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# Characterisation of 2-octen-1-ylsuccinylated waxy rice amylodextrins prepared by dry-heating

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ABSTRACT

Waxy rice amylodextrins substituted with 2-octen-1-ylsuccinic acid (OSA) were prepared by heating a dried mixture of waxy rice starch and OSA (3% OSA based on starch solids) at 130–150 °C and pH 3–5 for 1–3 h. The effects of the heating conditions, such as pH, temperature and heating time on the degrees of substitution (DS) and dextrinisation, and the physical properties of the dextrins were evaluated. Substitution and dextrinisation were more affected by the reaction pH than by the physical conditions (temperature and time). Heating under a more acidic condition resulted in a higher DS and a lower molecular weight ($M_w$). Pasting viscosity profiles showed reduced pasting temperatures but increased peak viscosities, due to the presence of OSA, compared to unsubstituted dextrin counterparts. The X-ray diffraction patterns of the OSA dextrins were similar to that of the native starch, indicating that the dextrinisation occurred mainly in the amorphous regions of the starch granules. Fat-free whipped creams were prepared with the OSA dextrins and skim milk powder to examine the fat-replacing effects of the dextrins. The OSA dextrins effectively substituted the fat in dairy cream, providing good foaming ability and storage stability.

## 1. Introduction

Native starches are often tailored by structural modification to develop desirable functional properties, such as solubility, texture, adhesion, dispersion and heat tolerance, in order to be suitable for industrial applications (Rutenberg & Solarek, 1984). One of the common chemical modifications is to substitute starch with organic acid esters using dicarboxylic acid anhydrides. By substituting with alkenylsuccinic anhydride, esters containing both hydrophilic and hydrophobic groups, starch acquires emulsifying properties. Amongst the alkenylsuccinic anhydrides, ocetenylsuccinic anhydride (OSA) is currently permitted in many countries for use in food products (Bhosale & Singhal, 2007), and the preparation of OSA starch by aqueous substitution was patented by Caldwell, Hills, and Wurgburg (1953). Since then, many studies on the preparation and characterisation of OSA starches produced in aqueous alkaline media have been carried out (Bhosale & Singhal, 2006; Cho, Lim, Park, Hwang, & Lim, 1999; Han & BeMiller, 2007; Shogren, Viswanathan, Felker, & Gross, 2000; Song, He, Ruan, & Chen, 2006; Thomas & Atwell, 1999). However, the aqueous reaction has disadvantages because the anhydride of OSA has poor water solubility, and the reaction requires a relatively long time. Some studies have been conducted to increase the reaction efficiency by using organic solvents (Viswanathan, 1999) and acidic conditions (Billmers & Mackerwicz, 2002). Applications of starches substituted with OSA include beverage emulsions, encapsulated flavours, clouding agents, salad dressings, creams, coatings and adhesives (Trubiano, 1986; Thomas & Atwell, 1999), as well as biodegradable plastics (Jane, Robert, Nidolov, & Roque, 1991). The OSA starches, from their emulsifying capacity, were suggested to be used as fat replacers in emulsified foods such as creams (Cho et al., 1999). It has also been reported that OSA substitution makes starch resistant to hydrolysis by digestive enzymes (Han & BeMiller, 2007; Viswanathan, 1999; Wolf et al., 2001), and thus increased the slowly digestible starch (SDS) and resistant starch (RS) contents (Han & BeMiller, 2007). Therefore, when used as fat replacers, the OSA starches provide additional benefits in reducing the total calories and in raising the content of the healthy starch components.

Starch dextrins are also often used as fat replacers (ADA reports, 2005). They are commonly manufactured by acidic and/or enzymatic hydrolysis of starch, but can also be produced by heating dry starch, which results in the thermal degradation of starch molecules. Translocation and repolymerisation often result when more highly branched and complex polysaccharides are treated (Thomas & Atwell, 1999). These structural changes are expected to result in reduced digestibility (Ohkuma, Matsuda, Katta, & Hanno, 1990).

