

# The effect of wild rose (*Rosa canina L.*) on the microbial contamination of ice cream

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## Abstract

Ice cream is a frozen mixture prepared from milk, sweeteners, stabilizer, emulsifiers and materials generating scent and taste that due to various components in its structure, such as ice crystals and scattered air bubbles in the serum phase has various colloidal properties. Dairy products and ice cream have high nutritional values, which provide significant portion of nutritional needs, cause neutralization of free radicals, prevent cancer, and increase safety resistance in children. Despite its significant nutritional values, ice cream, due to the nutrient medium, is a good environment for the growth of microorganisms. Wild rose (*Rosa canina L.*) is one of the most important medicinal plants; its fruits contain valuable medicinal and nutritional compounds. The aim of producing ice cream with the fruit extract of wild rose is to use its nutritional and therapeutic properties and to evaluate the effect of the extract on the microbial contamination of the produced ice cream. Therefore, ice cream samples were prepared with the fruit extract of wild rose in 2 levels of 5% and 15%, and then viscosity, hardness, overrun, drawing temperature, melting rate tests and microbial search were evaluated. The results were analyzed using SPSS software and Duncan test. By adding the extract, viscosity and hardness increased significantly ( $p \leq 0/05$ ), and overrun and drawing temperature decreased significantly ( $p \leq 0/05$ ). Melting rates were not significantly different between samples ( $p \geq 0/05$ ). The numbers of microorganisms in produced ice cream from fruit extract of wild rose were significantly lower than the control sample ( $p \leq 0/05$ ). By substituting the milk with fruit extract of wild rose, the physicochemical, rheological and microbial properties of the produced ice cream improved.

**Key words:** *Ice cream, Microbial contamination, Wild rose*

## Introduction

According to the statistics of the world health organization, annually, about 1.5 billion cases of diarrhea occur in children under 5 years old, 5 million of which lead to death (WHO, 1991) and hereof, contaminated food has an important role (WHO, 1990). Milk and dairy products are one of the most important sources of food-borne infections and poisonings (Barrette, 1986). Ice cream can be a very good environment for the growth of a variety of microorganisms because of its nutrient medium and pH 6-7 and long maintenance period (Kanbakan *et al.*, 2004). Pathogen bacteria of *Staphylococcus aureus*, Salmonella, Shigella, Brucella, coliform, Campylobacter, Pseudomonas, and Psychrotroph bacteria of *Listeria monocytogenes* and *Yersinia enterocolitica* are observed in contaminated ice cream (Azandnia

*et al.*, 2011). Aromatic plants rich in phenolic and antioxidant compounds are good examples which have been used in the pharmaceutical, food, perfumery, and non-alcoholic beverages industries from the past (Yasin and Abou-Taleb, 2007; Chun *et al.*, 2005; Qari, 2008). Wild rose with the scientific name *Rosa canina L (Rosaceae)* is a shrub plant, with several years old, which grows naturally on rocks and shrubs in dry areas. The fruit is rounded or ovoid jar-shaped, and smooth, in bright red color (at full fruiting stage it is in dark red color to brown) and the seeds are inside (Omid Beigi, 2005). The geopolitical dispersions of wild rose (*Rosa canina L.*) are in Europe, Turkey, Iran, the Soviet Union, Afghanistan, Pakistan and Iraq. Wild rose (*Rosa canina L.*) is distributed in north, northwest, west, southwest, center and eastern part of Iran. (Khatamsaz, 1992). This fruit is rich in sugars, fruit acids, pectin and oil essence. The pericarp of wild rose (*Rosa canina L.*) has been counted as one of the items in the European pharmacy guidebook and Hungary. This fruit is highly cultivated in central Europe as well as Hungary, with the production of 60 to 70 tons a year in this country. The main and valuable part of this fruit is the pericarp from which various products including pharmaceutical pharmaceutical products, herbal teas, jams, marmalade, syrups, jelly, soda and etc., can be made (Szentmihlyi *et al.*, 2002). Wild rose (*Rosa canina L.*) is rich in minerals such as phosphorus, potassium, calcium, zinc and iron (Artik and Eksi, 1998). One of the valuable and rich compounds of this fruit is vitamin C that is used in different regions to prevent cold (Rouhani *et al.*, 1976; Szentmihalyi *et al.*, 2002). This study is intended to enrich ice cream with fruit extract of wild rose and to investigate the viscosity, hardness, overrun, drawing temperature, melting rate tests and microbial search.

