

# **Maximizing alcohol yields from wheat and maize and their co-products for distilling or bioethanol production**

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1 **Maximizing Alcohol Yields from Wheat and Maize and their Co-products for Distilling or**  
2 **Bioethanol Production**

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9  
10 **Abstract**

11 The key to optimising alcohol production from cereals is a full understanding the physiology and  
12 processing characteristics of different cereals. This study examined the maximum alcohol yields that  
13 can be obtained from wheat and maize using different processing technologies. Lower processing  
14 temperatures (85°C) resulted in high alcohol yields from wheat (a temperate crop), whereas higher  
15 processing temperatures (142°C) gave maximum alcohol yields from maize (a tropical crop). Similar  
16 trends were also observed when the spent grains from these cereals were subjected to cellulolysis  
17 using commercial enzymes. Mill settings were additional factors in influencing alcohol production.  
18 Wheat has the potential to produce higher alcohol yields when compared with maize, when residual  
19 biomass (i.e. spent grains) saccharification using selected commercial enzymes is taken into account.  
20 While this approach is not applicable for the Scotch whisky industry due to strict legislation  
21 forbidding use of exogenous enzymes, this is pertinent for bioethanol production- to increase the  
22 alcohol yield obtained from both starch and lignocellulosic components of whole cereal grains.  
23 Wheat and maize processing temperatures and the use of processing aids are of potential economic  
24 benefit to bioethanol producers and to beverage alcohol producers seeking to understand the  
25 factors influencing the processing properties of different cereals.

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29 |  
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## 32 Introduction

33 The production of grain whisky has many parallels with the technology that is used in some  
34 bioethanol distilleries. Both products can be produced from starch-based substrates originating  
35 from cereals such as wheat and maize (corn). Many other cereals can be used for both bioethanol  
36 and whisky production and at the Scotch Whisky Research Institute (SWRI) work has been carried  
37 out on a range of different cereals such as wheat, maize, sorghum and millet to show how their  
38 properties can be used to optimize alcohol yield (1; 2). Under the legal definition of Scotch whisky  
39 (3), the use of commercial enzymes or other additives to increase alcohol yield and process  
40 efficiency are strictly forbidden, and process improvements must be derived from a better  
41 understanding of the physiological and processing characteristics of different cereals. However,  
42 distillers are also interested in the production of neutral spirits (Grain Neutral Spirit (GNS)), which is  
43 used for non-Scotch whisky products such as vodka and gin, which are not subject to these  
44 constraints. Similarly, the production of bioethanol is free from these requirements and a much  
45 wider range of options of both technologies and substrates are available, so that bioethanol  
46 producers can use many suitable raw materials and process aids for different alcohol production  
47 processes. In the longer term, there is interest from Scotch whisky distillers in evaluating the  
48 potential of alternative cereals and residual plant materials or the co-products deriving from them  
49 for additional production streams, such as biogas production, which may also be relevant to  
50 bioethanol production (4; 5). Since such treatment of co-products such as spent (or dark) grains  
51 (distillers dark grains with solubles (DDGS)) occurs after they are removed from the Scotch whisky  
52 production process, they can be further processed without restrictions, within the requirements of  
53 the end-user market for these co-products, which is primarily for animal feeds, but could also  
54 potentially be used for bioethanol production.

55 Bioethanol is derived from the microbial fermentation of biomass to produce fuel alcohol (6), which  
56 is chemically identical to synthetically produced ethanol by the petrochemical industry. Recently,

