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The incorporation of important traits underlying sustainable development of the oat crop through combining 'conventional' phenotypic selection with molecular marker technologies (OatLINK)

by

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1. ABSTRACT

The overall objective of OatLINK was to incorporate important traits underlying sustainable development of the oat crop through combining 'conventional' phenotypic selection with molecular marker technologies. Although conventional farmers, organic farmers, millers and poultry producers have different aims, there is also much in common in terms of the need for economic competitiveness, good agronomic and disease characteristics and sharing of molecular markers. The objectives of developing marker-assisted selection (MAS) and using it and phenotypic selection to develop and test oats for the milling and poultry industry have been brought together in a single project. Specific objectives were: i) to develop new molecular markers, UK mapping populations and contrasting bulk segregants for use in MAS of important traits, ii) to identify, incorporate, select and evaluate important traits for sustainable production and human consumption, iii) to identify, incorporate, select and evaluate important traits for sustainable production and premium livestock feed and iv) to identify, incorporate, select and evaluate important traits for organic production.

Progress has been made in the development and application of molecular markers and the first winter oat genetic linkage map developed. Phenotyping of populations developed for specific traits has been completed (low lignin husk, β-glucan) and markers have been identified associated with the dwarfing gene (*dw6*), components of height and yield, β-glucan content, oil content and naked character. The project confirmed that increasing oil content increases the energy (ME) value of naked oats, the potential of high oil oats in poultry diets and demonstrated the lower environmental impact per unit of energy provided compared with other cereals. NIR calibrations for oil and N content were developed and the suitability of oats for organic production systems was confirmed with yields of > 7t/ha achieved. Several potential varieties (winter, spring, husked and naked) have been tested for their end-user suitability in conjunction with the milling and poultry industry.

The development of markers for β-glucan will aid the development of oats that meet the needs of the milling industry and capitalise on the proven health benefit of oats. OatLINK has aided the promotion of oats as a low input cereal crop, a good break crop which aids cereal rotations with a lower environmental footprint than other cereals, as demonstrated by LCA (Life Cycle Assessment). It has also shown how future targeted research can further reduce the environmental footprint of the crop.

2. SUMMARY

2.1. Background

The development of improved oat varieties that meet the needs of different end-users has been the focus of OatLINK, a five year Defra and Rerad funded Sustainable Arable LINK project. The overall objective has been to incorporate important traits underlying sustainable development of the oat crop through combining 'conventional' phenotypic selection with molecular marker technologies. Although conventional and organic farmers, millers and poultry producers have different aims, there is also much in common in terms of the need for the development of oat varieties economic competitiveness, good agronomic and disease characteristics and sharing of molecular markers. OatLINK has demonstrated the added value from bringing together the different components of the oat production chain and the various end-users of oat and oat products within a single project. The industrial partners have been directly involved in the analysis and testing of selection lines and varieties developed within the project and providing important information that has been fed back into the breeding programme. The focus within OatLINK has been on the traits that impact on the agronomic performance of oats but also on those specific traits that are of particular importance to the end-users. For the milling industry the emphasis has been on traits associated with milling quality but also on β-glucan, the grain constituent associated with the health benefits of oats. For the poultry sector the emphasis has been on the development of high oil, naked oats that provide a high energy feed. Incorporating these characteristics into good agronomic backgrounds to produce oats with good yield, disease resistance and standing ability is essential to ensure their commercial acceptability. The following is a summary of some of the results from this project.

2.2. Development and application of molecular technologies

The primary objective of OatLINK has been to integrate molecular marker technology with conventional selection and demonstrate the value of molecular based approaches by applying markers to specific traits. One of many major achievements has been the development of the first winter oat genetic linkage map with 589 loci mapped onto 35

linkage groups and correspondence of linkage groups with those of the oat reference map (Kanota x Ogle) established.

Over 340 new polymorphic molecular markers developed in this project are applicable to oat lines in the breeding programme. In addition, new oat microsatellite markers developed elsewhere have been evaluated on a range of lines from the IBERS breeding programme. Marker development has continued both at Aberystwyth and elsewhere increasing the number of markers available. This includes the DArT consortium, of which we were active participants, that has developed over 2000 polymorphic DArT markers for oats; this has radically changed the possibilities for QTL mapping and identification of genes underlying traits of interest. DArT analysis of a number of populations from studies across the world has now resulted in the development of consensus genetic maps of oats for the first time and enables meta-QTL analysis. New collaborative opportunities for developing further markers are currently being actively explored.

2.3. Development of oats for human consumption

There is increasing demand across the UK for safe, high quality food that is produced sustainably and economically and oats and oat products are increasingly important in meeting that demand. Development of oats with the appropriate milling quality was a primary objective of the project and considerable effort was put into ensuring that methods of quantifying milling quantity were consistent between the breeding programmes and the different millers. There was also an opportunity to conduct some large scale pilot milling of advanced lines/varieties enabling 25t lots to be milled in 'commercial' mills. This has shown that the small scale techniques are good indicators of milling potential and correlate well with results from most commercial mills. One of the major successes of OatLINK has been bringing the whole production chain together from breeders to seed producers and then processors to ensure that varieties produced meet the end-users requirements.

The health benefits of oats are predominantly due to β-glucan, the major endospermic cell wall polysaccharide in the grain, thus a major target of the breeding programme at IBERS has been to develop UK adapted oat varieties with higher levels of β-glucan that meet the millers requirements in terms of yield and milling quality. As it is a

difficult and expensive trait to select, it is also an excellent trait for application of molecular technologies; within OatLINK significant progress has been made in the development of molecular markers and rapid screening methods for β-glucan content. These markers are now being tested in the oat breeding programme.

2.4. Improved agronomic performance

In conjunction with ADAS, field trials have been undertaken to dissect the components of lodging resistance, a key attribute for the growers and millers who increasingly demand grain produced without use of plant growth regulators. Straw height is not the only factor to prevent lodging, although shorter straw does usually reduce the risk of lodging, straw stiffness and rooting also play an important role. The difference in panicle structure also affects lodging as can be seen in Figure I below. Wheat has an ear area of approximately 15 cm^2. Currently plant growth regulators are routinely used, however the CEL RL data has demonstrated that some of the shorter, newer varieties such as Tardis, Brochan, Mascani and Balado suffer a yield penalty as a consequence. As some millers market oats as being PGR free, dwarf oats or lodging-resistant oats fit very well into their requirements.

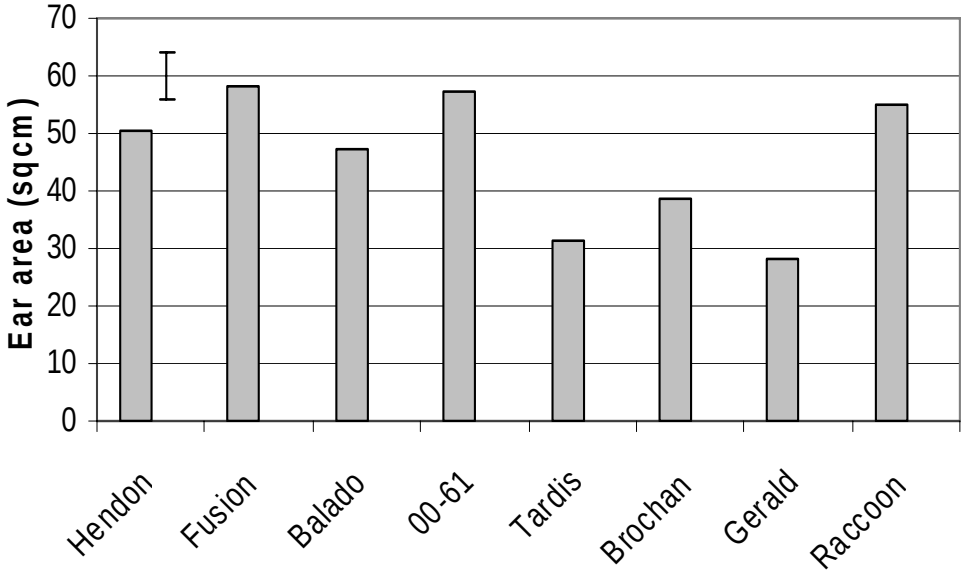


Figure I. Effect of oat variety on panicle area

2.5. Development of oats for sustainable livestock production

Oats have tremendous potential as a livestock feed. At the start of OatLINK the poultry industry expressed the need for oats which are economically competitive and/or have high oil/energy without jeopardising protein and amino-acid content. Both naked and husked oats have been studied with the focus on increasing the oil content and hence the energy (ME) value. Development of molecular markers for oil has been a major target. Over the course of the project approximately 500 crosses have been carried out to develop naked oats for animal feed. The development of a high oil oat with good agronomic attributes has been a key challenge. The variety Racoon, added to the National List in 2006, has an oil content of 12-14% but is tall (>140cm). The focus has therefore been on the development of a high yielding, high oil, short or dwarf naked oat.

Rapid assessment of breeding lines for oil and total N content has been possible by the application of NIR technology. Good correlations have been developed between laboratory and NIR analysis for oil content (Figure II) that is now being deployed within the programme to rapidly phenotype selection lines and potential crosses within the breeding programme.

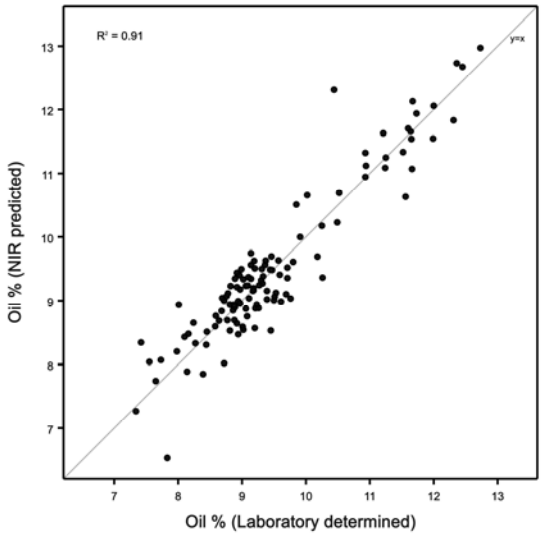


Figure II. Relationship between oil (%) as predicted from laboratory and NIR analysis.

Experiments carried out in conjunction with Roslin have confirmed that increasing the oil content increases the energy (ME) value of naked oats. Roslin ME bioassays have been carried out on broilers and ME values quantified in a range of genetic material throughout the project. Several large scale poultry feeding trials have been conducted and the impact of incorporating the high oil line Racoon in the feed ration on bird health, productivity and on lipid stability quantified. Energy and oil values assigned to Racoon (15.3 MJ/kg and 12.2%) respectively were confirmed by the study and bird performance was maintained when Racoon was included in the ration at the different inclusion rates.

2.6. Oats for organic production

A key objective of OatLINK was to quantify the agronomic performance of current and potential oat varieties in organic systems when grown as the first or second cereal. Experiments conducted by Organic Research Centre - Elm Farm confirmed the potential of oats as a cereal particularly suited to organic production with yields of >7t/ha achieved for some varieties. One of the main conclusions from the research in the 06/07 seasons was the difference in disease levels and yield between varieties in their position in the rotation as first and second cereals. The winter oat varieties Tardis and Mascani yielded 7.3t/ha and 7.1t/ha respectively as the first cereal and 6.6t/ha and 7.0t/ha as the second cereal and yielded more than Brochan and Gerald. Tardis and Gerald were notably less productive as second cereals. However, the reduction between the average yields of the naked oats in the first and second cereal experiments was larger at 19% than in the husked experiments (3%).

2.7. Life Cycle Assessment (LCA)

The environmental impact of crop and animal production is increasingly important. In this project we have carried out an LCA of the oat production chain and shown that oats have relatively less environmental impact compared to other arable feed crops per unit delivered feed energy and therefore offer a more sustainable feed source for the poultry industry. An LCA of the porridge oat production chain was completed to investigate the impact of plant varietal development on the sustainability of UK porridge production. Varietal improvements associated with the “downstream” cooking

process were most influential across the majority of impact categories investigated. However they also showed improvements in yield and nitrogen use efficiency correlated to a substantial reduction in the environmental footprint of porridge oats indicating potential targets for future research on oats.

2.8. Conclusions

Oats have important attributes that make them an increasingly relevant component of sustainable arable rotations. They are a valuable break crop, a lower input crop than wheat, perform well in marginal areas, are competitive against a range of weed species and are a high value feed that grows well in grassland rotations. There is increasing realisation that the grain composition of oats offers considerable health benefits as consumers shift towards healthier diets as well as opportunities for opportunity for industrial applications. Oats are also an environmentally sustainable animal feed across the livestock sector. A major focus for the future will therefore be the development and application of modern plant breeding tools for the identification of specific genes and molecular markers associated with key traits and exploitation of the wide range of oat genetic resources that are available to plant breeders.

3. TECHNICAL DETAIL

3.1. Project objectives

Aim: to incorporate important traits underlying sustainable development of the oat crop through combining 'conventional' phenotypic selection with molecular marker technologies.

Specific Objectives were:

- To develop new molecular markers, UK mapping populations and contrasting bulk segregants for use in marker-assisted selection (MAS) of important traits
- To identify, incorporate, select and evaluate important traits for sustainable production and human consumption
- To identify, incorporate, select and evaluate important traits for sustainable production and premium livestock feed
- To identify, incorporate, select and evaluate important traits for organic production

3.2. Background

Previous IGER (now IBERS) oat varieties have been produced using 'conventional' breeding methods. They have incorporated genes from cultivated oats worldwide, identifying and overcoming adverse associated traits. In many cases however, it has proved impossible to identify the underlying genes and to unravel the interactions between genes. For example, Gerald is a "one-in-a-million" segregant with shorter straw than either of its parents but the genetic basis of shortness is unknown. Even where important traits such as the naked and dwarf characters are controlled by major genes, considerable work involving several cycles of hybridisation and selection has been necessary in order to overcome associated poor agronomic traits. In the case of dwarf oats, the initial genotypes were late maturing, had incomplete extrusion of ears from flag leaves, largely have small grains, poor kernel content and poor resistance to mildew. Recent work on husked oats has centred on improving the yield, resistance to lodging, disease resistance and milling quality. Recent work on oats for livestock has focused on combining the naked and dwarf characters, and increasing metabolisable energy content by incorporating high levels of oil content from

unadapted spring husked germplasm from Iowa and incorporating the low husk lignin trait.

Molecular markers increase the speed and precision of plant breeding. This can save several years in breeding schemes and reduce ultimately the requirement for expensive phenotypic evaluation, especially for disease and quality traits, thereby substantially reducing the cost of cultivar development. Molecular markers are ideal for traits not easy to assess visually or for understanding the genetic control of traits so that it is possible to determine whether the apparent linkage between traits involves genetic linkage. Breeding for quality or processing traits can proceed with molecular markers without the need to grow out large quantities of seed or product for processing. In addition to these and other examples, molecular markers, maps and QTL (Quantitative Trait Locus) analysis identify and tag the genes involved in complex multi-gene traits. This permits breeders to construct specific genotypes for advancement of quantitative traits, a process that was very difficult or impossible to do without markers.

In this project the objective was to develop molecular marker technologies and combine them with the development of high throughput phenotyping to improve the economic competitiveness, resistance to diseases and quality of oats, to enhance end-use characteristics to meet consumer requirements, with particular focus on meeting the needs of the milling and poultry industries and to enhance economic prosperity. In this respect, oats deliver major economic and environmental benefits but relatively little work has as yet been undertaken on the development of rapid and precise methods of breeding based on molecular markers.

3.3. Materials and methods

OatLINK was a large multidisciplinary project that incorporated molecular biology, plant breeding, agronomy as well as a series of animal production trials carried out on poultry. Materials and methods relating to each project area are summarised and, where appropriate, reference is made to standard protocols or published papers where the methods are more fully described.

3.3.1. Marker development

From a collection of 9792 EST sequences, after clustering and *in silico* mining of di- to hexa- nucleotide repeats, 184 SSR primer sets were designed and tested on eight oat lines including the winter oat mapping family parents Buffalo and Tardis. Fifty-two SSRs that could reveal polymorphism among the eight oat lines tested were selected among the 128 SSRs that produced amplified products. Fluorescent labeled primers for running these SSRs on the ABI 3700 system were synthesized, and these markers were further validated for their robustness and to estimate the size of alleles observed in the eight oat lines. In addition, SSR markers developed for Lolium, Barley and Wheat were tested for use with oats using published methods. Transposon display markers were developed following published methods (Leigh et al 2003, Takagi et al 2003). Displays were established using both transposon and retrotransposon 'anchors'. All combinations of adapters were used for one transposon sub-family and polymorphic bands recovered for conversion to simple PCR markers. DArT markers were developed as part of an international consortium as described by Tinker et al 2009.

To construct a genetic linkage map and to identify QTL, a mapping population was developed by crossing two elite cultivars from the Aberystwyth breeding programme: the tall cultivar Tardis and the dwarf cultivar Buffalo. This population was designed to dissect components of yield particularly in relationship to the effect of the dwarfing gene *Dw6*. A genetic map was produced for this population using Mapmaker (Lander and Botstein 1989). The mapping population of 188 lines along with the parents were grown out in a replicated field trial in two field seasons, 2006/07 and 2007/08. Winter hardiness, flag leaf emergence, flowering time, height, yield components and disease resistance were scored in field. Quality analyses including kernel content and hectolitre weight were conducted using harvested grain. QTL analysis was conducted using PlabQTL (Utz and Melchinger, 1996).

In addition specific crosses were developed to identify markers associated with seed oil content, β-glucan content, and husk lignin via bulk segregant analysis (Michelmore et al, 1991).

3.3.2. Crossing

In work packages 1-4 crosses were made each year using glasshouse-grown plants. Crossing was completed according to a standard IBERS protocol. Healthy plants were selected, the female plant was emasculated and an inflorescence from a pollen donor cut and inserted in a test tube, then bagged to prevent any contamination and seed collected 4-5 weeks after crossing. Leaf material from both parents in the cross was harvested and stored to allow DNA extraction and then development and validation of molecular markers. Crossing was completed in each year of the project with crosses designed to meet the specific objective of each work package. For winter oats, the F1 seed was grown in a glasshouse for another generation to produce F2 seed. Some selected lines were selected for Advanced Pedigree Selection (APS), which was used to produce two generations in one growing season. The remaining F1s were routinely sown in the glasshouse in January, harvested in July and August and then sown in the field in October. For spring oats the crosses were made at a similar time however they were all harvested and sown in the same autumn, harvested in December or January ready to sow out as F2s in March. For some of the work package 1 objectives the F2 plants were sown in pots in December or January in the glasshouse and phenotyped for various characteristics, such as growth habit, ear emergence, height of main tiller, maturation and then tiller numbers. These were then harvested, cleaned and sown in the field for further phenotypic classification and bulking up of seed for yield trials and other quality parameters.

3.3.3. Agronomic evaluation of oats

In each of the 5 years of OatLINK a range of genetic material was produced in the field and glasshouse and analysed according to standard protocols. To meet the objectives of the different work packages material was grown and assessed in the field for agronomic performance and then harvested and re-sown as part of the ongoing breeding programme or as yield trials which were then subsequently used for quality analysis or as an animal feedstuff.

Earlier generation material (F3 and F4) was assessed mainly in Aberystwyth in unreplicated breeding nurseries. Replicated yield trials were carried out each year at different locations in the UK on winter and spring oats on material at a later stage of the breeding programme (usually F5 and F6 generations) using a balanced lattice

square design or complete randomised blocks. Specific disease nurseries for crown rust, mildew and OMV (oat mosaic virus) were all tested on the later generations. For all field based material the same protocols were followed. At IBERS, oats were grown in a grassland based rotation with oats grown one year in eight. A small area of approximately 0.2ha was used continually for the Soil-borne Oat Mosaic Virus (SBOMV) nursery. Husbandry methods were common to all field operations and are described in the following sections.

