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The study of the effect of oregano essence and Gellan gum on quality and shelf life of non-carbonated heat treated Doogh

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ABSTRACT

In present study, the effect of different levels of oregano essential oil (0, 250 and 500 ppm) and gellan gum (0, 0.25, 0.5 and 0.75%) on the physicochemical, microbial and sensory properties of doogh during 60 days of storage in refrigerator conditions. The results showed that gellan gum and oregano essential oil had no significant effect ( $p < 0.05$ ) on the chemical properties of doogh including acidity, pH, fat and dry matter. Adding gellan gum to doogh increased the viscosity and stability of Doogh. Oregano essential oil significantly increased ( $p < 0.05$ ) DPPH radical inhibition and reduced oxidation. The minimum inhibitory concentration and the minimum bactericidal concentration of oregano essential oil on *Escherichia coli* are 1.6 and 3.12 mg/ml, respectively, and for *Staphylococcus aureus*, 0.4 and 0.80 mg/ml, respectively. was reported. In terms of sensory characteristics, the use of oregano essential oil and gellan gum did not have a significant effect ( $p < 0.05$ ) on the color of doogh, but the essential oil improved the flavor of doogh, gellan gum improved the mouth feeling. The doogh containing 0.5% gellan gum and 250 ppm of oregano essential oil had more acceptability and stability in terms of sensory properties and was free of contamination, which was recognized as the best treatment.

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## 1- Introduction

Buttermilk is a dairy drink that has a special place among the drinks available in the market, and it is also a drink obtained from the lactic fermentation of milk and one of the old products of Iran, which can be considered one of the drinks made from yogurt with low viscosity. This resource is a suitable environment for the growth of spoilage microorganisms as it contains very nutritious substances. By producing compounds, these micro-organisms cause changes in flavor, bloating, reduction of shelf life and ultimately spoilage of this valuable product [1]. Spoilage of buttermilk, especially in the summer season, is one of the problems faced by today's society and it causes economic decline. Molds and yeasts in buttermilk can be briefly referred to cold-oriented bacteria, yeasts and coliforms. The initiators pointed to cold-oriented coliform bacteria as a possible source of contamination. Due to the non-standard design of buttermilk factories in Iran, there are naturally different pollution sources in these factories, and therefore it is necessary to determine critical control points in all factories and organize automatic control systems in order to eliminate or minimize the threat of pollution [1]. Mold and yeast growth is directly related to storage time; In such a way that it increases with the increase of storage time and due to the removal of competing bacteria from the environment and favorable conditions of mold and yeast in the environment. Today, the consumption of spoiled dairy products, including buttermilk, can cause many problems, such as diarrhea, abdominal cramps, vomiting, and even fever [1]. Bacteria such as *salmonella*<sup>1</sup> *Clostridium*<sup>2</sup>,

and *Escherichia coli*<sup>3</sup> as well as yeasts like *Rhodotorula*<sup>4</sup>, *Cloromyces Marcianos*<sup>5</sup> and *Saccharomyces cerevisiae*<sup>6</sup> It is one of the factors of pollution and spoilage in some food products, including yogurt and related products such as buttermilk drink in Iran, and it can cause the packaging to swell due to the production of buttermilk [1]. Various plant essential oils, including oregano essential oil, have been Alone or with each other in different doses to improve the sensory and quality characteristics of buttermilk, they have been investigated and used, and these yeasts and spoilage bacteria can sometimes be beneficial to achieve new immunization methods in the food industry and even beverages as preservatives. to be used naturally [1]. Oregano, whose extract is usually extracted by steam distillation in most researches and combined with a gas chromatography mass spectrometer GC/MC Determined. Oregano is one of the species of the genus *demand* from family *Labiatae* It grows in humid areas and its active ingredient is pulgun and its disinfectant is menthol, and other compounds such as piperitone oxide and cinerol etc. are found in abundance in it. The essential oils in oregano have strong antibacterial properties against microorganisms such as *assalmonella* And .... and yeasts like *Staphylococcus aureus*<sup>7</sup> and .... [2]. Therefore, the use of natural essential oils and even hydrocolloid in dairy products, including buttermilk, can increase the shelf life of products and reduce economic losses, and even prevent the appearance and value of the product and delay spoilage. or to destroy microorganisms in dairy products. One of

