



## Evaluation of the viability of starter bacteria and physicochemical properties in functional yogurt enriched with wheat germ powder and a mixture of processed plant essential oils

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### ABSTRACT

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According to the increasing demand of advanced societies for the consumption of functional foods, especially in dairy products, the use of natural-based materials such as plant essences in the field of enriching food products with their antimicrobial and antioxidant compounds, as well as the use of wheat germ powder as a rich source of dietary fibers and vitamins and minerals and their effects on the viability of starter bacteria (due to their prominent role in improving human health) are seem to be necessary. Investigating the effects of using processed essential oils and wheat germ powder simultaneously in yogurt is considered in this research. Sensory, physicochemical characteristics and bacterial counting of set yogurt including *Streptococcus thermophilus* and *Lactobacillus bulgaricus*, were tested during the storage period to study the simultaneous effect of plant essential oils and sprouted wheat powder. The essential oils used included tarragon and thyme with different proportions and at three levels (400, 1200, 2000 microliters per liter of yogurt). Yogurt physicochemical tests such as acidity and water holding capacity, hedonic sensory test, rheology test (apparent viscosity) and microbial count tests were conducted to check the viability of starter bacteria in yogurt during the 14-day storage period. Based on the results obtained from the analysis of variance of the tested samples, the concentration of essential oil significantly ( $p < 0.05$ ) reduced the acidity and viability of starter bacteria of the product, but it had not a significant effect on the viscosity the sensory characteristics. In this regard, thyme and wheat germ powder showed a negative and unfavorable effect, especially in high concentrations. Thyme had a higher antimicrobial effect on yogurt starter bacteria, while tarragon showed less antimicrobial activity. The sensory score of samples containing tarragon has increased slightly during storage, so in general tarragon is prone to be used in yogurt production. Wheat germ powder first had an increasing and then decreasing effect on the acidity of the product during the storage time of yogurt. Also, the addition of wheat germ powder increases the viscosity and water holding capacity of the product, but in high amounts it reduces the sensory score, and therefore its use in yogurt should be controlled. In general, wheat germ powder has a positive effect on the viability of both starter bacteria counted in this research, which can be due to the supply of nutrients for their further growth.

## 1. Introduction

Multipurpose foods are foods that contain one or more special compounds that have a practical effect on improving the health and well-being of the consumer. These useful components may be naturally increased in the food or may be intentionally added to it in the production process [1] and cause health effects such as regulation of metabolic activities, fitness, improving the functioning of the digestive system, heart and blood vessels. and so on. The people of Japan, America, and Europe are among the most consumers of such foods, and research has shown that gender, age, education level, and financial status are among the factors that are effective in the consumption of extra-beneficial foods [2].

Yogurt is one of the fermented products with a uniform texture and is consumed in most parts of the world under different titles. The popularity of this product is firstly due to its high amount of calcium, vitamins, minerals and low fat content, and secondly due to its health-giving effect and inhibition of harmful bacteria and increased life span [3]. Among probiotic food products, probiotic dairy products, especially fermented dairy products, have more acceptance and consumption. Today, probiotic fermentation products include about 25% of all fermentation products, among which yogurt is the most accepted and widely used probiotic product in the world [4].

According to their type, plant essential oils have many properties such as flavoring, antimicrobial, strengthening the immune system, antioxidant, biological properties, medical effects, etc. It has been done [5]. Garden thyme is one of the oldest medicinal plants and spices, whose active ingredients are widely used in the food industry as preservatives and flavors, and its essential oil has strong antibacterial and antifungal properties. The most important constituents of thyme essential oil are thymol, carvacrol and paracimol [6]. The most important components of tarragon essential oil are estragole, alpha and beta-pinene, sabinene and limonene. The effective ingredients of this plant are used in the pharmaceutical industry to prepare drugs to

lower blood pressure and help digest food in the digestive system. It has been reported that tarragon in animal models leads to a decrease in blood sugar and insulin [7]. Tarragon is also used to strengthen appetite and as a general body tonic. Tarragon is widely used as a flavoring agent in food and canning industries [8, 9].

Consumption of sprouted grains is increasing in different regions of the world. Sprouting grains in a certain period of time leads to an increase in the activity of hydrolyzing enzymes, an increase in the amount of essential amino acids, total sugar and group B vitamins and a decrease in dry matter, starch and anti-nutritional compounds. The digestibility of protein and starch increases due to partial hydrolysis during germination. However, the amount of nutritional value improvement is influenced by the type of grain, seed quality and germination conditions. In sprouted wheat flour, crude protein, crude fat, vitamin thiamine and riboflavin are significantly higher compared to normal flour [10].

In general, yogurt starters are not able to consume maltose *Streptococcus thermophilus* It can consume starch while *Lactobacillus bulgaricus* is not able to do this [11 and 12]. Consumers prefer yogurts with high and optimum sweetness to yogurts with low sweetness [13]. The consumption of wheat germ can have a favorable position due to increasing the nutritional value, improving the taste (sweetness) and rheological properties of yogurt. Wheat germ alone does not have a good taste and it is generally recommended to mix it with yogurt. Increasing the consumption of wheat germ is one of the appropriate solutions to reduce the anti-nutritional effects and increase the nutritional value of wheat flour. Considering the increase in yogurt consumption in the world and Iran and taking into account the importance of using natural plant compounds such as essential oils and wheat germ powder, in this research, the production of molded flavored yogurt using different essential oils and sprouted wheat powder and their effect on sensory properties. Its microbial and physicochemical properties

have been investigated. Also, the aim of this article is to investigate the effect of using the mentioned essential oils on the characteristics of the yogurt dairy product, so that the optimal concentration of these essential oils and wheat germ powder can be found in the product so that it does not have a negative effect on the marketability of the product. Also, while formulating a useful product, the effect of adding these essential oils on the survival of the initiator bacteria is also one of the research objectives.

## 2- Materials and methods

### 2-1- Materials

The cultivation environment of MRS<sup>1</sup> and TSA<sup>2</sup> from Merck company, processed essential oils (dissolved in water) including tarragon and thyme from Zardband Tehran company, Thermophil culture starter with CH1 code and in DVS form<sup>3</sup> They were purchased from Christian Hansen, Denmark.

