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Process optimization for the synthesis of octenyl succinyl derivative of waxy corn and amaranth starches

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Abstract

Modification of starch by dicarboxylic acid anhydrides to starch esters, containing both hydrophilic and hydrophobic groups are known to improve its emulsification properties, and can also be used for encapsulation after hydrolysis. Reports on the effect of process conditions on the extent of modification of starches by using *n*-octenyl succinic anhydride (*n*-OSA) are not readily available. In the present study, the process of manufacturing of OSA starches from waxy corn and amaranth starch were studied with respect to the OSA/starch ratio, pH, temperature and time of the reaction. The effects of these parameters were evaluated on the basis of degree of substitution (DS). The concluding conditions for amaranth-OSA starches was a reaction time of 6 h at 3% OSA/starch ratio at 30 °C and pH 8.0 at 25% starch concentration. For waxy corn-OSA starch, all parameters were identical except for the reaction time of 24 h. The maximum DS achieved for both the starches was 0.02. Emulsification capacity and oil absorption capacity of the OSA-modified starches were more or less similar within the parameter chosen and also independent of starch type.

Keywords: Amaranth starch; Octenyl succinic anhydride; Waxy corn starch; Degree of substitution

1. Introduction

Starch is widely used as food ingredient in many products. The partially crystalline nature in native starch is lost, when dispersed and heated in water. Under these conditions, starch granules swell irreversibly to form a viscous paste. Lack of pasting consistency and stability has limited the use of native starch in foods and paved the way for development of modified starch having desirable functional properties such as solubility, texture, adhesion, texture, dispersion and heat tolerance (Rutenberg & Solarek, 1984).

Modification of starch to meet requirements of properties for various applications has been of interest for many years. These modifications disrupt the hydrogen bonding and reduce retrogradation, while imparting other properties, in particular, introduction of hydrophobicity. Such modified starches find applications as emulsifiers in food systems. These can be prepared by treating starches with different alkenyl succinic anhydride, for example, dodecenyl succinic anhydride, octadecenyl succinic anhydride and octenyl succinic anhydride. Starch alkenyl ester contains both hydrophobic and hydrophilic groups. Among all modifications, starch modified with octenyl succinic anhydride (OSA) is permitted for food applications. The chemical structure of such starch is shown in Fig. 1. As per US FDA, addition of OSA for modification is restricted to 3% based on the weight of starch. The incorporation of bulky OSA grouping to hydrophilic starch molecules confers surface active properties to the modified starch so obtained (Trubiano, 1986, 1995). Unlike typical surfactants, OSA starch forms strong films at the oil–water interface giving emulsions that are resistant to reagglomeration. As a result, aqueous solutions of OSA starches have been used to stabilize flavour emulsion in beverages, oil in salad dressings and to encapsulate flavour and fragrances (Trubiano, 1986).

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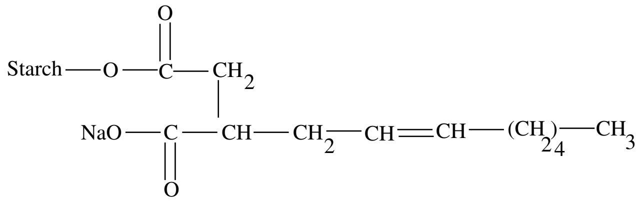

Fig. 1. Structure of starch octenyl succinate.

Cadwell and Wurzburg (1953) patented modification of starches with OSA. This groundbreaking work was based on the esterification of starch with OSA under mild alkaline conditions. More recently, an alternative method for modification of starch (DS up to 0.07) was documented by conducting reaction under harsh acidic conditions (Billmers & Mackewicz, 1997; Tessler & Billmers, 1996). Some recent work examines the distribution of OSA within the modified starch granule and also of the effect of extent of modification on different physicochemical properties of starches (Bao, Xing, Phillips, & Corke, 2003; Shorgen, Vishwanathan, Felker, & Gross, 2000). The physical properties of chemically modified starches are greatly influenced by electronic properties of introduced groups and DS. Hydrophilic substitution such as acetyl, hydroxy propyl and phosphate groups usually increase the gelatinization temperature and viscosity, but the magnitude of the change depends partially on the biological source of parent starches. Hydrophobic substituent such as benzyl, allyl and menthyl often results in more complicated changes (Cho & Lim, 1998).

