth mouth feel		*not to very*
	Starchy mouth feel	*floury coating, home-made sauce not cooked enough, sensation*	*not to very*
	Coats mouth	*clings to mouth/sticky*	*not to very*
	Leaves mouth dry	*after swallowing*	*not to very*

Appendix 1: Preliminary Experiment Details (cont.)

Table 2: Final questionnaire (cont.)

Category	*Attribute name*	*Further explanation*	*Anchors of scale*
Aftertaste	*Rancid aftertaste*		*not to very*
	Vinegary aftertaste	*acididc*	*not to very*
	Lemon aftertaste		*not to very*
	Cheesy/creamy		*not to very*
	Salty aftertaste		*not to very*
	Bitter aftertaste		*not to very*
	Dark specks		*not to very*
	Grey colour		*not to very*
	Green colour		*not to very*
	Yellow colour		*not to very*
	Darkness	*tone*	*light to dark*
	Lumpy	*overall impression on spoon, plate and in cup*	*not to very*
Texture when cold	*Set when cold*	*whether the sauce is set when the plate is tilted or whether the sauce moves*	*not to very (does not move)*
	Newly formed skin when cold	*using the back of the spoon, push the surface of the product and assess the newly formed skin*	*not to very*

Constraint

The biggest constraint of the study was the small quantity of oat flour available which made it more difficult for the panellists. The samples were prepared in 1Kg batches at a lab scale which generated variability between batches which made the rating more difficult for the panellists.

Statistical Methods Used

The mean scores for each attribute and each product can be found in Appendix 5. The analyses used included analysis of variance (ANOVA) and the Newman-Keuls test. The ANOVA divided the total variance into six components: assessor, sample, replicate, assessor x sample interaction, assessor x replicate interaction and residual. The first five components were then tested for significance against the residual term.

A significant result indicates the presence of real source effects over and above the base level of random variation. The results from the ANOVA analyses are shown in Appendix 6. To say a result is significant at the 5% level is to say with 95% certainty that there is a real difference between the products. This means that there is a chance of 5 in 100 (or 1 in 20) that there is no real difference between the products.

A significant interaction effect indicates that the relative rating of the samples differs between assessors. Some attributes indicated significant judge-product interaction, but this was mainly due to different rating of samples that were very similar for that attribute, as opposed to completely different sample orders for different panellists. In cases where the sample differences are considerably greater than the interaction effect the interaction effects may be overlooked. This was the case for the majority of the attributes in this profile that showed interactions of this nature. Some attributes, however, indicated that the panellists did not fully understand the protocol for assessment and these attributes have been ignored in the assessment of the results.

The Newman-Keuls test was employed to assess the significance of the differences between individual sample means using a standard error calculated from the residual mean square. It was used to identify groups of means that did not differ significantly at the 5% level. This test allows the statistical differentiation of samples in the groups as opposed to a simple mean comparison by eye.

Appendix 1: Preliminary Experiment Details (cont.)

Results

The panellists generated 61 attributes to describe the sauces. Only 30 of these attributes generated a significant difference between the samples and 22 attributes were inconclusive, as the statistical analyses performed highlighted problems with the panellists' use of these to differentiate the samples. The panellists had a limited sample awareness on the actual samples due to the limitation of oat flour provided. This could explain the high level of disagreement between the panellists and should improve in the large study.

The profiles are shown in Figures 1 to 4. The results from the Newman-Keuls tests is shown next to the attribute title in order of decreasing means: like samples are underlined. For example (Figure 1) for strength of aroma: samples O (prepared with oat flour) and sample V (prepared with Viceroy) are underlined indicating that they were not found to be statistically different during the Newman-Keuls analysis. These samples were found to be less strong in aroma than sample C (prepared with Colflo). In the discussion sections, similar samples, such as these, are grouped with an ampersand. There is a full discussion of the results at the end of this section. Samples prepared with Colflo, Viceroy and oat flour will be called respectively "Colflo", "Viceroy" and "Oat" in subsequent results' discussion.

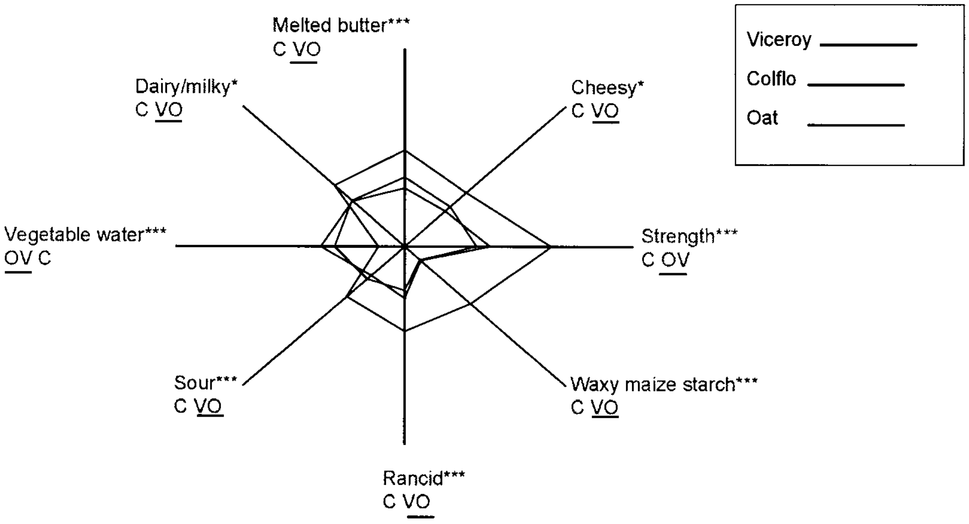
There was a significant interaction for the attributes "starchy, vinegar, lemon and sweet aroma", for the attributes "green colour, greasy looking, fissures in skin and bungee effect", for the attributes "strength of flavour, tangy, sour milk, rancid butter, starchy, peppery, bitter, and sweet flavour", for the attributes "starchy and greasy mouth feel" and for the attributes "rancid, vinegary, lemon, cheesy and bitter aftertaste". This means that the panellists were rating the samples in a different way, therefore no conclusion can be drawn for these attribute.

All the samples were perceived as similar for the attributes "breaks up skin, lumpy and newly formed skin, buttery flavour, milky flavour, vinegary flavour and leaves mouth dry".

Appendix 1: Preliminary Experiment Details (cont.)

Aroma discussion

Figure 1: Aroma of the three sauces

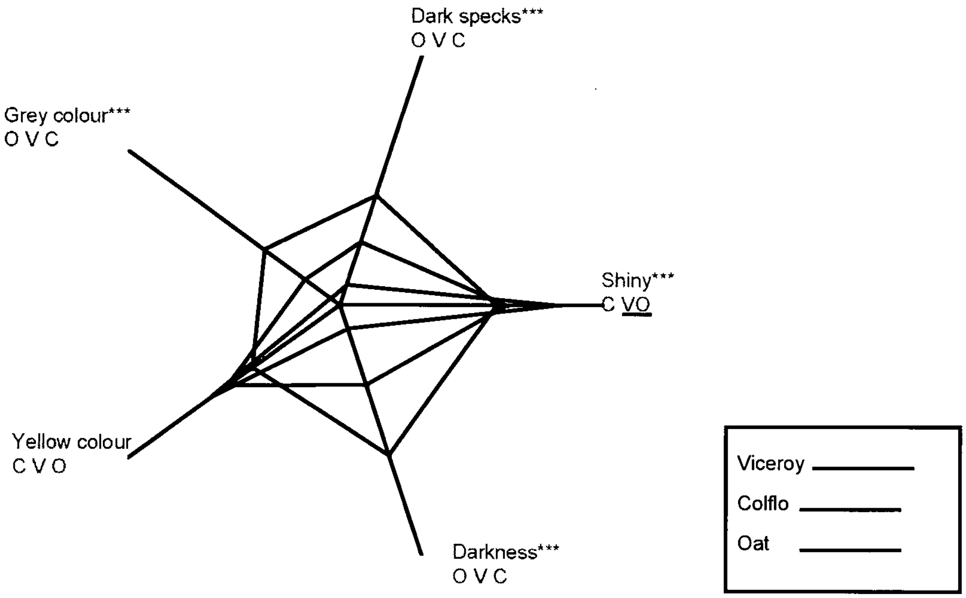


Attribute	*Discussion*
Strength	*Colflo (C) had a stronger aroma than Viceroy (V) and Oat (O) which were not perceived as being different from each other.*
Cheesy aroma	*C had a stronger cheesy aroma than O &V*
Melted butter	*C had a stronger butter aroma than O & V*
Dairy/milky	*C had a stronger dairy aroma than O & V*
Starchy	*C tended to have a stronger starchy aroma than O & V*
Vegetable water	*V & O had a stronger vegetable aroma than C*
Sour	*C tended to have a stronger sour/vinegary aroma than O & V*
Rancid	*C had a stronger rancid aroma than O & V*
Waxy maize starch	*C had a stronger waxy maize starch aroma than O & V*

Appendix 1: Preliminary Experiment Details (cont.)

Appearance discussion

Figure 2: Appearance of the three sauces



Attribute	*Discussion*
Shiny	*Colflo (C) was shinier than Viceroy (V) and the oat flour (O) which were not perceived as being different*
Dark specks	*O had more dark specks than V and V also had more than C*
Grey colour	*O was greyer than V which also was greyer than C*
Darkness	*O was the darkest sample in tone and sample C was the lightest*
Yellow	*C tended to be more yellow than V and O was the least yellow*

Appendix 1: Preliminary Experiment Details (cont.)

