Research Article

Modification of pineapple starch from stem and rhizome using multiple desired response optimization and its characterization

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ABSTRACT

The aim of this study was to modify pineapple starch from stem and rhizome using multiple desired response optimization. The octenyl succinic anhydride (OSA) modification conditions of pineapple starch were conducted using a fractional factorial central composite face-centered design. The selected independent variables included OSA concentration, pH, temperature, and time, whereas the desired multiple response variables related with emulsifying properties included the degree of substitution (DS), surface tension, and enthalpy of OSA-modified pineapple starch. The experimental and predicted values for the datasets of response variables exhibited good performance. The OSA concentration was found to be the principal factor for the DS, surface tension, and enthalpy model. The optimum conditions were 24% w/w OSA, pH 7.8, reaction temperature = 37.5° C, and reaction time = 2 h with a desirability value of 0.97. Furthermore, OSA-modified pineapple starch was characterized by FTIR, XRD, DSC and SEM comparing with native pineapple starch (NS). The OSA modification of pineapple starch resulted in an increase in viscosity and a decrease in surface tension, gelatinization temperature and enthalpy. For emulsifying property evaluation, the OSA-modified pineapple starch showed higher emulsifying activity index than NS.

1. INTRODUCTION

To reduce the impact of human activity on the environment, research into the development of green materials has increased in the past decade. Starch is one of the most promising materials for future use as it contains no toxic residues and is widely available, naturally abundant inexpensive, and has broad-ranging biocompatibility ¹. It is currently used in various applications as a gelling agent, thickener, suspending agent, diluent, filler, binder, and disintegrant ²⁻⁴. Several starches, including mango kernel ⁵, corn ⁶, *Tacca involucrate* ⁷ and plantain ⁸ have been structurally characterized and investigated for their physicochemical and rheological properties.

Starches found in legumes, rhizomes, herbs and seeds are considered as unconventional starches which may be used as ingredient materials in the same manner as starch from cereals and tuber because of their similar physicochemical properties and functional properties⁹. Modification treatment may be used to improve



Figure 1. Schematic diagram showing OSA modification of pineapple starch.

these properties, develop new process and consequently new products ¹⁰. However, there are limited studies on the use of unconventional starches ^{11,12}.

Pineapple (Ananas comosus L. Merr.) is a native plant that is widely grown in Prachuap Khiri Khan Province, Thailand. Recently, pineapple starch was extracted from stem and rhizome waste, which is normally disposed of via a burning process, thereby polluting the environment¹³. The application of pineapple starch in food and pharmaceuticals has been rarely reported ¹⁴. Since, starches have some limited properties in the native form, modified starches have therefore played a significant role in the food and pharmaceutical industries over the past few decades. Research on octenyl succinic anhydride (OSA) modified starches has been reported ¹⁵. Chemical reaction during OSA modification is shown in Figure 1. When OSA is introduced into the hydroxyl groups of starch glucose unit, the hydrophobicity of the starch is increased after modification. Various starches have been esterified with OSA, including Indica rice starch ¹⁶, sago starch ¹⁷, plantain starch ¹⁸ and potato starch ¹⁹. The functions of OSA-modified starches emulsion stabilizers as and encapsulating agents and their use in the pharmaceutical, food, and biodegradable plastics industries have been demonstrated ¹⁵.

In general, the quality of a product or process that has multiple quality characteristics, with one of them outside of some desired limits, is completely unacceptable. Response surface methodology (RSM) can be employed when a response or a set of responses of interest is influenced by several variables ²⁰. The optimization technique and statistical design provide an economical and efficient means of obtaining important information required for determining the effects of causal variables on response ²¹. The effects of the reaction conditions on DS of OSA-modified starches using RSM have been reported for *Indica* rice starch ¹⁶, corn starch ²² and starch from *Phaseolus lunatus* ²³. However, there was no report on optimization of the process for modification of starch with multiple responses. Moreover, there is a lack of information in the literature about the physicochemical and emulsifying properties of OSA-modified pineapple starch and no research on the optimization for OSA modification of pineapple starch has been conducted yet.

