Modification of Sago Starch for Edible Coating

D Sondari
Research Center for Biomatereials, Indonesian Institute of Sciences, Jl Raya Bogor km 46 Cibinong, Bogor, West Java 16911, Indonesia

E-mail: sondaridewi@gmail.com

Abstract. The edible coating film was prepared using native sago starch and modified sago starch with acetylation. The effects of acetic anhydride (7.5%, 15%, 30%, and 45% w/w) on swelling properties of wheat starch granules, amylose content, lipids content, protein content, a degree of substitution, percent acetylation, and gelatinization temperature were investigated. Modified sago starch and native starch were characterized by Fourier Transform Infrared (FTIR), X-ray Diffraction (XRD), Differential Scanning Calorimetry (DSC), Thermogravimetric Analysis (TGA) and Scanning Electron Microscopy (SEM). As a result, modified sago starch containing 45% w/w acetic anhydride have a degree of substitution 0.12 with acetylation 45%. All modified starch granules exhibited higher amylose content compared to native starch. Higher amylose contents inhibit the extensive granule swelling power, so swelling power decreased while increased amylose content. Edible coating film used were starch concentration 5.0% w/w, and glycerol content 1.0% v/v. Edible coating film made from native and modified sago starch have been studied to hydrophobicity, Fourier transforms infrared (FTIR), water vapor absorption, and scanning electron microscopy (SEM).

1. Introduction
Sago (Metroxylon sagu Rottb) variety of crops is one of the promising raw materials for the production of the biodegradable material. Most or around 50% of the world's sago plants are in Indonesia [1]. Sago starch is commonly used as a functional ingredient in food industries such as a thickener, stabilizer, and gelling agent. However, the utilization of sago starch is not optimal because if the sago starch cooked, the pasta is hard, not clear and very sticky. This is a constraint in the application as raw materials for food and non-food industries [2-4]. To increase starch added value is to make modifications starch to obtain properties suitable for a particular application [5, 6]. Modification of starch is the treatment given to starch in order to obtain better properties or to change several specific properties. Edible films or coatings could be used as an alternative way of conservation, because of their ability to reduce respiration, reduce moisture, solute migration, and transpiration rate, to maintain firmness and generally delay senescence [7]. Sago starch has great potential to be explored in the development of edible coatings because of the abundance of materials, easy to obtain, low price, edible, and easy to recycle (renewable) and its ease to be physically modified. In addition, the high amylose composition of sago starch that is 27% has potential as a strong film-forming material [8]. The objective in this study was a modification of sago starch using acetylation and characterization of an edible coating.
2. Experimental section

2.1. Sample Preparation
Acetylation was performed according to previous researcher [9]. 200 g of starch (native) were dispersed in 460 g of distilled. Then, acetic anhydride was added drop-wise to the stirred starch suspension at pH kept between 8 and 9 by adding 2% NaOH solution. When the whole amount of acetic anhydride was added, the suspension was further stirred for 15 min. After the reaction, the modified starch was washed with distilled water, dried at a temperature of 20°C for 48 h. Ten percent (w/v) solutions sago starch were prepared of stirring at 60°C. Glycerol was added as a plasticizer at 0.5-1.25% (w/w of sago). The solutions were then allowed to dry overnight at 45°C. They were then conditioned at 51.4% RH at room temperature (30 ± 2°C) for 48 h prior to any analysis [10].

2.2. Characterization
Amylose content was analyzed by the spectroscopic method, the solution was measured its absorbance by a UV-Vis spectrophotometer at a wavelength of 625 nm and amylose levels (in percent) was determined using the standard curve equation of amylose solution. Swelling power and solubility of starch was determined by previously developed method [11]. Soxhletation method was used to evaluated fat content according to SNI 01-2891-1992. Determination of degree of substitution using previously developed method [12]. The functional groups of native and modified sago starch were analyzed using FTIR at a wave number of 4000-400 cm⁻¹. The morphological analysis of the native and modified sago starch was investigated using a scanning electron microscope (SEM) HITACHI SU3500. X-ray diffraction pattern of native and modified sago starch was obtained by using x-ray diffractometer (PANalytical, X’pert PRO Multi-purpose X-ray diffractometer. Film moisture content (MC). MC of the film samples was determined gravimetrically by measuring water removed from the initial mass. The film samples were dried at 110°C for 24 h to attain a constant weight [13]. The contact angles of prepared films were assessed by the recommended procedure by previous researcher [14].

