As. J. Food Ag-Ind. **2012**, 5(04), 322-330

OPEN ACCESS

Asian Journal of Food and Agro-Industry

ISSN 1906-3040

Available online at www.ajofai.info

Research Article

Influence of pH and heat treatment on sesame oil emulsion stabilized by whey protein isolate–pectin membranes

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Abstract

Sesame oil is an edible vegetable oil derived from sesame seeds. Most people in Asia use sesame oil for seasoning, particularly in Asian cuisine such as sauce and salad dressing. However, the addition of this oil to food is limited by stability of the emulsion. The objective of this work was to investigate the influence of pH and heat treatment on the stability of multilayer emulsion consisting of whey protein isolate-pectin. A primary emulsion containing cationic whey protein isolate-coated droplets was prepared by homogenizing 20 wt% sesame oil with 80 wt% aqueous whey protein solution (0.5 wt% WPI, pH 3). This emulsion was mixed with aqueous pectin solution (0.5 wt%, pectin slow set 63%, pH 3) to yield emulsions containing 10 wt% sesame oil, 0.25 wt% WPI and 0.25 wt% pectin. The stability of secondary emulsion to pH (2–7) and heat treatment (30 min at 50–90^{0}C) was determined using particle size, zeta-potential and microstructure analysis. The results showed that secondary emulsions were stable to droplet aggregation and creaming at pH 3-5 and at 50–90^{0}C. The interfacial engineering technology used in this study could lead to the creation of food emulsions with improved stability to environmental stresses.

Keywords: multilayer emulsion, whey protein isolate, pectin, *Sesamum indicum,* Thailand.

Introduction

Sesame (*Sesamum indicum* L.) is an important oilseed which is cultivated in many tropical countries. In 2007, the Thai Office of Agriculture Economics reported that approximate sesame production and farm value in Thailand were 43,000 tons and 1,030 million baht, respectively [1]. Sesame seed is composed of about 47.8-52.2% oil, 26.9-25.8% protein and 4.7-5.6% ash [2, 3]. Sesame seed and sesame oil contain abundant lignans including sesamolin, sesamin and lignan glycosides. Sesamolin and sesamin have been reported to incorporate pharmacological properties, e.g. decreasing blood lipids [4], and arachidonic acid levels [5], increasing antioxidative ability [6] and γ-tocopherol bioavailability [7], and providing anti-inflammatory function [8] and estrogenic activity [9, 10]. Apart from lignans, sesame oil contains important biologically active compounds, such as vitamin E, especially γ-tocopherol, which has many beneficial properties, such as antiproliferative effects in human cancer cells [11], anti-inflammatory and partial prevention of age-associated transcriptional changes in heart and brain of mice [12]. Sesame seed is an important source of edible oil. In the sesame oil industry, sesame seed is commonly used as the raw material for oil extraction, either using organic solvents or by mechanical pressing. Sesame oil is very popular used in Korean, Japanese and Chinese food. In Thailand, sesame oil is wildly used for cooking and in cosmetic preparations such as body lotion, shampoo and soap.

The multilayer emulsion is a novel interfacial engineering technology. The emulsions formed contain oil droplets surrounded by multilayer interfacial membranes.

This interfacial engineering technology could be used by the food industry to create emulsion-based food products that have better resistance to environmental stresses (e.g. pH, temperature). Droplets coated with a relatively thick interfacial layer of emulsifier may also have better stability to aggregation and to lipid oxidation than those coated with thinner layers. However, information of sesame oil emulsion is still limited. The objective of this work is to investigate the influence of pH and heating on the stability of multilayer emulsion consisting of whey protein isolate-pectin.

Materials and Methods

Materials

Sesame oil was purchased from the Learning Organization and Development Center of Sesame for Sustainable Agro-Household Industry, Faculty of Agriculture, Ubonratchatani University, Thailand. Whey protein isolate (WPI) was obtained from Davisco Foods International Inc. (Le Sueur, MN). As stated by the manufacturer, the powdered WPI had a composition of 97.6 wt% protein, 2.0 wt% ash and 0.3 wt% fat (dry weight basis) and 4.7 wt% moisture (wet weight basis). Pectin slow set 63% was obtained from S.P.Y. Science Tech Ltd., Thailand.