Most of the earlier the work on modification of starches by OSA was done with waxy corn starch, although in recent times wheat, rice and potato has been also similarly studied. (Bao et al., 2003; Fred & Daigle, 2003). Hydrolyzed OSA starches from waxy corn starch are commercially available under the brand names HiCap, Capsul, NLok, EmCap and Cleargum, as gum arabic substitutes for encapsulation of sensitive food ingredients.

Earlier work in our laboratory has investigated the possibility of using amaranth starch from *Amaranthus paniculatus* Linn. seeds for preparation of useful starch derivatives (Chattopadhya, Singhal, & Kulkarni, 1997; Pal, Singhal, & Kulkarni, 2000). The waxy nature of amaranth starch prompted this study. Commercially available waxy corn starch was used in parallel for comparison.

In this study, we provide detailed investigation on the effect of reaction conditions such as OSA/starch ratio, temperature, pH and time on the synthesis of OSA starch from amaranth and waxy corn starch by evaluating DS. The modified OSA starch was evaluated for its emulsification capacity.

2. Materials and methods

2.1. Materials

Waxy corn starch (C∗ Gel 4320) was obtained as a gift from Cargill Inc, USA. Amaranth starch was isolated in the laboratory by the alkali steeping method (Yanez & Walker, 1986) from *Amaranthus paniculatus* seeds Linn. (Rajgeera). Octenyl succinic anhydride (OSA) 99.9% was obtained as a gift from Dixie Chemicals, USA. Other chemicals and reagents used were of analytical grade. Refined soybean oil was purchased from the local market (density = 0.91 g/ml).

2.2. Methods

2.2.1. Preparation of OSA starches

One hundred and twenty five grams of each of amaranth and waxy corn starch was introduced with agitation in distilled water (475 ml) in a glass reactor. pH of the slurry was adjusted by 2% NaOH solution. To this mixture, OSA at 1.0, 1.5, 2.0, 2.5, and 3.0% based on weight of starch was added dropwise for 2 h with addition after 30 min. During the addition, pH was maintained at 8.0 ± 0.2. The reaction was carried out for 6–24 h. At the end of reaction, pH of the reaction mixture was adjusted to 6.5 using 2% v/v HCl. The resulting starch-OSA derivative was washed with water and centrifuged at 5000 rpm. This was repeated three times after which OSA starches were dried in an air oven at 45 °C. After drying, the starch derivative was milled to 0.25 mm and kept in airtight container until further analysis. The process was optimized so as to obtain maximum DS.

2.2.2. Determination of DS of OSA starches

The DS is the average number of hydroxyl groups substituted per glucose unit. It was determined by alkali saponification followed by back titration of excess alkali. Octenyl succinylation level of the modified starches was determined using the titrimetric method of Whistler and Paschall (1967). Briefly, 25 ml of a 0.5 N aqueous NaOH solution was added to the suspension of the OSA starch (5 g of starch in 50 ml distilled water) and then shaken for 24 h. Excess of alkali was titrated with 0.5 N HCl, using phenolphthalein as an indicator. A blank was simultaneously titrated with native unmodified starch.

DS was determined from % OSA substitution. The calculation was as follows:

$$\%\ \text{OSA substitution} = \frac{(V_{\text{Blank}} - V_{\text{sample}}) \times 0.1 \times N \times 100}{W}$$

where V_{Blank}, volume of HCl required for blank titration; V_{sample}, volume of HCl required for sample titration; W, weight of sample taken (g); N, Normality of HCl solution.

$$DS = \frac{162 \times \%\ \text{OSA substitution}}{21,000 - (209 \times \%\ \text{substitution})}$$

where 162 = molecular weight of glucose unit; 21,000 = 100 × molecular weight of octenyl succinyl group; 209 = molecular weight of octenyl succinyl group.