3. Results
The effect of addition of acetic anhydride, at different levels, on moisture content (%), ash content (%), protein content (%), lipid content (%), swelling power (%), solubility (%), amylose content (%), acetyl (%) and degree of substitution (DS) of native and modification of sago starch are shown in Table 1.

| Proximate test                     | Native sago | Acetylation modification of sago starch |
|------------------------------------|-------------|---------------------------------------|
|                                    | 7.5%        | 15.0%                                 | 30.0% | 45.0% |
| Moisture content (%)               | 11.46       | 12.18                                 | 12.83  | 13.15  | 13.86  |
| Ash content (%)                    | 1.04        | 0.72                                  | 0.64   | 0.55   | 0.51   |
| Protein content (%)                | 0.016       | 0.005                                 | 0.006  | 0.011  | 0.016  |
| Lipid content (%)                  | 1.16        | 0.68                                  | 0.52   | 0.09   | 0.20   |
| Swelling power (%)                 | -           | 13.23                                 | 13.31  | 13.62  | 13.56  |
| Solubility (%)                     | -           | 1.03                                  | 1.18   | 1.96   | 1.85   |
| Amylose content (%)                | 25.85       | 25.59                                 | 28.35  | 29.41  | 29.79  |
| Acetyl group (%)                   | -           | 7.24                                  | 7.72   | 8.06   | 7.96   |
| Degree of substitution (%)         | -           | 0.09                                  | 0.11   | 0.11   | 0.12   |

The structure morphology native sago starch and sago starch were modified can be seen in Figure 1. Showed that the granule structure of native sago starch is round and oval. The modification acetylation of sago starch brought granule fusion in both starches. The fusion of starch granules is due to increased hydrogen bonds between starch molecules due to the substitution of hydrophilic compounds
into starch molecules, so that starch molecules combine to produce a fusion of starch granules [15].

[Acetylation modification does not cause damage to the starch granules, but the presence of cracks, scratches and partly occurring granular oval changes result in changes in their specific surface areas.

Figure 1. Scanning electron micrographs (SEM) of native (a) and acetylated sago starch (b)

The FTIR analyses were used to study the chemical structural change between native sago and modified sago starch. The chemical structure can be investigated from the change of functional group by means of the absorption peak characteristic.

Figure 2. Spectra FTIR of native sago (a) and acetylated sago starch (b)

Native sago starch is composed of amylose fraction (≈30%) and amylopectin fraction (≈70%). Amylose starch is linear molecule consisting of α-1,4- glucopyranose for amorphous region and amylopectin which is α-1 branch fraction, 4 and α-1,6-glucopyranose which have crystalline region. The x-ray pattern for native sago and modification with acetyl in Figure 3.

Figure 3. X-Ray patterns of native sago (a) and modified sago starch (b)
The determination of the thermal properties from native sago starch and acetylation was performed using DSC. The analysis results using DSC suggest gelatinization temperature difference where sago acetylation starch has lower gelatinization temperature (Table 2).

| Sample          | C₆H₁₀O₅ (ml) | Gelatinization Temperature | Gelatinization enthalpy (ΔHg) (J/g) |
|-----------------|--------------|----------------------------|-----------------------------------|
| Native sago     | 0            | 32, 102, 120               | 11.6                              |
| Acetylated sago | 45           | 40, 90, 120                | 5.2                               |

The effect of acetylation of starch on the thermal stability was studied by thermogravimetric analysis (TGA). TGA curves of starch and acetylation starch are shown in Fig. 4. Native sago starch (Fig. 4a) was degraded at 300 °C and ends at 450 °C with a loss percent weight of 95%. Further heating to a temperature of 550 °C produces carbonization and ash formation [16].