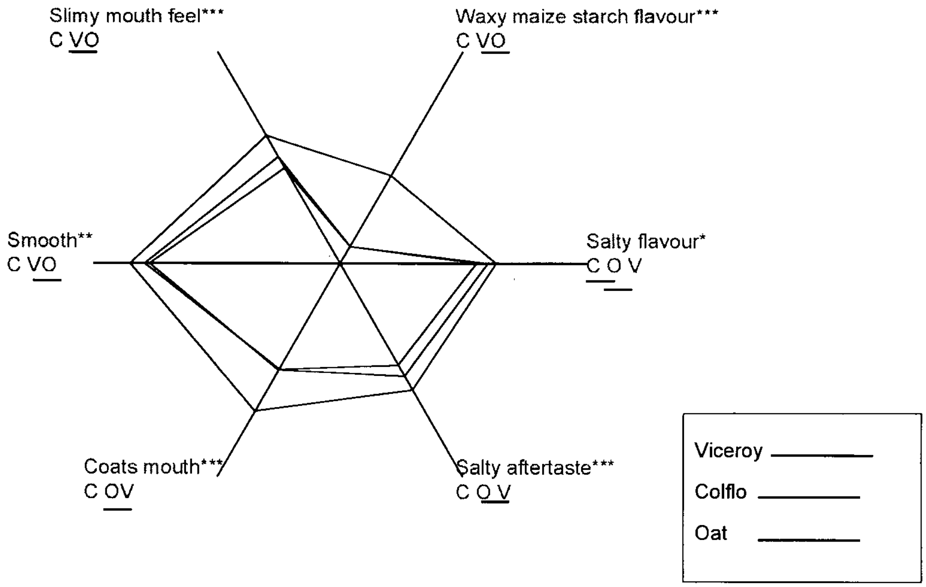
Texture discussion

Attribute	Discussion
Thickness	*Colflo (C) was the thickest sample by a long way (average of 9.20) whereas oat flour (O) was the thinnest (average of 1.14) but was not very different from Viceroy (V) which had an average score of 1.71*
Skin coverage	*C had more skin on the surface of the product than V & O*
Wobbles	*C wobbled more (was more gelatinous) when the cup was tapped than V & O*
Pourability	*When tilting the cup, C did not move as much as V; O was the runniest sample but again the difference between V and O was not really big (respective average of 8.22 against 8.82)*
Coats cup	*C coated the cup more than V & O*
Coats spoon	*In the same way, C coated the spoon the most. Sample O coated it less than sample V but the difference between these two samples was not very large*
Dropping consistency	*When dropped from the spoon to the plate, sample C dropped in blobs when sample V was runny and sample O even runnier*
Holds its shape	*C held its shape better than V & O*
Refills trough	*When drawing a line in the sauce on the plate, sample O refilled the trough quicker than V when sample C did not refill it at all. This attribute was not represented on the spider plot.*
How fast it moves	*O was the quickest to move when tilting the plate and sample C was the slowest sample*
Set sample when cold	*Once the samples had cooled down on the plate, sample C (average of 8.36) was more set than V (average of 6.14) which was slightly more set than sample O (average of 5.07)*

Appendix 1: Preliminary Experiment Details (cont.)

Flavour, mouth-feel and aftertaste

Figure 4: Flavour, mouth feel and aftertaste of the three sauces



Attribute	Discussion
Salty	*Colflo (C) was perceived as being saltier than Viceroy, the sample made with oat flour (O) being similar to C and V. In the aftertaste however, C was perceived as being saltier than O & V*
Waxy maize starch flavour	*Like in the aroma, sample C tasted more like a waxy maize starch than V & O*
Slimy mouth feel	*C had a slimier sensation in the mouth than V & O*
Smooth	*C was smoother in mouth than V & O but there was not much difference (average score of 8.6 for C, 8.0 for V and 7.8 for O)*
Coats mouth	*C coated the mouth more than O &V*

Conclusions

The oat flour Gerald gave an acceptable béchamel sauce which was not perceived as more rancid than the other sauces and the process of sauce making was optimised. The shear level of two minutes gave a good thickness to the béchamel sauce prepared with oat and this shear level will be used again in the large study. Sauces are a good application to promote the functionality of oat components.

The sample made with the oat flour was very similar sensorially to the sample made with a traditional flour Viceroy. The two samples had a similar aroma; they had similar flavour and mouth-feel and aftertaste but they were slightly different in appearance and texture. No off flavours or rancidity notes were detected in the oat flour sample. The béchamel sauce made with the oat flour was darker and greyer than the sample made with Viceroy. This was due to the colour of the flour itself. The sample prepared with the oat flour was slightly thinner, slightly more runny than Viceroy. It also coated the spoon less and was slightly less set when cold but the two samples Viceroy and oat held their shape in the same way on plate.

The sample made with the modified starch (Colflo) was very different when compared to wheat flour and oat four. It had a stronger aroma (more cheesy, more buttery, more dairy, more starchy and more sour), a stronger flavour (saltier, strong aroma of waxy maize starch).

The Colflo sample was much thicker, it formed more skin, coated the cup more, held its shape more and was more set when cold than the two other samples. It tended to be more yellow and less dark than the other samples. In the mouth, the Colflo béchamel was more slimy, smoother and coated the mouth more than the two other samples.

Future Work

The large study is already planned, its aim is to investigate the different functionality of six samples of oat cereal depending on their varieties and on their growing conditions (agriculture practices) in a béchamel sauce application. These sauces will be compared with common thickeners. The six samples of oat selected were: variety Image with two different levels for agriculture practices, variety Gerald with two different levels for agriculture practices, variety Chamois interesting for its rheological properties and variety Kynon interesting as it is a naked oat. This design will allow us to compare four different varieties and two different agriculture practices. The samples chosen are all fractions rich in starch and might have different levels of ß-Glucan. We decided not to work on fractions rich in ß-Glucan as this fraction was very coarse and showed a high grey/brown colour which would not have been suitable in a sauce application. Moreover, the fractions rich in starch showed lots of different textural properties, interesting sensorially.

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 1: Panel Organisation

Week Commencing	*Tuesday*	*Wednesday*	*Thursday*
27/7/98 *10-12 each day*	*Generation of attributes on pasteurised sauces:* *Commercial products* *COLD*	*Generation of attributes on pasteurised sauces:* *Commercial products* *COLD*	*Generation of attributes on pasteurised sauces:* *Commercial products* *COLD*
3/8/98 *10-12 each day*	*Generation of attributes on pasteurised sauces:* *Commercial products* *COLD*	*Generation of attributes on pasteurised sauces:* *Commercial products* *HOT*	*Generation of attributes on pasteurised sauces:* *Commercial products* *HOT*
10/8/98 *10-12 each day*	*Generation of attributes* *Wheat flour/ Colflo/Gem* *white wine and mushroom* *HOT*	*Generation of attributes on pasteurised sauces:* *Wheat flour/ Colflo/Gem* *HOT*	*Generation of attributes on pasteurised sauces:* *Wheat flour/ Colflo/Gem* *COLD*
17/8/98 *10-12 each day*	*Generation of attributes* *Oat flour/ Colflo / Viceroy* *Béchamel* *HOT*	*Generation of attributes* *Oat flour/ Colflo / Viceroy* *Béchamel* *HOT*	
24/8/98 *10-12 each day*	*Generation of attributes* *Oat flour/ Colflo / Viceroy* *Béchamel* *HOT*	*Generation of attributes* *Oat flour/ Colflo / Viceroy* *Béchamel* *HOT*	
31/8/98 *10-12 each day*		*Generation of attributes* *Oat flour/ Colflo / Viceroy* *Béchamel* ***HOT Rating 1***	*Generation of attributes* *Oat flour/ Colflo / Viceroy* *Béchamel* ***HOT Rating 2***
7/9/98 *10-12 each day*	*Rating of sample:* *Oat flour/ Colflo / Viceroy* *Béchamel* *COLD*	*Rating of sample:* *Oat flour/ Colflo / Viceroy* *Béchamel* *COLD*	*Rating of sample:* *Oat flour/ Colflo / Viceroy* *Béchamel* ***COLD: Rating 1 and 2***

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 2: Panel training on commercial sauces

These sauces were assessed by the panel (July, August 1998) to generate a "flexible" questionnaire for all sauces.

1- Sainsbury's white wine and mushroom sauce (in a tin)
Ingredients - water, white wine, wheat flour, mushrooms, onion, modified starch, vegetable oil, white wine vinegar, chicken stock, garlic, salt, sugar, flavour enhancer: mono-sodium glutamate, lactose, white pepper, caseinates, parsley, citric acid, emulsifier, di-Potassium Hydrogen Orthophosphate.

2- Chicken Tonight, Creamy mushroom sauce (Jar)
Water, mushrooms, sunflower oil, onion, cream, salt, modified starch, fructose, egg yolk, sugar, stabiliser (xantham gum), flavourings, lactic acid, parsley, potassium chloride, garlic powder, colour (sulphite ammonia caramel), white pepper

3- Sainsburys Saucery, Creamy Mushroom sauce (Jar)
Water, mushrooms, vegetable oil, onions, sugar, salt, skimmed milk powder, modified corn starch, egg yolk powder, double cream, stabiliser: xantham gum, lactic acid, flavourings, onion powder, parsley, roasted barley, malt extract, granulated garlic, colour: Beta-carotene

4- Homepride, White Wine & Cream sauce (Tin)
Water, onions, white wine, wheat flour, rapeseed oil, modified starch, sherry, salt, flavourings, double cream, milk, proteins, hydrogenated vegetable oil, skimmed milk powder, sugar, egg yolk powder, chicken bouillon, lactose, herbs, spices, colour: beta carotene.

5 Sainsburys Fungaiola sauce (Jar)
Water, mushrooms, vegetable oil, full fat processed cheese, hydrogenated vegetable fat, modified corn starch, lactose, salt, sugar, stabilisers: xanthan gum, chicken bouillon (with flavour enhancer: monosodium glutamate, sodium guanylate, sodium inosinate, flavouring) milk proteins, dried onion flavouring, dried parsley, white pepper.

6 Dolmio, Cheese Oven Bake Sauce (Jar)
Water, cream, cheeses (cheddar, gouda, regato), modified starch, white wine, salt, emulsifiers: polyphosphates & orthophosphates, flavourings, white pepper.

7 Ragu, White sauce for lasagne.
Water, sunflower oil, modified starch, egg yolk, fructose, sugar, salt,flavour enhancers (mono-sodium glutamate, sodium 5'-ribonucleotide), stabiliser (xanthan gum), onion powder, potassium chloride, phosphoric acid, spice extracts, flavouring and colour riboflavin, beta-carotene.

Appendix 3: Panel training - Samples assessed

Tuesday 28-7-98

1. *Sainsbury's White Wine & Mushroom Sauce (tin).*
2. *Chicken Tonight, Creamy Mushroom Sauce (jar).*
3. *Sainsbury's Saucery, Creamy Mushroom Sauce (jar).*

All products served cold in a polystyrene cup.