3.3.3.1. Ploughing, fertiliser and cultivations

Land to be used for winter oat experiments was sprayed with either gramoxone or glyphosate in July of each harvest year to kill grass and weeds before ploughing, for spring oats this was done in February. After ploughing all areas were soil sampled for pH, P, K and trace elements inducing Mn. For healthy oat crops a pH of 6.0 is required and if the pH was low lime was added at an appropriate rate. Fertiliser was applied at 375 kg/ha (0:24:24) to the seed bed for autumn sowing and 250 kg/ha (Sulphur Cut 22:4:14) to the seed bed for spring sowing. The seed bed was prepared with a power harrow to produce a fine enough tilth. Plots were sown with an Oyjord seed drill and F2 generation drills with a Monosem drill. Breeding nurseries and disease nurseries were sown with a Hege 90 drill. All layouts and field dimensions of nurseries and trials were designed to fit a 12m sprayer. All winter oat trials and breeding nurseries received a top dressing of fertiliser in the region of 70-90 units of N dependent on site. Field trials on dwarf oats received an additional 20 units of N. The yield trials were treated with fungicide to control crown rust and mildew. Plant growth regulators were not used except in the HGCA CEL trials. In general yield trials followed the guidelines given for the HGCA CEL and BSPB NL trials.

For the nurseries similar practices were adopted for land preparation, rotation and using appropriate chemicals for weed control. The oat breeding nurseries were left untreated and lines were scored for incidence of important diseases such as crown rust and mildew as well as other agronomic traits.

3.3.3.2. Disease evaluation

Disease nurseries were grown to screen genetic material for resistance to mildew and crown rust. An Oat Mosaic Virus (OMV) nursery was used to identify material which was resistant to OMV, a soil-borne virus. Higher levels of disease were attainable in disease nurseries by growing susceptible 'spreader' varieties, maintaining higher N

levels and controlling unwanted diseases, e.g. mildew was controlled in the crown rust nursery.

Genetic material screened in the nurseries included all IBERS entries in yield trials, new genetic material that may have had resistance and early generation breeding material where resistance had been incorporated. The mildew nurseries relied on natural infection whereas the crown rust nurseries were inoculated with known races. Seed of trial entries was measured into packets. 'Spreader' drills were sown that were 30m long and 1.65m apart. The 'spreader' drills were seed of a mixture of susceptible varieties to the particular disease to be encouraged in that nursery. The test material was sown in hill plots (clumps) 30cm apart and 30cm from spreader drills. The autumn and spring sown disease nurseries were treated in the same way as the trials with regard to herbicide and insecticide applications but also received a later application to control *Lema melinopa*. Disease nurseries received more N fertiliser than trials and breeding nurseries in several small applications to encourage the development of diseases.

3.3.3.3. Inoculation

The crown rust nursery was inoculated with spores of the required race which has been increased on a susceptible variety in a spore-proof glasshouse. Spores were suspended in water with a wetting agent to ensure an even suspension. An automatic syringe and needle were used to inject the suspension into the unfurled leaves of plants at 0.5m intervals along the spreader drills. When the leaf unfurled the infection site can be seen and in time spores were produced which will infect the spreader drills and the hill plots. Several primary inoculations were carried out at 3-4 day intervals to ensure conditions were right for secondary infection to occur. The plots were scored three times for % leaf area infected using the "Clive James" key.

3.3.3.4. Agronomic Traits

Various traits (winter hardiness, growth habit, ear emergence, height, lodging, maturity) were scored in the field following HGCA CEL guidelines.

3.3.3.5. Methods relating to specific objectives

Objective 2 – The material used for commercial evaluation was obtained from yield trials which were grown broadly in line with HGCA CEL trials protocols. Plots were sampled using a Sampo plot combine which enabled a bulk sample and a small sub-

sample of grain to be harvested from field plots for quality analysis. IBERS quality analysis was completed on the subsample and joint ring testing using laboratories of industry partners completed on the larger sample. For industry based samples ~1kg of oats was required. Material from agronomic trials carried out by ADAS Rosemaund and ORC Elm Farm (Objective 4) were also sent for industrial assessment. Protocols for ADAS field trials and lodging analysis are described in Appendix F.

Objective 3- The husbandry details are already described for field production of oat varieties to provide samples for chemical analysis, NIR determination of oil and protein and as feedstuff for poultry. The methods used for the poultry feeding trials at Roslin are detailed in MacLeod et al (2008).

Objective 4- Crop husbandry protocols for agronomic trials carried out by ORC Elm farm are described in Appendix G.

Life Cycle Assessment- approaches used in life cycle assessment are summarised in McDevitt, J E. (2009) and McDevitt, J E and Mila i Canals, L., (2009).

3.4. Results

3.4.1. Objective 1. To develop new molecular markers, UK mapping populations and contrasting bulk segregants for use in marker-assisted selection (MAS) of important traits

The OatLINK project has carried out extensive activities including development of new molecular markers and phenotyping methodologies, construction of linkage maps and marker assisted selection. Major achievements we have obtained are detailed below.

Over 340 new polymorphic molecular markers were developed in this project applicable to oat lines in the breeding programme, exceeding the number predicted at the start of the project. This exceeds the milestones for objective 1.1 (50). In addition, new oat microsatellite markers developed elsewhere (150) have been evaluated on a range of lines from the IBERS breeding programme.

We are part of an international consortium that has developed DArT markers in association with DArT PL based in Canberra, Australia. This has resulted in approximately 2000 DArT markers that show polymorphism in a panel of 200 cultivated oat varieties, over 300 of which display polymorphism between our winter oat mapping family parents, Buffalo and Tardis. The consortium's results have been submitted for publication in BMC Genomics.

The first winter oat genetic linkage map was developed, with 589 loci mapped onto 35 linkage groups (Figure 1) and correspondence of linkage groups with those of the reference map (Kanota x Ogle) was established.

Two years of replicated yield and quality trials have been conducted with the Buffalo x Tardis mapping family. Traits scored included winter hardiness, flag leaf emergence, flowering, height, panicle extrusion, yield components, disease resistance, kernel content, specific weight determined winter 2007.These have been analysed to dissect out the genetics of yield and its components and identify yield limiting factors in dwarf oats. For example, grain number per panicle showed a close correlation with yield in dwarf oats (but not in conventional height oats) with a number of progeny performing far better than the dwarf parent, Buffalo. This study not only identifies what the problems are in breeding high yielding dwarf oats but offers solutions to this problem. QTL for a large number of traits have been identified using this population.

Detailed data analysis has been conducted combining all the data for the Buffalo x Tardis mapping family from the two years of replicated field phenotyping and from earlier generations grown unreplicated in field and greenhouse. Good correlations between values obtained for individual traits between different years of study have been obtained. Consistently, this has revealed that for many traits segregation was bimodal and that the mapping population could be divided into two sub-populations based on the presence or absence of the dwarfing gene (Figure 2). Transgressive segregation was apparent for most traits. Dwarf plants tended to be later flowering, have fewer tillers, smaller grain, fewer grains per panicle, poor panicle extrusion and lower yielding (Figure 3). However dwarf plants with relatively early flowering, good panicle extrusion and good grain yield have been identified in the mapping population indicating that at least some of these linkages can be broken. QTL analysis conducted both on the complete population and separately on the sub-populations has revealed a large number of regions of the genome controlling these traits but also that certain

regions of the genome share QTL for a large number of traits. These regions of the genome are now being studied in more detail. At present over 100 traits have been assessed with a total of over 200 QTL identified. Many of these QTL were identified in more than one environment indicating their robustness.

Phenotyping of populations was developed for specific traits was completed and used for bulk segregant analysis. Candidate markers associated with both high and low β-glucan content have been identified using the 96-21Cn19 × SA99572 key trait population. In addition we have developed a high throughput phenotyping screen for β-glucan which will greatly assist in future validation of markers associated with β-glucan as well as being directly applicable to the breeding programme. This has been a major limitation in the past. Markers associated with β-glucan are being used to develop lines with high β-glucan content in both UK adapted winter and spring oat backgrounds. For example, β-glucan measurements for 22 F5 plants derived from one of the highest β-glucan 96-21Cn19 × SA99572 F2 lines identified a single very high (7%) β-glucan content plant, consistent with continuing segregation at the predicted major and/or minor gene loci.

Novel sources of high β-glucan content are also being assessed using both molecular markers and conventional phenotyping. This includes collaboration with Brian Rossnagel of the University of Saskatchewan using a spring oat mapping family between 2 sources of high β-glucan unrelated to SA99572. Analysis of β-glucan content of this population over a wide range of environments will enable the development of robust markers associated with this trait and provide understanding of the role of environment and genotype in controlling the expression of this trait. This population has been mapped using DArT markers enabling clear comparisons with results from Aberystwyth. A wide range of β-glucan content has been obtained in this population when grown both in Canada and in Aberystwyth.

Following screening of bulks contrasting for husk lignin content with over 200 polymorphic markers, candidate markers associated with the low lignin trait have been identified and are currently being verified in a wide range of genetic backgrounds and using crosses designed specifically for this trait.

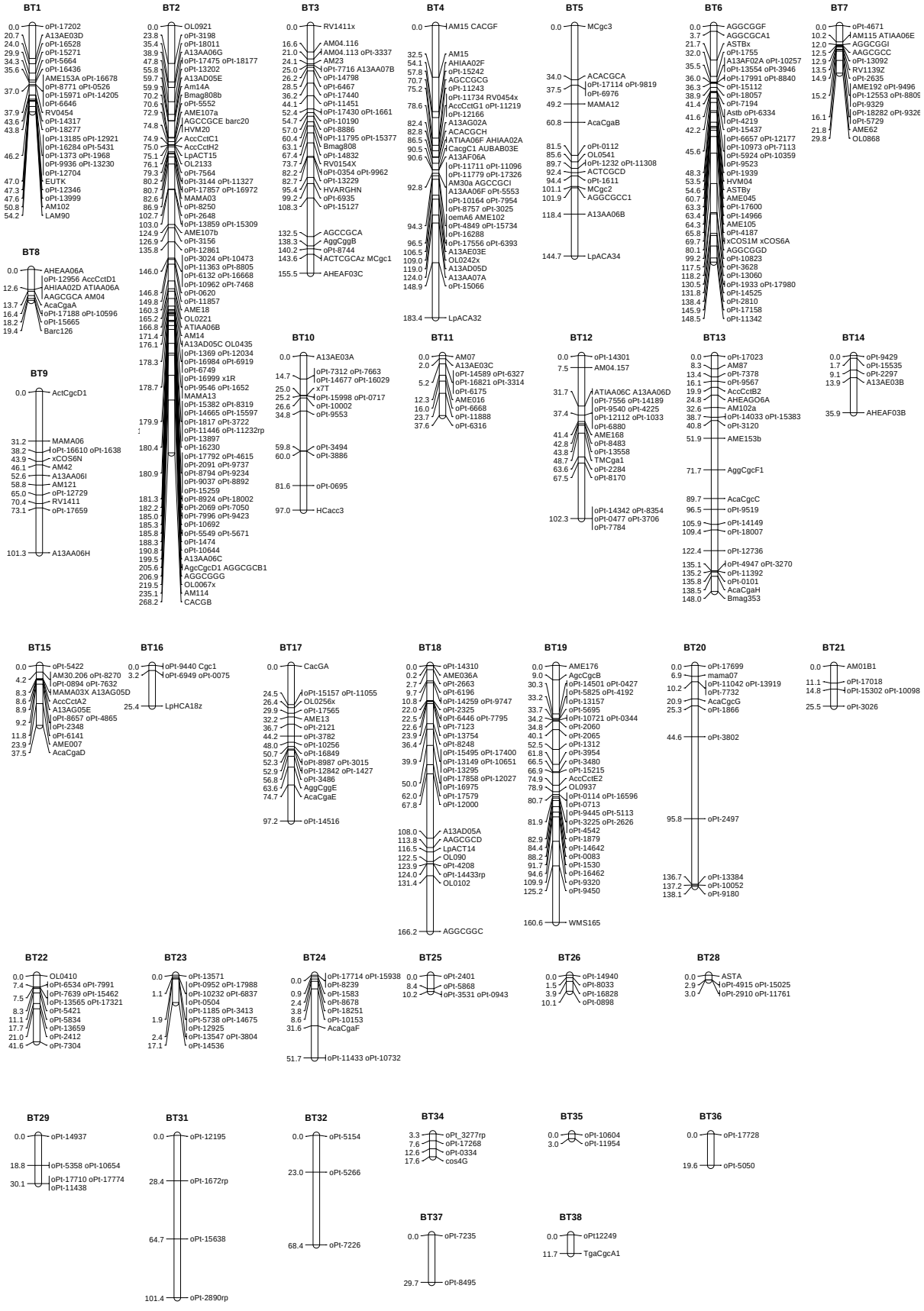


Figure 1. Genetic Linkage map of Buffalo x Tardis mapping population

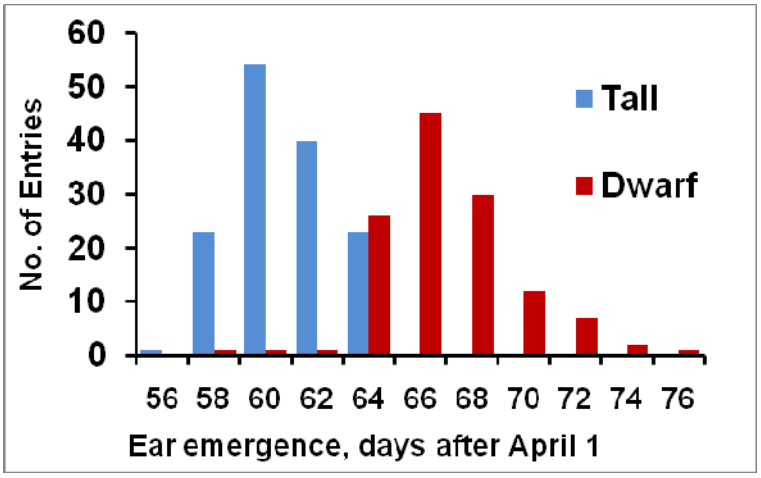


Figure 2. Range in flowering time in Buffalo x Tardis mapping population 2008 indicating the 2 sub-populations of dwarf and tall plants

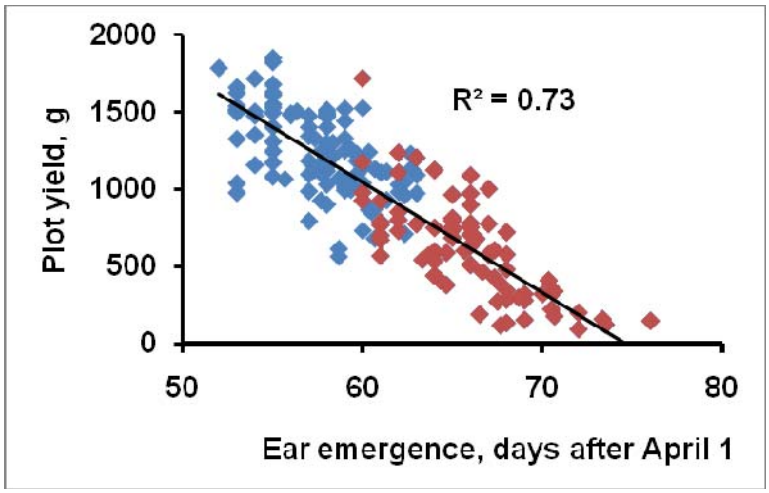


Figure 3. Relationship between flowering time and plot yield in Buffalo x Tardis mapping population 2008

Markers associated with the dwarfing gene (*dw6*), components of height and yield, β-glucan content, oil content and naked character were identified. Marker-trait associations for dwarfing gene (*dw6*), components of height, oil content and naked character were verified in a wider range of genetic backgrounds.

Selection of parents in the breeding programme was based on allele composition at key loci associated with traits of interest.

While a major objective of OatLINK was the development of molecular markers associated with key traits, the ultimate aim is the integration of these markers into the oat breeding programme and their application, with phenotypic analysis, for the selection of appropriate parental material or progeny for further development. As detailed above and in subsequent papers this has been successfully achieved for a number of traits. The value of marker-assisted selection (MAS) is dependent upon the effectiveness of the markers and the robustness of marker-trait associations. This will be explored further post OatLINK.

Following discussions within the IBERS oat breeding team we have already made some changes to previous practices to allow better integration of molecular marker and conventional approaches (for example, archiving material for DNA extraction from all crosses), which will be followed in 2009 by further improvements (for example in nursery lay-out) which will increase our potential molecular throughput in preparation for large-scale application of MAS.

3.4.2. Objective 2. Identify, incorporate, select and evaluate important traits for sustainable production and human consumption

The stated aims of the milling industry in supporting OatLINK were for:

- Oats which are economically competitive in order to meet the needs of growers, millers and consumers.
- Oats which have high milling quality in terms of high groat (kernel) content and low screenings.
- Oats which deliver PGR- and pesticide residue-free grain for human consumption as lodging resistant husked oats are needed in order to protect oats' image and markets.

OatLINK has worked towards these objectives by using conventional phenotypic selection together with the development of molecular markers (objective 1 above) that allow precise and rapid selection for important traits. In addition, by working together, breeders and millers aim to align their assessment of milling quality in order to maximise commercial acceptability.

During the project (2004 to 2009) over 600 successful crosses for winter oats and 240 for spring oats have been completed to produce oats which are environmentally sustainable and meet the requirements of the milling industry. These are, essentially, to produce oats which are high yielding with good resistance to disease (crown rust and mildew), are lodging resistant and have good grain quality in terms of kernel content and freedom from grain blackening. In each year of the project replicated yield trials at various locations of between 125 and 150 winter husked oats and 50 to 100 spring husked oats have been carried out. Each entry to trial was replicated 3 times and these were the sources of material used for quality analysis.

3.4.2.1. Quality testing

At the beginning of OatLINK a milling subcommittee was established comprising plant breeders, crop developers and millers with the aim of identifying the key traits for genetic improvement. Kernel content was identified as an important trait by the milling industry and therefore a major focus of OatLINK has been the selection of improved genetic material with high kernel content and the development and validation, with industry, of the methodologies to test for this important trait. A ring test for kernel content using IBERS, AFBI, Quaker and Grampian Oats at Banff has confirmed the close relationship between results at IBERS and AFBI, N. Ireland (who conduct the quality testing for the CEL Recommend List). Kernel content measurements have also been completed on the Buffalo x Tardis mapping population. Advanced selection lines and varieties have been analysed for a range of quality attributes according to the milling industry specifications. These can be briefly summarised as:

- Specific or bushel weight (industry requirement of 50)
- Kernel content (industry requirement of >70%)
- Screening losses (more than 6g in 100g)
- Discoloured Grains (maximum score of 30 per 100g)
- %Free groats (maximum of 5% per 100g)

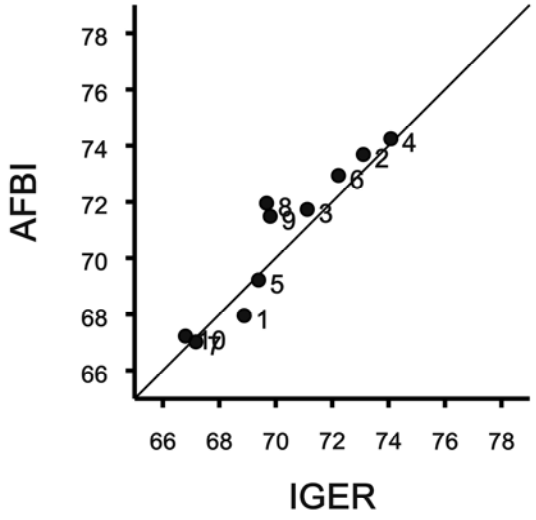
Ring testing by using different mills to analyse the same sample has shown the reliability of the IBERS quality assessment protocol (Figure 4). Results have highlighted certain issues relating to bushel weight which is an indication of how many grains can fit into a given volume. Obviously large grains will not pack as tightly as small grains hence some large grained varieties such as Brochan are shown to have good kernel content (KC, ~77%) but poor specific weight (~50). There was some slight difference in the absolute figures for KC but the ranking of varieties remains constant. The data also indicated the IGER protocol was slightly underestimating the kernel content compared to Quaker Oats. There was a very strong agreement between KC determined by IGER and AFBI who conduct the official quality analysis for CEL RL trials.

