



The influence of different sugars on corn starch gelatinization process with digital image analysis method



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ABSTRACT

The digital image analysis, integral optical density (IOD) method, combined with “the model of response difference of crystallite change (MRDCC)” was applied to dynamically analyze the influence of different sugars on the gelatinization of corn starch. All of the saccharides (ribose, fructose, glucose, sucrose, lactose, trehalose, maltose, raffinose and stachyose) proposed in our research showed protection effect on starch crystalline structure during gelatinization. The protection effect increased with the increase of sucrose concentration (0–20%). Trisaccharide and tetrasaccharide were more effective in inhibiting the gelatinization process than disaccharide; and the protection effect of disaccharides on starch was bigger than that of monosaccharide during gelatinization. The gelatinization inhibition effect had good relationship with n_{DHN} (dynamic hydration number), and the increase of the equatorial OH ($e-(OH)$) group number of saccharides might increase the inhibition effect on starch gelatinization. However, in addition to the $e-(OH)$ groups, the combination ability of sugar with water molecules might be also related to the size of the sugar molecules and their three-dimensional structure. We believed that owing to the helical structure which formed through hydrogen bonds, tetrasaccharide tended to decrease the hydration ability of saccharide and destabilized the water structure, thus the inhibitory effect of stachyose was almost the same as raffinose.

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1. Introduction

Starch is a kind of abundant renewable resources from plants. Within the category of starch, corn starch, which has a long history and unique advantages, is very representative. It is a typical A type starch and in its polymorphs, the double helices are closely packed together with a small amount of structural water (Bogacheva, Meares, & Hedley, 2006). When raw starch granules are heated in water, their semicrystalline nature is gradually eliminated, resulting in structural breakdown and starch polymer dispersion in solution. This heat-induced starch granule breakdown or the order to disorder phase transition is known as gelatinization (Jenkins & Donald, 1998; Ratnayake & Jackson, 2008).

Water plays an important role in the process of thermal starch gelatinization as the gelatinization temperature decreases with increasing water content of starch suspensions. Additionally the presence of alkali, salts, sugars, lipids, alcohol, organic acids and their salts etc. also influence starch gelatinization by rupturing

hydrogen bonds within the starch granule, or by forming soluble complexes with starch. They have an impact on the gelatinization temperature and thus affect the extent of gelatinization (Zobel, 1984).

Sugars have been shown to have a significant effect on the gelatinization and rheological properties of starches generally, and it has been found that they can change the gelatinization temperature (T_p) (Gonera & Cornillon, 2002; Wootton & Bamunuarachchi, 1980), gelatinization enthalpy change (ΔH) (Wootton & Bamunuarachchi, 1980) and similarly they might increase or decrease the rate and degree of gelatinization (Rumpold & Knorr, 2005) and retrogradation (Hoover & Senanayake, 1996). The effect of sugars on starch gelatinization has been studied by many researchers using a wide range of techniques. For example, differential scanning calorimetry (DSC) (Evans & Haisman, 1982; Hoover & Senanayake, 1996; Kohyama & Nishinari, 1991; Perry & Donald, 2002; Prokopowich & Biliaderis, 1995), rheological measurements (Ahmed, 2012; Prokopowich & Biliaderis, 1995; Sopade, Halley, & Junming, 2004), light microscopy (Gonera & Cornillon, 2002; Rumpold & Knorr, 2005), X-ray diffraction (XRD) (Hoover & Senanayake, 1996; Perry & Donald, 2002), nuclear magnetic resonance (NMR) (Le Botlan & Desbois, 1995; Rumpold & Knorr, 2005),

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viscometry with a Rapid Visco Analyzer (RVA) (Torley & Van der Molen, 2005) etc. With different parameters, these methods can characterize the effect of sugars on starch gelatinization properties from different perspectives.

Gelatinization has been shown to be a kinetically controlled process (Slade & Levine, 1988). Therefore, regardless of how gelatinization is monitored, any quantitative description of the process will be affected by changes in conditions such as the heating rate and extent of agitation. Some properties (viscosity, heat uptake, and loss of birefringence) may be monitored continuously during the process, while others (e.g. volume) are monitored more conveniently after interrupting the process (Leach, McCowen, & Schoch, 1959). However, this interruption, and the followed recovery of the experiment conditions confound the meaning of the results measured by these techniques (Ziegler, Thompson, & Casanovas, 1993). In order to make the information of starch gelatinization more accurately, the real-time detection instruments should be used to reduce various pre- and post-processing effects on starch structure.