Thus, this study aimed to optimize the reaction conditions for OSA-modified pineapple starch using RSM. The physicochemical properties of OSA-modified pineapple starch were characterized by scanning electron microscopy differential (SEM), scanning calorimetry (DSC), powder X-ray diffraction (XRD), Fourier transform infrared (FTIR) spectroscopy. The OSA concentration, reaction pH of the system, reaction temperature, and reaction time were chosen as the independent variables. Since the enthalpy is the factor indicating the efficiency on polymer swelling and one mechanism of emulsifying agent involved with the decrease in surface tension and related to high DS¹⁵. Optimizing OSA-modification of pineapple starch was conducted to obtain the multiple responses such as DS, surface tension and enthalpy. If the relationship was revealed, it could be trended as effective emulsifying agent. The emulsifying property of native pineapple starch (NS) and OSA-modified pineapple starch was also evaluated using emulsifying activity index (EAI).

2. MATERIALS AND METHODS

2.1. Materials

Pineapple starch from stem and rhizome obtained from Peintong research group (lot no.0117), Sam Roi Yot Districts, Prachuap Khiri Khan Province, Thailand was modified with 2-OSA (Tokyo Chemical Industry (TCI) Co., Ltd.,

		Levels				
Independent variables	Symbols	-1	0	+1		
OSA concentration (%)	X_1	1	15.5	30		
pH	\mathbf{X}_2	7	8	9		
Temperature (°C)	X_3	25	37.5	50		
Time (h)	X_4	2	3.5	5		

Table 1. Independent variables and the corresponding code variable levels used for the fractional factorial CCF design.

Tokyo, Japan). Sodium hydroxide (Merck, Germany) was used as catalyst to adjust pH of the alkaline medium and the solvent used to dilute the OSA for modification of pineapple starch was ethanol having 99.99% purity (Merck, Germany). Other chemicals and reagents used herein were of analytical grade.

2.2. Modification of pineapple starch with OSA

pineapple OSA-modified starch was prepared according to the procedure described by Hui et al ¹⁹. Briefly, aqueous suspension (30% w/w) of pineapple starch was prepared. The 3% w/v NaOH solution was added to starch suspension at the pH of 7-9 (pH meter, Mettler-Toledo, Switzerland) in order to initiate the reaction for starch in an alkaline medium. OSA (1-30%, in proportion to the starch weight; dry basis) was diluted five times with 99.99% v/v ethanol and was added dropwise to the previous mixture for 2 h. The reaction was continued for the time of 2-5 h at the temperature of 25-50 °C. After the reaction completed, the pH value was adjusted to 6.5 with 3% w/v HCl solution. Subsequently, the mixture was centrifuged at 8000 rpm for 5 min and washed twice with distilled water. The mixture was washed twice with 70% v/v ethanol. Finally, the solids were dried at 50 °C for 24 h and passed through a 100mesh sieve.

2.3. Optimization of the OSA modification conditions of pineapple starch

A fractional factorial central composite facecentered (CCF) design was generated using the Design-Expert[®] software package (version 8.0.7.1; Stat-Ease, Inc., USA) and used for optimizing the OSA modification conditions of pineapple starch. The four independent variables were OSA concentration (X₁), pH of the reaction system (X₂), reaction temperature (X₃), and reaction time (X₄). RSM was used to estimate the effects of independent variables on the response variables. Randomization of experiments is required to minimize the effects of unexplained variability in the observed responses due to the presence of extraneous factors ²⁴. A set of 21 experiments was performed, with each variable varied at three coded levels: -1, 0, and +1. The independent variables and the corresponding levels used in the fractional factorial CCF design are listed in Table 1. The design comprises eight factorial points, eight axial points, and five repetitions at the central points. The results were analyzed by analysis of variance (ANOVA). The quadratic equation for the four independent variables can be expressed as follows: ²⁰

