REVIEW

Structure-function relationships of starch components

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Starch continues to be an important renewable biopolymer in both the food and non-food industries. Its properties, which vary depending on the plant source, stem directly from its structure. The recent substantial increase in knowledge of the molecular structure of starch components and the architecture of starch granules has contributed significantly to an enhanced understanding of the structural base of starch functionality. This résumé provides an overview of the present views of the starch structure-function relationships with an emphasis on industrially important starch sources.

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1 Introduction

Starch exists in its native form as semi-crystalline granules that are essentially composed of two polyglucans. Amylopectin (AP) is the major component in most starches. The extensively branched structure consists of short chains of α-(1,4)-linked D-glucosyl units that are interconnected through α-(1,6)-linkages [1–4]. Amylose (AM) is the minor component, typically comprising 15–35% of the starch by weight, and is essentially linear with much longer chains than in AP, although a fraction of AM is slightly branched [5, 6]. In addition, many starches, especially those with extended AM content, contain intermediate materials with structures intermediate to those of AP and AM [7–11]. Starch granules also contain small quantities of proteins, fatty acids and minerals that influence the properties of starch [12–17]. Main sources of starch for commercial applications are maize, potato, cassava (also named tapioca and manioc), and wheat. Other commercial sources are e.g. rice, sweet potato, mung bean, sorghum, sago, and barley. In addition, certain mutant plants with little or no amylose, known as waxy samples (e.g. waxy maize and waxy potato), as well as high-amylose starches (e.g. amylomaize or Hylon) are of commercial value. Table 1 summarizes the properties and composition of some starches of industrial importance. It should be noticed that the table only gives a rough idea of the granule contents, because the composition and properties of starch granules vary considerably depending on year-to-year variations, place of growth, the plant variety, methods of starch isolation, as well as the analytical methods used [9, 18–22].

The properties of starch, which vary considerably between samples from different plants and between varieties within species, depend on the molecular composition and the structure of the components. These properties are of considerable importance for the diverse applications of starch. Herein follows a short overview of the molecular and granular characteristics of some selected native starches and the relationship with industrially interesting properties, namely the gelatinization and swelling of the starch granules, and the retrogradation, pasting, and freeze-thaw stability of the solubilized starch components. For information regarding starch digestibility and resistant starch, the reader is referred to recent excellent reviews by Sajilata *et al.*, Zhang and Hamaker, Sing *et al.* and Fuentes-Zaragoza *et al.* [23–26].

2 Molecular structures of amylose and amylopectin

AM has average degree of polymerization (DP) between approximately 900 and 3300 and consists of chains with an average length (CL) corresponding to 270–525 glucosyl units. The branched fraction of amylose has between 5 and 20

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Table 1. Properties and composition of starch granules

Starch source	Maize	Wheat	Barley	Tapioca	Potato
Type of starch	Cereal	Cereal	Cereal	Root	Tuber
Shape	Round, polygonal	Round, lenticular[a]	Round, lenticular[a]	Oval, truncated	Round, oval
Diameter (μm)	2–30	1–40[a]	2–40[a]	4–45	5–100
Crystallinity type	A	A	A	A or C	B
Amylose (%)	23–32	23–29	22–27	17–33	18–29
Lipids (%)	0.6–0.8	0.3–0.8	0.6–0.9	0.03–0.1	0.02–0.2
Proteins (%)	0.3–0.4	0.3	0.1	0.2	0.1–0.4
Phosphorus (%)[b]	0.03	0.06	0.05–0.06	0.01	0.04–0.09
Phosphate (nmol/mg)[c]	0.11	0.20	0.12	1.11	23.2

a Wheat and barley contain two populations of granules: lenticular, large (A-) granules with diameter >10 μm and round, small (B-) granules <10 μm.
b Phosphorus is primarily part of the lysophospholipids in cereals, but is covalently linked to AP as phosphate monoester groups in potato and tapioca.
c Phosphate in starch as glucose 6-phosphate, which generally comprises 2/3 of the phosphate bound to AP. The rest (1/3) is found as glucose 3-phosphate.
Values represent a summary of those reported in the literature [4, 9, 13, 15, 21, 22, 58, 66, 72, 73, 90–92, 95, 126, 164–170]. Note that the composition of starch also varies between varieties and due to environmental conditions, experimental methods, etc..

chains, depending on the source of starch [6]. AM in solution crystallizes readily, a phenomenon known as retrogradation, in the form of left-handed double helices that are packed in a parallel fashion forming either A- or B-type allomorphs [27]. The A-type allomorph consists of a unit cell with the monoclinic space group *B2* and dimensions of the a-, b- and c-axes being 2.083, 1.145, and 1.058 nm, respectively[28]. The B-type allomorph crystal has a unit cell with the hexagonal space group $P6_1$ and the dimensions a = b = 1.85 nm and c = 1.04 nm [29]. This allomorph contains 36 water molecules per unit cell, whereas the A-type only has eight water molecules [28]. Amylose also forms complexes with ligands, especially iodine, fatty acids and alcohols [30–34]. These helices are single stranded with dimensions depending on the type of ligand molecule. E.g., the characteristic blue complex with iodine is a left-handed helix with six residues per turn [35]. The crystalized single helices are known as V-types, as opposed to the either A- or B-type allomorphs of the double helices.

