Nigerian Journal of Polymer Science and Technology, Vol. 10, pp 46-53, 2015

Effect of Pregelatinisation on Selected Functional, Molecular and Pasting Properties of Legume Starches

A. O. Oladebeye[1*], J. E. Imanah[2], A. E. Eboreime[2], and A. A. Oladebeye[3]

[1]*Science Laboratory Technology Department, Auchi Polytechnic, P.M.B. 13, Auchi, Nigeria.*
[2]*Polymer Technology Department, Auchi Polytechnic, P.M.B. 13, Auchi, Edo State, Nigeria.*
[3]*Food Technology Department, Auchi Polytechnic, P.M.B. 13, Auchi, Edo State, Nigeria.*

ABSTRACT: Pregelatinised derivatives of the native starches isolated from pigeon pea (Cajanus cajan L.), lima bean (Phaseolus lunatus L.) and jack bean (Canavalia ensiformis) were prepared at 50 and 55 ^{o}C. Both the native and their pregelatinised derivatives were evaluated for functional, molecular and pasting properties. The pregelatinised starches possessed lower swelling profile and gel strength than their corresponding native forms whereas an opposite trend was obtained for their solubility profiles. The values M_w of the native starches ranged between 1.12 X 10^6 and 1.17 X 10^6, M_n ranged between 1.00 X 10^6 and 1.98 X 10^6 whereas those of the pregelatinised starches ranged between 3.11 X 10^6 and 6.44 X 10^6 for M_w and between 0.53 X 10^6 and 1.10 X 10^6. All the pregelatinised starches exhibited increase in polydispersity index (PDI) compared to their native forms. The native starches could, however, be more suitable in pharmaceuticals than their pregelatinised derivatives.

Keywords: Starch, pregelatinisation, swelling power, molecular weight

Introduction

Starch is a versatile biopolymer that has found numerous applications in many industries due to its environmental friendliness and availability. Such applications range from food to non-food industries. In its native form, starch suffers limitations under rigorous industrial processing. Such limitations can be eliminated or reduced significantly through modifications (Oladebeye et al., 2013). The effects of hydroxypropylation on the physicochemical properties, such as swelling power, solubility, pasting properties, rheological properties, of legume starches have been studied and reported by Oladebeye et al. (2013). Pregelatinisation is one of the physical methods used to modify starch, resulting in alteration of physicochemical and functional properties of the starch significantly (Lai, 2001). Due to starch granule disruption, pregelatinized starch can absorb water and increase viscosity immediately even with cold water (Wadchararat et al., 2006). Swelling power of starch, among other functional properties, has been considered to be of great significance in tablet and capsule formulations, as it is believed that disintegrants work through a swelling and wicking action (Adebayo and Itiola, 1998).

Starch is one of the traditional excipients used in the manufacture of tablets (Kottke et al., 1992). Depending on the application, specific starches are available for use as binders, diluents, and/or disintegrants. Starches are being used as disintegrants because of the swelling properties of starch granules in water. Studies have shown that starches, in their various forms, have a variety of swelling

---

**Corresponding author; E-mail: folabeye@yahoo.com, Tel.: +234 8033620279*

abilities (Herman et al., 1989; Sanchez et al., 1995; Alebiowu and Itiola, 2002), which may be responsible for the different disintegration and dissolution times reported by Kottke et al. (1992).

The present paper aims at: (a) sourcing starches from some selected legumes – pigeon pea (*Cajanus cajan L.*), lima bean (*Phaseolus lunatus L.*) and jack bean (*Canavalia ensiformis*), (b) modifying the starches via pregelatinisation process, and (c) examining the effects of pregelatinisation of some selected functional, molecular and pasting properties of the starches. This is with the view to proposing possible applications of these starches.

Experimental

Materials

Pigeon pea (*Cajanus cajan L.*) and lima beans (*Phaseolus lunatus*) were commercially purchased at Jattu Market, Etsako-West Local Government Area, Jattu, Edo State, Nigeria while jack beans (*Canavalia ensiformis*) were freshly from the orchard of one of the authors. All reagents were commercially purchased and were analar grade.

