

Rapid Synthesis of Acetylated Potato Starch by Microwave Heating with Iodine as Catalyst

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Potato starches were acetylated with acetic anhydride under different reaction times with the help of microwave heating and iodine as catalyst. Starch acetates with increasing degrees of substitution were obtained using microwave assistance when the reaction time varied from 3 to 25 min. The presence of absorption band in FT-IR at around 1750 cm^{-1} confirmed the carbonyl group attachment. SEM revealed exo-corrosion and fusion appeared after acetylation. After acetylation, the complex viscosity of starch reduced and G' , G'' of native potato starch and acetylated potato starch also showed different trends. The XRD pattern of acetylated potato starch was similar to the native potato starch may due to the low degree of substitution. Starch acetate with degree of substitution 0.01-0.2 is approved by the FDA for food applications, we assumed that the reaction time should be less than 10 min when preparing food grade acetylated potato starch by microwave heating.

Keywords: Acetylation, Potato starch, Microwave, Acetylated potato starch, Viscoelastic properties.

INTRODUCTION

For almost 200 years, starch has been considered an attractive material with its comparatively low price, availability and unique properties. It has been widely used in food and other industrial applications as thickener, colloidal stabilizer, gelling agent, bulking agent and water-holding agent¹. However, its hydrophilic and poor mechanical properties have seriously limited its application. To overcome these shortcomings, it is necessary to modify the native starch to improve its properties.

Acetylation is an effective way to modify the starch. It could significantly improve the physicochemical properties of the starch, even with a low degree of substitution². The acetylation is the process which hydrophilic hydroxyl groups are substituted by hydrophobic acetyl groups. It makes starch more hydrophobic and can prevent the formation of hydrogen bonding between hydroxyl groups and water molecules. Acetylation is primarily used to reduce syneresis and texture changes and improve the clarity of starch paste. Starch acetate with degree of substitution 0.01-0.2 is approved by the FDA for food use to improve binding, thickening, stability and texturizing³, food-grade acetylated starches are usually used in refrigerated and frozen foods and instant fried foods such as instant fried noodles⁴. Non-food applications of acetylated

starches include encapsulating agents⁵ and controlled drug release⁶.

Starch acetates are prepared commercially by reaction of starch granules with acetic anhydride for several hours with alkali as the catalyst which usually is sodium hydroxide, potassium carbonate⁷. With the development of microwave technology and application in the organic reactions⁸, microwave has been used in the acetylation of starch with its properties of fast and uniform heating. Microwave as a fast synthesis technique not only can reduce the reaction time, but also improve the yield by suppresses the side reactions⁹. In the current study, some papers have described to prepare starch acetates in the assistance of the microwave¹⁰⁻¹². Starch acetates were prepared at $180\text{ }^{\circ}\text{C}$ without added catalyst but under pressure which increase the risks of the experiment. Some researchers used iodine as catalyst during synthesis of acetylated starches¹³⁻¹⁵. Iodine as a Lewis acid catalyst and iodine combination works only in the absence of a solvent¹⁶. Furthermore, iodine was reported as a good microwave absorber.

In this work, the potato starch was chosen to prepare starch acetates because potato is one of the staples of the human diet throughout the world, potato in general are an excellent source of starch which contributes to the textural properties. Acetylated potato starches were prepared with the assistance of

microwave and iodine as catalyst. The effect of reaction time on starch acetate substitution and some characteristics (*e.g.*, morphological, rheological properties) of the modified starch were also investigated.

EXPERIMENTAL

Starch was provided by Gansu shujie potato Starch Co. Ltd. Starch was dried at 50 °C for 24 h before used. Acetic anhydride, Acetic acid (Tianjin Pharmaceutical Chemical Co. Ltd), Iodine (Sichuan West long chemical Co. Ltd).

Synthesis of starch acetate: The MAS-II microwave oven (2450 MHz, Sineo Microwave Chemistry Technology Company, Shanghai, China) was used. A sample of 9.36 g potato starch was put into a 50 mL three-neck round-bottom Teflon vessel. Potato starches blend with 10.81 g acetic acid and 18.38 g acetic anhydride. A magnetic stir bar was added, the mixture stirred for 6 min then a fiber optic was placed into the vessel to monitor the temperature and a reflux condenser was added. The mixture was heated to 100 °C under a microwave of 300 w for 2 min, then 0.3 mmol I₂ (0.0380 g)¹³ was added. The acetylation reaction was performed at 100 °C in five different times: 3 min, 5 min, 10 min, 15 min and 25 min for sample 1, 2, 3, 4 and 5, respectively. At the end of the reaction time, the vessel was removed from the microwave oven and immediately cooled to temperature. A saturated solution of sodium thiosulfate was added to the mixture. Then the mixture was precipitated in ethanol then filtered and washed with ethanol and distilled water. The starch acetates were dried for 12 h at 50 °C.

