1	<b>Title:</b> Food and bioenergy: exploring ideotype traits of a dual-purpose wheat cultivar
2	Running title: Assessing wheat cultivars for use for fuel and food
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10	Abstract

11 Lignocellulosic biofuels, such as those produced from crop residues, offer the 12 possibility of reducing GHG emissions in the transport sector. Wheat straw is one 13 potential feedstock for these fuels but grain yield has been prioritised over straw 14 yield in crop breeding as straw currently has limited value. Should a new market for 15 straw develop then dual-purpose cultivars (DPCs) that are optimised for food and 16 bioenergy may become desirable. Field experiments were used to assess four key 17 traits - grain yield, straw yield, lodging resistance and straw saccharification potential (i.e. the biofuel yield) - of a selection of modern and older UK wheat 18 19 cultivars (with release dates ranging from 1964 to 2010) with the aim of identifying dual-purpose cultivars and any trade-offs among the key traits. None of the cultivars 20 21 assessed were outstanding candidates for use as DPCs. Among the semi-dwarf 22 cultivars there were only minor relationships between traits; including the non-semi-23 dwarf cultivar showed trade-offs between grain and straw yields. The findings 24 suggest that selecting from among currently grown cultivars offers limited possibility 25 for growing truly DPCs. However, the results indicate that high straw yields can be 26 achieved by selecting high grain-yielding cultivars and managing these to maximise 27 grain yield will also result in high straw yields. Plant growth regulators should continue to be used as these do not significantly reduce straw yields but do decrease 28 29 lodging susceptibility.

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31 Keywords: Lignocellulosic Biofuel; Wheat (*Triticum aestivum*); Dual-Purpose
32 Cultivars; Agronomy; Crop Physiology

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Abbreviations: %GR (% glucose release); dual-purpose cultivars (DPC); field experiments 1-3 (FE1-3); fusarium head blight (FHB); growth stage (GS); harvest index (HI); nitrogen (N); nitrogen treatments 1-3 (N1-3); plant growth regulators (PGRs); root failure wind speed (RFWS); stem failure wind speed (SFWS); straw sections 1-4 (S1-4).

#### 39 **1. Introduction**

40 Second-generation biofuels, such as fuels produced from lignocellulosic material, 41 offer the possibility of reducing greenhouse gas emissions and ensuring energy 42 security (Demirbas, 2008). European Union legislation is mandating the use of biofuels in the transport sector; by 2020, 10% of transport fuel must be from 43 44 renewable sources, the majority of which is expected to be by biofuels (EU, 2009). 45 Currently the majority of biofuels are first generation liquid biofuels, produced from 46 starchy grains (such as wheat and maize); the use of these has been controversial due 47 to the potential land use competition between food and fuel production and 48 expansion of agricultural land (Sims et al., 2010). This has led to interest in biofuels 49 produced from lignocellulosic materials (e.g. crop and forestry residues, industrial 50 waste, and dedicated energy crops such as Miscanthus); as these are considered to be waste products or able to be grown on marginal land, competition with food 51 52 production is limited. The technology has developed to the point that commercial production of bioethanol from lignocellulosic material started in 2013 at Beta 53 54 Renewables' Crescentino plant in Italy and in 2014 at GranBio's Bioflex 1 plant in 55 Brazil and the POET facility in Emmetsburg, US.

Wheat straw is the biggest resource available for SGB production in the UK (Copeland & Turley, 2008); however, there are limitations in supply due to competition with other straw uses (Glithero et al., 2013b), some farmers' unwillingness to supply straw (Glithero et al., 2013a) and, as transporting lignocellulosic material is expensive (Miao et al., 2012), supply is limited to the local area. Dedicated energy crops could potentially be used but these would have to be grown on marginal land to limit competition with food production. As there is only limited marginal land available and farmers are often unwilling to grow these
crops (Glithero et al., 2015), their contribution is likely to be minor. This could mean
difficulties in matching a high enough density of supply for a biorefinery. Feasibility
of sufficient feedstock supply might, therefore, depend on increasing residue yields.
If this is possible without compromising grain yields then farmers might be
interested in growing cultivars with higher straw yields to increase the value of their
crops.

Dual-purpose cultivars (DPCs) offer the possibility of increasing the resources available for lignocellulosic biofuel production without compromising existing food production. The concept of a wheat DPC food and bioenergy ideotype is reviewed in Townsend et al. (2015); in that review four key traits are highlighted: grain yield, straw yield, straw saccharification potential (i.e. the amount of sugars made accessible during enzymatic hydrolysis for conversion to biofuel) and lodging resistance.

77 Improving all traits simultaneously might not be possible due to trade-offs among the 78 traits. The reason that lodging resistance is included in the traits of a DPC is because 79 of potential trade-offs with saccharification potential and straw yields. High straw yields could also lead to greater risk of lodging due to the relationship between plant 80 81 height and straw yield (Larsen et al., 2012) and the correlation between plant height 82 and lodging risk (Berry et al., 2000). For straw saccharification potential it has been 83 hypothesised that breeding for increased lodging resistance has lowered straw 84 saccharification potential through changing the stem material characteristics (Travis et al., 1996). It is also possible that this could increase the risk of disease 85 86 susceptibility making the plant more susceptible to pathogens and pests (Li et al.,

87 2008). Alongside these trade-offs is the possibility that high straw yields could come
88 at the expense of grain yield due to competition for a limited quantity of assimilates
89 (Austin et al., 1980).

As discussed in Townsend et al. (2015), other management practices might also influence these key traits and, therefore, could be used to maximise these traits. In particular, plant growth regulator (PGR) application, nitrogen (N) fertiliser rate and combine harvester cutter height could have impacts on straw yields and other characteristics. These management practices could potentially lead to trade-offs by improving some traits at the expense of others.

96 Variation among cultivars exists for all four traits identified but as straw yield and, in 97 particular, saccharification potential, are only very rarely quantified there is limited 98 data on these traits in modern cultivars. Larsen et al. (2012), exploring a similar 99 concept, assessed the biofuel potential of multiple wheat cultivars, measuring grain 100 and straw yields, and straw saccharification potential.

101 The aim of this paper is to quantify straw and grain yields, lodging susceptibility and 102 straw saccharification potential in a number of UK winter wheat cultivars 103 (introduced between 1968 and 2010) with the purpose of identifying potential 104 candidates for use as dual-purpose food and bioenergy cultivars. Field experiments 105 over three years were conducted to measure the traits in these cultivars and to 106 determine whether trade-offs exist among these desirable traits.

#### 107 **2. Materials and Methods**

#### 108 **2.1 Field experiments**

109 Three field experiments were conducted at the University of Nottingham's farm at 110 Sutton Bonington (52°50'N, 1°15'W) between 2009 and 2012. Key details of these 111 experiments are given in Table 1. Multiple winter wheat cultivars were assessed for 112 their traits for use as DPCs (see **Table 2** for a list of the cultivars assessed); these 113 cultivars were selected to provide a wide range of material in terms of date of 114 introduction, height, lodging resistance and grain end-use. These included a non-115 semi-dwarf cultivar (Maris Widgeon) while the rest were semi-dwarf cultivars. The 116 soil type was a stony sandy loam soil (Dunington Heath series). Cultivars were 117 grown in plots of size 24 x 1.6 m. Crop protection chemicals were used 118 prophylactically to minimise weeds, pests and diseases in all years. Soil fertility 119 levels were amended to ensure nutrient availability would not be limiting. Rainfall 120 and average temperature for the growing seasons are shown in Fig. 1a, b.

Field experiment 1 (FE1) took place in the 2009-2010 growing season. It consisted of four completely randomised blocks of 40 winter wheat cultivars, with a subsection of 14 cultivars used in the analysis. PGRs were not applied in order to allow full trait expression. Each block was separated by a discard plot.

Field experiment 2 (FE2) took place in the 2010-2011 growing season. An additional cultivar, Glasgow, was added to the 14 cultivars assessed in FE1. The cultivars were grown in a split-plot design with the plant growth regulator Chlormequat (PGR+) and without (PGR-) as the main plots. The experiment was organised in three blocks

with the cultivars randomly distributed within the main plots and the blocksseparated by discard plots.

