Received: 27 July 2023 Revised: 16 October 2023 Accepted article published: 16 November 2023 Published online in Wiley Online Library:

(wileyonlinelibrary.com) DOI 10.1002/jsfa.13137

Microfluidization improved hempseed yogurt's physicochemical and storage properties

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Abstract

BACKGROUND: Plant-based yogurts are suffering from the common problems, such as an unattractive color, stratified texture state and rough taste. Therefore, it is urgent to develop a novel processing method to improve the quality and extend the storage life of hempseed yogurt. In the present study, hempseed yogurt was microfluidized prior to fermentation. The effects of microfluidization on microstructure, particle size, mechanical properties, sensory acceptability, variations in pH and titratable acidity, lactic acid bacteria (LAB) counts, and stability of hempseed yogurt during 20 days of storage were investigated.

RESULTS: Microfluidization contributed to the production of hempseed yogurt as a result of the better physicochemical properties compared to normal homogenization. Specifically, microfluidization reduced the particle size of hempseed yogurt with a uniform particle distribution, increased water holding capacity, and improved texture and rheological properties. These advancements resulted in higher sensory scores for the yogurt. Furthermore, during storage, microfluidization effectively inhibited the post-acidification process of hempseed yogurt, and increased LAB counts and storage stability.

CONCLUSION: Microfluidization improved the physicochemical properties and storage stability of hempseed yogurt. Our findings support the application of microfluidization in hempseed yogurt and provide a new approach for enhancing the quality of plant-based alternatives that meet consumers' demands for high-quality food products.

Keywords: hempseed yogurt; microfluidization; physicochemical properties; storage stability

INTRODUCTION

Plant-based yogurts are usually made from fermented grains, legumes and nut flours.[1] The market for plant-based yogurt alternatives has been growing in recent years, and they are considered as healthy, natural and low-calorific, being particularly popular among young consumers.[2] The nutritional value of plant-based yogurts depends on the raw material. Grains can provide major macronutrients and micronutrients. Pseudo-cereals and legumes are suitable alternatives to animal proteins, characterized by rich high-quality proteins, bioactive compounds and fibers.[3] Hemp (*Cannabis sativa* L.) has hundreds of potentially biologically active compounds that can be used in food, feed, clothing and chemical industries.[4] Hempseed is rich in carbohydrates, oils, vitamins, proteins and minerals,[5] and is widely used in dairy products. In a previous study, 10% hempseed protein was added to soy yogurt and increased physicochemical properties, the number of lactic acid bacteria (LAB) and the desirable volatile component of the yogurt.[6] Wang *et al.*[7] subjected hemp milk to high pressure homogenization and pH displacement. They found that the structure of the hemp milk became more stable and the microbial population was significantly reduced, with no stratification during 3 days of storage at 4 °C. Hashemian and Nouri[8] found that the addition of hempseeds enhanced the phenolic component and antioxidant properties of camel yogurt, with a significant decrease in saturated fatty acids and an increase in unsaturated fatty acids.

Many manufacturers are developing plant-based dairy alternatives. Compared to fermented milk yogurts, plant-based alternatives have unsatisfied sensory attributes, stability and functionality, thus hindering their widespread use.[9] Moreover, yogurt is prone to whey precipitation, severe post-acidity and the generation of an unfavorable flavor during storage and transportation, adversely affectng the quality and sales of the product. Therefore, there is an urgent need to apply technical ways to improve the quality and storage stability of plant-based yogurts. Currently, the main methods to extend storage period of yogurt include the addition of food preservatives (such as antimicrobials, antioxidants and anti-browning agents), post-pasteurization and use of novel processing technology, etc. Microfluidization combines water jet technology, impinging stream technology and high pressure homogenization, with a working pressure up to

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30 000 psi, to create a distinctive environment of high pressure, high velocity, vibration, pressure drop, intense shear rate and hydrodynamic cavitation.[10] Microfluidization has been applied to colloidal products to change physical, chemical and microstructural properties, including an improved water holding capacity (WHC), apparent viscosity values and colloidal stability, but reduced microbial load.[11] Recently, microfluidization has been used in food processing and is generally considered as a suitable method for milk homogenization. For example, milk treated with microfluidization had a transparent layer of fat globules of approximately 10 nm in thickness instead of huge micellar clumps produced by the conventional homogenization, and the thickness decreased significantly with increasing pressure.[12] The larger fat globules created by microfluidization were coated with smaller fractions through protein bridges, and some of the tiniest fat globules were embedded within casein micelles.[13] Li *et al.*[14] produced whole soybean milk using an industrial-scale microfluidizer that was stable for storage at least 21 days at 4 °C without the addition of any stabilizer or emulsifier.

Although microfluidization has been widely used in dairy research, less research has focused on its application to hempseed fermented products to solve quality problems. In addition, it is necessary to access the physicochemical properties of plant-based yogurts, such as the appearance, texture and stability, which have great impacts on their processing, storage, quality attributes and functional performance.[15] Therefore, the present study investigated the effects of microfluidization on sensory quality, microstructure and physicochemical properties of hempseed yogurt to provide theoretical supports for the development of hempseed fermented products.

