

Size Reduction of Hydrophobically Modified Starch on Beads Mill: Effect of Dispersing Medium and Milling Time

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Abstract: The primary interest to this study was to investigate the effect of milling parameters on the size of hydrophobically modified starch particles, aiming to produce small, uniformly sized modified starch microspheres. Octie, a commercial product originated from cornstarch modified using Octenyl Succinate Anhydride (OSA), was dispersed (3 wt%) using different media (water or ethanol) and subsequently wet-milled using a beads mill with zirconium beads at a rotation of 6,000 rpm up to 30 min. It was found that milling Octie in water dispersion for 3 min resulted in the smallest mean particle size ($2.04 \pm 0.91 \mu\text{m}$), compared to unmilled modified starch granules ($15.2 \pm 6.0 \mu\text{m}$). Granular size and morphology changed considerably with further milling. For instance, very dense clusters with variable particle sizes ($20.6 \pm 10.0 \mu\text{m}$) were obtained after 30 min milling. As depicted by Scanning Electronic Microscopy, a large number of particles were apparently flattened during the milling process rather than broken, forming aggregates. Ultimately, within the range of experimental conditions tested, production of sub-micron modified starch particles was not possible.

Key words: Hydrophobic, octenyl, modified starch, milling.

1. Introduction

Starch is known as an abundant polysaccharide in nature, containing a mixture of two kinds of polyglucans, namely amylose, which is mainly linear and comprised primarily of α -1, 4 linkages with two to eight α -1, 6 linkages, and amylopectin, which is branched and consists of short α -1, 4 oligomers linked by α -1, 6 glycosidic bonds. Starch is found in form of rounded semi-crystalline granules, typically 2-20 μm in size. The size depends mainly on the botanical origin of the starch [1].

In addition to native starch, various modified starches are produced commercially to improve specific functional properties of the product. The desirable properties may be e.g. improved resistance

to mechanical and thermal degradation, and modifications of solubility such as introduction of hydrophobicity [2]. In fact, starch from different sources have been modified, using either chemical or physical approaches, aiming to control somehow their structure, properties and behavior. On these concerns, BeMiller and Lafayette have described some reasons for modifying native cornstarch [3], as follows: in order to modify cooking characteristics, decrease retrogradation, improve texture, adhesion and film formation, or to add hydrophobic groups foreseeing future application in emulsion stabilization, which is also the case in our study.

Hydrophobically modified starch is an amphiphilic macromolecule wherein while the water solubility is retained or enhanced, interactions with hydrophobic phases are likely to occur as well, offering properties that have many applications within the formation and

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stabilization of dispersed food systems such as emulsions [4]. For instance, Rayner et al. [5] demonstrated the use of starch granules isolated from Quinoa as emulsion stabilizing particles, upon hydrophobic modification. Generally, hydrophobically modified starch is prepared by derivatization of granular starch with octenyl-substituted succinic acid in aqueous alkaline medium below gelatinization temperature. Such treatment results in an ingredient with high emulsifying properties (Fig. 1) [6]. During this derivatization, the introduction of hydrophobic groups at low degree of substitution levels (0.01-0.1) imparts some hydrophobic properties to the starch without destroying the water dispersibility. Therefore, in order to disperse modified starch in water, it is necessary e.g. to moisten them with a water-miscible solvent such as ethanol or acetone [7].

The practice of using particles to stabilize emulsions, the so-called Pickering emulsions, has been used since many decades ago [9, 10], whereas this term refers to emulsions consisting of fine droplets stabilized by solid particles adsorbed at the interface between two immiscible phases. The use of particles, such as hydrophobically modified starch, to stabilize emulsions has attracted substantial research interest due to their distinctive characteristics and potential technological applications [5]. Emulsions

stabilized by solid particles are usually more stable against coalescence and Ostwald ripening compared to systems stabilized by surfactants [11].

On these regards, the purpose of this study was to investigate the effects of milling process variables on modified starch particles size, aiming to produce small, uniformly sized modified starch microspheres. Due to the presence of the highly hydrophilic starch backbone and the hydrophobic modification introduced to the chain, modified starch may find applications as stabilizer beyond foods, such as in pharmaceutical or cosmetics industries.

