STUDY OF SURFACTANT-CHITOSAN EFFECT ON EMULSION FORMATION

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Abstract

This work aims to study the formation of Surfactant Polyelectrolyte Complexes (SPECs) emulsion formed by mild and natural based anionic surfactant, Sodium Laurylglucosides Hydroxypropylsulfonate with cationic chitosan (CS). The suitable pH value for emulsion formation is at 5.00-5.70. The range of CS concentration to stabilize SPECs emulsion at 10%w/w jojoba oil, 0.5%w/w surfactant were between 0.5-0.7%w/w. The range of surfactant concentration to stabilize SPECs emulsion at 10%w/w jojoba oil, 0.5%w/w CS were between 0.2-1.5%w/w. The jojoba oil concentration that can be stabilized on SPECs emulsion at 0.5%w/w surfactant, 0.5%w/w CS were between 10-15%w/w. The NaCl electrolyte had an influence to deteriorate stabilization of emulsion system. In order to develop a stabilized emulsion, the use of mechanical apparatus such as high pressure homogenizer or ultrasonic homogenizer and the addition of stabilizer are recommended for future studies.

Keywords: Chitosan/Sodium Laurylglucosides Hydroxypropylsulfonate/
Surfactant Polyelectrolyte Complexes/Emulsion/Stability

Introduction

The new market trend for all-natural skin care products, by replacing synthetic ingredients with all natural or naturally derived ingredients is continuously increasing as the market trend respond quickly and favorably because consumers are better informed about the toxicity of chemicals used in food and cosmetic products (Łopaciuk & Łoboda, 2013). In the emulsion making process, it usually requires surfactants to decrease surface and interfacial tension between oil and water droplets in order to form emulsion; however, the synthetic surfactants may possibly cause irritation. In recent years many studies on Surfactant Polyelectrolyte Complexes (SPECs) of chitosan have been used to replace synthetic surfactant in emulsion making process due to chitosan which is a natural cationic polysaccharide has advantageous properties such as nontoxic, biodegradable. Chitosan (CS) is extensively utilized in various industries for instance in the water treatment, textile, food, and pharmaceutical industry. Chitosan works well with anionic surfactant to form multi-molecular film around inner phase of emulsion. It has been reported that the complex formation between sodium dodecyl sulfate (SDS) and chitosan help to stabilize emulsion (Onesippe & Lagerge, 2008) because the interaction of a strong electrostatic attraction force between polymer charges and opposite charges of surfactants were reinforced by relating the aggregation of bound surface molecules (Goddard, 1986). The emulsion was formed by complex of chitosan, a canonic polyelectrolyte interacting with anionic surfactant which was formed well below critical micelle concentration (CMC) (Desbrieres, Bousquet, & Babak, 2010). However, SDS is a synthetic and highly irritating substance. It was reported that SDS is safe with discontinuous application and rinsing off from skin. The skin irritation would appear when used in concentration more than 1% and prolonged contact with skin ("Final Report on the Safety Assessment of Sodium Lauryl Sulfate and Ammonium Lauryl Sulfate," 1983) Thus, it is interesting to use the alternative mild surfactant that can form complex and help stabilize emulsion. The current commercial surfactant, Sodium Laurylglucosides Hydroxypropylsulfonate (Suga®Nate NC160), is an anionic surfactant, naturally-derived, biodegradable with very low toxicity used in formulating shampoo, facial cleanser and baby cleaning products. It was proven as an extremely mild surfactant. It was chosen for this study due to its chemical structure which is partially similar to SDS. It might be able to form complex with chitosan and help stabilize emulsion. Jojoba oil as a part of emulsion system was chosen because it has excellent properties to help nourish the skin, prevent dryness and it is not easily oxidized. Hence, this study aims to use chitosan and Sodium Laurylglucosides Hydroxypropylsulfonate to produce stabilized SPECs emulsion by investigating ingredient concentrations that affect emulsion formation and the factors influence stability of SPECs emulsion are evaluated.

Materials and Methods

Materials

Chitosan 1,500 K dalton was obtained from Marine Bio Resources Co., Ltd., Thailand. Sodium Laurylglucosides Hydroxypropylsulfonate (Suga®Nate 160NC) was purchased from Colonial Chemical, Inc., USA. Jojoba oil was received from Desert Whale Jojoba Company, USA. Lactic acid was received from Purac Ltd., Thailand. Sodium hydroxide was received from TRS Thai Refined Salt Co.,Ltd., Thailand and all other chemicals used were cosmetic grade.

