Technical Report

Project title: Development of Soybean Oil Oleogels for Edible Applications

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1. Introduction

Cookies are one of the most popular snacks which generated about 30.62 billion dollars in transactions in the United States in 2018 and the trend of cookie market size is keeping increasing in the following years. The reasons why cookies are so popular include their special tastes, easy to eat feature, relatively moderate cost and long shelf life, and so on. Cookies are made by a variety of ingredients include flour, sugar, fat, baking soda, water, nuts, and chocolate. Among these ingredients, fat is a basic ingredient that not only quantitatively important but also has extensive functional properties. In general, fat provides lubrication and plasticity to the cookie dough, and influence the amount of air cells, the final texture, flavor, and shelf life. For most bakery products, shortening is the fat product which has been widely used for decades. The major ingredients to prepare shortening is partially hydrogenated oil (PHO). However, as the primary source of artificial *trans*-fat, PHO has been banned by the FDA due to the deleterious effects of trans-fat. Since PHO plays several important roles in shortening, it is very hard to find a desirable replacement for removing PHO in shortening. Due to the difficulty of fully remove PHO from the market, FDA extends the compliance date from June 18, 2018, to January 1, 2021. It is thus of great urgency to develop novel technique to fill the gap and guarantee the safety of consumers.

We set forth to develop solid-like soybean oil that can be used to replace partially hydrogenated soybean oil in bakery industry. Based upon the findings we received from the previous year study, we attempted to fabricate semisolid oleogels using crude soybean oil or high oleic soybean oil. Three objectives were pursued to achieve the goal: (i) comparing the physical, chemical, and rheological properties of oleogels prepared by two different soybean oil sources; (ii) elucidating the role of chemical differences of soybean oils types, i.e., crude, refined, and high oleic soybean oil on the physical properties of oleogels; and (iii) evaluating the effect of oleogels on the physical property of cookie dough and baked cookies.

2. Research methods

Preparation of soybean oil oleogels: We applied β-sitosterol (BS) or monoacylglycerol (MAG), or the combination of both as gelator to prepare commercial and crude regular soybean oil oleogels. This part of the research has been published on ***Food & Function*** journal and the results can be found at (https://pubs.rsc.org/en/content/articlehtml/2020/fo/c9fo02180a). In addition, we also used sunflower wax to fabricate expeller-pressed high oleic soybean oil oleogels and the results will be discussed hereafter.

Characterization of soybean oleogels: The thermal and physical characteristics of oleogels are investigated using differential scanning calorimetry (DSC), rheometer, x-ray diffraction (XRD), polarized light microscopy (PLM), and texture analyzer.

Cookie making: As a common aerated baked good, cookies are made with oleogels and the physical properties of which were compared with that made with commercial vegetable shortenings.

Microstructure of cookies with soybean oleogels: Confocal laser scanning microscopy was applied to visualize the microstructures of cookies made by the oleogels.

3. Research results

1) Color of high oleic soybean oil oleogels

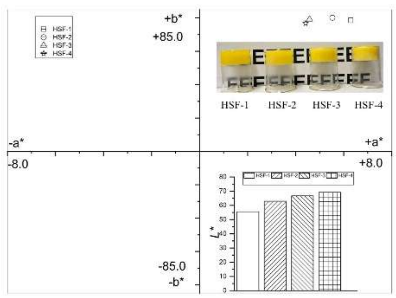

Fig. 1 The appearance of high oleic soybean oil oleogels

Four different concentration of sunflower wax (3, 5, 7, and 9%) were used to fabricate expeller-pressed (EP) high oleic soybean oil oleogels (HSF). 1% of gelator was unable to solidify soybean oils. It was obvious that the oleogel formed by EP oil was yellower which may due to the presence of pigment components such as carotenoids in the EP oil (**Fig.1**).

2) Thermal properties of soybean oleogels

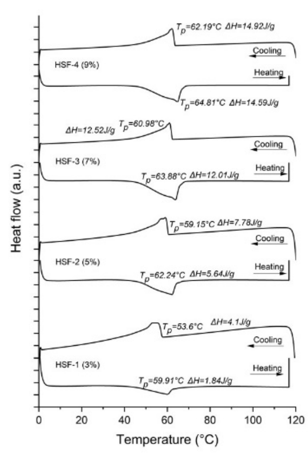

Fig. 2 DSC heat flow curves for high oleic soybean oil oleogels

Thermal behavior of oleogels including melting and crystallization behaviors are critical parameters when considering the potential application of oleogels in the food system. EP high oleic soybean oil oleogels have a melting point between 54.24°C and 62.22°C with an enthalpy between 4.26 J/g and 15.30 J/g (**Fig. 2**). The oleogel prepared with a greater concentration of sunflower wax has a higher endothermic and exothermic peak with higher enthalpy.

