

## Properties of Chitosan-Microencapsulated Orange Oil Prepared by Spray-Drying and Its Stability to Detergents

Yue Li,<sup>†,§</sup> Lianzhong Ai,<sup>†</sup> Wallace Yokoyama,<sup>#</sup> Charles F. Shoemaker,<sup>⊥</sup> Dong Wei,<sup>§</sup> Jianguo Ma,<sup>§</sup> and Fang Zhong<sup>\*,§</sup>

<sup>†</sup>State Key Laboratory of Dairy Biotechnology, Technology Center of Bright Dairy and Food Company Ltd., Shanghai 200436, People's Republic of China

<sup>§</sup>Key Laboratory of Food Colloids and Biotechnology, Ministry of Education, School of Food Science and Technology, Jiangnan University, Wuxi 214122, People's Republic of China

<sup>#</sup>Processed Food Research, Agricultural Research Service, U.S. Department of Agriculture, Albany, California 94710, United States

<sup>⊥</sup>Department of Food Science and Technology, University of California, Davis, California 95616, United States

**ABSTRACT:** Fragrance encapsulated in small particles of <20  $\mu\text{m}$  diameter is preferred for use in textiles. This study demonstrated that the proper combination of surfactants could produce small and heat-stable emulsion droplets with chitosan that could be spray-dried to produce microcapsules. The microcapsules were able to be deposited onto cotton using water or detergents. It was found that stable emulsion was obtained when Tween 40 and Span 20 were used as compound emulsifiers with the ratio of 4:1 (w/w). The optimum conditions were 1% (w/w) chitosan in acetic acid with the compound emulsifiers of 3–7% (w/w) in the oil, and the inlet temperature for spray-drying was 150  $^{\circ}\text{C}$ . The encapsulation efficiency for orange oil was >90% with a 1:2 (w/w) ratio of oil to chitosan. Microcapsules had a mean diameter of <20  $\mu\text{m}$  and regular particle morphology. The orange oil in the microcapsules was well retained in cotton fabrics after washing in normal detergent solution. The process and products are low in cost, nontoxic, biocompatible, and biodegradable.

**KEYWORDS:** microencapsulation, chitosan, orange oil, emulsion, encapsulation efficiency, retention properties

### ■ INTRODUCTION

The adsorption of perfumes and other fragrances onto solid carrier materials has been of interest in textile manufacturers because they can be more conveniently incorporated into textiles. However, volatile fragrances bound to solid carriers by adsorption are easily removed during ordinary washing processes by laundry detergents. To increase the persistence of fragrance on textiles, the microencapsulation technology widely used by the food and pharmaceutical industries was adapted by the detergent and fabric softener industries. Presently, the microencapsulated particles used in detergents have an average particle size of  $\leq 100 \mu\text{m}$ . It was reported that microencapsulated particles of  $\leq 20 \mu\text{m}$  are more easily incorporated into textiles during the final washing step.<sup>1</sup>

Materials used for microencapsulation intended for wearable textiles should be nontoxic, biocompatible, biodegradable, and low in cost. Currently, the preferred encapsulating polymers are prepared from melamine–formaldehyde or urea–formaldehyde condensates and other similar aminoplastics. Formaldehyde is a toxic chemical and probably should not be used in textiles that come in close contact with the body. Capsule-coating materials made with gelatin via simple or complex coacervation may also be preferred. Capsules microencapsulated by shell wall materials composed of polyurethane, polyamide, polyolefin, polysaccharide, protein, silicone, lipid, modified cellulose, gums, polyacrylate, polystyrene, and polyesters or combinations of these materials are also known.<sup>2–4</sup> The size of the microcapsules prepared by these polymers is often too large for textile applications.

Some small microparticles have been reported recently. Gordon prepared 5–10  $\mu\text{m}$  microcapsules containing volatile mint flavor core material;<sup>5</sup> however, melamine formalin polymerization was used. Interfacial polymerization was used to produce polyurethane/urea (PUU) microcapsules with a perfume-containing core for industrial application on a textile substrate for men's suits.<sup>6</sup> The mean particle size of those microcapsules was 10  $\mu\text{m}$ . Ethylcellulose has also been used as the wall material to produce microcapsules that were grafted to textiles with particle size range of 10–90  $\mu\text{m}$ .<sup>7</sup> Gelatin and polysaccharides have been used to produce microcapsules by coacervation with particle size ranging from 50 to 100  $\mu\text{m}$  for use in fabric softeners or detergents.<sup>8</sup>

Chitosan, a natural polysaccharide of glucosamine, and *N*-acetylglucosamine, prepared by partial deacetylation of chitin,<sup>9</sup> have been used by the pharmaceutical industry for controlled drug delivery systems of conventional drugs, protein drugs, and DNA and may have potential for wearable textile applications. Chitosan microparticles have attracted increasing attention in the past decade due to their unique properties including nontoxicity, biocompatibility, and biodegradation.<sup>10,11</sup> Numerous controlled-release delivery systems of chitosan for either implantation or oral delivery have been described in the literature.<sup>12–14</sup>

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In addition to its nontoxicity, chitosan is an attractive candidate for textiles because many processing techniques for the preparation of microcapsules have been developed since the 1980s. Some of the principal approaches are ionotropic gelation by oppositely charged, simple or complex coacervation; emulsification/solvent evaporation; and more recently, emulsification/spray-drying.<sup>15</sup> The latter process seems best suited for the development of controlled release volatile fragrances microcapsules based on the known properties of chitosan microparticles. Spray-dried chitosan microparticles are characterized by highly spheroid shape and high specific surface area, as well as small particle size. Fragrance encapsulated by chitosan may be controlled or slowly released in water and alkali conditions present in detergent solutions. In addition to useful performance characteristics of spray-dried microparticles, the spray-drying process can be applied on a large scale and is low in cost.

