Chapter 20

Acid Modification of Sago Hampas for Industrial Purposes

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Abstract Sago pith and sago hampas mainly consist of starch and fiber. In this research, acid modification of starch and fiber was conducted by high-temperature (autoclaving) and microwave-assisted treatments and slow or mild acid hydrolysis. Autoclaving and microwave-assisted treatments influenced the gelatinization and solubilization of starch granules to produce depolymerized starch and changed the fiber structure to become more amorphous forms. Heating in dilute acid produced high soluble total sugars with high dextrose equivalent, but the process also released hydroxymethylfurfural as undesired by-products. Slow or mild acid hydrolysis attacked the amorphous regions of starch and fiber. It did not change the starch and fiber crystallinity pattern but increased the degree of crystallinity. Acid modification techniques for sago starch and its fiber can be used for producing certain products such as starch sugar, fermentable sugars, and filler for biofoam production.

20.1 Introduction

Sago palm is considered an ideal crop for a starch-based sweetener and feedstock, since in Indonesia there is negligible competition between food and ethanol fuel and land resources are underutilized. Sago starch has been used for a long time, especially in Southeast Asia in the food industry for the production of sago pearl, vermicelli, bread, cake, biscuits, and many other traditional foods (Ahmad and Williams 1999). Sago palm (*Metroxylon* sp.) is a species from which starch-rich flour can be extracted from the pith of the trunk. The pith consists mainly of starch, which has to be separated from cellulosic materials by several stages of processing. Fujii et al. (1985) reported that the composition of dried sago pith was mainly starch (81.51–84.72%) and fiber (3.20–4.20%).

Sago starch is one of the least expensive starches but has low quality. Yatsugi (1985) has identified several problems associated with sago starch and manufacture, including whiteness, viscosity, starch particles, and starch content in starch pro-

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Table 20.1 Characteristics of acid and enzymatic hydrolysis products*

Characteristics**	Sago samples		
	Sago pith flour	Sago starch	Sago hampas
<i>Chemical components</i>			
Ash (%)	1.18	0.12	7.59
Crude lipid (%)	1.35	1.82	2.04
Crude protein (%)	0.49	0.19	0.86
Crude fiber (%)	3.34	0.19	15.34
Starch (%)	86.00	97.83	55.33
<i>Acid hydrolysis product</i>			
Total dry matter (%)	56.13 ^c	53.80 ^b	51.44 ^a
Yield (%)	52.56 ^b	63.04 ^c	32.67 ^a
Degree of polymerization	2.97 ^b	2.88 ^b	2.45 ^a
Dextrose equivalent	33.70 ^a	34.12 ^b	41.58 ^b
<i>Enzymatic hydrolysis product</i>			
Total dry matter (%)	57.74 ^a	60.67 ^b	69.81 ^c
Yield (%)	62.12 ^b	70.63 ^c	33.76 ^a
Degree of polymerization	1.35 ^a	1.40 ^c	1.38 ^b
Dextrose equivalent	72.69 ^b	71.52 ^a	73.88 ^c

*Means within a row related to particular parameter with the same superscript letter are not significantly different as $\alpha = 0.05$ confidence level; **measured as dry weight basis

duced by rural factories in the tropics. The color of the starch, which turns brown in the process of separation, is one of the factors responsible for low quality. When sago palm pith is chopped up and grated into a powder, colored substances are formed and tightly bound to the starch granules.

There are two methods for sago starch extraction from fresh sago pith, wet and dry. The dry process is conducted without water. In this method, fresh sago pith is shredded, oven-dried, ground, and sieved to produce fine starch particles. But as raw material for nonfood industry, the dry process can produce large and coarse particles passing through the 35 mesh sieve (Table 20.1); this is called sago pith flour. While from the wet method, the main product is sago starch, and its residue (sago hampas) is generated from the processing. Sago hampas is the fibrous by-product from crushing and sieving during the wet extraction process. In our research, samples of sago hampas were oven-dried, ground, and sieved to produce fine flour.

Table 20.1 shows that starch is the main component in all sago samples. Sago pith flour, as a part of the dry extraction production, contains a high amount of starch (86%), while, as a by-product from the inefficient wet extraction process, sago hampas contains approximately 55% starch and 15% fiber on a dry weight basis. Composition of this sago hampas was similar to that reported by Lingga et al. (2012) which showed 58% starch, compared to Awg-Adeni et al. (2013) who reported a lower starch content (30–45%) and higher fiber content (30–35%).