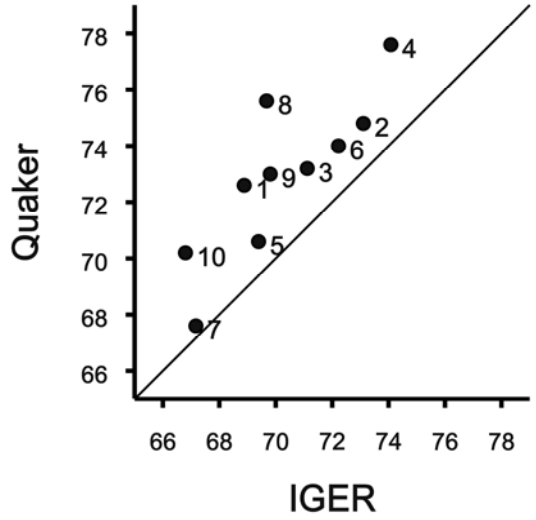
Screening losses are a major concern to the milling industry and this has necessitated screening potential lines using a series of slotted sieves to reject any where there are many small grains. Generally small grains are obtained from tertiary florets and visual selection is used to select against this trait. Occasionally, genetic material which has large tertiary grains has been utilised, in an attempt to improve quality however this has largely been unsuccessful.

Free groats are also an issue however this can be countered in some cases by attention to detail whilst combining the crop ensuring the correct concave setting and drum speed are used. An association between some sources of large grain size/ kernel content and incidence of free groats has been identified.

Kernel content (%)

Code	*Selection*
1	**Gerald**
2	**Millennium**
3	**Dalguise**
4	**Mascani**
5	**Tardis**
6	**96-21Cn7 (Brochan)**
7	**98-28Cn3/1**
8	**00-186ACn13**
9	**Kinross**
10	**Kinnell**





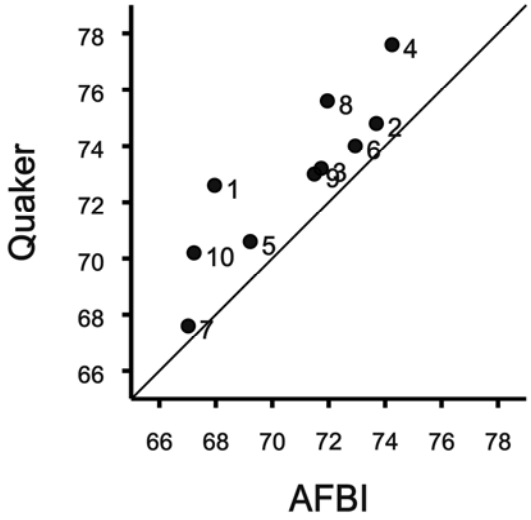


Figure 4. Comparison of test sites for kernel content determination

β -glucan content was identified by the milling industry as a "nice to have" trait but not at the expense of the previously mentioned quality issues of yield, kernel content and low screenings etc. Significant progress has been made in the development of markers for oat β-glucan content and in the development of rapid screening methods for this important trait which will enable selection of this trait in the future breeding programmes. The winter oat population 02-177ACnIII has been grown in the field for two years and selected progeny from this population have been identified using markers and utilised in the winter oat crossing programme to obtain UK adapted winter oats which have higher levels of β-glucan. We have also utilised material from collaborators in North America and grown some of their material in UK conditions. This has a wider range of β-glucan and is currently being assessed for a second season. Most of the sources of higher β-glucan have come from North America and these have proved difficult to transfer into UK adapted winter oats. Figure 5A shows the range in β-glucan content compared to current controls and within progeny of selected lines from the spring oat programme. Figure 5B shows yield in relation to β-glucan content. The highest yielding lines are the UK spring oat controls Firth and Ascot. The North American line Hifi, a high β-glucan control, has poor yield. There is a wide range of performance of progeny from lines with potentially high β-glucan content. It is obvious that there is a trade-off between high β-glucan content and yield and decisions on the development of this material will probably depend upon the relative value of β-glucan to the end user. Currently most farmers will be driven to grow highest yielding varieties as they are paid on yield/ha basis.

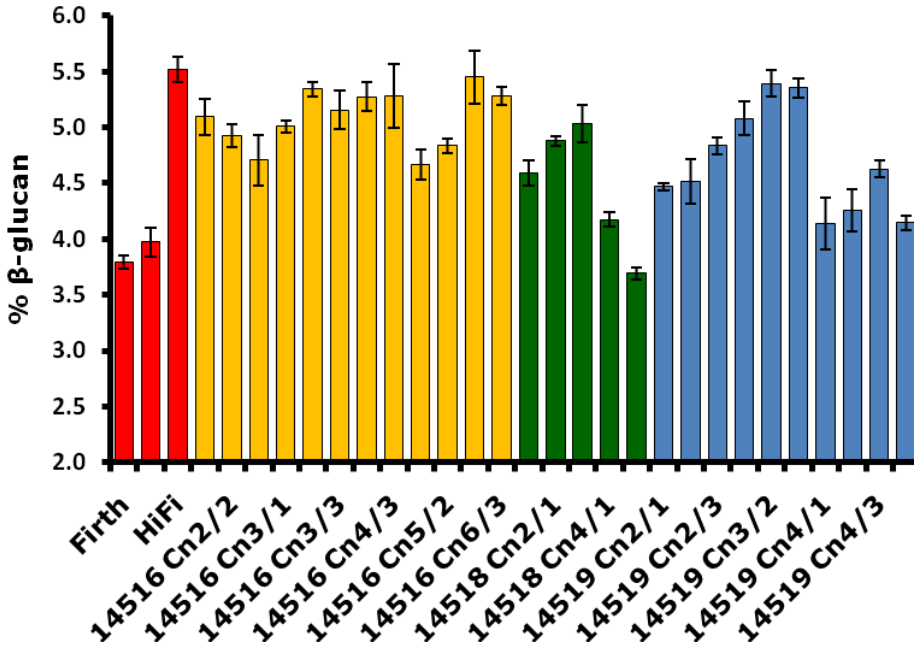


Figure 5 A. β-glucan content of selected spring oats from Morfa Mawr, Ceredigion (2008 harvest).

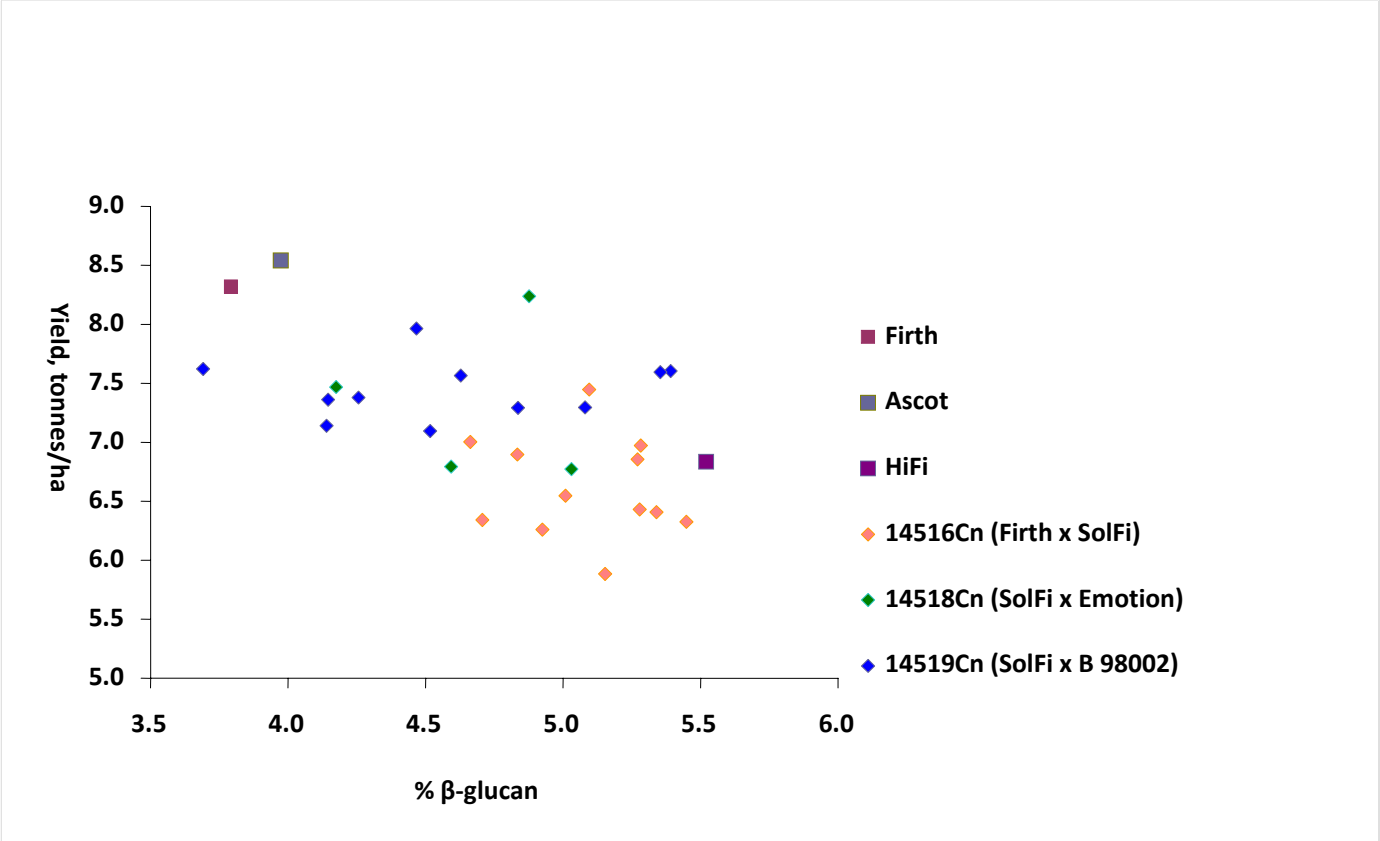


Figure 5 B. Yield t/ha in comparison with β-glucan content from Morfa Mawr, Ceredigion (2008 harvest).

3.4.2.2. Variety development

Agronomic evaluation is essential to identify potential varieties for entry into official trials (Figure 6). During the course of the project and in conjunction with Senova, we have added to the CEL Recommend List the winter oats Tardis and Brochan (see Appendix C for full details). The former has high yield and good disease resistance with grain quality on a par with Gerald which is the industry standard. Brochan has very high kernel content (77%) and yields 100% (105% no PGR) are slightly lower than Tardis. The dwarf oat Balado has been advanced to CEL RL testing and a decision on its entry is expected in December 2009. Further promising lines are in the early stages of development which have been designed to have higher levels of β-glucan incorporated into a UK adapted background. This project has seen a greater effort placed in breeding spring oats and has led to the selection of two spring oat varieties for advancement to NL trials in 2009.

Figure 6 Evaluation for agronomic performance of winter oat selection lines

3.4.2.3. Pilot milling of varieties

The **milling industry**, in supporting this project emphasised the need for oats which are:

- Economically competitive
- Have high milling quality in terms of high groat (kernel) content and low screenings.
- Deliver PGR- and pesticide residue-free grain for human consumption. The identification of new lodging resistant husked oats is needed in order to protect oats' image and markets. The first dwarf winter oat Buffalo has not lodged in three years of NL and RL trials, but we need to be able to radically improve kernel content and mildew resistance.

One of the benefits of OatLINK has been the close interaction between breeders, marketeers and the milling industry in pulling through varieties which are suitable for the industry and to provide information on commercial acceptability. Large crops of new varieties have been pilot milled by commercial millers to provide information on commercial acceptability of new material and to provide feedback on the value of breeder evaluations of milling quality. Using Gerald, the current most widely grown winter oat, as a standard against which to assess new varieties ensures that only varieties which meet the millers' requirements are introduced. This has been successful in identifying the new varieties Mascani, Brochan and Tardis as being suitable for the milling industry.

3.4.2.4. Agronomic characteristics of varieties

The needs of **farmers** identified prior to the start of OatLINK were identified as:

- Consistent returns
- Low inputs
- Improved ease of management including harvestability.

One of the stated aims of the milling industry was the ease of growing and harvesting oats. Lodging resistance usually results in better quality oats as there is less grain blackening and less lodging also makes the crop easier to combine. In conjunction with ADAS, field trials were undertaken at Rosemaund to further dissect the components of lodging resistance. From another Defra project investigating lodging resistance in wheat, the lodgemeter (Figure 7) was used to measure the force needed to "lodge "the plots. This has previously been published by Berry et al (2003a,

2003b). Figure 8 is a schematic model of lodging for wheat identifying various factors involved in lodging. There are two types of lodging, stem and root lodging. Root lodging can be affected by soil conditions with wet soil making root lodging more common. For stem lodging, straw height is not the only criteria to prevent lodging although shorter straw does usually reduce the risk of lodging. Straw stiffness and rooting also play an important role. Measurements were made on oats to assess the similarities and/or differences in oats to the wheat model.

Figure 7 Lodgemeter being used in field trials

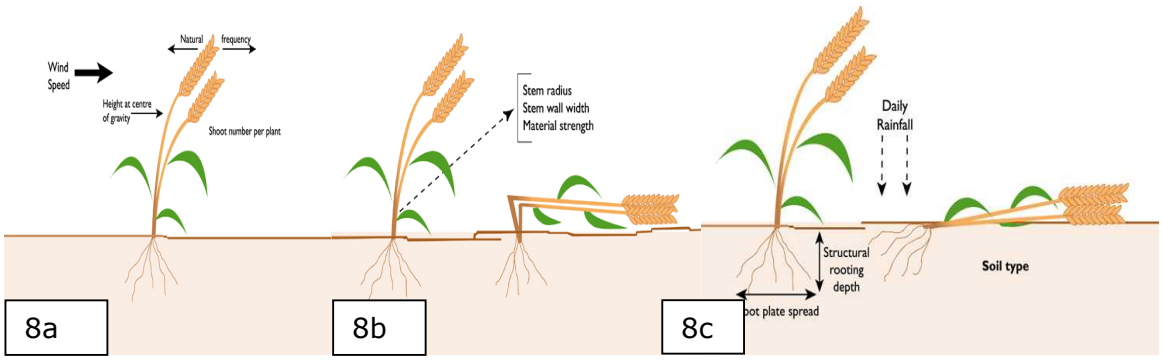


Figure 8a,b,c. Model describing factors involved in lodging a) shoot leverage force, b) stem strength and c) root from left to right

Two replicated split plot trials using four levels of N and 8 varieties were completed in successive years. Various characteristics were measured in oats and a considerable range of values were identified. In general terms the dwarf oats had a higher failure wind speed however Racoon, the tallest variety, had a similar failure speed indicating the complex nature of lodging resistance as this very tall variety is quite lodging resistant (Tables 1a, b). The stem strength per shoot is the force needed using the lodge meter to push the straw over. From Tables 1a and b it is clear that there are varietal differences with some of the dwarf lines e.g. Hendon and 00-61 having a low stem strength whilst Racoon has one of the highest stem strengths. Calculations (detailed in Berry et al 2003a and 2003b) were completed to give the final failure wind speed.

Table 1a shows the effect of two N fertiliser levels on lodging components in oats. In 2008 the higher N regime significantly increased the lodging risk mainly by producing taller weaker stemmed plants.

Individual differences were noted in the plant architecture and this influenced stem strength (Table 2). Some of the figures obtained appear to contradict information from field observations. Throughout its development Brochan had always been identified by short thick stiff straw and hardly any lodging had ever been observed in plots or yield trials. Demonstration plots of Brochan at Cereals 2006, Cereals 2007 and Cereals 2008 shows have always attracted farmers' attention and hand testing of straw strength had confirmed their opinion of the standing ability of Brochan. The data in Table 2 for 2007 appear to contradict this; however the results for 2008 are more in line with field observations.

Another important component of lodging resistance is the rooting ability of the crop as shown in Figure 8c. Table 3 shows statistically significant varietal differences in rooting depth and calculated anchorage. It does confirm the improved rooting capacity of oats, which are anecdotally harder to remove from plots than wheat plants. Other data from ADAS indicated that average root plate spread and depth for wheat is 41.3 and 44.3mm respectively (Berry et al 2003).

Table 1a. Shoots/m^2 and lodgemeter results showing stem strength and failure wind speed (2007).

Variety	Husked/ Naked	Height	Nitrogen	Field Crop Height (mm)	Stem strength per shoot (Nmm)	Failure wind speed (m/s)
Hendon	Naked	Dwarf	RB209	708.7	247.1	10.8
Fusion	Naked	Dwarf		855.3	275.6	10.1
Balado	Husked	Dwarf		845.3	338.4	11.3
00-61	Naked	Semi-dwarf		1030.7	394.3	10.7
Tardis	Husked	Conventional		1327.3	345.2	8.4
Brochan	Husked	Conventional		1293.3	310.1	8.1
Gerald	Husked	Conventional		1290.7	246.4	7.3
Racoon	Husked	Conventional		1536.0	494.7	9.11
Hendon	Naked	Dwarf	RB209 + 50 kgN/ha	738.7	191.6	9.3
Fusion	Naked	Dwarf		836.7	230.4	9.4
Balado	Husked	Dwarf		853.3	296.6	10.5
00-61	Naked	Semi-dwarf		960.7	400.3	11.3
Tardis	Husked	Conventional		1262.0	248.4	7.4
Brochan	Husked	Conventional		1263.3	280.0	7.9
Gerald	Husked	Conventional		1288.7	197.9	6.5
Racoon	Husked	Conventional		1500.7	305.1	7.3
		P-Value	Nitrogen	0.326 (NS)	(0.097 (NS)	0.044
			Variety	<0.001	<0.001	<0.001
			N x Variety	0.22 (NS)	0.008	0.041
		SED (28df)	Nitrogen	17.74	21.11	0.17
		SED (28df)	Variety	20.28	22.19	0.34
		SED (28df)	N x Variety	32.17	36.15	0.48

Table 1b. Lodgemeter results showing field crop height (mm), stem strength (Nmm) and failure wind speed (m/s) in 2008

Variety	Husked/ Naked	Height	Shoots/m^2	Stem strength per shoot (Nmm)	Failure wind speed (m/s)
Hendon	Naked	Dwarf	493.3	216	18.4
Fusion	Naked	Dwarf	408.3	262	16.0
Balado	Husked	Dwarf	389.8	325	21.3
00-61	Naked	Semi-dwarf	480.6	201	14.8
Tardis	Husked	Conventional	407.3	261	15.9
Brochan	Husked	Conventional	459.7	269	17.0
Gerald	Husked	Conventional	379.0	284	16.8
Racoon	Husked	Conventional	455.2	317	17.3
P-Value			**<0.01**	**<0.001**	**<0.001**
SED (28df)			**32.7**	**22.4**	**0.7**

Table 2. Cross year analysis of internode 1 stem strength components for 2007 and 2008.

