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By

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CONTENTS

ABBREVIATIONS

AMMI	Additive Main effects and Multiplicative Interaction
BG	β–glucan
DM	Dry matter
GS	Growth stage
HFN	Hagberg Falling Number
ICARDA	International Center for Agricultural Research in the Dry Areas
IBERS	Institute of Biological, Environmental and Rural Sciences, Aberystwyth University
N	Nitrogen
n	Number
nd	No data available
NS	Not significant
PC/PCA	Principal component/ Principal Components Analysis
PGR	Plant growth regulator
QTL	Quantitative trait locus
SENRGY	School of the Environment, Natural Resources and Geography, Bangor University
SpWt	Specific weight

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1. ABSTRACT

Under-utilisation of barley as a human food in the UK is potentially a missed opportunity for public health as components of barley grain, especially β-glucan soluble fibre, have the proven ability to ameliorate diet-related health problems, including obesity, type-2 diabetes and high cholesterol. The development of a health food market for barley could also benefit UK barley growers. All current UK barley varieties have covered grain and the hull must be removed by pearling to render the grain edible. Pearling also removes the nutritious bran and germ. Naked barley grains thresh freely from the *pales* (that form the hull) so that the whole grain can be used without processing.

This project evaluated the agronomic and grain quality characteristics of a collection of exotic naked barley varieties under field conditions in Wales over two years, and compared them with those of UK hulled varieties. Spring and autumn sowing were compared and grain β–glucan and amino acid content were measured. As a test of the potential to breed UK-adapted naked barley, selected naked lines from a cross between a conventional UK hulled and exotic naked barley were assessed for agronomic properties.

Many of the exotic naked barley varieties had high levels of foliar disease and lodging and only a limited yield response to fungicide. Conversely naked grain lines with modern UK parentage had stiff straw and good resistance to disease. The only agronomic problem specific to the naked grain trait was poorer crop establishment, due to the vulnerability of the exposed embryo to damage during harvesting, and to weaker coleoptile growth. Solutions for this are to ensure careful harvesting and handling of seed crops (e.g. reduce combine drum speed to 600-700rpm) and to delay sowing of spring varieties to ensure a warm seedbed. However, some naked Himalayan varieties, and progeny from Himalayan x UK crosses, had excellent seedling vigour, indicating that careful crossing and selection for early vigour may resolve establishment problems in future. There was wide variation in β-glucan concentration between genotypes (3.0 - 7.0g/100g DM), but also considerable variation between environments.

Lawina, a released German naked variety, was grown by four farmers to assess its possible use in a supply chain for food manufacturers. It showed a consistently low yield and failed to give a substantial yield response to fungicide, so the project concluded that it is not suitable for UK cultivation. Better-adapted varieties must be developed. Processors developed and evaluated a range of products (including speciality breads, flours and flakes) made from Lawina grain. Responses from the public to these were positive, indicating that there may be a viable market for UK naked barley.

2. SUMMARY

2.1. Background and objectives

Naked barley differs from covered barley in that the grains thresh freely from the outer covers of the flower, known as the *pales*. All current UK barley varieties are of the covered type, where the *pales* are tightly cemented to the grain and must be removed by pearling to render them edible for humans. No varieties of naked barley have been bred for UK conditions and food use, although naked barley has been grown in Asia for thousands of years. Asian varieties may possess variation for food and health traits that have been eliminated from European barley by selective breeding for malting quality. Exotic (including European) naked barley varieties are not well adapted to UK conditions. The German variety Lawina was identified in previous work as being the most appropriate for testing on a larger scale and with farmers, due to its good performance with food processors and being a variety released in a neighbouring European country.

The main aim of this project was to evaluate available exotic naked barley varieties alongside UK hulled varieties to identify agronomic requirements, develop a supply chain and produce best-practice agronomic guidance for the crop. A further aim was to identify pre-breeding material, lines and strategies for developing varieties better adapted to the UK climate.

The objectives of the trials were: i) to determine whether naked barley differs from covered barley in its agronomic management requirements; ii) to asses if it was viable to grow the available continental naked barley varieties in the UK climate; iii) to act as a pre-breeding screen to evaluate the usefulness of exotic varieties, including those from the Himalayas and Japan, as parents for crossing with UK covered varieties to introduce new variation into the UK barley gene pool; and iv) to give a better understanding of how genotype and environment interact to determine grain β-glucan content.

2.2. Materials and Methods

Replicated field trials were conducted at Bangor University's Henfaes Research Centre in North Wales during the 2008 and 2009 growing seasons. Varieties used were: the German naked barley varieties Lawina and Taiga; the covered UK control varieties, Cocktail, Optic, Static and Tipple; 26 naked lines from the breeder of Lawina, Cereal Breeding Research Darzau; three naked six-row semi-dwarf varieties bred by ICARDA, Aleppo, Syria; Koean, Himalayan and Japanese accessions from the John Innes Centre, Norwich. A small number of crosses were made between UK hulled and exotic naked varieties, and 21 selected lines were tested in 2009.

In order to assess the disease resistance of the different varieties all trials in 2008 were not treated with fungicides. In 2009, a fungicide programme based around triazole and strobilurin chemistry was used alongside untreated controls. Trials in 2008 showed that poor establishment and lodging were potential causes of loss of yield so that in 2009, a factorial experiment was carried out using Lawina sown at four seed rates, treated with trinexopac-ethyl (Moddus) PGR or untreated.

Laboratory tests were conducted for seed germination rate prior to sowing. Grain β-glucan content was measured with a standard Megazyme protocol.

2.3. Results and Discussion

2.3.1. Germination and establishment of naked barley

Germination percentages of naked barley were usually lower than those of covered barley varieties, possibly due to vulnerability of the exposed embryo to mechanical damage during harvesting. Increasing combine drum speed at harvest from 800rpm to 1200rpm decreased germination rate of Lawina from 95% to 85%.

Naked barley varieties consistently had lower establishment, compared to hulled barley, even when seed rates were increased to compensate for lower germination. Typically, only 80 plants were established for every 100 viable seeds sown. Many naked barley accessions had poor seedling vigour and weak coleoptile growth. Overall establishment rates were better in 2009 than in 2008 (Figure S1). This is likely to be due to the combined effect of very wet conditions prior to harvest in 2007 followed by a cold seed bed during spring sowing in 2008.

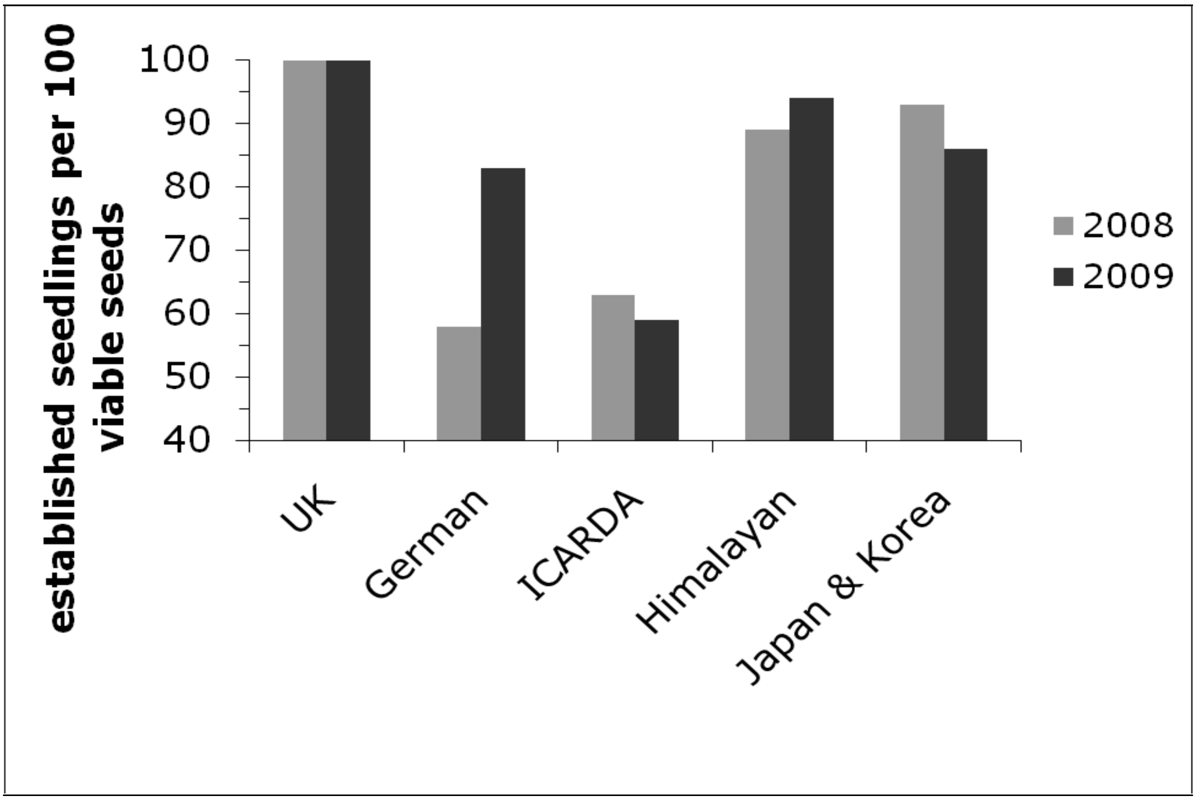


Figure S1. Establishment of spring barley varieties in 2008 and 2009. 'UK' are all covered-grain varieties (n = 4); 'German' are all naked-grain varieties (n = 28), 'ICARDA' are 6-row semi-dwarf naked varieties (n = 3); 'Himalayan' (n=13) and 'Japan & Korea' (n = 11) are naked-grain landrace varieties.

Although it was not formally tested under field conditions, the standard spring barley seed dressing Raxil-Pro (containing prothioconazole, tebuconazole and triazoxide) decreased laboratory germination of naked barley. It is strongly recommended not to use seed treatment of any kind on naked barley until further research is done.

2.3.2. Agronomy of naked barley under UK conditions

Yields of all varieties of naked barley were very low in the wet summer of 2008, illustrating their lack of adaptation to the wet UK climate and problems with disease and lodging. Yields were higher in 2009 (Figure S2, Table S1). The non-UK-adapted lines had a substantial yield penalty due to their poor agronomic characteristics, for example poor resistance to disease and lodging. However, it is not simply a case of comparing covered with naked varieties. The importance of modern agronomic traits to current UK cultivation was illustrated by the low yields of Haidd Enlli, a covered variety that lacks modern traits. In addition, naked barley usually yields around 15% less than otherwise equivalent covered barley as the weight of the hull is excluded from the yield.

