

## The Effect of Wild Leek (*Allium Ampeloprasum*) on Growth and Survival of *Lactobacillus Acidophilus* and Sensory Properties in Iranian White Cheese

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### Abstract

The survival ability of probiotic bacteria in food products is one of the most important challenges ahead. The wild leek (*Allium ampeloprasum*) herb of the Allium family contains various prebiotic compounds that can stimulate the growth of probiotic bacteria. In this research, sensory properties of cheese based on *Allium ampeloprasum* as a medicinal plant and flavoring was evaluated. For this purpose, after chemical tests on raw milk, different cheese treatments were prepared to determine the effect of fresh and dry concentrations of 1 and 2% of plants, as well as non-herb control containing probiotic bacteria *Lactobacillus acidophilus* (PTCC 1643) on the growth of *Lactobacillus acidophilus* bacteria. The results showed that probiotic bacteria *Lactobacillus acidophilus* had decreasing trend in all treatments and control during storage, but in the treatments containing *Allium ampeloprasum*, this trend was less. At the end of the 45<sup>th</sup> day, the lowest bacterial count (Log CFU/g) was observed in the treatment without plant (6.69) and the highest in the sample containing one percent dry plant (8.12). The results of pH assessment also showed that in all samples, the process of pH reduction observed naturally with time of the cheese ripening. However, in treatments containing plant, there was significant difference between 30 and 45 days with non- plant control ( $P \leq 0.05$ ). At the end of the 45<sup>th</sup> day, the lowest bacterial count (Log CFU/g) was observed in the treatment without plant (6.69) and the highest in the sample containing one percent dry plant (8.12). In sensory evaluation, samples containing 1% dry plant and probiotic bacteria had the highest score among different treatments and specified the addition of *Allium ampeloprasum* as a plant additive could increase the sensory properties of the product and could be successfully used to produce symbiotic cheese.

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### Keywords

*Lactobacillus acidophilus*

Prebiotic

White Cheese

Wild Leek

### Introduction

Probiotics are living microorganisms (bacteria or yeasts) that induce their

health effects in the host's body if fed through food and at the right number (at least  $10^6$ - $10^7$  living microorganisms per

g) (Mortazavian *et al.*, 2006, Pourahmad & Shaghaghi, 2016). Among the effects of probiotics, one can cite the anti-mutagenic and anticancer properties, immunosuppression and regulation, antimicrobial properties, reduction in serum cholesterol (Rosendale *et al.*, 2008; Saarela *et al.*, 2000), improved lactose intolerance and increased nutritional value (Hui, 1993).

Among the microorganisms, some *Lactobacillus acidophilus* species, with probiotic properties, are the most common bacterial species used in the production of probiotic products (Isolauri, 2004). Lactobacilli are fermentative bacteria, most of whose products are lactic acid. These bacteria have an inhibitory effect on the growth of undesirable bacteria. Many studies have been conducted on dairy products with this bacterium to evaluate its shelf life. For instance, the studies by Yilmaztekin *et al.* (2004) on white cheese in brine, Buriti *et al.* (2007) in fresh Minas cheese, Mazinani *et al.* (2015) in ultrafiltrated cheese and Dehnavi *et al.* (2013) (Yilmaztekin *et al.*, 2004; Buriti *et al.*, 2007; Mazinani *et al.*, 2015; Dehnavi *et al.*, 2013).

There are indigestible compounds such as inulin, fructans, oligosaccharides, and so on that stimulate selectively the growth or activity of one or more of the microorganisms in the intestines, which are known as such probiotic (Ozcan *et al.*, 2016). Many studies have been conducted for using plant probiotics in food products. In a study by Ghaemi *et al.* (2010), synbiotic ultrafiltrated white cheese with *Lactobacillus acidophilus* and inulin has been examined (Ghaemi *et al.*, 2010). Masihinezhad *et al.* (2014) also studied the effects of prangos on *Lactobacillus casei* in yogurt (Masihinezhad *et al.*, 2014). *Allium ampeloprasum* L.spp Iranicum is a plant known as Yaglica in Azerbaijan and Kurayeh and Konival in Kurdistan, and used to taste traditional cheeses in some

