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Effects of added enzymes on sorted, unsorted and sorted-out barley: A model study on realtime viscosity and process potentials using rapid visco analyser

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ABSTRACT

Barley sorting is an important step for selecting grain of required quality for malting prior to brewing. However, brewing with unmalted barley with added enzymes has been thoroughly proven, raising the question of whether traditional sorting for high quality malting-barley is still necessary. To gain more insight on this, we examine realtime viscosity of sorted-out and unsorted barley during downscaled mashing with added enzymes in comparison with malting quality sorted barley. A rapid visco analyser was used to simulate brewery mashing process at lab scale together with two commercial enzymes (Ondea®-Pro and Cellic®-CTec2). During downscaled mashing, viscosity profile of sorted-out barley was markedly different from others, irrespective of enzyme type, whereas a small difference was observed between the sorted and un-sorted barley. Furthermore, whilst sorted-out barley generated lowest sugar-concentration, unsorted and sorted barley resulted in higher sugar-content, regardless of the enzyme used. In terms of filterability, the Ondea®-Pro treatment resulted in significantly lower-turbidity and smaller particle-size compared to Cellic®-CTec2; however, this effect was observed in sorted and un-sorted barley but not in sorted-out barley. Consequently, we find that unsorted barley demonstrates great potential for brewing with added enzymes and its use may help to improve sustainability of the brewing process.

1. Introduction

Barley (*Hordeum vulgare* L.) is an important global food and ranked fifth worldwide. Crop production averaged over 134 million tons per year between the years 2010–2013; following maize, rice, wheat and soybean (FAO, 2014). Traditionally, barley is the choice for brewing but must adhere to specific quality standards requiring production under intensive management practices such as planting season, fertilizers, plant growth stages, harvest time/conditions and post-harvest storage practices (Nelson, 2005; Arnold, 2005; ANALYTICA – EBC, 2010; AMBA, 2011).

A typical brew starts with a malting process, where barley grains are converted into malt through controlled germination, providing essential enzymes for mashing process such as protease and α-amylase (Gupta et al., 2010). It is therefore very important that barley for malting has the right properties. Physical sorting (*e.g.* correct size, shape and colour, etc.) as well as properties related to germination are used to ensure that a well modified malt will be produced containing large amounts of hydrolytic enzymes. Sorted-out barley or batches of unsuitable quality are generally used as animal feed with a consequent lower price.

Brewing with unmalted barley and added enzymes has been documented in recent years (Kordialik-bogacka et al., 2014; Steiner et al., 2012; Aastrup, 2010; Lowe et al., 2004). More recently, our research group have shown that it is also possible to brew beers using 100% oat, wheat and rye with added enzyme Ondea® Pro (Zhuang et al., 2016). Completely avoiding the malting process can lead to several economic and social benefits, including total savings of EUR 0.5–1.0 per hectolitre of beer depending on local conditions, grain and enzyme prices, elimination of carbon footprint associated with malt production, reduction of CO_2 emission by 8 g per 33 cl of beer (Aastrup, 2010; Kløverpris and Spillane, 2010; Steiner et al.,

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2012), as well as the direct use of local crops, making the beer production more sustainable (Aastrup, 2010; Nelson, 2005; Arnold, 2005). In addition, exogenous use of enzymes is constantly increasing in order to increase efficiency of beer due to decreased barley quality for brewing because of climate change impact on poor harvest (Kløverpris and Spillane, 2010; Steiner et al., 2012). In fact, malting performance is no longer directly important when enzymes are added during mashing. Using unsorted barley may be a way to further reduce energy and production costs in brewing. Nevertheless, previous studies on brewing with unmalted barley were conducted based on high quality sorted barley, and the effect of added enzymes on process potentials derived from unsorted and sorted-out barley is not well understood.

The Rapid Visco Analyser (RVA) is normally used for viscosity measurements, but has been reported to be a useful tool to mimic and record the process of mashing in a small scale with tight control of mashing duration and temperature profile (Goode et al., 2005; Holmes, 1995). Studies have been conducted to predict the quality potentials of malt and cereals, such as enzymatic activities and degree of modification (Goode et al., 2005, 2006) by monitoring rheological profiles of samples in the RVA apparatus. However, most of the studies were conducted on rice, buckwheat, wheat and maize (Yilmaz et al., 2015; Martínez et al., 2015a, 2015b) and few studies can be found on the use of this instrument using barley samples.

Purpose of the current study was to investigate the effect of added enzymes on rheological profiles derived from different types of barley using the RVA apparatus, and subsequently to characterize the quality potential of the respective wort samples. It is anticipated that the data presented here will provide more functional information regarding raw material utilization, potentially leading to *improved* resource use and process efficiency in the future.