Acetyl content: The percentage of acetyl content and degree of substitution (DS) were determined by the known method¹⁷. Grounded sample (1 g) was added to 50 mL of 75 % ethanol solution (v/v). The slurry was kept in the water bath for 0.5 h. After the slurry was cooled to room temperature, 30 mL of 0.5 M (0.5 N) KOH were added and solution was stirred for 72 h. The excess alkali was back-titrated with 0.5 M HCl using phenolphthalein as an indicator.

Acetyl content (% A) was calculated according to the following equation:

$$\text{Acetyl group (\%)} = \frac{(\text{value for blank} - \text{value for}) (\text{mL})}{\text{sample weight (g)}} \times \text{normality of HCl} \times 0.043 \times 100$$

Acetyl content was used to calculate the degree of substitution, according to following equation:

$$\text{Degree of substitution (DS)} = \frac{162 \times \text{acetyl (\%)}}{4300 - (42 \times \% \text{ acetyl})}$$

Fourier transforms infrared spectra: IR spectra of the native starch and acetylated starch were measured using an IR spectrometer (FT-IR, Tensor 27 and Bruker Inc.). The dry sample was blended with KBr in a ratio of starch/KBr 1:50. Each spectrum was obtained at a wave number range of 4000-500 cm⁻¹.

Scanning electron microscopy: Granule morphology of the native potato starch and chemically modified potato starch were studied by scanning electron microscopy (SEM) (JSM-5601LV, JEOL, Japan). The dried samples were fixed to a conductive tape of copper of double glue and sputtered with

gold in order to make the sample conductive. Magnifications of 500×, 2000× were used.

Steady shear properties: Steady shear properties were measured by RS 6000, Haake (Germany) at 25 °C. In the experiment, 4 % (w/w) water suspensions of starches were prepared by heating the starch dispersions at boiling water bath for 10 min. The pastes were shared from 0.1 to 600 s⁻¹.

Dynamic oscillatory measurements: Dynamic properties of starches were performed using the Physica MCR 301 rheometer (Germany). All the experiments were conducted on 4 % starch pastes: 0.4 g starch were mixed with 9.6 g deionized water for 5 min at room temperature, then placed in a water bath at 100 °C. The storage modulus (G') was determined using parallel plates of 25 mm diameter at 25 °C. The angular frequency (ω, rad/s) were range from 0.1 to 100 rad/s.

X-ray diffraction analysis: Crystallographic patterns of native potato starch and acetylated potato starch were determined using an X-ray diffractometer (XRD; RigakuD/max 2400/PC, Japan) under the following conditions¹⁸: Graphite monochromatic copper radiation (CuKα, 1 ¼ 0.15418 nm); sodium iodide scintillation detector; scintillation counter; an accelerating voltage of 40 kV; an emission current of 100 mA; and a 2θ range of 5-30° for all powder samples at a scan rate of 10 °/min.

RESULTS AND DISCUSSION

Influence of reaction time on degree of substitution:

The effect of reaction time on the acetylation of potato starch under microwave heating was investigated. The results are shown in Table-1. An increase of the degree of substitution was noticed with the increasing of the reaction time. Under the similar condition, we could learn that potato starches are harder to acetylation compared to other starches^{13,14,19}. In this case, this study was to explore the time to acetylation. We could observe that after 15 min, the degree of substitution has a sharp rise. The surface of starch granule has fusion with the increase of reaction time. For that, it has more reaction sites and reaction was accelerated. The maximum permitted degree of substitution for acetylated starches are 0.2 [US FDA (food and drug administration), 1980], by means of this sense, we assumed that the reaction time should be less than 10 min when preparing food grade acetylated potato starch by microwave heating.