The molecular structure of AP is considerably more complex than AM and the DP of AP is much higher. Number average DP (DP_n) is between 4800 and 15,900 (Table 2) [3], whereas weight average molar mass (M_w) values are in the order of 10^7–10^8 [36–38] (which corresponds to DP_w 10^5–10^6). However, very high M_w values were found to be due to aggregates of AP in water, whereas in the good solvent dimethyl sulfoxide (DMSO) M_w was around 10^6 [39]. The average CL of AP is only 18–27 glucose residues [40–42]. The

Table 2. Structural parameters of amylopectin[a]

Starch source	Maize	Wheat	Barley	Tapioca	Potato
Structure type[b]	2	1–2	1	3	4
DP_n	5100–15,900	4800–9400	8700–12,000	6000–17,100	11,200
CL_n	18–22	17–21	18–20	19–21	22–24
ECL	11.9[c]	11.9	11.2	12.4	14.1
ICL	5.1[c]	4.4	5.3	5.3	8.0
TICL	12.0[c]	12.0	12.3	14.6	19.9
Ratio of S:L	13.5[c]	17.9	19.4	11.0	6.3
Ratio of A:B	0.9[c]	1.3	1.0	1.3	1.2
NC of AP	240–330	250–450	460–600	300–860	490
NC of clusters	11.6[c]	14.2	8.4–12.3	9.4–11.1	5.1–7.5
IB-CL	6.2[c]	6.4	5.8–6.1	6.8–7.5	7.1–8.5

a Values represent a summary of those reported in the literature [1, 3, 4, 22, 40, 41, 47, 50, 171–176]. DP_n = number average degree of polymerization; CL_n = number average chain length; ECL = external CL; ICL = internal CL; TICL = total internal CL; S:L = short:long chains; A:B = A:B-chains; NC = average number of chains; IB-CL = average interblock chain length.
b Structure of AP based on the internal unit chain distribution [4].
c Values are for waxy maize amylopectin.

chains are of two major types, namely short chains with CL 6∼36 and long chains with CL > 36 [4, 41–43]. About 5% of the residues are involved in (1,6)-linkages (the branch points) in the macromolecule. Chains that do not carry other chains through (1,6)-linkages are known as A-chains, whereas chains that carry other chains are B-chains [44]. The molar ratio of A: B-chains depends on the sample and is between 0.8∼1.4 (Table 2) [4, 45]. The so-called C-chain carries the sole reducing end-group of the macromolecule, but is otherwise similar to the B-chains [44]. All A-chains are external chains, i.e. they are completely situated outside the outermost branches. All B-chains, however, consist of an external chain segment and an internal part. The average external chain length (ECL) is between 10.7 and 15.0, depending on the starch sample [4]. The average internal chain length (ICL), defined as the segments between branches, is only 4.6–8.0. The average total internal chain length (TICL), however, is considerably longer (12.0–19.9) and includes the whole B-chain segment without the external part (Table 2) [4].

The structure of AP was divided into four characteristic types based on the size-distribution of the internal unit chains (i.e., the internal part of the B-chains) [4]: Type 1 APs have typically very little long B-chains and a broad size-distribution of short B-chains. Type 2 has considerably more long B-chains (especially so-called B2-chains) and a narrower size-distribution of the short chains. Type 3 has a little more long chains compared to type 2, whereas type 4 has considerably more long B-chains (especially B3-chains). Cereals typically belong to type 1 (e.g., barley, oat, and rye) and type 2 (e.g., maize and rice), whereas legumes, roots and tubers are of type 3 (e.g., cassava and mung bean) and type 4 (e.g., potato and canna) [4].

The organization of the unit chains in AP is critical for the understanding of the structure and architecture of the entire macromolecule. At present, the actual organization is not clarified, but two major hypothesis exist, namely the cluster model and the building block backbone model. The cluster model [46–49] suggests that the short unit chains are organized into clusters and the long chains interconnect them (Fig. 1). The actual structure of the clusters is not known, however, but the ratio of short to long chains (S:L-chains), which range between 5 and 20 in different samples (Table 2), would suggest the number of chains that are involved in a typical cluster [1]. Attempts to isolate clusters by using the enzyme α-amylase from *Bacillus amyloliquefaciens* suggest that clusters have a very broad size-range with average values corresponding to 5–16 chains, albeit most often not corresponding to the ratio of S:L-chains in a particular sample (Table 2) [50]. Nevertheless, the clusters obtained with this enzyme suggest that a cluster could be defined as a group of chains, in which the length of the segments between branches is <9 [51].

The building block backbone model [52] suggests that the clusters are built up from still smaller structural units called building blocks (Fig. 1). These units consist of up to 10 or more chains with ICL of only 1–3 residues [53]. The smallest blocks with only 2 chains are most common and the largest blocks are found in nearly trace amounts. The blocks inside the clusters are separated by segments corresponding to an average inter-block chain length (IB-CL) of approximately 5–8 residues (Table 2) [50]. Corresponding segments outside the clusters (i.e., inter-cluster segments) have been estimated to have at average up to 14 residues [54, 55]. The building blocks were suggested to be outspread along a backbone consisting mostly, but not entirely, of the long unit chains in AP. Very short internal chains, named "fingerprint" B-chains (B_{fp}-chains, TICL 3–7), are probably entirely included within the building blocks. Longer B-chains of the short chain category (usually the major part of the short B-chains, BS_{major}-chains with TICL 8∼23) were suggested to be involved to a large extent in the interconnection of two, or maybe three, building blocks and to a large part be found as branches to the backbone [52].