Previously we have reported that crude fiber was the second most common component in pith (Sunarti et al. 2011). Hampas also contains high amounts of cellulose (11%), hemicellulose (7.6%), and lignin (12%) (Utami et al. 2014); Abd-Aziz

(2002) reported similar results. With significant amounts of starch and fiber remaining in the hampas, it may be used for hydrolysis to make starch syrup, an alternative cheap substrate for fermentable sugar production (Lingga et al. 2012), a source for biohydrogen production (Jenol et al. 2014), and a composite material for biofoam production (Lai et al. 2013; Utami et al. 2014; Rao and Kumar 2015).

20.2 Acid-Catalyzed Hydrolysis

For industrial use, raw sago pith must be fresh and be processed quickly. With the passage of time, the separation yields less of a white product. Some importing countries are dissatisfied because each lot of starch received is variable in quality and in addition most are of inferior quality. Therefore, industrial application of the sago starch is limited. One way to overcome this inferior quality is to utilize the sago starch for glucose syrup production, which is not dependent on the properties of native starch.

Acid hydrolysis of starch to produce sugars has long been a commercial practice. Sago pith flour and hampas which contain mostly starch and lignocellulosic materials are good sources of glucose syrup and substrate for fermentation, either by enzymatic or acid hydrolysis (Table 20.1). In the acid process, a slurry containing about 30% (w/v) of starch or flour solid is suspended and acidified to a pH of about 2.0 by using diluted hydrochloric acid. The suspended starch or flour is then autoclaved at 121 °C for 1 h and neutralized by a 1 N NaOH solution. Although acid hydrolysis is a rather (but not completely) random process (BeMiller 1967), the hydrolysis produces syrup with a dextrose equivalent (DE) value range of 34–42, a lower value as compared to enzymatic (liquefaction and saccharification) hydrolysis (DE 71–74). Hebeda (1987) predicted that the carbohydrate profiles of the syrup in the 36–43 DE range consisted of 14–19% DP₁ (monosaccharides, dextrose), 11–14% DP₂ (disaccharides, primary maltose), 10–12% DP₃ (trisaccharides, primary maltotriose), and 55–65% DP₄₊ (oligosaccharides, maltotetraose, and higher saccharides). Lorenz and Johnson (1972) reported that at 120 °C, autoclaving treatment completely gelatinized the starch, as expected, but did not completely hydrolyze it.

Sweetness is an important and easily identifiable characteristic of glucose. When compared to sucrose, the relative sweetness value of 30–42 DE acid-converted syrup is between 30–35 and 45–50 (Pancoast and Junk 1980). Furthermore, the starch-derived sweetener also provides a highly fermentable substrate suitable as intermediate substrate for many industrial applications such as for making ethanol, lactic acid, and kojic acid (Shinghal et al. 2008).

Starch is hydrolyzed by acid to produce D-glucose, but other products are formed such as oligosaccharides and products of decomposition and dehydration of D-glucose, such as 5-hydroxymethylfurfural, levulinic acid, formic acid starch-derived products, and colored products. Lorenz and Johnson (1972) reported that in neutral pH hydrolysis, color change was very slight as the temperature of hydrolysis increased from 120 to 135 °C; the greatest color change was from 150 to 180 °C. Several reactions can also cause color development in starch because the

material contains reducing sugars via the nonenzymatic browning reaction between sugars and primary or secondary amines (Maillard reaction). Color development in the absence of nitrogenous compounds with the application of heat or acid is the result of caramelization (BeMiller and Whistler 2009).

20.3 Effect of Microwave-Assisted Hydrolysis in Dilute Acid

Conventional acid hydrolysis of starch is usually conducted under high-pressure and high-temperature treatment for several hours. Excessive heating of starch-derived products will result in partial caramelization and development of undesirable flavors. Pentose as a fiber-derived product will be converted to furfural, while hexose produces hydroxymethylfurfural (HMF), both giving a brown color solution. Furfural is more toxic than HMF. More browning compounds make the solution have less clarity.

The heating process using a microwave represents a different mechanism from that of using a conventional convection or conduction heating process. A microwave uses electromagnetic waves between 300 MHz (wavelength 1 m) and 300 GHz (wavelength 1 mm). Microwave heating is usually called dielectric heating since in the microwave electric field, dipole molecules such as water or other dielectric materials rotate vigorously to orient in the field (Gabriel et al. 1998). Microwave energy can penetrate into materials, so that it directly and uniformly heats the materials. Microwave heating avoids degradation of product strength and surface properties caused by excessive and long conventional heating.

