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Effect of heat–moisture treatment on the structure and physicochemical properties of tuber and root starches

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Abstract

Starch from tubers potato (*Solanum tuberosum*), taro (*Alocassia indica*), new cocoyam (*Xanthosoma sagitifolium*), true yam (*Dioscorea alata*), and root cassava, (*Manihot esculenta*) crops was isolated and its morphology, composition and physicochemical properties were investigated before and after heat–moisture treatment (HMT) (100 °C, for 10 h at a moisture content of 30%). Native starch granules were round to oval to polygonal with smooth surfaces. The granule size (diameter) ranged from 3.0 to 110 μm.The total amylose content ranged from 22.4 to 29.3%, of which 10.1–15.5% was complexed by native lipid. The phosphorus content ranged from 0.01 to 0.1%. The X-ray pattern of potato and true yam was of the 'B'-type. Whereas, that of new cocoyam and taro was of the 'A'-type. Cassava exhibited a mixed 'A + B'-type X-ray pattern. The relative crystallinity, swelling factor (SF), amylose leaching (AML), gelatinization temperature range and the enthalpy of gelatinization of the native starches ranged from 30 to 46, 22 to 54, 5 to 23%, 13 to 19 °C and 12 to 18 J/g, respectively. Susceptibility of native starches towards hydrolysis by 2.2N HCl and porcine pancreatic α-amylase were 60–86% (after 12 days), and 4–62% (after 72 h), respectively. Retrogradation was most pronounced in the B-type starches. Granule morphology remained unchanged after HMT. The X-ray pattern of the B-type starches was altered (B → A + B) on HMT. However, that of the other starches remained unchanged. HMT decreased SF, AML, gelatinization enthalpy and susceptibility towards acid hydrolysis, but increased gelatinization temperatures and enzyme susceptibility. Extent of retrogradation and relative crystallinity decreased on HMT of true yam and potato starches, but remained unchanged in the other starches. The foregoing data showed that changes in physicochemical properties on HMT are influenced by the interplay of crystallite disruption, starch chain associations and disruption of double helices in the amorphous regions.

Keywords: Tuber and root starches; Heat–moisture treatment

1. Introduction

Heat–moisture treatment of starches is defined as a physical modification that involves treatment of starch granules at low moisture levels (<35% moisture w/w) during a certain time period (15 min–16 h) and at a temperature (84–120 °C) above the glass transition temperature (T_g) but below the gelatinization temperature. Under the above conditions, changes in X-ray pattern, crystallinity, starch chain interactions, granule swelling, AML, viscosity, gelatinization parameters, retrogradation, acid and enzyme hydrolysis have been shown to occur in cereal (Donovan, Lorenz, & Kulp, 1983; Franco, Ciacco, & Tavares, 1995; Fukui & Nikuni, 1969; Hagiwara, Esaki, Kitamura, & Kuge, 1991; Hoover & Manuel, 1996; Kawabata, Takase, Miyoshi, Sawayama, Kimura, & Kudo, 1994; Kobayashi, 1993; Lorenz & Kulp, 1981, 1982, 1983; Maruta et al., 1994; Radosta, Kettlitz, Schierbacum, & Gernat, 1992; Sair, 1967; Schierbaum & Kettliz, 1994; Takaya, Sano, & Nishinari, 2000), tuber (Abraham, 1993; Collado & Corke, 1999; Collado, Mabesa, Oates, & Corke, 2001; Donovan et al., 1983; Hoover & Vasanthan, 1994; Hoover, Vasanthan, Senanayake, & Martin, 1994; Kobayashi, 1993; Kuge & Kitamura, 1985; Lorenz & Kulp, 1981, 1982; Sair, 1967; Schmiedel, Konig, & Jacobasch, 1998; Stoof, Schmiedl, Anger, Rehbrucke & Bergthaller, 1998; Stute, 1992) and legume (Hoover & Vasanthan, 1994; Hoover, Swamidas, & Vasanthan, 1993) starches. Changes to starch structure and properties on heat–moisture treatment have been found to vary with starch source. For instance, tuber starches have been shown to be more susceptible than legume or cereal starches towards heat–moisture treatment (Hoover & Vasanthan, 1994; Jacobs & Delcour, 1998). Most of the studies on heat–moisture treated (HMT) tuber and root starches have been on potato and sweet potato starches.

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Thus, it is difficult to ascertain whether changes observed during heat–moisture treatment of the above starches are truly representative of tuber and root starches. Furthermore, tuber and root starches exhibit different types of unit cell structures (A, B, A + B) (Hoover, 2001). Consequently, the magnitude of starch chain realignment and/or interactions during heat–moisture treatment may vary widely among these starches.

The objective of this study, was therefore to examine changes to starch structure and physicochemical properties on heat–moisture treatment (under identical time/temperature/moisture combinations) of some selected tuber and root starches. This study is of significance, since there is a growing interest in physical modification of starches for food and non-food applications.

2. Materials and methods

2.1. Materials

Dioscorea alata (true yam) and *Alocassia indica* (taro), *Manihot esculenta* (cassava), *Solanum tuberosum* (potato) and *Xanthosoma sagitifolium* (new coco yam) were grown on experimental plots (under identical environmental conditions). Crystalline porcine pancreatic α-amylase (EC 3.2. 1.1, type 1A), was purchased from Sigma Chemical Co., (St. Louis, MO, USA). All other chemicals and solvents were of ACS certified grade. Solvents were distilled from glass before use.

2.2. Methods

2.2.1. Starch isolation

All tubers and roots were divided into two lots representing whole sample. Each lot was subdivided into two lots. Each sub lot was further subdivided into two sub lots. Starch was extracted and purified using the procedure of Hoover and Hadziyev (1981).

2.2.2. Granule morphology

Granule morphology of native starches was studied by scanning electron microscopy. Starch samples were mounted on circular aluminum stubs with double sticky tape and then coated with 20 nm of gold and examined and photographed in a Hitachi (S 570) scanning electron microscope (Nissei Sangyo Inc., Rexdale, ON, Canada) at an accelerating potential of 20 kV.

2.2.3. Proximate analysis

Quantitative estimation of moisture, ash, nitrogen, and starch were performed by the standard AACC methods (1984). Starch lipids were determined by the procedure outlined by Vasanthan and Hoover (1992). Total phosphorus content were determined by the method of Morrison (1964).

2.2.4. Amylose content

Apparent and total amylose content was determined by a modification (Hoover & Ratnayake, 2001) of the method of McGrance, Cornell, and Rix (1998).

2.2.4.1. Apparent amylose content. Starch (20 mg, db) was dissolved in 90% dimethylsulfoxide (8 ml) [DMSO] in screw-cap reaction vials. The contents of the vials were vigorously mixed for 20 min and then heated in a water bath (with intermittent shaking) at 85 °C for 15 min. The vials were then cooled to ambient temperature and the contents were diluted with water to 25 ml in a volumetric flask. One millilter of diluted solution was mixed with water (40 ml) and 5 ml 1_2/KI solution (0.0025 M KI) and then adjusted to a final volume of 50 ml. The contents were allowed to stand for 15 min at ambient temperature, before absorbance measurements at 600 nm (Hoover & Ratnayake, 2001)

2.2.4.2. Total amylose content. The total amylose content of starch samples were determined by the above procedure, but with prior defating with hot *n*-propanol–water (3:1 v/v) for 7 h. In order to correct for over estimation of apparent and total amylose content (due to complex formation between iodine and the long outer branches of amylopectin), amylose content was calculated from a standard curve prepared using mixtures of pure potato amylose and amylopectin (over the range 0–100% amylose and amylopectin 100–0%)

2.2.5. Starch fractionation

Amylose and amylopectin were extracted from tuber and root starches utilizing the aqueous leaching procedure described by Montgomery and Senti (1958). The purity of isolated amylose and amylopectin were determined by the method of Jane and Chen (1992) using gel permeation chromatography (Sepharose CL-2B) chromatography.

2.2.6. Degree of polymerization (DP) of amyloses

Isolated amylose (0.01 g) was completely dissolved in 10 ml of DMSO by heating at 60 °C in a water bath. The resulting solution was divided into two equal volumes and the degree of polymerization (DP) was calculated (Jane & Robyt, 1984) using the equation shown below:

$$\mathrm{DP} = \frac{\text{Total carbohydrate } (\mu\text{g})}{\text{Reducing sugar (as } \mu\text{g of maltose)}} \times 2$$

Total carbohydrate and total reducing power were calculated according to the procedures outlined by Bruner (1964) and Dubois, Gilles, Hamilton, Rebers, and Smith (1956), respectively. Maltose (Fisher Scientific, Fair Lawn, NJ, USA) was used to prepare the standard curve for estimation of total carbohydrate and reducing power.

2.2.7. Fine structure of amylopectins

The β amylolysis limit of isolated amylopectins was determined by the method of Wursch and Gumy (1994).