### 2-2- Preparation of starter and wheat germ powder

1.1 gram of yogurt starter was dissolved in 80 ml of sterile milk, this process was done on a shaker at 80 revolutions per minute in aseptic conditions and at a temperature of about 30 degrees Celsius for about 30 minutes until the complete and uniform dissolution of the starter. . In order to prevent the precipitation of insoluble fibrous materials and help the appearance of yogurt, the particle size of wheat germ powder was reduced to a maximum of 50 microns using a home mill and sieving.

### 2-3- Yogurt production

Yogurt base mixture was prepared by adding non-fat dry milk (2% w/w) and milk. Then it was pasteurized at a temperature of 85 degrees Celsius for 30 minutes and immediately cooled down to a temperature of 42 degrees Celsius with stirring in ice water. Under the hood and hygienic conditions, one milliliter of the prepared starter solution was first transferred to polystyrene cups with a capacity of 280

milliliters. In the next step, about 150 grams of base milk and then the required essential oil separately or in combination (in a ratio of 1:1) of 400, 1200 and 2000 microliters per liter were added to the milk and stirred for 10 seconds. Finally, by adding milk, the weight of the samples was adjusted to 250 grams. After sealing, the prepared samples were placed in a greenhouse at a temperature of 42 degrees Celsius for 4 hours. After this period, it was transferred to a refrigerator equipped with an air circulation system at a temperature of 5 degrees Celsius and was kept at this temperature until the next tests on days 7, 1 and 14.

### 2-4- Viscosity measurement

The apparent viscosity of yogurt samples was determined in centipoise at a temperature of 5 degrees Celsius. According to the manufacturer's instructions, the torque value should be between 10 and 100%. Probe number 64 and 30 rpm was selected. Since yogurt is considered a non-Newtonian fluid, first the samples were mixed manually and gently for 40 seconds in the same direction and conditions, and then the data reading was done automatically after 15 seconds [14].

### 5-2- Measurement of acidity

Acidity was measured according to the national standard number 2852 (1), using 10 grams of yogurt sample and 10 grams of cold boiled distilled water and titration with 0.1 normal soda solution in the presence of phenolphthalein reagent until a pale pink color appeared. Acidity was calculated according to Dornick's degree [15].

### 6-2- Measurement of water storage capacity

The sample containing about 20 grams of yogurt (Y) was centrifuged for 10 minutes at 4 degrees Celsius and 1250 g. Then the separated water (W) was weighed and using formula number (1) the amount of water storage capacity was calculated in terms of grams of separated water per hundred grams of yogurt [16]. It should be mentioned that all measurements were done in two repetitions.

1. Man, Rogosa, Sharpe Agar  
2. Tryptic Soy Agar

3. Direct Vat Set

$$\text{WHC}^4 = (Y - W)/Y \times 100$$

## 2-7- counting bacteria in yogurt

One gram of yogurt sample was diluted with 9 ml of sterile 1.5% peptone water solution and homogenized on a vortex machine. Then, successive and suitable dilutions were prepared from it. One milliliter of the last three dilutions was added to the desired plates in two repetitions. Cultivation was done as a pore plate (mixed) and after the formation of bacterial colonies, counting was done. MRS agar culture medium with pH = 5.2 for counting *Lactobacillus bulgaricus* In anaerobic conditions, the temperature is 43 degrees Celsius and for 72 hours, as well as ST agar culture medium for counting *Streptococcus thermophilus* They were used in aerobic conditions with a temperature of 37 degrees Celsius for 24 hours [17].

## 2-8- Sensory evaluation

General sensory evaluation was done by the judgment of 15 selected evaluators of both sexes (in the age range of 20 to 40 years) who were selected based on the level of accuracy and sensitivity. The overall acceptance of the product was evaluated on a 9-point hedonic scale (extremely pleasant = 9 and extremely unpleasant = 1). The evaluators were asked to score the samples based on their overall acceptability [18 and 19].

## 9-2- Statistical analysis

From the mixed design<sup>5</sup> And incomplete factorial design was used to determine the number of experiments. The obtained data were statistically analyzed with Design Expert software (version 10). Analysis of variance test was used to determine significant effects at the 5% probability level.

## 3- Results and discussion

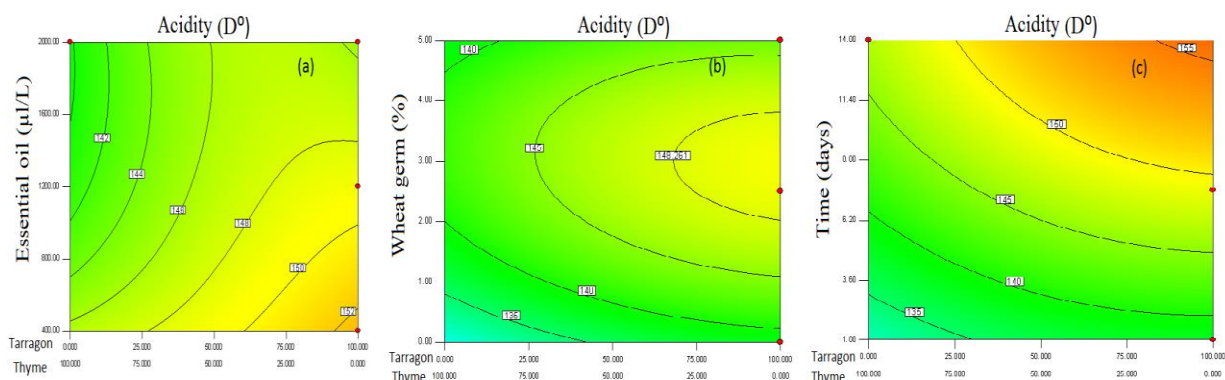
### 3-1- Acidity

By increasing the amount of essential oil in all samples, the acidity decreases. This decrease is more in the case of thyme essential oil, followed by horseradish essential oil, which is

justified due to the increase in the antimicrobial activity of the essential oil in high concentrations, so the graphs related to the microbial population of the starters also show this phenomenon. At low values of the ratio of these two essential oils, with the increase in the ratio of thyme to tarragon, the acidity decreases significantly, which can be justified considering the greater antimicrobial effects of thyme (Figure 1a). As the storage time increases, the acidity increases. Acidity is also dependent on the ratio between essential oils, so it is less in high ratios of thyme essential oil to tarragon essential oil. In other words, with increasing shelf life, the antimicrobial activity of thyme essential oil is still higher than that of tarragon essential oil, which can indicate that the essential oils are not metabolized during storage (Figure 1c). Increasing the acidity of the starter yogurt by using the plant essential oils of Calpura Maddi (*Your pole*) was also reported by Mahmoudi et al. (2014)[20].