One of the research challenges to spray-drying encapsulation is to minimize the amount of free or unencapsulated oil at the surface of powder particles.<sup>16–18</sup> Factors that affect the encapsulation efficiency of flavors and oils include the properties of wall and core materials and the spray-drying parameters, as well as the emulsifier characteristics.<sup>19–23</sup> Emulsification and the emulsifying ingredients play a key role in this regard. There are some reports that smaller oil droplets are more efficiently retained in the encapsulated product so that the resulting microcapsules will have less free oil on the surface.<sup>24,25</sup> The application of small molecule surfactants for microencapsulation development is an area that has not been studied extensively because these surfactants do not have desirable encapsulation properties such as film-forming abilities. The objective of this study is to determine the influence of different surfactants and their combinations on emulsifying effectiveness at high temperatures and the encapsulation efficiency of spray-dried microencapsulated orange oil. The retention of the orange oil in fabrics after washing in normal detergent solution was evaluated as well.

## MATERIALS AND METHODS

**Materials.** Chitosan (DE = 50000) was provided by Anhui Cigarette Factory (Hefei, China). Orange oil (Hua Bao Food Flavor and Fragrance Co. Ltd., Shanghai, China) was used as the core material. Nonionic surfactants, that is, Tween 20, 40, 60, and 80, Span 20, monoglyceride, and sucrose polyesters SE-11 and SE-15, were purchased from the National Chemical Co. (Shanghai, China). Analytical grade hexane and anhydrous ether were from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). Distilled water was used for the preparation of all solutions. All other chemicals used in this study were of analytical grade.

**Preparation of Emulsions.** Chitosan (wall material) solution was prepared by adding chitosan to 1% (w/w) aqueous acetic acid and stirring in a 45 °C water bath until completely dissolved. The core material, orange oil containing 3, 5, 7, or 10% surfactant by weight (in oil), was progressively added to the continuous phase (chitosan solution) during pre-emulsion preparation, and stirred for 1 min at 12500 rpm (T25, IKA Labortechnik, Germany). The surfactants used for the pre-emulsion preparations were Tween 20, 40, 60, or 80 as single emulsifier and Tween 40 with Span 20, monoglyceride, or sucrose polyesters SE-11 and SE-15 as compound emulsifiers at the ratio of 1:1, 7:3, 8:2, or 9:1. The coarse emulsions were then further homogenized at 40 MPa (APV-1000, SPX Corp., Charlotte, NC, USA) for two cycles. Thermal treatment was carried out by boiling the emulsions for 3 min and then incubating in a water bath for 30 min at 60 °C before cooling in an ice water bath.<sup>26,27</sup>

**Spray-Drying.** A high-speed centrifugal spray-dryer (model QZ-5, Lizhou Co., Wuxi, China) was used to convert the emulsions into encapsulated powders, as described by Jafari.<sup>22</sup> The operational conditions were as follows: air inlet temperature of 130, 150, or 170 °C; air outlet temperature of 80 °C; and sampling rate of material at 1.2 L/h. The dry powder was collected and stored in hermetically sealed packages in a desiccator at room temperature until analysis.

**Emulsion Droplet Size and Powder Particle Size Analysis.** Laser light scattering with the Mastersizer 2000 (Malvern Instruments, Worcestershire, UK) was used to determine the size distribution of oil droplets in the emulsions. Droplet size of the emulsion before and after thermal treatment was determined in triplicate. Analyses of powder particle size and specific surface area of the powder particles were performed in triplicate using the laser light scattering method by an analyzer with a batch cell unit (Mastersizer 2000). Encapsulated powders were dispersed in water for all of the analyses. In all cases, volume mean diameter or  $d_{43}$  was reported as the emulsion size or powder particle size.

**Viscosity Measurement.** The viscosity of emulsions and chitosan solutions (chitosan in aqueous acetic acid) was measured at 25 °C over a shear rate range of 4–400 s<sup>-1</sup> using a dynamic rheometer (AR-1000, TA Co., Elstree, UK) equipped with flat-plate geometry (CP 40). All measurements were performed in triplicate within 24 h after the emulsions were prepared.

**Scanning Electron Microscopy (SEM) Measurement.** Scanning electron micrographs of spray-dried samples were recorded with a Quanta-200 scanning electron microscope (FEI Co., Eindhoven, The Netherlands) at an accelerating voltage of 5 kV.<sup>28</sup> The powders were sprinkled onto double-backed cellophane tape attached to an aluminum stub before coating with gold–palladium in an argon atmosphere.

**Encapsulation Efficiency and Retention Determination.** The encapsulation efficiency (EE) during the process was determined as the amount of core material encapsulated inside the powder particles, which can be obtained from the following formula:

$$EE (\%) = \frac{\text{total oil content} - \text{surface oil content}}{\text{total oil content}} \times 100\%$$

Total oil is the internal and surface oil content of the powder, whereas surface oil is only the unencapsulated oil at the surface of the particles. The surface oil, or free or extractable oil, is determined by solvent extraction.

Due to the loss of volatiles during microcapsule preparation, the term “retention” has been proposed, which was calculated as

$$\text{retention} (\%) = \frac{\text{total oil content}}{\text{original oil content}} \times 100\%$$

**Total Oil Measurement.** Total oil was determined by solvent extraction.<sup>29</sup> Spray-dried powder (0.1 g) was combined with 10 g of 1% acetic acid in a 50 mL centrifuge tube and stirred until the powder was completely dissolved. The orange oil released from the microcapsules was extracted with 50 mL of *n*-hexane for 10 min three times. The extracts were collected in a 25 mL volumetric flask. The content of orange oil in the extract was then determined by measuring the absorbance at 202 nm (the maximum absorbance for orange oil in *n*-hexane by full wave scanning) in triplicate by a UV spectrophotometer (2802H, Unic Instruments Co., Ltd., Shanghai, China).