Wednesday 29-7-98

4. *Homepride White Wine & Cream Sauce (tin)*

1. *Sainsbury's White Wine & Mushroom Sauce (tin)*

All products served cold in a polystyrene cup

Thursday 30-7-98

5. *Sainsbury's Classic Italian Fungaiola Sauce with mushrooms (jar)*

Product served cold in a polystyrene cup.

Tuesday 4-8-98

6. *Dolmio - Cheese Oven Bake Sauce (jar).*
7. *Ragu, White Sauce for Lasagne.*

All products served cold in a polystyrene cup.

Wednesday 5-8-98

1 Sainsbury's White Wine & Mushroom Sauce (tin).

2 Chicken Tonight, Creamy Mushroom Sauce (jar)

Both products served hot in a polystyrene cup.

Thursday 6-8-98

3 Sainsbury's Saucery, Creamy Mushroom Sauce (jar).

4 Homepride White Wine & Cream Sauce (tin).

All products served hot in a polystyrene cup.

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 4: Sample presentation order

NJ	*C1*	*C2*	*C3*	*R1*	*R2*	*R3*	
1	*571*	*672*	*343*	*Colflo*	*Viceroy*	*Oat*	Replicate 1
2	*571*	*672*	*343*	*Colflo*	*Viceroy*	*Oat*	
3	*571*	*672*	*343*	*Colflo*	*Viceroy*	*Oat*	
4	*571*	*672*	*343*	*Colflo*	*Viceroy*	*Oat*	
5	*571*	*672*	*343*	*Colflo*	*Viceroy*	*Oat*	
6	*343*	*571*	*672*	*Oat*	*Colflo*	*Viceroy*	
7	*343*	*571*	*672*	*Oat*	*Colflo*	*Viceroy*	
8	*343*	*571*	*672*	*Oat*	*Colflo*	*Viceroy*	
9	*343*	*571*	*672*	*Oat*	*Colflo*	*Viceroy*	
32	*786*	*014*	*115*	*Oat*	*Colflo*	*Viceroy*	Replicate 2
33	*786*	*014*	*115*	*Oat*	*Colflo*	*Viceroy*	
34	*786*	*014*	*115*	*Oat*	*Colflo*	*Viceroy*	
35	*786*	*014*	*115*	*Oat*	*Colflo*	*Viceroy*	
36	*786*	*014*	*115*	*Oat*	*Colflo*	*Viceroy*	
37	*115*	*786*	*014*	*Viceroy*	*Oat*	*Colflo*	
38	*115*	*786*	*014*	*Viceroy*	*Oat*	*Colflo*	
39	*115*	*786*	*014*	*Viceroy*	*Oat*	*Colflo*	
40	*115*	*786*	*014*	*Viceroy*	*Oat*	*Colflo*	
63	*982*	*195*	*818*	*Viceroy*	*Oat*	*Colflo*	Replicate 3
64	*982*	*195*	*818*	*Viceroy*	*Oat*	*Colflo*	
65	*982*	*195*	*818*	*Viceroy*	*Oat*	*Colflo*	
66	*982*	*195*	*818*	*Viceroy*	*Oat*	*Colflo*	
67	*982*	*195*	*818*	*Viceroy*	*Oat*	*Colflo*	
68	*982*	*195*	*818*	*Viceroy*	*Oat*	*Colflo*	
69	*818*	*982*	*195*	*Colflo*	*Viceroy*	*Oat*	
70	*818*	*982*	*195*	*Colflo*	*Viceroy*	*Oat*	
71	*818*	*982*	*195*	*Colflo*	*Viceroy*	*Oat*	

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 5: Mean scores for each attribute

Attribute	*Viceroy*	*Coflo*	*Oat*
Aroma strength	*3.17*	*6.47*	*3.75*
Cheesy aroma	*2.81*	*3.82*	*2.52*
Melted butter aroma	*3.48*	*4.84*	*2.94*
Dairy/milky aroma	*3.27*	*4.33*	*3.21*
Starchy aroma	*1.23*	*4.45*	*1.35*
Vegetable water	*3.06*	*1.16*	*3.67*
Sour aroma	*2.34*	*3.61*	*2.06*
Vinegar aroma	*2.59*	*3.85*	*2.35*
Lemon aroma	*0.63*	*2.00*	*0.46*
Rancid aroma	*2.24*	*4.31*	*2.60*
Sweet aroma	*1.23*	*2.09*	*1.54*
Waxy maize starch	*0.97*	*4.07*	*1.01*
Greasy looking	*5.24*	*5.94*	*5.50*
Thickness	*1.71*	*9.20*	*1.14*
Skin coverage	*3.99*	*7.11*	*3.91*
Fissures in skin	*2.65*	*1.20*	*2.98*
Wobbles	*0.99*	*7.75*	*0.60*
Pourability	*8.22*	*3.13*	*8.82*
Skin breaks up	*3.14*	*1.91*	*3.13*
Coats cup	*2.80*	*8.08*	*2.59*
Shiny	*6.37*	*8.33*	*5.99*
Dropping consistency	*1.09*	*8.92*	*0.57*
Bungee effect	*3.94*	*3.58*	*3.71*
Greasy	*5.49*	*6.44*	*5.68*
Coats spoon	*5.19*	*8.31*	*4.27*
Holds its shape	*1.84*	*8.57*	*1.40*
Refills trough	*7.55*	*0.63*	*9.06*
How fast it moves	*7.09*	*3.13*	*8.71*
Flavour strength	*6.16*	*7.41*	*6.01*
Tangy/sharp flavour	*6.83*	*7.99*	*6.61*
Buttery flavour	*4.95*	*5.69*	*5.07*
Milky/creamy	*4.21*	*4.38*	*4.18*
Sour milk flavour	*4.61*	*5.76*	*4.61*
Rancid butter	*4.03*	*5.39*	*4.35*
Vinegar flavour	*5.34*	*6.00*	*5.58*
Starchy flavour	*2.79*	*4.50*	*2.49*
Salty flavour	*5.54*	*6.38*	*6.02*
Peppery flavour	*2.61*	*1.99*	*3.09*
Bitter flavour	*1.26*	*2.22*	*1.61*
Sweet flavour	*1.18*	*1.72*	*1.14*
Waxy maize starch	*0.81*	*4.15*	*0.79*
Slimy mouth feel	*5.05*	*6.01*	*4.52*
Greasy mouth feel	*5.91*	*6.24*	*6.14*
Smooth mouth feel	*7.96*	*8.55*	*7.77*
Starchy mouth feel	*3.22*	*4.54*	*2.76*
Coats mouth	*4.94*	*6.93*	*5.00*
Leaves mouth dry	*5.70*	*6.36*	*6.23*
Rancid aftertaste	*3.18*	*4.88*	*3.58*
Vinegary aftertaste	*4.81*	*5.89*	*4.97*
Lemon aftertaste	*0.86*	*2.11*	*0.53*

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 5: Mean scores for each attribute (cont.)

Attribute	*Viceroy*	*Colflo*	*Oat*
Cheesy/creamy	*3.11*	*4.49*	*3.09*
Salty aftertaste	*4.76*	*5.93*	*5.28*
Bitter aftertaste	*1.43*	*2.34*	*1.41*
Dark specks	*2.54*	*0.81*	*4.37*
Grey colour	*1.65*	*0.44*	*3.57*
Green colour	*1.17*	*1.04*	*1.71*
Yellow colour	*5.29*	*6.09*	*4.14*
Darkness	*3.22*	*0.96*	*6.06*
Lumpy	*2.18*	*1.84*	*2.54*
Set when cold	*6.14*	*8.36*	*5.07*
Newly formed skin	*1.26*	*1.20*	*1.20*

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 6: ANOVA results showing source of variation and its significance

AROMA

Attribute	*Source of variation*	*F-Ratio*	*Significance*
Strength	*Product (P)*	*32.46*	***
	Judge (J)	*1.52*	
	Replicate (R)	*0.03*	
	*P*J interaction*	*3.59*	***
	*J*R interaction*	*0.42*	
Cheesy	*Product (P)*	*4.77*	*
	Judge (J)	*7.27*	***
	Replicate (R)	*0.90*	
	*P*J interaction*	*1.64*	
	*J*R interaction*	*0.47*	
Melted butter	*Product (P)*	*10.03*	***
	Judge (J)	*4.17*	**
	Replicate (R)	*0.29*	
	*P*J interaction*	*1.26*	
	*J*R interaction*	*0.75*	
Dairy/milky	*Product (P)*	*4.22*	*
	Judge (J)	*9.27*	***
	Replicate (R)	*1.56*	
	*P*J interaction*	*1.81*	
	*J*R interaction*	*0.50*	
Starchy	*Product (P)*	*115.01*	***
	Judge (J)	*19.01*	***
	Replicate (R)	*5.10*	*
	*P*J interaction*	*14.81*	***
	*J*R interaction*	*2.55*	**
Vegetable water	*Product (P)*	*232.43*	***
	Judge (J)	*10.45*	***
	Replicate (R)	*0.99*	
	*P*J interaction*	*5.66*	***
	*J*R interaction*		
Sour	*Product (P)*	*15.52*	***
	Judge (J)	*25.26*	***
	Replicate (R)	*0.28*	
	*P*J interaction*	*4.41*	***
	*J*R interaction*	*1.73*	
Vinegar	*Product (P)*	*13.14*	***
	Judge (J)	*23.05*	***
	Replicate (R)	*1.97*	
	*P*J interaction*	*4.33*	***
	*J*R interaction*	*1.04*	
Lemon	*Product (P)*	*73.36*	***
	Judge (J)	*57.88*	***
	Replicate (R)	*0.91*	
	*P*J interaction*	*31.54*	***
	*J*R interaction*	*0.5*	
Rancid	*Product (P)*	*32.55*	***
	Judge (J)	*32.09*	***
	Replicate (R)	*0.32*	
	*P*J interaction*	*7.07*	***
	*J*R interaction*	*1.58*	
Sweet	*Product (P)*	*19.87*	***
	Judge (J)	*98.03*	***
	Replicate (R)	*1.01*	
	*P*J interaction*	*9.87*	***
	*J*R interaction*	*1.66*	

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 6: ANOVA results showing source of variation and its significance (cont.)