4. Water Holding Capacity

2. Mixture design



**Fig 1** The simultaneous effect of the ratio between thyme and tarragon and the concentration of essential oil (a), the percentage of wheat germ powder (b) and time (c) on acidity

Arslan and colleagues [21] have studied the effect of basil, marzah and coriander essential oils with concentrations of 10, 50 and 100 microliters per liter of yogurt as a flavoring agent. In his research on acidity and watering, a significant difference has been observed between the samples containing essential oil and the control. Yogurt samples containing basil essential oil had a lower level of acidity. The amount of watering increased with increasing the concentration of essential oil in the case of basil and coriander. Adding essential oil has no significant effect on viscosity. In relation to the total population of yogurt starters (lactic acid bacteria), the samples containing basil and coriander showed a lower number and the samples containing savory showed a higher number compared to the control sample. Marzah essential oil has less wateriness and a higher sensory score than the control, and in general, with the increase in the amount of essential oil, the characteristic of taste and smell has reached an unacceptable level.

Regarding the effect of sprouted wheat powder on acidity, according to Figure 3b, with the increase in the percentage of wheat germ, the acidity first increases and then decreases in higher amounts, and this increase or decrease has a significant dependence on the ratio of thyme-tarragon essential oil. The initial increase in acidity can be attributed to the increase in available nutrients and sugars and the subsequent decrease to the increase in osmotic pressure and therefore to the decrease in microbial activity.

Regarding the effect of storage time on acidity, according to Figure 3c, acidity increases with increasing storage time. According to this contour plot on different days, the acidity is also dependent on the ratio between essential oils, so

that it is less in high ratios of thyme essential oil to tarragon. In other words, with increasing shelf life, the antimicrobial activity of thyme essential oil is still higher. There is tarragon, which can indicate that essential oils are not metabolized during storage.

It should be noted that the minimum acidity (117.52) was obtained on the first day with the amount of essential oil of 2000 microliters per liter containing equal proportions of tarragon and thyme and without the presence of sprouted wheat. Based on this, tarragon essential oil has a lesser effect and thyme has a greater effect on reducing acidity. Also, the amount of essential oil and storage time have a direct relationship with the amount of acidity and wheat germ in the amount. Medium increases acidity.

### 2-3- Sensory evaluation

In high ratios of thyme essential oil to tarragon, with increasing concentration, the sensory score decreases significantly, but in low ratios of thyme to tarragon, the decrease in sensory score is small. In other words, in samples containing only tarragon, increasing the amount of essential oil from 400 to 2000 microliters per liter causes a slight decrease in the sensory score (Figure a2). The reason for the reduction of the sensory score by thyme, especially in high concentrations, is its bitter taste, while tarragon has no bitterness and has a cooling effect. In the case of samples containing 100% tarragon essential oil, the sensory score increases slightly but significantly with the increase in storage time.

In a similar study, improving the sensory properties of yogurt with the addition of 0.01% essential oil and 0.2% Chivil plant extract (*Ferulago angulata* (Schlecht.) Boiss) has been reported [22]. Also Ghalem et al. (2013) yogurt enriched with rosemary essential oil (*Rosemary officinalis*) subjected to sensory evaluation and

stated that the enrichment of yogurt with this essential oil with a concentration of 1.4 g/liter improved the sensory characteristics such as smell, taste and consistency (texture) [23].

Figure 2b shows the effect of the amount of wheat germ powder on the sensory evaluation, in which with the increase in the percentage of wheat germ powder, the sensory score decreases significantly in different ratios of

thyme to tarragon essence, which can be attributed to the bitter aftertaste and unfavorable color of the powder. Wheat germ is especially relevant in high amounts. Recently, the addition of wheat germ protein to yogurt has been tested by other researchers, including the research of Ghelich et al. But in higher concentrations, it has reduced the sensory properties of the product[24].

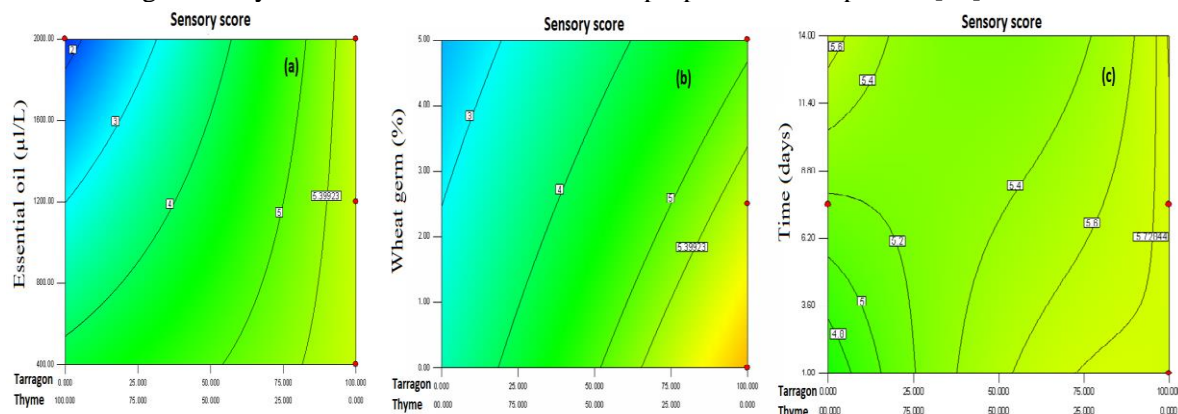


Figure 2- The simultaneous effect of the ratio between thyme and tarragon and the concentration of essential oil (a), the percentage of wheat germ powder (b) and time (c) on sensory characteristics

### 3-3- Viscosity

By increasing the ratio of thyme essential oil to tarragon, the viscosity decreases significantly, which is probably due to the decrease in acidity. In all samples, with the passage of time, the viscosity of yogurt has increased, which is probably due to the increase in acidity and casein hydration [25]. An increase in viscosity in yogurt due to the addition of essential oil and extract of the Chuil plant has also been reported by researchers [22]. In general, by increasing the amount of essential oil, the viscosity decreases in different ratios of thyme to tarragon, but in ratios close to 100% of tarragon, increasing the amount of essential oil up to about 800 microliters per liter has increased the viscosity, although the amount of this increase is not significant. An increase of more than 800 microliters per liter of tarragon essential oil has caused a significant decrease in viscosity (Figure a3). In general, the concentration of essential oil is much more important than the type of essential oil in reducing viscosity. In relation to the effect of storage time and composition of thyme and tarragon on viscosity, it should be noted that with increasing storage time, viscosity has increased in all ratios.

