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RESEARCH REVIEW

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Physicochemical Properties of Starch Affected by Molecular Composition and Structures: A Review

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Abstract Starches from different botanical sources differ in the ratio of amylose to amylopectin contents, molecular structures of amylose and amylopectin, granule morphology, and minor-component contents. These structural features result in different gelatinization, pasting, retrogradation properties, and enzyme digestibility of starch granules. In this review, compositions and molecular structures of starches and their effects on the physicochemical properties are summarized and discussed.

Keywords: starch, amylase, amylopectin, composition, gelatinization, retrogradation, pasting property, syneresis, enzyme digestibility

Introduction

Starch is the second most abundant carbohydrate, next to cellulose. Starch is a glucan, which is present in a semi-crystalline granular form. Starch is synthesized in many plant organs including leaves, seeds, stems, tubers, roots, and fruits. Degradation of starch provides energy for germination of seeds or sprouting of tubers.

Starch is the major energy source in human and animal diets. It provides 60 to 80% of the calories consumed by humans worldwide (1). Primary sources of starch are corn, potato, wheat, rice, and cassava. Starch is an economical commodity-product, which is widely used in many food and non-food industries. Functional characteristics of starch contribute greatly to various applications, including thickeners, gelling agents, stabilizers, binding agents, and moisture retention agents. Starch is also utilized as a renewable resource for production of ethanol and other biofuels and biomaterials.

Starch granules are dense with a specific density about 1.5 g/cm^3 (2), and are water insoluble at ambient temperatures. Morphology, compositions, and physico-chemical properties of starch granules vary with the botanical origin. Understanding the relationship between molecular structures and physicochemical properties of starch granules will greatly enhance the utilization of novel starches and provides guidelines for crossing and genetic modification of starch to produce desired properties.

In this article, the physicochemical properties of starch, including gelatinization, retrogradation and syneresis, pasting properties, and starch degradation, will be reviewed. Their relationships with the starch molecular compositions and molecular structures will be discussed.

Composition and Molecular Structure of Starch

Morphology of starch granules is characteristic of starch species. Starch granules display spherical, oval, disk, polygonal, elongated, kidney, and lobe shapes with diameters varying from submicron to more than 100 mm (3). Normal starch consists of two types of glucan: amylose and amylopectin. Amylose has essentially linear molecules of α(1-4)-linked D-glucopyranose, some with a few branches (4). Amylopectin, the major component of most starches, has highly branched molecules. Branch chains of α(1-4) D-glucose chains with different lengths are connected by α(1-6) glycosidic linkages organized into cluster structures (Fig. 1) (5). The weight-average molecular weight of amylopectin ranges from 7.0×10^7 to 5.7×10^9 g/mol (6). The semi-crystalline structure of starch is constituted by amylopectin molecules. Branch chains of amylopectin are packed into double helical structures in clusters (9-10 nm), which consist of alternating amorphous and crystalline lamellae (Fig. 2) (7-9). Branch chain length distributions of amylopectin vary with the botanical source of starch as shown in Table 1. X-ray-diffraction and crystalline-structure-modeling studies show that semi-crystalline structures of starch occur in two main polymorphic forms, the A- and the B-types, depending on the botanical sources. Left-handed double-stranded helices of amylopectin branch-chains are packed into either monoclinic or hexagonal unit cells and show the A- and the B-type X-ray diffraction

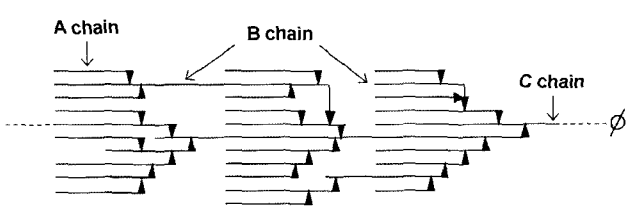

Fig. 1. Model of the cluster structure of amylopectin. The A-chains are branch chains whose reducing ends covalently link to other B- or C-chains but do not carry other chains. The B-chains link to other B- or C-chains and also carry other B- or A-chains. The C-chain is the only chain of the amylopectin molecule carrying a free reducing end.

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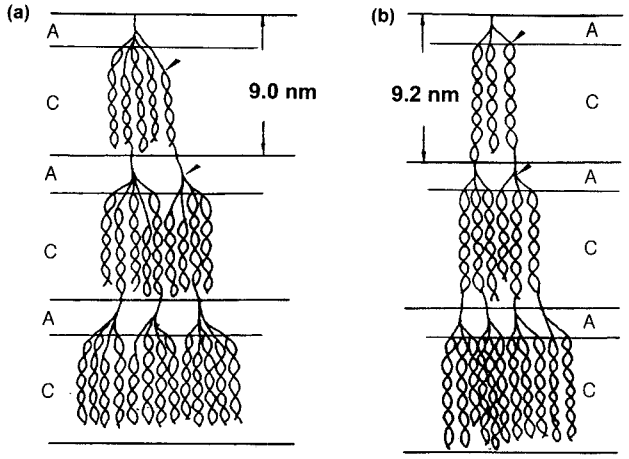

Fig. 2. Proposed models for branching patterns of (a) waxy corn starch and (b) potato starch. 'A' and 'C' stand for the amorphous and crystalline regions, respectively. The models indicate a 9-10 nm repeating distance of amylopectin cluster (47).

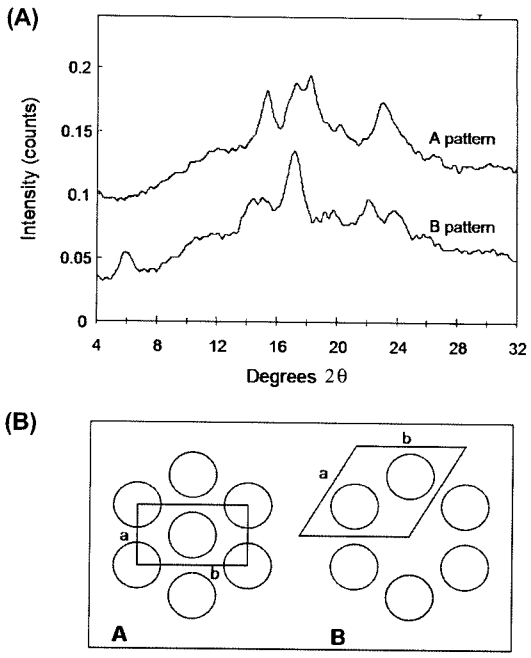

Fig. 3. X-ray diffraction pattern (A) and unit-cell dimension (B) of A- and B- type starches.

patterns, respectively (10, 11) (Fig. 3). The C-type polymorphic starch consists of a combination of the A-type and the B-type polymorphs. In general, the A-type starches, such as corn, wheat, rice, and cassava starches, consist of amylopectin with shorter chain length than do the B-type starches (potato, high-amylose corn, and wrinkle pea starches) (12). Under a polarized-light microscope, starch granules exhibit characteristic birefringence known as *Maltese cross*. The Maltese-cross birefringence reflects a radial orientation of the principle axis of the amylopectin crystallites arranged in the starch granule (7). Apparent and absolute amylose contents of selected starches are listed in Table 1. Most starches contain 15 to 30% of amylose. However, waxy cereals, such as corn and wheat, are virtually devoid of amylose, whereas some

Table 1. Amylose contents and branch chain-length distribution of amylopectin of starches

Source	Amylose content (%)		Amylopectin chain length distribution (%)				
	Apparent	Absolute	Average CL (DP)	DP 6-12	DP 13-24	DP 25-36	DP 37
A-type starch							
Normal corn	29.4	22.5	24.4	17.9	47.9	14.9	19.3
Waxy corn	0.00	0.00	23.5	17.0	49.4	17.1	16.5
Rice	25.0	20.5	22.7	19.0	52.2	12.3	16.5
Wheat	28.8	25.8	22.7	19.0	41.7	16.2	13.0
Barley	25.5	23.6	22.1	20.8	48.9	17.7	12.6
Cattail millet	19.8	15.3	21.5	20.2	53.8	12.7	13.3
Mung bean	37.9	30.7	24.8	15.6	47.6	18.3	18.5
Chinese taro	13.8	13.8	23.4	18.8	48.7	14.8	17.7
Tapioca	23.5	17.8	27.6	17.3	40.4	15.6	26.7
B-type starch							
Amylomaize V	52.0	27.3	28.9	9.7	43.9	20.3	26.1
Amylomaize VII	68.0	40.2	30.7	8.5	40.7	21.3	29.5
Potato	36.0	16.9	29.4	12.3	43.3	15.5	28.9
Green leaf canna	43.2	22.7	28.9	11.7	45.3	16.2	26.8
C-type starch							
Water chestnut	29.0	16.0	26.7	17.8	43.7	15.3	23.2

mutant cereals (e.g., high-amylose corn) have 50 to more than 70% amylose content (13). The molecular size of amylose varies between 100 anhydroglucose-units (AGU) and 10^4 AGU (14, 15).