## **Materials and methods**

One kg of wild rose fruits (*Rosa canina L.*) was bought from Sepidan city grocery and was kept at ambient temperature until the test day. Sterilized milk (3% fat) and sterilized cream (30% fat) were purchased from Mihan Company. Non-fat dry milk was bought from Mannymas Company, and salep with the brand sun rose, made in Japan, was prepared as well. Moreover, sugar, vanilla and plastic containers of 300 g were prepared from the confectionary accessories shop.

### **Preparation of the powder of wild rose fruits (*Rosa canina L.*)**

The fruits were milled after removing the seeds and internal lints.

### **Extraction of extracts using microwave**

Microwave mark anton par model SVM 3000 with voltage – frequency 230 - 240 W, 50/60 Hz, input power (microwave)1700 W, output power 1400 W, oven capacity 66 L, outer dimensions (L × W× H) 60 × 72 × 74 cm, net weight 74 kg was used.

2.5 g of the powder (of the fruit) was measured by the scales and was placed in a tubular microwave device, and then 50 ml of ethanol solvent was added to the tubular containing powder. Device parameters were set to the default; the temperature of 90 degrees of celsius for 20 minutes. Finally, extraction of the solution was filtered by a 150 mm filter paper. The supernatant material was centrifuged after collecting at 3000 rpm for 10 minutes. Then, it was passed from 0.45 millimeter filter. Finally, the extract was condensed using a rotary device and was kept at 4°C for the subsequent steps of the test (Chemat *et al.*, 2005).

### Preparation of the ice cream

Fat percentages, solid non-fat, sweeteners, flavors and stabilizers in the final product were adjusted at levels of 12, 10, 17, 0.2, and 0.3 percent, respectively. At first, the necessary materials for each of the formulas were calculated by serum point method, (Chegeni and Meshkat, 2006) and after weighing the materials the following procedures were done:

**Table1. Formulation of prepared samples**

Compounds	A (Control) (%)	B (%)	C (%)
Sugar	17	17	17
Vanilla	0.2	0.2	0.2
Salep	0.3	0.3	0.3
Dry milk	4.106	4.106	4.106
Creamy Milk	35.737	35.737	35.737
Milk Extract	42.657	37.657	27.657
	-	5	15

In this study, the production of ice cream was carried out according to the method(s) employed by Akalem *et al.* (2008). Milk, cream, and weighed extract were uniformed by mixer as well. The resulting mixture was heated to 40°C. Then, the other ingredients (a mixture of dry milk, sugar and salep) were slowly added to the mixture. The mixture was stirred for 15 minutes at a speed of 500 rpm and then pasteurized for 25 minutes at 75°C and immediately cooled down to 4°C with the aid of a mixture of water and ice. To achieve the consolidation, the mixture was placed in a refrigerator for 24 hours at 4°C. After the completion of the consolidation procedure, before freezing, vanilla was added. The ice cream mixture was frozen in the ice cream maker for

20 minutes. The ice cream was packaged and encoded in small plastic containers and transferred to the freezer  $-18^{\circ}\text{C}$  for doing tests (Akalm *et al.*, 2008).