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Recently, some studies have been conducted on the interaction between starch and chemical reagents induced by dry-heating. These include the production of starch phosphate and citrate (Landerito & Wang, 2005; Wing, 1996), as well as starch acetate and succinate (Shogren, 2003).

In this study, OSA dextrins were produced by dry-heating waxy rice starch premixed with OSA, and then the physicochemical properties and the feasibility as fat replacers of the dextrins were investigated.

## 2. Materials and methods

### 2.1. Materials

The waxy rice starch Remyline-AX DR was provided by Remy Industries (Leuven, Wijgmaal, Belgium), and the 2-octen-1-ylsuccinic acid anhydride (OSA) was purchased from Sigma–Aldrich Chemical Company (St. Louis, MO). Skim milk powder and sugar were purchased from a local market in Seoul, South Korea.

### 2.2. Preparation of OSA dextrins

The OSA (2.4 ml) was dispersed in water (120 ml) and stirred (280 rpm) in a temperature-controlled water bath (50 °C) for 1 h, to transform the anhydride to free acid form. The pH of the OSA solution was adjusted to 4.0 by adding NaOH solution (1 N), and then the waxy rice starch (80 g, dry basis) was slowly added to the solution. After mechanically stirred for 30 min, the slurry containing 3% of OSA based on the starch weight was adjusted to pH 3.0–5.0 with HCl (1 N) and NaOH (1 N) solutions. The slurry was then transferred to a glass dish and dried at 45 °C in a convection oven to a moisture content less than 10%. The dry cake was ground to powder and then sieved through 50 mesh. The powder samples of the OSA–starch mixtures were heated at 130–150 °C for 1–3 h in a convection oven. The pH of samples, measured using pH paper, did not change during the heat treatment (data not shown). A sample treated without OSA was also prepared under the same conditions and used as a control.

### 2.3. Degree of substitution

The degree of substitution (DS) of the OSA dextrin was determined following the titration method described by Song et al. (2006) with slight modifications. The OSA dextrin (5 g) was dispersed in 25 ml of an HCl–isopropanol solution (2.5 M) for 30 min, and then isopropanol (90%, 100 ml) was added. The dispersion was stirred for another 10 min, filtered through a glass filter, and then washed with 90% isopropanol until chloride was no longer detected by 0.1 M $AgNO_3$. After drying at 45 °C, the sample (0.5 g) was dispersed in distilled water (80 ml) and then autoclaved. The solution was then titrated with NaOH solution (0.01 N) until neutralised. The degree of substitution (DS) of OSA was calculated by the following equation:

$$\mathrm{DS} = \frac{0.162 \times ((A - A_0) \times N \times F)/W}{1 - [0.210 \times ((A - A_0) \times N \times F)/W]}$$

where $A$ is the titration volume (ml) for OSA dextrins; $A_0$ is the titration volume (ml) for the unsubstituted control; $N$ is the normality of NaOH; $W$ is the dry weight of the sample; and $F$ is the NaOH factor.

### 2.4. Molecular weight analysis

The molecular weight of the OSA dextrins was analysed using gel permeation chromatography (GPC, TSK G5000 and TSK G3000 columns; Tosoh Corporation, Tokyo, Japan) connected to a refractive index detector (RI). The mobile phase used for GPC analysis was an aqueous $NaNO_3$ solution (50 mM) that had been filtered through 0.1 μm cellulose acetate filters (Whatman plc, Maidstone, UK) and degassed with a vacuum pump. The flow rate was controlled at 0.4 ml/min. The dextrin solution was prepared following the procedure of You and Lim (2000), and was filtered through 5-μm cellulose acetate filters whilst hot (about 70 °C) before injection into the GPC system. Pullulan standards (Sigma–Aldrich Company, St. Louis, MO) were used for the standardisation of molecular weights.