areas. Medicinal herbs and their derivatives have long been used in the treatment of diseases and their side effects. There are many proofs for the beneficial effects of *Allium ampeloprasum* in treating some diseases. It has been shown that its active ingredients have a protective effect against injuries induced by harmful agents, serum cholesterol lowering agents, establishing a basic condition for some body functions and vasodilator (Nguansangiam *et al.*, 2003; Roghani & Aghaie, 2007). This plant has a large amount of cysteine sulfoxides, which has anti-diabetic and antioxidant properties (Kumari Kumari & Augusti, 2002). On the other hand, considering some active ingredients, it is similar to garlic, which can be beneficial for serum glucose and lipids (Fritsch & Keugen, 2006). However, the significant point about this plant, from allium family, is that like onions, it is considered probiotic compounds by having fructan. In one study, whole plant parts contain approximately 9% fructan compounds, and the degree of polymerization of fructan is about 8 (Muir *et al.*, 2007). However, the amount of fructan of this plant changes drastically, depending on the plant body and the harvest season, so that in another study, it was reported to vary from 2.5 to 83.5% per gram (Bernaert, 2013). Therefore, using it for producing probiotic foods can be interesting.

Recently, designing and producing herbal based probiotic preparations have received great attention given their nutritional value (protein, fiber, vitamin and minerals), and diversification in production and consumption. It seems that the industrial production of these products with proper qualitative properties would be a great section of the studies to be conducted in the future (Ozcan *et al.*, 2016). However, one of the most important challenges in the production and processing of probiotic products is the low survival probiotic

bacteria due to the sensitivity to specific conditions in the food product and the intestinal conditions for these organisms. One of the solutions to eliminate these factors is using probiotic compounds along with probiotic bacteria and creating a condition called symbiotic that increases the growth and survival of these bacteria (Zimer & Gibson, 1998). As *Allium ampeloprasum* is traditionally used in some regions of Azerbaijan for flavoring cheese, no studies have been done on the use of it in Iranian white cheese, and it is not studied on probiotic bacteria, the purpose of the study was to examine the effect of *Allium ampeloprasum* on the survival of *Lactobacillus acidophilus* and its effect on the sensory properties of Iranian white cheese. This is done to gain suitable efficacy as well as the desired effect on sensory properties; it can be used to produce cheese and new symbiotic products.

## Material and methods

### Chemical tests of raw milk used to produce cheese

After preparing the milk needed for cheese production, the chemical tests were done. In doing so, the fat value was considered according to the national standard of Iran 760 (Iranian National Standardization Organization [ISIRI], 2002a), protein according to standard 639 (Iranian National Standardization Organization [ISIRI], 2002b), pH and acidity measurements according to standard 2852 (Iranian National Standardization Organization [ISIRI], 2001).

### Preparation of probiotic bacteria and cheese starter

Probiotic bacteria (*Lactobacillus acidophilus* PTCC 1643) was bought from the Iranian Research Organization for Science and Technology as lyophilized ampoules. Lyophilized primer culture and DVS-R-704, including *Lactococcus lactis*, under the species of *Cremoris* and

*Lactococcus lactis* were purchased from Chr. Hansen Company. To prepare the probiotic bacteria, the contents of the lyophilized ampoules containing *Lactobacillus acidophilus* were transferred to a test tube containing 10 mL of MRS medium (Merck, Germany) and incubated at 37 °C for 24 h under anaerobic incubation conditions. Then, the bacterial cultures prepared were transferred into Erlenmeyer Flasks containing 95 mL of MRS liquid medium. This was repeated 2 to 3 times to reach a bacterial count of  $10^8$ - $10^9$  CFU/mL. Microbial cells were then obtained by centrifugation at 1500 rpm for 15 min. The obtained bacteria were washed again with 0.1% sterilized peptone water and used for inoculation in milk at cheese-making stage (Krasaeckoop *et al.*, 2004).

### Preparing *Allium ampeloprasum* L.spp *Iranicum*

The fresh plant was collected from mountains around Oshnaviyeh. After washing, some part of it was crushed and stored in the freezer, and the other part was dried in shade at 25 °C. Dried leaves were then stored in a freezer until the test was done. The fresh and dried plants were pasteurized in the oven and Bain-Marie at 65 °C for 30 min, respectively, and added to cheese by 1 and 2% (Ocak *et al.*, 2015).