APPEARANCE

Attribute	*Source of variation*	*F-Ratio*	*Significance*
Waxy maize starch	*Product (P)*	*113.24*	***
	Judge (J)	*14.83*	***
	Replicate (R)	*0.47*	
	*P*J interaction*	*24.93*	***
	*J*R interaction*	*0.69*	
Greasy looking	*Product (P)*	*1.86*	
	Judge (J)	*26.64*	***
	Replicate (R)	*0.27*	
	*P*J interaction*	*5.66*	***
	*J*R interaction*	*1.48*	
Thickness	*Product (P)*	*586.66*	***
	Judge (J)	*0.49*	
	Replicate (R)	*1.08*	
	*P*J interaction*	*2.00*	*
	*J*R interaction*	*0.34*	
Skin coverage	*Product (P)*	*34.07*	***
	Judge (J)	*17.09*	***
	Replicate (R)	*3.01*	
	*P*J interaction*	*2.75*	***
	*J*R interaction*	*0.70*	
Fissures in skin	*Product (P)*	*6.73*	**
	Judge (J)	*9.52*	***
	Replicate (R)	*1.07*	
	*P*J interaction*	*3.33*	**
	*J*R interaction*	*1.49*	
Wobbles	*Product (P)*	*392.72*	***
	Judge (J)	*12.26*	***
	Replicate (R)	*0.26*	
	*P*J interaction*	*5.58*	***
	*J*R interaction*	*0.78*	
Pourability	*Product (P)*	*408.34*	***
	Judge (J)	*10.90*	***
	Replicate (R)	*1.16*	
	*P*J interaction*	*9.07*	***
	*J*R interaction*	*0.46*	
Skin breaks up	*Product (P)*	*2.55*	
	Judge (J)	*10.94*	***
	Replicate (R)	*0.59*	
	*P*J interaction*	*0.83*	
	*J*R interaction*	*1.54*	
Coats cup	*Product (P)*	*233.52*	***
	Judge (J)	*11.67*	***
	Replicate (R)	*1.23*	
	*P*J interaction*	*3.75*	***
	*J*R interaction*	*1.60*	
Shiny	*Product (P)*	*55.15*	***
	Judge (J)	*48.55*	***
	Replicate (R)	*2.00*	
	*P*J interaction*	*11.27*	***
	*J*R interaction*	*3.39*	**
Dropping consistency	*Product (P)*	*1814.86*	***
	Judge (J)	*1.74*	
	Replicate (R)	*3.98*	*
	*P*J interaction*	*3.33*	**
	*J*R interaction*	*1.48*	

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 6: ANOVA results showing source of variation and its significance (cont.)

FLAVOUR

Attribute	*Source of variation*	*F-Ratio*	*Significance*
Bungee effect	*Product (P)*	*0.37*	
	Judge (J)	*24.89*	***
	Replicate (R)	*1.54*	
	*P*J interaction*	*3.79*	***
	*J*R interaction*	*1.12*	
Greasy	*Product (P)*	*4.41*	*
	Judge (J)	*29.42*	***
	Replicate (R)	*1.25*	
	*P*J interaction*	*4.78*	***
	*J*R interaction*	*2.59*	**
Coats spoon	*Product (P)*	*69.00*	***
	Judge (J)	*7.25*	***
	Replicate (R)	*3.12*	
	*P*J interaction*	*2.71*	**
	*J*R interaction*	*1.09*	
Holds its shape	*Product (P)*	*329.79*	***
	Judge (J)	*5.56*	***
	Replicate (R)	*0.03*	
	*P*J interaction*	*2.41*	*
	*J*R interaction*	*0.46*	
Refills trough	*Product (P)*	*353.16*	***
	Judge (J)	*0.54*	
	Replicate (R)	*0.66*	
	*P*J interaction*	*1.05*	
	*J*R interaction*	*0.73*	
How fast it moves	*Product (P)*	*127.83*	***
	Judge (J)	*6.09*	***
	Replicate (R)	*2.71*	
	*P*J interaction*	*4.16*	***
	*J*R interaction*	*1.93*	
Strength	*Product (P)*	*9.03*	***
	Judge (J)	*8.21*	***
	Replicate (R)	*0.01*	
	*P*J interaction*	*5.52*	***
	*J*R interaction*	*0.55*	
Tangy/sharp	*Product (P)*	*13.60*	***
	Judge (J)	*9.99*	***
	Replicate (R)	*0.57*	
	*P*J interaction*	*5.57*	***
	*J*R interaction*	*0.81*	
Buttery	*Product (P)*	*1.96*	
	Judge (J)	*12.42*	***
	Replicate (R)	*0.33*	
	*P*J interaction*	*3.03*	**
	*J*R interaction*	*0.56*	
Milky/creamy	*Product (P)*	*0.09*	
	Judge (J)	*10.34*	***
	Replicate (R)	*0.50*	
	*P*J interaction*	*0.74*	
	*J*R interaction*	*0.61*	
Sour milk	*Product (P)*	*9.41*	***
	Judge (J)	*28.76*	***
	Replicate (R)	*0.48*	
	*P*J interaction*	*7.49*	***
	*J*R interaction*	*1.19*	

Appendix 6: ANOVA results showing source of variation and its significance (cont.)

Attribute	*Source of variation*	*F-Ratio*	*Significance*
Rancid butter	*Product (P)*	*7.87*	**
	Judge (J)	*26.66*	***
	Replicate (R)	*1.32*	
	*P*J interaction*	*5.17*	***
	*J*R interaction*	*0.75*	
Vinegar	*Product (P)*	*2.02*	
	Judge (J)	*39.39*	***
	Replicate (R)	*0.03*	
	*P*J interaction*	*3.76*	***
	*J*R interaction*	*1.10*	
Starchy	*Product (P)*	*7.61*	**
	Judge (J)	*7.58*	***
	Replicate (R)	*0.60*	
	*P*J interaction*	*2.46*	*
	*J*R interaction*	*0.61*	
Salty	*Product (P)*	*3.73*	*
	Judge (J)	*67.28*	***
	Replicate (R)	*0.06*	
	*P*J interaction*	*1.64*	
	*J*R interaction*	*0.20*	
Peppery	*Product (P)*	*8.06*	**
	Judge (J)	*40.98*	***
	Replicate (R)	*11.16*	***
	*P*J interaction*	*4.88*	***
	*J*R interaction*	*11.77*	***
Bitter	*Product (P)*	*13.47*	***
	Judge (J)	*64.92*	***
	Replicate (R)	*0.82*	
	*P*J interaction*	*14.99*	***
	*J*R interaction*	*1.25*	
Sweet	*Product (P)*	*7.82*	**
	Judge (J)	*46.94*	***
	Replicate (R)	*0.93*	
	*P*J interaction*	*3.61*	***
	*J*R interaction*	*0.40*	
Waxy maize starch	*Product (P)*	*152.92*	***
	Judge (J)	*16.49*	***
	Replicate (R)	*0.18*	
	*P*J interaction*	*26.91*	***
	*J*R interaction*	*1.34*	
Slimy	*Product (P)*	*9.12*	***
	Judge (J)	*46.45*	***
	Replicate (R)	*1.41*	
	*P*J interaction*	*1.84*	
	*J*R interaction*	*0.42*	
Greasy	*Product (P)*	*0.62*	
	Judge (J)	*45.07*	***
	Replicate (R)	*3.89*	*
	*P*J interaction*	*2.02*	*
	*J*R interaction*	*0.51*	
Smooth	*Product (P)*	*7.23*	**
	Judge (J)	*45.88*	***
	Replicate (R)	*0.09*	
	*P*J interaction*	*1.80*	
	*J*R interaction*	*0.82*	

MOUTH FEEL

Appendix 1: Preliminary Experiment Details (cont.)

Appendix 6: ANOVA results showing source of variation and its significance (cont.)

	Attribute	*Source of variation*	*F-Ratio*	*Significance*
	Starchy	*Product (P)*	*10.60*	***
		Judge (J)	*24.97*	***
		Replicate (R)	*0.97*	
		*P*J interaction*	*3.39*	**
		*J*R interaction*	*2.60*	**
	Coats mouth	*Product (P)*	*22.86*	***
		Judge (J)	*30.51*	***
		Replicate (R)	*0.23*	
		*P*J interaction*	*1.19*	
		*J*R interaction*	*1.07*	
	Leaves mouth dry	*Product (P)*	*1.04*	
		Judge (J)	*17.11*	***
		Replicate (R)	*0.12*	
		*P*J interaction*	*0.89*	
		*J*R interaction*	*0.25*	
AFTERTASTE	*Rancid*	*Product (P)*	*20.06*	***
		Judge (J)	*53.64*	***
		Replicate (R)	*0.18*	
		*P*J interaction*	*7.60*	***
		*J*R interaction*	*0.67*	
	Vinegary	*Product (P)*	*4.82*	*
		Judge (J)	*29.49*	***
		Replicate (R)	*0.26*	
		*P*J interaction*	*3.54*	***
		*J*R interaction*	*0.43*	
	Lemon	*Product (P)*	*48.10*	***
		Judge (J)	*36.65*	***
		Replicate (R)	*0.10*	
		*P*J interaction*	*23.80*	***
		*J*R interaction*	*1.56*	
	Cheesy/creamy	*Product (P)*	*7.77*	**
		Judge (J)	*12.62*	***
		Replicate (R)	*0.60*	
		*P*J interaction*	*2.58*	**
		*J*R interaction*	*0.68*	
	Salty	*Product (P)*	*8.66*	***
		Judge (J)	*81.40*	***
		Replicate (R)	*0.48*	
		*P*J interaction*	*1.59*	
		*J*R interaction*	*1.49*	
	Bitter	*Product (P)*	*11.45*	***
		Judge (J)	*48.28*	***
		Replicate (R)	*0.36*	
		*P*J interaction*	*10.80*	***
		*J*R interaction*	*0.82*	
APPEARANCE	*Dark specks*	*Product (P)*	*31.44*	***
		Judge (J)	*11.51*	***
		Replicate (R)	*0.66*	
		*P*J interaction*	*3.03*	**
		*J*R interaction*	*0.35*	
	Grey colour	*Product (P)*	*25.33*	***
		Judge (J)	*7.50*	***
		Replicate (R)	*0.99*	
		*P*J interaction*	*1.88*	
		*J*R interaction*	*0.71*	

Appendix 6: ANOVA results showing source of variation and its significance (cont.)

