Formation and Physical Analysis of Oleogels Composed of Edible Oils and High-Melting Fat Crystals

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Abstract: This paper reports the preparation of oleogels composed of edible oils (olive oil, squalene, and caprylic/capric triglyceride) and high-melting fat crystals (tribehenoyl-glycerol (BBB)) to explore the potential use of BBB/edible oil mixtures as low-cost and stable gelators. These mixtures exhibited gel-like behaviors upon rapid cooling and subsequent heating. The mixtures of BBB in the liquid oils formed oleogels at BBB concentrations > 4.0 wt%. The thermal behaviors, crystal structures, and crystal morphologies of mixtures of BBB produced from 6.0 wt% BBB crystals in 94.0 wt% liquid oils were examined following the treatment of these systems according to different temperature regimes. In addition, rheological analysis was conducted to evaluate the physical properties and storage stabilities of the prepared oleogels. It was found that rapid cooling to the crystallization temperature ($T_c$) from 70°C and subsequent heating to the final temperature ($T_f$) were necessary to reveal the gel-like behavior. In addition, the crystals treated with rapid cooling were smaller and more uniform in size than those treated with a simple cooling procedure. The differential scanning calorimetry melting peaks were broad or split, and exhibited the eutectic mixing behavior of multi-component triacylglycerols. The X-ray diffraction spectra showed that the melt-mediated $\alpha$ to $\beta$ transformation of the mixtures was a prerequisite for revealing the gel-like behavior. Moreover, the tempering procedure was found to influence the physical properties of the oleogels, wherein no visible changes were observed for any of the oleogels after rapid cooling and storage for 6 months at 25°C.

Key words: oleogel, fat crystal, morphology, WAXD, DSC, rheology, tribehenoyl-glycerol

\section{Introduction}

Gels based on edible oils have attracted great interest in recent years because of their immense potential in formulation development\textsuperscript{7}. To date, two classes of gels have been described in the literature, namely oleogels and hydrogels. Oleogels are viscoelastic materials composed of organic gelators and liquid oils\textsuperscript{5}. These gels are formed by the immobilization of a liquid in a three-dimensional network based on self-assembly or interactions between particles. During the past decade, oleogels have been exploited for various common applications, including oil treatments, foods, pharmaceuticals, and cosmetics\textsuperscript{6}. Although significant efforts have been devoted to a wider range of applications for oleogels, the link between their structural features and their gelation properties is still not clearly understood\textsuperscript{4}.

Recently, research has focused on the relationship between the gelator structure, the nature of the oil, and the gelation efficiency\textsuperscript{5–8}. Indeed, a better understanding of these oleogel systems would help to improve the stability and rheological properties of cosmetic formulations\textsuperscript{9}, could lead to improved drug delivery and controlled release for pharmaceutical applications\textsuperscript{10}, and may provide alternative structural components for foods without the requirement for adding trans and saturated fats\textsuperscript{11,12}. In this context, the gelation of vegetable oils has garnered significant attention. For example, an ethyl cellulose (EC) gel can be formed by entrapping oil within the polymeric network after heating above its glass transition temperature (\textasciitilde 140°C) and subsequent cooling to return the polymer to a

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rigid state; the formation of intermolecular interactions via hydrogen bonding produces an entangled network that entraps the oil\(^8\), \(^{14}\). Waxes have also been shown to be excellent candidates for the formation of vegetable oil-based oleogels. For example, Daniel \textit{et al.} found that an increase in the chain length of the fatty acid reduced the minimum wt/wt concentration required to gel the vegetable oil. This concentration decreased from 15\% for lauric acid to 6\% for myristic acid, 4\% for palmitic acid, and 2\% for stearic acid\(^{15, 16}\). Moreover, Toro-Vazquez \textit{et al.} demonstrated the use of candelilla wax (CW) to structure vegetable oils such as safflower oil at concentrations as low as 2\% wt/wt\(^{17}\), while Sánchez-Becerrila, M. \textit{et al.} reported a detailed characterization of the CW crystal network in oleogels from the micro to the nanoscale by polarized light microscopy and scanning electron microscopy\(^{18}\). The vegetable oil present in CW oleogels is physically trapped in the pores formed by a continuous three-dimensional network of branched microplatelets. At the nanoscale, the building blocks of these CW microplatelets are irregularly-shaped nanoplates.

