Available online at www.sciencedirect.com
ScienceDirect
Journal of Food Engineering 79 (2007) 970–978
JOURNAL OF FOOD ENGINEERING
www.elsevier.com/locate/jfoodeng

Testing properties of potato starch from different scales of isolations—A ringtest

Bente Wischmann [a,*], Tina Ahmt [b], Ole Bandsholm [c], Andreas Blennow [d], Niall Young [e], Lotte Jeppesen [b], Lars Thomsen [f]

[a] *BioCentrum, DTU, Technical University of Denmark, DK-2800 Kgs. Lyngby, Denmark*
[b] *The Technological Institute, 10 Holbergsvej, DK-6000 Kolding, Denmark*
[c] *KMC, 60 Herningvej, DK-7330 Brande, Denmark*
[d] *Department of Plant Biology, Royal Veterinary and Agricultural University, 40 Thorvaldsensvej, DK-1871 Frb.C, Denmark*
[e] *Danisco A/S, 38 Edwin Rahrs Vej, DK-8220 Brabrand, Denmark*
[f] *Science Park, DK- 8000 Aarhus, Denmark*

Received 3 April 2004; received in revised form 28 September 2005; accepted 17 January 2006
Available online 3 April 2006

Abstract

Five different procedures were used to isolate potato starch from the same batch at three different scales in order to analyse what influence scaling up of the starch process had on the starch physical/chemical properties. Common to the five isolation processes was the steps of washing and maceration of potato tubers followed by separation of starch and cell debris by sieving, filtration or sedimentation. The properties of the processed starch were analysed both in water-based systems of 10 mM NaCl as well as in a milk based food model, dutch vla. Analysis of chemical and physical properties included content of phosphate, protein, ash and dry weight, pH and amylopectin chain length distribution. Other analysis included starch granule size distribution, melting properties by differential scanning calorimetry (DSC), rheological properties by small deformation testing, gel pasting characteristics by a rapid visco analyser (RVA) and freeze/thaw stability and retrogradation characteristics analysed by pulse-NMR. Various rheological flow properties were included in the analysis of the starch samples in the food model. Only the sample of starch isolated in distilled water showed significant difference from the others both when tested in water-based systems and in the food model dutch vla.

Keywords: Potato starch isolation; Ringtest; Granule size distribution; RVA; DSC; Pulse-NMR; Rheology; Food model

1. Introduction

Starch is isolated from numerous tuberous plants as well as cereal grains. It is a relatively cheap raw material with physical and chemical properties making it ideal in many food and non-food applications. Starch functionality has been boosted by the ability to modify starch granules chemically, genetically and enzymatically, but the variation in starch properties is also highly dependent on plant genotype and growth environment (Bay-Smidt, Wischmann, Nielsen, & Møller, 1997; Morrison et al., 2000; Wischmann et al., 2005). A large number of reports therefore aim on a characterisation of the properties of different regular starches. Some of these take into account the variation between plant species (Reddy & Seib, 2000), varieties (Hoover, Smith, Zhou, & Ratnayake, 2003; Sefa-Dedeh & Kofi-Agyir Sackey, 2002; Thitipraphunkul, Uttapap, Piyachomkwan, & Takeda, 2003; Yasui, Seguchi, Ishikawa, & Fujita, 2002) and growth conditions (Cottrell, Duffus, Paterson, & George, 1995; Madsen & Christensen, 1996; Morrison et al., 2000). Many properties have lately been re-examined by use of more advanced techniques or

* Corresponding author. Tel.: +45 45 25 27 74; fax +45 45 88 49 22.
E-mail address: bw@biocentrum.dtu.dk (B. Wischmann).

0260-8774/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.jfoodeng.2006.01.090

have included starches from less common plant sources (Betancur-Ancona, Chel-Guerrero, Bello-Pérez, & Dávila-Ortiz, 2002; Mélo, Stamford, Silva, Krieger, & Stamford, 2003; Sánchez-Hernández, Solorza-Feria, Méndez-Montealvo, Paredes-López, & Bello-Pérez, 2002). The latter may in part be in search of new interesting starch granule properties.

The physical and chemical properties are related to the structural and molecular features of the starch granules. The ratio of amylose to amylopectin, length of debranched amylopectin chains, starch granule size distribution, and phosphate content is among others some of the parameters known to influence the granule properties. In barley some reports state that the physical properties depend more on variety than on granule size (Vasanthan & Bhatty, 1995) but others find a relation between functional properties and granule size distribution due to a difference in amylose content (Tang, Watanabe, & Mitsunaga, 2002), as also found in wheat (Peng, Gao, Abdel-Aal, Hucl, & Chibbar, 1999). Potato starch granules show a unimodal size distribution. Several properties are correlated to the granule sizes as e.g. the viscosity (Bay-Smidt, Blennow, Bojko, & Møller, 1999) and phosphate content (Bay-Smidt, Wischmann, Olsen, & Nielsen, 1994) with corresponding influence on quality and properties of starch blends (Wang, Liu, & Sun, 2003) and food products (Chen, Schols, & Voragen, 2003).

Both morphology and molecular composition of starch granules vary between genotypes. While cereal starches as maize and wheat contain lipids of which the starch associated lipids are phosphorylated, potato starch granules contain no lipids and their phosphate is covalently bound to the glucose residues in the amylopectin molecules of the starch granules. Also potato starches contain hardly any protein compared to cereal starches. These differences have a great impact on the physical properties of the starches and also influence the processes by which starch granules are isolated both in industry and laboratory scale (Haase, Kempf, Tegge, & D'heur, 1987). Non-starch associated lipids in cereal grains are in laboratory processes removed by lipid extraction; in industrial scale via a wet milling step. Rice starch may be isolated successfully from rice flour by a physical disruption of the starch–protein agglomerates by use of various density gradient systems (Guraya, James, & Champagne, 2003). Three isolation methods of pea starches in a laboratory scale were compared in a study of Haase and collages (Haase et al., 1987). The initial incubation in NaOH gave high starch quality, but low yield. A similar NaOH steeping was also included in a study of various isolation procedures of starch from amaranth seeds (Radosavljevic, Jane, & Johnson, 1998). In this study the optimal procedure for starch isolation was a combined NaOH steeping and a protease treatment. In a study comparing barley starch isolation methods, McDonald and Stark (1988) concluded that loss of the fraction of small B starch granules could be avoided by a protease treatment of the starch slurry.

