

This study aimed to investigate the technological properties of kefir fortified with defatted sesame flour (DSF). We have developed a recipe for producing kefir with the addition of 0, 2, 4, 6, 8 % DSF. The physicochemical parameters (pH, titratable acidity) and rheological properties (water-holding capacity, viscosity) of the samples within 28 days of storage were studied. All the samples were stored at a temperature of 4 °C. Nuclear magnetic resonance and chromatographic analysis were performed to determine the optimal shelf life of the product. It was found that fermentation ends on the seventh day of storage, while a low content of free water is observed. The recommended optimal storage time for kefir enriched with DSF is within 14 days. The introduction of 2 % DSF in kefir increases the content of vitamin E by three times (from 0.32 to 0.93 mg/100 g) and the content of proteins – by 1 % (from 3.36 to 4.3 %). The result showed that the pH value decreases with the introduction of DSF and is within the normal range during the entire storage period of the product (from 4.38 to 4.07). In this case, the total titratable acidity increases from 112.07 to 163.52 °T. An increase in viscosity (by 3 Pa·s) and water-holding capacity (by almost 6 %) is observed with the addition of 2 % additive. Due to the high water-holding capacity and viscosity, dietary fiber content (up to 0.68 %), kefir supplemented with DSF showed high storage stability. The addition of 2 % DSF to kefir can increase the nutritional value and improve the structural stability of the product. DSF is a waste product with a bright prospect for use in the manufacture of fermented milk drinks

Keywords: drinks, defatted sesame flour, rheological properties, food additives, waste products, dietary fiber

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INFLUENCE OF SESAME FLOUR ON PHYSICOCHEMICAL PROPERTIES OF SOUR MILK DRINKS

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1. Introduction

Fermented milk drinks such as kefir are drinks that are prepared by adding kefir fungi to milk. It originates from the Caucasian mountains and has spread through the centuries in the Balkans and Eastern Europe. Kefir grains are small, gelatinous, yellowish, and irregularly shaped masses resembling individual miniature florets of a head of cauliflower. Kefir grains consist of a gel matrix in which yeasts and bacteria are embedded and live symbiotically [1]. The microbial content of kefir grains depends primarily on their source. It has been reported that kefir grains contain *Lactobacilli*, *Lactococci* and yeast, and sometimes acetic acid bacteria, depending on the source or country of origin [2].

Some results indicate that kefir and its constituents have pro-healthy properties that positively affect the immune and gastrointestinal systems [3]. Some studies have shown the positive effect of kefir in lactose intolerance [4]. Fermented milk drinks are a source of several vitamins [5] and may slow the development of breast cancer [6]. Besides, kefir cultures can be applied to promote food safety by inhibiting coliforms and numerous pathogens [5]. Kefir is regarded as a nutritious and healthy drink.

Surveys showed that consumers tend to buy thicker samples while most of the samples on the market are not thick enough, this leads to an increase in the amounts of stabilizers. Probiotic activity is an essential functional characteristic for yogurt [7], the introduction of stabilizers would negatively affect the population of “living microflora”, decrease the biological value of the product [8–10]. A way to overcome these difficulties can be the introduction of dietary fiber into fermented milk, for the strong water-holding capacity of dietary fiber, which is supposed to improve the viscosity and structural stability. DSF is a kind of raw material with high dietary fiber and protein content, which is expected to improve the viscosity of the product. Based on the data obtained, the expediency of using the proposed types of fillers, namely strawberry powder and candied beetroot, in the production of yoghurts without the use of stabilizers, flavors and other food additives was proved [11].

Sesame is one of the most important oil crops in the world, and its yield is the seventh in the world. It mainly grows in the tropics and subtropics. A large amount of sesame oil production brings a large amount of defatted sesame flour (DSF). DSF is a by-product of sesame oil production. On one hand, DSF contains 50 % protein, high calcium (1.5/100 g),

and crude fiber (10.8/100 g) [12]. DSF also contains phenolic compounds, which possess antioxidant, antimutagenic and antimicrobial activities [13]. Other nutraceutical compounds present in DSF are lignans [14] and several minerals, like potassium (4.6–5.3 g/kg), phosphorus (1.7–2.3 g/kg) magnesium (0.018–0.052 g/kg), and calcium in high concentration (9.6–12.8 g/kg) [15, 16]. Therefore, it is very beneficial to body health, especially its anti-aging, anti-fat, anti-tumor, anti-triple H (hypertension, hyperglycemia, hyperlipidemia), immunity-improving effects. On the other hand, DSF is mainly used as food of livestock or discarded, with low utilization rate. Therefore, we believe that the application of DSF in food processing will greatly withstand a series of problems caused by the current population's excessive nutrition, such as obesity, hypertension, hyperglycemia, hyperlipidemia, etc., also, it can promote intestinal health and improve the health level of the population. Thus, innovative facets regarding the usage of these wastes as co-products for further production of food additives or supplements or value-added products with high nutritional value are gaining increasing interest. Their further recovery and utilization are economically and ecologically attractive. Therefore, adding sesame flour to fermented milk drinks would be a very significant attempt.

2. Literature review and problem statement

Almost all types of dairy products can combine with different plant components. Many scientists are trying to create healthy food by considering natural plant components [17]. The paper [18] presents five different yogurts with a filler such as carrots, green peas, zucchini and beans. The results showed that vegetable puree addition had a noticeable effect on pH, titratable acidity, whey separation, sensory properties, as well as textural attributes such as firmness, cohesiveness, consistency and viscosity index in yogurt. It is shown that different raw materials have different effects on fermented yoghurt. The highest viscosity index, consistency and firmness were observed in yogurt with the carrot puree, while the samples with zucchini presented the less compact structure and textural properties. As a result, the present work showed that all the vegetable purees used had a beneficial role to improve textural properties of set type non-fat yogurt and could be used for the development of dairy products with functional ingredients. In the study [19], various anthocyanin-rich fruit and vegetable juices (black mulberry, pomegranate, strawberry and black carrot) were added to kefir at various concentrations (10, 25 and 50 %, w/w). The concentration of 50 % was chosen because Codex Standard [20] allows the addition of up to 50 % non-dairy ingredients to flavored fermented milk. The most preferred kefir by the sensorial panel were strawberry and pomegranate, followed by black mulberry and black carrot. Kefirs with 10 and 25 % pomegranate completed their shelf-lives microbiologically in a shorter time (8–11 and 5 weeks, respectively). Fortification with pomegranate at 25 % concentration reduced the shelf-life of plain kefir from 12 weeks to 5 weeks. Pomegranate at 10 % concentration is recommended for a longer shelf-life with high microbial stability. On the other hand, kefir fortified with the other juices retained sensorial, chemical and microbiological quality during 12 weeks of storage at 4 °C. Shelf-lives of the samples, except for pomegranate, ended after 12 weeks of storage, due to low sensorial scores. Additions of strawberry, pomegranate and black mulberry at 25 % concentration are recommended for