<sup>1</sup> -*salmonella*

<sup>2</sup> -*clostridium*

<sup>3</sup> -*AND. coli*

<sup>4</sup> -*Rhodotorula*

<sup>5</sup> -*cloramaysis marksiyanos*

<sup>6</sup> *saccharomyces cerevisiae*

<sup>7</sup> -*staphylococcus aureus*

the major problems in the production of acidic milk drinks is their two phases during production and storage, which is caused by low viscosity. pH Their effect on the sedimentation of proteins is low [3,4]. Basically, the stability of casein micelles in pH The naturalness of milk is due to the placement of kappa-casein on the surface of casein micelles, which prevents the micelles from approaching each other with the formation of hairy layers on their surface and spatial and electrostatic repulsion. Today, hydrocolloid has many uses, among which we can name suspension stabilization and suspension of solid particles in drinks. The results have shown that with the addition of all hydrochlorides, increased stability, prevention of biphasing and precipitation of protein in buttermilk [5]. Biphasing is one of the important problems of buttermilk, which is directly related to rheological properties. To prevent the accumulation of proteins and hydration in these products, various stabilizers are used, the most common of which are hydrocolloids. So, as mentioned, adding hydrocolloids or gums is one of the methods of increasing stability, preventing two phases and preventing protein precipitation in dairy products. Hydrocolloids or gums cause the stability of dairy products by increasing the apparent viscosity of the product or due to colloidal interactions of the type of spatial hindrance and electrostatic repulsion [6]; Therefore, the purpose of this research is to use hydrocolloid and essential oil and oregano in order to improve the quality of heat-treated buttermilk without gas.

## 2- Materials and methods

### 1-2- Buttermilk production method

Buttermilk production was carried out according to the national standard 10528 [7]. The produced buttermilk is heat-treated without gas. First, fresh cow's milk was standardized in terms of fat content and

different amounts of gellan gum were added to the milk according to the table below; Then heat treatment was applied at 95°C for 15 minutes, and after cooling at 45°C, it is our normal starter (*Streptococcus Thermophilus*<sup>8</sup> And *Lactobacillus Bulgaricus*<sup>9</sup>) Christian Hansen Model Company YOU It was inoculated. After being kept in a greenhouse for about 3 hours at a temperature of 45 degrees Celsius and ripening pH 50% by volume water and 0.8% salt were added to it, and after complete homogenization, it was pasteurized at 75 degrees Celsius for 1 minute and homogenized under 150 bar pressure, then oregano essential oil was added to it. And after packaging, it was stored in the refrigerator for 2 months.

### 2-2- buttermilk tests

#### 1-2-2- Physical and chemical tests

##### 1-1-2-2- Measurement of acidity in terms of lactic acid percentage

Acidity in terms of lactic acid Based on national standard 2852 It was measured by titration method using 0.1 normal sodium in the vicinity of phenolphthalein [8].

$$\text{Acidity \%} = \frac{N \times 0.009 \times 100}{M}$$

where in:

N: The amount of 1.0 milliliters of soda consumed is normal.

M: Sample weight

##### 2-1-2-2- pH measurement

It was measured according to the national standard 2852. For this purpose after calibrating pH m with buffer solutions 4 and 7, some buttermilk was poured into the bottle and the amount pH BypH meter was measured [8].

<sup>8</sup> -*Streptococcus thermophilus*

<sup>9</sup> -*Lactobacillus bulgaricus*

**3-1-2-2 percentfat**

Fat was measured by the Gerber method (Iranian National Standard No. 384) with 90% sulfuric acid and isoamyl alcohol and with the help of a butyrometer for buttermilk.[9].

**4-1-2-2- Amount of dry matter**

The amount of dry matter was measured according to the national standard 637 by heating in an oven with a temperature of 105 degrees Celsius until the complete evaporation of moisture.[10].

**6-1-2-2-Stability check of buttermilk**

In order to determine the stability of buttermilk samples, a 50 ml graduated cylinder was used. In this way, 50 milliliters of each buttermilk sample was poured into cylinders and covered with aluminum foil. After 15 days, their stability was calculated as a percentage using the following equation [11].

$$\text{(Percent)Buttermilk stability rate} = \frac{\text{Primary buttermilk volume} - \text{serum volume}}{\text{Primary buttermilk volume}} \times 100$$

**7-1-2-2- Viscosity measurement**

A viscometer was used to measure the viscosity of buttermilk samples; So that the buttermilk samples were poured into the cylindrical chamber of the machine. The apparent viscosity of the samples was also measured at a speed of 10 rpm. According to the instructions of the manufacturer, the suitable spindle for viscosity measurement is the spindle that shows a torque higher than 10% at the desired speed. All tests were performed at a temperature of 20 degrees Celsius and under the same conditions; So that the viscosity of the samples was read at a speed of 10 rpm and after 10 seconds of spindle rotation [12].