Physical modification of starch can be done by heat-treatment of the granules either at very low humidity and high temperature (i.e. heat-moisture treatment) or in excess water at low temperature for a prolonged period (i.e. annealing). The annealing treatment alters the molecular crystallinity of the granules, which causes enhanced viscosity properties and can be seen as a change in DSC melting profile of the starch (Wischmann & Adler-Nissen, 2002). In all starch processes the starch granules are soaked and washed in water for a period of time. The risk that starch granules are annealed during the process is therefore important to bear in mind.

Acid treatment of starch (i.e. acid thinning) is generally used to lower the viscosity of a starch paste in order to be able to disperse larger amounts of starch without excessive thickening of the paste. Even though the integrity of the granules is retained the intrinsic properties are changed (e.g. Wang, Truong, & Wang, 2003). It is therefore also important to consider the acidity of the water used in the process of starch isolation.

The properties of starches are therefore highly dependent of the history of the starch itself. Many studies of starch granule isolation are at laboratory scale (Andersson, Andersson, & Åman, 2001; Perez, Bahnassey, & Breene, 1993; Vasanthan, Bergthaller, Driedger, Yeung, & Sporns, 1999; Zhu, Haase, & Kempf, 1990). Some of them compare laboratory isolation methods with industrial isolation methods (Vasanthan, Bhatty, Tyler, & Chang, 1997). It is generally appreciated that there is inherent difficulty in scaling up processes. This also includes scaling up of a starch isolation process. It has however in practice not been possible before to address the nature of this problem.

We have previously studied the relation between the physical/chemical properties and molecular structure of potato starches in water-based systems (Blennow et al., 2005; Wischmann et al., 2005) and compared these starch properties with properties in more complex food model systems (Ahmt et al., 2004).

In the present study we have isolated potato starch from one and the same potato batch in four scales, that is a laboratory scale, a small and a medium pilot plant scale and finally in an industrial pilot plant scale. First, the isolated starch samples were characterised in water-based systems and results compared. Secondly, the starch samples were used in the production of a food model, dutch vla, and its rheological properties were tested. It was thus possible to analyse the influence of scaling up the starch isolation process and to see to what extent differences between the isolated starch samples were reflected in final food products.

2. Materials and methods

2.1. Plant material

Potatoes of the variety Kuras was grown in Denmark year 2002, collected by KMC (Brande, Denmark) and

fractions of potatoes collected at random by KMC were immediately distributed to the other collaborators for isolation of starch.

2.2. Starch isolation procedures

Common to all starch isolation procedures any immature or damaged potato tubers were removed and damaged part of tubers were initially cut away. Potato tubers were then rubbed or brushed in water to remove adhering dirt, surface infected skin and infected skin. Sodium bisulphate solution commonly used to inhibit browning was added in two of the processes i.e. *Laboratory b* and *Pilot b*.

Starch isolation method, *Laboratory a*. Potatoes (1 kg) were washed carefully in tap water. The potatoes were cut in smaller pieces, macerated with added tap water in a blender equipped with razor blades and filtered through two layer of gauze. The filtrate was washed extensively with cold tap water, in order to separate starch granules and potato cell debris. The residual containing the starch grains were further washed by several cycles of centrifugation (2000*g*, 10 min) and air dried over night at room temperature.

Starch isolation method, *Laboratory b*. Potatoes (1 kg) were washed carefully in tap water, dried and processed through a juice presser (Moulinex). The residual is filtrated through a sieve (mesh 125 μm) with addition of 1 L tap water removing cell wall material. To the residual starch slurry (final volume 2 L) is added 2 mL 38–40% sodium bisulphate solution and the slurry stands to settle for 1/2 h. The pellet of starch is washed two times in 1 L tap water and allowed to stand for 1/2 h. Finally the starch is dried at room temperature on filter paper over night.

Starch isolation method, *small pilot plant scale, Pilot a*. Potatoes (10 kg) were cleaned by an extensive wash in tap water. The potatoes were thereafter macerated in a Quadro Comill (model 194AS) by use of a series of sieves (meshes: 6350 μm, 812.8 μm, 475.2 μm and 228.6 μm) and rinsing with a total of 40 L of tap water. The starch was separated from the macerated potato slurries in a small hydrocyclone battery consisting of 14 hydrocyclones of which the 10 were blocked. The slurry was passed over the hydrocyclones in three cycles after which the starch was concentrated by centrifugation and dried at room temperature over night.

Starch isolation method, *medium pilot plant process, Pilot b*. Potatoes (100 kg) were disintegrated with a grater and the rasping was collected on a 250 μm screen. Starch was flushed through the screen with distilled water until the starch stream ebbed away. Discolouring of the juice was hampered by an immediate addition of 20 mL, 1% bisulphite solution to the crude starch milk. A series of sequential sedimentation, suspension and sieving was performed in order to remove potato cell debris. Starch was allowed to sediment, the supernatant was decanted and distilled water (three times the sediment volume) was added. The starch was brought in suspension by stirring, passed through a 125 μm screen flushed with distilled water. The slurry was collected in an Imhoff cone, allowed to rest for 3–5 min after which the supernatant was decanted leaving 5–10% of the starch in the cone. The supernatant was filtered through a 75 μm screen. The filtrate was combined with the residual sediment of the cone and the sedimentation and wash was repeated twice. The final sediment was approximately 25 Baumé (Bé°), where Baumé modulus 145: Bé° = 145 − 145/specific gravity at 60 °F (Cleland, Fauser, & Fetzer, 1943). Concentrations of large starch slurry's where achieved on a hydrocyclone battery to a concentration of not more that 21 Bé° in order to be able to pump the slurry.