MATERIALS AND METHODS

Materials

Soymilk powder made of soybean and malt syrup was received from Dragon Inc. (Heilongjiang, China). Hempseed protein, containing 7.1 g of total sugar, 7.4 g of dietary fiber, 14.8 g of total carbohydrate, 74.0 g of protein and 2.2 g of fat per 100 g, was received from Liaoning Qiaopai Biotech Co. Ltd (Liaoning, China). Sugar was purchased from Liaoning Zhongqi Agricultural Products Co., Ltd (Liaoning, China). Pectin was supplied by Xinjiang Fufeng Biotechnologies Co., Ltd (Xinjiang, China). Diacetyl tartaric acid ester of mono(di)glycerides was purchased from Henan Honest Food Co., Ltd (Henan, China). Agar was purchased from Shishi Globe Agar Industries Co., Ltd (Fujian, China). Active Ingredient distarch phosphate was purchased from Henan Wanbang Chemical Technology Co., Ltd (Henan, China).

Streptococcus thermophilus HCS07-002, *Bifidobacterium lactis* HCS04-001, *Lactobacillus plantarum* HCS03-084 and *Lactobacillus bulgaricus* HCS10-001 were donated from Hanchen's Children's Products Co., Ltd (Liaoning, China). Nile Blue was sourced from Beijing BoaoTuoDa Technology Co., Ltd (Beijing, China). NaOH was provided from Shanghai Aladdin Biochemical Technology Co., Ltd (Shanghai, China).

Preparation and storage condition of hempseed yogurt

Hot water was used to dissolve the soymilk powder (15% w/v), hempseed protein (10% w/v), sugar (7% w/v) and stabilizer (0.3% w/v). Then, the mixture was homogenized (AD300L-H; Shanghai Angni Instruments, Shanghai, China) at 8000 rpm for 10 min and filtered by a double gauze. The mixed slurry was passed through an 80-MPa NanoGenizer (Suzhou Microstream Nanobiotechnology, Suzhou, China) and sterilized at 90 °C for 10 min. After cooling to room temperature, the mixture was quickly inoculated with 0.8% v/w bacterial solution (*S. thermophilus* HCS07-002: *L. bulgaricus* HCS10-001: *B. lactis* HCS04-001: *L. plantarum* HCS03-084 = 1:1:1:1) to ferment at 42 °C for 8 h. Hempseed yogurts were kept at 4 °C for 20 days before being analyzed. Samples without microfluidization were referred to as control yogurt (CY) and samples after microfluidization were referred to as NanoGenizer (NG).

Particle size

Hempseed yogurt was dropped into the sample jar of a laser particle size analyzer (BT-9300ST; Baxter, Shanghai, China). The particle distribution was determined in the range of 0.1–1000 μm using water as dispersant at refractive indices up to 5%.[16]

Optical microscope and confocal laser scanning microscopy

A drop of yogurt sample was placed on a slide, covered with a coverslip, and placed under an inverted light microscope (BDS400; Chongqing Aote Optical Instrument, Chongqing, China) with a 40× objective lens.

Hempseed yogurt (1 mL) was stained with 100 μL of Nile Blue for 30 min, placed on a slide, and sealed with a coverslip and silicone oil. A confocal laser scanning microscope (LSM 510 META; Zeiss, Oberkochen, Germany) with a 40× objective was used to observe the protein structure of hempseed yogurt samples at 633 nm.[17]

WHC

WHC was measured as previously described[18] with slight adjustments. Briefly, the sample was weighed and centrifuged at $5000 \times g$ for 15 min. The precipitate was then weighed, and the WHC was calculated using:

$$\mathrm{WHC}\,(\%) = \frac{m_2}{m_1} \times 100 \tag{1}$$

where m_1 represents the sample weight before centrifugation and m_2 represents the precipitate weight after centrifugation.

Textural properties

The textural properties of hempseed yogurt were analyzed using a texture analyzer (TA-XT Plus; Stable Micio Systems, Godalming, UK) with a cylindrical P/0.5 probe (diameter 12.7 mm). The descent rate was 1 mm s^{-1}; the test and return rates were 2 mm s^{-1}; the strain was 30%; the relaxation time was 5 s; and the trigger force was 5 g.[6] Hardness, consistency, elasticity and cohesiveness were calculated using Texture Exponent Lite 32, version 4.9.8.0 (Stable Micio Systems).

Rheological properties

A rotational rheometer (Discovery HR-1; TA Instruments, New Castle, DE, USA) with a 40-mm parallel plate was used to determine the rheological characteristics according to Liu *et al.*[19] The apparent viscosity was measured at a shear rate of 0.1–100 s^{-1}, then fitted with the Herschel–Bulkley model. The model is represented by:

$$\tau = \tau_0 + \kappa \gamma^n \tag{2}$$

where τ is the shear stress, τ_0 is the yield stress, κ is the consistency coefficient, γ is the shear rate and n is the flow behavior index.

The dynamic rheology was scanned at an angular frequency of 0.1–100 rad s^{-1} ($\gamma = 2\%$) to estimate the elastic modulus (G'), viscous modulus (G'') and loss angle tangent (tan δ) of hempseed yogurts.

Sensory evaluation

According to Kim *et al.*,[20] 20 students or teachers (including 10 males and 10 females) aged between 20 and 45 years who had received sensory training before were convened to score the sample. The evaluation scale ranges from 1 (highly disliked) to 6 (strongly liked), covering five different categories: appearance, scent, texture, taste and overall acceptability.

pH and titratable acidity

A pH meter (PHS-3TC; Shanghai Precision Instrument, Shanghai, China) was used to measure pH values.