2. Materials and Methods

2.1 Materials

Octie (INCI Code: Aluminum Starch Octenylsuccinate; Nichidene Chemical Corp., Japan; Red Ball Brand) is a fine powder with average particles size of $15.2 \pm 6.0 \mu\text{m}$ originating from corn starch. Ethyl alcohol was purchased from Wako Pure Chemical Industries, Ltd. (Osaka, Japan). Deionized water (Milli-Q) with a resistivity of $18 \text{ M}\Omega\cdot\text{cm}$ was used for preparing all aqueous solutions.

2.2 Methods

The methods used in this study are described below:

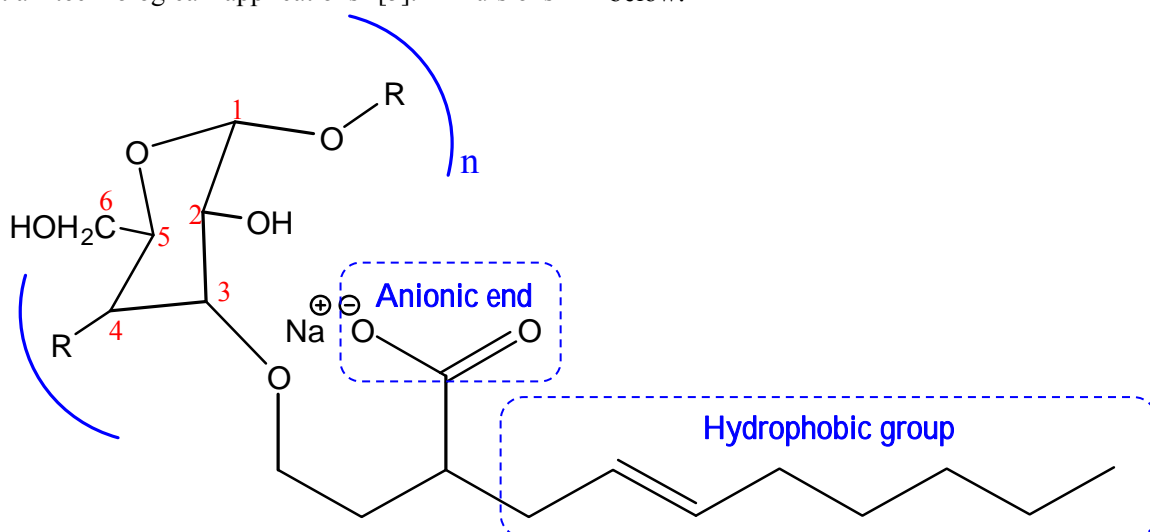


Fig. 1 Chemical structure of n-octenylsuccinate-derivatized starch. The substitution can occur at positions 2, 3 or 6 of starch molecule [8].

2.2.1. Wet Milling

Initially, Octie was pre-wetted with a minimum amount of acetone (0.1 mL/g-starch), and 100 mL of Octie dispersions in Milli-Q water (3 wt%) were prepared. The starch dispersions were wet-milled using a horizontal beads mill (Dispermat SL-C5, VMA-Getzman Reichshof), initially filled with 40 mL of zirconium oxide grinding beads (500 μm size), at rotation speed 6,000 rpm up to 30 min, while the temperature was kept below 25 °C by circulating cold water at 5 °C through the external mantle of the mill. All milling experiments were conducted in duplicate, and the wet-milled samples were kept at room temperature for posterior analyses.

2.2.2. Particle Size Measurement

The mean particle size and size distribution of unmilled and milled Octie collected at various milling times (1, 3, 7, 15 and 30 min), were measured by laser diffraction in combination with Polarization Intensity Differential Scattering (PIDS) technology (LS 13320, Beckman Coulter Ltd., Florida, US). The measurements were duplicated for all batches.