**8-1-2-2- Measurement of antioxidant activity**

Antioxidant activity of buttermilk samples by method DPPH was measured Samples at speed rpm 4000 was centrifuged for 10 minutes and 100 microliters of the supernatant solution was mixed with 2.9 milliliters of DPPH (2-diphenylpicrylhydrazyl)  $\mu\text{m}60$  mixed in methanol. The resulting mixture was kept in a dark environment for 30 minutes and finally the absorbance of the samples was read at 517 nm. Free radical inhibition percentage DPPH It was calculated from this relationship [13].

*Inhibition percentage*

$$= (1 - A_s/A_c) * 100$$

That  $A_c$  Adsorption of the control sample and  $A_s$  The absorption of the sample contains essential oils.

**3-2- Microbial tests****2-3-1-Determining the minimum inhibitory concentration of MIC (mg/mL)**

At this stage, oregano essential oil was prepared in concentrations of 25, 12.5, 6.25, 3.12, 1.60, 0.8, 0.4, 0.2, 0.1, 0.05 and 0 milligrams per milliliter. became. To determine the minimum inhibitory concentration, broth microdilution method was used in sterile 96-well microtiter plates. First, in each well, 50 microliters of brain heart infusion agar culture medium (BE) and 50 microliters of bacterial suspension cfu/ml  $10^6 1.5 \times$  was poured. The positive control sample of the well including culture medium and microbial suspension and the negative control sample of culture medium and essential oil were considered.

Microtiter plates for 30 seconds with round rpm 250 was mixed in a shaker. Then the cultures were incubated at 37 degrees Celsius for 24 hours. After the mentioned time in order to determine the minimum inhibitory concentration (MIC) the wells

were visually checked for turbidity and the minimum concentration that prevented the growth of bacteria or the absence of turbidity evident with the control group was determined as the minimum inhibitory concentration in mg/ml [14].

### 2-3-2- Determining the minimum lethal concentration of MBC (mg/mL)

Minimum lethal concentration based on results MIC Was determined. The amount of 50 microliters of the wells in which the growth of bacteria has stopped on Mueller Hinton Agar culture medium (MHA) sterilized, cultured; Then the plates were placed in a 37°C incubator for 24 hours. The last dilution of the extract, which was able to kill 99.9% of the primary living bacteria, was considered as the minimum lethal concentration of the essential oil [14].

**1-2-3-2- *Staphylococcus coagulase* Positive<sup>10</sup>**  
counting *Staphylococcus coagulase* Positive according to the national standard of Iran with number 6806-1 during enrichment in Canton Giolti medium in anaerobic conditions at 37 degrees Celsius and then transferring on Brad Parker agar at 37 degrees Celsius and counting shiny black colonies with a transparent halo and yellow sediment. [15].

### 2-3-2-2- *Escherichia coli*

counting *Escherichia coli* According to the national standard of Iran with number 2946, with enrichment in lauryl sulfate broth environments *Escherichia coli* Broth was made at 41.5 degrees Celsius and transferred to eosin methylene blue culture medium and kept in a greenhouse for 24 hours at 37 degrees Celsius. After performing the necessary biochemical tests on the green colonies with metallic polish in order to confirm the bacteria *Achrichiacle*, the number of colonies was counted [16].

### 4-2- Sensory evaluation of the product

Sensory evaluation of buttermilk was done using 10 evaluators who are familiar with buttermilk according to the five-point hedonic test on the characteristics of color, smell, texture, taste, and general opinion. In such a way that the maximum score of 5 means that the sample is very good and the score of 1 is the lowest score that indicates that the sample is very bad [12].

## 3- Results

### 1-3- Buttermilk physicochemical tests

#### 1-1-3- Acidity in percent

Natural acidity is caused by the presence of caseins, acid phosphates and citrates in milk, while the developed acidity is caused by the production of lactic acid by starters. The results showed that there was no significant difference between the treatments in terms of acidity on any of the investigated days, but during the storage period, the acidity of the buttermilk significantly increased ( $<0.05$ ). *P* Increased. At the end of the storage period, the lowest level of acidity (0.96%) was found in the control sample and the highest level of acidity (1.05%) was found in the sample containing 0.75% gellan gum. ppm500 oregano essential oils were observed (Table 1-3).

