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Physicochemical Properties of Job's Tears (*Coix lachryma-jobi* L.) Starch Modified with Different Levels of Acid Hydrolysis

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Abstract Physicochemical properties of native and acid-modified Job's tears (*Coix lachryma-jobi* L.) starches were investigated. Starch extracted from Job's tears was treated with 2.2 N hydrochloric acid for different length of time (3, 6, 12, and 18 hr). The hydrolysis pattern of starches with the acid proceeded rapidly up to 12 hr and then the approached constant values. The swelling power of acid-modified starches measured at all temperatures was lower than that of its native counterparts and the water solubility index increased as temperature and hydrolysis time increased. Rapid visco analyzer viscograms of acid-modified starches demonstrated a very low viscosity as compared with that of native starch. However, X-ray diffraction did not show any significant alteration in the crystallinity after acid-modification.

Keywords: Job's tears starch, acid-modified starch, physicochemical property, pasting property

Introduction

Carbohydrates constitute the principal component of cereals, legumes, tubers, and unripe fruits, accounting for up to 40-80% of dry matter (1). Starch is the main carbohydrate reserve in these plant sources and contributes to the appearance, structure, and quality of food products (2). However, native starches have many limitations such as low shear resistance and low cold storage stability that reduce its usefulness under processing conditions (e.g., temperature, pH, and shear) (3). Therefore, native starches have been modified by chemical, physical, and enzymatic methods to overcome the shortcomings and obtain desired attributes such as solubility, texture adhesion, dispersion, and heat tolerance (4,5). Among these methods, a chemical modification includes different chemical reaction: acid hydrolysis, oxidation, etherification, esterification, and cross-linking (6).

Acid hydrolysis changes the structure of starch granules and produces soluble materials (7). The typical procedure to manufacture acid-thinned starch is related to the treatment of treating concentrated starch slurry (36-40% solids) with mineral acid at a temperature below gelatinization temperature of the starch (40-60°C) for specific periods depending on the desired viscosity or degree of conversion (3,8). This hydrolysis process depends on the reaction condition such as reactant consistency, acid concentration, hydrolysis time, and temperature (9). Furthermore, it has advantages over other chemical methods, specially altering the physicochemical properties of starches without significantly destroying its granule shape (10).

Coix lachryma-jobi L. is known as well as Job's tears or adlay and a grain crop that mainly planted in Asia. The grain of Job's tears is pear-shaped, about 5 mm in diameter, and its protein content was similar to wheat starch. It has long been used as a nourishing food and is used in food such as soup and beverage in Asia (11-13). Recently, it has been reported that Job's tears grains may have physiological effects such as antitumor activity (14) and anti-obesity (15). Although, starches separated from Job's tears have been studied for its physicochemical properties, there are no reports regarding the effect of the degree of acid hydrolysis on the structural and physicochemical properties of Job's tears starches.

In this study, Job's tears starches were subjected to acid hydrolysis under different conditions. Then, the physico-chemical and structural properties of the resulting products were evaluated as a function of the degree of hydrolysis.

Materials and Methods

Materials Job's tears (*Coix lacryma Jobi* L.) used in this study were purchased at a grocery store and Job's tears starch was extracted according to the method of Li and Corke (11), and Ramirez (16). Flour of Job's tears was screened through a sieve of 100 mesh and was steeped in 0.45% $Na_2S_2O_5$ in a water bath at 40°C for 48 hr. The material was reground and rescreened several times through a 45 mesh openings. To remove protein and impurities, the residues were extracted with 0.05 M NaCl and 20% toluene. The residue was washed twice with distilled water and once with acetone, followed by drying at 40°C for 24 hr. After dried, the residue was ground and passed through 100 mesh sieve.

Acid hydrolysis Starches were suspended in 2.2 N HCl (15.0 g starch/120 mL acid) at 35°C for various periods of time (3, 6, 12, and 18 hr). The suspension was placed in a shaking incubator at 120 rpm. After hydrolysis, starch slurries were neutralized with 2.2 N NaOH and filtered through suction (Whatman filter #2). Carbohydrates content in the filtered liquid was assayed by phenol sulfuric acid method. The residues were washed 3 times with distilled water and dried at 60°C for 24 hr. The extent of hydrolysis was determined by expressing the solubilized

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carbohydrates as a percentage of the initial starch (7).