$$Y = \beta_0 + \sum_{i=1}^k \beta_i X_i + \sum_{i=1}^k \beta_{ii} X_i^2 + \sum_{1 \le i \le j}^k \beta_{ij} X_{ij}$$
(1)

where Y represents the predicted response variables; β_0 is the intercept; β_i , β_{ii} , and β_{ii} are the regression coefficients of the model; and X_i and X_i (i = 1, 2, 3, 4; i = 1, 2, 3, 4) represent OSA concentration, pH, reaction temperature, and reaction time, respectively. The correlation coefficient r^2 and the root-mean-square error (RMSE) were used for evaluating the polynomial model. The desirability function approach was used for the optimization of multiple response processes ²⁵. It is based on the idea that the quality of a product or process that has multiple quality characteristics, with one of them outside of some desired limits, is completely unacceptable. The method finds operating conditions x that provide the most desirable response values.

2.4. DS determination

DS is the average number of sites per anhydroglucose unit bearing substituent groups. The DS of OSA-modified pineapple starch was analyzed by the titrimetric method reported by Hui et al ¹⁹. An OSA-modified pineapple starch sample (5 g, dry weight) was accurately weighed and dispersed by stirring for 10 min in 100 mL of 90% v/v aqueous isopropyl alcohol solution. Then, 25 mL of 2.5 M HCl–isopropyl alcohol solution was added to the suspension. The suspension was stirred for additional 30 min and

filtered. The residue was washed with 90% v/v aqueous isopropyl alcohol solution until no more Cl⁻ could be detected using a 0.1 M AgNO₃ solution. Starch was redispersed in 300 mL of distilled water, and the dispersion was then heated for 20 min. The starch solution was titrated with 0.1 M standard NaOH solution using phenolphthalein as an indicator. NS served as a blank. DS was calculated using the following equation:

$$DS = \frac{0.162 \times (A \times M)}{1 - [0.210 \times (A \times M)/W]}$$
(2)

where 162 is the molecular weight of a glucosyl unit; 210 is the molecular weight of an octenylsuccinyl group; A is the titration volume of the NaOH solution (mL), M is the molarity of the NaOH solution, and W is the dry weight (g) of OSA starch. The reaction efficiency (RE) was calculated as follows:

$$RE = \frac{Actual DS}{Theoretical DS} \times 100$$
(3)

The theoretical DS value was calculated by assuming that all of the added anhydride reacted with starch to form an ester derivative.

2.5. Surface tension measurement

Surface tension was measured using drop shape analyzer (FTA 1000°, USA) at room temperature. The 3% w/w NS and OSA-modified pineapple starch were heated to form gel. Each determination was performed in triplicate.

2.6. Differential scanning calorimetry

Starch gelatinization was determined by differential scanning calorimetry (DSC; Sapphire DSC: PerkinElmer, Germany) using the technique described by Segura-Campos et al ²³. The data were analyzed using the Pyris software package (PerkinElmer, Germany). To study the starch gelatinization property, 2 mg (dry basis) of starch was placed in an aluminum pan and 8 mg of deionized water was added. The pan was sealed and left to keep for 2 h at room temperature. Then, it was placed in the calorimeter and heated from 30 °C to 120 °C at a heating rate of 10 °C/min under 30 mL/min nitrogen flow using an empty pan as a reference. The gelatinization temperature was determined automatically computing bv the onset

temperature (T_o), peak temperature (T_p), end temperature (T_e), and gelatinization enthalpy (Δ H) from the resulting thermograms.

2.7. Pasting properties

The pasting properties were evaluated according to the procedure described by Bello-Flores et al ¹⁸. NS and OSA-modified pineapple starch were heated in a stress rheometer (Kinexus; Malvern). In brief, 4% w/w (dry basis) starch suspension was heated from 30 °C to 90 °C at a rate of 2.5 °C/min, held at this temperature for 40 min, and cooled to 30 °C at the same rate. Additionally, the peak viscosity was analyzed.