### The performed tests

The viscosity of the ice cream mixture was tested by an Antonpar viscometer of the SVM 3000 model equipped with a thermal circulator. The mixture was evaluated at  $10^{\circ}\text{C}$ . It is to be mentioned that the viscometer has been equipped with computer software, and has controlled the operating conditions of the viscometer (Akin *et al.*, 2007).

The ice cream texture was evaluated by texture analyzer after 3 days of storage at  $-18^{\circ}\text{C}$ . This device was equipped with a cylindrical stainless steel probe with a diameter of 6 mm and a height of 35 mm. The probe of the device was entered twice into the test sample with the speed of 2 mm/s and up to 50% of the probe height. The results were recorded by the device software (Lu *et al.*, 2002). Overran measurements were carried out according to the institute of standard and industrial research of Iran No. 2450.

The ice cream drawing temperature was measured at the end of the freezing step by placing the alcohol thermometer inside the tank (Bahramparvar *et al.*, 2009).

The melting speed was done by measuring 30 grams of frozen ice cream and putting it on a sieve with a mesh 60, placed on a container of a specified weight. The dish was placed in an oven at  $24^{\circ}\text{C}$  for one hour. Every 10 minutes, the weight of the container and the melted ice cream was recorded and the melting speed was calculated (Alamprese *et al.*, 2002).

The salmonella search was carried out according to the Institute of standard and industrial research of Iran, No. 4413.

The *Escherichia coli* search was carried out according to the Institute of standard and industrial research of Iran, No. 5234.

The enumeration of microorganisms was done according to the Institute of standard and industrial research of Iran, No. 5272-1.

The enumeration of *Staphylococcus aureus* was carried out according to the Institute of standard and industrial research of Iran, No. 6806-3.

The enumeration of Enterobacteriaceae was performed according to the Institute of standard and industrial research of Iran, No.2461-2.

### Statistical analyses

All experiments were carried out in a completely randomized design with 3 replications. Appropriate statistical analyses, using SPSS software, were run to analyze the results, including analysis of variance and comparison of means by Duncan method at 5% level.

## Results

When analyzing viscosity of dynamics and kinematic, maximum amount were observed in sample C, sample B and control sample respectively. The differences between the samples were shown to be significant ( $p \leq 0/05$ ).

The highest hardness was observed in sample C (1880 g), sample B (1466 g) and control sample (1261 g) respectively. The differences between the samples were significant ( $p \leq 0/05$ ). Therefore, increasing the extract percentage has led to the higher viscosity and hardness of the ice cream.

The highest overran was observed in control sample (25.33 %), sample B (20.33 %) and sample C (13.33 %) respectively. The samples differed significantly ( $p \leq 0/05$ ).