Water solubility index and swelling power Water solubility index (WSI) and swelling power (SP) were determined by the method of Li and Yeh (17). A suspension of 0.1 g starch in 10 mL distilled water was heated at 55, 65, 75, 85, and 95°C in a shaking water bath for 1 hr and centrifuged (4,000×g) for 20 min. The supernatant was removed and the swollen granules weighed (Ws). The total solid carbohydrate (W_1) of supernatant was determined by using the phenol sulfuric acid method. WSI and SP were calculated as follows;

$$\text{WSI (\%)}=(W1/0.1)\times 100,$$
$$\text{SP (g/g)}=Ws/[0.1(100\%-\text{WSI})]$$

Pasting properties Pasting properties were determined using a rapid visco analyser (RVA, Series 4V; Newport Scientific Pty., Ltd, Warriewood, Australia). A native starch of Job's tears sample (3.0 g, d.b.) and its acid-hydrolyzed starches (10.0 g, d.b.) with 25 mL distilled water were mixed in an aluminum can with a paddle. The mixture was stirred for 1 min to facilitate dispersion before testing. The heating and cooling cycles were programmed in the following manner: The starch suspension was held at 50°C for 1 min, heated from 50 to 95°C at a rate of 12°C/min and held at 95°C for 2.5 min. Finally, it was cooled down to 50°C at a rate of 12°C/min and held at 50°C for 2 min.

X-ray diffraction measurements X-ray diffraction analysis was performed with an X-ray diffractometer (RINT 2000; Rigaku Co., Ltd., Tokyo, Japan) with target volatage-40 kV, electric current-40 mA, scanning range-4-45°, and scan speed-4°/min. Relative crystallinity of the starches was calculated using a peak fitting software (Origin-Version 7.5; Microcal Inc., Northampton, MA, USA). The starch samples were equilibrated at 25°C for 24 hr prior to the analysis.

Statistical analysis All experiments were performed in triplicate. The results were expressed as the mean±standard deviation (SD). One-way analysis of variance (ANOVA) followed by Duncan's multiple comparison test was performed. Statistical Package for the Social Science (SPSS, Version 12.0, 2004, SPSS Inc., Chicago, IL, USA) was used.

Results and Discussion

Degree of hydrolysis The extent of hydrolysis (X) vs. time is shown in Fig. 1. The hydrolysis data plotted as $\log_{10}$ [100/(100-X)] presented an evident 2-stage process. Acid hydrolysis of Job's tears starches proceeded rapidly up to 12 hr and then leveled off from 12 to 18 hr. Similar results were reported by Biladeris *et al.* (18) who suggested that the faster stage could be attributed to the hydrolysis of more amorphous parts of the starch granule and the later slower stage to the hydrolysis of the crystalline region. In acid modification, hydroxonium ion (H_3O^+) attacks the glycosidic oxygen atom and hydrolyzes the glucosidic linkage. Acid first acts on the surface of the starch granule before it gradually enters the inner region (3). Native

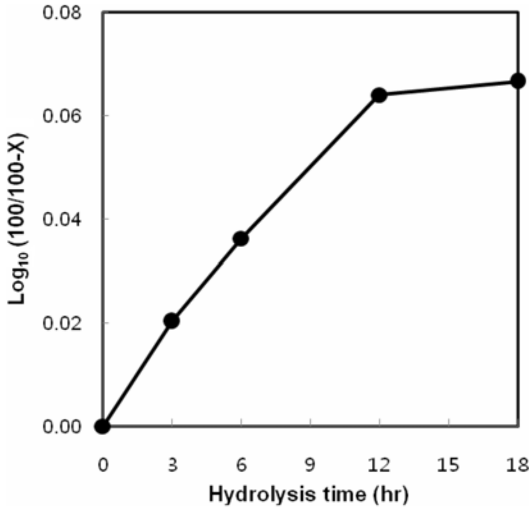

Fig. 1. Acid hydrolysis curve of Job's Tears starch which was plotted as $\log_{10}$ (100/100-X) vs time. X: The extent of hydrolysis

starches have some flaws on the surface of granules that can provide channels for the infiltration of hydrogen ions. These hydrogen ions primarily adhere to the amorphous areas and attack them (19). Wang *et al.* (20) also found that the amorphous areas of the starch granules were easier to attack with the hydrogen ions than the crystalline regions, which had a looser structure.

