Preparation and characterization of physicochemical properties of glacial acetic acid modified Gadung (*Dioscorea hispida* Dennst) flours

Andri Cahyo Kumoro · Rizka Amalia ·
Catarina Sri Budiati · Diah Susetyo Retnowati ·
Ratnawati Ratnawati

Revised: 30 December 2014 / Accepted: 7 January 2015 / Published online: 22 January 2015
© Association of Food Scientists & Technologists (India) 2015

Abstract In addition to the presence of antinutrients, the inferior physicochemical properties of flours has caused gadung (*Dioscorea hispida dennst*) becomes one of the underutilized tubers in the world. Acetylation is one of the starch modification methods to alter the physicochemical properties of starch, namely swelling power and solubility. The objective of this work was to investigate the effect of reaction time, glacial acetic acid/flour mass ratio and pH on gadung flour acetylation at ambient temperature. The acetylation was carried out by reacting gadung flour slurry of 20 % consistency with glacial acetic acid under alkaline condition. The results show that in general degree of substitution and swelling power increased with the increase of reaction time, while the solubility was not affected by reaction time after 10 min acetylation. Acetylation led to significant changes in morphology and structure of gadung flour starch granules. Overall, all the acetylated gadung flours obtained in this work were having higher swelling power and solubility than the native flour. Acetylation of gadung flour using glacial acetic acid with 1:3 mass ratio and pH 8.0 at ambient temperature for 30 min resulted gadung flours with swelling power and solubility similar to that of Korean wheat flour.

Keywords Gadung flour · Acetylation · Swelling power · Solubility · Degree of substitution · Morphology

Introduction

Gadung (*Dioscorea hispida* Dennst.) remains as one of the underutilized tubers, that is easily found in most Asia and Pacific regions during the dry season. Thanks to its carbohydrate content, this tuber has been used as a staple food during World War II, especially by people in the tropical and sub-tropical regions (Liu et al. 2006). The resistant starch contained in this food source promotes slow digestion in the lower parts of the human gastrointestinal tract, resulting in slow liberation and absorption of glucose. Aprianita et al. (2009) suggested that this digestive property has shown the potentials of gadung tuber in reducing the risks of being obesity, diabetes and other related diseases. In addition, gadung tuber also does not contain gluten, which makes it becomes an important substance in the reduction in the incidence of celiac disease (CD) or other allergic reactions (Rekha and Padmaja 2002). Apart to its superiorities, gadung tuber also possesses drawbacks, specifically related to its antinutritional substances contents, such as dioscorine and cyanides. To prevent serious dietary cyanides exposure, the glucosides and their derivative substances, popularly known as cyanogens, must be removed from the food source before being consumed. Effective processing modes may reduce all cyanogens in cassava and gadung tubers to below the safe level of 10 mg HCN equivalent per kg body weight as set by WHO 1988 (Mlingi et al. 1995; Kumoro et al. 2011).

A. C. Kumoro (✉) · R. Amalia · C. S. Budiati · D. S. Retnowati ·
R. Ratnawati
Department of Chemical Engineering, Faculty of Engineering,
Diponegoro University, Prof. H. Soedarto, SH Road-Tembalang,
Semarang 50275, Indonesia
e-mail: andrewkomoro@undip.ac.id

A. C. Kumoro · R. Amalia
Master of Chemical Engineering Program, Department of Chemical
Engineering, Faculty of Engineering, Diponegoro University, Prof.
H. Soedarto, SH Road-Tembalang, Semarang 50275, Indonesia

Tuber flours, which contain native starches, have been used since ancient times as raw material to prepare a variety of products. Starch is utilized for its various functionalities in thickening, stabilizing, texturizing, gelling, film forming, encapsulation, moisture retention and shelf life extension (Singh et al. 2004). They are also used to help retain moisture in baked goods, as anticaking agents in baking powder and as a molding medium in confections (Considine and Considine 1982). However, they have limitations i.e. low shear stress resistance, thermal decomposition, high retrogradation, and syneresis that reduce their use in industrial applications. The hydrophilic nature of starch is a major constraint that seriously limits the development of starch based products; so starch modification is a way to solve the problem and produce water-resistant materials (Peltonen and Harju 1996).