Previously, Matsumoto et al. (2011) and Khan et al. (1979) reported that microwave treatment can be an alternative method for starch hydrolysis in water; Warrand and Janssen (2007) and Yu et al. (1996) stated that starch in acid hydrolysis can produce oligosaccharide. Some researchers also reported the microwave-assisted hydrolysis can be applied to the mixture of starch and fiber from industrial waste such as cassava pulp (Hermiati et al. 2011) and waste from the corn starch industry (Yoshida et al. 2010). A study carried out by Palav and Seetharaman (2006) showed that gelatinization of starch during microwave heating did not produce the same phenomenon as using conduction heating. This can be observed in the loss of birefringent properties and in the swelling of the starch granules. Luo et al. (2006) conducted a study to investigate changes in crystallinity, swelling power and solubility, gelatinization parameters, retrogradation characteristics, and pasting properties of corn starches at 30% moisture after being subjected to microwave heating at 1 W/g microwave energy for 20 min. Microwave heating could convert starch directly to glucose in a relatively short time. Compared with conventional heating, the reaction rate of starch hydrolysis to glucose was accelerated 100 times under microwave irradiation (Kunlan et al. 2001).

Treatment or degradation of starch using microwave heating can use water or dilute acid, such as hydrochloric or sulfuric acid, as media. Kunlan et al. (2001) reported that the addition of inorganic salts containing Cl and SO₄ ions could

Fig. 20.1 Microscopic structures of (a) native sago pith, (b) microwave heating of 8% of sago pith in dilute acid, (c) 8% of sago pith in water, (d) 10% of sago pith in water, (e) fiber residue from microwave treatment, (f) autoclaving treatment after enzymatic saccharification. Magnification 200X (Sunarti et al. 2012)

enhance the hydrolysis of starch. However, the Cl ion gave better results. Yu et al. (1996) stated that 10% starch suspension in 0.5 M HCl could be completely converted to glucose in only 5 min without the formation of colored by-products, but if heated using the conventional method, it was not completely hydrolyzed. Figure 20.1 shows the effects of microwave treatment on the microscopic structure of sago pith, which contained starch and fiber, in water (c) and dilute acid (b) compared to native sago pith (a). Microwave-assisted heating in dilute acid (Fig. 20.1b) directly converted the starch and fiber in sago pith into depolymerized products and gave more simple sugars compared to the autoclaving treatment. Microwave heating of sago pith in water mainly caused gelatinized of the starch, as long as water remained in the slurry. Insufficient water produced partial or incomplete gelatinized starch as described from a higher concentration of sago pith (10%) (Fig. 20.1d) compared to 8% of suspended solids (Fig. 20.1c).

Microwave heating also affects the lignocellulosic materials. The effects of microwave heating on major chemical components of lignocellulosic biomass (lignin, cellulose, and hemicellulose) have been reported by Tsubaki et al. (2009), who stated that the required heating temperature for degradation of either lignin or cellulose corresponded with the glass transition (T_g) of the components. Some disruptions in fiber were detected after microwave treatment in water (Fig. 20.1e) compared to autoclave treatment (Fig. 20.1f).

Table 20.2 Characteristics of microwave-assisted hydrolysis of sago pith in dilute acid

Treatment	Total soluble sugar (g/l)	DE	Clarity ^a At λ_{660nm} (OD)	HMF (g/l)
<i>Autoclaving</i> (121 °C, 15 min)				
HCl 0.2 M	294	45	1.12	0.16
HCl 0.3 M	299	50	1.26	0.24
<i>Microwave</i>				
- HCl 0.2 M				
PL 50%, 1 min	32	40	0.81	0.0069
PL 50%, 2 min	40	39	0.63	0.0072
PL 50%, 3 min	66	45	(0.09)	0.0074
PL 70%, 1 min	201	81	0.78	0.0089
PL 70%, 2 min	261	69	0.37	0.0091
PL 70%, 3 min	282	84	(0.09)	0.0094
- HCl 0.3 M				
PL 50%, 1 min	27	50	1.09	0.0075
PL 50%, 2 min	41	40	0.86	0.0079
PL 50%, 3 min	69	45	(0.07)	0.0082
PL 70%, 1 min	266	57	0.05	0.0095
PL 70%, 2 min	272	71	0.25	0.0097
PL 70%, 3 min	285	90	(0.10)	0.0100