Swelling power and water solubility index The SP and WSI of native and acid-modified starches were measured at 10°C intervals from 55 and 95°C (Fig. 2). The swelling power of acid-modified starch decreased with an increase in hydrolysis time at all temperature tested in this study. It decreased significantly between 12 and 18 hr. These results were in good agreement with those of Wang and Wang (21) and Sánchez-Rivera *et al.* (5). It is well known that swelling power of starches have been influenced by various factors such as amylopectin molecular structure and amylose content (22,23). Morrison *et al.* (24) suggested that swelling factor is primarily a property of amylopectin and amylose acts as an inhibitor of swelling, especially in the presence of lipids. Zhou *et al.* (25) suggested that lower swelling factor of wrinkled pea starches could be attributed to its lower amylopectin content and/or strong interactions between amylose chains. Therefore, the modification of Job's tears starches with acid led to the depolymerization of amylopectin, appearing to make the starch lose the ability to hold absorbed water (21). The WSI increased as both temperature and hydrolysis time increased. A similar pattern was reported by Wang *et al.* (26) for corn starches modified with acid at different chlorine concentrations. This result indicated that the prolonged acid hydrolysis allowed the starch to be decomposed greatly, resulting in increased solubility (2). John *et al.* (27) also suggested that acid-modified starch granules could be comprised of a high proportion of soluble dextrins of both small and medium chain lengths, which increased solubility.

Pasting properties The pasting properties of native and acid-modified starches are shown in Table 1. Acid-

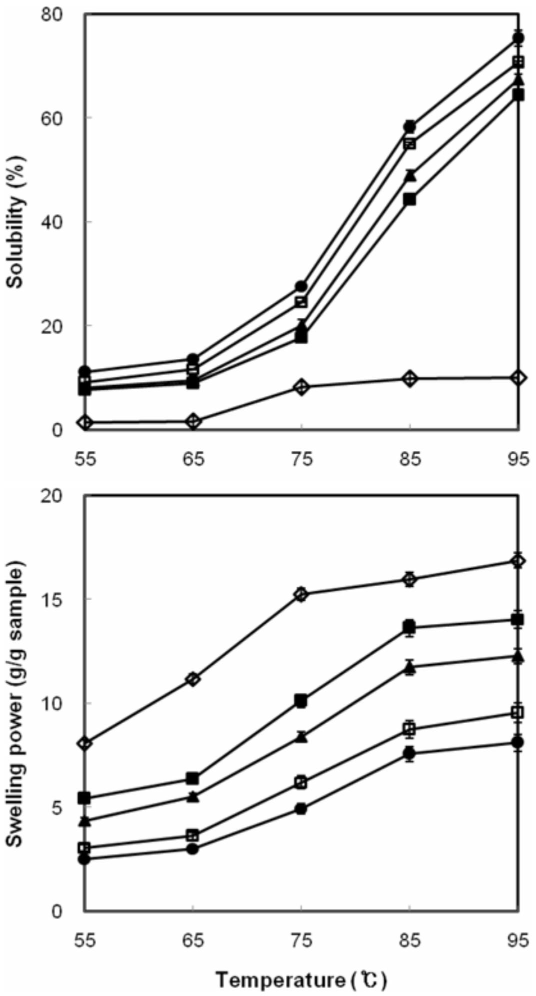

Fig. 2. Solubility and swelling power of native and acid-modified Job's tears starches. ◇, 0 hr; ■, 3 hr; ▲, 6 hr; □, 12 hr; ●, 18 hr.