**Surface Oil Measurement.** Spray-dried microcapsules (0.1 g each) were suspended in a known volume of anhydrous ether and extracted for 10 min three times. The total extracts were collected by centrifugation. The content of orange oil in the extract was determined by measuring the absorbance at 219 nm (the maximum absorbance for orange oil in anhydrous ether by full wave scanning) in triplicate.<sup>29</sup>

**Storage Stability.** Spray-dried microcapsules (3 g each) or free orange oil were placed in open glass beakers, and the beakers were placed in desiccators with relative humidity (RH) of 33 or 52% maintained by saturated MgCl<sub>2</sub> or Mg(NO<sub>3</sub>)<sub>2</sub> solutions, respectively. The desiccators were then incubated at 25 and 45 °C. For the free

**Table 1. Droplet Size of Emulsions Formed with Different Emulsifiers before and after Heating<sup>a</sup>**

single emulsifier 10% (w/w) in oil	size of original emulsion ( $d_{43}$ , $\mu\text{m}$ )	size of heated emulsion ( $d_{43}$ , $\mu\text{m}$ )	$\Delta\mu^b$ ( $d_{43}$ , $\mu\text{m}$ )	compound emulsifiers (10% in oil, ratio 1:1)	size of original emulsion ( $d_{43}$ , $\mu\text{m}$ )	size of heated emulsion ( $d_{43}$ , $\mu\text{m}$ )	$\Delta\mu^b$ ( $d_{43}$ , $\mu\text{m}$ )
Tween 20	0.410 ± 0.009	2.277 ± 0.020	1.867 ± 0.030	Tween 40 + Span 20	0.178 ± 0.015	0.218 ± 0.005	0.040 ± 0.020
Tween 40	0.196 ± 0.003	0.988 ± 0.011	0.792 ± 0.015	Tween 40 + Span 40	0.244 ± 0.009	0.381 ± 0.008	0.137 ± 0.015
Tween 60	0.794 ± 0.015	2.106 ± 0.089	1.312 ± 0.020	Tween 40 + Span 60	0.185 ± 0.008	0.304 ± 0.009	0.119 ± 0.015
Tween 80	0.261 ± 0.019	2.542 ± 0.075	2.281 ± 0.020	Tween 40 + Span 80	0.198 ± 0.011	0.274 ± 0.013	0.076 ± 0.020
				Tween 40 + monoglyceride	0.211 ± 0.021	0.781 ± 0.025	0.570 ± 0.045
				Tween 40 + SE-11	0.196 ± 0.025	0.325 ± 0.036	0.129 ± 0.055
				Tween 40 + SE-15	0.191 ± 0.019	0.234 ± 0.031	0.043 ± 0.050

<sup>a</sup>Values are the mean of triplicate measurements. Core material was orange oil, and 1 wt % chitosan in 1% acetic acid solution was wall material. The weight ratio of oil to chitosan was 1:1. <sup>b</sup> $\Delta\mu$  = size of heated emulsion – size of original emulsion.

**Table 2. Droplet Size of Emulsions Formed with Compound Emulsifiers before and after Heating<sup>a</sup>**

ratio of compound emulsifier <sup>c</sup>	size of original emulsion ( $d_{43}$ , $\mu\text{m}$ )	size of heated emulsion ( $d_{43}$ , $\mu\text{m}$ )	$\Delta\mu^b$ ( $d_{43}$ , $\mu\text{m}$ )	ratio of compound emulsifier <sup>c</sup>	size of original emulsion ( $d_{43}$ , $\mu\text{m}$ )	size of heated emulsion ( $d_{43}$ , $\mu\text{m}$ )	$\Delta\mu^b$ ( $d_{43}$ , $\mu\text{m}$ )
Tween 40/Span 20				Tween 40/SE-15			
1:1	0.178 ± 0.015	0.218 ± 0.005	0.040 ± 0.020	1:1	0.191 ± 0.005	0.234 ± 0.010	0.043 ± 0.015
7:3	0.211 ± 0.001	0.220 ± 0.002	0.009 ± 0.003	7:3	0.194 ± 0.009	0.240 ± 0.010	0.046 ± 0.020
8:2	0.224 ± 0.001	0.225 ± 0.001	0.001 ± 0.002	8:2	0.205 ± 0.006	0.225 ± 0.007	0.020 ± 0.015
9:1	0.200 ± 0.003	0.216 ± 0.005	0.016 ± 0.008	9:1	0.229 ± 0.010	0.253 ± 0.015	0.024 ± 0.025

<sup>a</sup>Values are the mean of triplicate measurements. Core material was orange oil, and 1% chitosan in 1% acetic acid solution was wall material. The weight ratio of oil to chitosan was 1:1. <sup>b</sup> $\Delta\mu$  = size of heated emulsion – size of original emulsion. <sup>c</sup>Compound emulsifier was 10% in oil.

orange oils, samples were withdrawn every 2 h and analyzed using ultraviolet spectrophotometry at 202 nm. For the microencapsulated orange oils, samples were withdrawn every 2 days and completely dissolved in 1% aqueous acetic acid. Then the orange oil was extracted with *n*-hexane and analyzed using ultraviolet spectrophotometry at 202 nm. The retention of orange oil was defined as the ratio of the amount of orange oil remaining to the amount at the start. The analysis was carried out in triplicate.

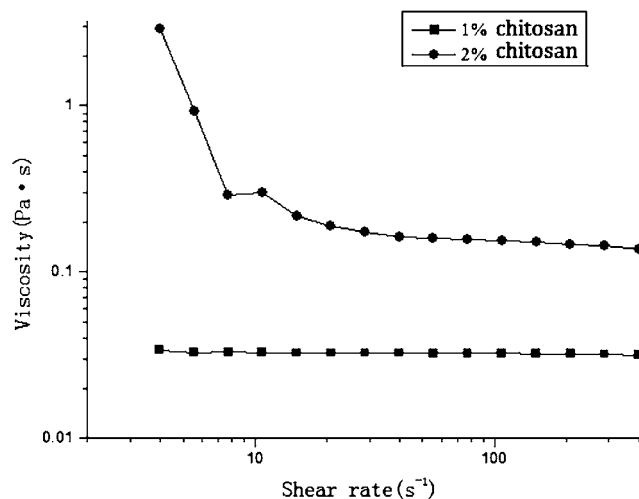
**Retention of Orange Oil in Fabrics.** Orange oil or microencapsulated products (1 g each) were added to 800 g of water containing 8 g of a common household laundry detergent. Small swatches, 3 cm × 3 cm, of cotton fabric or terylene fabric were placed into the orange oil or microencapsulated oil laundry detergent solutions, stirred for 2 h, and rinsed in water three times (10 min each). The washed fabrics were air-dried at room temperature for 12 h. After drying, the fabrics were stored at room temperature and taken out for oil measurement every day. The fabrics were immersed in 1% (w/w) acetic acid and stirred for 2 h. The orange oil was then exhaustively extracted with *n*-hexane. The oil content was determined using UV spectrophotometry at 202 nm. The retention of orange oil was defined as the ratio of the amount of orange oil remaining after washing during storage to the amount before washing. The analysis was carried out in triplicate.