Methods

Solution preparation

An emulsifier solution was prepared by dispersing 0.5 wt% whey protein isolate in 5 mM phosphate buffer at pH 7.0 containing 0.04% sodium azide (as an antibacterial agent). Whey protein solution was stirred for at least 1 h. A pectin solution was prepared by dispersing 1 wt% pectin (slow set 63%) in 5 mM sodium citrate buffer at pH 3 and stirring for at least 1 h. to ensure complete hydration.

Emulsion preparation

A primary emulsion was prepared by homogenizing 20 wt% sesame oil and 80 wt% emulsifier solution. The oil and emulsifier solution were blended using a blender for 1 min (HR1357 300, Watt, Philips) and then passed through a high-pressure valve homogenizer twice at 1200 psi (TA 18/D, Didacta, Italy). The pH of the emulsion was adjusted to 3.0 (1.0 using 0.1 N HCl). Secondary emulsions were prepared by diluting primary emulsion with pectin solution to obtain a final emulsion composition of 10 wt% sesame oil, 0.25 wt% WPI and 0.5 wt% pectin. The emulsions were then stored for 24 h. at ambient temperature prior to analysis.

Influence of pH and heat treatment on sesame oil emulsion stabilized by whey protein isolate–pectin membranes

The stability of the secondary emulsions to pH (2-7) and heat treatment (50-90°C) was determined using a variety of analytical techniques; laser light scattering, zeta-potential and optical microscopy as described below.

Particle size determination
The particle size distribution of the emulsions was measured using Zetasizer Nano (Malvern Instruments Ltd., Malvern, UK). The emulsions were diluted in a buffer solution at pH of the sample (10^{-3}). The particle size was reported as mean particle diameter.

ζ-Potential measurements
Emulsions were diluted to a droplet concentration of approximately 0.001% wt using buffer solutions of pH from 3 to 8 to avoid multiple scattering effects. The ζ-potential of the droplets was then determined using a particle electrophoresis instrument (Zetasizer Nano, Malvern Instruments Ltd., Malvern, UK) that measures the direction and velocity of droplet movement in the applied electric field. The ζ-potential provides an estimate of the net charge on a particle measured at the 'shear plane', which depends on the charge on the actual particle (in this case droplets, emulsifier and biopolymer) plus the charge associated with any ions that move along with the particle in the electric field.

Optical microscopy
The microstructures of the emulsions were observed using an optical microscope. Emulsion samples were slightly vortexed in a glass test tube before analysis. A drop of emulsion was then placed on a microscope slide, covered by a cover slip. The emulsion structure was then observed at a magnification of 40x. An image of the sample was acquired using digital image processing software and stored on a personal computer.

Statistical analysis
All experiments were carried out using at least two freshly prepared emulsion samples and three replicates of each sample were measured. The obtained data were analyzed using one way analysis of variance (ANOVA). Means were compared by Duncan's multiple range test with mean square error at 5% probability (SPSS 11.0 for Windows statistical software).

Results and Discussion

Influence of pH on emulsion stability
The aim of this part was to investigate the influence of pH on the stability of secondary emulsion. The influence of pH on ζ- potential, mean particle diameter and microstructure of secondary emulsion was measured after 1 day of preparation (Figures 1-3).

The ζ-potential of the whey protein stabilized droplets in primary emulsions was positively charged (49.7 mV) at pH 3 (data not shown). The ζ-potential of secondary emulsions were negatively charged (-12.0 to -15.0) at all pH 2-7 (Figure 3). The magnitude of the ζ-potential was lower at pH levels close to *pKa* of pectin (pH 3-5). This was expected as it has been previously reported by Guzey *et.al.,* [13]. At pH above 5, pectin is expected to be desorbed from the surfaces of whey protein isolate, which is a major component of β-lactoglobulin, stabilized oil droplets because both polymers are negatively charged at these pH levels.