Source: Sunarti et al. 2011

^aData in parenthesis express negative values

There are some distinct characteristics of 10% sago pith slurry in dilute acid when poured into a glass jar with a lid and then heated in a microwave oven with varied power levels (50–70% of maximum output 1000 watts) for 1–3 min heating duration (Table 20.2). Interaction between heating level and its duration influenced the hydrolysis yield (Sunarti et al. 2012). Bej et al. (2008) also stated that acid hydrolysis of starch follows the first-order reaction. Time-dependent conversion was observed with variation of temperature, pH, and initial starch concentration. Similar results have been reported by Khan et al. (1979) who found that liquefying and coloring were the most obvious visible changes during starch hydrolysis. With extended heating times, temperature, and pressure, total acidity increased causing darker hydrolyzates. Starch sugar syrups of high dextrose equivalent can be produced within a short time by using microwave energy.

Heating in dilute acid produced high soluble total sugars with high dextrose equivalent; the process also released HMF as an undesired by-product, and a high amount of salt was produced from neutralization of acid. Long exposure to microwave heating increased the formation of brown coloring as detected in low syrup clarity. Compared to autoclaving treatment, microwave heating treatment produced less furfural compound. Autoclaving for a longer heating time (15 min) influenced the formation of furfural and HMF compounds, as 0.24 g/l compared to the highest HMF content from microwave treatment (0.01 g/l). As substrate for ethanol production, the presence of HMF and furfural negatively influenced productivity. Nigam

(2001) found that a furfural concentration of more than 1.5 g/l inhibits the yeast (*Pichia stipites*) growth and influences ethanol production. Furfural and HMF show different inhibitory effects to yeast growth in bioethanol production (Sunarti et al. 2011). Modig et al. (2002) reported that 1 g/l of furfural can inhibit the microbial enzymes for ethanol production, such as alcohol dehydrogenase, aldehyde dehydrogenase, and pyruvate dehydrogenase, while 2 g/l of HMF shows the same effects.

20.4 Slow Acid Hydrolysis

For many years, slow and mild acid hydrolyses have been applied to starch by industries to improve its solubility. This process involves suspending starch in an aqueous solution of hydrochloric or sulfuric acid at certain temperatures, since in the presence of strong acid and heat, the glycosidic bonds between the monosaccharides in the starch polymer are cleaved (Yiu et al. 2008) and produce glucose and oligosaccharides. Prolonged mild acid hydrolysis can be performed to produce lintnerized starches. Odeku et al. (2009) stated that acid modification has been proven to change the physicochemical properties of starch without destroying its granular structure, yielding starch with increased solubility and gel strength but decreased viscosity. The extent of hydrolysis depends on starch consistency, acidity of the medium, hydrolysis temperature, and duration of hydrolysis. The effects of acid hydrolysis of various starches, such as corn, potato, and rice, have been studied previously (Wang and Wang 2001; Wang et al. 2003; Thirathumthavorn and Charoenrein 2005). Moreover, acid treatment in alcohol, especially for sago starch, has been conducted by Yiu et al. (2008), and their study showed that sago starch can be modified through hydrolysis and alcoholysis to give various limit dextrans with high solubility.

Acid treatment also can be applied to fiber-rich starch such as sago hampas. Ning et al. (1991) reported that acid modification of fiber at high temperature (90 °C) can produce oligosaccharides and changes in its crystallinity. It has been shown that sulfuric or trifluoroacetic acid has been used to hydrolyze the hemicellulose in fiber (Englyst and Cumming 1984). In the present research, sago hampas was modified by the acid-methanol treatment according to the Chung and Lai (2006) methods. Sago hampas flour was acid-treated with a low concentration of hydrochloric acid in methanol (4 ml of HCl concentrated in 1 liter of methanol) solution at room temperature (30 °C) for 0, 60, 120, 180, and 240 h.

