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# Developing enhanced breeding methodologies for oats for human health and nutrition (InnovOat)

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# CONTENTS

1.	ABSTRACT1						
2.	INTRODUCTION2						
3.	MATE	MATERIALS AND METHODS4					
	3.1.	Plant material	4				
	3.2.	Field trials	5				
	3.2.1.	Experimental design, treatments and management	5				
	3.2.2.	Field Assessments	6				
	3.2.3.	Grain quality analysis	7				
	3.2.4.	Data analyses	8				
4.	RESU	RESULTS8					
	4.1.	2014/15 nitrogen response experiment	8				
	4.1.1.	Grain and biomass yield	8				
	4.1.2.	Nitrogen Use Efficiency	10				
	4.2.	2016/17 nitrogen response experiment	14				
	4.2.1.	Grain and Biomass Yields	14				
	4.2.2.	Nitrogen Use Efficiency	16				
	4.3.	Grain Quality	20				
	4.3.1.	Specific Weight and Thousand Grain Weight	24				
	4.3.2.	Kernel Content and Hullability	25				
	4.3.3.	Grain size	31				
	4.3.4.	Grain composition	34				
	4.3.5.	Relationship between traits	34				
	4.4.	QTL-NIL analyses	40				
	4.4.1.	Plant height and Flowering time	45				
	4.4.2.	Grain yield	47				
	4.4.3.	Grain quality	50				
5.	DISCU	JSSION	54				
6.	TAKE HOME MESSAGES60						
7.	ON-GOING AND FUTURE WORK						

8.	REFERENCES	61
9.	ACKNOWLEDGEMENTS	64
10.	APPENDIX 1 WEATHER DATA	65
11.	APPENDIX 2 PROJECT DISSEMINATION AND OUTREACH ACTIVITIES	70

## 1. Abstract

The objectives of this project were three-fold. Firstly, to determine the effect of nitrogen (N) rate on selected modern winter oat varieties and provide information to farmers on the optimal N to use – for enhanced yield and grain quality and, ultimately, milling performance. Secondly, to use precisely developed genetic stocks, in which specific regions of the genome controlling key grain quality and yield related traits have been introgressed into a common genetic background, to identify the effect of specific QTL on grain quality, yield and agronomic performance in a range of environments. Thirdly, to communicate results and recommendations to stakeholders, academics and the public.

Results have revealed the large effect of the environment on grain yield and on all grain quality measures analysed, with a significant interaction between variety and environment. The winter oat variety Mascani was found to have the most stable grain quality across all treatments. Dissection of grain quality traits has revealed the importance of grain roundness on milling quality.

A series of QTL-NILs, in which key QTL for adaptive traits such as flowering time and height have been introgressed into either a Buffalo or Tardis background, were used to validate QTL identified in previous studies and to understand the physiological basis of these traits. The results have provided new insights into the interaction of height and flowering time genes on stem extension and heading date, and on grain quality. In the process, new genetic markers have been identified and shared with breeders to facilitate more efficient selection of these regions, along with understanding as to how traits are closely linked or pleiotropic to these QTL.

Detailed analysis of N response has identified that, although higher levels of N fertilisation increased grain yield over all varieties, there was a variety dependent positive response to increased N for kernel content and hullability, but a negative response of both specific weight and screenings. At high N, some varieties may have specific weights and screenings below milling specifications, despite meeting kernel content and hullability requirements, and having higher β-glucan content.

Results have been incorporated into the recent review of recommendations for farmers (RB209) on N applications in winter oats, although it was concluded that there was insufficient data to derive robust N recommendations for winter oats. This knowledge gap is now being addressed in the AHDB and industry-funded project 'Optimising nitrogen rates and timings and sulphur in winter and spring oats for yield and milling quality' (NoatS – 21140039). As varieties differed in the extent of response to N fertiliser, it is necessary to develop variety-specific N management plans. This emphasises the importance of variety choice, as well as management strategies for specific cultivars. These findings highlight the complementary use of breeding and management in the production of milling oats.

## 2. Introduction

The oat (*Avena sativa* L.) is a high quality cereal, currently experiencing resurgence in its use for human consumption (Marshall *et al.*, 2013), due to the recognised health benefits attributed to the nutritional qualities of the oat grain (Andersson and Borjesdotter, 2011). Dietary benefits associated with phytochemicals within the oat grain, such as  $\beta$ -glucan, and approved health claims for oat  $\beta$ -glucan has contributed to the increased interest in oats as a food ingredient and led to incorporation into an increasing number of food products (Yao *et al.*, 2007). As the interest in oat products increases, so the demand for raw materials with particular health benefits requires oat breeding programmes aimed at the release of improved oat cultivars which meet the needs of food industries (Redaelli *et al.*, 2013) and at the same time have the milling qualities which are required to ensure their uptake by the milling industry.

Grain yield and quality determine much of the value of an oat crop to the producer with several grain characteristics routinely used to define grain quality and mill yield (Doehlert et al., 2001). Although alternative methods for quantifying grain milling quality through detailed analysis of grain size and shape using image analysis are now being developed (Marshall et al., 2015), oats for milling are currently traded on the basis of their hectolitre weight, screenings and a subjective assessment of condition (White and Watson, 2010). Hectolitre weight is a measure of the density or specific weight and although regarded as a poor indicator of milling quality (Burke et al., 2001) is still routinely used in analysing oat crops ex farm. Kernel content (the proportion of groat to whole grain) and ease of hull removal (hullability) are the most important traits for milling quality (Burke et al., 2001) as kernel content is the characteristic most closely associated to the millers extract yield of product (Burke et al., 2001; White and Watson, 2010) and hullability has important implications for mill efficiency. Varieties with poor hullability require greater impact speeds within the dehuller during the milling process and result in greater kernel breakage, thereby depressing the millers extract yield. Improving the physical characteristics of the oat grain to maximise milling yield has become a major target of many oat breeding programmes (Marshall et al., 2013) and therefore understanding the genetic and environmental effect on these characteristics is increasingly important.

Agro-environmental adaptation is critical to many aspects of crop performance and yield. The model winter oat population (Buffalo x Tardis) has been used to identify Quantitative Trait Loci (QTL) for a wide range of traits including flowering time, height and yield component traits as part of the QUOATS project which was partly funded by AHDB (PR543). Many of these traits are quantitatively inherited, displaying continuous variation due to the contribution of multiple genes, each with relatively small effects. Although the use of segregating populations or association panels derived from well adapted varieties have been able to identify QTL and provide linked markers for use in plant breeding, there are often large genomic intervals associated with those

2

QTL. More detailed understanding into the function of these QTL can be conducted by converting them into discrete Mendelian factors by the production of Near Isogenic Lines (NILs). These have otherwise identical genetic backgrounds except at one or a few genetic loci and have been used intensively for detailed mapping and characterization of individual loci in a range of crops (Montfort and Tanksley, 2000, Bai *et al.*, 2012; Zikhali *et al.*, 2014, Farré *et al.*, 2016, Brinton *et al.*, 2017, Kumari *et al.*, 2019)

A series of QTL-NILs have been developed in in which key QTL for these adaptive traits were introgressed into either a Buffalo or Tardis background. These pairs of NILs have been developed using marker assisted selection (MAS) to have contrasting genotypes at the selected QTL regions within an otherwise isogenic recombinant genetic background. Such lines provide a very effective strategy both for the validation of selected QTL, and the identification of their physiological and genetic bases. It is also possible to conduct more detailed analysis of such lines and determine the effect of such QTL on other important traits such as grain quality. Moreover, it is possible to transfer these regions of the genome to other varieties to determine the effect of genetic background on QTL expression.

Compared to other cereals (e.g. wheat and barley), oats are seen as a low input cereal needing lower fertilization levels (Weightman *et al*, 2004; Kindred *et al.*, 2008). Although nitrogen fertilisation is well recognised for enhancing crop yield, nitrogen is a considerable environmental burden in arable crop production (Stark and Richards, 2008; Rütting *et al.*, 2018). For example, improving nitrogen recovery and utilisation by cereals is seen as essential to protect water quality. Moreover, the effects of nitrogen application during oat yield development on grain quality parameters are poorly understood. Provision of a sufficient amount of nitrogen has great bearing on the plant height, quality and oat yield (Chalmers *et al.* 1998; Givens *et al.* 2004, Yan *et al.* 2017). Oat quality and yield, in turn, impact upon the processing quality of oats, with respect to dehulling, as well as the later processing steps of flaking and milling (Browne *et al*, 2003). Current agricultural guidelines on the levels of nitrogen application. It is important also to understand the role of variety choice and of crop management in determining yield and quality of oats.

The specific aims of this project were to:

- Determine the optimum N rate for selected modern winter oat varieties and provide information to farmers on the optimal N to use for both enhanced yield and grain quality.
- Utilise QTL-NILs in which specific regions of the genome controlling key grain quality and yield related traits have been introgressed into a common genetic background to identify the effect of these QTL on grain quality, yield and agronomic performance in a range of

environments and to determine the effect of genetic background on selected QTL by crossing into UK adapted germplasm.

• Communicate results and recommendations to stakeholders, academics and the wider public.

## 3. Materials and methods

## 3.1. Plant material

The nitrogen response trials included six winter oat varieties from the Aberystwyth University winter oat breeding programme. These included two of the most widely grown winter oat varieties in the UK over the last 20 years, Gerald and Mascani. Five varieties were of conventional height (Mascani, Gerald, Tardis, Maestro and Griffin) and the sixth was a dwarf variety (Balado).

Grain quality was also analysed in this project from trials in 2013/14 that had been harvested immediately prior to the start of this project. These nitrogen response trials had been conducted at Lydbury North and at ADAS Rosemaund in the QUOATS project and methodology of those trials are presented in AHDB report for project RD-2008-3556.

A series of near-isogenic lines (QTL-NILs) developed at Aberystwyth University in which contrasting haplotypes for specific major quantitative trait loci (QTL) have been fixed in common winter oat genetic backgrounds were also grown in field trials. The QTL had been previously identified from a recombinant inbred line (RIL) mapping population that was produced from a cross between the two winter oat varieties Buffalo and Tardis at Aberystwyth University, UK (Mellers et al., 2020). The population was created to capture key differences between the parents: Buffalo is a dwarf variety with low kernel content, small grains and late flowering whereas Tardis is a conventional height variety with higher kernel content, large grains and mildew resistance. Each QTL-NIL was developed following at least 3 generations of marker assisted backcrossing (MAS), using microsatellite markers flanking each QTL, to either the Buffalo or the Tardis parent. RILs from the Buffalo x Tardis mapping population which were as close to the desired genotype as possible were selected as donor genotypes. At least 2 versions of each QTL-NIL were developed from independent backcrossing. In addition, MAS of selected QTL from Buffalo and Tardis into Mascani was conducted. The genotype of each QTL-NIL was verified by using the oat 6k SNP chip (Tinker et al., 2014).

## 3.2. Field trials

## 3.2.1. Experimental design, treatments and management

In each of the 2014/15 and 2016/17 seasons, nitrogen (N) response experiments were carried out near ADAS Rosemaund, Herefordshire on sites with sandy clay loam soil and winter wheat as the previous crop. In both seasons, a split-plot design was used. There were three replicates within which were randomised the N treatments and within which were randomised the variety treatments. The N mainplots were separated with nil-N discard plots.

In 2014/15, four winter oats varieties (Gerald, Mascani, Tardis and Balado) were each tested at six N rates. Before any N treatments were applied, soil samples were taken to 90 cm depth and tested for soil mineral N. An estimate of N in the crop was made at the same time. This indicated that SMN was 24 kg N/ha with approx. 20 kg N/ha in the crop. The N rates applied to each of the varieties were therefore as per Table 1.

**Table 1.** Details of amounts and timings of nitrogen (N) treatments applied to the 2014/15 N response experiment.

	Early March application (kg N/ha)	Early stem extension – 1 <sup>st</sup> split (kg N/ha)	Early stem extension – 2 <sup>nd</sup> split (kg N/ha) approx 2 weeks after 1 <sup>st</sup> split	Total N applied (kg N/ha)
1	0	0	0	0
2	0	30	30	60
3	40	40	40	120
4	40	70	70	180
5	40	95	95	230
6	40	120	120	280

The crop was managed as per local commercial practice. A PGR programme was applied to all varieties apart from the dwarf variety Balado.

In 2016/17, three winter oats varieties (Mascani, Maestro and Griffin) were each tested at six N rates (Table 2). Before any N treatments were applied, soil samples were taken to 90 cm depth and tested for soil mineral N. An estimate of N in the crop was made at the same time. This indicated that SMN was 47 kg N/ha. Along with the N already in the crop, this gave an SNS index of 1.

The crop was managed as per local commercial practice. A PGR programme was applied to all varieties.

	Early March application (kg N/ha)	Early stem extension – 1 <sup>st</sup> split (kg N/ha)	Early stem extension – 2 <sup>nd</sup> split (kg N/ha) approx. 2 weeks after 1 <sup>st</sup> split	Total N applied (kg N/ha)
1	0	0	0	0
2	0	30	30	60
3	40	40	40	120
4	40	60	60	160
5	40	90	90	220
6	40	120	120	280

**Table 2.** Details of amounts and timings of nitrogen (N) treatments applied to the 2016/17 N response experiment.

In each of the 2015/16 and 2016/17 seasons, selected QTL-NILs were grown in a randomised trial with three replicate plots of each QTL-NIL along with control lines including Buffalo, Tardis and a range of modern winter oat varieties at both ADAS Rosemaund and at Gogerddan, Aberystwyth University. Fungicides and weed control followed standard practise for winter oats and fertiliser application followed RB209 guidelines.

## 3.2.2. Field Assessments

In the 2014/15 season, the number of tillers per  $m^2$  was assessed at GS59 in 5 places per plot by counting the number of tillers on either side of a 0.5m rod which had been placed between 2 rows of plants. The number of tillers per  $m^2$  was then calculated, taking account of row width.

In 2015/16 and 2016/17, in addition to the assessments described above, flowering time was assessed as well as height at GS75 to the flag leaf ligule and the tip of the panicle.

The percentage of each plot that was affected by leaning (to 45°) and lodging (45-90°) was assessed when lodging first occurred and before harvest.

Just before harvest, grab samples of c. 50 shoots were taken per plot and separated into straw and panicles. The panicles were threshed and the straw + chaff and grain were dried and weighed separately and Harvest Index calculated. Thousand grain weight (g) was measured and samples of the straw + chaff and grain sent for analysis of %N. N uptake and partitioning were then calculated.

All plots were combined, and grain yield was calculated, adjusted to 15% moisture content.

Daily rainfall (mm), minimum and maximum temperatures (°C) and relative humidity (%) were recorded for each site using in-field weather stations. Results are presented in Appendix 1.

#### 3.2.3. Grain quality analysis

Harvested grain was cleaned through a 3.5 mm and 2 mm sieve to remove broken grain and residual chaff and straw prior to analysis of grain quality. The proportion of grain passing through the 2 mm sieve was weighed and used to determine the screenings value.

A 25 g sample of grain was measured using a Marvin Seed Analyser (GTA Sensorik GmbH, Germany) prior to de-hulling. Seeds were placed on the analysing tray and spread out so that no seeds were touching. All seeds in the sample were measured for individual grain length (mm), width (mm) and area (mm<sup>2</sup>). The ratio of grain width to grain length was used as an indicator of grain roundness where 0 is very elongated and 1 is perfectly round. Individual grain data from each plot sample was also extracted to examine distributions of grain size. This analysis was conducted by Pilar Martinez as part of her PhD studies which were part funded by AHDB.

Kernel content (KC) was determined by passing 25 g of whole grain through a Laboratory Oat Huller (Codema Model LH5095; Maple Grove, Minneapolis, USA) set at 100 bar for 60 seconds and then separating the output into groats and whole grain.

Kernel content was calculated as:-

KC (%) = 100 x (Groat weight (g) / (Initial weight (g) – Whole grain weight (g)))

Hullability (%) was calculated as:-

Hullability (%) =  $100 - (100 \times Whole grain weight (g) / Initial weight (g))$ 

Thousand grain weight (TGW; g) was determined using the mean weight of 3 samples of 250 seeds. Grain numbers per m<sup>2</sup> and per panicle were calculated using grain yield and TGW results.