Although recent successes in relation to the edible applications of oleogels will continue to increase interest and research in the field, it is necessary to implement low-cost (e.g., without specific processing conditions for the respective products) and stable systems to meet consumer expectations in foods and cosmetic products. Thus, despite the fact that EC has an excellent capacity to give structure to oils, its application in large-scale operations is unfavorable due to the high temperatures required for its dispersion in the oil. CW provides ideal characteristics that are comparable to those of saturated and trans fats; however, problems arise regarding its stability over extended periods. In the context of personal applications, there is currently a distinct gap in knowledge in terms of identifying ideal fatty acids and fatty alcohols in relation to the structuring oils, crystal morphologies, and the resulting product stabilities and textures. Although carbohydrate-based gelators have been extensively studied, they have received little attention in the field of edible oleogels. From a physical stability point of view, recent publications into oleogels have failed to consider the stability of such systems over several weeks to months at temperatures greater than refrigerator temperatures. Indeed, many open questions remain regarding the gelling properties of these systems.

Recently, Sato \textit{et al.} reported the crystallization of a network of triacylglycerol (TAG) crystal particles upon rapid cooling of their liquid oil mixtures, which plays a decisive role in the formation of\(^{6, 19}\). By controlling the temperature, they were able to obtain crystals of high melting triglycerides with low melting triglycerides. They found that these oleogels possessed a smooth texture and a good stability over a wide temperature range.

This study therefore aims to explore a potentially low-cost oleogel system with a high stability at ambient temperatures without the requirement for specific processing conditions. In this context, although previous studies\(^{20, 21}\) have reported that behenic acid possesses the potential to exhibit a gel-like behavior, no studies into the preparation of oleogels using tribehenoyl-glycerol (BBB) have been conducted to date. Thus, we herein provide insights into the potential use of BBB and edible oil mixtures as low-cost and stable gelators by examining the gel behaviors and the influence of processing on the crystal properties, wherein we focus on β-fat crystals that do not require any additives. The stability and physical properties of the prepared oleogels are also evaluated using a model system.

2 Experimental Procedures

2.1 Materials

BBB (cosmetic grade, an average of 10 of saponification = approximately 180, \(T_{m} = 53.2^\circ C\), \(T_{g} = 57.7^\circ C\), \(T_{m} = 60.6^\circ C\)) and high purity (>99\%) olive oil (\(T_{m} = 2.0\)–4.0\(^\circ C\)) were purchased from Croda Japan KK (Tokyo). Caprylic/capric triglyceride (non-GMO grade, \(T_{m} = -20^\circ C\)) was purchased from BASF Japan (Tokyo, Japan). Vegetable squalane (derived from sugar cane, \(T_{m} = -37^\circ C\)) was purchased from Nikko Chemicals Co. Ltd. (Tokyo, Japan).

2.2 Preparation of the oleogels

All samples (30 mL) were placed in clean and dry glass bottles. The oleogels were formed by adding 2–10 wt\% BBB to the liquid oils at 70\(^\circ C\) in a water bath. When BBB was completely dissolved, the sample was maintained for 5 min at 70\(^\circ C\). After heating, the samples were followed by tempering upon cooling from 70 ± 1 to 5 ± 1\(^\circ C\) and subsequent heating. All measurements were performed after 24 h storage at 25\(^\circ C\). The gel-like behavior was observed visually by placing the mixtures in glass bottles and tilting them 180° from the vertical, and was defined as the state where no deformation occurred, and where the liquid solid phases did not separate after at least 24 h.