Unfortunately, to-date there has been no individual experiment presented that was able to distinguish between the two structural models of AP. However, the structural features of clusters isolated by the α-amylase of *Bacillus amyloliquefaciens* are in favour of the building block backbone model [50, 56]. The iodine binding properties of acid treated starches and β-limit dextrins were also found to support the backbone model [57]. In addition, as we will see, certain functional properties of starch have been proposed to be better explained by this model.

3 Starch granules

The size and morphology of starch granules varies considerably between plant species [58]. The diameter ranges from less than 1 μm to about 100 μm. Generally, cereals have small granules compared to roots or tubers (Table 1). The granules are semi-crystalline, i.e., they consist of both crystalline and amorphous structures. Observed in light microscope or in scanning electron microscope (after a slight acid or enzyme treatment), the granules possess generally rings, or shells, commonly known as "growth rings", with alternating amorphous and semi-crystalline structures and typical thickness of 100–400 nm [59–64] (Fig. 1). The structure of the amorphous rings is poorly known, but it is generally believed that a large part of AM in the granules is in the amorphous state and, thus, found in these rings [65, 66]. However, AP is probably also a component in these rings, because the rings also exist in amylose-free (waxy) granules [67].

The semi-crystalline rings consist of stacks of alternating crystalline and amorphous lamellae with a universal repeat distance of approximately 9 nm [68]. It is generally accepted that it is the AP component that is responsible for this