Grain hectolitre weight (often referred to as specific weight) was measured using a chondrometer (Nileme, C288) on 3 replicate samples (approximately 500 ml) per field plot. Chondrometers are cylindrical devices containing a column in which grains are isolated from the cylinder of known volume underneath by means of a level blade or metal bar (Manley *et al.*, 2009). The blade separates a precise volume of grain (below the blade) from excess grains above the blade (ISO, 1986). This known volume of grain was weighed and the mass converted to kg hL-1.

Groat nitrogen and oil content were predicted using near infrared spectroscopy (NIRS) and βglucan content was determined with a Megazyme<sup>™</sup> kit (McCleary and Codd, 1991 (AOAC method 995.16, 1999)) as described in Allwood *et al.* (2019).

#### 3.2.4. Data analyses

All data were analysed with ANOVA in GenStat (VSN International).

The response of yield to N was estimated for each experiment individually using the linear plus exponential function (LEXP). This has been used as the standard method since a comparison of approaches by George (1984), including in the preparation of RB209.

y = a + b.r N + c.N

where y is yield in t/ha at 85%DM, N is total fertiliser N applied in kg/ha, and a, b, c and r are parameters determined by statistical fitting. Occasionally there is a difficulty in estimating the parameter r. Therefore, if r was outside an acceptable range, the function was re-fitted using an r value of 0.99.

Optimum N rates (Nopt) were then derived from the fitted LEXP parameters using:

Nopt = [ln(k-c)-ln(b(ln(r)))]/ln(r)

where k is the breakeven price ratio between fertiliser N ( $\pounds$ /kg) and grain ( $\pounds$ /tonne). The breakeven ratio used in this study was 0.005 (tonnes grain per kg N) so that direct comparisons could be made with RB209 (both 8<sup>th</sup> and 9<sup>th</sup> editions).

## 4. Results

## 4.1. 2014/15 nitrogen response experiment

## 4.1.1. Grain and biomass yield

There was no lodging present in this trial, but overall the yields in this experiment were low (Table 3). The overall average yield at the 180 kg N/ha N rate was 4.97 t/ha (at 15% mc; Table 3). There was a significant (P<0.001) difference in yields among the varieties, caused by the 1.1 t/ha lower yield of Gerald compared to the average of the other varieties, whose yields did not significantly differ from each other (Table 3). There was a significant (P<0.001) yield response to N in this experiment and also a significant (P<0.001) variety x N rate interaction (Table 3).

Fertiliser N					
rate (kg N/ha)	Balado	Gerald	Mascani	Tardis	Mean
0	2.28	1.83	1.85	2.14	2.02
60	3.43	2.67	2.64	3.88	3.15
120	4.90	3.66	4.65	4.78	4.50
160	5.05	2.72	4.45	4.86	4.27
220	5.07	3.81	5.57	5.42	4.97
280	4.80	3.65	4.92	4.70	4.52
Mean	4.25	3.06	4.01	4.30	3.91
		N rate	Variety	N Rate*Variety	
	F Pr.	<0.001	< 0.001	<0.001	
	LSD	0.842	0.250	0.955	
LSD when a	comparing same	levels N		0.607	

**Table 3.** Yields (t/ha @ 15% moisture content) of variety and N rate treatments in the 2014/15 trial at ADAS Rosemaund.



**Figure 1.** Linear plus exponential curves fitted to the yield response to N of Balado, Gerald, Mascani and Tardis in the 2014/15 trial at ADAS Rosemaund. Economic optimum N rates for each variety is represented by a cross.

As well as low yields, overall the treatments produced low total biomass and low harvest indices (HI); at the 180 kg N/ha N treatment, the average biomass was 7.77 t/ha @ 100% dm and HI was 46.7% (Table 4). There was a significant (P<0.001) effect of variety on both biomass and HI. On average Mascani gave the highest biomass (9.16 t/ha) but lowest HI (37.1%) with Gerald giving the reverse (5.47 t/ha biomass and 47.4% HI). There was a significant (P = 0.022) effect of N rate on HI, but the only treatment which significantly differed from the others was the 60 kg/ha N rate where the HI was 4.3% kg/HI lower than the average of the rest of the treatments (Table 4). There was a significant (P = 0.045) variety x N rate interaction in the biomass results (Table 4). The biomass of Gerald gave a very shallow response to N rate up to a maximum response of 3.56 t/ha, whereas the biomass of Mascani increased more steeply, with a maximum response of 7.30 t/ha; the responses of Tardis and Balado were between these two extremes (Table 4).

#### 4.1.2. Nitrogen Use Efficiency

As with the yields and biomass, the amount of N that was taken up was low in this experiment with 133.76 kg N/ha taken up, on average, at the 180 kg/ha N rate (Table 4). At the start of the experiment, it was estimated that there was 44 kg N/ha in the soil and crop before any N was applied. The actual amount that was taken up by Mascani with no N applied was 40.21 kg N/ha (Table 4). Balado took up a similar amount with no N (39.18 kg/ha), but Tardis, and particularly Gerald, took up less of the available N in the soil at 34.03 and 29.91 kg/ha, respectively (Table 4). There was a significant (P = 0.038) variety x N rate interaction on total N uptake, with differences between variety responses to N rates similar to those found with the biomass results (Table 4).

The grain, biomass and N yield figures were used to calculate N use, uptake and utilisation efficiencies (Table 5). N use efficiency (NUE; kg grain per kg N) generally mirrors yield responses, and this was the case in this experiment. There was a significant (P = 0.04) variety x N rate interaction with the NUE of Balado and Tardis highest at the 0 kg/ha N rate and that of Gerald lower at most N rates than the other varieties (Table 5, Figure 2).

There was no significant interaction seen in the N uptake efficiency (NUpE; kg N in crop/kg N from soil and fertiliser) results, but there were significant (P<0.05) main effects of variety and N rate (Table 5). This showed that the least of the available N from the soil and fertiliser was taken up by Gerald and the most taken up by Mascani (Table 5). The significant (P = 0.024) effect on N rate was due to the higher NUpE of the 0 kg/ha N rate compared to the rest of the treatments (Table 5).

The N utilisation efficiency (NUtE; kg grain/kg N taken up) shows how well the N taken up is partitioned to the grain. There was a significant (P = 0.013) variety x N rate interaction (Table 5) whereby Balado and Gerald had similar NUtE and were better at partitioning the N taken up to the grain than Mascani and Tardis.



**Figure 2** Effect of Nitrogen on Nitrogen Use Efficiency of 4 winter oat varieties at ADAS Rosemaund 2014-15



**Figure 3** Mean ± s.e.m Nitrogen uptake efficiencies (NUpE) for 4 winter oat varieties at ADAS Rosemaund 2014-15.

**Table 4.** Harvest Index (%), total biomass (t/ha @ 100% dm) plus grain, straw/chaff and total N yield (kg ha) of the varieties and N rates in the 2014/15 trial at ADAS Rosemaund.

Variety	N rate (kg	Harvest	Total	Grain N	Straw/ chaff N	Total N yield
,	N/ha)	index (%)	biomass	yield (kg/ha)	yield (kg/ha)	(kg/ha)
	-		yield (t/ha)			
Balado		44.3	8.33	72.15	29.68	101.83
Gerald		47.4	5.47	51.56	18.13	69.69
Mascani		37.1	9.16	73.67	42.48	116.15
Tardis		46.6	7.87	72.57	28.00	100.15
Mean	0	44.5	3.97	26.41	9.43	35.83
	60	40.4	6.82	41.39	19.29	60.68
	120	44.3	8.88	66.98	26.95	93.93
	180	46.7	7.77	77.97	35.79	113.76
	230	43.1	10.03	98.29	43.25	141.54
	280	44.8	8.67	95.84	42.72	139.24
Balado	0	46.8	4.20	28.98	10.20	39.18
	60	43.8	6.67	41.93	17.71	59.64
	120	44.2	9.39	69.82	28.36	98.17
	180	47.7	9.00	90.56	29.04	119.60
	230	37.2	11.74	97.06	52.22	149.28
	280	45.9	8.96	104.56	40.55	145.11
Gerald	0	47.3	3.27	23.04	6.87	29.91
	60	48.4	4.73	32.92	10.46	43.38
	120	49.5	6.29	53.02	14.96	67.98
	180	46.6	4.89	50.53	18.14	68.67
	230	47.2	6.82	74.38	27.55	101.93
	280	45.5	6.82	75.50	30.78	106.29
Mascani	0	34.9	4.72	27.21	13.00	40.21
	60	28.9	7.80	40.70	24.22	64.91
	120	37.5	11.00	73.75	37.17	110.91
	180	42.7	9.12	84.24	68.49	152.73
	230	39.6	12.02	111.90	54.84	166.74
	280	40.8	10.29	104.23	57.16	161.39
Tardis	0	49.0	3.68	26.39	7.64	34.03
	60	40.6	8.07	50.03	24.77	74.80
	120	46.0	8.86	71.34	27.32	98.66
	180	48.5	8.51	86.55	27.49	114.03
	230	48.5	9.53	109.81	38.40	148.21
	280	46.8	8.59	100.68	42.39	146.66
N rate	P-value	0.022	<0.001	<0.001	0.001	<0.001
	LSD	2.883	1.934	15.512	13.34	24.09
Variety	P-value	<0.001	<0.001	<0.001	<0.001	<0.001
-	LSD	3.025	0.685	4.069	8.32	9.81
N rate*Variety	P-value	0.107	0.045	<0.001	0.284	0.038
	LSD	6.886	2.318	17.13	21.29	30.46
LSD to compare a	same rate <mark>s N</mark>	7.410	1.677	9.968	20.37	24.04

**Table 5.** Nitrogen (N) Use efficiency (kg grain/kg N), Uptake efficiency (kg N in crop/kg N from soil and fertiliser) and Utilisation efficiency (kg grain/kg N taken up) of the varieties and N rates in the 2014/15 trial at ADAS Rosemaund.

Variety	N rate (kg		N Uptake efficiency			
2	N/ha)	N Use efficiency	(kg N in crop/kg N	N utilisation efficiency		
		(kg grain/kg N)	from soil and fert)	(kg grain/kg N taken up)		
Balado		24.15	0.598	38.97		
Gerald		17.97	0.420	40.69		
Mascani		21.43	0.667	31.96		
Tardis		24.23	0.608	39.50		
Mean	0	39.10	0.814	48.44		
	60	25.78	0.583	45.03		
	120	23.32	0.573	41.37		
	180	16.19	0.508	33.10		
	230	15.41	0.517	30.05		
	280	11.85	0.430	27.70		
Balado	0	13 08	0.800	49.60		
Dalado	60	28.04	0.030	48.00		
	120	20.04	0.575	40.70		
	120	20.09	0.599	42.34		
	100	19.14	0.554	30.02		
	230	10.70	0.040	20.90		
	280	12.60	0.448	28.10		
Gerald	0	35.31	0.680	51.49		
	60	21.83	0.417	52.63		
	120	18.98	0.414	45.88		
	180	10.32	0.307	33.26		
	230	11.81	0.372	31.60		
	280	9.57	0.328	29.27		
Mascani	0	35.82	0.014	30 50		
Mascalli	60	21 50	0.624	34.84		
	120	21.09	0.024	36.05		
	120	16.99	0.070	26.02		
	100	10.00	0.002	20.92		
	230	17.29	0.009	20.40		
	280	12.90	0.490	25.97		
Tardis	0	41.30	0.773	53.19		
	60	31.67	0.719	43.86		
	120	24.80	0.602	41.20		
	180	18.44	0.509	36.20		
	230	16.82	0.541	31.15		
	280	12.33	0.453	27.34		
N rate	P-value	<0.001	0.024	<0.001		
	LSD	8.532	0.1977	3.659		
		0.002				
Variety	P-value	<0.001	<0.001	<0.001		
	LSD	1.775	0.0556	2.269		
N rate*Varietv	P-value	0.04	0.409	0.013		
·······	LSD	9.069	0.2217	5.819		
LSD to compare	e same rates N	4.348	0.1363	5.559		

## 4.2. 2016/17 nitrogen response experiment

## 4.2.1. Grain and Biomass Yields

The overall average yield of the 2016/17 experiment at the 160 kg/ha N rate was 6.74 t/ha @ 15% m.c. (Table 6). There was a significant (P = 0.005) effect of N rate on yield but the variety effect was not significant at the 5% level (P = 0.088) and there was no significant interaction (Table 6). The overall average variety yields indicated that Maestro gave the lowest yields and Griffin the highest, with Mascani slightly lower than Griffin (Table 6). Yields increased with N rate up to 220 kg N/ha but reduced at the highest N rate (Table 6) due to brackling and lodging, an effect that was exacerbated because poor weather led to a delayed harvest.

**Table 6.** Yields (t/ha @ 15% moisture content) of variety and N rate treatments in the 2016/17 trial at ADAS Rosemaund.

Fertiliser N	Yie	Yield (t/ha @ 15% moisture content)						
rate (kg N/ha)	Griffin	Maestro	Mascani	Mean				
0	4.58	4.16	4.08	4.27				
60	6.31	6.45	6.78	6.51				
120	6.87	6.26	7.02	6.72				
160	7.07	6.54	6.63	6.74				
220	7.01	6.37	6.03	6.47				
280	5.38	5.19	6.00	5.52				
Mean	6.20	5.83	6.09	6.04				
		N rate	Variety	N Rate*Variety				
	F Pr.	0.005	0.088	0.244				
	LSD	0.524	0.1663	0.6206				
LSD when c	0.407							



**Figure 4.** Linear plus exponential curve fitted to the yield response to N of Griffin, Maestro and Mascani at ADAS Rosemaund in 2016/17. From statistical analysis, there was no justification to fit more than one curve over the three varieties. Economic optimum N rate for this curve is represented by a cross.

When the yield response to N was examined, there was no statistical justification to fit separate curves to the varieties. The calculated economic optimum N rate for the N response curve was 98 kg N/ha (Figure 4).

Brackling results (Figure 5) showed the differing responses of the three varieties. Griffin suffered most from brackling with nearly 100% of plot area affected when any amount of N was applied. Maestro and Mascani suffered less than Griffin at lower N rates, but the brackling of Mascani did not increase when more than 120 kg N/ha was applied whereas brackling in Maestro continued to increase until it reached 100% brackled at 220 kg N/ha (Figure 5).



**Figure 5.** Effect of N rate on brackling (% of plot area brackled) of three varieties at ADAS Rosemaund in 2016/17: Griffin, Maestro and Mascani.

The brackling results generally reflected those of crop height (Table 7). There was a significant (P<0.001) effect of variety on both height measured to the top leaf ligule and the top of the panicle (Table 7) with Griffin 7.3 cm taller to the flag leaf ligule than the average of the other two varieties. Height increased (P<0.01) with increasing N rates (Table 7) with the main increases occurring over the first three N rates, but also a large (7.5 cm) increase between the highest two N rates.

The date of panicle emergence significantly (P<0.001) differed with variety but was not affected by N rate (Table 7); Maestro was the earliest variety and Griffin the latest.

There were no significant variety or N rate effects on harvest index and no variety effect on total crop pre-harvest biomass (Table 8). There was a significant (P = 0.007) effect of N rate on pre-harvest biomass (t/ha @ 100% dm) with the least biomass when no N was applied, and also reduction at the highest N rate compared to the other rates where N was applied (Table 8).

## 4.2.2. Nitrogen Use Efficiency

At the start of the experiment the SMN was measured as 47 kg N/ha and it was estimated that there was around 20 kg N/ha already in the crop. The actual amount of N taken up by the crop was, on average over varieties, 82.0 kg N/ha. Maestro took up the most N from the soil (98.1 kg/ha) followed by Mascani (75.8 kg/ha) then Griffin (72.0 kg/ha; Table 8).