**Statistical Analysis.** All of the data were analyzed by SPSS 13.0 (SPSS Institute Inc., Cary, NC, USA) and expressed as the mean ± standard deviation (SD). The calculations and graphs were made using the computer program Microcal Origin 7.5 (Microcal Software Inc., Northampton, MA, USA).

## RESULTS AND DISCUSSION

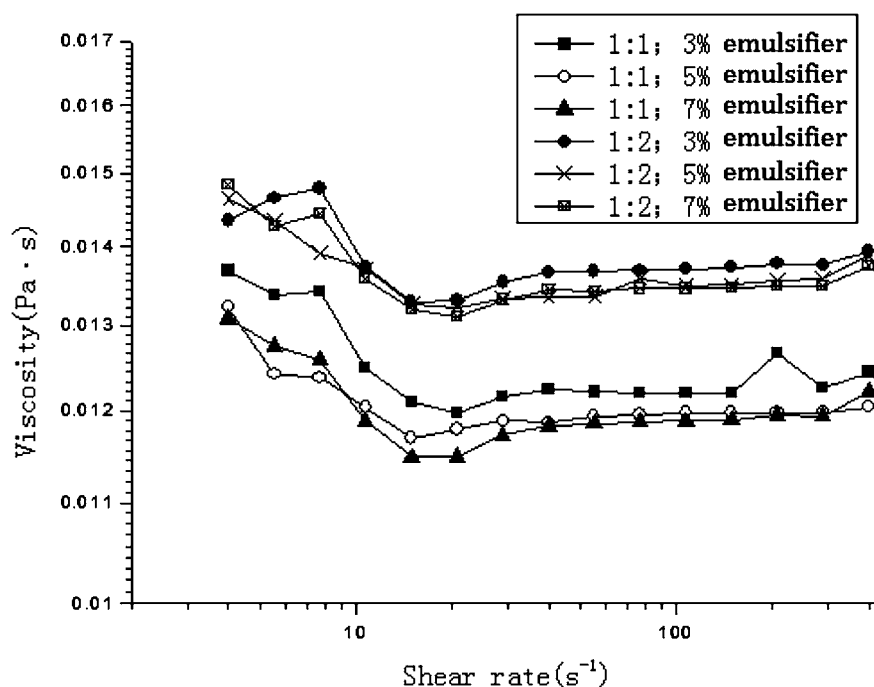
### Effect of Emulsifiers on Emulsion Size and Stability.

The chitosan molecules have both hydrophobic and hydrophilic groups, and it is possible that they may exist in the interface of the emulsion by acting as coemulsifiers. The emulsifying properties of Tween and Span on the size and stability of emulsions at conventional temperatures are well-known, but the effects of high temperatures have not been well studied. Because the emulsions would be subject to thermal processes during spray-drying, their stability to high temperatures were



**Figure 1.** Flow curves of 1 and 2% (w/w) chitosan in 1% acetic acid solution.

examined in this study. We found that there was an increase in emulsion droplet volume after heating briefly at 100 °C and holding at 60 °C (Table 1). The droplet diameter of all emulsions prepared with single emulsifiers increased by nearly 2  $\mu\text{m}$  after heating except for Tween 40. This is presumably because the molecular geometry of surfactants changed differently in response to raised temperature. It is now widely recognized that the molecular geometry of a surfactant plays a major role in determining its functional performance in colloidal systems.<sup>30</sup> The molecular geometry is characterized by a packing parameter ( $p$ ), which is the ratio between the effective cross-sectional areas of the tail group and the head group of the surfactant. The packing parameter determines the optimum curvature of a monolayer that a particular surfactant tends to form. The optimum curvature influences parameters such as the interfacial tension and rheology, which may then



**Figure 2.** Flow curves of emulsions with 1:1 and 1:2 ratios of orange oil to chitosan at 3, 5, and 7% emulsifier concentrations. (Core material was orange oil, and 1% chitosan in acetic acid solution was wall material. The emulsifiers were Tween 40 and Span 20 with ratio of 4:1.)

**Table 3.** Influence of Inlet Temperature on Encapsulation Efficiency and Retention of Orange Oil<sup>a</sup>

emulsifier (10% in oil)	inlet temperature (°C) (outlet temperature was 80 °C)	size of original emulsion ( $d_{43}$ , $\mu\text{m}$ )	size of heated emulsion ( $d_{43}$ , $\mu\text{m}$ )	$\Delta\mu^b$ ( $d_{43}$ , $\mu\text{m}$ )	encapsulation efficiency (%)	retention (%)
Tween 40	130	0.410 ± 0.010	2.277 ± 0.190	1.867 ± 0.2	88.1 ± 0.04	5.9 ± 0.04
Tween 40/Span 20 = 4:1	130	0.224 ± 0.001	0.225 ± 0.001	0.001 ± 0.002	91.5 ± 0.02	56.4 ± 0.06
Tween 40/Span 20 = 4:1	150	0.224 ± 0.001	0.225 ± 0.001	0.001 ± 0.001	90.7 ± 0.03	45.7 ± 0.03
Tween 40/Span 20 = 4:1	170	0.224 ± 0.001	0.225 ± 0.001	0.001 ± 0.002	89.3 ± 0.03	43.9 ± 0.03