Isolation of Native Legume Starches

The native starches were isolated, using the method described by Oladebeye et al. (2013), which is illustrated in Fig. 1. The fully pregelatinised starches were prepared by adopting the method described by Herman et al. (1989) with slight modifications. An aqueous slurry of each starch was made with 100 g of starch powder in 100 ml of deionised water and then heated at 50 °C with stirring for 10 min. The resulting paste was crisp-dried in a hot-air oven at 60 °C for 48 h, followed by grinding and sieving through a 250-mesh (250 μm) sieve before use. The same procedure was repeated for slurry of each starch at 55 °C to obtain two pregelatinised starch samples for each native starch type. The starch samples were labeled as PPS (native pigeon pea starch), PPS$_{50}$ (pregelatinised pigeon pea starch at 50 °C), PPS$_{55}$ (pregelatinised pigeon pea starch at 55 °C), LBS (native lima bean starch), LBS$_{50}$ (pregelatinised lima bean starch at 50 °C), LBS$_{55}$ (pregelatinised lima bean starch at 55 °C), JBS (native jack bean starch), JBS$_{50}$ (pregelatinised jack bean starch at 50 °C) and JBS$_{55}$ (pregelatinised jack bean starch at 55 °C),

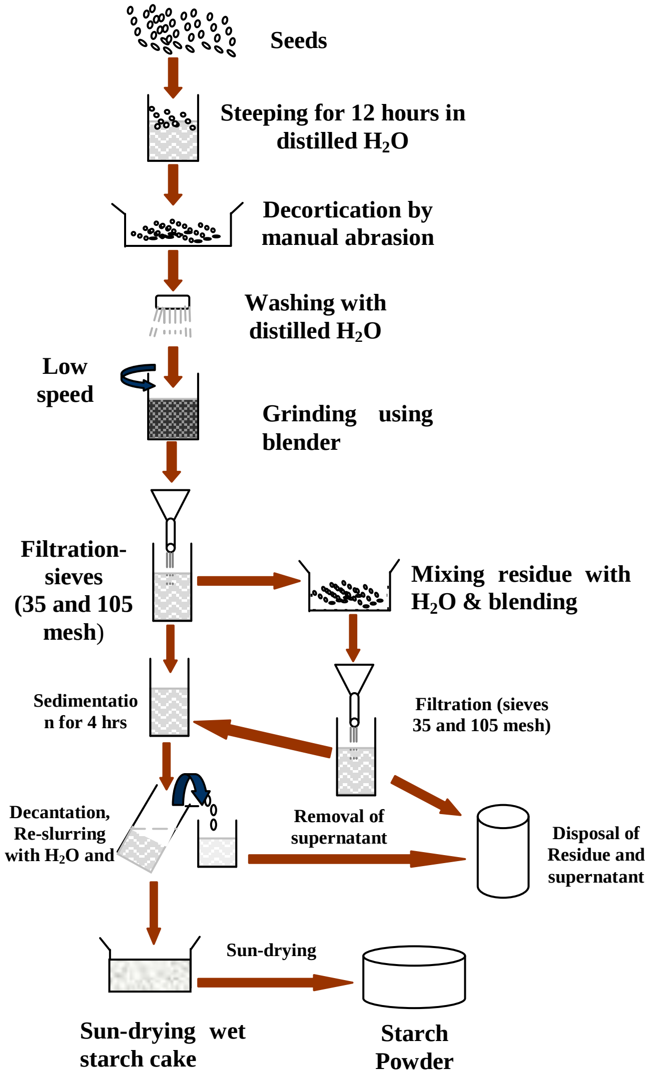

Figure 1: Isolation of Legume Starches

Functional Properties

The chemical method described by Leach et al. (1959) was adopted for the determination of both swelling power and solubility of starch samples. Swelling power was expressed as the ratio of the weight of the difference between the weight of swollen sediment and dry test tube to the initial weight of dry starch. The solubility was expressed as the ratio of dried supernatant weight initial dry starch weight.

The method of Gunaratne and Corke (2007) was adopted for the determination of gel strength of the samples. Starch paste (8%, w/v) was covered by parafilm wrap and stored at 4°C for 7 days for proper gel formation. The gel was allowed to equilibrate to room temperature and was compressed at a pre-set speed of 5 mm/s, test speed of 0.5mm/s and post speed of 5 mm/s to a distance of 10 mm with a 6-mm cylindrical probe and trigger force of 5g, using a TA-XT2 texture analyzer (Stable Micro Systems, Godalming, England). The maximum force peak (hardness) in the TPA profile represented the gel strength..