2.8. Powder X-ray diffraction analysis

The powder X-ray diffraction (XRD) patterns of NS and OSA-modified pineapple starches were analyzed by using an X-ray diffractometer (D8, Bruker, Germany) operated at 40 mA and 40 kV. The scanning region of the diffraction angle (2 Θ) was from 5° to 40° using Cu K α radiation wavelength of 1.5406 A°. The starch samples were kept at 50°C for 24 h prior to the analysis. The crystallinity was calculated using the following equation ²⁶,

$$Crystallinity = \frac{area \ of \ crystalline \ peak}{(crystalline + amorphous)area} \quad (4)$$

2.9. Scanning electron microscopy

The NS and OSA-modified pineapple starch samples were attached to the sample holders with conducting glue and sputtered with a layer of gold. These samples were then examined via scanning electron microscopy (SEM; LEO 1450 VP; Zeiss, USA) using beam voltage 10.00 kV and working distance (WD) of 9mm. A magnification of 2000× was used.

2.10. Fourier transform infrared (FTIR) spectroscopy

The changes in the chemical structure of OSA-modified pineapple starch were qualitatively analyzed using FTIR spectroscopy (NICOLET 4700; Thermo Electronic Corporation, USA). The NS and OSA-modified starch samples preparation were carried out using KBr disc method. Each sample was grinded together with KBr powder and compressed to form a pellet at a pressure of 5 tons before placing



Figure 2. Effect of OSA concentration on DS and RE under the following conditions: starch concentration =30% w/w, pH 8, reaction temperature = 35°C, and reaction time = 5 h.

in the sample holder. The spectra were recorded over a wavenumber range of $400-4000 \text{ cm}^{-1}$ with 32 scans at a resolution of 4 cm⁻¹.

2.11. Emulsifying property

Aqueous suspension (3% w/w) of NS and OSA-modified pineapple starch were prepared and heated to form gel. The gel was mixed with the combination of castor oil and oleic acid (1:1) and homogenized at 20,000 rpm for 20 min using homogenizer (T25 Digital ultra turrax, IKA, Germany). The EAI of both starch was determined and calculated with the method described by Pearce and Kinsella²⁷. Briefly, 10 μ L of freshly prepared emulsion was pipetted into 5 mL of 0.1 % w/v sodium dodecyl sulphate and was measured the absorbance using spectrophotometer (T60 UV/Vis spectrophotometer, PG Instruments Ltd, UK) at 500 nm. Each measurement was conducted in triplicate.

Table 2. A fractional factorial CCF design with independent variables and observed responses.