131 Field experiment 3 (FE3) took place in the 2011-2012 growing season. Only three 132 cultivars were considered to allow a focus on management practices. The PGR 133 treatments matched those of the previous field experiment. The N treatments were 134 based on the N requirements of the field (based on RB209, 2010); all plots were 135 given the first two splits of 40 kg ha<sup>-1</sup> and 80 kg ha<sup>-1</sup> with a final split of 0 kg ha<sup>-1</sup> 136 (N1), 50 kg ha<sup>-1</sup> (N2) or 100 kg ha<sup>-1</sup> (N3; fertiliser application timing is given in 137 Table 1). The N2 treatment matched the recommended N application rate for the 138 particular field, rotation and crop conditions. The three cultivars assessed – Cordiale, 139 Grafton and Xi19 – were from the previous year's experiment and were selected 140 based on current use (all three were in AHDB's 2011 Recommended List for wheat 141 cultivars), high grain yields and differing characteristics in terms of height and 142 lodging susceptibility. A split-split plot design was used whereby each block was 143 divided into three main plots (N treatment) that were each subdivided into two sub-144 plots (PGR treatment). Cultivars were then randomised within the sub-plots.

## 145 **2.2 Biomass assessments**

Stem number was counted prior to harvest in FE1 and FE2 while in FE3 plant number and average tiller number were counted at GS61 (based on the decimal system described in Zadoks et al., 1974). When the crops were fully mature, a 0.5 x 0.5 m area was randomly sampled from each plot and cut at soil level. Plant height was measured to the tip of the ear. The above ground dry matter (AGDM) of the sample was determined by drying to constant mass, and then the ears were separated 152 from the stems and threshed so that straw, grain and chaff weight could be 153 determined. In FE3, the stem was split into sections to investigate the effect of 154 combine harvester cutter height on straw yield; firstly, the leaf blades were removed 155 then the stem was split into the first 10 cm from ground level (S1), 10-15 cm from ground level (S2), 15-20 cm from ground level (S3) and the remainder (S4). These 156 157 simulated combine cutter heights of 10 cm, 15 cm and 20 cm and allowed 158 determination of the amount of harvestable straw (i.e. the straw amount excluding 159 the stubble and friable material that would be left on the field during straw 160 harvesting).

## 161 **2.3 Saccharification potential assessments**

Sugar composition of the straw was assessed for a subset of cultivars from FE1 and FE2. These assessments followed the methodology given in Ibbett et al. (2011). Whole plant samples from ground level to the top of the peduncle were milled to obtain homogenous samples with particle sizes of 200-700 microns. The milled straw (30 mg) was subjected to a Saeman acid hydrolysis and the glucose and xylose content of the resultant hydrolysate were measured using high-performance anion exchange chromatography with pulsed amperoetric detection (HPAEC-PAD).

Milled samples (1 g) were then subjected to a standardised dilute acid pre-treatment (10% w/v, 1% H<sub>2</sub>SO<sub>4</sub>, 121°C, 15 m), the residue was washed in distilled water then dried and subjected to a Saeman hydrolysis before HPAEC-PAD was used to quantify the glucose present. Dried residue (200 mg) was then incubated with cellulase cocktail (Cellitec2) and saccharification potential assessed as the amount of monomeric sugars released during hydrolysis for 72 h, which were quantified usingHPAEC-PAD.

Saccharification potential is given as the glucose yield released during enzymatic hydrolysis per unit of pre-treated straw (expressed as mg of glucose released per g pre-treated straw). Percentage glucose release (%GR) is expressed as the percentage of total available glucose in the pre-treated residue released during enzyme hydrolysis.

### 181 **2.4 Lodging assessments**

182 To determine lodging resistance for the different treatments, the lodging model 183 outlined in Baker et al. (1998) and Berry et al. (2003) was used to determine the 184 failure wind speed (i.e. the minimum wind speed at which lodging will occur). The 185 model uses measurements of plant characteristics to allow calculation of the material 186 strength of the lower internodes, the strength of the root-soil interface and the stem 187 leverage force (Berry et al., 2000). When the stem leverage force exceeds the 188 material strength of the lower internodes, the stem bends or breaks leading to stem 189 lodging; the wind speed at which stem lodging occurs is called stem failure wind 190 speed (SFWS). When the plant leverage force exceeds the anchorage strength the roots are displaced within the soil. The wind speed at which root lodging occurs is 191 192 called root failure wind speed (RFWS).

Ten plants were carefully removed from each plot on 1 July 2010 (FE1), 28 June 2011 (FE2) and 17 July 2012 (FE3) when the plants were at approximately GS75. The plants were placed in polythene bags and stored at 4°C for no more than 2 weeks until laboratory analysis. In the laboratory, eight of these plants were taken for 197 measurement of lodging traits, as detailed in Berry et al. (2000). Structural rooting 198 depth and average root plate spread were measured to calculate the strength of the 199 root-soil interface. Stem leverage force was calculated by isolating the main stem and 200 measuring natural frequency, number of ears per plant, ear area and height at centre 201 of gravity. Stem material strength for internodes one and two were determined by 202 measuring internode length, diameter, wall width and breaking strength. The equations for calculating stem material strength and stem leverage force, as well as 203 204 SFWS and RFWS are given in Berry et al. (2003). For each of the eight plants the 205 SFWS and RFWS and these eight failure wind speeds were averaged to give a SFWS 206 and RFWS for each plot.

## 207 **2.5 Statistical analysis**

208 Analysis of variance (ANOVA) with blocking procedures appropriate for the experimental design was carried out using GenStat for Windows, 15th Edition (VSN 209 210 International Ltd.). Data was checked to see if it met the assumption of constant 211 variance and normal distribution of residuals. All yield data was converted into 212 tonnes per hectare and 15% moisture content to enable comparison to standard 213 cultivar trial data. ANCOVA was used to investigate the relationships between traits. 214 For comparison across years, all treatments not common to the three field 215 experiments were excluded.

#### 216 **3. Results**

The weather varied greatly between the years. In FE2 there was low rainfall throughout the growing season, but especially in spring, and a colder than average December. In FE3 rainfall was low at the start of 2012, but other than May, monthly rainfall was far higher than average; because of the high rainfall during June and July, the plants developed considerable fungal disease. High levels of fusarium head blight (FHB) were observed as well as other fungal diseases.

## 223 **3.1 Biomass production and partitioning**

The cultivar Battalion had a greater number of stems compared to the other cultivars (P = 0.002). In FE2, stem number did not significantly differ with cultivar or PGR. In FE3, stem number was assessed at GS61 and Xi19 had significantly fewer stems than Grafton or Cordiale (P < 0.001) but neither PGR nor N affected stem number.

In FE1 and FE2 there were no significant differences in AGDM (**Tables 3** and **4**, respectively). Although there was a large range of mean AGDMs, the high variation among replicates meant significant differences were not detected. In FE2, PGR application did not significantly affect AGDM. In FE3, Xi19 had significantly lower AGDM than Grafton and Cordiale (P = 0.017) but neither PGR nor N had an influence (**Table 5**). AGDM was significantly higher in 2012 than 2010, with 2011 having an intermediate AGDM (P = 0.035).

In FE1, there were significant differences in grain yield between the highest yielding cultivar, Cordiale, and the lowest yielding cultivar, Maris Widgeon (P = 0.048) but not among the other cultivars (**Table 3**). In FE2, there was a significant difference in 238 grain yield (P < 0.001) with Maris Widgeon having a significantly lower grain yield 239 than the majority of the other cultivars (Table 4). PGR application had no effect on 240 grain yields. In FE3, grain yield for Xi19 was significantly lower than that of Grafton 241 and Cordiale (*P* <0.001; **Table 5**); grain yield was not affected by N application level 242 but was significantly higher with PGR application (P = 0.040). When comparing 243 years, there was a significant interaction between cultivar and year (P = 0.036); grain 244 yields were highest in FE2 for all cultivars but Grafton had a much lower grain yield 245 than the other cultivars in FE1 while Xi19 had a much lower grain yield in FE3.