57 emphasis has been placed on partial or total replacement of energy derived from fossil fuel to  
58 bioethanol derived from plant materials (7) and global production is dominated by processes using  
59 maize in USA and sugarcane in Brazil (8; 9). Ethical concerns are raised when bioethanol derived  
60 from cereals (wheat, corn, sorghum, millet) and tubers (such as cassava, yam and potato) leads to  
61 direct conflict with food production (10; 11; 12). In order for a biofuel to be sustainable it should  
62 have a net energy gain, provide environmental benefits, be economically competitive and be  
63 capable of being produced on large scale without disrupting agricultural food crops (13). It is also  
64 essential that a sustainable bioethanol process will not upset the balance between the greenhouse  
65 gas emissions generated as the carbon released by the burning of plant derived fuels, and those  
66 which can be captured by growing plants (i.e. carbon neutral). The production of biofuels from more  
67 diverse feedstocks, for example, based on co-products such as cellulosic agricultural residues and  
68 brewer's and distiller's spent grains will eventually offer more sustainable alternatives to cereals and  
69 other food crops, and should help to ameliorate food versus fuel arguments (14). Commercial scale  
70 production of cellulosic ethanol is now a reality with, for example, Beta Renewables plant now  
71 operational in Italy, and several US plants on stream in 2014 (Lane, 2013). It is estimated that other  
72 large scale production plants capable of producing 50,000 – 150,000 tonnes of cellulosic ethanol will  
73 be built in 2013/14 (15). In Germany an estimated 22 million tonnes of straw could be used to meet  
74 around 25% of Germany's current petrol requirements (15). In the UK, nine bioethanol plants that  
75 are likely to use wheat as raw material have been planned, each with a potential total capacity of  
76 approximately  $2 \times 10^6$  tonne bio-ethanol per annum (16; 17). There are at present three bio-ethanol  
77 plants in the UK – 'British Sugar' in Wisington, 'Ensus' (now owned by Crop Energies) in Teeside and  
78 'Vivergo' in Hull. The latter two plants each can potentially produce 400 million litres of bioethanol  
79 per annum (18). Although previous studies on the feasibility of bioethanol in the UK concluded that  
80 production costs were uncompetitive compared with petrol (17), the economics have been gradually  
81 improving over the last two decades with the use of improved commercial enzymes to liberate  
82 fermentable sugars.

83 [Agu et al. \(2\)](#) studied the effects of starch liberation and breakdown conditions and observed that  
84 wheat can produce a significantly higher spirit yield when processed at a lower temperature of 85°C,  
85 compared with the higher cooking temperatures required for other cereals such as maize which will  
86 normally produce its maximum alcohol yield when processed at much higher temperatures (typically  
87 142°C – 145°C). This observation was in agreement with other studies focussed on bioethanol  
88 production ([19; 20; 21](#)). These studies confirmed that the processing procedure should be tailored to  
89 the cereal being used. The lower processing temperature of wheat also lowered the residue viscosity  
90 which causes problems in downstream recovery of the distillery co-products ([2](#)). The higher viscosity  
91 of wheat is due, in part, to the presence of high levels of pentosan (8% w/w) compared to lower  
92 levels (3% w/w) in maize, sorghum and millet (Palmer, 1989). In particular, pentosans such as  
93 arabinoxylans and other polymers such as  $\beta$ -glucans are known to cause processing problems ([22;](#)  
94 [23; 24; 25; 26; 27](#)).

95 The present study was undertaken to investigate the effects of process conditions and enzyme  
96 processing aids on maximising alcohol yields from wheat and maize starch and lignocellulose (in the  
97 form of spent grains).

## 98 **Materials and Methods**

### 99 **Cereal samples**

100 Samples of cereals (wheat and maize) were obtained from two sources (I) soft, low nitrogen wheat  
101 (cv Viscount) from a trial site producing wheat for assessment for Scotch grain whisky production; (II)  
102 a commercial yellow maize sample (variety not specified) obtained from a Scotch whisky grain  
103 distillery.

### 104 **Alcohol yield from wheat and maize flours (142°C process)**

105 The following procedure based is on the work of [Brosnan et al \(28\)](#) and described fully by [Agu et al](#)  
106 ([1](#)) and simulates the production process in a “typical” Scotch whisky grain distillery. Cereal flour