Starch isolation method, *factory pilot plant process, Industry*. Fresh potatoes were carefully cleaned before starch extraction. Soils were removed on rotating bar screens and adhering impurities were removed by intensive washing with tap water. High-speed raspers open all cells in the potato tissue and the starch granules and juice were separated from cell walls on rotating conical screens using undiluted juice as flushing medium. The crude starch milk was washed on multi-stage hydrocyclones in counter current with tap water added to the last stage. The resulting purified starch milk had a concentration and viscosity of 22 Bé°. Most of the water was removed on a rotating vacuum filter and the dewatered filter cake was dried in a stream of hot air in a flash dryer. The dried starch and drying air were separated on cyclones, the starch was cooled in a stream of cold air and the cooled dried starch was screened on centrifugal screens.

2.3. Physical/chemical and structural analysis

pH was measured in starch slurries of 25 g starch in 50 mL distilled water, dry weight was determined using a Mettler weight equipped with heating device, ash content measured according to ISO 3593 and total protein using Kjeldahl, ISO 3188. Granule size distribution of starch samples was measured by use of a Malvern Mastersizer 2000 (Malvern Instr. Ltd., Malvern, UK). Starch bound phosphate was determined as released Glc-6-P after acid hydrolysis following the method of Bay-Smidt et al. (1994). Determination of amylose was done colorimetrically as described in Bay-Smidt et al. (1999). The preparation of debranched amylopectin chains and formation of chain length distribution of debranched amylopectin was carried out as described by Blennow, Bay-Smidt, Wischmann, Olsen, and Møller (1998). Solubilised starch was incubated with isoamylase (Megazyme, Sidney, Australia) and the obtained linear debranched amylopectin chains were separated on a Dionex DX 500 system. Peaks were integrated and corrected for detector response factors as described earlier (Blennow et al., 1998). The thermal properties of the native starch samples under dilute conditions were analysed by differential scanning calorimetry (DSC) using a Seiko DSC220 calibrated with indium and operated from 25 °C to 100 °C at 5 °C/min. All starch samples were analysed in slurries of 2 mg sample and 8 μl 10 mM NaCl in triplicates as described in Wischmann et al. (2005).

Freeze/thaw stability describes how well the starch gel is able to resist exudation of water by molecular reorganisation. As described in Wischmann et al. (2005) the freeze/thaw stability of the starch samples measured at 20 °C after 1, 2 or 3 cycles of freeze/thaw (−18 °C to +20 °C) was estimated using low field pulsed NMR spectrometry (Bruker Minispec NMS 120) operating at 20 MHz—0.47 T. The NMR method is based on the principle that the signals from protons in the "solid-like" state decay at a quicker rate than the protons in a liquid state, following a 90° pulse, and is described in detail by Teo and Seow (1992). It is thus a measure of the amount of dry matter. As molecular association or recrystallisation proceeds during aging of a starch gel, the proportion of the "solid-like" component of the system increases. With the same measurement technique the extent of retrogradation was measured at 5 °C at day 1, day 3 and day 7. Measurements were carried out with six replicates, respectively using 5% starch on dry weight basis suspended in 10 mM NaCl (Wischmann et al., 2005).

2.4. Rheological analysis

Pasting profiles of the starches were acquired using a Rapid Visco Analyser (RVA) model 4 (Newport Scientific, Warriewood, Australia). The starch concentration was 4% starch on dry weight basis in slurries of 10 mM NaCl. A programmed heating and cooling cycle was used, where the sample was held at 50 °C for 1 min, heated to 95 °C in 31/2 min, held at 95 °C for 21/2 min, cooled to 50 °C in 4 min and finally held at 50 °C for 2 min. Paddle stirring speed was 160 rpm. Peak viscosity, Through 1, Final viscosity, Setback, Peak time and Pasting time were recorded.

Oscillatory measurements were performed using a Stress Tech controlled stress rheometer (Reologica AB, Lund, Sweden). The measurement geometry used was a 4 cm plate–plate. All measurements were carried out in triplicates at 25 °C on 5% starch gels in 10 mM NaCl. Oscillatory measurements were carried out within the linear viscoelastic region, over a frequency range of 0.1–10.0 Hz. From the data, the storage modulus, G'_{lin}, the loss modulus G''_{lin} and the phase angle, δ at 1 Hz were calculated. Finally the slope of the plot of log G' versus log frequency was calculated and used to quantify the gel strength character of the samples.

2.5. Production and rheological analysis of vla

Vla was manufactured as described in Wischmann, Norsker, and Adler-Nissen (2002) and Ahmt et al. (2004), in 2000 mL triplicate batches using a Stefan mixer (Stefan UMC5 electronic, Hameln, Germany) connected to a heating bath circulating glycerol at 120 °C and a cooling bath circulating water at 0 °C. Each batch of vla contained on dry weight basis 0.70% starch, 12.00% full milk powder, 8.00% sugar, 2.80% modified waxy maize starch and 0.04% carrageenan. All ingredients were heated to 92 °C and cooled to 22 °C while stirring continuously. Products were kept at 5 °C for further analysis. Texture analysis was performed with a Back Extrusion setup on a TA-XT2 texture analyser (Stable Micro Systems, Godalming, UK) equipped with a 25 mm plunger, which was pressed 85% into the sample contained in a 40 mL beaker (40 mm diam.) at a speed of 3 mm/s. The extrusions force (Max Force) and the corresponding extrusion work (Extrusion Work) was quoted.