246 For straw yield, there were significant differences between cultivars in FE1 (P =247 (0.005) and FE2 (P < 0.001; Tables 3 and 4). However, in FE1, the difference was 248 only significant between the highest yielding cultivar, Maris Widgeon, and the four 249 lowest yielding cultivars, Grafton, Quartz, Zebedee and Sterling, while in FE2, Maris 250 Widgeon had significantly more straw than all the other cultivars. There was a 251 general trend for higher straw yields when PGRs were not used but this was not 252 significant. In FE3 straw yield was not significantly influenced by cultivar, PGR or 253 N. Excluding the leaf blades (leaving just stem and leaf sheath) did not lead to 254 significant differences between the treatments. Straw yields were significantly higher 255 in FE3 than the first two field experiments (P < 0.001) but across the three years there 256 was no significant difference in straw yield between the cultivars.

Splitting the stem into sections allows an assessment of the influence that combine harvester cutter height might have on straw yields. At a cutter height of 10 cm there was a significant cultivar effect (P = 0.036) with Xi19 having significantly higher straw yield than Grafton, with Cordiale having an intermediate yield, matching height order (**Table 6**). At a cutter height of 15 cm PGR application significantly 262 lowered straw yield (P = 0.036), while percentage difference in yield between Xi19 263 and Grafton increased from 4.1% to 5.5% (P = 0.007). At a cutter height of 20 cm 264 the percentage difference in yield between Xi19 and Grafton increased to 7.0% (P =265 0.001) and the percentage difference in yield between PGR treatments increased 266 from 5.5% to 7.3% (P = 0.009). However, from a practical viewpoint, these 267 significant differences are actually relatively minor. For example, the difference in straw yield between Grafton and Xi19 at a cutter height of 10 cm is 0.40 t ha<sup>-1</sup> but 268 this only increases to 0.49 t ha<sup>-1</sup> at a cutter height of 20 cm. 269

270 Assuming that the lower 10 cm remain on the field as stubble and the leaf blades are 271 lost during baling, on average 61% of the total leaf and stem material is harvested. 272 The proportion of total straw that is 'harvestable straw' varied with cultivar (P 273 <0.001) ranging from 58% for Grafton to 63% for Xi19. Although PGR application 274 lowered this ratio (P = 0.013) the actual difference was inconsequential with 61% 275 collected when no PGRs were applied compared to 60% for treated plants. These 276 results suggest that assuming 60% recovery of straw is a suitable approximation 277 regardless of cultivar and PGR treatment; however, it is unclear whether this percentage recovery would be seen with the cultivars from FE1, which were 278 279 considerably shorter.

In FE1, harvest index (HI) significantly varied with cultivar (P < 0.001) with Maris Widgeon having a much lower HI than other cultivars (**Table 3**). In FE2, HI significantly varied with cultivar (P < 0.001) ranging from 0.43 for Maris Widgeon to 0.61 for Grafton (**Table 4**). In FE3, HI significantly varied with cultivar (P < 0.001), being significantly lower in Xi19 than Cordiale and Grafton (**Table 5**). When comparing years, HI was similar in FE1 and FE2 but significantly lower in FE3 (P 286 <0.001). Xi19 also had a significantly lower HI than the other cultivars in all three 287 years (P = 0.001).

#### 288 **3.2 Saccharification potential**

In FE1, glucose yields and xylose yields for untreated straw did not significantly differ with cultivar (**Table 7**). After pretreatment, glucose yield did not significantly differ with cultivar. Straw saccharification potential differed significantly between cultivars (P < 0.001) with Maris Widgeon showing much lower saccharification potential than the other cultivars in both years. This is reflected in %GRwith Maris Widgeon having a lower value than the other cultivars (P < 0.001).

As with FE1, in FE2 glucose yields and xylose yields for untreated straw did not significantly differ with cultivar (**Table 8**) or with PGR. After pretreatment, glucose yield did not significantly differ with cultivar or PGR. Straw saccharification potential differed significantly between cultivars (P < 0.001) with Maris Widgeon having a much lower saccharification potential, which is also reflected in %GR with Maris Widgeon having a lower value than the other cultivars (P < 0.001). PGR did not significantly influence saccharification potential or %GR.

## 302 **3.3 Lodging assessments**

In all three years, SFWS was lower than RFWS indicating that stem lodging was more likely to occur. Average SFWS in FE3 was 10.62 m s<sup>-1</sup> (**Table 10**), which was much lower than the 18.65 m s<sup>-1</sup> in FE1 (**Table 8**) but very similar to the 10.66 m s<sup>-1</sup> in FE2 (**Table 9**). FE1 had lower stem leverage force and greater stem material strength than FE2 and FE3, leading to greater lodging resistance.

In FE1, SFWS differed significantly between cultivars (P < 0.001) with Zebedee 308 309 having a significantly lower SFWS and Grafton and Quartz having significantly 310 higher SFWS (Table 8). Maris Widgeon had the second lowest SFWS. In FE2 311 SFWS differed significantly among cultivars (P < 0.001) with Grafton having 312 significantly higher SFWS than the majority of the other cultivars (Table 9). 313 Although Maris Widgeon was much taller than the other cultivars it was less susceptible to lodging than some of the semi-dwarf cultivars; its high leverage force 314 315 was partly offset by a high material strength. As expected, PGR application 316 significantly increased SFWS (P = 0.026), due to lower leverage force. In FE3, Xi19 317 had significantly lower SFWS than Cordiale or Grafton (P < 0.001; Table 10). As 318 with FE2, PGR application significantly lowered the risk of lodging (P = 0.001).

In FE1, leverage force on internodes 1 and 2 significantly differed with cultivar (P <0.001), with Maris Widgeon having the greater leverage force. Height at centre of gravity (HCG) is a major determinant of leverage force and this significantly varied with cultivar (P <0.001; **Table 8**).

323 In FE2, leverage force on internode 1 was influenced by cultivar and PGR 324 application (P <0.001) as was leverage force on internode 2 (P <0.001 and P = 0.001, respectively; **Table 9**). In FE2 and FE3 there was a significant interaction 325 326 between PGR and cultivar (P = 0.002 and P = 0.005, respectively); leverage force 327 was lower after PGR application for all cultivars but the extent of this varied with 328 cultivar. This reflected the significant interaction between PGR and cultivar in 329 determining the HCG (P = 0.002) with some cultivars having much greater 330 reductions in HCG with PGR application; interestingly, percentage reduction in plant 331 height was not related to original plant height.

332 In FE3, leverage force for internodes one and two was much higher for Xi19 (P <0.001 for both) reflecting the greater height of Xi19 and the lower natural 333 334 frequency. Leverage force was also greater for plants without PGRs (P = 0.001 and 335 <0.001 for internodes one and two respectively). Interestingly leverage force was 336 lower for N1 treatment compared to the other N treatments (P = 0.041 and 0.035 for 337 internodes one and two respectively); however, when considering individual parameters that combine to give leverage force, N did not significantly affect these. 338 339 For material strength of internode 1 there was a significant interaction between 340 cultivar, nitrogen and PGR; this resulted from the material strength of internode 1 of 341 Cordiale without PGRs being much higher than for N1 and N3, while for other 342 cultivars with and without PGRs the material strength did not vary with nitrogen 343 treatment.

Differences between cultivars in stem material strength were not significant for internodes 1 and 2 in FE1 (**Table 9**). Cultivar significantly affected stem material strength for internode 1 in FE2 (**Table 10**; P = 0.002) but not internode 2.

In FE3 the material strength of internode 1 was significantly higher for Cordiale than the other cultivars (P < 0.001) but was not influenced by PGR or N. For internode 2, Grafton was significantly lower than Xi19, which in turn was significantly lower than Cordiale (P < 0.001). Neither PGR nor N had an influence.