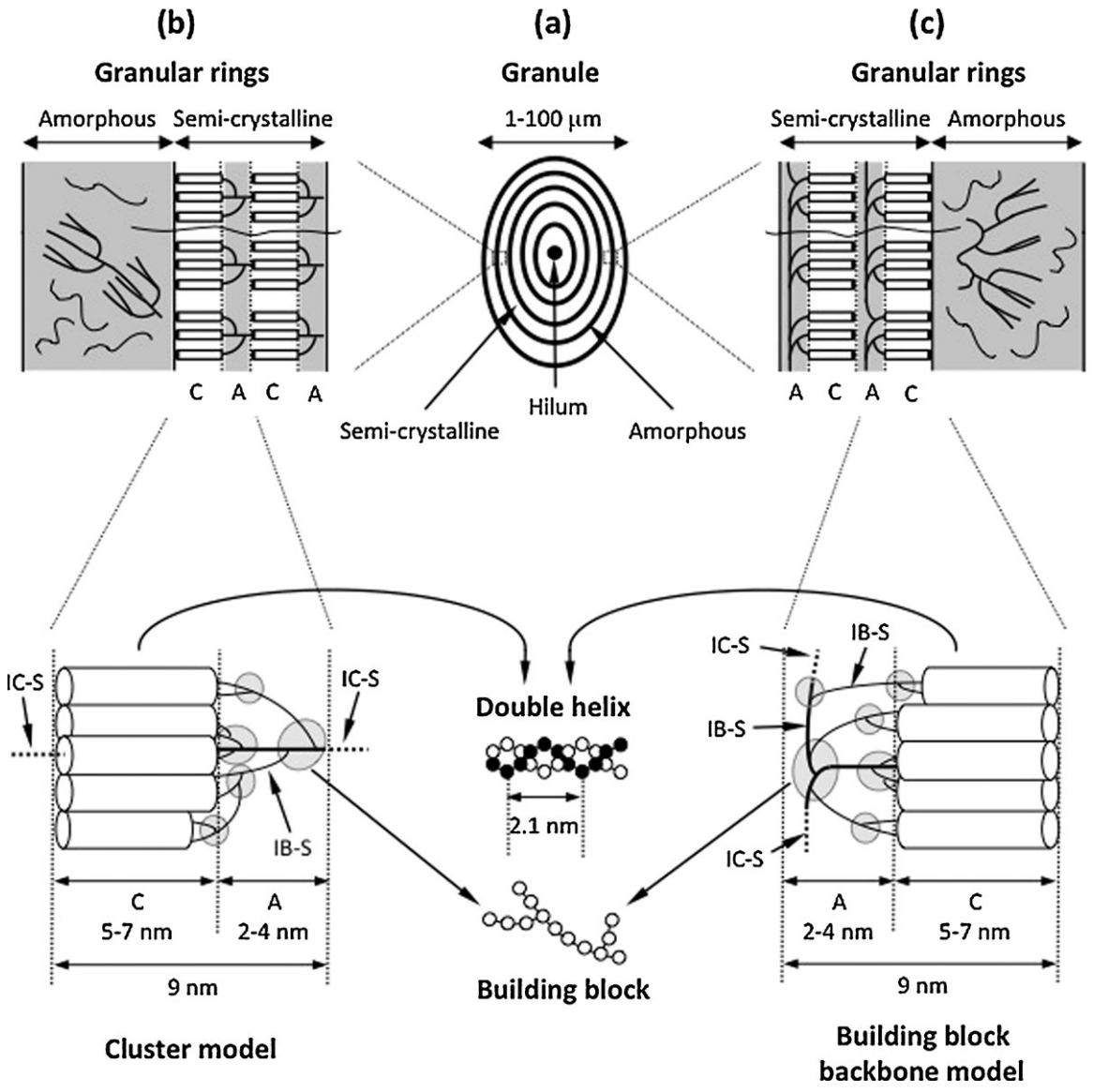

Figure 1. From starch granules to building blocks; a schematic showing the different structural levels of starch granules. (a) The granule consisting of alternating rings with a hilum region, normally considered as amorphous and centered to their middle. (b) The principal arrangement of the semi-crystalline rings according to the cluster structure of AP. (c) The principal arrangement of the semi-crystalline rings according to the building block backbone structure of AP. The structure of the amorphous rings is not established, but consists of AM as well as AP. The semi-crystalline rings consist of alternating crystalline (C) and amorphous (A) lamellae, which are enlarged in the lower figures. The details of double helices (cylinders) and building blocks (encircled) are depicted in the centre lower figure (circles depict glucose residues). Interblock segments (IB-S) and intercluster segments (IC-S) are indicated and are found in both models, but the principal unit in (b) is the cluster and in (c) is the much smaller and more tightly branched building block. Note that a major difference between the models is that in (b) the AP molecules penetrate the stacks of lamellae, whereas in (c) the AP molecules do not penetrate the stacks. The blocklets or super helices of AP are not shown, but are supposed to be structures in between the granular rings and the molecular levels.

structural organization, albeit AM probably also is involved and interfere with its properties [69, 70]. The short, external chains of AP form double helices [71] that in certain starches, particularly from cereals, form the A-type allomorph (of the same type as discussed for AM above), whereas in most tuber and many root starches they form the B-type allomorph (Table 1) [72–74]. These double helices build up the crystalline lamellae [27], which have a thickness of approximately 4.1–6.4 nm [75–77] and correspond largely to the ECL of AP. Some plant species, particularly from legumes, have mixed type of crystals with B-type in the centre and A-type at the periphery of the granule, and are then known as C-type granules [78]. The amorphous lamellae contain most of the branches of AP and the internal chains. The actual structure of this region is poorly known. The building block backbone model of AP suggests that the backbone is found here and

that layers of AP molecules build up the stacks of the lamellae [52] (Fig. 1c). The cluster model, however, suggests that the AP molecules penetrate the stacks of lamellae, so that the long, inter-cluster chains are only partly amorphous, and partly crystalline [47, 49] (Fig. 1b). Both models, however, consider that most of the branches are in the amorphous lamellae, whereas some branches might be scattered into the crystalline lamellae [79].

A structural level between growth rings and lamellae has been observed by scanning electron microscopy and atomic force microscopy. This level consists of "blocklets" with diameters roughly from 20–100 nm [80–86]. The exact nature of the blocklets is still unclear, but they appear to exist in both the amorphous and the semi-crystalline rings. This suggests that blocklets in the latter rings contain the stacks of lamellae. Blocklets in these parts of the granules were pictured as having a more perfect structure, whereas blocklets in amorphous growth rings would have more defect structures [87]. Still another molecular structure, a super-helix, was suggested for the AP component [88]. In this structure, the entire AP molecule, or molecules, are turned into a super-helical arrangement with tilted lamellae [89]. The actual existence of the super-helix is still uncertain and the relation with the blocklets, as well as the actual relation of blocklets to the rest of the granular structures, remains a matter of speculation.

4 Granular swelling

The water absorption capacity, solubility pattern and swelling behaviour of starch granules are crucial in many industrial applications. Starch granules are generally insoluble in cold water, but when heated in excess water, they absorb water and swell. Starches from different genetic backgrounds exhibit characteristic swelling pattern [90–92]. The swelling patterns of some commercial starches are shown in Fig. 2. Potato and tapioca display rapid swelling pattern compared to cereal starches (maize, barley, and wheat). Glucan composition (amylopectin to amylose ratio), granule morphology, fine structure of AP [91, 92] and non-carbohydrate constituents such as protein, monoacylglycerol, phospholipid [90–94] and phosphate monoester [95] have all been shown to influence the swelling pattern of starch granules. Starch granule hydration, swelling, and solubility are also influenced by the extent of starch damage [96] that occurs primarily as a result of milling and starch isolation methods.

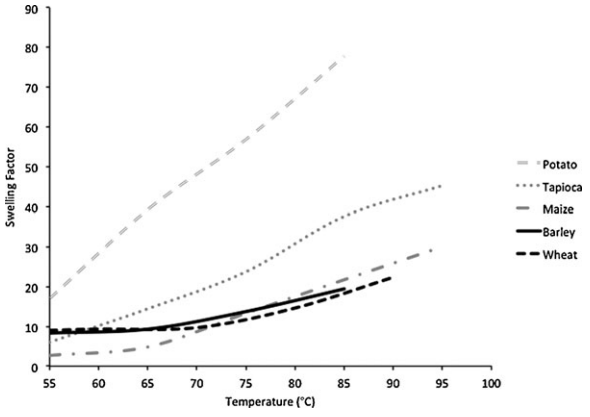

Figure 2. Swelling pattern of starches in the range of 55–90 °C. The swelling factor was determined according to the method employed by Tester and Morrison [92].

Two different methods, namely swelling power, which is the ratio of the wet weight of the sedimented gel to its dry weight [97], and the swelling factor [92], which is the ratio of the volume of swollen granules to the initial volume, have been widely used to measure the degree of swelling.