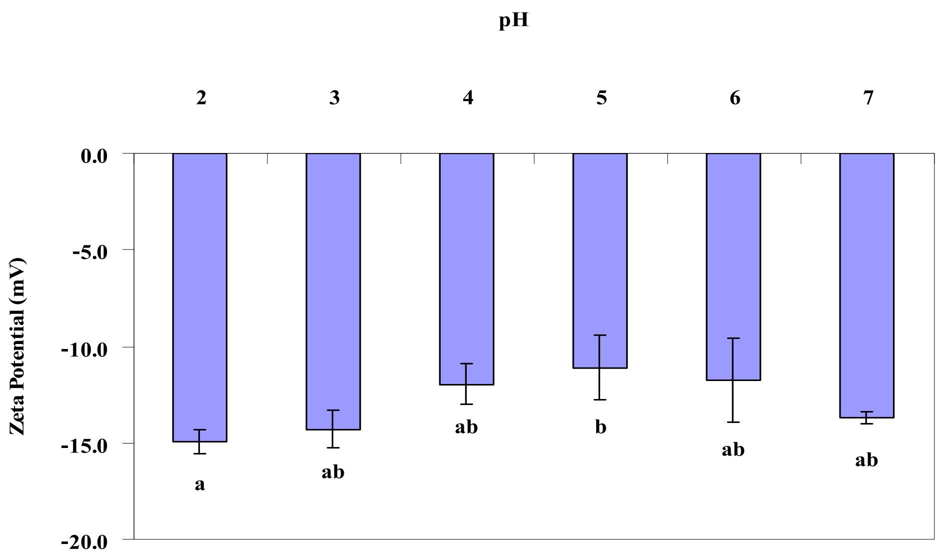

Figure 1. ζ-potential of 10% wt oil emulsion stabilized by 0.25% wt whey protein isolate (WPI) and 0.25 wt% pectin in sodium citrate buffer at pH 2-7.

Mean droplet size and microstructure of the secondary emulsion provides further information to understand the desorbed state of polymer layers (Figures 2-3). In secondary emulsions, mean droplet size was small at pH 2-5, but was bigger at pH 6-7. However, microscopic pictures showed aggregation at pH 6 and 7. This was probably because of depletion flocculation associated with the non-adsorbed pectin. The stability of secondary emulsions at pH 5 is an important advantage of multilayer emulsions over the primary emulsion because Blg could not stabilize the emulsion at this pH alone [14, 15].

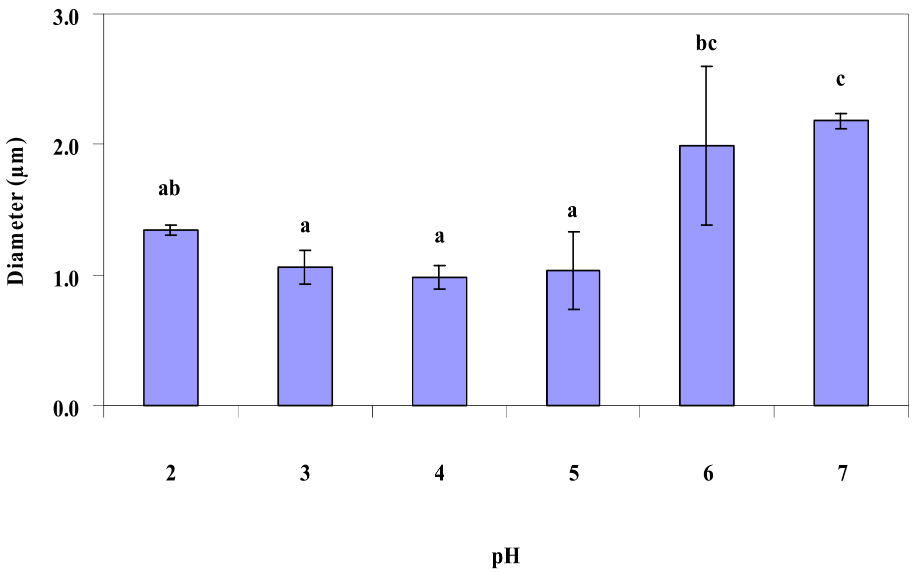

Figure 2. Changes in mean particle size of 10% wt oil emulsion stabilized by 0.25% wt whey protein isolate (WPI) and 0.25 wt% pectin in sodium citrate buffer at pH 2-7.