#### 351 **3.4 Relationships between traits**

## 352 3.4.1 Grain and straw yields

For FE1 and FE2 (without PGRs) there was a strong positive relationship between straw yield and grain yield (P < 0.001) with a significant difference between FE1 and FE2 due to the difference in yield between the field experiments (adj.  $R^2 = 0.44$ . Regression lines: y = 0.3264x + 2.197 [FE1]; y = 0.3264x + 2.971 [FE2]). This relationship is skewed slightly by the inclusion of Maris Widgeon, which had high straw yields but low grain yield.

## 359 **3.4.2** Straw saccharification potential and lodging resistance

SFWS was positively related to saccharification potential (P < 0.001; adj.  $R^2 = 0.86$ . 360 Regression lines: y = 0.01685x + 12.71 [FE1]; y = 0.01685x + 3.30 [FE2]). This was 361 362 despite the negative relationship between saccharification potential and material strength of internode 1 (P = 0.004; adj.  $R^2 = 0.17$ . Regression lines: v = -0.728x + 0.004363 358.2 [FE1]; y = -0.728x + 392.5 [FE2]) and internode 2 (*P* < 0.001; adj.  $R^2 = 0.27$ . 364 Regression line: y = -2.488x + 399.6). However, as leverage force increases, 365 366 saccharification potential decreases for both internode 1 (P <0.001; adj.  $R^2 = 0.57$ . 367 Regression lines: y = -0.4406x + 272.2 [FE1]; y = -0.4406x + 340.53 [FE2]) and 368 internode 2 (P < 0.001; adj.  $R^2 = 0.62$ . Regression lines: y = -0.4119x + 245.2 [FE1]; y = -0.4119x + 315.3 [FE2]) reflecting the reduction in saccharification potential 369 with increasing plant height (P < 0.001; adj.  $R^2 = 0.74$ . Regression lines: y = -0.2254x370 371 + 492.6 [FE1]; y = -0.2254x + 536.4 [FE2]).

372 Maris Widgeon's much lower saccharification potential than the semi-dwarf cultivars 373 skews these relationships and without the inclusion of Maris Widgeon, the 374 relationship between saccharification potential and material strength is not 375 significant. The relationship between saccharification potential and leverage force is 376 still significant without Maris Widgeon but the regression line is much shallower 377 showing only a minor change in saccharification potential with increasing leverage force. The relationship between height and saccharification potential is also 378 379 significant when excluding Maris Widgeon.

## 380 3.4.3 Lodging resistance and straw yield

In FE1 and FE2 (without PGRs) there was a strong positive relationship between straw yield and plant height (P < 0.001; adj.  $R^2 = 0.62$ . Regression lines: y = 0.005495x + 0.904 [FE1]; y = 0.010755x - 1.356 [FE2]) with a difference between field experiments (P < 0.001) but with a greater increase in straw yield per unit height in FE2 (P = 0.003). This reflected the difference in growing conditions and the greater straw yields seen in 2011. Maris Widgeon had high leverage on the results reflecting it being much taller than the other cultivars.

As plant height (given as height at centre of gravity in the model) strongly influences lodging resistance it would be expected that there would be a negative correlation between straw yield and lodging resistance. However, regression analysis demonstrated a positive relationship (P < 0.001; adj.  $R^2 = 0.84$ . Regression lines: y =0.081x + 18.263 [FE1]; y = 0.081x + 9.423 [FE2]), albeit, only a very small increase in SFWS is seen when straw yield increases. There was a difference between the

- 394 field experiments, representing the large difference in mean SFWS between the two
- 395 field experiments.

#### 396 **4. Discussion**

The field experiments demonstrated variation in the key traits of interest, allowing cultivar selection for individual traits, but when considering all traits no cultivar stood out as an ideal candidate for use as a DPC. The results give some insight into the relationship between key characteristics, which will be important when selecting or breeding a DPC.

402 Maris Widgeon had a similar AGDM yield to the semi-dwarf cultivars but the 403 biomass was partitioned differently, with Maris Widgeon consistently having the 404 lowest grain yield and highest straw yield. Among the semi-dwarf cultivars the yields 405 tended to be inconsistent between years though this is common (Austin et al., 1980; 406 Shearman et al., 2005) due to variation in weather. For example, regional statistics showed average yields of 7.9, 8.0 and 6.4 t  $ha^{-1}$  for 2010, 2011 and 2012, 407 408 respectively (Defra, 2015). The high grain yields in FE2 suggest that rainfall did not 409 limit grain yield though may have retarded stem growth giving lower straw yields 410 and high HI. Growing conditions were initially favourable to high yields in FE3 (as 411 seen by the AGDM) but high rainfall in summer 2012 led to fungal disease that 412 lowered grain yield, which is reflected in the lower HI.

Excluding Maris Widgeon, there was a positive relationship between grain and straw yields, which was also seen by Larsen et al. (2012). This may represent variation in productivity among the cultivars with some cultivars better suited to the specific field conditions present; this is supported by the inconsistency in the relative performance of cultivars between years. HI tends to be conservative (Hay, 1995), so as conditions favour higher AGDM, straw yield would increase alongside grain yield. Each 419 cultivar had similar HIs in FE1 and FE2, supporting this conservative HI though420 FHB in FE3 led to inconsistent and lower HIs.

The lack of significant difference in AGDM between the non-semi-dwarf cultivar and the semi-dwarf cultivars supports that AGDM has not increased with breeding. This is in contrast to some studies (Shearman et al., 2005) but in agreement with others (Slafer & Andrade, 1989; Brancourt-Hulmel et al., 2003). An explanation for the lack of significance could be that the current study used a fairly narrow range of release dates, with only one non-semi-dwarf cultivar, whereas other studies have had much wider ranges.

428 The positive correlation between grain and straw yields suggests that managing for 429 higher grain yields will give higher straw yields so limitations on straw yield will 430 depend on the limitations for higher grain yields. Increasing straw yield without compromising grain yield is a central idea for developing DPCs. However, as 431 432 discussed in Townsend et al. (2015), HI is reaching its upper limits and, therefore, 433 further increases in grain yield will necessitate increases in AGDM. In fact, the HIs 434 for some cultivars in FE2 are approaching the hypothesised upper limit for HI of 0.64 435 (Foulkes et al., 2011) and similar HIs at this location have been recorded in previous 436 work (Whaley et al., 2000).

In agreement with the majority of the literature, saccharification potential of pretreated straw residue after enzyme hydrolysis varied among cultivars. Interestingly, Maris Widgeon, the only non-semi-dwarf cultivar, had the lowest straw saccharification potential in both years; as its overall glucose yield for untreated and pre-treated straw did not significantly differ from the other cultivars this suggests 442 that the material was more difficult to break down to release the glucose. This is in 443 agreement with other studies that found that saccharification potential is not 444 correlated with the cellulose present in the material (Murozuka et al., 2015). Capper 445 (1988) suggested that taller cultivars would have lower saccharification potential due 446 to having more stem relative to leaf than shorter cultivars; however, this was not 447 measured in the current study. In contrast to the current study Bellucci et al. (2015) 448 found an increase in saccharification potential with increasing plant height. 449 Lindedam et al. (2012) did not find a relationship between leaf-to-stem ratio and 450 saccharification potential, suggesting that cultivar-specific relationships of leaf and 451 stem sugar yield is more important in predicting the overall saccharification 452 potential. One difficulty in considering leaf-to-stem ratio is that in the senesced state, 453 the leaf blades are friable and are likely to be lost during harvesting. There are many 454 factors that can influence saccharification potential (Townsend et al., 2015) but the 455 current study did not attempt to determine reasons for differences observed in this 456 trait. Interestingly, even though Maris Widgeon had the lowest saccharification potential, because it had much higher straw yield it is likely to have a greater 457 458 bioethanol yield per unit area of crop.

There was little difference in lodging susceptibility among cultivars in each FE even with Maris Widgeon, which, as the only non-semi-dwarf cultivar, would be expected to have much greater risk of lodging. Maris Widgeon had the highest straw yield and was the tallest cultivar yet it was less susceptible to lodging than some of the semidwarf cultivars at that growth stage. This resulted from it having a small ear area, which reduced its leverage force, and a slightly higher stem material strength. The 465 higher lodging resistance in FE1 is due to a slightly smaller ear area resulting in466 lower stem leverage force.