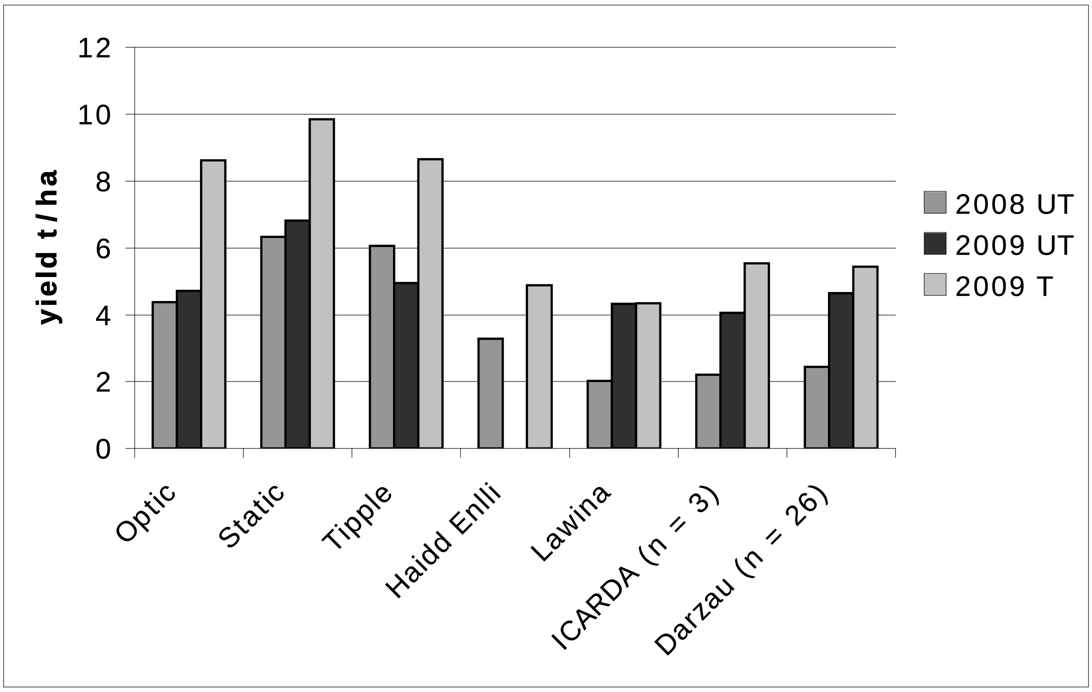


Figure S2. Grain yields of selected varieties and mean grain yields of the Darzau and ICARDA naked barley lines. Plots were untreated (UT) in 2008: and fungicide treated (T) or untreated (UT) in 2009.

Many naked barley accessions were very susceptible to powdery mildew (*Blumeria graminis*). It is likely that they lack the resistance genes found in modern UK varieties. Rhynchosporium (*Rhynchosporium secalis*) and brown rust (*Puccinia hordei*) infected both covered and naked barley types. The naked six-row 93.747, bred by

ICARDA, showed the lowest level of infection by Rhynchosporium of any of the accessions tested.

Use of fungicides increased yield and specific weight and decreased brackling in most naked barley lines and covered UK control varieties, although it increased lodging in many of the tall, non-UK-adapted lines.

An experiment to test the effects of seed rate and PGR on Lawina showed that Moddus (trinexapac-ethyl) significantly reduced straw length by 6cm and increased ear number, grain yield and specific weight.
There was wide variation in β-glucan concentration between genotypes (Last column in Table S1), but also considerable variation between environments. There was no evidence to support β-glucan concentration being diluted by higher grain yields, contrary to the situation for grain protein concentration. Foliar fungicide generally decreased β-glucan concentration, although in some accessions there was an increase. Sequential harvesting showed that grain β-glucan concentration increased initially as the grain matured, then decreased rapidly due to weather damage and sprouting, suggesting an optimum harvest window to ensure maximum β-glucan concentration.

2.3.3 Pre-breeding screening of 'exotic' germplasm and progeny of crosses between UK hulled and exotic naked types

Many of the Himalayan accessions showed much greater seedling vigour and stronger coleoptile growth. This translated into higher rates of crop establishment, typically 90 plants for every 100 viable seeds sown. Naked grain lines derived from Himalayan parentage appeared to retain this vigour, suggesting that it may be possible to overcome the establishment problems of naked barley by selective breeding.

Many of the non-UK-adapted naked barleys were very susceptible to lodging. Himalayan lines had very weak straw and Japanese and Korean lines were susceptible to root lodging. When Japanese and Korean lines were sown in autumn the plants were taller but there was no lodging, probably as a result of stronger crown root growth over the longer season.

Himalayan lines are adapted to the short growing season at high altitude by having a very short vegetative development phase and producing very few tillers, so that in these trials they could not take advantage of the longer UK growing season and hence had low grain yields. However, naked lines derived from crosses with stiff-strawed UK varieties such as Static and Tipple were resistant to lodging and had higher grain yield than Lawina.

Table S1. Summary of agronomic traits and β-glucan content of hulled and naked varieties tested in 2009 spring-sown trials. Disease assessment was made at GS 61, and uses the key for NL trials. Data for the Darzau (German) and ICARDA (Syrian) lines are either means of all varieties or the range of all varieties. Haidd Enlli was only grown with fungicide treatment and Line 15 (a naked line derived from a cross between Static (UK hulled) and a naked variety from Pakistan) was only grown with no fungicide treatment (nd = no data).

Variety	**Covered (C) or Naked (N)**	**Ear emergence (days +/- Static)**	**Mildew (1 = no infection, 9 = dead)**	**Rhycho-sporium (1 = no infection, 9 = dead)**	**Lodging (%)**	**Treated Specific Weight (kg/hl)**	**Untreated Specific Weight (kg/hl)**	**B-glucan (g/100g dry matter)**
Optic	C	+3	8	8	3	60.8	47.5	4.0
Static	C	0	1	8	0	57.7	53.3	4.9
Tipple	C	+3	1	8	0	57.2	51	3.9
Haidd Enlli	C	0	nd-	nd	100	63	nd	5.4
Lawina	N	+3	7	6	5	73.4	73.4	5.4
ICARDA (n = 3)	N	-2	8	2-4	20	69.4	70.9	3.0 - 7.0
Darzau (n = 26)	N	-3 to +3	1-8	2-8	60	70.7	72.7	3.0 - 6.2
Line 15	N	+1	1	4	0	nd	70.3	5.0

2.3.4 Prospects for barley as a functional food

Awareness amongst farmers, processors and consumers of the potential of naked barley has increased during this project. Bread made from a mixture of naked barley and wheat flour by a bakery based near Bangor has sold very well and won second place in the national 'True Taste of Wales' competition in 2009. Any future development of a supply chain depends on the development of UK-adapted varieties. Breeding priorities must focus on maximising the health benefits of naked barley in processed food products and at the same time they must address improving production under UK conditions. It should be possible to improve β–glucan and amino acid content in UK naked barley through conventional breeding.

2.4. Key Conclusions

- Naked barley varieties from Europe, Middle East and Asia are low yielding, susceptible to lodging and foliar diseases when grown in the UK
- Establishment rate of naked barley is lower than for hulled barley, however, establishment of 85% can be obtained by reducing combine drum speed at harvest.
- The German naked variety Lawina is not suitable for UK agriculture.
- Pre-breeding screening and selection trials have shown that 'exotic' varieties of naked barley have high β-glucan levels that can be incorporated into higher yielding genetic backgrounds by crossing with UK varieties.
- A pre-breeding programme has identified promising novel varieties from crosses between naked and hulled varieties.
- With appropriate management naked barley lines can produce yields as high as 70% of those of covered varieties in the UK.
- Concentration of β–glucan in barley grains varies due to genetic and environmental factors and changes over time during grain development.

3. TECHNICAL DETAIL

3.1. Introduction

3.1.1. Statement of objectives

The overall goal of this project was to identify and develop strategies that could lead to profitable adoption of naked barley by UK farmers. The aims were

1) To evaluate the crop in a range of agronomic environments in order to elucidate agronomic requirements of naked barley varieties and produce best practice agronomy advice.

2) To develop end uses of naked barley in conjunction with food manufacturers.

3) To identify germplasm suitable for further development.

The objectives of the trials were: i) to determine whether naked barley differs from covered barley in its agronomic management requirements; ii) to asses if it is viable to grow the available Continental naked barley varieties in the UK climate; iii) to act as a pre-breeding screen to evaluate the usefulness of exotic varieties, including those from the Himalayas and Japan, as parents for crossing with UK covered varieties to introduce new variation into the UK barley gene pool; and iv) to give a better understanding of how genotype and environment interact to determine grain β-glucan content.

Joint funding for these four objectives was received from the Welsh Assembly Government (WAG) Supply Chain Efficiencies Scheme. Objective 1 was the major focus for the HGCA-funded work.

3.1.2. Background

Barley is an appealing source of dietary fibre that has a wider range of culinary applications than oats. Unlike oats, it requires no specialist processing and can be harvested and processed with the same equipment as wheat. It can be used as flour for bread, and as a whole grain cereal as flakes or porridge.

In the UK some hulled barley is pearled for food uses, but the bran and endosperm are lost during the pearling process. Naked barley has homozygous recessive alleles at the *nud* locus (chromosome 7H) conferring hulless grain which requires no pearling. All currently cultivated UK barley varieties are hulled.

Malting barley has low β–glucan content compared to naked barley varieties from Europe and Asia. In malting barley, β–glucan content is low (typically 3 - 5%) and β-glucanase activity is high, while in naked barley varieties β–glucan content is high (4.5 - 7%).

Unbranched, mixed-linkage (1,3:1, 4)-β-D-glucans (β-glucan) are key components in the cell walls of the Poaceae (Fincher, 2009). The endosperm of oats and barley are especially rich sources of water-soluble β-glucan (Wood, 2007). The consumption of both oat and barley β-glucan has beneficial effects on glucose metabolism, lipid levels, cholesterol and blood pressure (Anderson et al., 2004) and clinical trials suggest that increased consumption of β-glucan may reduce the risk of insulin resistance and type 2 diabetes in adults (Gary Frost, Personal communication).