### 2.5. Pasting viscosity

The pasting viscosities of the starch and dextrins were evaluated using a Rapid Visco Analyzer (RVA, Newport Scientific, Warriewood, Australia). The viscosity profiles of the OSA dextrins [16.7% solids in a sodium phosphate buffer (0.1 M, pH 7.0)] were determined using the method described by Lim, BeMiller, and Lim (2003).

### 2.6. X-ray diffraction analysis

X-ray diffraction analysis was performed using an X-ray diffractometer (Philips, X′pert MPD high resolution XRD, Almelo, The Netherlands) that was operated at 40 kV, 30 mA, and $\lambda = 0.154$ nm. Diffractograms were obtained from 3° ($2\theta$) to 43° ($2\theta$), using a scanning speed of 4°/min.

### 2.7. Preparation of fat-free cream

Whipped creams containing OSA dextrins and no fat were prepared following the ingredients formulations described in Table 1. The dairy cream (20 g) used in commercial whipped creams contains 35–40% fat (0.7–0.8 g) with some carbohydrates (1.4 g) and proteins (0.4 g). To make the protein content equal 0.4, 1.2 g of skim milk was added. Excluding the sugar added, the total solids content in the formulation was adjusted to 40% by using 6.8 g of the OSA dextrins, which was similar to that of commercial whipped cream. The aqueous dispersions containing the dextrins were whipped with a high shear (10,000 rpm, Ultra-Turrax T25, IKA Werke GmBH & Co., Staufen, Germany) for 1 min, and then stored at 4 °C for 1 h prior to testing.

### 2.8. Overrun of fat-free cream

The fat-free creams containing OSA dextrins prepared at different heating times (30 min, 1, 3 and 5 h) at pH 3 and 140 °C were used in the tests of overrun and foam stability during a cold storage (24 h at 4 °C). Overrun was measured using the following equation:

$$\mathrm{Overrun}(\%) = \frac{W_\mathrm{b} - W_\mathrm{a}}{W_\mathrm{b}} \times 100$$

where $W_\mathrm{b}$ and $W_\mathrm{a}$ were weights of the cream before and after shearing, respectively.

### 2.9. Dynamic oscillatory test

The rheological properties of the fat-replaced creams prepared with OSA dextrins were compared to those of commercial cream. To measure the viscoelastic properties of the creams, a frequency sweep test was performed with a rheometer (HAAKE RheoStress 150, Thermo Fisher Scientific Inc., Waltham, MA) using a parallel plate (35 mm diameter, 1 mm gap). After stress sweeps over the range of 0.1–10 Pa at 1 Hz, 2 Pa was determined from the linear viscosity region. The storage and loss moduli ($G$ and $G''$) of the

**Table 1**
Compositions of fat-replaced creams prepared with OSA dextrins.

| | Ingredients | Lipids (g) | Protein (g) | Carbohydrate (g) |
|---|---|---|---|---|
| Whipping Cream | 20 g dairy cream | 7.0–8.0 | 0.4 | 1.4 |
| | 1.0 g sugar | | | 1.0 |
| Total | 21.0 g | 7.0–8.0 | 0.4 | 2.4 |
| | 6.8 g OSA dextrin | | | 6.8 |
| Fat-replaced cream | 1.2 g skim milk powder | 0.01 | 0.4 | 0.6 |
| (42.9% solids) | 12.0 g water | | | |
| | 1.0 g sugar | | | 1.0 |
| Total | 21.0 g | 0.01 | 0.4 | 8.4 |

cream samples were obtained from a frequency sweep test (0.1–10 Hz at 10 °C).

### 2.10. Statistical analysis

All tests in this study were performed in triplicate, except for the X-ray diffraction analysis. Duncan's multiple-range test was performed using SAS software system (Version 8.10, SAS Inst., Cary, NC).