the production of more palatable kefir with high antioxidant activity. This paper laid emphasis on the antioxidant activity of kefir, the number of microorganisms, and sensorial properties, specifically analysis on the physicochemical property of kefir lacked. Dietary fiber was used in [21], experimental yoghurts were made with 13 % reconstituted skim milk supplemented with 1, 2, 3 % rice bran (RB) and inoculated with a probiotic culture of *Lactobacillus casei* 431. The products were stored at 4 °C for 3 weeks. Probiotic viability increased significantly by increasing the amount of added RB. *L. casei* 431 counts remained high in all of the RB enriched yoghurts during the overall storage period of 21 days. The highest *S. thermophilus* numbers were found in yogurt containing 3 % RB at the end of storage. Enrichment of RB resulted in an increase in water-holding capacity and pH and a decrease in syneresis of yoghurt base. RB fortification improved DPPH radical scavenging activities of yoghurt samples. Unfortunately, acceptability was lower for all enriched yoghurts compared to plain yoghurt. In the paper [22], the rolled oats were floured and then used to prepare oat milk with a concentration of 13 g in 100 mL water. Kefir samples with 20, 40 and 60 % of oat milk were produced. The effect of oat-milk ratio on physicochemical, rheological, microbiological and sensory characteristics of the kefir samples was studied during 21 days storage in refrigerated conditions. Increasing oat milk concentration affected the whey-off and apparent viscosity by higher whey-off and lower viscosity results. An increase in the oat milk concentration led to a decrease in the pH of the samples. In the study [23], skim milk (9.5 % w/v solid content) was supplemented with 1–3 % (w/v) lentil flour or skim milk powder, inoculated with a yogurt culture, fermented and stored at 4 °C. Acid production during the fermentation, microbial growth, physical properties (pH, syneresis, and color), rheological properties (dynamic oscillation temperature sweep test at 4–50 °C) during 28 days of refrigerated storage and also sensory properties (flavor, mouth feel, overall acceptance and color) after production were studied. Milk supplementation with 1–3 % lentil flour enhanced acid production during fermentation, but the microbial population (CFU) of both *S. thermophilus* and *L. delbrueckii subsp. bulgaricus* were in the same range in all lentil flour and skim milk powder supplemented yogurts. The average pH of the samples decreased from 4.5 to 4.1 after 28 days storage. Syneresis in 1–2 % lentil flour supplemented yogurts was significantly higher than in all other samples; however, greater lentil supplementation (3 %) resulted in the lowest syneresis during the 28 days storage. With respect to color, “a” and “L” values did not significantly differ in all samples and remained constant after 28 days whereas “b” value increased as a result of lentil supplementation. Yogurt with 3 % lentil flour showed higher storage (G') and loss (G'') moduli in comparison with the samples supplemented with 1–3 % skim milk powder and the non-supplemented control yogurt. Storage modulus (G') was higher than loss modulus (G'') in all samples and at all temperatures between 4 and 50 °C and they showed a hysteresis loop over this temperature range when the samples were heated and cooled. 1–2 % lentil flour supplemented yogurt showed comparable sensory properties in comparison with 1–2 % skim milk powder supplemented yogurt and the control sample.

There were unresolved issues related to a deterioration of rheological parameters: decrease of viscosity, separation of whey, deterioration of taste. The reason for this may be the high moisture content of fillers, an odorous substance produced by a biochemical reaction that occurs during fer-

mentation, the microstructural balance of fermented milk is disturbed, which makes relevant research impractical. All this suggests that it is advisable to conduct a study on other fillers, which rich in dietary fiber and have lower water content and smaller particles, and have no negative effects on the rheological parameters, stabilization of structural-mechanical properties, water-holding capacity, and acidity of kefir. A waste-free technology for processing vegetables is shown, according to which powders obtained from vegetable waste can be used as functional additives [24].

In the paper [25], mixtures of defatted sesame cake (DSC) (0–20 %) and corn grits were processed in a single screw extruder at a screw speed ranging from 324 to 387 rpm to improve the nutritional value of corn expanded extrudates. Chemical composition of raw and extruded materials, sectional expansion index (SEI), texture properties, color, paste viscosity, microstructure and sensory analysis of the extrudates was performed. The addition of DSC increased the protein, fat and ash content of corn extrudates, whereas carbohydrate content was reduced. The addition of DSC reduced the sectional expansion of the corn extrudates and increased puncture force. DSC-corn extrudates were darker than non-DSC-corn extrudates. Increasing DSC increased the number of cells similar to those of commercial corn extrudates with small cells. Sensory analysis showed 20 % SDSC-corn extrudates to be acceptable and nutritionally balanced. The use of DSC on corn extrudates up to 20 % is an alternative to improve nutritional value keeping good sensory characteristics. In the paper [26], fifty percent incorporation of defatted white sesame and black sesame into biscuits improved the protein, fat, crude fiber, mineral, and reduced the carbohydrate level. The textural attributes and color characteristics of the white sesame-amended biscuits (WSB) were similar to that of the control biscuits. WSB scored highest upon subjecting to sensory evaluation over black sesame-amended biscuits (BSB) and panelists scored them equal to the control biscuits. The likability of WSB was also revealed in the consumer preference survey. Both WSB and BSB fortification improves the storability of biscuits by reducing microbial contamination. Also, the aqueous and methanolic extracts of 90-day stored biscuits were non-toxic to the cell line (L929). In the paper [27], cookies were prepared from blends of wheat flour with 10, 20, 30 and 40 % dehulled sesame seed meal. The protein content increased from 12.52 % with wheat flour to 16.86 % with 40 % substitution, but fortification did not affect the color, flavor, texture or overall acceptability of the cookies. In the paper [28], the acceptability of using sesame meal in baked products was estimated. The defatted sesame meal was supplemented into white wheat flour to get composite flours. An increase in protein content due to the addition of partially defatted sesame meal results in an increase in farinographic parameters. The mixographic peak height percentages and mixing time of the combined flours decreased significantly by the supplementation. In vitro protein digestibility of breads prepared from composite flours increased gradually as the level of supplementation increased. The sensory scores of the breads were significantly affected by the difference in supplementation levels. The partially defatted sesame enhances the chemical components of composite flour. The breads prepared from sesame meal supplemented composite flour indicated a significant reduction in internal and

external sensory characteristics by the raising level beyond 12 % supplementation of sesame meal. The scores assigned to aroma and taste of breads prepared from freshly prepared composite are significantly higher. Sesame meal supplemented breads should be advised for diabetic and heart patients. Sesame should be used in various other products based on dairy and beverages.