In different proportions of thyme and tarragon essential oil, increasing the percentage of wheat germ powder has caused a very large and significant increase in viscosity, which is probably due to the presence of high amounts of starch and its compounds in wheat germ powder. As shown in Figure b3, in fixed amounts Wheat germ powder By increasing the ratio of tarragon to thyme, the viscosity decreased significantly but slightly He finds that this issue is probably related to the very low antimicrobial effect of tarragon and therefore the decomposition of some of the polymers of wheat germ powder, especially starch, by bacteria and therefore the reduction of viscosity. In one of the most recent researches, Ghelich et al. (2022) evaluated the functional properties of hydrolyzed wheat germ protein and its effect on the physicochemical properties of frozen yogurt, which reported an increase in viscosity and firmness of the product as a result of increasing the concentration of wheat germ.[24].

Figure 3c shows the effect of storage time on viscosity, which in different proportions of essential oil, with increasing time storage, the viscosity has increased significantly, the reason for this can be the increase in water retention capacity of milk proteins [26].

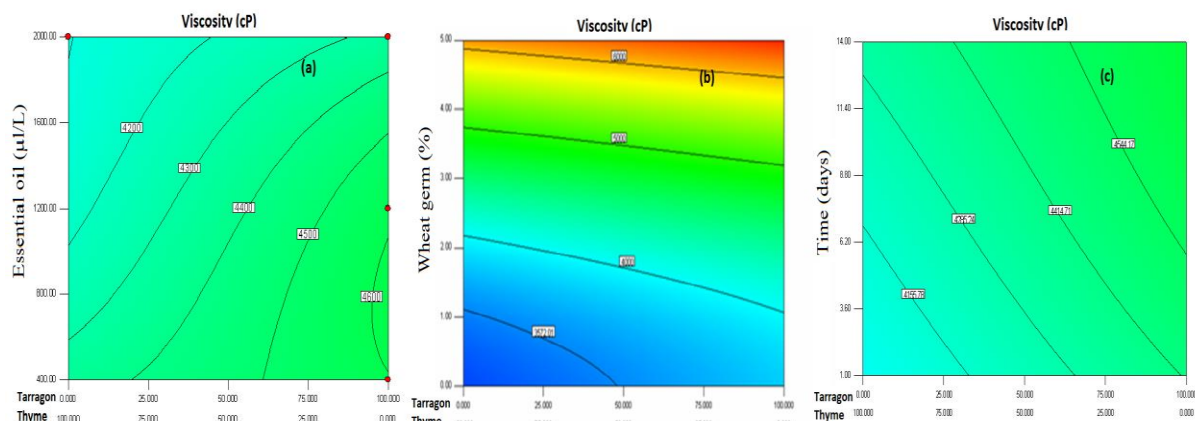


Figure 3- The simultaneous effect of the ratio between thyme and tarragon and the concentration of essential oil (a), the percentage of wheat germ powder (b) and time (c) on viscosity

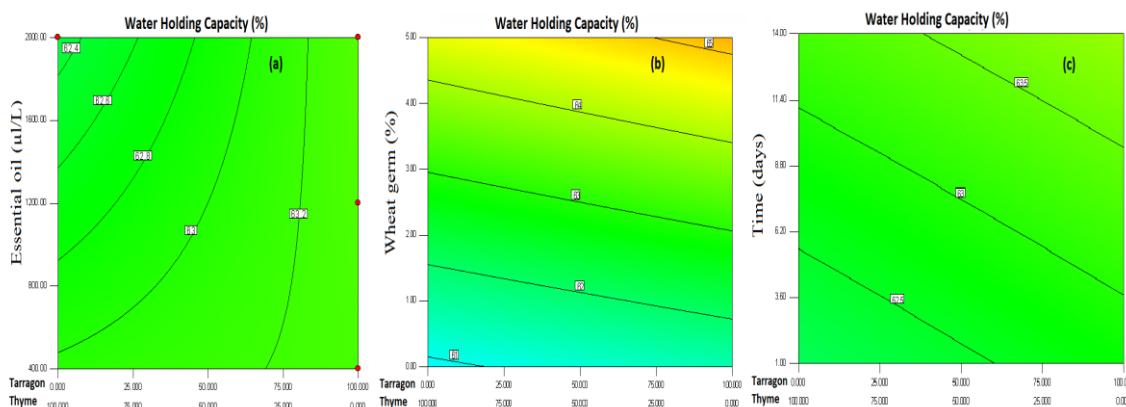
### 3-4- Water holding capacity (WHC)

As shown in Figure 4, the increase in the ratio of thyme essential oil to tarragon decreases the water holding capacity. In general, with the increase in the amount of essential oil, the water holding capacity decreases. 3:1 tarragon to thyme is very little. Probably, the small amount of tarragon essential oil, whose main composition is estragole, increases the water holding capacity indirectly (microbial production). There are also some indications of the action of essential oils on proteins [27]. By increasing the concentration of tarragon essential oil, the amount of water holding capacity increases, but in the case of thyme, with increasing concentration of essential oil, a significant decrease in water holding capacity has been obtained. Also, with the increase of storage time, the capacity of water storage increases, which can be due to the increase and completion of water absorption of proteins [25]. In another research, the essential oil of chivil plant also had a positive effect on the amount of water holding capacity in yogurt [22]. In another study by Mahmoodi et al. (2021), the reduction of synergism in starter yogurt

enriched with caraway essential oil (*Bunium peachBoiss*) has been reported [28].