Tester and Morrison [92] have shown by studies on maize and AM-free maize starches, that swelling is primarily a property of AP. Compared to their regular counterparts, AM-free starches, including waxy potato [98–103], waxy maize [104], and waxy barley [92] exhibit a steeper swelling pattern with a narrow temperature range. However, waxy potato shows a gradual increase of expansion over a wide range of temperature after modification through heat-moisture treatment (100°C, 25% moisture, 16 h) [98, 99] or through cross-linking [100]. Granular swelling of starches from various botanical sources has also been shown to decrease after hydrothermal modifications (heat-moisture treatment and annealing) [105, 106]. These findings indicate that the swelling pattern is governed primarily by structural integrity, which is influenced mainly by the magnitude of the interactions between the glucan chains (AM-AM, AM-AP, AP-AP) within the crystalline and amorphous region and by the packing arrangement of the glucan chains within the crystalline lamellae [107]. Sasaki and Matsuki [108] observed that wheat starches with a higher proportion of long AP chains (DP $\geq$35) are characterized by a greater degree of swelling power. On the other hand, based on their studies of potato and cassava starches, Goman *et al.* [103] have shown that longer AP chains (DP >18) inhibit swelling, whereas short chains (DP <14) promote swelling. It is interesting to notice that the starches studied by Goman *et al.* [103] contained a significant proportion of very short AP chains (DP 6–9). Vamadevan *et al.* [107] postulated that the amount of so-called "fingerprint" A_{fp}-chains (DP 6–8) that are too short to form double helices, the length of the external chains, and the internal organization of AP (especially IB-CL) determine the type and degree of crystalline defects that could facilitate the entry of water into the crystalline lamellae by weakening the double helix packing. Since granular swelling is related to the structural integrity, the swelling pattern cannot be related simply to the proportion of long and short chains of AP. Significant consideration must be given to how different chain categories (e.g. external chains and A_{fp}-chains) and their arrangement influence the crystalline stability and thereby the granular swelling pattern.

AM-extender (*ae*) maize starches [90, 109], high-AM potato starches [90] and pulse starches [91, 105] all exhibit restricted granular swelling. This effect is attributable primarily to the glucan composition, the interaction between the AM/intermediate material and the AP, and the tie chains (AM/intermediate material that pass through the crystalline and amorphous lamellae).

The presence and amount of non-carbohydrate constituents of starches are dependent on the botanical sources and starch isolation methods. The removal of the surface protein and lipid from the starch granules increases the rate of swelling in maize and wheat starch granules but has less effect on starches with a relatively low amount of protein/lipid content (waxy maize, potato, tapioca) [90]. Potato starch shows a greater degree of granular swelling than that from other botanical sources [91]. This is generally attributed to the presence of high levels of phosphate monoesters, which confer the properties of an anionic polyelectrolyte when they are dispersed into aqueous solutions and then repel one another. However, shotii (*Curcuma zedoaria*) starch, which has high phosphate content similar to that of potato starch, is characterized by a lower swelling volume [110, 111]. This disparity again suggests that the packing arrangement of double helices plays a dominant role in granular swelling and raise further questions about the actual effect of the phosphate content on granular swelling and on other thermal properties.

The swelling of starch granules is accompanied by leaching of the glucan components. AM of mainly low molecular weight diffuses out at low temperatures, whereas larger molecules leach at higher temperatures. However, complete exudation of the AM molecules occurs at higher temperatures (> 90°C) [112]. Pérez and Bertoft [113] postulated that larger AM chains, which are not easily leached, may participate in double helices with AP or may be entangled within the intricate architecture of the starch granule. In addition to temperature, other influences that have been demonstrated to affect the extent of AM leaching include (i) the extent of the interaction between glucan chains (AM-AM and/or AM-AP), (ii) the amount of lipid-complexed AM chains, (iii) the phosphate content [105], and (iv) the granule size [114].

5 Gelatinization

Gelatinization is an endothermic reaction that requires an aqueous medium, which acts as a plasticizer. When a starch granule is heated in the presence of moisture, it undergoes a glass transition in the amorphous background prior to the gelatinization, which is an irreversible phase transition [115]. During this transition, the starch granule loses its crystalline order and therefore its birefringence [116]. A number of techniques, including differential scanning calorimetry (DSC), thermo-mechanical analysis (TMA), wide angle X-ray scattering (WAXS), small angle X-ray scattering (SAXS), small angle neutron scattering (SANS), Fourier transform infrared spectroscopy (FTIR), nuclear magnetic resonance spectroscopy (NMR), light scattering, and optical microscopy have been used to monitor the phase transition or structural changes during gelatinization [117–120]. A Kofler hot stage microscope with polarized light has been employed as a means of detecting the loss of birefringence in starch granules [121]. DSC is widely used to detect the phase transition. DSC measures gelatinization transition temperatures (onset [To], melting [Tm], and conclusion [Tc]), and the enthalpy ($\triangle$H) of gelatinization. DSC thermograms of native starches are shown in Fig. 3. Tm is generally used as a measure of crystalline perfection [21], and $\triangle$H represents the rupture of the H-bonds between glucan strands (loss of double helical order) [122]. Generally, a high gelatinization temperature and a narrow endothermic peak suggest a higher molecular order or more stable crystals [107]. This assumption can be rationalized by the event called annealing. When starch is annealed with an excess (>65%) or an intermediate amount of water (40–55%) at a temperature between that of glass transition and the To, the DSC endotherm peak associated with AP gelatinization shifts to a higher temperature and becomes narrower [106, 123]. DSC thermograms of cereal starches reveal an additional endothermic peak at a higher temperature (90–115°C) (Fig. 3), which can be ascribed to the melting of the AM-lipid complex [124].