### Preparation of white cheese stored in brine

To prepare the cheese, after milk pasteurization, the temperature was adjusted to 35 °C and calcium chloride was added at a rate of 0.01% (v/w). For probiotics containing *Lactobacillus acidophilus*, 1 mL of the prepared suspension containing  $10^9$ - $10^8$  CFU/mL bacteria described above was added to the milk and mixed up. After that, lyophilized primer culture was inoculated according to the recommended dose of the manufacturer for 1 L of milk at 0.005% (v/w). After that, the milk was

kept at 35 °C to keep it at a pH of 6.4. Then, various concentrations of *Allium ampeloprasum* were added to the milk and mixed. Then rennet was added to the milk as much as 0.001% (v/w). The clot was kept after 15 min for the removal of whey, and the clot was then compressed into a sterile bag of carobs and a press machine, and pressed gradually for two hours at a load of weights. After the cheese had reached the intended consistency, 100 g cheese pieces were first placed in 20% sterilized brine for 8 h at ambient temperature (20 to 24 °C). After that, they were transferred to sterilized 8% brine and stored for 15 days at a temperature of 12-14 °C. Ultimately, after the initial consistency period, the cheese pieces were stored in a refrigerator for 8% at 4 °C for 45 days (Iranian National Standardization Organization, No. 5772, 2011).

#### **Preparation of cheese samples for counting probiotic bacteria**

A specific culture medium was used for counting lactobacilli in cheeses produced from MRS-BC agar (Merck, Germany) containing 2% green bromoceresol (Sigma Company) and clindamycin (Sigma Company). In doing so, 5 mg of clindamycin was mixed in 100 mL of water and 2 mL per liter of MRS was added using sterile syringe filter. After dilution and surface cultivation, incubation was done at 37 °C for 48 to 72 h in an anaerobic way and counted from day zero to 45 every 5 days (Phillips *et al.*, 2006).

#### **Sensory evaluation**

Although the main value of probiotic products is their biotic features, their

sensory properties are important as well, since the consumer has the main role in selection. Taste and odor are of the effective features in cheese market. Therefore, sensory testing was performed simultaneous to bacterial counting. Odor, taste, color and texture (consistency) of the samples were determined by a group of 10 trained sensory evaluators using the consumer inclination test and the 5-point Hedonic method. From each treatment, 5 identical samples were prepared and along with the form submitted to the referees. In doing so, 5 was given to excellent and 1 to very poor quality. The overall consumer evaluation was presented as a numerical value (IDF, 1997).

#### **Statistical Analysis**

The experiments were performed in 3 iterations. Data analysis was done in SPSS (version, 19) and mean comparison was done using Duncan's test with p-value less than 0.05 considered significant. The corresponding charts were drawn by Microsoft Excel software (version, 2016).

#### **Result and discussion**

##### **Physicochemical analysis of raw milk**

The results of the physicochemical properties of milk used to produce cheese are shown in Table (1). As milk compounds and its properties affect the properties of the produced cheese and may even affect the durability of the probiotic bacterium, analyses were done on the raw milk used for producing cheese.

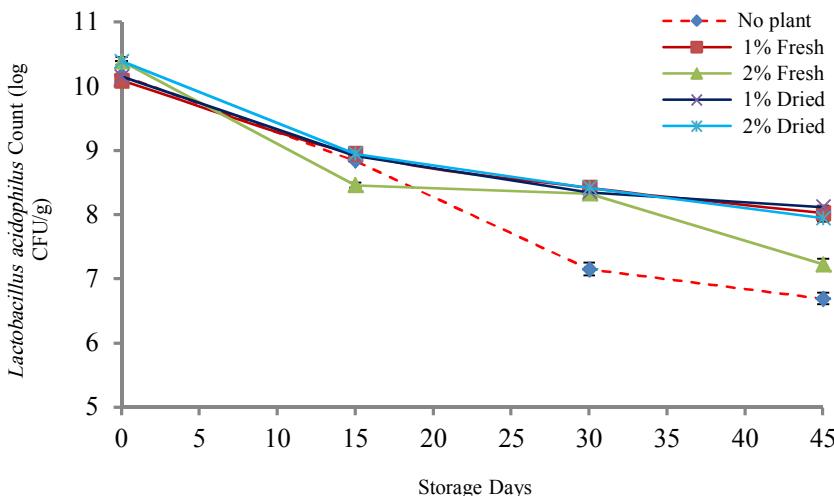
**Table 1.** Physicochemical properties of raw milk used for cheese production

Test	Fat (%)	Dry matter (%)	pH (1:2)	Acidity (°D)	Protein (%)
Results	3.7±0.6	11.9±0.7	6.2±0.02	17.1±0.8	3.01±0.6

### Counting the probiotic bacteria

The results of counting probiotic bacteria in Iranian white cheese are shown in Figure (1). As indicated in the

chart, the probiotic bacteria *Lactobacillus acidophilus* had decreasing trend in all treatments and during storage.