2.3 The specific temperature procedures

Based on the mixing phase behaviors of BBB and the liquid oils, we tested four tempering processes that differed in the cooling rate \(V_{c}\) from 70\(^\circ C\), the crystallization temperature \(T_{c}\), the final temperature \(T_{f}\), and the subsequent heating process, as outlined in Fig. 1.

2.4 Differential scanning calorimetry (DSC) measurements

DSC experiments were conducted for the BBB and liquid oil mixtures, with the exception of the whipped oil samples, using a DSC7000X (Hitachi High-Tech Science Co., Tokyo). The sample (10 mg) was sealed in an aluminum pan, and Al\(_2\)O\(_3\) was used as the reference material for
calibration. The samples were cooled from 80 to 10°C at a rate of 5°C/min, maintained for 5 min at 10°C, and then heated to 80°C at a rate of 2°C/min.

2.5 Polarized light microscopy (PLM) observations
To observe the crystal morphology, the sample was placed on a glass microscope slide with a cover slip and observed using a digital microscope (VHX-6000, Keyence, Osaka). All optical observations were performed at magnifications of 200× and 500×. To kinetically characterize the crystal growth and transformation, the sample was placed on a glass microscope slide with a cover slip and observed using an optical microscope (BX-63, Olympus, Tokyo). The temperature of the slide was controlled using a LinkamT-96-controller. All optical observations were performed at 200× magnification and held for 5 min at each temperature.

2.6 X-ray diffraction (XRD) analysis
A wide-angle diffractometer (X’Pert PRO MPD, PANalytical, Netherlands) was used to analyze the polymorphic forms of the BBB/liquid oil mixtures with CuKα radiation (λ = 0.154 nm, 45 kV, 40 mA) at 25°C. The camera length, which is the distance between the samples and detectors, was 115 mm.

2.7 Rheological properties
The rheological properties of the oleogels and whipped oils were measured at 25°C using an AR-G2 rheometer (TA Instruments, Delaware, USA) equipped with a Peltier system for temperature control. A parallel plate (40 mm) was applied with a measuring gap of 1000 μm. To obtain the variation in the elastic modulus (G’) and the viscous modulus (G’’) with stress, an oscillatory sweep was conducted at a frequency of 1 Hz, and an oscillatory stress from 0.1 to 500 Pa was applied. The variations in the elastic modulus (G’) and the viscous modulus (G’’) with frequencies ranging from 0.01 to 100 Hz were measured within the linear viscoelastic region. The viscosities of all samples were measured at shear rates of 0.1 to 1000 s⁻¹ under steady shear conditions.

3 Results and Discussion
3.1 Visual observations
Figure 2 shows the gel-like behaviors of the BBB/olive oil (6.0/94.0 wt %) liquid mixtures that were subjected to the conditions of Tempering Procedures 1–3. When Tempering Procedure 1 was employed through cooling the mixture from 70 to 36°C followed by storage at 36°C, a sol-like fat
mixture was formed. Under the conditions of Tempering Procedure 2, when the mixture was slowly cooled from 70 to 5°C at a rate of 2°C/min and subsequently heated to 36°C at a rate of 2°C/min and subsequently heated to 36°C at a rate of 2°C/min, the sample showed sol-like behavior (Fig. 2a). Under the conditions of Tempering Procedure 3, a gel-like behavior was observed when the mixture was rapidly cooled from 70 to 5°C at a rate of 10°C/min and subsequently heated to 36°C at a rate of 2°C/min (Fig. 2b).

A similar gel-like behavior was confirmed not only for the olive oil system, but also for the caprylic/capric triglyceride (Fig. S1a) and squalane (Fig. S1b) systems. Moreover, it became clear that the optimum final temperature ($T_f$) was different for the different systems, i.e., 32°C for the caprylic/capric triglyceride system, and 36°C for the squalane system.