The number average chain length (Cl_n) of amylopectins were determined by assaying the reducing power after debranching with *pseudomonas* isoamylase (Shi & Seib, 1992). The number average chain length (Cl_n) was calculated as 2 × weight of carbohydrate (μg)/reducing sugar (as μg of maltose). The average exterior chain length (ECL) and the average interior chain length (ICL) were calculated (Manners, 1989) as shown below:

$$\mathrm{ECL} = \bar{\mathrm{C}}\mathrm{l}_n \times (\%\beta\text{-limit}/100) + 2$$

$$\mathrm{ICL} = \bar{\mathrm{C}}\mathrm{l}_n - \mathrm{ECL} - 1$$

2.2.8. X-ray diffraction

X-ray diffractograms of the starches were obtained with a Rigaku RU X-ray diffractometer (Rigaku-Denki Co., Tokyo, Japan) with operating conditions as: target voltage 40 kV, current 100 mA, aging time 5 min, scanning range 3–350, scan speed 2.000°/min, step time 4.5 s, divergence slit width 1.00, scatter slit width 1.00 and receiving slit width 0.60. Relative crystallinity of starches was calculated using the method of Nara, Mori, and Komiya (1978) using a peak-fitting software (Origin-version 6.0, Microcal Inc., Northampton, MA, USA). Amorphous starch was prepared by heating a 10% starch solution at 95 °C for 30 min with continuous agitation and then drying it at 100 °C for 24 h. The dried starch sample was then ground into a free flowing powder.

2.2.9. Swelling factor

The SF of the starches (50 mg db) when heated to 50–90 °C in water (5 ml) was determined according to the method of Tester and Morrison (1990). The SF was reported as the ratio of the volume of swollen starch granule to the volume of the dry starch. This method measures only intragranular water and hence the true SF at a given temperature.

2.2.10. Amylose leaching

Starch (20 mg, db) in water (10 ml) was heated (50–90 °C) in volume calibrated sealed tubes for 30 min. The tubes were then cooled at ambient temperature and centrifuged at 2000*g* for 10 min. The supernatant liquid (1 ml) was withdrawn and its amylose content was determined as described by Hoover and Ratnayake (2001).

2.2.11. Gelatinization parameters

Gelatinization parameters were measured using a Seiko DSC (Seiko Instrument Inc., Chiba, Japan) differential scanning calorimeter equipped with a thermal analysis data station and data recording software. Water (11 μl) was added with a microsyringe to starch (3.0 mg) in the DSC pan, which were then sealed, reweighed and allowed to stand for 2 h at room temperature in order to attain an even distribution of water. The scanning temperature range and the heating rates were 20–120 °C and 10 °C/min, respectively. In all measurements, the thermogram was recorded with an empty aluminum pan as the reference. The transition temperatures reported are the onset (T_o), peak (T_p), and conclusion (T_c). The enthalpy of gelatinization (ΔH) was estimated by integrating the area between the thermogram and a base line under the peak and was expressed in terms of joules per unit weight of dry starch (J/g).

2.2.12. Differential scanning calorimetry of retrograded starch

Water (3 μl) was added with a microsyringe to starch (3.0 mg) in DSC pans, which were then sealed, reweighed an allowed to stand for 2 h at room temperature for moisture equilibration. The sealed pans were then heated to 120 °C at 10 °C/min to gelatinize the starch. The gelatinized samples were stored at 4 °C for 24 h (to increase nucleation), and then at 40 °C for seven days (to increase propagation). Subsequently, the samples were equilibrated at room temperature for 2 h, and then rescanned in the calorimeter from 20 to 120 °C at 10 °C/min to measure retrogradation transition temperatures and enthalpy.

2.2.13. Enzymatic hydrolysis with α-amylase

Enzymatic hydrolysis were done using a crystalline suspension of porcine pancreatic α-amylase in 0.5 M saturated sodium chloride containing 3 mM calcium chloride, in which the concentration of α-amylase was 30 mg/ml, and the specific activity was 790 units per milligram of protein. Hydrolysis was carried out for 6, 12, 24 and 72 h. Details of the procedure were as outlined by Hoover and Vasanthan (1994).

2.2.14. Acid hydrolysis

The starches were hydrolyzed with 2.2N HCl at 35° (1.0 g starch/40 ml acid) for 12 days. The starch slurries were shaken by hand daily to resuspend the deposited granules. At 24 h intervals, aliquots of the reaction mixtures were neutralized and recentrifuged (2000*g*) and the supernatant liquid was assayed for total carbohydrate (Dubois et al., 1956). The extent of hydrolysis was determined by expressing the solubilized carbohydrates as a percentage of the initial starch.

2.2.15. Heat–moisture treatment

Starch (15 g, db) were weighed into glass containers. Starch moisture content was brought to 30%. The sealed samples (in glass jars) were heated in a forced air oven (Fisher Scientific, Isotemp 615g), USA) at 100 °C for 10 h. After cooling, the jars were opened and starch samples were air-dried to a moisture content of ~10%.

2.2.16. Statistical analysis

All determinations were replicated three times and mean values and standard deviations reported. Analysis of variance (ANOVA) were performed and the mean separations were done by Tukey's HSD test ($p < 0.05$) using

Table 1
Chemical composition (%) and some properties of tuber and root starches

Characteristic	Composition (%)[a]				
	True yam	Taro	New cocoyam	Cassava	Potato
Moisture	10.2 ± 0.20^{p}	$11.2 \pm 0.10^{q,r}$	11.3 ± 0.03^{r}	13.5 ± 0.24^{s}	$13.1 \pm 0.15^{t,s}$
Ash	$0.12 \pm 0.01^{p,q}$	0.14 ± 0.01^{q}	$0.15 \pm 0.02^{r,p,q}$	$0.11 \pm 0.02^{s,q,p}$	0.25 ± 0.01^{t}
Nitrogen	$0.05 \pm 0.01^{p,t,r}$	$0.03 \pm 0.01^{q,p}$	0.08 ± 0.03^{r}	$0.02 \pm 0.01^{s,p,q}$	$0.09 \pm 0.02^{t,r}$
Lipid					
Solvent extracted					
Chloroform–methanol[b]	$0.02 \pm 0.01^{p,r,t,s}$	$0.08 \pm 0.02^{q,r}$	0.09 ± 0.04^{r}	$0.06 \pm 0.02^{s,r,q,t}$	$0.08 \pm 0.01^{t,r,q}$
n-Propanol–water[c]	$0.01 \pm 0.00^{p,s,q}$	$0.04 \pm 0.01^{q,s}$	0.30 ± 0.05^{r}	$0.08 \pm 0.03^{s,t}$	0.12 ± 0.02^{t}
Acid hydrolyzed[d]	0.03 ± 0.01^{p}	$0.15 \pm 0.01^{q,t,s}$	0.40 ± 0.03^{r}	0.12 ± 0.02^{s}	0.20 ± 0.04^{t}
Amylose content					
Apparent[e]	24.6 ± 0.30^{p}	26.1 ± 0.21^{q}	22.3 ± 0.12^{r}	19.8 ± 0.20^{s}	25.2 ± 0.22^{t}
Total	$28.5 \pm 0.70^{p,q}$	29.3 ± 0.62^{q}	26.4 ± 0.20^{r}	22.4 ± 0.13^{s}	$28.1 \pm 0.50^{t,p,q}$
Amylose complexed with lipid[f]	$10.1 \pm 0.40^{p,q,t}$	$10.9 \pm 0.30^{q,s}$	15.5 ± 0.60^{r}	11.6 ± 0.32^{s}	$10.4 \pm 0.30^{t,q}$
Phosphorus	0.03 ± 0.01^{p}	$0.02 \pm 0.01^{q,p}$	$0.02 \pm 0.00^{r,p,q}$	$0.01 \pm 0.00^{s,p,q,r}$	0.1 ± 0.05^{t}
Starch damage	$1.23 \pm 0.20^{p,t}$	$0.85 \pm 0.30^{q,p,r}$	$1.20 \pm 0.02^{r,p}$	0.25 ± 0.01^{s}	$1.5 \pm 0.04^{t,r}$
Granule shape	Oblong to oval	Round to variable	Polygonal to variable	Round to variable	Oval to spherical
Granule size (diameter) (μm)	12–100	10–50	3.0–10	5.0–45	10–110

[a] All data reported on dry basis and represent the mean of three determinations. Means within a row with different superscripts are significantly different ($p < 0.05$).

[b] Lipids extracted from chloroform–methanol 2:1 (v/v) at 25 °C (mainly unbound lipid).

[c] Lipids extracted by hot *n*-propanol–water 3:1 (v/v) from the residue left after chloroform–methanol extraction (mainly bound lipid).

[d] Lipids obtained by acid hydrolysis (24%) HCl of the native starch (total lipid).

[e] Apparent and total amylose determined by I_2-binding before and after removal of bound lipids, respectively.

[f] (Total amylose − apparent amylose/Total amylose)100.

Sigmastat Version 2.0 (Jandel Scientific/spss science, Chicago,IL, USA).

3. Results and discussion

3.1. Morphological granular characteristics

The starch granules ranged from oval to round to spherical to polygonal in shape with characteristic dimensions in the range 3–110 μm (Table 1). On the basis of starch granule size, new cocoyam had a very large surface area per unit weight compared to that of the other starches. The granule surface of all starches appeared to be smooth and showed no evidence of pin holes under the scanning electron microscope. Heat–moisture treatment did not alter the size or shape of the starch granules. Similar observations have been made on HMT maize (Hoover & Manuel, 1996) and wheat (Hoover & Vasanthan, 1994) starches.