Starches can be modified by chemical, physical, and enzymatic methods to improve desirable functional properties (Agboola et al., 1991). Among chemically modified starches is the acetylated starch, which is obtained by esterification of native starch with acetic anhydride (Jarowenko 1986). Acetylated starch is a granular starch ester with the CH_3CO group introduced at low temperature (Singh et al. 2004). The modified starches generally show better paste clarity, stability, increased resistance to retrogradation, and increased freeze-thaw stability (Agboola et al., 1991). Rutenberg and Solarek (1984) reported that the introduction of acetyl groups upon acetylation reduces the bond strength between starch molecules and thereby increases the swelling power and solubility of the starch granule, decreases the coagulation of the starch, and provides improved freeze-thaw stability. Phillips et al. (1999) found that the extent of physicochemical property changes in the acetylated starch compared to the native starch is proportional to the degree of acetylation or degree of C=O substitution incorporated into the starch molecules. Acetylated starches are widely used in a large variety of foods including baked goods, canned pie fillings, sauces, retorted soups, frozen foods, baby foods, salad dressings, and snack foods (Wurzburg 1978). As in all chemical reactions, acetylation depends upon factors such as reactant concentration, reaction time, pH, and the presence of catalyst (Whistler and Daniel 1995), which finally determine the number of acetyl groups incorporated into the molecule. The objective of this study was to evaluate the effect of reaction time, glacial acetic acid:flour mass ratio and pH on the swelling power and solubility of glacial acetic acid modified gadung tuber flour.

Materials and methods

Materials

Native gadung flour used in this work was obtained from milling and sieving of treated gadung tuber chips using a

method suggested by Kumoro et al. (2011) and Kumoro et al. (2012). Glacial acetic acid and other chemicals used in this work were of analytical grade ($\geq 99.90\%$ w/w) were purchased from Sigma-Aldrich Pte. Ltd. (Singapore) and directly used without further purification.

Apparatus

The acetylation of gadung flour study in this work employed mainly a 500 mL capacity beaker glass and a hot plate magnetic stirrer. While the swelling power and solubility analyses used glass tubes, centrifuge and analytical balance.

Procedure

The acetylation was carried out by dispersing 20 g of gadung flour in 100 mL distilled water to obtain gadung flour slurry with 20 % consistency. To facilitate flour dispersion, a magnetic stirrer was employed for agitation. The pH of the dispersed system was then adjusted to a certain value (8, 9 or 10) by addition of 1 M NaOH. Then, a predetermined mass of glacial acetic acid was added to the reaction mixture which caused a drastic reduction of pH. The pH was brought back to the initial condition (8, 9 or 10) by addition of 100 mL 1 M NaOH. The acetylation reaction was let to occur for 10 to 60 min before being terminated by bringing the pH to 5.5 through addition of 1 M HCl. The acetylated flours were obtained by filtration of the reaction mixture and followed by threefold washing of the flours using distilled water with 1:3 (w/v). The acetylated flours were then dried in an electric oven at 50 °C till dryness before subjected to degree of substitution, swelling power and solubility analysis.

Analyses

The degree of substitution (DS) was determined titrimetrically, following the method of Sodhi and Singh (2005). Acetylated starch (1.0 g) was placed in a 250 ml flask and 50 ml of 75/100 mL ethanol in distilled water were added. The loosely stopper flask was agitated, warmed to 50 °C for 30 min, cooled and 40 ml of 0.5 mol/L KOH were added. The excess alkali was back-titrated with 0.5 mol/L HCl using phenolphthalein as an indicator. A blank, using the original unmodified starch, was also used. Degree of substitution is defined as the average number of sites per glucose unit that possess a substituent group.

$$\text{Acetyl \%} = \frac{[(\text{blank}(\text{ml}) - \text{sample}(\text{ml})) \times \text{Molarity of HCl} \times 0.043 \times 100]}{\text{Sample weight (g)}} \quad (1)$$

$$\text{DS} = \frac{(163 \times \text{Acetyl}(\%))}{[4300 - (42 \times \text{Acetyl}(\%))] } \quad (2)$$

Solubility (WS) and swelling power (SP) of gadung flours were determined by modifying the method of Li and Yeh (2001). A starch sample 1.0 g (W_0) was accurately weighed and quantitatively transferred into a clear dried test tube and re-weighed as W_1 . The starch was then dispersed in 50 mL of distilled water. The resultant slurry was heated at 60 °C for 30 min, with constant mixing. The mixture was cooled to 30 °C and centrifuged at 100×g for 15 min in a Superspeed centrifuge (Sorvall® RC-6, Kendro laboratory products, NC, USA). Aliquots (5 mL) of the supernatant were dried to a constant weight at 110 °C and denoted as W_c . The residue obtained after drying the supernatant represented the amount of starch solubilized in water. Solubility was calculated as g per 100 g of starch on a dry weight basis.