TABLE I
ACETYL PERCENTAGE AND DEGREE SUBSTITUTION OF
ACETYLATED POTATO STARCH UNDER
DIFFERENT REACTION TIME

Reaction time (min)	Acetyl percentage	Degree substitution
3	4.07 ± 0.003	0.16 ± 0.0001
5	4.23 ± 0.01	0.17 ± 0.0005
10	5.03 ± 0.005	0.20 ± 0.0002
15	9.69 ± 0.009	0.40 ± 0.0004
25	17.42 ± 0.02	0.79 ± 0.0011

Results reported are means of triplicate samples ± SD

Fourier transform-infrared (FTIR): The FT-IR spectra were recorded to detect that whether acetyl group has incorporation into the starch molecule. The spectra of native and acetylated potato starch are, respectively shown in Fig. 1.

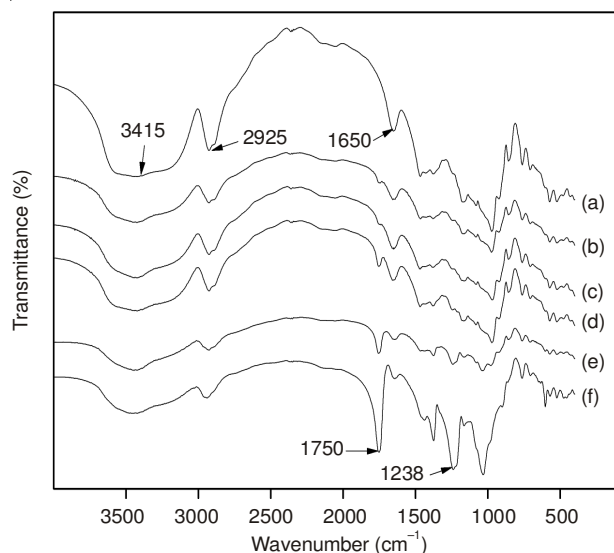


Fig. 1. FTIR spectrum of native potato starch (a) and acetylated potato starch: (b) 3 min, (c) 5 min, (d) 10 min, (e) 15 min, (f) 25 min

The native potato starch shows peak at 3415 cm^{-1} , which were attributed to the vibration of the associated hydroxyl group (O-H)²⁰. The peak at 2925 cm^{-1} was resulting from C-H stretch vibration and absorption bands at around 1650 cm^{-1} were assigned to the tightly bound water (H_2O)²¹. There are two peaks at 1079 and 1170 cm^{-1} which were signed to the C-O bond stretching vibration²². Due to the entire anhydroglucose ring stretching vibrations, peaks at 572 , 761 , 856 , 923 and 971 cm^{-1} were recorded.

After acetylation by acetic anhydride, new absorption bands at around 1750 , 1240 and 1373 cm^{-1} were recorded which were attributed to the formation of an ester carbonyl group (C=O), to the CH_3 symmetry deformation vibration, to the carbonyl C-O stretch vibration, respectively. That confirms the incorporation of the acetyl group.

Morphological properties: SEM has played an important role in increasing understanding of granule structure of modified starches. It has been used to detect structural changes caused by chemical modifications^{23,24}.

For potato starch, the average granule size ranges from 1 to $20\text{ }\mu\text{m}$ for small and 20 to $110\text{ }\mu\text{m}$ for large granules. As Fig. 2 shows that the small potato starch granules are spherical or oval in shape, whereas the large ones are generally ellipsoidal to cuboidal or irregular in shape. Surface of potato starch granules are smooth and shown the small protuberances and fragmentation as reported by Singh *et al.*²⁵. Native potato starch granules do not have channels, so the reagent diffuses supposing from the exterior to the interior. This suggests that the surface of potato starch granules may be the first to be modified, as our study observed. After acetylation, the significant changes have happened in the surface of potato starch granules but still possess well-defined edges. Fig. 2 showed that acetylation result in granule fusion mainly occurred on small-size granules of potato starch. The fusion and fold of starch granules after acetylation could be attributed to the introduction of acetyl groups on the starch molecules, which resulted in the increase in hydrogen bonding²⁶. Aggregation and cluster formation of granules was also observed²⁷.