3.2. Chemical composition

The data on chemical composition are presented in (Table 1). The purity of the starches was judged on the basis of composition (low nitrogen and low ash level) and microscopic observation (absence of any adhering protein). The low nitrogen content was in the range 0.01–0.09%. The low nitrogen content indicated the absence of non-starch lipids (lipids associated with endosperm protein). Therefore, the total lipids (0.03–0.4%) (Table 1) obtained by acid hydrolysis mainly represent free and bound starch lipids (Vasanthan & Hoover, 1992). The total lipid content was in the range reported for most tuber and root starches (0.1–1.14%) (Hoover, 2001). The free lipid obtained by extraction with chloroform–methanol (2:1 v/v at 25 °C) was in the range 0.02–0.09% of the total weight. The bound lipid content obtained by extraction of the chloroform–methanol residue with *n*-propanol–water (3:1 v/v for 7 h) was in the range 0.01–0.3% (Table 1). The apparent amylose content (determined by I_2 before removal of bound lipid) was in the range 19.8–26.1% (Table 1). The total amylose content (determined by I_2 binding after removal of bound lipids by *n*-propanol–water was in the range 22.4–29.3% (Table 1). A comparison of the apparent and total amylose content (Table 1) showed that the percentage of total amylose complexed by native starch lipids ranged from 10.1 to 15.5% (Table 1). New cocoyam differed from the other starches in exhibiting a significantly higher content of total lipid and a much higher proportion of lipid complexed amylose chains (Table 1). The phosphorus content ranged from 0.01 to 0.10% (Table 1). Potato contained more phosphorus (0.10%) than any of the other starches (0.01–0.03%) (Table 1). The extent of starch damage (0.25–1.50%) was low in all starches (Table 1).

3.3. Molecular structure

The average degree of polymerization (DP_n) of isolated

Table 2
Structure of amylose and amylopectins of tuber and root starches (means within a column with different superscripts are significantly different ($p < 0.05$))

Starch source	Amylose[a]	Amylopectin			
	DP$_n$[b]	CL$_n$[c]	ECL[d]	ICL[e]	β amylolysis (%)
True yam	1800 ± 45^{a}	29.0	18.5	9.5	57 ± 4^{a}
Taro	2200 ± 60^{b}	26.1	16.6	8.4	56 ± 2^{a}
New cocoyam	2775 ± 59^{c}	24.2	15.8	7.4	57 ± 2^{a}
Cassava	2500 ± 62^{d}	24.5	16.2	7.3	58 ± 2^{a}
Potato	4850 ± 75^{e}	28.1	18.6	8.5	59 ± 3^{a}

[a] Amylose from all starches were free of amylopectin (confirmed by gel permeation chromatography [GPC]). The yeild of amylose (by GPC) was 26.6, 28.8, 26.0 and 26.7%, respectively, in true yam, taro, new cocoyam, cassava, and potato.

[b] Number average degree of polymerization.

[c] Average chain length.

[d] (External chain length = $\bar{\mathrm{Cl}}_n \times \beta$ amylolysis limit (%)/100) + 2.

[e] Internal chain length = $(\mathrm{Cl}_n - \mathrm{ECL}) - 1$.

amylose followed the order: potato > new cocoyam > cassava > taro > true yam. The DP$_n$ of potato (4850) and cassava (2500) was close to the values reported by Takeda, Shiraska, and Hizukuri (1984). The DP$_n$ of the other starches have not been reported previously, and therefore, no comparisons are possible. The average chain length ($\bar{\mathrm{Cl}}_n$) of isolated amylopectins followed the order: Potato ~ true yam > taro ~ new cocoyam ~ cassava. The $\bar{\mathrm{Cl}}_n$ of potato (28) and cassava (24) were close to reported values (Hizukuri, 1985; Suzuki, Takeda, & Hizukuri, 1985). The $\bar{\mathrm{Cl}}_n$ of the other starches have not been reported previously, and therefore, no comparisons are possible. The average ECL of amylopectin followed the order: potato > true yam > taro > new cocoyam > cassava. The corresponding order for the average ICL was: true yam > taro > potato > new cocoyam > cassava (Table 2).

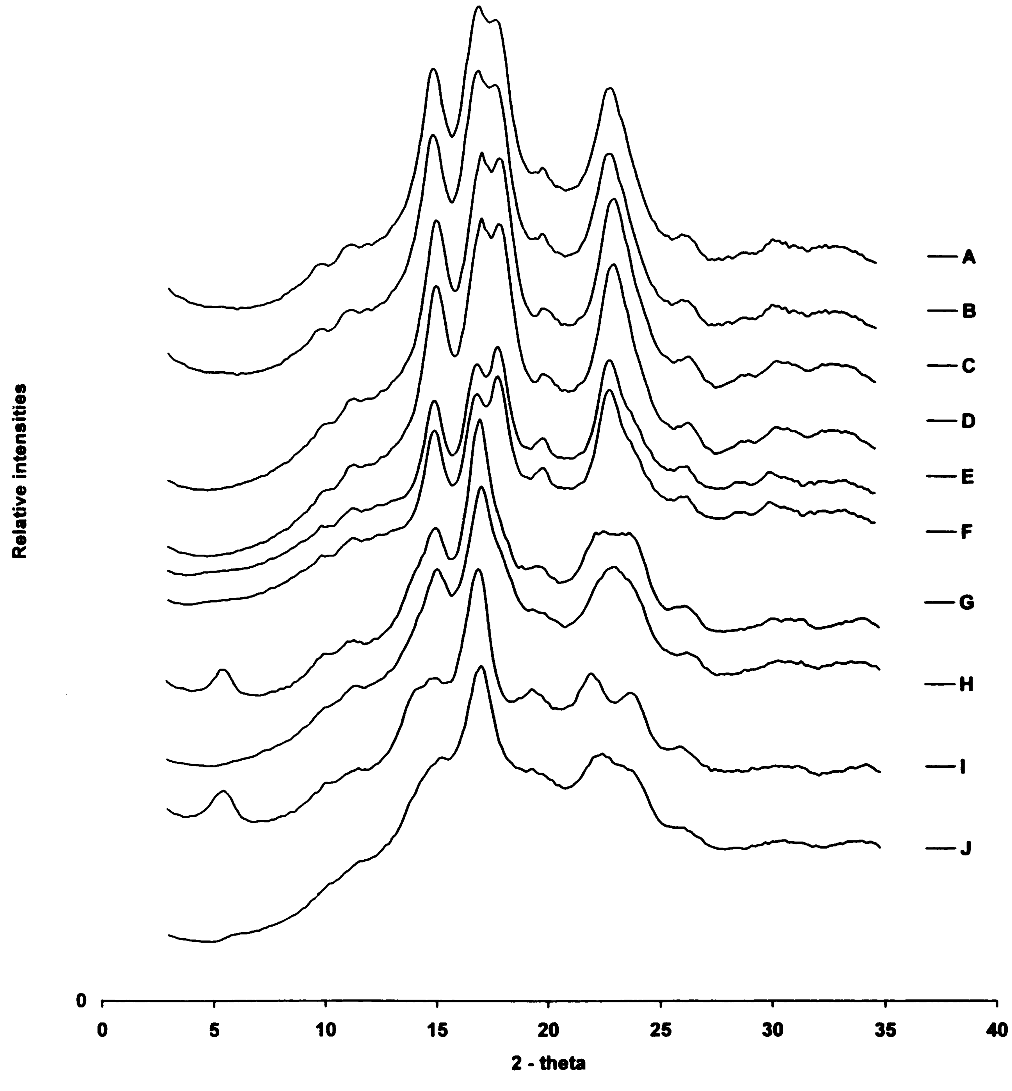

Fig. 1. X-ray diffraction patterns of native and HMT tuber and root starches (10 h, 30% moisture, 100 °C). (A) native *M. esculenta*, (B) HMT *M. esculenta*, (C) native *A. indica*, (D) HMT *A. indica*, (E) native *X. sagitifolium*, (F) HMT *X. sagitifolium*, (G) native *D. alata*, (H) HMT *D. alata*, (I) native *S. tuberosum*, (J) HMT *S. tuberosum*.

Table 3
X-ray pattern and relative crystallinity (%) of native and heat–moisture treated tuber and root starches (means within a column with different superscripts (for native starch and its heat–moisture treated counterpart) are significantly different ($p < 0.05$))

Starch source and treatment[a]	X-ray pattern	Relative crystallinity[b] (%)
True yam		
Native	B	32.0 ± 0.2^{p}
HMT[c]	A + B	23.0 ± 0.8^{q}
Taro		
Native	A	31.0 ± 0.5^{p}
HMT[c]	A	30.0 ± 0.1^{q}
New cocoyam		
Native	A	45.0 ± 0.3^{p}
HMT[c]	A	43.5 ± 0.2^{q}
Cassava		
Native	A	37.0 ± 0.5^{p}
HMT[c]	A	36.0 ± 0.1^{q}
Potato		
Native	B	30.0 ± 0.7^{p}
HMT[b]	A + B	22.0 ± 0.4^{q}

[a] Moisture content ~16% (w/w).

[b] Relative crystallinity $= \sum|I_s - I_a| / \sum|I_c - I_a| \times 100$, where $I_s - I_a =$ difference between the sample and morphous intensities and $I_c - I_a +$ difference between the crystallinity (quartz) and amorphous intensities.

[c] Heat–moisture treated (100 °C, 30% moisture, 10 h).