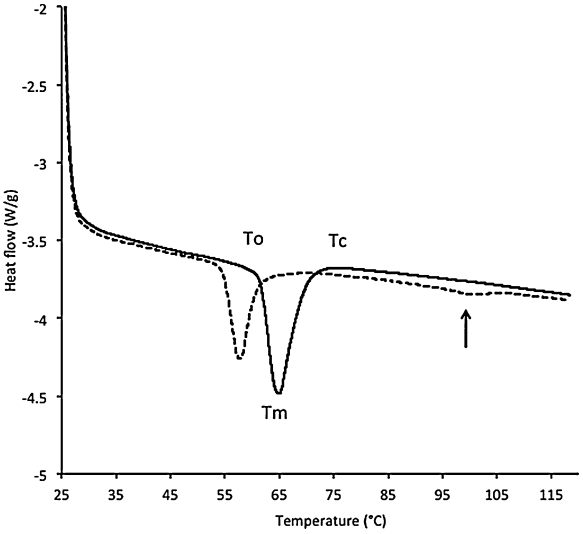

Figure 3. DSC thermograms of barley (--) and potato (—) starches obtained using a TA instruments Q 2000 differential scanning calorimeter. The gelatinization transition temperatures are onset (To), melting (Tm) and conclusion (Tc) temperatures (as indicated for the thermogram of potato starch). The enthalpy of gelatinization (ΔH) was estimated by integrating the area between the thermogram and a baseline over the peak and is expressed as J/g of dry starch. The arrow points to the AM-lipid transition endotherm of barley starch.

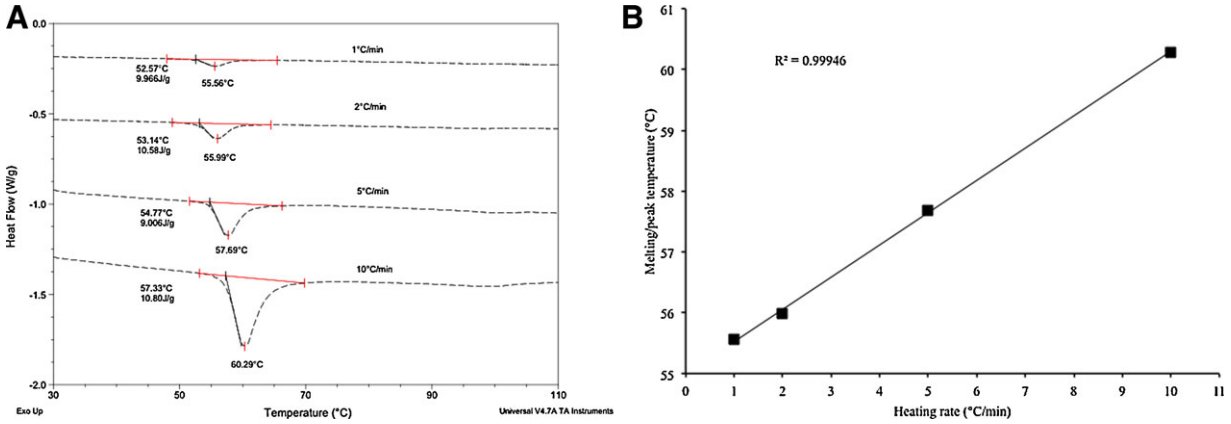

Figure 4. **a.** DSC thermogram of barley starch at four different heating rates obtained using a TA Instruments Q2000 differential scanning calorimeter. Starch dispersions in water (1:3) were equilibrated for 3 h at room temperature before DSC analysis, and the thermogram was then recorded with an empty alodined aluminum pan as a reference. Sensitivity, which is the ability to detect phase transitions in a sample, increased with increasing scanning rates, whereas resolution decreased as the temperature gradient rose. The temperature gradient correlated with delays in the melting temperatures. **b.** Linear relationship between melting temperature (Tm) and heating rate for native barley starch.

It should be noted that the gelatinization transition temperatures obtained using DSC are dependent on the heating rate. A higher heating rate increases the thermal lag, which delays the transition temperatures (Fig. 4a, b). Therefore, a slower heating rate is preferable in order to attain a transition event that is closer to the true thermodynamic event. However, at the slowest heating rate (<2°C), the possibility of annealing also becomes a concern [125]. Water to starch ratio in the system also influence the gelatinization transition temperatures [116]. When the moisture content of the system decreases, the gelatinization endotherm peak shifts to a higher temperature.

Starches from different genetic backgrounds exhibit varied gelatinization temperature attributes (To, Tm, Tc), gelatinization temperature range (Tc-To) and △H (Table 3) [107, 126]. A relationship between average chain length of AP and gelatinization attributes has been reported in numerous studies [103, 126–129]. However, because chain length values *per se* do not represent the crystalline stability of the starch granule, it is highly unlikely that average AP chain length values explain the melting parameters. Instead, gelatinization parameters are more probably influenced primarily by the organization of the glucan chains in the crystalline lamellae of the granules. In the crystalline lamellae, the double helix packing arrangement is determined based on the chain length between the building blocks (IB-CL), the length of the external chains (ECL), the amount of A_{fp}-chains (DP 6–8) [107], and the environmental conditions during the growth period [21, 130].