107 (30g), obtained by milling the grains in a Buhler Miag disc mill set at either 2 (0.2mm), or 11 (1.1mm)  
108 was transferred into a stainless steel mashing beaker and slurried with water (81ml), and 25 $\mu$ L of  
109 Termamyl 120L Type L (a bacterial  $\alpha$ -amylase supplied by Novozymes France S.A.) was added. This  
110 was slowly heated up to 85°C (temperature rise 2°C /min) in a water bath, before pressure cooking  
111 in an autoclave, temperature programmed with a maximum temperature of 142°C for 15 minutes.  
112 The cooked slurry was cooled to 85°C and given a second treatment with Termamyl (25 $\mu$ L) for 30  
113 min to prevent starch retrogradation. The mash was then transferred to a water bath at 65°C and  
114 mashed for 1 hour with a predetermined amount of high enzyme grain distilling malt grist (Miag mill  
115 setting 2 (0.2mm) and 11 (1.1mm)), equivalent to a malt inclusion rate of 20% dry weight basis (dwb)  
116 to 80% wheat. After cooling to room temperature, the mash was pitched with distiller's yeast  
117 (*Saccharomyces cerevisiae* 'M' type, supplied by Kerry Ingredients & Flavours) at a pitching rate of  
118 0.4 (w/w) pressed yeast, transferred to a fermentation vessel and the weight adjusted to 250g with  
119 water. The mash was then fermented at 30°C for 68 hours and distilled to collect the alcohol. The  
120 alcohol yield was determined from the alcohol strength of the distillate, which was measured using  
121 an Anton Paar 5000 density meter. The alcohol yield was quoted as litres of alcohol per tonne (LA/t)  
122 on a dry weight basis (dwb).

### 123 **Alcohol yield from wheat and maize flours (85°C process)**

124 The procedure is similar to that described above except that the pressure cooking step where the  
125 slurry was transferred to the autoclave, as well as the second treatment with Termamyl was  
126 bypassed.

### 127 **Alcohol yield from wheat and maize spent grains (SG) (142°C process)**

128 The procedure was similar to that described above for the cereal flour, except that the cereal spent  
129 grains (30g), were slurried with acidified water (0.005M H<sub>2</sub>SO<sub>4</sub>) (130mL) rather than distilled water,  
130 and 25 $\mu$ L of Termamyl 120L Type L (a bacterial  $\alpha$ -amylase supplied by Novozymes France S.A.) and

131 heated up to 85°C before pressure cooking in an autoclave at 142°C for 15 minutes. The cooked  
132 slurry was transferred to a 85°C water bath and given a second treatment with Termamyl (25µL) for  
133 30 min to prevent starch retrogradation, after which the mash was then transferred to a water bath  
134 at 65°C and incubate for 1 hour with the addition of 25µL each of the commercial enzymes  
135 Bioglucanase ME 250, Bioprotease NL 100 and Promalt 4TR (supplied by Kerry Bioscience Ltd). The  
136 dose rate for all the enzymes is shown in **Table 1**. After cooling to room temperature, the mash was  
137 pitched with distillers yeast and then fermented as described earlier. The alcohol yield was also  
138 determined from alcohol strength of the distillate as described above.

### 139 **Alcohol yield from wheat or maize spent grains (SG) using cellulosic enzymes**

140 The alcohol yield from spent grains using cellulosic enzymes is essentially as described for the  
141 alcohol yield from milled cereal spent grains except that the commercial enzymes Bioglucanase ME  
142 250, Bioprotease NL 100 and Promalt 4TR were replaced with Cellic HTech and Cellic CTec (cellulosic  
143 and xylan hydrolysing enzymes, supplied by Novozymes France S.A.) and the mashing time was  
144 extended overnight to 24 hours. The effect of the addition of the commercial enzymes at different  
145 stages, before (upstream) and after (downstream) cooking, was also investigated using maize spent  
146 grains.

### 147 **Rheological properties of the spent grains using Rapid Visco-Analysis (RVA)**

148 The physiological properties of the spent grains were studied using a Newport Scientific Rapid Visco  
149 Analyser (RVA) instrument supplied by Calibre Control. The Rapid Visco-Analyser is a rotational,  
150 continuously recording viscometer, with heating, cooling and variable shear capabilities, specifically  
151 configured for starch-based materials. The aim was to confirm that limited or no starch was present  
152 in the spent grains, and the alcohol yield was obtained from the spent grains. Here, a slurry of milled  
153 cereal spent grain (approximately 3.0g spent grain and a measured amount of water or acid total

154 weight 28g) with water or 0.005M sulphuric acid (H<sub>2</sub>SO<sub>4</sub>) was processed in the RVA analyser using a  
155 programme designed for un-malted cereals (29; 1).