Molecular Mass Distribution

The standard method described by Timpa (was adopted for the GPC analysis of the starch samples with some modifications. 45mg of the starch sample were added to 5ml of dimethylacetamide (DMAC) in 10ml Reacta-Vials (Pierce, Rockford, IL) in a heating block. The temperature was raised to 150°C for 1 hr 15 mins, cooled to 100°C and dried LiC1 (to 8% w/v) was added. Further temperature lowering to 50°C was ensured with continuous stirring overnight before incubating until clear solutions were obtained. The resulting solutions were quantitatively diluted to 50 ml with DMAC. Prior to injection, solutions were filtered *in vacuo* through Teflon solvent-resistant disposable filters (Millex SR, 0.5pm, Millipore) using 4ml glass vials (WISP, Waters) in a Baker 10 extraction apparatus fitted with glass syringes (10 cm^3). The mobile-phase solvent for GPC was DMAC containing 5% LiCl. The GPC system consisted of an automatic sampler (Waters WISP) with an HPLC pump (Waters Model 590), detected by multi-angle laser light scattering (MALLS) (DAWN EOS, Wyatt Technology Corp, Santa Barbara, CA) and differential refractive index (RI) (Waters Corp., Milford, MA) detectors. The RI calibration constant was measured with a series of NaCl standards. The 90^o photodiode detector of MALLS was calibrated using toluene (HPLC grade). This system was equipped with four columns (Ultra-styragel 10^3, 10^4 and 10^5 (Baxter, Muskegon, MI) and 10^6 (Phenomenex, Torrance, CA)) connected in series and preceded by a guard column (Phenogel, linear, Phenomenex). The system was maintained at 50°C. Standard injection volume was 40 μL and the mobile phase was pumped at a rate of 1.0 ml min^{-1}. Run times were 60 min. The software package Unical based upon ASYST (Unical, Version 3.02, Viscotek) was used for data acquisition and analysis. The system was calibrated with the polystyrene standards. Data were obtained from two dissolutions per sample with two GPC runs per dissolution.

Pasting Properties

The pasting profiles (pasting temperature, peak viscosity, hot paste viscosity, breakdown, cold paste viscosity, setback and peak time) of the starches were determined using the Rapid Visco Analyzer (model RVA Series 4, Newport Scientific Pvt. Ltd., Warriewood, Australia) following the method described by Karim et al. (2008) with some modifications. The samples (8% w/w, db) were equilibrated at 50 °C for 1 min and then raised to 95 °C in 3.75 min, held for 2.5 min, cooled to 50 °C in 3.75 min, and held for 5 min. The paddle speed was set at 960 rpm for the first 10 s to evenly disperse the starch slurry and reduced to 160 rpm throughout the remainder of the experiment. The units of viscosity were expressed as rapid visco units (RVUs).

Results and Discussion

Functional Properties

Swelling power, solubility and gel strength of the native and pregelatinised starches presented in Table 1. The starches are significantly different ($p<0.05$) in terms of the values of their swelling power, solubility and gel strength. All the pregelatinised starches exhibit lower values of swelling power than the native starches.

This observation may suggest possible disruption in granular network of the starch during the process of modification. Lowering in the extent of granular swelling has been attributed to the increase in granular stability of the starch, when the long amylopectin chains recrystallise (Singh et al., 2004). This may suggest an opinion that pregelatinisation, though a physical process of modification, could disrupt structural network of starch. The behaviours of starches, to treatment and processing, among other factors, depend on their botanical sources. However, among the pregelatinised starches, the swelling power increases with increase in pregelatinised temperature, except for LBS_{50} and LBS_{55}. The solubility values of the pregelatinised starches are significantly higher than their corresponding native starches with deviations observed for LBS_{50} and LBS_{55}. Inhibition of granular swelling and lowered solubility of starch have been adduced to possible increase in amylose-lipid complex formation (Hoover and Manuel, 1996). Conversely, all the pregelatinised starches are relatively weaker than their corresponding native starches with the exception of PPS_{55}. The gels of pregelatinised starches appear weaker than their corresponding native starches with the exception of PPS_{55} (Table 1). This observation may inform the possibility of pregelatinisation process loosening the starch granule structure, leaving weaker cohesive swollen particles in the gel matrix. The observations obtained in this present work are in agreement with the previous report of Adedokun and Itiola (2010) that the gels of natural starches are stronger than their pregelatinised derivatives.