Stress sweep, frequency sweep and viscosity were performed as described above for the water-based starch gels. The flow curves obtained over a shear range of 1–300 s^{-1} and a time period of 115 s were fitted to the power law equation: $\eta = K\dot{\gamma}^{(n-1)}$ from which the consistency index, K and the power law index, n were calculated. Spreadability analysis was performed using a USDA consistometer with a concentrical cylinder, on which the diameter of the sample was read after 1 min. All rheological analyses were performed in triplicates on each of the three batches of vla after 7 days incubation of the product at 5 °C.

2.6. Data treatment

The computer program Statgraphics plus v4.1 (Manugistics Inc., Rockville, Maryland, USA) was used to analyse the data by two-factor (sample, replication) analysis of variance (ANOVA), When significant differences were revealed ($P < 0.001$, $P < 0.05$), mean scores were compared by multiple comparison using Duncan's multiple range test ($P < 0.05$).

3. Results and discussion

3.1. Properties of starch in water-based systems

Starch was isolated from potatoes of the same variety and batch, but at different scale ranging from laboratory scale of approximately 1 kg fresh weight (FW) potatoes (samples Lab. a and Lab. b), to pilot plant isolations from approximately 10 kg FW potatoes (sample Pilot a) and 100 kg FW potatoes (sample Pilot b), to finally the large industrial scale of approximately 1 tons FW potatoes (sample Industry). The starch isolation processes varied primarily in the method used for maceration, filtration and concentration, but the most striking difference turned out to be the use of distilled water for sample Pilot b as opposed to tap water for the rest of the samples. Following the starch isolations the properties of the starch samples were analysed. The data on the chemical analysis of the five potato starch samples are shown in Table 1. Whereas the content of phosphate does not vary significantly between the samples as expected the pH is surprisingly low in sample Pilot b. This very low pH is likely to be a result of the isolation and washing of this starch sample in distilled water rather than in tap water as the rest of the presented starch samples.

Fig. 1 shows the distribution of the length of debranched amylopectin chains. Surprisingly one of the samples (Pilot

Table 1
Chemical analyses of the five starch samples showing phosphate content determined as nmol Glucose-6-P per mg starch; dry matter, protein, and ash in percent of total and pH

Sample	Phosphate[a] [nmol Glc6P/mg starch]	Dry matter[b] [%]	Protein[b] [%]	Ash[b] [%]	pH[b] [–]
Lab. a	21.8 ± 0.8	82.9	0.169	0.45	7.1
Lab. b	22.5 ± 1.2	84.2	0.104	0.47	6.6
Pilot a	22.6 ± 0.8	86.7	0.121	0.45	7.7
Pilot b	22.4 ± 0.4	82.6	0.064	0.38	4.8
Industry	21.4 ± 0.1	81.8	0.171	0.43	7.4

Number of replicates is indicated.
[a] Mean ± SD, $n = 2$.
[b] Mean, $n = 2$.

b) contained a higher relative amount of short chains compared to the other samples. This may be the result of the low pH of this sample (Table 1) that may have led to a partial acid hydrolysis of the starch.

One of the difficulties in isolating starch in a laboratory scale is to ensure that none of the smallest granules are lost during the thorough wash of the starch slurry (McDonald & Stark, 1988). As is seen in the granule size distribution plot (Fig. 2) the starch sample isolated by the industry (Industry) contain a higher volume percent of the smallest and the largest granules, respectively and therefore have a more flat distribution curve. The starch sample Lab. a shows the most narrow size distribution of starch granules.

The melting properties were analysed by DSC in 10 mM NaCl, such that all five starch samples had the same ionic strength. The low pH of sample Pilot b is therefore not reflected in the DSC results (Table 2). All five samples contain the same amount of covalently bound phosphate (see Table 1) and as expected the DSC profiles therefore do not differ much from each other. The width of the DSC peak [T_c and T_o] is a measure of the crystallinity of the starch granules. No difference in width was found between samples, which indicate either that none of them has undergone any annealing during processing or they are all annealed to the same extent. There is however significant difference ($P < 0.001$) between the parameters T_m and ΔH (Table 2), which may be explained partly by the difference in granule size distribution found between the samples. Fig. 3 shows the RVA pasting profiles of the starch samples. As is seen the sample Pilot b has a significant lower peak viscosity as well as pasting profile in general compared to the other samples. The sample shows a pasting profile as of an "acid thinning" starch, which are low viscosity starches that are acid treated.

The gelling properties of starch ingredients are crucial for their application in food products. All five samples were analysed in 5% gels in 10 mM NaCl by non-destructive oscillatory measurements. The storage modulus G'_{lin} were for all samples approximately three times higher than the corresponding loss modulus, G''_{lin} (Table 4), which is typical for normal potato starch (Blennow et al., 2005). Taken together with the rather small values of slope G'_{lin} the results indicate that the starch gels had the network forming characteristics expected for normal potato starch. The starch samples were all significantly different from each other, but the difference could not be assigned to one single starch sample or parameter. Two of the samples, Pilot a and Pilot b were however significantly different from the rest of the samples considering G''_{lin} and slope G'_{lin}, respectively, as indicated by the ranking in Table 4. The results of the phase angle in Table 4 furthermore shows that the starch samples Pilot b and Industry are significantly different from each other and from the rest of the samples.