## 156 **Results and Discussion**

157 Results shown in **Tables 2** and **3** verify the precision and the robustness of a high temperature  
158 pressure cook for the determination of alcohol yield from cereals. The results presented in **Tables 2**  
159 **and 3**, were obtained when the Miag mill was set at 2 (0.2mm) to produce finely milled flour from  
160 the cereals. **Table 2** shows the results obtained when wheat and maize were processed at the higher  
161 temperature of 142°C. This represents the traditional, high temperature process used for grain  
162 whisky production. **Table 3** shows the results obtained when the same wheat and maize samples  
163 were processed at a lower temperature of 85°C which represents a potential way of reducing the  
164 energy requirement for the process. Alcohol yields obtained from each of the cereals were  
165 reproducible whether the cereals were processed at either 142°C or 85°C, and confirm the reliability  
166 and robustness of the method.

167 When both cereal types were processed at the higher temperature of 142°C, wheat gave a much  
168 lower alcohol yield compared with maize. The differential of at least 15 litres alcohol per tonne is  
169 fairly typical for wheat and maize. On the other hand, when both cereal types were processed at the  
170 lower temperature of 85°C, maize gave a much lower alcohol yield than wheat. This is because maize  
171 requires higher temperatures than wheat to fully gelatinise the starch (1; 30). These results are  
172 consistent with observations by other researchers (19; 20; 21). In order to establish the maximum  
173 extractable alcohol yield potential from wheat and maize, the spent grains (after initial processing of  
174 the cereal flours) were re-processed using various commercial enzyme preparations (Bioglucanase  
175 ME 250, Bioprotease NL 100 and Promalt 4TR). **Table 4** shows alcohol yields obtained from the spent  
176 grains when re-processed at temperatures of 142°C or 85°C. The results showed that maize spent  
177 grains yielded more alcohol than wheat spent grain when they were processed at 142°C in the  
178 presence of the enzyme mixture. In contrast, at the lower temperature wheat spent grains gave a



179 much higher alcohol yield (84 litres of alcohol /tonne), almost 5 orders of magnitude, compared with  
180 the maize spent grains. This observation is important because it shows that the additional substrate  
181 necessary to generate the increased alcohol yield from wheat and maize is present in the spent  
182 grains, although the maize spent grains contain much less available substrate than wheat at 85°C.

183 When the total alcohol yield from maize flour and spent grains processed at 142°C were combined,  
184 the overall alcohol yield was much higher than that for the wheat flour and spent grains (**Figure 1**),  
185 by about 24 litres of alcohol per tonne. This would suggest that the high temperature has released  
186 the starch more efficiently from the maize kernel (endosperm) reflecting the generally higher starch  
187 content of maize. It is well known that maize requires more vigorous processing conditions for  
188 optimum performance in the distilling industry, while wheat needs less severe conditions to  
189 efficiently extract and solubilise the starch. Again, as well as its starch having higher gelatinization  
190 temperature compared with wheat, maize can be prone to developing resistant starch during  
191 processing. In contrast, the gelatinization temperature of wheat starch is much lower (**30; 1; 2**).

192 The opposite trend was seen at the lower processing temperature (85°C), for wheat yielded about  
193 80 litres of alcohol per tonne more than for maize (see **Figure 1**). The difference between the  
194 relative contribution of the components of the wheat and maize spent grains (about 70 litres of  
195 alcohol per tonne) accounted for a large proportion of the differential between the combined  
196 alcohol yields of the flour and spent grains components. This shows that at the lower temperature of  
197 85°C wheat has a bigger 'untapped' substrate in the spent grains (e.g. as cell wall materials),  
198 compared with maize. In wheat these can be released at the lower temperature by the combination  
199 of enzymes that were used. The lower temperature process would also be expected to produce  
200 fewer Maillard reaction products, which are known to significantly reduce alcohol yield. Therefore,  
201 there is additional potential for wheat to produce more alcohol if the processing conditions are  
202 adjusted to take account of both flour and spent grains. This is an important observation because it  
203 shows that during "normal" processing of wheat flour, materials residing in the spent grains were