**Figure 1.** Changes in counting *Lactobacillus acidophilus* in different days at 4 °C

However, in the treatments containing *Allium ampeloprasum*, this trend was less so that on the 30<sup>th</sup> control day, the bacterial count was lower 7.15 CFU/g with a significant difference with other treatments ( $P \leq 0.05$ ). On day 45, counts of the control and 2% fresh plants treatment reached below 8 CFU/g, whereas in other treatments, probiotic bacteria were higher with a significant difference with these 2 treatments ( $P \leq 0.05$ ). Additionally, the results of day 45 showed no significant differences between the treatments containing dry matter 1 and 2% in terms of probiotic bacteria ( $P \geq 0.05$ ). The small differences between the effects of dry and fresh plants should be found in the mechanism for the release of fructan compounds and the effective ingredients in the growth of probiotic bacteria were distinguished. Many studies have been conducted on the use of different compounds as probiotic in dairy products. Araujo et al. (2009) used inulin to enhance the livestock recovery and tissue alteration of cottage cheese containing *Lactobacillus delbrueckii* and increase the viability of the probiotic bacteria mentioned (Araujo et al., 2009). In another study, Ghaemi et

al. (2010) studied the symbiotic white ultrafiltrated cheese containing *Lactobacillus acidophilus* and inulin, and the results showed that the addition of inulin increased the bacterial survival (Ghaemi et al., 2010). The probiotic effects of garlic fructan in a laboratory environment were examined by Zhang et al. (2013). Their results showed that garlic fructans increased the count of bacterial bacteria in the intestine (Zhang et al., 2013). China et al. (2012) examined the antimicrobial activity of *Sesbania grandiflora* extract on some pathogen bacteria and evaluated the stimulatory effect of growth on the probiotic organism *Lactobacillus acidophilus*. Their results showed that *Sesbania grandiflora* polyphenol extract has inhibitory effect on pathogens and stimulates the growth of probiotic bacteria (China et al., 2012). Masihinezhad et al. (2014) studied the effects of prangos on *Lactobacillus casei* in yogurt, and the results showed that the maximum bacterial survival was at 20% concentration of the plant (Masihinezhad et al., 2014). Zomorodi et al. (2015) studied the survival of bacteria *Lactobacillus acidophilus* in symbiotic

yogurt using apple and wheat fiber, and the results showed that the use of fiber of wheat and apple can cause an increase in the number of probiotic bacteria (Zomorodi *et al.*, 2010). Hap & Gutierrez (2012) showed that the extracts of Kiwi, Strawberries, Blackberry and *Acca sellowiana* significantly increased the probiotic bacteria except *Bifidobacterium* (Hap & Gutierrez, 2012). Marhamatizadeh *et al.* (2009) examined the survival of *Lactobacillus acidophilus* and *Bifidobacterium* and their count in probiotic milk and yogurt. The results showed that during the 20 days of probiotic preservation, the bacteria remained alive and the growth rate and survival rate of bacteria in the sample containing *Lactobacillus acidophilus* were higher than that of *Bifidobacterium bifidum* (Marhamatizadeh *et al.*, 2009). Kasimoglu *et al.* (2004) showed that the lactoobacillus survival rate declined during the first 15 days of arrival, the researchers reported the decrease in moisture content, increased salt content and reduced storage temperature, which are in line with the results of this study (Kasimoglu *et al.*, 2004). Another study by Sutherland *et al.* (2009) showed that from among about 100 extracts studied, garlic and pepper extracts had an increasing effect on the growth of the probiotic bacteria *Lactobacillus Reuteri* (Sutherland *et al.*, 2009). As the results showed, the bacterial survival was greater than  $10^7$  CFU/g in samples of different concentrations of *Allium ampeloprasum*. If this cheese is consumed at a dose of 30 g per day, the probiotic amount will be between  $10^9$  to  $10^{10}$  CFU/g, higher than the recommended amount for therapeutic effects (Boylston *et al.*, 2004; FAO/WHO, 2002). Many researcher have reported similar results in various types of cheese (Bergamini *et al.*, 2005; Buriti *et al.*, 2007). As the results of most studies indicate, using a specific

concentration of the plant in most cases increased the survival and survival of probiotic bacteria that is in agreement with the results of the present study. The mechanism of action of *Allium ampeloprasum* extract is similar to that of garlic and onion from the allium family, and the effect of increasing the extract of *Allium ampeloprasum* on the growth of *Lactobacillus* bacteria is due to the presence of some active compounds such as phenolic compounds and fructans in it. Similar results have been reported by other researchers on other plant extracts (Molan *et al.*, 2009; Zomorodi *et al.* 2015; Rosendale *et al.*, 2008). Concerning the mechanism of action of active compounds in the growth of the growth of probiotic bacteria, there is no complete understanding, but active compounds appear to act as an energy source and also as antioxidants.