Methods

1. Stock Solution and Complex Preparation

Stock solutions of chitosan were prepared by dissolving an appropriate concentration of chitosan into lactic acid solution (1%w/w) under stirring until all chitosan was dissolved. The pH was adjusted to desired value (pH 5.00 – 5.50) by slowly adding NaOH solution (10%w/w) while continuously stirred until all NaOH solution was dispersed into chitosan solution. Sodium Laurylglucosides Hydroxypropylsulfonate (Suga®Nate 160NC) was added at appropriate concentration into solution under stirring in order to obtain surfactant-chitosan complex. Lastly, preservative (0.2%w/w) was added into complex to prevent microbial growth.

2. Preparation of Emulsion

Oil-in-water emulsions were prepared by mixing jojoba oil at appropriate concentration into SPECs by using high speed homogenizer at 5,000 rpm for 8 min. The effect of chitosan concentration was studied by preparing emulsion with the conditions of 0.5%w/w surfactant, 10%w/w oil and 0-0.9%w/w chitosan. The effect of surfactant concentration was studied by preparing emulsion with the conditions of

0.5%w/w CS, 10%w/w oil and 0-2%w/w surfactant. The effect of oil concentration was studied by preparing emulsion with the conditions of 0.5%w/w CS, 0.5%w/w surfactant and 0-25%w/w oil. The effect of pH value was studied by preparing emulsion with the conditions of 0.5%w/w CS, 0.5%w/w surfactant, 10%w/w oil and pH range of 3.75-6.50. The effect of NaCl was studied by preparing emulsion with the conditions of 0.7%w/w CS, 0.7%w/w surfactant, 10%w/w oil and 0-5%w/w NaCl.

3. Evaluation of the Emulsion

The physical appearances of emulsions such as color, texture, were visually observed. The pH values of emulsion were controlled and measured by digital pH meter with 2 decimal precision. The viscosities were differentiated by scale number to range viscosities which were done by observation. The higher number means higher viscosity. The percentage of creaming layer were calculated by measurement of height of cream layer dividing by total height of emulsion in test tube and multiplying by 100 to obtain the percentage of creaming layer. Usually, the higher percentage of creaming layer implies that the emulsion has more stabilization of emulsion. The levels of emulsion stability in this study were determined by considering on 3 factors; the time of phase separation, %creaming layer and turbidity of lower part of emulsion.

4. Microscopic Observations

Optical micrograph of emulsions were captured at magnify 10x, 40x and 100x on by using microscope MOTIC BA300, China. The size distributions were observed. The dispersed phase and continuous phase were identified.

Results and Discussion

1. Preparation of Emulsion

There are several techniques and advance equipment such as high pressure homogenizer and ultrasonic homogenizer which can be used in preparation of SPECs emulsion. The emulsion preparation procedure of this study was adapted from Zinoviadou, Scholten, Moschakis, and Biliaderis (2012) which used conventional mixing process by high speed homogenizer. The chitosan (CS) has a strong hygroscopic property, thus it was prepared by dissolving the required amount in 1% lactic acid to obtain stock solution. The maximum concentration of CS that will give the preparable solution is 1.5%w/w because polymers absorbed all aqueous and became very viscous gel. The NaOH (10%w/w) was used to adjust pH of CS solution to 5.00-5.50. It must be slowly adjusted to desired pH values. Complexes were formed when polycationic CS solution was mixed with anionic surfactant (Suga®Nate 160NC). The emulsion of SPECs could be formed by homogenizing without heating.

2. Effect of Chitosan Concentration

The effect of CS concentration from 0-0.9%w/w on emulsion formation was studied. The oil concentration was fixed at 10%w/w and the surfactant concentration was fixed at 0.5%w/w. The final pH value of emulsion was 5.00. The order of phase separation time was observed. It was found that the formulation without CS was rapidly separated at 1 hour after formulating. When CS concentrations were increased from 0.1 to 0.5%w/w, the phase separation times were delayed from 1 day to 2 days. Once CS concentrations were more than 0.5%.w/w, the phase separation times became faster again to 1 day and 3 hours for CS concentration at 0.7% and 0.9%w/w, respectively. In addition, when the concentration of CS increased, the viscosities of emulsions were higher accordingly. It was also found that when the CS concentration increased, the emulsion became thicker (Table 1). The thickness of creaming layer increased when CS concentration increased. These results were consistent with research reported by Mun, Decker and McClements (2006). The emulsion without CS was formed and stable for only short period then transparent layer at bottom appeared. The emulsion textures were homogenous when CS concentrations increase from 0.3-0.7%w/w. Then the texture became non-homogenous when concentration reached 0.9%w/w. The percentage of creaming layer dictated the stability of emulsion, the higher number represented higher stability. However, the migration mechanism of emulsion by gravity occurred at different rate on each formulation, thus slower separation phase would imply higher stability as well. These 2 criteria tell us that either 0.5 %w/w or 0.7%w/w of CS emulsion could provide more stable system, Table 1.