Hempseed yogurt (10 g) was mixed with 20 mL of distilled water and two drops of phenolphthalein, and then titrated with 0.1 M NaOH.[21] Titratable acidity was calculated as:

$$^{\circ}T = \frac{c \times V \times 100}{m \times 0.1} \tag{3}$$

where c is the concentration of NaOH, V is the volume of NaOH consumed for titration and m is the mass of the sample.

LAB counts

According to Peng *et al.*,[22] 25 g of hempseed yogurt was mixed with 225 mL of saline and gradually diluted to a suitable concentration. The diluted samples were pipetted onto MRS Agar and incubated at 37 °C for 72 h to record the total number of colonies.

Apparent stability and storage stability

Hempseed yogurt was observed and photographed at 0, 5, 10, 15 and 20 days during storage.

The inoculated mixture was fermented in a special Turbiscan test flask and scanned in a Turbiscan stability analyzer (Lab Expert; Formulaction, Toulouse, France).[23] Turbiscan stability index (TSI) was calculated with TurbiSoft (https://turbisoft.software.informer.com) using:

$$TSI = \sqrt{\frac{\sum_{i=1}^{n}(x_i - x_{BS})^2}{n-1}} \tag{4}$$

where x_i and x_{BS} are backscattered light values, and n is the scanning time.

Statistical analysis

Each group of experiments was repeated three times. Data are expressed as the mean ± SD. An independent samples *t*-test was used to compare the differences between CY and NG. $P < 0.05$ was considered statistically significant. The results were analyzed with SPSS, versus 26 (IBM Corp., Armonk, NY, USA) and plotted graphically with Origin 2019 (OriginLab, Northampton, MA, USA).

RESULTS AND DISCUSSION

Effects of microfluidization on the particle size and distribution of hempseed yogurt

Particle size influences the physicochemical characteristics and sensory qualities of the yogurt, such as appearance, texture, taste, etc.,[24] As depicted in Figure 1(a), CY had a multi-peaked distribution with an unstable state, whereas NG had a single-peaked distribution with the particle peak at 8 μm. Microfluidization significantly decreased the $D_{4,3}$ value of hempseed yogurt from 15.19 ± 0.29 μm to 6.44 ± 0.39 μm (Figure 1b). Microfluidization can disrupt protein micelle aggregates in the pre-fermentation mixture, thus achieving homogeneous dispersion of the components and reducing the particle size distribution.[25] Ronkart *et al.*[26] measured the particle size of microfluidized inulin–water system gels and found that microfluidization treatment reduced particle size and formed a network of agglomerates. With respect to improving the storage stability of soymilk, Mukherjee *et al.*[27] found that combining ultra-high-pressure homogenization with 0.05% κ-carrageenan increased surface energy and absolute ζ-potential, and also reduced the mean spherical particle size of soymilk.

Effects of microfluidization on the microstructure of hempseed yogurt

By observing food microstructure, we can better understand its rheological properties, WHC and organoleptic properties.[28] There was a difference in the network density of hempseed yogurt before and after microfluidization (Figure 1c). The hempseed protein particles were large and aggregated in CY, whereas the proteins were relatively small and uniformly distributed in NG. The result was further confirmed by optical microscopy (Figure 1d). Microfluidization combines water jet technology, impinging stream technology and high pressure homogenization technology to propel a liquid through a well-designed chamber.[10] Ozturk and Turasan[29] reported that microfluidization disintegrated the large hydrophobic structures to form micro- and nanoparticles, and even modified the smooth surfaces into branched networks by forming tissues, cavities and micropores through the structure. Bernat *et al.*[30] found that high-pressure homogenization reduced droplet size and promoted protein solubilization, thus modifying the microstructure of almond milk. Luo *et al.*[31] discovered that the quinoa protein isolate gels with high-pressure homogenization at a pressure of 30/50 MPa had more homogenous, finer and denser protein networks compared to the control group. Microfluidization effectively reduced the particle size and formed a homogeneous distribution of proteins after microbial fermentation.

Effects of microfluidization on the WHC of hempseed yogurt

The WHC is used to assess the interactions between water and protein, which affects the viscosity and texture of foods.[32] As shown in Figure 2(a), a significant higher WHC value was observed in NG (71.68 ± 3.05%) compared to CY (63.68 ± 2.31%), confirming that microfluidization improved the WHC of hempseed yogurt. The WHC of yogurts has close connections with the internal network structure.[33] The formation of a robust gel network in NG might be a result of microfluidization that ruptures protein micelles in the pre-fermentation mixture, thereby increasing the quantity of protein particles and increasing the total surface area accessible for interaction during fermentation.[34] The improved WHC by microfluidization was consistent with previous research.