The UK lacks suitably adapted high-yielding naked barley varieties, and the health benefits of barley β–glucan need to be clarified. We carried out an assessment of the agronomic properties and grain traits of a range of hulled and naked barley varieties. The Menterra Project at Bangor University (menterra.org) had previously evaluated two German released varieties, Lawina and Taiga, of which Lawina was most preferred by processors. A certified seed crop of Lawina was grown prior to the start of this project. The hulled variety Haidd Enlli ("Bardsey Barley") is a Welsh variety that was grown up to the mid-20th Centaury but is no longer cultivated, and was included in the study as a hulled variety that has not been developed through a modern breeding programme. A collection of naked barley varieties that are used for food included 24 samples from Asia (Pakistan, Nepal, Tibet, Japan and Korea) and a high lysine variety (Hiproly) from Ethiopia. The Asian collection included examples of uzu varieties (brassinolide-insensitive dwarf barleys carrying the *uzu* gene for brassinolide-insensitivity), waxy varieties (low amylose) and hooded barley with very short awns that terminate in a reversed sterile floret. Breeding lines from a German naked barley breeding programme (Darzau lines) and from ICARDA were also tested.

3.2 Materials and Methods

3.2.1. Plant materials

UK hulled varieties: Optic, Tipple and Cocktail were supplied by W. Handley, HGCA; Static was supplied by New Farm Crops, UK (in 2003). German naked varieties: Lawina and 26 naked breeder's lines (coded Darzau 1- 26) were supplied by Cereal Breeding Research, Darzau, Germany; Taiga was supplied by Ackermann, Germany. Three ICARDA naked semi-dwarf 6-row lines (92.321, 93.747, 93.855) were obtained from ICARDA, Syria. A collection of Himalayan accessions (in 2005) and a collection of Japanese and Korean accessions (in 2007) were obtained from the John Innes Centre, Norwich, UK (Table 1). Further information is available at http:www.jic.ac.uk/GERMPLASM/Collections.htm

Table 1. Details of barley accessions from the John Innes Centre (JIC).

Origin	Name	Row	JIC accession	Comments
Japan	Seto Hadaka	6	3709	uzu
Japan	Kitagawa Chobo	6	3717	uzu
Japan	Mochimugi	6	7977	waxy
Japan	Iyatomi Mochi	6	7983	waxy, uzu
Japan	Tokushima Mochimugi	6	8028	waxy, uzu
Japan	Kairyo Shiri Hadaka	6	19255	uzu
Korea	Paik Dong	6	3969	uzu
Korea	Juk Ha	6	3970	uzu
Korea	Na Zai Rai	6	3972	
Korea	Baec-Dong	6	4109	uzu
Korea	Koyane-Chae-Rae	6	20280	
UK	Hen Haidd Enlli	2	3482	hulled
UK	Hen Gymro	2	7053	hulled
Ethiopia	Hiproly	2	7979	
Afghanistan	Hindukusch	2	4439	purple grain
Tibet	Tibet 37	2	4218	black grain
Tibet	Tibet	2	4438	
Afghanistan	Afghan R1169	6	4066	
Bhutan	Sinclair & Long 5125	6	18763	
China	Huang Yen	6	19395	purple grain
China	Men Jun	6	19406	grey grain
Pakistan	Pakistan 23B BN-4	6	6439	grey grain
Pakistan	Pakistan 29 BN-8	6	6500	
Nepal	Nepal A84BN	6	8697	Hooded
Nepal	Nepal 92A BN-1	6	4777	Hooded
Nepal	Nepal 102A BC-5	6	5582	
Nepal	Nepal 15A BC-3	6	5614	Hooded

3.2.2. Field Trials

Trial Dz08

In 2008, a trial of 35 varieties was conducted, without fungicide. The varieties were: the German naked barley varieties Lawina and Taiga; the covered UK control varieties, Cocktail, Optic, Static and Tipple; 26 naked lines from the breeder of Lawina, Cereal Breeding Research Darzau; and three naked six-row semi-dwarf varieties bred by ICARDA, Aleppo, Syria.

Trial Dz09

In 2009, the same 35 varieties were trialled again and a fungicide programme based around triazole and strobilurin chemistry, based on that used in HGCA RL trials was used, alongside an untreated trial.

The experimental designs used in both years were orthogonal row-column designs, with two replicates, from those published by Patterson and Robinson (1989).

Trial Lawina09

In 2009, a factorial experiment tested Lawina sown at four seed rates (300, 350, 400 and 450 seeds/m^2), either untreated or treated with trinexopac-ethyl (Moddus) plant growth regulator (PGR). Experimental design was a randomised complete block design with three replicates.

Trial sequential harvest 09

Four contrasting varieties were sown in 2009 and sequential samples taken to assess the effects of weather damage on β–glucan content. Experimental design was a randomised complete block design with three replicates.

Trials World08 and World09

For the pre-breeding screen of the diversity of naked barley available from Asia a trial of 30 accessions was sown in spring 2008, and of 25 accessions in spring 2009. These were both orthogonal row-column designs with two replicates (Patterson and Robinson, 1989).

Trials Autumn08 and Autumn09

Thirteen of the Asian accessions, which earlier experiments had suggested to have at least a small vernalisation requirement (e.g. Japanese uzu types) or were expected to be winter hardy (e.g. Tibet) were sown in autumn 2008 and 2009, along with the three ICARDA varieties.

Selections from Skardu/Static cross

The possibility of improving the yield and grain quality of naked barley by crossing naked lines with adapted UK varieties was studied using material derived from an earlier DFID funded project. Crosses between naked barley landraces from the Skardu region of northern Pakistan and the UK feed variety Static were made in 2004. Static was chosen for its excellent straw strength and mildew resistance. The Skardu/Static population was advanced to the F_5 generation with no deliberate selection under glasshouse conditions, except the generation F_3-F_4 which was grown under field conditions. The F_5 harvest was divided into two bulks according to row type: two-row bulk (2.4kg) and six-row bulk (3.05kg). In 2007, naked grains were selected from each of the F_5 bulks and sown in small plots in the field without fungicide. Single ears were selected (for uniform, good grain size, no disease symptoms and good stem strength) from these plots in the field in 2007 and advanced one generation over winter in the glasshouse. Twenty-nine lines generated in this way were sown in metre long double rows in the field in 2008. Disease resistance, straw strength and grain quality were assessed again, and eight lines were rejected.

Trial Selections09
Twenty-one selected Skardu/Static lines were sown in plots with no replication (5m x 16 rows x 0.12 inter-row spacing) on 19 March 2009 at a rate of 300 seeds/m^2. Seed in this trial was treated with Raxil-Pro (containing prothioconazole, tebuconazole and triazoxide). The trial was in blocks of 4 plots, with Static as a control in each block.

3.2.3. Field operations

All agronomy trials were carried out at Bangor University's Henfaes Research Centre in 2008 and 2009. Previous cropping was rotational pasture in both years. Seedbeds were prepared by ploughing and power harrow, and Cambridge rolled after drilling. Sowing depth was approximately 30mm. All plots (5m x 16 rows) were sown using a Wintersteiger drill, with 12cm inter-row spacing unless otherwise stated.

Spring-sown trials were sown on 6 March 2008 and 19 March 2009. Seed rate for naked varieties was 350 seeds/m^2 in 2008 and 425 seeds/m^2 in 2009. Seed rate for covered varieties was 350 seeds/m^2 in both seasons.

Autumn-sown trials were sown on 26 September 2008 and 8 October 2009. Seed rate was 350 seeds/m^2.

For determination of establishment, plants were counted as soon as it was judged that emergence was complete, at the first leaf emerged stage (GS11 [GS = growth stage, referring to the Zadoks decimal system outlined in Tottman et al., 1979]). A 250mm marker was placed at random within the central 10 rows and plants counted either side. Five sampling points were used in each plot.

To measure over-winter survival of autumn-sown plots, plants were counted in March using the same protocol as the establishment counts, above. Over-winter survival was expressed as the number of plants in March as a percentage of the number of seedlings established in autumn.

In both years a total of 100-120kg/ha N was applied, split approximately 50:50 between emergence and approximately 3-4 weeks later at the start of tillering (GS21). P and K were applied at 40-50kg/ha as 17:17:17 N P K compound fertiliser with the first N application. Weed control was achieved using commercial formulations of metsulfuron-methyl and mecoprop-P applied at the manufacturers' recommended dose and timings. The fungicide programme was based around triazole and strobilurin and applied at the standard T1 (GS30-31) and T2 (GS39-49) timings for spring barley. Products were used at the manufacturers' full dose recommendations and no significant foliar disease was observed on treated plots in any season.

Foliar disease was scored at GS61 and GS75 using the VCU scoring system (http://www.fera.defra.gov.uk/plants/plantVarieties/nationalListing/documents/protocolGrain09.pdf, accessed in 2008 & 2009). Scores were on a scale of 1-9, for disease progression: i.e. 1 = no disease observed, 9 = no leaf green area remains. Diseases observed and scored were powdery mildew (*Blumeria graminis*) Rhynchosporium (*Rhynchosporium secalis*) and brown rust (*Puccinia hordei*). The proportion of green area remaining was also assessed.

Lodging was assessed at GS61 and GS75. The proportion (%) of each plot lodged (stem displaced > 45^o) was recorded.

In 2009, a sequential harvest trial was conducted to determine the effect of weather damage on β-glucan and HFN. Sequential 'grab samples' were made on 27 July, 5 August, 13 August, 21 August and 9 September. Samples were taken between 9 and 11am. Sample size was 25-35 ears 'grabbed' at random, and cut at the collar. Ears were air-dried, hand threshed and the grains hand cleaned. By the 13 August, large areas of each plot were lodged following a squall on 10 August. Separate samples were taken from the lodged and standing areas on 13 August and 21 August. By the 9th September the plots were completely lodged.

3.2.4. On-farm trials and seed crop of Lawina

2008

Two neighbouring farmers on the Llyn Peninsula, North Wales, grew commercial crops of Lawina following agronomy guidelines that were updated annually (Appendix 1). Plots were 4 ha and 2 ha and seed rate was 200kg/ha. Fungicide was used and N applied early with some in the seed bed. Bad summer weather delayed harvest until mid-September. A seed crop (1.5 ha) of Lawina was sown at Henfaes on 6 March 2008.

2009

Two farmers grew Lawina in 2009 following the guidelines in Appendix 1. A farmer on Anglesey grew 4 ha and a farmer in Cheshire grew 8ha. Seed rate was 425 seeds per m^2. A seed crop (2 ha) of Lawina was sown at Henfaes on 31 March 2009.