modified starches were very thin, thus its RVA viscogram could be observed only at high concentration (10 g starch/ 25 mL distilled water) while the viscosity of native starch was too high to be determined by the RVA at same concentration. This result was similar to that reported by Atichokudomchai *et al.* (28) in which higher acid hydrolysis time provided tapioca starches with a very low viscosity. Pasting temperatures of native starches were reduced by the acid modification. However, the pasting temperature after 6 hr acid hydrolysis could not be detected. The peak and final viscosity of acid-modified starches decreased with increasing hydrolysis time. This observation was in agreement with the finding of Chung *et al.* (29) who mentioned that acid hydrolysis decreased paste viscosity, possibly due to the shorter chains produced. The decrease in viscosities was caused by partial cleavage glucosidic linkage form extensive hydrolysis, resulting in a decrease in molecular weight of starch molecules (4). Kuakpetoon and Wang (30) also proposed that the partially degraded network was not resistant to shear and thereby produced a lower viscosity. The breakdown viscosity (BV) decreased with an increase in hydrolysis time. The BV of samples treated for 18 hr was 1.3 RVU compared to 139.3 RVU for native starch. A higher BV presents granule disruption or the tendency of starch to succumb to shear force during heating (31). As hydrolysis time increased, the peak time (time required to peak viscosity) and setback decreased. All RVA viscograms of acid modified starches decreased with an increase in the hydrolysis time. This observation was in agreement with the finding of Koksel *et al.* (32) who reported that all the viscosity values of acid-modified corn starches are lower than native corn starch, and those of the hydrolysates progressively decreased to a significant extent with an increase in the hydrolysis time.

X-ray diffraction measurements The X-ray diffraction patterns of native and acid-modified starches are shown in Fig. 3. There was no pronounced difference between the native and modified starches. That is, the native and modified starches showed diffraction patterns with peaks at 15, 17, 18, and 23° (2θ), which are typical diffraction lines for A-type crystallites (33). This result was in accordance with that of other acid-hydrolyzed starches which exhibited the same crystalline type comparison to the unmodified starches (34). However, as the starches were subjected to a longer period of acid hydrolysis (i.e., 18 hr), the crystallinity of acid-modified starches increased slightly with sharper peaks at 2θ=17°. These results are consistent with reports by Wang *et al.* (20), Chung *et al.* (29), and Wang and Wang (34) who found that the crystallinity of starches with acid hydrolysis increased slightly with increasing extent of acid modification. It has been suggested that cleavage of starch chains in the amorphous regions allows extensive recording of the chain segments to give a more crystalline structure with a sharper X-ray pattern (35). Therefore, it may be that starch chains modified with more hydrolysis time showed an increased intensity of peak because of more extensive hydrolysis and subsequently more recording (27).

In this study, native Job's tears starches were modified

Table 1. Pasting properties of native and acid-modified Job's tears starches

Hydrolysis time (hr)	Pasting temp. (°C)	Peak viscosity (RVU)	Peak time (min)	Final viscosity (RVU)	Breakdown (RVU)	Setback (RVU)
0	74.68±0.33	323.96±19.57$^{a1)}$	4.07±0.25^{a}	211.28±0.39^{a}	139.31±1.83^{a}	26.56±0.20^{a}
3	70.45±0.05	71.09±3.36^{b}	3.50±0.15^{b}	68.02±1.78^{b}	64.50±2.21^{b}	13.58±2.55^{b}
6	ND	8.33±1.47^{c}	3.44±0.08^{bc}	4.26±0.11^{c}	6.52±0.18^{c}	2.50±1.57^{c}
12	ND	4.22±1.01^{c}	3.23±0.03^{cd}	1.17±0.02^{d}	3.03±0.10^{d}	1.10±0.60^{c}
18	ND	1.78±0.55^{c}	3.13±0.03^{d}	0.58±0.04^{d}	1.32±0.03^{d}	0.72±0.23^{c}

[1)]Different subscripts within the same column are significantly different among samples α=0.05 level by Duncan's multiple range test; ND, not detected.

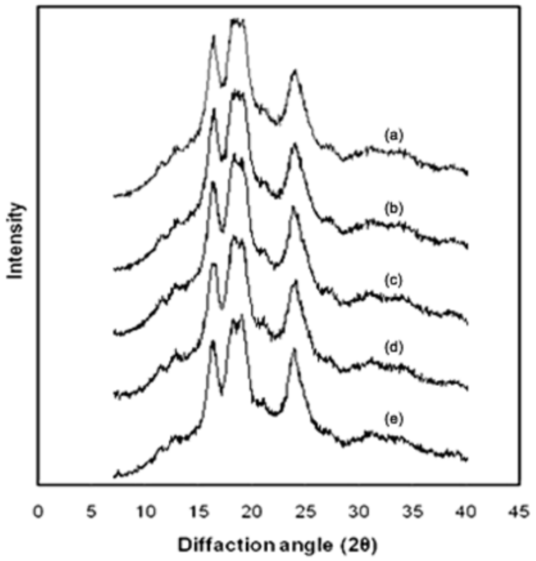

Fig. 3. X-ray diffraction pattern of Job's tears starches after acid modification. (a) native, (b) 3 hr, (c) 6 hr, (d) 12 hr, and (e) 18 hr.