Variety	Year	Internode 1				
		Length (mm)	Diameter (mm)	Wall width (mm)	Material strength (Mpa)	Overall strength (Nmm)
Hendon	2007	83.6	5.09	0.83	27.85	14.11
Fusion	2007	84.0	5.52	0.93	19.07	12.00
Balado	2007	88.4	5.88	0.95	17.86	12.75
00-61	2007	95.7	4.30	0.78	24.03	6.90
Tardis	2007	85.1	4.91	0.77	24.35	10.59
Brochan	2007	101.3	4.72	0.61	19.63	5.69
Gerald	2007	113.4	5.39	0.62	21.16	7.40
Racoon	2007	110.0	5.28	0.69	23.05	8.68
Hendon	2008	56.7	5.35	1.01	24.38	22.05
Fusion	2008	53.7	6.23	1.05	21.36	30.52
Balado	2008	62.0	6.30	1.05	23.21	29.62
00-61	2008	67.4	6.15	1.09	23.23	25.95
Tardis	2008	63.8	5.54	0.98	25.38	22.16
Brochan	2008	67.6	7.87	0.87	18.52	23.00
Gerald	2008	57.1	5.44	0.80	23.61	19.44
Racoon	2008	71.1	5.91	1.01	29.40	27.39
P-Value	**Year**	0.064(NS)	0.145(NS)	0.088(NS)	0.450(NS)	0.009
	Variety	<0.001	0.251(NS)	<0.001	0.043	0.005
	Year x Variety	0.028	0.081(NS)	0.120(NS)	0.534	0.089(NS)
SED (2df)	**Year**	8.67	0.413	0.067	1.621	1.416
SED (28df)	**Variety**	4.69	0.509	0.041	2.497	2.076
SED (28df)	**Year x Variety**	10.66	0.790	0.086	3.679	3.090

Table 3. Cross Year analysis of root plate spread, root plate depth and anchorage failure wind speed.

Variety	Year	Root plate spread (mm)	Root plate depth (mm)	Anchorage failure wind speed (m/s)
Hendon	2007	32.35	81.36	7.75
Fusion	2007	31.13	81.43	7.80
Balado	2007	30.82	79.07	7.74
00-61	2007	25.17	81.43	5.00
Tardis	2007	27.87	77.93	5.42
Brochan	2007	29.62	77.92	6.72
Gerald	2007	29.95	81.47	6.34
Racoon	2007	28.45	84.47	5.68
Hendon	2008	47.77	65.87	10.83
Fusion	2008	38.83	53.80	6.58
Balado	2008	40.67	62.67	8.69
00-61	2008	41.53	59.13	6.47
Tardis	2008	37.37	61.83	7.05
Brochan	2008	40.27	60.27	6.90
Gerald	2008	45.50	65.70	10.29
Racoon	2008	40.08	67.00	5.43
P-Value	Year	0.027	0.004	0.172
	Variety	0.017	0.352(NS)	<0.001
	Year x Variety	0.253(NS)	0.664(NS)	0.048
SED(2df)	Year	2.014	1.229	0.586
SED(28df)	Variety	1.966	3.562	0.779
SED(28df)	Year x Variety	3.289	4.870	1.185

Lodging is still regarded as a reason not to grow oats, even though modern varieties are much less prone to lodging. Currently plant growth regulators are routinely used on oats, however the CEL RL data demonstrated that some of the shorter, newer varieties such as Tardis, Brochan, Mascani and Balado suffer a yield penalty as a consequence. In addition some millers market oats as being PGR free so dwarf oats or lodging resistant oats fit very well into their requirements. We are using lodging resistance in the selection process in advancing lines to variety trials. Figure 9 shows height differences amongst oat varieties.

Figure 9 Quantifying height differences in oats

3.4.2.5. Fertiliser requirements

RB209 shows oats require ~120kg /ha of N compared to wheat at ~200kg/ ha of N. This confirms the lower input status of oats. Nitrogen fertiliser production and use is one of the major components of the carbon footprint of agriculture. A replicated trial with 4 levels of N and 8 varieties was conducted at ADAS over 2 years to examine the response of oats to additional levels of N (Figure 10a, b and Table 4). This was designed to ensure some lodging by applying supra optimal levels of N to a range of varieties chosen for their standing ability. There were 4 treatments, 1) no N applied, 2) N to RB209 levels, 3) RB 209 plus 50kg N and 4) RB209 plus 70kg N. Soil N status of the field was high (SNS=4 (121-160kg/ha) in 2006/07 and 3 (101-120kg/ha) in

2007/08). As can be seen in the husked oats (Figure 10a), at levels above those in treatment 3 there was no improvement in yield for two varieties indicating a plateau had been reached in terms of N response and any further addition of N would be unproductive in terms of yield due to lodging. There was a slight indication that for varieties Balado and Tardis further N application (e.g. treatment 4) resulted in greater yield. This response was also observed for 3 of the naked oats varieties and it is clear that they were still responding well to additional N with the exception of Racoon which had mainly lodged at the exceptionally high levels of N.

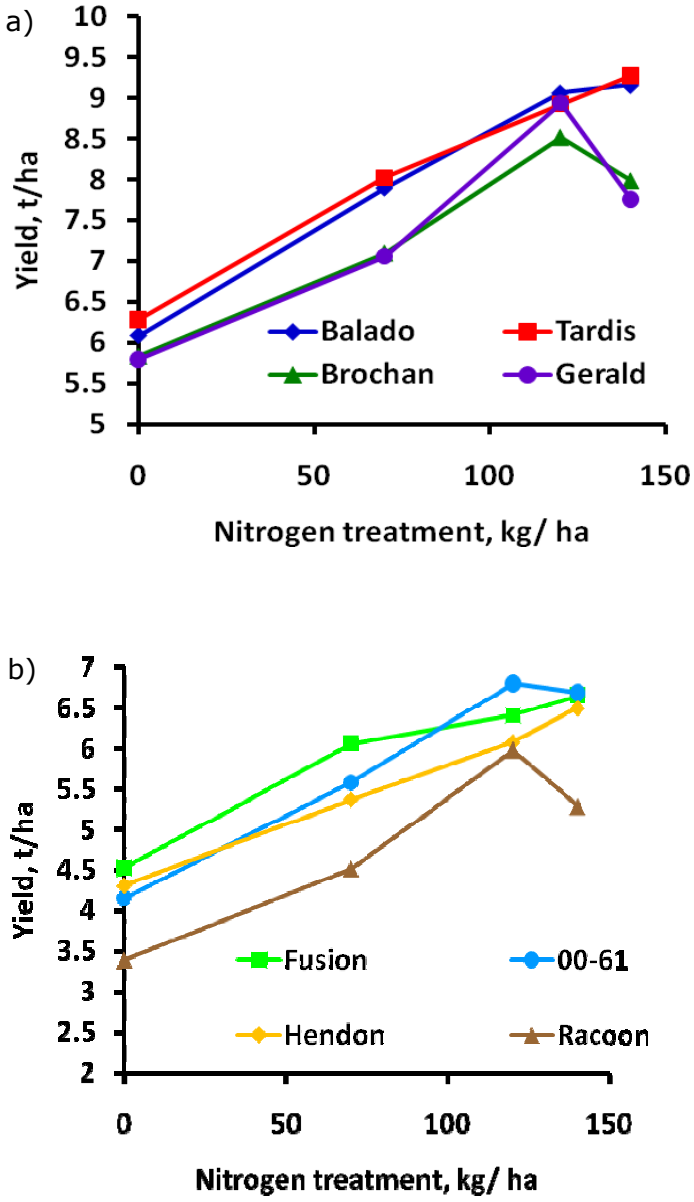


Figure 10. Yield response of a) husked and b) naked oats to applied N (mean of 2 years results)

These data indicated that RB209 may have been underestimating the N requirements of modern oats especially the newer dwarf varieties which are similar in height to wheat. Concern was raised at the project management committee about the revision of RB209 and whether the requirements of modern oats would be truly reflected.

Table 4. Yield results for oats under 4 different N regimes (2007 harvest) (t/ha @ 85% DM).

	Nitrogen treatment				Variety Average
Variety	Nil	RB209	RB209 + 50kg N/ha	RB209 + 70kg/ha N	
Hendon	4.11	4.33	5.89	6.24	5.14
Fusion	4.59	6.02	6.57	5.66	5.71
Balado	5.75	7.20	9.46	9.45	7.97
00-61	4.02	5.14	6.98	6.53	5.67
Tardis	6.55	8.43	9.46	9.91	8.59
Brochan	5.24	6.33	7.96	8.29	6.96
Gerald	5.53	6.06	8.70	8.50	7.19
Racoon	3.12	4.16	6.29	5.33	4.73
Nitrogen Average	4.86	5.96	7.66	7.49	
Nitrogen P-Value	<0.001				
Nitrogen SED (64df)	0.274				
Variety P-Value	<0.001				
Variety SED (64df)	0.387				

3.4.3. Objective 3. To identify, incorporate, select and evaluate important traits for sustainable production and premium livestock feed

In supporting this project the poultry industry expressed the need for oats which are economically competitive and/or have high oil/energy without jeopardising protein and amino-acid content. Within OatLINK both naked and husked oats have been studied with the focus on increasing the oil content and hence the energy (ME) value. Development of markers for oil have been summarised under objective 1. In parallel to the molecular approach, we have also been developing other high throughput analysis using NIR facilities. Progress is detailed below.

Various crosses have been carried out to validate markers associated with oil; for example 06-153 was grown in the glasshouse in years 2006-2007 and then in the field in 2007. The progeny ranged from 7.52 to 11.12% oil with both parents being about 9.9 % oil.

Over the course of the project approximately 500 crosses have been carried out to develop naked oats for animal feed. In general the high oil sources are from North America material that is agronomically poor and unadapted to UK conditions. The development of a high oil oat in a good genetic background with good agronomic attributes has therefore been challenging. The variety Racoon was added to the National List in 2006. This has an oil content of 12-14% but is very tall (>140cm). The objective is a high oil short or dwarf naked oat which is high yielding. For example in 2007 we completed 97 crosses with naked oats, of which 45 of these crosses were designed to provide high oil dwarf oats. In the course of the project over 8000 ex F3 lines have been assessed for oil with a range from 6-13% determined by NIR. In 2008 1165 ex F3 naked oats had a range of 5.98 to 11.34% and 63 husked oats had a range between 6.26-10.85%.

An NIR calibration for oil and total N content has been developed for whole groat samples (Figure 11) based on the cold soxtec method enabling the rapid analysis of oil content. This is routinely used for the naked oats. Higher levels of oil can be verified using AHEE wet chemistry method.

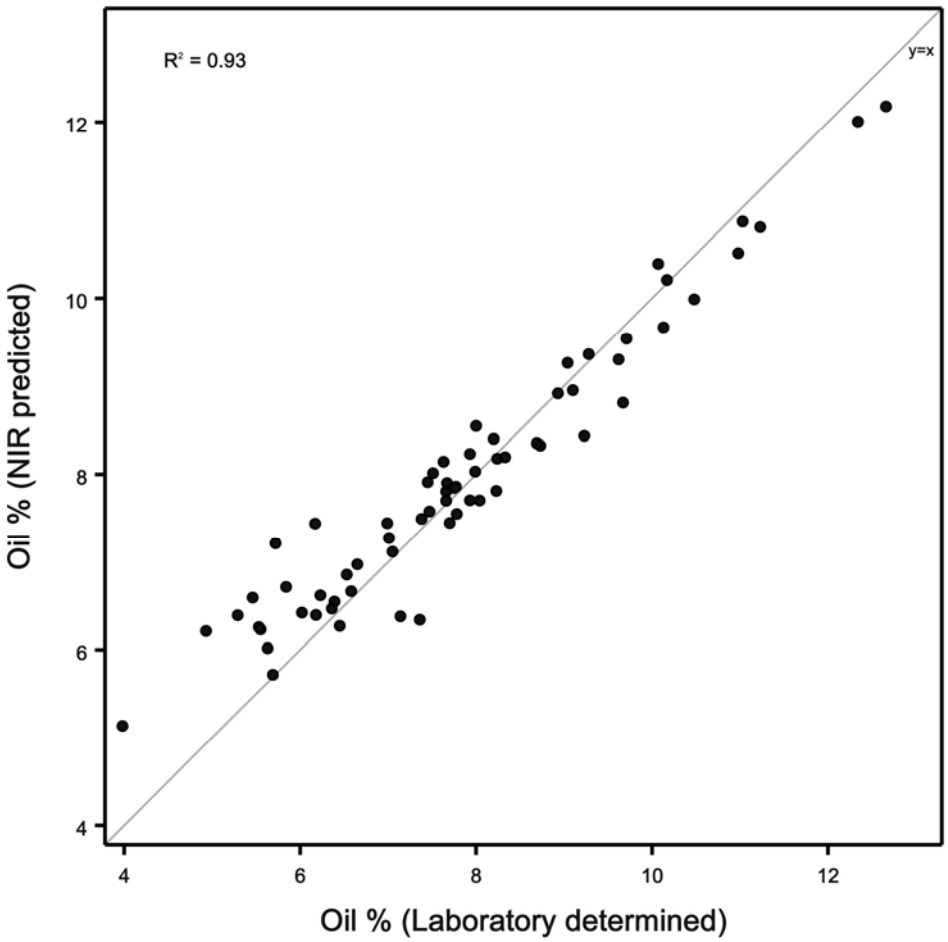


Figure 11. Predicted oil % versus lab determined oil using Foss NIRSystems 6500

In 2006 a Static field NIR system was purchased from Haldrup which utilises a Zeiss Corona diode array NIR instrument and calibrations for its use are currently being developed (see below). The field NIR can scan larger sample bulks (300-400 g rather than the 20g used in lab based NIR) (FOSS NIRSytems 6500) and we are in the process of calibrating this as a high throughput screening tool for both the naked and husked oats (Figures 12 and 13).

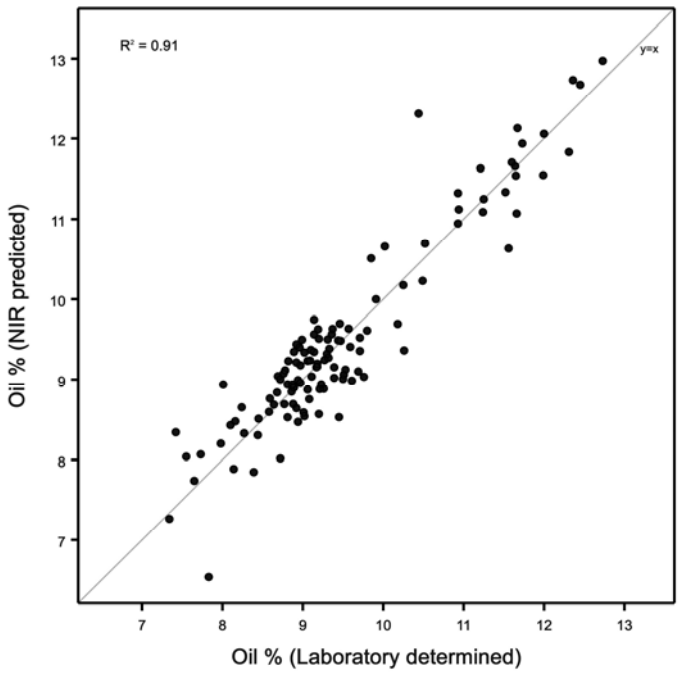


Figure 12 Predicted oil using Static NIR against lab determined oil

Figure 13 Static field NIR and comparison of sample sizes.

In 2006-07 50 high oil lines (>10% oil by NIR) were tested for agronomic quality, and 17 continued for further study in 2008. A high oil dwarf oat (01-145Cn1) was entered into NL trials in 2008-2009. This has progressed to its second year of National List testing and in breeding trials has out yielded Racoon and Hendon.

The project has gathered confirmation to show that increasing oil content increases the energy (ME) value of naked oats. Within OatLINK, Roslin ME bioassays have been carried out on broilers. ME values were quantified in a range of genetic material in a number of experiments from 2004-2008. This included advanced lines selected for high oil content (naked and husked oats) in comparison with conventional husked oats and feed wheat as well as some feed wheat used by commercial feed compounders. Chemical analysis of the lines has also been undertaken to identify impact on ME. Tables 5 and 6 summarise results from this analysis of the energy value of oats in comparison with standard feed wheat. It is clear that naked oats have a much greater feed value and this has been confirmed by large scale feeding trials completed by Bernard Matthews. Husked oats were equivalent or slightly lower than wheat.

Table 5. Metabolisable energy values of oats and reference wheats in growing broiler chicks (2004-2007 harvests)

Variety		TME_N MJ/kg as fed 2004		TME_N MJ/kg as fed 2005		TME_N MJ/kg as fed 2006		TME_N MJ/kg as fed 2007		
		Mean	SD	Mean	SD	Mean	SD	No	Mean	SD
[1]95-240-Cn	*high oil*	16.11	*0.38*							
[1]Racoon *ex Lydbury*	*high oil*			15.68	*0.51*	15.69	*0.46*			
[1]Racoon *ex Morspan*	*high oil*					16.21	*0.50*			
[1]Racoon *Gogerddan*	*high oil*							3	15.98	*0.43*
[1]Racoon *Rosemaund 1*	*high oil*							11	16.04	*0.47*
[1]Racoon *Rosemaund 2*	*high oil*							12	16.06	*0.39*
[1]Racoon *Rosemaund 3*	*high oil*							13	16.04	*0.57*
[1]Racoon *Rosemaund 4*	*high oil*							14	15.65	*0.42*
99-76-Cn	*high oil*			15.27	*0.35*					
00-171-Cn3	*high oil*			15.45	*0.18*					
01-126-Cn1	*high oil*					15.83	*0.44*			
01-126-Cn2	*high oil*					15.92	*0.46*			
01-126-Cn5	*high oil*					15.46	*0.45*	6	15.12	*0.19*
01-145-Cn1	*high oil*					15.54	*0.38*	7	14.84	*1.04*
[2]01-145A-Cn1/1	*high oil*							10	15.23	*0.36*
[2]02-146-Cn1	*high oil*							15	16.27	*0.20*
[2]02-233-Cn2	*high oil*							16	16.29	*0.49*
[2]02-227-Cn1	*high oil*							17	16.07	*0.44*
[2]02-217-Cn2	*high oil*							18	16.08	*0.31*
01-146-Cn5	*high oil*					15.84	*0.16*			
01-146-Cn7	*high oil*					15.73	*0.41*	8	15.59	*0.58*
[2]01-116-Cn7	*high oil*							9	15.35	*0.25*
Zuton	*naked spring*					15.40	*0.36*			
Lennon	*naked spring*					15.71	*0.32*			
Hendon	*naked dwarf*	15.68	*0.37*	14.38	*0.44*	15.32	*0.56*	4	15.37	*0.40*
Expression	*naked*	15.59	*0.40*							
[2]00-61-Cn3	*dwarf naked*							5	14.77	*0.54*
[2]01-171-Cnl 24	*dwarf covered*							19	12.68	*0.63*

Gerald	*covered*	12.17	*0.45*	12.38	*0.23*	11.50	*0.42*	1	11.77	*0.71*
Millennium	*covered*	12.33	*0.48*							
Brochan	*covered*					12.40	*0.44*			
[2]Mascani	*covered*							2	12.46	*0.44*
[2]02-133A-Cn6	*covered*							20	12.63	*0.42*
[2]01-171-CnV 49	*covered*							21	13.50	*0.66*
Consort	*wheat*	14.33	*0.41*							
Robigus	*wheat*	14.21	*0.37*							
Robigus 3	*wheat*			13.88	*0.37*			22	13.51	*0.85*
Robigus 5	*wheat*			13.76	*0.31*			23	13.76	*0.49*
Commercial feed Wheats (Bernard Matthews)										
Barnes+Maney	*wheat*					13.91	*0.33*			
"Combined"	*wheat*					13.97	*0.36*			
Fengrain	*wheat*					13.79	*0.21*			
Frontier	*wheat*					13.94	*0.57*			

[1]95-240-Cn was renamed Racoon in 2005.

[2]ME assayed for first time in 2007 harvest.

Several large scale poultry feeding trials have been conducted by Bernard Matthews. The impact of incorporating the high oil line Racoon in the feed ration on bird health and productivity and on lipid stability was quantified. Energy and oil values assigned to Racoon (15.3 MJ/kg and 12.2%) respectively were confirmed by the study and bird performance was maintained when Racoon was included in the ration at the different inclusion rates.