To the best of our knowledge, there is no information available about the effects of DSF addition on the rheological, physicochemical properties of kefir.

3. The aim and objectives of the study

The aim of this study was to explore the possibility of using DSF as a filler for kefir.

To achieve this aim, the following objectives are accomplished:

- determination of appropriate addition amount of DSC into kefir;
- determination of physicochemical and rheological properties (pH, acidity, color, water retention capacity, nuclear magnetic resonance, viscosity) of fermented milk drinks containing DSF;
- determination of the chemical composition of products with DSF (content of fat, proteins, dietary fiber, vitamin E).

4. Materials and methods

Kefir grains were obtained from private households in Tibet, China. Cow milk was supplied from Mengniu Dairy Group Co, Neimenggu, China. All chemicals were of analytical grade.

Sesame seeds (white) were procured from the local market (Hezhou, Guangxi, China). DSF was obtained from sesame, which was roasted at 120–130 °C for 10 min at first and then pressed to separate the fat and flour. The flour was finely powdered and homogenized by sieving through a fine screen (a fraction with a crushing degree of 100), then frozen in a refrigerator at 18 °C in sealed plastic bags until use. The aim of DSF refrigeration was to prevent microbial growth and to preserve the active health components, such as phenols. Microbiological examination on DSF showed that it was sterile due to the high-temperature treatment and low-temperature freezing treatment.

The kefir grain was activated before use and added to sterilized cow milk in a 3/100 (w/v) ratio and fermented at 28 °C for 22 hours until the pH reached 4.7. Kefir grain was obtained by filtration of kefir, the process was repeated 7 times. At the 7th time, the kefir grain was frozen in a refrigerator, the fermented milk drink was used as a start culture at a ratio of 10 % (V/V). Five raw materials formulae were investigated: sterilized cow milk added with 0 (control), 2 % (A), 4 % (B), 6 % (C), 8 % (D) DSF. First, the processed DSF was added into the sterilized milk in proportion; second, the mixture with kefir was inoculated, the total volume of the mixture was 200 ml; last, fermented under the condition of ventilation. All the operations above are carried out on a benchtop to ensure no contamination. The cultural condition was the same as activation. Kefir samples were stored at 4 °C for 28 days.

The pH value was determined by a pH meter (METTLER TOLEDO, Switzerland), shown in Fig. 1.



Fig. 1. pH meter for pH measuring

The acidity was measured by titrating 10 g of the sample with 0.1 N NaOH using a phenolphthalein indicator.

Water-holding capacity (WHC) of fermented milk drinks was determined using a centrifuge (cence L550, China), shown in Fig. 2.



Fig. 2. Centrifuge for water-holding capacity

By this technique: 10 g of fermented milk drink (X) samples were weighed into a 50 mL test tube and centrifuged at 3,000 rpm for 20 min at 4 °C. The separated whey (Y) was removed and weighed. The water-holding capacity was calculated as:

$$WHC = \frac{X - Y}{X_s} 100\% \quad (1)$$

The apparent viscosity of the samples was measured with an NDJ-8S digital viscometer (LICHEN NDJ-8S, China), shown in Fig. 3.

Nuclear magnetic resonance was determined by NMR (NIUMAI, Shanghai, China, Fig. 4) as follows: a sample of about 1.500 g was weighed and placed into a glass tube (caliber: 18 mm), the sample tube was placed in the NMR probe (maintain temperature: 32 °C), and the transverse relaxation time T_2 was measured using the CPMG sequence, where the receiver bandwidth was SW=100 kHz.

The control parameters of sampling starting point $RFD=0.150$ ms, repeat sampling interval $TW=4500.000$ ms, analog gain $RG1=20.0$ dB, 90° and 180° pulse widths were $P90=10.00$ μ s, $P180=19.52$ μ s, sampling points $Td=800034$, digital gain $DRG1=1$, data radius $DR=1$, cumulative scanning times $NS=2$, echo number $NECH=8000$. The test measured 3 copies.

A chroma meter (CR-400, Japan) was used to determine Hunter L , a and b color parameters of the fermented milk drink samples. The source of light and the observation angle are D65 and 10° . The chroma meter is shown in Fig. 5. “ L ” denotes brightness, the larger the “ L ” value, the brighter

the sample; “ A ”, as a substitute for red and green, a positive value as a substitute for red, the higher the value, the redder the sample; a negative value as a substitute for green, the larger the absolute value, the greener the sample; “ B ”, as a substitute for blue and yellow, a positive value as a substitute for yellow, the higher the value, the yellower the sample; a negative value as a substitute for blue, the larger the absolute value, the greener the sample.

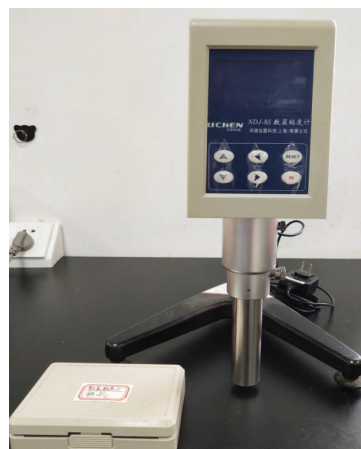


Fig. 3. Digital viscometer for apparent viscosity



Fig. 4. NMR (Nuclear Magnetic Resonance)

The amount of dietary fiber was determined by the measurement method using the AOAC system, the instrument (CSF 6 Filtration System, Italy) is shown in Fig. 7.

Vitamin E content – using a set for the analysis of vitamin E by the colorimetric method (Institute of Bioengineering Nankin Jianchen, Nankin, China), the detection kit is shown in Fig. 8.