2.2.3. Morphological Analyses

Octie samples (unmilled or milled at various residence time) were mounted onto stubs using double-sided adhesive carbon tape. A thin layer of gold (100 Å) was applied to the surface of the particles, in a sputter coater (JFC-1500 Quick Auto Coater, JEOL, Japan) during 1 min at 7 mA, to make samples conductive. The coated samples were transferred to the scanning electron microscope (SEM) (JEOL JSM-94 5600 LV, JEOL, Japan) and micrographs were recorded at various magnification.

3. Results and Discussion

3.1 Effect of Milling Time on the Particle Size Distribution of Modified Starch

The primary interest of this study was to decrease the modified starch particles size, for the potential application of modified starch as emulsifier in

oil-in-water emulsion systems. The original modified starch particles (before milling) are depicted in Fig. 2a. After milling the modified starch dispersion for 15 min., a few partially broken particles were observed (Fig. 2b). Generally, the particle size increased with milling time, after 3 min milling.

The apparent laser scattering based mean particle size of unmilled and milled Octie were analyzed and the size distribution is depicted in Fig. 3. Unmilled Octie (3 wt% dispersed in water) showed a bimodal particle size distribution with average size of $17.21 \pm 10.57 \mu\text{m}$ (mean \pm SD, $n = 3$). Reduced granule size could be observed after 3 min milling. Beyond 15 min, further increase on milling time resulted in considerably large mean particle size.

Based on the particle size distribution and microscopic observations, changes on starch granules size and shape with increasing milling time were represented schematically in Fig. 4. Apparently, milling Octie particles beyond 3 minutes resulted in deformation of single particles initially, followed by the formation of larger clusters of particles, regardless of the dispersion medium. This hypothesis was further supported during the morphological characterization of the milled Octie particles, especially upon 30 min milling (refer to the following section).

3.2 Morphological Characterization of Milled Octie

The results indicated that the milling time affected not only the granules size, but also granules morphology, as indicated in the following micrographs obtained by SEM (Figs. 5 and 6).

Fig. 5 depicts unmilled Octie granules. Generally, the particles presented a well-defined polygonal, rounded or spherical shape, and were isolated from each other. While the dimension of most of the granules was in the order of 10 μm , a few particles had dimensions situated between 15 μm and 20 μm , characterizing a bimodal particle size distribution, which agrees well with the data shown above for unmilled Octie particle size distribution (Fig. 3).

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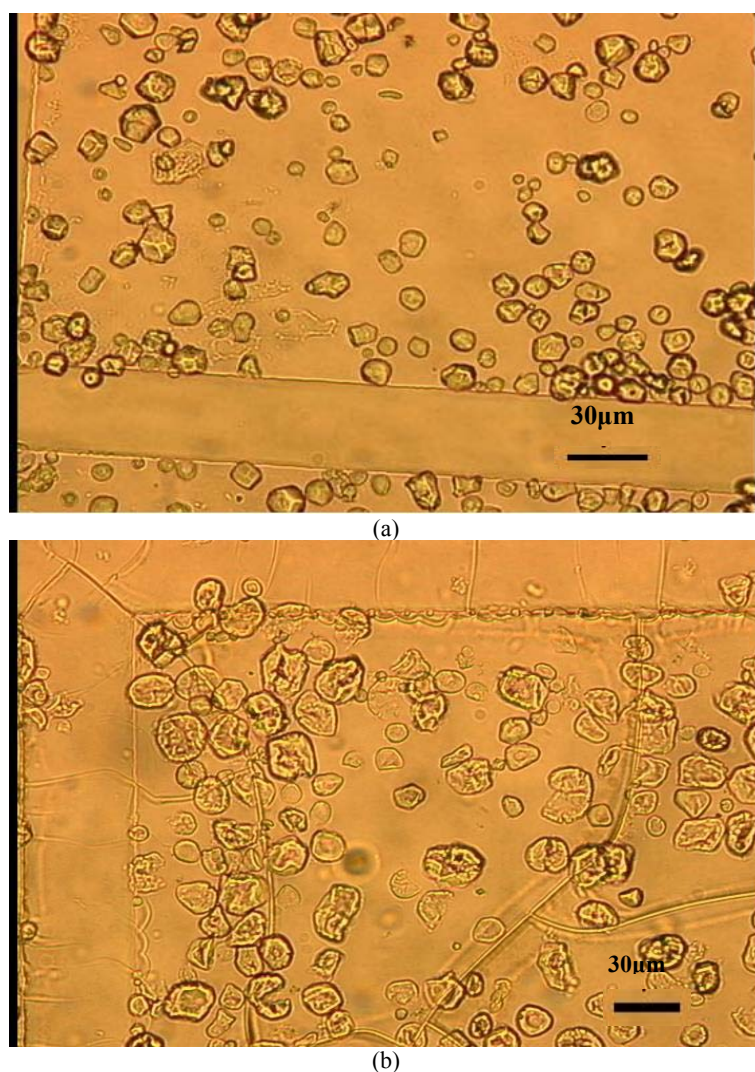


