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Characterization of Starch from Ginger Root (Zingiber officinale)*

By F. G. R. Reyes, B. L. d'Appolonia, C. F. Ciacco and M. W. Montgomery

Starch has been isolated from ginger root in 12.3% yield. The starch that contained 22.2% amylose had a density of 1.517 g/cm³ and an A type x-ray diffraction pattern. Solubility studies revealed low swelling power and solubility in water, and reduced solubility in dimethyl sulfoxide and in potassium hydroxide, suggesting homogeneous and strong bonding forces maintaining the granular matrix. Information from the Brabender amylogram indicated a relatively high initial pasting temperature (80°C) and resistance to mechanical shear upon gelatinization, resembling those starches modified by crosslinking.

Charakterisierung von Stärke aus Ingwerwurzel (Zingiber officinale). In 12,3% iger Ausbeute wurde Stärke aus Ingwerwurzeln isoliert. Die Stärke enthielt 22,2% Amylose, hatte eine Dichte von 1,517 g/cm³ und zeigte ein Röntgenbeugungsbild des A-Typs. Wie Löslichkeitsstudien ergaben, wies die Stärke eine geringe Quellfähigkeit und Löslichkeit in Wasser sowie eine reduzierte Löslichkeit in Dimethylsulfoxid und in Kaliumhydroxid auf, weshalb zu vermuten ist, daß homogene und starke Bindungskräfte die Kornmatrix zusammenhalten. Das Brabender-Amylogramm zeigte eine relativ hohe Anfangs-Verkleisterungstemperatur (80°C) und Widerstandsfähigkeit gegenüber mechanischen Scherkräften beim Verkleistern, wie es bei Stärken beobachtet wird, die durch Vernetzung modifiziert sind.

1 Introduction

Ginger (Zingiber officinale) is a spice valued for its characteristic aroma and pungency. The major pungent compounds of ginger are the gingerols, and the related dehydration products shogaols [1, 2]. Even though ginger root has been reported to have a high content (70-79%) of total carbohydrates [3] and a starch content between 40.4 to 59% [4] no previous studies have dealt with the characterization of this starch. The purpose of this study was to isolate and examine the physical and chemical properties of ginger root starch.

2 Materials and Methods

2.1 Source of Starches

Ginger roots were obtained commercially at a local supermarket, Corvallis, Oregon. Tapioca starch was isolated from tapioca flour, distributed by Giusto's Specialty Foods Inc., South San Francisco, CA., and purchased locally. Corn starch was obtained from Matheson, Coleman and Bell.

2.2 Starch Isolation

Ginger starch was isolated from the roots according to the procedure of *Badenhuizen* [5] using mercuric chloride as an amylase inhibitor.

2.3 Chemical Composition

Protein and ash content were determined according to AACC methods 46-11 and 08-01, respectively [6]. Fiber was determined by the acid detergent method of the

AOAC [7]. Fat was determined by extraction with methanol in a soxhlet for 24 h.

2.3.1 Amylose Content

The amylose content was determined by the iodine colorimetric method of McCready and Hassid [8]. A standard curve prepared with different amylose: amylopectin ratios, isolated from ginger starch [9] was used for this determination.

2.3.2 Fatty Acid Composition

The fatty acid composition of the methanol extracted lipids was determined by gas liquid chromatography (GLC), of the methyl ester derivatives [10].

2.3.3 Hydrolysis and Identification of the Hydrolysis Products of Starch

Starch (0.5g/50ml) was suspended in 0.5-N sulfuric acid solution and refluxed until a constant value for reducing power was obtained (8 h). The reducing power was determined by the *Nelson-Somogyi* micro colorimetric method [11]. The hydrolyzed product was neutralized with barium hydroxide, centrifuged, and the supernatant percolated through a column containing cationic exchange resin (Biorad AG 50W-X4, 200 – 400 mesh in the hydrogen form) and anion exchange resin (Biorad AG 1-X8, 200 – 400 mesh acetate form). The system was washed with deionized water. Aliquots were pipetted into 3 ml vials, and the samples were evaporated to dryness on a rotary evaporator (45 °C) and stored under vacuum over phosphorous pentoxide. The hydrolyzed products were silylated and analyzed by GLC according to the procedure of *Wrolstad* et al. [12].

2.4 Microscopic Examination of Starch Granules

The starch granules were examined microscopically with the granules suspended in water, 2% potassium hydroxide solution and anhydrous dimethylsulfoxide (DMSO). Photomicrographs of the starches were obtained using a magnification of $400 \times \text{under normal}$ and polarized light.