## 3. Results and discussion

### 3.1. Degree of substitution (DS) and molecular weight ($M_w$)

The dry-heating process for the simultaneous substitution and dextrinisation was controlled by three variables: pH, temperature and heating time. The degrees of substitution (DS) for OSA under different heating conditions are presented in Table 2. When the heating was done at 130 °C for 1 h, the OSA dextrin produced at pH 3.0 had a DS of 0.0171, which was higher than that of the dextrin produced at pH 5 (DS = 0.0151). The same trend was observed when the heating was done at higher temperatures (140 and 150 °C). Therefore, the OSA dextrins were produced with higher substitution yields when the reaction occurred in more acidic conditions. A higher reaction yield of starch citrate has been reported with the dry-heat treatment at a lower pH value (Wing, 1996). The DS could be raised by increasing the heating temperature. The effect of the temperature on the DS increased when the reaction pH was lower (Table 2). Consequently, the reaction affected OSA substitution, so that using a lower pH for the dry-heat treatment was favoured to achieve a greater substitution yield.

**Table 2**
Degree of substitution (DS) and molecular weight ($M_w$) of OSA dextrins prepared under different dry-heating conditions[A].

| Heating | | | DS | $M_w$ ($\times 10^3$ g/mol) |
|---|---|---|---|---|
| pH | Temp. (°C) | Time (h) | | |
| 3 | 130 | 1 | $0.0171^{ef}$ | $269^{e}$ |
| 3 | 130 | 3 | $0.0198^{c}$ | $149^{f}$ |
| 3 | 150 | 1 | $0.0216^{b}$ | $116^{g}$ |
| 3 | 150 | 3 | $0.0232^{a}$ | $60^{h}$ |
| 5 | 130 | 1 | $0.0151^{gh}$ | $1468^{a}$ |
| 5 | 130 | 3 | $0.0149^{h}$ | $1379^{ab}$ |
| 5 | 150 | 1 | $0.0169^{f}$ | $1349^{abc}$ |
| 5 | 150 | 3 | $0.0174^{def}$ | $1199^{bc}$ |
| 3 | 140 | 2 | $0.0187^{cd}$ | $155^{f}$ |
| 4 | 140 | 2 | $0.0168^{f}$ | $713^{d}$ |
| 5 | 140 | 2 | $0.0166^{f}$ | $1319^{abc}$ |
| 4 | 130 | 2 | $0.0164^{fg}$ | $1225^{bc}$ |
| 4 | 140 | 1 | $0.0150^{h}$ | $1179^{c}$ |
| 4 | 140 | 3 | $0.0182^{de}$ | $672^{d}$ |
| 4 | 150 | 2 | $0.0189^{cd}$ | $656^{d}$ |

[A] Data are mean ± standard deviation. Values followed by the same letter in the same column are not significantly different ($p < 0.001$).

The average molecular weight of the OSA dextrins, as determined by gel permeation chromatography, ranged from $60 \times 10^3$ to $1468 \times 10^3$ g/mol (Table 2). Since the glycosidic linkages are susceptible to acids, the chain degradation during the heat treatment was more intense when the pH value was lower. Similar to the case of the OSA substitution, the susceptibility to temperature increase was higher when the pH was lower. Thus, controlling the reaction pH was effective in controlling the thermal degradation as well as the substitution yield for the preparation of OSA dextrins. However, the negative relation between DS and $M_w$ should be considered: the OSA dextrin having the highest DS (0.0232) had the lowest $M_w$ ($6.0 \times 10^4$ g/mol).

The $M_w$ range of the OSA dextrins prepared in this experiment ranged from $10^4$ to $10^6$ which was equivalent to that of amylodextrins consisting of hundreds of anhydroglucose units. Considering the average molecular size of native waxy rice starch ($3.4 \times 10^8$ g/mol; Chung, Han, Yoo, Seib, & Lim, 2008), the thermal degradation induced in this experiment was not minor. It was also expected the molecular size could be further reduced by using a pH less than 3.0. However, heating at a temperature higher than 150 °C to raise the reaction efficiency may induce undesired side-reactions, resulting in colour and flavour changes. The overall results demonstrated that both OSA substitution and dextrinisation could be achieved simultaneously and effectively by dry-heating the starch and OSA mixtures.