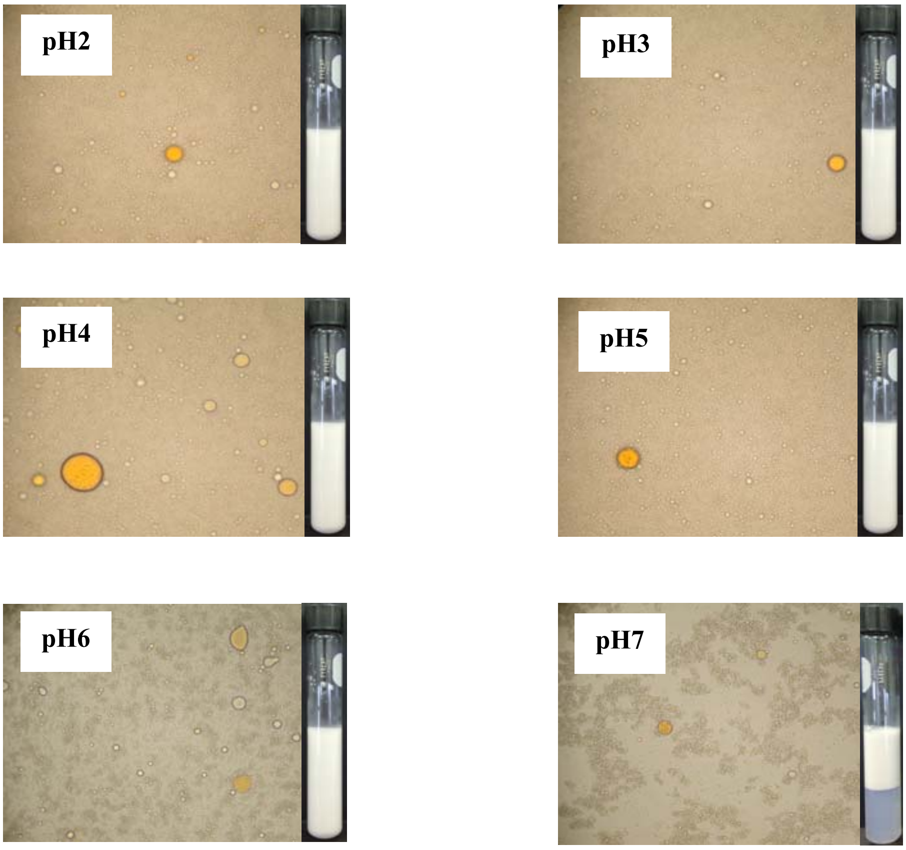

Figure 3. Micrographs of 10% wt oil emulsion stabilized by 0.25% wt whey protein isolate (WPI) and 0.25 wt% pectin in sodium citrate buffer at pH 2-7.

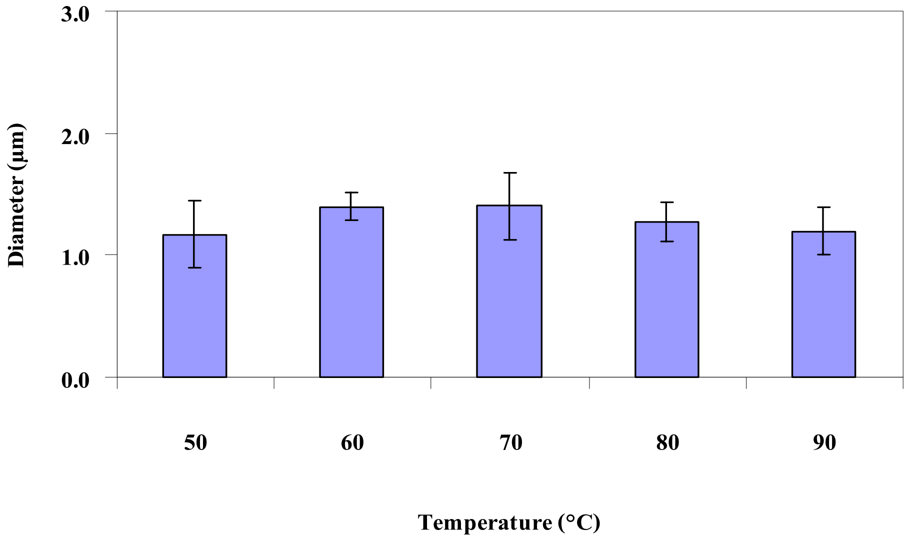

Figure 4. Changes in mean particle diameter of 10% wt oil emulsion stabilized by 0.25% wt whey protein isolate (WPI) and 0.25 wt% pectin in sodium citrate buffer at 50-90^{0}C.

Influence of heat treatment on emulsion stability

The aim of this part was to examine the influence of heat treatment on the stability of whey protein isolate-pectin (secondary emulsion). There was no significant difference in particle size of the secondary emulsions (~1.0-1.4 µm). The secondary emulsions were stable against heat treatment at all temperatures, which was observed as a single population of relatively small droplets and good creaming stability (Figures 4-6). These result suggested that the overall composition of interfacial layers was unaffected by heating.

