



## Molecular structure and digestibility of banana flour and starch



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### ABSTRACT

In this study, the *in vitro* digestibility of flour and starch from green *Musa Dwarf Red banana* (MDR), *Musa ABB Pisang Awak* (MPA) and *Musa AAA Cavendish* (MC) were examined. The results showed that raw MC flour and MPA starch had the lowest digestion rates, whereas cooked MDR flour and starch had the highest resistant starch contents and cooked MPA starch had the highest slowly digestible starch contents. It was explored that many factors affected the digestibility, including chemical composition, granule morphologies, and molecular structures. The pectin in banana flour could significantly affect the *in vitro* digestibility of flour. Scanning electron micrographs showed that granules had a flat, oval shape. X-ray diffraction patterns revealed that flour and starch of MDR exhibited the C<sub>A</sub>-type, whereas those of the other bananas exhibited the C<sub>B</sub>-type. MPA flour and starch had the highest amylose contents. MC starch had greater proportions of long chains, whereas MPA starch had the highest proportion of short chains. The molecular weight and radius of gyration of MDR starch were the lowest.

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### 1. Introduction

Bananas (*Musa paradisiac*), belonging to *Musaceae*, *Musa*, are perennial herbs that are widely distributed in tropical and subtropical regions (Pelissari, Andrade-Mahecha, Sobral, & Menegalli, 2012). Banana plantations, which are low-investment, high-efficiency, and rapid-income, have been developing rapidly and have become an essential industry in the rural economy of southern China. The area of banana orchards was up to  $3.92 \times 10^5$  ha and the annual production was up to  $1.18 \times 10^7$  tons in 2014. China has been the second largest producer inferior to India (FAO, 2014). Bananas are rich in carbohydrates, vitamin B, vitamin C, potassium, magnesium, and other mineral elements (Vatanasuchart, Niyomwit, & Wongkrjang, 2012). However, bananas are a type of respiration climacteric fruit and are perishable, leading to serious economic

losses during the concentrated maturation period. In addition, 10–15% of all harvested bananas cannot be sold as commercial bananas but can be used as raw materials for extraction of resistant starch (RS) and other functional ingredients. Therefore, it is necessary to make full use of green banana resources (Zhang et al., 2014). Starch is the main component of green bananas, accounting for more than 60% of the dry weight of the fruit (Zhang, Whistler, BeMiller, & Hamaker, 2005). Importantly, starch from green bananas provides resistance to digestion because it cannot be digested in the small intestine, similarly to dietary fibers, which are fermented by gut bacteria found in the large intestine (Englyst, Kingman, & Cummings, 1992).

Several intrinsic and extrinsic factors affect starch digestion. For example, modifications of starch and alteration in methods for processing and storage of foods can affect the digestibility of starch. In contrast, intrinsic factors include the starch granule structure and interactions between the granule and other components, such as the starch source, starch granule size, crystalline structure, native  $\alpha$ -amylase inhibitors, non-starch polysaccharides (NSPs), and amylose-lipid complexes (Zhang, Hong, Gu, & Wang, 2012; Zhao & Gu, 2007). Fibers, the main components of NSPs, can form a physical barrier to limit access of the enzymes to starch (Snow &

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O'Dea, 1981). Amylose-lipid complexes have been shown to reduce the susceptibility to  $\alpha$ -amylase (Holm et al., 1983). Guraya, James, and Champagne (2001) demonstrated that starch digestibility was directly related to granule size; larger granules exhibit lower digestibility. Additionally, cereal starch exhibit an A-type structure, whereas tubers, fruits, and high-amylose corn starch exhibit B-type structures, as distinguished by X-ray diffraction patterns. A-type starch are generally more easily digested than B-type starch, and legume starch show a C-type structure, having an intermediate digestion rate between A-type and B-type starch (Biliaderis, 1991; Zhao & Gu, 2007). Starch with higher crystallinity also shows lower digestion rates (Gu, 2009). In addition, the fine structure of amylopectin is closely related to digestion, and the high content of long-chain amylopectin is associated with lower degradation rates (Srichuwong, Sunarti, Mishima, Isono, & Hisamatsu, 2005).

The physicochemical characteristics of banana starch have been extensively studied. Zhao, Bao, and Yang (2006) studied starch from three varieties of bananas and confirmed that the shapes and sizes of banana starch granules were irregular. The crystalline structure resembled C-type starch, and most granules had fine, dense striations. Additionally, in a study on the digestion of bananas carbohydrates in the human small intestine, the content of indigestible starch was shown to be eight times higher than the NSPs and was closely related to the maturity of bananas (Englyst & Cummings, 1986). Several researchers have reported that the indigestible fraction contents of bread (Juarez-Garcia, Agama-Acevedo, Sáyago-Ayerdi, Rodríguez-Ambríz, & Bello-Pérez, 2006), cookies (Agama-Acevedo, Islas-Hernández, Pacheco-Vargas, Osorio-Díaz, & Bello-Pérez, 2012), and pasta (Ovando-Martínez, Sáyago-Ayerdi, Agama-Acevedo, Goñi, & Bello-Pérez, 2009), increased significantly when prepared with unripe banana flour.