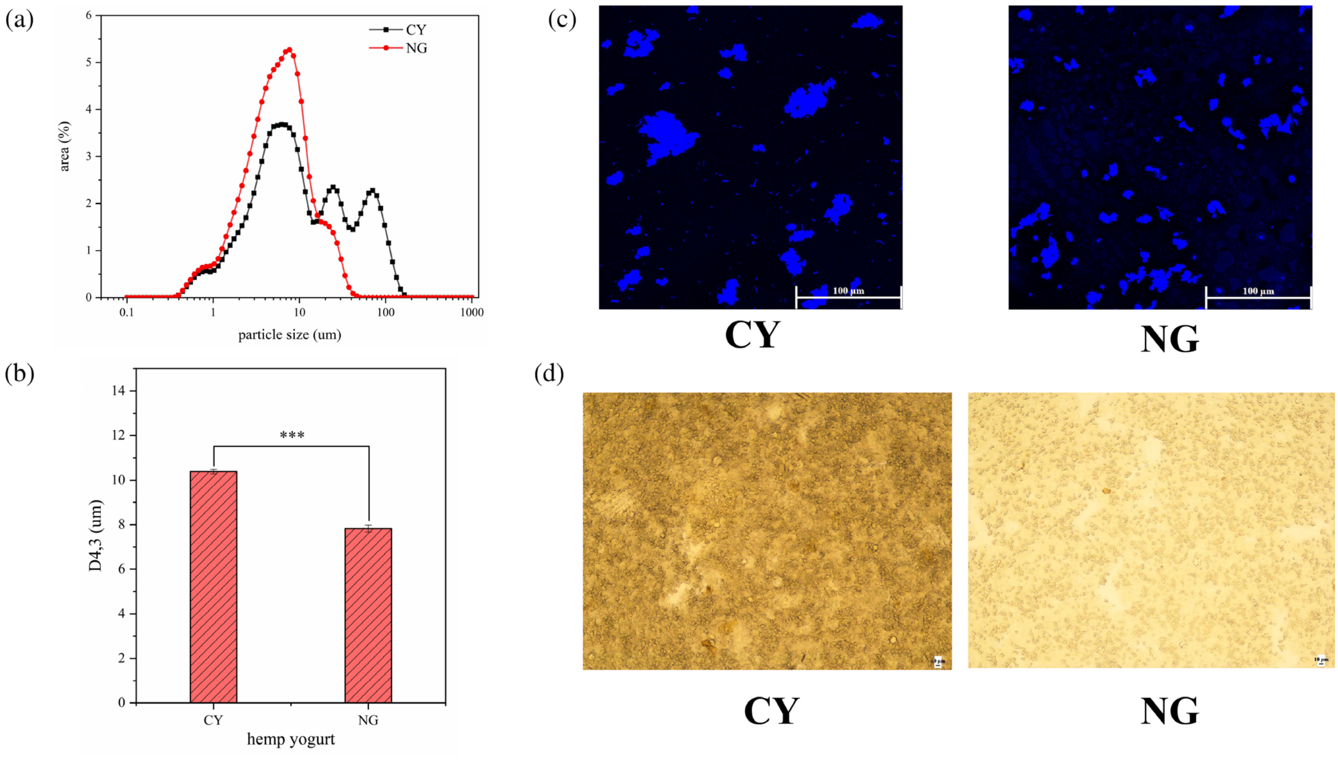

Figure 1. Effects of microfluidization on (a) particle size distribution, (b) volume surface mean diameter $D_{4,3}$, (c) confocal laser scanning microscope images, and (d) optical microscope images of control yogurt (CY) and yogurt processed with NanoGenizer (NG). $^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.001$.

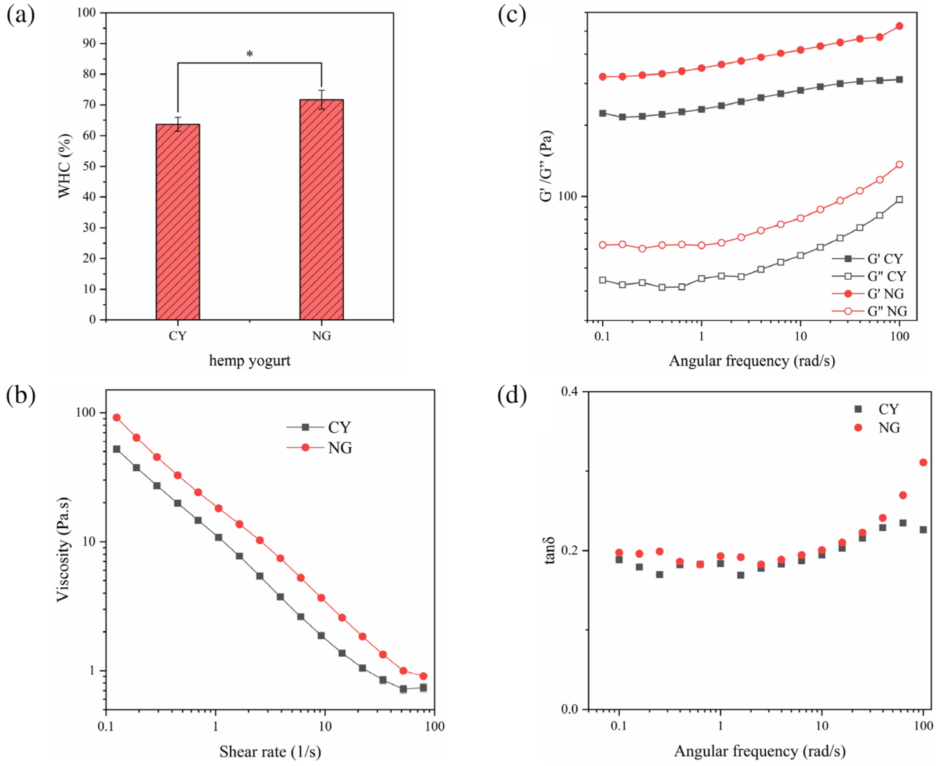

Figure 2. Effects of microfluidization on (a) WHC, (b) apparent viscosity, (c) storage modulus G' and loss modulus G'', and (d) loss tangent (tan δ) of control yogurt (CY) and yogurt processed with NanoGenizer (NG). $^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.001$.