Molecular Mass Distribution

Table 2 presents the molecular weight (MW), that is, average weight (M_w), number average (M_n), weight average degree of polymerisation (DP_w), number average degree of polymerisation (DP_n) and polydispersity index (PDI) of the native and pregelatinised starches. The values of M_w of the native starches increase after pregelatinisation process. An opposite trend is obtained for their values of M_n. The percentage increases in the values of M_w of the pregelatinised starches relative to their native forms are 456.00% for PPS_{50}, 451.75% for PPS_{55}, 211.00% for LBS_{50}, 216.00% for LBS_{55}, 471.43% for JBS_{50} and 475.00% for JBS_{55}. The percentage relative decrease in the values of M_n ranges from -47.00% (in PPS_{50}) to -24.07% (in JBS_{55}). These observations may account for the results obtained for the starches in terms of swelling power and solubility (Table 1). DP_w values of the pregelatinised starches are higher than those of their corresponding native forms. An opposite trend is observed for their values of DP_n. PDI, which is an indication of broadness of molecular weight distribution (Gowariker et al., 1986), increases upon pregelatinisation process. The percentage increases in PDI values of the pregelatinised starches, relative to the native starches, are 437.31%, 430.85%, 44.08%, 46.45%, 359.45%, and 372.81% for PPS_{50}, PPS_{55}, LBS_{50}, LBS_{55}, JBS_{50} and JBS_{55} respectively. Oladebeye et al. (2013) have attributed arbitrarily high increment in PDI and MW to the reorganisation and reinforcement of the fragments of the starch granules.

Pasting Properties

Table 3 shows the pasting profiles of the native and pregelatinised starches. Pasting properties depend on the rigidity of starch

Table 1: Swelling power, solubility and gel strength of native and pregelatinised starches.

Starch	**Swelling Power (g/g)**	**Solubility (%)**	**Gel Strength (N)**
PPS	9.61 ± 0.04^c	3.76 ± 0.05^a	5.23 ± 0.02^b
PPS_{50}	7.32 ± 0.15^a	4.87 ± 0.04^c	0.79 ± 0.01^a
PPS_{55}	8.02 ± 0.23^b	4.70 ± 0.01^b	5.30 ± 0.02^c
LBS	11.61 ± 0.03^c	4.79 ± 0.02^c	5.28 ± 0.02^c
LBS_{50}	10.70 ± 0.22^b	4.19 ± 0.02^a	3.53 ± 0.02^b
LBS_{55}	10.41 ± 0.15^a	4.39 ± 0.01^b	2.76 ± 0.05^a
JBS	10.74 ± 0.07^c	6.22 ± 0.07^a	3.79 ± 0.01^c
JBS_{50}	9.32 ± 0.11^a	7.89 ± 0.03^b	2.64 ± 0.02^b
JBS_{55}	9.56 ± 0.05^b	8.11 ± 0.05^c	2.13 ± 0.01^a

PPS: native pigeon pea starch; PPS_{50}: pregelatinised pigeon pea starch at 50 ^oC; PPS_{55}: pregelatinised pigeon pea starch at 55 ^oC; LBS: native lima bean starch; LBS_{50}: pregelatinised lima bean starch at 50 ^oC; LBS_{55}: pregelatinised lima bean starch at 55 ^oC; JBS: native jack bean starch; JBS_{50}: pregelatinised jack bean starch at 50 ^oC; JBS_{55}: pregelatinised jack bean starch at 55 ^oC; Results are expressed as means±standard deviations ($n = 3$). Values in the same column with the same superscript letters are not significantly different ($p < 0.05$).

Table 2: Molecular mass distribution of native and pregelatinised starches.

Starch	$M_w \times 10^6$	$M_n \times 10^6$	$DP_w \times 10^3$	$DP_n \times 10^3$	**PDI**
PPS	1.14	1.00	7.04	6.17	1.14
PPS_{50}	6.34 (456.00)	0.53 (-47.00)	39.14	3.27	11.96 (949.12)
PPS_{55}	6.29 (451.75)	0.63 (-38.00)	38.83	3.88	9.98 (775.44)
LBS	1.17	1.98	7.22	12.22	0.59
LBS_{50}	3.11 (211.00)	1.05 (-46.97)	19.20	6.48	2.96 (401.69)
LBS_{55}	3.16 (216.00)	1.10 (-46.97)	19.51	6.79	2.87 (386.44)
JBS	1.12	1.08	6.93	6.67	1.04
JBS_{50}	6.40 (471.43)	0.76 (-29.63)	39.51	4.69	8.42 (709.62)
JBS_{55}	6.44 (475.00)	0.82 (-24.07)	39.75	5.06	7.85 (654.81)