2. Materials and methods

2.1. Source of barley and enzymes

Sorted, unsorted and sorted-out barley (Quench: spring variety) were purchased from Danish Malting Group and used within 6 months of harvest. Each kind of barley was ground using a coffee grinder (DeLonghi KG40 coffee grinder, USA) to achieve a uniform particle size of approximately 1 mm.

Two commercial enzymes (Ondea® Pro and Cellic® CTec2) were provided by Novozymes (Denmark) as gifts. Ondea® Pro is designed for brewing purposes, containing alpha-amylase, cellulase, xylanase (endo-1-4), protease (neutral) and lipase. Cellic® CTec2 is a blend of aggressive cellulases with high level of beta-glucosidases and hemicelluloses, mainly used for degradation of cellulose to obtain fermentable sugars (Novozymes.com).

2.2. Downscaled mashing process

The downscaled mashing process was carried out using a rapid visco analyser (Newport Scientific, Australia). 'Mash-in' was prepared by mixing 6.75 g of ground barley grist with 25 mL of brewing water (pH 5.8) in the coated cans with paddles (Newport Scientific, Australia). Subsequently enzymes (Ondea® Pro or Cellic® CTec2) were added in the concentration of 2.00 g/kg or 1.85 g/kg, respectively, as recommended by the manufacturer. A pre-established mashing regime (Aastrup, 2010) was used as follows: 54 °C and rest for 30 min; heating up to 64 °C and rest for 60 min; a further heating up to 80 °C and rest for 10 min. At each stage the heating rate was at least 1° per minute.

At the end of the programme, each wort sample (ca. 25 mL) was collected and filtered by making use of a filter of *muslin* cloth. Subsequently 5 mL of the brewing water was sparged onto the spent grain and collected by filtration with pressure application (a metal weight 938 g and an area of 22.1 cm^2). The two collections above were combined and stored at −20 °C until further analysis.

2.3. Analytical methods

Moisture content, germination energy and thousand corn weights of barley were determined according to European Brewery Convention (EBC) method 3.2, 3.6.2 and 3.4, respectively. Particle distribution of ground barley materials were determined by a Mastersizer 2000 (Malvern Instruments, UK).

Rheological profile of each sample was monitored by the rapid visco analyser throughout the mashing process. Total sugar concentration, turbidity and particle size distribution in the final wort samples were determined using a refractometer (Atago®, Master-α, Japan), a spectrophotometer at an absorbance of 600 nm (Thermo Fisher, Denmark), and a Mastersizer 2000 (Malvern Instruments, UK), respectively. Free amino nitrogen (FAN) content was determined according to ANALYTICA – EBC (2010) method 8.10.

3. Results and discussion

3.1. Characterisation of barley

Prior to mashing, typical quality parameters of barley were

Table 1
Characteristics of barley used in this study (Data is presented as the mean of duplicate samples ± range of the duplicates).

Parameters (unit)	Barley		
	Sorted	Unsorted	Sorted-out
Moisture content (%)	8.7 ± 0.3	9.4 ± 0.9	8.7 ± 0.5
Germination capacity (%)	96.7 ± 2.1	82.7 ± 2.5	80.3 ± 2.5
Thousand corn weight (g)	44.9 ± 0.5	44.9 ± 0.4	19.5 ± 0.4

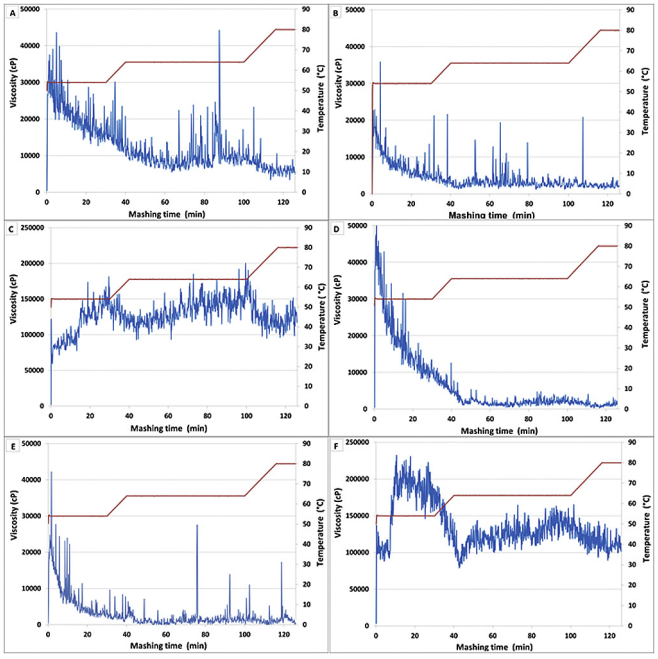

Fig. 1. The effect of enzyme Ondea® Pro or Cellic® CTec2 on rheological behaviour of sorted (A, D), unsorted (B, E) and sorted-out (C, F) barley, respectively. The profiles represent one of two replicates that are virtually identical.