![Figure 4. Thermal stability analysis of native (a) and modified cassava starch (b)](image)

The edible film of acetylated sago starch showed higher moisture content (%) than acetylated sago starch (Table 3).

| Formula                      | Glycerol (%) | Mc (%) | Contact Angle (°) |
|------------------------------|--------------|--------|-------------------|
| Native sago                  | 1            | 0.16   | 27.43             |
| Modified of sago (45% acetic| 1            | 0.46   | 60.41             |
| anhydride)                   | 0.5          | 0.23   | 68.75             |
|                              | 0.75         | 0.29   | 63.29             |
|                              | 1.25         | 0.51   | 50.24             |

4. Discussion
Native sago starch showed higher ash content than a modified sago starch, but moisture content of sago decreased after acetylation reaction at different levels. The moisture content and ash content of native sago starch and modified sago are in accordance with SNI requirements is for water content...
<14.5 and ash content <7. According to Winarno (1989), the level of food with a moisture content of less than 15% is classified as dry food [17]. Fellow (2000) explained the influence factors of the drying process of foodstuffs are the initial moisture content of the material, the humidity of the environment, and the heat transfer [18]. Protein and lipid content decreased with increasing levels of acetyl anhydride as agent acetylation.

The acetylation of modification sago starch increased the swelling power and solubility. This may be due to the substitution of hydrophilic compounds from the acetyl group in the hydroxyl group of sago starch [19]. Swelling power and solubility of oxidation starch increased to 30.0% acetic anhydride concentration. The correlation between solubility and lipid content, high lipid content in sago starch causes lower solubility as report previous researcher [20]. The various factors influenced to swelling power, granule disintegration, and release of amylose depends on the type of starch, starch concentration, temperature, solute, and agitation during heating [21]. Acetylated sago starches had slightly higher amylose contents than native starches, the amylose content increased from 25.85% to 29.79% in sago starch during acetylation with the highest concentration of acetic anhydride. The similar research on the amylose content of starch acetylation which has been previously observed [19].

The presence of acetyl groups in sago starch affects the function of amylose and amylopectin fractions in iodine absorption during amylose testing [19, 22]. The effect of modification using acetic anhydride on acetyl (%) and degree of substitution (DS) of sago starch showed that the increasing of both acetyl and degree of substitution. The degree of substitution (%) increased with increase in the level of acetic anhydride, this research was reported other researcher [23]. The level of acetic anhydride up to 45.0% with a degree of substitution at 0.12. Similar trends were observed for acetyl (%), acetyl increased with increase in the level of acetic anhydride up to 45.0% (8.06), but the level of acetic anhydride up to 45.0%, the acetyl content decreased (7.96). Acetate starch with a low degree of substitution (<0.1) commonly used in the food industry because it provides structural consistency and stability and starch with a degree of substitution (> 1.0) used as a substitute for thermoplastic cellulose acetate [24].

In Figure 2, shows that modified starch and native starch have a similar functional group structure. The same functional group because the main constituent components of starch are amylose and amyllopectin [25]. The intensity of the hydroxyl groups absorption band at range 3000 - 3300 cm\(^{-1}\), and 2,950 cm\(^{-1}\) correspond to OH and CH stretchings, while the peaks at 1,647 cm\(^{-1}\) and 1,427 cm\(^{-1}\) correspond to (OH) and (CH) bendings was observed of the native sago starch. Compared to native starch, starch acetates presence sorption band at 1,750 cm\(^{-1}\) that was attributed to the stretching of the ester carbonyl C=O and indicated the acetylation of starch [26].