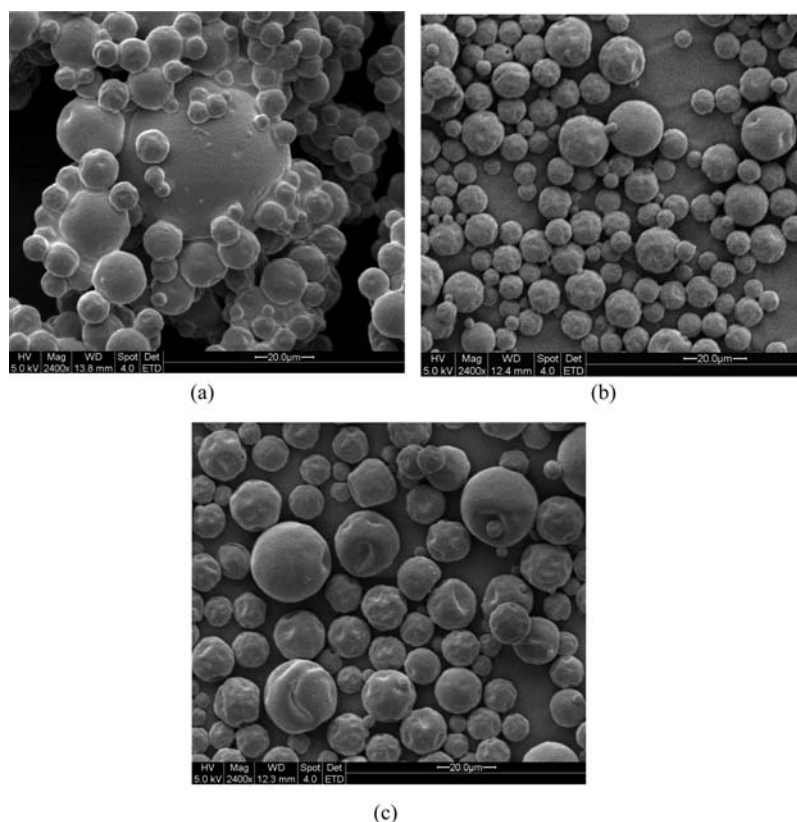
<sup>a</sup>Values are the mean of triplicate measurements. Core material was orange oil, and 1% chitosan in 1% acetic acid solution was wall material. The weight ratio of oil to chitosan was 1:1. <sup>b</sup> $\Delta\mu$  = size of heated emulsion – size of original emulsion.

affect the stabilities of emulsions formed. As the temperature rises, the headgroup becomes progressively dehydrated, resulting in altered packing parameter. The single surfactants used in this study (Tween 20, 40, 60, and 80) all have similar polar head groups, but have different nonpolar tail groups. It was hypothesized that the packing parameter changed differently during heating because of the various tail groups of these surfactants, which may be responsible for the different heating stabilities of the emulsions. The emulsions prepared with compound emulsifiers were found to be more stable to heat treatment, especially for the two combinations, Tween 40 with Span 20 and Tween 40 with SE-15 (Table 1). Because chitosan has little emulsifying properties, the nonionic surfactants are responsible for the emulsification of orange oil in chitosan solution to form O/W emulsion. Others have also found that the emulsifying ability of a single emulsifier is usually inferior to that of compound emulsifiers.<sup>31</sup> Two different surfactants in combination can not only emulsify water and oil more effectively but also interact to form more stable emulsions that prevent the droplets from agglomeration or flocculation.

The combinations of Tween 40 with Span 20 and Tween 40 with SE-15 gave the lowest  $\Delta\mu$  values (Table 1), so these two best combinations were chosen for further investigation of the

effects of weight ratio of the compound emulsifiers to chitosan wall material and thermal treatment on droplet size. As from Table 2, the combination of Tween 40 with Span 20 was superior to Tween 40 with SE-15 (value of  $\Delta\mu$  was small overall), although all combinations of Tween 40 with either Span 20 or SE-15 resulted in small particles after heating. SE-15 may not be as good an emulsifying agent in this application because it is a solid, and its solubility in water may not be as good as that of liquid emulsifier Span 20 or the stronger interactions between SE-15 molecules that make it a solid do not allow it to interact as much with the oil core as Span 20. The droplet sizes of all the ratios of Tween 40 to Span 20 were relatively similar after heating. Although the cold emulsion prepared with 80% Tween 40 and 20% Span 20 had the relatively largest droplet size, the droplets were stable to heat treatment ( $\Delta\mu = 0.001$ ). Thus, 80% Tween 40 and 20% Span 20 was the best combination for the preparation of small, stable emulsions in our experiments.

**Emulsion Viscosity Determination.** Viscosity is an important property of fluids in processing because it can affect the selection of equipment and the processing conditions. A lower viscosity is desirable for spray-drying. At the concentrations of chitosan we used as the continuous phase of our



**Figure 3.** SEM photograph of encapsulated powders produced from emulsions by different spray-drying conditions ( $\times 2400$ ): (a) inlet temperature =  $130\text{ }^{\circ}\text{C}$ ; (b) inlet temperature =  $150\text{ }^{\circ}\text{C}$ ; (c) inlet temperature =  $170\text{ }^{\circ}\text{C}$ . Wall material was 1% chitosan in 1% acetic acid solution. The weight ratio of oil (core material) to chitosan was 1:1. The emulsifiers (10% in oil) were Tween 40 and Span 20 with a ratio of 4:1.

**Table 4.** Influence of Different Concentrations of Compound Emulsifiers and the Weight Ratios of Core Material to Wall Material on Encapsulation Efficiency and Retention of Orange Oil<sup>a</sup>