Interestingly, thermal properties have been shown to be related to the structural type of the amylopectin component in the starch granule [107]: starches with short IB-CL (DP <6) and high amount of A_{fp}-chains have been observed to melt at lower temperatures, whereas starches with long IB-CL (DP >6) and fewer A_{fp}-chains melt at higher temperatures. Vamadevan *et al.* [107] postulated that short IB-CL restricts the parallel alignment of chains within the crystalline lattice and A_{fp}-chains induce crystalline defects, thereby reducing crystalline stability. This hypothesis has been well supported by annealing studies [131], which have indicated a positive correlation between IB-CL and change in enthalpy after

Table 3. Gelatinization parameters of native starches[a]

Starch source	Maize	Wheat	Barley	Tapioca	Potato
To (°C)	62.1±0.2	57.9±0.1	54.6±0.1	62.3±0.2	58.2±0.0
Tm (°C)	68.9±0.1	61.6±0.1	58.0±0.0	67.5±0.1	62.6±0.0
Tc (°C)	75.3±0.1	66.5±0.1	63.4±0.1	75.9±0.1	70.3±0.1
Tc–To (°C)	13.2±0.1	8.6±0.0	8.8±0.1	13.6±0.1	12.2±0.1
ΔH (J/g)	15.7±0.0	9.9±0.1	10.9±0.1	15.7±0.0	18.7±0.3

a To, Tm, and Tc represent onset, mid-point, and conclusion temperatures, respectively; Tc–To = gelatinization temperature range; ΔH = gelatinization enthalpy based on weight dry starch. Values for barley, tapioca and potato are adopted from [131]. Note that the gelatinization of starch is affected by several circumstances, such as environmental conditions and experimental parameters (as illustrated in Fig. 4).

annealing. Starches with long external chains display higher gelatinization enthalpy [107]. This connection seems plausible, since enthalpy reflects the unwinding of the double helices. Overall, these observations are in perfect agreement with the chiral side-chain liquid–crystalline polymer model of starch, in which short side-chains are attached to backbone chains through flexible "spacer" chains [132]. Indeed, this structural model is directly applicable to the building block backbone model of AP discussed above (Fig. 1).

A- and B-type polymorphs, developed from the recrystallization of debranched glycogen chains [122] and spherulites from AM [133] have exhibited different melting behaviours: the A-type polymorph melts at a higher temperature than the B-type. This observation indicates that the A-type polymorph has greater crystalline stability than the B-type. However, the crystalline stability of starch granules cannot be predicted based on the polymorphic pattern, because the packing of the double helices in the crystalline lamellae is influenced primarily by the nature of the branching pattern (distance between the branching points and number of chains in the building blocks) and the length of external chains. In addition to the structural organization, other factors that have been shown to affect the gelatinization parameters are the AM [134] and lipid content [92], and the starch damage [65].

6 Pasting properties

Pasting is the phenomenon that follows gelatinization and involves granular swelling, the exudation of macromolecules, and the eventual disintegration of the starch granules [135]. Starch paste is a mixture of solubilized AM and/or AP, which forms the continuous phase, and granule ghosts and fragments, which represent the discontinuous phase [136].

Rapid Visco Analyser (RVA) and Micro Visco Amylograph (MVAG) are the instruments most commonly employed for measuring the pasting properties of starch. These instruments are configured to simulate the heating and cooling profile of real processes. The technique entails heating a starch-water suspension in a bowl under constant shear applied with a paddle and measuring the changes in viscosity as torque on the paddle. A pasting profile is a record of the change in viscosity as a function of temperature and time [137, 138]. It should be noted that the change in temperature in the sample is delayed compared to that measured by RVA (Fig. 5a). MVAG, however, measures the sample temperature directly, thus providing a temperature for the current viscosity that is more precise than that measured using RVA.

Starches show characteristic pasting curves (Fig. 5b), which provide a multitude of information. The pasting temperature (PT) is the temperature at which a detectable viscosity can be measured: the point at which starch begins to thicken. Peak viscosity (PV) is the highest viscosity level reached during the heating cycle. The viscosity of the starch slurry decreases after the PV due to the continued shearing that occurs during the 95°C holding period, during which highly swollen granules disintegrate. Breakdown is derived by subtracting the trough viscosity (minimum viscosity after holding at 95°C) from the PV; it indicates the stability of the paste during cooking. When the hot paste is cooled down to 50°C, glucan chains begin to reassociate, which increases the viscosity. Setback, which is a measure of the retrogradation and syneresis of the starch upon the cooling of the cooked starch paste, is calculated by subtracting the trough viscosity from the final viscosity.

Studies have shown that the PV of starch is influenced by its AM content [134, 139], the extent of the AM leaching, the