3) Viscoelastic properties of soybean oleogels

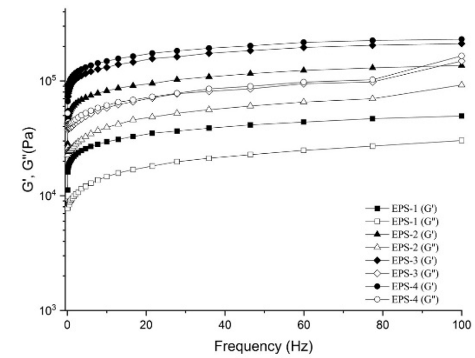

Fig. 3 Frequency dependence of (A) storage modulus (G'), and (B) loss modulus (G") for EP high oleic soybean oil oleogel at 25 °C

In general, oscillatory measurements could be used to distinguish gel and sol status and measure the deformation and flow properties of samples. It is clear that the strength of oleogel is gelator concentration dependent (**Fig.3**). As more sunflower wax is added into the oil phase, both G' and G" increase indicating stronger gel which would be more resistant to deformation. EPS-4 had a G' about 10^5 which was about 10 times higher than EPS-1, indicating the great influences of gelator concentration on the strength of oleogel.

4) Polymorphism of soybean oleogel

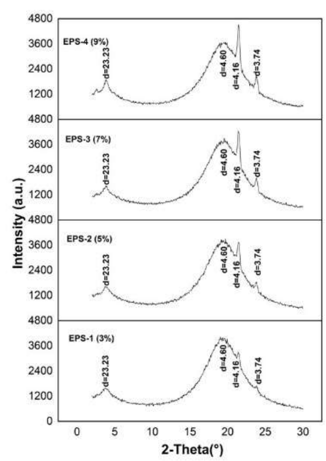

Fig. 4 XRD patterns of soybean oil oleogels

Crystal structure is very important when considering the actual application of solid fat products (**Fig.4**). All the EPS oleogels had two peaks at 4.16 Å and 3.74 Å. When comparing the amount of peak based on wax concentration, EPS oleogels were not affected. This result indicated that EPS could produce oleogel that had β' crystal at a sunflower wax concentration ⩾ 3% wt.

5) Crystal morphology of soybean oil oleogels

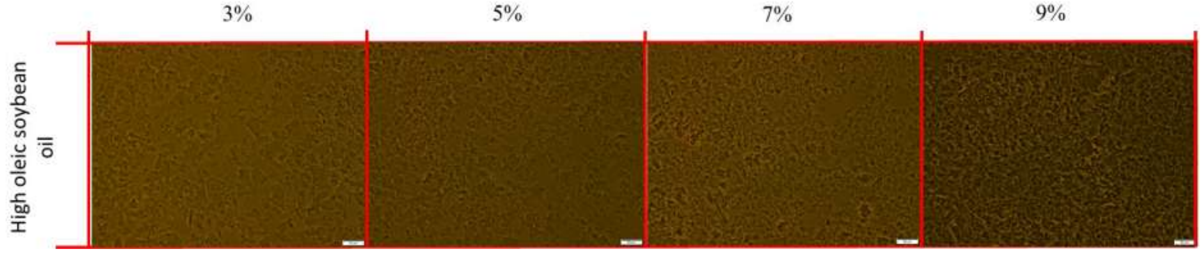

Fig.5 Polarized light microscopy images of soybean oil oleogels (scale bar = 20 μm).

In order to have a better visualization of crystal morphology of oleogel, polarized light microscopy (PLM) was used to get more information about oleogel structure (**Fig.5**). All the oleogel fromed thin platelet-like crystals. In addition, more yellow background could be seen in the micrograph for oleogel prepared with a lower concentration of sunflower wax. This result indicated the addition of gelator increased the formation of crystals in the oleogel, thus, harder and stronger gel was produced by higher amount of gelator.