Fig. 2 Optical micrographs of Octie dispersed in water (0.3 wt%). (a) Before milling; (b) Milled at 6,000 rpm for 15 min in beads mill.

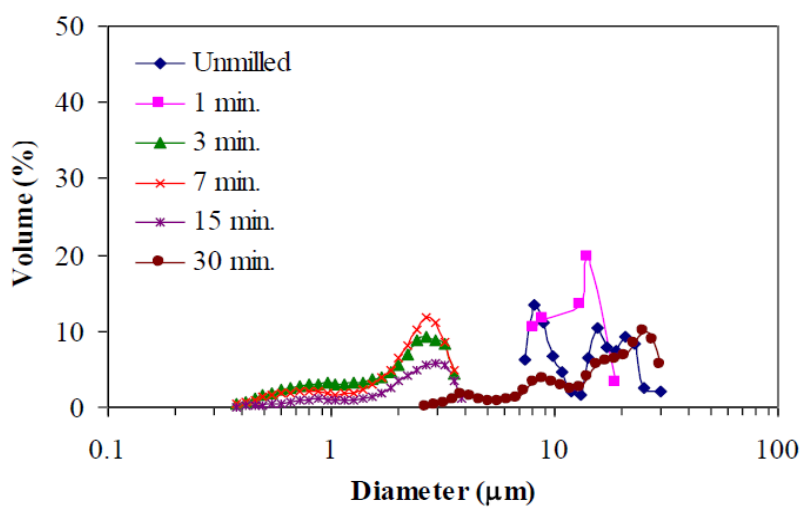


Fig. 3 Effect of milling time on the particle size distribution of modified starch (3 wt% dispersed in water).

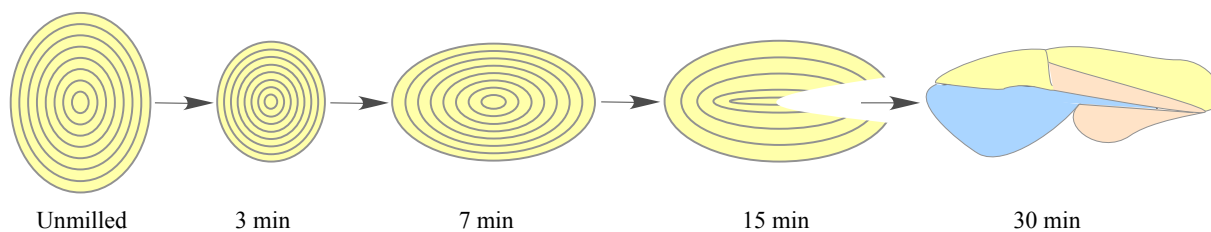


Fig. 4 Scheme representing the effect of milling time on starch granule size and shape, from round (unmilled) stretched to a large cylindrical form (after 30 min milling); in this case, various particles were agglomerated.