467 To reduce future lodging risk Berry et al. (2007) proposed a lodging resistance wheat 468 ideotype; they suggested more biomass would be required in the lower stem to 469 increase stem material strength. Interestingly, this would require higher straw yield. 470 One important consideration is how this would impact on saccharification potential 471 and conversely whether changes to increase saccharification potential might lower 472 lodging resistance. Selecting cultivars for higher saccharification potential has been 473 suggested to lead to lower lodging resistance due to a negative relationship between 474 saccharification potential and stem material strength; the results of our study also 475 suggest that as saccharification potential increases, material strength decreases but 476 overall lodging resistance (i.e. SFWS) increases. This is partly explained by the 477 decrease in saccharification potential with increasing plant height. The current study 478 did suggest that material strength would be lower with higher saccharification 479 potential. This did not follow through to overall lodging susceptibility due to the 480 negative correlation between saccharification potential and plant height/stem 481 leverage force. It has been suggested that improving saccharification potential could 482 lead to greater risk of lodging but the opposite was found in this study. An important 483 caveat from this study is that some relationships among traits were only seen because 484 of the inclusion of the non-semi-dwarf cultivar Maris Widgeon.

Alongside cultivar choice, other management practices might influence the key traits
of a DPC. Application of the PGR chlormequat did not significantly reduce straw
yields, as previously reported in a number of crops (Bragg et al., 1984; Cox & Otis,
1989; Naylor, 1989; Rajala & Peltonen-Sainio, 2001). There is a caveat to this

489 though, as at higher cutter heights there was a significant reduction in straw yield 490 when chlormequat was applied (the lack of significance in published studies could be 491 due to the inclusion of straw that would normally be left on the field as stubble); 492 however, the actual difference in yield was minor and unlikely to warrant changing 493 farming practices. Considering that chlormequat did not have a significant effect on 494 overall straw yield but increased the SFWS while not affecting saccharification 495 potential suggests that farmers should continue to use chlormequat even when 496 supplying straw for biofuel production. There is anecdotal evidence that some 497 farmers are not applying PGRs in order to have higher straw yields for livestock but 498 it may be providing little benefit while increasing lodging risk.

499 Nitrogen application rate only had very limited effect on grain and straw yields; it is 500 likely that dry weather after the final nitrogen fertiliser application, meant that the 501 fertiliser was not washed into the soil and therefore N availability was similar for all 502 three treatments.

503 Decreasing the cutter bar height increased straw yields but there is a trade-off 504 between having these higher straw yields and the higher fuel costs; energy in straw 505 and the energy required to collect that additional straw (Špokas & Steponavičius, 506 2010). However, there are potential benefits of removing as much as possible if a 507 lower-intensity tillage practice is used afterwards as crop residue can harbour pests 508 and disease (Carter, 1994). Cutter height determined whether significant differences 509 in straw yield were seen among cultivars but that actual differences in yield were 510 very small.

511 As this study only captured cultivar characteristics for a limited number of cultivars 512 grown under a limited range of conditions, and there is only limited data from other 513 studies, it is possible that among currently grown cultivars there are those with 514 characteristics suited to the role of a DPC. However, based on our study, should 515 farmers wish to grow straw for bioenergy, they should base their cultivar choice on 516 grain yield potential for their location. Currently, farmers in the UK are provided with metrics on cultivar characteristics (for example, the AHDB's Recommended 517 518 Lists) but these do not include straw metrics. This lack of availability reflects the 519 limited demand for these metrics. From this study, the limited variation in straw 520 yields among cultivars, but variation between years, suggests that assigning values to 521 individual cultivars would be difficult.

522 Breeding techniques offer the opportunity for improving key characteristics. As the relationships found in this paper suggest, breeding crops for higher grain yields 523 524 might lead to higher straw yields. Breeding for higher saccharification potential has 525 been suggested as a possibility due to the variation seen among cultivars (Jensen et 526 al., 2011) though Bellucci et al. (2015) found only a limited genetic effect. Greater 527 saccharification potential did not lead to a reduction in the other key traits for the 528 cultivar assessed so targeted breeding for this trait might have potential. Genetic modification techniques could provide the best means of increasing saccharification 529 530 potential although there would be significant barriers to growing these crops in the 531 EU. One consideration is that although differences in saccharification potential were 532 seen in this study, as discussed in Townsend et al. (2015), it is unclear how these 533 differences would relate to industrial-scale processing. As with other studies, the pretreatment conditions were selected to achieve 50% subsequent saccharification of 534

- 535 glucose from a standard wheat cultivar. This allowed the identification of variations
- in saccharification potential between cultivars yet possibly does not reflect theindustrial process.

#### 538 **5.** Conclusions

The field experiments did not identify any outstanding DCPs as, among the high 539 540 grain-yielding cultivars, straw yields were similar. While there were no outstanding 541 DPC candidates, our data suggest that growers supplying straw should select high 542 grain-yielding cultivars and do not need to change management practices for existing cultivars because higher grain yield gives higher straw. PGRs should be used as they 543 544 have only a minimal impact on straw yields but reduce lodging risk. Although 545 saccharification potential did vary among cultivars, currently growers and breeders 546 should not consider saccharification potential; for growers there is no financial 547 incentive for growing higher saccharification potential material while for breeders it 548 will be necessary to see the pretreatment methods utilised at the commercial-scale in 549 order to determine the merit of developing higher saccharification potential cultivars.

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659

# 661 Figure captions

Figure 1a,b: a) average monthly temperature for FE1 (solid line), FE2 (large dashes)
and FE3 (small dashes), and b) total monthly rainfall for FE1 (black bar), FE2 (dark
grey bar) and FE3 (light grey bar). Measured at Sutton Bonington meteorological
centre.



**Table 1** 

671 Field experiments key information

		Field experiment	
	FE1	FE2	FE3
Sowing date	20/10/2009	13/10/2010	06/10/2011
Previous crop	Winter oats	Winter oats	Winter oats
SNS N Index	101.4 kg ha <sup>-1</sup> , SNS Index 2 (12/11/09)	32.9 kg ha <sup>-1</sup> , SNS Index 0 (09/09/10)	18.9 kg ha <sup>-1</sup> , SNS Index 0 ( $23/02/12$ )
Soil Indices	P:5, k:4, Mg:6, pH:6.8	P:4, K:3, Mg:4, pH:7.2	P:4, K:4, Mg:4, pH:7.6
Cultivations	Plough (16/09/09); Power harrow	Plough (16/09/10); Power harrow	Plough (13/09/10); Power harrow
	(16/10/09); Roll after drilling (22/10/09)	(11/10/10); Roll after drilling (14/10/10)	(22/09/10); Roll after drilling (06/10/10)
Seed rate	$250 \text{ seeds m}^{-2}$	$250 \text{ seeds m}^{-2}$	$250 \text{ seeds m}^{-2}$
Design	Randomised block design	Split plot	Split-split plot
Fertiliser	1.0 L ha <sup>-1</sup> Manganese Jett (09/11/09);	87 kg ha <sup>-1</sup> 34.5% Nitram (30 kg ha <sup>-1</sup> N;	2.0 L ha <sup>-1</sup> Headland Jet (24/02/12); 116
	148 kg ha <sup>-1</sup> 27N 9SO <sub>3</sub> (40 kg ha <sup>-1</sup> N,	08/03/11); 1 L ha <sup>-1</sup> Human Extra	kg/ha 34.5% Nitram (40 kg ha <sup>-1</sup> N;
	13.3 kg ha <sup>-1</sup> SO <sub>3</sub> ; 03/03/10); 1 L ha <sup>-1</sup>	(08/03/11); 1 L ha <sup>-1</sup> Human Extra	08/03/12); 2 L ha <sup>-1</sup> Headland Jett
	Human Extra, 232 kg ha <sup>-1</sup> 34.5% Nitram	$(25/03/11); 232 \text{ kg ha}^{-1} 34.5\% \text{ Nitram (80)}$	(20/03/12); 232 kg/ha 34.5% Nitram (80 kg
	(80 kg ha <sup>-1</sup> N; 08/04/10); 1 L ha <sup>-1</sup>	kg ha <sup>-1</sup> N; $06/04/11$ ); I L ha <sup>-1</sup> Human Extra	$ha^{-1}$ N; 11/04/12); Manganese 15% @1.5 L
	Human Extra (28/04/10); 159 kg ha <sup>-1</sup>	(20/04/11); 1/4 kg na <sup>-</sup> 34.5% Nitram (60 kg ha <sup>-1</sup> N: 06/05/11); 1 J ha <sup>-1</sup> Magnar	$na^{-1}(50/04/12)$ ; various rates of N (see trial plan for rates; $10/05/12$ ); Magnor @ 1 L ha
	34.5% Nitram (55 kg ha <sup>-1</sup> N; 14/05/10);	(24/05/11), 1 L ha Magnor $(24/05/11)$	$^{1}(23/05/12)$ : Magnor @ 1 L ha <sup>-1</sup> (25/05/12)
	1.23 L ha <sup>-1</sup> Magnor (27/05/10).		$(25/05/12)$ , wagnor $\approx 1 L$ na $(25/05/12)$ .
Herbicide	3.0 L ha <sup>-1</sup> Picona C (09/11/09); 1.2 L ha <sup>-</sup>	1 L ha <sup>-1</sup> Hatra, 1.7 L ha <sup>-1</sup> Picona, 1 L ha <sup>-1</sup>	0.6 L ha <sup>-1</sup> Liberator (09/11/2011); 25g ha <sup>-1</sup>
	<sup>1</sup> Hatra, 1.0 L ha <sup>-1</sup> Biopower (27/04/10);	Biopower (08/03/11); 1 L ha <sup>-1</sup> Spitfire	Lorate (20/03/12); 1 L ha <sup>-1</sup> Foxtrot & 1 L