TEXTURE when cold

Attribute	*Source of variation*	*F-Ratio*	*Significance*
Green colour	*Product (P)*	*2.64*	
	Judge (J)	*16.46*	***
	Replicate (R)	*0.26*	
	*P*J interaction*	*8.51*	***
	*J*R interaction*	*0.56*	
Yellow colour	*Product (P)*	*21.25*	***
	Judge (J)	*26.87*	***
	Replicate (R)	*0.66*	
	*P*J interaction*	*6.50*	***
	*J*R interaction*	*2.46*	*
Darkness	*Product (P)*	*72.15*	***
	Judge (J)	*4.26*	**
	Replicate (R)	*1.70*	
	*P*J interaction*	*2.24*	*
	*J*R interaction*	*0.77*	
Lumpy	*Product (P)*	*0.71*	
	Judge (J)	*5.54*	***
	Replicate (R)	*0.42*	
	*P*J interaction*	*2.37*	*
	*J*R interaction*	*1.35*	
Set	*Product (P)*	*29.20*	***
	Judge (J)	*15.06*	***
	Replicate (R)	*0.75*	
	*P*J interaction*	*5.56*	***
	*J*R interaction*	*1.64*	
Newly formed skin	*Product (P)*	*0.01*	
	Judge (J)	*3.64*	**
	Replicate (R)	*0.06*	
	*P*J interaction*	*1.44*	
	*J*R interaction*	*1.36*	

	Not significant
*	*Significant at the 5% level*
**	*Significant at the 1% level*
***	*Significant at the 0.1% level*

Appendix 2: References

References are used to ensure that the panellists are assessing and rating the same aroma and flavour. The list below gives examples of some of the types of sample assessed.

Béchamel sauce
Béchamel sauce made with vinegar
Roux mixes - butter and flour mixes
Water from cooked potatoes and peas
Sour milk
Béchamel sauce made with citric acid
Stewed apples
Rancid butter
Condensed milk
Range of milk samples (no fat to Jersey milk)
Curd cheese
Béchamel sauce made with corn flour
Artificial lemon flavour
Soured cream

Appendix 3: Sample presentation order

Replicate 1:

No.	Code								Sample							
1	155	046	784	982	519	375	818	128	V	C	V2	V7	G1	V10	V5	I1
2	155	046	784	982	519	375	818	128	V	C	V2	V7	G1	V10	V5	I1
3	155	046	784	982	519	375	818	128	V	C	V2	V7	G1	V10	V5	I1
4	155	046	784	982	519	375	818	128	V	C	V2	V7	G1	V10	V5	I1
5	155	046	784	982	519	375	818	128	V	C	V2	V7	G1	V10	V5	I1
6	784	982	818	155	046	128	375	519	V2	V7	V5	V	C	I1	V10	G1
7	784	982	818	155	046	128	375	519	V2	V7	V5	V	C	I1	V10	G1
8	784	982	818	155	046	128	375	519	V2	V7	V5	V	C	I1	V10	G1
9	784	982	818	155	046	128	375	519	V2	V7	V5	V	C	I1	V10	G1
10	046	375	128	818	155	982	519	784	C	V10	I1	V5	V	V7	G1	V2
11	046	375	128	818	155	982	519	784	C	V10	I1	V5	V	V7	G1	V2
12	046	375	128	818	155	982	519	784	C	V10	I1	V5	V	V7	G1	V2

Replicate 2:

No.	Code								Sample							
32	884	598	787	672	343	075	133	939	V2	V7	V5	V	C	I1	V10	G1
33	075	672	343	133	598	939	884	787	I1	V	C	V10	V7	G1	V2	V5
34	075	672	343	133	598	939	884	787	I1	V	C	V10	V7	G1	V2	V5
35	075	672	343	133	598	939	884	787	I1	V	C	V10	V7	G1	V2	V5
36	075	672	343	133	598	939	884	787	I1	V	C	V10	V7	G1	V2	V5
37	939	884	672	343	598	787	075	133	G1	V2	V	C	V7	V5	I1	V10
38	939	884	672	343	598	787	075	133	G1	V2	V	C	V7	V5	I1	V10
39	939	884	672	343	598	787	075	133	G1	V2	V	C	V7	V5	I1	V10
40	939	884	672	343	598	787	075	133	G1	V2	V	C	V7	V5	I1	V10
41	939	884	672	343	598	787	075	133	G1	V2	V	C	V7	V5	I1	V10

Appendix 4: Mean scores for each attribute

Attribute	C	V	G1	V2	V5	V7	V10	I1
Strength	7.68	3.93	4.66	4.02	4.85	5.67	4.28	4.46
Skin coverage	8.71	4.63	4.14	4.44	4.03	5.60	5.35	4.47
Cheesy	0.66	2.67	2.09	2.20	2.44	2.17	1.18	1.31
Milky/sweet	0.88	3.23	3.19	3.03	2.11	1.99	2.42	2.24
Roux	0.51	2.61	1.53	1.81	1.85	1.83	2.09	1.78
Vegetable	0.69	1.66	1.67	1.74	1.30	1.50	1.68	1.89
Sour	3.23	2.08	1.86	2.24	2.69	2.80	1.44	1.74
Lemon	0.96	1.77	1.66	1.58	2.07	1.63	1.17	1.77
Stewed apples	1.02	1.57	3.19	3.04	2.37	2.96	2.97	3.12
Waxy maize starch	8.87	1.58	1.36	1.22	1.57	2.21	1.75	1.31
Thin to thick	9.49	4.58	2.73	2.59	2.72	2.86	3.06	2.64
Wobbles	9.44	2.33	1.68	1.71	1.83	1.87	1.77	1.63
Pourability	1.04	6.23	7.89	8.18	7.92	7.36	7.37	7.95
Coats cup	9.38	4.60	3.88	4.96	3.65	4.47	4.67	4.42
Dropping consistency	9.40	2.98	1.73	1.43	2.14	2.27	2.31	1.36
Coats spoon	9.35	5.88	4.38	5.10	5.24	4.76	4.62	4.44
Holds it shape	9.33	3.03	2.20	2.02	2.82	2.41	2.62	2.14
Shiny	7.98	7.20	6.98	6.64	7.01	6.87	6.49	6.77
Refills trough	0.44	4.80	7.95	7.05	7.58	7.34	6.82	8.16
Bland-strong	8.36	5.09	5.38	4.76	5.86	5.77	5.46	5.52
Tangy	7.09	5.60	6.61	5.45	6.92	6.49	6.06	6.23
Buttery	2.52	4.21	2.90	3.19	2.70	2.23	2.66	3.20
Milky/creamy	2.87	4.23	3.04	3.42	3.16	2.60	2.67	3.71
Sour milk	5.63	4.53	3.68	3.56	3.84	4.37	4.31	3.31
Vinegar	4.39	3.69	2.97	3.97	3.87	4.22	3.82	2.97
Starchy	6.00	3.31	2.81	3.07	3.33	2.80	2.97	3.11
Salty	4.99	4.24	4.36	4.09	4.42	3.97	4.14	4.10
Peppery	3.48	2.81	2.73	2.80	3.32	3.29	3.47	3.06
Sweet	0.68	1.26	1.42	1.51	0.77	0.77	0.81	1.32

Appendix 4: Mean scores for each attribute (cont.)

Attribute	**C**	**V**	**G1**	**V2**	**V5**	**V7**	**V10**	**I1**
Waxy maize starch	8.64	1.37	1.41	1.34	1.78	1.34	1.60	1.33
Lemon	4.00	4.53	3.23	2.94	3.92	3.62	3.72	4.04
Slimy	7.45	3.78	3.23	3.48	3.93	3.51	3.16	3.37
Greasy	5.16	4.74	5.12	4.93	4.04	3.97	4.50	3.39
Smooth	8.23	7.37	6.58	6.89	6.63	6.62	6.16	6.85
Starchy/floury	5.66	3.39	3.33	4.17	3.78	4.17	3.89	3.07
Coats mouth	7.33	5.32	4.52	4.43	3.84	3.59	3.96	3.52
Vinegary	3.13	2.98	3.20	2.83	2.79	2.63	3.41	2.50
Acidic	5.84	4.86	5.00	4.66	5.38	5.25	5.14	5.11
Lemon	3.55	3.80	3.68	2.63	3.43	3.18	2.66	3.25
Cheesy/creamy	2.64	3.08	2.42	2.02	2.79	2.46	2.18	2.61
Salty	4.16	3.76	4.26	3.41	4.55	3.72	4.17	4.24
Pepper	3.34	3.18	2.85	3.56	3.82	3.79	2.99	3.36
Leaves mouth dry	6.54	4.56	5.54	4.37	5.78	5.16	5.44	5.53
Dark specks	1.71	2.45	3.72	5.61	4.78	6.22	5.70	5.34
Shade	0.46	2.09	4.63	5.91	5.03	6.27	6.56	5.93
Lumpy	2.17	2.31	2.25	3.17	3.71	3.52	4.41	3.59
Newly formed skin	2.67	3.95	3.12	2.94	1.89	3.16	3.89	3.00
Set	8.44	7.17	5.12	5.84	5.01	3.78	6.74	5.06

File : T:\PROJECTS\INNOVA~1\largestu.frs : Large study September-October 1998
Profile 1
Number of repetitions : 2
Number(s) of the repetitions taken : All
Option : Descriptive statistics
Raw data
Computation of the statistics : global

Appendix 5: ANOVA results showing source of variation and its significance

	Attribute	Source of variation	F-Ratio	Significance
AROMA	Strength	Product (P)	8.89	***
		Judge (J)	4.91	***
		P*J interaction	1.74	*
	Cheesy	Product (P)	4.07	***
		Judge (J)	5.21	***
		P*J interaction	0.89	NS
	Milky/sweet	Product (P)	5.44	***
		Judge (J)	13.44	***
		P*J interaction	1.40	NS
	Roux	Product (P)	3.91	**
		Judge (J)	12.74	***
		P*J interaction	1.04	NS
	Vegetable	Product (P)	2.64	*
		Judge (J)	22.11	***
		P*J interaction	1.63	*
	Sour	Product (P)	3.39	**
		Judge (J)	12.79	***
		P*J interaction	1.38	NS
	Lemon	Product (P)	1.04	NS
		Judge (J)	8.77	***
		P*J interaction	1.20	NS
	Stewed apples	Product (P)	3.88	**
		Judge (J)	11.50	***
		P*J interaction	1.05	NS
	Waxy Maize Starch	Product (P)	32.36	***
		Judge (J)	3.49	**
		P*J interaction	0.74	NS
APPEARANCE	Thickness	Product (P)	108.50	***
		Judge (J)	40.56	***
		P*J interaction	3.13	***
	Skin coverage	Product (P)	9.53	***
		Judge (J)	3.27	**
		P*J interaction	1.94	**
	Wobbles	Product (P)	891.41	***
		Judge (J)	786.28	***
		P*J interaction	19.16	***
	Pourability	Product (P)	97.51	***
		Judge (J)	21.22	***
		P*J interaction	1.95	**
	Coats cup	Product (P)	18.32	***
		Judge (J)	6.65	***
		P*J interaction	1.21	NS
	Dropping consistency	Product (P)	73.32	***
		Judge (J)	11.55	***
		P*J interaction	1.39	NS

Appendix 5: ANOVA results showing source of variation and its significance (cont.)