	Independent variables		Respons	es									
Runs	X_1	X_2	X3	X_4	DS	RE	Peak	Surface	%Cryst	Gelatinization			Enthalpy
						(%)	viscosity	tension	allinity	temperatures		(J/g starch)	
							(mPa·s)	(mN/m)		T_0	Tp	Te	
1	30	7	50	5	0.0301	13	23	56.28	26	73.56	79.1	88.21	3.1201
2	1	9	50	5	0.0006	8	12	68.04	29	83.79	86.87	92.75	2.8623
3	15.5	7	37.5	3.5	0.0212	18	21	61.49	30	74.92	82.4	89.6	2.5664
4	15.5	8	50	3.5	0.0270	23	23	57.48	28	79.50	84.29	90.71	1.9522
5	15.5	9	37.5	3.5	0.0085	7	15	60.09	21	82.59	85.89	91.26	2.3744
6	1	9	25	5	0.0013	16	13	67.81	31	83.15	86.34	92.17	2.5056
7	30	7	25	5	0.0161	7	18	59.78	26	80.81	84.79	89.39	1.3217
8	15.5	8	37.5	3.5	0.0262	22	28	55.00	28	78.83	83.47	89.45	1.2212
9	30	9	50	2	0.0162	7	27	56.05	26	74.15	76.98	81.12	4.4990
10	1	7	50	2	0.0024	31	21	67.67	26	83.83	86.76	90.98	2.3016
11	1	8	37.5	3.5	0.0019	26	18	68.54	32	83.86	86.57	90.62	2.0220
12	15.5	8	37.5	3.5	0.0236	20	19	55.47	29	79.38	83.61	89.38	1.5422
13	15.5	8	25	3.5	0.0108	9	21	60.17	26	81.59	85.24	90.62	2.1626
14	15.5	8	37.5	2	0.0247	21	21	55.86	27	80.47	83.99	89.11	1.0497
15	15.5	8	37.5	3.5	0.0244	20	16	55.83	23	79.76	84.5	89.94	1.4281
16	1	7	25	2	0.0028	37	12	67.14	23	83.64	86.88	91.92	2.4479
17	30	8	37.5	3.5	0.0311	13	19	56.64	26	80.36	85.07	89.44	2.1027
18	15.5	8	37.5	5	0.0262	22	22	58.67	26	78.05	84.36	91.35	2.6088
19	15.5	8	37.5	3.5	0.0244	20	20	56.23	22	79.56	84.05	89.5	1.1911
20	15.5	8	37.5	3.5	0.0246	21	18	55.05	28	79.2	83.94	91.19	2.0815
21	30	9	25	2	0.0227	10	24	53.12	23	82.77	85.78	90.76	2.3304

 X_1 : octenyl succinic anhydride (OSA) concentration (% w/w), X_2 : pH, X_3 : temperature, X_4 : time (h), T_o : onset temperature, T_p : peak temperature, and T_e : end temperature

Source	Model	df	Sum square	Mean square	F-value	P-value	r^2	RMSE
DS-model	Quadratic	10	0.00206	0.000206	12.45	0.0002	0.9256	0.0028
Residual		10						
Total		20						
Surface tension-model	Quadratic	9	487.56	54.17	16.87	< 0.0001	0.9324	1.297
Residual		11						
Total		20						
Enthalpy-model	Quadratic	9	10.5	1.17	7.18	0.0017	0.8544	0.2919
Residual		11						
Total		20						

Table 3. ANOVA results for the response surface models.

df: degrees of freedom; RMSE: root-mean-square error

3. RESULTS AND DISCUSSION

3.1. Optimization OSA modification conditions of pineapple starch

The pineapple starch was analyzed by the Cassava and Starch Technology Research Unit through methods of AOAC ²⁸. It is composed of amylose (21.85%) and amylopectin (78.15%). Starch content was 90.22 % and protein, fat, crude fiber, and moisture contents were 1.25%, 0.14%, 0.40% and 5.57%, respectively. Figure 2 shows the effect of OSA concentration on DS and RE under the following conditions: starch concentration = 30% w/w, pH 8, reaction temperature = 35 °C, and reaction time = 5 h. An increase in OSA concentration resulted in an increase in DS and a decrease in RE. Similar trend was also reported by Hui et al¹⁹. The effects of OSA concentration, pH, reaction temperature, and reaction time on DS were preliminarily investigated using a one-factor-ata-time method. Finally, OSA concentrations of 1-30% w/w at pH 7-9 and reaction temperatures of 25-50 °C for a 2-5 h reaction time were selected as independent variables for pineapple starch modification using RSM. A fractional factorial CCF design of 21 experimental runs with the independent variables and the observed responses is summarized in Table 2.