#### Measuring the pH of probiotic-containing cheeses

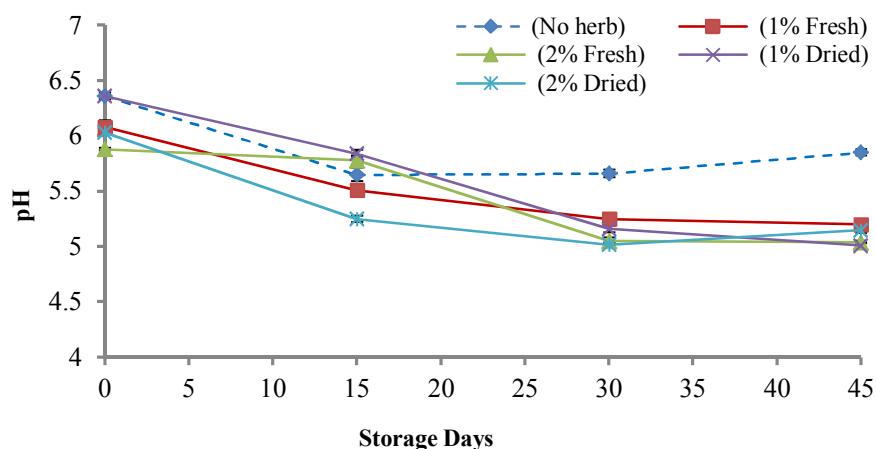
The pH of the probiotic bacteria is one of the most important factors in its survival. The effects of various concentrations of *Allium ampeloprasum* and control samples on pH of the cheese produced with acidophilus bacteria are shown in Figure (2). As the results show, in all samples, pH decreases naturally with cheese ripening time. However, there was a significant difference in planting treatments between days 30 and 45 ( $P \leq 0.05$ ). There was a significant difference in different pH treatments between different treatments during days of storage, so that on day 15 and 30, the lowest pH of the cheese sample containing dry matter was 2%, and on day 45, the sample had 1%. Furthermore, on day 45, the highest pH was related to the treatment without plant with a significant difference with other treatments. The pH reduction while storage may be due to the acidic

activity of probiotic strains and commercial starter (Ong *et al.*, 2007). The pH decreased due to the production of organic acids by carbohydrate fermentation bacteria and decreased more in the treatments with the plant samples, which can be due to the effect of increasing the growth of probiotic compounds of *Allium ampeloprasum* on the bacteria producing acid. Reductions in pH have also been reported by other researchers (Buriti *et al.*, 2007; Akin *et al.*, 2003). Thus, following increased acidity production and reduction in pH, probiotic bacteria are affected as well.

In a study to examine the effects of commercial probiotics Raftiline and Raftilose on pH changes in the skim milk and the peptone-containing

*Lactobacillus acidophilus* model, Olson & Aryana (2012) found that adding probiotics reduced pH compared to the control sample (Olson & Aryana, 2012), in line with the results of this study. Similar results were also obtained by Zhang *et al.* (2013) in examining fructan extracted from garlic on the pH changes of the Bifidobacterium in the culture medium (Zhang *et al.*, 2013).

The results of the present study showed the effect of increasing the growth of the probiotic composition of *Allium ampeloprasum*, but given the important effect of fructan in the plant as the key determining factor, it is necessary that more studies be done to determine the amount of fructan in the whole cheeses produced.



**Figure 2.** Changes in pH in cheese containing *Lactobacillus acidophilus* in different treatments during storage days at 4 °C

### Sensory evaluations

Sensory properties of food products are of the most significant effects on the part of consumers. The results of the flavor evaluation in cheese samples containing *Lactobacillus acidophilus* during the storage days at 4 °C are presented in Table (2). Overall, adding *Allium ampeloprasum* to cheese has changed the taste of cheese samples, so that on the first day of the experiment, the flavored scores decreased and a significant difference was observed with the control ( $P \leq 0.05$ ). On the 15<sup>th</sup> day, the planting treatments increased and on the 30<sup>th</sup> and 45<sup>th</sup> days, this trend

continued. Therefore, the highest score in these days was for treatments containing 2% dry matter, 1% dry and fresh plants and 1% dry plant. Thus, adding *Allium ampeloprasum* significantly improved the taste of cheeses from the viewpoint of evaluators. The results of tissue evaluation in cheese samples containing *Lactobacillus acidophilus* during the storage days are shown in Table (3) as well. The results show that there are no significant differences between treatments at zero and 15 days ( $P \geq 0.05$ ). Nevertheless, from the 30<sup>th</sup> and 45<sup>th</sup> days of cheese ripening, the score for

control sample tissue decreased, whereas the proportion of specimens containing *Allium ampeloprasum* increased. Overall, the addition of *Allium ampeloprasum* as an additive to cheese causes tissue alteration, and this is evident in the results of the sensorytest as well.