There was a near-significant (P = 0.06) variety effect of the total amount of N taken up by the crop, due to a difference (P = 0.029) in the amount taken up into the grain rather than the straw/chaff (P = 0.442; Table 8). Overall, Mascani took up 5.5 kg/ha more N than Griffin and 13 kg/ha more N than Maestro. The amount of N taken up increased as the N rate increased, although this differed with variety (P = 0.045), with a steeper response to N shown by Mascani than the other two varieties (Table 8).

NUE results mirrored the yield results: Griffin gave the highest of the varieties, and Maestro the lowest, although differences weren't significant (P = 0.073); and NUE decreased with increasing N rates (P<0.001; Table 9). There were no significant variety differences in NUpE or NUtE (Table 9), but there was a significant (P = 0.043) variety x N rate interaction in the NUtE (kg grain/kg N taken up) results; NUtE generally decreased with increasing N rate for all varieties, but with Maestro there was a slight increase in NUtE between the 0 and 60 kg/ha N rates (Table 9).

Variety	N rate (kg N/ha)	Date of panicle emergence (Julian	Height (ground to	Height (ground to
Griffin		145.3	75.5	114 0
Maestro		143 1	67.5	97.9
Mascani		144 0	69.0	101.6
Maoballi		111.0	00.0	101.0
Mean	0	144.1	51.3	78.4
	60	143.7	64.6	100.3
	120	144.1	73.8	107.9
	180	144.3	78.2	113.0
	230	143.9	74.3	110.3
	280	144.8	81.8	117.1
Griffin	0	145.0	53.8	85.8
	60	145.0	70.0	111.0
	120	145.0	81.0	119.2
	180	145.0	84.8	123.9
	230	145.0	78.2	116.9
	280	147.0	85.4	127.3
Maestro	0	143.0	50.0	72.6
	60	143.0	58.1	90.5
	120	143.0	68.2	98.8
	180	143.7	74.0	104.5
	230	143.0	73.8	107.7
	280	143.0	81.0	113.0
Magaani	0	111 2	F0 1	76.9
Mascarii	60	144.5	50.1 65.6	70.0
	120	143.0	00.0	99.4 105 7
	120	144.3	72.1	100.7
	100	144.3	70.0	110.4
	230	143.7	70.9	100.3
	280	144.3	79.1	110.9
N rate	P-value	0.528	0.002	<0.001
	LSD	0.577	5.266	5.819
Variety	P-value	<0.001	<0.001	<0.001
-	LSD	0.327	1.027	1.698
Ν				
rate*Variety	P-value	0.505	0.078	0.519
5	LSD	0.872	5.652	6.738
LSD to compare	same rates N	0.801	2.516	4.16

**Table 7.** Date panicle emergence (Julian days) and Height at GS75 to flag leaf ligule and tip of panicle (cm) of the varieties and N rates of the 2016/17 experiment at ADAS Rosemaund.

**Table 8.** Harvest Index (%), total biomass (t/ha @ 100% dm) plus grain, straw/chaff and total N yield (kg ha) of the varieties and N rates of the 2016/17 experiment at ADAS Rosemaund in 2016/17.

Variety	N rate (kg N/ha)	Harvest index (%)	Total biomass yield (t/ha)	Grain N yield (kg/ha)	Straw/ chaff N yield (kg/ha)	Total N yield (kg/ha)
Griffin		53.0	9.96	109.8	41.8	151.5
Maestro		51.7	9.59	104.6	39.4	144.0
Mascani		50.8	10.21	113.2	43.8	157.0
	0	49.7	7.44	57.9	24.1	82.0
	60	53.2	10.44	100.0	32.4	132.3
	120	53.8	10.66	119.3	37.5	156.8
	180	51.8	11.10	130.0	50.9	180.9
	230	52.5	10.48	129.0	52.6	181.6
	280	50.0	9.40	118.8	52.5	171.3
Griffin	0	54.8	7.09	56.3	15.6	72.0
	60	52.7	10.19	94.9	32.4	127.3
	120	54.6	10.69	121.7	37.3	158.9
	180	52.1	11.53	136.0	55.8	191.8
	230	51.9	11.47	136.0	60.1	196.0
	280	51.8	8.81	113.7	49.4	163.1
Maestro	0	43.8	8.33	60.5	37.6	98.1
	60	56.0	9.78	96.7	28.3	125.0
	120	51.8	10.36	106.4	37.1	143.5
	180	54.0	10.29	124.3	43.0	167.3
	230	54.4	9.97	126.1	47.4	173.4
	280	50.1	8.81	113.4	43.0	156.4
Mascani	0	50.5	6.89	56.7	19.0	75.8
	60	50.9	11.35	108.4	36.3	144.7
	120	55.1	10.94	129.9	38.2	168.0
	180	49.4	11.48	129.8	53.8	183.6
	230	51.2	10.00	125.0	50.4	175.4
	280	47.9	10.57	129.2	65.1	194.3
N rate	P-value	0.357	0.007	<0.001	<0.001	<0.001
	LSD	2.145	0.751	7.31	4.89	10.52
Variety	P-value	0.343	0.210	0.029	0.442	0.06
	LSD	1.467	0.339	3.02	3.39	5.18
N rate*	P-value	0.232	0.123	0.125	0.077	0.045
Variety	LSD	3.635	1.012	9.48	8.36	14.77
LSD to c	ompare same rates N	3.594	0.83	7.39	8.31	12.7

**Table 9.** Nitrogen (N) Use efficiency (kg grain/kg N), Uptake efficiency (kg N in crop/kg N from soil and fert) and Utilization efficiency (kg grain/kg N taken up) of the varieties and N rates of the 2016/17 experiment at ADAS Rosemaund.

Variety	N rate (kg	N Use efficiency	N Uptake efficiency	N utilisation efficiency
	N/ha)	(kg grain/kg N)	(kg N in crop/kg N from soil and fort)	(kg grain/kg N taken
Griffin		34.2	0.87	<u> </u>
Maestro		30.9	0.86	35.4
Mascani		31.1	0.86	34.7
Mascani		51.1	0.00	54.7
	0	57.5	1.29	46.2
	60	44.8	1.07	42.1
	120	31.1	0.86	36.6
	180	25.7	0.81	31.7
	230	19.4	0.64	30.4
	280	13.7	0.50	27.5
	_			
Griffin	0	65.4	1.21	54.1
	60	44.9	1.07	42.3
	120	32.5	0.89	37.0
	180	27.4	0.87	31.2
	230	21.3	0.70	30.4
	280	13.5	0.48	28.1
Maestro	0	55.4	1.54	38.1
	60	44.2	1.01	44.1
	120	28.9	0.78	37.1
	180	24.8	0.75	33.2
	230	19.1	0.61	31.5
	280	12.8	0.45	28.4
Mascani	0	51.8	1.13	46.3
	60	45.4	1.14	40.0
	120	31.9	0.90	35.7
	180	24.8	0.81	30.8
	230	17.9	0.61	29.3
	280	14.7	0.56	26.1
N rate	P-value	<0.001	<0.001	<0.001
i i i i i i i i i i i i i i i i i i i		3 809	0.084	1 771
	LOD	0.000	0.004	1.771
Variety	P-value	0.073	0.956	0.182
	LSD	1.523	0.046	1.35
N rate*Variety	P-value	0.373	0.080	0.043
it i allo valioty		4 876	0 125	3 228
LSD to compare s	ame rates N	3.73	0.113	3.306

## 4.3. Grain Quality

In addition to the nitrogen response trials conducted in 2014-15 and 2016-17 described above, grain quality was also analysed in this project from trials in 2013-14 that had been harvested immediately prior to the start of this project. These nitrogen response trials had been conducted at Lydbury North and at ADAS Rosemaund in the QUOATS project and methodology of those trials are presented in AHDB report for project RD-2008-3556 along with grain yield results. The same 4 varieties were used in all trials (Mascani, Gerald, Tardis and Balado) except for 2016-17 where the newer varieties Griffin and Maestro were included instead of Gerald, Tardis and Balado.

Results for the milling quality traits for the four trials are presented in Tables 10, 11, 12 and 13. To summarise, in all trials there was a significant difference both between varieties and between nitrogen rates for the milling quality traits specific weight, kernel content and hullability, except for kernel content in the 2017 N response trial. The highest values for all these traits were obtained by the variety Mascani in all trials except for specific weight at the 2013-14 trial at Lydbury North. Specific weight decreased at higher N rates (Figure 7) although a significant genotype by nitrogen interaction was obtained (Tables 10, 11, 12). Conversely, kernel content and hullability increased in response to nitrogen (Figures 7, 8, 9). Along with Mascani, the newer varieties Griffin and Maestro displayed much more stable kernel content and hullability across nitrogen levels (Figures 6, 8, 9, Table 13).

		0		TOW	TOW	<b>D</b> 1 · 0/	0.1	<u> </u>
	Hullability,	Specific	Kernel	IGW	IGW	Protein %	Oil	β-glucan
	%	Weight,	Content %	grain, g	groat, g	DM	% DM	% DM
		kg/hl						
Variety								
Gerald	94.14 b	54.10 c	73.90 b	38.94 a	29.59 a	10.70	7.66 b	3.31 a
Mascani	99.27 c	52.10 a	77.23 c	45.85 d	37.33 d	10.71	7.31 a	3.96 c
Tardis	86.90 a	50.30 b	72.64 a	42.57 b	32.18 b	11.01	8.19 c	3.75 b
Balado	88.75 a	49.10 b	72.22 a	43.76 c	35.14 c	10.82	7.68 b	4.50 d
Nitrogen level (k	(g/ha)							
0	88.70 a	52.97 c	72.98 a	41.49 a	32.47 a	9.11 a	8.00 e	3.72 a
50	90.50 ab	52.47 bc	73.47 ab	44.11 c	33.46 b	9.64 b	7.89 d	3.83 ab
100	90.09 bc	51.56 b	73.94 bc	42.96 bc	33.55 b	10.67 c	7.70 c	3.86 ab
150	93.70 c	50.33 a	74.10 c	42.03 ab	33.66 b	11.65 d	7.57 b	3.93 bc
200	96.68 d	49.58 a	75.50 d	43.32 c	34.65 c	12.97 e	7.39 a	4.07 c
N treatment p value	<.001	<.001	<.001	<.001	0.002	<.001	<.001	0.002
Variety (V) p value	<.001	<.001	<.001	<.001	<.001	0.277	<.001	<.001
V x N p value	0.002	0.083	0.011	0.015	0.023	0.760	0.222	0.563

**Table 10.** Mean values by variety and by nitrogen application for thousand grain weight (TGW) of grain and de-hulled groat, milling quality and grain composition traits from 2013-14 experiment grown at Lydbury North. Letters indicate significant differences between mean values at p < 0.05.

**Table 11** Mean thousand grain weight (TGW) of grain and de-hulled groat, milling quality and grain composition traits from 2013-14 experiment grown at ADAS Rosemaund by variety and nitrogen treatment.

Variety N applied (kg/ha)		Specific Weight (kg/hl)	Kernel Content (%)	Hullability (%)	Protein (% DM)	Oil (% DM)	β- glucan (%DM)	TGW whole grain	TGW groat (g)
	,				•			(g)	
Balado	0	50.8	72.7	73.0	9.63	7.50	4.69	41.0	30.0
	50	51.4	73.7	81.2	8.48	7.75	4.17	43.5	31.9
	100	51.5	73.9	87.2	9.75	7.54	4.31	43.9	32.4
	150	51.3	74.5	87.9	10.21	7.61	3.99	44.0	32.7
	200	51.3	73.8	85.4	11.48	7.48	4.31	45.6	33.4
	250	50.2	74.0	83.6	13.17	7.25	4.28	42.5	31.5
	Mean	51.1	73.8	83.1	10.45	7.52	4.29	43.4	32.0
Gerald	0	52.0	70.3	75.0	9.91	7.32	3.04	37.1	26.1
	50	52.6	71.3	88.2	8.52	7.58	3.26	37.2	26.4
	100	52.5	72.0	85.4	9.15	7.69	3.19	35.5	25.2
	150	52.5	74.2	94.1	10.50	7.51	3.31	35.7	26.2
	200	52.8	71.7	94.1	11.31	7.52	3.53	36.4	26.5
	250	52.3	73.8	93.5	12.75	7.54	3.48	33.8	24.5
	Mean	52.4	72.2	88.4	10.36	7.53	3.30	36.0	25.8
Mascani	0	53.4	76.2	97.4	9.47	6.98	3.72	43.5	33.3
	50	52.3	76.5	98.3	8.77	7.12	3.86	45.7	35.0
	100	52.9	76.9	98.3	9.79	7.04	3.92	45.3	34.9
	150	53.9	76.3	99.1	10.44	6.96	3.87	45.7	35.6
	200	53.7	79.1	99.1	11.81	6.86	4.19	47.0	35.9
	250	53.4	79.0	99.6	13.06	6.49	4.49	45.0	34.4
	Mean	53.3	77.3	98.6	10.56	6.91	4.01	45.4	34.9
Tardis	0	47.3	69.6	61.2	10.52	7.81	3.44	39.9	28.3
	50	49.4	71.7	78.7	9.77	7.95	3.56	41.3	30.0
	100	49.7	72.7	82.6	9.73	8.07	3.33	40.8	30.5
	150	50.4	74.7	67.4	11.33	7.90	3.48	42.7	30.3
	200	50.7	73.1	82.3	12.19	7.76	3.83	42.4	30.8
	250	50.3	73.5	88.2	13.98	7.42	3.91	41.4	30.5
	Mean	49.7	72.5	76.7	11.25	7.82	3.59	41.4	30.1
N treatment <i>p</i> value		<.001	<.001	<.001	<.001	<.001	<.001	0.001	<0.001
Variety (V) p value		<.001	<.001	<.001	<.001	<.001	<.001	<.001	<0.001
V x N p value		<.001	<.001	<.001	0.355	0.009	<.001	0.094	0.005

**Table 12** Thousand grain weight (TGW) of grain and de-hulled groat, milling quality and grain composition traits from 2014-15 experiment grown at ADAS Rosemaund by variety and nitrogen treatment.

Variety	Ν	Specific	Kernel	Hullability	Protein	Oil	β-	TGW	TGW
	applied	Weight		(%)	(%)	(%)	glucan	whole	groat
	(kg/na)	(Kg/III)	(%)				(%)	(a)	(g)
Balado	0	49.7	71.0	86.8	10.17	6.81	4.43	40.4	31.4
	60	50.5	71.9	88.0	9.65	6.74	4.56	41.6	32.2
	120	50.0	72.3	95.1	11.27	6.49	4.62	40.8	31.7
	180	49.2	73.7	95.4	14.27	5.99	5.08	40.2	31.5
	230	48.9	74.4	97.1	15.10	5.77	4.94	41.5	33.4
	280	47.8	73.8	95.9	17.25	5.74	5.10	39.4	31.1
		49.4	72.9	93.1	12.95	6.26	4.79	40.7	31.9
Gerald	0	50.3	70.4	82.4	10.06	6.53	3.68	34.2	25.9
	60	51.0	71.8	86.3	9.73	6.43	3.58	34.5	26.2
	120	51.1	71.2	90.8	11.44	6.50	3.63	33.3	25.6
	180	49.3	70.9	90.3	14.77	6.29	4.28	31.8	23.7
	230	48.3	71.3	94.0	15.44	6.15	4.06	31.6	23.4
	280	48.0	70.9	95.2	16.35	6.11	4.15	30.7	22.5
		49.7	71.1	89.9	12.97	6.34	3.90	32.7	24.6
Mascani	0	51.1	72.9	94.2	11.71	5.91	4.37	39.2	31.8
	60	51.1	73.3	96.3	12.17	5.57	4.48	41.2	34.1
	120	52.9	77.4	98.1	12.58	5.47	4.35	43.8	35.6
	180	52.8	77.5	99.3	15.02	4.97	4.52	43.3	35.2
	230	53.0	78.4	99.2	15.88	4.82	4.63	44.1	36.3
	280	52.5	77.9	99.3	16.75	4.90	4.68	43.2	34.5
		52.2	76.2	97.7	14.02	5.28	4.51	42.5	34.6
Tardis	0	47.6	69.5	77.1	9.73	7.01	3.94	39.3	29.5
	60	47.9	70.4	86.5	10.10	6.76	3.83	40.1	30.4
	120	48.1	71.6	88.8	11.79	6.50	3.99	39.9	29.9
	180	47.1	71.7	91.5	14.08	6.34	4.24	37.7	28.6
	230	45.5	71.6	90.6	16.02	6.09	4.45	37.1	27.7
	280	44.3	71.5	91.1	16.97	6.11	4.80	35.3	26.5
		46.8	71.1	87.6	13.12	6.47	4.21	38.2	28.8
N treati	ment <i>n</i>								
value		<.001	<.001	<0.001	<.001	<.001	<.001	0.001	0.002
Variety (V) p		<.001	<.001	<0.001	<.001	<.001	<.001	<.001	<0.001
V x N p value		<.001	<.001	0.037	0.001	0.002	0.082	<.001	0.001

**Table 13** Mean values for milling quality traits, thousand grain weight (TGW), screenings (grain less than 2mm),  $\beta$ -glucan content and grain dimensions of grain from 2016-17 experiment grown at ADAS Rosemaund by variety and nitrogen treatment.