Table 6. Metabolisable energy values of oats and reference wheats in growing broiler chicks (2008 harvest, with 2007 results for comparison).

Sample code	Variety		TME_N MJ/kg as fed 2008		TME_N MJ/kg as fed 2007	
			Mean	SD	Mean	SD
1	Dalguise	*covered*	12.65	*0.22*		
2	Balado	*covered*	12.55	*0.40*		
3	Mascani	*covered*	12.78	*0.46*	1 12.46	*0.44*
4	Tardis	*covered*	12.42	*0.59*		
5	02-133A-Cn6/1	*covered high oil*	12.64	*0.30*	1 12.63	*0.42*
6	01-171-Cn1 24	*covered high oil*	12.46	*0.30*		
7	01-171-Cn11 13	*covered high oil*	12.15	*0.54*		
8	01-171-CnV 49	*covered high oil*	13.31	*0.75*	1 13.50	*0.66*
9	02-97Cn1	*covered low lignin*	12.40	*0.55*		
10	02-93Acn4	*covered low lignin*	12.34	*0.59*		
11	Racoon 1	*high oil*	15.86	*0.47*	16.04	*0.47*
12	Racoon 2	*high oil*	16.13	*0.41*	16.06	*0.39*
13	Racoon 3	*high oil*	16.15	*0.25*	16.04	*0.57*
14	Racoon 4	*high oil*	15.72	*0.31*	15.65	*0.42*
15	Gerald 1	*covered*	12.89	*0.55*		
16	Gerald 4	*covered*	12.49	*0.48*		
17	Brochan 1	*covered*	13.08	*0.41*	12.40^{2006}	*0.44*
18	Brochan 4	*covered*	13.20	*0.94*		
19	Gerald	*covered*	12.28	*0.44*	11.77	*0.71*
20	Brochan	*covered*	12.87	*0.24*		
21	Racoon	*high oil*	15.38	*0.35*	15.98	*0.43*
	Commercial-standard wheats (IBERS)					
22	Well grain	*wheat*	14.02	*0.42*	13.51	*0.85*
23	Open field	*wheat*	13.73	*0.22*	13.76	*0.49*

In addition to developing naked oats with high oil mainly for poultry feed crosses have also been carried out to produce high oil husked oats specifically for ruminant animal feed. This has been a minor part of the project as we have avoided increasing oil content in husked oats since they have generally been produced as milling oats and the milling industry have generally wanted low oil. Table 6 shows some high oil husked oats; as can be seen entry 8 is on average at least > 0.5% in TME value which is an improvement above other husked oats. Oil content was 11.45% on a dry matter

basis compared with other husked varieties such as Mascani at 8.7%, Dalguise 10.6% and Gerald 10.05%. The Canadian variety AC Assiniboia which has a low lignin husk has been included in the crossing programme. The low lignin husk is easily determined by staining with phloroglucinol as lignin stains a deep purple colour in the presence of phloroglucinol.

The low lignin husk has been shown to be more digestible to rumen microbes and in the future oat varieties that combine low lignin husk with a high oil groat will be a major target. The potential of a low lignin high oil husked oat has been shown by mixing ratios of high oil groats and low lignin husks (Cowan et al, 2008). This will form an important part of future research as combinations of high oil and low lignin husks have been shown *in vitro* to reduce methane emissions from ruminants.

3.4.4. Objective 4. To identify, incorporate, select and evaluate important traits for organic production

The needs of the organic sector are to capitalise on the high yield and stability of oats in organic systems both for their agronomic potential and as a livestock feed e.g. in mixtures with peas. In addition, the current annual demand for organic milling oats is around 10,000t, with up to 2,000t being imported. The development of oats with high oil and protein levels will improve the suitability of oats as an on-farm feed; this is especially important for organic producers who are increasingly being required to produce more feed on-farm and to provide completely organic rations.

A key objective of OatLINK was to quantify the agronomic performance of oat varieties and advanced selection lines in organic systems. Experiments have been conducted by Organic Research Centre - Elm Farm at two sites (Wakelyns and Sheepdrove) during the course of the project. A number of winter oat varieties (husked and naked) and advanced selection lines have been grown in experiments that compared methods of establishment, sowing rate and yield of these oats as both the 1st and 2nd cereal in the organic rotation.

The first differences detected among the husked varieties, mixture and populations in 2006/07 the main (second cereal) experiment were in early crop cover; there were no differences in the number of plants that emerged or established. Tardis had a

significantly (P = 0.015) higher level of crop cover than the other varieties and mixture. However, this trend was not repeated later in the season when maximum Leaf Area Index (LAI) was assessed. Unlike in the previous season (2005/06), when Tardis had a significantly higher LAI than the other varieties and went on to have the highest yield, in 2006/07 there were no significant differences in LAI among the varieties, the mixture and populations. Instead, the yields in 2006/07 correlated with disease levels. Unlike 2005/06 which was a low disease year, 2006/07 was notable in terms of the large amounts of disease present, especially crown rust. There were significant (P < 0.001) differences in total disease levels on the flag leaf among the varieties, mixture and populations. Gerald had significantly higher levels of disease than the other varieties, with Mascani having slightly, but not significantly, lower levels than Brochan and Tardis. These results are reflected in the yields of the varieties with Gerald having the lowest and Mascani the highest yields of the second cereal experiments.

The experiments confirmed the potential of oats as a cereal particularly suited to organic production with yields of > 7t/ha achieved. One of the main conclusions from the work in 06/07 was the difference in disease levels and yield between varieties as either first or second cereals. The winter oat varieties Tardis and Mascani yielded 7.3t/ha and 7.1t/ha respectively as the first cereal and 6.6t/ha and 7.0t/ha as the second cereal and yielded more than Brochan and Gerald. Tardis and Gerald were notably less productive as second cereals (Figure 14). However, the reduction between the average yields of the naked oats in the first and second cereal experiments was 19% compared with 3% in the husked oats (Figure 15).

An additional treatment included a mixture of the individual varieties. The mixture had 18% less disease than the average of its component varieties confirming data from 05/06, and yielded as well as the best individual variety as a first and second cereal. The naked oat Expression yielded 5t/ha as the 1st cereal but there was no significant advantage from the mixture over the individual varieties either in disease or yield. These experiments have been repeated in 07/08 to analyse performance of additional lines in organic systems.

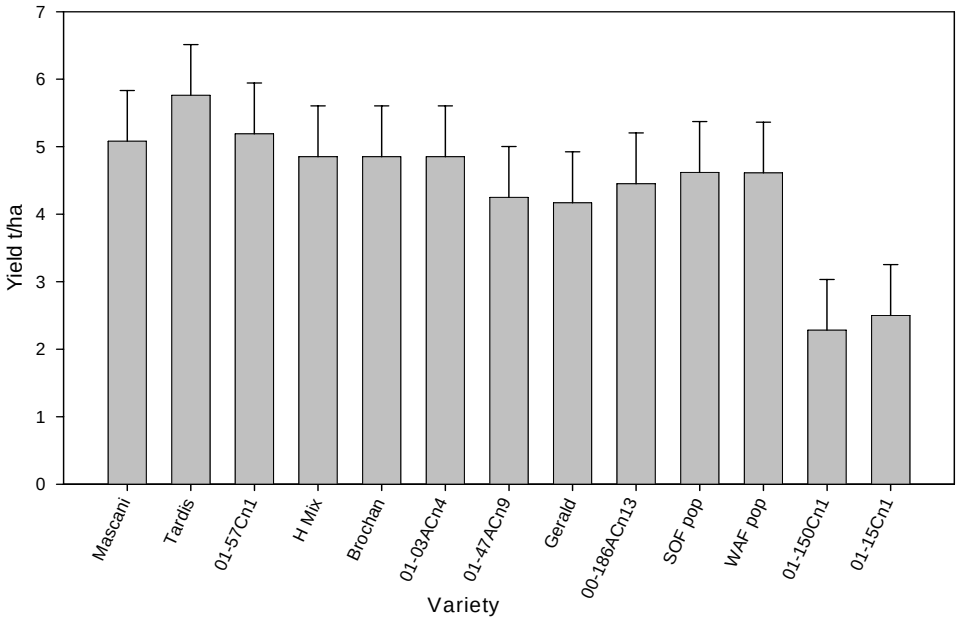


Figure 14. Yield results husked oats grown as second cereals at SOF 2007/08. Error bars indicate the l.s.d. at 5%.

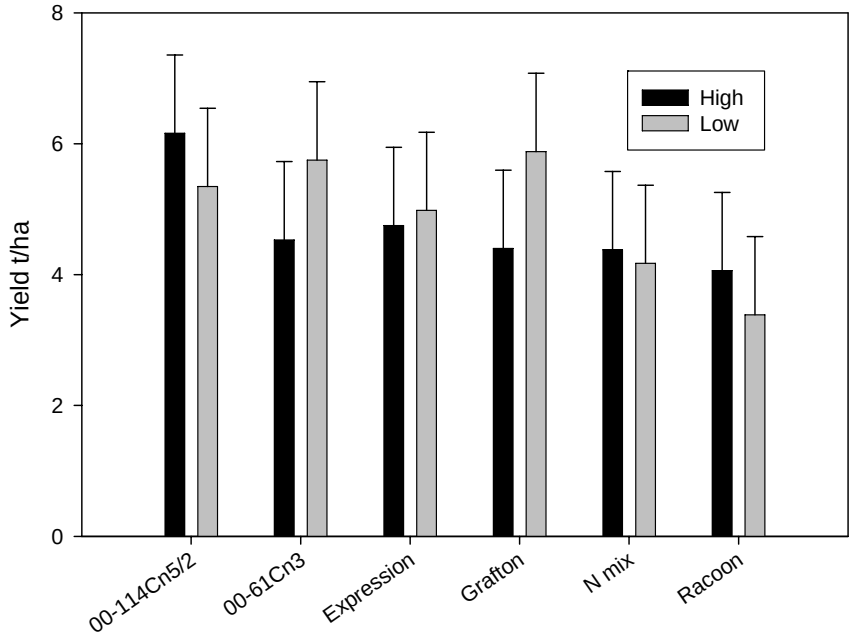


Figure 15. Yield of six naked oat varieties each at 150 and 200 kg seed per ha. Error bars are l.s.d for variety x rate effects.

3.5. Life Cycle Assessment in OatLINK

Life Cycle Assessment (LCA) were completed in accordance with international standards (ISO, 1997) for UK arable feed crops used in the poultry sector and for the UK porridge oat production chain. It was found that oats have relatively less ecological impact compared to other arable feed crops per unit delivered avian feed energy and therefore offer a more sustainable feed source for the poultry industry, however further work is needed in terms of the avian mass balance under different feeding regimes. Poultry feed is a growing market where the cereal input is currently fulfilled predominantly by wheat. The LCA technique was employed to assess the impact of naked oat cultivation compared to the cultivation of barley and wheat for feed. It was found that naked oat cultivation was more environmentally benign when compared directly to wheat and barley.

The LCA of the porridge oat production chain was completed to investigate the impact of plant varietal development to the sustainability of UK porridge production. In order to quantify the impact of different available variety traits a plough to plate life cycle assessment (LCA) of the flaked oat production chain was undertaken. The reduction of inputs which may be associated with multiple phenotypic characteristics was assessed using this technique. Significant progress has been made in two areas, and summarised below.

It was found that varietal improvements associated with the "downstream" cooking process were most influential across the majority of impact categories investigated. Furthermore traits relevant to the agricultural stage were especially important in the production chain, in particular, improvements in yield and nitrogen use efficiency correlated to a substantial reduction in the ecological footprint of porridge oats (McDevitt and Mila i Canals, 2009).

It was found that the cooking process was a dominant resource consumer and producer of emissions in the oat production chain (Figure 16) although the agricultural component of the oat production chain was the principal source of eutrophication.

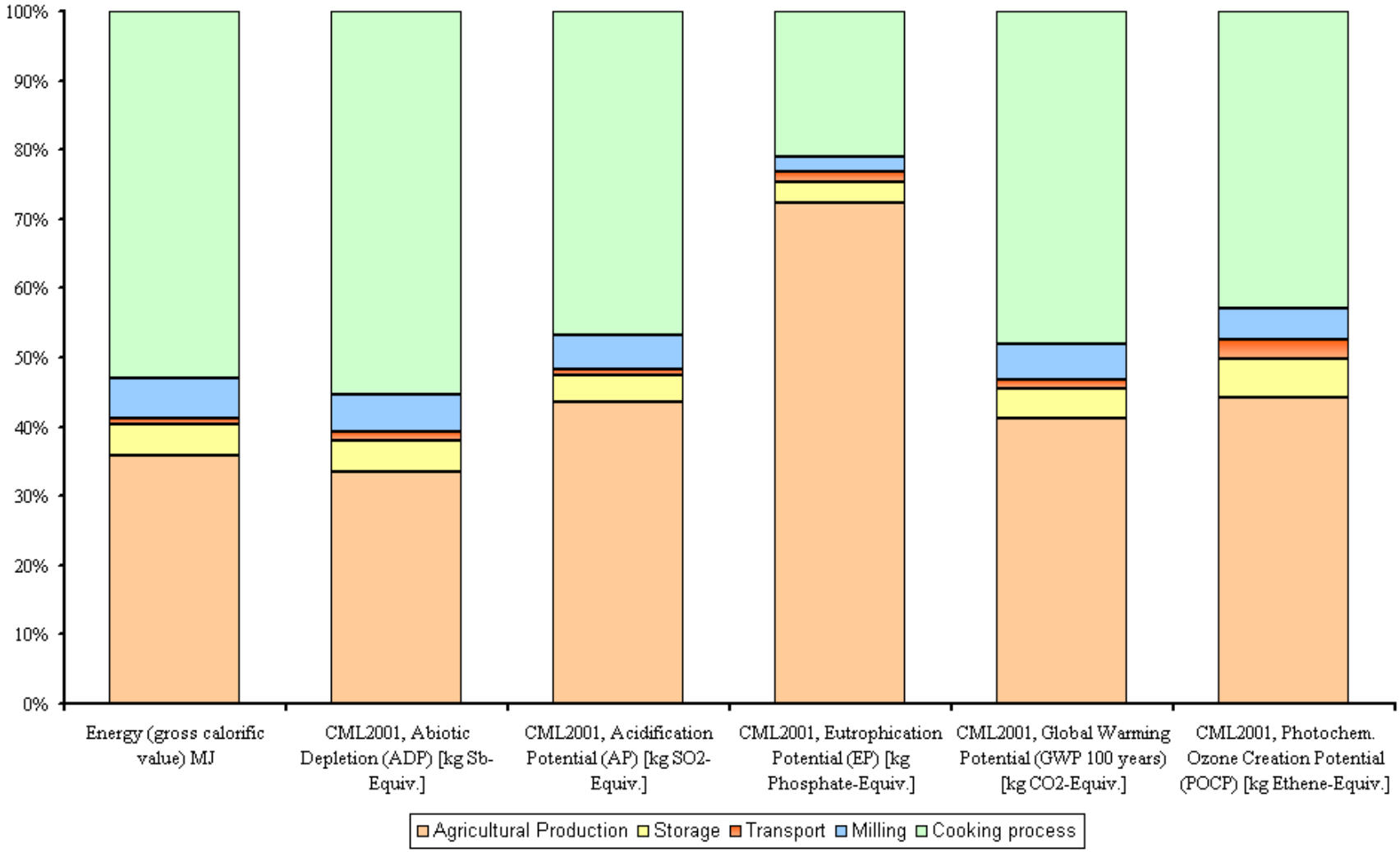


Figure 16. Stacked bar chart showing the relative contribution of the different components of the porridge oats production chain.

A sensitivity analysis was conducted in order to quantify the potential impact of a 10% change in resource use (Table 7). Accordingly with Figure 16 it was found that a reduction in cooking energy would have the most wide ranging impact on reducing the environmental footprint of the porridge oat production chain and improvements made in yield and nitrogen use efficiency (NUE) would have the most significant effect on eutrophication. Although it is noteworthy that improvements in NUE and yield also have a substantial effect on the other impact categories.

Fortunately, there are plant traits which affect the cooking process, for example variety improvements associated with enhanced viscosity or liquid absorption in cooking. Such improvements also show potential for varietal development which confers health improvements alongside environmental impact reduction. The traits relevant to this study are assessed for their potential effect on resource use, although few are immediately obvious and subject to phenotypic assessment. Advances in molecular markers and other targeted DNA technologies that facilitate non phenotypic assessment could be utilised for this purpose. Consequently varietal development should be in conjunction with the whole production chain and integration of conventional ecophysiological and phenotypic profiling with targeted DNA strategies may be a powerful methodology to produce enhanced varieties which fulfil the criteria of a multifunctional landscape.

Table 7. % effect on the studied impact categories caused by a 10% reduction in resource use through characteristics that may be affected by varietal development.

	Reduction in applied nitrogen	Reduction in applied potassium	Reduction in applied phosphorus	Reduction in cooking energy	Improvement in yield per hectare	Reduction in applied PGR	Reduction in applied insecticide	Reduction in applied fungicide	Reduction in applied herbicide
Energy (gross calorific value) [MJ]	-2.24	-0.37	-0.27	**-7.04**	-2.99	-0.03	-0.11	-0.12	-0.09
Abiotic depletion [kg Sb-Eq.]	-1.96	-0.33	-0.24	**-6.17**	-2.79	-0.03	-0.09	-0.10	-0.08
Acidification Potential [kg SO_2-Eq.]	-3.32	-0.33	-0.24	**-6.22**	-3.63	-0.03	-0.10	-0.11	-0.08
Global warming Potential$_{100\ years}$ [kg R11-Eq.]	-2.86	-0.34	-0.25	**-6.40**	-3.43	-0.03	-0.10	-0.11	-0.08
Ozone Layer Depletion Potential [kg CO_2-Eq.]	-2.40	-0.40	-0.29	**-7.54**	-2.81	-0.03	-0.12	-0.13	-0.10
Photochem. Ozone Creation Potential [kg Ethene-Eq.]	-2.70	-0.30	-0.22	**-5.70**	-3.67	-0.03	-0.09	-0.10	-0.07
Eutrophication Potential [kg Phosphate-Eq.]	**-5.98**	-0.15	-0.11	-2.81	**-6.01**	-0.01	-0.04	-0.05	-0.04

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Appendix A. Publications, conference presentations, papers and posters

2006

Clarke, S (2006) Bumper oat yields – Tardis trials top ten tonnes. The Organic Research Centre, Elm Farm Bulletin, December 2006

Jones, H., Clarke, S., Hinchsliffe, K. and Wolfe, M (2006) Husked and naked oat varieties and mixtures for organic farming systems In: Proceedings of the COST SUSVAR workshop on implications for production and products. La Besse, France 12-15th June 2006

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Griffiths, I (2007) Dissecting the components of yield in oats. Paper presented at Plant Science Wales, December 2007,

Howarth, Langdon, Cowan and Valentine, (2007) Poster "Development of markers associated with traits of agronomic importance in oats" at the Plant and Animals genome XV Conference, January, 2007.

2008

Clarke, S.M.,Jones, H. , Haigh, Z on disease levels, crop cover and their resulting yield. Boyd, H. & Wolfe, M.S. (2008) Effects of husked oat varieties, variety mixtures and populations ds. The 16th IFOAM Organic World Congress Modena 16-20 June 2008 Research Centre, Elm Farm Bulletin, November 2006

Cowan et al., (2008). Can methane emissions of ruminant animals be reduced by altering composition of feed oats. Proceedings of the Livestock and Global Climate Change Conference Hammamet, Tunisia 17-20 May, 2008 192-194.

Cowan et al, (2008). Can we breed feed oats which lower methane production by ruminants. Poster at International Oat Conference, Minneapolis, 28 June-2 July.