Figure 3 shows the C-type Crystallin structure consisting of about 65% of type A and 35% of type B of the crystalline form [8, 27]. According to the Figure 3. Can be known that the intensity of modified sago starch decrease compare to native sago. This result indicated that acetylation reduced the crystallinity of sago starch. The crystalline structure is affected by the intra- and intermolecular hydrogen bonds. In the acetylation process, the hydroxyl groups were replaced partially by the acetyl group. Therefore, acetylation reduce the formation of intermolecular hydrogen bonds and decrease the crystalline structure [24, 28].

The molecular structure of amyllopectin, and granule structure can be a control for starch gelatinization (Table 2). Acetylation of starch leads to the opening of double helices and fusion of lamella starch crystals from faster amyllopectin which results in decreasing gelatinization temperature [29]. The acetylation reaction with anhydrous acetate results in a decrease in \(\Delta H\) from the acetylated sago starch. The decrease in \(\Delta H\) represents the loss of the double helix sequence, and the loss of crystallinity in the granules. The native sago starch has a high degree of substitution and can increase the larger reduction in \(\Delta H\). The decrease in \(\Delta H\) from acetylated sago starch is the result of double helices present in the crystal and non-crystalline regions of the broken granules during the acetylation reaction. The \(\Delta H\) value of native sago starch shows a lower percentage of regular or less stable crystal structures [29].

As for acetylated sago starch (Fig. 4b) degrades at the same temperature as native sago starch at 300 \(^{0}\)C but ends at higher temperatures at 500 \(^{0}\)C and with a lower percent weight loss of 93%. The high
thermal stability of acetylation modified starch is due to the presence of ester groups. The TGA results show that acetylation modification on starch can reduce thermal stability to 330 °C. However, thermal stability increases at temperatures over 330 °C. The degree value of substitution (DS) affects the value of thermal stability. An increase in thermal stability with increased DS is associated with a small number of hydroxyl groups remaining after acetylation. Starch decomposition is the result of inter or intramolecular dehydration reactions of starch molecules with water as the main product of decomposition [24]. The condensation reaction becomes slower after the hydroxyl group is replaced by the acetyl group. Therefore, a high DS has a beneficial effect on the thermal stability of the sample. The edible film of acetylated sago starch showed higher moisture content (%) than acetylated sago starch (Table 3). The presence of glycerol in the film formulation causes an increase in the hydrophilic properties of the edible film. While edible film from native sago starch has a lower water content. Chain of starch molecule is disconnected by acetic anhydride so that its water content value decreases. The water content of the film affects the ability to absorb water of a film. The high water content causes the quality of the film to have a shorter lifetime due to bacterial disorders [30]. Edible films made from native sago and acetylation modifications using glycerol with a concentration of 0.5, 0.75, 1.0 and 1.25 % weight, glycerol as plasticizer to impart the thermoplasticity to the starch film. The quality of the edible film can be seen from the surface hydrophilicity of edible film that using contact angles determination. The results of testing the contact angles edible film of native sago and acetylation modifications can be seen in Table 3. An increase in the concentration of glycerol as a plasticizer decreases the contact angle value of a film due to the hydrophilicity of glycerol. Addition glycerol to impart the thermoplasticity to the starch film, but the addition of excess amount caused the drop contact angle, bringing about the deteriorating mechanical properties of the film [31].

5. Conclusion
The optimum conditions of the acetylation reaction were carried out at a concentration acetic anhydride of 45% with degree of substitution 0.12%. The presence of the ester carbonyl C=O indicated the acetylation of starch has been done. Generally, the physicochemical properties of modified sago starch better than native sago starch.

Conflict of interest
No conflict of interest

Acknowledgment
The authors would like to thank the Ministry of Research, Technology and Higher Education for Program Insentif riset Sistem Inovasi Nasional (INSINAS) 2018.