Ge *et al.*[35] found that the WHC of *Eucommia ulmoides* Oliv. seed meal proteins was enhanced by microfluidization in acidic circumstances. Adjei-Fremah *et al.*[36] concluded that microfluidization altered the hydration characteristics of whole cowpea flour, with a significant increase in WHC.

Effects of microfluidization on the textural properties of hempseed yogurt

Texture is an important indicator of yogurt quality, including hardness, consistency, elasticity, cohesiveness, etc. As shown in Table 1, the hardness, consistency and elasticity of NG were significantly higher than that of CY, increased by 33.60%, 48.80% and 0.21%, respectively. However, there was no significant difference in cohesiveness between NG and CY. Hardness, as a measure of yogurt firmness, refers to the force required to induce a certain deformation.[37] A previous study found that the force-induced cavitation phenomenon, higher shear and turbulence generated by dynamic high-pressure homogenization disrupted the non-covalent interactions of whey protein isolate,[38] resulting in the exposure of some reactive groups that caused more intermolecular interactions influencing the gel hardness.[39] Particle size and its distribution influenced the consistency of plant-based food dispersions.[40] During microfluidization, the turbulence, shear and cavitation processes reduced the particle size thereby resulting in higher product consistency. It was also found that microfluidization affected the surface tension and flexibility of pea albumin aggregates[41] and ovalbumin[42] as a result of the reduced particle size and unfolded protein structure. Overall, microfluidization contributed to the improvement of the textural properties of hempseed yogurt.

Effects of microfluidization on the rheological properties of hempseed yogurt

Rheological properties characterized the deformations and flows of the substance under a certain stress rate.[43] A study on food rheology and microstructure enables to minimize food defects and improve consumer satisfaction.[44] According to pseudoplastic fluid characteristics, the apparent viscosity of hempseed yogurts decreased with the increase of shear rate (Figure 2b). The result was in line with previous yogurt research.[45] Microfluidization caused changes in protein structure, mainly denaturation and aggregation, and exposed more hydrophilic groups, thus creating more water-binding sites and a stronger protein gel network.[46] The apparent viscosity of NG was consistently higher than that of CY. This might be a result of the reduced protein macromolecule particles and stretched peptide chains caused by microfluidization, which exposes numerous chemical bonds and forms a stable protein network. The Herschel–Bulkley model was used to fit the apparent viscosity, and the results are presented in Table 2. The fitted correlation coefficient $R^2 > 0.97$ suggested that the model accurately captured the sample's rheological characteristics during the shear scan. The flow behavior index $n < 1$ indicated that the hempseed yogurt was a pseudoplastic fluid and shear-thinning fluid.[47] The yield stress τ_0 of NG was lower than that of CY, suggesting that the fluidity of hempseed yogurt was improved by microfluidization. The consistency factor κ of NG was higher than that of CY, suggesting a stronger molecular interaction by means of microfluidization.[48] Therefore, microfluidization improved the fluidity and consistency of hempseed yogurt, resulting in a smoother texture.

Frequency scans characterized the viscoelastic properties of yogurt products.[49] The gel network's elastic behavior is represented by the storage modulus (G') and the viscous behavior is represented by the loss modulus (G'').[50] As shown in Figure 2(c,d), the storage moduli of all hempseed yogurts were higher than the corresponding loss moduli, and both variables enhanced with the increase of angular frequency. The results indicated that hempseed yogurt was primarily elastic rather than viscous in nature, which stayed generally stable at low frequencies and displayed a solid-like quality.[51] The tan δ value, which is the ratio of G'' to G', being less than 0.4 confirmed the above results. Microfluidization increased the storage modulus and loss modulus of hempseed yogurt, showing strong viscoelastic properties. The moduli are correlated with the amount of covalent bonds in the gel network, and thus with the gel strength.[52] Microfluidization may decompose the large protein molecules into small particle structures in hempseed yogurts, which helps to form a homogeneous and dense gel network structure. As a result, the gel exhibits enhanced tensile strength, imparting the hempseed yogurt with great viscoelasticity.[34]

Effects of microfluidization on the sensory of hempseed yogurt

Sensory assessment is an experimental technique that evaluates and interprets the responses to foods through sight, smell, touch, taste and hearing.[53] As shown Figure 3(a), the scores of appearance, texture and overall acceptability were greatly improved in NG, reaching 5.37, 5.63 and 5.53, respectively. According to volunteer feedback, hempseed yogurt with microfluidization process had a smooth surface with no whey precipitation. This was probably a result of the disintegration of protein micelles caused by microfluidization, resulting in a denser and more uniform protein gel network structure.[54] Therefore, the yogurt had a non-grainy and fine texture with high acceptability. An earlier study concluded that microfluidization improved the perception of buttermilk and soft cheese flavors, as well as the flavor of natural yogurt.[55] Although there was not an obvious difference in flavor

Table 1. Textural properties of hempseed yogurt

	Samples			
	CY	NG	*t*	*P*
Hardness	12.53 ± 0.25	16.74 ± 0.29	−19.069***	0.000
Consistency	33.22 ± 0.43	49.43 ± 0.58	−39.044***	0.000
Elasticity	0.952 ± 0.002	0.954 ± 0.001	−2.828*	0.047
Cohesiveness	0.450 ± 0.006	0.447 ± 0.004	0.723	0.510

t, significance test value for the regression parameter. *$P < 0.05$; ***$P < 0.001$.