M_w: weight average; M_n: number average; DP_w: weight average degree of polymerization (calculated by dividing M_w by 162 i.e. the molecular weight of anhydrous glucose); DP_n: number average degree of polymerization (calculated by dividing M_w by 162 i.e. the molecular weight of anhydrous glucose); PDI: Polydispersity index (M_w/M_n); Values in parentheses are percentage increase (+) and decrease (-) relative to native starch

---

**Corresponding author; E-mail: folabeye@yahoo.com, Tel.: +234 8033620279*

Table 3: Pasting properties of native and pregelatinised starches.

Starch	**Peak Viscosity (RVU)**	**Breakdown (RVU)**	**Setback (RVU)**	**Peak Time (mins)**	**Pasting Temperature (^{o}C)**
PPS	45.42 ± 2.02^{c}	1.67 ± 0.14^{b}	15.11 ± 0.93^{c}	6.94 ± 0.03^{b}	86.54 ± 0.83^{b}
PPS_{50}	7.92 ± 0.04^{a}	1.42 ± 0.08^{a}	5.75 ± 0.12^{a}	6.98 ± 0.01^{b}	85.22 ± 0.01^{b}
PPS_{55}	11.25 ± 0.12^{b}	$2.00\ \pm0.15^{c}$	8.25 ± 0.50^{b}	6.92 ± 0.01^{a}	84.25 ± 0.30^{a}
LBS	84.67 ± 2.47^{c}	21.03 ± 2.44^{c}	10.64 ± 0.97^{c}	5.01 ± 0.10^{a}	86.54 ± 0.75^{a}
LBS_{50}	60.67 ± 0.37^{b}	1.83 ± 0.09^{a}	5.67 ± 0.10^{a}	6.72 ± 0.01^{b}	89.55 ± 0.02^{b}
LBS_{55}	54.67 ± 0.23^{a}	3.33 ± 0.30^{b}	7.50 ± 0.29^{b}	6.92 ± 0.01^{c}	91.95 ± 0.01^{c}
JBS	32.52 ± 1.80^{c}	3.03 ± 0.65^{b}	9.42 ± 0.82^{c}	5.65 ± 0.88^{a}	86.31 ± 1.96^{a}
JBS_{50}	29.67 ± 0.13^{b}	0.83 ± 0.06^{a}	4.25 ± 0.04^{b}	6.92 ± 0.01^{c}	88.85 ± 0.02^{b}
JBS_{55}	27.17 ± 0.22^{a}	0.67 ± 0.30^{a}	2.95 ± 0.23^{a}	6.85 ± 0.03^{b}	91.00 ± 0.01^{c}

Results are expressed as means±standard deviations (n = 3). Values in the same column with the same superscript letters, within the same sample type, are not significantly different (p < 0.05)

granules, which in turn affect the granule swelling potential, and the amount of amylose leaching out in the solution (Wadchararat et al., 2006). The pasting temperatures of the starches are significantly different ($p<0.05$). Upon pregelatinisation, the pasting temperatures of the native starches increase with deviations observed for PPS, PPS_{50} and PPS_{55}.

The peak pasting temperature (91.95 ± 0.01 ^{o}C) is observed in LBS_{55} while the least pasting temperature (84.25 ± 0.30 ^{o}C) in PPS_{55}. The increase in pasting temperature could be adduced to the stability of the starch granule network after the modification process. The values of the peak viscosity, hot paste viscosity and cold paste viscosity of the native starches significantly decrease after pregelatinisation process. These observations may substantiate the justifications for the previous observations given for the reduced swelling profiles of these starches (Table 1). Breakdown is a measurement of the starch granules stability during the pasting process. Extensive breakdown occurs when the starch granules swells to maximum volume, but lacks the ability to retain its structure and subsequently collapses. All pregelatinised starches, except PPS_{50}, exhibit lowered breakdown values than their corresponding native starches. This may suggest the reason for the stability of the granules of the pregelatinised starches to withstand swelling. Setback, a measure of retrogradation, decreases after pregelatinisation of the native starches. Lowering of setback viscosity has been reported as the less tendency of the starch to retrograde (Oladebeye et al., 2013). The results obtained show that the pregelatinisation process inhibits retrogradation of starch. The peak times for the pregelatinised starches used in this study are significantly higher those of their corresponding native starches. The differences in peak time of the starches could indicate differences in intrinsic behaviours and responses of the starch granules to paste formation and botanical variations.