#### 2-1-3-pH

The results of investigating the effect of different concentrations of oregano essential oil and gellan gum on the amount of pH Buttermilk during the storage period is given in Table 1-3. The results showed that there was a significant difference between the treatments on each of the investigated days

<sup>10</sup> -Positive *staphylococcus coagulase*

In terms of amount pH There was not, but during the maintenance period of the rate pH buttermilk significantly ( $>0.05$ ). *P*) Decreased. At the end of the minimum maintenance period pH (4.14) in the sample containing 0.75% gellan gum + 500 ppm oregano essential oil and the highest acidity (4.20) was observed in the control sample.

### 3-1-3-Total solids in percentage

The results of investigating the effect of different concentrations of oregano essential oil and gellan gum on total buttermilk solids during the storage period are given in Table 1-3. The results showed that there was no significant difference between the treatments in terms of the amount of total solids on any of the investigated days. Also, during the storage period, the amount of total buttermilk solids changes significantly ( $>0.05$ ). *P*) did not have. At the end of the storage period, the lowest amount of total solids (5.24%) in the control sample and the highest amount (5.32%) in the sample containing 0.75% gellan gum. ppm500 oregano essential oils were observed.

### 4-1-3-Fat by percentage

The results of investigating the effect of different concentrations of oregano essential oil and gellan gum on buttermilk fat content during the storage period are given in Table 1-3. The results showed that there was no significant difference between the treatments in terms of the amount of fat in each of the investigated days. Also, during the storage period, there were significant changes in butterfat content ( $<0.05$ ). *P*) did not have.

### 5-1-3-Stability of buttermilk in terms of percentage

The results of investigating the effect of different concentrations of oregano essential oil and gellan gum on the stability of buttermilk during the storage period are given in Table 1-3. The results showed that there was a significant difference between the treatments in terms of stability on each of the investigated days; In such a way that there was always a significant difference between the control treatment and the treatments containing gellan gum. On the first day, there was no significant difference between the treatments containing gellan gum, but with the passage of time and during the other days examined (20th and 40th day), the treatment containing 0.75% gellan gum had the most stability with a significant difference. In the control treatment, during the storage period, the buttermilk stability decreased significantly, in the treatment containing 0.25% gellan gum, no significant change in the buttermilk stability was observed from the 20th day until the end of the storage period. In the treatment containing 0.75% of olean gum, no significant change in buttermilk stability was observed during the storage period. At the end of the storage period, the lowest level of stability (46.30%) in the control sample and the highest level of stability (99.16%) in the sample containing 0.75% gellan gum+ppm500 oregano essential oils were observed.

**Table 3-1. The mean of physicochemical and stability characteristics of Doogh\* during the storage\*\* period**

Days of Storage	Treatments	Acidity %	pH	Total Solid %	Fat%	Stability%
1	A(Control)	0.00aD±0.78	0.02aD±4.61	0.06aA±5.18	0.05aA±1.00	1.00bA±99.36
	B	0.01aD±0.78	0.02aD±4.57	0.03aA±5.21	0.05aA±0.96	0.500aA±99.50
	C	0.01aD±0.77	0.02aD±4.55	0.08aA±5.24	0.05aA±1.00	0.00aA±100.00
	D	0.01aD±0.77	0.02aC±4.53	0.08aA±5.26	0.02aA±1.03	0.00aA±100.00
	A(Control)	0.02aC±0.83	0.02aC±4.42	0.09aA±5.20	0.05aA±1.05	1.12dB±64.76

20	B	0.02aC±0.84	0.03aC±4.41	0.10aA±5.23	0.02aA±0.96	1.89cB±94.83
	C	0.02aC±0.85	0.03aC±4.38	0.09aA±5.25	0.08aA±1.00	0.76bAB±97.16
	D	0.02aB±0.87	0.04aB±4.36	0.07aA±5.27	0.07aA±1.01	0.28aA±99.83
40	A(Control)	0.02aB±0.91	0.04aB±4.33	0.11aA±5.21	0.07aA±1.01	1.41cC±52.50
	B	0.01aB±0.93	0.03aB±4.31	0.07aA±5.23	0.07aA±0.98	1.89bB±93.83
	C	0.03aB±0.94	0.04aB±4.27	0.12aA±5.26	0.07aA±0.98	1.00bB±96.00
	D	0.04aA±0.97	0.05aB±4.25	0.09aA±5.28	0.05aA±1.03	0.50aA±99.16
60	A(Control)	0.01aA±0.96	0.04aA±4.20	0.07aA±5.22	0.10aA±1.00	0.75cD±46.30
	B	0.05aA±0.99	0.02aA±4.18	0.05aA±5.25	0.12aA±1.01	2.00bB±92.93
	C	0.03aA±1.01	0.06aA±4.17	0.05aA±5.28	0.02aA±0.96	1.52abB±95.66
	D	0.07aA±1.05	0.06aA±4.14	0.08aA±5.28	0.05aA±0.96	0.50aA±99.16

\* Means with small common letters in each column and caps lock common letters in each row do not have a significant difference at the 5% probability level.