Cowan et al., (2008). Development of high oil winter and spring naked oats and release of variety Raccoon. Poster at International Oat Conference, Minneapolis, 28 June-2 July.
Cowan et al. (2008). Use and development of NIR spectroscopy for quality assessment on oats. Poster at International Oat Conference, Minneapolis, 28 June-2 July.
Griffiths et al. (2008). Dissecting the components of yield in oats. Poster at International Oat Conference, Minneapolis, 28 June-2 July.
Griffiths et al (2008) Genetic diversity and yield in oats. Invited paper at Molecular Mapping and Marker Assisted Selection in Plants Conference, Vienna 3-6 February 2008
Howarth et al. (2008). The use of genetic mapping to access and understand valuable traits in wild relatives of the cultivated oat. Poster at International Oat Conference, Minneapolis, 28 June-2 July.
Howarth et al. (2008). Development of markers associated with traits of agronomic interest in winter oats. Poster at International Oat Conference, Minneapolis, 28 June-2 July.
Howarth et al (2008) Introgression of traits from wild ancestors into cultivated oat. Invited paper, Plant and Animal Genomes XVI conference, January 12-15 San Diego USA
McDevitt (2008). Life Cycle assessment for Plant Breeding: An example using porridge oats. Poster at International Oat Conference, Minneapolis, 28 June-2 July.
McDevitt (2008) "Life Cycle Assessment for plant breeding" in Plant Genetic Resources for Food and Agriculture 22nd May 2008. Warwick HRI, Wellesbourne, Warwick.
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2009

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Appendix B. Mapping populations grown for OatLINK

Winter oat mapping populations grown for OATLINK

01-171 ***Hendon x N327-6 dwarf naked oat crossed with high oil source***

This was used to generate the markers for oil and has been grown in the field for 3 seasons with selected lines being continued and trialled for yield characters in 2008 harvest. High oil selections from this cross have been used extensively as parents in the crossing programme. High oil husked lines are also being developed mainly form this source although also from other lines obtained from the Iowa recurrent selection programme.

02-177 ***SA 99752 x 96-21Cn19 a cross to incorporate β-glucan into winter oat background.***

This has been grown to use for BSA and phenotyped for two years in the field. Selections within this have also been grown in the field and used in the crossing programme. New sources of higher β-glucan have been obtained and are being tested using the markers developed in the project. We have also utilised these sources in the breeding programme.

Av6118 ***Buffalo x Tardis cross.***

This cross was designed to cover the traits of interest to the millers with yield , kernel content, disease resistance and dwarf trait all potentially varying. This was grown in 05-06 as F3 rows and 06-07 and 07-08 as replicated yield plots at Gogerddan. It has been phenotyped extensively to provide data to allow the association of phenotypic traits with genetic markers.

04-52 was a cross designed to incorporate the low lignin trait identified in AC Assiniboia into winter oats. Bulk segregant analysis was used to identify progeny with low lignin. Low lignin crosses are at various stages in the breeding programme. We have crossed this low lignin husk type with high oil husked lines to produce a potential ruminant feed.

06-87 and **06-90** were crosses designed to incorporate large grain from the wild type B443. Both populations were phenotyped in the glasshouse and 06-90 was grown in the field in 2008. This was to identify features which can contribute to yield and harvest index. 06-87 was not grown in the field as it exhibited the wild type characteristic of shedding seed.

06-153 is a naked dwarf cross with different high oil sources and was utilised to verify molecular markers for oil. It was also grown in the field in 2008 and will be tested for oil content during the winter.
Currently we have 4 populations which will be used to verify markers
06-78 is a cross which incorporates low lignin husk and high oil husk dwarf

06-81 is high oil husk x high oil husk dwarf

06-159 naked oat x high oil naked oat dwarf

06-165 tall high oil naked x dwarf high oil naked

Spring oat mapping populations used in OatLINK

The Swedish Freja x Matilda population has been grown in the field for two seasons and the oil content determined.
A new population Solfi x Hifi from Canada which is a high β-glucan cross has been grown in the field for phenotyping.

Appendix C. Winter oat varieties entered to National List from 2004-present

Variety name	Husked or naked	Entered NL	Entered CEL RL testing
96-21Cn7 Brochan	Husked	September 2003	2005-06
96-41Cn3 Tardis	Husked	September 2003	2005-06
95-240Cn3 Racoon	Naked	September 2003	
98-28Cn3 Balado	Dwarf Husked	September 2005	2007-08
98-82Cn1 Fusion	Dwarf Naked	September 2005	2007-08
01-03ACn4	Husked	September 2007	2009-10
00-186ACn13	Husked	September 2007	withdrawn
00-114Cn5	Naked	September 2007	2009-10
00-61Cn3	Naked	September 2007	withdrawn

Appendix D. Spring oats entered into national List trials from 2004-present

14603Cn Husked oat passed through NL testing but marketing decision not to proceed to Recommended List in 2005

Spring naked oats, there is no category for spring naked oats in NL

13914Cn named Zuton added to NL in 2006

14149Cn named Lennon added to NL in 2007

Appendix E. Oat varieties on the CEL Recommended List

Winter husked oats Gerald, Mascani, Brochan, Tardis, and Balado added dec2009

Winter naked oats Hendon, Grafton, Expression and Fusion added Dec 2009.

Spring oats Bullion (naked) and Banquo (husked) which are not on the Recommended List but still grown.

Appendix F. ADAS Protocols

R & D EXPERIMENT PROTOCOL

1. TITLE

Advanced breeders' Lodging/Nitrogen trial.

2. REFERENCE NUMBER

XAA1412

3. STUDY DIRECTOR

Philip Bounds, ADAS Rosemaund, Preston Wynne, Hereford HR1 3PG

Tel: (01432) 820444 Fax: (01432) 820121

Mob: 07725 360388 Email: Philip.Bounds@adas.co.uk

4. CUSTOMER/SPONSOR

IGER project leader of 'OATLINK' SAL Link project

Sandy Cowan, IGER, Plas Gogerddan, Aberystwyth

Tel: 01970 823193 Fax: 01970 828357

5. COMPLIANCE STATUS

None

6. AUTHORISATION

The undersigned have read, agreed and will comply with this protocol:

Study director:…….. Date:……..

Site Manager:…… Date:……..

7. SITES, SITE MANAGERS AND OTHER TEST FACILITIES

Site: ADAS Rosemaund

Site Manager: Philip Bounds, ADAS Rosemaund, Preston Wynne, Hereford HR1 3PG

Tel: (01432) 820444 Fax: (01432) 820121

Mob: 07880 786979 Email: Philip.Bounds@adas.co.uk

8. OBJECTIVES

To evaluate lodging in advanced breeders' lines of winter sown oat varieties.

9. TIMETABLE

9.1 Duration

The project fieldwork will commence in October 2006 and end in September 2007.

9.2 Milestones

(a)	October 2007	Drill plots
(b)	November 2007	Assess plant population
(c)	June/July 2008	Lodging Scores as required
(d)	July 2008	Rapid lodging resistance assessments
(e)	July 2008	Lodging component assessments
(f)	July/Aug 2008	Assess crop height
(g)	August 2008	Harvest plots to measure yield
(h)	October 2008	Data summaries sent to customer

10. TREATMENTS

There will be eight varieties, four conventional height types and four dwarf height types.

	Varieties
	Dwarf
1	Hendon (naked dwarf)
2	Fusion (98-68Cn1) (naked dwarf)
3	00-61Cn3 (semi dwarf naked)
4	Balado (98-28C23) (husked dwarf)
	Conventional Height
1	Gerald (husked conventional height)
2	Brochan (husked conventional height)
3	Tardis (husked conventional height)
4	Racoon (naked conventional height)

There will be four nitrogen treatments which will be confirmed once soil mineral nitrogen supply status of the field is known in the spring.

Nitrogen (TBC)	
1	Nil
2	RB209 recc
3	RB209 + 50kg N/ha
4	RB209 + 70kg N/ha

All agrochemicals to follow normal farm practice except *PGRs that should not be applied to experiments.*

11. EXPERIMENT DESIGN, DATA HANDLING AND ANALYSIS

11.1 Experimental Design

The experiment will be a split plot design with three replicates, the nitrogen as main plots (to avoid nitrogen scavenging) and varieties as sub plots. Plots will be 2m wide x 24m long.

11.2 Experimental records (paper)

All paper records should be kept as detailed in DATA/200 and DATA/201.

11.3 Experimental records (electronic)

All records should be kept as detailed in DATA/202.

11.4 Collation

Hand-written data will be transferred to Excel worksheets as soon as practicable after collection and in accordance with DATA/022. Template excel worksheets will be provided for recording the lodging character data. Data should then be verified and validated in accordance with DATA/300 and DATA/500.

11.5 Analysis

Data will be initially analysed by analysis of variance, or if assumptions of this test are not met, by another appropriate statistical method (DATA/600).

12. MATERIALS

12.1 Seed

Seed will be provided by the customer.

13. METHODS, ASSESSMENTS AND RECORDS

13.1 Field operations

13.1.1 Sites should be selected with as little as possible variation in soil type or drainage. Any variation should be minimised within the blocking of the experiment.

13.1.2 Weed control should remove any competition with the crop. Other treatments (micronutrients, molluscicides, insecticides) should follow standard farm practice. **No nitrogen fertilisers or PGRs to be applied to the experiments.**

13.1.3 Plots will be harvested with a plot combine harvester (CER/037).

13.2 Assessment of Plant Population

Assess plant population in autumn (Nov/Dec) once 100% establishment is achieved in all varieties from one nitrogen main plot per replicate. Count plants in 5 x 0.5m row lengths per plot (CER/011).

13.3 Assessments of lodging

At the first sign of lodging carry out the whole plot % score lodging according to SOP CER/017. After this, score lodging in the same procedure as and when further lodging occurs.

13.4 Assessment of Crop height

Measure final crop height (ground level to top of ear) at five places within each plot (CER/013) just prior to harvest.

13.5 Rapid lodging resistance assessment techniques

Two nitrogen treatments (RB209 recc and RB209 + 50) and all varieties (48 plots in total) will be assessed for lodging resistance in-situ at GS65 using the 'lodgemeter' in three locations per plot (see lodgemeter protocol). The plots will be split into two 12m areas, one for the destructive lodgemeter measurements and one for harvesting to determine yield.

13.6 Lodging component assessment

On one nitrogen treatment (RB209 recc) and all varieties (24 plots in total) both stem and root lodging components will be measured at GS65 (see lodging component assessment protocol).

13.7 Harvest

Harvest plots with a combine harvester and take a minimum of 2 kg samples for determination of moisture content and specific weight (CER/037; CER/008). Sub-sample 500g of sieved grain and send to Alex Cowan, IGER, Plas Gogerddan, Aberystwyth, SY2 3EB.

13.8 Other records

A study diary detailing all study activities and observations will be kept. Also the following site details will be recorded: soil type and series, previous cropping (4 years), straw disposal method, pre-sowing cultivations, sowing date, seed rate, and all pesticides and fertilisers applied (dates and application rates).

14. REPORTS

A bullet point summary report will be produced by September 2004.

15. RECORD RETENTION

The data and documents relating to the study will be formally archived at ADAS Rosemaund in accordance with SOP DATA/014.

16. SOP LIST

ADMIN/008	Production of R & D reports
AGRON/002	Preparing experiment site plans
AGRON/003	Calculating the seed rates of combinable crops
AGRON/005	Measuring specific weight in cereals, pulses and oilseeds using DICKEY-JOHN GAC2000 grain analysis computer
AGRON/017	Marking out experiment plots
CER/017	Assessing leaning, lodging, brackling and necking in cereals
CER/037	Measuring plot yields and taking grain samples with a combine harvester
CER/040	Cleaning harvested samples of wheat, barley and rye

CER/037	Weighing plot yields and taking plot grain samples on the combine harvester
CER/050	Cereals, assessing winter hardiness in experimental plots
DATA/014	Preparation and archiving of study specific raw data packages
DATA/019	Guidelines for the backup and archive of experimental data held on computer
DATA/020	Guidelines for keeping manual file records of experiments
DATA/023	Transferring data from Husky portable computers to office computer using the HCOM programme
DATA/201	Manual recording of data
DATA/200	Data capture - generic principles
DATA/202	Manually keyed electronic data capture
DATA/203	Semi-automated electronic data capture
DATA/300	Data verification - principles and procedures
DATA/500	Data validation - principles and procedures
DATA/600	Guidelines for data manipulation and statistical analysis
MECH/001	Calibration and use of Oyjord tractor-mounted seed drill

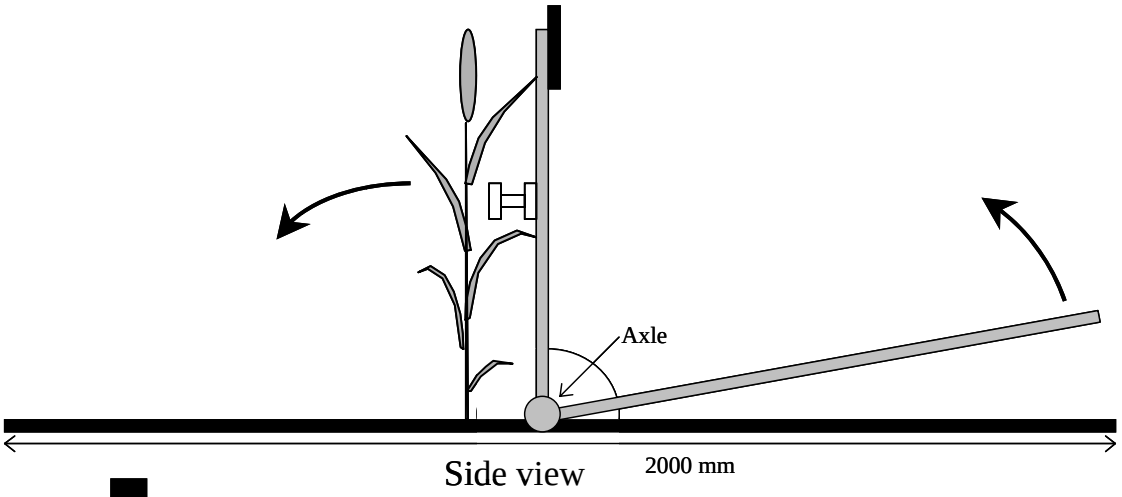
17. DISTRIBUTION

Copies of this protocol have been distributed to:

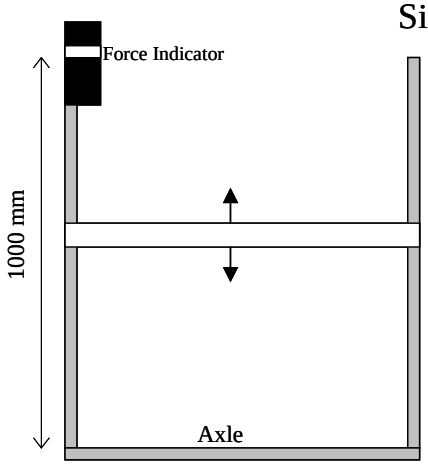
Study Director/Site Manager	P Bounds
Customer	S Cowan

Lodgemeter Protocol

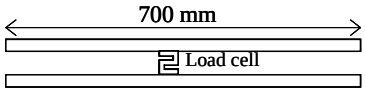
1. Assemble lodgemeter as in the diagram and photo below. NB be careful with the metal 'S' shaped load cell because it is quite sensitive.
2. Adjust the height of the pushing bar so that when in a vertical position its height (as measured to the centre of the bar) is 40cm above ground level.
3. Select one of the central 6 rows of a plot for testing. Flatten down the rows on one side of the testing row
4. Set the lodgemeter to its lowest setting with the arms resting close to the ground.
5. Position the lodgemeter so the pushing bar is just touching the wheat plants that are to be tested (see photo). The plants will be pushed across the direction of drilling.
6. Count the number of shoots to be tested.
7. Cut off the shoots at 50 cm about ground level
8. Switch on the Advanced Force & Torque Indicator (AFTI), then Zero the AFTI by pressing the 'ZERO' button.
9. Move the lodgemeter slowly to the 20^{o} position (3^{rd} notch) and leave for 10 seconds before recording the value on the AFTI in Newtons (N).
10. Repeat instruction 9 at lodgemeter positions 35^{o} (6^{th} notch), 50^{o} (9^{th} notch), 65^{o} (12^{th} notch) and 75^{o} (14^{th} notch).
11. At 75^{o} check whether any of the stems have buckled. This is best done when the stems are displaced.
12. After all the tests on a particular row, record whether or not the shoots return to their vertical position.



Side view



Front view



Plan view

Adjustable pushing plate & load cell

Rotating handles and upright struts

Base frame

Stem and root lodging component assessment protocol

All data should be inputted directly into a computer. At the end of each day, save the data to the f drive and print out a hard copy.

3.6.1.1. Shoot sampling

Select ten plants from 3-4 different areas of the plot and carefully remove each plant from the soil making sure that the whole stem is recovered. The base of the stem extends about 2cm into the soil so some levering with a hand fork will probably be necessary, particularly in dry soil. Avoid sampling from the outer 3 rows and any rows next to a missing coulter. Do not sample plants with 8 or more shoots. The shoots must not be damaged in any way because the strength of the stem base will be measured on them. Once each plant has been recovered, separate the main shoot of each plant and store in a plastic bag. The main shoot is identified as having the largest ear and it is often the tallest. Discard the remaining shoots. Refrigerate the main shoots as soon as possible to stop them from drying out as this affects the strength of the stem. Take care not to bend or buckle the stems when transporting the plants. Do not store shoots for more than TWO days. This means that a team of 2 people should not sample more than two–three sub-blocks (22-33 plots) at a time.

3.6.1.2. Shoot measurements

Do not carry out measurements on any internodes with severe eyespot or fusarium. Use CER002 for a definition of 'severe' infection.

Determine the shoot height (mm) from the stem base to the tip of the panicle. Determine the height at centre of gravity (mm) of the main shoot by cutting off the roots and balancing the isolated shoot on a ruler (leaves and ear still attached). Record the distance from the point of balance to the base of the stem. Measure the length of the panicle (mm). Clamp the base of the main shoot at the point where the soil surface would have been. Ensure that the shoot is vertical. Pull the shoot back (at the collar of the panicle) 5-10 cm from the vertical and release. Record the time for three complete oscillations to occur in the line of displacement. Natural frequency (Hz) = (timed period (s))/ (the number of oscillations observed during the timed period

(3)). Remove the panicle and measure the area by passing it through a green leaf area machine.

3.6.1.3. Stem base measurements

The first job is to identify internode one. This is defined as the first internode of more than 20 mm, which originates at or just below the ground surface and which does not have any crown roots emerging from its upper node. Subsequent internodes up the stem are numbered two, three, four etc., with the uppermost internode referred to as the peduncle. Measurements of the stem base are carried out on internode one and internode two of each main shoot. Measure the stem diameter (mm) at the middle of each internode using digital callipers. Measure the lengths of internodes one and two (mm) from the mid-point of their adjacent nodes. Determine the breaking strength (N) of internodes one and two using a three-point bending test. Support the nodes adjacent to the internode on the 'Y' frame which is clamped to the bench. Place the hook of a digital spring balance around the mid-point of the internode and apply a pulling pressure at an even rate. Record the force just before the internode buckles as its breaking strength. Finally, cut the internodes at their centre point and note whether the stem is hollow. NB a stem with pith is regarded as hollow. Use digital callipers to measure the stem wall width (mm). Take two measurements of stem wall width at right angles to each other, and record a mean value.

Place internodes one and two from each of the ten main shoots into a single bag and freeze. Make sure that the stem material saved does not include any stem material below internode one or above internode two.

3.6.1.4. Root sampling

Choose a day when the soil is wet to make this job easier. This can be done any time between GS61 and GS83. All plots can be sampled in one go because the root samples will be frozen before being measured at a later date.