$$WS = \frac{W_c}{W_0} \times \frac{50}{5} \times 100 \quad (3)$$

The supernatant was separated and swollen starch as the precipitate was weighted. The residue obtained from the above experiment (after centrifugation) with the water it retained was quantitatively transferred to the clean dried test tube used earlier and weighed (W_2). To calculate the swelling power, the weight of residue was divided by the original weight after solubility subtraction (Osundahunsi et al. 2003).

$$SP = \frac{W_2 - W_1}{W_0 \times (100 - WS)} \quad (4)$$

Results and discussion

The proximate analysis of native gadung flour

Proximate analysis of food materials is aimed to determine their macro components content, such as carbohydrate, protein, fat, crude fiber, ash and moisture. Proximate composition of gadung tubers, gadung tuber flour and its comparison with other tuber crops are presented in Table 1.

As seen in Table 1, the carbohydrate content in the gadung tuber is on par with other tubers, which is about 20/100 g edible portion. Carbohydrate content in gadung tuber flour is high and makes gadung tubers can be one of reliable food sources ensure food security, especially during drought. Gadung tuber protein content was 1.81/100 g, slightly higher than that of its closest relatives, the *Dioscorea esculenta* tuber (Polycarp et al. 2012). Unfortunately, gadung tuber protein content is less than half of the protein content in taro (*Colocasia esculenta*) tuber (Sefa-Dedeh and Agyir-Sackey

Table 1 Chemical composition of gadung tuber and flour and their comparison with other tuber flours (g/ 100 g)

Component	Gadung tuber	Gadung flour	Colocasia esculenta flour ^a	Dioscorea esculenta flour ^b
Carbohydrate	18	92.3	17,8	19,38
Protein	1.81	1.1	4,69	1,26
Fat	1.6	0.9	0,97	0,18
Fiber	1.9	2.4	2,8	0,49
Ash	0.7	0.3	1,88	1,19
Moisture	77	3	72,1	77,15

^a Sefa-Dedeh and Agyir-Sackey (2004)

^b Polycarp et al. (2012)

2004). Therefore, protein enrichment is necessary if gadung tubers will be used as food material. High fat content in gadung tubers is offset by its high crude fiber content. Therefore, gadung tubers are best consumed by people suffering from indigestion. Although the ash content is less than half of that of taro tubers, but the ash content in tubers of yam tuber is also almost equivalent to *Dioscorea esculenta* tuber. However, high moisture content in the gadung tuber (77/100 g) makes this tuber vulnerable to deterioration by fungal attacks. That is why; this research suggests gadung tubers to be stored in the form of flour with moisture content not exceeding 3/100 g so that tuber damage by fungi and other microfloral activities can be avoided. Table 1 also shows that attempt to transform gadung tubers into gadung tuber flour causes only a slight decrease in of protein, fat and ash content, but increased carbohydrates and crude fibre contents.

Effect of glacial acetic acid/gadung flour mass ratio on water solubility (WS), swelling power (SP) and degree of substitution (DS) of acetylated gadung flours at pH 8

While most acetylation of starch is conducted using nucleophilic agents such as acetic anhydride of vinyl acetate, this work employs glacial acetic acid as a modifying agent. The formers are more reactive, but they are also more expensive and hazardous to human. Acetylation of gadung flours was carried out using slurry method employed distilled water as a dispersing agent and sodium hydroxide as a catalyst at ambient temperature. Effect of glacial acetic acid:gadung flour mass ratios on water solubility and swelling power of acetylated gadung flour is presented in Table 2.

As seen in Table 2, raw gadung flour has low swelling power and water solubility, which are 6.9 (g/g) and 4 (g/100 g), respectively. These values are slightly higher than those reported by Tattiyakul et al. (2007) for taro tuber flour, which were 2.5 (g/g) and 3.5 (g/100 g). While Adebawale et al. (2002) reported swelling power and water solubility of Bambarra groundnut flour, which were 2.59 g/g and 5.5 (g/100). The low swelling power and water solubility of

Table 2 Effect glacial acetic acid/gadung flour mass ratio on water solubility (WS), swelling power (SP) and degree of substitution of acetylated gadung flours at pH 8