Figure 4b shows the effect of wheat germ powder on the water holding capacity, where different proportions of tarragon and thyme essential oils increase the amount of wheat germ powder, the water holding capacity increases significantly, mainly by increasing the amount of related starch polymers. Is. By increasing the ratio of tarragon to thyme, the amount of water holding capacity decreases, which is due to the low antimicrobial effects of tarragon compared to thyme, and therefore the decomposition of polymers, especially starch, in ratios above tarragon, Water storage capacity decreased.

Figure 4c, Increase Water storage capacity During the storage time, it shows that it can be due to the increase and completion of protein hydration [26]. Also, by increasing the ratio of tarragon to thyme, the amount Water storage capacity increase It is found that due to the low amount of essential oil (the main amount is estragole, which is an unsaturated compound), this issue is probably due to the specific effects of essential oil on microbial products such as exopolysaccharides and therefore increasing water retention capacity.



**Fig 4** The simultaneous effect of the ratio between thyme and tarragon and the concentration of essential oil (a), the percentage of wheat germ powder (b) and time (c) on water holding capacity

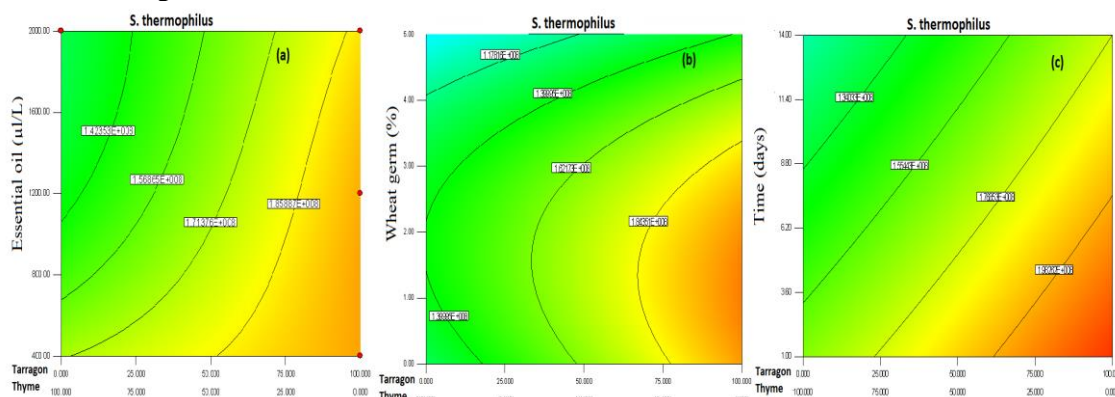
### 3-5- Counting *Streptococcus thermophilus*(St)

By increasing the ratio of thyme to tarragon, the population of St has decreased significantly, which can be justified considering the greater antimicrobial effects of thyme compared to tarragon. Also, in different proportions of tarragon and thyme essential oil, with the increase in the amount of essential oil, a significant decrease in the population of this bacterium has been observed, which can be justified considering the high antimicrobial effects of thyme. However, its antimicrobial effect against St. is not very high ( $>10^8$  cfu/ml) which is probably related to the presence of compounds such as fats and proteins in milk that have a reducing effect on the antimicrobial activity of essential oils. Finally, during the storage time, the population of St has shown a significant decrease, and this population decrease is significant with the increase in the ratio of thyme to tarragon on constant and the same days.

As shown in Figure 5b, with the increase of

wheat germ powder, the population increased to about 2% and after that, a decreasing trend was observed. It is wheat germ powder. Also, in addition to the existence of a significant interaction between the amount of wheat germ powder and the ratio of thyme to tarragon essential oil, with the increase in the ratio of thyme to tarragon, the population of St decreases significantly, which can be justified considering the greater antimicrobial effects of thyme compared to tarragon.. In a relatively similar study, Kamel et al. (2021) investigated the effect of adding inulin with ratios of 0.2 to 0.6% on the viability of yogurt starter bacteria and reported that inulin significantly reduced the viability of starter bacteria. *Bifidobacterium bifidum* It has become curd during storage and has also acted as an anti-mold[29].

According to Figure 5c, with increasing storage time, there is a significant decrease in the population of St. It has been shown that on constant and the same days, with the increase in the ratio of thyme to tarragon, this decrease in population is also greater.



**Fig 5** The simultaneous effect of the ratio between thyme and tarragon and the concentration of essential oil (a), the percentage of wheat germ powder (b) and time (c) on the number of *Streptococcus thermophilus* bacteria

In various researches, the reduction of St population due to increasing acidity with increasing storage time has been reported [30].

The effect of essential oils of clove, cinnamon, cardamom and peppermint on the growth of yogurt bacteria has been investigated [31]. For



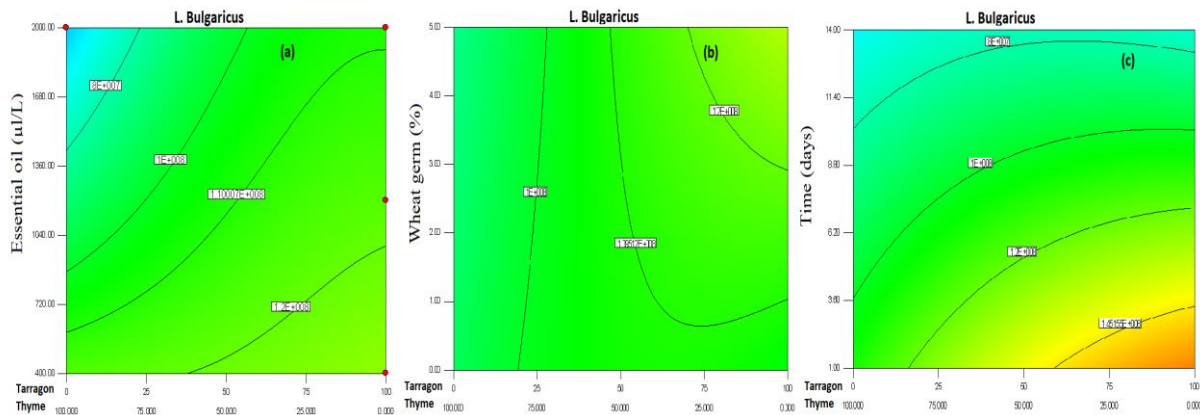
example, cinnamon is the most effective antibacterial against *Streptococcus termophilus* and *Lactobacillus bulgaricus*. It has been reported that it completely stops the growth of yogurt bacteria at a concentration of 0.005-0.5% in milk, and in this regard, cardamom, cloves and mint are ranked next.

