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Evaluation of long-term stability of milk beverages by a novel method for rapid determination of aggregation forces between colloidal particles

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ABSTRACT

Long-term stability of milk-based emulsions in canned coffee or tea varies with the formulation of small-molecule emulsifiers aiming at bacteriostatic effects or stability-enhancing effects. To predict the long-term stability of the emulsions in short-time period, we developed a novel method for rapid determination of aggregation forces between colloidal particles using model emulsions and suspensions with different stability to aggregation for the first step. The novel method was based on an idea that particles with stronger aggregation forces tend to form aggregates and cannot be readily redispersed. While the milk-based emulsions were subjected to long-term storage test with coffee extract, the same emulsions, but not including coffee extract, were rapidly evaluated by both the novel method and a common method, Turbiscan analysis often applied to the evaluation of emulsion stability. Statistical regression analysis according to the datasets obtained by the two rapid assays revealed that the long-term stability of the milk-based emulsions can be better predicted by the aggregation forces evaluated by the newly developed method than the initial aggregating process evaluated by Turbiscan method. We named the novel method "vibration-redispersion method".

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1. Introduction

Milk and its ingredients are often used for food products such as beverages, particularly for coffee or tea. Canned coffee or tea with milk normally includes two types of emulsifiers; one is for bacteriostatic effects and the other is for stability-enhancing effects (Matsumiya, Takahashi, Inoue, & Matsumura, 2010). Since bacteriostatic emulsifiers tend to destabilize protein-based emulsions despite of the name "emulsifier" (Matsumiya, Nakanishi, & Matsumura, 2011), stability-enhancing emulsifiers are necessary for manufacturing stable products, that is, products with long-term shelf life.

When both types of emulsifiers are added to canned coffee or tea with milk, the product shows different long-term stability with respect to creaming or aggregation according to different combinations of the emulsifiers. To examine the different long-term stability, manufactures have to spend much time and efforts toward designing optimum formulations in developing new commercial products. In practice, it is quite difficult to predict the stability of the milk-based emulsions during long-term storage and is, therefore, strongly required to develop a simple and rapid method to estimate the long-term stability of the emulsions in short-time period.

An emulsion is thermodynamically unstable and undergoes a time-dependent change of the droplet size distribution based on instability processes such as creaming, flocculation, aggregation and coalescence (Dickinson, 1992, pp. 79–122). Creaming can be readily confirmed in destabilized emulsions by visual observation and optical methodology, while aggregation of oil droplets can be also detected in macro or micro-ordered scale by visual or microscopic observation and particle size analysis (McClements, 2005, pp. 269–339). These analytical techniques, however, cannot be directly applied to the prediction of long-term stability of emulsions in kinetically stable state, i.e., with no creaming or no aggregation in most cases.

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In our previous research, we investigated potential factors affecting the creaming velocity and aggregation efficiency in milk-based emulsions including bacteriostatic and stability-enhancing emulsifiers; for example, particle size, zeta potential, steric effects of adsorbed layer and so on (Matsumiya et al., 2010). We clarified that the main factors affecting stability of the milk-based emulsions can be the amount and composition of milk proteins on the oil droplet surfaces. Although our objective for potential factors affecting the destabilization process was, therefore, attained in the previous research, the development of a practical tool for predicting emulsion stability is still required.

Recently, many researchers have been used an optical technique, Turbiscan for evaluation of the stability of emulsions such as creaming or flocculation, which is originally developed by Mengual, Meunier, Cayré, Puech, and Snabre (1999a,b). Juliano et al. (2011) analyzed creaming of fat globules in raw and recombined milk by Turbiscan, and reported that creaming was more evident in raw milk than that of recombined one. Grotenhuis, Tuinier, and Kruif (2003) applied Turbiscan method to concentrated dairy products that can be models of whipped cream or evaporated milk to evaluate a kind of colloidal attractive force, depletion interactions. This method was also utilized for measuring sedimentation velocity of destabilized milk proteins in acidified milk beverages (Laurent & Boulenguer, 2003; Sedlmeyer, Brack, Rademacher, & Kulozik, 2004). The Turbiscan techniques are common and advantageous to rapid measurements, and widely applied for food emulsions consisting of milk or its ingredients.

In our previous research, we performed a centrifugal process on the milk-based emulsions in order to obtain the cream layer for subsequent analyses (Matsumiya et al., 2010). We found a tendency at that process that the collected cream layer, i.e., concentrated oil droplets after centrifugation treatments varied with the formulation of the milk-based emulsions; that is, the more unstable an emulsion was, the more tightly packed and aggregated oil droplets were. This implies some relationship between the aggregation strength of oil droplets and the long-term stability in the milk-based emulsions.