Table 1 shows the results of the visual evaluation of the BBB/olive oil fat mixtures obtained at different concentrations of BBB and with variations in $T_f$ under the conditions of Tempering Procedure 3. Sol-like mixtures were formed when the concentration of BBB did not exceed 4.0 wt%, while at BBB concentrations of 4.0 and 6.0 wt%, the mixtures became gel-like below a $T_f$ of 35°C (4.0 wt%) and below a $T_f$ of 38°C (6.0 wt%).

These results indicate the importance of the crystallization of the $\alpha$-form BBB crystals in olive oil at $T_c \geq 5^\circ$C through quenching from 70°C at a rate of 10°C/min at BBB concentrations >4.0 wt%, as previously reported by Higaki et al. These $\alpha$-form crystals are then converted to $\beta$-form crystals through $\alpha$-melt mediation after subsequent rapid heating. From the above visual observations, it was confirmed that a gel state was formed in natural oil, ester oil, and hydrocarbon systems, wherein the gelation temperature was found to differ depending on the melting point and the molecular structure of the liquid oil. This seemed to be related to the high solubility of the liquid oil in the BBB. Of these liquid oils, caprylic/capric triglyceride, which is the most polar and low-molecular-weight oil, is known to dissolve oils and fats well, and so the gelation temperature would be expected to be low; indeed, a sol state was formed at ~36°C. Furthermore, variation in the cooling rate is known to influence the crystallization process; Higaki et al. reported that fully hydrogenated rapeseed oil rich in behenic acid (FHR-B) formed a specific crystal size and shape, and exhibited $\beta$-polymorphism when the appropriate cooling rate was employed. In our case, it is likely that the $\beta$-polymorphic crystals present in the BBB/liquid oil mixtures formed a dense network structure. It was therefore considered that a uniform gel state could form upon maintaining the state of holding the liquid oil. For the 6.0 wt% BBB system, a gel-like behavior was observed when $T_c$ was set at 20°C.

### Table 1: Effect of $T_f$ on the gel-like behaviors of BBB/olive oil mixtures under the conditions of Tempering Procedure 3

| Concentration of BBB (wt%) | $T_f$ (°C) | 30 | 32 | 35 | 38 | 41 | 45 |
|---------------------------|-----------|----|----|----|----|----|----|
| 0                         | -         | -  | -  | -  | -  | -  | -  |
| 2.0                       | -         | -  | -  | -  | -  | -  | -  |
| 4.0                       | ○         | ○  | ○  | △  | △  | △  | -  |
| 6.0                       | ○         | ○  | ○  | △  | △  | △  | △  |
| 8.0                       | ○         | ○  | ○  | ○  | ○  | ○  | ○  |
| 10.0                      | ○         | ○  | ○  | ○  | ○  | ○  | ○  |

*The mixtures were maintained at $T_f$ for 60 min. ○: gel-like, △: somewhat gel-like, - : liquid; $T_c$: crystallization temperature; $T_f$: final temperature; for other abbreviations see Fig. 1.
peaks were obtained upon heating at a rate of 2°C/min. As shown in Fig. 3a, the DSC melting peaks were broad or split due to the eutectic mixing behavior of the multi-component TAGs. Therefore, we indicated the peak maximum temperatures for the broad melting peaks in addition to the average temperatures for the split melting peaks. The melting temperature of the BBB crystals in olive oil was the highest of the various systems examined herein, i.e., 49.7°C (peak I) and 52.4°C (peak II), as shown in Table 2; the presence of two melting peaks is discussed later. In the crystallization thermogram (Fig. 3b), all samples exhibited a sharp exothermic peak, revealing that the initial lipid nucleation and crystal growth were complete within a short time. The corresponding crystallization onset temperatures for the olive oil, squalane, and caprylic/capric triglycerides were 32.6, 33.1, and 31.5°C, respectively (Table 2); we believe that these results can be attributed to intermolecular hydrogen bonding. In this context, Lupi et al. reported that oleogels based on monoglycerides of fatty acids and edible oils formed a network structure involving hydrogen bonding and van der Waals interactions Ref[23]. Thus, the higher crystallization onset observed for oleogels composed of BBB crystals in squalane indicated that the incorporation of squalane facilitated the crystallization of the BBB through the formation of strong van der Waals interactions between the fatty acids of the BBB and the alkyl groups of squalane.