with hydrochloric acid under various hydrolysis times. Then, the physicochemical and pasting properties of the modified starches were investigated as a function of the degree of hydrolysis. The acid modification significantly altered the physicochemical properties of Job's tears such as water solubility and swelling power which depended on the extent of acid hydrolysis. Also, significant reduction in pasting profiles of Job's tears starches could be readily observed, depending on the degree of acid hydrolysis. The X-ray pattern of the granule, however, remained unchanged as a result of acid modification. Thus, the physicochemical properties of the modified Job's tears starches measured in this study can be a clue for its practical use in food products. Further research would be necessary to evaluate its rheological and textural performances in a food system when incorporated into food formulations.

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References

1. Skrabanja V, Liljeberg H, Hedley C, Kreft I, Bjorck I. Influence on genotype and processing on *in vitro* rate starch hydrolysis and resistant starch formation in peas (*Pisum sativum* L.). J. Agr. Food Chem. 47: 2033-2039 (1999)
2. Aparicio-Saguilan A, Flores-Huicochea E, Tovar J, Garcia-Suarez F, Gutierrez-Meraz F, Bello-Perez LA. Resistant starch-rich powders prepared by autoclaving of native and lintnerized banana starch: Partial characterization. Starch/Stärke 57: 405-412 (2005)
3. Lawala OS, Adebowaleb KO. Physicochemical characteristics and thermal properties of chemically modified jack bean (*Canavalia ensiformis*) starch. Carbohyd. Polym. 60: 331-341 (2005)
4. Rutenberg MW, Solarek D. Starch derivatives: Production and uses. pp. 311-388. In: Starch: Chemistry and Technology. Whistler RL (ed). Academic Press, Inc., New York, NY, USA (1984)
5. Sánchez-Rivera MM, Garcýa-Suárez FJL, Valle MV, Gutierrez-Meraz F, Bello-Pérez LA. Partial characterization of banana starches oxidized by different levels of sodium hypochlorite. Carbohyd. Polym. 62: 50-56 (2005)
6. Santacruz S, Koch K, Svensson E, Ruales J, Elisson AC. Three underutilised sources of starch from the Andean region in Ecuador. Part I. Physico-chemical characterization. Carbohyd. Polym. 49: 63-70 (2002)
7. Jayakody L, Hoover R. The effect of linterization on cereal starch granules. Food Res. Int. 35: 665-680 (2002)
8. Jiping P, Shujun W, Jinglin Y, Hongyan L, Jiugao Y, Wenyuan G. Comparative studies on morphological and crystalline properties of B-type and C-type starches by acid hydrolysis. Food Chem. 105: 989-995 (2007)
9. BeMiller JN. Acid hydrolysis and other lytic reactions of starch. p. 495. In: Starch: Chemistry and Technology. Whistler RL, Paschall EF, BeMiller JN, Roberts HJ (eds). Academic Press, Inc., New York, NY, USA (1965)
10. Bentacur AD, Chel GL, Canizares HE. Acetylation and characterization of canavalia ensiformis starch. J. Agr. Food Chem. 45: 378-382 (1997)
11. Li J, Corke H. Physicochemical properties of normal and waxy Job's tears (*Coix lachryma-jobi* L.) starch. Cereal Chem. 76: 413-416 (1999)
12. Schaaffhausen RV. Adlay or Job's tears- A cereal of potentially greater economic significance. Econ. Bot. 6: 216-222 (1952)
13. Lee M-Y, Lin H-Y, Cheng F, Chiang W, Kuo Y-H. Isolation and characterization of new lactam compounds that inhibit ling and colon cancer cells from adlay (*Coix lachrymal-jobi* L. var. *ma-yuen* Stapf) bran. Food Chem. Toxicol. 46: 1933-1939 (2008)
14. Numata M, Yamamoto A, Moribayashi A, Yamada H. Anti-tumor components isolated form the Chinese herbal medicine *Coix lachryma-jobi*. Planta Med. 60: 356-359 (1994)
15. Kaneda T, Hidaka Y, Kashiwai T, Tada H, Takano T, Nishiyamas MK. Effect of coix seed on the changes in peripheral lymphocyte subsets. Rinsho Byori 40: 179-181 (1992)
16. Ramirez JL. Characterization of Job's tears starch. I. Extractions and physical properties of the starch granules. Alimentaria 276: 97-100 (1996)
17. Li JY, Yeh AI. Relationships between thermal, rheological characteristics, and swelling power for various starches. J. Food Eng. 50: 141-148 (2001)
18. Biliaderis CG, Grant DR, Vose JR. Structural characterization of legume starches. II. Studies on acid-treated starches. Cereal Chem. 58: 502-507 (1981)
19. Franco CM, Cabral RAF, Tavares DQ. Structural and physicochemical properties of lintnerized native and sour cassava starches. Starch/Stärke 54: 469-475 (2002)
20. Wang S, Yu J, Gao Wenyuan, Pang J, Liu H, Yu J. Granule structural changes in native Chinese yam (*Dioscorea opposite* Thunb var. Anguo) starch during acid hydrolysis. Carbohyd. Polym. 69: 286-292 (2007)
21. Wang Y, Wang L. Physicochemical properties of common and waxy corn starches oxidized by different levels of sodium hypochlorite. Carbohyd. Polym. 52: 207-217 (2003)
22. Tester RF, Morrison WR. Swelling and gelatinization of cereal starches I. Effects of amylopectin, amylose, and lipids. Cereal Chem. 67: 551-557 (1990)
23. Sasaki T, Matsuki J. Effect of wheat structure on swelling power. Cereal Chem. 74: 525-529 (1998)
24. Morrison WR, Tester RF, Snape CE, Law R, Gidley MJ. Swelling and gelatinization of cereal starches. IV. Some effects of lipid-complexed amylose and free amylose in waxy and normal barely starches. Cereal Chem. 70: 385-389 (1993)
25. Zhou Y, Hoover R, Liu Q. Relationship between α-amylase degradation and the structure and physicochemical properties of legume starches. Carbohyd. Polym. 57: 299-317 (2004)
26. Wang YJ, Truong VD, Wang L. Structures and rheological properties of corn starch as affected by acid hydrolysis. Carbohyd. Polym. 52: 327-333 (2003)
27. John JK, Raja KCM, Rani S, Moorthy SN, Eliasson A. Properties of arrowroot starch treated with aqueous HCl at ambient temperature. J. Food Sci. 67: 10-14 (2002)

