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Effects of L-Cysteine on some characteristics of wheat starch

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ABSTRACT

In this study the effects of L-Cysteine as a food additive on wheat starch characteristics before and after gelatinization were studied. L-Cysteine (63 mg/kg, starch basis) was added to slurry of wheat starch in water (30%, w/w). One set of samples was prepared by mixing it at 40 °C for 45 min. Another set was gelatinized at 100 °C for 45 min. The scanning electro-micrographs of the samples prepared at 40 °C in the presence of L-Cysteine showed some spots on the granules. However, thermal properties, X-ray patterns and the degree of crystallinity of the samples did not obviously change (P > 0.05); while a reduction in intrinsic viscosity, peak and final viscosities of the samples was observed. After gelatinization, intrinsic, peak and final viscosities of the samples were reduced. Some of these changes may indicate degradation of starch molecules in the presence of L-Cysteine, particularly after gelatinization.

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1. Introduction

The most important storage carbohydrate produced in plants is starch which is deposited in the form of granules in storage tissues. Starch granules have semi-crystalline structure and vary in shape and size depending on the starch origin (Copeland, Blazek, Salman, & Tang, 2009). The interior structure of starch granules and starch constituents; amylose and amylopectin, have received much attention (see e.g. Copeland et al., 2009). The external structure of starch granules is of particular significance since it is at the interface between the granules and its surroundings. Some components such as proteins and lipids are present on the surface of starch granules. The proteins on the surface of the granules are referred to as starch granules associated proteins and may be associated with lipids on the granule surface. In wheat, these proteins known as "friabilins" are of particular interest since they have connections with grain hardness (Baldwin, 2001).

Using electron microscopy it has been shown that the surface of the starch granules had some pores which are the openings to the interior of the granules (Buléon, Colonna, Planchot, & Ball, 1998; Fannon, Shull, & BeMiller, 1993). The structural characteristics of the surface of the starch granules (crystallinity, absorbed and non-starch materials and porosity) were suggested to be responsible for the variation in starch granules susceptibility to enzymes and chemicals (Copeland et al., 2009; Majzoobi, Radi, Farahnaky, Jamalian, & Tongdang, 2009).

Starch is a major component of many foods and provides 70-80% of daily calories consumed by human. It also has a great influence on the physicochemical properties of food products by acting mainly as thickening and gelling agents (Copeland et al., 2009). Different parameters affect starch functionality in a food system. For instance, intrinsic differences between starches obtained from different sources including starch crystallinity, molecular weight of either amylose or amylopectin, degree of polymerization of starch molecules, chain length of amylopectin side chains and the ratio of amylose to amylopectin as well as plant growing conditions have great impacts on starch characteristics and its role in food products (Singh, Singh, Kaur, Sodhi, & Gill, 2003; Srichuwong, Cadra Sunarti, Mishima, Isono, & Hisamatsu, 2005; Srichuwong et al., 2005). On the other hand, the properties of starch can be influenced by other components in a food system. For example, the presence of lipids such as fatty acids, lysophospholipids and monoacylglycerols can result in the formation of amylose-lipid complex which can alter starch gelatinization temperature, type and degree of starch crystallinity (Copeland et al., 2009; Kaur & Singh, 2000). Also, hydrocolloids such as proteins and non-starch polysaccharides can interact with starch molecules and hence alter starch properties (Ferrero, Martino, & Zaritzky, 1996; Funami, Kataoka, Omoto, Goto, & Asai, 2005; Kohyama & Nishinari, 2006; Ozcan & Jackson, 2002).

Food additives may have some effects on starch characteristics. For instance, gaseous chlorine as a bleaching agent for cake flour can oxidize starch molecules and therefore change their characteristics (Pomeranz, 1984). Ascorbic acid has been recognized as a highly reactive agent, which can be easily oxidized and is capable of decomposing polysaccharides. Its degradation effects on cassava starch (Sriburi, Hill, & Mitchell, 1999) and cereals β-glucans

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(Kivela, Gates, & Sontag-Strohm, 2009) have been reported. Valler-Pamise, Barclay, Hill, Mitchell, Paterson, & Blanshard, (1997) and Paterson, Mitchell, Hill, and Blanshard (1996) showed that addition of ascorbic acid to starches caused changes in their pasting properties (Valler-Pamise et al., 1997). The effects of other food additives such as enzymes (e.g. α -amylase) (Pomeranz, 1984), sour food seasonings (e.g. acetic, lactic, malic, tartaric and ascorbic acids) (Hirashima, Takahashi, & Nishinari, 2005), salt (Farahnaky, Farhat, Mitchell, & Hill, 2009) and sucrose (Carvalho & Mitchell, 2001) on the physicochemical properties of starch have been studied. The findings of these studies indicate that food additives even at low concentrations may have some effects on the starch structure and functionality.