The most significant factors affecting the tissue of the cheese are the total

solids content and the gradual decomposition of proteins (Çelik *et al.*, 2008). In the present study, it seems that the addition of the plant has changed the total amount of solids in the cheese and thus affects the tissue results. However, it is recommended that tissue analysis methods be used in later studies by devices.

**Table 2.** Results (mean  $\pm$  standard deviation) of flavor evaluation in cheese samples containing *Lactobacillus acidophilus* during storage days at 4 °C

Treatment	Day			
	0	15	30	45
No plant	3.6 $\pm$ 0.54 <sup>a*</sup>	3.4 $\pm$ 0.54 <sup>a</sup>	3.2 $\pm$ 0.44 <sup>a</sup>	3.0 $\pm$ 0.00 <sup>a</sup>
Fresh plant 1%	3.4 $\pm$ 0.54 <sup>ab</sup>	3.4 $\pm$ 0.54 <sup>a</sup>	5.0 $\pm$ 0.00 <sup>c</sup>	4.6 $\pm$ 0.54 <sup>cd</sup>
Fresh plant 2%	3.2 $\pm$ 0.44 <sup>ab</sup>	4.0 $\pm$ 0.00 <sup>a</sup>	4.4 $\pm$ 0.54 <sup>bc</sup>	3.6 $\pm$ 0.54 <sup>b</sup>
Dry plant 1%	3.0 $\pm$ 0.00 <sup>ab</sup>	3.4 $\pm$ 0.54 <sup>a</sup>	5.0 $\pm$ 0.00 <sup>c</sup>	5.0 $\pm$ 0.00 <sup>d</sup>
Dry plant 2%	2.8 $\pm$ 0.44 <sup>ab</sup>	3.8 $\pm$ 0.44 <sup>a</sup>	3.8 $\pm$ 0.44 <sup>ab</sup>	4.2 $\pm$ 0.44 <sup>c</sup>

\* Non-similar letters in each column show significant differences ( $P\leq 0.05$ ).

**Table 3.** Results (mean  $\pm$  standard deviation) of tissue evaluation in cheese samples containing *Lactobacillus acidophilus* during storage days at 4 °C

Treatment	Day			
	0	15	30	45
No plant	5.0 $\pm$ 0.54 <sup>a*</sup>	4.4 $\pm$ 0.54 <sup>a</sup>	2.4 $\pm$ 0.54 <sup>a</sup>	3.0 $\pm$ 0.00 <sup>a</sup>
Fresh plant 1%	5.0 $\pm$ 0.54 <sup>a</sup>	5.0 $\pm$ 0.00 <sup>b</sup>	5.0 $\pm$ 0.00 <sup>b</sup>	4.8 $\pm$ 0.44 <sup>b</sup>
Fresh plant 2%	5.0 $\pm$ 0.44 <sup>a</sup>	4.6 $\pm$ 0.54 <sup>b</sup>	4.6 $\pm$ 0.54 <sup>b</sup>	4.6 $\pm$ 0.54 <sup>b</sup>
Dry plant 1%	5.0 $\pm$ 0.00 <sup>a</sup>	5.0 $\pm$ 0.00 <sup>b</sup>	4.8 $\pm$ 0.44 <sup>b</sup>	5.0 $\pm$ 0.44 <sup>b</sup>
Dry plant 2%	5.0 $\pm$ 0.44 <sup>a</sup>	4.4 $\pm$ 0.54 <sup>a</sup>	4.6 $\pm$ 0.44 <sup>b</sup>	4.6 $\pm$ 0.54 <sup>b</sup>

\* Non-similar letters in each column show significant differences ( $P\leq 0.05$ ).