1.23 L na <sup>2</sup> Starane XL (27/05/10).	(24/05/11)	ha <sup>-1</sup> Toil (24/04/12); 1 L ha <sup>-1</sup> Spitfire (23/05/12)
0.75 L ha <sup>-1</sup> Alto Elite (08/04/10); 0.65 L ha <sup>-1</sup> Proline, 0.75 L ha <sup>-1</sup> Amistar Opti (28/04/10); 1.23 L ha <sup>-1</sup> Brutus, 1.23 L ha <sup>-1</sup> Amistar Opti, 0.5 L ha <sup>-1</sup> Corbel (27/05/10); 0.75 L ha <sup>-1</sup> Folicur, 0.5 L ha <sup>-1</sup> <sup>1</sup> Corbel, 0.15 L ha <sup>-1</sup> Justice (09/07/10)	0.75 L ha <sup>-1</sup> Alto Elite, 0.15 L ha <sup>-1</sup> Vegas (25/03/11); 0.5 L ha <sup>-1</sup> Proline, 0.5 L ha <sup>-1</sup> Alto Elite (20/04/11); 0.5 L ha <sup>-1</sup> Comet, 0.1 L ha <sup>-1</sup> Justice, 0.5 L ha <sup>-1</sup> Proline (24/05/11); 0.75 L ha <sup>-1</sup> Caramba (15/06/11).	0.75 L ha <sup>-1</sup> Opus, 1.0 L ha <sup>-1</sup> Bravo, 0.4 L ha <sup>-1</sup> Instinct (20/03/12); 0.75 L ha <sup>-1</sup> Cortez, 1.3 L ha <sup>-1</sup> Phoenix (30/04/12); 0.75 L ha <sup>-1</sup> Opus, 1.3 L ha <sup>-1</sup> Phoenix (23/05/12); 0.85 L ha <sup>-1</sup> Orius, 0.15 L ha <sup>-1</sup> Vegas (25/06/12)
$0.25 \text{ L ha}^{-1} \text{ Permasect } (09/11/09)$	0.25 L ha <sup>-1</sup> Permasect (08/03/11); 0.25 L ha <sup>-1</sup> Aphox (15/06/11)	0.25 L ha <sup>-1</sup> Permasect (09/11/11); 0.28 kg ha <sup>-1</sup> Aphox (25/06/12)
None	1 L ha <sup>-1</sup> Chlormequat (+PGR plots only; 25/03/11); 0.8 L ha <sup>-1</sup> Chlormequat (+PGR plots only; 20/04/11)	1 L ha <sup>-1</sup> Chlormequat (+PGR plots only; 22/03/12); 0.8 L ha <sup>-1</sup> Chlormequat (+PGR plots only; 30/04/12).
	0.75 L ha <sup>-1</sup> Alto Elite (08/04/10); 0.65 L ha <sup>-1</sup> Proline, 0.75 L ha <sup>-1</sup> Amistar Opti (28/04/10); 1.23 L ha <sup>-1</sup> Brutus, 1.23 L ha <sup>-1</sup> Amistar Opti, 0.5 L ha <sup>-1</sup> Corbel (27/05/10); 0.75 L ha <sup>-1</sup> Folicur, 0.5 L ha <sup>-1</sup> <sup>1</sup> Corbel, 0.15 L ha <sup>-1</sup> Justice (09/07/10). 0.25 L ha <sup>-1</sup> Permasect (09/11/09) None	$0.75 L ha^{-1}$ Alto Elite (08/04/10); 0.65 L ha^{-1} Proline, 0.75 L ha^{-1} Amistar Opti (28/04/10); 1.23 L ha^{-1} Brutus, 1.23 L ha^{-1} Amistar Opti, 0.5 L ha^{-1} Brutus, 1.23 L ha^{-1} Amistar Opti, 0.5 L ha^{-1} Corbel (27/05/10); 0.75 L ha^{-1} Folicur, 0.5 L ha^{-1} Alto Elite (20/04/11); 0.5 L ha^{-1} Comet, 0.1 L ha^{-1} Justice, 0.5 L ha^{-1} Corbel (27/05/10); 0.75 L ha^{-1} Folicur, 0.5 L ha^{-1} Corbel, 0.15 L ha^{-1} Justice (09/07/10). 0.25 L ha^{-1} Permasect (09/11/09)0.75 L ha^{-1} Alto Elite, 0.15 L ha^{-1} Vegas (25/03/11); 0.5 L ha^{-1} Comet, 0.1 L ha^{-1} Justice, 0.5 L ha^{-1} Comet, 0.1 L ha^{-1} Justice, 0.5 L ha^{-1} Proline (24/05/11); 0.75 L ha^{-1} Caramba (15/06/11).None0.25 L ha^{-1} Permasect (08/03/11); 0.25 L ha^{-1} Aphox (15/06/11)None1 L ha^{-1} Chlormequat (+PGR plots only; 25/03/11); 0.8 L ha^{-1} Chlormequat (+PGR plots only; 20/04/11)

Cultivars assessed in field experiments. Source: AHDB Recommended Lists for
cereals and oilseeds 2009, 2010 and 2011. N.B. Maris Widgeon predates AHDB
Recommended Lists so data is not available on these key parameters; the date refers
to the year of introduction (Austin et al., 1980). Field experiments: 1, 2 and 3. Nabim
groups refer to the grain end-use (i.e. whether it is suited to milling or animal feed).
A rating of 1 refers to milling quality while 4 is for feed wheat.