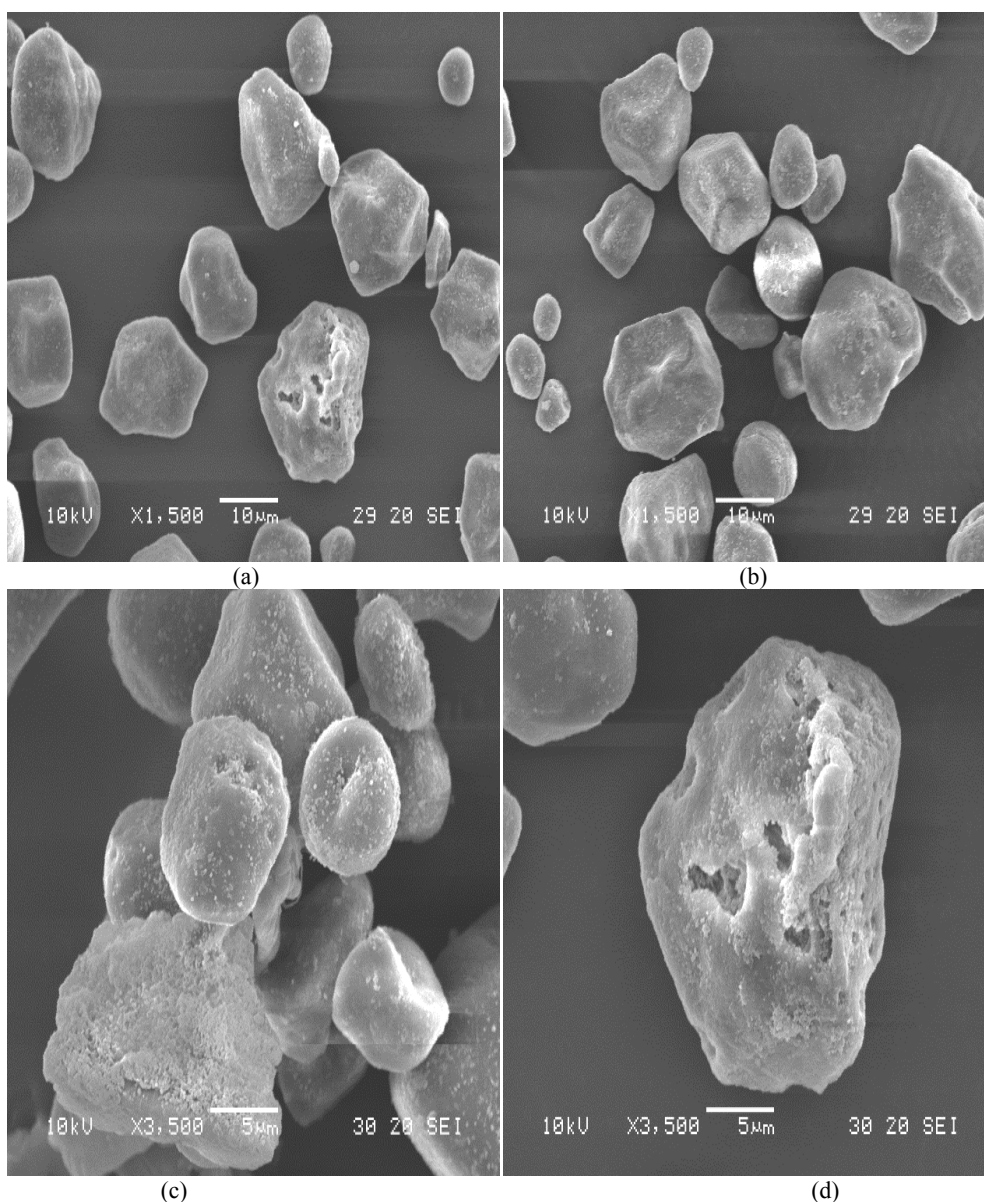


Fig. 5 SEM micrographs of Octie particles before milling.