2.2.3. Scanning electron microscopy (SEM) of OSA modified starches

Samples of both the amaranth and waxy corn and its modified starches were examined by SEM. Starch samples were mounted on the circular specimen holders with adhesives (electrically conductive aluminum plate) and coated with 150 °A gold palladium. The samples were examined in a Philips 505 scanning electron microscope (Kaur, Singh, & Singh, 2004).

2.2.4. *Emulsification capacity of OSA modified starch*

Emulsification capacity was determined by the procedure of Beuchat (1977) at room temperature (30 ± 2 °C). Starch sample (0.05%) was taken in distilled water, heated to 90 °C for 10 min to gelatinize it, and then cooled to room temperature. The above solution was mixed with shear homogenizer (Model type–SPM-9, Indofrench Industries Engineers, Mumbai) at 3000 rpm. Refined soybean oil was added simultaneously with burette at 2 ml/min. The mixing was continued until the emulsion separated in two layers. Emulsification capacity was expressed as g oil/ 0.05 g of OSA-modified starch. Native starches were used for comparative purpose.

2.2.5. *Determination of oil absorption capacity of modified OSA starch*

It was determined as per the procedure Sathe and Salunkhe (1981). Starch (1 g) was mixed with 15 ml refined soy-

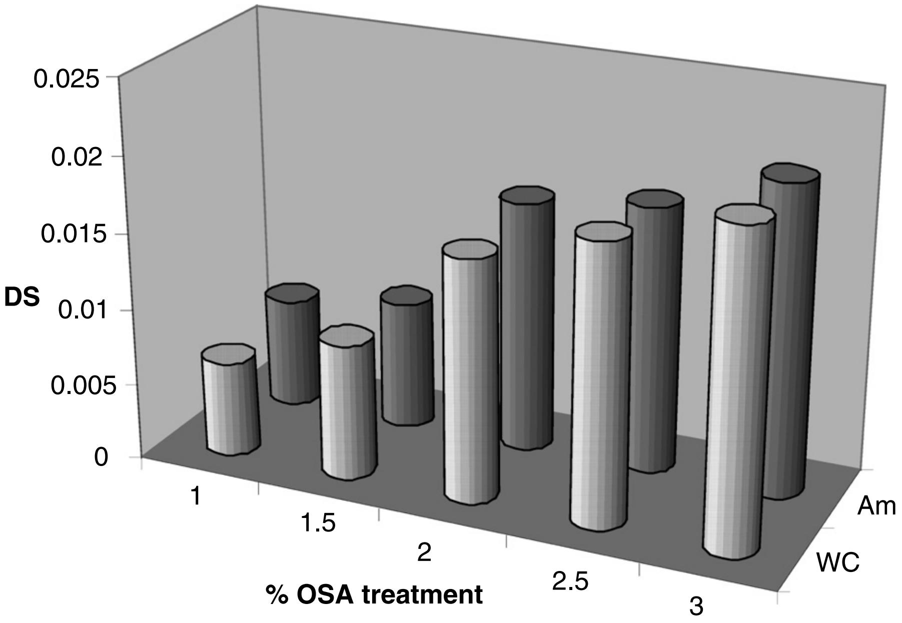

Fig. 2. Effect of OSA concentration (w/w) relative to starch on DS of OSA starches; at pH 8.0, 25% w/w starch concentration, 30 °C for 6 h.