\*\* A:Control sample; B: 0.25 GG+ 100 ppm OEO; C: 0.5 GG+ 250 ppm OEO; D: 0.75 GG+ 500 ppm OEO

### 2-3-DPPH free radical inhibition

The results of investigating the effect of different concentrations of oregano essential oil and gellan gum on radical inhibition DPPH in buttermilk during the storage period is given in Figure 1-3. The lowest rate of radical inhibition DPPH (54.4%) in the control

sample and the highest amount (71.9%) in the sample containing 0.75% gellan gum+ppm500 oregano essential oils were observed and the results showed that there was a significant difference between the treatments in terms of radical inhibition.DPPH There was.

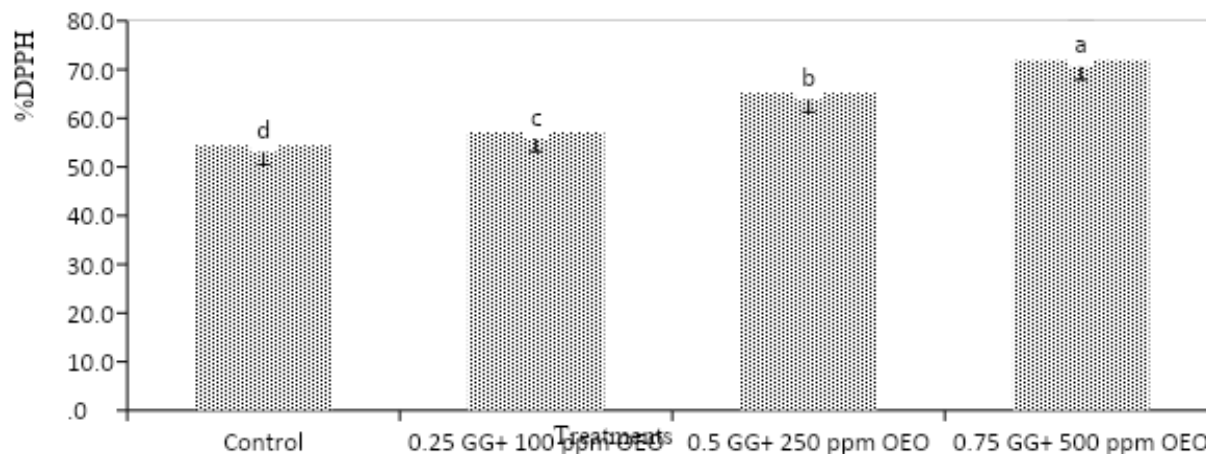


Fig. 3-1. Comparison of DPPH free radical inhibition percentage in different treatments\*

\* Columns with common letters are not significantly different from each other at  $p>0.05$

### 3-3- Viscosity (m pa.s)

Complex viscosity is an important parameter to describe the rheological properties of a material. The value of the complex viscosity of the samples is shown as a function of the angular frequency in Figure 2-3. As can be

seen, the viscosity of the complex of all the samples decreases with the increase in angular frequency, which is the highest in the control sample and in the sample containing 0.75% gellan gum.ppm500 oregano essential oil is the least; In other words, adding gellan gum increased the viscosity of buttermilk.