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Granule size was determined using a 7.5μ eyepiece lens calibrated against a micrometer.

Scanning electron microscopy was performed by placing the starch sample on a stub coated with gold using a diode sputter coater and photographed using a JEOL JSM-35 Scanning Electron Microscope (× 1000).

2.5 X-Ray Diffraction Pattern

X-Ray diffractometer traces were obtained using the following experimental conditions; CuKa radiation, voltage 35 K v, current 18 ma, scanning speed $1^{\circ}2\theta$ per inch of chart. Values of intensities were read from the curves over the angular range $10^{\circ}-29^{\circ}$ which includes most of the crystalline peaks.

2.6 Gelatinization Temperature Range

The gelatinization temperature range of the starch slurries (0.2%) was determined by microscopic observation under polarized light. Samples were observed after heating at different temperatures [13].

2.7 Swelling Power and Solubility

The method of *Leach* et al. [14] was used to study the swelling and solubility of ginger starch.

2.8 Solubility in Dimethylsulfoxide

A modification of the method of *Leach* and *Schoch* [15] was used to determine the solubility of starch in DMSO. Starch samples (100mg, dry basis) were dispersed in 20.0 ml anhydrous DMSO in 50 ml erlenmeyer flasks. The flasks were stoppered and shaken gently at 25 ± 0.01 °C so as to maintain the starch in suspension. After a specified period of time a flask was removed and the starch suspension centrifuged for $15 \, \text{min}$ at $2000 \times g$. An aliquot of the clear supernatant was removed, properly diluted with distilled water and the total carbohydrates measured by the phenol sulfuric acid method [16].

2.9 Starch Pasting Properties

Pasting Properties of ginger starch were investigated utilizing the Brabender amylograph with and without addition of carboxymethyl cellulose (CMC). Twenty grams of starch (dry basis) and 3.6 grams of CMC [17] or 40 g of starch (dry basis) in 450 ml of water were used. The starch slurry was heated at $1.5\,^{\circ}$ C/min in the amylograph bowl up to $95\,^{\circ}$, kept at this temperature for 15 min and then cooled to $50\,^{\circ}$ C.

2.10 Absolute Density

Absolute density was determined by the xylene displacement method of *Schoch* and *Leach* [18].

3 Results and Discussion

3.1 Chemical and Physicochemical Properties

The chemical composition of the isolated root starch is presented in Table 1. The isolated starch contained 99.57% carbohydrate, and no detectable fiber. GLC analysis of the trimethylsilyl derivatives of the starch hydrolyzate revealed the presence of only glucose. This result in conjunction with the low protein, ash and fiber content of the isolated starch indicated a starch with a high degree of purity. The amylose

content of ginger starch (22.2%) was similar to other root starches [19]. The comparatively high absolute density (1.517 g/cm³) of ginger starch indicated a compact starch granule [17, 19].

Table 1. Chemical Composition of Ginger Root Starch¹⁾.

Component	Composition (%)	
Protein ²⁾	0.18	
Fat	0.10	
Fiber	nil	
Ash	0.15	
Amylose	22.2	

¹⁾ Results expressed on a dry basis.

The fatty acid composition of the fat extracted from the ginger starch is presented in Table 2. Approximately 50% of the acids were saturated, with palmitic being the most abundant. Among the unsaturated fatty acids oleic and linoleic were present in the highest amounts.

Table 2. Fatty Acid Composition of Ginger Root Starch.

Fatty Acid	Composition (%)	
Lauric	7.8	
Myristic	5.0	
Pentadecanoic	Trace	
Palmitic	30.5	
Margaric	Trace	
Stearic	5.2	
Palmitoleic	4.1	
Oleic	16.1	
Linoleic	21.1	
Linolenic	3.2	
Arachidonic	6.1	

3.2 Starch Granules

Photomicrographs of starch granules under ordinary and polarized light are shown in Figure 1. Ginger starch granules had an eccentric hilum with no fissures being observed. Scanning electron microphotographs revealed disk shaped granules of almost uniform size and smooth surface. The average granule size was $24.4 \pm 8.7 \mu$ in length and $21.1 \pm 7.0 \mu$ in width.

3.3 X-Ray diffraction

X-Ray diffraction pattern of ginger starch is shown in Figure 2. Ginger starch showed diffraction patterns with Bragg reflection angle characteristics of the A pattern. The A pattern is the common pattern for ordinary cereal starches while root and tubers give C or B patterns [20].

3.4 Gelatinization Temperature Range

The gelatinization temperature of ginger starch ranged from 76° to 85°C, which is higher than values reported for other root starches [19, 21].