3.4. X-ray pattern and crystallinity

Potato and true yam showed the typical B-type X-ray pattern (Zobel, 1988) with reflection intensities at 5.5, 17, and 22–24°2θ angle (Fig. 1). However, the other three starches showed a A-type X-ray pattern (Fig. 1). Both A and B-type starches are based on parallel standard double helices, in which the helices are more closely packed in the A-type starch. Furthermore, they also differ in the content of intra-helical water (B > A) (Imberty, 1988; Imberty, Chanzy, Perez, Buleon, & Tran, 1988). The type of crystalline polymorph has been shown (Hizukuri, Takeda, Yasuda, & Suzuki, 1981) to be influenced by the chain length (CL) of amylopectin (A-type $CL < 19.7$, B-type $CL \geq 21.6$), growth temperature (Hizukuri, Fujii, & Nikuni, 1961) and fatty acids (Hizukuri, 1996). The relative crystallinity (Table 3) of potato (30%) and cassava (31%) were comparable to the values reported by Zobel (1988) for the above two starches. However, the relative crystallinity of new cocoyam (45%) was much larger than the value reported (24%) by Takeda, Takeda, and Hizkuri (1983). The differences in relative crystallinity (Table 3) among the starches probably represent differences in: (1) crystallite size, (2) orientation of double helices (within the crystallite), (3) average chain length of amylopectin (Cl_n), and (4) mole percentage of the short chain fraction of amylopectin (DP 10-13).

Heat–moisture treatment changed the X-ray pattern (B → A + B) of both potato and true yam starches (Fig. 1). A shift from B to A crystal type as a result of heat–moisture treatment was confirmed by ^{13}CCP/MAS NMR based on variations in C-1 multiplicity (Gidley & Bociek, 1985). The X-ray pattern of taro, cassava and new cocoyam, however, remained unchanged on heat–moisture treatment (Fig. 1). The relative crystallinity of potato and true yam decreased by 9 and 8%, respectively after heat–moisture treatment (Table 3). However, the relative crystallinity of the other starches remained practically unchanged (Table 3).

Imberty and Perez (1989) and Imberty et al. (1988) have shown that double helices of A and B-type starches are packed in a pseudohexagonal array. The lattices of B-type starches have a large void (channel) in which 36 water molecules can be accommodated. However, in A-type starches, the lattices contain a helix in the center rather than a column of water. In both A and B-type starches, there is a spacing of double helices that corresponds to 1.1 nm distance between the axes of the two double helices. The change in X-ray pattern (B → A + B) on heat–moisture treatment can be attributed to: (1) dehydration (vaporization

Table 4
Swelling factor (starches (50 mg) were heated with water (5 ml) at the specified temperatures for 30 min) of native and heat–moisture treated tuber and root starches (means within the same column with different superscripts (for native starch and its heat–moisture treated counterpart) are significantly different ($P < 0.05$))

Starch Source	Treatment	Temperature (°C)				
		50	60	70	80	90
True yam	Native	0.0	0.0^{p}	5.3 ± 0.3^{p}	26 ± 0.2^{p}	33 ± 0.1^{p}
	HMT[a]	0.0	1.2 ± 0.2^{q}	3.3 ± 0.2^{q}	7.9 ± 0.4^{q}	19.5 ± 0.6^{q}
Taro	Native	0.0	2.5 ± 0.1^{p}	8.5 ± 0.2^{p}	36 ± 0.1^{p}	34.2 ± 0.0^{p}
	HMT[a]	0.0	0.0^{q}	6.0 ± 0.5^{q}	13.4 ± 0.2^{q}	23.3 ± 0.4^{q}
New cocoyam	Native	0.0	5.0 ± 0.2^{p}	10.6 ± 0.3^{p}	18.0 ± 0.6^{p}	22 ± 0.62^{p}
	HMT[a]	0.0	4.0 ± 0.5^{q}	9.2 ± 0.4^{q}	12.6 ± 0.1^{q}	13.1 ± 0.2^{q}
Cassava	Native	0.0	4.6 ± 0.2^{p}	31.0 ± 0.7^{p}	43.0 ± 0.4^{p}	36.5 ± 0.2^{p}
	HMT[a]	0.0	2.1 ± 0.1^{q}	16.4 ± 0.3^{q}	24.5 ± 0.2^{q}	23.5 ± 0.5^{q}
Potato	Native	0.0	37.6 ± 0.4^{p}	57.4 ± 0.5^{p}	60.0 ± 0.5^{p}	54.0 ± 0.3^{p}
	HMT[a]	0.0	10.2 ± 0.2^{q}	17.2 ± 0.6^{q}	18.5 ± 0.8^{q}	16.4 ± 0.4^{q}

[a] Heat–moisture treated (100 °C, 30% moisture,10 h).

Table 5
Amylose leaching (starches (20 mg) were heated with water (10 ml) at the specified temperatures for 30 min) (%) of native and heat–moisture treated tuber and root starches (means within the same column with different superscripts (for native starch and its heat–moisture treated counterpart) are significantly different ($P < 0.05$))

Starch Source		Temperature				
		50 (°C)	60 (°C)	70 (°C)	80 (°C)	90 (°C)
True yam	Native	0.0	0.0	1.1 ± 0.5^{p}	13.0 ± 0.3^{p}	20.1 ± 0.3^{p}
	HMT[a]	0.0	0.0	0.1 ± 0.0^{q}	2.0 ± 0.5^{q}	5.5 ± 0.1^{q}
Taro	Native	0.0	0.3 ± 0.3^{p}	2.3 ± 0.2^{p}	22.1 ± 0.2^{p}	23.0 ± 0.6^{p}
	HMT[a]	0.0	0.0^{q}	0.2 ± 0.0^{q}	7.3 ± 0.4^{q}	12.2 ± 0.1^{q}
New cocoyam	Native	0.0	0.1 ± 0.1^{p}	0.6 ± 0.1^{p}	2.9 ± 0.6^{p}	5.4 ± 0.4^{p}
	HMT[a]	0.0	0.0^{q}	0.4 ± 0.1^{q}	2.0 ± 0.2^{q}	2.2 ± 0.5^{q}
Cassava	Native	0.0	7.0 ± 0.2^{p}	15 ± 0.3^{p}	16.6 ± 0.5^{p}	17.2 ± 0.1^{p}
	HMT[a]	0.0	6.3 ± 0.2^{q}	11.5 ± 0.4^{q}	15.3 ± 0.1^{q}	16.1 ± 0.1^{q}
Potato	Native	0.0	4.5 ± 0.5^{p}	18.1 ± 0.1^{p}	22 ± 0.3^{p}	22.2 ± 0.2^{p}
	HMT[a]	0.0	1.9 ± 0.1^{q}	6.7 ± 0.2^{q}	8.7 ± 0.6^{q}	9.2 ± 0.3^{q}

[a] Heat–moisture treated (100 °C, 10 h, 30% moisture).

of the 36 water molecules in the central channel of the B-unit cell), and (2) movement of a pair of double helices into the central channel, (that was originally occupied by the vaporized water molecules). Double helical movement during heat–moisture treatment could disrupt starch crystallites and/or change crystallite orientation. This would then explain the observed changes in crystallinity on heat–moisture treatment of B-type starches (Table 3).

3.5. Swelling factor and amylose leaching

The SF and AML at different temperatures are presented in Tables 4 and 5, respectively. SF and AML of native and HMT starches increased with increase in temperature. The extent of this increase was more pronounced at temperatures beyond 70 °C. The, SF of native starches followed the order: Potato > cassava > taro > true yam > new cocoyam. Whereas, AML followed the order: potato ~ taro > true yam > cassava > new cocoyam. The interplay between the extent of interaction between starch chains (in the amorphous and crystalline domains), phosphate content (Table 1), and the amount of lipid complexed amylose chains (Table 1) may have been responsible for the observed differences in SF (Table 4) and AML (Table 5). The decrease in AML on heat–moisture treatment (Table 5) suggests that additional interactions may have occurred between amylose–amylose (AM–AM) and amylose–amylopectin (AM–AMP) chains during heat–moisture treatment. This type of mechanism may also be partly responsible for the observed decrease in SF on heat–moisture treatment (Table 4). Tester and Morrison (1990) have shown by comparative studies, on non-waxy and waxy maize starches, that swelling is primarily a property of amylopectin and that amylose is a diluent. Furthermore, Cooke and Gidley (1992) have suggested that the forces holding the granule together are mainly at the double helical level and that the starch 'crystallinity' functions as a means of achieving dense packing rather than as a primary provider of structure. This implies

Table 6
DSC characteristics of native, and heat moisture treated tuber and root starches (starch: water ratio is, 1:3 (w/w) dry basis; means within the same column with different superscripts (for native starch and its heat–moisture treated counterpart) are significantly different ($P < 0.05$)

Starch source	Treatment	Transition temperatures				
		T_0[a] (°C)	T_p[a] (°C)	T_c[a] (°C)	$T_c - T_0$[a] (°C)	ΔH[b] (J/g)
True yam	Native	75.0 ± 0.3^{p}	80.0 ± 0.5^{p}	91.2 ± 0.3^{p}	$16.5 \pm 0.^{p}$	17.8 ± 0.1^{p}
	HMT[c]	79.8 ± 0.2^{q}	88.2 ± 0.6^{q}	98.5 ± 0.5^{q}	18.9 ± 0.3^{q}	15.5 ± 0.1^{q}
Taro	Native	76.8 ± 0.2^{p}	83.0 ± 0.6^{p}	95.2 ± 0.7^{p}	18.4 ± 0.2^{p}	14.5 ± 0.3^{p}
	HMT[c]	79.2 ± 0.3^{q}	89.5 ± 0.1^{q}	98.5 ± 0.2^{q}	19.5 ± 0.2^{q}	12.2 ± 0.2^{q}
New cocoyam	Native	71.5 ± 0.6^{p}	77.2 ± 0.3^{p}	85.4 ± 0.9^{p}	13.6 ± 0.3^{p}	13.1 ± 0.4^{p}
	HMT[c]	75.4 ± 0.6^{q}	83.1 ± 0.2^{q}	90.3 ± 0.8^{q}	14.7 ± 0.5^{q}	11.5 ± 0.4^{q}
Cassava	Native	63.0 ± 0.5^{p}	71.5 ± 0.2^{p}	81.5 ± 0.2^{p}	18.7 ± 0.6^{q}	12.3 ± 0.2^{p}
	HMT[c]	66.4 ± 0.5^{q}	79.1 ± 0.2^{q}	87.0 ± 0.3^{q}	20.6 ± 0.3^{q}	11.7 ± 0.3^{q}
Potato	Native	59.6 ± 0.4^{p}	66.3 ± 0.4^{p}	76.0 ± 0.5^{p}	16.5 ± 0.4^{p}	16.3 ± 0.1^{p}
	HMT[c]	61.2 ± 0.3^{q}	75.6 ± 0.5^{q}	86.5 ± 0.4^{q}	25.3 ± 0.2^{q}	11.5 ± 0.2^{q}

[a] T_0, T_p, T_c, $T_c - T_0$ represent the onset, mid-point, end of gelatinization, and the gelatinization temperature range, respectively.
[b] Enthalpy of gelatinization.
[c] Heat–moisture treated (100 °C, 30% moisture, 10 h).