Table 2. Viscosity of hempseed yogurt fitted to the Herschel–Bulkley model

	Samples			
	CY	NG	*t*	*P*
τ_0 (Pa)	9.51 ± 0.37	7.70 ± 0.13	7.80**	0.001
K (Pa·s^n)	0.81 ± 0.07	10.85 ± 0.70	−24.61***	0.000
n	0.93 ± 0.02	0.38 ± 0.01	38.40***	0.000
R^2	0.98 ± 0.00	0.97 ± 0.00	3.19	0.083

τ_0, yield stress; κ, viscosity; *n*, rate index; R^2, correlation factor. *t*, significance test value for the regression parameter. **$P < 0.01$; ***$P < 0.001$.

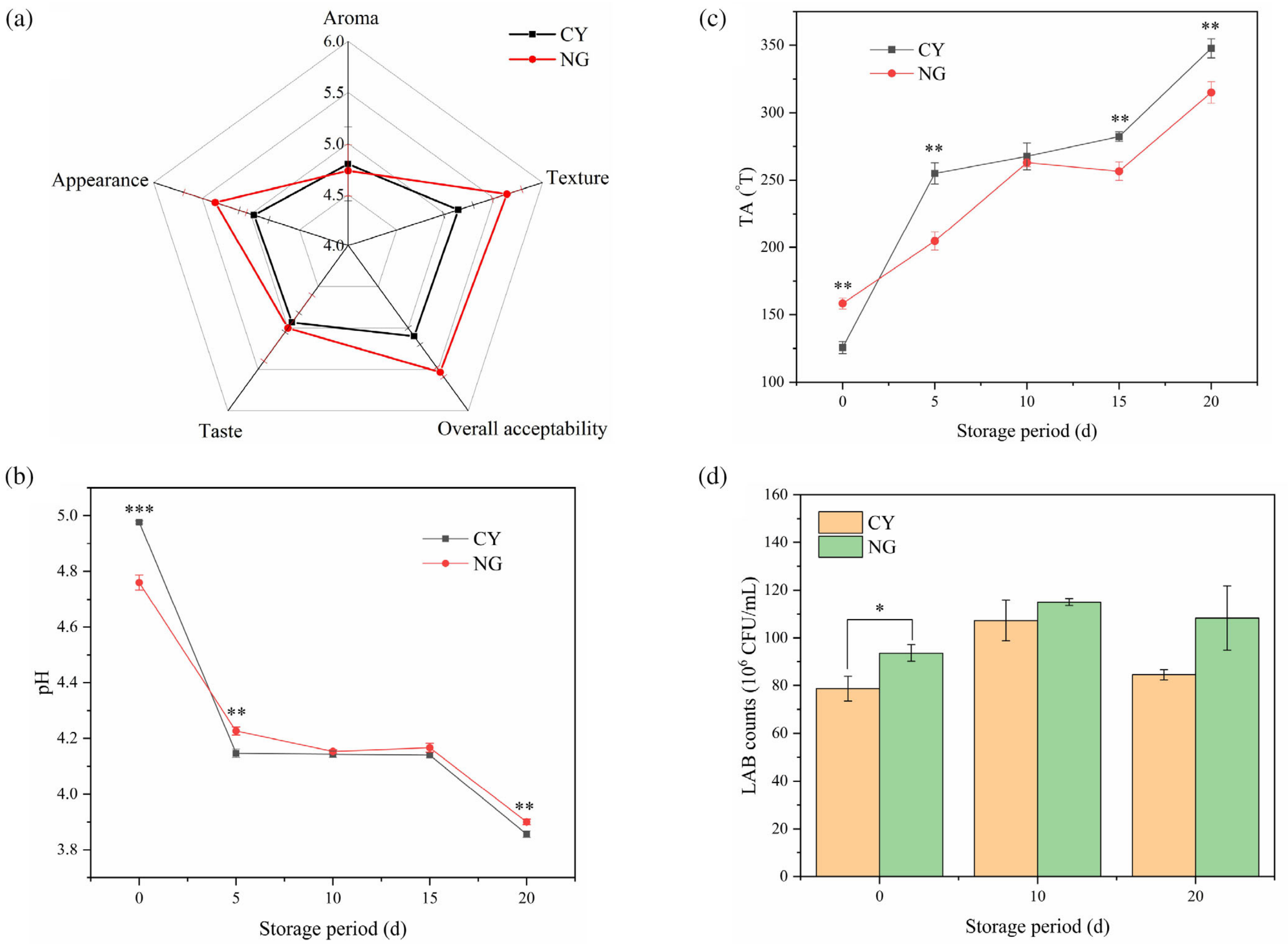

Figure 3. Effects of microfluidization on (a) sensory evaluation, (b) changes in pH, (c) changes in titratable acidity, and (d) LAB counts of control yogurt (CY) and yogurt processed with NanoGenizer (NG). *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

evaluation between two groups, NG scored slightly lower than CY. This may be because microfluidization mixed the hempseed proteins more evenly with other ingredients, allowing the herb-like aroma of the hempseed proteins more pronounced. Kavinila *et al.*[10] reported that microfluidization offered better texture and sensory properties than conventional pressure homogenization under the same working pressure. In conclusion, microfluidization improved the sensory quality and the acceptability of hempseed yogurt.