Modification of sago hampas by acid in methanol cleaved the amorphous site of fiber and starch. The sago hampas was immersed in acid in methanol solution and produced a dark filtrate color. The dark color of sago pith flour comes from an oxidative reaction of phenolic compounds. This acid condition causes solubilization of phenolic compound and dissolves some reducing sugars and produces 91–94% yield of acid modified sago hampas as described on Table 20.3. This made the acid modified sago hampas's color or performance brighter compared to that before the treatment. Lignins are amorphous and noncarbohydrate compounds containing phe-

Table 20.3 Starch and fiber characteristics of native and acid modified sago hampas*

Treatment of hydrolysis	Total soluble solid in filtrate (ppm)	Yield (%)	Starch (%)	Lignin (%)	Cellulose (%)	Hemicellulose (%)	Crystallinity (%)
Native hampas		100	49.94	4.97	15.10	5.33	
0 h	0.39	93.81	48.07 ^a	4.32 ^a	15.31 ^b	5.87 ^a	57.33
60 h	51.70	93.67	42.94 ^b	4.70 ^a	14.00 ^{ab}	4.98 ^a	64.37
120 h	72.21	91.62	40.02 ^c	4.83 ^a	11.37 ^a	4.58 ^a	62.68
180 h	79.62	91.48	25.72 ^d	5.71 ^a	13.04 ^{ab}	5.04 ^a	52.22
240 h	87.73	91.35	19.38 ^c	3.11 ^a	14.20 ^{ab}	5.91 ^a	58.55

Source: Sunarti et al. 2015

*Means within a column related to particular parameter with the same superscript letter are not significantly different as $\alpha = 0.05$ confidence level

nolic substances which were diluted in acid solution. This condition also causes small changes in fiber composition (lignin, hemicellulose, and cellulose) but has significant effects on changes in molecular weight and composition of starch.

Sathaporn et al. (2005) reported that some branched points in amylopectin were hydrolyzed and remained crystalline parts of starch. Soluble carbohydrate produced from acid hydrolysis can be glucose, maltose or malto-oligosaccharides as products from starch hydrolysis, and soluble hemicellulose or its derived products. It was detected that a small amount of soluble carbohydrate (not more than 90 ppm) (Table 20.3) was produced from acid hydrolysis.

Previously, we reported that the crystalline type of starch and fiber were not markedly changed after acid modification (Sunarti et al. 2016). Higher diffraction intensities were clearly obtained after acid treatment. An increase in starch and fiber crystallinity is attributed to preferential hydrolysis of the amorphous domains by acid (Gerard et al. 2002). Ahmad and Williams (1999) and Pukkahuta and Varavinit (2007) found that sago hampas has a crystallinity type between cereal (A-type) and tuber (B-type). The profiles also expressed the mixture of starch and fiber crystalline fractions. Typical diffraction angle for starch can be detected on 2θ 15.1, 17.2, 17.8, and 23.4°; Lai et al. (2013) also reported diffraction angle of 22.5 ° for fiber.

Changes of molecular structures are also expressed on surface morphology of starch granules and fiber structures. Compared to native sago hampas, internal fissures in starch granules appeared after 180 and 240 h. The fiber also degraded into small parts and became transparent which showed the increment of amorphous fraction of fiber (Sunarti et al. 2016). Some data in Table 20.3 also proved that the destruction of fiber and starch took place after acid modification.

Hampas, sago pith residue, a solid waste of sago wet extraction industry, is a potential source for use as fiber reinforcement on starch-based foam production due to its composition that contains starch and fiber which are necessary for producing biofoam. Utami et al. (2014) also reported that partial acid hydrolysis of sago ham-

pas modified the structural and physico-mechanical properties of the resulting foam. Compared to styrofoam, biofoam produced from acid modified sago hampas has better mechanical properties but is very sensitive to moisture making biofoam ideal for trays of dry food products (Sunarti et al. 2016).

20.5 Conclusion

Sago palm (*Metroxylon sagu*) is a species from which starch-rich flour can be extracted from the pith of the trunk. Sago hampas is a fibrous by-product from the wet extraction process. Although it is inefficient for starch production, sago hampas consists of starch and fiber as major components. Some conclusions can be summarized about acid modified treatment of hampas:

1. Direct hydrolysis of starch and fiber using dilute acid and heat treatment (autoclaving or microwave-assisted treatments) degraded the starch into glucose and oligosaccharide and increased the amorphous region of fiber.
2. Slow or mild acid hydrolysis of starch and fiber attacked the amorphous region. Mild acid hydrolysis did not change the crystallinity pattern but increased the degree of crystallinity.
3. Products revealed from acid hydrolysis of starch and fiber in sago samples (starch, sago pith, and sago hampas) can be applied as sweetener, substrate for fermentation, and raw materials for food and nonfood industries.

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