	Attribute	Source of variation	F-Ratio	Significance
	Coats spoon	Product (P)	28.53	***
		Judge (J)	34.06	***
		P*J interaction	1.64	*
	Holds its shape	Product (P)	71.45	***
		Judge (J)	38.51	***
		P*J interaction	1.66	*
	Shiny	Product (P)	5.26	***
		Judge (J)	141.26	***
		P*J interaction	7.91	***
	Refills trough	Product (P)	33.89	***
		Judge (J)	3.92	***
		P*J interaction	0.58	NS
FLAVOUR	Strength	Product (P)	4.81	***
		Judge (J)	3.45	**
		P*J interaction	0.95	NS
	Tangy	Product (P)	1.79	NS
		Judge (J)	10.05	***
		P*J interaction	1.26	NS
	Buttery	Product (P)	3.59	**
		Judge (J)	32.11	***
		P*J interaction	1.49	NS
	Milky/creamy	Product (P)	1.99	NS
		Judge (J)	30.23	***
		P*J interaction	0.88	NS
	Sour milk	Product (P)	4.44	***
		Judge (J)	38.82	***
		P*J interaction	1.16	NS
	Vinegar	Product (P)	1.72	NS
		Judge (J)	40.91	***
		P*J interaction	1.50	NS
	Starchy	Product (P)	5.12	***
		Judge (J)	11.41	***
		P*J interaction	1.24	NS
	Salty	Product (P)	0.83	NS
		Judge (J)	51.03	***
		P*J interaction	0.77	NS
	Peppery	Product (P)	0.56	NS
		Judge (J)	21.53	***
		P*J interaction	0.58	NS
	Sweet	Product (P)	2.26	* (NK=NS)
		Judge (J)	8.83	***
		P*J interaction	1.80	*
	Waxy maize starch	Product (P)	70.81	***
		Judge (J)	9.30	***
		P*J interaction	1.13	NS
	Lemon	Product (P)	0.90	NS
		Judge (J)	12.34	***
		P*J interaction	0.99	NS
MOUTH FEEL	Slimy	Product (P)	17.69	***
		Judge (J)	51.72	***
		P*J interaction	2.38	***
	Greasy	Product (P)	3.11	**
		Judge (J)	47.16	***
		P*J interaction	2.41	***

Appendix 5: ANOVA results showing source of variation and its significance (cont.)

	Attribute	Source of variation	F-Ratio	Significance
	Smooth	Product (P)	5.51	***
		Judge (J)	97.38	***
		P*J interaction	2.85	***
	Starchy/floury	Product (P)	2.68	*
		Judge (J)	5.26	***
		P*J interaction	2.03	**
	Coats mouth	Product (P)	7.47	***
		Judge (J)	11.46	***
		P*J interaction	1.33	NS
	Vinegary	Product (P)	0.42	NS
		Judge (J)	28.93	***
		P*J interaction	0.72	NS
	Acidic	Product (P)	0.90	NS
		Judge (J)	51.44	***
		P*J interaction	0.67	NS
	Lemon	Product (P)	1.06	NS
		Judge (J)	32.82	***
		P*J interaction	0.82	NS
	Cheesy/creamy	Product (P)	1.15	NS
		Judge (J)	31.85	***
		P*J interaction	1.08	NS
	Salty	Product (P)	0.93	NS
		Judge (J)	49.16	***
		P*J interaction	0.61	NS
	Pepper	Product (P)	0.48	NS
		Judge (J)	20.22	***
		P*J interaction	0.54	NS
	Leaves mouth dry	Product (P)	1.43	NS
		Judge (J)	13.34	***
		P*J interaction	0.94	NS
APPEARANCE	Dark specks	Product (P)	18.97	***
		Judge (J)	12.98	***
		P*J interaction	2.47	***
	Shade	Product (P)	77.52	***
		Judge (J)	12.56	***
		P*J interaction	2.60	***
TEXTURE when cold	Lumpy	Product (P)	2.53	*
		Judge (J)	12.27	***
		P*J interaction	1.67	*
	Set	Product (P)	15.24	***
		Judge (J)	20.36	***
		P*J interaction	1.92	**
	Newly formed skin	Product (P)	2.18	*
		Judge (J)	16.20	***
		P*J interaction	1.81	*

	Not significant
*	Significant at the 5% level
**	Significant at the 1% level
***	Significant at the 0.1% level

Sourcing added value food ingredients from home grown oats
'Innovation'

Part 5

Assessment of oat emulsifier: Studies carried out 1987-8

by

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Analysis of polar lipids of oats carried out 1987-8

This work originated when it was found that oats contained a high proportion of various polar lipids. One of these was an unusual glycolipid composed of digalactosyl-diacyl-diglyceride containing long chain interesterified hydroxy fatty acids on two positions of the glycerol. This is a particularly active component amongst the other polar lipid species. This, with the other components, gives the polar fraction viscosity-reducing properties in chocolate which were intermediate between those of soya lecithin and the artificial emulsifier polyglycerol polyricinoleate. This fraction thus had the advantages:

1. It was an entirely "natural " surface active agent.
2. It had properties that were superior in chocolate to soya lecithin.
3. At the levels used in chocolate (and probably for any other use) it would be used at a level at or below that naturally occurring in oats.
4. It was also a satisfactory emulsifier for use in other products such as bread and frying oils.

Several methods were used to extract the polar fraction. Initially the non-polar lipids were removed from the oats with hexane, and then the polar fraction was extracted with ethanol or isopropanol. It was then found that the entire lipid fraction could be extracted with isopropanol or ethanol, the solvent removed, and the polar lipid fraction extracted from the initial product with methanol. This was likely to give a more commercially viable process.

This initial work was carried out on rolled oats purchased from the supermarket. We proceeded to investigate other crops and also obtained oats of different varieties and from different growing sites. The presence of the active polar glycolipid in each case was assessed by thin layer chromatography.

No other cereal crop that we examined was found to contain this component. Studying the different varieties and growing sites did show considerable differences. A total of 191 samples of oats were examined. Of these 83 were winter oats and 108 were spring oats.

Winter Oats

The winter oats were divided between four sites:

Inkberrow
Berriew Powys
Peithyll
Dundee

Eleven types of winter oats were studied, five of them commercial varieties at the time (Pennal, Peniarth, Image, Luster and Solva). Two were naked oats.

The yield of total lipid (including nonpolar lipid) for the samples in general varied with the site. Inkberrow in almost every case gave lower oil yields than any other site, in some cases considerably lower. The order of yields for the other sites, however, varied with variety. The varieties which gave high total oil yield (9-10%) were a non-commercial variety (88/55 79-77Cn5) and Kynon. The high yield here would have been partially accounted for by the fact that they were both naked oats, where the yield obtained is expressed with no reference to the husk. Other non-naked varieties gave no large difference in oil content, being on average about 6 - 7 %

The levels of the active polar lipid in the winter oats were, in all but one case, lower than that present in the sample of rolled oats which we used as our standard. The exception was a non-commercial variety listed as 88/11 80-30Cn16/2/2, which on average contained twice the level of our standard.

Spring Oats

The spring oats were divided between five sites:

Morfa
Sarn Powys
Cambridge
Dundee
Brynllys

Fifteen types of spring oats were studied, four of them commercial (Commander, Dula, Rollo, and Rhiannon). One variety (Rhiannon) was a naked oat.

The total oil yields for spring oats varied by site and variety. Dundee and Cambridge tended to give higher yields, while Sarn Powys and Morfa tended to give low yields, but this was only a trend. Rhiannon consistently gave the highest yield for a commercialised variety (>10%), and other non-commercialised varieties gave only a slightly higher yield. As with the winter oats however, this was a naked oat. Again average yield of oil was in the region of 6%.

The highest levels of active polar lipid were obtained in the following varieties:
Avalanche = Keeper >88/3 08523Cn11 > Dula = Rollo

Rhiannon, which gave a high oil yield was found to contain none of this component. With two exceptions (including Rhiannon), spring oats contained higher levels of the active polar lipid than winter oats.

In both winter and spring oats there appeared to be little, if any, correlation between oil extracted and active polar lipid level. Presumably this means that the formation of the two are not related.

Conclusion

The only large variations in oil content were found in naked oats, where the comparison is not genuine. Inkberrow appeared to give consistently lower oil yields than other sites.

The active polar lipid level could not be correlated with oil level, but varied with variety. One variety (Rhiannon) gave high oil level but contained no active polar lipid. Spring oats tended to give higher yields of the active polar lipid than winter oats.

Sourcing added value food ingredients from home grown oats
'Innovation'

Part 6

Review of oat fractionation technology

by

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Introduction

Traditional processing of oats consists primarily of cleaning, grading, dehulling, cutting and rolling into flakes, with the product aimed primarily at the human food sector. However, with increasing interest in specific oat fractions, several new processes have been developed which are capable of producing much purer fractions of individual components. Many of these processes have only been demonstrated on laboratory or plant scale, though a few have reached some degree of commercial success.

Research has concentrated on fractions containing high levels of bran, since these are enriched in oat β-glucan, which is the component widely believed to be responsible for lowering cholesterol levels in humans. Until recently these processes have only been able to enrich the β-glucan content by 2-3 times but new technology means that it is now possible to increase this to a much higher degree of purity, opening new markets for the product.