Conclusion

Pregelatinisation of pigeon pea, lima bean and jack bean was successfully carried out. The study has revealed that the swelling profiles of the native starches were lowered after pregelatinisation whereas an opposite trend was observed for their solubility profiles. All the gels of the native starches were stronger than their pregelatinised derivatives with the former possessing less tendency to retrograde upon storage. This was evident in the possibility of their starch granule network becoming more stable than their native forms. The native starches could, however, be more suitable in pharmaceuticals than their pregelatinised derivatives. In addition, in textiles, the pregelatinsed starches could be suitable.

References

Adebayo, A.S., Itiola, O.A. (1998) Evaluation of breadfruit and cocoyam starches as exo-disintegrants in a paracetamol tablet formulation. *Pharmacy and Pharmacology Communications*, **4**:385–389.

Adedokun, M., Itiola, O.A. (2010) Material properties and compaction characteristics of natural and pregelatinized forms of four starches. *Carbohydrate Polymers*, **79**:818–824.

Alebiowu, G., Itiola, O.A. (2002) Compressional characteristics of native and pregelatinized forms of sorghum, plantain, and corn starches and the mechanical properties of their tablets. *Drug Development and Industrial Pharmacy*, **28**:663–672.

Gowariker, V.R., Viswanathan, N.V., Sreedhar, J. (1986) Polymer Science. Wiley Eastern Limited, New Delhi.

Gunaratne, A., Corke, H. (2007) Functional properties of hydroxypropylated, cross-linked, and hydroxypropylated cross-linked tuber and root starches. *Cereal Chemistry*, **84(1)**:30–37.

Herman, J., Remon, J.P., DeVilder, J. (1989) Modified starches as hydrophilic matrices for controlled oral delivery I: Production and characterization of thermally modified starches, *International Journal of Pharmaceutics*, **56**:51–66.

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 starch. *Journal of Cereal Science*, **23**:153–162.

Karim, A.A., Sufha, E.H., Zaidul, I.S.M. (2008) Dual modification of starch via partial enzymatic hydrolysis in the granular state and subsequent hydroxypropylation. *Journal of Agricultural and Food Chemistry*, **56**:10901–10907.

Kottke, M.K., Chueh, H.R., Rhodes, C.T. (1992) Comparison of disintegrant and binder activity of three corn starch products. *Drug Development and Industrial Pharmacy*, **18**:2207–2223.

Leach, H.W., McCowen, L.D., Schoch, T.J. (1959) Structure of the starch granule I. Swelling and solubility patterns of various starches. *Cereal Chemistry*, **36**:534–544.

Lee, J.S., Kumar, R.N., Rozman, H.D., Azemi, B.M.N. (2004) Flow behaviour of sago starch-g-poly(acrylic acid) in distilled water and NaOH–effect of photografting. *Carbohydrate Polymers*, **56**:347–354.

Oladebeye, A.O., Oshodi, A.A., Amoo, I.A., Karim, A.A. (2013) Hydroxypropyl derivatives of legume starches: Functional, rheological and thermal properties. *Starch/Starke*, **65**:762–772.

Sanchez, L., Santiago, T., Lastres, J.L. (1995) Gelatinized–freeze-dried starch as excipient in sustained-release tablets.

International Journal of Pharmaceutics, **115**:201–208.

Singh, N., Singh, J., Kaur, L. (2004) Factors influencing the properties of hydroxypropylated potato starches. *Carbohydrate Polymers*, **55**:211–223.

Timpa, J.D. (1991) Application of universal calibration in gel permeation chromatography for molecular weight determination of plant cell wall polymers: cotton fiber. *Journal of Agricultural and Food Chemistry*, **39**:270–275.

Wadchararat, C., Thongngam, M., Naivikul, O. (2006) Characterisation of pregelatinised and heat moisture treated rice flouyrs. *Kasetsart Journal (Nature and Science)*, **40:**144–153.