Based on polarizing microscopy, the IOD method was of advantage compared with the previous traditional method, because it is able to characterize the starch granules which stay in the partially gelatinized stage. At the same time, it is a method of real-time monitoring that dispenses with the pre- and after-treatment of various samples and its resulting date can reflect the real situation of the gelatinization process. Our previous works (Li, Xie, Yu, & Gao, 2013, 2014) have confirmed that IOD method has the following advantages: 1) IOD value is an integral function that is related to the area and the OD (optical density) value, the area is corresponding to the number of starch crystalline structure, and the OD is proportional to its intensity, so it is more accurate compared with traditional methods such as counting the particle number and calculating polarization area, as a result; 2) By repetitively measuring the optical density of the same digital image, the error result is less than 0.1%, which indicates that the systematic errors can be controlled in a small range with robustness; 3) good reproducibility. The parallel experiments show that the measurement error of a sample is less than 5%; 4) more than 1000 starch granules can be observed in a digital image and higher density of microscope observation can be achieved, which lead to the reduction of experiment workload and the raise statistical significance. The model of response difference of crystallite change (MRDCC) (Li et al., 2013, 2014) is a new characterization of the crystallization change degree in the starch gelatinization process. It characterizes that the starch gelatinization speed changes with the temperature. MRDCC is more sensitive and accurate compared with DSC, even the subtle expansion in the pre-gelatinized stage could be detected.

The water structure surrounding starch chains is stabilized by saccharides in starch–water–saccharide systems (Katsuta, Nishimura, & Miura, 1992a). If water around starch chain is stabilized by saccharides, the motion of the starch chains will be affected. Hence the gelatinization process of starch is controlled by saccharides. As a result, the stabilization degree of saccharides on water structure increases with the increase of e–(OH) group numbers. The saccharide molecule which has a large number of e–(OH) groups possesses a stronger stabilizing effect on water structure (Uedaira & Uedaira, 1985).

Although a major consequence of sugars on the gelatinization of starch granules has been described by many theories, a universally accepted explanation that caters for all the measurable changes is still not available. The objective of this work, therefore, has been to undertake a comprehensive study of the effect of different sugars on the gelatinization of corn starch, by combining information from hot stage microscopy with image analysis of starch granules. Meanwhile, a new method, integral optical density (IOD) method and MRDCC were employed in this work.

2. Materials and method

2.1. Materials

Corn starch (food grade) was purchased from Guangdong Pengjin Industrial Ltd. (Guangdong, China), starch composition was: corn starch 84%, water 12.35%, protein 0.3%, ash 0.3%, lipid 0.87% and phosphate 0.02% which were measured in our laboratory. Ribose (D-ribose) was obtained from Sigma Chemical Co. (St. Louis, MO, USA); Sucrose, glucose (D-glucose), fructose (D-fructose), lactose, trehalose (α-trehalose), and maltose were purchased from Shanghai Boao biological technology Co., Ltd. (Shanghai, China). Raffinose and stachyose were obtained from Hefei bomei biotechnology Co., Ltd. (Hefei, China). All sugars were of analytical grade except stachyose with a content of 85% and the main impurity was raffinose.

2.2. Preparation of sample

Corn starch slurries were prepared at starch concentrations of 10% (starch:distilled water = 1:9, dry basis). Starch slurries with 5%, 10%, 15%, 20% (w/w) of sucrose and 10% (w/w) of glucose, ribose, fructose, lactose, trehalose, maltose, raffinose, and stachyose were prepared separately to evaluate the effects of sugar type and concentration on gelatinization properties. The measured starch-sugar slurries were equilibrated for 2 h and then sealed between two glass cover slip using Dow Corning 732 Sealant before replaced in the hot stage (model THMS600, Linkam Scientific Instruments Ltd., Britain). Each measurement was carried out in triplicate.

2.3. Hot stage-light microscopy

Each specimen in the hot stage was observed under a polarization microscope (Vanox BHS-2, Olympus Corp., Japan) equipped with a digital camera, which can display live video of birefringence granules in a real time. A temperature programmer was connected with the hot stage to control the heating progress from 40 °C to 80 °C at a rate of 2 °C/min. Live pictures were captured every 5 °C when below 60 °C, while 2 °C above 60 °C. Fifteen digital pictures of each sample were used and each image (2048 × 1536, 12 bits) was saved as TIFF image file, without data compression. All of the samples were observed under the same aperture (maximum), light intensity (fixed at 9), and exposure time (40 ms). The combination of eyepiece and objective lens were selected with a magnification of 200 times, as described in our early research (Li et al., 2013).

2.4. IOD method

It is a new method to measure the degree of gelatinization (DG). The IOD value of each digital picture was calculated by the Image-pro plus 5.0 software (Li et al., 2013).

The DG based on the IOD value (DG_I) was calculated as defined in our early research (Li et al., 2013).

$$\text{Background correction} : C = A - B \quad (1)$$

$$DG_I\% = (1 - C/C_0) \times 100\% \quad (2)$$

where A is the original IOD value (IOD value calculated from the original digital image when all of the birefringence remain unchanged), B is the background IOD value (IOD value calculated from the original digital image when all of the birefringence disappeared), and C_0 is the initial real IOD value (IOD value of birefringence light derived from the specific crystal structure of starch

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