Processes also exist for extraction of oil, principally using existing hexane-extraction technology which has been modified to cope with the extraction properties of oats. Development of new varieties of oats containing high levels of oil has increased interest in this area and led to at least one commercial facility for extraction of oat oil.

Protein and starch are seen in some processes as by-products but new markets for these fractions means that processes have been developed in which they are the primary product, thus increasing the purity with which they can be obtained. This has resulted in added value, improving the overall economy of processing, and combined with the development of new markets for oat products has been the driving force behind development of commercial processing plants.

The following review discusses current and possible new technologies concerned with the processing of oats into fractions enriched in some of these functional components.

Extraction of β-glucan

Technologies for producing fractions enriched in oat β-glucan have been in existence for some time, although early processes were aimed at producing a bran fraction rather than a pure preparation of β-glucan itself. Bran was first commercially produced by the Quaker Oats Company in the late 1970's, containing double the starting amount of β-glucan. Commercial oat bran is a mixture of components, which has led to some confusion concerning its definition. In an attempt to clarify matters, the American Association of Cereal Chemists has produced a definition which states that the bran fraction is not more than 50% of the original starting material, has a β-glucan content of at least 5.5% and a total dietary fibre content of at least 16.0% of which at least one third is soluble fibre. More traditional methods for producing a bran-enriched fraction have been adapted from processes for production of wheat bran, consisting essentially of roller-milling and sieving, with the bran being collected as the coarse fraction. One of the simplest processes for extraction of a more purified form of bran involved pin-milling into a flour, defatting with pentane-hexane, and using air-classification to give a range of flours; one of the fractions was enriched in fibre and protein and although β-glucan was not measured it can reasonably be assumed that it was also enriched in this fraction. However, the yield of this fraction was only 22-24% of the whole flour.

With further significance of β-glucan in the human diet in the mid 1980's, more attention was paid to extracting β-glucan itself rather than as a by-product of the extraction of other components. An early process patent for extraction of purified β-glucan was issued to the Quaker Oats Company (Hohner and Hyldon, 1977) which described a process using dry-milling and air-classification of hexane-defatted oat flakes. This yielded a bran fraction comprising 28% of the original oats which was processed further by extraction of the β-glucan in sodium carbonate solution, centrifugation to remove the extracted bran, precipitation of protein and starch, and finally concentration and precipitation of the resultant β-glucan in the supernatant using propan-2-ol. Unfortunately, no details were given of the purity of the β-glucan, since it was not the primary product, although it is believed to be in excess of 60%. Although this was an early process patent, some of the methods used still show much promise for future processes since both protein and β-glucan components are extracted to a high degree of purity following solvent extraction to remove the oil. Studies by Oughton (1980a,b) and Boczewski (1980) also involved defatting oats with hexane but then used a series of hydrocyclones and screening techniques to separate protein and bran-enriched fractions. Although the β-glucan content of the bran was not quantified it is likely to be considerably enriched compared to the starting material.

Other processes based on aqueous steeping have been proposed but have not been adopted commercially. Inglett (1992) described a process for production of a soluble fibre fraction using amylolysis of an aqueous slurry at pH 6.0 and centrifugation of the remaining solids to produce a product which contains 7-8% β-glucan, at least as much as commercial oat brans; the commercial product was named Oatrim, and a number of applications were identified, including as a fat substitute in ice cream (Beaudette, 1991).

In a patent assigned to Alko AB (Finland) Lehtomaki *et al.* (1990) purified β-glucan further using cold water steeping followed by successive wet-screening. Few process details were given but it is claimed that the final product contained 15-30% β-glucan. However, it was not the primary product of these processes. The process was commercialised by Alko but production ceased in 1989. The equipment is currently for sale together with the patents associated with the process.

Some processes have been proposed which combine both solvent extraction with aqueous processing techniques. Wood *et al.* (1989) described a pilot scheme for extraction of large quantities of β-glucan for use in nutritional studies. The bran which formed the basis of the pilot extraction scheme was prepared by defatting and air-classification and contained 11-17% β-glucan. The bran was refluxed in 75% ethanol to inactivate enzymes and the β-glucan extracted using sodium carbonate to give a product which contained 78% β-glucan. Pilot plant material was shown to have a lower viscosity than that produced in the laboratory due to shear forces during centrifuging leading to lower molecular weight, though no major structural differences were found. Myllymaki *et al.* (1989) also described a process, incorporating a number of technologies in order to extract a range of components including one enriched in β-glucan as well as others containing protein and starch. It is believed that some attempt is currently underway to commercialise the process in Finland. A very pure β-glucan fraction (92-99%) was prepared by Westerlund *et al.* (1993) by fat extraction, enzymic removal of starch and protein and subsequent precipitation of water-soluble polysaccharides with 60% aqueous ethanol and 20% ammonium sulphate. Alkaline treatment of flour and removal of starch residue, isoelectric precipitation of protein, and alcoholic precipitation of gum were used by Dawkins and Nnanna (1993) who found that 50-68% β-glucan was extractable from rolled oats, and 70-89% from oat bran, depending on temperature and pH of extraction (this was the procedure used for extraction of β-glucan in the current study). Bhatty (1996) examined sodium hydroxide as an alternative to sodium carbonate for extraction and obtained 69-72% β-glucan with 87-95% recovery from oat bran as the starting material. Finally, Beer *et al.* (1996) compared different processing technologies for their efficiency in extraction and found alcoholic precipitation to be superior to dialysis or ultrafiltration in terms of yield, but the resultant β-glucans differed in viscosity depending on extraction method.

A novel process was proposed by Burrows *et al.* in 1984 using an aqueous steeping technique. This formed the basis of the only process for extracting relatively pure β-glucan known to have reached commercial success. The research described in the patent was based on an observation by Burrows that if whole oats are steeped in water at 50°C, after 24 hr the kernel can be squeezed to express the endosperm contents from the bran layers rather like toothpaste being squeezed from a tube. Further work was carried out to determine the basis of the earlier observations, leading to the finding that the digestion of the endosperm cell walls was responsible for the separation of the endosperm contents. At this time the emphasis was on the production of a bran-free flour fraction rather than β-glucan, the significance of which in terms of the human diet was yet to be realised. A patent for the steeping process was granted in 1984 but the process was far from ideal at this stage since significant lipase activity led to poor quality of the final product with fatty acids being present in the bran and flour fractions. Further work led to an improved process in which the oats were steeped in aqueous alcohol (Collins and Paton, 1992), effectively preventing lipase activity and stabilising the final product. The alcohol also facilitated the separation of the flour from the bran. Primary products are a bran-free oat flour and a flour-depleted bran. Maceration of the steeped oats and sieving were used to separate the bran from the endosperm, and further maceration and sieving yielded a range of bran and flour fractions of varying composition. The flour was further refined into starch and a protein concentrate, and the bran extracted to produce a range of β-glucan powders of 65-85% purity. The β-glucan has particularly good water hydration properties which is believed to originate from the steeping in aqueous ethanol. Further downstream processing of the waste alcohol includes anionic and cationic exchange methods to produce a wide range of minor but economically valuable products, including phenolic acids, alkaloids, fatty acids, organic acids, uronic acids, amino acids, free sugars and avenathramides. The process was originally licensed to Canamino Inc. of Saskatoon for the production of a range of fractions marketed for use in the cosmetics sector with the establishment of a facility capable of processing up to 15,000 tonnes of oats per year. However, during 1998 the processing plant was closed down due to financial difficulties and is currently being offered for sale.

Extraction of oil

Much laboratory research has been carried out aimed at optimising the extraction of lipids from cereal grains and the results of this must be considered carefully in relation to any proposed pilot-scale or industrial processes. In general, extraction of lipids has been shown to depend on a number of factors including solvent type, extraction time, ratio of solvent to sample weight, temperature, and extraction equipment grain moisture, particle size, permeability of tissue, and age of grain (due to lipase activity). Lipids are generally classed as 'free' or 'bound', based on their extractability in various solvent types. 'Free' lipids are extracted with non-polar solvents such as ethyl ether, hexane or petroleum ether and lipid classes are mainly triglycerides, diglycerides, free fatty-acids, and sterols. Following extraction of 'free' lipids, 'bound' lipids can be extracted with polar solvents such as chloroform, methanol, ethanol, propanol, butanol, usually as water-alcohol mixtures; lipid classes are mainly glycolipids and phospholipids. A number of points should be considered in any extraction scheme for oat oil. It should be remembered that even solvents classed as non-polar have some degree of polarity and this can lead to variable extraction when studies with different solvents are compared, and polar solvents often extract components other than lipids, which may need further processing to remove.

Several laboratory studies have demonstrated the specific need to avoid the deterioration of lipids in oats due to lipase and lipoxygenase. Liukkonen *et al.* (1992a) showed that the most significant effect of steeping of oats in water was the appearance of free fatty-acids at the expense of triglyceride. Hence prevention of lipid hydrolysis rather than oxidation should be the main aim in any manufacturing process in prevention of lipid deterioration. Liukkonen *et al.* (1992b, 1993) subsequently reported that lipid stability was improved in a wet-steeping process by steeping in alkaline conditions, at pH 9 or above as opposed to water. Acid steeping conditions were also effective but not as much as alkaline. In addition, in alkaline steeping, loss of linoleic acid was also reduced. Ekstrand *et al.* (1993) found that steam treatment was effective in destroying lipase activity during wet-steeping but not dry heat treatment, although the latter gave some protection during storage. Surprisingly in this study, it was reported that lipolysis was more pronounced in whole grain than flour samples. Molteberg *et al.* (1995) found that storage at high relative humidity resulted in lower lipid content and proposed that the reduction in free fatty acids was due to complexing whereas the reduction in linoleic acid was due to production of volatile oxidative products. Collins and Paton (1990) also noted the inhibition of lipase activity by the use of alcoholic solvents in steeping of oats. The results of these studies indicate that oats should be steam-treated, stored at low relative humidity and processed in either alcoholic or alkaline conditions to limit deterioration of lipid components.