Effects of microfluidization on the pH and titratable acidity of hempseed yogurt during storage

The hydrogen ion activity in a substance is measured by the pH value, which determines the quality of the product.[56] Figure 3(b) shows the pH variations of hempseed yogurt kept at 4 °C for 20 days. As the storage time extended, the pH of hempseed yogurt continued to decrease, with a rapid decline during the first 5 days and a slow decline from 5 to 20 days. Particularly, the pH of NG exhibited a smaller decrease than that of CY. Titratable acidity

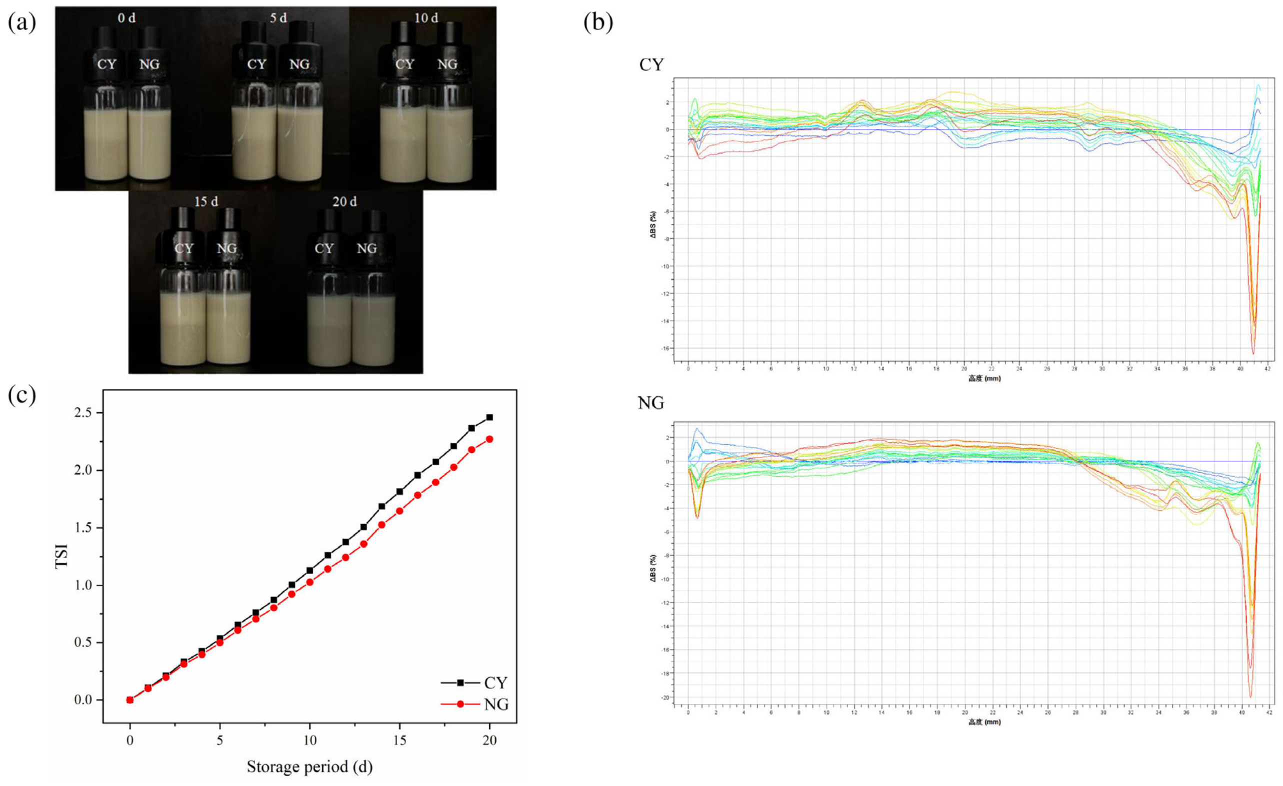

Figure 4. Effects of microfluidization on (a) apparent stability, (b) backscattering intensity, and (c) TSI values of control yogurt (CY) and yogurt processed with NanoGenizer (NG) during storage.

is used to measure the total acid concentration in foods and is a better indicator of acid's impact on flavor compared to pH.[57] As shown in Figure 3(c), the titratable acidity increased continuously with the extending of storage period. The titratable acidity of NG increased less during the storage period compared to that of CY, consistent with the pH results.

In the present study, microfluidization decreased the pH and increased titratable acidity of hempseed yogurt at the beginning of storage. Microfluidization may disrupt the structure of proteins to form stretching polymer polypeptide chains. Therefore, it was conducive to the degradation of probiotics during fermentation, thus promoting the growth of LAB and increasing the acidity of hempseed yogurt. During storage, the pH of hempseed yogurt decreased and the acidity increased, which was the result of post-acidification in yogurt.[58] This was ascribed to the continuous digestion of carbohydrates by microorganisms, which produced a large amount of organic acids to reduce the pH.[59] At the end of storage, microfluidization inhibited the post-acidification process.