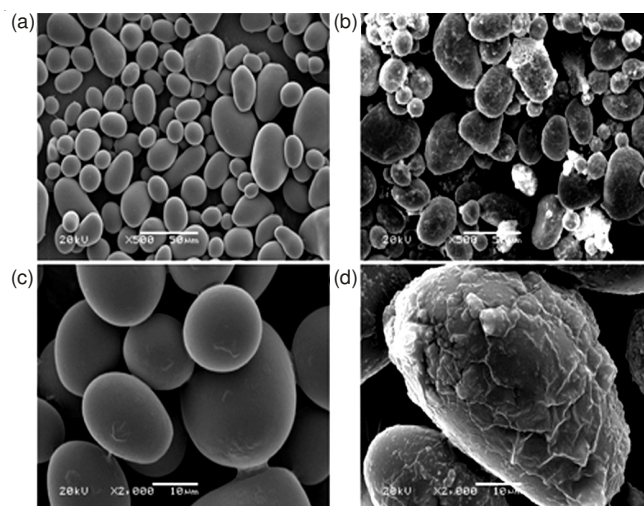


Fig. 2. SEM of native potato starch (a) 500 \times , (c) 3000 \times and acetylated potato starch (b) 500 \times , (d) 3000 \times

Steady flow properties: The flow behaviours of the native and modified starches at $25\text{ }^\circ\text{C}$ were investigated. Both the native potato starch and acetylated potato starch are found to be non-Newtonian, shear-thinning (pseudoplastic) fluids (Fig. 3). The pastes were sheared from 0.1 to 600 s^{-1} . The relationship between shear rate and steady shear stress could be analyzed by using a power law model to describe the flow behaviour of the pastes, as shown in the following: $\sigma = K\gamma^n$. σ is the shear stress (Pa), γ is the shear rate (s^{-1}). K is the consistency coefficient (Pa s^n), n is the flow behaviour index (dimensionless), R^2 coefficients of determination are shown in Table-2. The R^2 are range from 0.979 to 0.999 which means the power law is appropriate to describe the behaviour of the starch pastes. The n reflect the difference between the Newtonian flow and samples. The pastes are Newtonian flow when $n = 1$ and the lower n values means a higher degree of pseudoplastic properties of the fluid²⁸. Therefore, all the pastes are pseudoplastic behaviour because of $n < 1$. With the increase of degree of substitution, lower K value for starch pastes was observed. This phenomenon was due to the weaker of the starch molecular pastes with the reduction of hydroxyl reduction.

Dynamic properties of native and modified starches: Rheological properties of starch pastes determine their possible application such as thickeners or as gelling agents. The most common rheological characteristic of starch paste is viscosity, which changes in broad range upon applied shearing. Complex viscosity decreases with increase in angular frequency. Both native potato starch and degree of substitution 0.79 acetylated starch showed a decrease in viscosity with angular frequency (Fig. 4). These results showed that potato starch behaves as

TABLE-2
POWER LAW PARAMETERS FOR 4% STARCH PASTES

Sample ^a	k	n	R ²
NPS	23.508	0.52499	0.9884
1	16.228	0.60782	0.9937
2	9.505	0.50758	0.9992
3	9.995	0.5569	0.9941
4	7.658	0.54593	0.9809
5	4.011	0.46972	0.9796

^aSamples 1, 2, 3, 4 and 5 are acetylated potato starch with degree of substitution of 0.16 , 0.17 , 0.20 , 0.40 and 0.79 , respectively

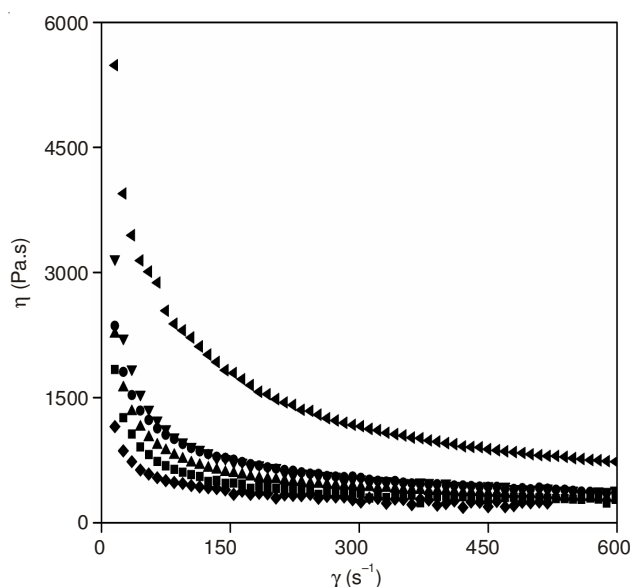


Fig. 3. Shear rate dependence of the steady shear viscosity of native potato starch (\blacktriangle) and acetylated potato starch with different degree of substitution: (\blacktriangledown) 0.16; (\bullet) 0.17; (\blacktriangle) 0.20; (\blacksquare) 0.40; (\blacklozenge) 0.79