Fig. 5. Chroma meter for color measurement



Fig. 6. Kjeldahl apparatus for protein measuring



Fig. 7. Dietary fiber determination system

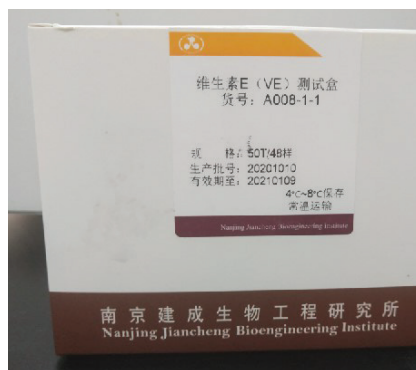


Fig. 8. Vitamin E assay kit (Colorimetric method)

Above were the materials and methods for the research.

5. Results of the study of fermented milk drinks with the addition of DSF

5.1. Preparation of samples of fermented milk drinks with different amounts of DSF

Sesame seeds are sorted as white, black, yellow and other sesame according to the color. According to our survey, kefir with white color was highly accepted by consumers than others. Therefore, we chose white sesame meal, which has little influence on the color of kefir, as the raw material.

According to the preliminary experiment, when the addition amount of DSF reached 10 %, the sensory quality of kefir declined seriously. At the same time, when the addition amount

of DSF was between 0 and 8 %, the physical and chemical properties of the product changed greatly. So 0, 2 %, 4 %, 6 % and 8 % addition amounts of DSF into kefir were fitting to conduct the research. Compared to other similar researches with adding by-product into kefir (mango peel, 5 % (w/v); apple fiber (AF) and lemon fiber (LF), 1 % (w/v)) [29, 30], the addition amount range of by-product improved significantly.

5.2. Results of physical and chemical analysis of fermented milk drinks with DSF additive

5.2.1. Results of acidity

The values of pH and acidity in 28 days are shown in Table 1. The histogram of pH for different samples is shown in Fig. 9. The histogram of titratable acidity for different samples is shown in Fig. 10.

Table 1

Acidity of fermented milk drink samples during storage for 28 days ($n=3, \pm SD$)

Indicator	Sam-ple	Day 0	Day 7	Day 14	Day 21	Day 28
pH	K	5.01± ±0.02 ^{a, X}	4.83± ±0.05 ^{a, Y}	4.72± ±0.02 ^{a, Z}	4.54± ±0.00 ^{a, W}	4.51± ±0.00 ^{a, W}
	A	4.38± ±0.03 ^{b, X}	4.16± ±0.02 ^{b, Y}	4.14± ±0.02 ^{b, Y}	4.06± ±0.00 ^{b, Z}	4.07± ±0.02 ^{b, Z}
	B	4.25± ±0.04 ^{c, X}	4.05± ±0.04 ^{c, Y}	4.03± ±0.00 ^{c, YZ}	3.97± ±0.00 ^{c, ZW}	3.95± ±0.00 ^{c, W}
	C	4.11± ±0.03 ^{d, X}	4.06± ±0.02 ^{c, Y}	4.05± ±0.02 ^{c, Y}	3.94± ±0.00 ^{d, Z}	3.93± ±0.00 ^{d, Z}
	D	4.14± ±0.05 ^{d, X}	4.06± ±0.01 ^{c, Y}	4.03± ±0.00 ^{c, Y}	3.94± ±0.00 ^{d, Z}	3.93± ±0.00 ^{d, Z}
Acidity (°T)	K	67.78± ±0.38 ^{d, M}	75.44± ±1.10 ^{d, W}	82.92± ±1.75 ^{e, Z}	92.83± ±1.17 ^{e, Y}	99.22± ±2.25 ^{d, X}
	A	112.07± ±0.92 ^{c, Z}	125.88± ±4.75 ^{c, Y}	133.46± ±1.62 ^{d, Y}	157.43± ±4.03 ^{d, X}	163.52± ±6.04 ^{c, X}
	B	117.34± ±2.18 ^{c, M}	155.27± ±1.38 ^{b, W}	167.25± ±2.77 ^{c, Z}	191.12± ±3.53 ^{c, Y}	205.57± ±8.26 ^{b, X}
	C	138.88± ±5.26 ^{b, M}	179.23± ±3.89 ^{a, W}	192.53± ±5.33 ^{b, Z}	208.41± ±1.04 ^{b, Y}	222.74± ±3.26 ^{a, X}
	D	163.13± ±6.02 ^{a, W}	188.61± ±9.84 ^{a, Z}	209.48± ±0.71 ^{a, Y}	222.00± ±2.52 ^{a, XY}	234.91± ±1.76 ^{a, X}
<i>a, b, c, d</i> – means in the same column with different superscripts significantly differ ($P<0.05$)						
<i>X, Y, Z, W, M</i> – means in the same row with different superscripts among fermented milk drink samples significantly differ ($P<0.05$)						
SD (Standard deviation): K=control sample, A=fermented milk drink with 2 % DSF, B=fermented milk drink with 4 % DSF, C=fermented milk drink with 6 % DSF, D=fermented milk drink with 8 % DSF						

The pH of the samples ranged between 5.01 and 4.07. The addition of DSF could significantly decrease the pH of products, similar to the research [30]. The substantial decrease occurred in active acidity (pH) in the first week for all the samples, and no further changes for the next 3 weeks of storage, because of post acidification of lactobacillus by using remaining lactose within 7 days.

Above were the results of pH and titratable acidity of the samples.

The addition amount of DSF had a significant effect on the acidity of fermented milk drinks. With the increase of addition amount, the acidity of fermented milk drinks increased significantly. It could be related to the stimulation of lactic acid bacteria by fiber. Previous studies have also

demonstrated that kefir as a probiotic and dietary fiber had a potential prebiotic effect [30]. Storage time had a significant effect on the acidity of products, with the increase of storage time, the acidity of fermented milk drinks increased significantly, which was due to the catabolism of lactose.