204 limiting alcohol production. Such materials may also be responsible for higher residual viscosities  
205 found when processing wheat compared with maize (1; 2; 31). When the spent grains were re-  
206 processed using commercial enzyme preparations at 85°C, over 84 LA/t (dry basis) of extra alcohol  
207 yield was obtained, which indicates that wheat spent grains have potential for bioconversion to  
208 bioethanol. We additionally investigated the effects of using acidified process liquor and adding  
209 enzymes at different stages. The results in Table 5 (for maize spent grains) show that the effect of  
210 adjusting the process liquor to give a low acid concentration (0.005M H<sub>2</sub>SO<sub>4</sub>) gave higher alcohol  
211 yields from spent grains when the enzymes were added at the downstream process (after cooking)  
212 rather than upstream process (before cooking). The higher acid concentration (0.05M H<sub>2</sub>SO<sub>4</sub>) was  
213 less effective in this regard

214 Particle size of milled cereal samples also influenced subsequent alcohol yields. The results discussed  
215 above were obtained when the 'fine' Miag mill setting 2 (0.2mm) was used to give flours with small  
216 particle sizes. However, in many industrial processes, grains are bruised or 'cracked' rather than  
217 finely hammer milled. Bruised grains will produce very coarse grists, depending on the mill settings.  
218 We evaluated alcohol yields from coarse (1.1mm Miag mill setting) versus finely milled (0.2mm)  
219 wheat and maize and the results are shown in Table 6. Average alcohol yields obtained from coarse  
220 wheat and maize grist were higher than those obtained for the fine flour when both cereal types  
221 were processed at the higher temperature of 142°C. The higher alcohol yields from coarse-ground  
222 wheat or maize did lead to lower residual alcohol recoverable from the respective spent grains. A  
223 similar effect was also found when the cereals were processed at 85°C. Coarse milling is preferred in  
224 industrial bioprocesses to avoid the problem of "balling" when the cereal is slurried prior to cooking  
225 in the distillery. This phenomenon can reduce the hydration of the starch and prevent it from being  
226 released properly. Whilst, under ideal conditions, fine milling would result in larger surface areas to  
227 provide easier access to enzymes, in practice some potentially fermentable material would be lost  
228 through Maillard reactions during the cooking process. In contrast, the smaller surface area provided  
229 by coarse milling may limit loss of sugars and amino acids by Maillard reactions, and at the same

230 time release gelatinized starch for enzymolysis during the mashing process to improve yeast  
231 fermentation performance.

232 It was important to establish that alcohol obtained from spent grains of processed wheat and  
233 maize was not produced from residual starch To confirm this, pasting properties of the spent grains  
234 were analysed using a rapid visco-analyser (RVA) using 0.005M H<sub>2</sub>SO<sub>4</sub> which would release some  
235 fermentable non-starch polysaccharides (31). The results obtained for maize spent grains are shown  
236 in **Figure 2** and **Figure 3** for wheat spent grains. Both RVA profiles show that the peak and final  
237 viscosities were effectively zero, indicating the absence of starchy materials after processing. These  
238 results confirm that the additional alcohol yields obtained from the cereal spent grains emanated  
239 from lignocellulosic-derived sugars following pretreatment using dilute acids and hydrolysis with  
240 cellulolytic enzymes

## 241 **Conclusions**

242 This investigation has highlighted that understanding the physicochemical properties of different  
243 cereals used in industrial processes is the key in optimising alcohol yields, both from starch in the  
244 grains and lignocellulose in the spent grains A relatively low processing temperature (85°C) was  
245 found to be sufficient to obtain a high alcohol yield from wheat (a temperate crop), while a higher  
246 processing temperature was required to produce optimum yields from maize (a tropical crop). The  
247 study further showed that cereal mill settings influenced alcohol yields, which is pertinent for both  
248 potable spirits such as Scotch whisky as well as bioethanol. When the spent grains from these  
249 cereals were processed using commercial enzymes at the higher and lower temperatures, there  
250 were significant differences between wheat and maize. When the residual lignocellulosic material in  
251 spent grains is taken into account, then wheat has the potential to produce a higher alcohol yield  
252 compared with maize. However, this necessitates wheat spent grains being treated under more  
253 vigorous processing conditions, including acid pretreatment and hydrolysis using commercial  
254 enzymes. This is currently prohibited for Scotch whisky production but co-products such as spent

255 grains could be re-processed into neutral spirits or bioethanol that are not directed at the Scotch  
256 whisky market. The bioethanol industry is not faced with such restrictions and our findings may  
257 prove beneficial with regard to maximizing alcohol yields from whole cereals when correct mill  
258 settings, processing temperatures and enzymes are adequately combined.