3.2.5. Laboratory germination tests (pre-sowing)

Germination of seed lots was tested at Henfaes prior to sowing and because of large differences between varieties seed rates were adjusted so that the rates quoted are the number of viable seeds per m^2. Germination tests in Petri dishes were conducted on Whatman No. 1 filter paper, moistened with deionised water. Germination was measured after 48 hours and again after 7 days in an incubator at 18 ºC.

Germination tests were conducted on seed harvested from plots of Lawina grown at IBERS in 2009 and harvested using different drum speeds. Germination was tested in trays of sand.

3.2.6. Grain handling and testing

Grain was harvested using a Hege plot combine harvester; air-dried and cleaned using a tabletop cleaner (Clipper 400, Seedburo) with circular sieve apertures of 6 mm on upper screen and 3 mm on lower screen. Specific weight was measured using a Sinar AP 6060 Moisture Meter and chondrometer tube.

Grain was milled using a hammer mill with 0.8mm screen (Mill 31000, Perten Instruments). Dry matter of flour was determined by weighing of samples (approx 0.5 g) after oven drying at 80 °C for 20 hours.

β–glucan (1,3:1,4-ß-D-glucan) was measured using β-Glucan (Mixed Linkage) Assay Kits (Magazyme, Co. Wicklow, Ireland) based on the McCleary method (McCleary & Glennie-Holmes, 1985) with no deviation from the kit instructions. Sprouted grains gave high absorbance readings for the reaction blanks with the standard Megazyme method for barley flour. Samples harvested on the 9th September 2009, as part of the sequential harvesting experiment, which contained a high proportion of sprouted grains were assayed using the malt method (EBC Method 4.16.1). Grain β–glucan concentration was calculated as for dry matter (β–glucan g/100 gDM).

Grain nitrogen content was determined by a semi-automated Kjeldahl 2300 distillation unit (Foss UK, Warrington, England) using a method modified from the Agricultural Development and Advisory Service (1986). Amino acid content of grain was analysed at Nottingham University with a protocol based on that of Mason et al. (1980).

3.2.7 Data analysis

All statistical analysis was carried out using Genstat Twelfth Edition (Release 12.1, VSN International, 2009). Row-and-column designs were analysed using the appropriate REML analysis. Complete randomised block designs were analysed using ANOVA. Multi-environment trials (MET) allowed analysis of cultivar x environment (Smith et al., 2005) using AMMI (Additive Main effects and Multiplicative Interaction) for some traits. AMMI uses a Principle Components Analysis (PCA) to analyse the cultivar x environment interactions matrix generated by ANOVA (Gauch, 1992), and allows the cultivar means to be plotted against the first principle component (PC1) as a measure of stability.

3.3 Results

3.3.1. Germination and establishment

In laboratory tests naked barley varieties germinated faster than covered varieties (data not shown). Final germination percentage of naked barley was generally lower than that of covered barley and crooked coleoptile growth was common.

Increasing combine threshing drum speed at harvest decreased germination percentage of naked barley (Figure 1). Data presented in Figure 1 below are single samples. Further work using different naked barley varieties, harvest conditions and types of harvester is required.

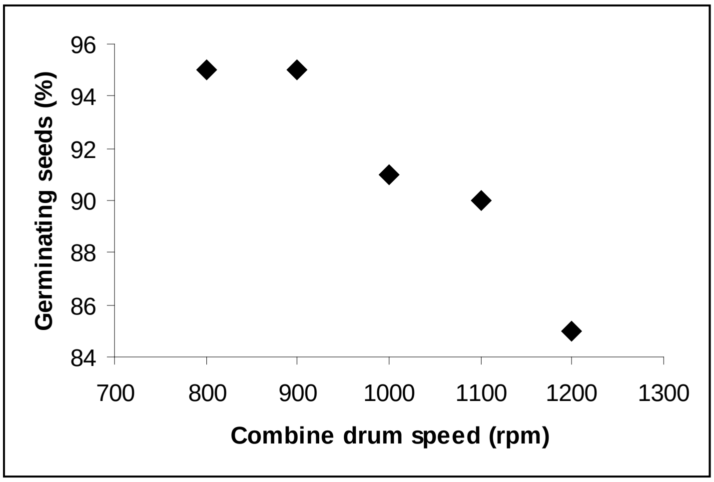


Figure 1. Germination percentage of Lawina naked barley harvested at different combine drum speeds.

A laboratory test was carried out on filter paper using the naked variety Lawina to compare untreated seed and seed treated with Raxil-Pro. This showed that the seed dressing treatment decreased germination from 85% to 65%.

In the field there were large differences in establishment between varieties. Establishment was consistently lower in naked than in hulled varieties. Establishment rates for Syrian and Asian varieties were higher after autumn sowing than after spring sowing (Table 2). Soil temperatures at autumn sowing were substantially higher than in spring (Table 3).

Dormancy is likely to be a factor influencing germination. Seed tested prior to autumn sowing in the same calendar year in which it was harvested had generally lower germination rates than seed from the same seed lot re-tested the following spring. Japanese accessions showed greater dormancy than Himalayan accessions.

Table 2. Percent establishment (number of plants established per 100 viable seeds tested in the lab) in spring and autumn trails at Henfaes. The seed rate for each variety was calculated from prior lab germination tests. 'Hulled' are UK hulled varieties Optic, Static, Tipple and Cocktail. 'European Naked' are German varieties Lawina, Taiga and Darzau lines 1 - 26; 'ICARDA', 'Himalayan' and Japan & Korea' refer to groups of naked accessions, see section 3.2.1. Mean, standard deviation (SD), min and max are given for each group.

				Establishment (%)			
Season	**Year**	**Collection**	**N**	**Mean**	**SD**	**min**	**max**
Spring	2008	UK Hulled	4	103*	9.5	88	113
		European Naked	28	58	18.3	14	87
		ICARDA naked	3	63	1.4	61	65
		Himalayan naked	16	89	18.9	59	115
		Japan & Korea naked	11	93	14.7	71	115
	2009	UK Hulled	4	136*	4.3	131	142
		European Naked	28	83	15.6	33	116
		ICARDA naked	3	59	18.1	34	77
		Himalayan naked	14	94	21.0	63	139
		Japan & Korea naked	8	86	30.2	43	143
Autumn	2008	ICARDA naked	3	71	3.4	67	75
		Himalayan naked	7	121*	25.9	84	168
		Japan & Korea naked	6	119*	13.1	106	146
	2009	ICARDA naked	3	115*	24.3	92	149
		Himalayan naked	7	128*	14.8	108	152
		Japan & Korea naked	6	118*	29.4	89	177

*Field germination rate was higher than lab germination rate.

Table 3. Sowing date, min and max soil temperature at 10 cm during sowing and crop establishment (data are means for 28 day period, taking sowing date as day 7) and the cumulative rainfall and sun hours for eight weeks up to harvest when the seed was grown (i.e. the previous summer).

Season	Sowing date	Soil temp. at 10 cm (deg.C)		Rainfall (mm)	Sun hours
spring		**Min**	**Max**		
2008	06/03/2008	5.4	7.7	217.4	263
2009	19/03/2009	7.4	8.7	135.4	259
autumn					
2008	26/09/2008	12.7	14.1	135.4	259
2009	08/10/2009	12.9	13.6	127.8	381

3.3.2. Foliar disease

Foliar disease (Table 4) was scored in experiments that received no fungicide: DZ08 and DZ09 (UT treatment). Only powdery mildew was scored in 2008 because the other diseases were observed to be at very low levels. Brown rust developed quickly in 2009, during hot humid weather in the last week of June. Levels of Rhynchosporium were high in 2009, especially in Optic, Static and Tipple. Mildew appeared earlier in 2008 than 2009 and the level of infection in 2008 at GS61 was slightly higher than 2009. In both years mildew infection levels were high on all but the most resistant varieties. Lawina, Taiga, all three ICARDA lines and some of the Darzau lines were as susceptible to powdery mildew as Optic. Some Darzau lines (for example lines 6 and 22) possessed a higher degree of resistance, equivalent to Tipple and Static.

Table 4. Disease scores of 4 hulled and 31 naked spring sown barley varieties. Powdery mildew, Rhyncosporium and brown rust were scored on a 1-9 scale where 1 shows least symptoms and 9 shows most symptoms. In 2009 disease scores were taken at two growth stages: GS61 (anthesis) and GS75 (medium milky ripe).

	2008	2009							
Genotype	Mildew GS61	Mildew GS61	Mildew GS75	Rhyncho-sporium GS61	Rhyncho-sporium GS75	Brown rust GS61	Brown rust GS75	Green area (%) GS61	Green area (%) GS75
Optic (hulled)	8	5	8	4	8	1	2	81	8
Static (hulled)	2	1	1	5	8	2	1	97	17
Cocktail (hulled)	8	4	7	3	5	3	2	63	20
Tipple (hulled)	6	1	1	4	8	2	2	95	16
Lawina	8	6	7	1	6	2	3	72	16
Taiga	8	4	7	1	4	2	2	93	38
Darzau 1	5	4	5	2	5	2	4	85	9
Darzau 2	3	3	3	1	3	2	6	100	24
Darzau 3	4	4	5	2	4	2	2	90	39
Darzau 4	4	4	4	3	5	1	2	81	37
Darzau 5	6	1	6	5	5	4	9	82	2
Darzau 6	2	2	2	1	4	2	5	97	29
Darzau 7	5	4	4	2	5	2	5	98	20
Darzau 8	4	1	2	1	3	2	2	93	36
Darzau 9	3	2	7	2	2	2	3	74	0
Darzau 10	6	2	4	2	5	3	4	94	39
Darzau 11	6	3	4	5	6	2	3	85	16
Darzau 12	6	2	2	5	8	2	4	84	21
Darzau 13	6	4	6	3	6	1	2	74	39
Darzau 14	4	3	4	3	5	2	4	94	34
Darzau 15	6	3	7	1	4	2	2	91	40
Darzau 16	4	1	8	3	7	1	3	100	9
Darzau 17	8	7	8	4	3	1	1	53	2
Darzau 18	8	6	8	2	3	2	2	50	0
Darzau 19	7	6	7	4	7	2	3	74	8
Darzau 20	7	5	8	1	6	2	2	78	0
Darzau 21	8	6	8	2	6	2	4	70	7
Darzau 22	6	2	4	2	4	2	3	91	31
Darzau 23	7	5	6	2	6	2	3	65	9
Darzau 24	7	3	5	2	5	2	3	76	29
Darzau 25	1	2	8	2	6	2	2	88	0
Darzau 26	5	4	8	2	6	2	4	86	0
ICARDA 92.321	8	5	8	2	3	2	1	76	51
ICARDA 93.747	8	5	8	1	2	2	3	71	28
ICARDA 93.855	8	6	8	1	4	2	3	65	4

3.3.3. Grain yield

Yields of all naked varieties were low in 2008, both in comparison to the covered controls grown in the same trial, and to the same naked varieties grown untreated in 2009 (Table 5). In 2009, differences between fungicide-treated and untreated yields were significant (Table 5), as was the interaction between variety and treatment. The grain yields of the UK covered control varieties were substantially increased by the use of fungicide, but Lawina and many of the Darzau lines showed very little yield response.