²⁾ $N \times 6.25$.

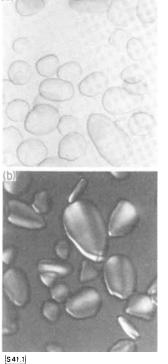




Figure 1. Photomicrographs of ginger root starch: (a) under ordinary light (about × 400); (b) under polarized light (about × 400); (c) scanning electron micrograph.

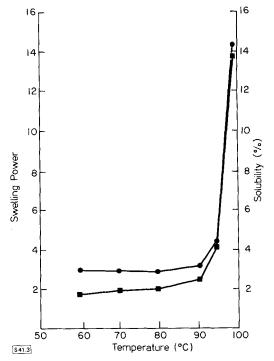


Figure 3. Swelling power $(\bullet - \bullet)$ and solubility $(\blacksquare - \blacksquare)$ of ginger root starch in water.

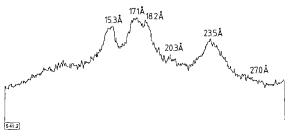
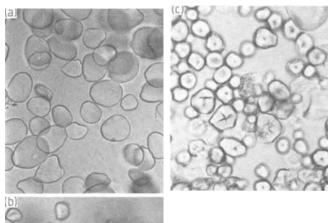


Figure 2. X-ray diffraction pattern of ginger root starch. Units are expressed as $2\,\theta$.



3.5 Swelling Power and Solubility

The swelling and solubility properties of ginger starch are illustrated in Figure 3. The ginger starch showed essentially no increase in swelling and solubility between 60° to 90°C, but a sharp increase above 90°C. The low swelling and solubility associated with a sharp increase after 90°C suggest the presence of homogeneous and strong bonding forces maintaining the granular matrix.

To ascertain the degree of association in the ginger starch granule, it was dispersed in a solution of 2% K OH and after 10 min the starch was examined under a microscope. Tapioca and corn starches were used for comparison purposes. Figure 4 illustrates that unlike tapioca and corn, ginger starch swells to a limited extent. This observation confirms the high degree of association among the molecules in the ginger starch granule.

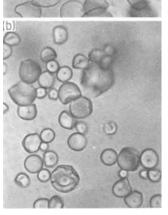


Figure 4. Photomicrographs of: (a) ginger root; (b) tapioca; and (c) corn starches suspended in 2% potassium hydroxide solution for 10 min.

3.6 Solubility in Dimethylsulfoxide

Solubilization rates of ginger, tapioca and corn starches in DMSO are presented in Figure 5. Initially corn and tapioca starches were solubilized at a faster rate than the ginger root starch. After 25 h of digestion the rate of solubilization of the corn starch decreased and it was the least soluble of the

starches after 50 h. The reduced solubility of ginger starch in DMSO may be due to poor solvent penetration because of the relatively homogeneous and strong bonding forces within the granule.

The low solubility of ginger starch, compared with tapioca and corn starches during the initial stages of incubation can be visualized in Figure 6.

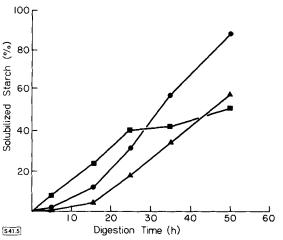
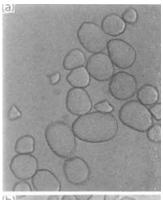
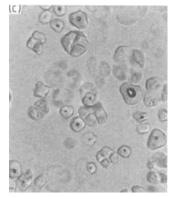


Figure 5. Solubility in dimethylsulfoxide of ginger root $(\triangle - \triangle - \triangle)$, tapioca $(\bullet - \bullet - \bullet)$ and corn $(\blacksquare - \blacksquare - \blacksquare)$ starches.





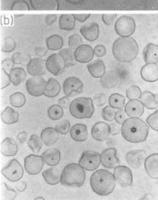


Figure 6. Photomicrographs of (a) ginger root; (b) tapioca and (c) corn starches after 50 min in dimethylsulfoxide.

3.7 Pasting Properties

Pasting characteristics of ginger starch are shown in Figure 7. Ginger starch showed a relatively high initial pasting temperature (80°C) compared to cereals or other root starches and a sharp increase in viscosity, once the initial pasting was reached, confirming the presence of homogeneous bonding forces. The single stage of gelatinization was also indicated with incorporation of CMC. No distinct peak was observed with or without CMC. During the isothermal holding period at 95°C the viscosity continued to increase indicating high resistance of the gelatinized ginger root to mechanical shear. The ginger starch amylogram without CMC showed considerable set back on cooling, reaching a viscosity of 1050 B. U. at 50°C.