Variety	N applied	Specific	Kernel	Hullability	Screenings	TGW (g)	grain	grain	grain	β-
	(kg/ha)	(kg/bl)		(%)	(%)		length	(mm)	roundness	Glucan
		(Kg/III)	(70)				(11111)	(11111)		(70 DIVI)
Griffin	0	52.02	78.83	96.8	0.48	45.71	13.41	3.15	0.24	3.49
	60	49.10	77.58	96.0	1.76	41.98	13.87	2.98	0.22	4.26
	120	48.42	76.77	98.4	4.61	37.92	13.41	2.89	0.22	4.28
	160	48.47	76.88	98.0	4.89	37.49	13.31	2.89	0.22	4.73
	220	48.07	76.77	97.8	3.99	39.51	13.55	2.91	0.22	4.95
	280	48.26	77.32	99.6	5.30	37.29	13.13	2.87	0.22	4.97
	Mean	49.06	77.36	97.8	3.50	39.99	13.45	2.95	0.22	4.45
Maestro	0	52.70	77.44	98.7	3.79	35.26	10.98	2.92	0.27	3.69
	60	50.93	77.37	99.0	7.26	34.31	11.36	2.84	0.25	3.77
	120	51.00	76.98	99.6	12.74	31.58	11.16	2.76	0.25	3.93
	160	49.34	76.59	99.6	17.91	30.50	11.43	2.75	0.24	4.37
	220	48.38	76.83	99.8	18.77	30.63	11.54	2.75	0.24	4.63
	280	47.13	75.74	99.8	24.71	29.27	11.55	2.73	0.24	4.37
	Mean	49.91	76.82	99.40	14.20	31.93	11.34	2.79	0.25	4.13
Mascani	0	53.23	78.30	98.9	0.74	44.67	12.09	3.18	0.27	4.44
	60	53.05	78.64	99.5	0.91	42.03	12.00	3.10	0.26	4.25
	120	52.35	78.83	99.6	2.03	39.42	11.72	3.03	0.26	4.38
	160	51.86	78.24	99.8	2.45	38.96	11.84	3.01	0.26	4.55
	220	51.99	78.57	99.8	2.08	39.50	11.95	3.04	0.26	4.65
	280	51.94	78.60	99.8	2.68	37.70	11.66	2.99	0.26	5.00
	Mean	52.40	78.53	99.6	1.81	40.38	11.88	3.06	0.26	4.55
N treatmer	nt <i>p</i> value	<.001	0.514	<.001	<.001	<.001	0.053	<.001	<.001	<.001
Variety (V)	p value	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	0.002
V x N p value		0.008	0.859	0.056	<.001	0.250	0.038	0.008	<.001	0.199





#### 4.3.1. Specific Weight and Thousand Grain Weight

Specific weight is routinely used as a measure of grain quality in the grain trade and is the mass of grain which fits into a specified volume. A significant effect of variety, nitrogen treatment and interaction between the two was found (Tables 10, 11, 12, 13, Figure 6). In general, specific weight decreased at higher nitrogen application levels, as found in previous studies (Marshall et al., 1987; Browne et al., 2003; Finnan et al., 2019). The finding that at high N application rates specific weights are compromised could mean as a result that some varieties are below milling industry specifications when nitrogen is applied to maximise yield. It was not possible to determine the Nopt for grain yield in both the 2013-14 trials because yields were continuing to increase at the highest N rates, but for the trials in 2014-15 and 2016-17 grain yield Nopt were calculated. Table 15 indicates both the specific weights obtained for each variety at the grain yield N opt and the N application rate at which the maximal value of specific weight was obtained. With respect to the lowest available nitrogen treatments, specific weight was also reduced suggesting that there is an optimal level of nitrogen required for maximal hectolitre weight. However, the specific weight of Mascani was not only high in all trials but was enhanced in the Rosemaund 2014- 15 trial with increasing levels of nitrogen (Table 12). Specific weight is influenced by both grain density and packing density components. These results suggest that conditions that result in enhanced grain filling and thus higher thousand grain weight can increase hectolitre weight. A summary of the mean specific results for each trial is shown in Figure 7. Across all trials and treatments, Mascani had the highest mean specific weight of 52.5 kg/hL and Tardis the lowest of 48.9 kg/hL (Table 14). Mascani also had the smallest range in specific weights across all trials and treatments (Figure 8)



**Figure 7** Effect of Nitrogen (N supplied plus soil N measured using SMN tests) on mean specific weight. IBERS 2014 (Lydbury North), ADAS 2014 (Rosemaund), ADAS 2015 (Rosemaund) mean of Mascani, Gerald, Tardis and Balado; ADAS2017(Rosemaund) mean of Mascani, Griffin and Maestro

Thousand grain weight (TGW) values showed statistically significant differences between varieties (Tables 10, 11, 12, 13), levels of nitrogen applied and for the interaction between the two factors but there was not a consistent effect across the 4 sites studied. In some varieties, higher levels of nitrogen can result in a greater proportion of smaller grain (and groats) and this can result in an increase in screenings. A significant interaction between variety and nitrogen treatment was also found. Mascani not only had the highest mean TGW in each trial, but its TGW also increased with the addition of nitrogen in 3 out 4 of the trials, whereas Gerald had the lowest TGW across each trial and its TGW decreased with increasing levels of nitrogen. A similar result was found with Maestro in the 2016-17 trial with TGW decreasing with increasing levels of nitrogen. Balado and Tardis displayed a varying response depending on the trial, with their TGW decreasing in the 2014-15 at higher nitrogen levels, and a less clear effect revealed in the other two trials. Previous studies have shown that increasing nitrogen application results in lower individual lower individual grain weight (Peltonen-Sainio and Peltonen, 1995; Chalmers et al., 1998; Weightman et al., 2004; Ma et al., 2017) but this study suggests that the response of TGW to nitrogen is oat variety dependent. TGW is often used as an indicator of grain quality since it is related to grain plumpness, with high values reflecting well-filled grains.

## 4.3.2. Kernel Content and Hullability

Kernel content (the proportion of groat to whole grain) is the characteristic most closely related to the miller's extract yield of product (Browne *et al.*, 2003). Kernel content displayed significant differences between varieties and nitrogen application with the highest values obtained in all trials by Mascani (Tables 10, 11, 12, 13). Mean kernel content increased with increasing levels of nitrogen applied (Figure 9) except at the 2016-17 Rosemaund trial where the effect of nitrogen on kernel content was not significant (Table 13). Thus, nitrogen applications that favour grain yield also result in enhanced kernel content (Table 15). Kernel content did not show a strong correlation with specific weight (Figure 10) which suggests that specific weight is not a good predictor of milling yield. Across all treatments and trials, Mascani had the highest kernel content at 77.3% across treatments (Figure 8) with the newer varieties Griffin and Maestro also displaying high kernel contents, albeit that they were only grown in one trial (Table 14).

Variety	Kernel Content (%)	Hullability (%)	Specific Weight (kg/hl)	β-Glucan (% DM)	Oil (% DM)	TGW (g)
Mascani	77.35	98.80	52.49	4.22	6.35	43.04
Gerald	72.30	90.86	51.93	3.51	7.13	35.69
Balado	73.00	88.88	49.85	4.52	7.11	42.60
Tardis	72.06	84.48	48.92	3.84	7.48	40.74
Griffin	76.71	97.94	49.06	4.36	6.77	39.03
Maestro	76.82	99.40	49.91	4.17	6.96	30.40

**Table 14** Variety means for grain quality traits across all treatments and trials. Griffin and Maestro only grown in 2016-17 Rosemaund trial in place of Gerald, Tardis and Balado.



**Figure 8.** Box plot of A, specific weight, B, hullability, C, Kernel Content, D  $\beta$ -glucan values for four winter oat varieties, Mascani, Gerald, Balado and Tardis, across all treatments at Lydbury North 2013-14, ADAS Rosemaund 2013-14 and ADAS Rosemaund 2014-15. The box plot represents the first and third quartiles, with the horizontal line inside the box indicating the median and the x representing the mean. Whiskers indicate variability outside the upper and lower quartiles, and any point outside those lines or whiskers is considered an outlier.

**Table 15** Values at fitted grain yield economic optimum N rate (Nopt) and the maximal values obtained for specific weight, kernel content, hullability and  $\beta$ -glucan content of grain from 2013-14, 2014-15 and 2016-17 trials at Rosemaund and 2013-14 trial at Lydbury North for each variety tested. N applied to obtain the maximal value indicated except where there was no significant effect of nitrogen (n.s.). N opt not able to be calculated for either trial in 2013-14.

		S	Specific Wt., kg/hl			Kernel Cont	ent, %		Hullability	r, %		β-glucan, %	
	Nopt kg/ha	at yield N <sub>opt</sub>	Maximal value	N applied for maximal value kg/ha	at yield N <sub>opt</sub>	Maximal value	N applied for maximal value kg/ha	at yield N <sub>opt</sub>	Maximal value	N applied for maximal value kg/ha	at yield N <sub>opt</sub>	Maximal value	N applied for maximal value kg/ha
2013-14 Ros	semaund, l	N range (	)-250 kg/ha	a									
Mascani	n/a		53.9	150		79.1	200		99.6	250		4.49	250
Tardis	n/a		50.7	150		74.7	150		88.2	250		3.91	250
Gerald	n/a		52.8	200		73.8	250		94.1	200		3.53	200
Balado	n/a		51.5	100		74.5	150		87.9	150		4.31	200
2013-14 Lyd	bury Nortl	h, <mark>N rang</mark>	e 0-200 kg	/ha									
Mascani	n/a		53.2	0		78.1	200		99.8	200		4.25	200
Tardis	n/a		52.7	0		73.4	150		93.6	200		3.89	200
Gerald	n/a		55.1	0		75.2	200		98.1	200		3.53	200
Balado	n/a		51.4	50		74.9	200		95.2	200		4.62	150
2014-15 Ros	emaund, l	N range (	)-280 kg/ha	a									
Mascani	207	52.9	53.0	230	77.9	78.4	230	99	99.4	230	4.55	4.70	280
Tardis	170	47.2	48.1	60	71.7	71.7	180	91.4	91.6	180	4.15	4.81	280
Gerald	160	50.0	51.0	120	71.3	71.3	160	91.6	95.0	280	3.92	4.22	280
Balado	172	49.6	50.2	60	73.4	74.4	230	95.4	97.1	230	4.87	5.11	280
2016-17 Ros	emaund, l	N range (	)-280 kg/ha	a									
Mascani	98	52.5	53.4	0	78.6	79.0	n.s.	99.6	99.9	230	4.16	4.84	280
Maestro	98	50.8	52.5	0	77.2	77.6	n.s.	99.3	99.9	280	4.01	4.70	280
Griffin	98	48.9	51.7	0	76.7	77.4	n.s.	97.5	99.5	280	4.31	4.84	230



**Figure 9.** Effect of nitrogen applied on mean kernel content of individual varieties at A, IBERS 2014 (Lydbury North), B, ADAS 2014 (Rosemaund), C, ADAS 2015 (Rosemaund) and D, ADAS 2017 (Rosemaund)



**Figure 10.** Relationship between specific weight and kernel content of grain from all varieties grown at Lydbury North 2013-2014 and ADAS Rosemaund in 2013-14, 2014-15 and 2016-17.



**Figure 11.** Effect of nitrogen applied on mean hullability of individual varieties at A, Lydbury North 2013-14, B, ADAS Rosemaund 2013-14 and C, ADAS Rosemaund 2014-15

Hullability, or the ease of husk removal, has important implications for mill efficiency. Removal of the hull from the groat, the first stage in the oat milling process, is energy consuming so costs for milling are increased when oat grain is harder to dehull and if it fails to de-hull on the first pass through the dehuller. A variety with poor hullability will require greater impact speeds within the dehuller during milling and result in greater kernel breakage, thereby reducing the miller's extract yield. Hullability was assessed in this project as the percentage of grain remaining unhulled when determining the kernel content with a value of 100% representing complete removal of the hull during the procedure. Significant differences in hullability were obtained both between varieties and between nitrogen rates and the interaction between them (Tables 10, 11, 12, 13). This was particularly apparent with the varieties used in the 2013-14 and 2014-15 trials with Tardis having a mean hullability across treatments of only 84.5% (Table 14) compared to Mascani with a hullability of 98.9%. Increasing nitrogen application resulted in higher hullability in all varieties (Figures 6, 11, 12). This response was not linear, however, with little significant improvement in hullability in some varieties between the higher rates of nitrogen applied (Table 15). For Mascani and the newer varieties Maestro and Griffin very high hullabilities were found at all nitrogen application rates. Another way to express the stability of traits across treatments is to conduct joint regression analysis (Finlay & Wilkinson, 1963) in which variety performance is plotted against mean performance of all varieties at that treatment (Figure 13). The stability in performance of the hullability of Mascani is clearly apparent with a value of 0.14 for the gradient of the regression line compared to 1.75 for Tardis. The narrow range of hullabilities obtained for Mascani as compared to Tardis, Gerald and Balado is also apparent in Figure 8.



**Figure 12** Effect of nitrogen (N supplied plus soil N measured using SMN tests) on mean hullability. IBERS 2014 (Lydbury North), ADAS 2014 (Rosemaund), ADAS 2015 (Rosemaund) mean of Mascani, Gerald, Tardis and Balado, ADAS2017 (Rosemaund) mean of Mascani, Griffin and Maestro



**Figure 13** Joint regression analysis of hullability (%) across nitrogen treatments of four winter oat varieties grown at Rosemaund and Lydbury North in 2013- 2014 and at Rosemaund in 2014-2015.

#### 4.3.3. Grain size

To investigate the effect of grain size on grain quality traits, grain area (mm<sup>2</sup>), length (mm) and width (mm) values were determined using image analysis (Tables 13, 16). In addition, the ratio between the length and width (referred to as grain roundness) was calculated as a measure of grain shape. Area, width and length of the grain displayed significant differences between varieties and levels of nitrogen applied in most trials (Tables 13, 16). In general, mean grain area and length values increased with increasing levels of nitrogen applied whereas mean grain width displayed lower values at higher levels of nitrogen. The consequence of an increase in grain length and a decrease in mean grain width at higher levels of nitrogen was that the mean grain roundness decreased significantly with increasing nitrogen fertiliser levels (Tables 13, 16). Grain roundness

was also significantly different between varieties with both Mascani and Gerald having rounder grain than Tardis and Balado.

Frequency distribution analysis of the individual grain data was conducted by site, season and variety. The datasets were analysed to determine their bi-modality and to establish the mean, standard deviation and the proportion, between any subpopulations observed (Symons & Fulcher, 1988). A bimodal frequency distribution was found for grain and groat area and length representing the primary and secondary grain found in each oat spikelet (Figure 14). At the nil nitrogen rate, a clear bimodal distribution was obtained. However, in the variety Tardis a wider range of grain sizes was obtained at a higher N treatment. This change in bimodality was not obtained to the same extent with Mascani.