compound emulsifier concentration (wt % in oil)	wt ratio of orange oil to chitosan (w/w)	size of original emulsion ( $d_{43}$ , $\mu\text{m}$ )	size of heated emulsion ( $d_{43}$ , $\mu\text{m}$ )	$\Delta\mu^b$ ( $d_{43}$ , $\mu\text{m}$ )	size of powder particle ( $d_{43}$ , $\mu\text{m}$ )	X (powder size – original emulsion size)/powder size	encapsulation efficiency (%)	retention (%)
3	1:1	$0.573 \pm 0.003$	$0.670 \pm 0.009$	$0.097 \pm 0.012$	$20.981 \pm 0.020$	$0.973 \pm 0.020$	$85.5 \pm 0.02$	$41.7 \pm 0.03$
5	1:1	$0.298 \pm 0.002$	$0.387 \pm 0.008$	$0.089 \pm 0.010$	$16.857 \pm 0.015$	$0.982 \pm 0.015$	$80.5 \pm 0.03$	$52.5 \pm 0.02$
7	1:1	$0.243 \pm 0.002$	$0.273 \pm 0.010$	$0.03 \pm 0.010$	$17.132 \pm 0.010$	$0.986 \pm 0.01$	$80.5 \pm 0.02$	$72 \pm 0.03$
3	1:2	$0.476 \pm 0.003$	$0.499 \pm 0.009$	$0.023 \pm 0.012$	$24.522 \pm 0.019$	$0.980 \pm 0.02$	$93.7 \pm 0.02$	$80.3 \pm 0.02$
5	1:2	$0.332 \pm 0.001$	$0.334 \pm 0.007$	$0.002 \pm 0.007$	$22.610 \pm 0.010$	$0.985 \pm 0.01$	$92.9 \pm 0.02$	$86.8 \pm 0.02$
7	1:2	$0.214 \pm 0.001$	$0.229 \pm 0.007$	$0.015 \pm 0.007$	$21.091 \pm 0.010$	$0.910 \pm 0.01$	$90.1 \pm 0.01$	$86.9 \pm 0.01$

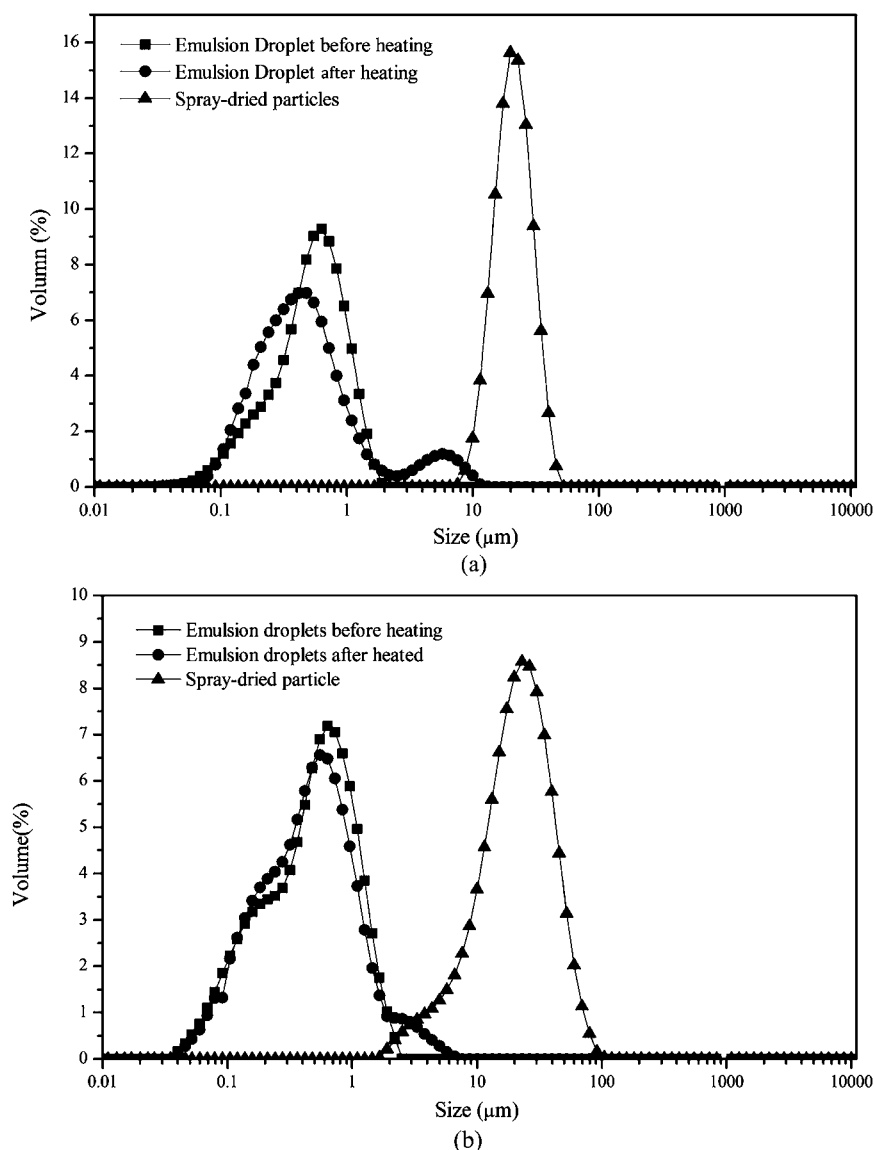
<sup>a</sup>Values are the mean of triplicate measurements. Core material was orange oil, and 1% chitosan in 1% acetic acid solution was wall material. The weight ratio of oil to chitosan was 1:1 or 1:2. Tween 40 and Span 20 were used as compound emulsifier with the ratio of 4:1. Spray-drying inlet temperature was  $150\text{ }^{\circ}\text{C}$ , and outlet temperature was  $80\text{ }^{\circ}\text{C}$ . <sup>b</sup> $\Delta\mu$  = size of heated emulsion – size of original emulsion.

emulsified mixtures, the flow curves of chitosan in acetic acid solution showed that at 1% (w/w) concentration, chitosan behaved like a Newtonian fluid, but at 2%, it exhibited shear-thinning behavior (Figure 1). Interestingly, the viscosity of the emulsion in 1% chitosan was lower than 1% chitosan alone (Figures 1 and 2) suggesting that the emulsion droplets may interact with chitosan effectively lowering the concentration in the aqueous phase. The viscosity was increased when the ratio of oil to chitosan was decreased from 1:1 to 1:2. The increasing concentration of chitosan in the aqueous phase would be responsible for the raised viscosity.