Amylose molecules with α(1-4) linked D-glucose chains consist of a continuous hydrocarbon moiety, which enables amylose to form an inclusion helical complex with molecules consisting of a hydrophobic tail, such as 1-butanol and fatty acids. Amylose molecules are amorphous in the starch granule, and are present as either free-amylose in random coil or as lipid-complexes in single helices. Studies on cross-linking of starch granules show that amylose molecules are cross-linked to amylopectin but not cross-linked between amylose molecules. These results indicate that amylose is not present in bundles but is interspersed among amylopectin in the granule (16, 17). Results of surface gelatinization studies using saturated neutral-salt solutions, such as LiCl and $CaCl_2$, have shown that amylose molecules are more concentrated at the periphery of the starch granule than in the inner part of the granule (18, 19). These results agree with the fact that the amylose content of starch increases with the increase in granule size and maturity (20).

Some mutants of starches (e.g., high-amylose corn and *sugary*-2 corn starches) contain the intermediate materials, which are the branched molecules with smaller molecular weight than amylopectin (21-24). Starches also contain minor components, such as lipids, phospholipids, and phosphate-monoester derivatives. Waxy starch contains little lipid. Normal cereal starches contain approximate 1% of lipids (25). The structures of lipids vary with botanical sources (26, 27). Normal corn starch contains mainly free fatty acids and triglycerides, whereas normal wheat, rice, barley, and rye starches contain a substantial amount of phospholipids. In contrast, phosphate-monoester derivatives are found extensively in potato (0.02-0.1%) and other root and tuber starches (0.004-0.015%) (28). Although these minor components are present in trace amounts, they have profound effects on physical behaviors of starch. Table 2 shows phosphorous contents, i.e., phosphate-monoester derivatives and phospholipids, of selected starches determined using ^{31}P nuclear magnetic resonance (NMR) spectroscopy (26).

Table 2. Phosphorus content in starches determined using ^{31}P nuclear magnetic resonance spectroscopy[1)]

Starches	Phosphate monoesters	Phospholipids	Inorganic phosphate
Potato	0.086	nd	0.0048
Wheat	nd	0.058	trace
Mung bean	0.0083	0.0006	nd
Tapioca	0.0062	nd	Trace
Maize	0.003	0.0097	0.0013
Amylomaize V	0.005	0.015	0.0076
Waxy maize	0.0012	nd	0.0005

[1)]Percentage of phosphorus in starch (dsb, w/w); Ref. 26.

Effects of Molecular Structures of Starch on Functional Properties

Gelatinization

The starch granule is not soluble in water at ambient temperatures because of its semi-crystalline structure. When starch is placed in sufficient water, starch granules absorb a small amount of water and swell to a limited extent (ca. 30-50% dry starch basis, dsb) (7). This process is reversible before the temperature reaches the gelatinization temperature. By heating starch granules in a sufficient amount of water above the gelatinization temperature, starch granules lose their molecular order manifested in irreversible changes in properties, such as granular swelling, loss of native crystalline structure and birefringence, and starch solubilization (29, 30). This process is known as *Gelatinization*. Water acts as a plasticizer in the gelatinization process (31). A water content at a ratio of water:starch ≥2 is required to obtain the characteristic gelatinization temperature of starch. When the water content is less than two times that of the dry starch weight, the gelatinization temperature and the temperature range increase. Gelatinization of starch is an important physical transformation, which converts semi-crystalline starch to the amorphous conformation and, thus, increases the viscosity of starch paste and the enzyme digestibility.

Starch gelatinization is an endothermic reaction that corresponds to the dissociation of starch molecules from a double helical structure in starch granules to an amorphous confirmation (7, 32). The gelatinization temperature and the enthalpy change of starch gelatinization can be determined using a differential scanning calorimeter (DSC). Gelatinization occurs over a temperature range of 6.6 (barley starch) to 25.7°C (*ae* waxy maize) for the entire population of starch granules with enthalpy changes of 10 (barley starch) to 22 J/g (*ae* waxy maize) (13, 33). High-amylose corn starches have broader gelatinization ranges of 41.6-58.8°C. The gelatinization temperature of starch can also be determined using a polarized light microscope equipped with a hot stage. The temperature that starch granules lose the Maltese-cross birefringence is known as the gelatinization temperature.

Experimental results have shown that gelatinization properties of starch are related to the structure of amylopectin, the amylose content, polymorphisms, and the concentrations of phosphate-monoester derivatives and lipids. Starches of low gelatinization temperatures often show that amylopectin chain-length distribution profiles consist of an obvious shoulder at DP 18-21. This is a result of the amylopectin consisting of a large proportion of short chains (13). Noda *et al.* (34) reported that gelatinization temperatures and enthalpy changes of buckwheat and sweet potato starches negatively correlate with a proportion of short amylopectin branch-chains of DP 6-10. Corn starches, containing a smaller proportion of intermediate chain of DP 15-24, display lower onset gelatinization-temperatures (35). Studies on rice starch varieties also show that the proportion of short branch-chains (DP<10) negatively correlates with the gelatinization temperatures (36-38). Similar trends are also reported for taro and waxy corn starches (39-42). Studies with starches isolated from

large numbers of different botanical sources have shown that the gelatinization temperatures negatively correlate with the proportion of branch-chains of DP 6-12 (13, 43) and positively correlate with that of DP 13-24 (43). The length of 6 nm is proposed for the thickness of the crystalline lamellae of starch (44), which is in agreement with the crystalline thickness (ca. 5 nm) of acid-treated waxy corn observed on transmission electron microscopic (TEM) images (9). The crystalline thickness of 5-6 nm corresponds to branch chains of DP 14-17 (on the basis of 0.35 nm per anhydroglucose unit) (45, 46). Therefore, a large proportion of short branch-chains (e.g., DP 6-12) is likely to result in a crystalline defect and decreases the melting temperature of crystallites (13, 47). Amylodextrin of DP 10 is required for double helix formation in a pure oligosaccharide solution (48, 49). However, short chains of DP 6 can co-crystallized with other longer chains (48). This fact agrees with the shortest branch chain-length of DP 6 found in amylopectin.

Calorimetric studies of the A- and B-type crystalline spherulites, prepared from short chain amylose, show that the B-type spherulite melts at lower temperatures than the A-type counterparts (50, 51). A monoclinic packing cell of the A-type polymorphic form consists of 7 sets of double helices closely packed in the unit, and each unit consists of a few water molecules (2). Whereas the B-type polymorph has a hexagonal unit cell and consists of 36 molecules of water per unit cell (Fig. 3) (2, 52). A larger amount of water in the B-type polymorph may lower the gelatinization temperature of the B-type spherulites. Potato starch contains a large amount of phosphate-monoester derivatives that are covalently bounded to long branch-chains (ca. DP 42) of amylopectin (53). The repulsion force between negative charges of the phosphate-monoesters derivatives also reduces the gelatinization temperature (13, 30). Waxy starches display larger gelatinization enthalpy-changes than their normal starch counterparts. This is attributed to the presence of amylose in normal starch, and amylose is amorphous. The enthalpy changes of starch gelatinization are negatively correlated with the amylose contents (54-56). High amylose corn, *ae* waxy corn, and potato starches containing long amylopectin branch-chains also require larger energy for crystalline melting.

Gelatinization properties of starch suspensions are also affected by the presence of other solutes and physical treatments. The gelatinization temperature of starch increases with the increase in sucrose concentration (57, 58). A study on sweet potato starch shows that sucrose is the most effective in increasing the gelatinization temperatures. Fructose is the least effective and glucose is in between. The increase of gelatinization temperature is attributed to the stabilization of the crystalline region and the immobilization of water by sugars (59). Hydrothermal treatments, including heat-moisture treatments and annealing, modify the physicochemical properties of starch without destroying starch granules. Heat-moisture treatment is the exposure of starch to the moisture content lower than that require for gelatinization at an elevated temperature (e.g., above 100°C) (32, 60). The treatment causes a change from the B- to the A-type crystalline polymorph, increases the gelatinization temperature, and broadens the gelatinization temperature range. Annealing is to subject starch in excess water at a temperature above the glass transition temperature, but below the gelatinization temperature of starch. The process results in an improved crystalline order of the starch granule without affecting the polymorphic form (32). Annealing of starch increases the gelatinization temperature and narrows the gelatinization-temperature range with increased or unchanged enthalpies changes of gelatinization (60, 61).

Non-thermal gelatinization of starch can be achieved using dimethyl sulfoxide, aqueous alkali solutions, and neutral salt solutions. Dimethy sulfoxide functions as a hydrogen bond acceptor, which is capable of dissociating the starch double helices. Water-structure breaking compounds, such as KI and KSCN, break hydrogen bonds between water, which enhance the penetration of water molecules into starch granules and gelatinize the starch at room temperature. Strong alkali, such as NaOH and KOH, can react with the hydroxyl groups of starch and generate a negative charge on the hydroxyl groups. The repelling force between the negative charges dissociates the starch chains from the double helical structure (62). Neutral salts consisting of cations with a large positive charge density, such as $CaCl_2$ (18, 63) and LiCl (19, 64), can interact with the hydroxyl groups of starch molecules and release heat. The heat released can melt the double helical starch and results in the gelatinization of starch granules from the periphery of starch granule.