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339 44.
- 340

341 **Table 1**

342

343 Commercial enzyme preparations and dose rate used in this study

344

| Commercial enzyme   | Type                          | Supplier              | Dose rate  |
|---------------------|-------------------------------|-----------------------|------------|
| Termamyl 120L       | Heat-stable $\alpha$ -amylase | Novozymes France S.A. | 25 $\mu$ L |
| Promalt 4TR         | $\alpha$ -amylase             | Kerry Bioscience Ltd  | 25 $\mu$ L |
| Bioprotease N100L   | Proteolytic enzyme            | Kerry Bioscience Ltd  | 25 $\mu$ L |
| Bioglucanase ME 250 | Endo-beta glucanase           | Kerry Bioscience Ltd  | 25 $\mu$ L |
| Cellic Htech        | Cellulase Complex             | Novozymes Denmark     | 25 $\mu$ L |
| Cellic Ctec         | Endoxylanase                  | Novozymes Denmark.    | 25 $\mu$ L |

345

346

347 **Table 2**

348

349 Alcohol yield results of wheat cooked at 142°C - High Temperature (HT) (Miag mill setting 2)

350

| Wheat (cv Viscount) /<br>Maize (unspecified variety) | Weight of sample<br>flour | Alcohol yield (LA/t)<br>dry | Average alcohol<br>yield (LA/t) dry |
|--|---------------------------|-----------------------------|-------------------------------------|
| Wheat 1  | 29.9999                   | 448.71                      |                                     |
| Wheat 2  | 30.0000                   | 449.39                      |                                     |
| Wheat 3  | 29.9998                   | 448.09                      |                                     |
| Wheat 4  | 29.9999                   | 445.03                      |                                     |
| Wheat  |                           |                             | 447.81                              |
| Maize 1  | 30.0003                   | 463.08                      |                                     |
| Maize 2  | 29.9998                   | 465.49                      |                                     |
| Maize 3  | 30.0003                   | 464.86                      |                                     |
| Maize 4  | 29.9998                   | 463.21                      |                                     |
| Maize 5  | 29.9996                   | 461.67                      |                                     |
| Maize  |                           |                             | 463.66                              |

351

352

353 **Table 3**

354

355 Alcohol yield results of wheat cooked at 85°C - Low Temperature (LT) (Miag mill setting 2)

356

| Wheat (cv Viscount) /<br>Maize (unspecified<br>variety) | Weight of sample<br>flour | Alcohol yield (LA/t) dry | Average alcohol yield<br>(LA/t) dry |
|---|---------------------------|--------------------------|-------------------------------------|
| Wheat 1   | 29.9999                   | 459.13                   |                                     |
| Wheat 2   | 29.9996                   | 454.32                   |                                     |
| Wheat 3   | 30.0000                   | 459.74                   |                                     |
| Wheat   |                           |                          | 457.73                              |
| Maize 1   | 30.0000                   | 443.03                   |                                     |
| Maize 2   | 30.0003                   | 441.06                   |                                     |
| Maize 3   | 30.0003                   | 441.43                   |                                     |
| Maize 4   | 29.9999                   | 441.06                   |                                     |
| Maize   |                           |                          | 441.65                              |

357



358 **Table 6**

359

360 Alcohol yield (LA/t) dry obtained from spent grains at different processing temperatures (142°C and  
361 85°C)

362

|                    | Alcohol yield (LA/t) dry |       |
|--------------------|--------------------------|-------|
|                    | 142°C                    | 85°C  |
| Wheat spent grains | 36.13                    | 83.93 |
| Maize spent grains | 43.60                    | 17.60 |

363

364

365 **Table 7**

366

367 Effect of adding 0.005M and 0.05M Sulphuric Acid (H<sub>2</sub>SO<sub>4</sub>) at different stages of stage of enzyme  
368 addition on the alcohol yield (LA/t) from maize spent grains. (Pre-cooking = upstream; Post cooking  
369 =downstream)