Emulsification of orange oil by surfactants in 1% chitosan resulted in shear-thinning behavior that was not observed in 1% chitosan alone, suggesting that the emulsified droplets may

form aggregates and these associations were broken down by shear (Figure 2). The emulsion viscosity also decreased with increasing concentration of emulsifier in oil. The viscosity reached a minimum when 5 or 7% surfactant in oil was incorporated. All viscosity curves had the same shear-thinning behavior. The shear -thinning behavior is explained by the increased shear rate overcoming Brownian motion, resulting in the emulsion droplets becoming more ordered along the flow field and offering less resistance to flow and hence the lower viscosity.<sup>32</sup> The viscosity leveled out with further increases in shear rate above  $10\text{ s}^{-1}$ .

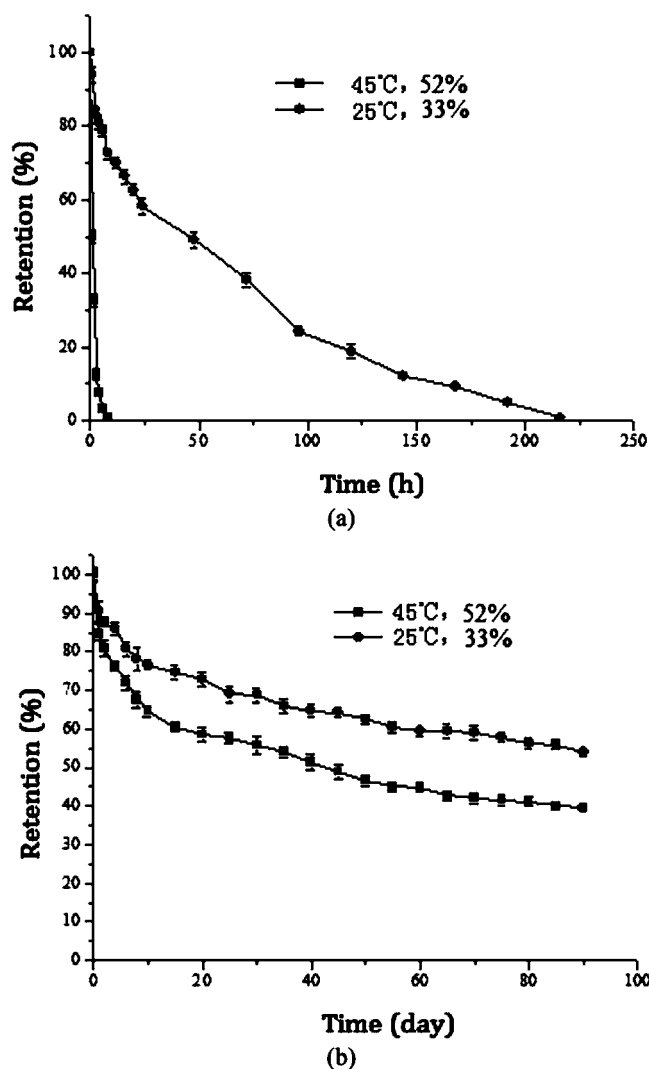
**Effect of Spray-Drying Conditions and Emulsifiers on Microencapsulation.** A high amount of encapsulated oil is desirable, and retention is one of the most important



**Figure 4.** Emulsion droplet size distribution before and after thermal treatment as well as the microencapsulated powder size after spray-drying: (a) weight ratio of oil and chitosan was 1:1; (b) weight ratio of oil and chitosan was 1:2. The emulsifiers (3% in oil) were Tween 40 and Span 20 with a ratio of 4:1. Thermal treatment was carried out by boiling the emulsions for 3 min and then incubating in a water bath for 30 min at 60 °C.

parameters of spray-dried microencapsulated powders. The use of compound surfactants did not improve encapsulation efficiency (EE) significantly, but improved oil retention almost 10-fold and resulted in smaller droplet size compared to the use of a single surfactant (Table 3). Emulsions with little change of the droplet size after thermal treatment also produced spray-dried product with higher EE and retention. As expected, the EE value was reduced when the inlet temperature increased (Table 3). However, low inlet temperatures (130 °C) resulted in insufficient water volatility and adhesion of the particles (Figure 3a). Too high inlet temperatures (170 °C) resulted in more drapes and microgaps at the surface of the microencapsulated particles (Figure 3c). Total retention of orange oil in spray-dried powders decreased from 56.4 to 43.9% when the inlet temperature was increased from 130 to 170 °C (Table 3). Thus, 150 °C was selected as the optimal inlet temperature, at which the microcapsules had regular particle morphology (Figure 3b), although it had slightly lower retention and encapsulation efficiency than at 130 °C.

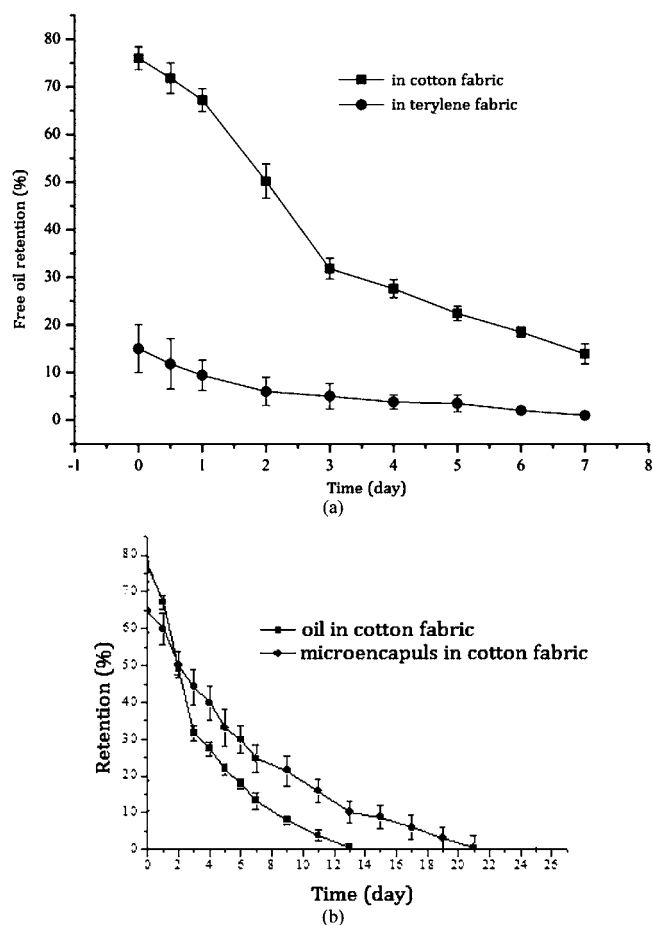
The retention of orange oil was found to be negatively correlated to emulsion size when the weight ratio of orange oil to chitosan was kept constant (Table 4). Because higher retention was desirable, this suggested that a fine emulsion was advantageous and could be stable during both atomization and spray-drying. In a previous study it was observed that decreasing surface oil is related to the difference between emulsion size and particle size of the spray-dried microcapsules. Re and Liu defined a parameter  $X = (\text{powder size} - \text{emulsion size}) / \text{powder size}$ , to relate retention to powder and emulsion size.<sup>33</sup> They showed that by increasing  $X$  from 0.2 to 0.8, retention of volatiles increased significantly, so it could be possible to improve the retention by increasing the difference between emulsion and powder particle size. Although  $X$  was greater than 0.8 in this study (Table 4), the results were consistent with the previous paper. It was also found that EE was positively correlated to the powder particle size. The larger powder particle size of microcapsules was related to higher values of EE. The explanation is that emulsion droplets are