Pasting Properties

Starch pasting involves granular swelling, leaching of starch components from the granule, and eventually total disruption of the granules (29) as shown in Fig. 4. Amylopectin is primarily responsible for granular swelling and viscosity (65). During granular swelling, the hydrogen bonds between starch chains are dissociated and replaced with hydrogen bonds with water molecules, resulting in an increase in viscosity. Amylose is the main component of the materials leached out from the granules, and the concentration of solubilized amylopectin increases as the temperature increases (66, 67). In general, pasting properties are affected by starch concentration, rate of heating, and shear force (68). Pasting properties are characteristic for a starch variety, and the pasting properties are governed by the amylose content, the molecular structure of amylopectin, granule size, and the contents of minor components.

Waxy cereal starches typically have lower pasting temperatures and higher peak viscosity than their normal starch counterparts because of their larger amylopectin contents (13). Studies have shown that larger amylose contents correlate with smaller peak and breakdown viscosities (13, 69-73) and higher pasting temperatures (74). Several studies have shown that pasting properties of starch correlate with the branch chain-length distribution of amylopectin. Starch consisting of amylopectin with more short branch-chains of DP 6-12 displays a lower pasting temperature, lesser peak viscosity, and larger breakdown viscosity (38, 72-77). Short branch-chains do not provide strong interaction to hold the integrity of the swollen granules. Thus, the granules disrupt during heating and result in a lower peak viscosity.

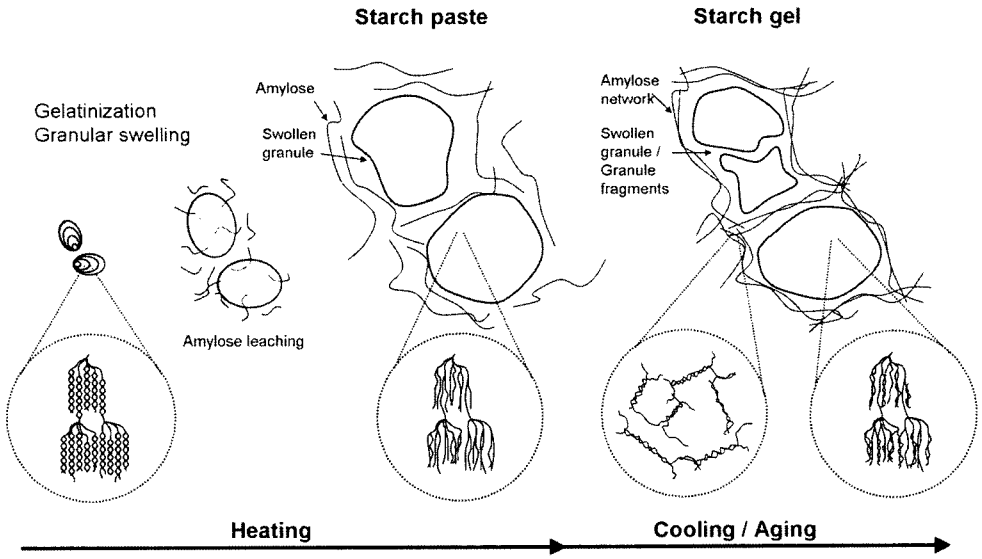

Fig. 4. Schematic representation of pasting properties of starch granules.

In general, normal cereal starches develop opaque pastes and have high pasting temperatures, small peak and breakdown viscosity. The features are attributed to the helical complexes formed between lipids and amylose molecules or long branch chains of amylopectin, which reinforce the interactions between entangled amylose and amylopectin molecules and significantly restrict the swelling of starch (13, 65, 78, 79). Phospholipids are more potent complexing agents than free fatty acids and triglycerides. Thus, swelling of wheat, barley, rye, and triticale starch granules, which contain phospholipids, are restricted. Swelling of corn starch granules is less restricted because it consists of mainly free fatty acids and triglycerides (26, 27, 80, 81). Removal of surface lipids from normal cereal starches using surfactant reagents increases the viscosity of starch pastes (82).

Tuber and root starches contain little lipids and develop clear starch pastes with lower pasting temperatures and greater peak viscosities. The extremely large peak viscosity and large breakdown viscosity of potato starch are attributed to the great granular swelling because of its large amount of phosphate-monoester derivatives (13, 53, 83). Cations of salts, such as Na^+ and Ca^{2+}, mask the negative charges of phosphate groups and reduce the repulsion force between negative charges on starch chains, thus significantly lower the viscosity of potato starch. Larger granules of potato starch display a slightly lower pasting temperature and a larger peak viscosity, compared with the smaller granules (84, 85).

During cooling of hot starch paste, leached amylose molecules rapidly aggregate. Formation of amylose junction zones is believed to be responsible for a setback viscosity. Waxy starch has a smaller setback viscosity compared with its normal starch counterpart because of the absence of amylose (13). Tsai *et al.* (86) suggested that the gel strength formation is governed by the rigidity of swollen granules, and the components leached into the solution reinforce the structure of the gel. An increase in setback viscosity with a larger amylose content is reported in wheat (56) and rice (73) starches. In normal cereal starches, amylose-lipid complexes in the granule prevent the dispersion of starch molecules and disruption of starch granules during cooking. The presence of swollen granules also contributes to the higher setback viscosity.

Retrogradation and Syneresis

Retrogradation occurs during aging of starch dispersion. Amorphous starch molecules gradually reassociate to develop double helical crystallites. The double helix formation is driven by hydrophobic interaction and hydrogen bond formation between starch chains. Retrogradation causes an increase in gel firmness and a loss of water-binding capacity of starch gel, and results in the quality deterioration of starch-containing foods. Degree of retrogradation increases with storage time. Amylose molecules retrograde much faster than amylopectin molecules. Double helix formation of amylose molecules occurs immediately after gelatinization of starch (87). Amylose is responsible to the initial rheological changes of starch paste, such as gel hardness. An increase in gel hardness with increased amylose content has been reported in rice starches (88). Amylose molecules of DP 80-100 have been reported to display the fastest retrogradation (49, 89). Retrograded amylose requires a high gelatinization of ca. 145-153°C (87, 90) to dissociate the double helices. Retrograded short-chain amylose crystallites form spherical particles with diameters of 10 to 15 μm (50). Crystalline regions of retrograded amylose, which contain ca. 31 glucose units, are highly resistant to enzyme hydrolysis (91).

Retrogradation of amylopectin proceeds slowly over several days or weeks because of its highly branched structure (92, 93). Co-crystallization between amylopectin and amylose is likely enhanced, when amylose is present in a large concentration (94). Retrogradation of amylopectin slowly transforms amorphous molecules to a weak crystalline state, and a B-type X-ray diffraction pattern is developed (95). The rate of retrogradation depends on the amylopectin

branch-chain lengths and the concentration of lipids and phosphate-monoester derivatives (13, 96). Negative correlations have been reported between the content of very short branch-chains of DP 6-9 or 6-12 and degree of retrogradation of waxy corn starch varieties (40, 41), rice starch varieties (88) and starches of differed botanical sources (13, 43, 98). Those results suggest that short branch-chains reduce the retrogradation. Studies on waxy maize and potato amylopectin show that hydrolysis of starch by β-amylase shortens the branch chain-length of amylopectin, and decreases the retrogradation (99). In general, cereal amylopectins, having shorter average chain-lengths, retrograde to a lesser extent than pea and potato amylopectins (13, 96). Retrogradation of normal cereal starches could be enhanced by the presence of amylose-lipid complexes. Amylose-lipid complexes restrict the swelling of starch molecules and dispersion of starch granules, which keep starch molecules closely located, and provide ample opportunities for starch molecules to crystallize (13). The presence of phosphate-monoester derivatives in tuber, root, and chemically modified starches tends to suppress retrogradation because of the repulsion force between negative charges of the phosphate groups (100, 101). Rate of retrogradation is also affected by the storage temperature and the water content. Three step mechanisms of nucleation, propagation, and maturation is proposed for starch retrogradation (102). The nucleation rate increases exponentially with decreasing temperature down to the glass transition temperature, whereas the propagation rate increases exponentially with the increase in temperature up to the melting temperature. The maximal crystallization occurs at 50 to 55% starch concentration (103, 104). In addition, adding salts to starch gels has been reported to decrease the percent of retrogradation in amaranth (105) and rice (106) starches.

Freezing is widely used to preserve the quality of food products. However, freezing and thawing of starch-containing products introduce severe changes to the texture of the products. During freezing, water in the starch gel forms ice crystals and enhances starch retrogradation. Temperature fluctuation above the glass transition temperature during storage in a freezer accelerates the retrogradation of starch, and a sponge-like structure is created. Upon thawing, ice melts to water and is released from the sponge-like structure. The water loss from the product is known as *syneresis*. Repeated freeze-thaw treatments introduce more syneresis. Starch with a high freeze-thaw stability is essential for frozen food products and other applications. Freeze-thaw stability of starch varies with species (107-109). Waxy starches have better freeze-thaw stability than their normal starch counterparts. Several freeze-thaw cycles are required to obtain a significant syneresis of waxy corn starch gels (110, 111). Starch with less amylose-content displays greater freeze-thaw stability (112). Molecular structure of amylopectin also affects the syneresis of the products. Jobling *et al.* (113) have shown that genetically modified potato starches containing no amylose and a large proportion of short branch-chains of amylopectin (DP 6-12) exhibit excellent freeze-thaw stabilities.