Time (minutes)	Glacial acetic acid/gadung flour ratio								
	1:3			1:4			1:5		
	WS (g/100 g)	SP (g/g)	DS	WS (g/100 g)	SP (g/g)	DS	WS (g/100 g)	SP (g/g)	DS
0	4.0	6.9	0	4.0	6.9	0	4.0	6.9	0
10	8.0	7.2	0.16	6.0	7.1	0.17	4.0	7.1	0.17
20	9.0	7.2	0.17	6.0	7.2	0.17	4.0	7.2	0.18
30	9.3	7.3	0.18	6.0	7.6	0.18	4.0	7.3	0.18
40	6.0	7.9	0.18	6.0	7.8	0.18	4.0	8.6	0.20
50	6.0	8.1	0.19	6.0	8.1	0.19	4.0	8.5	0.18
60	6.0	8.4	0.17	6.0	8.3	0.18	4.0	8.8	0.17
^a Korean wheat flour	WS: 7.8–9.3			SP: 7.3–8.5					
^a American wheat flour	WS: 6.8–7.9			SP: 6.3–7.3					

^a Chung et al. (2010)

gadung flour is mainly related to its high amylose content. The amylose content of gadung starch and flour are 39.30 and 34.72 %, respectively (Tattiyakul et al. 2006; Kumoro et al. 2012). High amylose content in the starch granules causes the amylose molecules in the form of crystalline become more compact and interwoven by amylopectin. These structures form the surface of the starch granules become almost solid and hinder the diffusion of water molecules into starch granules (Violeta et al. 1998). High fat content in the gadung flour also allows the formation of complexes with amylose, which also inhibit amylose leaching and granules swelling. In addition to amylose-amylopectin ratio, swelling power and water solubility is also influenced by the molecular weight distribution of amylose and amylopectin, the degree and length of branches and conformation (Hoover 2001).

Table 2 also shows that the more glacial acetic acid used as shown by larger mass ratio of glacial acetic acid: gadung flour increased the water solubility of gadung flour. Similar result was also reported by Singh et al. (2004) on acetylation of corn and potato starches, and Raina et al. (2006) who studied acetylation of rice starch. As seen in Table 2, prolonging reaction time also improve water solubility of gadung starch in accordance with increasing degree of substitution. High amylose content in gadung flour slower the acetylation reaction speed and finally slower the increase of degree of acetylation. At low degree of substitution, the starch granules of gadung flour are predominantly in the form of crystalline or retrograded, which are insoluble in water at ambient temperature. Low acetylation reaction speed with an increase in amylose content might be due to amylose crystal or complexes. In its crystalline form,

Table 3 Effect of pH on water solubility (WS) and swelling power (SP) and degree of substitution (DS) of acetylated gadung flours at glacial acetic acid/gadung flour mass ratio 1:3

Time (minutes)	pH								
	8			9			10		
	WS (g/100 g)	SP (g/g)	DS	WS (g/100 g)	SP (g/g)	DS	WS (g/100 g)	SP (g/g)	DS
0	4.0	6.9	0	4.0	6.9	0	4.0	6.9	0
10	8.0	7.2	0.16	4.0	6.5	0.20	4.0	8.1	0.20
20	9.0	7.2	0.17	4.0	6.7	0.21	4.0	8.7	0.23
30	9.3	7.3	0.18	4.0	6.9	0.22	4.0	7.8	0.23
40	6.0	7.9	0.18	4.0	7.4	0.23	4.0	7.4	0.24
50	6.0	8.1	0.19	4.0	7.2	0.21	4.0	7.5	0.23
60	6.0	8.4	0.17	4.0	7.6	0.20	4.0	8.9	0.22
^a Korean wheat flour	WS: 7.8–9.3			SP: 7.3–8.5					
^a American wheat flour	WS: 6.8–7.9			SP: 6.3–7.3					

^a Chung et al. (2010)

Fig. 1 Morphology of gadung flours before (a) and after acetylation (b) as analysed using SEM imaging (100× magnification)

amylose usually only melts at higher temperature than amylopectin (Shogren 1992), therefore amylose is more resistant to swelling and reaction. At reaction time longer than 40 min and glacial acetic acid:gadung flour ratio 1:3, a reduction in water solubility of acetylated gadung flour was observed. Shogren and Biswas (2006) reported that at low temperature, acetylation occurred at slow speed until the degree of substitution 0.3 was achieved at 40 min. They also reported that water solubility of acetylated high amylose corn starch decrease if degree of substitution is higher than 1. This is possibly due to an increase in hydrophobicity of acetyl groups. As the degree of substitution increases, the number of substituted hydroxyl groups by acetyl groups also increased. This condition increases starch hydrophobicity and reduces hydrogen bonding in the starch molecules and finally retards hydration of starch (Luo and Shi 2012). Incorporation of acetyl groups to starch molecules reduces the interaction between starch molecules which increase water solubility and swelling power of starch granules. The introduction of acetyl groups into starch molecules also reduces the interaction between outer chains of amylopectin and interaction between amylose chains (Jarowenko 1986). Acetylation also provides a way for water molecules to move toward the amorphous parts of starch caused by changes in the arrangement of intragranular structure as a result of steric effects and destruction of hydrogen bonds in starch grains (González and Perez 2002). In the starch molecule, there are three free hydroxyl groups with different levels of reactivity. Primary OH group located at