3.1.1. DS model

The DS values of OSA-modified pineapple starch ranged from 0.0006 to 0.0311, and RE ranged from 7 to 37%, as summarized in Table 2. The experimental data were used to determine the relation between the independent variables and DS. RSM was employed to produce a regression equation for the DS value in terms of actual parameters as follows:

DS =	-0.3629 + 0.0026×OSA concentrati	on+
	$0.1003 \times pH + 0.0014 \times Temperature$	_
	0.0269× <i>Time</i> – 0.0002× <i>OSA</i>	
	$concentration \times Time - 0.0002 \times pH \times$	
	<i>Temperature</i> + $0.0032 \times pH \times Time$ +	
	0.0001×Temperature×Time –	
	0.00002×OSA concentration×OSA	
	$concentration - 0.0068 \times pH \times pH$	(5)

The ANOVA results for the response surface models are summarized in Table 3. The full model was reduced by excluding the nonsignificant interaction terms. The *p*-value was used as a tool to examine the significance of each variable, mutual interactions, and regression model. The DS model was significant according to an Ftest value of 12.45 and p < 0.05. The r^2 value was 0.9256 and the RMSE was 0.0028. The OSA concentration, pH, and quadratic terms of OSA concentration x OSA concentration and pH x pH were the significant factors affecting DS. Figure 3(a) shows the combined effects of the OSA concentration and pH on the DS of OSA-modified pineapple starch. The reaction temperature and time were fixed at 37.5 °C and 3.5 h, respectively. It can be clearly observed that the DS value increased with increasing OSA conentration. It was similar to Wang et al ²². This may be due to the fact that the OSA molecule is insoluble in water and the dispersed OSA molecules can enter immobile hydroxyls at the carbon-2, 3, and 6 positions along the starch chains when the OSA concentration is increased. At a high OSA concentration (30% w/w), the maximum DS value was obtained when the optimum pH was at 8, as shown in Figure 3(a). This indicates the interaction effects of OSA and pH on DS¹⁷. When the pH was increased to 9, DS slightly decreased. The similar trend has been reported that pH > 8 promotes undesired side reactions ²⁹, whereas pH<7.5 does not sufficiently activate the hydroxyls of starch for a nucleophillic



Figure 3. Response surface plots showing the interaction effects of reaction condition on (a) DS, (b) surface tension and (c) enthalpy, and contour plot of (d) Graphical optimization displaying the area of feasible response values in the factor space.

attack by anhydride moieties, leading to decreased DS ^{16,19}.

3.1.2. Surface tension model

The surface tension data for OSAmodified pineapple starch ranged from 53.12 to 68.54 mN/m, as summarized in Table 2, whereas the surface tension of NS was 69.8 mN/m. All surface tension values of OSA-modified pineapple starch were decreased when compared with the NS. It may be due to an increase in hydrophobicity after esterification with OSA. The similar pattern was demonstrated by the significantly decreased water sorption capacity of OSA-modified cassava starch ³⁰. The significant quadratic model for surface tension is summarized in Table 3. The model terms were significant (p < 0.05), and the model exhibited a good fit to the experimental data. The r^2 and RMSE value were 0.9324 and 1.297, respectively. The regression equation of the actual model can be expressed as follows:

 $\begin{aligned} \text{Surface tension} &= 223.4526 - 0.99646 \times OSA \\ & \textit{concentration} - 39.45107 \times pH - \\ & 0.35337 \times Temperature + \\ & 6.28208 \times Time + 0.0613 \times pH \times \\ & Temperature - 0.044867 \times \end{aligned}$

 $\begin{array}{l} Temperature \times Time + 0.01939 \times \\ OSA \ concentration \times OSA \\ concentration + 2.27671 \times pH \times \\ pH - 0.5548 \times Time \times Time \end{array} \tag{6}$

The ANOVA results indicated that the OSA concentration, the quadratic terms of OSA x OSA and pH x pH were the significant factors affecting the surface tension of OSA-modified pineapple starch. Figure 3(b) shows the effects of OSA concentration and reaction pH on surface tension, with reaction temperature and reaction time fixed at 37.5°C and 3.5 h, respectively. It was clearly observed that the surface tension of OSAmodified pineapple starches decreased when the OSA concentration was increased to 30% shown in Figure 3(b). This may be due to the fact that increasing hydrophobicity of starch modified by OSA lead to adsorb air-water interface, thereby decreasing air-water surface tension of OSAmodified pineapple starches. This finding was complied with Prochaska et al ³⁰. On the contrary, the surface tension increased at 8<pH<7.5 with respect to lower OSA concentration shown in Figure 3(b). These results correspond with those of DS model. This indicates that pH is a critical factor affecting DS and surface tension of OSA-modified pineapple starches.