**Figure 14** Frequency diagram indicating distribution of individual grain size (grain area, mm<sup>2</sup>) in a 25 g sample of grain of Tardis (A, B) and Mascani (C, D).grown at 0 applied N (A, C) and at 250 kg N/ha (B, D) at Rosemaund 2014-15
	Width Grain (mm)	Length Grain (mm)	Roundness Grain (mm)	Area Grain (mm²)		
IBERS 2014 Variety						
Ralado	3 208bc	12 363c	0.260a	28 312h		
Gerald	3 141a	10.915a	0.2004	24 763a		
Mascani	3 2390	11 724b	0.2000 0.277h	27.290h		
Tardis	3 178h	12 339c	0.258a	27.200b		
Nitrogen level (kg/ha)	0.1705	12.0000	0.2004	21.0005		
( <b>iiii: ogoi: iovoi</b> ( <b>iiig</b> /iiii)	3 181	11 279a	0.283c	25 731a		
50	3 217	11.675ab	0.276c	26.877ab		
100	3 186	11.807abc	0.271ab	26.878ab		
100	3 185	12 189bc	0.262a	27.687h		
200	3 190	12.10000 12.226c	0.262a	27.865b		
200	5.150	12.2200	0.2028	27.0000		
N treatment p value	0.138	<0.001	<0.001	<0.001		
Variety (V) p value	<0.001	<0.001	<0.001	<0.001		
V x N p value	0.023	0.267	0.241	0.151		
ADAS 2014 Variety						
Balado	3.164c	12.610c	0.251b	28.657c		
Gerald	3.016a	11.109a	0.272d	24.445a		
Mascani	3.196d	12.184b	0.262c	28.006b		
Tardis	3.117b	13.040d	0.239a	28.638c		
Nitrogen level (kg/ha)						
0	3.109ab	12.279bc	0.254b	27.206a		
50	3.147c	12.210ab	0.258c	27.469ab		
100	3.134bc	10.044a	0.261c	27.183a		
150	3 136bc	12 122a	0.2590	27 346a		
200	3 14c	12 414c	0.254b	27 990b		
250	3.075a	12.345c	0.250a	27.425ab		
N treatment p value	<0.001	<0.001	<0.001	<0.001		
Variety (V) p value	<0.001	<0.001	<0.001	0.001		
V x N p value	0.101	0.031	<0.001	0.218		
ADAS 2015 Variety						
Balado	3.059c	12.173b	0.251b	27.091b		
Gerald	2.838a	11.176a	0.254b	23.566a		
Mascani	3.063c	12.100b	0.253b	27.253b		
Tardis	2.994b	12.925c	0.232a	27.589b		
Nitrogen level (kg/ha)						
0	3.008	11.922	0.253b	25.977		
60	3.024	12.049	0.252b	26.415		
120	2.998	11.983	0.251ab	26.190		
180	2.962	12.086	0.245ab	26.254		
230	2.977	12.221	0.244a	26.674		
280	2.960	12.300	0.241a	26.738		
N treatment p value	0.002	0.008	<0.001	0.117		
Variety (V) p value	<0.001	<0.001	<0.001	<0.001		
V x N p value	0.035	0.913	0.004	0.957		

**Table 16**. Mean area ( $mm^2$ ), width (mm) and length (mm) from grain of the four winter oat varietiesunder each level of nitrogen fertiliser (kg/ha) at ADAS2014, ADAS2015 and IBERS2014 trials.Letters indicate significant differences between mean values at p < 0.05.</td>

#### 4.3.4. Grain composition

Proximal grain composition was assessed by NIR on dehulled groats. Total protein content significantly increased in a linear manner with nitrogen application (Tables 10, 11, 12). In the ADAS Rosemaund 2013-14 and 2014-15 trials there was also a significant difference between varieties but protein content was affected more by nitrogen application than by variety. The highest values were obtained at the lowest yielding trial, Rosemaund 2014-15, with a maximum value of 17.25% protein for Balado at the 280 kg N/ha treatment. In the Rosemaund 2014-15 trial there was also a significant interaction between nitrogen and variety with at the lowest nitrogen levels, Mascani having the highest protein content whereas Balado had the highest value at the highest nitrogen level. Total oil content was significantly different between varieties in all trials (Tables 10, 11, 12) and decreased significantly in response to nitrogen as also found by Yan *et al.* (2017). Mascani had the lowest mean oil content across treatments and trials with a value of 6.35% (Table 15).

The grain  $\beta$ -glucan content was significantly different between varieties and also significantly increased with nitrogen application in all four trials (Tables 10, 11, 12 and 13), with Balado displaying the highest average value across trials (4.5%) and Gerald the lowest (Table 14). A wide range in  $\beta$ -glucan contents was obtained across treatments for each variety (Figures 8, 15).Maximal levels of  $\beta$ -glucan were obtained at the highest nitrogen application rates (table 15) although for some trials and varieties there was no significant increase between the 2 highest nitrogen application rates used.

### 4.3.5. Relationship between traits

Correlation analysis was conducted by site and by variety of all physical and chemical quality traits, for grain size and shape parameters and grain yield. A number of correlations were obtained that were specific to a particular trial or variety and only those that were found across all trials are reported here. Total nitrogen applied had a positive correlation with yield and protein content at all sites, showing increasing values of both traits with higher levels of nitrogen. Yield displayed a curvilinear response to nitrogen whereas grain protein content increased with applied nitrogen in a linear manner in each trial. Although thousand grain weight was significantly affected by nitrogen it was not correlated with grain yield. Grain yield was, however, highly correlated with the number of grains m<sup>-2</sup> (Figure 16). The grain number m<sup>-2</sup> is a combination of panicle number m<sup>-2</sup> and grain number per panicle, both of which increased significantly as nitrogen application increased (Table 17).



**Figure 15.** Effect of nitrogen on mean  $\beta$ -glucan content of individual varieties at A, IBERS 2014 (Lydbury North), B, ADAS 2014 (Rosemaund), C, ADAS 2015 (Rosemaund) and D, ADAS 2017 (Rosemaund)



**Figure 16** Relationship between grain number per m<sup>2</sup> and grain yield at ADAS Rosemaund 2013-14, 2014-15 and Lydbury North 2013-14 sites for four winter oat varieties.

Thousand grain weight was, for all sites, significantly correlated (<0.001) with grain width (Figure.17) and also displayed strong positive correlation coefficients with grain area. Both thousand grain weight and thousand groat weight were significantly correlated with kernel content (Figure 18). Thousand grain weight displayed correlation with specific weight in a variety specific manner (Figure 19) with a significant correlation only found for Gerald and Tardis. Specific weight, however, displayed a significant strong positive correlation (<0.001) with grain roundness across all varieties and trials (Figure 20).



**Figure 17** Relationship between grain width and thousand grain weight (TGW) at Rosemaund 2013-14 (ADAS2014), 2014-15 (ADAS2015), 2016-17 (ADAS2017) and Lydbury North 2013-14 (IBERS2014) sites



**Figure 18** Relationship between kernel content and thousand groat weight at Rosemaund 2013-14 (ADAS2014), 2014-15 (ADAS2015) and Lydbury North 2013-14 (IBERS2014) sites



**Figure 19** Relationship between specific weight and thousand grain weight (TGW) for the winter oat varieties Mascani, Gerald, Tardis and Balado at ADAS Rosemaund 2013-14 and 2014-15 and Lydbury North 2013-14 sites



**Figure 20** Relationship between grain roundness (as determined by the ratio of grain length to width) and Specific Weight for all varieties at all sites (Rosemaund 2013-14, 2015-16, 2016-17 and Lydbury North 2013-14 sites)

**Table 17.** Mean panicle number per m<sup>2</sup>, grain number per m<sup>2</sup> and grain number per panicle. Mean values by variety and by nitrogen application level for Lydbury North 2013-14 (LN14, N levels 0, 50, 100, 150, 200 kg/ha), ADAS Rosemaund 2013-14 (RM14, N levels 0, 50, 100, 150, 200, 250 kg/ha) and ADAS Rosemaund 2014-15 (RM15, N range 0, 60, 120, 160, 220, 280 kg/ha).

	panicles/m2			grain no/m2						grain no per panicle								
	LN1	4	RM	14	RM1	5	LN14	RM14		RM15	115 L'		14 RM		14 RM15		15	
Variety																		
Balado	300.2	а	300.1	а	400.9	а	17128.9	b	13301.3	b	9006.3	ab	56.6	d	43.4	d	22.0	С
Gerald	392.0	bc	378.5	b	432.7	а	18942.6	с	14867.0	d	8014.4	ab	48.3	С	38.2	с	18.4	ab
Mascani	399.8	с	415.2	b	502.7	b	15307.6	а	12117.5	а	7938.7	ab	38.0	а	28.5	а	15.4	а
Tardis	363.2	b	406.3	b	482.5	b	17563.8	b	13310.0	с	9628.9	b	47.8	b	32.1	b	19.6	bc
Nitrogen level																		
1	251.8	а	278.6	а	321.9	а	10096.8	а	5580.8	а	4487.1	а	40.4	а	20.5	а	13.9	а
2	353.7	b	358.5	b	425.6	b	15603.2	b	10723.9	bc	6843.9	b	44.8	b	31.2	b	16.3	ab
3	414.6	с	378.4	bc	482.1	С	18426.9	с	12772.8	ab	9732.0	с	46.0	bc	34.7	bc	20.6	bc
4	408.1	с	422.6	с	480.0	С	19772.1	d	15860.3	bc	9400.6	с	49.2	с	38.3	с	19.9	bc
5	390.8	с	404.1	bc	514.0	С	22279.8	е	17301.7	с	11083.8	с	58.0	d	43.6	d	21.7	С
6			407.9	bc	504.6	С			18453.1	ab	10334.9	с			45.8	d	20.8	С
N treatment <i>p</i> value	<.001		<.001		<.001		<.001		<.001		<.001		<.001		<.001		<.001	
Variety (V) p value	<.001		<.001		<.001		<.001		<.001		0.002		<.001		<.001		<.001	
V x N p value	0.506		0.451		0.079		0.101		0.02		0.826		0.114		0.147		0.815	

### 4.4. QTL-NIL analyses

Flowering time indicates the transition from the vegetative to the reproductive phase in plants and is a major determinant for biomass accumulation and grain filling period length which affect subsequent grain yield and grain quality. Flowering time is a complex trait. Generally, flowering time is accelerated by longer days conditions associated with the more favourable conditions of spring and summer. The timing of flowering is a decisive factor in the adaptation of oats to different growing environments. For example, some oat varieties require low temperatures for floral initiation, a process called vernalisation. These adaptation processes enable crops to form different growth habits, such as winter and spring types.

QTL analysis of the Buffalo x Tardis winter oat mapping population previously conducted identified a number of regions of the genome associated with flowering time and yield component traits with consequent effects on grain quality. For example, a major flowering time QTL in which the Buffalo parent provided the late flowering allele co-located with the dwarfing gene (dw6) on linkage group Mrg04. The effect of this QTL was such that panicle emergence for progeny homozygous for the dwarf alleles from Buffalo was approximately 8 days later than for those with the Tardis alleles. QTL analysis suggested that the effect on flowering time of this QTL was dependent on both photoperiod and vernalisation; if the population was sown in the spring, no significant QTL on Mrg04 for flowering time was obtained. However, a major QTL on Mrg21 was found in which the Buffalo parent provided the early flowering allele. This QTL on Mrg21 was not expressed when the population was sown in the autumn. This region of the genome was also associated with winterhardiness. An additional QTL associated with flowering time was also obtained on Mrg20, again dependant on date of sowing. Mrg20 and Mrg21 represent homeologous chromosomes (Chaffin et al., 2016). Nava et al. (2012) mapped an ortholog of the vernalisation gene Vrn1 to markers now placed on Mrg20, and QTL for vernalisation response have also been mapped to this linkage group (Holland et al., 2002). Similarly, the Mrg21 linkage group contains markers associated with a vernalisation response QTL (Holland et al., 1997, 2002; Nava et al., 2012).

The set of QTL-NILs for height and flowering time used in this project comprise 6 genotypes plus the 2 parental control lines as outlined in Table 18. Each QTL-NIL was developed following at least 3 generations of marker assisted backcrossing to either the Buffalo and Tardis parent using RILs from the Buffalo x Tardis mapping population selected to be as close to the desired genotype as possible as donor genotypes. At least 2 versions of each genotype were developed from independent backcrossing. Each QTL-NIL was verified by using the oat 6k SNP chip (Tinker *et al.*, 2014). Examples of genotypes developed are shown in Figures 21-23. These lines represent a considerable resource for understanding the physiological basis of these QTL and to provide a foundation for the identification for the genes underlying them and of pleiotropic effects. In addition,

selected QTL have also been introgressed from either Buffalo or Tardis into the elite winter oat cultivar Mascani using marker assisted backcrossing.



**Figure 21** Genotype of line 2012-137/5/1 in which the QTL on Mrg04 containing *dw6* has been introgressed from Buffalo into a Tardis background. Markers with alleles from Tardis indicated in blue whereas those from Buffalo indicated in red. Marker order and linkage group numbering as in Chaffin *et al.*, 2016.

**Table 18** Summary of QTL-NILs used in field trials at Rosemaund and Aberystwyth indicating background genotype, QTL introgressed and associated traits. Mrg designations refer to the consensus linkage group numbering (Chaffin *et al.* 2016). Designation refers to the allelic complement at Mrg20, Mrg21 and Mrg04 respectively. A represents alleles from the Tardis parent and B, alleles from the Buffalo parent.

Breeders Codes	Designation	Name	Background genotype	QTL introgressed							
				Allele	Buffalo x Tardis Linkage Group	Nearest marker to peak QTL	Trait targeted	Other traits associated with QTL			
	AAA		Tardis (A)	None (parental control)							
	BBB		Buffalo (B)	None (pa	arental cont	rol)					
2012-137/5/1 2012-137/5/5	AAB	Tardis +B_Mrg04	A	В	17 (Mrg04)	CC2606_241	Height (Dw6)	Flowering time Yield			
2012-139/6/25 2012-125/1/27	BBA	Buffalo +A Mrg04	В	A				Grain Quality			
2012-134/1/35 2012-134/1/36	ABA	Tardis +B_Mrg21	A	В	13		Flowering time (spring sown)	Winter hardiness, Juvenile plant stature,			
2013-214ACnX/4 2012-124/23/19	BAB	Buffalo +A Mrg21	В	A	(Mrg21)	AM87		Flowering time (autumn sown) Grain Quality			
2012-130/5/2 2012-130/5/5	BAA	Tardis +B_Mrg20	A	В	1	Vrp1	Flowering time	Winter hardiness			
2013-212ACnl/23 2013-212ACnVII/11	ABB	Buffalo +A Mrg20	В	A	(Mrg20)			Grain Quality			



**Figure 22** Genotype of line 2012-134/1/35 in which QTL associated with flowering time on Mrg21 has been introgressed from Buffalo into a Tardis background. Markers with alleles from Tardis indicated in blue whereas those from Buffalo indicated in red Linkage group numbering as in Chaffin *et al.*, 2016



**Figure 23** Genotype of line 2013-214ACnX/4 in which QTL associated with flowering time on Mrg21 has been introgressed from Tardis into a Buffalo background. Markers with alleles from Tardis indicated in blue whereas those from Buffalo indicated in red. Linkage group numbering as in Chaffin *et al.*, 2016.

These QTL-NILs along with control varieties were grown in replicated trials at both ADAS Rosemaund and at Aberystwyth over 2 field seasons (2015-16 and 2016-17) to validate the identified QTL and to further elucidate the role of selected QTL on a wide range of traits including flowering time, height, flag leaf width, grain size, grain number per panicle, tillering, grain quality and disease resistance. A separate glasshouse experiment in the phenomics facility in Aberystwyth compared vernalised and un-vernalised plants and confirmed the involvement of the Tardis allele on Mrg21 in the vernalisation response and the interaction of height and flowering time genes on Mrg20, Mrg21 and Mrg04 on both stem extension and heading date and other yield components. The QTL-NILs have also been grown at Harper Adams University in an associated studentship partly funded by AHDB to validate and dissect QTL associated with mycotoxin accumulation and developed understanding of the infection process of *Fusarium langsethiae*.