### 3.3 PLM observations

Figure 4 shows the results of PLM imaging of the 6.0 wt% BBB crystals in 94.0 wt% olive oil treated under the conditions of Tempering Procedures 2 and 3. As can be seen from these images, the crystals treated under the conditions of Tempering Procedure 3 were smaller in size and more uniform than those treated under the conditions of Temperature procedure 2. All other samples gave similar results, which can be consulted in the Supporting Information (Fig. S2).

### 3.4 Effect of Vc

Figures 5a, 5b and S3 show PLM images of the 6.0 wt% BBB crystals in 94.0 wt% olive oil solvent treated under the conditions of Tempering Procedures 2–4. As shown, small BBB crystals with dimensions of 10 μm were formed in the BBB/olive oil mixture at Tc = 5°C (Fig. S3a). After heating to Tc = 36°C (Fig. 5a) and further heating to 48°C (Fig. S3b), these crystals did not melt completely, and

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### Table 2

| Oil                  | Endothermic peak (°C) | Exothermic peak (°C) |
|----------------------|-----------------------|----------------------|
| Olive oil            | 49.7, 52.4            | 32.6                 |
| Squalane             | 49.2, 51.7            | 33.1                 |
| Caprylic/capric triglyceride | 48.9, 51.6            | 31.5                 |

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**Fig. 3** (a) Melting and (b) cooling curves of 6.0 wt% BBB crystals in 94.0 wt% liquid oil solvents. Symbols: olive oil (solid line), squalane (dashed line), and caprylic/capric triglyceride (dotted line).

**Fig. 4** PLM images of 6.0 wt% BBB crystals in 94.0 wt% olive oil: (a) under the conditions of Tempering Procedure 2; (b) under the conditions of Tempering Procedure 3. Scale bar = 50 μm. Top right images: 500 × magnification, others: 200 × magnification.
were dispersed non-uniformly, resulting in the formation of crystal aggregates. In this case, a gel-like behavior was not observed. However, under the conditions of Tempering Procedure 3, a gel-like behavior was observed, as shown in Fig. 2, and large, aggregated crystals with dimensions of 20 μm were formed at Tc = 5°C (Fig. S3c). After heating to Tc = 36°C, the crystals of partially aggregated lumps began to dissolve (Fig. 5b). After further heating to 48°C, only needle-like crystals with dimensions of 10 μm remained unmelted (Fig. S3d). These crystals were also more densely dispersed than those treated under the conditions of Tempering Procedure 2. It was revealed that the difference in the cooling rate affects the crystal size and uniformity of the mixtures. The gelation behavior exhibited by quenching was therefore considered to result from the following process. Initially, greater numbers of small α crystals were formed under the conditions of Tempering Procedure 3. During subsequent heating to 48°C, i.e., above the melting points of all forms, the large, aggregated crystals melted and an α-melt-mediated crystallization took place to give needle-like crystals. Adjacent crystals then combined with one another to form a network structure, which increased the hardness of the gel. As a result, the mixture treated under the conditions of Tempering Procedure 3 exhibited a gel-like behavior, as shown in Figs. 2 and S1a–c.