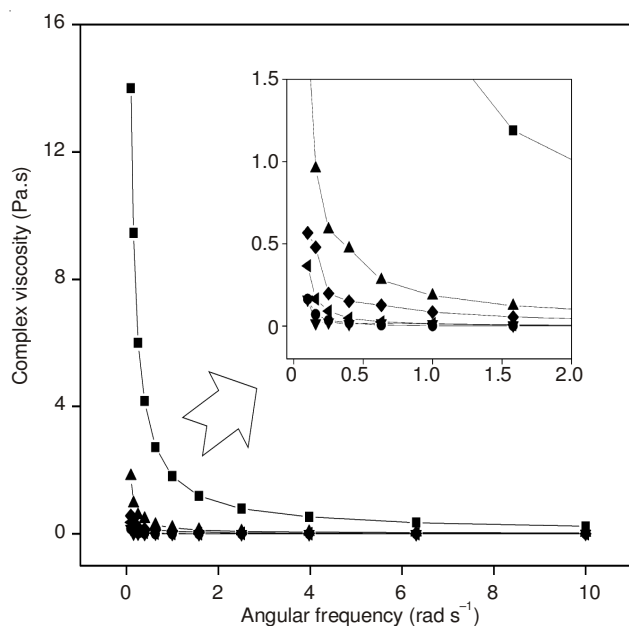


Fig. 4. Complex viscosity of native potato starch (\blacksquare) and acetylated potato starch with different degree of substitution: (\bullet) 0.16; (\blacktriangle) 0.17; (\blacktriangledown) 0.20; (\blacktriangleleft) 0.40; (\blacktriangleright) 0.79

a non-Newtonian fluid with pseudoplastic nature, as in the case of other starches. Modification caused a decrease in viscosity of pastes. The viscosity of the acetylated paste was lower than that of the native potato starch pastes throughout the angular frequency (0.1–10 rad/s). Moreover, all samples showed shear-thinning flow behaviour in this studied range. The higher the shear rate, the lower the apparent viscosity. Degree of substitution in the range of samples do not have obvious effects on complex viscosity.

The storage dynamic modulus (G') is a measure of the energy stored in the material and recovered from it per cycle while the loss modulus (G'') is a measure of the energy dissipated or lost per cycle of sinusoidal deformation. As Fig. 5

shows native potato starch G' increases with frequency to a certain limit and then it decreases. G'' presents the same trend. The G' in 0.79 degree of substitution of acetylated starch demonstrate the same trend. However, the G'' has a sharp decrease after about 80 rad/s. The storage modulus (G') and loss modulus (G'') both are dependent on frequency and a crossover between these two modulus was observed throughout the tested frequency range, demonstrating that pastes are typical weak gel structure as classified rheologically by Clark and Ross-Murphy²⁹. Moreover, the dynamic mechanical loss tangent ($\tan \delta = G''/G'$) values for two gels tested increases with frequency to 1, indicating predominantly elastic or solid-like behaviour.

The dependence of G' and G'' to frequency is important to the characterization of the structure. A material which is frequency-independent over a large time scale range is solidlike; a true gel system is such a material. In contrast, frequency dependence suggests a material structure with molecular entanglements that behaves more like a solid at higher frequencies and more like a liquid at lower frequencies.