3.2.1 Milling with Water

After 3 min milling at 6,000 rpm, the mean particle size was $2.04 \pm 0.91 \mu\text{m}$, which is substantially smaller compared to the unmilled Octie. A few

particles showed some deformation, with small disk-like flattened regions or holes (indicated by arrows in Fig. 6a). The mean particle size after milling for 7 min was $2.25 \pm 0.94 \mu\text{m}$, which is slightly higher

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than the size obtained at 3 min milling. On the other hand, the particles started to form agglomerates (indicated by white arrows in Fig. 6b). In addition, most granules showed a characteristic deformation. As the milling time was increased to 15 min, the formation of clusters with variable size was observed. Fig. 6c shows an individual starch granule, which had been flattened during milling, resulting in a granule size relatively larger, compared to the unmilled Octie. Further increase on milling time up to 30 min resulted in denser network, of relatively large size. As depicted in Fig. 6d, generally no individual granules were

observed. The white arrow indicates a typical flattened structure, which was supposedly polygonal or round shaped before milling, stretched to a large cylindrical shape. This behavior is consistent with the particle size distribution results, indicating that the milling for prolonged time caused disintegration of the granules, and was represented schematically in Fig. 4.

Native starch granules are known to have ordered crystalline regions formed by amylopectin and less ordered amorphous regions (Fig. 7). The granules deformation observed in this study are more likely to have occurred in the amorphous rather than in the