Cultivar	Field	NABI	Resistanc	Resistanc	Height	Heigh	Year
	experimen	Μ	e to	e to	withou	t with	first
	t	group	lodging	lodging	t PGR	PGR	liste
			without	with PGR	(cm)	(cm)	d
			PGR				
Herewar	1, 2	1	8	9	88	-	1991
d							
Mascot	1, 2	1	6	8	93	84	2006
Xi19	1, 2, 3	1	4	6	97	88	2002
Battalion	1, 2	2	7	8	88	82	2007
Cordiale	1, 2, 3	2	8	9	82	76	2004
Sterling	1, 2	2	6.7	8.3	80	-	2010
Invicta	1, 2	3	7.2	7.5	93	86	2010
Riband	1, 2	3	8	8	89	-	1989
Zebedee	1, 2	3	6	6	87	84	2007
Ambrosi	1, 2	4	7	8	88	80	2005
а							
Glasgow	2	4	6	8	85	74	2005
Grafton	1, 2, 3	4	9	9	79	72	2009
Istabraq	1, 2	4	6	7	96	88	2004
Quartz	1, 2	4	9	9	75	-	2009
Maris	1, 2	-	-	-	-	-	1964
Widgeon							

- 682 Yield components of cultivars grown in FE1. ANOVA statistical output with degrees
- 683 of freedom (d.f.) and standard error of the differences between means (SED).

Cultivars	AGDM	Grain Yield	Straw Yield	Harvest Index
	(t ha⁻¹)	(t ha⁻¹)	(t ha⁻¹)	
Ambrosia	14.50	8.28	4.76	0.57
Battalion	14.57	8.21	4.81	0.56
Cordiale	17.07	9.98	5.20	0.58
Grafton	12.86	7.43	3.98	0.58
Hereward	14.22	7.80	4.76	0.55
Invicta	14.24	7.68	5.05	0.53
Istabraq	13.84	7.82	4.58	0.56
Maris Widgeon	13.56	5.61	6.68	0.42
Mascot	14.02	8.13	4.46	0.58
Quartz	13.19	7.67	4.00	0.58
Riband	15.87	9.05	5.17	0.57
Sterling	12.84	7.22	4.15	0.56
Xi 19	16.65	9.30	5.47	0.56
Zebedee	13.05	7.51	4.13	0.57
Mean	14.32	7.98	4.80	0.56
Р	NS	0.048	0.005	<0.001
SED	1.720	1.039	0.594	0.014
df	39	39	39	39

686	Yield comp	onents of c	ultivars g	rown in	FE2. ANO	VA statis	tical outp	ut with	degrees
	1			/					0

687	of freedom (d.f	) and standard er	for of the differences	s between means (	(SED).
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Treatment	t	AGDM	Grain Yield	Straw Yield	Harvest Index
		(t ha⁻¹)	(t ha⁻¹)	(t ha <sup>-1</sup> )	
Cultivar	Ambrosia	20.10	11.83	6.59	0.59
	Battalion	18.18	10.51	6.05	0.58
	Cordiale	17.23	10.34	5.30	0.60
	Glasgow	20.45	12.34	6.15	0.61
	Grafton	18.05	10.93	5.52	0.61
	Hereward	18.88	10.36	6.55	0.55
	Invicta	19.24	11.04	6.54	0.57
	Istabraq	20.13	11.50	6.81	0.57
	Maris Widgeon	18.19	7.89	8.75	0.44
	Mascot	18.97	10.87	6.43	0.57
	Quartz	17.15	10.41	5.41	0.59
	Riband	18.58	11.25	5.82	0.61
	Sterling	16.41	9.66	5.26	0.59
	Xi 19	18.96	11.33	5.88	0.60
	Zebedee	18.74	11.13	5.99	0.59
PGR	PGR+	18.00	10.53	5.85	0.58
	PGR-	19.23	10.95	6.55	0.57
Mean		18.62	10.74	6.2	0.58
Cultivar	Р	NS	<0.001	< 0.001	<0.001
	SED	1.357	0.780	0.508	0.007
	df	56	56	56	56
PGR	Р	NS	NS	NS	NS
	SED	0.645	0.286	0.318	0.004
	df	2	2	2	2

690 Yield components of cultivars grown in FE3. ANOVA statistical output with degrees

691	of freedom (	d.f.) a	nd standard	error of the	differences	between	means (SED).
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Treatmen	t	AGDM (t ha <sup>-1</sup> )	Grain Yield (t ha <sup>-1</sup> )	Straw Yield (t ha⁻¹)	Harvest Index
Cultivar	Cordiale	20.60	8.15	9.14	0.47
	Grafton	20.54	8.10	9.16	0.46
	Xi19	19.08	6.76	9.11	0.42
PGR	PGR+	20.36	7.89	9.07	0.45
	PGR-	19.79	7.45	9.20	0.44
Ν	N1	19.97	7.73	8.93	0.46
	N2	20.13	7.51	9.47	0.44
	N3	20.11	7.76	9.01	0.45
Mean		20.07	7.67	9.14	0.45
Cultivar	Р	0.017	< 0.001	NS	<0.001
	SED	0.550	0.256	0.272	0.009
	d.f	24	24	24	24
PGR	Р	NS	0.040	NS	NS
	SED	0.363	0.169	0.231	0.006
	d.f	6	6	6	6
Ν	Р	NS	NS	NS	NS
	SED	0.578	0.559	0.218	0.022
	d.f	4	4	4	4

695 Straw section yields (t ha<sup>-1</sup>) from FE3. ANOVA statistical output with degrees of

696 freedom (d.f.) and standard error of the differences between means (SED).

Treatmer	nt		Cutter heigh	t
incutinent		10 cm	15 cm	20 cm
Cultivar	Cordiale	4.71	4.12	3.57
	Grafton	4.50	3.90	3.33
	Xi19	4.90	4.34	3.82
PGR	PGR+	4.62	4.01	3.45
	PGR-	4.79	4.23	3.70
Ν	N1	4.51	3.93	3.40
	N2	4.91	4.31	3.75
	N3	4.69	4.12	3.58
Mean		4.70	4.12	3.57
Cultivar	Р	0.036	0.007	0.001
	SED	0.146	0.127	0.178
	d.f	24	24	24
PGR	Р	NS	0.036	0.009
	SED	0.107	0.082	0.066
	d.f	6	6	6
Ν	Р	NS	NS	NS
	SED	0.160	0.159	0.159
	d.f	4	4	4

700 Saccharification potential traits of cultivars in FE1. ANOVA statistical output with degrees of freedom (d.f.) and standard error of the

701 differences between means (SED).

Cultivars	Untreated glucose	Untreated xylose	Pretreated glucose	Saccharification potential	% saccharification potential
	(mg g⁻¹ straw)	(mg g⁻¹ straw)	(mg g <sup>-1</sup> preteated straw)	(mg g <sup>-1</sup> pretreated straw)	-
Cordiale	265	167	639	344	54.0
Hereward	244	161	643	324	50.6
Maris Widgeon	282	123	634	244	38.7
Quartz	241	165	623	374	60.3
Riband	251	157	633	336	53.1
Zebedee	234	156	631	339	54.0
Mean	253	155	634	327	51.8
Р	NS	NS	NS	<0.001	<0.001
SED	15.0	24.5	15.0	9.7	1.78
df	15	15	15	15	15

705 Saccharification potential traits of cultivars in FE2. ANOVA statistical output with degrees of freedom (d.f.) and standard error of the

706 differences between means (SED).

Treatment		Untreated glucose	Untreated xylose	Pretreated glucose	Saccharificat	% glucose release
					ion potential	
		(mg g <sup>-1</sup> )	(mg g⁻¹)	(mg g <sup>-1</sup> )	(mg g⁻¹)	-
Cultivar	Cordiale	302	155	581	391	67.5
	Hereward	277	148	571	376	66.0
	Istabraq	289	154	593	370	62.7
	Maris Widgeon	288	143	584	315	54.1
	Quartz	265	153	605	405	67.1
	Riband	288	150	598	381	64.0
	Zebedee	266	150	579	381	66.0
PGR	PGR+	290	149	591	381	64.7
	PGR-	275	149	584	367	63.2
	Mean	282	149	587	374	63.9
Cultivar	Р	NS	NS	NS	< 0.001	<0.001
	SED	18.4	4.2	13.1	10.7	2.52
	df	24	24	24	24	24
PGR	Р	NS	NS	NS	NS	NS

SED	34.64	7.36	5.98	6.23	0.991
df	2	2	2	2	2

708 Lodging components from FE1. I1 and I2 refer to internodes 1 and 2, respectively. ANOVA statistical output with degrees of freedom (d.f.) and

standard error of the differences between means (SED).

