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Effect of ageing-induced changes in rice physicochemical properties on digestion behaviour following storage

Zhongkai Zhou [a, b, *], Xue Yang [a], Zhe Su [a], Dandan Bu [a]

[a] Key Laboratory of Food Nutrition and Safety, Ministry of Education, Tianjin University of Science and Technology, Tianjin 300457, China
[b] ARC Functional Grains Centre, Charles Sturt University, Wagga Wagga, NSW 2678, Australia

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ABSTRACT

The changes in rice grains structure and digestion behaviours were investigated following storage at 4 °C and 37 °C, respectively. Pasting study indicated that rice samples stored at 37 °C demonstrated a consistent increase in the time to peak viscosity of the Rapid ViscoAnalysis parameters, implying a quick ageing progress. Compared to the rice stored at 4 °C, aged rice (stored at 37 °C) showed a coarser morphology after cooking by SEM, suggesting a limited starch gelatinization. Consistently, ageing process led to a decrease in the leaching of starch molecules in cooking residual water, which further confirmed that starch granules in aged rice grain caused less hydration and swelling. The analysis of the amount of cell wall remnants showed that rice stored at 37 °C caused a significant increase in the amount of cell wall remnants along the storage at 37 °C, which might suggest that the cell wall structure of the rice grains became more lignified because of the ageing. Furthermore, the ageing process significantly reduced rice digestion kinetics both in rate and extent. Thus, it is assumed that ageing process leads to the cell walls becoming more strengthened and lignified, which makes the rice grain more organized in its structure, and subsequently reduces starch granules disruption and molecules leaching during the cooking. Therefore, this study suggests that the changes in digestion behaviours of rice are highly associated with the changes in rice physical and chemical properties occurred during storage and the ageing process might be another option for manipulating rice digestion properties.

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1. Introduction

Rice is mostly consumed by humans as cooked rice and only a small portion of rice is used to prepare ingredients for processed foods. This pattern of utilization leads to the requirement to store rice over varying periods. It has been confirmed that rice chemical and physical properties changed during storage, and this process is usually termed rice ageing. Rice ageing starts before harvest and continues afterwards (Perdon et al., 1997; Zhou et al., 2002). The previous studies found that ageing-induced changes occurred in rice composition, pasting properties, thermal properties and texture (Chrastil and Zarins, 1992; Noomhorm et al., 1997; Park et al., 2012; Perdon et al., 1999; Sowbhagya and Bhattacharya, 2001; Suzuki et al., 1999; Zhou et al., 2007, 2010). Attempts to explain the changes in rice functionality associated with the ageing have focused on the properties of rice components, such as starch, protein, and lipids during storage (Chrastil and Zarins, 1992; Limpisut and Jindal, 2002; Matsukura et al., 2000). As with functionality, changes in those components were most apparent at an elevated storage temperature (Zhou et al., 2010). For example, Chrastil and Zarins (1992) studied the changes in the properties of rice protein during storage and found that the number of disulfide bridges increased and the lower molecular weight peptides decreased with increase of the higher molecular weight peptides following the storage. Changes in fatty acid profiles and an increase of free fatty acids during storage have been noted as well.

Recently, the interactions between rice components during storage have attracted more interest (Martin and Fitzgerald, 2002; Teo et al., 2000). Previous investigation suggests that interactions between starch and non-starch components play important roles on rice properties during the ageing process (Zhou et al., 2010). Nevertheless, the effect of aging process on rice property is complex and the mechanism of rice ageing is still being examined. More recently, there have been considerable interests in the potential for improving diabetic control by altering the glycemic impact of the

* Corresponding author. School of Food Engineering and Biotechnology, Tianjin University of Science and Technology, Tianjin 300457, China.
E-mail address: zkzhou@tust.edu.cn (Z. Zhou).

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carbohydrates ingested. The nutritional advantages could be anticipated with foods of low glycaemic index (GI) and this type of foods is usually related to a lower postprandial response of glucose and insulin (Schwingshack and Hoffmann, 2013; Youn et al., 2012). In recent years, a considerable number of studies have focused on the importance of foods containing resistant starch as a substrate for colonic fermentation (Zhou et al., 2013). These kinds of food products will provide substrate to the colonic microflora, thus promoting short-chain fatty acid production in the colon with potential health benefits (O'Callaghan et al., 2012). Our preliminary survey suggested that the people consuming aged rice would have a lower risk to have diabetes mellitus compared the community of consuming fresh rice (data not shown), but the reason is known yet. To the best of our knowledge, the information regarding the effect of ageing process on rice digestion behaviour is very rare. Thus, this study extends our previous work on the effect of storage on rice thermal and cooking properties (Zhou et al., 2007, 2010) and examines the effect of ageing process on rice digestion property.

2. Materials and methods

2.1. Rice samples

Commercially available rice samples of three Australian cultivars, Koshihikari, Keeyma and Doongara were selected in this study. Koshihikari is a medium rice grain with 18% amylose content; Keeyma, an aromatic rice grain with 26% amylose content; and Doongara, a long rice grain having 28% amylose content. The three varieties were grown in the Murrumbidgee Irrigation Area of New South Wales, Australia.