#### 4.4.1. Plant height and Flowering time

Validation of the QTL for plant height are shown in Figure 24 which presents the mean results from the four field trials at ADAS Rosemaund and Aberystwyth. Buffalo contains the dwarfing gene, *dw6*, and is significantly shorter than Tardis. Tardis +B\_Mrg04 represent lines that have the Buffalo alleles on Mrg04 associated with *dw6* introgressed into Tardis and on average they were 49 cm shorter than the Tardis parent. Similarly, the reciprocal lines, Buffalo +A\_Mrg04, are 53 cm taller than the Buffalo parent. The Mrg21 reciprocal introgression lines, however, have little effect on plant height (Figure 24).



**Figure 24** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on plant height. Buffalo +A represents a line in which the Tardis alleles at a particular QTL have been introgressed into Buffalo, and Tardis +B represents a line in which the Buffalo alleles at a particular QTL have been introgressed into Tardis. Mean ( $\pm$  s.e.m) results of 4 field trials (ADAS Rosemaund 2015-16, 2016-17 and Aberystwyth 2015-16, 2016-17). Letters indicate significant difference between mean values at p < 0.05.

Mean panicle emergence results of these QTL-NILs and the parents from the 4 autumn sown trials at ADAS Rosemaund and Aberystwyth were significantly different (p <0.001, Figure 25). Flowering time was estimated based on observations of panicle emergence as is usual in cereals because floral initiation and anthesis are difficult to detect. The effect of the QTL on Mrg04 containing *dw6* is clear with the Buffalo +A\_Mrg04 line having a mean panicle emergence date 4 days earlier than the Buffalo parent whereas the reciprocal line, Tardis +B\_Mrg04, was 7.5 days later than the Tardis parent. The introgression of the Tardis alleles on Mrg04 into Buffalo resulted in a similar panicle emergence to the Tardis parent. The effect of the flowering time QTL on Mrg21 is also apparent with the Buffalo +A\_Mrg21 lines having a mean panicle emergence 1.4 days later than Buffalo and the reciprocal Tardis +B\_Mrg21 lines having a mean panicle emergence 1.4 days earlier than Buffalo and the reciprocal Tardis +B\_Mrg21 lines having a mean panicle emergence 1.4 days



**Figure 25** Mean ( $\pm$  s.e.m) results of 4 autumn sown field trials showing the effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on panicle emergence. Letters indicate significant differences between mean values at p < 0.05





The effect of the QTL on Mrg20 and Mrg21 was much more apparent when these genotypes were sown in the spring (Figure 26). These trials were conducted in Aberystwyth over 2 growing seasons. In Figure 26, the time to panicle emergence is expressed in growing degree days which

are a cumulative measure of temperature. The introgression of the Tardis *dw6* alleles into Buffalo had little effect on panicle emergence when spring sown although there was still a delay in panicle emergence found in the reciprocal introgression line. Date of sowing also affected final plant height with spring sown lines always shorter than those sown in the autumn (Figure 27). The effect of Mrg04 on plant height was apparent irrespective of when the plants were sown suggesting that the effect of this region of the genome on flowering time is under photoperiodic control.



**Figure 27** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on plant height when trials sown in the autumn or spring. Mean of 2 trials conducted in 2016-17 and 2017-18 at Aberystwyth

### 4.4.2. Grain yield

These QTL also significantly affected grain yield and grain quality (Figures 28, 29). The mean yield of Buffalo across the 4 autumn sown trials was significantly less than Tardis. The Mrg04 introgression from Tardis into Buffalo increased yield by an average of 0.9 t/ha. Conversely the Mrg21 introgression from Tardis resulted in a decrease in yield of approximately 1.2 t/ha. Comparable reciprocal results were found with the introgressions from Buffalo into Tardis. A similar result was found for thousand grain weight (TGW, Figure 29) with the Buffalo + A\_Mrg04 line having a TGW 5g higher than the Buffalo parent. The Buffalo + A\_Mrg21 line did not display a significant change in TGW compared to Buffalo. The TGW results mirrored that of the panicle emergence data with earlier flowering lines having a higher TGW due to potentially a longer grain filling period.



**Figure 28** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on grain yield. Mean ( $\pm$  s.e.m) results of 4 autumn sown field trials at Rosemaund and Aberystwyth. Letters indicate significant differences between mean values at p < 0.05.



**Figure 29** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on thousand grain weight. Mean ( $\pm$  s.e.m) results of 4 autumn sown field trials at Rosemaund and Aberystwyth. Letters indicate significant differences between mean values at p < 0.05.

The grain yield of a cereal crop like oats can be split into three major components: the panicle population density, the number of grains per panicle and the individual grain weight. Although there was no significant difference in the number of grains m<sup>-2</sup> between Buffalo and Tardis, they display contrasting tiller and panicle architectures. Tardis produces more panicles m<sup>-2</sup> and fewer grains per panicle than Buffalo (Figure 30, 31). The QTL introgression lines did not display significant

differences from their recurrent parent lines for these traits. The enhanced grain yield in Buffalo +A\_Mrg04 and in Tardis +B\_Mrg21 was as a result of a combination of a higher TGW as well as an increased grain number per panicle.



**Figure 30** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on panicles per m<sup>2</sup>. Mean ( $\pm$  s.e.m) results of 2 autumn sown field trials at Rosemaund. Letters indicate significant differences between mean values at p < 0.05.



**Figure 31** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on grain number per panicle. Mean ( $\pm$  s.e.m) results of 2 autumn sown field trials at Rosemaund. Letters indicate significant differences between mean values at p < 0.05.

#### 4.4.3. Grain quality



**Figure 32** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on screenings (% grain passing through a 2mm sieve). Mean ( $\pm$  s.e.m) results of 4 autumn sown field trials at Rosemaund and Aberystwyth. Letters indicate significant differences between mean values at p < 0.05.



**Figure 33** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on kernel content. Mean (± s.e.m) results of 4 autumn sown field trials at Rosemaund and Aberystwyth. Letters indicate significant differences between mean values at p < 0.05.

The percentage of the harvested grain passing through a 2mm sieve (screenings %) was highest in the Buffalo parent which was also significantly higher than the Tardis parent (Figure 32). The effect of the QTL on Mrg04 containing dw6 is clear with the Buffalo +A\_Mrg04 line having significantly lower screenings and the Tardis +B\_Mrg04 line having higher screenings than their respective parental lines. The lowest screenings were found in the Tardis +B\_Mrg21 QTL-NIL. These results mirrored those found for TGW (figure 29).

The low kernel content of Buffalo was significantly enhanced with both Mrg04 and Mrg21 introgressions from Tardis (Figure 33). Although the kernel content of Tardis was lowered by the introgression of the Mrg04 QTL from Buffalo, a decrease was not found for the Mrg21 introgression from Buffalo. The highest kernel content was obtained with the Mrg21 introgression from Tardis into Buffalo. The mean hullability of Buffalo across the 4 trials was higher than that of Tardis and the Mrg04 introgression from Tardis resulted in a significant lowering of hullability. Conversely the Mrg 21 introgression from Tardis resulted in higher hullability and the reciprocal was found in the introgressions from Buffalo into Tardis (Figure 34).

Although Buffalo had a lower specific weight than Tardis this was not significant (Figure 35). The Buffalo + A\_Mrg04 QTL\_NIL however had a significantly higher specific weight than the Buffalo parent and the converse was found with the Tardis +B\_Mrg04 QTL-NIL.



**Figure 34** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on Hullability. Mean ( $\pm$  s.e.m) results of 4 autumn sown field trials at Rosemaund and Aberystwyth. Letters indicate significant differences between mean values at p < 0.05.



**Figure 35** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on Specific Weight (kg/hl). Mean ( $\pm$  s.e.m) results of 4 autumn sown field trials at Rosemaund and Aberystwyth. Letters indicate significant differences between mean values at p < 0.05.

Grain length was not affected by the Mrg04, Mrg20 and Mrg21 introgressions with Tardis grain being 8 % longer than Buffalo irrespective of the presence of introgressed QTL (Figure 36). Grain width, however, was significantly different between parental lines and the Mrg04 introgressions (Figure 37). This increase in grain width suggests an increased capacity for grain filling as a result of the introgression of the Mrg04 Tardis alleles into Buffalo. The results in figure 37 mirror that for panicle emergence in figure 25 with an increase in grain width being associated with a decrease in panicle emergence and thus a longer duration of grain filling.



**Figure 36** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on mean grain length. Mean (± s.e.m) results of 4 autumn sown field trials at Rosemaund and Aberystwyth



**Figure 37** Effect of introgression of QTL on Mrg04 and Mrg21 into either Buffalo or Tardis on mean grain width. Mean (± s.e.m) results of 4 autumn sown field trials at Rosemaund and Aberystwyth.



**Figure 38** Relationship between mean plant height and thousand grain weight in Buffalo (green), Tardis (red) and 16 individual QTL-NIL (blue) grown at ADAS Rosemaund and in Aberystwyth in 2016-17.Tardis +B\_Mrg04, Tardis +B\_Mrg21 and Buffalo +A\_Mrg04 lines highlighted.

The Buffalo parent is not only shorter and later flowering than Tardis but also has lower grain yield, kernel content and thousand grain weight as well as higher screenings. This study has confirmed that alleles from Buffalo on Mrg21 confer earlier panicle emergence. These alleles are also associated with enhanced grain yield, higher thousand grain weight, higher kernel content, higher specific weight, higher hullability, and lower screenings. When introgressed into Tardis these result

in lines with higher thousand grain weight than would be predicted from the height of these lines (Figure 38). Conversely, the Buffalo alleles on Mrg04 when introgressed in Tardis result in lines that have greatly decreased height, grain yield, TGW, kernel content and specific weight and later panicle emergence. The results have provided new insights into the interaction of height and flowering time genes on both stem extension and heading date and on grain quality in oats. In the process new genetic markers have been identified and shared with breeders to facilitate more efficient selection of these regions along with understanding as to how traits are closely linked or pleiotropic to these QTL. Ultimately, identifying the genes and pathways underlying these QTL and how they interact will allow breeders to manipulate and fine-tune grain quality, yield and adaptation of oats to specific environments in innovative ways.

## 5. Discussion

Optimum nitrogen fertilisation levels are one of the main management tools in oats to enhance its competitiveness among other cereals. Oats due to its long stems, are considered prone to lodging, which might result in a loss in yield and grain quality particularly at high levels of N application (Chalmers *et al.*, 1998). It is crucial to obtain the maximum yield and grain and groat quality to maximise milling industry and farmer benefits, minimising at the same time lodging, the cost to the producers and environmental impacts. The strategy for breeders is to develop varieties with better yield and stability, avoiding lodging that may cause losses.

This project continued from AHDB project RD-2008-3556 (part of the Sustainable Arable Link project QUOATS, LK09124) in which two N response experiments were conducted in the 2013-14 season. As N response curves could not be fitted to the 2013-14 yield data because yields were still increasing at the highest N rates, despite a high SNS Index and a low RB209 (8th edition) recommendation, additional N response trials were completed in 2014-15 and 2016-17 with winter oats grown at six N rates, from 0 kg N/ha to 280 kg N/ha. The N response analysis has shown that for these trials the economic optimum N rate varied from 98 to 207 kg N/ha depending on variety and season. It was concluded in the recent review of RB209 (Roques *et al.*, 2016), that there was insufficient data to derive robust N recommendations for winter oats, although it was clear that the RB209 (8th edition) recommendations were too low. This knowledge gap is now being addressed in the AHDB and industry-funded project 'Optimising Nitrogen rates and timings and Sulphur in winter and spring oats for yield and milling quality' (NoatS; 21140039).

By sampling both biomass and grain and conducting tiller counts, it was possible to calculate Nitrogen Use Efficiency (NUE) and its components as well as identifying the response of individual yield components to the addition of N. The increase in yield with N application is driven by an increase in the number of grains m<sup>-2</sup>. An interaction was found between N rate and variety for both the number of panicles m<sup>-2</sup> and the number of grains panicle<sup>-1</sup> suggesting a differential response

between varieties in how grain number m<sup>-2</sup> is determined at higher N levels. Nitrogen Uptake Efficiency and NUE were affected by both variety and N treatment with Balado on average having the highest value for both in 2014. Gerald had the lowest Nitrogen Uptake Efficiency in both years. Overall, it was found that Mascani is better at capturing N from the soil and fertiliser than Gerald, but less efficient at partitioning that N to the grain.

The yields of both the 2014-15 and 2016-17 N response experiments were low, with the 2014-15 yields particularly so. The reason for the low yield in 2014-15 was a lot of wheat volunteers in the crop which were rogued out once the ears had emerged. These competed with the oats that were drilled, resulting in low yields, biomass, and low N uptake for the oats in this trial. The volunteers affected all treatments and so, once taken out, the results could still be analysed and compared. Despite the low yields, the economic optimum N rates for all varieties were above those currently recommended in RB209, at between 160 and 207 kg N/ha.

The yields of the 2016-17 N response experiment were not as low as those in 2014-15 but were affected by brackling and lodging because poor weather conditions delayed harvest. Having said that, the variety which was tallest and most affected by brackling, Griffin, gave a slightly (but not significantly) higher yield than Mascani and Maestro. In this season, the response of yield to N rate did not differ among varieties, with the economic optimum N rate fitted as 98 kg N/ha.

The varieties tested differed between the two N response experiments, with the four varieties in 2014-15 (Balado, Gerald, Mascani and Tardis) a continuation of varieties tested in the previous AHDB project QUOATS. The only variety in common across both seasons was Mascani, with newer varieties Griffin and Maestro replacing the other three from 2014-15. It could be seen that in 2014-15, Mascani was the best variety at taking up N from the soil and fertiliser but was less efficient at partitioning it to the grain. In 2016-17, Maestro was better than Mascani at taking up N from the soil and crop, with Griffin the best at partitioning the N taken up into the grain.

As would be expected, in both seasons, the biggest response of yield to N occurred between the nil and the first N fertiliser rate applied. In 2016-17 there was a more marked reduction in yield at the highest N rate (280 kg N/ha) than in 2014-15, probably due to the wet weather that delayed harvest (see appendix 1 for weather data). The grain yield of a cereal crop like oats can be split into three major components: the panicle population density, the number of grains per panicle and the individual grain weight. In all trials, although thousand grain weight was significantly affected by nitrogen it was not correlated with grain yield. Grain yield was highly correlated with the number of grains m<sup>-2</sup>. Grain number m<sup>-2</sup> is a combination of panicle number m<sup>-2</sup> and grain number per panicle, both of which increased significantly as nitrogen increased in all trials. These results suggest the productive tiller survival rate increases as greater levels of nitrogen are provided, as previously

reported (Weightman et al., 2004; Browne et al., 2006; Ma et al., 2017; Finnan et al., 2019). However, it was found in this study that the panicle number m<sup>-2</sup> did not increase beyond applications of 100 kg N/ha, and that the grain number per panicle continued to increase with higher applications of nitrogen. Oats display phenotypic plasticity in response to soil-climate conditions, but this is strongly influenced by variety. Although differences in total grain yield between oat varieties were only significant at the Rosemaund 2014-15 site, significant differences were found for the three yield component traits. There was, however, no significant interaction between variety and nitrogen treatment for these traits. Mascani was observed to have the lowest grain number per panicle and the highest panicle number m<sup>-2</sup> as well as the highest TGW under all treatments, whereas Balado had the highest grain number per panicle and lowest panicle number m<sup>-2</sup> in all three trials. Large increases in grain numbers with nitrogen fertilisation have been found previously (Peltonen-Sainio and Peltonen, 1995; Finnan et al., 2019). This may either be due to increased initiation or survival of spikelet primordia. When a greater number of grains are formed, competition can result in incomplete grain filling, reducing the final grain size (Marshall et al., 2013). The results suggest that whilst the different oat varieties can change yield component structure in response to changing nitrogen levels, the actual grain yield remains unchanged.