Bananas are typically marketed fresh; however, post-harvest losses are often very serious during the storage period. Approximately 10–15% of bananas do not have commercial value, resulting in major wasting of resources. The above factors cause losses of income and are not beneficial for development of the banana industry. Thus, making full use of the abundant starch resources of green bananas would be helpful for improving the added value, extending the industry chain, and enhancing farmers' interests. Additionally, with the rapid development of the global economy, consumers have become more health conscious. The high content of RS in green bananas can help alleviate constipation, control diabetes and obesity, and prevent colon cancer. Therefore, studies of the digestive performance, molecular structure of banana flour and starch are helpful for producing healthy foods and conforming to the developing trends of people's health needs. Currently, domestic and foreign researches have mainly focused on the preparation, physicochemical properties, digestibility, and health effects of banana flour and starch. However, systematic analysis of relationships among digestibility, chemical composition, granule morphology and molecular structure of banana flour and starch have not yet been performed.

Therefore, in this study, the *in vitro* digestibility of green *Musa Dwarf Red banana*, *Musa ABB Pisang Awak* and *Musa AAA Cavendish* flour and starch was examined. In addition, the chemical composition, granule morphologies, and molecular structures were evaluated to identify the factors affecting the digestion of banana flour and starch.

## 2. Materials and methods

### 2.1. Materials

Three varieties of banana flour, made from fresh green *Musa Dwarf Red banana Var. Guihongjiao No. 1* (MDR), *Musa ABB Pisang*

*Awak. Jinfen No. 1* (MPA) and *Musa AAA Cavendish Var. Williams B6* (MC) at ripening stage 1 (judged with a standardized color chart), were obtained from Guangxi Academy of Agricultural Science, China. Banana starch was isolated from the banana flour according to the method described by Jiang et al. (2015).

### 2.2. *In vitro* digestion properties of raw and cooked samples

The *in vitro* digestion properties of flour and starch samples were determined by the method of Englyst et al. (1992), with a few modifications. Briefly, samples (200 mg), guar gum (20 mg), and distilled water (2 mL) were added to 50-mL polypropylene centrifuge tubes and mixed fully using a magnetic stirrer. Each sample was left uncooked or cooked in a boiling water bath for 10 min. The tubes were then placed in a water bath at 37 °C with the addition of 4 mL pepsin ( $\geq 250$  U/mg solids, P7000, EC 3.4.23.1; Sigma, St. Louis, MO, USA)/hydrochloric acid solution (5 mg/mL) and allowed to hydrolyze for 30 min with shaking. Thereafter, six glass beads (diameter: 3–4 mm) and 2 mL sodium acetate buffer (0.5 M, pH 5.2) were added to the tubes, and the reaction was continued for 30 min. Starch digestion was initiated by adding 2 mL enzyme mixture (pancreatin extract [P7545, EC Number 232-468-9; Sigma], amyloglucosidase [ $\geq 300$  U/mL, A7095, EC 3.2.1.3; Sigma], and invertase [ $\geq 300$  U/mg solids, I4504, EC 3.2.1.26; Sigma]). The released glucose content at different times (0, 10, 20, 30, 45, 60, 90, and 120 min) was measured using a glucose oxidase-peroxidase kit (Leadman Biochemistry Co. Ltd, Beijing, China). Values for the digested starch fractions were expressed as mg glucose  $\times$  0.9. Values for rapidly digestible starch (RDS), slowly digestible starch (SDS), and RS were calculated from the measured G20 and G120 values (G20 and G120 are the glucose content hydrolyzed within 20 min and 120 min, respectively).

$$\%glucose = \frac{(A_t - A_b) \times c \times V \times D}{A_s \times w} \times 100$$

where  $A_t$  is the absorbance of the test solution at 520 nm,  $A_b$  is the absorbance of the blank solution at 520 nm,  $c$  is the concentration of the standard solution (in mg glucose/mL, offered by the glucose oxidase-peroxidase kit),  $V$  is the total volume of the test solution,  $D$  is a dilution factor,  $A_s$  is the absorbance of the standard solution at 520 nm, and  $w$  is the weight (in mg) of sample taken for analysis, which may be corrected for moisture.

$$RDS = (G20 - FG) \times 0.9$$

$$SDS = (G120 - G20) \times 0.9$$

$$RS = TS - RDS - SDS$$

where FG is the free glucose content and TS is the total starch content (% dry basis) of the sample.

### 2.3. Chemical composition

Moisture was determined by drying to a constant weight at 105 °C. Ash, protein, fat contents were analyzed according to AACC (1983) methods 08-01, 46-13, 30-25, respectively. The conversion factor used for protein determination was N g/100 g  $\times$  6.25. Insoluble dietary fiber was determined using neural detergent according to Van Soest, Robertson and Lewis (1991). Pectin was determined using a standard method in China (NY 2016-2011). Briefly, the pectin in the sample was precipitated with absolute ethanol and was hydrolyzed to galacturonic acid that could react with carbazole in the sulfuric acid and produce a magenta

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