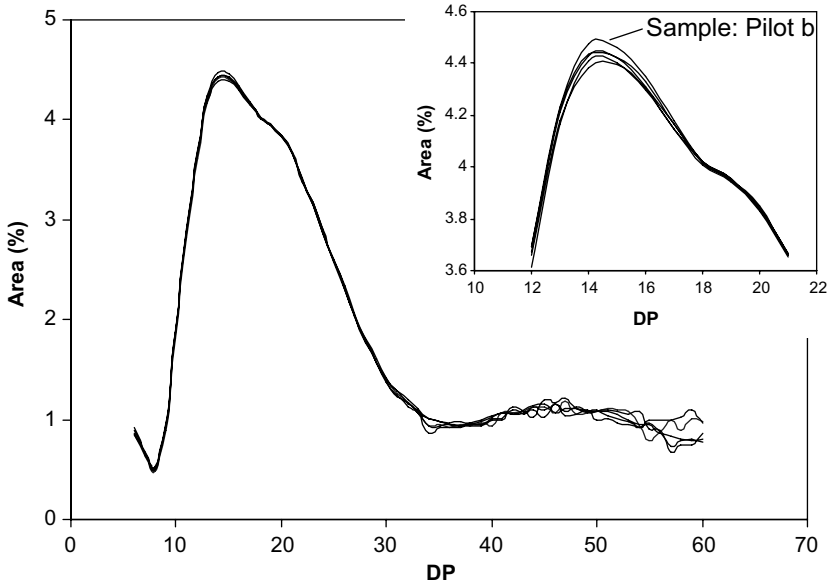

Fig. 1. Amylopectin chain length distribution of the starch samples Lab. a, Lab. b, Pilot a, Pilot b and Industry, isolated from the same potato batch. The sample with significant higher number of smaller amylopectin chain lengths is indicated in the figure (Pilot b).

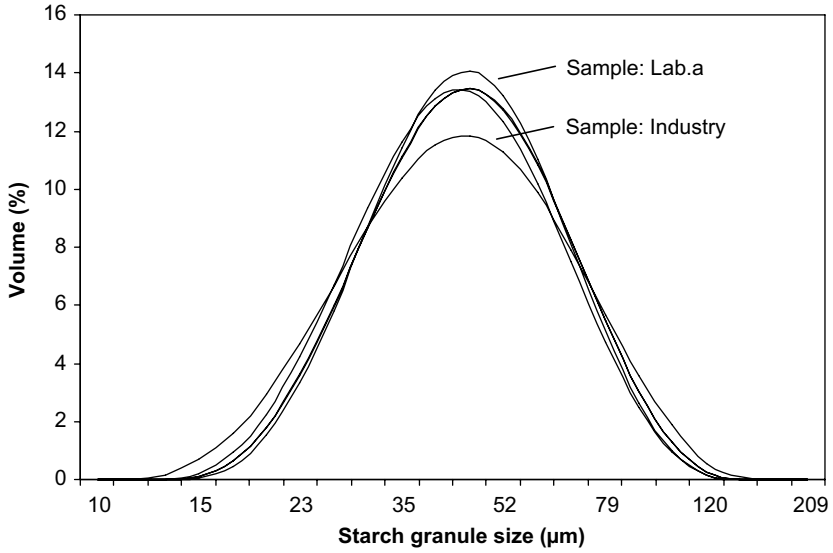

Fig. 2. Starch granule size distribution of the starch samples, Lab. a, Lab. b, Pilot a, Pilot b and Industry. The two samples with distinguishable different size distribution profile are indicated in the figure (Lab. a and Industry).

Table 2
DSC gelatinisation parameters showing the onset temperature (T_o), mid temperature (T_m), completion temperature (T_c) and enthalpy change (ΔH)

Sample	T_o [°C]	T_m [°C]	T_c [°C]	$T_c - T_o$ [°C]	ΔH [J/g]
Lab. a	60.67 ± 1.68	62.87 ± 0.23^{ab}	67.50 ± 0.70	4.63 ± 0.65	15.64 ± 1.41^{abc}
Lab. b	59.70 ± 0.53	63.43 ± 0.32^{bc}	69.57 ± 0.23	6.13 ± 0.25	16.90 ± 0.86^{bc}
Pilot a	59.85 ± 0.30	63.23 ± 0.10^{abc}	68.08 ± 1.00	4.85 ± 1.06	15.14 ± 5.63^{a}
Pilot b	60.20 ± 0.20	63.67 ± 0.12^{c}	69.17 ± 1.20	5.50 ± 1.21	18.19 ± 2.4^{c}
Industry	58.13 ± 0.76	62.60 ± 0.17^{a}	67.47 ± 0.32	4.87 ± 0.42	15.58 ± 0.26^{ab}

Number of replicates is indicated.
Mean $\pm$ SD, $n = 3$.
[a,b,c] Indicates the significant ranging of measurements at $P < 0.05$.

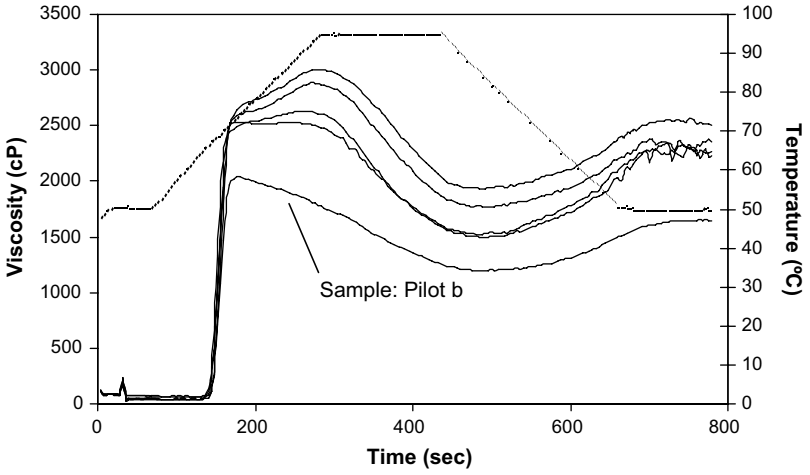

Fig. 3. RVA pasting profiles of the five starch samples, Lab. a, Lab. b, Pilot a, Pilot b and Industry. The starch sample with the lowest peak viscosity and pasting profile is indicated in the figure (Pilot b).