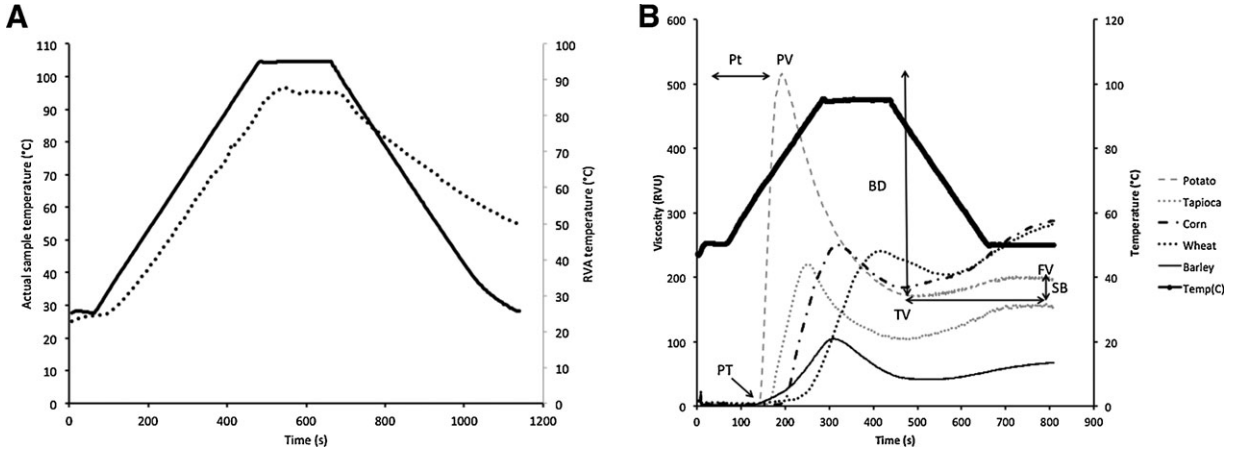

Figure 5. **A.** Comparison of the actual sample temperature (···) and RVA temperature (—). **B.** Pasting profiles of potato, tapioca, corn, wheat, and barley starches obtained using Rapid Visco Analyser (RVA). Pasting temperature (PT), peak time (Pt), peak viscosity (PV), breakdown (BD), trough viscosity (TV), setback (SB), and final viscosity (FV) are indicated in the potato starch profile.

granular swelling [134], the friction between the swollen granules, the phosphate monoester content, and/or the proportion of long AP branch chains [42, 140]. Potato starch displays a higher PV and greater clarity than cereal starches, a difference attributable to the esterified phosphate groups that are present in potato starch AP chains. The presence of phospholipids in cereal starches reduces clarity of the paste [42]. Potato [99] and wheat starches [134] exhibit higher PV and greater setback than do their waxy counterparts. This is generally ascribed to the extensive AM leaching and superior granule integrity in potato and wheat starches, which enable the granules to swell to a greater extent and to attain a higher PV. In contrast, the PV has been shown to be lower and the setback greater in maize starch than in waxy maize [109].

7 Retrogradation

One reason baked products become stale during storage is starch polymer retrogradation [141], which is the recrystallization process of glucan chains in gelatinized starch. During this process glucan molecules dispersed in the paste begin to reassociate through H-bonding [13, 135]. Because linear AM chains can reassociate at a faster rate than can the highly branched AP molecules [142, 143], higher AM/intermediate material content has generally been linked to a greater retrogradation tendency in starches. Investigations have been shown that starches with longer AP chains (potato, pea) retrograde more quickly than those with short AP chains (cereals) [126, 144, 145]. Starch-lipid complexes have a lower tendency toward retrogradation [146, 147]. However, how retrogradation is affected by phosphates that are esterified with AP is unclear [95, 148].

The retrogradation process can be explained as three successive steps: nucleation, propagation and maturation. The rate of retrogradation is dependent on the concentration of the paste, the molecular size, the pH and the temperature [149, 150]. Retrogradation cannot take place in a strong alkaline medium in which glucan chains bear a negative charge and repel one another [151]. The storage temperature has been shown to influence the rate of both nucleation and propagation [152]. During storage, retrogradation drives changes in molecular order, turbidity, crystallinity, gel strength, the gel network, and the intermolecular distance between glucan chains [145, 153]. These changes can be monitored with the use of WAXS, ^{13}C-NMR, DSC, FTIR, and spectrophotometry [13, 154]. Retrograded starch exhibits a B-type crystalline pattern [155], and retrograded AM melts at high temperatures [156] between 130–160°C. Retrograded AP, however, melts at lower transition temperatures than does its native counterpart [157, 158].

8 Freeze-thaw stability

The freeze-thaw stability of starches plays a paramount role in the quality of the sensory attributes of starch-based frozen foods. Studies have shown that freeze-thaw cycles enhance starch retrogradation [159, 160], which is associated with the release of bound water from the glucan polymer network. When a frozen food is thawed, water is readily separated from the matrix. This separation of the gel and water phases leads to syneresis, which causes deterioration of the overall quality of frozen food products. The freeze-thaw stability of starch gels can be evaluated through DSC or by measuring the amount of liquid that has separated from the gel [160].

As discussed above, starches with high AM content have a greater propensity for retrogradation, whereas AM-free starches have less tendency toward this behavior. However, the syneresis of waxy starch gel escalates with an increasing number of freeze-thaw cycles [160, 161]. The freeze-thaw stability of starch has been shown to be improved by chemical substitution [162], which prevents reassociation of the glucan chains, or by alterations in the molecular structure of starch through genetic modification. AM-free potato starch with short AP chains produced by antisense down-regulation of starch synthases (GBSSI, SSII, and SSIII) displayed increased stability after freeze-thaw cycles [163]. Hydrocolloids have also been shown to reduce syneresis and thus improve the freeze-thaw stability of starches, an improvement that can be explained as a result of the water-holding capacity of the hydrocolloids [136].

9 Conclusions

The useful properties of starch are dependent on its granular and molecular structure, the details of which, while largely understood, still continue to be a topic of investigation and debate. Nevertheless, many of the functional properties are well-understood to-date based on the structural characterization of starch. The thermal properties of starch are highly dependent on the organization of the glucan chains, the length and number of each category of chains, and the glucan composition. New insights into the molecular structure of starch components and the architecture of the starch granule can be expected to provide continued opportunities for the increased use of starch.

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