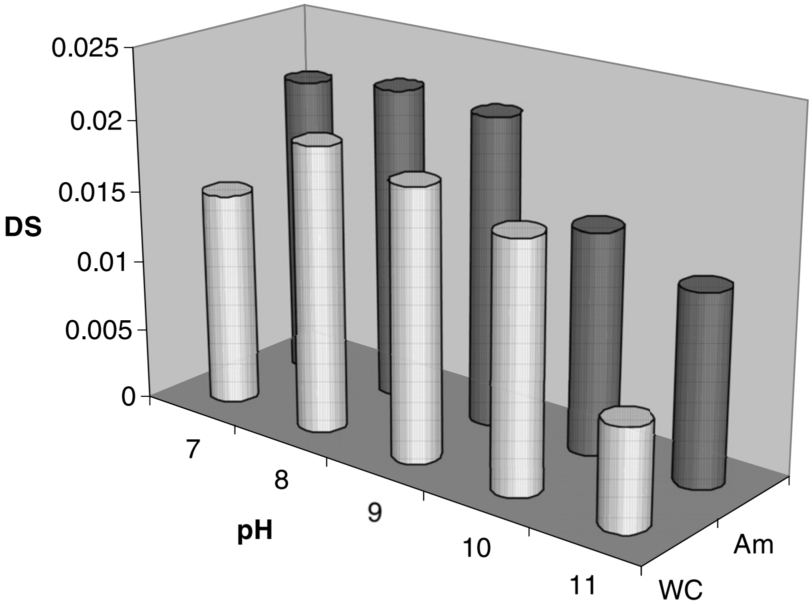

Fig. 3. Effect of reaction pH on DS of OSA starches; 25% starch concentration, 3% OSA addition, at 30 °C for 6 h.

bean oil for 30 s in vortex mixer. The contents were allowed to stand for 30 min at room temperature (30 ± 2 °C), and then centrifuged at 4000 rpm for 20 min. The supernatant oil was decanted until no further oil separated. The amount of oil absorbed by the sample was calculated by the difference in the weight of the sample after centrifugation. Oil absorption capacity was expressed in terms of g oil/g sample.

3. Results and discussion

For modification of starches in an aqueous slurry, it was necessary to preset the concentration of starch slurry for the reaction. From initial trials, 25% w/w starch slurry concentration was decided for both amaranth and waxy corn starch for modification. A concentration higher than 25% w/w, created mixing problem due to the high viscosity. Besides, the energy requirement to disperse the modifying agent would also increases. The other conditions were a temperature 30 ± 2 °C, pH of 8.0 ± 0.2 and reaction time of 6 h.

3.1. Effect of OSA/starch ratio

As per US FDA, the levels of the OSA addition as modifying agent for starch used in food is restricted to 3% w/w

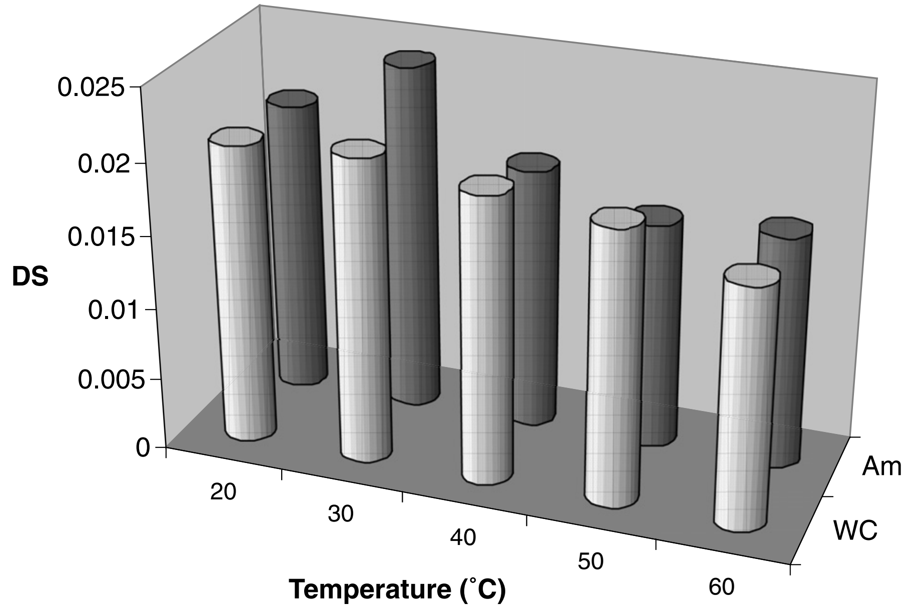

Fig. 4. Effect of temperature on DS of OSA starches; at 25% starch concentration, 3% OSA addition, pH 8.0 for 6 h.