By low field pulse NMR it is possible to estimate the stability of starch gels upon repeated cycles of freezing and thawing as well as the degree of retrogradation with time. Table 3 shows the results from these analyses and as expected from previous work (Wischmann et al., 2005) a rise was found in relative dry matter both after up to three cycles of freezing at −18 °C and thawing at +20 °C and after up to 7 days stand at 5 °C. Unfortunately both the

Table 3
Estimation by pulse-NMR of freeze–thaw stability (F/T) after 0, 1, 2 and 3 cycles of freeze/thaw (−18 °C to +20 °C) as well as estimation of retrogradation at 5 °C (Retr.) measured at day 1, 3 and 7

Sample	F/T 0 cycles [%]	F/T 1 cycles [%]	F/T 2 cycles [%]	F/T 3 cycles [%]	Retr. day 1 [%]	Retr. day 3 [%]	Retr. day 7 [%]
Lab. a	1.26 ± 0.27	3.35 ± 0.31	4.57 ± 0.33	4.95 ± 0.13	1.18 ± 0.22	1.41 ± 0.25	1.67 ± 0.24^{b}
Lab. b	1.56 ± 0.28	4.04 ± 0.32	4.59 ± 0.33	5.07 ± 0.40	0.86 ± 0.29	1.06 ± 0.32	1.27 ± 0.29^{a}
Pilot a	1.33 ± 0.21	4.27 ± 0.39	4.43 ± 0.33	5.25 ± 0.33	1.26 ± 0.23	1.50 ± 0.15	1.78 ± 0.24^{b}
Pilot b	1.31 ± 0.19	3.56 ± 0.12	4.28 ± 0.18	4.66 ± 0.52	1.20 ± 0.28	1.51 ± 0.34	1.75 ± 0.18^{b}
Industry	1.39 ± 0.11	3.64 ± 0.29	4.77 ± 0.21	4.89 ± 0.26	1.25 ± 0.39	1.26 ± 0.37	1.60 ± 0.40^{b}

Number of replicates is indicated.
Mean ± SD, $n = 6$.
[a,b] Indicates the significant ranging of measurements at $P < 0.05$.

Table 4
Oscillation parameters of 5% starch gels in 10 mM NaCl measured at 25 °C: storage modulus G'_{lin}, loss modulus G''_{lin}, phase angle δ and slope of $\log G'_{lin}$ versus log frequency at 1 Hz, slope G'_{lin}

Sample	G'_{lin} (Pa)	Slope G'_{lin} (Pa s)	G''_{lin} (Pa)	Phase angle δ (°)
Lab. a	52.95 ± 3.58^{ab}	0.189 ± 0.003^{a}	16.05 ± 0.90^{a}	16.07 ± 0.20^{a}
Lab. b	47.68 ± 6.22^{bc}	0.194 ± 0.012^{ab}	14.88 ± 1.49^{a}	17.38 ± 0.55^{a}
Pilot a	63.60 ± 6.70^{a}	0.212 ± 0.009^{bc}	20.72 ± 1.37^{b}	18.10 ± 0.76^{a}
Pilot b	37.39 ± 8.12^{c}	0.257 ± 0.010^{d}	15.85 ± 3.03^{a}	23.06 ± 0.68^{c}
Industry	43.00 ± 7.96^{bc}	0.220 ± 0.013^{c}	15.73 ± 1.36^{a}	20.32 ± 1.86^{b}

Number of replicates is indicated.
Mean ± SD, $n = 3$.
[a,b,c,d] Indicates the significant ranging of measurements at $P < 0.05$.

replicates and the samples were significantly different from each other at cycle 1 and cycle 2 making it impossible to make conclusions after these cycles of freezing and thawing. After three cycles, however no difference was found either between replicates or between the five starch samples. The analysis of degree of retrogradation showed as indicated by the ranking in Table 3, that one of the laboratories samples Lab. b, was different from the rest of the samples after day 7. Half of the replicates from these analyses were however insignificantly different from each other, which makes this interpretation vague.

3.2. Properties of starch in a food model, vla

Table 5 shows the results from the rheological analyses of the food model prepared with the five starch samples are shown. One of the five samples differed from the rest of the samples. The Duncan multiple range test showed that starch sample Pilot b was significantly different from the rest of the samples for six out of the eight rheological parameters ($P < 0.05$ for Extrusion Work, G'_{lin}, Max Force, n_up, K_up and n_down). Vla prepared with starch sample Pilot b was firmer with higher values for storage modulus G'_{lin}, consistency index (K_up), Max Force and Extrusion Work. This is possibly a direct effect of a slight hydrolysis taking place during the isolation process due to the low pH giving shorter chains with a higher tendency to retrograde during cold storage of vla.

The physical and chemical properties of the starch sample Pilot b is thus reflected in the food product model, even though this product consists of a complex food matrix of full milk, sugar and carrageenan. The concentration of the potato starch samples in the food model is furthermore as low as 0.7% based on dry matter and the food model product contains additionally 2.8% modified waxy maize starch.