In a research on buttermilk dairy product, it was shown that the number *Streptococcus termophilus* and *Lactobacillus bulgaricus* in local buttermilk samples produced with mint, thyme, garlic spices and the control sample, it decreased significantly during the storage time, but the effect of the mentioned spices on the number of initiator bacteria was not significant compared to the control sample [32]. Karajhian and colleagues (2005) have shown that the effect of different concentrations of kakuti extract (from the thyme family) on the number of initiator bacteria was not significant until the 16th day, and from that time on, only the extract at a concentration of 4000 microliters per liter had a significant effect on the number of initiator bacteria in Yogurt, while different concentrations of kakuti essential oil did not have a significant effect on the number of yogurt initiator bacteria during the storage time [33]. In another study, Mahmoudi et al. (2014) investigated the effect of adding the essential oil of Calpura Maddi (*Your pole*) addressed starter yogurt and reported that the highest viability of starter bacteria was observed in yogurt samples containing 60 ppm of this essential oil [20].

### 3-6-Counting *Lactobacillus bulgaricus*(Lb)

By increasing the amount of essential oil and the ratio of thyme to tarragon, the population of Lb decreases significantly, meanwhile, the antimicrobial effect of tarragon on Lb is insignificant, while thyme essential oil has a significant decreasing effect. In the case of tarragon, by increasing the concentration of essential oil to a certain extent, population *Lactobacillus* First, it increases and then decreases with the further increase of the

concentration of essential oil. In different ratios of tarragon and thyme essential oils, with increasing storage time, a significant decrease in Lb population is observed. Also, population changes at the end of the storage period between high and low proportions of thyme are less than this population difference in the first days of storage. In this regard, other researchers have also investigated the effect of adding different essential oils such as ginger and chamomile at different levels (0.2 and 0.4%) on the survival rate of the initiator bacteria and have declared that the highest survival rate of The initiator has been observed in samples containing 0.4% essential oil [34]. Also, Mahmoudi et al. (2021) investigated the physicochemical characteristics and viability of yogurt starter bacteria and stated that the highest viability of *Lactophilus acidophilus* in samples enriched with cumin essential oil (*Bunium peach* Boiss) has been observed [28]. Figure 6b shows the effect of wheat germ powder on Lb, in which, with the increase in the percentage of wheat germ powder in high ratios of thyme to tarragon, a statistically significant increase in the population of Lb is shown, and also in low ratios of thyme, this population increase It is more likely that this issue can be due to the increase in nutrients, the decrease in antimicrobial activity of essential oils due to the presence of wheat germ powder proteins. It should be noted that Lb is resistant to high amounts of wheat germ powder (low amounts of water activity). In this regard, Demirci et al. (2017) investigated the effect of adding rice bran up to 3% on the viability of the initiator bacteria. *Lactobacillus casei* in yogurt and reported a significant positive effect of rice bran on the survival of this bacterium in yogurt [35]. In Figure 6a, in different proportions of tarragon and thyme essential oils, with increasing storage time, a significant decrease in Lb population can be seen, and the population changes at the end of the storage period between high and low proportions of thyme are less than the population difference in the first days of storage.



**Fig 6** The simultaneous effect of the ratio between thyme and tarragon and the concentration of essential oil (a), the percentage of wheat germ powder (b) and time (c) on the number of *Lactobacillusbulgaricus* bacteria Sarabi-Jamab et al. (2008) The effect of thyme on activity*Lactobacillus acidophilus* have been investigated as the starter bacteria of the starter yogurt and obviously the survival*Lactobacillus acidophilus* During the storage of yogurt at 4 degrees Celsius and during specific time intervals, they have been examined. These researchers have reported that the number of initiator bacteria is significantly reduced after 7 days of storage. Also, no significant difference was observed between the control sample and the samples containing different concentrations of thyme essential oil (zero to 300 microliters/liter) at the 95% confidence level [36]. In another study, Mehdizadeh et al. (2019) investigated the viability of initiator bacteria*Bifidobacteriumbifidum* And*Lactobacilluscaseidill* in yogurt enriched with essential oil (*Anethumgraveolens*) and presented yogurt samples containing 100 ppm of dill essential oil as samples with the highest survival rate of the mentioned bacteria[17].

#### 4- Conclusion

The results of this research showed that thyme and tarragon essential oils have strong antimicrobial effects on*Lactobacillus bulgaricus* They showed that, especially in the case of thyme, it was used in the highest concentration and also during storage, population reduction.*Lactobacillus bulgaricus* It is less than one logarithmic cycle. In general, the different characteristics of the yogurt samples produced showed that the combination of tarragon and thyme with a concentration of 2000 microliters per liter is suitable. Also, in this regard, high concentrations of tarragon and the minimum concentration studied did not have much difference in sensory evaluation. Thyme essential oil is not suitable even in low

#### 5- Resources

[1] Villaño, D., Gironés-Vilapana, A., GarcíaViguera, C. & Moreno, D. A. 2016. Development of Functional Foods. Innovation Strategies in the Food Industry, 191–210.

[2] Tur, J. A., and Bibiloni, M. M. 2016. Functional Foods. Encyclopedia of Food and Health, 157–161.