### 3.2. Pasting viscosity

The OSA dextrins produced by the dry-heat treatment showed different pasting viscosity profiles, compared to those prepared without OSA, proving the effect from the OSA substitution. Due to the substantially reduced molecular weight, the viscograms could be measured with relatively high solid content (16.5%), and some dextrins, such as those prepared by heating at pH 3 and 150 °C showed no viscograms. When a dextrin was prepared by heating at pH 4 and 140 °C for 2 h, the pasting temperature (PT) of the OSA dextrin (69.0 °C) was lower but the peak viscosity (3508 cP) was substantially higher than those of the unsubstituted dextrin (71.4 °C and 3047 cP) (Fig. 1).

Pasting properties are characteristic for a starch variety and affected by the molecular structure of starches (Srichuwong & Jane, 2007). It was reported that OSA substitution itself raised the viscosity of starch (Bhosale & Singhal, 2007), whereas the dry-heat treatment reduced the peak viscosity for pasting (Lim, Han, Lim, & BeMiller, 2002). However, in this study, the dextrins, regardless of OSA substitution, displayed a readiness to swell even though a significant breakdown was observed (Fig. 1). The setback, which indicates the re-association of the starch chains, was reduced by OSA substitution, indicating that the substituents hindered the chain association.

The viscosity increase by OSA substitution was previously reported for starches, which was supposed to be a result from the formation of amylose–OSA inclusion complexes (Ortega-Ojeda,

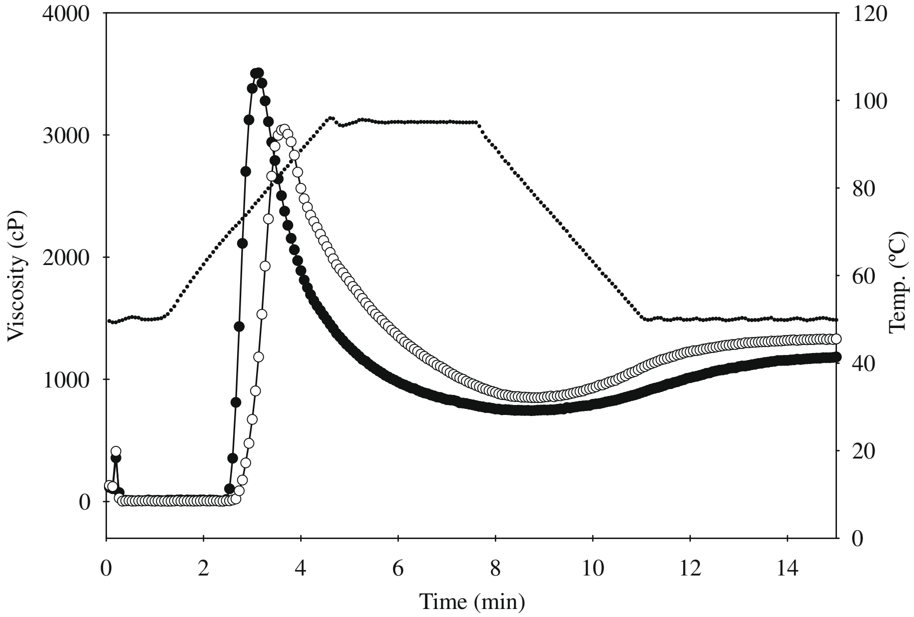

**Fig. 1.** Comparison of Rapid Visco Analyzer (RVA) profiles of 16.7% dispersions of waxy rice starch dry-heated with and without OSA. Waxy rice starch treated at pH 4/140 °C/2 h with OSA (●) and without OSA (○).