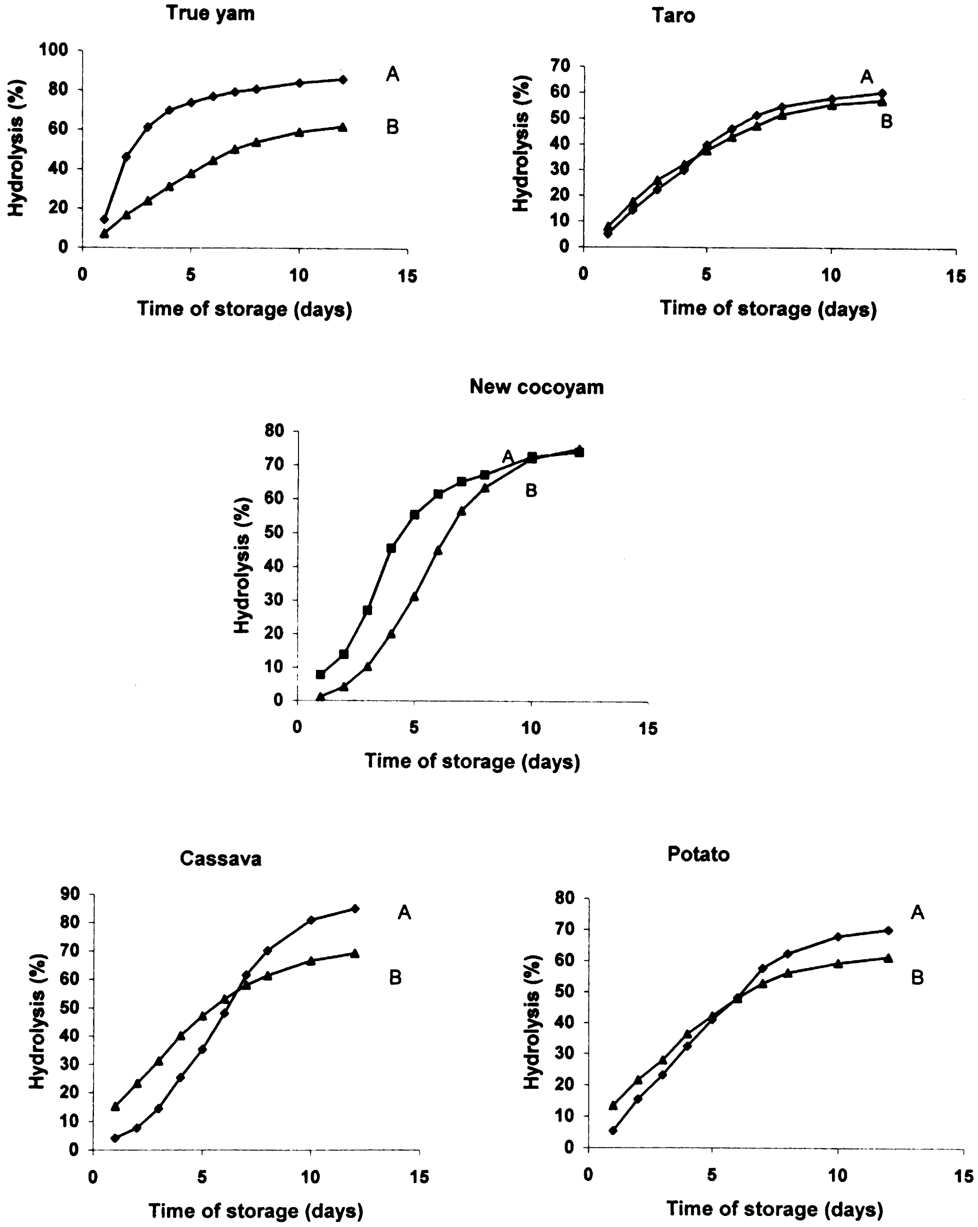

Fig. 2. Acid hydrolysis of native and HMT tuber and root starches; A, native; B, HMT.

that the decrease in SF on heat–moisture treatment (Table 4) could also be due to a decrease in granular stability, resulting from unraveling of double helices that may have been present in a crystalline array in the native granule. Disruption of crystallites on heat–moisture treatment (Table 3) could have contributed to the large decrease in SF observed for true yam and potato starches (Table 4).

3.6. Gelatinization parameters

The gelatinization temperatures, onset [(T_0), mid point (T_p), and conclusion (T_c)] and gelatinization enthalpy (ΔH) are presented in (Table 6). The gelatinization temperatures of native starches followed the order: taro > true yam > new cocoyam > cassava > potato. Whereas, the gelatinization temperature range (T_c–T_o) followed the order: cassava > taro > true yam > potato. The ΔH followed the order: true yam > potato > taro > new cocoyam > cassava.

The differences in gelatinization temperatures among the starches can be attributed to the interplay of three factors: (1) molecular structure of amylopectin (unit chain length, extent of branching (2) starch composition (amylose to amylopectin ratio, amount of lipid complexed, amylose chains, phosphorous content), and (3) granular architecture (crystalline to amorphous ratio). Difference in T_c–T_o (Table 6) suggests that the degree of heterogeneity of crystallites within the granules of the five starches are different. The ΔH values have been shown to represent the number of double helices that unravel and melt during gelatinization (Cooke & Gidley, 1992). Thus the higher ΔH values for true yam and potato starches (Table 6) could be attributed to the presence of a higher number of double helices (due to their longer amylopectin chain length (Table 2)) and/or weaker interaction between adjacent amylopectin double helices within the crystalline domains of the native granule (this postulate seems plausible, since crystallite disruption on heat–moisture treatment occurs only in potato and true yam (Table 3).

Heat–moisture treatment increased T_0, T_p, T_c and T_c–T_o (potato > true yam > cassava > taro > new cocoyam) (Table 6), but decreased ΔH (potato > true yam > cassava > taro > new cocoyam) (Table 6).

The melting temperatures (T_o, T_p, T_c) of the starch crystallites are controlled indirectly by the surrounding amorphous region. The reduction in granular swelling on heat–moisture treatment (Table 4) would reduce the destabilization effect of the amorphous region on crystallite melting. Consequently, a higher temperature would be required to melt crystallites of HMT starches. This would then explain the increase in T_o, T_p, and T_c on heat–moisture treatment (Table 6). The decrease in ΔH (Table 6) on heat–moisture treatment suggests that some of the double helices present in crystalline and in non-crystalline regions of the granule may have disrupted under the conditions prevailing during heat–moisture treatment. Thus, fewer double helices would unravel and melt during gelatinization of HMT starches.

In B-type starches, the packing of helices is less compact than in A-type starches, (Gidley, 1987). Furthermore, there are 36 water molecules per B-type unit cell, whereas only four water molecules are present within the A-type unit cell (Sarko & Wu, 1978). Consequently, on heat–moisture treatment, the double helical chains forming the crystallites of B-type starches would be more mobile, and hence more prone to disruption than those of A-type starches. This would then explain, the large difference in ΔH between native and HMT B-type starches (Table 6). The changes in gelatinization parameters on heat–moisture treatment are more pronounced in potato starch due to its higher phosphate monoester content (Table 1). Phosphate groups are mainly located on C_2, C_3, and C_6 of the glucose unit of potato starch (Hizukuri, Tabata, & Nikuni, 1970). Repulsion between negatively charged phosphate groups on adjacent amylopectin chains would hinder strong interaction between double helices. Consequently, crystallites of potato starch would be very susceptible to disruption on heat–moisture treatment. This would then explain the large decrease in T_o, T_p, T_c and ΔH on heat–moisture treatment of potato starch (Table 6).