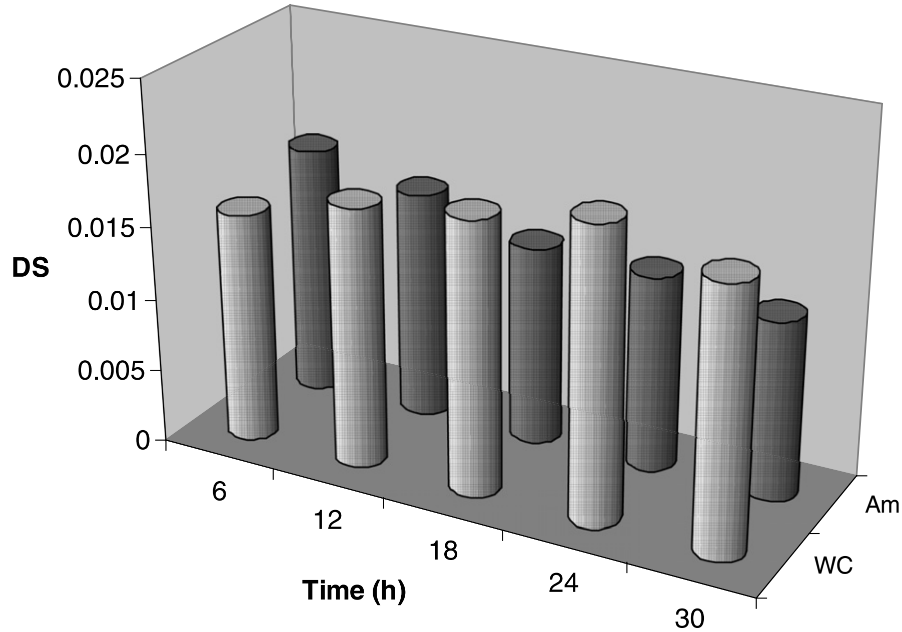

Fig. 5. Effect of reaction time on the DS of OSA starches; at 25% starch concentration, 3% OSA addition, pH 8.0, 30 °C.

based on starch (Trubiano, 1986). Hence the effect of the level of addition of OSA was studied from 1.0–3.0%, keeping all the other condition given above. A linear increase in the DS of the OSA-modified amaranth and waxy corn starches was observed with increase in concentration of OSA (Fig. 2). 3% OSA gave maximum DS to both modified OSA-amaranth and OSA-waxy corn starch. For amaranth starch, treatment with 3% OSA gave a DS of 0.022, whereas for waxy corn starch, it was 0.021 under similar reaction conditions. The increase in DS with increase in OSA concentration could be interpreted in terms to greater availability of the OSA molecules in the proximity of the starch molecule. It is well known that starch hydroxyls are immobile and their reaction will therefore rely on the availability of OSA molecules in the vicinity of the hydroxyl groups. Similar reasoning is reported for the modifiers such as dodecnyl succinic anhydride, decadecenyl succinic anhydride and octadecenyl succinic anhydride (Jeon, Vishwanathan, & Gross, 1999).

3.2. *Effect of pH*

The next parameter that was optimized was the pH of the reaction conditions. In this case, the conditions were an OSA/starch ratio of 3%, temperature of 30 ± 2 °C for 6 h. The modification process was carried out at pH 7.0–11.0. For both the starches, % OSA content was maximum at pH 8.0 (Fig. 3). The DS values for modified amaranth and waxy corn starch was 0.020 and 0.022. The maximum DS at pH 8.0 was explained by the fact that a pH >8.0 favor anhydride hydrolysis, whereas pH <8.0 does not sufficiently activate the hydroxyl group of the starch for the nucleophilic attack of the anhydride moieties (Jeon et al., 1999). Hence pH 8.0 was selected for further studies.