It is also essential that grain quality is maintained as well as enhanced yield. Grain quality analyses of the four N response trials conducted over 3 years (2013-14, 2014-15 and 2016-17) have been completed as part of project 2113004. The trials revealed variety dependent effects on grain quality, and it is suggested that these are related in part to the different response of yield components to the addition of nitrogen of the varieties used. Whereas grain yield was not associated with thousand grain weight, many of the grain quality traits measured were associated with thousand grain weight.

Specific weight was, in accordance with previously reported results (Givens, Davies, & Laverick, 2004; Ohm, 1976), significantly reduced with higher levels of nitrogen at all sites. An interaction between variety and N treatment for specific weight was found with Mascani not displaying a reduction in specific weight at higher levels of N fertilisation. For the other varieties, the specific weight at high levels of N application fell below that which is usually acceptable for the grain milling market (50kg/hL). At the ADAS Rosemaund 2013-14 and 2014-15 sites however a slight increase in specific weight values was observed at lower levels of nitrogen. This was not found at the Rosemaund 2016-17 and Lydbury North 2013-14 sites but these sites had a much higher spring soil nitrogen content. Specific weight is influenced by both grain density and packing efficiency components (Wychowaniec et al., 2013). Incomplete grain filling and therefore less dense grains, due to competition because of an increased shoot number have previously been linked to higher levels of nitrogen applied (Browne *et al.*, 2004; Chalmers *et al.*, 1998; Muurinen, Slafer, & Peltonen-Sainio, 2006) and this might explain lower specific weight values. Poor grain filling also affects

grain dimensions with the resulting grain being less round and not packing so efficiently. The timing of nitrogen applications in these trials followed standard RB209 recommendations but issues in poor specific weight at high nitrogen rates could potentially be resolved by optimising the timing of the N applications to optimise the shoot number. This is certainly an area where more study is required.

At the same time, increased levels of nitrogen resulted in lower thousand grain weight with higher values of grain length but lower values of grain width, i.e. longer and thinner grains, indicating poor grain filling. This decrease in grain roundness was associated with a reduction in specific weight. The specific weight of Mascani was not only high in all trials but was enhanced in the Rosemaund 2014-15 trial with increasing levels of nitrogen. Mascani has a high thousand grain weight which was relatively stable across the treatments in these trials. This was also evident when examining the bimodality in grain size with an increased proportion of small grain found with increasing application of nitrogen in the varieties Balado and Tardis but not in Mascani. This variety specific response to nitrogen must be considered when decisions are made about the level of nitrogen to use on farm. The finding that at high N application rates that specific weights are compromised could mean as a result that some varieties are below milling industry specifications when nitrogen is applied to maximise yield. With respect to the lowest available nitrogen treatments, specific weight was also reduced suggesting that there is an optimal level of nitrogen required for maximal hectolitre weight.

Higher levels of nitrogen had a positive effect on kernel content and hullability, with significant differences between varieties and levels of nitrogen and interaction between the two factors, for both traits. Maximal kernel content and hullability was obtained at the higher levels of nitrogen applied except at the 2016-17 Rosemaund trial where the effect of nitrogen on kernel content was not significant. Thus, nitrogen applications that favour grain yield also result in enhanced kernel content. Overall, the difference between varieties was greater than the difference between nitrogen treatments. A similar result was found by Yan *et al.* (2017). Kernel content did not show a strong correlation with specific weight which suggests that specific weight is not a good predictor of milling yield.

Although all varieties had maximal hullability at higher nitrogen application levels, Mascani, along with the newer varieties Griffin and Maestro, displayed very high values of hullability at all levels of applied nitrogen. Gerald, Balado and Tardis, however, had low hullability and kernel content at low nitrogen application rates. These hullabilities were calculated using the same sample as used for the kernel content determination. A high percentage of grain remaining unhulled indicated that the hull was difficult to remove and, therefore, has poor hullability. More work is required to determine how this relates to hullability within a commercial mill. Collaboration with the oat milling industry

has enabled a coordinated ring test of oat grain quality parameters. These results confirm the robustness of kernel content determination but indicate a greater level of variation in the determination of hullability. This suggests that the current method of hullability determination using a lab dehuller is not a suitable measure to be included in the AHDB Recommended List criteria as a numerical value, particularly as the actual methods used by each mill vary considerably. There was a consensus in the ranking of lines for hullability which could be an indicator of how a load would perform in the mill.

Groat protein and  $\beta$ -glucan content increased in response to nitrogen whereas oil content decreased. Mascani was revealed to have the lowest total oil content of the four oat varieties across all three trials and Tardis had the highest. Total oil content was negatively correlated with both total protein content and  $\beta$ -glucan content. Balado and Mascani had the highest  $\beta$ -glucan contents. Previous studies reporting the effects of nitrogen application on  $\beta$ -glucan content have given conflicting results (Saastamoinen *et al.*, 1992; Brunner and Freed, 1994; May *et al.*, 2004; Yan *et al.*, 2017). This is partly due to the different levels of nitrogen application used in these various studies and also suggests that variety is potentially having a great influence on this trait. Oil content was predominately determined by variety than by nitrogen application whereas protein content was predominately determined by N fertiliser rather than by variety. Differences obtained between trials also indicate that other environmental variables such as temperature and rainfall also impact on these traits

The weather cannot be controlled by oat growers. In contrast, oat growers do have the freedom to choose varieties and N fertiliser rates and the results obtained in this study provide valuable data to assist in this process. These results suggest that quality traits can be more easily improved through breeding, whereas yield can be more easily increased by use of N fertiliser. Therefore, as proposed by Yan *et al.*, 2017, when yield and quality are in negative genetic associations and cannot be simultaneously improved through breeding, it may be advisable to focus genetic improvement on the quality traits and to accept genotypes with a superior package of quality parameters at some expense of yield, and to depend on N fertiliser to achieve the required yield.

This will only be a successful strategy if the increased yield as a result of N fertiliser application does not compromise milling quality. In this study simultaneous and significant improvement of grain yield, milling quality (higher kernel content and hullability), and compositional quality (higher levels of  $\beta$  glucan and protein and lower oil content) were obtained. Specific weights of some varieties were reduced at high N fertiliser rates and this may need to be considered by growers depending on end user requirements. Due to the decrease in grain size at high N application rates, screenings (grains that pass through a 2mm sieve) may also increase.

The four nitrogen response field trials reported here have revealed robust changes associated with winter oat grain yield and quality, total protein, oil, and ß-glucan levels, with respect to increased levels of nitrogen supplementation and oat variety. Although nitrogen application significantly increased grain yield, protein and  $\beta$ -glucan content, some traits, such as specific weight,  $\beta$ -glucan, oil content, kernel content and hullability were affected by the variety used as much as by the addition of nitrogen. There were also significant variety by treatment interactions for several traits indicating variety specific responses to nitrogen. This emphasises the importance of variety choice as well as management regime strategies for specific cultivars. These findings suggest the complementary use of breeding and management in production of milling oats.

In the second part of the project, the effect of QTLs previously identified were verified by field testing QTL-NILs at Aberystwyth and ADAS Rosemaund confirming the interaction of height and flowering time genes on both stem extension and heading date and providing insight into their role in determining both grain yield and quality. Although the Buffalo parent is later flowering than Tardis, alleles from Buffalo on both Mrg20 and Mrg 21 confer earlier flowering. The highest yields (and largest grain size) at both sites were obtained when the Buffalo flowering time QTL on Mrg21 was introgressed into the conventional height cultivar Tardis. However, this resulted in low hullability despite enhanced kernel content and specific weight. Grain quality analyses revealed the association of the dw6 dwarfing gene on milling quality traits. Alleles from the Buffalo parent in the region of the genome containing dw6 were associated with much later flowering, lower grain yield, lower TGW, kernel content and specific weight as well as much shorter plants. Crosses of selected QTL-NILs with advanced winter oats have been successfully completed along with genotyping with the 6k single nucleotide polymorphism (SNP) chip.

The results from the N response trials have been disseminated to growers at ADAS farming association meetings and open days as well as annually at Cereals on the Just Oats stand. All the data from these trials have also been incorporated into the database of oats nutrition datasets as part of the AHDB project 'Optimising Nitrogen rates and timings and Sulphur in winter and spring oats for yield and milling quality' (NoatS; 21140039) and will used to update future versions of RB209. In addition to demonstrations at the Royal Welsh Show and Cereals this work has been publicised at a number of open days and meetings including invited papers at a range of conferences and meetings. Full details of outreach activities of the InnovOat project are provided in Appendix 2.

# 6. Take Home Messages

- Low inputs of N result in low grain yield and quality
- Higher levels of N fertilisation increase yield and had a variety-dependent positive response on all grain quality parameters, except specific weight and screenings. The variety Mascani displayed very stable grain quality across all nitrogen treatments.
- Economic N optima for grain yield range from 98–207 kg N/ha, depending on trial and variety
- Grain protein and β-glucan content increase in response to nitrogen, whereas oil content decreases
- Important that grain meets market requirements. For example:
  - At high N, some varieties may have specific weights and screenings below milling specifications
  - $\circ$  A lower  $\beta$ -glucan variety could reach acceptable levels with higher N application
- As varieties differed in the extent of response to N fertiliser, it is necessary to develop variety-specific N management plans. Plant breeding can complement the optimisation of N fertiliser to improve yield and grain quality
- The project has developed and characterised a series of reciprocal genetic stocks (QTL-NILs) that contrast at key chromosomal regions associated with traits related to adaptation, yield, and grain quality. These will help further refine and investigate the underlying genes controlling the targeted traits
- QTL for height, flowering time and grain size were validated. Interaction of QTL underlying yield determining traits were revealed along with impact on grain quality.
- Understanding of these QTL and how they interact will allow breeders to manipulate and fine-tune grain quality, yield and adaptation of oats to specific environments
- Dissection of grain quality traits reveals the importance of grain roundness and grain filling on grain quality traits

# 7. On-going and Future Work

The effect of vernalisation and photoperiod on flowering time QTL expression is currently being investigated under controlled conditions and using the phenomics facility at Aberystwyth University. Further backcrossing of QTL-NIL lines will reduce the interval surrounding the introgressed QTL, provide suitable germplasm for fine mapping, enable more accurate identification of genes underlying traits and of phenotypic effects. Candidate genes within refined QTL intervals can then be used to develop breeder friendly markers and to develop reliable predictions of milling quality based on genotype to allow selection in early generations of oat breeding programmes. Pyramiding these QTL will result in lines with an optimal ideotype. Such NILs provide a powerful resource to establish mechanisms behind QTL with controlled environments experiments looking at critical

growth periods. The ability to manipulate flowering time and plant height in lines with a common genetic background has many applications. For example, these QTL-NIL lines have been used in a pilot UK-Canada root phenotyping project (BB/S020926/1) which used precision root phenotyping technologies to identify diversity in important root system architecture traits in oats. An associated studentship partly funded by AHDB joint with Harper Adams University is currently using these QTL-NILs to develop understanding of the infection process of *Fusarium langsethiae* and of mycotoxin accumulation. QTL-NILs have also been developed in the winter oat variety Mascani and these could be used to dissect the components of plant and panicle architecture which contribute to its resilience of grain quality. The aim is to define an ideotype which may be selected in early generations of the breeding programme.

The nitrogen response experiments reported here showed improvements with increasing N rates of yield and most quality parameters apart from specific weight and, in some cases, screenings. This may have resulted from higher N rates leading to greater production of tillers and formation of grains, which could not be subsequently filled. When nitrogen is applied there is a large influence on the number of shoots produced and also grain filling. Research on the timing of nitrogen applications, as well as rates, is key to optimising fertilisation for maximising all aspects of quality of oats as well as the efficiency of the use of nitrogen. This knowledge gap is now partly being addressed in the AHDB and industry-funded project 'Optimising Nitrogen rates and timings and Sulphur in winter and spring oats for yield and milling quality (NoatS; 21140039). Breeding for varieties with enhanced nitrogen use efficiency rather than just through indirect selection for yield would have considerable benefits as well as understanding root architecture in the uptake and use of nitrogen.

# 8. References

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## 9. Acknowledgements

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# 10. Appendix 1 Weather data



Figure A1 Daily mean air temperature from day of sowing (01/10/14) until harvest (08/08/15) at ADAS Rosemaund in 2014-15



Figure A2 Daily rainfall at ADAS Rosemaund in 2014-15 from day of sowing (01/10/14) until harvest (08/08/15) Total rainfall in season 485mm



Figure A3 Daily minimum and maximum air temperature from day of sowing (02/10/15) until harvest (11/08/16) at ADAS Rosemaund in 2015-16.



Figure A4 Daily minimum and maximum air temperature at Aberystwyth in 2015-16 from day of sowing (28/09/15) until harvest (03/08/16)



Figure A5 Daily rainfall at ADAS Rosemaund in 2015-16 from day of sowing (02/10/15) until harvest (11/08/16) Total rainfall in season 538mm



Figure A6 Daily rainfall at Aberystwyth in 2015-16 from day of sowing (28/09/15) until harvest (03/08/16) Total rainfall in season 1290mm



Figure A7 Daily minimum and maximum air temperature from day of sowing (11/10/16) until harvest (22/08/17) at ADAS Rosemaund in 2016-17.



Figure A8 Daily minimum and maximum air temperature at Aberystwyth in 2016-17 from day of sowing (03/10/16) until harvest (10/08/17).


Figure A9 Daily rainfall from day of sowing (11/10/16) until harvest (22/8/17) at ADAS Rosemaund in 2016-17. Total rainfall in season 472mm



Figure A10 Daily minimum and maximum air temperature at Aberystwyth in 2016-17 from day of sowing (3/10/16) until harvest (10/8/17). Total rainfall in season 898mm

## 11. Appendix 2 Project dissemination and outreach activities.

Activity	Date: (mm/yy)	<b>Organiser</b> (and participants):
Radio interview on value of oats for human health on "Good Morning Wales"	2015	IBERS
InnovOat Project website set up http://www.innovoat.uk/	2015	IBERS
EIT Health Workshop - Oat Innovation (Cardiff)	2015	IBERS
Invited presentation at Genetics Society conference http://www.genetics.org.uk/Conferences/tabid/84/Filter/0%20gs/MeetingNo/gts _sm2015/MeetingID/88/ClientID/1/view/Programme/Default.aspx	Apr-15	IBERS
Participation in Cereals 2015 "Just Oats – Breeding in the 21 <sup>st</sup> Century", Boothby Graffoe, Lincs	Jun-15	IBERS/ BOBMA/ Senova
NIAB-TAG Sutton Scotney Open Day https://www.niab.com/shop/civicrm/event/info?reset=1&id=1840	Jun-15	NIAB
ADAS Rosemaund Open Day	Jun-15	ADAS
Milan EXPO Agri-Tech Week – various roundtables	Jul-15	IBERS
Cereals in Practice (organised by JHI)	Jul-15	JHI/ IBERS
Attendance at launch of NutriWales business cluster	Oct-15	IBERS
Canada Partnering Award Workshop, Aberystwyth	Nov-15	IBERS
Oats2020 presentations	Nov-15	IBERS
Oats2020 presentations	Nov-15	ADAS
Presentation to AHDB monitoring meeting	Dec-15	IBERS
Boyle, R. Corke, F and Howarth, C.J. (2015) Oat panicle detection using generalised LTPs. Functional Plant Biology 42: 433-443	2015	IBERS
Montilla-Bascón, G, Rispail, N, Sánchez-Martín, J, Rubiales, D, Mur, LAJ, Langdon, T, Howarth, CJ, Prats, E (2015) Genome-wide association study for crown rust and powdery mildew resistance in an oat collection of commercial varieties and landraces. Frontiers in Plant Science doi. 10.3389/fpls.2015.00103	2015	IBERS
Ward J, Rakszegi M, Bedő Z, Shewry PR, Mackay I. (2015). Differentially penalized regression to predict agronomic traits from metabolites and markers in wheat. BMC genetics, pp. 19	2015	NIAB
Jones H, Mackay I. (2015). Implications of using genomic prediction within a high-density SNP dataset to predict DUS traits in barley. TAG. Theoretical and applied genetics.128 (12), pp. 2461-70.	2015	NIAB
New varieties launched		
New winter oat variety <b>Maestro</b> added to RL 2015		
New winter oat variety <b>Coracle</b> added to NL 2015		
New winter oat variety <b>Coracle</b> added to NL 2015		
New spring oat variety <b>Spurtle</b> added to NL 2015		
	2016	
Invited speaker at Plant and Animal Genome XXIV, San Diego	Jan-16	IBERS
Westminster Food & Nutrition Forum: 'The role of science in food and agriculture - novel foods, sustainable intensification and challenges for policy.' During discussion, the (potential and actual impact of oats in the diet was reinforced and the growth in the associated breakfast products over the last several years was identified. Oats as a model for other crops was discussed	Jan-16	James Hutton Institute
Presentation to Genetic Improvement Network stakeholder meeting, Norwich	Feb-16	IBERS
Presentation to Rosemaund Farming Association meeting	Feb-16	ADAS