Methods to improve freeze-thaw stability of starch-containing products have been reported. For example, hydrocolloids, such as xanthan gum, can significantly enhance the freeze-thaw stability (114, 115). The authors proposed that xanthan gum does not affect ice crystal formation and amylopectin retrogradation, but prevents amylose molecules from retrogradation. A faster freezing rate has shown to improve the freeze-thaw stability (112, 116). Modified starch, such as hydroxypropyl starch, resists the retrogradation and provides excellent freeze-thaw stability (117, 118). Adding sugars to starch products, in general, improves the stability of frozen starch gels (119). Sodium chloride is shown to increase retrogradation in wheat starch gels (120), however, the contrary finding is reported in rice starch gels (121). Schoch and French (122) also reported that salts have little influence on retrogradation of wheat starch paste.

Degradation of Starch Granules

Acid hydrolysis Acid hydrolysis of starch is commonly used to reduce molecular size and to produce soluble starch for industrial applications. Susceptibility of starch granules to acid hydrolysis varies with botanical origins. Acid hydrolysis has been performed with either sulfuric acid or hydro-chloric acid to produce Naegeli dextrins (123) or Lintnerized starch (124), respectively. It has been shown that the iodine-binding capacity of starch decreases during acid hydrolysis (125, 126) and the rate of acid hydrolysis is not constant throughout the time course of hydrolysis. Most starches exhibit two-stage hydrolysis kinetics (125, 126). The faster rate of hydrolysis taking place at the early period of hydrolysis corresponds to rapid hydrolysis of amorphous materials. Crystalline materials are resistant to the acid hydrolysis and some are slowly hydrolyzed. Acid hydrolysis of a glycosidic bond requires a change in conformation of the α-D-glucosidic unit from a chair to a half chair. When the glucose units are packed in a crystalline structure, such a conformation change is hindered (7, 8). Two main groups of dextrins are found in the Naegeli dextrins and Linterized starch. The first fraction (DP 13-17) is the linear chains or branched chains with a glucosyl- or maltosyl- branch unit near the reducing end. The second fraction is the singly-branched molecules of DP ca. 25 carrying two linear chains linked by an α(1-6) linkage near the reducing end (47, 127-129). Linear dextrins in the first fraction correspond to double helices present in the crystalline lamellae that are 5-6 nm-thick (9, 44). The findings confirm a preferential attack on amorphous regions that consist of branch linkages. In addition, acid-resistant materials, such as retrograded amylose (91) and amylose-lipid complexes (7, 47, 130), may also develop during the acid-modified process and remain in the residues. X-ray diffraction studies show that acid-modified starches retain the same type crystalline pattern as their native structure but the percentage crystallinity increases (8, 125, 126). The increased crystallinity results from the removal of amorphous materials (126) and/or the annealing of crystalline materials during the process (8, 45).

Starches of different polymorphisms produce Naegeli dextrins of different chain-length distributions as shown in Fig. 5 (47). The chain length of the linear fraction of the B-type Naegeli dextrins (DP 15-17) is longer than those of

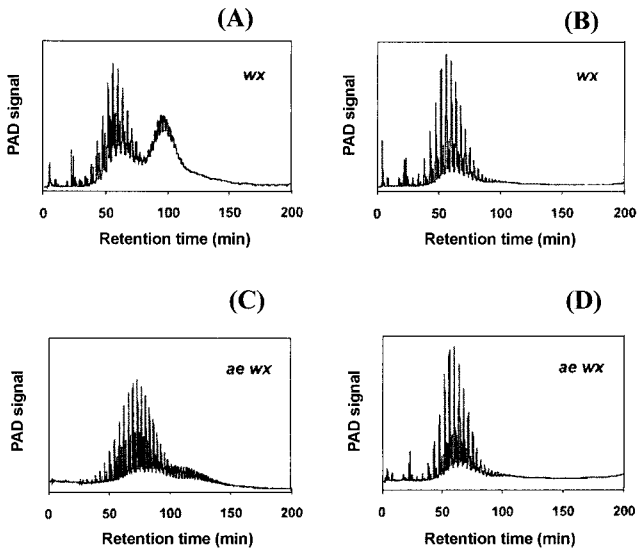

Fig. 5. Naegeli dextrins produced from the A- and B-type polymorphic starches. (A) waxy maize starch (A-type polymorphic starch) Naegeli dextrin; (B) debranched waxy maize starch Naegeli dextrin; (C) *ae* waxy maize starch (B-type polymorphic starch) Naegeli dextrin; and (D) debranched *ae* waxy maize starch Naegeli dextrin.

the A-type (DP 13-14). Naegeli dextrins of the A-type starches show a greater proportion of the singly-branched molecules. The findings suggest that large numbers of short A and B1 chains of the A-type amylopectin have branch linkages scattered in the amorphous and the crystalline regions. Those branch linkages located within the crystalline region are protected from acid hydrolysis. This result agrees with the report that hydroxypropyl derivative groups are concentrated in the region of branch linkages but a few are located at the non-reducing ends of short branch chains of manioc starch (131). The same authors also find that 50% of the short branch-chains (DP 15) do not carry any derivative groups, indicating that they are present within the crystalline region. Naegeli dextrins of B-type starches, however, produce mostly linear chains, indicating that clusters of branch linkages are mainly located in the amorphous regions and are more easily hydrolyzed by acid (Fig. 2). In high-amylose corn starch (B-type), a large amount of amylose molecules can be retrograded and protect some branch linkages of amylopectin from acid hydrolysis.

Upon heating in water, the acid-treated granules are fragile and tend to split instead of swelling. Dextrins in acid-modified high-amylose corn starch tend to easily associate with each other and form a rigid gel upon cooling, which is essential for confectionery products, such as jelly beans and gum candies. A wide range of dextrins can be produced by acid hydrolysis in different alcohols. Alcohols with longer hydrocarbon chains, such as propanol and butanol, facilitate the acid hydrolysis (132-134).

Enzymatic hydrolysis Hydrolysis of starch granules to glucose as the energy source is essential for metabolism in plants (e.g., germination of grains, sprouting of tubers), animal feeds and also for bioethanol production. Different α-amylases hydrolyze dispersed amylose and starch in different reaction patterns, including multiple attack and multi-chain attack (135, 136). Because of its semi-crystalline nature, the intact starch granule is hydrolyzed by α-amylases much more slowly than gelatinized and dispersed starch, but given time, some amylases, such as glucoamylase I, can disentangle double helical starch chains and completely hydrolyze starch molecules. The rate of enzymatic hydrolysis varies with the source of α-amylase (135, 136) and botanical origin of starch (Fig. 6) (137-139). Waxy starches display greater enzyme digestibilities than their normal starch counterparts (137, 140-142). As shown in Fig. 6, starches of the A-type polymorph are more susceptible to enzyme hydrolysis than

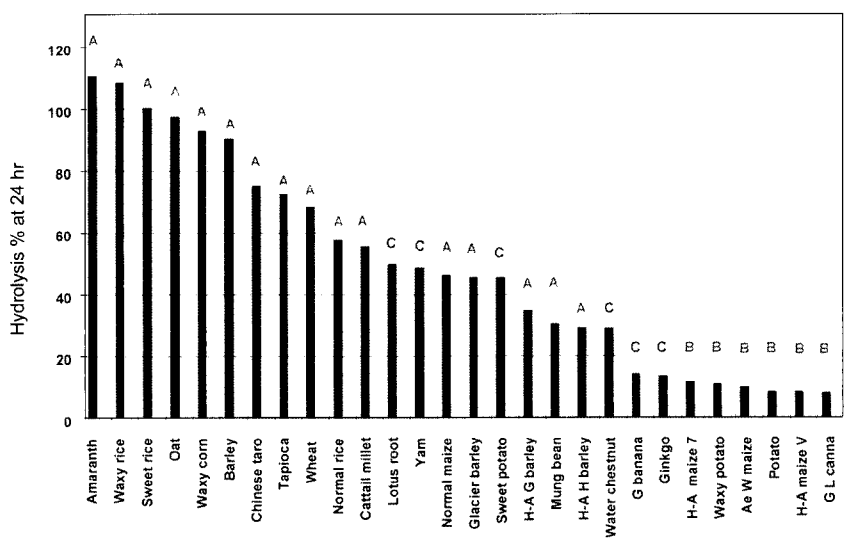

Fig. 6. Relative enzyme digestibilities of selected uncooked granular starches of different crystalline structures. A, B, and C stand for the types of crystallinity (137).

those of the B-type (137, 143, 144). In contrast to the acid hydrolysis, studies have shown that the X-ray diffraction pattern and percentage crystallinity of starch are not significantly changed after enzymatic hydrolysis (138, 145, 146). The findings indicate no preferential hydrolysis on the amorphous materials of the starch granules. The degradation pattern of starch granules is species-specific. Granule pitting and appearance of alternating growth rings are common morphological appearances for the A-type starch granules. The B-type starches are much more resistant to hydrolysis and display a minor exo-corrosion (147, 148). Amylose-lipid complexes and retrograded starch are also resistant to enzyme hydrolysis (149, 150).