Larsson, & Eliasson, 2005; Park, Chung, & Yoo, 2004). However, more research should be carried out to clarify this claim.

### 3.3. Crystallinity

The X-ray diffraction data showed that the OSA dextrins had slightly reduced crystallinity compared to the native waxy rice starch (Fig. 2). As the temperature and time for heating were increased, the decrease in the intensity of diffraction peaks became greater. The OSA dextrins showed three major peaks at 15, 17 and 23° ($2\theta$), which were typical for A-type crystals in cereal starches (Eliasson, 2004). It was reported that there were no significant differences in X-ray patterns between native and OSA starches (Shogren et al., 2000; Song et al., 2006). However, the starch in this experiment underwent thermal degradations to become amylodextrins. Threrefore it may be concluded that the thermal degradation of the starch during the dry-heat treatment occurred mostly in the amorphous regions in the starch granules with little variations in the crystalline structure.

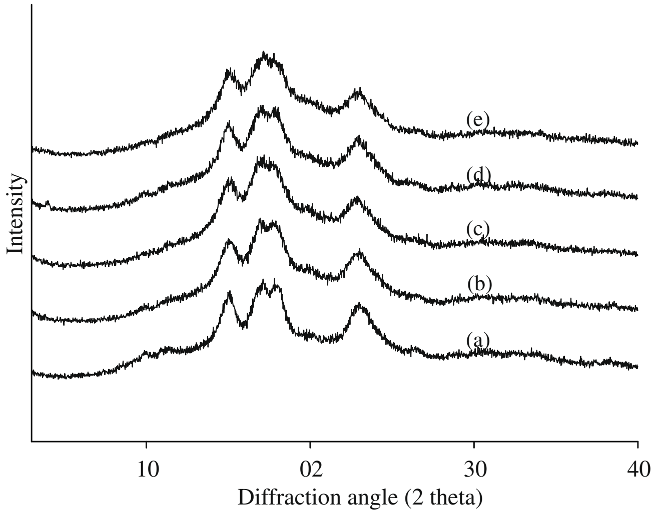

**Fig. 2.** X-ray diffraction patterns of: (a) native waxy rice starch and OSA dextrins; (b) pH 3/130 °C/1 h; (c) pH 3/150 °C/3 h; (d) pH 5/130 °C/ 1 h; and (e) pH 5/150 °C/3 h.

### 3.4. Use in fat-free creams

Fat-free creams were formulated by using the OSA dextrins in the compositions shown in Table 1, and their characteristics were compared with a commercial full fat cream. The changes in the moduli ($G'$ and $G''$) of the creams *vs.* frequency are shown in Fig. 3. Two OSA dextrins (pH 5/130 °C/1 h and pH 3/150 °C/3 h), which had the lowest and highest DS values, were selected for the comparison. As the frequency increased from 0.1 to 10 Hz, both $G$ and $G''$ of all the samples tested increased. It has been reported that $G$ and $G''$ of pasted starches increased as the DS for OSA substitution increased (Park et al., 2004). The creams containing the OSA dextrins displayed higher moduli than did the creams containing unsubstituted counterparts, indicating that the substitution raised the moduli. The cream prepared with the OSA dextrin that had the highest DS (pH 3/150 °C/3 h) displayed the highest stability (least change in modulus) against the frequency change. The surface activity provided by the OSA substituents might result in the increased stability of the cream. It was noteworthy that the two OSA dextrins tested (pH 3/150 °C/3 h and pH 5/130 °C/1 h) displayed modulus values that were not much different, although there was a significant difference in $M_w$. The unsubstituted dextrins treated under the same conditions (Uns-pH 3/150 °C/3 h and Uns-pH 5/130 °C/1 h), however, had a marginal difference in the moduli (Fig. 3). These results indicate that the DS rather than the $M_w$ has a greater affect on the rheological behaviour of the creams, which proves that the OSA groups play an important role.