28. Atichokudomchaia N, Shobsngob S, Chinachotic P, Varavinita S. A study of some physicochemical properties of high-crystalline tapioca starch. Starch/Stärke 53: 577-581 (2001)
29. Chung HJ, Jeong HY, Lim ST. Effects of acid hydrolysis and defatting on crystallinity and pasting properties of freeze-thawed high amylose corn starch. Carbohyd. Polym. 54: 449-455 (2003)
30. Kuakpetoon D, Wang YJ. Characterization of different starches oxidized by hypochlorite. Starch/Stärke 53: 211-218 (2001)
31. Bhosale R, Singhal R. Effect of octenylsuccinylation on physicochemical and functional properties of waxy maize and amaranth starches. Carbohyd. Polym. 68: 447-456 (2007)
32. Koksel H, Ozturk S, Kahraman K, Basman A, Ozbas OO, Ryu GH. Evaluation of molecular weight distribution, pasting and functional properties, and enzyme resistant starch content of acid-modified corn starches. Food Sci. Biotechnol. 17: 755-760 (2008)
33. Zobel HF. Molecules to granules: A comprehensive starch review. Starch/Stärke 40: 44-55 (1988)
34. Wang L, Wang YJ. Structures and physicochemical properties of acid-thinned corn, potato, and rice starches. Starch/Stärke 53: 570-576 (2001)
35. Kainuma K, French D. Naegeli amylodextrin and its relationship to starch granule structure. I. Preparation and properties of amylodextrins from various starch types. Biopolymers 10: 1673-1680 (1971)