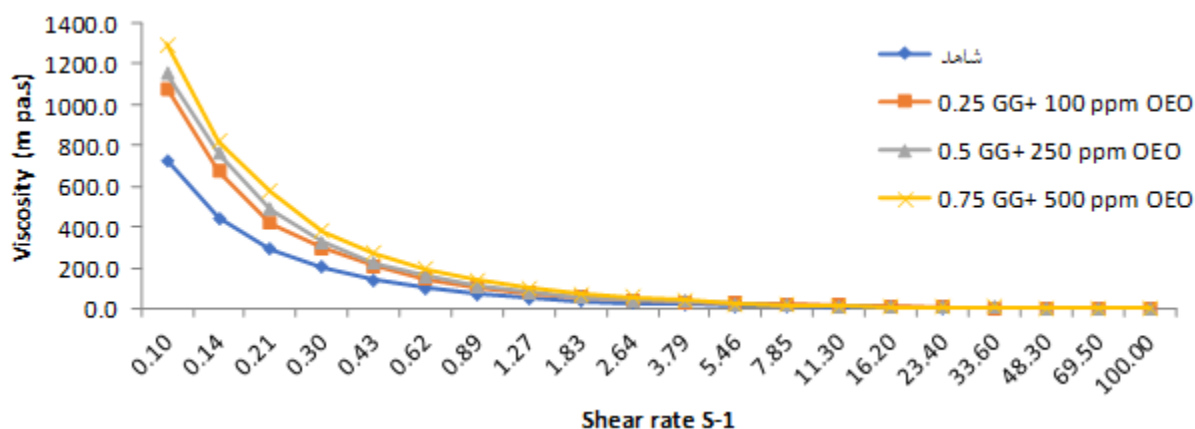


Fig. 3-2. Comparison of viscosity in different treatments

#### 4-3- Microbial characteristics

In the present study, the absence of coliforms, *Staphylococcus Oreos* And *Escherichia coli* It confirms the proper hygiene measures during the preparation of buttermilk and proper storage conditions.

#### 1-4-3-minimum inhibition concentration and minimum bactericidal concentration(mg/mL)

Table 3-2. Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) of oregano essential oil (mg/ml)

Bacteria	MIC	MBC
<i>Escherichia coli</i>	1.6	3.12
<i>Staphylococcus aureus</i>	0.8	0.4

#### 5-3- Sensory evaluation

##### 1-5-3- color sensory score

The results of investigating the effect of different concentrations of oregano essential oil and gellan gum on buttermilk color score during the storage period are given in Table 3-3. The results showed that there was no

ResultsMBC AndMIC *Escherichia coli* And*Staphylococcus Oreos* It is given in Table 2-3. Minimum inhibitory concentration and minimum bactericidal concentration of oregano essential oil on zinc*Escherichia coli* 1.6 and 3.12 mg/ml and direction, respectively*Staphylococcus Oreos* 0.4 and 0.80 mg/ml were reported respectively.

significant difference between the treatments in terms of color score on any of the investigated days. Also, during the maintenance period, there were significant changes in the buttermilk color score ( $<0.05$ ).P) did not have (Table 3-3).

Table 3-3. Color and taste scores\* of different treatments\*\* during the storage period

Days of Storage	Treatments	Color	Flavor
	A(Control)	0.52aA±4.5	0.63bA±3.8



1	B	0.52aA±4.5	0.66abA±4.0
	C	0.51aA±4.4	0.52aA±4.5
	D	0.51aA±4.4	0.56abA±4.1
30	A(Control)	0.52aA±4.5	0.48bA±3.7
	B	0.51aA±4.5	0.56abA±3.9
	C	0.48aA±4.3	0.51aA±4.4
	D	0.67aA±4.3	0.66abA±4.0
60	A(Control)	0.51aA±4.4	0.51bA±3.6
	B	0.48aA±4.3	0.42abA±3.8
	C	0.48aA±4.3	0.63aA±4.2
	D	0.78aA±4.2	0.56abA±3.9

\*Means with small common letters in each column and caps lock common letters in each row do not have a significant difference at the 5% probability level.

\*\*A:Control sample; B: 0.25 GG+ 100 ppm OEO; C: 0.5 GG+ 250 ppm OEO; D: 0.75 GG+ 500 ppm OEO

### 2-5-3-sensory rating of taste

The results of investigating the effect of different concentrations of oregano essential oil and gellan gum on buttermilk taste score during the storage period are given in Table 3-3. The results showed that in each of the examined days, the treatment always contained 0.5 GG+ 250 ppm OEO. It had the highest taste score with a significant difference compared to the control treatment. Also, during the maintenance period, there were significant changes in the buttermilk taste score ( $>0.05$ ). *P* did not have.

### 4-Discussion

The use of oregano essential oil stimulates the activity of initiator bacteria and reduces the amount of pH. Sánchez-Zamora, Colleagues (2022) observed that the use of oregano essential oil had a significant effect on the amount of pH. There was no cheese [19]. Also, Shahbazi and Shavizi (2019) observed that during the storage period pH of Yogurt decreased. Decrease in pH can be due to the post-acidification process<sup>11</sup>. During storage, it is due to the activity of beta-galactosidase enzyme, which can remain

active at temperatures from zero to five degrees Celsius [18]. Some researchers reduce pH. It is known because of the remaining enzymes from the activity of the initiator bacteria during the fermentation process [20]. The results of the total solids of the buttermilk samples showed that the addition of oregano essential oil and gellan gum did not have a significant effect on the total solids of the buttermilk. Quintana Martinez et al. (2022) reported that pumpkin puree hydrocolloid had no effect on the approximate composition of the acidic milk drink [21]. The results of buttermilk fat showed that the addition of oregano essential oil and gellan gum had no significant effect on buttermilk fat. Yousefi et al. (2017) reported that angelica essential oil had no significant effect on buttermilk fat content [20].