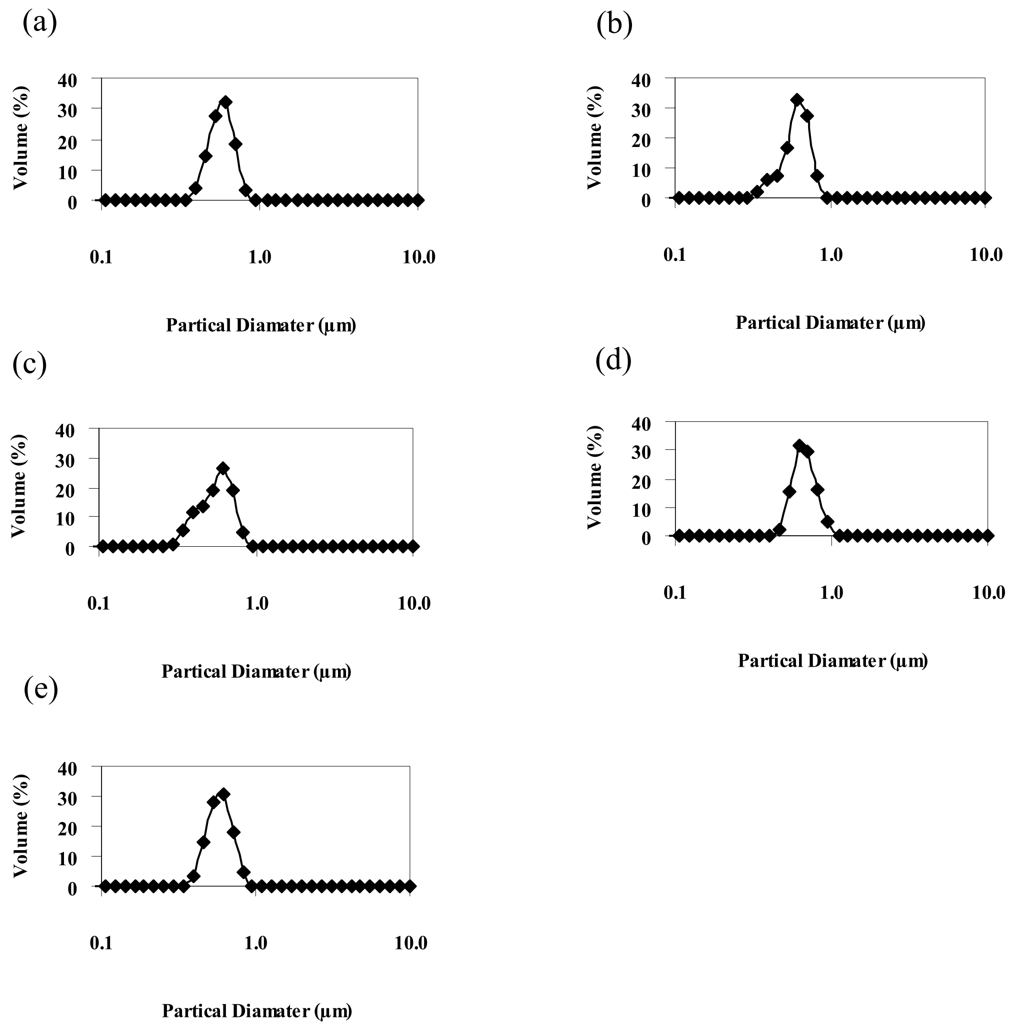

Figure 5. Particle size distribution of 10% wt oil emulsion stabilized by 0.25% wt whey protein isolate (WPI) and 0.25 wt% pectin in sodium citrate buffer at (a) 50^{0}C, (b) 60^{0}C, (c) 70^{0}C, (d) 80^{0}C and (e) 90^{0}C.