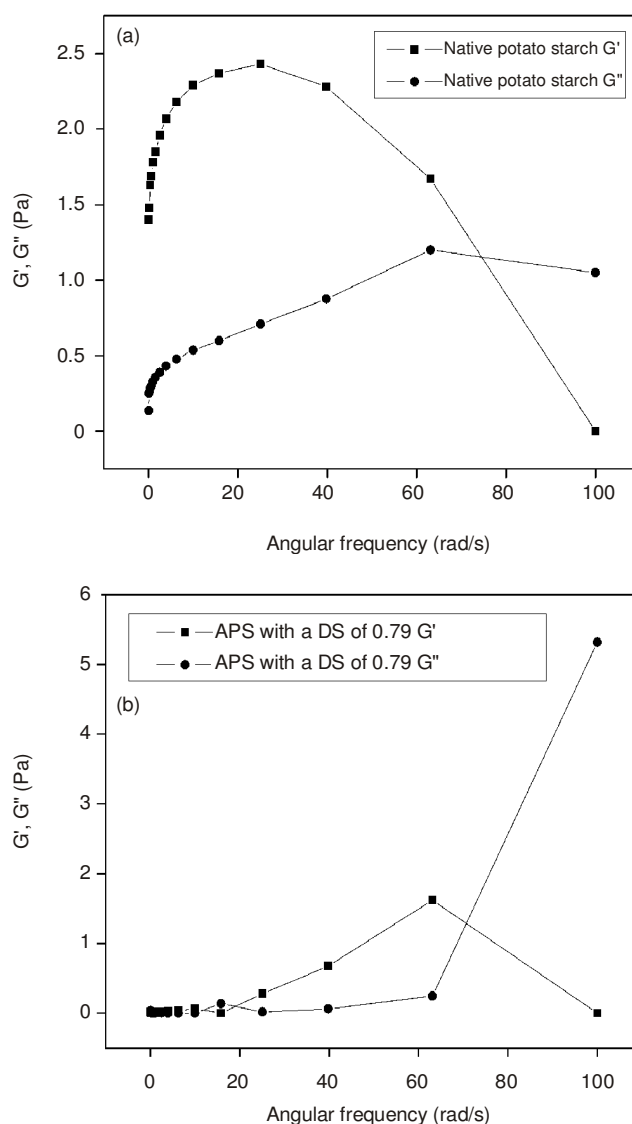


Fig. 5. Examples of mechanical spectra of native potato starch (NPS) (a) and acetylated potato starch (APS) with a degree of substitution of 0.79 (b)

Wide angle X-ray diffractometry: Starch granules are partially being crystalline which have been given distinct X-ray diffraction patterns by Sarko and Wu³⁰. Both native and modified potato starch granules are shows peaks at around 5, 17, 22 ° (2 θ) which belong to the B style however has a little difference described by Kim and Lee³¹ (Fig. 6). The XRD of both starches are similar. When compared with the native potato starch, there was no obvious difference between them which means crystal area of the starch was not greatly affected by acetylation. This phenomenon may be due to the low degree of substitution of the acetylated potato starch and can be confirmed by the SEM photos (Fig. 2), the acetylation occurred only on the surface of the potato starch granules.

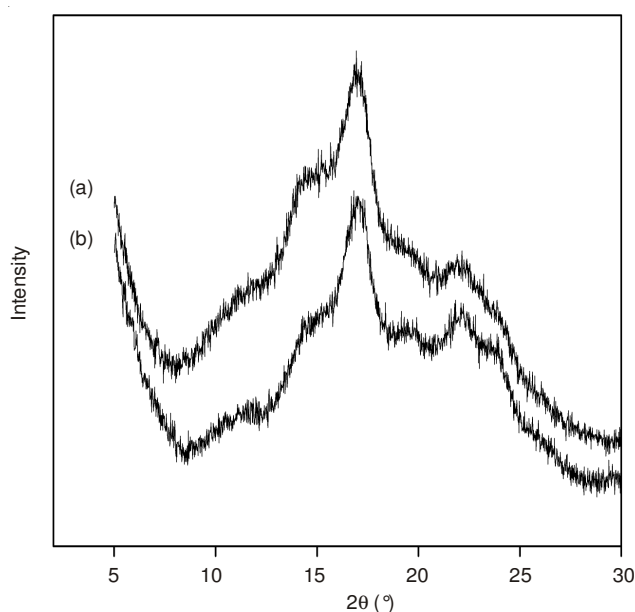


Fig. 6. Wide angle XRD pattern of native potato starch (a) and acetylated potato starch with a degree of substitution of 0.79 (b)

Concluding

A reaction time of 10 min at 100 °C in a microwave reactor using 0.41% I₂ (based on starch) was sufficient to obtain acetylated potato starch with a degree of substitution of 0.20. The increases in the reaction time contribute to the augmentation of the degree of substitution. The FTIR analysis showed higher band at above 1750 cm⁻¹, when the reaction time increased, pattern that was corroborated with the degree substitution. The SEM showed the obvious change in morphology but still retain well-defined edges. The XRD pattern of acetylated potato starch with low degree of substitution was similar to the native potato starch. After acetylation, the complex viscosity of starch reduced and G', G'' of native potato starch and acetylated potato starch also showed different trends. It is possible to use microwave to produce food grade acetylated potato starch and the reaction time should be less than 10 min.

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