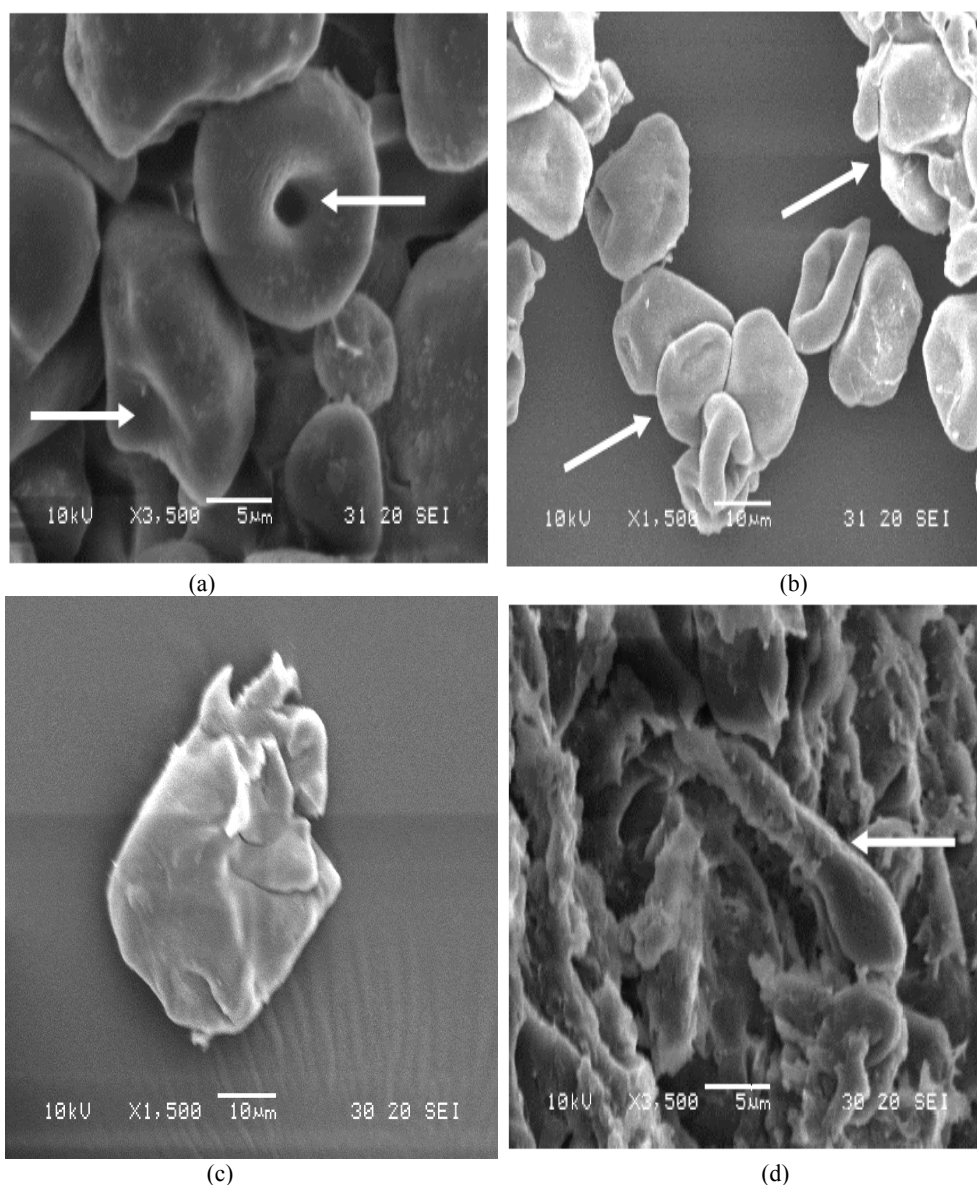


Fig. 6 SEM micrographs of Octie dispersed in water milled at 6,000 rpm for: (a) 3 min; (b) 7 min; (c) 15 min; (d) 30 min.

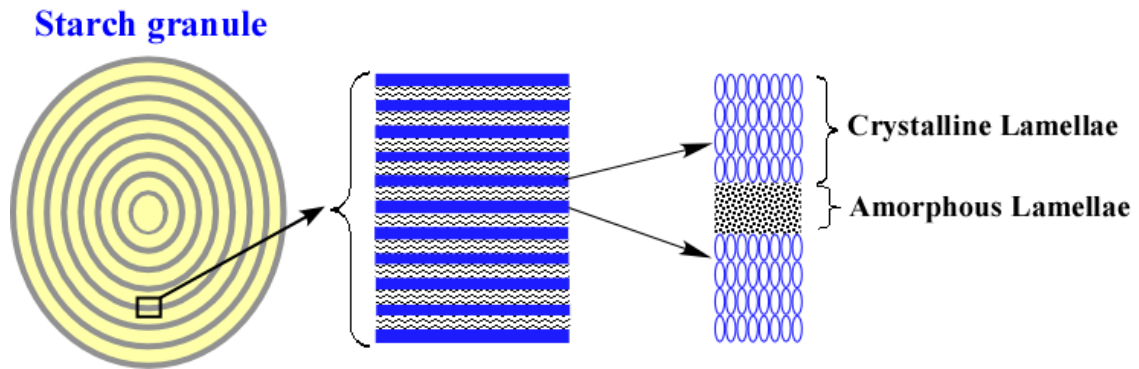
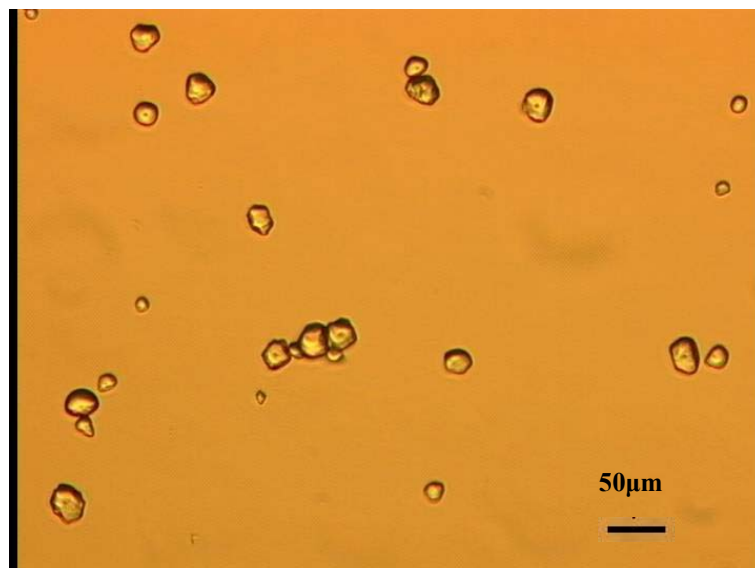