A number of potential commercial schemes have been proposed for the extraction of oil from oats, though generally the oil is not the primary product of the process, which in most cases is a means of defatting the oats prior to purification of protein, starch or bran. Hohner and Hyldon (1977) on behalf of the Quaker Oats Company, described a process in which oat flakes were extracted for 45 min with hexane as a preliminary to milling and air-classification for the preparation of a bran fraction. Both normal and high oil varieties were used in the study, but no indication was given of the yields of crude oil from the flakes. Oughton (1980a,b) also described a process in which hexane was used to extract oil. In this process high protein fractions were produced using a variety of stages including hexane extraction, liquid cyclones, and screening techniques. Oats were slurried with hexane following dehulling and pin-milling. Further hexane was then used for extraction of oil from solids separated by hydrocyclones and sieving. No details were given of the oil recovered at each stage since this was not the primary product. Boczewski (1980) developed this process further, again using hexane to extract oil from crushed oats, but modifying the pre-extraction roller-milling stage. Potter *et al.* (1993) also described an extraction process using hexane which produced an oat oil which was subsequently shown to have good dermatological properties. It is believed that this process has been commercialised by a US company Nurture, although details of the production process and confirmation of the existence of the company are difficult to obtain.

Other processes have been described in which polar solvents were used for extraction of oil. Myllmaki *et al.* (1989) described a number of extraction schemes containing both dry and wet milling in which lipids were extracted with polar solvents (propan-2-ol, ethanol, acetone in various water/solvent proportions) at temperatures of 50-90°C. These were reported to increase the amount of polar lipids extracted, and the total lipid yield. The scheme also favoured the extraction of antioxidant components into the oil fraction, due to the high polarity of the solvents used. Lipase and lipoxygenase were inactivated both by heating and from inhibition by the alcoholic solvents used. Another process was described in which oats were extracted with either ethanol or isopropanol, this extract was further extracted with methanol and then the methanol evaporated to leave a surfactant which had uses as an emulsifying agent (Evans *et al.*, 1990). One further method has been described, using supercritical carbon dioxide (Fors and Eriksson, 1990). Using this process, lower levels of free fatty-acids were produced than with a comparable extraction with hexane, though it is not clear whether this was due to lower levels of lipase activity or incomplete extraction of free fatty-acids.

On an industrial scale solvent extraction is a well-established technique for extraction of oil from many vegetable materials, originating in Europe in 1870. Continuous solvent-extraction systems were developed just after World War 1 and these proved to be efficient in extracting oil from low-oil sources. Modern systems extract using several countercurrent washes with hexane. Starting material for the extraction is generally cracked, flaked, ground, or pressed (to pre-extract some oil in the latter case). Two fractions thus result, the hexane, containing oils, and the residual meal. The extracted meal is carried in sealed conveyors for solvent recovery in enclosed vessels using steam, while hexane is removed from the oil in rising film evaporators and eventually by vacuum distillation. The crude extracted oil may then be refined in a number of steps including degumming, alkali-refining, bleaching, and high-vacuum steam-distillation. Using hexane, oil yields of over 95% are possible, with over 95% recovery of hexane solvent. these systems are widely used for extraction of oil from materials such as rape and soya.

Other commercial scale industrial processes developed for extraction of oils from vegetable material are based on pressing or aqueous enzymatic techniques. An overview of the processes is given by Rosenthal *et al.* (1996). Pressing, using either hydraulic or expeller methods, is generally only used on sources high in oil content (<35%) so is not considered further in this review. An alternative method for oil extraction is using an aqueous enzymatic process, a relatively new technique (Rosenthal *et al.*, 1996). It utilises the principle of hot water flotation in which oil floats on the surface of hot water and can be skimmed off for recovery. Significant advantages of this technology are that it is safer, more environmentally friendly, and results in less deleterious effects of extraction on the residual protein in the oil source, releasing this as a valuable by-product. However, as yet there is no full scale industrial utilisation of this technology and its use has not been demonstrated using oats as the starting material.

The only plant known to be extracting oat oil on a commercial scale is that run by Con Agra Speciality Grain Products at Sough Sioux City, Nebraska, USA. The plant was commissioned in February 1996 and has the capacity to extract oil from 100 tonnes oats per day (approx. 8000 kg oil). The plant cost $25 million to construct and is situated next to ConAgra's oat mill. Only limited details of the extraction process are known since it has not been patented, in an effort for the details to remain confidential, but it is reported to comprise of a hexane extraction facility originally used for cocoa extraction which has been modified to cope with the specific properties of oat grains. Oats are cleaned and dehulled in the ConAgra oat mill, and then processed into an extruded product prior to oil extraction. The extruded oat pellets are transported through the extraction vessel on a slowly moving chain belt, oats entering at the top with hexane slowly draining through the oats. Six recirculators pump hexane back into the system for efficient extraction. The defatted oat pellets then pass into a further vessel which is a steel drum containing layers of gauze through which steam is injected to extract oil impregnated hexane. The defatted oats are removed from this vessel, cooled and passed on for further processing. Approximately 70% of the total oat oil is extracted in the first vessel and 20% in the second. The oil-impregnated hexane then passes through a further vessel where hexane is stripped out using a heating process, with the recovered hexane returning to the start of the system. It is claimed that the recovery of hexane is approximately 98%. The oil is further purified by removal of residual water and starch, the whole system operating as a closed loop. Although the process has been modified by using extruded oats instead of flakes as a starting material, it is assumed that in all other respects standard hexane extraction technology is employed.

Facilities also exist for the commercial extraction of oat oil at Swedish Oat Fiber, a company producing a range of high fibre oat flours for the food industry. This process utilises ethanol for extraction of oil but is not believed to routinely producing oil due to lack of markets for the product.

Extraction of starch

Extraction of starch in pure form from oats is more difficult than from other cereals from which starch is commonly extracted (maize, wheat, potato and rice) because the starch granules are much smaller and do not separate cleanly from the protein on centrifugation. However, some laboratory studies have shown that under suitable conditions slurried oat flour can be centrifuged to produce layered sediments with starch concentrated in the heaviest fractions. Repeated washing and centrifuging can produce a starch with approximately 0.5% protein and 1% lipid (Youngs, 1974). This was used as the basis for laboratory scale extraction by Adkins and Greenwood (1966), Morrison *et al.* (1984) and Tester and Karkalas (1996). Water-washing has benefits for laboratory studies of starches since, unlike more rigorous procedures in which the protein is solubilised by alkaline solutions (Wu *et al.*, 1973; Paton, 1977), the starch is isolated in a native form containing true starch lipids and proteins. This process would not be feasible on a larger scale due to the difficult separation of the starch from the protein and bran fragments.

A number of techniques have been proposed for use on an industrial scale, though these have often been in combination with processes for isolating other oat components. One of the few processes described in which starch is the primary product is that of Wilhelm *et al.* (1989). In this process the oats were dehulled, dry-milled, and then steeped in a solution containing cellulolytic and hemicellulolytic enzymes. Residual fibrous material was removed by screening and then the starch and protein separated using a series of hydrocyclones to give a prime starch with lower than 1% protein. A secondary starch was obtained by further fractionation, containing 10-15% protein. The protein fraction was also relatively pure but the bran was highly degraded due to the enzymes earlier in the process. It is unlikely that this approach would be suitable commercially due to the loss of the valuable β-glucan fraction. The Hohner and Hyldon (1977) process produced the starch and protein as a co-product. Removal of lipid and β-glucan in this process may have facilitated separation of protein and starch but no attempt was made to purify either starch or protein any further. In the process described by Oughton (1980b) the groats were simultaneously defatted and fractionated using a number of hydrocyclones but since the primary aim was to extract the oat protein, the starch was relatively impure, containing up to 7% contaminating protein.

One of the main features of oat starch is the presence in its native form as compound granules. Disintegration of these compound granules results in a starch with extremely fine texture and good flow properties. Malkki *et al.* (1995) described a method in which the compound granules are disintegrated, by treatment with either a surface-active agent or a lipolytic enzyme. As well as production of individual granules, it is claimed that the functional properties of the starch are altered, such as the water-binding, gelatinisation and viscosity properties, and its enzymic susceptibility. It is claimed that the resultant starch has uses in biodegradable plastics and as surface-active agents. The process has the potential to be used as an add-on to another process in which relatively pure starch is extracted.

Some processes for extraction of oat starch have been used commercially, though more on a pilot scale than true industrial scale. The original process of Burrows *et* *al.* (1984), modified by Collins and Paton (1992), was used by Ceapro Inc. to produce a pure oat starch. This process uses a series of sieving and centrifugation stages but little detail is given regarding the separation of starch from protein, despite marketing what is regarded as a pure starch fraction. It is claimed that the starch has uses in a range of cosmetic and skin care products such as eye blushes and talcum powder.

Oat starch was produced commercially by a Finnish company Alko AB although the plant ceased operation in 1989 and is currently being offered for sale. The processing details were covered by the patent by Lehtomaki *et al.* (1990) and consisted of cold water steeping followed by successive wet screening. The starch produced was relatively pure and would have been likely to have contained true starch lipids and proteins. The properties of this starch must thus have been quite different to that produced by Ceapro.

Expansion of current processing of oats specifically for starch will only occur if further markets for the starch are developed. It is not likely that oat starch can compete in commodity starch markets with potato and maize starch so specialised markets must thus be established. In this respect the direct competitor is rice starch which also has small starch granules. Successful commercial development of oat starch production is thus likely to be wholly dependent on specialist uses for the starch and suitable markets for the other oat components.

Extraction of protein

During the early 1970's the main interest was in producing protein-enriched fractions, though more recently there has been much less interest in the extraction of protein than β-glucan, oil or starch. A number of workers proposed schemes which gave protein-rich fractions, involving processes of milling, defatting with pentane/hexane and either air-classification or use of hydrocyclones and screens (Wu and Stringfellow, 1973; Hohner and Hyldon, 1977; Boczewski, 1980; Oughton, 1980a, 1980b). Although the protein contents were high, at 55-83%, the recovery of original protein was rather low. The only process producing protein at relatively high purity (50-55%) and also with reasonable recovery is that of Wilhelm *et al.* (1989) but the use of cellulolytic enzymes during processing renders the bran fraction of low value and is hence less amenable to commercial development due to poor overall economics.

Most interest has been in the production of soluble protein extracts. Lenz and Paisley (1994) described a process using proteolysis whereby a soluble oat protein fraction is produced, along with soluble fat and soluble carbohydrate by-products. Ceapro Inc. have also produced a hydrolysed protein extract which is used in hair care products. This process utilises non-specific hydrolysis using acidic conditions.

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