6) Replacement of commercial shortening using soybean oleogels in cookie making

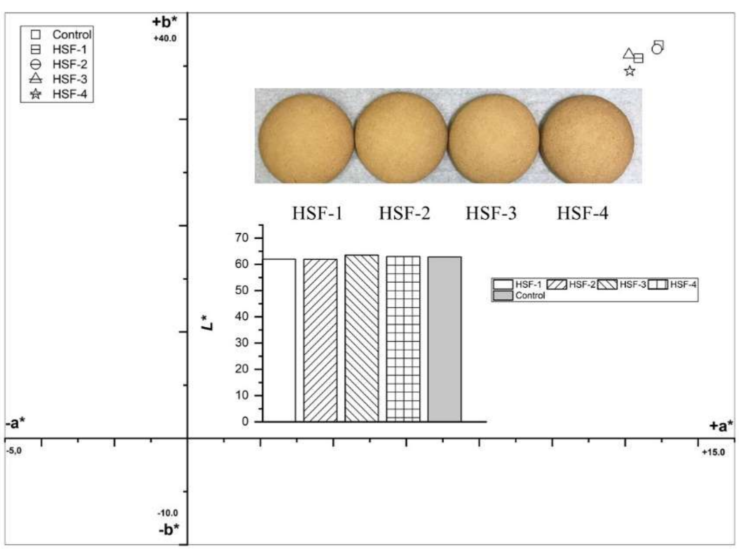

Fig. 6 Cookies prepared by high oleic soybean oil oleogels

We determined the appearance and colorimetric results for cookies prepared with EP high oleic soybean oleogels and shortening (**Fig.6**). In general, the lightness of oleogel cookies was slightly lower but still very close to the control cookie. Although the EP high oleic soybean oleogels are much yellower than shortening, all the cookies showed similar yellowness and redness tone which indicated that the color of oleogel did not have great effects on the color of cookies.

7) Texture analysis of the soybean oleogels and the cookies prepared using soybean oleogels

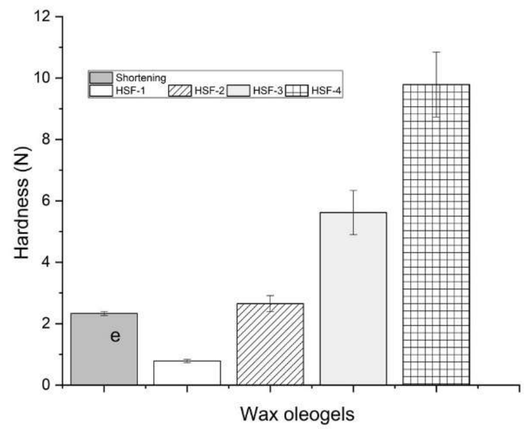

Fig. 7 Hardness of (A) soybean oil oleogels and (B) cookies made by oleogels

All the oleogel cookies were harder than control cookie with no statistically significant differences were found between EPS-4 cookie and control cookie (**Fig.7**). The control cookie had a hardness of 81.75 N while EPS-4 cookie had a similar hardness of 88.29N, and the hardness of EPS oleogel cookies decreased as the gelator concentration increased.

8. Microstructure of cookies made by high oleic soybean oleogels

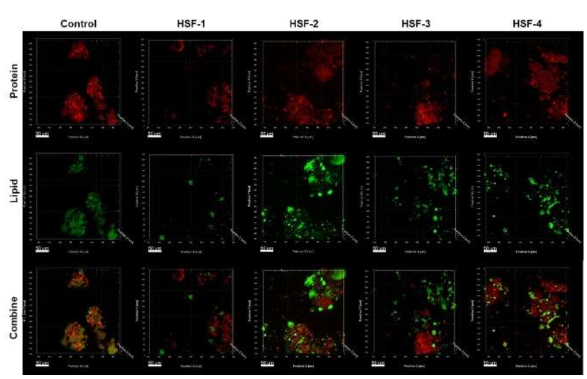

Fig. 8 CLSM result of cookies prepared with shortening and EP high oleic soybean oil oleogels

To understand the lipid distribution in cookies, CLSM was used to visualize the microstructure of cookies (**Fig.8**). Unlike normal microscope, the laser of CLSM is strong and easy to penetrate thick samples which makes it is an effective technology to measure low-moisture foods because most of them are crumbly and hard to cut into thin pieces.

CLSM indicated that proteins and lipids were associated with each other and dispersed evenly in control cookies. It could be noticed that there was almost no single lipid droplet in control cookies, almost all the lipid components were highly associated with protein components. In general, all the oleogel cookies especially EPS-2 and EPS-3 showed similar patterns to the control cookies where most lipids were combined with proteins. The similar lipid distribution may be one of the reasons that EP high oleic soybean oil oleogel cookies have comparable properties compared to control cookies.

4. Conclusion

The physical properties of oleogel prepared with expeller-pressed high oleic soybean oil with the incorporation of sunflower wax at different concentration (1, 3, 5, 7, and 9%) were evaluated. Liquid soybean oil could form yellow oleogel at a gelator concentration $\geqslant$ 3% wt. An increased melting point, crystallization point, and corresponding enthalpy was observed as the concentration of gelator increased. Other physical characteristics included rheological, hardness, crystal structure also showed a dependence on the concentration of gelator. An increase in the addition level of gelator resulted in stronger oleogel. The potential application of EP high oleic soybean oil oleogel was investigated by replacing shortening in cookie production. All the EP oleogel cookies showed similar properties to cookies prepared with commercial shortening. EP soybean oil oleogels had better performance in cookie production. Also, the microstructure of EP soybean oil oleogel showed similar lipid and protein distribution to the control cookies. For hardness, EP soybean oil oleogel cookies were softer as the concentration of gelator increased. Overall, EP high oleic soybean oil oleogels were able to produce stronger oleogel and the results showed a great possibility of using EP high oleic soybean oil oleogels to replace shortening in real food processing.