3.5 Effect of Tc

Figure 5c shows the differences in the BBB crystal morphologies of the 6.0 wt% BBB crystals in 94.0 wt% olive oil processed under the conditions of Tempering Procedure 4 (Tc = 25°C, Tf = 36°C). More specifically, under the conditions of Tempering Procedure 4, needle crystals were observed after heating to Tc = 36°C; in this case, no gel-like behavior was observed. However, under the conditions of Tempering Procedure 3, the crystal morphology was different; two kinds of crystals, namely needle-like and lump-like, were observed after heating to Tc = 36°C, as shown in Fig. 5b. In this case, a gel-like behavior was observed.

In the microscopy experiments, the 6.0 wt% BBB crystals in 94.0 wt% olive oil that were treated under the conditions of Tempering Procedure 3 (Tc = 5°C) exhibited a larger crystal size and a more rounded shape than those treated under the conditions of Tempering Procedure 4 (Tc = 25°C). From this result, it was considered that α crystals formed in the BBB/olive oil mixture, and upon heating, the morphology changed to be uniform and needle-like. Therefore, it seemed that the 6.0 wt% BBB crystals in the 94.0 wt% olive oil solvent system allowed the selective crystallization of β-form crystals through an α-melt-mediated transition. It was clarified that the lump-type crystals were formed not only upon quenching, but also upon cooling to 5°C. In addition, there was a difference in the closeness of the crystal structures. Through a comparison of Figs. 5b and 5c, it became apparent that to obtain a gel-like behavior, it was necessary to form a strongly interacting network structure by mixing the lump-like β’ crystals and the needle-type β crystals, as shown in Figs. 2 and S1. The consideration that crystals of partially aggregated lumps could be derived from the β-form will be discussed later, based on the obtained polymorphic and thermal behaviors.

3.6 XRD observations of the BBB/liquid oil mixtures under different tempering conditions

The polymorphic behaviors of the crystals were observed using wide-angle XRD to determine the possible mechanism contributing to the sol- and gel-like behaviors. As shown in Fig. 6, the β and β’ crystal polymorphs of the 6.0 wt% BBB crystals in 94.0 wt% olive oil treated under the conditions of Tempering Procedures 2 and 3 were represented by weak short-spacing patterns of 0.46 and 0.41 nm, respectively, although the patterns were subtle. Weak short-spacing patterns of 0.38 and 0.37 nm corresponding to the β and β’ crystal polymorphs, respectively, were also observed. All other samples gave similar results, which can be consulted in the Supporting Information (Fig. S4). These properties may be related to the crystallinity (size and perfection) of the β-form crystals of BBB that exhibit a gel-like behavior. More specifically, the β-form crystals that

Fig. 5 In situ PLM images of the crystallization behavior of 6.0 wt% BBB crystals in 94.0 wt% olive oil as determined by hot-stage microscopy at Tc. (a–c) Under the conditions of Tempering Procedures 2–4, respectively. Scale bar = 50 μm.
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Fig. 6 XRD profiles of 6.0 wt% BBB crystals in 94.0 wt% olive oil. Solid line: under the conditions of Tempering Procedure 3, dotted line: under the conditions of Tempering Procedure 2.

exhibited a gel-like behavior were very small, as revealed by XRD, visual observations (Figs. 2 and S1), and PLM (Figs. 4 and S2). These results indicate that the crystals can form a gel-like fat mixture. Moreover, considering the result of XRD measurements (Figs. 6 and S4) in comparison with the results obtained by DSC (Fig. 3) and PLM (Fig. S5), the following can be considered.