Table 5. Grain yield (t/ha) and specific weight (kg /hl)for European and ICARDA naked barley varieties and four UK hulled varieties in 2008 and 2009 (Trials Dz 08 and Dz09). T = fungicide treated trial; UT = untreated trial.

	2008		2009			
Genotype	Yield (t/ha)	Specific weight (kg/hl)	Yield (t/ha)		Specific weight (kg/hl)	
	UT	UT	T	UT	T	UT
Optic	4.38	64.4	8.62	4.76	60.8	47.5
Static	6.33	62.9	9.72	6.83	59.7	53.3
Cocktail	3.77	63.1	8.82	7.07	61.7	58.2
Tipple	6.07	62.6	8.88	4.93	57.2	51.0
Lawina	2.02	73.4	4.34	4.26	73.5	73.4
Taiga	1.96	70.2	5.78	4.92	72.3	71.1
Darzau 1	1.57	69.3	5.47	4.62	70.1	69.9
Darzau 2	3.03	73.7	5.00	4.61	73.1	73.3
Darzau 3	3.35	75.4	5.41	5.08	73.8	71.5
Darzau 4	2.30	74.6	5.60	4.97	74.3	72.7
Darzau 5	2.57	73.0	6.89	4.05	71.8	66.5
Darzau 6	2.98	72.8	4.21	3.51	69.0	64.8
Darzau 7	3.06	73.8	5.60	4.59	75.7	72.6
Darzau 8	2.05	70.3	5.11	4.98	68.4	68.5
Darzau 9	2.71	70.7	5.41	3.94	72.1	70.9
Darzau 10	2.57	74.3	4.99	5.11	70.9	72.0
Darzau 11	2.93	73.9	6.59	5.63	74.0	73.7
Darzau 12	1.37	73.8	6.00	4.42	75.8	71.2
Darzau 13	2.32	72.8	5.23	4.77	71.8	71.6
Darzau 14	3.38	73.3	5.70	4.55	72.4	68.3
Darzau 15	3.22		6.55	4.73	72.5	68.3
Darzau 16	2.47	72.3	5.24	5.78	74.9	73.1
Darzau 17	1.25	71.6	5.94	3.78	74.3	71.5
Darzau 18	1.39	71.1	5.41	4.24	73.9	72.9
Darzau 19	1.40	72.7	6.27	5.57	74.4	73.8
Darzau 20	1.98	72.3	6.26	5.38	74.7	72.1
Darzau 21	1.73	70.2	5.31	4.70	73.2	70.4
Darzau 22	3.00	70.9	3.87	4.02	71.6	71.6
Darzau 23	2.26	74.3	5.04	3.99	73.3	68.4
Darzau 24	2.55	70.6	5.12	4.80	70.9	71.0
Darzau 25	2.87	71.0	4.32	3.24	68.7	65.0
Darzau 26	3.01	74.1	5.03	6.05	75.1	73.8
ICARDA 92.321	2.40	71.8	6.19	4.68	73.7	72.5
ICARDA 93.747	2.06	70.4	4.20	3.92	70.7	71.3
ICARDA 93.855			6.26	3.51	68.3	64.4
Treatment means	2.72	71.3	5.83	4.74	71.1	68.6

Autumn sowing did not increase yields of the Himalayan or ICARDA naked barley accessions, but the semi-dwarf 'uzu' accessions 'Iyatomi Mochi', 'Tokushima Mochimugi', 'Paik Dong' and 'Juk Ha' were much higher yielding when sown in autumn than spring (Table 6).

Table 6. Winter survival for autumn-sown naked barley varieties and the effect of time of sowing on their grain yield and β–glucan content. All data is for harvests in 2009. No data (nd) indicates that variety was not sown in Spring 09 due to poor yield from Spring 08 plots.

		Winter survival (%)		Yield (t/ha)		Beta glucan (%)	
Genotype	Vernalisation	2008	2009	winter	spring	winter	spring
ICARDA 92.321	N	66	48	7.02	6.29	6.2	5.9
ICARCA 93.747	N	88	71	3.95	4.15	6.3	7.4
ICARDA 93.855	N	59	79	6.17	6.2	5.7	3.2
Hiproly	N	54	92	3.88	3.31	6	5.3
Hindukusch 62	Y	73	74	4.24	4.69	6	5.7
Tibet 37	N	66	69	4.7	4.09	6.3	4.7
Tibet	N	53	53	4.53	3.99	6.3	5.6
Bhutan	Y	54	49	5.2	3.86	6.1	6.8
Nepal A84BN	Y	65	54	3.76	2.43	5.8	5.5
Nepal 102A BC-5	Y	100	61	3.25	3.89	6.1	6.1
Kitagawa Chobo	N	57	61	4.52	5.08	5.1	5.8
Iyatomi Mochi	Y	68	56	5.15	nd	5.2	nd
Tokushima-Mochimugi	Y	89	60	6.36	nd	5.7	nd
Paik Dong	N	57	74	6.27	4.12	4.9	5.0
Juk Ha	N	53	73	7.88	4.63	5.3	5.4
Koyane-Chae-Rae	Y	54	83	4.72	nd	4.3	nd
Mean		68.6	63.6	4.8	4.4	5.9	5.6
Standard deviation		15.1	12.8	1.1	1.2	0.4	1.1

3.3.4. Grain quality

Results from the Dz08 and Dz09, World08 and World09 trials showed that naked barley varieties consistently had higher specific weights (kg/hl) than covered varieties grown in the same season. In contrast, the untreated specific weights for the four covered varieties in 2009 were very low. Differences in specific weight between genotypes and treatments were significant (Table 5), and the interaction between genotype and treatment was significant. AMMI (Figure 2) showed that specific weights of naked grain accessions were more stable between environments than those for covered accessions: this was the case both in comparing year (08 versus 09) and fungicide treatment (T versus UT in 2009; Figure 2a). The specific weight of Optic was very unstable, in contrast to that of Cocktail, which was very stable (PC 1 value close to zero).

a

b

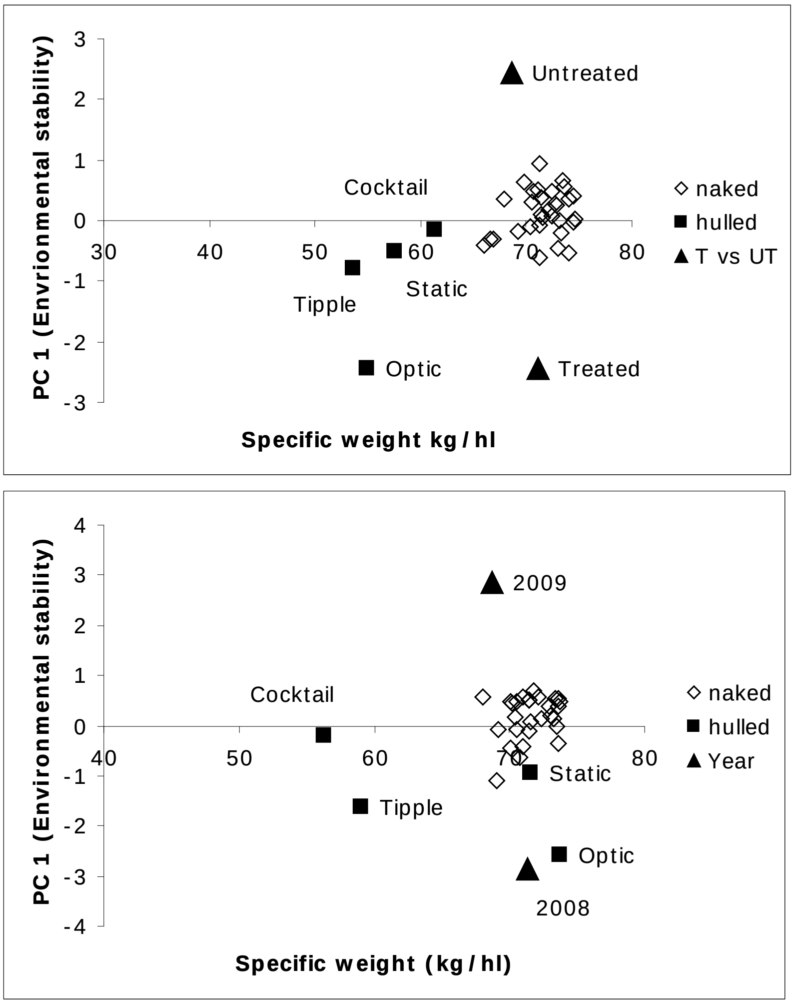


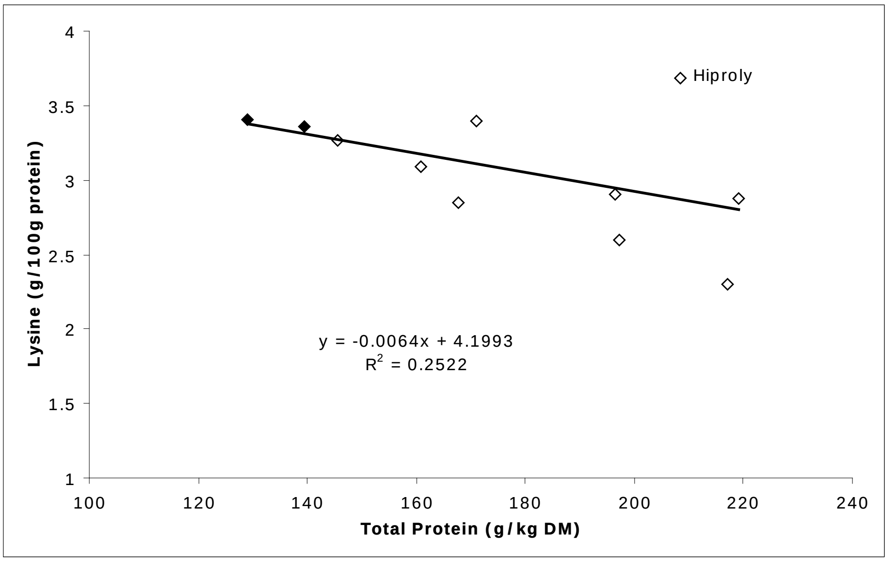
Figure 2. AMMI plots for specific weight of 31 naked and 4 hulled barley accessions (see Table 4 for genotypes) grown (a) with and without fungicide in 2009, and (b) without fungicide in two years 2008 and 2009. Principal component 1 (PC1), environmental stability between treatments is plotted against the specific weight of plots.