position C₆ is highly reactive and will be immediately react with the acetyl group when compared with the two secondary OH groups at positions C₂ and C₃ as a result of steric hindrance. Primary OH groups are located on the outer surface of the starch molecules so that they can immediately react with the acetyl group. Meanwhile, two secondary OH groups located in the inner surface of the starch molecules form hydrogen bonds with the OH groups owned by nearby glucose units. Among the secondary OH groups, the OH group on C₂ is more reactive than the OH group at C₃. This happens because the OH group at C₂ is more acidic and closer to the hemiacetal (Fedorova and Rogovin 1963).

Gadung flour swelling power after being modified with glacial acetic acid is higher than the native flour. However, acetylated gadung flour swelling power is not significantly influenced by the amount of acetic acid used (mass ratio of glacial acetic acid/gadung flour) in the process of acetylation. As expected, acetylated gadung flour swelling power continues to increase with increasing reaction time. As shown in Table 2, the swelling power values of acetylated gadung flour at certain reaction time (0–30 min) but with different ratio of glacial acetic acid/gadung flour are very close to each other. Similar result is also reported by Singh et al. (2004) on the acetylation of corn starch. This phenomenon is thought to be caused by the presence of fat in the gadung flour (Galliard and Bowler 1987). Because acetylation on the study was carried out at low temperatures, the acetylation occurred only in the amorphous starch grains, causing swelling in that section.

Fig. 2 Morphology of gadung flours before (a) and after acetylation (b) as analysed using SEM imaging (1000× magnification)

Leaching of amylose into water almost impossible at low temperatures, but with the strong bonds between the water molecules with the outer branches of the starch molecules will form a gel (Shogren and Biswas 2006).

Gadung flour would be modified to suit industrial applications with very low level of acetate group donors. de Graaf et al. (1998) suggested that acetylated starches with degree of substitution (DS) between 0.01 and 0.2 are approved by the FDA for used as food materials. Their usage is based on properties with respect to film forming, binding, adhesivity, thickening, stabilizing and texturing. With that point of view, acetylation of gadung flour using glacial acetic acid is targeted to obtain acetylated gadung flour with DS within 0.01–0.2. The degree of substitution increases with reaction time. However, the degree of substitution did not significantly affected by mass ratio of glacial acetic acid to starch. This finding is consistent with that reported by Singh et al. (2004) who reported that DS increased progressively with increase in level of acetic anhydride (up to 8 %) during the acetylation of potato starch using acetic anhydride. However, any further increase in level of acetic anhydride (up to 12 %) did not result in an increase of DS. With glacial acetic acid:gadung flour mass ratios of 1:3, 1:4 and 1:5, the glacial acetic acid concentrations used in this work were corresponding to between 16.67 and 25 %, which were higher than 12 %. The longer the reaction time, the more time is available for an acetyl group to diffuse and undergo adsorption to the surface of starch molecules (Khalil et al. 1995). However, this increase is not linear. The reaction takes place very rapidly at the beginning of the reaction (first 10 min) and then slowed down after the reaction lasted for 20 min. This can be explained by the fact that as the acetylation reaction proceeded, the glacial acetic acid is depleted. In addition, the reactive sites on the starch molecules are also increasingly reduced as a result of structural modification of the starch molecule backbone (Waly et al. 1998). Instead of being affected by esterification reaction, the lower the concentration of acetyl groups in the reaction medium with increasing reaction time can also be caused by hydrolysis of the ester starch (starch acetate) produced or elimination reactions as a result of excess water (from starch and byproducts esterification) are not discharged from the reactor (Luo and Zhou 2012) during the reaction. However, the highest degree of substitution obtained from gadung tuber flour acetylation with glacial acetic acid at 40 min was only 0.24, or still lower when compared with the degree of substitution of corn starch, which is about 0.27 (Luo and Shi 2012). This is likely due to differences in amylose content and structure, grain size and fragility of yam tuber starch in the flour and corn starch.

Based on the value of swelling power and water solubility of acetylated gadung tuber flour obtained, the best reaction conditions chosen were the ratio of glacial acetic acid/gadung tuber flour 1:3, pH 8, and the reaction time of 30 min. Acetylated gadung flour obtained has swelling power

and water solubility 7.3 (g/g) and 9.3 (g/100 g), and meet the standards of water solubility and swelling power of Korean wheat flour (Chung et al. 2010).