Unlike acid hydrolysis, amylase with a larger molecule size (approximate 4 nm) can not penetrate freely into the molecular lattice of the granules. Thus, amylases attack starch granules on the surface (145). Exo-corrosion and pitting are apparently the sole mechanisms during the initial stage of hydrolysis. Svensson (151) suggested that hydrolysis of starch granules depends on the ability of enzymes to adsorb onto the granule surface; however, some bacterial amylases digest raw starch granules without granule adsorption (152, 153). Once the enzyme hydrolysis generates large pores to the internal structure, successive endo-corrosion occurs. Starch with larger granules has been shown to have less enzyme digestibility because of their smaller relative surface area (141, 144, 154, 155). Minor components may also influence enzyme adsorption. Surface proteins and lipids may reduce surface accessibility to the enzyme by blocking the adsorption sites (156).

The less organized zones on the granule surface (e.g., pores, spongy-like, and cracks) are more susceptible to enzyme hydrolysis (157, 158). Surface pores (approximately 0.1 μm in diameter) are commonly found distributed over the surface of some granules of A-type cereal starches, but little or none is found on the B-type starch granules (159, 160). Starch isolated from corn kernels harvested on 30 days after pollination (DAP) show little or no surface pores; however, starch isolated from that of 45 DAP (mature and dried in the field) show a large number of surface pores (20). Pitted starch granules are observed around the germ in dormant maize kernels (137). Damaged starch that occurred during isolation can develop faults or dislocations on the granule surface, allowing the enzyme greater access to the amorphous and internal structure (161).

Confocal laser-scattering micrographs (CLSM) (Fig. 7) show that starch granules have specific internal structures, depending on botanical sources (162). The presence of voids inside the granule provides more surface area for the enzyme attack. The locations of internal voids are related to the sites of enzyme hydrolysis. The A-type starches, having loosely packed internal structures and voids, are more susceptible to enzyme degradation. Normal corn granules show random and deep pitting canals through the granules. Large granules of wheat starches show voids around the equatorial groove (Fig. 7c), which is the most susceptible to enzyme hydrolysis.

B-type starch granules, such as potato and high-amylose corn, possess a homogenous internal-structure (162). The enzyme resistance of B-type starch is likely resulted from its large proportions of long branch-chains that extend through multiple clusters of amylopectin (163). These long branch-chains prohibit rearrangements of starch molecules in the granule and maintain the homogeneous internal structure. The long and stable double helices of amylopectin chains and the homogeneous internal structure without voids in the granule make the B-type starch granule highly resistant to enzyme hydrolysis.

References

1. Whisler RL, BeMiller JN. Starch. pp. 117-151. In: Carbohydrate Chemistry for Food Scientists. Whisler RL, BeMiller JN (eds). American Association of Cereal Chemists, St. Paul, MN, USA (1997)
2. Imberty A, Buleon A, Vinh T, Perez S. Recent advances in knowledge of starch structure. Starch-Stärke 43: 375-384 (1991)

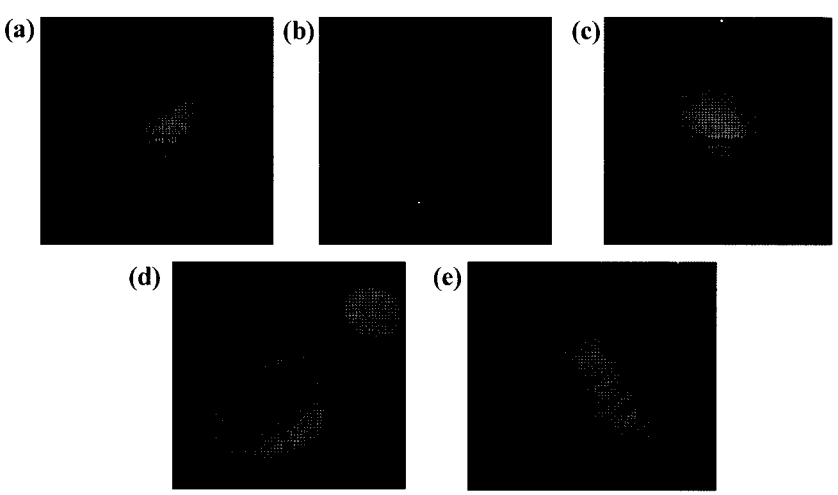

Fig. 7. Confocal laser-light scanning micrographs of starch granules. Starch was stained with rhodamine B, and unbound dye was removed by rising with water and centrifuged immediately. (a) waxy maize starch; (b) normal maize starch; (c) wheat starch; (d) potato starch; and (e) banana starch.