3.7. Acid hydrolysis

Acid hydrolysis of native and HMT starches are presented in (Fig. 2). The extent of hydrolysis of native starches during the first few days (corresponding mainly to the degradation of amorphous regions) followed the order: true yam > potato > taro > cassava > new cocoyam. Thereafter, the extent of hydrolysis (corresponding mainly to the degradation of crystallites) followed the order: true yam > cassava > new cocoyam > potato > taro. Differences in the extent of acid hydrolysis between native starches has been attributed to granule size, extent of starch chain interactions (within the amorphous and crystalline regions), amount of double helical structures (in the amorphous area) and starch composition (extent of phosphorylation, amylose/amylopectin ratio, and amount of lipid complexed amylose chains (Hoover, 2000). Jane, Wang, and Mcpherson (1997) postulated that difference in susceptibility of potato (B-type X-ray pattern) and maize (A-type X-ray pattern) starches towards acid hydrolysis can be attributed to a certain extent to the mode of distribution of α (1 → 6) branch points between the amorphous and crystalline domains. In B-type starches the α (1 → 6) branch points are located mainly in the amorphous region and thus very susceptible to acid hydrolysis. Whereas, in A-type starches the α (1 → 6) branches which are located in the crystalline region are resistant to hydrolysis by H_30^+. The observed data on hydrolysis of native starches (Fig. 2) cannot be explained on the basis of granule surface area or granule composition. Therefore, it is plausible, that the interplay of the magnitude of starch chain interactions (within the amorphous domain), the amount of double helical structures (in the amorphous domain), and the mode of distribution of α (1 → 6) branch linkages are the factors that influence the observed differences (during the first six days) in acid hydrolysis among native starches (Fig. 2). However, differences in hydrolysis beyond the seventh day (Fig. 2) reflects the degree of accessibility of H_30^+ to the glycosidic oxygens buried within the starch crystallites. The change in the extent of acid hydrolysis on heat–moisture treatment (Fig. 2) can be explained on the basis of the interplay of four factors: (1) crystallite disruption (increases acid hydrolysis by making available more amorphous regions); (2) interactions between starch chains (decreases hydrolysis by reducing chain flexibility, and thereby hindering the conformational change (chair → half chair) required for efficient protonation of glycosidic oxygens; and (3) disruption of double helices in the amorphous region (increases hydrolysis by making glycosidic oxygens more accessible to protonation). The influence of heat–moisture treatment on acid hydrolysis varied among the starch sources. For instance, throughout the time course of hydrolysis, HMT true yam starch was hydrolyzed to a much lesser extent than that of its native counterpart (Fig. 2). However, HMT taro, cassava and potato showed increased hydrolysis until the fourth, sixth, and fifth day respectively. Thereafter, hydrolysis was less than their native counterpart (Fig. 2). The decreased susceptibility of HMT true yam starch towards acid hydrolysis (Fig. 2) suggests that extensive amylose chain interactions on heat–moisture treatment (Table 5) probably negates the influence of crystallite disruption on acid hydrolysis. In potato and true yam

Table 7
Enzyme hydrolysis (%) of native and heat–moisture treated tuber and root starches by porcine pancreatic α-amylase (means (for the different stages of hydrolysis) within the same column (for native starch and its heat–moisture treated counterpart) with different superscripts are significantly different ($P < 05$))

Starch source	Treatment	Number of hours			
		6	12	24	72
True yam	Native	1.2 ± 0.2^{p}	1.5 ± 0.2^{p}	3.1 ± 0.2^{p}	4.9 ± 0.3^{p}
	HMT[a]	4.2 ± 0.3^{q}	8.5 ± 0.3^{q}	11.3 ± 0.5^{q}	17.2 ± 0.5^{q}
Taro	Native	8.2 ± 0.4^{p}	15.3 ± 0.1^{p}	22.1 ± 0.2^{p}	38.0 ± 0.2^{p}
	HMT[a]	12.3 ± 0.5^{q}	19.2 ± 0.4^{q}	28.1 ± 0.3^{q}	45.4 ± 0.1^{q}
New cocoyam	Native	18.8 ± 0.6^{p}	24.2 ± 0.1^{p}	36.1 ± 0.4^{p}	62.5 ± 0.1^{p}
	HMT[a]	20.2 ± 0.7^{q}	29.1 ± 0.6^{q}	40.2 ± 0.1^{q}	67.3 ± 0.3^{q}
Cassava	Native	15.3 ± 0.4^{p}	22.1 ± 0.2^{p}	34.2 ± 0.6^{p}	56.2 ± 0.4^{p}
	HMT[a]	19.4 ± 0.5^{q}	38.4 ± 0.3^{q}	48.1 ± 0.5^{q}	69.5 ± 0.1^{q}
Potato	Native	1.8 ± 0.6^{p}	2.1 ± 0.3^{p}	3.8 ± 0.1^{p}	5.9 ± 0.5^{p}
	HMT[a]	8.3 ± 0.5^{q}	14.7 ± 0.6^{q}	23.1 ± 0.8^{q}	32.8 ± 0.1^{q}

[a] Heat–moisture treated (100 °C, 30%miosture, 10 h).

starches, the extent of crystallite disruption (Table 2) and amylose–amylose interactions (Table 5) during heat–moisture treatment are nearly similar. Therefore, the different hydrolysis patterns shown by HMT true yam and potato starches (Fig. 2), suggests that double helical structures that may have been present in the amorphous regions of potato starch are probably disrupted on heat–moisture treatment, and are thus rendered more accessible to attack by H_30^+. This would then explain the increase in hydrolysis shown by HMT potato starch during the first five days of hydrolysis (Fig. 2). The results suggest that double helical structures are either absent and/or are present only in trace amounts in the amorphous regions of true yam (due to its shorter amylose chain length (Table 2)). The increased susceptibility of HMT new cocoyam starch towards acid hydrolysis (during the first 10 days) can be attributed to the interplay of the following factors: (1) action of H_30^+ on disrupted double helices in the amorphous region; (2) weak interaction between amylose chains during heat–moisture treatment (Table 5); and (3) action of H_30^+ on free amylose chains (that were originally complexed with lipids in the native granule). The increase in acid hydrolysis on heat–moisture treatment is more pronounced in cassava than in taro due to weaker amylose–amylose interactions (Table 5) in the former.

3.8. Enzyme hydrolysis

The susceptibility of tuber and root starches towards hydrolysis by porcine pancreatic α-amylase followed the order: New cocoyam > cassava > taro > potato ~ true yam (Table 8). Differences in vitro digestibility of native starches by α-amylase has been attributed to the interplay of many factors such as: granular size, surface area, type of unit cell (A, B or A + B), amylose to amylopectin ratio, amount of lipid complexed amylose chains, crystallinity and extent of distribution of α (1 → 6) branch points between the amorphous and crystalline regions of amylopectin (Holm et al., 1983; Hoover & Sosulski, 1991; Jane et al., 1997; Planchot, Colonna, Buleon, & Gallant, 1997). The results indicate that differences in granule size (potato > true yam > taro > cassava > new cocoyam) and the presence of α (1 → 6) branch points (The location of α (1 → 6) branch points in the crystallite regions would weaken the crystalline structure thereby increasing the accessibility of α-amylase into the granule interior.) in the crystalline regions of A-type starches (new cocoyam, taro, cassava) are the factors that influence hydrolysis of native starches. This postulate is based on the fact that differences between starches with respect to the level of amylose content (Table 1), amylose lipid complexes (Table 1) and crystallinity (Table 2) are too small to account for the observed differences on enzyme hydrolysis.

In all starches, enzyme susceptibility increased on heat–moisture treatment (potato > true yam > cassava > taro > new cocoyam). Gallant (1974) has shown that one of the limiting factors in α-amylolysis could be the nature of the granule surface with respect to crystallinity. Furthermore, Planchot et al. (1997) have postulated, that the fraction of total crystalline material is an important factor defining the rate and extent of α-amylase hydrolysis. The initial step of α-amylolysis corresponds to adsorption of α-amylase on the granule surface. Thus, crystallite disruption near the granule surface on heat–moisture treatment of true yam and potato (Table 3) starches, could facilitate the rapid entry of α-amylase into the granule interior. This would then explain the more pronounced increase in enzyme hydrolysis observed on heat–moisture treatment of the above starches (Table 7). The extent of crystallinity disruption during heat–moisture treatment was nearly the same in both true yam and potato starches (Table 3). Therefore, the more pronounced increase in hydrolysis after heat–moisture treatment of potato starch (Table 7) could be attributed to interactions (during heat–moisture treatment) involving amylose chains (Table 5) being of a lower order of magnitude than in true yam (Interaction between amylose chains (within the

Table 8
The enthalpy of retrogradation (ΔH_R) of native and heat–moisture treated tuber and root starches monitored by differential scanning calorimetry (at 40°C)