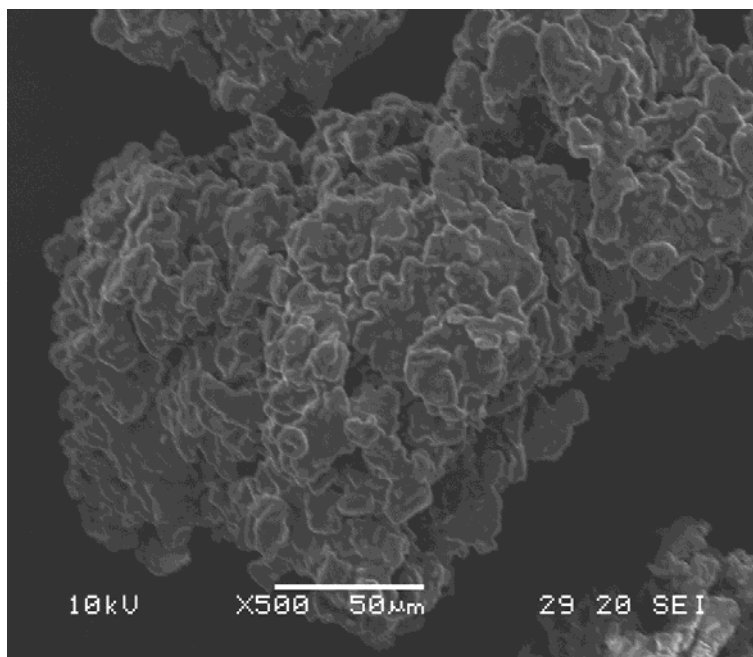
Fig. 7 Schematic diagram of crystalline and amorphous layer structure of starch granules (Adapted from Ref. [1]).



(a)



(b)



(c)

Fig. 8 Optical and SEM micrographs of Octie dispersed in ethanol (10 wt%). (a) Before milling; (b) Milled at 6,000 rpm for 30 min in beads mill; (c) SEM image of milled Octie (under the same conditions as Fig. 8b).

crystalline lamellae, assuming that the hardened structure on the crystalline regions is supposed to cause the granules to break down.

3.2.2 Milling with Ethanol

Initially, using water as the dispersing medium was thought to soften the starch granules, causing them to flatten and agglomerate, as the milling time increases. Based on this supposition, and taking into account that Octie is poorly wetted by water, but disperses evenly in a less polar water-soluble solvent such as ethanol or acetone, ethanol dispersions (10 wt%) were prepared for milling under the same experimental conditions used previously, for water dispersions. The results of these experiments are presented in Fig. 8. Milling in presence of ethanol for 30 min resulted in relatively large clusters, as previously observed during milling with water.

Based on the results above, the medium used to disperse the modified starch granules did not have apparent effect on the mean particle size after milling.

In order to produce oil-in-water droplets stabilized by hydrophobic starch in absence of any other surfactant agent, the starch particles size should be

considerably smaller than the droplet size. For instance, initially Octie granules have roughly 15 μm , which was considerably reduced to c.a. 2 μm , after 3 min milling. Meanwhile, the use of microchannel emulsification devices enables production of monodisperse emulsions with average droplet size ranging from 3 μm to 90 μm [12].

As reported in previous literature, unlike typical surfactants, hydrophobic starch forms strong films at the oil-water interface giving emulsions that are resistant to reagglomeration [2].

Potential future work would include conducting the milling utilizing different starch crop varieties such as amaranth (genus *Amaranthus*), reported to have small sized starch granules (0.80-1.0 μm), compared to waxy corn starch granules ranging from 20 μm to 50 μm [2].

4. Conclusion

The smallest mean particle size of modified starch was $2.04 \pm 0.91 \mu\text{m}$, obtained after milling for 3 min using the beads mill at 6,000 rpm. Beyond this time, the particles size increased with milling time. Milling

starch granules for prolonged time, e.g. 30 min caused the granules to flatten, resulting in considerable increase on the mean particle size, as well as the formation of a large number of agglomerates.

Based on the results above, the dispersing medium, i.e. water or ethanol, does not seem to have influence on the mean particle size after milling.

Sub-micron particles were unattainable within the range of experimental conditions tested. Further reduction on modified starch granules size is a determinant factor to succeed on emulsion stabilization using hydrophobized octenyl succinate starch.

Acknowledgement

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