Table 1 Properties of emulsion with different concentration of chitosan

Physical properties	Chitosan concentration (%w/w)					
	0.0	0.1	0.3	0.5	0.7	0.9
Appearance and texture	White, transparent, very thin	White, translucent, thin	White, lotion, fine emulsion	White, lotion, fine emulsion	Milky white, thick	Milky white, coarse and jelly
Viscosity[a]	0	1	2	2	3	4
First separation time	1 hr	1 day	2 days	2 days	1 day	3 hr
Creaming layer[b] (%)	18.3	14.4	14.4	19.2	23.1	30.8

Note. [a]Every formula has low viscosity. The higher number represents higher viscosity

[b]Determined at day 3

3. Effect of Surfactant Concentration

The effect of surfactant (Suga®Nate 160NC) concentration from 0 to 2%w/w on emulsion formation was studied. The oil concentration was fixed at 10%w/w and the CS concentration was fixed at 0.5%w/w. The final pH valve of emulsion was 5.00. The Suga®Nate 160NC concentration had slight effect to emulsion texture and viscosity. As Suga®Nate 160NC concentration increased, the emulsions texture become a little thicker and the viscosities were also higher accordingly. The percentage of creaming layer was highest without Suga®Nate 160NC. It was slightly thinner at the concentration of 0.2-1.5%w/w. At the concentration at 2%w/w, the percentage of creaming was in between the previous concentrations. The order of phase separation time was also observed. It was found that the formulation without Suga®Nate 160NC was fast separated at 3 hours after formulating (Table 2). When Suga®Nate 160NC concentrations were increased to 0.2%w/w, the phase separation time was delayed to 2 days. When Suga®Nate 160NC concentration increased to 0.5%w/w, the separation time became shorter to 1 day only. Once Suga®Nate 160NC concentrations were more than 0.5%.w/w, the separation times became delayed again at 1% and 1.5%. When Suga®Nate 160NC concentration was over 1.5%w/w, the first phase separation time was faster again to 1 day at 2%w/w. Zinoviadou et al.(2012) reported that the most stable system in his experiment occurred when molecular weight of CS and surfactant (sodium stearoyl lactylate) ratio was equal to 1.25 : 1. It implied that the proper amount of surfactant might fall between 0.2-0.5%w/w. However, it might not be comparable due to surfactant chemical structure difference.

Thus, longest time of first phase separation time occurred would interpret as the most stable formulations. Therefore, the stable emulsion could be able to prepare within the range of 0.2-1.5%w/w Suga®Nate 160NC in formulation, Table 2.

Table 2 Properties of emulsion with different concentration of surfactant

Physical properties	**Surfactant concentration (%w/w) (Chitosan : Surfactant ratio)**					
	0 (0.5 : 0)	**0.2 (2.5 : 1)**	**0.5 (1 : 1)**	**1.0 (0.5 : 1)**	**1.5 (0.33 : 1)**	**2.0 (0.25 : 1)**
Appearance and texture	White, lotion, fine emulsion, very thin	White, lotion, fine emulsion, thin	White, lotion, fine emulsion, thin	White, lotion, fine emulsion, thicker	White, lotion, fine emulsion, thicker	Milky white, lotion, fine emulsion, thickest
Viscosity[a]	1	2	2	3	3	3
First separation time	3 hr	2 days	1 day	2 days	2 day	1 day
Creaming layer[b] (%)	19.2	14.4	14.4	14.4	14.4	16.3

Note. [a]Every formula has low viscosity. The higher number represents higher viscosity

[b]Determined at day 4

4. Effect of Oil Concentration

Jojoba oil was selected for emulsion formation in this study due to its function as natural moisturizing factor. It closely resembles skin's natural oil and resistant to oxidation which is interesting for emulsion formulation of skin care product. The effect of jojoba oil concentration from 0 to 25%w/w on emulsion formation was studied. The CS concentration was fixed at 0.5%w/w and the surfactant concentration was fixed at 0.5%w/w. The final pH value of emulsion was 5.00. It was found that the formulation without oil looks translucent as emulsion was not formed. The order of phase separation time of emulsion was observed and it was found that the separation is at 1 day for system with 5 to 15%w/w oil. Moreover, the striped texture was noticeable at 3 hours after emulsion was prepared as a sign of flocculation for 15%w/w. Once oil concentrations were more than 15%.w/w, the phase separation times become faster to 1 hour only. Moreover, when the oil concentration was increased, the emulsion textures became thicker (Table 3) and the thickness of creaming layer increased when oil concentration increased as well. These means that the oil concentration had direct effect to the percentage of creaming layer. The results were the same for viscosity as oil concentration increased, the viscosity of emulsions also increased accordingly. The appearance of emulsion with 5%w/w oil was translucent and when concentrations increased from 5 to 25%w/w, it showed homogenous texture. Therefore, the stable emulsion could be prepared at jojoba oil between 10-15%w/w, Table 3.