The results of odor evaluation in cheese samples containing *Lactobacillus acidophilus* during the storage days at 4 °C are shown in Table (4). In the days 0 and 15 of cheese ripening the lowest scores related to odor was for the samples of fresh plant of 2% sample, whereas 1% sample had the highest score and the other samples did not differ significantly ( $P\leq 0.05$ ). On days 30 and 45, the highest score was for the sample containing 1% dry matter. The lowest score was for the control sample without plant, with the other samples not differing significantly ( $P\leq 0.05$ ). Finally, for a better comparison, the mean of total results of sensory tests in cheese containing *Lactobacillus acidophilus* in different treatments during storage days at 4 °C is shown in

Figure (3). The results showed no significant differences between treatments on days zero and 15, but on days 30 and 45, the treatments containing *Allium ampeloprasum* had significantly higher sensory scores than the control group. The highest score was obtained by fresh 1% treatment on day 45 with a significant difference with non-plant treatment, whereas the lowest was for control treatment ( $P\leq 0.05$ ). Overall, due to different compounds in the case of addition to food products as additive, aromatic herbs such as *Allium ampeloprasum* cause organoleptic changes in the product, which in some cases is a satisfactory change. Few studies have been done regarding the effect of plant probiotics on the sensory properties of

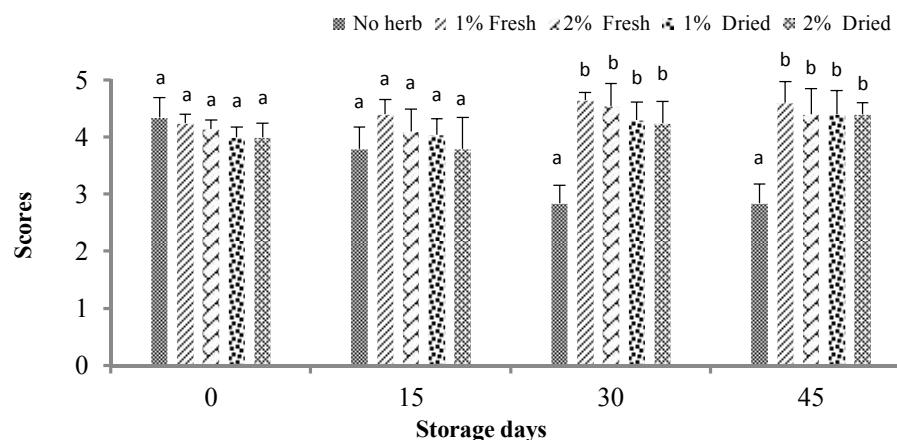
synbiotic cheeses. In one case, Mazinani *et al.* (2015) studied the sensory features of synbiotic ultrafiltrated white cheese containing *Origanum vulgare* powder and *Spirulina plasticum* algae. The results showed that the highest acceptance among algae samples was for 0.3% algae samples and 1% *Origanum vulgare* (Mazinani *et al.*, 2015). On the contrary, a review by Araujo *et al.* (2009) showed that adding inulin to cottage synbiotic cheese does not cause any organoleptic alteration (Araujo *et al.*, 2009). Azambuja *et al.* (2013) examined the sensory properties of fresh cheese containing *Bifidobacterium* as probiotic and polydextrose probiotic. The results showed that sensory evaluators found no significant differences between treatments (Azambuja *et al.*, 2013). In another study on cheese with *Allium ursinum*, the results showed that the addition of

garlic to other than the change in cheese texture did not have a significant effect on other sensory properties (Tarakci *et al.*, 2011). Furthermore, El-Khalek *et al.* (2016) on probiotics with probiotics containing zinger as a prebiotic showed that ginger containing treatments had a higher sensory score than the control group (El-Khalek *et al.*, 2016). Overall, as the results of similar studies and the present study show, the additions of plant compounds to dairy products not only produce a variety of products, but also in many cases, in addition to the probiotic properties added to the product, increase the sensory scoring. In the present study, although the positive effects of *Allium ampeloprasum* on the sensory properties of Iranian white cheese were determined according to the participants in the sensory test, proving this requires more tests and conducting the exact device tests.

**Table 4.** Results (mean  $\pm$  standard deviation) of odor evaluation in cheese samples containing *Lactobacillus acidophilus* during storage days at 4 °C