Table 7. Grain β–glucan content (g per 100g) with and without fungicide treatment in 2009 for four UK hulled, 28 German and 3 Syrian (ICARDA) varieties.

Genotype	Fungicide treated	Untreated
Optic (hulled)	4.0	5.1
Static (hulled)	4.9	4.3
Cocktail (hulled)	4.2	4.1
Tipple (hulled)	3.9	4.9
Lawina	5.5	4.9
Taiga	5.0	5.1
Darzau 1	5.3	5.3
Darzau 2	5.0	5.4
Darzau 3	5.1	6.5
Darzau 4	5.8	5.9
Darzau 5	6.3	4.9
Darzau 6	4.5	4.9
Darzau 7	6.3	7.7
Darzau 8	3.6	6.3
Darzau 9	4.2	5.8
Darzau 10	3.7	5.2
Darzau 11	3.5	4.5
Darzau 12	5.3	7.2
Darzau 13	4.4	5.4
Darzau 14	3.0	5.4
Darzau 15	4.3	5.7
Darzau 16	5.4	4.7
Darzau 17	4.3	5.3
Darzau 18	3.9	5.8
Darzau 19	3.8	5.2
Darzau 20	3.2	4.6
Darzau 21	4.0	6.3
Darzau 22	3.5	4.1
Darzau 23	3.7	5.4
Darzau 24	3.8	4.3
Darzau 25	4.4	4.7
Darzau 26	4.4	4.8
ICARDA 92.321	5.9	5.0
ICARDA 93.747	7.4	6.6
ICARDA 93.855	3.2	5.9
Mean	4.5	5.4
Standard deviation	0.90	0.85

REML analysis of the fungicide experiment showed significant differences between genotypes ($P = 0.004$) in β–glucan content, and between fungicide treatments ($P = 0.049$), but the genotype x fungicide interaction was not significant ($P = 0.375$) (Table 7). The mean β–glucan concentration was higher in the untreated plots (5.4 vs 4.5 g/100 g DM), although some genotypes (e.g. Darzau 5, Static and ICARDA 92.747) had higher β–glucan when fungicide was used. When AMMI was used to measure environmental stability, in contrast to the situation for yield and specific weight, the naked genotypes were generally less environmentally stable in their β–glucan content than the hulled controls.

a



b

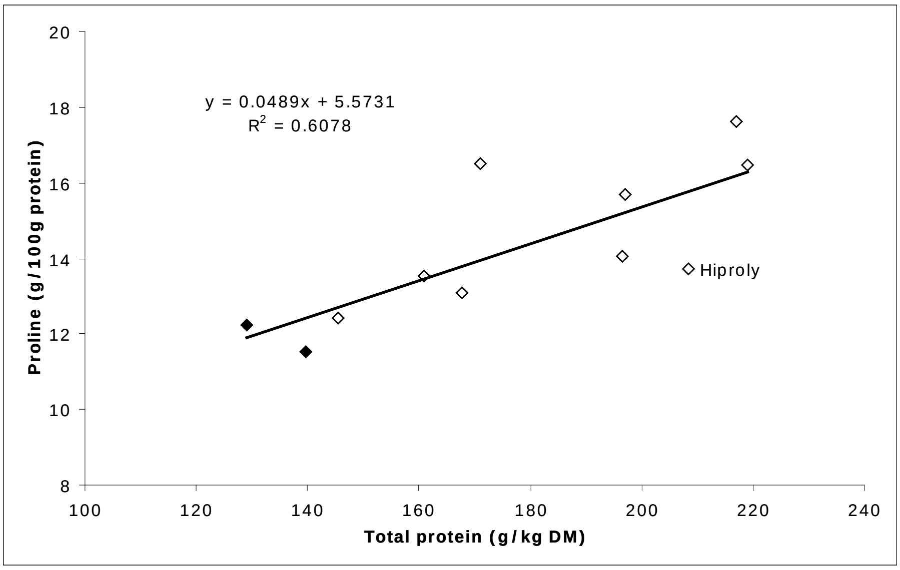


Figure 3. Regressions of amino acids (a) lysine and (b) proline against total protein in barely grains of 2 hulled (closed symbols) and 9 naked (open symbols) varieties.

The amino acid profiles varied for varieties but the profiles of the UK hulled varieties Static and Tipple were almost identical. Hiproly had more lysine, threonine and methionine, and it was an outlier in regressions against total protein (Figure 3). Lysine (Figure 3a) and threonine content were negatively correlated with total protein while proline was positively correlated to total protein (Figure 3b). A PCA (not shown) showed proline accounts for the greatest amount of variation.

3.3.4.1 Effect of delayed harvest on β–glucan and N content

Sequential harvesting of grain from two naked and two covered accessions revealed that β–glucan concentration increased linearly over the first three sampling dates and then declined (Figure 4a). HFN followed a similar pattern (Figure 4b). Differences in grain β–glucan concentration between cultivars were not significant (P = 0.581). For clarity, only β–glucan data for standing areas of the plots are shown (Figure 4), but grain from lodged areas had less β–glucan than the standing areas (significant on 13 August, not significant on 21 August).

A B

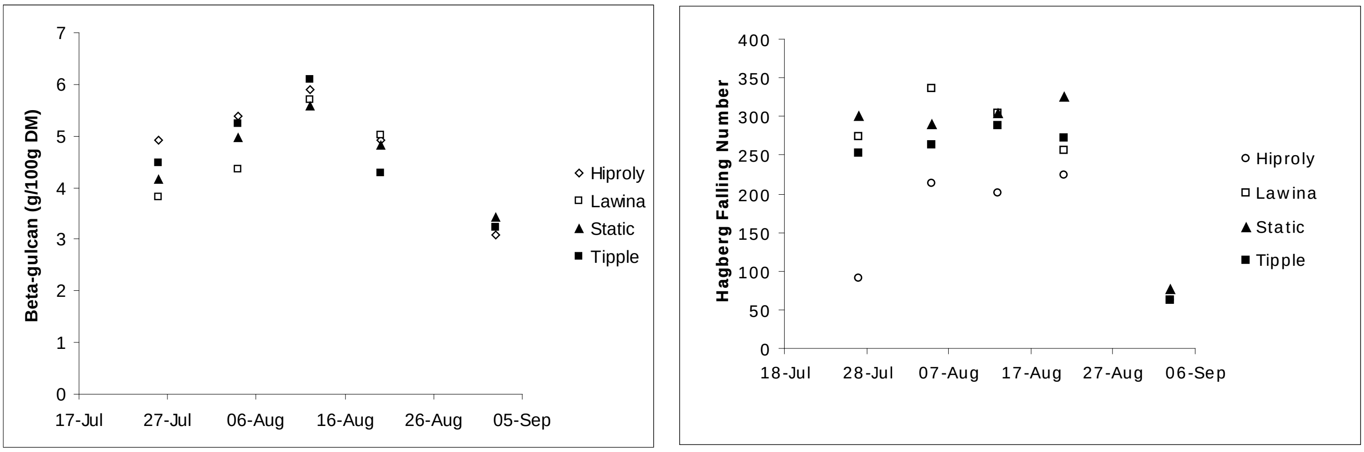


Figure 4. **Sequential harvest dates in 2009 plotted against (A) B-glucan (g/100g w/w) S.E.D: sample date = 0.21., cultivar = NS and (B) Hagberg falling number of four spring barley cultivars from 'grab samples' of ears. Closed symbols = hulled cultivars, open symbols = naked cultivars. By 9 Sept the all plots were 100% lodged.**

There were significant differences between sampling dates and cultivars in grain dry weight (data not shown). Grain N content varied significantly between cultivars (P<0.001) with Hiproly (2.7% N) having the highest N concentration. There was no clear pattern in grain N concentration over time (data not shown).

3.3.5. Lodging

Across all trials the naked barley accessions were more prone to lodging than the UK covered controls. In the Dz09 trial the four UK controls did not lodge under either treatment, whereas fungicide treatment significantly (P = 0.001, Figure 5) increased lodging of the European naked barley lines. This effect was not seen for the ICARDA lines, and two of them did not lodge. In the World08 and World09 trials the Himalayan and Japanese naked barley accessions showed high levels of lodging. Of the Himalayan accessions, 6-row types (e.g. Men Jun and Afghan 1169) were very susceptible to lodging, whilst 2-row accessions (e.g. Tibet37 and Hindukusch) had similar straw strength to Lawina. Japanese accessions tended to root lodge and the Himalayan 6-rows had very weak, thin-walled stems, but root and stem lodging were not scored separately. In the Dz09 trial, fungicide appeared to decrease pre-harvest brackling in UK covered and European naked lines. However, because many Darzau lines were already lodged before brackling occurred it was impossible to make accurate measurement and statistical analysis of brackling.

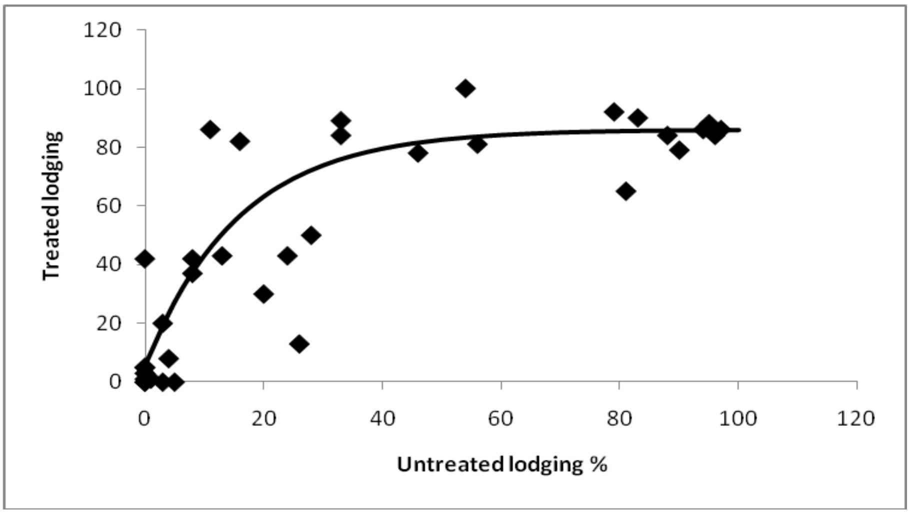


Figure 5. Percent of plot area lodged (>45°) recorded at GS75 in trial Dz 09 showing fungicide-treated plotted against untreated. Curve fitted by Genstat non-linear regression, $y = 86.05-80.25(0.9539^{x})$, P <0.001, variance accounted for = 74.3%.