Table 3 Properties of emulsion with different concentration of jojoba oil

Physical properties	**Oil concentration (%w/w)**					
	0	**5**	**10**	**15**	**20**	**25**
Appearance and texture	White, translucent, very thin	White, less translucent, thin	White, lotion, fine emulsion, thin	White, lotion, fine emulsion, thicker	Milky white, lotion, fine emulsion, thicker	Milky white, lotion, fine emulsion, thickest
Viscosity[a]	1	2	2	3	3	4
First separation time	N/A[c]	1 day	1 day	1 day	1 hr	1 hr
Creaming layer[b] (%)	N/A	6.7	17.3	27.9	45.2	43.3

Note. [a]Every formula has low viscosity. The higher number represents higher viscosity
[b]Determined at day 6
[c]Not applicable

5. Effect of pH of Emulsion

The effect of pH values from 3.75 to 6.51 on emulsion formation was studied. The CS concentration was fixed at 0.5%w/w. The surfactant concentration was fixed at 0.5%w/w and the oil concentration was fixed at 10%w/w. The order of first phase separation time was observed. It was found that the formulation with pH values from 3.75-4.50 rapidly separated at 1 hour after formulating. When the pH values were increased to 5.09, the phase separation times were delayed to 4 days. Once the pH value was more than 5.09, the separation times became faster again to 3 days and 1 day for the pH value at 5.35 and 5.70 respectively. When the pH values were more than 5.7, the separation times became much shorter to 3 hours and 1 hour for pH value of 6.09 and 6.51 respectively. The texture of emulsion at pH value of 6.50 was non homogeneous and coarse. The viscosities of emulsions were highest at the pH values at 5.35 and 5.70. The viscosities became less when the pH values was higher than 5.70 and less than 5.35. Therefore, the stable conditions of pH values could be ranging from 5.00 to 5.70 (Table 4). The result was the same explanation from Onesippe and Lagerge (2008) that at higher pH value the emulsion tend to loss their charge and tend to separate.

Table 4 Properties of emulsion with different pH value

Physical properties	**pH**							
	3.75	**4.10**	**4.50**	**5.09**	**5.35**	**5.70**	**6.09**	**6.51**
Appearance and texture	Fine, thin	Fine, thin	Fine, thin	Fine, thin	Fine, thicker	Fine, thicker	Fine, thin	Coarse
Viscosity[a]	1	1	2	2	3	3	2	1
First separation time	1 hr	1 hr	1 hr	4 days	3 days	1 day	3 hr	1 hr
Creaming layer[b] (%)	27.5	27.5	27.5	20.1	20.1	22.9	27.5	55.0

Note. [a]Every formula has low viscosity. The higher number represents higher viscosity

[b]Determined at day 6

6. Effect of NaCl Concentration

NaCl is an electrolyte substance that dissociates into ion. It usually helps increasing viscosity of anionic solution for surfactant cleaning system when used at low concentration by increasing ionic density which affect the size and shape of micelle formation. The electrolyte also improves stabilization of emulsion (Leelapornpisid, 1997a, 2001b). The effect of NaCl concentration from 0 to 5%w/w on emulsion formation was studied. The CS concentration was fixed at 0.7%w/w. The surfactant was fixed at 0.7%w/w and the oil concentration was fixed at 10%w/w. The final pH value of emulsion was 5.00. The appearance of emulsion was thin with fine texture for all concentration of NaCl from 0-5%w/w. The viscosities did not change by the presence of NaCl as well. The emulsions separate rapidly within 30 minutes when NaCl was added in the formulation (0.5-5%w/w). The emulsion stayed stable for 1 day without NaCl. Therefore, it was clearly found that NaCl has direct impact on emulsion stability. The emulsions in this study lost their stability rapidly when NaCl was added into the formulation (Table 5) whereas in ordinary emulsion the electrolyte improves stability of the emulsion system. This could be explained that the decreased electrostatic repulsion force of polyelectrolyte complex was due to ionic charges of NaCl electrolyte.