It was reported that the stability of whipped creams containing dairy protein and starch increased as the concentrations of protein increased because they enhance the hydrophobic interactions with other ingredients (Anderson & Brooker, 1988). It was hypothesised that the hydrophobic nature of the OSA groups in the dextrins interacted with the milk proteins, so that the OSA-containing creams formed a more stable structure than did the unsubstituted dextrin creams. Furthermore, instead of using whisk or wire cage, the mechanical whipping (1 min, 10,000 rpm) was used for inducing swelling or gelatinisation of the OSA dextrins. Microscopic

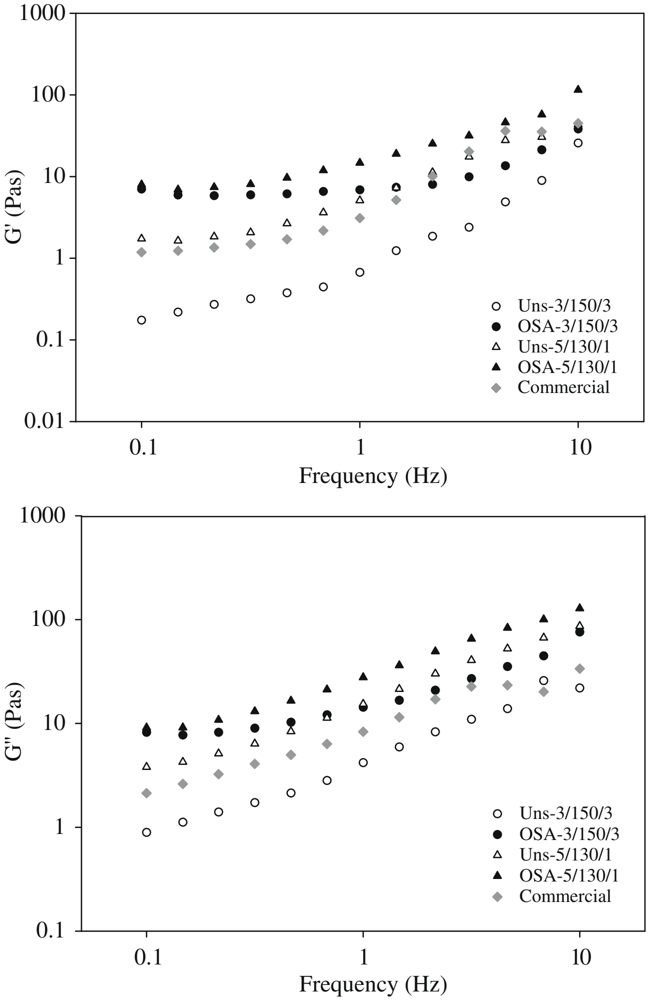

**Fig. 3.** Changes in $G'$ and $G''$ *vs.* frequency for the fat-free creams containing OSA and unsubstituted dextrins, in comparison with a commercial cream.

observation of the creams prepared with the highly substituted dextrin (pH 3/150 °C/3 h) revealed that the dextrin granules were highly swollen with a partial loss of birefringence, whereas the OSA dextrin having a low DS (pH 5/130 °C/1 h) had its granule form not much changed (data not shown). The swelling and partial gelatinisation allowed some of the OSA dextrins to leach out of the granules and to readily interact with the other ingredients such as milk proteins.

The overall behaviour of the dextrins in whipped cream, such as swelling and leaching, may be important in providing the fat-like texture of the cream. In addition, the inherent characteristics of the dextrins, including the chain size and structure (chain length, degree of branching) are expected to influence these interactions and the formation of cream matrices.

The cream containing the OSA dextrins, tested in this study, displayed higher modulus values than did the commercial cream. We assume that using less amount of the OSA dextrins than that in the formulation used in this experiment can give the rheological properties similar to that of the commercial cream. A further study for optimising the formation should be conducted if the OSA dextrins are commercially used as fat substitutes in cream.