3.1.3. Enthalpy model

The gelatinization temperature such as To, Tp, and Te characterized by DSC are summarized in Table 2. The gelatinization temperature was 84.36 °C for OSA-modified pineapple starch and 87.11°C for NS. The enthalpy value of OSA-modified pineapple starch was in the range 1.0497–4.499 J/g starch and that of NS was 4.5939 J/g starch. It was found that the enthalpy values of OSA-modified pineapple starch samples were lower than that of NS, indicating that OSA-modified pineapple starch did not require high energy to melt after modification with OSA. It may be possible that the OSA groups weaken the interactions between starch macromolecules, thereby the starch granules swell and easily melt at low temperature¹⁸. The enthalpy model yielded a significant quadratic fit, and the obtained actual enthalpy model equation is expressed as follows:

Enthalpy = $53.044 - 0.6072 \times OSA$ concentration $- 12.289 \times pH - 0.0084 \times Temperature$ $+ 0.8559 \times Time + 0.0690 \times OSA$ concentration $\times pH + 0.0026 \times OSA$ concentration $\times Temperature - 0.0217$ $\times OSA$ concentration $\times Time + 0.0014 \times$ OSA concentration $\times OSA$ concentration $+ 0.6952 \times pH \times pH$ (7) Among the model terms, the reaction temperature, reaction time, and their interaction effects such as OSA concentration x pH, OSA concentration x temperature, and quadratic effect of pH x pH, were significant model terms for the enthalpy model. The correlation between the actual and predicted values based on the experimental data yielded an r^2 value of 0.8544 and an RMSE value of 0.2918 (Table 3). Figure 3(c) demonstrates the effects of the reaction temperature and time on the enthalpy, with OSA concentration and pH fixed at 15.5% w/w and 8, respectively. The increase in the reaction time and temperature slightly affected the enthalpy of OSA-modified pineapple starch.

3.1.4. Optimization model

The predictive ability of each model was validated by a set of four compounds that were not included in the training dataset. The predicted and experimental values for DS, surface tension, and enthalpy were well correlated with the standard predicted errors of 0.0132, 2.2297, and 1.395, respectively. Thus, it was suggested that the obtained model could exhibit good performance for predicting the DS, surface tension, and enthalpy values. To obtain design space for multiple responses, graphical optimization was performed by specify the



Figure 4. (a) Effect of DS on the pasting profiles (b) XRD patterns of NS and OSA-modified pineapple starch with DS of 0.0006, 0.0244, and 0.0311.



Figure 5. SEM images of (a) NS and OSA-modified pineapple starch granules with DS of (b) 0.0006, (c) 0.0244, and (d) 0.0311. The magnification was $2000 \times$.

desired response limits. In this study, modified pineapple starch with high DS, low surface tension and low enthalpy were required for effective emulsifying property. Figure 3(d) shows the graphical optimization, which displays the area of feasible response values in the factor space. The area that satisfies the constraints is indicated in yellow, whereas the area that does not meet the criteria is indicated in gray. The optimum conditions were 24% w/w OSA, pH 7.8, reaction temperature = 37.5°C, and reaction time = 2 h, with a predicted DS value of 0.03, surface tension of 53.49 mN/m, enthalpy of 1.3417 J/g starch, and desirability value of 0.97.