2.2. Rice storage

Previous studies suggest that storage at 37 °C could accelerate the ageing process, whereas storage at 4 °C retards the process. Thus, the rice sample stored at the two different temperatures provides a valid comparison of rice ageing (Meullenet et al., 1999; Rajendra and Zakiuddin, 1991; Zhou et al., 2007, 2010). Therefore, in this study, two storage temperatures, 4 °C and 37 °C were selected for investigating the changes in rice property following storage.

White rice samples (3 kg) were placed in air-tight glass bottles, sealed and stored in the dark at 4 °C and 37 °C in thermostatically controlled incubators. Samples were withdrawn from the same bottle at the beginning of storage and at different intervals. Prior to analysis, rice grains were ground immediately after being withdrawn from the storage containers using a Cyclone Sample Mill (UDY Corporation, Fort Collins, CO) and passed through a 0.5 mm sieve screen.

2.3. Cell wall remnants isolation by amylases digestion

After ground, rice flour (3.0 g; 12% moisture) was mixed with 25 mL sodium acetate buffer (0.005 M, pH 6.5) and then 50 μL of thermostable α-amylase (Source: *Bacillus licheniformis*) was added. The mixture was incubated in a boiling water bath for 4 min to hydrolyse starchand quietly cooled in an ice bath. The hydrolysate was centrifuged at 2095 × *g* for 10 min and the supernatant was discarded. The residue was washed using 50 mL distilled water, stirred and centrifuged. The washing procedure was repeated twice. The residue was combined with 20 mL of sodium acetate – acetic acid buffer (0.005 M, pH 5.0) and then 50 μL glucoamylase and 20 μL isoamylase were added. The mixture was incubated at 50 °C for 20 h.

After incubation, the mixture was centrifuged at 2095 × *g* for 10 min and the residue was collected and washed using 50 mL distilled water twice. The collected residue was then washed twice with ethanol followed by acetone. The residue was dried to constant weight in an oven at 105 °C and the cell wall remnant was calculated and expressed as per cent of dry rice grain.

2.4. Pasting analysis by Rapid ViscoAnalysis (RVA)

The pasting properties of the rice samples were studied using a RVA (Newport Scientific, Warriewood, NSW, Australia). In brief, rice flour (2.8 g; 12% moisture) was added into 25 mL distilled water. The temperature profile involved an initial 10 s high-speed (960 rpm) stir that dispersed the sample prior to the beginning of the measuring phase at 160 rpm. The temperature was held at 50 °C for 1 min and then increased to 95 °C in 3.75 min, held for 2.5 min, cooled to 50 °C in 3.75 min, and held for 5 min.

Moreover, an "extending temperature profile" was also designed for further investigating the pasting properties of the rice samples.

2.5. Size-exclusion high performance liquid chromatography (SE-HPLC)

After the gelatinization of rice flour using a RVA under a normal heating profile (described above), the paste in the RVA canister was instantly collected. 0.5 g of the rice pasting was mixed with 0.5 mL sodium acetate buffer (0.05 M, pH 5.0), vigorously vortexed and centrifuged at 10,000 rpm for 10 min. The superannuant was used for measuring soluble starch molecular profile using a SE-HPLC system. The system includes a Waters 2690 pump equipped with an auto sampler and a differential refractive index detector (Waters, Model 410, Milford, MA). An Ultrahydrogel™ 250 column (Waters, 7.8 mm × 300 mm), a guard column (Phenomenex Inc., Australia) and the detector were maintained at 37 °C and injection was at 25 °C. Sodium acetate buffer (0.05 M; pH 5.0) containing 0.02% sodium azide was used as mobile phase at a flow rate of 0.4 mL min^{-1}.

2.6. Rice grain cooking

White rice grain (2.5 g, dry basis) was mixed with 25 mL RO water in a quick-fit conical flask fitted with a glass stopper. The flask was immersed in a boiling water bath and cooked for 15 min. The flask was quickly cooked down in an ice bath for 5 min. The cooked rice was freeze dried, and was visualized using a scanning electron microscopy (SEM).

2.7. Rice grain digestion in vitro

The digestion procedure of the cooked rice was described as followings: 0.5 g of the cooked rice (on dried basis) was added into 25 mL of pH 5.2 NaAc buffer (0.1 mol L^{-1}). Prior to the digestion, 0.55 ml $CaCl_2$ solution (0.1 mol L^{-1}) was added into the mixture. The suspension was equilibrated at 37 °C for 1 h with magnetic stirring, and then 0.15 mL of enzyme solution containing 2.3U of porcine pancreatin α-amylase and 24 U of amyloglucosidase was added. The digestion was carried out at 37 °C with magnetic stirring, and 0.3 mL aliquots of hydrolysed solution were withdrawn at different time intervals. The aliquots were immediately put in a boiling water bath for 10 min to deactivate the enzymes. The glucose content in each digestion slurry was determined using the Megazyme glucose assay kit (GOPOD method, manufacture instruction). Analysis was performed in triplicates.

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