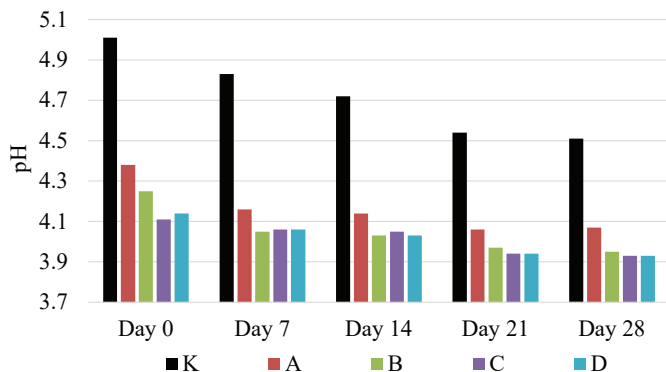


Fig. 9. Results of research of active acidity of fermented milk drinks with DSF additive

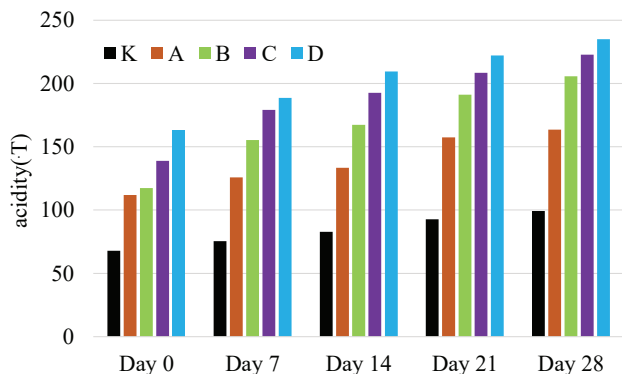


Fig. 10. Titratable acidity of kefir samples during storage for 28 days (n=3, ±SD)

5. 2. 2. Result of WHC

The results of determining the water-holding capacity of fermented milk drinks with the addition of DSF additives are presented in Table 2. The histogram of WHC for different samples is shown in Fig. 11.

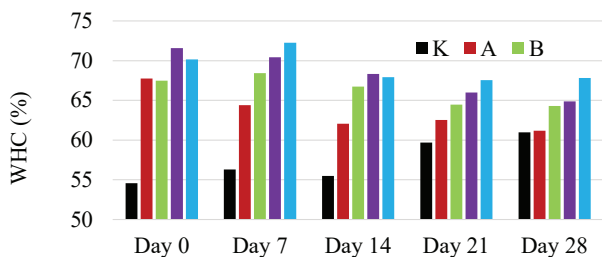


Fig. 11. WHC of fermented milk drink with DSF

The WHC of fermented milk drink supplemented with DSF was significantly increased compared to blank one ($p<0.05$), which was due to the high WHC of dietary fiber and protein, the trend was similar as in the research [30]. There was no significant change in WHC among fermented milk drinks added with 2 %, 4 %, 6 %, 8 % DSF. There was no significant change in the WHC of all the samples during

the storage period, which showed that the storage period had no significant effect on all the samples.

Table 2

WHC of fermented milk drink samples during storage for 28 days (n=3, ±SD)

Indicator	Sample	Day 0	Day 7	Day 14	Day 21	Day 28
WHC (%)	K	54.57± ±4.91 ^{b,X}	56.31± ±1.30 ^{d,X}	55.49± ±1.90 ^{c,X}	59.69± ±1.42 ^{d,X}	60.97± ±0.93 ^{d,X}
	A	67.76± ±0.85 ^{a,X}	64.4± ±1.28 ^{c,Y}	62.07± ±0.45 ^{b,YZ}	62.56± ±1.198 ^{c,YZ}	61.21± ±1.36 ^{cd,Z}
	B	67.49± ±2.26 ^{a,XY}	68.44± ±0.41 ^{b,X}	66.73± ±1.23 ^{a,XY}	64.49± ±0.28 ^{bc,YY}	64.30± ±0.79 ^{bc,YY}
	C	71.60± ±1.02 ^{a,X}	70.44± ±0.46 ^{ab,XY}	68.35± ±1.06 ^{a,YZ}	65.99± ±0.52 ^{ab,ZW}	64.87± ±1.35 ^{ab,W}
	D	70.17± ±0.97 ^{a,XY}	72.27± ±0.08 ^{a,X}	67.94± ±1.01 ^{a,YZ}	67.58± ±0.63 ^{a,YZ}	67.84± ±1.33 ^{a,Z}

a, b, c, d – means in the same column with different superscripts significantly differ ($P<0.05$)
x, y, z, w – means in the same row with different superscripts among fermented milk drink samples significantly differ ($P<0.05$)
 SD (Standard deviation): K=control sample, A=fermented milk drink with 2 % DSF, B=fermented milk drink with 4 % DSF, C=fermented milk drink with 6 % DSF, D=fermented milk drink with 8 % DSF

Above were the results of WHC of the samples. Versus a beverage without additives, 2 % addition amount of DSF significantly increased the apparent viscosity ($p<0.05$).

5. 2. 3. Result of viscosity

The results of the study of viscosity are presented in Table 3 and Fig. 12.

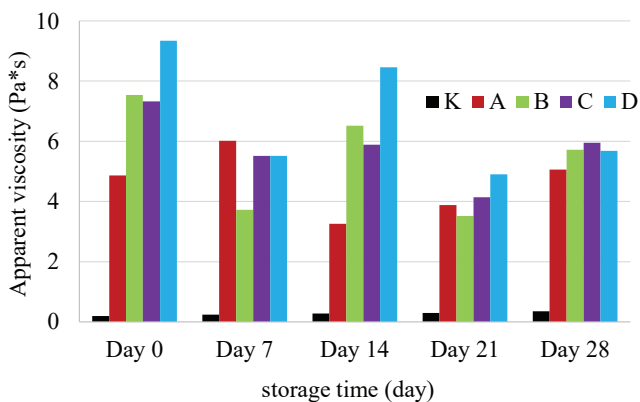


Fig. 12. Apparent viscosity of fermented milk drink with DSF

Above were the results of apparent viscosity of the samples. The apparent viscosity of fermented milk drink supplemented with DSF was significantly increased compared to blank one ($p<0.05$).

The increased viscosity of the fiber-enriched kefir seems to be caused both by the contribution of the soluble matter to the composition of the aqueous phase and by the contribution of insoluble fibers to the increase of total solids affecting the three-dimensional conformation of the hydrated biopolymers [31]. There was no significant difference of

apparent viscosity among the samples added with 2 %, 4 %, 6 %, 8 % DSF. The storage time had no significant influence on the apparent viscosity of all the samples.