3.3. *Effect of reaction temperature*

Fig. 4 shows that effect of reaction temperature in the range of 20–50 °C. For both amaranth and waxy corn starch, DS was maximum at 30 °C. As the temperature increased from 30 to 60 °C, there was decrease in the DS. For waxy corn starch as temperature increased from 30 to 60 °C, DS decreased from 0.021 to 0.017 whereas for amaranth starch the DS decreased from 0.024 to 0.016. While increase in the reaction temperature may enhance the solubility of OSA and its hydrolysis in aqueous phase, its higher temperature would also expected to enhance

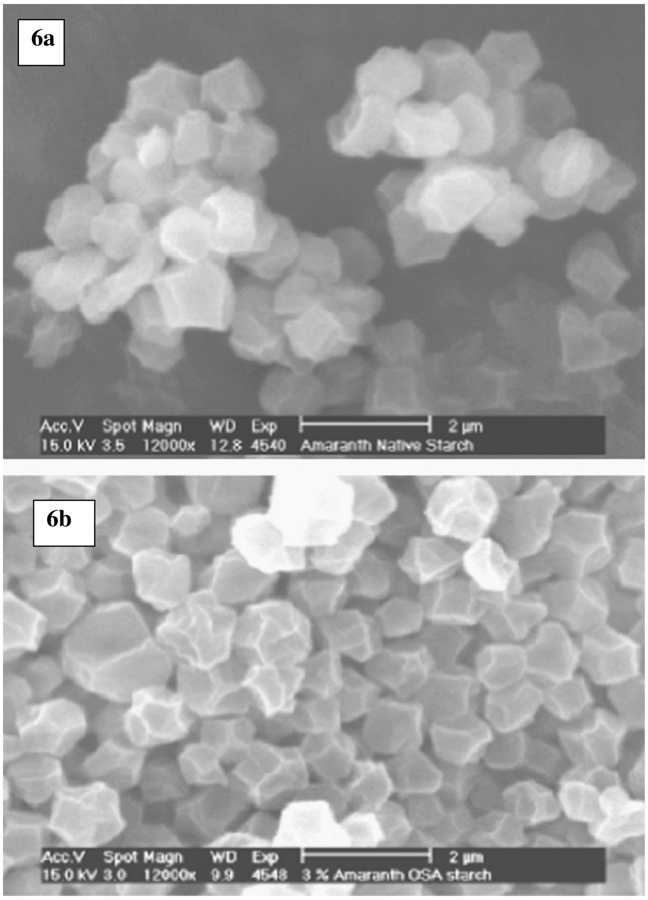

Fig. 6. Scanning electron micrographs: (a) Native amaranth and (b) amaranth OSA starch (DS 0.02) at 12,000× magnification.

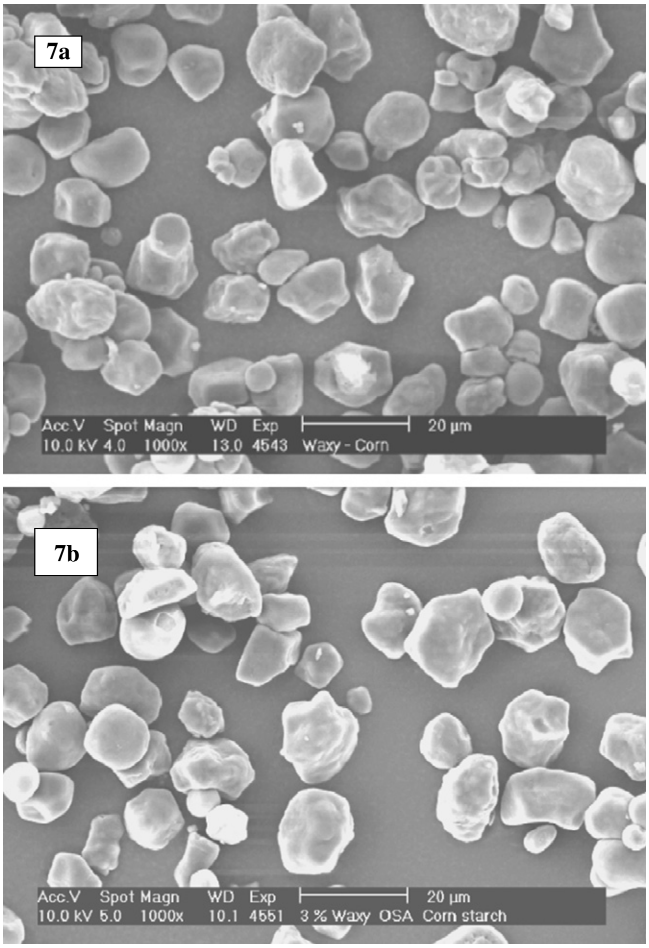

Fig. 7. Scanning electron micrographs: (a) Native waxy corn starch and (b) Waxy OSA corn starch (DS 0.021) at 2000× magnification.