Table 5 Properties of emulsion with different NaCl concentration

Physical properties	**NaCl concentration (%w/w)**								
	0	**0.5**	**1**	**1.5**	**2**	**2.5**	**3**	**4**	**5**
Appearance and texture	Fine, White emulsion	Fine, White emulsion	Fine, White emulsion	Fine, White emulsion	Fine, White emulsion	Fine, White emulsion	Fine, White emulsion	Fine, White emulsion	Fine, White emulsion
Viscosity[a]	2	2	2	2	2	2	2	2	2
First separation time	1 days	<30 min	<30 min	<30 min	<30 min	<30 min	<30 min	<30 min	<30 min
Creaming layer[b] (%)	21.0	21.0	21.0	21.0	21.0	21.0	21.0	21.0	21.0

Note. [a]Every formula has low viscosity. The higher number represents higher viscosity

[b]Determined at day 1

The microscopic property of emulsion was investigated using microscope MOTIC BA300, China. The SPECs emulsion was taken via the optical microscope after 1 day of preparation (Figure 1). This formulation consisted of 0.7%w/w CS, 0.7%w/w surfactant and 10%w/w jojoba oil. The emulsion was stable for 1 day then the phase separation was observed on the formulation without NaCl. It can be seen that the particle sizes were aggregated and did not distribute evenly. The dispersed phase was being coalescence. The emulsion was identified as oil-in-water emulsion.

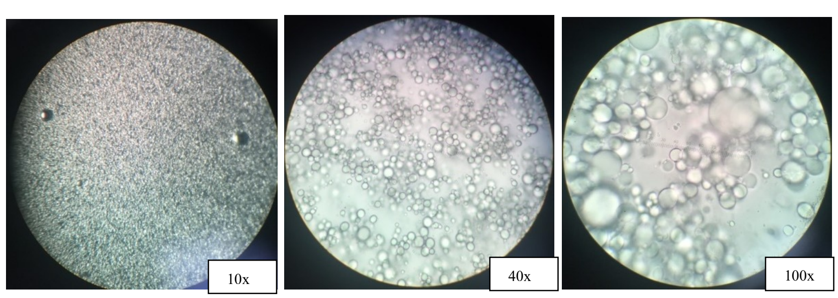

Figure 1 Microscopic images of surfactant-chitosan emulsion at different magnification

The stability of different emulsion systems such as emulsion with only CS, emulsion with only surfactant and SPECs emulsion of CS and surfactant from the previous experiment were observed on phase separation after 1 day of emulsion produce (Figure 2). It can be seen that emulsion that composed only CS showed phase separation very fast (3 hours). The same is observed in system with only surfactant, as the phase separation occurred in 1 hour. When the system composed of CS and surfactant, the emulsion could last longer (1-4 days). Therefore, the Surfactant Polyelectrolyte Complexes was formed and can improve emulsion stability.

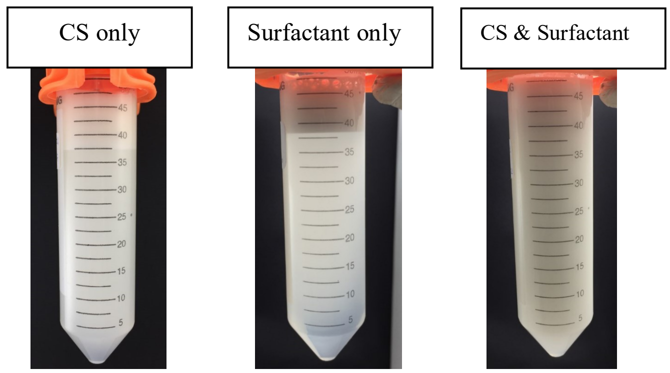

Figure 2 Appearance of emulsion

Conclusion

In this study Suga®Nate 160NC and chitosan complexes were used as emulsifiers in SPECs emulsion preparation. The range of CS concentration to stabilize the emulsion were between 0.5-0.7%w/w, on specific condition of 10%w/w oil, 0.5%w/w surfactant and pH value of 5.00. The range of surfactant concentration were between 0.2-1.5%w/w, on specific condition of 10%w/w oil, 0.5%w/w CS, pH value of 5.00. The jojoba oil concentration were between 10-15%w/w, on specific condition of 0.5%w/w surfactant, 0.5%w/w CS and pH value of 5.00. The suitable pH value for emulsion formation is at 5.00-5.70. The NaCl electrolyte had an influence to deteriorate stability of emulsion system. For future studies, it is recommended to use mechanical apparatus such as high pressure homogenizer or ultrasonic homogenizer to reduce the particle size of emulsion or use additional stabilizer in order to develop a more stable emulsion.

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