Table 3

Viscosity of fermented milk drink samples during storage for 28 days ($n=3, \pm SD$)

Indicator	Sample	Day 0	Day 7	Day 14	Day 21	Day 28
Apparent viscosity (Pa*s)	K	0.19± ±0.02 ^{c,Z}	0.23± ±0.02 ^{b,YZ}	0.27± ±0.05 ^{d,XY}	0.30± ±0.02 ^{b,XY}	0.35± ±0.03 ^{b,X}
	A	4.87± ±0.37 ^{b,Y}	6.01± ±1.47 ^{a,X}	3.26± ±0.34 ^{c,Z}	3.88± ±0.11 ^{a,Z}	5.06± ±0.64 ^{a,Y}
	B	7.53± ±0.92 ^{ab,X}	3.72± ±0.19 ^{a,Z}	6.52± ±0.09 ^{b,XY}	3.52± ±0.94 ^{a,Z}	5.72± ±0.67 ^{a,Y}
	C	7.33± ±1.12 ^{ab,X}	5.52± ±0.63 ^{a,XY}	5.88± ±0.29 ^{b,XY}	4.14± ±0.31 ^{a,Y}	5.95± ±1.31 ^{a,XY}
	D	9.33± ±1.69 ^{a,X}	5.52± ±1.11 ^{a,YZ}	8.45± ±1.31 ^{a,XY}	4.90± ±0.62 ^{a,Z}	5.68± ±0.60 ^{a,YZ}
<i>a, b, c, d</i> – means in the same column with different superscripts significantly differ ($P<0.05$)						
<i>X, Y, Z, W</i> – means in the same row with different superscripts among fermented milk drink samples significantly differ ($P<0.05$)						
SD (Standard deviation): K= control sample, A=fermented milk drink with 2 % DSF, B=fermented milk drink with 4 % DSF, C=fermented milk drink with 6 % DSF, D=fermented milk drink with 8 % DSF						

5. 2. 4 Result of color parameters

The values of color parameters of the samples in 28 days are shown in Table 4.

Above were the results of color parameter of the samples.

The introduction of DSF into fermented milk drink significantly decreased the brightness of the samples, and the red and yellow colors of the sample are significantly deepened, which was similar to the research [30]. But when the addition amount of DSF reached 4 %, the brightness of the products kept steady. The storage period had no significant effect on the brightness of all the samples. With the prolongation of storage time, the red color of all the samples showed a trend of deepening at first and then weakening, and the red color was the deepest on the 14th day. On the 7th and 21st days of storage, the yellow color of the samples decreased significantly.

5. 2. 5. Result of NMR

The spin-spin relaxation time of SDF fermented milk drink during storage was measured by NMR, the representative spin-spin relaxation time of SDF fermented milk drink is shown in Fig. 13. The signal amplitude corresponding to T_{23} is shown in Fig. 14.

With the extending of storage time, the signal amplitude corresponding to T_{23} presents a down-up-down fluctuation. From the completion of fermentation to the seventh day of storage, the signal amplitude decreased significantly, indicating that the content of free water was the lowest, the retention power of the sample was the highest, and the solidification state was the best. Then on the 21st day, the signal amplitude increased significantly, indicating that the content of free water increased, the water holding capacity of the samples decreased, and the solidification structure also decreased. Until the 28th day of storage, the water-holding capacity of the samples increased again, and the solidification structure increased too.

Table 4

Color parameter changes during cold storage

Sample	Storage period (days)	<i>L</i>	<i>a</i>	<i>b</i>
K	0	97.08±0.02 ^{a,Y}	1.96±0.09 ^{d,Z}	2.25±0.02 ^{d,X}
	7	97.42±0.33 ^{a,XY}	2.06±0.05 ^{d,YZ}	2.00±0.11 ^{d,Y}
	14	97.71±0.13 ^{a,X}	2.23±0.05 ^{e,X}	2.02±0.03 ^{e,Y}
	21	97.73±0.13 ^{a,X}	2.14±0.05 ^{e,XY}	1.84±0.05 ^{e,Z}
	28	97.38±0.10 ^{a,XY}	2.05±0.04 ^{d,YZ}	1.94±0.01 ^{e,YZ}
A	0	95.6±0.40 ^{b,Y}	3.04±0.04 ^{c,Y}	4.03±0.09 ^{c,X}
	7	96.06±0.30 ^{b,XY}	3.21±0.06 ^{c,XY}	3.92±0.03 ^{c,X}
	14	96.35±0.03 ^{b,X}	3.34±0.08 ^{d,X}	3.91±0.10 ^{d,X}
	21	96.45±0.08 ^{b,X}	3.05±0.05 ^{d,Y}	3.55±0.08 ^{d,Y}
	28	96.41±0.11 ^{b,X}	3.27±0.09 ^{c,X}	3.62±0.11 ^{d,Y}
B	0	94.35±0.32 ^{c,X}	3.43±0.06 ^{b,Z}	5.53±0.11 ^{b,X}
	7	94.94±0.57 ^{c,X}	3.61±0.08 ^{b,X}	5.25±0.03 ^{b,Y}
	14	94.59±0.11 ^{c,X}	3.6±0.04 ^{c,XY}	5.21±0.12 ^{b,Y}
	21	95.15±0.09 ^{c,X}	3.46±0.02 ^{c,YZ}	5.12±0.09 ^{c,YZ}
	28	94.90±0.06 ^{c,X}	3.49±0.04 ^{b,XYZ}	4.89±0.04 ^{c,Z}
C	0	93.53±0.38 ^{c,X}	3.88±0.23 ^{a,Y}	6.1±0.49 ^{ab,Y}
	7	93.47±0.29 ^{d,X}	4.14±0.07 ^{a,XY}	6.62±0.72 ^{a,Y}
	14	93.48±0.25 ^{d,X}	4.28±0.06 ^{a,X}	6.78±0.47 ^{a,Y}
	21	91.47±0.27 ^{e,Y}	3.99±0.12 ^{a,XY}	9.25±0.03 ^{a,X}
	28	91.55±0.09 ^{e,Y}	4.16±0.06 ^{a,XY}	8.79±0.06 ^{a,X}
D	0	93.79±0.40 ^{c,X}	3.72±0.13 ^{ab,XY}	6.45±0.33 ^{a,X}
	7	94.29±0.03 ^{cd,X}	3.79±0.09 ^{b,XY}	5.94±0.10 ^{ab,Y}
	14	94.33±0.17 ^{c,X}	3.81±0.05 ^{b,X}	5.85±0.04 ^{b,Y}
	21	94.16±0.06 ^{d,X}	3.73±0.05 ^{b,XY}	5.99±0.03 ^{b,Y}
	28	93.99±0.21 ^{d,X}	3.56±0.11 ^{b,Y}	5.93±0.03 ^{b,Y}

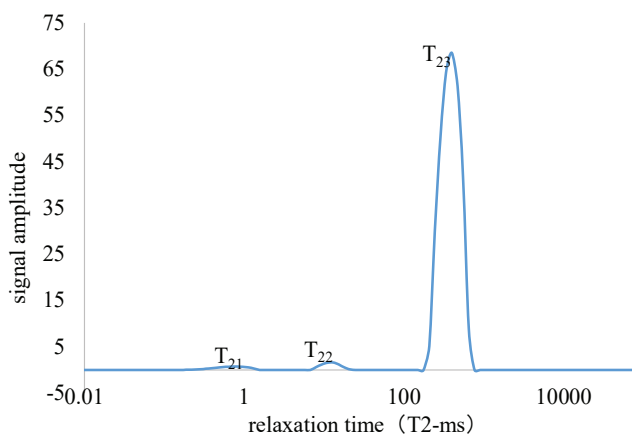


Fig. 13. Transverse relaxation time distribution of fermented milk drink added with DSF

Above were the results of NMR of the samples.