3. Jane J, Kasemsuwan T, Leas S, Zobel H, Robyt JF. Anthology of starch granule morphology by scanning electron microscopy. Starch-Stärke 46: 121-129 (1994)
4. Takeda Y, Hizukuri S, Takeda C, Suzuki A. Structures of branched molecules of amyloses of various origins, and molar fractions of branched and unbranched molecules. Carbohyd. Res. 165: 139-145 (1987)
5. Hizukuri S. Polymodal distribution of the chain lengths of amylopectins, and its significance. Carbohyd. Res. 147: 342-347 (1986)
6. Yoo S-H, Jane J. Molecular weights and gyration radii of amylopectins determined by high-performance size-exclusion chromatography equipped with multi-angle laser-light scattering and refractive index detectors. Carbohyd. Polym. 49: 307-314 (2002)
7. French D. Organization of starch granules. pp. 183-247. In: Starch Chemistry and Technology. Whistler RL, Bemiller JN, Paschall EF (eds). Academic Press, New York, NY, USA (1984)
8. Kainuma K, French D. Naegeli amylodextrin and its relation to starch granule structure. I. Preparation and properties of amylodextrins from various starch types. Biopolymers 10: 1673-1680 (1971)
9. Yamaguchi M, Kainuma K, French D. Electron microscopic observations of waxy maize starch. J. Ultrastruct. Res. 69: 249-261 (1979)
10. Imberty A, Perez S. A revisit to the three-dimensional structure of B-type starch. Biopolymers 27: 1205-1221 (1988)
11. Imberty A, Chanzy H, Perez S, Buleon A, Tran V. The double-helical nature of the crystalline part of A-starch. J. Mol. Biol. 201: 365-378 (1988)
12. Hizukuri S. Relationship between the distribution of the chain length of amylopectin and the crystalline structure of starch granules. Carbohyd. Res. 141: 295-306 (1985)
13. Jane J, Chen YY, Lee LF, McPherson AE, Wong KS, Radosavljevic M, Kasemsuwan T. Effects of amylopectin branch chain length and amylose content on the gelatinization and pasting properties of starch. Cereal Chem. 76: 629-637 (1999)
14. Jane J, Chen JF. Effect of amylose molecular size and amylopectin branch chain length on paste properties of starch. Cereal Chem. 69: 60-65 (1992)
15. Takeda Y, Shirasaka K, Hizukuri S. Examination of the purity and structure of amylose by gel-permeation chromatography. Carbohyd. Res. 132: 83-92 (1984)
16. Jane J, Xu A, Radosavljevic M, Seib PA. Location of amylose in normal starch granules. I. Susceptibility of amylose and amylopectin to cross-linking reagents. Cereal Chem. 69: 405-409 (1992)
17. Kasemsuwan T, Jane J. Location of amylose in normal starch granules. II. Locations of phosphodiester crosslinking revealed by phosphorus-31 nuclear magnetic resonance. Cereal Chem. 71: 282-287 (1994)
18. Jane J, Shen JJ. Internal structure of the potato starch granule revealed by chemical gelatinization. Carbohyd. Res. 247: 279-290 (1993)
19. Pan DD, Jane J. Internal structure of normal maize starch granules revealed by chemical surface gelatinization. Biomacromolecules 1: 126-132 (2000)
20. Li L, Blanco M, Jane J. Physicochemical properties of endosperm and pericarp starches during maize development. Carbohyd. Polym. 67: 630-639 (2007)
21. Inouchi N, Glover DV, Takaya T, Fuwa H. Development changes in fine structure of starches of several endosperm mutants of maize. Starch-Stärke 35: 371-376 (1983)
22. Kasemsuwan T, Jane J, Schnable P, Stinard P, Robertson D. Characterization of the dominant mutant amylose-extender (Ae1-5180) maize starch. Cereal Chem. 72: 457-464 (1995)
23. Perera C, Lu Z, Sell J, Jane J. Comparison of physicochemical properties and structures of *sugary-2* cornstarch with normal and waxy cultivars. Cereal Chem. 78: 249-256 (2001)
24. Takeda Y, Preiss J. Structures of B90 (*sugary*) and W64A (normal) maize starches. Carbohyd. Res. 240: 265-275 (1993)
25. Morrison WR. Starch lipids and how they relate to starch granule structure and functionality. Cereal Food World 40: 437-438 (1995)
26. Kasemsuwan T, Jane J. Quantitative method for the survey of starch phosphate derivatives and starch phospholipids by ^{31}P nuclear magnetic resonance spectroscopy. Cereal Chem. 73: 702-707 (1996)
27. Lim S, Kasemsuwan T, Jane J. Characterization of phosphorus in starch by ^{31}P-nuclear magnetic resonance spectroscopy. Cereal Chem. 71: 488-493 (1994)
28. Hizukuri S. Starch: Analytical aspects. pp. 305-390. In: Carbohydrates in Food. Eliasson AC (ed). CRC Press, Boca Raton, FL, USA (2006)
29. Atwell WA, Hood LF, Lineback DR, Varriano-Marston E, Zobel HF. The terminology and methodology associated with basic starch phenomena. Cereal Food World 33: 306-311 (1988)
30. Jane J. Starch: Structures and properties. pp. 81-101. In: Chemical and Functional Properties of Food Saccharides. Tomasik P (ed). CRC Press, Boca Raton, FL, USA (2004)
31. Donovan JW. Phase transitions of the starch-water system. Biopolymers 18: 263-275 (1979)
32. Eliasson A-C, Gudmundsson M. Starch: Physicochemical and functional aspects. pp. 391-469. In: Carbohydrates in Food. Eliasson AC (ed). CRC Press, Boca Raton, FL, USA (2006)
33. Liu H, Lelievre J. A model of starch gelatinization linking differential scanning calorimetry and birefringence measurements. Carbohyd. Polym. 20: 1-5 (1993)
34. Noda T, Takahata Y, Sato T, Suda I, Morishita T, Ishiguro K, Yamakawa O. Relationships between chain length distribution of amylopectin and gelation properties within the same botanical origin for sweet potato and buckwheat. Carbohyd. Polym. 37: 153-158 (1998)
35. Ji Y, Seetharaman K, Wong K, Pollak LM, Duvick S, Jane J, White PJ. Thermal and structural properties of unusual starches from developmental corn lines. Carbohyd. Polym. 51: 439-450 (2003)
36. Nakamura Y, Sakurai A, Inaba Y, Kimura K, Iwasawa N, Nagamine T. The fine structure of amylopectin in endosperm from Asian cultivated rice can be largely classified into two classes. Starch-Stärke 54: 117-131 (2002)
37. Vandeputte GE, Vermeylen R, Geeroms J, Delcour JA. Rice starches. I. Structural aspects provide insight into crystallinity characteristics and gelatinisation behaviour of granular starch. J. Cereal Sci. 38: 43-52 (2003)
38. Wong K-S, Kubo A, Jane J, Harada K, Satoh H, Nakamura Y. Structures and properties of amylopectin and phytoglycogen in the endosperm of *sugary-1* mutants of rice. J. Cereal Sci. 37: 139-149 (2003)
39. Jane J, Shen L, Chen J, Lim S, Kasemsuwan T, Nip WK. Physical and chemical studies of taro starches and flours. Cereal Chem. 69: 528-535 (1992)
40. Shi Y-C, Seib PA. The structure of four waxy starches related to gelatinization and retrogradation. Carbohyd. Res. 227: 131-145 (1992)
41. Shi Y-C, Seib PA. Fine structure of maize starches from four *wx*-containing genotypes of the W64A inbred line in relation to gelatinization and retrogradation. Carbohyd. Polym. 26: 141-147 (1995)
42. Yuan RC, Thompson DB, Boyer CD. Fine structure of amylopectin in relation to gelatinization and retrogradation behavior of maize starches from three *wx*-containing genotypes in two inbred lines. Cereal Chem. 70: 81-89 (1993)
43. Srichuwong S, Sunarti TC, Mishima T, Isono N, Hisamatsu M. Starches from different botanical sources I: Contribution of amylopectin fine structure to thermal properties and enzyme digestibility. Carbohyd. Polym. 60: 529-538 (2005)
44. Blanshard JMV. Starch granule structure and function: A physicochemical approach. pp. 16-54. In: Starch: Properties and Potential. Galliard T (ed). John Wiley & Sons , Chichester, OH, USA (1986)
45. Wu H-CH, Sarko A. Packing analysis of carbohydrates and polysaccharides. VIII. The double-helical molecular structure of crystalline B-amylose. Carbohyd. Res. 61: 7-25 (1978)
46. Zobel HF. Molecules to granules: A comprehensive starch review. Starch-Stärke 40: 44-50 (1988)
47. Jane J, Wong K-S, McPherson AE. Branch-structure difference in starches of A- and B-type x-ray patterns revealed by their Naegeli dextrins. Carbohyd. Res. 300: 219-227 (1997)
48. Gidley MJ, Bulpin PV. Crystallization of malto-oligosaccharides as models of the crystalline forms of starch: Minimum chain-length requirement for the formation of double helixes. Carbohyd. Res. 161: 291-300 (1987)