[3] Hosseini, M. Lashkari, F. and Qanbarzadeh, B. 2019. The effect of the combination of inulin-gelatin and polydextrose-gelatin hydrocolloids on the rheological and sensory properties of prebiotic low-fat yogurt. Food Industry Research. 30(4), 123-135. [In Persian]

[4] Mohammadi, R. Rouzitalab, A. Shahabbaspour, Z. and Mortazavian, A. 2013. Study of microbiological, biochemical and organoleptic properties in the starter soy yoghurt. Iranian J Nutr Sci Food Technol. 7 (5):149-158 [In Persian]

[5] Shakerian, A. Sohrabi, M. J. and Ghasemi Pirbalouti, A. 2012. Effect of Bakhtiari celery (*Kelussia odoratissima* Mozaff) on sensory properties and shelf life of set yogurt. Journal of Medicinal Herbs. 3(1): 41-48. [In Persian]

[6] Habibi, S. Ehteshamnia, A. Fatehi, F. and Ghaderi, A. 2020. Investigation of  $\gamma$ -Terpinene synthesis and  $\alpha$ - Terpinene synthesis reductoisomerase genes expression

- and its relation to monoterpene carvacrol biosynthesis in *Thymus vulgaris* cv. 'Varico 3'. *Genetic Engineering and Biosafety Journal*. 9 (2) :161-169 [In Persian]
- [7] Nowrozi, F. Hojjati, M. Joyndeh, H. and Barzegar, H. 2017. Using tarragon essential oil in mayonnaise as a natural antioxidant. *Food Industry Research*. 28(3), 85-99 [In Persian]
- [8] Khajeh, N. and Mohammadi, N. 2022. Investigating the effect of degumming coating containing two essential oils of Shirazi thyme and tarragon on the chemical composition and texture of fried potato wedges. *Food industry engineering research*. Doi: 10.22092/FOODER.2021.353821.1298 [In Persian]
- [9] Hassanzadeh, H., Alizadeh, M., and Bari, M. R. 2016. Kinetic Modeling and Optimization of Milk Coagulation Affected by Several Prevalent Cheesemaking Factors and Essence Addition. *International Journal of Food Engineering*. 12(5): 421-428.
- [10] Hassanzadeh, H., Ghanbarzadeh, B., Galali, Y., and Bagheri, H. 2022. The physicochemical properties of the spirulina-wheat germ-enriched high-protein functional beverage based on pear-cantaloupe juice. *Food Science & Nutrition*. <https://doi.org/10.1002/fsn3.2963>
- [11] Aryana, K. J., & Olson, D. W. 2017. A 100-Year Review: Yogurt and other cultured dairy products. *Journal of Dairy Science*. 100(12): 9987-10013.
- [12] Bintsis, T. 2018. Lactic acid bacteria as starter cultures: An update in their metabolism and genetics. *AIMS microbiology*. 4(4): 665-684.
- [13] Chollet, M., Gille, D., Schmid, A., Walther, B., and Piccinali, P. 2013. Acceptance of sugar reduction in flavored yogurt. *Journal of dairy science*. 96(9): 5501-5511.
- [14] Pang, Z., Xu, R., Luo, T., Che, X., Bansal, N., & Liu, X. 2019. Physicochemical properties of modified starch under yogurt manufacturing conditions and its relation to the properties of yogurt. *Journal of food engineering*. 245: 11-17.
- [15] Iranian National Standard, Number 2852. 2006. Milk and its products, the determination of acidity and pH, the Institute of Standards and Industrial Research of Iran. [in Persian]
- [16] Santiago-García, P. A., Mellado-Mojica, E., León-Martínez, F. M., Dzul-Cauich, J. G., López, M. G., and García-Vieyra, M. I. 2021. Fructans (agavins) from *Agave angustifolia* and *Agave potatorum* as fat replacement in yogurt: Effects on physicochemical, rheological, and sensory properties. *LWT*. 140, 110846.
- [17] Mehdizadeh, T., Mojaddar Langroodi, A., Shakouri, R., and Khorshidi, S. 2019. Physicochemical, microbiological, and sensory characteristics of starter yogurt enhanced with *Anethum graveolens* essential oil. *Journal of food safety*. 39(5): e12683..
- [18] Bahrami, B. Alizadeh, M., and Hassanzadeh, H. 2017. Kinetic Analysis of Antioxidant Changes in Domestic Cheese with Haven Extract Made in Clay Jugs during the Proteolysis Progress. *Iranian Journal of Nutrition Sciences & Food Technology*. 12(2): 87-95. [in Persian]
- [19] Hasanzadeh, H. Alizadeh, M., and Rezazad Bari, M. 2017. Production and assessment of physicochemical characteristics and encapsulation efficiency of garlic essential oil nanoemulsions. *Journal of Food Research*. 27(4): 159-170. [in Persian]
- [20] Mahmoudi, R., Zare, P., Hassanzadeh, P., and Nosratpour, S. 2014. Effect of T eucrium polium Essential Oil on the Physicochemical and Sensory Properties of Starter Yoghurt. *Journal of Food Processing and Preservation*, 38(3), 880-888.
- [21] Arslan, D., Ünver, A., and Özcan, M. 2005. Essential oil flavored yoghurt: physicochemical, microbiological and sensory, 36th International Symposium on Essential Oils, 4-7 September, Budapest, Hungary, Properties.p88.
- [22] Keshavarzi, M., Sharifan, A. and Yasini Ardakani, S. A. 2021. Effect of the ethanolic extract and essential oil of *Ferulago angulata* (Schlecht.) Boiss. on protein, physicochemical, sensory, and microbial characteristics of starter yogurt during storage time. *Food Science & Nutrition*. 9(1): 197-208.
- [23] Ghalem, B. R., and Zouaoui, B. 2013. Microbiological, physico-chemical and sensory quality aspects of yoghurt enriched with *Rosmarinus officinalis* oil. *African Journal of Biotechnology*, 12(2): 192-198.
- [24] Ghelich, S., Ariaii, P., and Ahmadi, M. 2022. Evaluation of Functional Properties of Wheat Germ Protein Hydrolysates and Its