Starch source	Treatment	Number of days of storage[b]						
		1	2	3	4	5	6	7
True yam	Native	7.4 ± 0.5^{p}	7.2 ± 0.9^{p}	7.6 ± 0.6^{p}	8.7 ± 0.3^{p}	9.2 ± 0.5^{p}	9.3 ± 0.8^{p}	9.8 ± 0.6^{p}
	HMT[a]	7.2 ± 0.8^{p}	7.1 ± 0.3^{p}	7.4 ± 0.5^{p}	8.1 ± 0.6^{p}	8.3 ± 0.6^{p}	8.4 ± 0.6^{p}	8.7 ± 0.1^{q}
Taro	Native	4.1 ± 0.6^{p}	5.2 ± 0.6^{p}	5.7 ± 0.8^{p}	6.3 ± 0.4^{p}	6.8 ± 0.8^{p}	7.1 ± 0.8^{p}	7.1 ± 0.8^{p}
	HMT[a]	3.9 ± 0.9^{p}	4.9 ± 0.2^{p}	5.7 ± 0.2^{p}	6.2 ± 0.9^{p}	6.9 ± 0.2^{p}	6.7 ± 0.1^{p}	6.9 ± 0.5^{p}
New cocoyam	Native	3.6 ± 0.7^{p}	3.4 ± 0.6^{p}	3.8 ± 0.6^{p}	4.1 ± 0.3^{p}	4.9 ± 0.3^{p}	5.2 ± 0.6^{p}	5.3 ± 0.7^{p}
	HMT[a]	3.5 ± 0.5^{p}	3.1 ± 0.2^{p}	3.8 ± 0.2^{p}	4.5 ± 0.4^{p}	4.8 ± 0.9^{p}	5.0 ± 0.8^{p}	5.4 ± 0.8^{p}
Cassava	Native	1.3 ± 0.5^{p}	1.4 ± 0.2^{p}	2.2 ± 0.3^{p}	2.3 ± 0.6^{p}	2.5 ± 0.8^{p}	3.4 ± 0.4^{p}	3.6 ± 0.3^{p}
	HMT[a]	1.5 ± 0.6^{p}	1.6 ± 0.2^{p}	2.5 ± 0.8^{p}	2.2 ± 0.3^{p}	2.7 ± 0.5^{p}	3.1 ± 0.5^{p}	3.2 ± 0.4^{p}
Potato	Native	4.9 ± 0.8^{p}	5.3 ± 0.4^{p}	5.9 ± 0.5^{p}	6.6 ± 0.3^{p}	7.2 ± 0.3^{p}	7.6 ± 0.3^{p}	7.9 ± 0.7^{p}
	HMT[a]	4.8 ± 0.3^{p}	5.6 ± 0.5^{p}	5.3 ± 0.6^{p}	5.5 ± 0.9^{p}	5.9 ± 0.8^{p}	7.2 ± 0.8p	7.1 ± 0.5^{p}

[a] Heat–moisture treated (100 °C, 30%moisture, 10 h).

[b] Means (for the different days of storage) with different superscripts (for native starch and its heat–moisture treated counterpart) are significantly different ($p < 0.05$)

amorphous region) would decrease the accessibility of α-amylase towards the α-(1 → 4) glycosidic linkages). In A-type starches, crystallites are not disrupted on heat–moisture treatment (Table 3). Therefore, the extent of increase in hydrolysis on heat–moisture treatment (cassava > taro new new cocoyam) mainly reflects the interplay between: (1) the number of double helices that may have disrupted in the amorphous regions during heat–moisture treatment (disrupted double helices would increase accessibility of the unraveled chains to the binding sites of α-amylase), and (2) the extent of interaction that occurs between amylose chains (taro > new cocoyam > cassava) during heat–moisture treatment (Table 5).

3.9. Retrogradation

The melting enthalpies (ΔH_R) of amylopectin recrystallization are presented in (Table 8). ΔH_R reflects the extent of retrogradation during the storage period (seven days at 40 °C). ΔH_R followed the order: True yam > potato > taro > new cocoyam > cassava. This result confirms earlier reports that amylopectin from B-type starches retrogrades to a greater extent than amylopectins from A-type (cereal) and A + B-type (legume) starches (Kalichevsky, Orford, & Ring, 1990; Silverio, Svensson, Eliasson, & Olofsson, 1996). This was attributed to the shorter average amylopectin chain length of the A-type starches (Kalichevsky et al., 1990; Orford, Ring, Carroll, Miles, & Morris, 1987). Ward, Hoseney, and Seib (1994) postulated that differences in retrogradation between cereal amylopectins is influenced by: (a) an increased molar proportion of unit chain with DP 14-24 (increases retrogradation, and (b) an increased molar proportion of short chains with DP 6-9 (inhibits retrogradation). A similar finding was also reported by Wursch and Gumy (1994).

In this study, differences in the extent of retrogradation between taro, new cocoyam and cassava (Table 8) can be explained on the basis of differences in their external chain length (Table 2). However, differences in retrogradation between true yam and potato are probably influenced to a greater extent by differences in their amylopectin chain length distribution (not determined in this study) rather than the external chain length. Heat–moisture treatment decreased retrogradation in B-type starches, but caused no significant changes to the retrogradation of A-type starches (Table 8). As discussed earlier, crystallites are disrupted in B-type starches, but remain unchanged in A-type starches on heat–moisture treatment (Table 2). Thus, after heat–moisture treatment, the degree of separation between the outer branches of adjacent amylopectin chains would be greater in the B-type starches, but would remain practically the same in A-type starches. Consequently, during gel storage, the formation and lateral association of double helices involving amylopectin chains, would be much slower, more difficult and less stronger for HMT B-type starches. This would then explain the observed decrease in ΔH_R for true yam and potato, and the unchanged ΔH_R for new cocoyam, cassava and taro starches (Table 8).

4. Summary and conclusion

The results showed that starch chain interaction, crystalline disruption and dissociation of double helical structures (in the amorphous region) occur on heat moisture treatment. The extent of these structural changes and the accompanying changes to crystallinity, amylose leaching, granular swelling, acid and enzyme susceptibility, gelatinization and retrogradation were more pronounced in the B-type starches (true yam and potato).

Many tuber and root starches are not widely used in food applications due to their poor functional properties. Presently, chemical modification is widely used to tailor the properties of potato and cassava starches. This study has shown that heat–moisture treatment may be an alternative to chemical modification for altering the gelatinization and retrogradation properties of tuber and root starches. Many tuber and root crops are endemic to less developing countries. Thus, scientists in these countries need to tailor the properties of tuber and root starches by heat–moisture treatment (using different/temperature/time combinations) to a level that is presently met by chemical modification. Such a study would help these countries to compete more effectively in the markets in both the food and non-food sectors.

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References

Abraham, T. E. (1993). Stabilization of paste viscosity of cassava starch by heat–moisture treatment. *Starch*, *45*, 131–135.

American Association of Cereal Chemists (1984). *Approved methods of the AACC* (8th ed.). St Paul, MN.

Bruner, R. L. (1964). Determination of reducing value. In R. L. Whistler, *Methods in carbohydrate chemistry* (pp. 67–71). Vol. 4. New York: Academic Press.

Collado, L. S., & Corke, H. (1999). Heat–moisture treatment effects on sweet potato starches differing in amylose content. *Food Chemistry*, *65*, 329–346.

Collado, L. S., Mabesa, L. B., Oates, C. G., & Corke, H. (2001). Bihon type noodles from heat–moisture treated sweet potato starch. *Journal of Food Science*, *66*, 604–609.

Cooke, D., & Gidley, M. J. (1992). Loss of crystallinity and molecular order during starch gelatinization: Origin of the enthalpic transition. *Carbohydrate Research*, *227*, 103–112.

Donovan, J. W., Lorenz, K., & Kulp, K. (1983). Differential scanning calorimetry of heat–moisture treated wheat and potato starches. *Cereal Chemistry*, *60*, 381–387.

Dubois, M., Gilles, K. A., Hamilton, J. K., Rebers, P. A., & Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, *28*, 350–352.

Franco, C. M. L., Ciacco, C. F., & Tavares, D. Q. (1995). Effect of heat–moisture treatment on the enzymatic susceptibility of corn starch granules. *Starch*, *47*, 223–228.

Fukui, T., & Nikuni, Z. (1969). Heat–moisture treatment of cereal starch observed by X-ray diffraction. *Agricultural Biological Chemistry*, *33*, 460–462.

Gallant, D. J. (1974). Contribution a' l' etude de la structure et de l' ultrastructure de l' amidon. Th'ese de Doctorat d' Etat: Univ., Paris.

Gidley, M. J. (1987). Factors affecting the crystalline type (A–C) of native starches and model compounds. A rationalization of observed effects in terms of polymorphic structure. *Carbohydrate Research*, *161*, 301–304.

Gidley, M. J., & Bociek, S. M. (1985). Molecular organization in starches. A ^{13}CCP/MAS NMR study. *Journal of the American Chemical Society*, *107*, 7040–7044.

Hagiwara, S., Esaki, K., Kitamura, S., & Kuge, T. (1991). Observation by photo microscopic and X-ray diffraction method of heat–moisture treatment on starch granules. *Denpun Kagaku*, *38*, 241–247.

Hizukuri, S. (1985). Relationship between the distribution of the chain length of amylopectin and the crystalline structure of starch granules. *Carbohydrate Research*, *141*, 295–306.

Hizukuri, S. (1996). Starch analytical aspects. In A. C. Eliasson, *Carbohydrates in foods* (pp. 342–429). New York: Marcel Dekker.

Hizukuri, S., Fujii, M., & Nikuni, Z. (1961). Effect of temperature during germination on the crystalline type of starch in soya bean seedling. *Nature*, *192*, 239–242.

Hizukuri, S., Tabata, S., & Nikuni, Z. (1970). Studies on starch phosphates. Part 1. Estimation of glucose-6 phosphates residues in starch and the presence of other bound phosphates. *Starch/Starke*, *22*, 338–343.

Hizukuri, S., Takeda, Y., Yasuda, M., & Suzuki, A. (1981). Multibranched nature of amylose and the action of debranching enzymes. *Carbohydrate Research*, *94*, 205–213.

Holm, J., Björck, I., Ostrowska, S., Eliasson, A. C., Asp, N. G., Larrson, K., & Lundquist, L. (1983). Digestibility of amylose–lipid complexes in vitro and in vivo. *Starch*, *35*, 294–297.

Hoover. R. (2000). Acid treated starches. Food Reviews International, 16, 369–392.