49. Pfannemuller B. Influence of chain length of short monodisperse amyloses on the formation of A- and B-type x-ray diffraction patterns. Int. J. Biol. Macromol. 9: 105-108 (1987)
50. Ring SG, Miles MJ, Morris VJ, Turner R, Colonna P. Spherulitic crystallization of short chain amylose. Int. J. Biol. Macromol. 9: 158-160 (1987)
51. Whittam MA, Noel TR, Ring SG. Melting behavior of A- and B-type crystalline starch. Int. J. Biol. Macromol. 12: 359-362 (1990)
52. Sarko A, Wu H-C. The crystal structures of A-, B-, and C-polymorphs of amylose and starch. Starch-Stärke 30: 73-78 (1978)
53. Takeda Y, Hizukuri S. Studies on starch phosphate. Part 6. Location of phosphate groups in potato amylopectin. Carbohyd. Res. 102: 321-327 (1982)
54. Fujita S, Sugimoto Y, Yamashita Y, Fuwa H. Physicochemical studies of starch from foxtail millet (*Setaria italica Beauv.*). Food Chem. 55: 209-213 (1996)
55. Kosson R, Czuchajowska Z, Pomeranz Y. Smooth and wrinkled peas. 1. General physical and chemical characteristics. J. Agr. Food. Chem. 42: 91-95 (1994)
56. Sasaki T, Yasui T, Matsuki J. Effect of amylose content on gelatinization, retrogradation, and pasting properties of starches from waxy and nonwaxy wheat and their F1 seeds. Cereal Chem. 77: 58-63 (2000)
57. Ahmad FB, Williams PA. Effect of sugars on the thermal and rheological properties of sago starch. Biopolymers 50: 401-412 (1999)
58. Lee HA, Kim NH, Nishinari K. DSC and rheological studies of the effects of sucrose on the gelation and retrogradation of acorn starch. Thermochim. Acta 322: 39-46 (1998)
59. Kohyama K, Nishinari K. Effect of soluble sugars on gelatinization and retrogradation of sweet potato starch. J. Agr. Food. Chem. 39: 1406-1410 (1991)
60. Nakazawa Y, Wang Y-j. Acid hydrolysis of native and annealed starches and branch-structure of their Naegeli dextrins. Carbohyd. Res. 338: 2871-2882 (2003)
61. Yoon J-I, Kim S-K. Effect of annealing on pasting properties and gel hardness of mung bean starch. Food Sci. Biotechnol. 12: 526-532 (2003)
62. Maher GG. Alkali gelatinization of starches. Starch-Stärke 35: 226-234 (1983)
63. Koch K, Jane J. Morphological changes of granules of different starches by surface gelatinization with calcium chloride. Cereal Chem. 77: 115-120 (2000)
64. Ahmad FB, Williams PA. Effect of salts on the gelatinization and rheological properties of sago starch. J. Agr. Food. Chem. 47: 3359-3366 (1999)
65. Tester RF, Morrison WR. Swelling and gelatinization of cereal starches. I. Effects of amylopectin, amylose, and lipids. Cereal Chem. 67: 551-557 (1990)
66. Derek R, Prentice M, Stark JR, Gidley MJ. Granule residues and "ghosts" remaining after heating A-type barley-starch granules in water. Carbohyd. Res. 227: 121-130 (1992)
67. Doublier JL. Rheological studies on starch - flow behavior of wheat starch pastes. Starch-Stärke 33: 415-420 (1981)
68. Rasper V. Theoretical aspects of amylographology. pp. 1-6. In: The Amylograph Handbook. Shuey WC, Tipples KE (eds). American Association of Cereal Chemists, St. Paul, MN, USA (1980)
69. Araki E, Miura H, Sawada S. Differential effects of the null alleles at the three Wx loci on the starch-pasting properties of wheat. Theor. Appl. Genet. 100: 1113-1120 (2000)
70. Collado LS, Mabesa RC, Corke H. Genetic variation in the physical properties of sweet potato starch. J. Agr. Food. Chem. 47: 4195-4201 (1999)
71. Reddy KR, Subramanian R, Ali SZ, Bhattacharya KR. Viscoelastic properties of rice-flour pastes and their relationship to amylose content and rice quality. Cereal Chem. 71: 548-552 (1994)
72. Srichuwong S, Sunarti TC, Mishima T, Isono N, Hisamatsu M. Starches from different botanical sources II: Contribution of starch structure to swelling and pasting properties. Carbohyd. Polym. 62: 25-34 (2005)
73. Vandeputte GE, Derycke V, Geeroms J, Delcour JA. Rice starches. II. Structural aspects provide insight into swelling and pasting properties. J. Cereal Sci. 38: 53-59 (2003)
74. Noda T, Tohnooka T, Taya S, Suda I. Relationship between physico-chemical properties of starches and white salted noodle quality in Japanese wheat flours. Cereal Chem. 78: 395-399 (2001)
75. Edwards A, Fulton DC, Hylton CM, Jobling SA, Gidley M, Rossner U, Martin C, Smith AM. A combined reduction in activity of starch synthases II and III of potato has novel effects on the starch of tubers. Plant J. 17: 251-261 (1999)
76. Franco CML, Wong K-S, Yoo S-H, Jane J. Structural and functional characteristics of selected soft wheat starches. Cereal Chem. 79: 243-248 (2002)
77. Han X-Z, Hamaker BR. Amylopectin fine structure and rice starch paste breakdown. J. Cereal Sci. 34: 279-284 (2001)
78. Galliard T, Bowler P. Morphology and composition of starch. pp. 55-78. In: Starch: Properties and Potential; Critical Reports on Applied Chemistry. Galliard T (ed). Wiley Interscience, New York, NY, USA (1987)
79. Jane J, Kasemsuwan T, Chen JF, Juliano BO. Phosphorus in rice and other starches. Cereal Food World 41: 827-832 (1996)
80. Morrison WR, Law RV, Snape CE. Evidence for inclusion complexes of lipids with V-amylose in maize, rice, and oat starches. J. Cereal Sci. 18: 107-109 (1993)
81. Yoo S-H, Jane J. Structural and physical characteristics of waxy and other wheat starches. Carbohyd. Polym. 49: 297-305 (2002)
82. Debet MR, Gidley MJ. Three classes of starch granule swelling: Influence of surface proteins and lipids. Carbohyd. Polym. 64: 452-465 (2006)
83. Noda T, Tsuda S, Mori M, Takigawa S, Matsuura-Endo C, Hashimoto N, Yamauchi H. Properties of starches from potato varieties grown in Hokkaido. J. Appl. Glycosci. 51: 241-246 (2004)
84. Kainuma K, Yamamoto K, Suzuki S, Takaya T, Fuwa H. Studies on structure and physico-chemical properties of starch. Part IV: Structural, chemical, and rheological properties of air classified small and large granule of potato starch. J. Jpn. Soc. Starch Sci. 25: 3-11 (1978)
85. Yamamoto K, Sugai Y, Onogaki T. The rheological properties of starch pastes and gels obtained from air classified potato starches. J. Jpn. Soc. Starch Sci. 29: 277-286 (1982)
86. Tsai M-L, Li C-F, Lii C-Y. Effects of granular structures on the pasting behaviors of starches. Cereal Chem. 74: 750-757 (1997)
87. Sievert D, Wuersch P. Amylose chain association based on differential scanning calorimetry. J. Food Sci. 58: 1332-1334 (1993)
88. Vandeputte GE, Vermeylen R, Geeroms J, Delcour JA. Rice starches. III. Structural aspects provide insight in amylopectin retrogradation properties and gel texture. J. Cereal Sci. 38: 61-68 (2003)
89. Gidley MJ, Bulpin PV. Aggregation of amylose in aqueous systems: The effect of chain length on phase behavior and aggregation kinetics. Macromolecules 22: 341-346 (1989)
90. Eberstein K, Hoepcke R, Konieczny-Janda G, Stute R. DSC studies on starches. Part I. Possible thermoanalytical methods for characterizing starches. Starch-Stärke 32: 397-404 (1980)
91. Jane J, Robyt JF. Structure studies of amylose-V complexes and retrograded amylose by action of alpha amylases, and a new method for preparing amylodextrins. Carbohyd. Res. 132: 105-118 (1984)
92. Eliasson AC. Retrogradation of starch as measured by differential scanning colorimetry. pp. 93-98. In: New Approaches to Research on Cereal Carbohydrates. Hill RD, Munck L (eds). Elsevier, Amsterdam, Netherlands (1985)
93. Ring SG, Colonna P, l'Anson KJ, Kalichevsky MT, Miles MJ, Morris VJ, Orford PD. The gelation and crystallization of amylopectin. Carbohyd. Res. 162: 277-293 (1987)
94. Russell PL. The aging of gels from starches of different amylose/amylopectin content studied by differential scanning calorimetry. J. Cereal Sci. 6: 147-158 (1987)
95. Brennan JG, Sodah-Ayernor G. A study of the kinetics of retrogradation in a starch-based dough made from dehydrated yam (*Dioscorea rotundata* L. *Poir*). Starch-Stärke 25: 276-280 (1973)
96. Kalichevsky MT, Orford PD, Ring SG. The retrogradation and gelation of amylopectins from various botanical sources. Carbohyd. Res. 198: 49-55 (1990)