- Effect on Physicochemical Properties of Frozen Yogurt. *International Journal of Peptide Research and Therapeutics*. 28(2): 1-12.
- [25] Afonso, I. M., and Maia, J. M. 1999. Rheological monitoring of structure evolution and development in stirred yogurt, *Journal of Food engineering*. 12: 183– 190.
- [26] Pang, Z., Deeth, H., Prakash, S., and Bansal, N. 2016. Development of rheological and sensory properties of combinations of milk proteins and gelling polysaccharides as potential gelatin replacements in the manufacture of stirred acid milk gels and yogurt. *Journal of Food Engineering*, 169, 27-37.
- [27] Pol, I.E., Mastwijk, H.C., Slump, R.A., Popa, M.E., and Smid, E.J. 2001. Influence of food matrix on inactivation of *Bacillus cereus* by combinations of nisin, pulsed electric field treatment and carvacrol, *Journal of Food Protection*. 64(7): 1012– 1018.
- [28] Mahmmodi, P., Khoshkhoo, Z., Basti, A. A., Shotorbani, P. M., and Khanjari, A. 2021. Effect of *Bunium persicum* essential oil, NaCl, Bile Salts, and their combinations on the viability of *Lactobacillus acidophilus* in starter yogurt. *Quality Assurance and Safety of Crops & Foods*. 13(1): 37-48.
- [29] Kamel, D. G., Hammam, A. R., Alsaleem, K. A., and Osman, D. M. 2021. Addition of inulin to starter yogurt: Viability of starter bacteria (*Bifidobacterium bifidum*) and sensory characteristics. *Food Science & Nutrition*. 9(3): 1743-1749.
- [30] Tamime, A. Y., and Robinson, R.K. 1999. *Yogurt: science and technology* (2nd ed). Boca Raton, FL: CRC Press.
- [31] Bayoumi, S. 1992. Bacteriostatic effect of some spices and their utilization in the manufacture of yoghurt, *Chem. Mikrobiol. Technol. Lebensm*. 14(2): 21-26.
- [32] Simsek, B., Sagdic, O., and Ozcelik, S. 2007. Survival of *Escherichia coli* O157:H7 during the storage of Ayran produced with different spices, *Journal of Food Engineering*. 78(2):676-680.
- [33] Karajhian R. 2005 The effect of essential oils and extracts *Ziziphora clinopodioides* on pathogenic bacteria and increase the possibility of shelf life of yogurt by adding them. Ferdowsi university of mashhad, M. C. Faculty of Agriculture; [in Persian].
- [34] Yangilar, F., and Yildiz, P. O. 2018. Effects of using combined essential oils on quality parameters of bio-yogurt. *Journal of Food Processing and Preservation*. 42(1): e13332.
- [35] Demirci, T., Aktaş, K., Sözeri, D., Öztürk, H. İ., and Akın, N. 2017. Rice bran improve starter viability in yoghurt and provide added antioxidative benefits. *Journal of Functional Foods*. 36: 396-403.
- [36] Sarabi M, Niazmand R, Abedinia A R. 2008. Thyme essential oil effect on the activity of *Lactobacillus acidophilus*, a starter yoghurt starter bacteria; Eighteenth National Congress of Food Science and Technology Iran, Mashhad . [in Persian]



ارزیابی زنده‌مانی باکتری‌های آغازگر و خصوصیات فیزیکوشیمیایی در ماست فراسودمند غنی شده با

پودر جوانه گندم و مخلوط اسانس‌های فرآوری شده گیاهی

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## چکیده

## اطلاعات مقاله

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با توجه به تقاضای روز افزون جوامع پیشرفته برای مصرف غذاهای فراسودمند بویژه در محصولات لبنی استفاده از مواد بر پایه طبیعی مثل اسانس‌های گیاهی در زمینه غنی سازی محصولات غذایی با ترکیبات ضد میکروبی و آنتی اکسیدانی آن ها و همچنین به کارگیری پودر جوانه گندم به عنوان یک منبع غنی فیبر های رژیمی و ویتامین ها و مواد معدنی و اثرات آن‌ها بر روی زنده مانگی باکتری‌های آغازگر (به دلیل نقش بارز آن ها در بهبود سلامتی انسان) امری ضروری به نظر می رسد. بررسی اثرات استفاده از اسانس‌های فرآوری شده و پودر جوانه گندم به صورت همزمان در ماست در این تحقیق مد نظر قرار گرفته است. ویژگی‌های حسی، فیزیکوشیمیایی و شمارش باکتری‌های آغازگر ماست قالبی شامل استرپتوکوکوس ترموفیلوس و لاکتو باسیلوس بولگاریکوس در طی مدت نگهداری برای مطالعه اثر همزمان اسانس های گیاهی و پودر جوانه گندم مورد آزمون قرار گرفتند. اسانس‌های به کار رفته شامل ترخون و آویشن با نسبت های مختلف و در سه سطح (۴۰۰، ۱۲۰۰، ۲۰۰۰ میکرولیتر در لیتر ماست) بودند. آزمون‌های فیزیکوشیمیایی ماست مثل اسیدیته و ظرفیت نگهداری آب، آزمون حسی به روش هدونیک، آزمون رئولوژی (ویسکوزیته ظاهری) و آزمون‌های شمارش میکروبی برای بررسی زنده مانگی باکتری های آغازگر خود ماست در طی مدت نگهداری ۱۴ روزه در این تحقیق انجام گرفتند. بر اساس نتایج به دست آمده از آنالیز واریانس نمونه های مورد آزمون، غلظت اسانس به طور معنی داری ( $p < 0.05$ )، اسیدیته محصول، ویژگی های حسی و زنده مانگی باکتری های آغازگر را کاهش داده است اما بر روی ویسکوزیته و ظرفیت نگهداری آب ماست اثر معنی داری نداشته است. آویشن و پودر جوانه گندم به ویژه در غلظت‌های بالا، تاثیر منفی و نامطلوب از خود نشان دادند. آویشن اثر ضد میکروبی بالاتری را روی باکتری‌های آغازگر ماست داشتند در حالی که ترخون فعالیت ضد میکروبی کمتری را از خود نشان داد. امتیاز حسی نمونه های حاوی ترخون در حین نگهداری اندکی افزایش داشته است لذا در مجموع ترخون مستعد به کارگیری در تولید ماست می‌باشد. پودر جوانه گندم ابتدا اثر افزایشی (تا حدود ۳ درصد) و سپس کاهش را بر روی اسیدیته محصول در طی زمان نگهداری ماست داشته است. همچنین، افزودن پودر جوانه گندم باعث افزایش ویسکوزیته و ظرفیت نگهداری آب در محصول شده اما در مقادیر بالا امتیاز حسی را کاهش می دهد و بنابراین مصرف آن در ماست باید به صورت کنترل شده باشد. به طور کلی پودر جوانه گندم بر روی زنده مانگی هر دو باکتری آغازگر شمارش شده در این تحقیق اثر مثبتی داشته که می‌تواند به دلیل تامین مواد مغذی برای رشد بیشتر آن‌ها باشد.

## کلمات کلیدی:

زنده مانگی، باکتری‌های آغازگر، ماست، اسانس‌های گیاهی، پودر جوانه گندم، غذای فراسودمند

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