In previous work the effect of chemical and genetic modification on the functional properties of starch were analysed in gels and in a vla (Ahmt et al., 2004). Here as expected, chemical modification of potato starch by substitution, which dramatically changes the molecular structure of the starch strongly affected the functional properties of both starch gels as well as the rheological and sensory properties of vla prepared with these starches. But also slight alteration of the amylopectin chain length and phosphorous content in potato starch by genetic modification affected the functional properties of starch gels, and gave

Table 5
Rheological properties determined in the food model dutch vla produced with the five different potato starch samples

Sample	Storage modulus G'_{lin} [Pa]	Consistency index K_up [Pa s^n]	Power law index n_up [–]	Consistency index K_down [Pa s^n]	Power law index n_down [–]	Spreadmeter [mm]	Max Force [N]	Extrusion Work [N/a]
Lab. a	72.5 ± 0.6^{a}	25.1 ± 0.3^{a}	0.3 ± 0.0^{a}	20.2 ± 0.0	0.3 ± 0.0^{a}	49.8 ± 0.5	40.9 ± 0.9^{a}	689.6 ± 17.7^{a}
Lab. b	73.5 ± 1.4^{a}	24.9 ± 0.8^{a}	0.3 ± 0.0^{a}	19.9 ± 0.9	0.3 ± 0.0^{a}	50.3 ± 0.5	41.5 ± 0.9^{a}	705.6 ± 15.5^{a}
Pilot a	70.5 ± 2.1^{a}	24.6 ± 1.3^{a}	0.3 ± 0.0^{a}	19.7 ± 1.1	0.3 ± 0.0^{a}	50.1 ± 1.0	40.0 ± 1.6^{a}	680.2 ± 31.4^{a}
Pilot b	88.4 ± 1.3^{b}	28.6 ± 0.7^{b}	0.3 ± 0.0^{b}	19.9 ± 0.5	0.3 ± 0.0^{b}	49.2 ± 0.9	48.5 ± 1.7^{b}	834.6 ± 38.4^{b}
Industry	75.3 ± 4.9^{a}	25.7 ± 0.5^{a}	0.3 ± 0.0^{a}	20.4 ± 0.5	0.3 ± 0.0^{a}	49.8 ± 0.1	41.8 ± 0.5^{a}	705.1 ± 12.8^{a}

Number of replicate is indicated.
Mean ± SD, $n = 3$.
[a,b] Indicates the significant ranging of measurements at $P < 0.05$.

detectable changes in the rheological and sensory properties of vla. The results from these earlier findings and the present work confirm the role of starch as a tool to design different textures in food and stress the importance of knowing the history of the food ingredients used.

4. Conclusion

In this work the difficulty of scaling up isolation process of potato starch was addressed. The degree to which starch properties revealed in water-based systems would be reflected in the rheology of real food systems was studied. The starch samples were characterised physically, chemically and structurally. Granule size distribution reflected some of the mechanical differences in starch isolation procedures. The industrially isolated starch contained a relatively higher volume percent of the smallest and largest granules whereas the sample with the most narrow size distribution was isolated in a laboratory scale.

One of the starch samples was outstandingly different from the others. This sample (Pilot b) was the only one processed in distilled water. This starch sample had an acidic pH, contained a higher number of short chains in the amylopectin molecules and had a lower peak viscosity as well as general pasting profile. It was concluded that a mild but detectable acid hydrolysis had taken place in this starch sample. Only the properties of this starch sample could be reflected in the rheological properties of the food model vla. No significant difference was found between the other samples. The importance of the history of the starch samples is therefore stressed.

Acknowledgements

Nancy Kjøbæk, Anne-Marie Nepper and Rene Thrane are thanked for excellent technical support. This work is part of the research project ''Centre for development of new starches'' financed by The Danish Research Council, and supported by The Danish Directorate for Development (Centre for Development of Improved Food Starches) and The Committee for Research and Development of the Öresund Region (Öforsk).

References

Ahmt, T., Wischmann, B., Blennow, A., Madsen, F., Bandsholm, O., & Thomsen, J. (2004). Sensory and rheological properties of transgenic and chemically modified starch ingredients as evaluated in a food product model. *Nahrung/Food, 48*(2), 149–155.

Andersson, A. A. M., Andersson, R., & Åman, P. (2001). Starch and by-products from a laboratory-scale barley starch isolation procedure. *Cereal Chemistry, 78*(5), 507–513.

Bay-Smidt, A. M., Blennow, A., Bojko, M., & Møller, B. L. (1999). The effect of phosphate and amylopectin molecular size on peak viscosity of starch pastes. *Annual Transactions of the Nordic Rheology Society, 7*, 31–38.

Bay-Smidt, A. M., Wischmann, B., Nielsen, T. H., & Møller, B. L. (1997). Phosphorylation of potato starch. In P. L. Frazier, P. Richmond, & A. M. Donald (Eds.), *Starch—Structure and Functionality* (pp. 266–267). Cambridge: Royal Society of Chemistry.

Bay-Smidt, A. M., Wischmann, B., Olsen, C. E., & Nielsen, T. H. (1994). Starch bound phosphate in potato as studied by a simple method for determination of organic phosphate and ^{13}P-NMR. *Starch/Stärke, 46*, 167–172.

Betancur-Ancona, D. A., Chel-Guerrero, L. A., Bello-Pérez, L. A., & Dávila-Ortiz, G. (2002). Isolation of velvet bean (*Mucuna pruriens*) starch: physicochemical and functional properties. *Starch/Stärke, 54*, 303–309.

Blennow, A., Bay-Smidt, A. M., Wischmann, B., Olsen, C. E., & Møller, B. L. (1998). The degree of starch phosphorylation is related to the chain length distribution of the neutral and the phosphorylated chains of amylopectin. *Carbohydrate Research, 307*, 45–54.

Blennow, A., Wischmann, B., Houborg, K., Ahmt, T., Jørgensen, K., Engelsen, S. B., et al. (2005). Structure function relationships of transgenic starches with engineered phosphate substitution and starch branching. *International Journal of Biological Macromolecules, 36*, 159–168.

Chen Schols, Z. H. A., & Voragen, A. G. J. (2003). Starch granule size strongly determines starch noodle processing and noodle quality. *Journal of Food Science, 68*(5), 1584–1589.

Cleland, J. E., Fauser, E. E., & Fetzer, W. R. (1943). Baume-dry substance tables for starch suspensions. *Industrial and Engineering Chemistry, 15*(5), 334–336.