The results of buttermilk stability showed that adding gellan gum to buttermilk significantly increased the stability. Basically, the stability of casein micelles in pH. The naturalness of milk is due to the placement of capcaseins on the surface of casein micelles, which prevent the micelles

<sup>11</sup> Postacidification

from approaching each other by forming hairy layers on their surface and spatial and electrostatic repulsion mechanisms. If for any reason the hairy layers are separated (broken by milk scaling enzymes) or disintegrated (loss of effective net charge with reduction pH, increase in ionic strength and decrease in solubility), instability occurs in casein micelles. Because with the acidification of the environment, calcium phosphate is gradually removed from the micelle, the negative electric charge of the micelle decreases and the casein micelle disintegrates. The prevention of serum separation is related to the molecular structure of the hydrocolloid used. If the used gum is pregnant, it causes the stability of the fermented beverages through spatial hindrance and electrostatic repulsion. It seems that the mechanism that increases the stability of buttermilk in this study is the increase in viscosity and the trapping of protein particles in a molecular network created by the gum used [22]. In samples containing gellan, two phases in The length of the storage period was significantly reduced compared to the control sample, which is due to the formation of gel particles resulting from the accumulation of gellan molecules. If the fluid has a yield stress, the yield stress prevents the movement of particles. The use of gellan leads to the production of a product that has a yield stress. This yield stress is caused by gellan connections either alone or connected with proteins, which by forming a three-dimensional network and trapping protein and fiber particles in this network, makes the product stable [23]. Boroskiet al. (2012) observed that the addition of oregano extract or essential oil did not affect the physical stability of dairy beverages during storage [24]. Casein particles in fermented products due to pH being down and close to their isoelectrics begin to accumulate and precipitate during the storage period. One of

the ways to prevent this problem is to use stabilizers [25]. Adding oregano essential oil to buttermilk significantly increases radical inhibition. DPPH and reduced oxidation. The phenolic compounds in the essential oil act as antioxidants by donating a hydrogen atom (which acts as a free radical receptor) by interrupting oxidation chain reactions or by chelating metals. Oregano essential oil is rich in phenolic compounds. These compounds play a vital role in neutralizing and inhibiting free radicals [26]. Boroskiet al. (2012) observed that oregano extract contained 269 mg gallic acid equivalent per gram and by radical method DPPH indicator It was given that it has strong antioxidant activity. Oregano essential oil with  $IC_{50}$  Equivalent to 26.7 micrograms/ml of high radical scavenging potential DPPH It showed practicality in dairy drink [24]. Sahin et al. (2004) rosmarinic acid, caffeic acid, coumaric acid, quercetin and carvacrol are the compounds responsible for the antioxidant activity of oregano [27]. Gomes and Colleagues (2016) observed that oregano essential oil is rich in phenolic compounds with antioxidant activity and works well to protect mayonnaise against oxidation reactions [26]. The high viscosity of buttermilk containing gellan gum can be attributed to the formation of an electrostatic complex by increasing the interaction between positively charged protein groups and anionic groups of gellan gum during the pasteurization process. The presence of a bridge between casein particles by gellan-casein aggregates creates a strong network against flow [28]. Khannieri et al. (2015) reported that the use of 0.6% carboxymethyl cellulose increased the viscosity of buttermilk [26]. Quintana Martinez et al. (2022) reported that pumpkin puree hydrocolloid increased the viscosity and improved the rheological properties of acidic milk drink [21]. *Escherichia coli* Than *Staphylococcus Aureus* It had a higher minimum inhibitory