References
[1] Flach M 1997. Sago palm. In Promoting the conservation and use of underutilized and neglected crops, report No.13, 1–61, Rome, Italy:Institute of Plant Genetics and Crop Plant
[2] Mirmoghtadaie L, Kadivar M, Shahedi M 2009 Food. Chem. 116 709–13.
[3] Cock J H 1982 Sci. 218(4574) 755-62.
[4] Miyazaki M R, Hung P V, Maeda T, Morita N 2006 Trends in Food. Sci. Technol. 17 591-99.
[5] Singh S, Gamlath S, Wakeling L 2007 Int. J. Food. Sci. Technol. 42(8) 916-29.
[6] Huang J, Schols H A, Jin Z, Sulmann E Y, Voragen A G J 2007 Carbohydr. Polym. 67 11–20.
[7] Tezotto-Uliana JV, Fargoni GP, Geerdink GM, Kluge RA 2014 Postharvest Biol. Technol. 91 72–77.
[8] Ahmad F B, Williams P A 1998 J. Agricul. Food. Chem. 46(10) 4060–65.
[9] Fortun T, Juszczak L, Gałkowski D, Małgorzat B, Aczkowicz, Łabanowska M, Kurdziel, M 2018 Int. J. Biol. Macromol. 106 57–67
[10] Moreno O, Cardenas J, Atarés L, Chiralt A 2017 Carbohydr. Polym. 178 147–158.
[11] Onyango C, Mowa EA, Mutahi AW, Okoth MW 2013 African. J. Food. Sci. 7(5) 80-86.
[12] Shah N N, Vishwasrao C, Singhal R S, Ananthanarayan L 2016 Food Hydrocol. 55 179-188.
[13] Thakura R, Saberia B, Pristijonoa P, Goldinga J, Stathopoulos C, Scarlett C, Bowyer M, Vuong Q 2016 Int. J. Biol. Macromol. 93 952–960.
[14] Rezaei S M, Razavi S H, Hosseini S M H 2010 J. Biotechnol. 122 161–66.
[15] Singh N, Chawla D, Singh J 2004 Food. Chem. 86 601–08.
[16] Qiao L 2016 Chem. Eng. Trans. 55 235-40.
[17] Winarno F G 1989 Kimia Pangan dan Gizi. PT. Gramedia, Jakarta.
[18] Fellow P J 2000. Food Processing Technology, Principle and Practice. CRC Press. Cambridge, England Research, Gatersleben/International Plant Genetic Resources Institute.
[19] Betancur D, Chel G L, Canizares H E 1997 J. Agricul. Food. Chem. 45 378-382.
[20] Galliard T, Bowler P 1987. Starch properties and potential, In T. Galliard (Ed.), Chichester: Wiley, 57–78.
[21] Biliaderis C G 1991 Water relationships in food In H. Levine, & L. Slade (Eds.) New York: Plenum Press. 251–273.
[22] Whistler R L, Daniel J R 1995 Food. Chem. 69–137. New York: Marcel Decker
[23] Rusman R, Majid R A, Abd Rahman W A W, Low J H 2017 Chem. Eng. Trans. 56 1171-76.
[24] Xu Y, MiladinovV, Hanna M 2004 Cereal. Chem. 81(6) 735–40.
[25] Fleche G 1985. Chemical Modification and degradation of Starch. In G.M.A.V. Beynum and J.A Roels (eds.). Starch Conversion Technology. Marcel Dekker, Inc., New York
[26] Mano J F, Koniarova D, Reis R L 2003 J. Mater. Sci. Mater. Med. 14 127-35.
[27] Zobel H F, Young S N, Rocca L A 1988 Cereal. Chem. 66 443–46.
[28] Colussi R, Pinto V, Halala S, Vanier N, Villanova F, Silva R, Zavareze E, Dias A 2014 Carbohydr. Polym. 163 405–413.
[29] Colussi R, Mello S, Pinto V, Bartz J, Carlos L, Zavareze E, Dias A 2015 Food. Sci. Technol. 62 1076-82.
[30] Malia S, Victo’ria M, Grossmann, Maria A, Garcia, Miriam N, Martino E, Zaritzky 2002 Carbohydr. Polym. 50 379–86.
[31] Bangyekan C, Aht-Ong D, Srikulkit K 2006 Carbohydr. Polym. 63 61–71.