L-Cysteine (L-Cys), a non-essential amino acid, is an accepted food additive which is added to bread flour (up to 90 mg/kg, wheat flour basis) to improve dough and bread quality (Pomeranz, 1984). It may also be present naturally or added to many other food products. Therefore, understanding the effects of this chemical on starch properties is of great importance. It has been well established that L-Cys can interact with dough proteins (gluten) by splitting the disulfide bonds. Accordingly, it reduces the mixing time of the dough and aids faster dough development (Pomeranz, 1984; Paterson et al., 1996; Angioloni & Rosa, 2007; Elkhalifa & El-Tinay, 2002). However, the possible effects of L-Cys on starch granules, as the main component of flour, have received less attention in the literature. Previous studies showed that addition of sulfites as a reducing agent can change the pasting properties of starch (Valler-Pamise et al., 1997). Accordingly, the addition of L-Cys (a reducing agent) might have similar effects on starch molecules.

The main objective of this study was to investigate the effects of L-Cys on some physiochemical properties of wheat starch before and after gelatinization. The results may be helpful to explain the changes in functional properties of the food systems such as bakery products containing starch and L-Cys.

2. Materials and methods

2.1. Materials

Pure wheat starch was purchased from Fars-Glucosin, Co. Shiraz, Iran. Other chemicals including L-Cys and KOH were of analytical grade and were obtained from Merck Co., Germany.

2.2. Methods

Slurries of wheat starch (30 g) were made with distilled water (100 g) containing L-Cys (63 mg/kg, starch basis). The first set of samples were mixed well with a magnetic stirrer for 8 min at 40 °C (under these conditions the starch was not gelatinized). Then the samples were poured into large metallic trays with thickness of 2–3 mm and dried in a cabinet drier at 50 °C for 2 h. The dried starch was hammer–milled and sieved to obtain particle sizes in the range of 120–200 μm . These samples were designated as st + Cys. A control sample (C) was prepared under the same conditions without L-Cys.

To prepare the second set of samples, first the samples were mixed and heated in the same manner as explained previously for st + Cys samples. However, these samples were further heated in a boiling water bath (at 98 °C) for 40 min. Observation of the sample under the light microscope showed no intact starch granules at this stage indicating full gelatinization of wheat starch. A thin layer (2–3 mm) of each sample was spread in individual metallic trays and dried in a cabinet drier at 50 °C for 2 h. After drying, the samples were hammer–milled and sieved to obtain particle sizes in the range of 120–200 μm . These samples were named

as gel st + Cys. A control was prepared under the same conditions without L-Cys and named gel C.

2.3. Determination of the moisture content of the samples

The moisture content of the samples was assessed by drying 5 g of the samples in an oven at 105 $^{\circ}$ C until a constant weight was obtained, which was 5.0 \pm 0.1% dry weight basis.

2.4. Microscopic structure of starch

To study the microscopic structure of the starch samples, a scanning electron microscope (SEM) (Model 5526, Cambridge, UK) was used. To prepare the samples, a tiny amount of each sample was sputter coated with gold/palladium. Finally the samples were transferred to the microscope where they were observed at 20 kV.

2.5. Intrinsic viscosity

To determine the intrinsic viscosity of the samples, first solutions of 6 mg/ml starch in 1 M KOH were prepared according to Anastasiades, Thanou, Loulis, Stapatoris, and Karapantsios (2002). Since it is crucial to obtain a fully solubilized sample before being used in this experiment, the solubility of each sample in 1 M KOH was checked by light transmission method according to Lawal, Adebowale, Ogunsanwo, Barba, and Ilo (2005). Therefore, light transmittances of the solutions were measured at 640 nm against the blank (1 M KOH). Then a U-tube viscometer at (20 ± 0.1) °C was used to measure the relative, reduced and intrinsic viscosities of the samples using the Eqs. (1)–(3), respectively (Harding, 1997).

$$\eta_{rel} = \frac{\eta}{\eta_0} = \left(\frac{t}{t_0}\right) \cdot \left(\frac{\rho}{\rho_0}\right) \tag{1}$$

where η_{rel} is relative viscosity, t and t_0 are the times required for the sample and solvent (KOH 1 M) to pass through the U-tube, respectively. ρ and ρ_0 are the densities of sample solution and solvent, respectively.

Then reduced viscosity, η_{red} , was determined according to the Eq. (2):

$$\eta_{red} = \frac{\eta_{rel} - 1}{C} \tag{2}$$

where *C* is the concentration of the sample.

Then intrinsic viscosity; $[\eta]$, was determined using the Eq. (3):

$$[\eta] = \lim_{\epsilon \to 0}^{\eta_{red}} \tag{3}$$

2.6. Wide angle X-ray diffraction

To determine the X-ray diffraction pattern of the starch samples, approximately 2 g of each powder equilibrated at relative humidity of 75% were pressed into a sample holder. Diffractograms were recorded using an X-ray diffractometer (Model D8 Advance, Germany). The scattered X-ray radiation was recorded by a proportional moving detector over a 4–38° (20) angular range with an angular velocity of 0.05/s. The degree of starch crystallinity was determined using the Eq. (4). The area was calculated using the software developed and supplied by the instrument manufacturer (EVA, Version 9.0).

Degree of crystallinity(%) =
$$\left(\frac{\textit{Area of the peaks}}{\textit{Total curve area}}\right) \times 100$$
 (4)

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