97. Kim N-H, Yoo S-H. Molecular structure and gelatinization properties of turnip starch (*Brassica rapa* L.). Food Sci. Biotechnol. 14: 470-473 (2005)
98. Silverio J, Fredriksson H, Andersson R, Eliasson AC, Aman P. The effect of temperature cycling on the amylopectin retrogradation of starches with different amylopectin unit-chain length distribution. Carbohyd. Polym. 42: 175-184 (2000)
99. Würsch P, Gumy D. Inhibition of amylopectin retrogradation by partial β-amylolysis. Carbohyd. Res. 256: 129-137 (1994)
100. Banks W, Greenwood CT. Starch and Its Components. Edinburgh University Press, Edinburgh, Great Britian. pp 77-79 (1975)
101. Thygesen LG, Blennow A, Engelsen SB. The effects of amylose and starch phosphate on starch gel retrogradation studied by low-field ^{1}H NMR relaxometry. Starch-Stärke 55: 241-249 (2003)
102. Slade L, Levine H. Recent advances in starch retrogradation. pp. 387-430. In: Industrial Polysaccharides. The Impact of Biotechnology and Advanced Methodologies. Stivala SS, Crescenzi V, Dea ICM (eds). Gordon and Breach, New York, NY, USA (1987)
103. Longton J, LeGrys GA. Differential scanning calorimetry studies on the crystallinity of ageing wheat starch gels. Starch-Stärke 33: 410-414 (1981)
104. Zeleznak KJ, Hoseney RC. The role of water in the retrogradation of wheat starch gels and bread crumb. Cereal Chem. 63: 407-411 (1986)
105. Baker LA, Rayas-Duarte P. Retrogradation of amaranth starch at different storage temperatures and the effects of salt and sugars. Cereal Chem. 75: 308-314 (1998)
106. Lee S-W, Han S-H, Rhee C. Effects of various salts and emulsifiers on retrogradation rate of rice starch gel. Food Sci. Biotechnol. 11: 48-54 (2002)
107. Bhandari PN, Singhal RS. Effect of succinylation on the corn and amaranth starch pastes. Carbohyd. Polym. 48: 233-240 (2002)
108. Praznik W, Mundigler N, Kogler A, Pelzl B, Huber A. Molecular background of technological properties of selected starches. Starch-Stärke 51: 197-211 (1999)
109. Thitipraphunkul K, Uttapap D, Piyachomkwan K, Takeda Y. A comparative study of edible canna (*Canna edulis*) starch from different cultivars. Part I. Chemical composition and physico-chemical properties. Carbohyd. Polym. 53: 317-324 (2003)
110. Yuan RC, Thompson DB. Freeze-thaw stability of three waxy maize starch pastes measured by centrifugation and calorimetry. Cereal Chem. 75: 571-573 (1998)
111. Zheng GH, Han HL, Bhatty RS. Physicochemical properties of zero amylose hull-less barley starch. Cereal Chem. 75: 520-524 (1998)
112. Varavinit S, Anuntavuttikul S, Shobsngob S. Influence of freezing and thawing techniques on stability of sago and tapioca starch pastes. Starch-Stärke 52: 214-217 (2000)
113. Jobling SA, Westcott RJ, Tayal A, Jeffcoat R, Schwall GP. Production of a freeze-thaw-stable potato starch by antisense inhibition of 3 starch synthase genes. Nature Biotechnol. 20: 295-299 (2002)
114. Ferrero C, Martino MN, Zaritzky NE. Stability of frozen starch pastes: Effect of freezing, storage, and xanthan gum addition. J. Food Process Pres. 17: 191-211 (1993)
115. Ferrero C, Martino MN, Zaritzky NE. Corn starch-xanthan gum interaction and its effect on the stability during storage of frozen gelatinized suspensions. Starch-Stärke 46: 300-308 (1994)
116. Navarro S, Martino MN, Zaritzky NE. Effect of freezing rate on the rheological behavior of system based on starch and lipid phase. J. Food Eng. 26: 481-495 (1995)
117. Pal J, Singhal RS, Kulkarni PR. Physicochemical properties of hydroxypropyl derivative from corn and amaranth starch. Carbohyd. Polym. 48: 49-53 (2002)
118. Van Hung P, Morita N. Physicochemical properties of hydroxypropylated and crosslinked starches from A-type and B-type wheat starch granules. Carbohyd. Polym. 59: 239-246 (2005)
119. Baker LA, Rayas-Duarte P. Freeze-thaw stability of amaranth starch and the effects of salt and sugars. Cereal Chem. 75: 301-307 (1998)
120. Ciacco CF, Fernandes JLA. Effect of various ions on the kinetics of retrogradation of concentrated wheat starch gels. Starch-Stärke 31: 51-53 (1979)
121. Chang SM, Liu LC. Retrogradation of rice starches studied by differential scanning calorimetry and influence of sugars, sodium chloride, and lipids. J. Food Sci. 56: 564-566, 570 (1991)
122. Schoch TJ, French D. Studies on bread staling. I. The role of starch. Cereal Chem. 24: 231-249 (1947)
123. Naegeli CW. Contributions to the recent knowledge of the starch group. Liebigs Ann. Chem 173: 218-227 (1874)
124. Lintner CJ. Study over diastase. J. Prakt. Chem. 34: 378-394 (1886)
125. Biliaderis CG, Grant DR, Vose JR. Structural characterization of legume starches. II. Studies on acid-treated starches. Cereal Chem. 58: 502-507 (1981)
126. Robin JP, Mercier C, Charbonniere R, Guilbot A. Lintnerized starches. Gel filtration and enzymic studies of insoluble residues from prolonged acid treatment of potato starch. Cereal Chem. 51: 389-406 (1974)
127. Jacobs H, Eerlingen RC, Rouseu N, Colonna P, Delcour JA. Acid hydrolysis of native and annealed wheat, potato, and pea starches - DSC melting features and chain length distributions of lintnerized starches. Carbohyd. Res. 308: 359-371 (1998)
128. Srichuwong S, Isono N, Mishima T, Hisamatsu M. Structure of lintnerized starch is related to X-ray diffraction pattern and susceptibility to acid and enzyme hydrolysis of starch granules. Int. J. Biol. Macromol. 37: 115-121 (2005)
129. Umeki K, Kainuma K. Fine structure of Nageli amylodextrin obtained by acid treatment of defatted waxy-maize starch - structural evidence to support the double-helix hypothesis. Carbohyd. Res. 96: 143-159 (1981)
130. Morrison WR, Tester RF, Gidley MJ, Karkalas J. Resistance to acid hydrolysis of lipid-complexed amylose and lipid-free amylose in lintnerized waxy and non-waxy barley starches. Carbohyd. Res. 245: 289-302 (1993)
131. Hood LF, Mercier C. Molecular structure of unmodified and chemically modified manioc starches. Carbohyd. Res. 61: 53-66 (1978)
132. Fox JD, Robyt JF. Modification of starch granules by hydrolysis with hydrochloric acid in various alcohols, and the formation of new kinds of limit dextrins. Carbohyd. Res. 227: 163-170 (1992)
133. Ma WP, Robyt JF. Preparation and characterization of soluble starches having different molecular sizes and composition, by acid hydrolysis in different alcohols. Carbohyd. Res. 166: 283-297 (1987)
134. Robyt JF, Choe J, Hahn RS, Fuchs EB. Acid modification of starch granules in alcohols: Effects of temperature, acid concentration, and starch concentration. Carbohyd. Res. 281: 203-218 (1996)
135. Robyt JF. Enzymes in the hydrolysis and synthesis of starch. pp. 87-123. In: Starch: Chemistry and Technology. Whistler RL, Bemiller JN, Paschall EF (eds). Academic Press, London, UK (1984)
136. Robyt JF, French D. Multiple attack hypothesis of α-amylase action. Action of porcine pancreatic, human salivary, and *Aspergillus oryzae* α-amylases. Arch. Biochem. Biophys. 122: 8-16 (1967)
137. Jane J, Ao Z, Duvick SA, Wiklund M, Yoo S-H, Wong K-S, Gardner C. Structures of amylopectin and starch granules: How are they synthesized? J. Appl. Glycosci. 50: 167-172 (2003)
138. Leach HW, Schoch TJ. Structure of the starch granule. II. Action of various amylases on granular starches. Cereal Chem. 38: 34-46 (1961)
139. Lee Y-T, Chang H-G. The effect of heat treatments on *in vitro* starch digestibility and resistant starch of selected cereals. Food Sci. Biotechnol. 13: 810-813 (2004)
140. Evers AD, Juliano BO. Varietal differences in surface ultrastructure of endosperm cells and starch granules of rice. Starch-Stärke 28: 160-166 (1976)
141. Fuwa H, Nakajima M, Hamada A, Glover DV. Comparative susceptibility to amylases of starches from different plant species and several single endosperm mutants and their double-mutant combinations with *opaque-2* inbred *Oh43* maize. Cereal Chem. 54: 230-237 (1977)
142. Noda T, Kimura T, Otani M, Ideta O, Shimada T, Saito A, Suda I. Physicochemical properties of amylose-free starch from transgenic sweet potato. Carbohyd. Polym. 49: 253-260 (2002)

143. Kimura A, Robyt JF. Reaction of enzymes with starch granules: Kinetics and products of the reaction with glucoamylase. Carbohyd. Res. 277: 87-107 (1995)
144. Valetudie JC, Colonna P, Bouchet B, Gallant DJ. Hydrolysis of tropical tuber starches by bacterial and pancreatic α-amylases. Starch-Stärke 45: 270-276 (1993)
145. Colonna P, Buleon A, Lemarie F. Action of *Bacillus subtilis* α-amylase on native wheat starch. Biotechnol. Bioeng. 31: 895-904 (1988)
146. Lauro M, Forssell PM, Suortti MT, Hulleman SHD, Poutanen KS. α-Amylolysis of large barley starch granules. Cereal Chem. 76: 925-930 (1999)
147. Gallant DJ, Bouchet B, Buleon A, Perez S. Physical characteristics of starch granules and susceptibility to enzymatic degradation. Eur. J. Clin. Nutr. 46: S3-S16 (1992)
148. Gallant DJ, Derrien A, Aumaitre A, Guibot A. In *vitro* degradation of starch by pancreatic juice. Starch-Stärke 25: 56-64 (1973)
149. Cui R, Oates CG. The effect of retrogradation on enzyme susceptibility of sago starch. Carbohyd. Polym. 32: 65-72 (1997)
150. Cui R, Oates CG. The effect of amylose-lipid complex formation on enzyme susceptibility of sago starch. Food Chem. 65: 417-425 (1999)
151. Svensson B. Regional distant sequence homology between amylases, α-glucosidases, and transglucanosylases. FEBS Lett. 230: 72-76 (1988)
152. Hamilton LM, Kelly CT, Fogarty WM. Raw starch degradation by the non-raw starch-adsorbing bacterial alpha amylase of *Bacillus* sp. IMD 434. Carbohyd. Res. 314: 251-257 (1998)
153. Hayashida S, Teramoto Y, Inoue T, Mitsuiki S. Occurrence of an affinity site apart from the active site on the raw-starch-digesting but non-raw-starch-adsorbable *Bacillus subtilis* 65 α-amylase. Appl. Environ. Microb. 56: 2584-2586 (1990)
154. Noda T, Takahata Y, Nagata T. Factors relating to digestibility of raw starch by amylase. J. Jpn. Soc. Starch Sci. 40: 271-276 (1993)
155. Snow P, O'Dea K. Factors affecting the rate of hydrolysis of starch in food. Am. J. Clin. Nutr. 34: 2721-2727 (1981)
156. Oates CG. Towards an understanding of starch granule structure and hydrolysis. Trends Food. Sci. Tech. 8: 375-382 (1997)
157. Badenhuizen NP. Chemistry and biology of the starch granule. pp. 1-74. In: Protoplasmatologia. Heilbrunn LV, Weber F (eds). Springer, Verlag, Vienna, Austria (1959)
158. Fuwa H, Glover DV, Sugimoto Y, Tanaka M. Comparative susceptibility to amylases of starch granules of several single endosperm mutants representative of floury-opaque, starch-deficient, and modified starch types and their double-mutant combinations with *opaque-2* in four inbred lines of maize. J. Nutr. Sci. Vitaminol. 24: 437-448 (1978)
159. Fannon JE, Gray JA, Gunawan N, Huber KC, BeMiller JN. The channels of starch granules. Food Sci. Biotechnol. 12: 700-704 (2003)
160. Fannon JE, Hauber RJ, BeMiller JN. Surface pores of starch granules. Cereal Chem. 69: 284-288 (1992)
161. Lauro M, Poutanen K, Forssell P. Effect of partial gelatinization and lipid addition on α-amylolysis of barley starch granules. Cereal Chem. 77: 595-601 (2000)
162. Jane J, Atichokudomchai N, Suh DS. Internal structures of starch granules revealed by confocal laser-light scanning microscopy. pp. 147-156. In: Starch: Progress in Structural Studies, Modifications and Applications. Tomasik P, Yuryev VP, Bertoft E (eds). Polish Soc. Food Technologists, Cracow, Poland (2004)
163. Hizukuri S, Kaneko T, Takeda Y. Measurement of the chain length of amylopectin and its relevance to the origin of crystalline polymorphism of starch granules. BBA-Gen. Subjects 760: 188-191 (1983)