Effect of pH on water solubility (WS), swelling power (SP) and degree of substitution (DS) of acetylated gadung flours at glacial acetic acid/gadung flour mass ratio 1:3

The pH of the flour slurry determines the successfulness of esterification reaction between starch and carboxylic acid molecules. The effect of pH on the swelling power and water solubility of acetylated gadung flours is detailed in Table 3. Table 3 shows that at pH 9 and 10, the water solubility of acetylated gadung flours were the same as the native one. Acetylation at moderate alkalinity did not change the water solubility of gadung flour. However, acetylation of gadung flours at low alkalinity (pH 8) results flours with higher water solubility. Similar phenomenon was also found by Adebawale et al. (2002) during their study on acetylation of Bambarra groundnut flour. The increase in water solubility of flour is a result of an increase in starch hydrophobicity. In addition, the water solubility of the starch also depends on degree of substitution and polymerization (Krucer and Rutenberg 1976).

Table 3 also reported that in general the swelling power of acetylated gadung flours were higher than that of native gadung flour. Highest increase in swelling power was found for acetylation at pH 10, while the lowest was observed during acetylation at pH 9. Adebawale et al. (2002) reported that pH did not significantly affect the swelling power of acetylated Bambarra groundnut flour and starch. Only slight changes in swelling power values were found during acetylation of Bambarra groundnut in either acidic or basic conditions. They also reported that the increase in swelling power was more pronounced under basic condition (pH 8–10) than acidic condition (pH 2–6). This phenomenon might be due to interaction between protein and starch. Under basic condition, both starch and protein bring positive charge, while under acidic condition only protein brings positive charge (Shieldneck and Smith 1971). Therefore, swelling of starch granules is more pronounced under alkaline condition, and depends on the protein content of the starch.

Table 3 shows that the degree of substitution increases with pH of the reaction medium. As a catalyst, sodium hydroxide initiates the reaction by forming alcoholate ions along the starch polymer, resulting in activated starch molecules (Funke and Lindhauer 2001). Song et al. (2006) reported that at pH 9.5, side reaction (acetate starch hydrolysis) will be the dominant reaction. In contrast to that, at pH 7.5 hydroxyl groups in the starch molecules are not activated enough to perform nucleophilic substitution attack to the unsaturated electrophilic carbon of acetic acid molecules (Hui et al. 2009). The nucleophilic reaction involves addition and elimination mechanisms (Roberts 1965). Jeon et al. (1999) dan

Song et al. (2006) found that the optimum condition for esterification of rice starch was at pH 8.5.

As seen in Table 3, pH 8 is the preferred esterification condition for gadung flour as at this condition the reaction yielded gadung flour with similar swelling power and water solubility with Korean wheat flour (Chung et al. 2010). Hui et al. (2009) also found that pH 8 is the best reaction condition for esterification of potato starch with anhydride octenyl succinate. In addition, Bhosale and Singhal (2006) also reported pH 8 as the optimum pH for esterification of corn and amaranth starches with anhydride octenyl succinate.

Morphology of the native and acetylated gadung flours

The micrographs of native and acetylated gadung flours obtained from *Scanning Electron Microscopy* analysis are presented in Figs. 1 and 2. As seen in those figure, the structure and morphology of gadung flours changed significantly in their size and shape after being modified by acetylation. Similar results were reported by Singh et al. (2004) on acetylated corn and potato starch.

Figure 2 shows that the surface of acetylate gadung flours resembles flat flower petals. The wavy surface of acetylated gadung flours was a result of gelatinization of the starch surface when NaOH added to the reaction system to maintain the pH during the introduction of glacial acetic acid as reported by Luo and Shi (2012) on their corn starch acetylation study. In addition, as the acetylation took place the starch granules developed blisters or small protuberances on their surfaces. The acetylated gadung flour starch granules size was within 25–70 μ , and far bigger than the native ones. Acetylation causes aggregation or cluster formation of starch granules in the gadung flours. The agglomeration takes place as a result of introduction of hydrophilic acetyl groups into starch molecules, which increases the number and strength of hydrogen bonding. Then, the starch molecules are very close to each other forming starch clusters and finally combine to form aggregates (Singh et al. 2004).