Hoover, R. (2001). Composition, molecular structure and physicochemical properties of tuber and root starches: A review. *Carbohydrate Polymers*, *45*, 253–267.

Hoover, R., & Hadziyev, D. (1981). Characterization of potato starch and its monoglyceride complexes. *Starch*, *33*, 290–300.

Hoover, R., & Manuel, H. (1996). The effect of heat–moisture treatment on the structure and physicochemical properties of normal maize, waxy maize, dull waxy maize and amylomaize v starches. *Journal of Cereal Science*, *23*, 153–162.

Hoover, R., & Ratnayake, R. M. W. S. (2001). Determination of total amylose content of starch. In R. E. Wrolstad, T. E. Acree, H. An, E. A. Decker, M. A. Penner, D. S. Reid, S. J. Schwartz, C. F. Shoemaker & P. Sporns, *Current protocols in food analytical chemistry* New York: Wiley (Section E, Unit 2–3).

Hoover, R., & Sosulski, F. W. (1991). Composition, structure, functionality and chemical modification of legume starches. A review. *Canadian Journal of Physiology and Pharmacology*, *69*, 79–92.

Hoover, R., & Vasanthan, T. (1994). Effect of heat–moisture treatment on the structure and physicochemical properties of cereal, tuber, and legume starches. *Carbohydrate Research*, *252*, 33–53.

Hoover, R., Swamidas, G., & Vasanthan, T. (1993). Studies on the physicochemical properties of native, defatted and heat–moisture treated pigeon pea (*Cajanus cajan* L.) starch. *Carbohydrate Research*, *246*, 185–203.

Hoover, R., Vasanthan, T., Senanayake, N., & Martin, A. M. (1994). The effect of defatting and heat–moisture treatment on the retrogradation of starch gels from wheat, oat, potato, and lentil. *Carbohydrate Research*, *261*, 13–24.

Imberty, A. (1988). A revisit to the three dimensional structure of B-type starch. *Biopolymers*, *27*, 1205–1221.

Imberty, A., & Perez, S. (1989). Conformational analysis and molecular modeling of the branching point of amylopectin. *International Journal of Macromolecules*, *11*, 177–185.

Imberty, A., Chanzy, H., Perez, S., Buleon, A., & Tran, V. (1988). The double helical nature of the crystalline part of A-starch. *Journal of Molecular Biology*, *201*, 365–378.

Jacobs, M., & Delcour, J. A. (1998). Hydrothermal modifications of granular starch with retention of the granular structure. A review. *Journal of Agriculture and Food Chemistry*, *46*, 2895–2905.

Jane, J. L., & Chen, J. F. (1992). Effect of amylose molecular size and amylopectin branch chain length on paste properties of starch. *Cereal Chemistry*, *69*, 60–65.

Jane, J. L., & Robyt, J. F. (1984). Structure studies of amylose-v complexes and retrograded amylose by action of alpha-amylase, and a new method for preparing amylodextrins. *Carbohydrate Research*, *132*, 105–118.

Jane, J. L., Wang, K. S., & Mcpherson, A. E. (1997). Branch structure difference in starches of A and B-type X-ray pattern revealed by their Naegeli dextrins. *Carbohydrate Research*, *300*, 219–227.

Kalichevsky, M. T., Orford, P. D., & Ring, S. G. (1990). The retrogradation and gelatinization of amylopectins from various botanical sources. *Carbohydrate Research*, *198*, 49–55.

Kawabata, A., Takase, N., Miyoshi, E., Sawayama, S., Kimura, T., & Kudo,

K. (1994). Microscopic observation and X-ray diffractometry of heat–moisture treated starch granules. *Starch*, *46*, 463–469.

Kobayashi, T. (1993). Susceptibility of heat–moisture treated starches to pancreatic α-amylase, and the formation of resistant starch by heat–moisture treatment. *Denpun Kagaku*, *40*, 285–290.

Kuge, T., & Kitamura, A. S. (1985). Annealing of starch granules warm water treatment and heat–moisture treatment. *Journal of Japanese Society of Starch Science*, *32*, 65–83.

Lorenz, K., & Kulp, K. (1981). Heat–moisture treatment of starches. ii. Functional properties and baking potential. *Cereal Chemistry*, *58*, 49–52.

Lorenz, K., & Kulp, K. (1982). Cereal and root starch modification by heat–moisture treatment. 1. Physicochemical properties. *Starch*, *34*, 50–54.

Lorenz, K., & Kulp, K. (1983). Physicochemical properties of defatted and heat–moisture treated starches. *Starch*, *35*, 123–129.

Manners, D. J. (1989). Recent developments in our understanding of amylopectin structure. *Carbohydrate Polymers*, *11*, 87–112.

Maruta, I., Kurahashi, Y., Takano, R., Hayashi, K., Yoshino, Z., Kamaki, T., & Hara, S. (1994). Reduced pressurized heat–moisture treatment: A new method for heat–moisture treatment of starch. *Starch*, *46*, 177–181.

McGrance, S. J., Cornell, H. J., & Rix, C. J. (1998). A simple and rapid colorimetric method for the determination of amylose in starch products. *Starch*, *50*, 158–163.

Montgomery, E. M., & Senti, F. R. (1958). Separation of amylose from amylopectin of starch by extraction sedimentation procedure. *Journal of Polymer Science*, *28*, 1–9.

Morrison, W. R. (1964). A fast simple and reliable method for the micro determination of phosphorous in biological materials. *Analytical Biochemistry*, *7*, 218–224.

Nara, Sh., Mori, & Komiya, T. (1978). Study on relative crystallinity of moist potato starch. *Starch*, *30*, 11–114.

Orford, P. D., Ring, S. G., Carroll, V., Miles, M. J., & Morris, V. J. (1987). The effect of concentration and botanical source on the gelation and retrogradation of starch. *Journal of Science and Food Agriculture*, *39*, 169–177.

Planchot, V., Colonna, P., Buleon, A., & Gallant, D. (1997). Amylolysis of starch granules and α-glucan crystallites. In R. J. Frazier, A. M. Donald & P. Richmond, *Starch structure and functionality* (pp. 141–152). Cambridge, UK: Royal Society of Chemistry.

Radosta, S., Kettlitz, B., Schierbacum, F., & Gernat, C. (1992). Studies on rye starch properties and modification part 11. Swelling and solubility behavior of rye starch granules. *Starch*, *44*, 8–14.

Sair, L. (1967). Heat–moisture treatment of starch. *Cereal Chemistry*, *44*, 8–26.

Sarko, A., & Wu, H. C. H. (1978). The crystal structures of A, B, and C—polymorphs of amylose and starch. *Starch*, *30*, 73–78.

Schierbaum, F., & Kettlitz, B. (1994). Studies on rye starch properties and modification. Part 11. Viscograph pasting characteristics of rye starches. *Starch*, *46*, 2–8.

Schmiedel, D., König, B., & Jacobasch, G. (1998). Hydrothermische behandlung von starke in gegenwart von maltosirup. Teil 1 Einflußauf DSC-parameter und de konsistenz von kartoffelstarkege len. *Starch*, *50*, 297–305.

Shi, Y. C., & Seib, P. (1992). The structure of four waxy starches related to gelatinization and retrogradation. *Carbohydrate Research*, *227*, 131–145.

Silverio, J., Svensson, E., Eliasson, A. C., & Olofsson, G. (1996). Isothermal microcalorimetric studies on starch retrogradation. *Thermal Analysis*, *47*, 1179–1200.

Stoof, G., Schmiedl, D., Anger, H., Rehbrucke, B., & Bergthaller, W. (1998). Hydrothermische behandlung von starke in gegenwart von α-amylase Teil 4. Änderungen der eigenschaften von kartoffelstarke durch hydrothermisch-enzymetische behandlung. *Starch*, *50*, 108–114.

Stute, R. (1992). Hydrothermal modification of starches: The difference between annealing and heat–moisture treatment. *Starch*, *44*, 205–214.

Suzuki, A., Takeda, Y., & Hizukuri, S. (1985). Relationship between the molecular structures and retrogradation properties of tapioca, potato, and kuzu starches. *Denpun Kagaku*, *32*, 205–210.

Takaya, T., Sano, C., & Nishinari, K. (2000). Thermal studies on the gelatinization and retrogradation of heat–moisture treated starch. *Carbohydrate Polymers*, *41*, 97–100.

Takeda, C., Takeda, Y., & Hizukuri, S. (1983). Physicochemical properties of lily starch. *Cereal Chemistry*, *64*, 313–316.

Takeda, Y., Shiraska, K., & Hizukuri, S. (1984). Examination of the purity and structure of amylose on gel permeation chromatography. *Carbohydrate Research*, *132*, 83–86.

Tester, R. F., & Morrison, W. R. (1990). Swelling and gelatinization of cereal starches. l. Effects of amylopectin, amylose and lipids. *Cereal Chemistry*, *67*, 551–559.

Vasanthan, T., & Hoover, R. (1992). A comparative study of the composition of lipids associated with starch granules from various botanical sources. *Food Chemistry*, *43*, 19–27.

Ward, K. E. J., Hoseney, R. C., & Seib, P. A. (1994). Retrogradation of maize and wheat starches. *Cereal Chemistry*, *71*, 150–155.

Wursch, P., & Gumy, D. (1994). Inhibition of amylopectin retrogradation by partial β-amylolysis. *Carbohydrate Research*, *256*, 129–137.

Zobel, H. F. (1988). Molecules to granules. A comprehensive starch review. *Starch*, *40*, 44–50.