Invited speaker "Global warming and the effects on our rural estates - the farm" at Northern Briefing 2016. Oats were featured in the talk building on the QUOATS work http://www.caav.org.uk/Events/Conferences/Northern_Briefing_2016.aspx	Mar-16	JHI
Discussion were held and followed up with multiple partners to develop and then submit an application to the Newton UK-China Bridges call (funded by Innovate UK). Proposal submitted on time and entitled "Enhancing the application of Chinese soybean by developing soy protein / beta glucan conjugates as sustainable meat and fat replacers for the Chinese sausage industry (SoyGluSaus)".	Mar-16	JHI
Keynote speaker at Westminster Food & Nutrition Forum - Priorities for the UK food and drink industry - implementing the 25 year food and farming plan.	Mar-16	JHI
Discussion were held with the local councils regarding the Tayside Cities deal. As part of the to-be bid-for package to stimulate the local economy and environment, the benefits so JHI Cereals research was highlighted as the potential fora international Barley Hub, although cereals would be a broad target including oats. The regions target also includes North east fife where Quaker oats are situated.	Mar-16	JHI
Presentation to Monogram meeting	Apr-16	IBERS
Invited speaker at XXXVI Brazil Annual Oat Research Meeting, Pelotas	Apr-16	IBERS
Publication: Population genomics related to adaptation in elite oat germplasm. The Plant Genome doi: 10.3835/plantgenome2015.10.0103	May-16	IBERS
Cereals Event 2016, Included Just Oats, BBSRC and AHDB stands. Large project display and oats discussion forum/variety plot 'tour with industry representatives' on AHDB stand. ADAS and IBERS also representing project.	Jun-16	IBERS/AHD B/ADAS/ BOBMA/Sen ova
NIAB-TAG Morley Open day	Jun-16	NIAB
NIAB-TAG Sutton Scotney Open Day	Jun-16	NIAB
Royal Welsh Show, Builth Wells	Jul-16	IBERS
International Oat Conference, St Petersburg		IBERS
•Paper: An oat nested associated mapping population (IBERS)		
•Paper: Developing enhanced breeding methodologies for oats (IBERS)		
•Paper: The use of disease resistance alleles in the UK oat breeding programs (IBERS)	Jul-16	
•Paper: Oats the first 32,000 years and the next ten (Chris Green/ Green Resources)		
•Poster: Effects of environment on yield and grain quality parameters of winter oats in different growing seasons in the UK		
•Poster: Effect of grain development on quality parameters in oats		
cereals in Practice (organised by JHI) http://www.hutton.ac.uk/events/cereals- practice-2016	Jul-16	IBERS & JHI
Discussions with Wynnstay group	Jul-16	IBERS
Publication: A consensus map in cultivated hexaploid oat reveals conserved grass synteny with substantial sub-genome rearrangement. Accepted by The Plant Genome doi: 10.3835/plantgenome2015.10.0102	Jul-16	IBERS
Invited speaker (Alison Bentley) at the 1st International Symposium on Genomic Selection for Crop Breeding, Rabat, Morocco, http://www.cropgs.com/international-symposium-on-genomic-selection-for- crop-breeding/	Sep-16	NIAB
Participation in Food Matters Live 2016 by Prof D Stewart		
	Nov 2016	JHI

Publication: High-density marker profiling confirms ancestral genomes of Avena species and identifies D-genome chromosomes of hexaploid oat Theor Appl Genet (2016). doi:10.1007/s00122-016-2762-7 online first	Nov-16	IBERS
Discussion with Frontier re grain quality	Dec-16	IBERS
Meeting - Portuguese FASTBREED Project	2016	IBERS
Bentley AR, Mackay IJ. (2016). Advances in wheat breeding techniques. Achieving sustainable cultivation of wheat. Cambridge, UK:Burleigh Dodds Science Publishing	2016	NIAB
Chaffin A, Huang Y, Smith S, Bekele W, Babiker E, Gnanesh BTinker N. (2016). A Consensus Map in Cultivated Hexaploid Oat Reveals Conserved Grass Synteny with Substantial Subgenome Rearrangement. The Plant Genome, 9 (2)	2016	IBERS
Esvelt Klos K, Huang Y, Bekele W, Obert D, Babiker E, Beattie A, Tinker N. (2016). Population Genomics Related to Adaptation in Elite Oat Germplasm. The Plant Genome, 9 (2),	2016	IBERS
Seminars on genomic selection	Ongoing	NIAB
Project data used in review of RB209	2016	ADAS/AHDB
Regional pitch to the BioBased Industry Consortium by Prof D Stewart	2016	JHI
New varieties launched		
Winter oats Griffin and Peloton (naked oat) added to AHDB RL list	Dec-16	IBERS
	2017	
Senova stand at AICC (Association of Independent Crop Consultants) annual conference	Jan-17	Senova
Provided info to Ricochet TV about oats for "Superfood" programme	Jan-17	IBERS (SC)
Publication: The agronomic performance and nutritional content of oat and barley varieties grown in a northern maritime environment depends on variety and growing conditions Article reference: YJCRS2268 Journal title: Journal of Cereal Science Corresponding author: Dr. Karen P. Scott First author: Dr. Andrew Chappell DOI information: 10.1016/j.jcs.2017.01.005	Jan-17	JHI/IBERS
Attendance and participation at International Plant & Animal Genome Conference XXV, San Diego http://www.intlpag.org/ Speaker, Oats Workshop: "Oats - Contacting the Uncontacted Genome" Speaker, Introgression Workshop: "Introgressions in an Oat Nested Association Mapping Population" Poster: "Fine Mapping of the Dw7 Dwarfing Gene in Oat" Poster: "Identifying Avena-specific genes"	14th- 18th Jan 2017	IBERS
Named authors on presentation given by Elena Prats, CSIC, Cordoba, Spain, at oats workshop during PAG XXV https://app.core-apps.com/pag- 2017/abstract/1a72fbe5da697beb0993e254b2ca5369	14th Jan 2017	CSIC/IBERS
Participation in the "Taste of Wales" Event - Celtic Manor Hotel (event linking academia, industry, with the food supply chain)	23rd March 2017	IBERS
Participation and poster at the MonoGram event in Bristol http://www.monogram.ac.uk/MgNW2017.php	4th-6th April 2017	IBERS
Oat production chain and rural development in Central Asia, workshop, Baku	2017	IBERS
Plant Breeding Workshop, IBERS (oat delegates from Morocco, Kazakhstan)	2017	IBERS
New edition of RB209	May-17	AHDB/ADAS
Cereals event 2017 http://www.cerealsevent.co.uk/ Included Just Oats and AHDB stands	13th- 15th June	IBERS/AHD B/ADAS/ BOBMA/ Senova
ARGRAIN Open Day	Jun-17	IBERS

NIAB Croft Open Day, Darlington - project demo and role/uses of oats	29th June	IBERS/NIAB/ Senova
Visit by Sandy Cowan to Hogarth Mills	June	IBERS (SC)
Participation by Greg Mellers (NIAB) in Genetic Analysis course organised by WHEALBI (WUR)	June	NIAB (GM)
Canadian Partnering Award, BBSRC - trip by IBERS team to Canadian oat breeders. Developing links and discussing strategies and implementation of molecular techniques into breeding programmes.	7th-13th July	IBERS (SC/IG)
Cereals in Practice 2017	Jul-17	IBERS/ JHI
Attendance at Royal Welsh Show 2017	Jul-17	IBERS
PlantGen 2017, Almaty – Conference presentation, national botanic garden meeting	2017	IBERS (TL)
BEWS 2017, Antalya – Conference presentation, meeting with Turkish genebank management	2017	IBERS (TL)
Nicosia – Meeting with ECPGR Avena Chair, collection trip	2017	IBERS (TL)
Gatersleben – Exome capture & genetic resources workshop, meeting/seminar at JKI	2017	IBERS (TL)
Lund – Meeting with Crop Tailor, ScanOat (genomics, dihaploid)	2017	IBERS (TL)
Tbilisi (twice), Yerevan – Meetings and collection trips, Georgian and Armenian national botanic gardens	2017	IBERS (TL)
Warsaw – Meeting with U. Lublin (domestication, rust and NAM field trials)	2017	IBERS (TL)
Rome – Meeting with ENEA (chromosome isolation and sequencing	2017	IBERS (TL)
Hickey, J.M., Chiurugwi, T., <u>Mackay, I.</u> and Powell, W., 2017. Genomic prediction unifies animal and plant breeding programs to form platforms for biological discovery. <i>Nature Genetics</i> , <i>49</i> (9), p.1297. DOI: 10.1038/ng.3920	Aug-17	NIAB
JHI (WA) attendance and poster presentation of the oats nitrogen trial work at the Scottish Metabolomics Network Annual Meeting: http://scottishmetabolomics.net/events/	2nd-3rd Nov	JHI (WA)
IBERS (CH) presentation at Global Oat Rust Forum, University of Cornell	Nov-17	IBERS (CH)
IBERS (CH) presentation at AAFC-AgriFood Canada strategic oat planning forum, University of Cornell	Nov-17	IBERS (CH)
Visit to IBERS from Chris Price-Jones, NutriWales, to discuss possible uses of oats in human health	2017	IBERS
New varieties launched		
New winter oat variety Galloway added to NL 2017		
New winter oat variety <b>Penrose</b> added to NL 2017		
New winter oat variety <b>Coracle</b> added to RL 2017		
New spring oat variety 15056CnI added to NL 2017		
	2018	
Publication accepted in Plant Biotechnology Journal: Haplotype based genotyping-by-sequencing in oat genome research DOI: 10.1111/pbi.1288	18th Jan	IBERS (CH)
Attendance at TEAGASC Tillage Conference, Kilkenny	31st Jan	IBERS
Presentation at Conservation Agriculture meeting, JIC, Norwich	January	IBERS (CH)
Presentation at ADAS High Mowthorpe Farming association meeting	March	ADAS (SC)
MonoGram Meeting - Norwich	April 24th- 26th	IBERS/JHI/N IAB
Oat genomics workshop held, Birmingham, attended by Canadian, US, German, JIC colleagues - developed BBSRC RM application submitted 04/18	April	IBERS (TL)
Plant Disease publication accepted; Characteristics of resistance to <i>Puccinia coronata</i> f. sp. <i>avenae</i> in <i>Avena fatua</i> L.	May	IBERS (TL)

	12th- 14th	IBERS/ Senova/
Cereals 2018	June	BOBMA
visit by Graze to IBERS options on oats	21/6/18	IBERS
Croft open day NIAB TAG Darlington	28/6/18	IBERS
Presentation at American Oat Workers Conference	June	IBERS (CH)
Discussions at Marshfield Bakery on potential of oats: follow up from Graze visit	3rd July	IBERS
	24-26	
Royal Welsh Show, Builth Wells	July	IBERS
Research visit by Dr Bisaga to Morden and Saskatoon to extend disease resistance collaboration (OECD fellowship application submitted 9/18) (BBSRC funded)	July	IBERS (MB/TL)
Plant Disease publication accepted; Diversity within <i>Avena sterilis</i> L. populations provides new sources of resistance to crown rust	August	IBERS (TL)
New Phytologist publication accepted; Towards take-all control: a C-21β oxidase required for acylation of triterpene defense compounds in oat	August	IBERS (TL)
Poster presented at the Scottish Metabolomics Network, Glasgow City College: Application of metabolomics to study the effect of nitrogen elevation on winter oat metabolite composition and quality traits: J. William Allwood, Yun Xu, Raphaelle Palau, Catherine Howarth, Athole Marshall, Roy Goodacre, Derek Stewart	Nov	JHI/IBERS
Radio interview for Radio Wales Science Café programme		IBERS (CH)
AHDB monitor farm visit, Pembs	June	IBERS (SC/IG)
Stakeholder discussions on milling oats		IBERS (CH/IG/SC)
Discussions with ORC and Gaia Foundation on developing new project		IBERS (CH)
Presentation to BOBMA AGM	May	IBERS (SC)
Presentation to AHDB	March	IBERS (CH)
Participation in Grain-genes Liaison Committee meeting	May	IBERS (CH)
Supply chain discussions, Northumberland.	30th March	IBERS (SC)
Presentation at Food & Drink Wales - Beer and Cider Special Interest Group meeting	1st Nov	IBERS (SC)
Successful Grant application: Optimising Nitrogen rates and timings and Sulphur in winter and spring oats for yield and milling quality (NoatS) 1/08/2018 - 31/05/2022 This includes collaboration with ADAS, Teagasc (Ireland), Seges (Denmark), PepsiCo, Richardsons Milling, BOBMA, Saaten Union, Senova, KWS, RAGT Seeds, Camgrain, Gowlett Grain, Frontier, CF Fertilisers, Omex, Chadacre Agricultural Trust, Felix Thornley Cobbold Agricultural Trust	1/8/18	ADAS/ IBERS
Bekele, W.A., Wight, C.P., Chao, S., Howarth, C.J., Tinker, N.A. (2018) Haplotype based genotyping-by- sequencing in oat genome research Plant Biotechnology Journal 16: 1452-1463 DOI: 10.1111/pbi.1288	2018	IBERS
	2019	
Presentation at Food & Drink Wales - Future Foods	14th Jan	IBERS (SC/CH/DL)
Presentation on oats at "Cows on Tour" IBERS	7th Feb	IBERS (SC)
Presentation to AHDB	January	IBERS (CH)
Participation in mycotoxin meeting at Harper Adams	Februar y	IBERS (CH)
Participation in crop diversity meeting Brussels	27/03/2 019	IBERS (CH)

Grant application submitted: Understanding the genetic control of milling quality in oats Howarth, Langdon, Cowan, Griffiths	30/04/2 019	IBERS
Grant awarded: UKRI-NRC Prototyping Root System Architecture in Avena: Technologies for Environmental Sustainability and Food Security (£133k) Howarth, Langdon, Doonan 1/3/19 - 31/10/19	01/12/2 018	IBERS
Grant awarded: BBSRC Responsive Mode: Origin of a European cereal (£540k) Tim Langdon 17/12/18 - 16/6/21	01/12/2 018	IBERS
IBERS Centenary of Plant Breeding	02/07/2 019	IBERS
IBERS BioBank seed store launch activities	25/10/2 019	IBERS
Visit to potential industrial partner (Chuckling Goat) to discuss oat milk products	14/05/2 019	IBERS
Cereals Event 2019, Boothby Graffoe, Lincoln	12th- 13th June	SENOVA/ IBERS/ AHDB/ BOBMA
Presentation at BIC Innovation event, Anglesey on green protein and oat products	16th July	IBERS
Oat Field tour and demonstration, Farming Connect	01/08/2 019	IBERS
Scottish Metabolomics Network, Discover Point, Dundee	Nov	JHI
Publication in Springer Metabolomics journal. "Rapid UHPLC-MS metabolite profiling and phenotypic assays reveal genotypic impacts of nitrogen supplementation in oats" Doi:10.1007/s11306-019-1501-x J. William Allwood, Yun Xu, Pilar Martinez-Martin, Raphaëlle Palau, Alexander Cowan, Royston Goodacre, Athole Marshall, Derek Stewart, Catherine	January	JHI/ IBERS
Howarth		
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