Cultivar	SFWS	Leverage for	orce (Nmm)	Stem material	strength (MPa)	HCG
	(m s⁻¹)	11	12	11	12	(mm)
Ambrosia	19.27	127.6	108.7	43.1	27.8	427.9
Battalion	17.80	103.0	86.5	33.9	24.8	414.4
Cordiale	19.71	93.1	79.0	47.4	33.2	393.7
Grafton	21.02	103.2	88.6	38.5	29.2	384.9
Hereward	19.05	116.1	100.7	53.1	30.3	436.1
Invicta	17.72	153.9	132.3	41.2	32.2	472.8
Istabraq	18.22	161.6	138.6	44.1	32.4	473.2
Maris Widgeon	16.08	184.1	160.9	49.0	35.3	582.5
Mascot	19.02	133.8	115.7	39.7	23.2	458.5
Quartz	20.27	99.4	85.5	34.9	27.1	380.5
Riband	19.47	125.3	107.8	39.0	27.8	440.4
Sterling	18.97	112.5	96.1	38.2	30.3	399.0
Xi 19	19.72	137.8	117.9	44.9	34.0	465.6
Zebedee	14.78	149.8	127.9	30.1	21.5	439.7
Mean	18.65	128.7	110.4	41.2	29.2	440.7
Р	<0.001	< 0.001	<0.001	NS	NS	<0.001
SED	1.011	8.83	7.86	7.83	4.27	11.46

df	39	39	39	39	39	39
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711 Lodging components from FE2. I1 and I2 refer to internodes 1 and 2, respectively. ANOVA statistical output with degrees of freedom (d.f.) and

512 standard error of the differences between means (SED).

Cultivar	PGR treatment	SFWS	Leverage force (Nmm)		Stem material strength (MPa)		HCG (mm)
		(m s <sup>-1</sup> )	11	12	11	12	<b>,</b>
Ambrosia	PGR-	9.83	147.7	147.7	29.12	11.45	452.7
	PGR+	11.20	127.1	127.1	33.49	12.57	422.8
Battalion	PGR-	10.76	137.0	137.0	32.37	16.09	461.5
	PGR+	11.25	97.8	97.8	28.86	12.06	406.8
Cordiale	PGR-	10.08	159.6	159.6	51.69	15.60	458.5
	PGR+	10.39	110.8	110.8	30.97	14.89	415.5
Glasgow	PGR-	10.52	158.9	158.9	27.36	14.81	473.8
	PGR+	10.51	124.0	124.0	26.90	12.22	432.5
Grafton	PGR-	12.64	128.7	128.7	38.60	15.10	416.2
	PGR+	13.50	100.8	100.8	28.86	16.29	389.5
Hereward	PGR-	10.38	127.0	127.0	42.11	13.95	456.0
	PGR+	11.27	107.5	107.5	31.79	12.08	417.3
Invicta	PGR-	10.64	180.7	180.7	30.87	15.56	496.9
	PGR+	11.34	171.3	171.3	28.81	13.46	479.0
Istabraq	PGR-	9.19	184.2	184.2	30.22	12.39	501.8
	PGR+	11.64	166.1	166.1	41.86	20.08	465.3
Maris Widgeon	PGR-	8.52	201.8	201.8	40.19	15.97	591.6

	PGR+	10.83	166.6	166.6	41.64	23.73	550.0
Mascot	PGR-	10.18	179.3	179.3	28.20	14.06	491.0
	PGR+	11.02	117.5	117.5	21.42	12.61	421.7
Quartz	PGR-	9.70	128.5	128.5	28.37	9.45	408.3
	PGR+	12.98	107.6	107.6	28.31	21.62	387.0
Riband	PGR-	9.73	159.8	159.8	26.70	9.38	467.8
	PGR+	11.62	128.6	128.6	25.90	9.74	433.2
Sterling	PGR-	9.24	152.5	152.5	34.71	11.79	429.3
	PGR+	11.74	128.7	128.7	32.16	21.83	417.3
Xi 19	PGR-	9.08	194.5	194.5	35.00	16.23	511.5
	PGR+	11.75	139.8	139.8	26.77	17.92	454.0
Zebedee	PGR-	8.79	187.7	187.7	24.92	12.86	464.0
	PGR+	9.43	160.3	160.3	26.63	14.64	445.4
Mean		10.66	159.4	146.1	31.83	14.68	453.9
Cultivar	Р	<0.001	<0.001	<0.001	0.002	NS	<0.001
	SED	0.659	6.61	6.35	4.333	3.114	6.48
	df	56	56	56	56	56	56
PGR	Р	0.026	0.001	<0.001	NS	NS	<0.001
	SED	0.230	1.13	0.90	0.967	1.664	0.59
	df	2	2	2	2	2	2
PGRxCultivar	Р	NS	0.002	0.005	NS	NS	0.002
	SED	0.930	9.11	8.73	5.999	4.568	8.87
	df	56	56	56	56	56	56

716 Lodging components from FE3. I1 and I2 refer to internodes 1 and 2, respectively.

717 ANOVA statistical output with degrees of freedom (d.f.) and standard error of the

718 differences between means (SED).

Cultivar	Nitrogen	PGR	Material	strength	Levera	ge force	SFWS	HCG
			(M	Pa)	(Nn	nm)	(m s⁻¹)	(mm)
			11	12	11	12		
Cordiale	N1	With	44.3	22.3	130.1	118.1	12.69	476.3
	N2		41.6	21.6	139.1	125.6	12.51	484.0
	N3		42.1	22.8	148.8	138.4	11.60	496.8
	N1	Without	35.3	17.9	159.9	147.6	10.36	527.0
	N2		48.9	25.0	173.5	156.9	11.16	540.5
	N3		36.4	22.9	174.7	160.6	9.95	537.7
Grafton	N1	With	26.3	15.7	137.2	123.4	11.79	466.0
	N2		34.4	17.9	143.6	132.4	12.30	465.9
	N3		30.4	16.8	148.6	135.3	11.60	466.2
	N1	Without	30.2	16.9	156.7	144.7	11.28	505.3
	N2		29.1	15.8	165.2	148.7	10.07	508.7
	N3		29.4	14.9	171.7	155.4	10.31	506.8
Xi19	N1	With	28.2	19.2	222.6	202.4	10.4	546.7
	N2		29.5	19.7	266.8	242.6	9.18	582.2
	N3		32.2	20.2	269.4	245.4	9.28	565.0
	N1	Without	30.2	20.4	291.8	267.9	8.78	615.3
	N2		27.9	19.8	311.8	286.1	9.09	620.6
	N3		35.4	19.3	308.8	283.9	8.85	604.9
Mean			34.0	19.4	195.6	178.6	10.62	528.7
Cultivar	Р		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
	SED		1.56	0.99	7.49	7.17	0.255	4.08
	df		24	24	24	24	24	24
PGR	Р		NS	NS	0.001	<0.001	0.001	<0.001
	SED		1.42	0.66	5.98	5.19	0.217	4.32
	df		6	6	6	6	6	6
Nitrogen	Р		NS	NS	0.041	0.035	NS	NS
Ū	SED		2.92	0.97	5.52	4.82	0.328	8.43
	df		4	4	4	4	4	4
C x PGR	Р		NS	NS	NS	NS	NS	NS
	SED		2.30	1.32	10.51	9.77	0.365	6.39
	df		24	24	24	24	24	24
CxN	Р		NS	NS	NS	NS	NS	NS

	SED	3.67	1.70	11.95	11.22	0.487	10.22
	df	24	24	24	24	24	24
N x PGR	Р	NS	NS	NS	NS	NS	NS
	SED	3.41	1.26	9.17	7.98	0.422	9.96
	df	6	6	6	6	6	6
N x PGR x C	Р	0.024	NS	NS	NS	NS	NS
	SED	4.63	2.35	17.56	16.40	0.662	12.87
	df	24	24	24	24	24	24