### *3.5. Overrun of OSA dextrin-formulated cream*

The overrun of the fat-replaced creams with selected OSA dextrins, which were prepared by heating at pH 3 and 140 °C for different heating times (30 min, 1 and 3 h), were compared (Fig. 4). The OSA dextrin heated for 3 h, which had the highest DS values and the lowest $M_w$, absorbed water rapidly and formed the softest cream. No significant difference in the overrun (%) for the fresh creams (0 h) was observed among the samples prepared at different heating times. However, the dextrin heated for 3 h showed the most stable foam and the least change in overrun after a cold storage for 1 day. The interactions between the OSA groups and the other cream components resulted in formation of a stable matrix structure. Dairy foams are defined as “structures in which a gas is stabilised in a matrix, where a significant proportion of the principal components are of milk origin” (Anderson & Brooker, 1988). Fat globules that are dispersed by shear partially coalesce, which results in the formation of a network, and firm and rigid foam structure (Walstra, 1987). Adding a stabiliser, such as gum, increases the viscosity of cream, thus, the initial overrun of the creams becomes lower than normal cream. However, the stabilisers retard the movement of air bubbles during storage so that the foam structure remains longer (Stanley, Goff, & Smith, 1996). The data in this experiment revealed that the OSA dextrins function as a stabiliser in the cream tested and may be used as fat substitutes. Replacing fat with dextrins reduced the total amount of calories in the cream without significantly changing its rheological properties. We assume that increasing DS with OSA raises the surface activity, which may increase the aeration capacity; optimisation studies in DS and level of thermal degradation should be considered for the utilisation of the dextrin.

## 4. Conclusions

Dry-heating with starch premixed with some carboxylic acids such as octenylsuccinate produces dextrins by simultaneous substitution and thermal degradation. Comparing the reaction in aqueous media, dry-heat treatment of the mixture is simpler and cost-saving. Acidity was favoured for these thermally-induced reactions. For the preparation of OSA dextrins from waxy rice starch in this study, controlling pH (3–5) appeared more critical for both substitution and dextrinisation, compared to the physical heating conditions (temperature and time). The thermal reactions occurred selectively in the amorphous regions of starch, and thus the crystalline nature was maintained in the dextrin. The OSA dextrins prepared by dry-heating in this study could readily swell and be dispersed by mechanical forces, such as whipping, and effectively substitute the dairy fat in whipped creams in a preli-

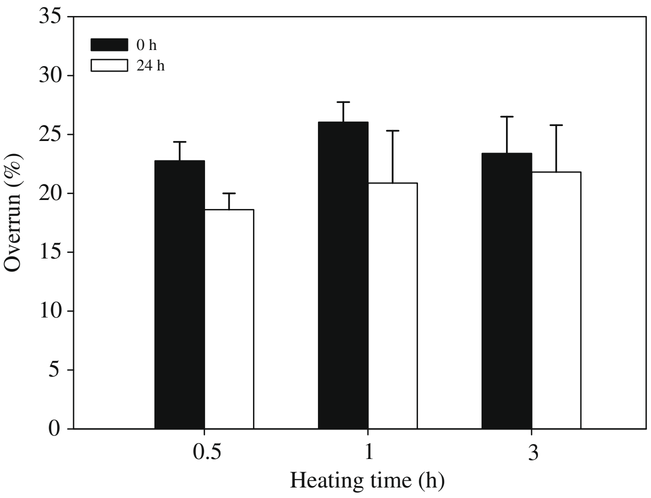

**Fig. 4.** Overrun (%) of fresh and stored (24 h at 4 °C) fat-free creams containing OSA dextrins prepared by dry-heating at pH 3 and 140 °C for different times (0.5–3 h).

minary experiment. However, additional research on the optimisation of the levels of substitution and dextrinisation is needed for the practical use of OSA dextrins as fat replacers.

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