We assumed that melting peak I was derived from the β'-form crystals, while melting peak II was derived from the β-form crystals. Furthermore, from the results of the PLM observations shown in Fig. S5, it was considered that melting peak I arose from the lump-like crystals, and melting peak II arose from the needle-like crystals. We therefore considered that the lump-type crystals were derived from the β' crystal polymorph, while the needle-type crystals were derived from the β crystal polymorph. In addition, the β and β' crystal polymorphs were distinctly observed in the X-ray diffractogram of the 6.0 wt% BBB crystals in 94.0 wt% olive oil that were treated under the conditions of Tempering Procedures 2 and 3, respectively. In this context, Callau et al. reported that a mixture of fatty acids and fatty alcohols with longer alkyl chains (i.e., behenyl alcohol and behenic acid) in sunflower and soybean oils formed oleogels, while Lupi et al. also reported that policosanol oleogels exhibit a gel-like behavior, which was attributed to the large number of interactions between the long alkyl chains of the fatty alcohols. Based on these studies, we assumed that BBB formed a closed-packed intermolecular system in oils due to the presence of numerous interactions between the long alkyl chains of behenic acid, thereby accounting for the formation of the β and β' crystal polymorphs. From the results of the PLM observations shown in Figs. 5 and S3, it was apparent that Tempering Procedure 3 led to needle-type crystals, while Tempering Procedure 2 gave lump-type crystals. These results suggest that the mixtures treated under the conditions of Tempering Procedure 3 possess a higher crystallinity and uniformity than those treated under the conditions of Tempering Procedure 2. These differences in crystallinity caused differences in the diffraction peak shapes and strengths, and as a result, sharper and stronger diffraction peaks were observed for the crystals treated using Tempering Procedure 3, as shown in Fig. 6.

From the results of the DSC, PLM, and XRD observations, we considered the mechanism responsible for the gel-like behavior. Due to the fact that a large number of α-form crystals were uniformly dispersed in the mixed fat and oil upon quenching, it was considered that the β-form crystals generated by melting of the α-form crystals would be uniformly dispersed in the mixed fat and oil, which could result in the construction of a crystal network structure in which the β-form crystals exhibited gel-like behavior. On the other hand, during crystallization by the slow cooling procedure, the number of β crystals formed was small, and they tended to be larger in size. As a result, the β crystals aggregated and dispersed in a state of separation from the liquid oils to give a sol state in the absence of a network structure.

3.7 Rheological analysis

To explore the effects of different cooling methods on the physical properties of the oleogels, dynamic rheological analysis (i.e., strain sweep, angular frequency sweep, and shear sweep) was carried out. The results for the BBB/olive oil (6.0/94.0 wt %) mixture are shown in Fig. 7, and those of the other samples, which gave similar results, are provided in the Supporting Information (Figs. S6 and S7). In terms of the strain sweeps at a fixed frequency of 1.0 Hz (Fig. 7a), the elastic modulus (G') was higher than the loss modulus (G''), confirming the formation of an elastic network of oleogels during cooling. It was also found that the tempering procedure influenced the physical properties of the oleogels. More specifically, the oleogel treated under the conditions of Tempering Procedure 3 was initially stiff with a shear modulus of ~1000 Pa, and a limited linear viscoelastic plateau was observed until the oscillation stress reached ~5 Pa. Above the critical strain level, G' and G'' decreased sharply, and exhibited crossover around an oscillation stress of ~10 Pa. In comparison, the oleogels treated under the conditions of Tempering Procedure 2 did not exhibit a linear viscoelastic plateau, with its elasticity decreasing initially with crossover at an oscillation stress of ~5 Pa. The frequency sweep shown in Fig. 7b demonstrates that the oleogels are viscoelastic materials and exhibit solid-like behavior. For most samples, G' was higher than the loss modulus G'', and the crossover where the rheo-
ological behavior shifted from the elastic state to the viscous state seemed to occur at particularly low frequencies (i.e., <0.01 Hz). In the oleogels treated under the conditions of Tempering Procedure 3, $G'$ and $G''$ increased gradually until the frequency reached approximately 10 Hz, and then increased rapidly over the measured range. In addition, the $G''$ values were always smaller than the $G'$ at frequency over the measured range. The behaviors of $G'$ and $G''$ were the same as that of the typical gel state; in general, the $G'$ and $G''$ values in the gel state are larger than those in the liquid state, and $G'$ tends to be larger than $G''$. In the liquid state, $G'$ and $G''$ increase linearly with increasing frequency, where $G''$ dominates the viscoelasticity parameter of the liquid state. In the oleogels treated under the conditions of Tempering Procedure 2, it was found that $G'$ and $G''$ increased with increasing frequency, although their values were smaller than those achieved under the conditions of Tempering Procedure 3. Furthermore, $G'$ dominated the viscoelasticity parameter, corresponding to a sol-like behavior. As shown in Fig. 7c, the viscosity of the oleogels decreased with an increase in the shear rate, thereby reflecting their shear-thinning characteristics. The oleogels obtained under the conditions of Tempering Procedures 2 and 3 possessed similar viscosities, although the viscosity of the oleogel obtained using the Tempering Procedure 3 was slightly higher. In general, the high gel rigidity accounts for its high storage stability, which is of particular importance in the contexts of industrial food products and cosmetics, which are often stored for long periods after preparation. It is interesting to note that the difference in the cooling rate influences the crystallization, polymorphism, rheological, and morphological properties of the prepared oleogels. In this context, we note that Higaki et al. reported that FHR-B formed a specific crystal size and shape, and exhibited $\beta$-polymorphism upon careful control of the cooling rate. In our case, it is possible that the $\beta$-polymorphic crystals in the BBB and liquid oil mixtures formed a dense network structure. It was considered that a uniform gel state would be formed by maintaining the state of holding the liquid oil. As shown by the results of the PLM observations (Figs. 4, 5, S2, and S3), smaller crystals were formed during rapid cooling, thereby resulting in a significant interaction between the crystals, which then contributed to a higher network rigidity. The observed differences were therefore attributed to the rheologies of the BBB crystals formed by the different tempering procedures with varying cooling rates.