The high initial pasting temperature in conjunction with the high absolute density (1.517 g/cm³) of ginger starch would support the theory of it having a compact granule.

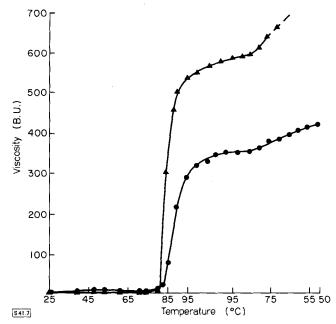


Figure 7. Brabender amylogram of ginger root starch at 4.4% (w/v) with $(\bullet - \bullet - \bullet)$ and at 8.8% (w/v) without $(\blacktriangle - \blacktriangle - \blacktriangle)$ incorporation of carboxymethyl cellulose.

4 Conclusions

Evidence presented suggests that ginger starch has a high degree of association between the starch components that maintain the granular matrix. These bonding forces between the starch components are not easily disrupted by alkali, DMSO or heat.

The relatively high initial pasting temperature of ginger starch associated with its resistance to mechanical shear upon gelatinization resembles those starches modified by crosslinking [22]. Since these modified starches have been used in the food industry [23] ginger root starch may also find similar food uses.

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Properties of Residual Starches of Sugary-2 and Sugary-2 Opaque-2 Maize Following Amylase Hydrolysis*

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Properties of residual starches of sugary-2 opaque-2 and sugary-2 maize starch granules hydrolyzed with glucoamylases were investigated. A crude and two crystalline glucoamylases were used. The amylopectin fractions of both starches hydrolyzed easier than that of amylose with all enzymes. Residual starches hydrolyzed by the crude glucoamylase accumulated low-molecular weight materials, which was not observed in residual starches attacked by crystalline glucoamylase. It was suggested that in the crude enzyme the contaminating α -amylase caused the accumulation of the minified fraction. It is also suggested that the crystalline region of sugary-2 opaque-2 starch may consist of a mixture of A-type and B-type patterns. Evidence for this was from observation of the changes in X-ray diffraction patterns of residual starch following amylase and acid hydrolysis.

Eigenschaften der nach Amylasehydrolyse verbleibenden Stärkereste von Zucker-2- und Zucker-2-opaque-2-Mais. Die Eigenschaften der mit Glucoseamylasen hydrolysierten Stärkeresten von Zucker-2-opaque-2- und Zucker-2-Maisstärkekörnern wurden untersucht. Dafür wurden eine rohe und zwei kristalline Glucoamylasen verwendet. Die Amylopektin-Fraktionen beider Stärken wurden durch alle Enzyme rascher hydrolysiert als die Amylosefraktionen. Die durch rohe Glucoamylase hydrolysierten Reststärken führten zu überwiegend niedrigmolekularem Material; dies wurde nicht beobachtet, wenn Stärkereste durch kristalline Glucoamylase angegriffen wurde. Es wird vermutet, daß die in dem Rohenzym als Verunreinigung enthaltene a-Amylase die Ansammlung der zerkleinerten Fraktion verursacht. Weiterhin wird angenommen, daß der kristalline Bereich der Zucker-2-opaque-2-Stärke aus einer Mischung des A- und des B-Typs besteht.

1 Introduction

Granular starches have various susceptibilities to amylase action according to their origin and pretreatments of the granules [1]. In maize (Zea mays L.) starches, endosperm genes affect starch-granule susceptibility to amylase action. Starch-granules of sugary-1 (su_1), sugary-2 (su_2) and waxy (wx) were more susceptible than those of normal, dull (du) starch was similar to normal in susceptibility and starch-granules of amylose-extender (ae) were lower than normal in

susceptibility to amylase hydrolysis [2-7]. Sandstedt et al. [2] suggested that the different starch-granule susceptibilities to amylase attack might lie in the structure of the starch-granules. However, properties of the residual starch-granules after enzymatic hydrolysis are not fully understood except for a few starches [8,9]. It is not known what changes actually occurred in the granules following different degrees of hydrolysis.

Exhaustive degradation of maize starch-granules by glucoamylase (a crude preparation of *Rhizopus amagasakiens*) showed that *ae* starches were more resistant to amylase attack than normal starches and were hydrolyzed very slowly after 80-85% degradation [9]. The residual starch-granules had different properties than the original *ae* starches. With

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