3.2. Viscosity

The effect of DS on the pasting profiles of OSA-modified pineapple starch is shown in

Figure 4(a). After modification, the pasting temperature decreased, whereas the peak viscosity (at 90°C), final viscosity (at 30°C), and breakdown values increased compared with NS. The viscosity of OSA-modified pineapple starch increased with increasing DS. It may be possible that the hydrophobic groups of OSA being octenyl moieties disrupt hydrogen bonding along starch chains which make the starch chains more easily water-soluble. A similar pattern was found for OSA-modified plantain starch ¹⁸.

3.3. Crystallinity

Figure 4(b) shows the XRD patterns of NS and OSA-modified pineapple starch. The % crystallinity of OSA-modified pineapple starch is listed in Table 2. Both NS and OSA-modified pineapple starch exhibit crystalline



Figure 6. FTIR spectra of NS and OSA-modified pineapple starch.



Figure 7. EAI of emulsions containing NS and OSA-modified pineapple starches with DS of 0.0006 and 0.0311.

A-type XRD patterns, as shown by the strong reflections at approximately 15.1° , 17.5° , and 23° . The diffraction pattern of OSA-modified pineapple starch did not change up to a DS value of 0.0311 because esterification primarily occurred in the amorphous region²².

3.4. Morphology

The SEM results reveal that pineapple starch exhibited Y-shaped granules, with the granule sizes of $5-10 \mu m$. The clustering of granules increased with increasing DS as shown in Figure 5. This could be due to the swelling and disruption of the starch particles during the reaction ²². Similar to potato starch, OSA modification moderately changed the morphology of starch ^{19,22}.

3.5. FTIR spectra

Figure 6 illustrates the FTIR spectra of NS and OSA-modified pineapple starch having different DS. The five absorption peaks of finger print regions were observed between 800-1200 cm⁻¹ which are attributed to C-O bond stretching. The bands approximately appeared at 2928 cm⁻¹ and 3417 cm⁻¹ are characteristics of C-H stretching vibration of glucose unit and O-H stretch vibration of starch, respectively ¹⁹. The band occurred at 1646 cm⁻¹ was due to residual bound water ³¹. Compared with NS, a new peak was found at 1716 cm⁻¹ for OSA-modified pineapple starch, which can be attributed to the formation of ester carbonyl groups ^{19,31}. In the modification process, sodium octenyl succinate starch was formed under alkaline conditions, leading to the appearance of a new peak at 1568 cm⁻¹ that can be attributed to the asymmetric

stretching vibration of carboxylate¹⁹. The absorption peaks' intensity at 1716 and 1568 cm⁻¹ increased with increasing DS ³¹.

3.6. Emulsifying property

The EAI of emulsions containing OSAmodified pineapple starch was higher than that of emulsion containing NS as shown in Figure 7. The EAI was relatively increased with increasing DS of OSA-modified pineapple starch. It is group possible that OSA increases hydrophobicity of pineapple starch and decreases in interfacial tension of emulsion. This was similar to that observed by Miao et al^{32} . Therefore, it could be possible to further study the application of OSA-modified pineapple starch as effective co-emulsifier.

4. CONCLUSIONS

Pineapple starch from stem and rhizome waste was successfully modified with OSA. FTIR spectroscopy confirmed the substitution of the hydroxyl groups in the pineapple starch molecules with the carbonyl groups of OSA. XRD revealed that there was no change in the Atype crystallinity pattern of pineapple starch after OSA modification. The DS value of OSAmodified pineapple starch increased with increasing OSA concentration. The OSA modification of pineapple starch resulted in an increase in the viscosity; however, the surface tension, gelatinization temperature and enthalpy were found to decrease. The OSA-modification conditions of pineapple starch were successfully optimized using RSM. The proposed model could predict the DS, surface tension, and enthalpy values with a reasonable degree of accuracy. The multiple responses were obtained in factor space. The OSA-modified pineapple starch showed better emulsifying property than NS. These finding would be useful for OSA modification of pineapple starch with desirable properties utilizing economical and efficiency optimization technique.

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Conflict of interest

None to declare

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