3.3.6. Effect of seed rate and PGR on Lawina

A factorial experiment to test the effects of seed rate and PGR on Lawina showed that Moddus (trinexapac-ethyl) reduced straw length by 6 cm (P = 0.015) (Table 8). There was a small, non-significant, increase in ear density due to Moddus (data not shown). Differences in yield and grain specific weight between the sowing densities were not significant (P = 0.134 and P =0.221, respectively) but Moddus increased yield by approximately 0.5 t/ha (P = 0.047), and increased specific weight by around 1 kg/hl (P = 0.043). There was no significant interaction between sowing density and PGR for any variable measured.

Table 8. Grain yield (t/ha), grain specific weight (kg/hl) and crop height (cm) of naked barley, cv. Lawina, at four sowing densities (300,350, 400, 450 seeds /m^2) and with (M) or without (U) PGR (trinexapac-ethyl, Moddus).

Treatment	Level	Yield (t/ha)	Specific weight (kg/hl)	Crop height (cm)
Seed rate	300	4.29	76.53	96.9
	350	4.58	76.00	94.5
	400	4.99	77.58	91.9
	450	4.97	76.68	95.9
PGR	M	4.95	77.27	91.8
	U	4.46	76.13	97.8

3.3.7. Selected naked Static/Skardu Lines

Several lines from the cross between Static (hulled) and Skardu (naked) performed well and could be useful for further testing or in breeding. Line 4 had short stiff straw, good resistance to mildew and a high concentration of β-glucan. Line 15 had a very impressive ability to tiller and short and stiff straw similar to Static. However, in this trial it had very poor establishment. Line 20 showed a high level of vigour and would be a good candidate to back-cross with further UK varieties as a source of vigour and naked grain without such a large agronomic disadvantage as Skardu. The yield data in this trial should be treated with caution due to the very poor establishment, which was believed to be an adverse effect of seed treatment.

Table 9. Agronomic traits of 21 naked barley lines selected from the Static x Skardu cross, sown in March 2009 and not treated with fungicide.

Line	Plants /m2	Days to ear	Ears /m2	Ears /plant	Mildew (GS 75)	Rhynco-sporium (GS 75)	Brown rust (GS 75)	% Green area (GS 75)	Lodging (%)	Yield (t/ha)	B glucan (%)	SpWt (kg/hL)
2	80	88	263	3.3	5	4	3	75	0	3.53	5.2	71.3
4	110	82	430	3.9	1	4	3	60	0	3.97	6.5	67.0
5	133	75	446	3.3	1	4	3	80	50	3.55	5.5	69.7
6	130	75	263	2.0	1	4	3	70	5	3.67	6.3	70.0
7	93	89	396	4.2	1	3	2	90	30	2.93	5.0	70.0
10	97	85	513	5.3	1	3	4	80	10	3.38	6.0	69.7
12	97	83	780	8.1	1	5	4	50	100	4.29	5.2	68.1
14	103	78	813	7.9	4	5	4	50	100	3.48	5.6	70.7
15	30	89	613	20.4	1	4	4	80	0	2.79	5.0	70.3
16	103	81	546	5.3	4	4	3	60	100	3.09	4.6	67.0
18	143	71	446	3.1	1	4	2	50	80	4.11	5.1	70.7
19	30	81	263	8.8	1	4	4	80	10	2.70	5.9	65.6
20	147	81	738	5.0	2	4	3	70	90	3.68	5.5	66.6
21	77	88	571	7.5	1	4	3	70	90	2.09	5.7	68.9
22	137	74	771	5.6	1	4	3	20	50	4.94	5.4	71.0
23	210	75	463	2.2	1	4	3	40	80	3.23	5.9	67.9
24	223	75	630	2.8	1	3	3	60	0	3.73	5.2	70.0
25	73	78	413	5.6	1	4	3	70	0	3.07	5.3	68.3
26	17	91	330	19.8	6	4	4	60	0	2.31	4.9	62.6
29	40	87	396	9.9	1	4	4	50	90	3.89	5.6	64.7
31	43	81	430	9.9	1	5	5	40	80	4.16	5.0	66.3
Static	308	88	743	2.4	1	5	1	80	0	7.46	4.3	55.3

3.3.8. On-farm plots of Lawina

2008

Seedling growth was poor in the cold spring, and a dry period in May restricted tillering. June and July were dull with little sunshine. Lawina showed yellowing of leaf tips, an undesirable characteristic trait of the variety which concerned the farmers. Heavy rain during August delayed harvest until mid-September, by which time the crops were heavily brackled and a substantial proportion of ears and grain shed. Yields were 2 t/ha. Some grain was dried and approximately 5 t was sold for milling to Pobty Cae Groes, with the rest used for animal feed.

The seed crop at Henfaes showed all the problems seen by the farmers and yielded 2.5 t/ha.

2009

To compensate for poor seed quality after the wet harvest of 2008 a higher seed rate was used, but crop establishment was still poor. Although the crops looked significantly better by ear emergence than in 2008, yields were still disappointing at 2.5 t/ha and neither crop was suitable for human consumption.

The seed crop at Henfaes was of much better quality than 2008. Drilling was delayed until April and establishment was better in the warmer soil. Metrafenone (Flexity, BASF) was used at the start of tillering (Zadoks GS 21), in addition to the T1 and T2 triazole and strobilurin fungicides and provided good control of mildew. Trinazopac-ethyl (Moddus) was used, and appeared to strengthen the stem base, although the crop was still taller than conventional spring barley. Despite improved crop establishment, ear population was still much lower than would be expected in a UK variety. Lawina tends to self-shade due to large broad leaves, limiting ear population. Yield was 3.7 t/ha. By decreasing the combine drum speed to c.700 rpm, seed quality was maintained. Germination was >85%.

3.4. Discussion

3.4.1. Agronomy of naked barley

All conclusions concerning the agronomy of naked barley should be made with the caveat that there are no varieties of naked barley specifically developed for the UK: comparisons are between UK-adapted hulled barley and non UK-adapted naked barley. The Lawina and the Darzau lines were bred in Germany for organic growing on sandy soils, and are generally taller with longer, broader leaves for enhanced competition with weeds. When grown with fertilizer and fungicide, lodging became a serious problem in these lines. The Himalayan accessions were also tall and 'leafy' and suffered from high rates of lodging. Additionally, these had a very short vegetative phase of development, producing very few tillers, as an adaptation to a short growing season. This limited ear population and grain yield when grown in the UK, as they were unable to take full advantage of the longer growing season. Therefore, although the naked barley varieties tested were generally lower yielding, more susceptible to disease and lodging than modern UK covered barley varieties, this is most likely to be due to lack of adaptation, rather than traits linked to the naked grain type.

For Lawina, there was a much bigger yield response to PGR than to fungicide. This is due to Lawina's unusually (compared to UK varieties) tall straw and large leaves.

3.4.2. Establishment

Establishment rates of naked barley were generally lower than for covered barley. Plant populations were lower, even though seed rates were increased to compensate for the lower germination rate of naked barley.

Germination rates of both naked and covered barley were lower for the seed produced in the wetter summers of 2007 and 2008 than that produced in 2009.

Establishment appeared to be influenced both by soil temperature after sowing and weather conditions before the seed was harvested. The poorest establishment was in spring 2008, in a cold wet seedbed using seed produced in the very wet summer of 2007.

The finding that there were no significant differences in grain yield between seed rates in the Lawina 09 trial, suggests that the lower ear population and yield of Lawina and other Darzau lines were not just due to poorer crop establishment. The long, broad leaf lamina, intended to shade weeds in an organic system, are the antithesis of the short, erect leaves that are the ideotype for an efficient canopy. A degree of self-shading thus restricts the potential ear population of Lawina.

Poorer establishment of naked barley has been reported in Australia (Box et al., 1999) and Canada (Choo et al., 2001), so the finding of poor establishment in UK conditions suggests that it is a general problem with naked barley. However, Himalayan accessions of naked barley had a consistently higher rate of establishment than the European naked barleys.

Seed treatment with Raxil-Pro (a standard seed dressing for spring barley) appeared to reduce the establishment of the naked lines selected from Skardu x Static when sown in March 2009. Untreated seed from the same lines harvested from the same plots in 2008 was sown in April 2009 and establishment increased to 85%. Seed dressing treatment decreased germination from 85% to 65% in Lawina in laboratory tests. Treated seed was also slower to germinate and showed weaker coleoptile growth, suggesting that the effects on establishment in the field could be greater.

Therefore it is strongly recommended not to use seed treatment of any kind on naked barley until further research is done to determine the extent of the problem.

Data from an ongoing PhD project at Bangor to investigate the establishment issues suggest that the higher establishment rate of Himalayan naked barley is linked to longer coleoptiles. In field trials in 2010, some of the naked-grain Skardu-Static (Himalayan x UK) had coleoptile growth as strong as the best Himalayan accession, Tibet 37, but also included the mildew resistance and straw strength of Static. Thus there is good potential that it will be possible to breed naked barley with acceptable rates of crop establishment and full agronomic adaptation to the UK.

3.4.3. Disease

The disease susceptibility observed in many naked lines was probably due to a lack of the resistance genes found in modern UK cultivars. Some of the naked Skardu/Static lines showed mildew resistance equal to the UK parent Static. Resistance of the Skardu/Static lines to Rhynchosporium was poor, but so was that of Static. Further crosses with varieties with Rhynchosporium resistance, such as Westminster and Quench, may be able to produce naked grain lines with disease resistance profiles equal to any covered variety currently on the HGCA Recommended List.

3.4.4. Grain quality

Naked lines generally had higher β-glucan concentration than the covered barleys. This is partly because the hull contains virtually no β-glucan so its inclusion dilutes the β-glucan of covered varieties. UK covered barley is also bred to have a low β-glucan concentration due to its negative effects on malting quality and its anti-nutritional effect on animal feed value. Fungicide decreased β-glucan concentration of most varieties but increased it in a few, especially Static, ICARDA 93.747 and Darzau 5, although the reasons for this are unclear.