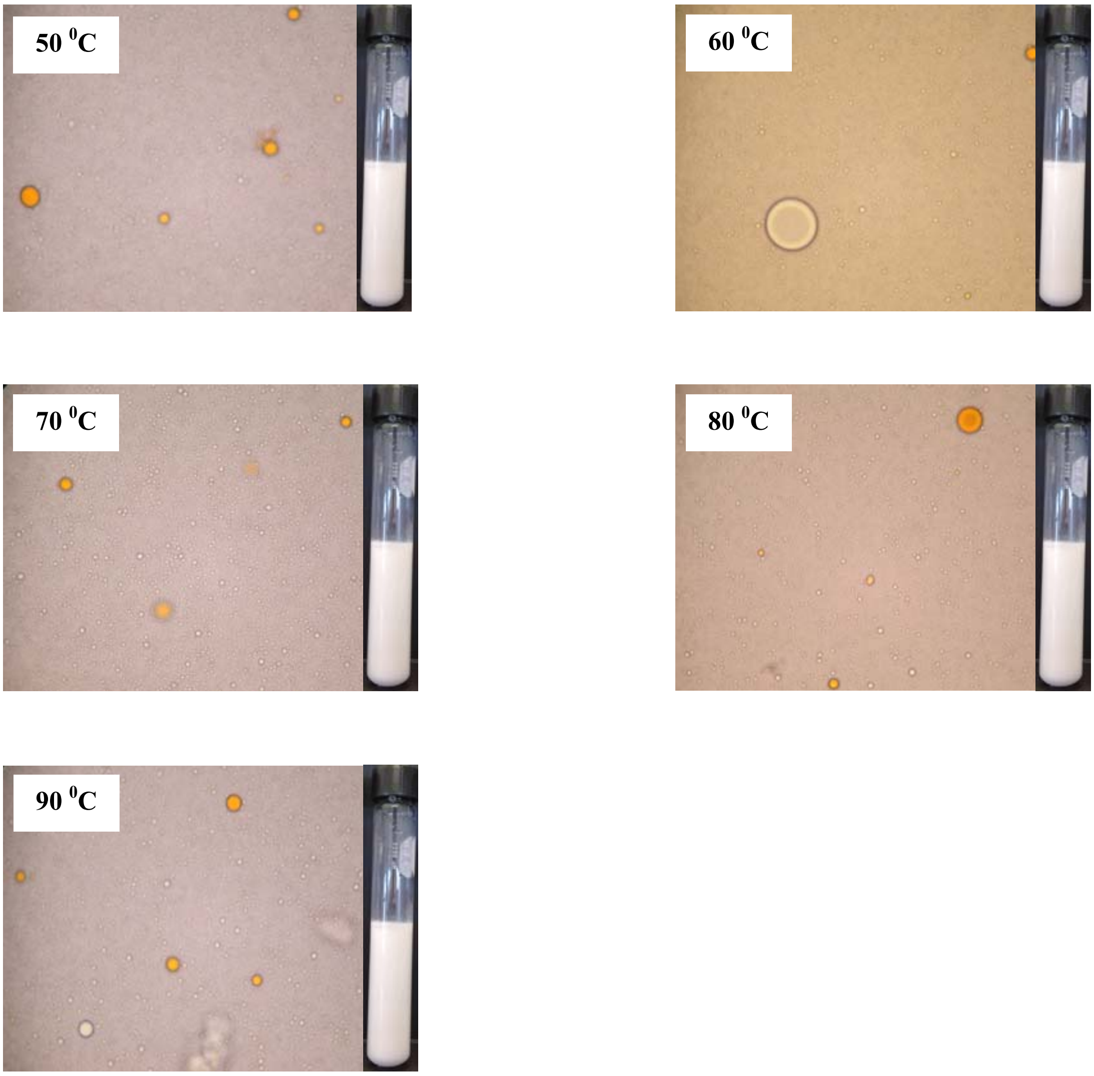

Figure 6. Micrographs of 10%wt oil emulsion stabilized by 0.25% wt whey protein isolate (WPI) and 0.25 wt% pectin in sodium citrate buffer at 50-90^{0}C.

Conclusion

This work has demonstrated that multilayer emulsions (whey protein isolate-pectin) can be used to prepare sesame oil-in-water emulsions using high pressure valve homogenization. The multilayer emulsion containing anionic lipid droplets coated with a whey protein isolate-pectin membrane was produced by mixing an anionic polysaccharide (pectin at pH3) with the primary emulsion (whey protein at pH3). These emulsions had relatively good stability to particle aggregation at pH 3-6 and at 50-90^{0}C.

The fact that the multilayer emulsions are mainly stable at lower pH values (3–6), suggests that they would be most suitably used in acidic food, such as salad dressings, beverages and yogurt drinks. It is therefore recommended that future studies should examine in greater detail the influence of environmental conditions on stability of multilayer emulsions.