Within 21 days of storage, the signal amplitude of the samples significantly decreased as the addition amount of DSF rose, indicating that the content of free water was significantly decreased, and the solidification state of the samples was enhanced. On the 28th day of storage, the signal amplitude of all the samples was completely consistent, indicating that the solidification state of all the samples tended to be consistent at the later stage of storage. When the amount of DSF exceeded 6 %, the amount of DSF had little effect on the signal amplitude of the samples.

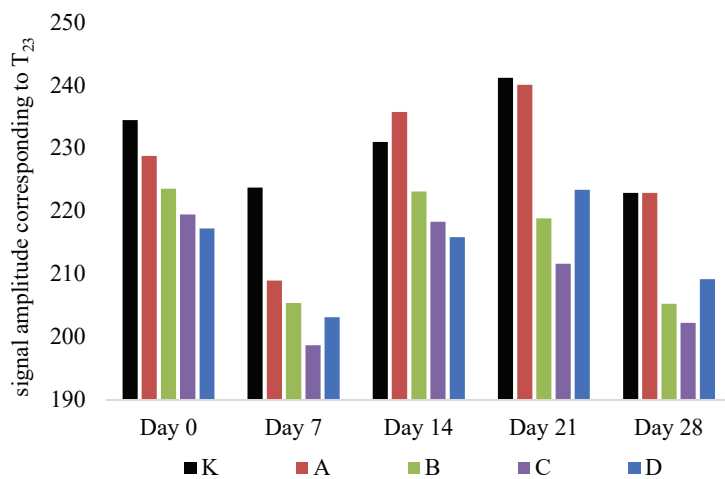


Fig. 14. Signal amplitude corresponding to T₂₃

5. 3. Results of the study of the chemical composition of fermented milk drinks with a DSF additive

The study of the chemical composition of fermented milk drinks with the DSF additive consisted in determining the content of fat, proteins, dietary fiber and vitamins. The results and studies are presented in Table 5.

Above was the chemical composition of fermented milk drinks samples.

The content of protein and dietary fiber increased with the addition of DSF. Fermented milk drink with 2 % DSF has a significantly increased protein and dietary fiber content. This is mainly because of the high level of dietary fiber and protein of DSF. With the increase of DSF amount, the content of VE in fermented milk drink also increased significantly, which was mainly due to the rich content of VE

of DSF. The addition of DSF had no significant effect on the fat content of fermented milk drink, because most of the fat in sesame seeds was separated as sesame oil when pressed.

Table 5

Chemical composition of fermented milk drinks samples (n=3, ±SD)

Sample	Fat, %	Protein, %	Dietary fiber,%	Vitamin E, mg/100 g
K	3.99±0.08 ^d	3.36±0.08 ^d	0.02±0.00 ^c	0.32±0.02 ^d
A	4.10±0.25 ^{cd}	4.30±0.13 ^c	0.68±0.11 ^c	0.93±0.01 ^{cd}
B	4.45±0.14 ^{bc}	4.89±0.13 ^{bc}	1.14±0.00 ^{bc}	2.02±0.16 ^c
C	4.85±0.06 ^b	5.74±0.37 ^{ab}	2.12±0.48 ^b	5.53±0.83 ^b
D	5.29±0.15 ^a	6.31±0.62 ^a	4.52±0.44 ^a	9.07±0.61 ^a

a – Values expressed as mean ± standard deviation of duplicate samples;
 b – K=control sample, A=fermented milk drink with 2 % DSF, B=fermented milk drink with 4 % DSF, C=fermented milk drink with 6 % DSF, D=fermented milk drink with 8 % DSF;
 c – Different superscript letters within a column indicate a significant difference (p<0.05) according to Tukey test

6. Discussion of the results of the study of fermented milk drinks with a DSF additive

The research results showed that the addition of DSF to a fermented milk drink has a positive effect on the physico-chemical and rheological properties.

The change of pH is shown in Table 1. In the sample without additives, the active acidity (pH) level immediately after fermentation exceeded the standard values by 0.2. With further storage, the pH decreased and corresponded to the normative indicators. The pH value in all the samples with the addition of DSF for 14 days was within the normal range (in accordance with DSTU 4417: 2005 pH=4.8–4.0). After 21 days of research, in the samples containing 4, 6 and 8 % additives, the pH decreased slightly and did not correspond to standard parameters. In a fermented milk drink containing 2 % additive, the pH level corresponded to standard values throughout the study (28 days). The pH value decreased significantly (P≤0.01) with the increasing amount of DSF, which could be related to the stimulation of lactic acid bacteria by fiber. Previous studies have also demonstrated that fermented milk drinks as a probiotic [29, 30] and dietary fiber had a potential prebiotic effect [31, 32].

No significant difference existed in pH among the samples with 4 %, 6 % and 8 % DSF, which may be caused by the high water-holding capacity of dietary fiber, the more DSF was added, the less free water there was, which would limit the vitality of probiotics. The substantial decrease occurred in active acidity (pH) in the first week for all the samples, and no further changes for the next 3 weeks of storage, because of post acidification of lactobacillus by using remaining lactose within 7 days.

The titratable acidity of the studied samples corresponded to the standard indicators (85–130 °T) only in the sample containing 2 % DSF additive in the first 7 days of storage. The rest of the indicators differed significantly from the norm. The addition amount of DSF had a significant effect on the acidity of fermented milk drinks. With the increase

of addition amount, the acidity of fermented milk drinks increased significantly. Storage time had a significant effect on the acidity of the products, with the increase of storage time, the acidity of fermented milk drinks increased significantly.

The WHC of fermented milk drink supplemented with DSF was significantly increased compared with blank one ($p < 0.05$), which was due to the high hydrophilic properties of dietary fiber and protein. There was no significant change in WHC among fermented milk drinks added with 2 %, 4 %, 6 %, 8 % DSF. There was no significant change in the WHC of all the samples during the storage period, which showed that the storage period had no significant effect on all the samples.