Fungicide increased grain specific weight, which should increase the endosperm content and flour yield. This may increase the yield of β-glucan, even if fungicide use does decrease β-glucan concentration. The unusually high lodging rate of the Darzau lines when fungicide was used was a complicating factor in interpreting the results of this trial.

The time of harvest affects β-glucan content. It increased with time as the endosperm cell walls thickened, suggesting that a longer grain filling period could be advantageous in ensuring a high β-glucan concentration. It then declined in later harvested samples. The most likely cause of this decline is activation of endogenous β-glucanase during the first stages of germination, as grain was subject to weather damage. These data agree with the findings of Tiwari and Cummins (2008), whose models predicted decreases in β-glucan content of around 30% if grain was harvested early in physiological maturity (GS92), rather than allowing time for β-glucan to develop. Tiwari and Cummins (2008) also warn that there is a 'trade off' between delaying harvest to allow full β-glucan development and a delayed harvest risking sprouting and loss of β-glucan. The data reported here support this, especially as the differences in β-glucan between sampling dates were larger than those between the four genotypes used. Data presented here for percentage β-glucan concentration were for whole grain, including the husk for hulled varieties. Therefore, a more comprehensive study using a range of milling techniques is recommended to fully assess these differences between hulled and naked varieties.

The amino acid results showed that the pattern of storage protein deposition is fairly well conserved despite big differences in morphology between varieties. Our results agreed well with the results of Lange et al. (2007). Varieties that have extra protein had relatively less lysine and threonine, with the variety Hiproly being an outlier, as expected. Varieties with more overall protein have extra hordeins that are rich in proline and glutamate. Antisense technology has been shown to suppress C-hordein in barley and enhance lysine and threonine content (Lange et al., 2007). The variation that exists in the exotic germplasm suggests that lysine and threonine content could be enhanced through conventional breeding.

3.4.5. Breeding UK-adapted naked barley

The Static/Skardu lines that emerged from several seasons of adaptation and selection showed adaptive traits to the UK climate, such as mildew resistance and straw strength, equal to the UK controls. F_2 from further crosses, especially

Static/Skardu lines backcrossed to other UK varieties, showed further improvements, but data are not presented here as insufficient seed was available for replicated trials. Some of these mixed covered-naked populations out-yielded the UK controls, indicating that it could be possible to select high yielding naked barleys from these populations in future generations. This further emphasises the case that *nud* does not cause a pleiotropic effect on agronomic characteristics, and that the poor agronomic performance observed in these trials is a result of non-adapted material.

3.4.6. Steps towards developing a marker-assisted selection programme

A supply of UK-adapted barley grain with an optimum content of biologically active, high molecular weight, highly soluble β-glucan is required if a UK supply chain is to flourish for naked barley. There is still a lack of understanding of the genetic control of β-glucan. Advances in molecular screening of germplasm collections have opened up a new resource for barley breeders. Mapping of β-glucan content and related traits is required. Candidate genes include the CSL superfamily including HvCslF6 on 7H, linked to *nud* (Burton et al., 2006 and 2008). These could be targeted in future marker assisted selection. Selectable markers can be developed following the identification of biochemical components involved in its synthesis and degradation. Other useful markers that could be selected for were published by Varshney et al., 2008 and four of these markers have been tested on 12 of the varieties used in this project (Steele, unpublished). Selection for the appropriate QTLs and genes using molecular markers in a conventional breeding programme could then enable linkages to be broken and new germplasm to be introduced into UK barley without compromising yield and agronomy.

An initial step would be to asses traits related to health benefits in a mapping population that we have developed from the Static/Skardu cross (229 lines at the F_6 generation) and identify QTLs. The exotic parents could also be assessed with a larger number of markers at loci linked to these traits, and at loci linked to published QTLs for β-glucan, to see which loci could be targeted in selection.

3.4.7. Conclusions

- None of the varieties tested in this project were suitable for UK growing; all had substantial agronomic disadvantages and low yield potential.

- The wet summers during the project exacerbated the agronomic weaknesses of available varieties, leading to lodging and disease. Wet weather and delayed harvest caused poor grain quality – staining, low specific weights and low β-glucan.

- Crop establishment of naked barley was poor, compared to covered varieties, but this alone did not account for the low yields of Lawina. The ear population of Lawina was limited by self-shading, so that increased seed rates failed to significantly increase yield.

- Better establishment was achieved by sowing into a warmer seedbed. The recommendation for spring naked barley is to sow later (ideally 20 March to 10 April). Autumn sowing gave good establishment, therefore development of winter varieties of naked barley should be investigated, as this may be the optimum strategy

- Lawina and most other Darzau lines did not give a yield response to fungicide.

- The agronomic problems of naked barley are caused by non-adaptation, not a pleiotropic effect of the *nud* locus. It may possible to bred UK-adapted naked barley by crossing naked and UK covered barleys.

- The lower yield potential of naked barley may be compensated by the development of cultivars with grain with specific health characteristics that would attract a premium price in the market, for example, BARLEYmax in Australia and Glucagel in New Zealand. This is likely to require the development of barley-based foods (such as bread or breakfast cereals) that have demonstrable health advantages.

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5. APPENDIX

AGRONOMY GUIDELINES FOR LAWINA NAKED BARLEY

2010 Version

By Edward Dickin

Disclaimer: This report was written for, and distributed to, farmers involved in the trials. The following agronomy information is based on the data and experience from field trials at Henfaes Research Centre since 2005 and every effort has been made to ensure that it is as accurate as possible. However, Bangor University can accept no responsibility for any losses arising from the use of this information.

Any crop protection products mentioned are suggestions only. For specific recommendations concerning crop protection products consult a BASIS registered agronomist.

The variety Lawina

There are currently no varieties of naked barley bred specifically for UK growing conditions. The naked barley variety Lawina was bred in Germany and can be grown in the UK under the provisions of the EU Common Catalogue for agricultural seeds. As a continental variety, Lawina is not ideally adapted for UK conditions. Trials over four years have shown that with careful agronomy, yields of around 4-5t/ha can be achieved, but yields may be as low as 2.5 t/ha in adverse conditions. Grain quality of this variety has been generally high.

Sowing

Establishment of naked barley is generally only 70% of viable seeds sown and seed rates should be adjusted accordingly. For example, where 350 seeds/m^2 would be used for covered barley, use 425 seeds/m^2. The grower is strongly advised not to sow at below the recommended seed rate. Seed treatment should not be used on naked barley as trials indicate that standard barley seed dressings such as Raxil-pro decrease establishment. Further research is needed. Trials have shown that best yields come from crops where 280-300 plants/m^2 were established, although a plant population as low as 200 will still give an acceptable crop.

Ideal sowing date for Lawina is 20 March – 10 April, though this is weather-dependant. It is better to wait for good seedbed conditions than try to establish a crop in wet cold soil. Lawina produces fewer tillers than most UK varieties. For most UK varieties of spring barley the extra tillering from an early sowing more than compensates for lower establishment. This may not be the case for Lawina, so sowing must be done when seedbed conditions are good to ensure optimum establishment. Trials in 2007 and 2009 (warm spring) showed better establishment than 2008 (cold spring). Lawina matures 5-10 days before most UK spring barley varieties, so is suitable for later sowing.

Sowing depth should not be too deep. Optimum is probably around 1” (25mm). Later sown crops can be sown deeper. Trials have shown that some European naked barleys appear to have weak coleoptile growth.

Naked barley will flow through the drill faster than covered barley. Drills should be put on a ‘wheat’ setting rather than a ‘barley’ setting, to correct for flow rate.

Fertilizer

Lawina has similar nutrient requirements to covered spring barley, but a small decrease in N fertiliser may be considered as the variety is susceptible to lodging. Lawina has a tendency to be a shy tillerer early in the season, and then produce secondary tillers, which can cause green grains at harvest. This is one of this variety’s agronomic drawbacks. N should be applied early (a proportion in the seedbed) to encourage tillering, forming a denser canopy that will discourage late tillering. As the end-use is not malting, ensuring low grain-N is not a requirement for Lawina.

Our experience has shown that a foliar application of manganese is often helpful in ensuring green healthy growth. This can be applied tank-mixed with fungicide (consult a BASIS registered agronomist for tank-mix advice. Manganese should never be tank-mixed with hormone herbicides, e.g. Mecoprop-P).

Crop protection

Lawina has poor resistance to UK strains of powdery mildew. This is a problem of its origin, not its naked grain. A similar fungicide programme to that for the malting variety Optic (poor mildew resistance) should be used. A protectant fungicide with activity against mildew, such as Flexity (metrafenone) is a good option.

Lawina has shown a tendency for the tips of leaves 2 and 3 (2^{nd} and 3^{rd} from top) to yellow at flag leaf emergence (GS39). This is not due to disease or nutrient deficiency, but is a natural trait of this variety. Experience in 2009 suggested that fungicides that help green leaf retention such as strobilurins (eg Amistar) or chlorothionil (Bravo 500) may be helpful. Late applications of morpholene (e.g. Corbel) should be avoided as this can cause dieback of leaf green area. Lawina failed to give a significant yield response in 2009 to fungicide, so there may be an opportunity to decrease rates.

Lawina has longer straw than most UK spring barley varieties. If early N is used and a good crop established, a PGR may be advisable. Moddus may have an effect of strengthening rooting as well as reducing straw length so is a good option. Avoid Terpal (it stresses the plants too much) and chlormaquat which does not work well on barley and residues may be an issue. The straw of Lawina is strong, but the strength of the root crown is only moderate.

Grain quality

The real strength of Lawina is that it produces clean bright grains, ideal for flaking. Unlike some other naked varieties it is less likely to become dull and stained in wet conditions, but to ensure quality crops should be harvested as soon as possible after maturity.

We would suggest managing the crop for grain quality in a similar way to milling wheat. Pre-harvest glyphosate will even ripening and ease harvesting (check with processor if residues are an issue). In our trials, Lawina has appeared to be more resistant to *Fusarium* ear blight and sooty mould than many UK spring barley varieties. However an earwash of tebuconazole (Folicur), as used on milling wheat may be advisable to ensure clean grain (again check with processor). Lawina has been moderately susceptible to ergot in trials. Control grass weeds (especially blackgrass) and consider keeping grain from infected field margins separate.

For flaking, grains need to be clean, bright and intact. Slow combine drum speed to 2/3 of normal barley setting to avoid cracking grains. The grains should thresh easily out of the ear without the need for higher drum speeds normally needed to remove the awns.

If there are green grains present in the sample it must be dried immediately after harvest. The grain is for human consumption so appropriate storage protocols must apply.