Table 1
Emulsification Capacity[a] and oil absorption capacity of OSA modified starches of amaranth (Am) and waxy corn starches (WC)

% OSA treatment	Emulsification capacity (g)		Oil absorption capacity (g of oil/g of sample)	
	Am	WC	Am	WC
Control	—	—	1.16 ± 0.02	0.92 ± 0.01
1.0	79.62 ± 2.8	75.25 ± 1.92	1.30 ± 0.05	0.96 ± 0.03
2.0	81.17 ± 3.82	76.99 ± 1.25	1.31 ± 0.21	1.04 ± 0.06
3.0	80.53 ± 3.16	79.21 ± 2.96	1.45 ± 0.16	1.00 ± 0.08

All values are means ± SD of three determinations.
[a] Concentratin of starch (0.0125 g/25 ml).

OSA diffusion into starch granule as well as swelling of starch granule that would increase the rate of reaction. But for both the starches, DS decreased as the temperature increased from 30 to 60 °C. These results indicate hydrolysis of OSA to be more pronounced at higher temperatures, and also similar to the results obtained by Jeon et al. (1999). The effect of temperature on modification is a net result of two contradictory phenomenon. A better understanding of effect of temperature on the OSA solubility and starch granule swelling is wanting, and would help in explaining above results.

3.4. Effect of reaction time

Previous experiments were carried out for reaction time of 6 h. To understand the effect of reaction time on the % OSA content and DS, reaction was carried out at different time intervals (6, 12, 18, 24 and 30 h.). The other reaction conditions are 25% starch concentration, 3% OSA addition, 30 °C at pH 8.0. It was observed that for amaranth-OSA starches, DS was maximum at the end of 6 h (Fig. 5). As the reaction time increased from 6 to 48 h, DS decreased from 0.018 to 0.013. For waxy corn starch, maximum DS was observed at reaction time of 24 h. Enhancement of the octenyl succinylation by prolonging the duration of reaction is direct consequence of favorable effect of time on diffusion and adsorption of the reactants between the modifying agents and starch molecule (Khalil, Hashem, & Hebeish, 1995). Effect of reaction time on DS for both the starches are different, can be attributed to the difference in morphology and botanical origin of both starch granules.

3.5. Scanning electron microscopy

Figs. 6 and 7 illustrates the size and shape distribution of amaranth, waxy corn and their OSA derivatives prepared by reacting the parent starch with 3% OSA. Amaranth starch granules showed small granule size (0.80–1.0 μm) were polygonal in shape. Waxy corn starch granules were oval in shape with sizes range from 20 to 50 μm. The modified OSA starches did not show any morphological changes, indicating that the condition for octenyl succinylation did not cause any detectable change in the structural characteristics. These results are in confirmation that of other workers (Kaur et al., 2004; Lawal, 2004; Shorgen et al., 2000).

3.6. Emulsification capacity and oil absorption capacity of modified OSA starches

After optimization of process parameters both starches were modified by treating with 1.0%, 2.0%, and 3.0% of OSA, and evaluated for emulsification capacity (Table 1). The emulsification capacity for the OSA starches were more or less similar for both amaranth and waxy corn starch and did not vary much with the concentration of OSA used for modification in this both study. The native starches did not show any emulsification capacity and oil separated immediately after addition from burette. Similarly, no correlation between the extent of modification and oil absorption was observed with both waxy corn and amaranth starches.

4. Conclusion

Both amaranth and waxy corn starches were successfully modified with octenyl succinic anhydride. All reaction parameters are evenly influenced the reaction DS. No correlation between % OSA treatment and emulsification capacity was observed for both the starches. Further studies to understand the effect of modification on the physicochemical properties necessary to utilize them in food systems and formulations are warranted.