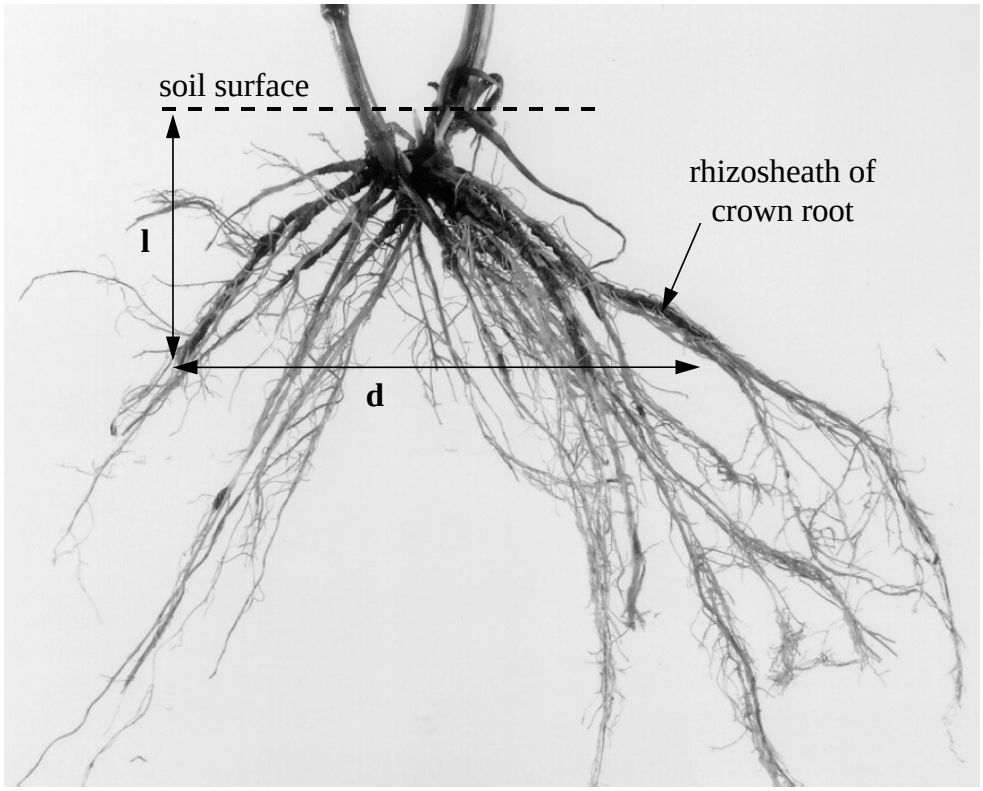
Select ten plants from 3-4 different areas of the plot. Avoid sampling from the outer 3 rows and any rows next to a missing coulter. Avoid any plants with 8 or more shoots. Before excavating snip the shoots off 5 cm above soil level. The shoots must be snipped off precisely at this height because this 5 cm distance will be used to estimate where the soil surface was during the lab measurements of root plate depth. Perhaps

take out a block of wood 5 cm thick and use this as a gauge for where to snip the shoots. Carefully excavate each whole plant with its upper root system. A fork should be used to ensure that the root system with associated soil is excavated to a depth of about 8 cm. Tease away any obviously excess soil that does not contain any roots. Place the ten plants from each plot in one plastic bag and freeze. The root measurements below can then be done during a less busy time.

3.6.1.5. Root measurements

In the lab, wash all remaining soil from the roots and isolate individual plants. Count the number of shoots on each plant. Identify the crown roots by their inherent rigidity and by the tendency for soil particles to adhere to their dense covering of root hairs, or rhizosheath (Figure 1). This distinguishes them from seminal roots, which emerge directly from the seed, number six or less, are much less rigid and usually have no adhering soil.

The section of crown root with rhizosheath is termed the 'rigid root length' for which there is usually only small variation between the roots of individual plants. Occasional plants do have very variable rigid root lengths, so that determination of the spread of the root plate and its depth is more subjective. The points at which the majority of rigid root portions terminate (Figure 1) are estimated visually to define 'root plate spread' (mm). Both the maximum root plate spread and the root plate spread at 90° to the maximum, usually the smallest spread, should be measured. Structural rooting depth (mm) is measured as the distance from base of the root plate to the soil surface, identified as the point 5cm below where the shoots were snipped off. For simplicity simply measure the distance from the base of the root plate to the end of the stems (5cm will then be subtracted during the data analysis). Place the roots and stem bases from each plot in a single bag and freeze.



Crown root system of a wheat plant at GS 73 (*d* - root plate spread, *l* - structural rooting depth).

The measurements given in the table below should now have been recorded.

Plant characters associated with lodging.

Plant Character	Units of measurement
*Natural frequency	Hz
Spread of the root plate	mm
Structural rooting depth	mm
Shoot number per plant	
*Shoot height	mm
*Shoot height at centre of gravity	mm
*Panicle length	mm
*Panicle area	cm^2
*Length of internodes one and two	mm
*Breaking strength of internodes one and two	(g)
*Diameter of internodes one and two	mm
*Wall width of internodes one and two	mm

* Carried out on the main shoot of each plant only.

R & D EXPERIMENT PROTOCOL

1. TITLE

Advanced breeders' variety trials.

2. REFERENCE NUMBER

XAA1412

3. STUDY DIRECTOR

Philip Bounds, ADAS Rosemaund, Preston Wynne, Hereford HR1 3PG

Tel: (01432) 820444 Fax: (01432) 820121

Mob: 07725360388 Email: Philip.Bounds@adas.co.uk

4. CUSTOMER/SPONSOR

Senova Limited

Alison Barrow, Senova Limited, 49 North Road, Great Abington, Cambridge, CB21 6AS.

Tel: 01223 890777 Fax: 01223 890666

5. COMPLIANCE STATUS

None

6. AUTHORISATION

The undersigned have read, agreed and will comply with this protocol:

Study director:…….. Date:……..

Site Manager:…… Date:……..

7. SITES, SITE MANAGERS AND OTHER TEST FACILITIES

Site : ADAS Rosemaund

Site Manager: Philip Bounds, ADAS Rosemaund, Preston Wynne, Hereford HR1 3PG

Tel: (01432) 820444 Fax: (01432) 820121

Mob: 07725360388 Email: Philip.Bounds@adas.co.uk

8. OBJECTIVES

To evaluate advanced breeders lines of winter sown Oat varieties.

9. TIMETABLE

9.1 Duration

The project fieldwork will commence in October 2007 and end in September 2008.

9.2 Milestones

(a)	October 2007	Drill plots
(b)	November 2007	Assess plant population
(c)	June/July 2008	Lodging scores as required
(d)	July 2008	Assess foliar disease
(e)	July/Aug 2008	Assess crop height
(f)	August 2008	Harvest plots
(g)	October 2008	Data summaries sent to customer

10. TREATMENTS

Two trials each of 25 advanced breeders varieties as detailed below.

Trial 1H – 25 varieties coded 1-25

Code	Selection code/name	Code	Selection code/name	Code	Selection code/name
1	Gerald	10	02-17Cn4	19	02-45Cn4
2	Dalguise	11	02-19Cn5	20	02-45Cn5
3	Kinross	12	02-19Cn7	21	02-56Cn2
4	2001-15Cn1	13	02-24Cn4	22	02-56Cn5
5	2001-57Cn1	14	02-25Cn1	23	02-56Cn9
6	03-77ACn24	15	02-25Cn2	24	02-56Cn10
7	03-77ACn27	16	02-34Cn3	25	02-168Cn1
8	02-04Cn2	17	02-45Cn1		
9	02-07Cn1	18	02-45Cn2		

Trial 6N – 25 varieties coded 126 - 150

Code	Selection code/name	Code	Selection code/name	Code	Selection code/name
126	Grafton	135	99-76CnI-II	144	02-58Cn2
127	Expression	136	01-146ACn1/1/1	145	02-66Cn2
128	Fusion	137	2001-116Cn7/1	146	*02-187Cn7*
129	2001-110Cn3/1	138	03-90ACn4	147	02-206Cn7
130	2001-92Cn6/2	139	03-90ACn7	148	*00-52Cn6/2/2*
131	2001-126Cn5/2	140	02-210Cn4	149	*02-225Cn1*
132	2001-145Cn1/2	141	02-213Cn2	150	*02-213Cn3*
133	2001-145Cn2/1	142	03-122ACn3		
134	2001-146Cn7/2	143	03-122ACn6		

The treatment numbers/codes shown must be used on all appropriate records and as unique identifiers for residue grain samples.

All agrochemicals to follow normal farm practice except PGRs that should not be applied to experiments.

11. EXPERIMENT DESIGN, DATA HANDLING AND ANALYSIS

11.1 Experimental Design

Experiments will be a randomised block with three replicates. Plots will be 2m x 6m.

11.2 Experimental records (paper)

All paper records should be kept as detailed in DATA/200 and DATA/201.

11.3 Experimental records (electronic)

All records should be kept as detailed in DATA/202.

11.4 Collation

Hand-written data will be transferred to Excel worksheets as soon as practicable after collection and in accordance with DATA/022. Template excel worksheets will be provided for recording the lodging character data. Data should then be verified and validated in accordance with DATA/300 and DATA/500.

11.5 Analysis

Data will be initially analysed by analysis of variance, or if assumptions of this test are not met, by another appropriate statistical method (DATA/600).

12. MATERIALS

12.1 Seed

Seed will be provided by the customer.

13. METHODS, ASSESSMENTS AND RECORDS

13.1 Field operations

Sites should be selected with as little as possible variation in soil type or drainage. Any variation should be minimised within the blocking of the experiment.

Weed control should remove any competition with the crop. Other treatments (micronutrients, molluscicides, insecticides) should follow standard farm practice. **No PGRs to be applied to the experiments.**

Plots will be harvested with a plot combine harvester (CER/037).

13.2 Assessment of foliar disease

Assess foliar diseases present within each plot at GS 75, score on a 1-9 scale (1 high disease, 9 no disease) on a whole plot basis. Assess plots by opening canopy and assessing at three points per plot.

13.3 Assessments of lodging

At the first sign of lodging carry out the whole plot % score lodging according to SOP CER/017. After this, score lodging in the same procedure as and when further lodging occurs.

13.4 Assessment of crop height

Measure final crop height (ground level to top of ear) at three places within each plot (CER/013) just prior to harvest.

13.6 Harvest

Harvest plots with a combine harvester and take a minimum of 2 kg samples for determination of moisture content and specific weight (CER/037; CER/008). Sub-sample 500g of sieved grain and send to Alex Cowan, IGER, Plas Gogerddan, Aberystwyth, SY2 3EB.

13.7 Other records

A study diary detailing all study activities and observations will be kept. Also the following site details will be recorded: soil type and series, previous cropping (4 years), straw disposal method, pre-sowing cultivations, sowing date, seed rate, and all pesticides and fertilisers applied (dates and application rates).

14. REPORTS

A bullet point summary report will be produced by September 2004.

15. RECORD RETENTION

The data and documents relating to the study will be formally archived at ADAS Rosemaund in accordance with SOP DATA/014.

16. SOP LIST

ADMIN/008	Production of R & D reports
AGRON/002	Preparing experiment site plans
AGRON/003	Calculating the seed rates of combinable crops
AGRON/005	Measuring specific weight in cereals, pulses and oilseeds using DICKEY-JOHN GAC2000 grain analysis computer
AGRON/017	Marking out experiment plots
CER/017	Assessing leaning, lodging, brackling and necking in cereals
CER/037	Measuring plot yields and taking grain samples with a combine harvester
CER/040	Cleaning harvested samples of wheat, barley and rye
CER/037	Weighing plot yields and taking plot grain samples on the combine harvester
CER/050	Cereals, assessing winter hardiness in experimental plots
DATA/014	Preparation and archiving of study specific raw data packages
DATA/019	Guidelines for the backup and archive of experimental data held on computer
DATA/020	Guidelines for keeping manual file records of experiments
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DATA/203	Semi-automated electronic data capture
DATA/300	Data verification - principles and procedures
DATA/500	Data validation - principles and procedures
DATA/600	Guidelines for data manipulation and statistical analysis
MECH/001	Calibration and use of Oyjord tractor-mounted seed drill

17. DISTRIBUTION

Copies of this protocol have been distributed to:

Study Director/Site Manager	P Bounds
Customer	A Barrow

Appendix G. ORC Elm Farm Protocols

Agronomic Assessments for OatLINK Trials 2004-2005

All assessments must be completed meticulously and identically on each site. Assessors must ensure they train together to calibrate techniques. Labelled photographs (i.e. site, date, variety, growth stage and assessment score) of the two extremes must be taken for each agronomic assessment at each site. This will then illustrate any differences that may be within the data set. It will also be important to take a set of photos regularly of each site to be used in hindsight (if necessary) to determine crude differences between sites (e.g. growth rate/weed dispersion). When conducting agronomic assessments the outer rows and 1m from plot ends should always be excluded.

Proposal suggests: EFRC will assess plant habit, plant height, maturity, lodging/leaning, winter damage, diseases (including seed-borne diseases), weed prevalence, yield and moisture content %, and milling quality.

Site Crop History

The previous year's crop and the current surrounding crop (species and variety if possible) will be recorded.

Site Management

A diary of any management practices likely to influence the trial shall be kept (i.e. spray/fertilizer regime, minimum till, weeding practice etc.).

Crop Emergence

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
10-12	Late October-Early November	Crop emergence	To be completed once on all sites.	

Randomly throw a sectioned 0.25m^2 quadrat on to the plot, count and record the number of individual wheat plants within the quadrat. Repeat this twice per plot. Multiply each count by 4 to calculate plants per m^2. The germination rate can then be calculated from the seed rate.

Crop Establishment

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
12-20	March	Crop establishment	To be completed once on all sites.	

Randomly throw a sectioned 0.25m^2 quadrat on to the plot, count and record the number of individual plants within the quadrat. Repeat this twice per plot. Multiply each count by 4 to calculate plants per m^2.

Early Crop Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
20-29	Late April-Early May	Early Crop Cover	To be completed once on all sites.	

The aim of this assessment is to provide an accurate measurement of the percentage of ground cover by plants at an early stage of crop growth. Randomly throw a sectioned 0.25m^2 quadrat on to the plot – Assess only five diagonal squares. For each of the five squares assess and record the percentage of ground cover by plants (i.e. 0%, 1-10%, 10-30%, 30-50%, 50-70%, 70-90% and 90-100%. Repeat this process twice per plot.

Early Weed Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
20-29	Late April-Early May	Early Weed Cover	To be completed once on all sites.	

The aim of this assessment is to provide an accurate measurement of the percentage of ground cover by weed plants at an early stage of crop growth. Randomly throw a sectioned 0.25m^2 quadrat on to the plot – Assess only five diagonal squares. For each of the five squares assess and record the percentage of ground cover by wheat plants (i.e. 0%, 1-10%, 10-30%, 30-50%, 50-70%, 70-90% and 90-100%. Repeat this process twice per plot. The weed data should be benchmarked by assessing weed cover on bareground/wheelings between plots where necessary.

Pests and Diseases

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
57-93	June-August	Pests and diseases	To be completed on a minimum of three occasions at all sites	

The plants will need regular observation to determine if/when the start of infection begins. Once a disease has been observed the plants should be assessed a minimum of three times. The number and timimg of assessments required will be at the assessor's discretion, as this will vary depending on the rate of spread of infection. Diseases assessed will be Oat Mosaic Virus (OMV), Barley Yellow Dwarf Virus (BYDV), Mildew and Crown Rust. 10 random plants per plot will be assessed according to "NIAB Assessment Key 11" to generate % Infection. It is important that senescent leaf material is not included in assessments.

There may be other diseases and pests that require assessment depending on the season.

If aphid infestation occurs ten random heads per plot will be scored. The score will be none, low, medium and high (0, 1, 2 and 3 respectively, which will be calibrated with a photo of heads at each of the levels, from which a percentage aphid cover may later be calculated).

Late Canopy Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
41-47	Early June	Canopy Cover (LAI)	To be completed once on all sites.	

The Sunscan Canopy Analysis system should be used to generate canopy expansion data. For technical details refer to Delta-T manuals. The aim is to measure interception of solar radiation. The SunData software will help to plan appropriate times for measurements and how to take/record measurements. (refer to Word file, Sunscan details). Two under canopy (ground level and horizontal) measurements are required per plot, with the meter at 45 degrees to rows (figure 1).

Crop height

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
90-93	Early-Mid August	Crop height	To be completed on one occasion at all sites	

Crop height (cm) should be measured from the soil surface to the top of panicle for 10 random plants per plot.

Lodging

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
90-93	Mid-Late August (one week previous to harvest).	Lodging	To be completed on one occasion at all sites	

Degree of lodging within the plot should be scored. This will be recorded as the percent of each plot that is upright (0-30 degrees), the percent of the plot lodged (30-60 degrees) and the percent of the plot fallen (60-90 degrees).

Grain Parameters

The Grain will be assessed in a variety of ways to determine quality/yield parameters

Grain yield

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Grain Yield	To be completed once on all sites.	

Each plot will be harvested (avoiding contamination) from which the yield (t/ha @25%mc) will be calculated.

Milling Quality

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Protein/oil content	To be completed once on all sites.	

A 200g sample from each plot (to be numbered) will be sent to IGER/NRM (?) for analysis of milling quality.

Specific weight and TGW

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Specific weight and TGW	To be completed once on all sites.	

Specific weight will be assessed with Hectolitre easy weigh (to correct for moisture content add 0.35Kg/hl for each % over 15%mc or subtract 0.35Kg/hl for each % below 15%mc). Thousand grain weight will also be assessed and corrected to 15%mc.

Seed borne disease

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Seed borne disease	To be completed once on all sites.	

A 200g sample from each plot (to be numbered) will be sent to NIAB for analysis of Michrodochium nivale seed borne disease levels. (Be aware of ergot).

The comparison of types in pure stands should direct the selection of lines from segregating populations appropriate for monocultural organic production.

The comparison of varieties in mixtures should permit the selection of lines appropriate for mixing.

Identification of genotypes with appropriate grain and plant traits to be compared in mixture, intercrop and different sites/soil type trials against promising variety types identified in 2004-2005.

Agronomic Assessments for OatLINK Trials 2005-2006

All assessments must be completed meticulously and identically on each site. Assessors must ensure they train together to calibrate techniques. Labelled photographs (i.e. site, date, variety, growth stage and assessment score) of the two extremes must be taken for each agronomic assessment at each site. This will then illustrate any differences that may be within the data set. It will also be important to take a set of photos regularly of each site to be used in hindsight (if necessary) to determine crude differences between sites (e.g. growth rate/weed dispersion). When conducting agronomic assessments the outer rows and 1m from plot ends should always be excluded.

Proposal suggests: EFRC will assess plant habit, plant height, maturity, lodging/leaning, winter damage, diseases (including seed-borne diseases), weed prevalence, yield and moisture content %, and milling quality.

Site Crop History

The previous year's crop and the current surrounding crop (species and variety if possible) will be recorded.

Site Management

A diary of any management practices likely to influence the trial shall be kept (i.e. spray/fertilizer regime, minimum till, weeding practice etc.).

Crop Emergence

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
10-12	Late October-Early November	Crop emergence	To be completed once on all sites.	

Randomly throw a sectioned 0.25m^2 quadrat on to the plot, count and record the number of individual wheat plants within the quadrat. Repeat this twice per plot. Multiply each count by 4 to calculate plants per m^2. The germination rate can then be calculated from the seed rate.

Crop Establishment

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
12-20	March	Crop establishment	To be completed once on all sites.	

Randomly throw a sectioned 0.25m^2 quadrat on to the plot, count and record the number of individual plants within the quadrat. Repeat this twice per plot. Multiply each count by 4 to calculate plants per m^2.

Early Crop Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
20-29	Late April-Early May	Early Crop Cover	To be completed once on all sites.	

The aim of this assessment is to provide an accurate measurement of the percentage of ground cover by plants at an early stage of crop growth. Randomly throw a sectioned 0.25m^2 quadrat on to the plot – Assess only five diagonal squares. For each of the five squares assess and record the percentage of ground cover by plants (i.e. 0%, 1-10%, 10-30%, 30-50%, 50-70%, 70-90% and 90-100%. Repeat this process twice per plot.