370

| Stage of Enzyme Addition | Weight of spent grain sample (g) | Acid concentration                    | Alcohol yield (LA/t) dry |
|--------------------------|----------------------------------|---------------------------------------|--------------------------|
| Upstream                 | 29.9999                          | 0.005M H <sub>2</sub> SO <sub>4</sub> | 8.33                     |
| Downstream               | 30.0000                          | 0.005M H <sub>2</sub> SO <sub>4</sub> | 23.53                    |
| Upstream and downstream  | 30.0002                          | 0.005M H <sub>2</sub> SO <sub>4</sub> | 18.53                    |
| Upstream                 | 30.0000                          | 0.05M H <sub>2</sub> SO <sub>4</sub>  | 0.00                     |
| Downstream               | 30.0003                          | 0.05M H <sub>2</sub> SO <sub>4</sub>  | 1.53                     |

371

372

373 **Table 8**

374

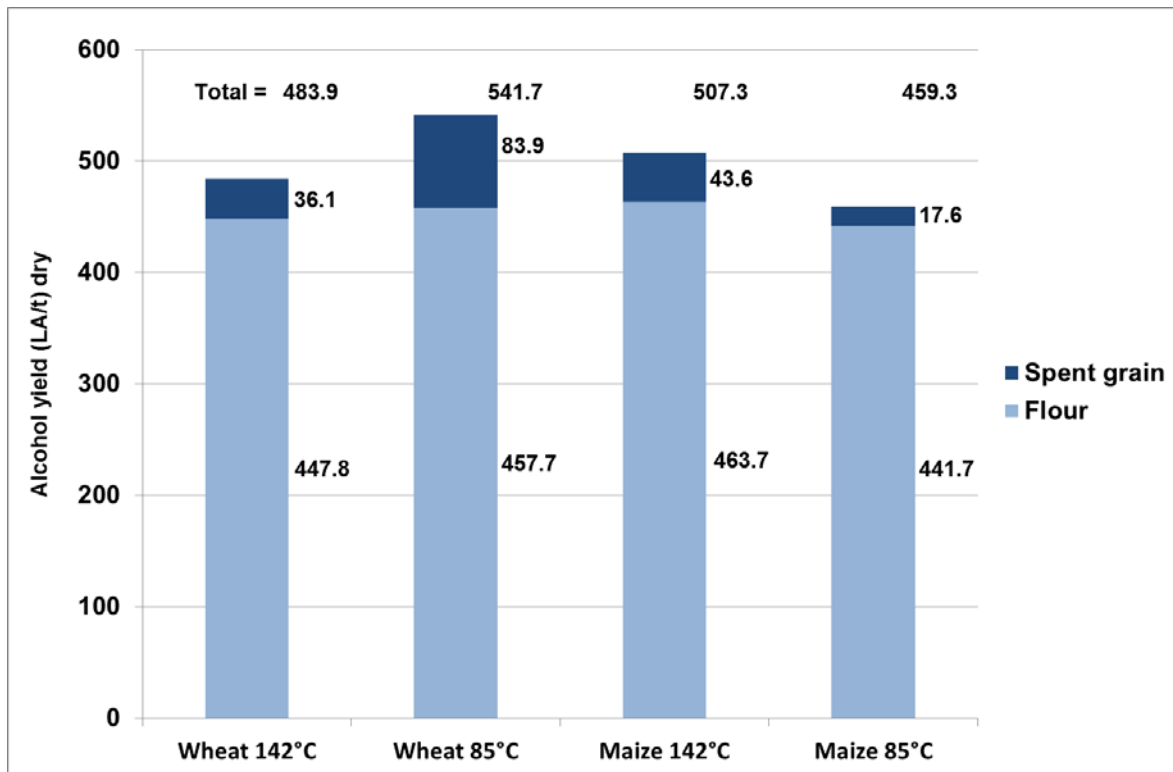
375 Alcohol yield (LA/t) dry obtained from wheat and maize flour and spent grains at different cook  
376 temperatures and a coarse mill setting (Miag setting 1.1 mm)