References

Bao, J., Xing, J., Phillips, D. L., & Corke, H. (2003). Physical properties of octenyl succinic anhydride modified rice, wheat and potato starches. *Journal of Agricultural and Food Chemistry, 51*, 2283.

Beuchat, L. R. (1977). Functional and electrophoretic characteristics of succinylated peanut flour. *Journal of Agricultural and Food Chemistry, 25*, 258–261.

Billmers, R. L. & Mackewicz, V. L. (1997). Process for the preparation of hydrophobic starch derivatives. European Patent Application EP 0761691.

Cadwell, C. G., & Wurzburg, O. B., (1953). Polysaccharide derivatives of substituted dicarboxylic acids. U.S. Patent 2, 661, 349 National Starch Products Inc., USA.

Chattopadhya, S., Sinhgal, R. S., & Kulkarni, P. R. (1997). Optimisation of conditions of synthesis of oxidized starch from corn and amaranth for use in film-forming application. *Carbohydrate Polymers, 34*, 203–212.

Cho, K. Y., & Lim, S. T. (1998). Preparation and properties of benzyl corn starches. *Starch/Starke, 50*, 250–257.

Fred, S. H., & Daigle, K. M. (2003). Gelatinization and pasting properties of rice starch modified with 2-octen-1-yl succinic anhydride. *Nahrung/Food, 47*(1), 64–67.

Jeon, Y., Vishwanathan, A., & Gross, R. A. (1999). Studies of starch esterification: Reactions with alkenyl succinates in aqueous slurry systems. *Starch/Starke, 51*, 90–93.

Kaur, L., Singh, N., & Singh, J. (2004). Factors influencing the properties of hydroxypropylated potato starches. *Carbohydrate Polymers, 55*, 211–223.

Khalil, M. I., Hashem, A., & Hebeish, A. (1995). Preparation and characterization of starch acetates. *Starch/Starke, 47*(10), 394–398.

Lawal, O. (2004). Composition, physicochemical properties and retrogradation characteristics of native, oxidized, acylated and acid thinned new cocoyam (*Xanthosoma sagittifolium*) starch. *Food Chemistry, 87*, 205–218.

Pal, J., Singhal, R. S., & Kulkarni, P. R. (2000). A comparative account of conditions of synthesis of hydroxypropyl derivative from corn and amaranth starch. *Carbohydrate Polymers, 43*, 155–162.

Rutenberg, M. W., & Solarek, D. (1984). Starch derivatives: Production and uses. In R. L. Whistler (Ed.), *Starch: Chemistry and technology* (pp. 312–388). New York: Academic Press.

Sathe, S. K., & Salunkhe, S. K. (1981). Functional properties of great northern bean *(Phaseolus vulgaris L.)* proteins: emulsion, foaming, viscosity and gelation properties. *Journal of Food Science, 46*, 70–74, 81.

Shorgen, R. L., Vishwanathan, A., Felker, F., & Gross, R. A. (2000). Distribution of octenyl succinate groups in octenyl succinic anhydride modified waxy maize starch. *Starch/Starke, 52*, 196–204.

Tessler, M. M., & Billmers, R. L. (1996). Preparation of starch esters. *Journal of Environment and Polymer Degradation, 4*, 85–89.

Trubiano, P. C. (1986). Succinate and substituted succinate derivatives of starch. In O. B. Wurzburg (Ed.), *Modified starch: Properties and uses* (pp. 131–147). Boca Raton, FL: Press.

Trubiano, P. C. (1995). The role of speciality food starches in flavour encapsulation. *ACS Symposium Series, 610*, 244–253.

Whistler, R. L., & Paschall, E. F. (1967). *Starch chemistry and technology*. New York, London: Academic Press, pp. 369–389.

Yanez, G. A., & Walker, C. E. (1986). Effect of tempering parameters on extraction and ash of prosomillet flours and partial characterization of proso starch. *Cereal Chemistry, 63*, 164–170.