Early Weed Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
20-29	Late April-Early May	Early Weed Cover	To be completed once on all sites.	

The aim of this assessment is to provide an accurate measurement of the percentage of ground cover by weed plants at an early stage of crop growth. Randomly throw a sectioned 0.25m^2 quadrat on to the plot – Assess only five diagonal squares. For each of the five squares assess and record the percentage of ground cover by wheat plants (i.e. 0%, 1-10%, 10-30%, 30-50%, 50-70%, 70-90% and 90-100%. Repeat this process twice per plot. The weed data should be benchmarked by assessing weed cover on bare ground/wheelings between plots where necessary. The weed data for plots should then be expressed as a % of weed cover relative to bare ground (positive or negative).
Record the weed species present, noting those that are dominant for each block.

Pests and Diseases

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
57-93	June-August	Pests and diseases	To be completed on a minimum of three occasions at all sites	

The plants will need regular observation to determine if/when the start of infection begins. Once a disease has been observed the plants should be assessed a minimum of three times. The number and timing of assessments required will be at the assessor's discretion, as this will vary depending on the rate of spread of infection. Diseases assessed will be Oat Mosaic Virus (OMV), Barley Yellow Dwarf Virus (BYDV), Mildew and Crown Rust. 10 flag leaves per plot will be assessed according to "MAFF Assessment Key" to generate % Infection. Score the % of each disease, % senescent material (and calculate the % green in excel) of each flag leaf.

There may be other diseases and pests that require assessment depending on the season.

It is not necessary to score % diseases/senescent for overall plot, unless there is a hotspot, or the random flag leaves are felt to be unrepresentative.

1 assessor to assess 1 whole block. Assessors should calibrate with each other at the end of each row.

If aphid infestation occurs ten random heads per plot will be scored. The score will be none, low, medium and high (0, 1, 2 and 3 respectively, which will be calibrated with a photo of heads at each of the levels, from which a percentage aphid cover may later be calculated).

Late Canopy Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
41-47	Early June	Canopy Cover (LAI)	To be completed once on all sites.	

The Sunscan Canopy Analysis system should be used to generate canopy expansion data. For technical details refer to Delta-T manuals. The aim is to measure interception of solar radiation. The SunData software will help to plan appropriate times for measurements and how to take/record measurements. (refer to Word file, Sunscan details). Two under canopy (ground level and horizontal) measurements are required per plot, with the meter at 45 degrees to rows (figure 1).

Crop height

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
90-93	Early-Mid August	Crop height	To be completed on one occasion at all sites	

Crop height (cm) should be measured from the soil surface to the base of panicle for 10 random plants per plot.

Lodging

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
90-93	Mid-Late August (one week previous to harvest).	Lodging	To be completed on one occasion at all sites	

Degree of lodging within the whole plot should be scored. This will be recorded as the percent of each plot that is upright (0 degrees), the percent of the plot partly lodged (1-30 degrees) lodged (31-60) and the percent of the plot fallen (61-90 degrees).

Grain Parameters

The Grain will be assessed in a variety of ways to determine quality/yield parameters

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Moisture content	To be completed once on all sites.	

The aim of this assessment is to record the moisture content of the grain at the same time that other post harvest assessments (yield, TGW, specific weight etc) are undertaken. This is so results can be adjusted to 15% mc for fair comparison.
Moisture content of the grain will be determined using the Protometer. For full instructions refer to the manual.
Several random samples should be taken for each trial at each site. If the results are within 3% of one another, an average should be taken and used. If the results are more varied a sample should be taken from each plot.

Grain yield

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Grain Yield	To be completed once on all sites.	

Each plot will be harvested (avoiding contamination) from which the yield (t/ha @15%mc) will be calculated. Record the size of harvested area (combine width times plot length). Subtract weight of sack.

Milling Quality

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Protein/oil content	To be completed once on all sites.	

A 200g sample from each plot (labelled with IGER, EFRC, project, site, trial year, block, plot, cultivar, seed rate and undersown/not) will be sent to IGER for analysis of milling quality. Also send IGER plot plan in case of any queries.

Specific weight and TGW

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Specific weight and TGW	To be completed once on all sites.	

Specific weight of each plot will be assessed with Hectolitre easy weigh (to correct for moisture content, add 0.35Kg/hl for each % over 15%mc or subtract 0.35Kg/hl for each % below 15%mc). Thousand grain weight for each plot will also be assessed using grain counter/grain card and corrected to 15%mc.

Seed borne disease

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Seed borne disease	To be completed once on all sites.	

Obtain quote for finance department to determine number/bulking of samples to be sent (27 samples sent in 2004/05)
A 200g sample from each plot (labelled with NIAB, EFRC, project, site and plot number) will be sent to NIAB for analysis of seed borne disease levels. (Be aware of ergot, do not touch but do include in samples).

Agronomic Assessments for OatLINK Trials 2006-2007

All assessments must be completed meticulously and identically on each site. Assessors must ensure they train together to calibrate techniques. Labelled photographs (i.e. site, date, variety, treatments, growth stage) should be taken on each assessment occasion, one of each 'variety' and drill arrangement, and any other interesting photos - differences between plots etc. When conducting agronomic assessments the outer rows and 1m from plot ends should always be excluded.

Proposal suggests: EFORC will assess plant habit, plant height, maturity, lodging/leaning, winter damage, diseases (including seed-borne diseases), weed prevalence, yield and moisture content %, and milling quality.

The following assessments are for the main trial only (second cereal). The mini trial (1st cereal) will have only yield assessed. If there is funding, some quality assessments may be performed. To be confirmed.

Site Crop History

The previous year's crop and the current surrounding crop (species and variety if possible) will be recorded.

Site Management

A diary of any management practices likely to influence the trial shall be kept (i.e. spray/fertilizer regime, minimum till, weeding practice etc.).

Soil Fertility

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
00-09	October	Soil fertility	To be completed once on all sites.	

A soil sample will be taken from each alley at all sites. Sample will be sent to EFORC for analysis.

Crop Emergence

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
10-12	Late November	Crop emergence	To be completed once on all sites.	

Randomly throw a sectioned 0.25m^2 quadrat on to the plot, count and record the number of individual wheat plants within the quadrat. Repeat this twice per plot. Multiply each count by 4 to calculate plants per m^2. The germination rate can then be calculated from the seed rate.

Crop Establishment

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
12-20	March	Crop establishment	To be completed once on all sites.	

Randomly throw a sectioned 0.25m^2 quadrat on to the plot, count and record the number of individual plants within the quadrat. Repeat this twice per plot. Multiply each count by 4 to calculate plants per m^2.

Early Crop Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
20-29	Late April-Early May	Early Crop Cover	To be completed once on all sites.	

The aim of this assessment is to provide an accurate measurement of the percentage of ground cover by plants at an early stage of crop growth. Randomly throw a sectioned 0.25m^2 quadrat on to the plot – Assess only five diagonal squares. For each of the five squares assess and record the percentage of ground cover by plants (i.e. 0%, 1-10%, 10-30%, 30-50%, 50-70%, 70-90% and 90-100%. Repeat this process twice per plot.

Early Weed Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
20-29	Late April-Early May	Early Weed Cover	To be completed once on all sites.	

The aim of this assessment is to provide an accurate measurement of the percentage of ground cover by weed plants at an early stage of crop growth. Randomly throw a sectioned 0.25m^2 quadrat on to the plot – Assess only five diagonal squares. For each of the five squares assess and record the percentage of ground cover by wheat plants (i.e. 0%, 1-10%, 10-30%, 30-50%, 50-70%, 70-90% and 90-100%. Repeat this process twice per plot. The weed data should be benchmarked by assessing weed cover on bare ground/wheelings between plots where necessary. The weed data for plots should then be expressed as a % of weed cover relative to bare ground (positive or negative).

Record the weed species present, noting those that are dominant for each block.

Pests and Diseases

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
57-93	June-August	Pests and diseases	To be completed once at all sites.	

The plants will need regular observation to determine if/when the start of infection begins. The timing of the assessment will be at the assessor's discretion, as this will vary depending on the rate of spread of infection. Check regularly every 1-2 days from May if possible. Diseases assessed will be Mildew, crown, and *Septoria tritici. 10 random* flag leaves per plot will be assessed according to "MAFF keys 1.1.2-1.7, 1976" to generate % infection. Score the % of each disease, % senescent material (and calculate the % green in excel) of each flag leaf.

It is not necessary to score % diseases/senescent material for overall plot, unless there is a hotspot, or the random flag leaves are felt to be unrepresentative.

1 assessor to assess 1 whole block. Assessors should calibrate with each other at the end of each row.

When recording data on the score sheet - where senescence is 100%, diseases for that flag leaf must be marked with a *, not as 0%, as it is not possible to assess how much disease had been present.

If aphid infestation occurs ten random heads per plot will be scored. The score will be none, low, medium and high (0, 1, 2 and 3 respectively, which will be calibrated with a photo of heads at each of the levels, from which a percentage aphid cover may later be calculated).

Late Canopy Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
55	Early June	Canopy Cover (LAI)	To be completed once on all sites.	

The Sunscan Canopy Analysis system should be used to generate canopy expansion data. For technical details refer to Delta-T manuals. The aim is to measure interception of solar radiation. The SunData software will help to plan appropriate times for measurements and how to take/record measurements (refer to Word file, Sunscan details). Two under canopy (ground level and horizontal) measurements are required per plot, with the meter at 45 degrees to rows (figure 1).

Crop height

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
90-93	Early-Mid August	Crop height	To be completed on one occasion at all sites	

Crop height (cm) should be measured from the soil surface to the base of panicle for 7 random plants per plot.

Lodging

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
90-93	Mid-Late August (one week previous to harvest).	Lodging	To be completed on one occasion at all sites	

Degree of lodging within the whole plot should be scored. This will be recorded as the percent of each plot that is upright (0 degrees), the percent of the plot partly lodged (1-30 degrees) lodged (31-60) and the percent of the plot fallen (61-90 degrees).

Grain Parameters

The Grain will be assessed in a variety of ways to determine quality/yield parameters

Moisture content

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Moisture content	To be completed once on all sites.	

The aim of this assessment is to record the moisture content of the grain at the same time that other post harvest assessments (yield, TGW, specific weight etc) are undertaken, so results can be adjusted to 15% mc for fair comparison.

Moisture content of the grain will be determined using the Protometer. For full instructions refer to the manual.

Take five moisture readings for three random varieties/mixtures for each site (unless harvested on different days). Samples should be taken from the middle of sacks rather than the top as grain there will have dried more than the rest of the sack. If moistures are variable take more samples.

Grain yield

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Grain Yield	To be completed once on all sites.	

Each plot will be harvested from which the yield (t/ha @15%mc) will be calculated. Record the size of harvested area (combine width x plot length). Subtract weight of sack.

Milling Quality

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Protein/oil content	To be completed once on all sites.	

A 200g sample from each plot (labelled with IGER, EFRC, project, site, trial year, block, plot, cultivar, seed rate and undersown/not) will be sent to IGER for analysis of milling quality. Also send IGER plot plan in case of any queries.

Specific weight and TGW

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Specific weight and TGW	To be completed once on all sites.	

Specific weight of each plot will be assessed with Hectolitre easy weigh (to correct for moisture content, add 0.35kg/hl for each % over 15%mc or subtract 0.35kg/hl for each % below 15%mc). Thousand grain weight for each plot will also be assessed using grain counter/grain card and corrected to 15%mc.

Seed borne disease

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Seed borne disease	To be completed once on all sites.	

Obtain quote for finance department to determine number/bulking of samples to be sent (27 samples sent in 2004/05). A 200g sample from each plot (labelled with NIAB, EFORC, project, site and plot number) will be sent to NIAB for analysis of seed borne disease levels. (Be aware of ergot, do not touch but do include in samples).

Agronomic Assessments for OatLINK Trials 2007-2008

All assessments must be completed meticulously and identically on each site. Assessors must ensure they train together to calibrate techniques. Labelled photographs (i.e. site, date, variety, treatments, growth stage) should be taken on each assessment occasion, one of each 'variety' and drill arrangement, and any other interesting photos - differences between plots etc. When conducting agronomic assessments the outer rows and 1m from plot ends should always be excluded.

Proposal suggests: ORC will assess plant habit, plant height, maturity, lodging/leaning, winter damage, diseases (including seed-borne diseases), weed prevalence, yield and moisture content %, and milling quality.

HEALTH AND SAFETY

Assessors must read and comply with the ORC's stated Health & Safety Policy and Regulations (found in the Employee Handbook in the office and on the server (EFRC admin > Health & Safety)

Assessors should use common sense when carrying out assessments.

- Appropriate PPE to be worn at all times: dust mask, ear protectors, goggles and gloves where necessary. Long sleeved shirt, wide brimmed hat and sun cream to be used on sunny days outdoors. Long hair to be tied back or covered.
- Tetnus jabs should be kept up to date, cuts covered, touching ergot avoided, hands washed with soapy water regularly.
- Only trained operators to use machinery in compliancy with Machinery Use procedure (found in the Employee Handbook in the office and on the server (EFRC admin > Health & Safety).
- Adequate ventilation used in dusty environments. Excess dust swept up regularly.
- Care taken when lifting heavy objects – use two people where necessary.

The following assessments are for the main trial only (second cereal). The mini trial (first cereal) will have only yield assessed. If there is funding, some quality assessments may be performed. To be confirmed.

Site Crop History

The previous year's crop and the current surrounding crop (species and variety if possible) will be recorded.

Site Management

A diary of any management practices likely to influence the trial shall be kept (i.e. spray/fertilizer regime, minimum till, weeding practice etc.).

Soil Fertility

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
00-09	October	Soil fertility	To be completed once on all sites.	

A soil sample (minimum 300g) will be taken from each alley at all sites. Sample will be sent to NRM for analysis.

Crop Emergence

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
10-12	Late November	Crop emergence	To be completed once on all sites.	

Randomly throw a sectioned 0.25m^2 quadrat on to the plot, count and record the number of individual wheat plants within the quadrat. Repeat this twice per plot. Multiply each count by 4 to calculate plants per m^2. The germination rate can then be calculated from the seed rate.

Crop Establishment

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
12-20	March	Crop establishment	To be completed once on all sites.	

Randomly throw a sectioned 0.25m^2 quadrat on to the plot, count and record the number of individual plants within the quadrat. Repeat this twice per plot. Multiply each count by 4 to calculate plants per m^2.

Early Crop Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
20-29	Late April-Early May	Early Crop Cover	To be completed once on all sites.	

The aim of this assessment is to provide an accurate measurement of the percentage of ground cover by plants at an early stage of crop growth. Randomly throw a sectioned 0.25m^2 quadrat on to the plot – Assess only five diagonal squares. For each of the five squares assess and record the percentage of ground cover by plants (i.e. 0%, 1-10%, 10-30%, 30-50%, 50-70%, 70-90% and 90-100%. Repeat this process twice per plot.

Early Weed Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
20-29	Late April-Early May	Early Weed Cover	To be completed once on all sites.	

The aim of this assessment is to provide an accurate measurement of the percentage of ground cover by weed plants at an early stage of crop growth. Randomly throw a sectioned 0.25m^2 quadrat on to the plot – Assess only five diagonal squares. For each of the five squares assess and record the percentage of ground cover by wheat plants (i.e. 0%, 1-10%, 10-30%, 30-50%, 50-70%, 70-90% and 90-100%. Repeat this process twice per plot. The weed data should be benchmarked by assessing weed cover on bare ground/wheelings between plots where necessary. The weed data for plots should then be expressed as a % of weed cover relative to bare ground (positive or negative).

Record the weed species present, noting those that are dominant for each block.

Pests and Diseases

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
57-93	June-August	Pests and diseases	To be completed once at all sites.	

The plants will need regular observation to determine if/when the start of infection begins. The timing of the assessment will be at the assessor's discretion, as this will vary depending on the rate of spread of infection. Check regularly every 1-2 days from May if possible. Diseases assessed will be Mildew, crown, and *Septoria tritici. 10 random* flag leaves per plot will be assessed according to "MAFF keys 1.1.2-1.7, 1976" to generate % infection. Score the % of each disease, % senescent material (and calculate the % green in excel) of each flag leaf.

It is not necessary to score % diseases/senescent material for overall plot, unless there is a hotspot, or the random flag leaves are felt to be unrepresentative.

1 assessor to assess 1 whole block. Assessors should calibrate with each other at the end of each row.

When recording data on the score sheet - where Green Leaf Area = 0%, diseases for that flag leaf should still be assessed.

If aphid infestation occurs ten random heads per plot will be scored. The score will be none, low, medium and high (0, 1, 2 and 3 respectively, which will be calibrated with a photo of heads at each of the levels, from which a percentage aphid cover may later be calculated).

Late Canopy Cover

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
55	Early June	Canopy Cover (LAI)	To be completed once on all sites.	

The Sunscan Canopy Analysis system should be used to generate canopy expansion data. For technical details refer to Delta-T manuals. The aim is to measure interception of solar radiation. The SunData software will help to plan appropriate times for measurements and how to take/record measurements (refer to Word file, Sunscan details). Two under canopy (ground level and horizontal) measurements are required per plot, with the meter at 45 degrees to rows (figure 1).

Crop height

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
90-93	Early-Mid August	Crop height	To be completed on one occasion at all sites	

Crop height (cm) should be measured from the soil surface to the base of panicle for 7 random plants per plot.

Lodging

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
90-93	Mid-Late August (one week prior to harvest).	Lodging	To be completed on one occasion at all sites	

Degree of lodging within the whole plot should be scored. This will be recorded as the percent of each plot that is upright (0 degrees), the percent of the plot partly lodged (1-30 degrees) lodged (31-60) and the percent of the plot fallen (61-90 degrees).

Grain Parameters

The Grain will be assessed in a variety of ways to determine quality/yield parameters

Moisture content

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Moisture content	To be completed once on all sites.	

The aim of this assessment is to record the moisture content of the grain at the same time that other post harvest assessments (yield, TGW, specific weight etc) are undertaken, so results can be adjusted to 15% mc for fair comparison.

Moisture content of the grain will be determined using the Protometer. For full instructions refer to the manual.

Take three moisture readings for five random varieties/mixtures for each site (unless harvested on different days). Samples should be taken from the middle of sacks rather than the top as grain there will have dried more than the rest of the sack. If moistures are variable take more samples.

Grain yield

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Grain Yield	To be completed once on all sites.	

Each plot will be harvested from which the yield (t/ha @15%mc) will be calculated. Record the size of harvested area (combine width x plot length). Subtract weight of sack.

Milling Quality

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Protein/oil content	To be completed once on all sites.	

A 200g sample from each plot (labelled with IGER, ORC, project, site, trial year, block, plot, cultivar, seed rate and undersown/not) will be sent to IGER for analysis of milling quality. Also send IGER plot plan in case of any queries, & check 200g is sufficient.

Specific weight and TGW

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Specific weight and TGW	To be completed once on all sites.	

Specific weight of each plot will be assessed with Hectolitre easy weigh (to correct for moisture content, add 0.35kg/hl for each % over 15%mc or subtract 0.35kg/hl for each % below 15%mc). Thousand grain weight for each plot will also be assessed using grain counter/grain card and corrected to 15%mc.

Seed borne disease

GROWTH STAGE	APPROX. TIME OF YEAR	ASSESSMENT NAME	ASSESSMENT PROTOCOL	COMPLETION
91-93	September (post harvest)	Seed borne disease	To be completed once on all sites.	

Obtain quote for finance department to determine number/bulking of samples to be sent (14 samples sent in 2006/07)

A 200g sample from each bulk to be analysed (labelled with NIAB, ORC, project, site and plot number) will be sent to NIAB for analysis of seed borne disease levels. (Be aware of ergot, do not touch but do include in samples).