Cottrell, J. E., Duffus, C. M., Paterson, L., & George, R. M. (1995). Properties of potato starch: effects of genotype and growing conditions. *Phytochemistry, 40*, 1057–1064.

Guraya, H. S., James, C., & Champagne, E. T. (2003). Physical basis for separation of rice starch using various density gradient systems and its effect on starch recovery, purity, and pasting properties. *Starch/Stärke, 55*, 450–456.

Haase, N. U., Kempf, W., Tegge, G., & D'heur, U. (1987). Comparative investigations on the wet-separation of pea starch. *Starch/Stärke, 39*(12), 416–421.

Hoover, R., Smith, C., Zhou, Y., & Ratnayake, R. M. W. S. (2003). Physicochemical properties of Canadian oat starches. *Carbohydrate Polymers, 52*, 253–261.

Madsen, M., & Christensen, D. H. (1996). Change in viscosity properties of potato starch during growth. *Starch/Stärke, 48*, 245–249.

McDonald, A. M. L., & Stark, J. R. (1988). A critical examination of procedures for the isolation of barley starch. *Journal of the Institute of Brewing, 94*, 125–132.

Mélo, E. M., Stamford, T. L. M., Silva, M. P. C., Krieger, N., & Stamford, N. P. (2003). Functional properties of yam bean (*Pachyrhizus erosus*) starch. *Bioresource Technology, 89*, 103–106.

Morrison, I. M., Cochrane, M. P., Cooper, A. M., Dale, M. F. B., Duffus, C. M., Ellis, R. P., et al. (2000). Potato starches: variation in composition and properties between three genotypes grown at two different sites and in two different years. *Journal of the Science of Food and Agriculture, 81*, 319–328.

Peng, M., Gao, M., Abdel-Aal, E.-S. M., Hucl, P., & Chibbar, R. N. (1999). Separation and characterisation of A- and B-type starch granules in wheat endosperm. *Cereal Chemistry, 64*, 361–375.

Perez, E., Bahnassey, Y. A., & Breene, W. M. (1993). A simple laboratory scale method for isolation of amaranth starch. *Starch/Stärke, 45*(6), 211–214.

Radosavljevic, M., Jane, J., & Johnson, L. A. (1998). Isolation of amaranth starch by diluted alkaline-protease treatment. *Cereal Chemistry, 75*(2), 212–216.

Reddy, I., & Seib, P. A. (2000). Modified waxy wheat starch compared to modified waxy corn starch. *Journal of Cereal Science, 31*, 25–39.

Sánchez-Hernández, L., Solorza-Feria, J., Méndez-Montealvo, G., Paredes-López, O., & Bello-Pérez, L. A. (2002). Isolation and partal characterization of Okenia (*Okania hypogaea*) starch. *Starch/Stärke, 54*, 193–197.

Sefa-Dedeh, S., & Kofi-Agyir Sackey, E. (2002). Starch structure and some properties of cocoyam (*Xanthosoma sagittifolium* and *Colocasia esculenta*) starch and raphides. *Food Chemistry, 79*, 435–444.

Tang, H., Watanabe, K., & Mitsunaga, T. (2002). Structure and functionality of large, medium and small granule starches in normal and waxy barley endosparms. *Carbohydrate Polymers, 49*, 217–224.

Teo, C. H., & Seow, C. C. (1992). A pulsed NMR method for the study of starch retrogradation. *Starch/Stärke, 44*(8), 288–292.

Thitipraphunkul, K., Uttapap, D., Piyachomkwan, K., & Takeda, Y. (2003). A comparative study of edible canna (*Canna edulis*) starch from different cultivars. Part I. Chemical composition and physicochemical properties. *Carbohydrate Polymers, 53*, 317–324.

Vasanthan, T., Bergthaller, W., Driedger, D., Yeung, J., & Sporns, P. (1999). Starch from Alberta potatoes: wet-isolation and some physicochemical properties. *Food Research International, 32*, 355–365.

Vasanthan, T., & Bhatty, R. S. (1995). Starch purification after pin milling and air classification of waxy, normal, and high amylase barleys. *Cereal Chemistry, 72*(4), 379–384.

Vasanthan, T., Bhatty, R. S., Tyler, R. T., & Chang, P. (1997). Isolation and cationization of barley starches at laboratory and pilot scale. *Cereal Chemistry, 74*(1), 25–28.

Wang, Y.-J., Liu, W., & Sun, Z. (2003). Effects of granule size and shape on morphology and tensile properties of LDPE and starch blends. *Journal of Materials Science Letters, 22*, 57–59.

Wang, Y.-J., Truong, V.-D., & Wang, L. (2003). Structures and rheological properties of corn starch as affected by acid hydrolysis. *Carbohydrate Polymers, 52*, 327–333.

Wischmann, B., & Adler-Nissen, J. (2002). Melting properties and lintnerisation of potato starch with different degrees of phosphorylation. *Starch/Stärke, 54*, 499–507.

Wischmann, B., Blennow, A., Madsen, F., Jørgensen, K., Poulsen, P., & Bandsholm, O. (2005). Functional characterisation of potato starch modified by specific in planta alteration of the amylopectin branching and phosphate substitution. *Food Hydrocolloids, 19*(6), 1016–1024.

Wischmann, B., Norsker, M., & Adler-Nissen, J. (2002). Food product models developed to evaluate starch as a food ingredient. *Nahrung/Food, 46*(4), 167–173.

Yasui, T., Seguchi, M., Ishikawa, N., & Fujita, M. (2002). Starch properties of a waxy mutant line of hull-less barley (*Hordeum vulgare* L.). *Starch/Stärke, 54*, 179–184.

Zhu, J. H., Haase, N. U., & Kempf, W. (1990). Investigations on the laboratory scale separation of mung bean starch. *Starch/Stärke, 42*(1), 1–4.