377

| Sample Fraction | Alcohol yield (LA/tonne) dry<br>142°C cooking temperature |             |        | Alcohol yield (LA/tonne) dry<br>85°C cooking temperature |             |        |
|-----------------|---|-------------|--------|--|-------------|--------|
|                 | Flour   | Spent grain | Total  | Flour  | Spent grain | Total  |
| Wheat           | 460.11  | 15.13       | 475.24 | 448.98   | 11.67       | 460.65 |
| Maize           | 474.05  | 16.27       | 490.32 | 429.73   | 8.20        | 437.93 |

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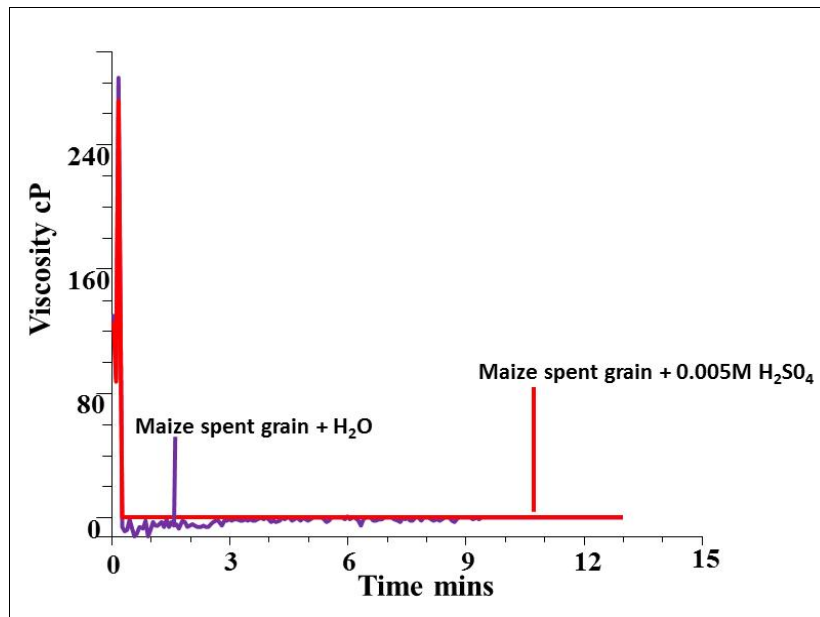
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**Figure 1**

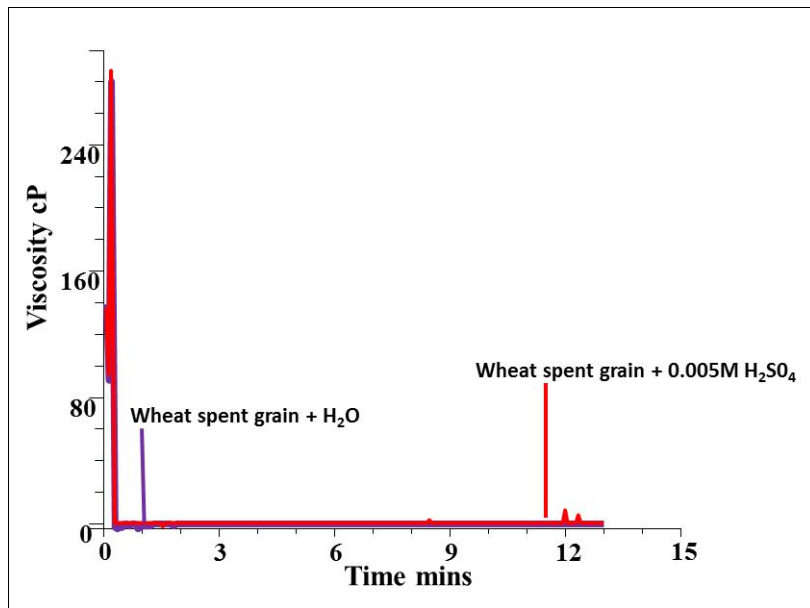
Combined spirit yields obtained from wheat and maize flours and respective spent grains (Miag mill setting 0.2mm) processed at 142°C and 85°C



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**Figure 2**

RVA pasting profile of maize spent grain using water or 0.005M H<sub>2</sub>SO<sub>4</sub> as process liquor



393  
394

395 **Figure 3**

396

397

398 RVA pasting profile of wheat spent grain using water or 0.005M H<sub>2</sub>SO<sub>4</sub> as process liquor

399

399