Compared with the samples without DSF, 2 % addition amount of DSF significantly increased the apparent viscosity ($p < 0.05$). No significant difference of apparent viscosity existed among the samples added with 2 %, 4 %, 6 %, 8 % DSF. The storage time had no significant influence on the apparent viscosity of all the samples.

The transverse relaxation time during storage was measured by NMR, the result is shown in Fig. 14. Three peaks were obtained after software fitting. The transverse relaxation time ranges T_{21} (0.051~1.831) and T_{22} (5.722~24.771) of fermented milk drink are small, the signal amplitudes are also small, they showed that T_{21} and T_{22} water molecules may be the water that is not easy to flow and less easy to flow in the pores that form the network structure, hydrogen bond strength was strong, or it could be the parts of the water that bind more tightly to large molecules like protein. The transverse relaxation time range of T_{23} (174.753~756.463 ms) is higher than that of T_{21} and T_{22} . The signal amplitude of M_{23} was also high, this indicates that the combination of T_{23} water molecules is relatively loose and has high fluidity [33, 34]. It can be seen from the peak ratio that the peak ratio of T_{21} , T_{22} and T_{23} was 5.364: 6.777: 330.257, T_{21} and T_{22} were not significant ($P > 0.05$), therefore, the peak area change of T_{23} was mainly studied. T_{23} and the signal amplitude M_{23} were taken as the main indexes, the moisture transfer characteristics of samples were compared.

The results of magnetic resonance imaging showed that fermentation ends before 7 days of storage. In this case, the content of free water was minimal. After 21 days, the fermented milk product began to lose its structure, and free water formed. Thus, it is desirable to limit the storage of the product for a period of 14 days.

The “*L*” value of the samples introduced with DSF was significantly decreased compared to the samples without DSF (*K*), the more DSF was added, the lower the “*L*” value was, which showed that the introduction of DSF significantly decreased the brightness of fermented milk drink. The “*a*” value of the samples introduced with DSF was significantly increased compared to the samples without DSF (*K*), the more DSF was added, the higher the “*a*” value was, which showed that the introduction of DSF significantly deepened the red color of fermented milk drink. The “*b*” value of the samples introduced with DSF was significantly increased compared to the samples without DSF (*K*), the more DSF was added, the higher the “*b*” value was, which showed that the introduction of DSF significantly deepened the yellow color of fermented milk drink. A factor that influenced the color of the products was the color of the ingredients. As the data showed in Table 3, the storage period had no significant effect on the brightness of all the samples ($P > 0.05$). With the prolongation of storage time,

the red color of all the samples showed a trend of deepening at first and then weakening ($P < 0.05$), and the red color was the deepest on the 14th day.

The introduction of DSF into fermented milk drink significantly decreased the brightness of the samples, and the red and yellow colors of the samples are significantly deepened. But when the addition amount of DSF reached 4 %, the brightness of the products kept steady. The storage period had no significant effect on the brightness of all the samples. With the prolongation of storage time, the red color of all the samples showed a trend of deepening at first and then weakening, and the red color was the deepest on the 14th day. On the 7th and 21st days of storage, the yellow color of the samples decreased significantly.

The analysis of the chemical composition showed that when the additive is added to the fermented milk drink, the content of fat, protein, dietary fiber and vitamins increases. Fermented milk drink with 2 % DSF significantly increased the protein and dietary fiber content. This is mainly because of the high level of dietary fiber and protein of DSF. With the increase of DSF amount, the content of VE in fermented milk drink also increased significantly, which was mainly due to the rich content of VE of DSF. The addition of DSF had no significant effect on the fat content of fermented milk drink, because most of the fat in sesame seeds was separated as sesame oil when pressed.

In addition, the need to add a large amount of additive has not been identified. In terms of physicochemical and rheological parameters, it is advisable to use from 2 to 4 % of the additive. At the same time, an increase in the DSF content up to 4 % allows an almost twofold increase in the content of dietary fiber and vitamin.

So far, there has been no research on adding DSF to kefir. The features of this method are to study the influence of DSF addition on the water-holding capacity of kefir by combining the NMR method and the influence of DSF on the color of kefir by combining chromaticity measurement.

The limitation of this study is the lack of the microstructure analysis of DSF kefir, which leads to the lack of in-depth research on changes in DSF kefir's rheological properties.

The disadvantage of this study is that DSF was treated at a high temperature (200–300 °C) in practice, which leads to a slight scorched taste. This problem can be solved by adding DSF without high-temperature treatment. However, the effect of DSF without high-temperature treatment on the characteristics of kefir remains to be further explored.

7. Conclusions

1. In terms of physicochemical and rheological parameters, it is advisable to add from 2 to 4 % DSF into kefir, within this range, the product has good viscosity, antioxidant capacity, WHC and appropriate acidity.

2. The addition of DSF can significantly improve the acidity, water-holding capacity and viscosity of kefir ($P < 0.05$); significantly reduce the pH value and brightness of the product ($P < 0.05$), but when the addition amount of DSF reached 4 %, the brightness of the products kept steady; significantly deepen the red and yellow colors of the sample; significantly affect the free water content, the signal am-

plitude decreased significantly in the first week, indicating that the content of free water was the lowest, the retention power of the sample was the highest, and the solidification state was the best.

The storage period showed no significant effect on WHC, apparent viscosity and brightness in 4 weeks, while showed a significant effect on pH, acidity during the first week, ($P < 0.05$) and no further changes for the next 3 weeks of storage for the lack of lactose; significant effect on the red and yellow colors of the products, the red color of the products was the deepest on the 14th day, the yellow color of the samples decreased significantly on the 7th and 21st days of the storage period. The WHC and apparent viscosity of fermented milk drink supplemented with DSF were significantly increased compared to blank one ($p < 0.05$), which was due to the high hydrophilic properties of dietary fiber and protein. There was no significant change in WHC and apparent viscosity among fermented milk drinks added with 2 %, 4 %, 6 %, 8 % DSF.

3. The addition of DSF could significantly increase the protein content, dietary fiber content and VE content of the

product ($P < 0.05$). The more DSF was added, the higher the content of protein, dietary fiber and VE were. 2 % addition amount of DSF can significantly improve the chemical content of products.

The dairy industry should focus on the fortification of milk products with DSF. DSF-supplemented milk should be advised for diabetic, heart patients and people who pursue health. Sesame should be used in various other products based on dairy and beverages. Active ingredients from sesame should be extracted and used as nutraceuticals in different food products including chapattis and other traditional products.

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