In this research, we propose a novel method for rapid determination of aggregation forces between colloidal particles that is developed based on this previous work. The central idea of the novel method is that particles with stronger aggregation forces tend to form aggregates and should not be redispersed easily. Our objective is to test the usefulness of our newly developed method as a tool for predicting the long-term stability of emulsions in short-time period, especially, in comparison with Turbiscan method that is common and widely accepted in the fields of food science and food industry.

2. Materials and methods

2.1. Materials

Deionized water was used for the preparation of all the solutions. Corn oil was obtained from Nacalai Tesuque, Inc. (Kyoto, Japan). Fatty-acid free bovine serum albumin (BSA) was purchased from Wako Pure Chemical Industries, Ltd. (Osaka, Japan). Polystyrene latex suspension with particle diameter of 0.5 μm (Polybead polystyrene microspheres, 2.5% solids-latex) was made by Polysciences, Inc. (Pennsylvania, the USA). Powdered milk was obtained from Snow Brand Milk Products Co., Ltd. (Tokyo, Japan). Emulsifiers, P-1670 (sucrose palmitate, HLB 16), S-570 (sucrose stearate, HLB 5) and S-770 (sucrose stearate, HLB 7) were manufactured by Mitsubishi-Kagaku Foods Corporation. Q-14Y (decaglycerol esters of myristic acid, HLB 14.5) was produced by Taiyo Kagaku Co., Ltd. TRP-97RF (triglycerol esters of palmitic acid, HLB 10), DP-95 (diglycerol esters of palmitic acid, HLB 8) and BS-20 (succinate mono-glyceride, HLB 5.5) were manufactured by Riken Vitamin Co., Ltd. Of these emulsifiers, P-1670, DP-95 and TRP-97RF are with bacteriostatic effects. Powdered milk and all the emulsifiers were stored in a refrigerator prior to use. All other general chemicals used were of analytical grade purchased from Nacalai Tesuque, Inc. (Kyoto, Japan) and Wako Pure Chemical Industries, Ltd. (Osaka, Japan).

2.2. Development of a novel method

2.2.1. Preparation of model emulsions and suspensions

Protein solution was prepared by dissolving BSA into 200 mM sodium phosphate buffer (pH 7.0). Stock emulsion was prepared by homogenizing corn oil and the protein solution with a high-speed blender at 20 °C at 14,000 rpm for 3 min (Physcotron, NS-51, Microtec Co., Ltd., Japan). The contents of oil, protein and buffer in the stock emulsion were 2.5, 0.5 and 97.0 wt% respectively. The stock emulsion and the purchased polystyrene latex suspension were mixed with various concentrations of NaCl solutions at the volume ratio of 1:4 to prepare model emulsions and model suspensions, respectively. The final concentration of NaCl in these model emulsions and suspensions was from 0 to 400 mM. The model emulsions were heated at 90 °C in a water bath for 30 min to promote emulsion destabilization. They were cooled down in trash ice and then kept at room temperature for a while. Both the model emulsions and suspensions were transferred into a plastic sample tube with a volume of 1.5 ml.

2.2.2. Forcible aggregation

Colloidal particles in the model emulsions and suspensions were forced to form aggregates through centrifugal treatments. The model emulsions in the sample tube were centrifuged using an ultracentrifuge (CS120, Hitachi Koki Co., Ltd., Japan) at 20 °C at 140,000 *g* for 20 min. The model suspensions in the sample tube were centrifuged at 20 °C at 1960 *g* for 5 min and then centrifuged at 7830 *g* for 10 min using a centrifuge (MR-150, Tomy Seiko Co., Ltd., Japan). This two-step treatment was aimed at collecting the aggregates in the tightly packed state into the bottom part of the tubes. The two-step treatments were enough to make the colloidal particles thoroughly aggregated.

2.2.3. Redispersion

As a preliminary examination, the centrifuged model emulsions and suspensions were subjected to shaking process by a vortex-type mixer (Vortex genie 2, Scientific Industries Inc., USA) at room temperature at a shaking level of 6, which is just an indication of the mixing speed, in order to redisperse the forcibly aggregated colloidal particles.

As a more sophisticated method controlling the power of redispersion, the following process was employed. The tubes containing the centrifuged model emulsions and suspensions were placed into a tube rack with no bottom and placed on the vibrating part of an active contact speaker (GY-1, Fostex Company, a division of Foster Electric Co., Ltd., Japan) with the top gently pressed by a weight (100 g), and subsequently vibrated to redisperse the forcibly aggregated colloidal particles by the speaker at 100 Hz for various times in an incubator at 20 °C.

2.2.4. Turbidimetry

After the redispersion treatment, the middle part of the capped sample tube was bored with a flame-heated sharp needle followed by insertion of another injection needle of a syringe. The redispersed layer of the both model dispersions was carefully taken out by the needle-attached syringe with the cap open, and then

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