concentration and a higher minimum bactericidal concentration. Molodi et al. (2018) minimum inhibitory concentration and minimum bactericidal concentration of oregano essential oil on *Escherichia coli* reported 1.25 and 2.5 mg/ml respectively [29]. Also, Karimi Nik et al. (2019) minimum inhibitory concentration and minimum bactericidal concentration of oregano essential oil on *Staphylococcus Oreos* reported 0.15 and 0.3 mg/ml, respectively [30]. Thymol and carvacrol are known as bioactive compounds of oregano essential oil and they break apart the outer cell wall of bacteria, increase the permeability of the cytoplasmic membrane and cause death. They become cells. Oregano essential oil usually has more inhibitory activity on gram-positive bacteria than gram-negative bacteria, because gram-negative bacteria have a lipopolysaccharide layer on the outer membrane [19]. The significant difference in the sensitivity of bacteria to natural antimicrobial compounds can be caused by the outer cytoplasmic membrane covering the thin peptidoglycan structure of Gram-negative microorganisms, which limits the diffusion of hydrophobic components through its lipopolysaccharide coating. The results showed that the addition of oregano essential oil and gellan gum had no significant effect on the buttermilk color score. Khodashanas and Joki (2020) observed that the use of gellan gum, xanthan gum and seed gum did not have a significant effect on the color of buttermilk [31]. treatment containing ppm 500 essential oil due to the strong taste of oregano essential oil, less acceptance than the treatment containing ppm 250 showed. Since the treatments were stored in suitable temperature conditions during the storage period, no significant decrease in the taste score was observed, also due to the low fat content of the buttermilk, the treatments do not undergo severe oxidation and taste changes. Taste is one of the most important

qualitative aspects of foods and beverages, It will follow the consumer. The most important factors affecting the taste of dairy products are the amounts of acetaldehyde and diacetyl, which are produced by the fermentation of dairy products. The results showed that the addition of oregano essential oil and gellan gum had a significant effect on the buttermilk taste score. The reason for this is people's interest in flavored drinks; Since ancient times, the flavoring of dairy products, especially yogurt and buttermilk, has been done with plant powder. The positive effect of plant extracts such as mint on the taste of dairy products has been proven in some experiments that have already been done. Tavakli et al. (2013) observed that the use of aloe vera and thyme essential oils improved the taste of buttermilk [32].

#### 5- General conclusion

Treatments of gellan gum and oregano essential oil have a significant effect on acidity. pH It did not have fat or dry matter. The addition of gellan gum to buttermilk increased the viscosity and stability of buttermilk, so that in the treatment containing 0.75% gellan gum, almost no biphasing was observed during the storage period. The use of oregano essential oil causes a significant increase ( $p < 0.05$ ) Radical inhibition DPPH and reduced oxidation. Application Oregano essential oil and gellan gum did not have a significant effect on the color of buttermilk, however, it improved the taste score of buttermilk and gellan gum improved the consistency and mouthfeel.

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بررسی تأثیر اسانس پونه کوهی و صمغ ژلان بر ویژگی‌های کیفی و ماندگاری دوغ گرمادیده بدون گاز

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## چکیده

## اطلاعات مقاله

در تحقیق حاضر تأثیر سطوح مختلف اسانس پونه کوهی (۰، ۲۵۰ و ۵۰۰ ppm) و صمغ ژلان (۰، ۰/۲۵، ۰/۵ و ۰/۷۵ درصد) بر خصوصیات فیزیکوشیمیایی، میکروبی و حسی دوغ در طی ۶۰ روز نگهداری در شرایط یخچال بررسی گردید. نتایج نشان داد که صمغ ژلان و اسانس پونه کوهی تأثیر معنی‌داری ( $p < 0.05$ ) بر خصوصیات شیمیایی دوغ شامل اسیدیته، pH، چربی و ماده خشک نداشت. افزودن صمغ ژلان به دوغ موجب افزایش ویسکوزیته و پایداری دوغ گردید. اسانس پونه کوهی موجب افزایش معنی‌دار ( $p < 0.05$ ) مهار رادیکال DPPH و کاهش اکسیداسیون گردید. حداقل غلظت بازدارندگی و حداقل غلظت باکتری‌کشی اسانس پونه کوهی بر روی *اشرشیاکلا*ی به ترتیب ۱/۶ و ۳/۱۲ میلی‌گرم/میلی‌لیتر و جهت *استافیلوکوکوس اورئوس* به ترتیب ۰/۴ و ۰/۸۰ میلی-گرم/میلی‌لیتر گزارش شد. از نظر خصوصیات حسی، کاربرد اسانس پونه کوهی و صمغ ژلان تأثیر معنی‌داری ( $p < 0.05$ ) بر رنگ دوغ نداشت اما اسانس موجب بهبود امتیاز طعم دوغ و صمغ ژلان موجب بهبود احساس دهانی گردید. دوغ حاوی ۰/۵ درصد صمغ ژلان و ۲۵۰ ppm اسانس پونه کوهی از نظر خصوصیات حسی دارای مقبولیت و پایداری بیشتر و فاقد آلودگی بود که به عنوان تیمار برتر شناخته شد.

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