Conclusion

From the experimental results, a relatively good processing condition for the production of acetylated gadung tuber flour is through acetylation at a weight: volume ratio of gadung tuber flour: water (1:4), the mass ratio of glacial acetic acid: gadung tuber flour (1:3), pH 8.0 and reaction time 30 min. Implementation of this process condition results acetylated gadung flours of 0.18° of substitution with swelling power and water solubility values similar to Korean wheat flour. The morphology and structure of gadung tuber flours are altered by acetylation.

Acknowledgments The authors would like to express their gratitude to the Directorate General of Higher Education, Ministry of National Education The Republic of Indonesia for its financial support through Hibah Penelitian Strategis Nasional/ National Strategic Research Grant 2013 under contract No. 96/SP2H/PL/Dit. Litabmas/TV/2013.

References

- Adebowale KO, Afolabi TA, Lawal OS (2002) Isolation, chemical modification and physicochemical characterisation of Bambarra groundnut (*Voandzeia subterranean*) starch and flour. *Food Chem* 78:305–311
- Agboola SO, Akingbala JO, Oguntimein GB (1991) Physicochemical and functional properties of low DS cassava starch acetates and citrates. *Starch/Stärke* 43(2):62–66
- Aprianita A, Purwandari U, Watson B, Vasiljevic T (2009) Physicochemical properties of flours and starches from selected commercial tubers available in Australia. *Int Food Res J* 16:507–520
- Bhosale R, Singhal R (2006) Process optimization for the synthesis of octenyl succinyl derivative of waxy corn and amaranth starches. *Carbohydr Polym* 66:521–527
- Chung SY, Han SH, Lee SW, Rhee C (2010) Physicochemical and bread-making properties of air flow pulverized wheat and corn flours. *Food Sci Biotechnol* 19(6):1529–1535
- Considine PE, Considine GD (1982) *Foods and food production encyclopedia*. Van Nostrand Reinhold Company (VNR), New York
- de Graaf RA, Broekroelofs A, Janssen LPBM (1998) the acetylation of starch by reactive extrusion. *Starch Stärke* 50(5):198–205
- Fedorova AF, Rogovin ZA (1963) Relative reactivity of cellulose hydroxyls on esterification in an acid medium. *Vysoko molekulyamyte Soedineniya* 5:519–523
- Funke U, Lindhauer MG (2001) Effect of reaction conditions and alkyl chain lengths on the properties of hydroxyalkyl starch ethers. *Starch Stärke* 53:547–554
- Galliard T, Bowler P (1987) Morphology and composition of starch. In: Galliard T (ed) *Starch properties and potential*. Wiley, Chichester, pp 57–78
- González Z, Perez E (2002) Effect of acetylation on some properties of rice starch. *Starch Stärk* 54:148–154
- Hoover R (2001) Composition, molecular structure, and physicochemical properties of tuber and root starches: a review. *Carbohydr Polym* 45: 253–267
- Hui R, Chen QH, Fu ML, Xu Q, He GQ (2009) Preparation and properties of octenyl succinic anhydride modified potato starch. *Food Chem* 114:81–86
- Jarowenko W (1986) Acetylated starch and miscellaneous organic esters. In: Wurzburg OB (ed) *Modified starches: properties and uses*. CRC Press, Boca Raton, pp 55–77
- Jeon YS, Viswanathan A, Gross RA (1999) Studies of starch esterification: reactions with alkenylsuccinates in aqueous slurry systems. *Starch Stärke* 51:90–93
- Khalil MI, Hashem A, Hebeish A (1995) Preparation and characterization of starch acetate. *Starch* 47:394–398
- Kruger LH, Rutenberg MW (1976) Production and uses of starch acetates. In: Whistler RL, Paschall EF (eds) *Starch: chemistry and technology*. Academic, New York, pp 369–401
- Kumoro AC, Retnowati DS, Budiyati CS (2011) Removal of cyanides from Gadung (*Dioscorea hispida* Dennst) tuber chips using leaching, steaming techniques. *J Appl Sci Res* 7(12):2140–2146
- Kumoro AC, Retnowati DS, Budiyati CS, Manurung T, Siswanto (2012) Water solubility, swelling and gelatinization properties of raw and ginger oil modified Gadung (*Dioscorea hispida* Dennst) flour. *Res J Appl Sci Eng Technol* 4(17):2854–2860