Treatment	Day			
	0	15	30	45
No plant	3.4 $\pm$ 0.54 <sup>b*</sup>	3.0 $\pm$ 0.00 <sup>b</sup>	2.8 $\pm$ 0.44 <sup>a</sup>	2.2 $\pm$ 0.44 <sup>a</sup>
Fresh plant 1%	4.0 $\pm$ 0.00 <sup>c</sup>	4.0 $\pm$ 0.00 <sup>c</sup>	4.6 $\pm$ 0.54 <sup>bc</sup>	4.6 $\pm$ 0.54 <sup>b</sup>
Fresh plant 2%	2.2 $\pm$ 0.44 <sup>a</sup>	2.4 $\pm$ 0.54 <sup>a</sup>	5.0 $\pm$ 0.00 <sup>c</sup>	4.8 $\pm$ 0.44 <sup>b</sup>
Dry plant 1%	3.0 $\pm$ 0.00 <sup>b</sup>	3.6 $\pm$ 0.54 <sup>b</sup>	5.0 $\pm$ 0.00 <sup>c</sup>	5.0 $\pm$ 0.00 <sup>b</sup>
Dry plant 2%	3.4 $\pm$ 0.54 <sup>b</sup>	3.2 $\pm$ 0.44 <sup>b</sup>	4.4 $\pm$ 0.54 <sup>b</sup>	4.6 $\pm$ 0.54 <sup>b</sup>

\* Non-similar letters in each column show significant differences ( $P \leq 0.05$ ).



**Figure 3.** Mean of the overall results of sensory tests in cheese containing *Lactobacillus acidophilus* in different treatments during storage days at 4 °C

Non-identical Latin letters per day show a significant difference ( $P \leq 0.05$ ).

## Conclusion

The results indicated that tertiary as a prebiotic in cheese, *Allium ampeloprasum* L.spp *Iranicum* can contribute to the survival of probiotic and beneficial microorganisms in unsuitable storage conditions and improve the tissue and sensory properties of the product. Thus, it is

suggested that in the production of probiotic white Iranian cheese *Allium ampeloprasum*, with prebiotic, antimicrobial and nutritional features, be used to increase the probiotic shelf life, to reduce the risks of food-related diseases, and to create the organoleptic desirable characteristics.

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## تأثیر گیاه تره کوهی بر رشد و ماندگاری باکتری پروبیوتیک لاکتوباسیلوس اسیدوفیلوس و ویژگی‌های حسی پنیر سفید ایرانی

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### چکیده

قابلیت زنده‌مانی باکتری‌های پروبیوتیک در محصولات غذایی یکی از مهم‌ترین چالش‌های پیشرو می‌باشد. گیاه تره کوهی از خانواده آلیوم دارای ترکیبات مختلف پری‌بیوتیکی بوده که می‌تواند منجر به تحیریک رشد باکتری‌های پروبیوتیک شود. در این پژوهش ویژگی حسی پنیر سفید ایرانی بر پایه تره کوهی به عنوان یک گیاه دارویی و طعم‌دهنده مورد ارزیابی قرار گرفت. بدین‌منظور پس از انجام آزمون‌های شیمیایی روی شیر خام، تیمارهای مختلف شامل پنیر حاوی غلاظت‌های ۱ و ۲ درصد گیاه تازه و خشک و نیز نمونه کنترل بدون گیاه حاوی باکتری پروبیوتیک لاکتوباسیلوس اسیدوفیلوس تهیه شدند. نتایج نشان داد که باکتری پروبیوتیک لاکتوباسیلوس اسیدوفیلوس (PTCC 1643) در تیمارهای حاوی گیاه و کنترل بدون گیاه در طول زمان نگهداری (۴۵ روز) روند کاهشی را داشته ولی در تیمارهای حاوی گیاه تره کوهی این روند کاهشی کمتر بوده است. به طوری که در پایان روز ۴۵ آم کمترین میزان باکتری (CFU/g) در نمونه بدون گیاه (۶/۶۹) و بیشترین در نمونه حاوی ۱ درصد گیاه خشک (۸/۱۲) بود. نتایج ارزیابی pH نیز نشان داد که در تمام نمونه‌ها با طی زمان رسیدن پنیر روند کاهش pH به طور طبیعی مشاهده گردید. ولی در تیمارهای حاوی گیاه در روزهای ۳۰ و ۴۵ اختلاف معنی‌داری با نمونه کنترل بدون گیاه دیده شد ( $P \leq 0.05$ ). در ارزیابی حسی نیز نمونه حاوی گیاه ۱ درصد تازه و باکتری پروبیوتیک بیشترین امتیاز را بین تیمارهای مختلف داشت (۴/۶) و مشخص نمود که افزودن تره کوهی به عنوان یک افزودنی گیاهی می‌تواند موجب بهبود خصوصیات حسی محصول شده و به طور موفقیت‌آمیزی برای تولید پنیر سین‌بیوتیکی مورد استفاده قرار گیرد.

**واژه‌های کلیدی:** پری‌بیوتیک، پنیر سفید، تره کوهی، لاکتوباسیلوس اسیدوفیلوس