Acknowledgments

This research was supported by the Grant-in-Aid for senior project from the Faculty of Agriculture, Ubonratchathani University, Thailand. The authors also thank Davisco International for the whey protein isolate, as well as S.P.Y. Science Tech Ltd., Thailand for kindly donating pectin used in this study.

References

1. Office of Agricultural Economics (2008). Agricultural Statistics of Thailand 2006. Bangkok, Ministry of Agriculture and Co-operatives.

2. Elleuch, M., Besbes, S., Roiseux, O., Blecker, C. and Attia, H. (2007). Quality characteristics of sesame seeds and by-products. ***Food Chemistry,*** 103 (2), 641-650.

3. Gandhi, A.P. and Srivastava, J. (2007). Studies on the production of protein isolates from defatted sesame seed (*Sesamum indicum*) flour and their nutritional profile. ***ASEAN Food Journal,*** 14 (3), 175-180.

4. Hirata, F., Fujita, K., Ishikura, Y., Hosoda, K., Ishikawa, T. and Nakamura, H. (1996). Hypocholesterolemic effect of sesame lignan in humans. ***Atherosclerosis,*** 122, 135–136.

5. Shimizu, S., Akimoto, K., Shinmen, Y., Kawashima, H., Sugano, M. and Yamada, H. (1991). Sesamin is a potent and specific inhibitor of delta 5 desaturase in polyunsaturated fatty acid biosynthesis. ***Lipids,*** 26, 512–516.

6. Hemalatha, S., Raghunath, M. and Ghafoorunissa (2004). Dietary sesame oils inhibits iron-induced oxidative stress in rats. ***British Journal of Nutrition,*** 92, 581–587.

7. Lemcke-Norojarvi, M., Kamal-Eldin, A., Appelqvist, L.A., Dimberg, L.H., Ohrvall, M. and Vessby, B. (2001). Corn and sesame oils increase serum gamma-tocopherol concentrations in healthy Swedish women. ***Journal of Nutrition,*** 131, 1195–1201.

8. Utsunomiya, T., Chavali, S.R., Zhong, W.W. and Forse, R.M. (2000). Effects of sesamin-supplemented dietary fat emulsions on the *ex vivo* production of lipopolysaccharide-induced prostanoids and tumor necrosis factor A in rats. ***American Journal of Clinical Nutrition,*** 72, 804–808.

9. Coulman, K.D., Liu, Z., Hum, W.Q., Michaelides, J. and Thompson, L.U. (2005). Whole sesame seed is as rich a source of mammalian lignan precursors as whole flaxseed. ***Nutrition and Cancer,*** 52(2), 156–165.

10. Wu, W.H., Kang, Y.P., Wang, N.H., Jou, H.J. and Wang, T.A. (2006). Sesame ingestion affects sex hormones, antioxidant status and blood lipids in post-menopausal women. ***Journal of Nutrition,*** 136, 1270–1275.

11. Guthrie, N., Gapor, A., Chambers, A.F. and Carroll, K.K. (1997). Inhibition of proliferation of estrogen receptor-negative MDA-MB-435 and -positive MCF-7 human breast cancer cells by palm oil tocotrienols and tamoxifen, alone and in combination. ***The Journal of Nutrition,*** 127(3), 544S–548S.

12. Rangkadilok, N., Pholphana, N., Mahidol, C., Wongyai, W. Saengsooksree, K., Nookabkaew, S. and Satayavivad, J. (2010). Variation of sesamin, sesamolin and tocopherols in sesame (*Sesamum indicum L.*) seeds and oil products in Thailand. ***Food Chemistry****,* xxx.

13. Guzey, D., Kim, H.J. and McClements, D.J. (2004). Factors influencing the production of o/w emulsions stabilized by beta-lactoglobulin-pectin membranes. ***Food Hydrocolloids****,* 18, 967–975.

14. Onsaard, E., Vittayanont, M., Sringam, S. and McClements D. (2005). Properties and stability of oil-in-water emulsions stabilized by coconut skim milk proteins. ***Journal of Agricultural and Food Chemistry,*** 53*:* 5747-5753.

15. Onsaard, E., Vittayanont, M., Sringam, S. and McClements, D. (2006). Comparison of properties of oil-in-water emulsions stabilized by coconut cream proteins with those stabilized by whey protein isolate. ***Food Research International,*** 39: 78-86.