- Li YJ, Yeh AI (2001) Relationships between thermal, rheological characteristics and swelling power for various starches. *J Food Eng* 50: 141–148
- Liu Q, Donner E, Yin Y, Huang RL, Fan MZ (2006) The physicochemical properties and in vitro digestibility of selected cereals, tubers, and legumes grown in China. *Food Chem* 99:470–477
- Luo ZG, Shi YC (2012) Preparation of acetylated normal, waxy and high amylose maize starch with intermediate degrees of substitution in aqueous solution and their properties. *J Agric Food Chem* 60:9468–9475
- Luo ZG, Zhou Z (2012) Homogeneous synthesis and characterization of starch acetates in ionic liquid without catalyst. *Stärke* 64:37–44
- Mlingi NLV, Bainbridge ZA, Poulter NH, Rosling H (1995) Critical stages in cyanogen removal during cassava processing in southern Tanzania. *Food Chem* 53:29–33
- Osundahunsi OF, Fagbemi TN, Kesselman E, Shimoni E (2003) Comparison of the physicochemical properties and pasting characteristics of flour and starch from red and white sweet potato cultivars. *J Agric Food Chem* 51:2232–2236
- Peltonen S, Harju K (1996) Application and methods of preparation of fatty acid esters of polysaccharides. US Patent 5:589577
- Phillips DL, Huijium L, Duohai P, Harold C (1999) General application of raman spectroscopy for the determination of level of acetylation in modified starches. *Cereal Chem* 76:439–443
- Polycarp D, Afoakwa EO, Budu AS, Otoo E (2012) Characterization of chemical composition and anti-nutritional factors in seven species within the Ghanaian yam (*Dioscorea*) germplasm. *Int Food Res J* 19(3):985–992
- Raina C, Singh S, Bawa AS, Saxena DC (2006) Some characteristics of acetylated, cross-linked and dual modified Indian rice starches. *Eur Food Res Technol* 223:561–570
- Rekha MR, Padmaja G (2002) Alpha-amylase inhibitor changes during processing of sweet during processing of sweet potato and taro tubers. *Plant Food Hum Nutr* 52:285–294
- Roberts JH (1965) Nondegradative reaction of starch. In: Whistler RL, Paschall EF (eds) *Starch chemistry and technology*. Academic, New York, pp 1439–1487
- Rutenberg MW, Solarek D (1984) Starch derivatives: production and uses. In: Whistler RL, BeMiller JN, Paschall EF (eds) *Starch chemistry and technology*, 2nd edn. Academic, New York, pp 311–388
- Sefa-Dedeh S, Agyir-Sackey EK (2004) Chemical composition and the effect of processing on oxalate content of cocoyam *Xanthosoma sagittifolium* and *Colocasia esculenta* cormels. *Food Chem* 85: 479–487
- Shieldneck P, Smith CE (1971) Production and uses of acid modified starch. In: Whistler RL, Paschall EF (eds) *Starch: chemistry and technology*. Academic, New York, pp 173–215
- Shogren RL (1992) Effect of moisture content on the melting and subsequent physical aging of corn starch. *Carbohydr Polym* 19:83–90
- Shogren RL, Biswas A (2006) Preparation of water-soluble and water-swelling starch acetates using microwave heating. *Carbohydr Polym* 64:16–21
- Singh N, Chawla D, Singh J (2004) Influence of acetic anhydride on physicochemical, morphological and thermal properties of corn and potato starch. *Food Chem* 86:601–608
- Sodhi NS, Singh N (2005) Characteristics of acetylated starches prepared using starches separated from different rice cultivars. *J Food Eng* 70: 117–127
- Song X, He G, Ruan H, Chen Q (2006) Preparation and properties of octenyl succinic anhydride modified early Indica rice starch. *Starch Stärke* 58:109–117
- Tattiyakul J, Naksriarporn T, Pradipasena P, Miyawaki O (2006) Effect of moisture on hydrothermal modification of yam *Dioscorea hispida* Dennst starch. *Starch Stärke* 58:170–176
- Tattiyakul J, Pradipasena P, Asavasaksakul S (2007) Taro colocasia *esculenta* (L.) Schott amylopectin structure and its effect on starch functional properties. *Starch Stärke* 59:342–347
- Violeta BS, Lawrence R, Mal S, Harold C (1998) Genetic variation in physical properties of flour from selected Asian yams (*Dioscorea* spp.). *Trop Agric* 75:212–216
- Waly A, Abdel-Mondy FA, Hebeish A (1998) Chemical modification of starch- poly (vinyl acetate) materials. *Poly Polym Comp* 61:161–170
- Whistler RL, Daniel JR (1995) Carbohydrates. In: Fennema OR (ed) *Food chemistry*. Marcel Decker, New York, pp 69–137
- Wurzburg OB (1978) Starch, modified starch and dextrin, proceeding of seminar in products of the corn refining industry. Corn Refiners Association, Inc., Washington, DC, pp 23–32