Effects of microfluidization on the LAB counts of hempseed yogurt during storage

The survival of probiotics during food storage is critical to the production of probiotic products. As shown in Figure 3(d), the number of LAB initially increased during 0–10 days of storage because LAB can still grow slowly and engage in metabolic activities during the storage period.[60] At 10–20 days of storage, LAB counts declined. This may be because most of the nutrients were utilized at the early stages of storage, thus leading to a shortage of nutrients for LAB growth in the end. During storage, the LAB counts in NG remained higher than CY, which was consistent with our previous speculation that microfluidization promoted the proliferation of LAB in hempseed yogurt. LAB produce lactic acid as the final product of carbohydrate fermentation metabolism, as well as organic substances that are contributing factors to the texture, flavor and aroma resulting in unique sensory characteristics.[61] The ingestion of LAB has been recognized to confer a range of health benefits, including immune system regulation and enhanced resistance against malignant tumors and infectious disease.[62]

Effects of microfluidization on the apparent stability of hempseed yogurt during storage

As shown in Figure 4(a), hempseed yogurt prepared by conventional homogenization method exhibited particle sedimentation. This was because, during the extraction of hempseed protein, the green membrane inside the hard shell of the hempseed cannot be completely removed, resulting in some visible particles in the raw material. By contrast, the microfluidized hempseed yogurt was in a homogeneous state and no precipitation of hempseed protein particles was observed during the whole storage period, indicating a better apparent stability for NG yogurt. Microfluidization can modify the properties of several macromolecules, evenly distributing large particles into the fibrous structures, thus producing a unprecipitated, highly gelatinous and stable product.[63] Additionally, hempseed protein contained a small amount of dietary fiber, where the particle size was effectively reduced by microfluidization, causing the redistribution of fiber components from insoluble to soluble fractions.[64]

Effects of microfluidization on the storage stability of hempseed yogurt during storage

Backscatter imaging is a spectral imaging technique that provides high accuracy to detect the quality attributes of food products and further identifies the relationships between the quality characteristics and the observed physiochemical properties of the commodity.[65] The central portion of the backscatter pattern represents the middle of the cuvette, and the left and right portions represent the bottom and top of the cuvette, respectively.[66] As shown in Figure 4(b), the overlap in the middle part of NG was superior to CY, indicating that the sample with microfluidization had no particle aggregation and flocculation, and exhibited good stability during storage.

TSI value is used to describe the motion and interaction of particles,[67] and reflects the stability of an emulsion, where a low TSI value indicates a stable system.[68] As shown in Figure 4(c), the TSI values increased with extended storage period, suggesting a decrease stability of hempseed yogurt. This might be a result of the slow metabolic activity of LAB, which disrupted the internal network structure and reduced the stability of hempseed yogurt. TSI values of NG were consistently lower than CY. At the end of storage, the TSI values for CY and NG were 2.46 and 2.27, respectively, indicating that microfluidization improved the storage stability of hempseed yogurt. This was because the intense mechanical forces generated by microfluidization unfolded the protein structure and exposed more hydrophilic groups, which improved the interaction with water and increased the solubility of protein in water,[69] resulting in a more uniform hempseed slurry. Santos et al.[70] produced nanoemulsions for the eco-friendly formulation prepared by microfludization and found that the TSI value of the emulsion gradually decreased with an increasing homogenization pressure. Overall, microfluidization improved the storage stability of hempseed yogurt within 20 days.

CONCLUSIONS

Microfluidization significantly affected the physicochemical properties, microstructure, sensory characteristics and storage stability of hempseed yogurt compared to traditional homogenization. Specifically, hempseed yogurt with microfluidization had a small and uniform particle distribution, which enhanced its gel structure and increased the hardness, consistency, elasticity and viscoelasticity, making it more acceptable to consumers. During storage, microfluidization suppressed the post-acidification process, increased the number of LAB and reduced the TSI values of hempseed yogurt. The present study demonstrates that microfluidization has the potential to provide new opportunities for the development of high-quality and stable hempseed yogurt. It also offers new insights into solving the problem of dissatisfied quality of food products and helps food manufacturers create competitive products to meet the demands of health-conscious consumers. Further future research should focus on exploring the effects of microfluidization on the volatile components and small-molecule nutrients in hempseed yogurt.

ACKNOWLEDGMENTS

This study was supported by the National Natural Science Foundation of China (No. 31972031), LiaoNing Revitalization Talents Program (XLYC2002039), Liaoning BaiQianWan Talent Program and Liaoning Xing Liao Talent Program (LNBQW2019B0089), Unveiling project of Liaoning.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

AUTHOR CONTRIBUTIONS

JX and HL were responsible for conceptualization. JX and XF were responsible for methodology. JX was responsible for data curation and formal analysis. JX, XF, XX, DD and HS were responsible for software. JX was responsible for writing the original draft. XF was responsible for visualization XF, XX, DD and LY were responsible for investigations. HL was responsible for resources, supervision, reviewing and editing, and funding acquisition.

ETHICAL STATEMENT

Ethical approval was not required for this research.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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