3.8 Stability

For the oleogels prepared under the conditions of Tempering Procedure 3, no visible changes were observed over the course of a six-month storage at 25°C. The elastic
modulus ($G'$) and loss modulus ($G''$) were also relatively constant over this period. In this context, the stress curves for the BBB/olive oil (6/94.0 wt %) liquid mixtures are presented in Fig. 8, while those of the other samples, which showed similar results, are provided in the Supporting Information (Figs. S8 and S9).

These results and the observed stability over 180 d for all oleogels at 25°C were attributed to the formation of a strong crystal network structure in the mixtures. From the results of the PLM and XRD observations presented in Figs. 5, 6, S3, and S4, respectively, it could be concluded that a large number of needle-type β crystals are uniformly dispersed, and that a β-form crystal network structure holds the liquid oil strongly within the mixtures. Therefore, it was considered that the gel structure should be stabilized even after 180 d.

4 Conclusion

In this study, the crystallization and gelling activity of tribehenoyl-glycerol (BBB) were explored through the preparation of oleogels composed of edible oils (olive oil, squalene, and caprylic/capric triglyceride) and high-melting fat crystals (BBB) to explore the potential use of BBB and edible oil mixtures as low-cost and stable gelators. It was found that BBB exhibited polymorphic behavior in the crystalline state, with both α- and β-forms being present.

We also observed that the rapid cooling of BBB/liquid oil mixtures led to the crystallization of BBB in the α-form, while subsequent transformation to the β-form was achieved via an α-melt-mediated transformation. This thermal treatment resulted in the formation of oleogels. From an applied aspect, the present study provides information that could be employed to improve the properties of oleogels through variation in the processing conditions, such as the cooling rate and temperature. From a physical stability point of view, the prepared oleogels demonstrated good stabilities (i.e., no oil drainage, and constant rheological properties) over 6 months. Based on our results, it is therefore expected that oleogels of BBB crystals in liquid oils should be suitable for storage above refrigerator temperatures for significant periods of time. These two factors are particular importance in the development of commercial applications for oleogels in foods and cosmetic products. We hope that this work will pave the way for novel technological developments, especially in the food and cosmetic arena, where this type of material is highly sought after.

Supporting Information

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