**Figure 5.** Retention of orange oil at 25 °C and 33% humidity or at 45 °C and 52% humidity: (a) free orange oil; (b) microencapsulated orange oil. The weight ratio of oil to chitosan was 1:2. The emulsifiers (3% in oil) were Tween 40 and Span 20 with a ratio of 4:1.

more likely to adhere to the surface of smaller particles than to the surface of larger ones. In other words, fine oil droplets will be covered by the wall material more efficiently in bigger particles than in smaller ones. The ratio of wall material to core material also had an effect on the spray-dried particle size, EE, and the total oil retention (Table 4). The EE was >90% at the ratio of orange oil to chitosan of 1:2. Increasing the wall material weight resulted in more encapsulated oil but increasing powder particle size. The retention of orange oil was also markedly increased when the ratio of orange oil to chitosan decreased from 1:1 to 1:2. The optimum ratio of wall materials to oil and the emulsion droplet size were found to be the most important for the retention of orange oil. This observation is in agreement with Risch and Reineccius, who reported in a similar study that smaller emulsion size yields a higher retention of orange oil.<sup>24</sup>

Emulsion droplet size distributions before and after thermal treatment as well as microencapsulated powder size distributions were determined. A new peak about 6  $\mu\text{m}$  in diameter after heating was observed for the emulsion with the oil to chitosan ratio of 1:1 (Figure 4a), which was interpreted as the



**Figure 6.** Retentions of free orange oil and microencapsulated oil in fabrics: (a) free orange oil retention in cotton and terylene polyester after washing; (b) free orange oil and microencapsulated oil retention in cotton after washing. The weight ratio of oil to chitosan was 1:2. The emulsifiers (3% in oil) were Tween 40 and Span 20 with a ratio of 4:1.

aggregation of emulsion droplets. The emulsion size of oil to chitosan ratio at 1:2 had similar diameters after heating (Figure 4b) and had higher stability to spray-drying and higher retention and EE (Table 4). This confirmed the observation that a higher proportion of wall material components in emulsion resulted in particles that were more stable during thermal treatment and spray-drying. However, with the increased proportion of wall material, the distribution range of powder particle size after spray-drying was expanded and the mean particle size was increased. The microcapsules had a mean diameter below 20  $\mu\text{m}$  (Figure 4). It was also shown that emulsions with less stability resulted in encapsulated powders with higher surface oil content and lower retention of volatiles (Table 4), which was consistent with previous studies.<sup>34–37</sup>

**Storage Stability of Orange Oil Microcapsules.** The storage stability of orange oil by encapsulation in chitosan was markedly improved compared to free orange oil. Free orange oil was extremely volatile and was almost completely volatilized in 8 h at 45 °C and 52% relative humidity as seen in Figure 5a. Orange oil encapsulated in chitosan under the same conditions volatilized over a period of months (Figure 5b). Temperature and relative humidity also had a significant effect on volatility. Retention was lower when microcapsules were stored at 45 °C and 52% humidity than when stored at 25 °C and 33%

humidity. Higher temperature caused the volatilization of oil. Another possible explanation is that at higher humidity the chitosan wall material may absorb water and swell; the swelling may create stresses resulting in cracks at the surface of the microcapsule, allowing the oil to volatilize through the cracks.

**Retention of Orange Oil in Fabrics.** Free oil retention in two kinds of fabrics, cotton and terylene polyester, after washing was determined (Figure 6a). The oil retention in cotton fabric decreased gradually to 20% in 1 week. The terylene fabric retained only 15% of the oil initially, and the oil was essentially gone by the fifth day. Cotton fabric has a coarse surface and interspaces that are capable of absorbing oil droplets or adsorbing microcapsules applied with detergent. Terylene fabric has a smoother surface and more compact fibers so that fewer oil or microencapsulated particles would be adsorbed. As seen from Figure 6b, the original retention for microencapsulated orange oil (about 65%) was lower than that for free orange oil (about 75%) in cotton fabric, which was probably due to the larger particle size for the microcapsules. Small particles are desirable because they fit into more of the interspaces of the fabric and can adsorb onto it. However, the retention of free oil decreased rapidly in the first few days, so that only 30% of the oil was retained by the third day. The wall material chitosan is water insoluble (when pH >6) and protects the core oil during water washing and storage. The oil retention of microcapsules on cotton fabric decreased gradually to 0 in 21 days, which was 8 days longer than for unencapsulated orange oil (Figure 6b). These results confirmed that the microencapsulated orange oil was slowly released during storage, whereas free oil is volatilized in hours.

## AUTHOR INFORMATION

### Corresponding Author

\*Phone: 86-138-12536912. Fax: 86-510-85197876. E-mail: fzhong@jiangnan.edu.cn or yue\_li1@yahoo.com.cn.

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### Notes

The authors declare no competing financial interest.

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