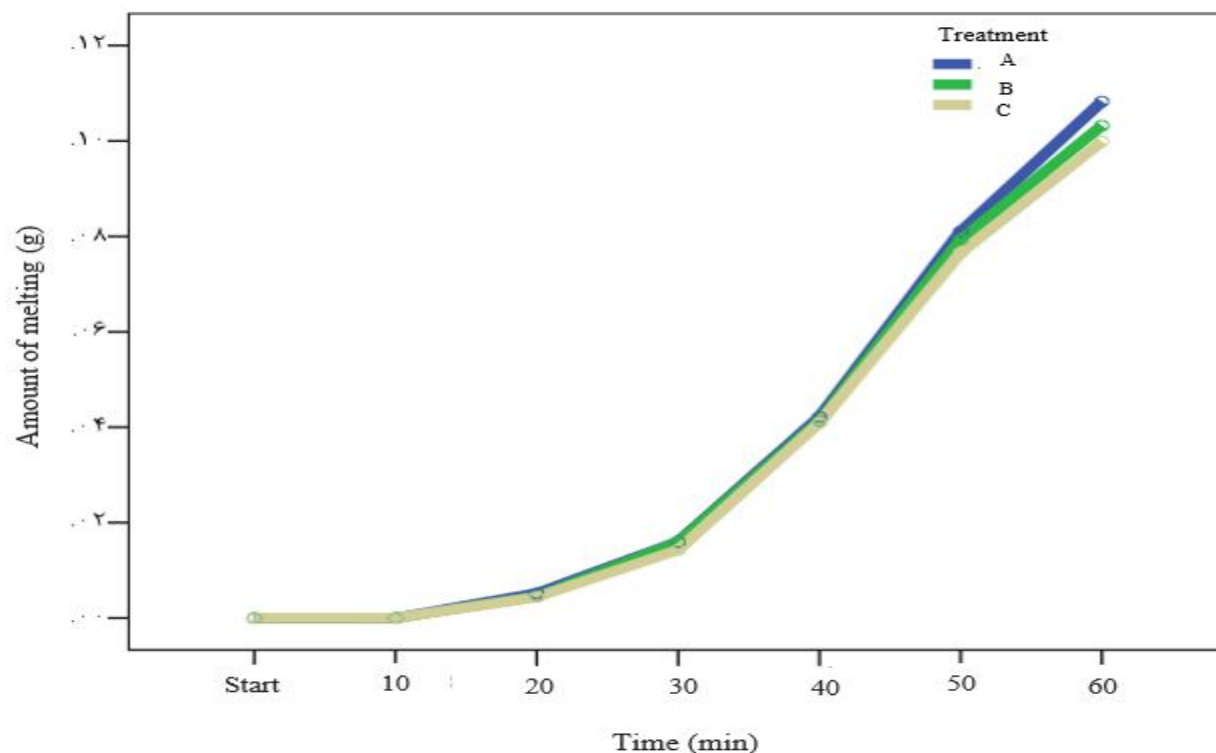
Drawing temperature of products in sample C was significantly lower than the other samples ( $p \leq 0/05$ ).

**Table 2. Study of viscosity, Hardness, Overran and Drawing temperature**

Treatment type	Dynamic viscosity (mpa.s)	Kinematic viscosity (mm <sup>2</sup> /s)	Hardness (g)	Overran (%)	Drawing temperature (°C)
A	1241 <sup>c</sup>	1128 <sup>c</sup>	1261 <sup>c</sup>	25.33 <sup>a</sup>	-1
B	1668 <sup>b</sup>	1530 <sup>b</sup>	1466 <sup>b</sup>	20.33 <sup>b</sup>	-1
C	1849 <sup>a</sup>	1709 <sup>a</sup>	1880 <sup>a</sup>	13.33 <sup>c</sup>	-2

A (Control treatment), B (5 % Extract), C (15 % Extract). The same letters indicate no significant differences ( $p \geq 0/05$ ).

The melting speed in different samples did not differ significantly ( $p \geq 0/05$ ). However, the melting speed was shown to be lower in samples containing the extract than the control sample.



**Diagram 1.** Study the melting speed. A (Control treatment), B (5 % Extract), C (15 % Extract).

*Escherichia coli*, *Salmonella*, *Staphylococcus aureus* and *Enterobacteriaceae* not found in none of the ice cream samples. In the enumeration of microorganisms, significant differences were observed among the samples ( $p \leq 0.05$ ). The number of microorganisms in the control sample was more than the two other samples.

**Table 3. Average of microbial count (cfu/ml)**

Treatment type	<i>Escherichia coli</i>	<i>Salmonella</i>	<i>Staphylococcus aureus</i>	<i>Enterobacteriaceae</i>	Microorganisms
A	Negative	Negative	Negative	Negative	$2.567 \times 10^{3a}$
B	Negative	Negative	Negative	Negative	$1.967 \times 10^{3b}$
C	Negative	Negative	Negative	Negative	$1.300 \times 10^{3c}$

A (Control treatment), B (5 % Extract), C (15 % Extract). The same letters indicate no significant differences ( $p \geq 0.05$ ).

## Discussion

Awareness of viscosity values, in addition to helping determine the most suitable ice cream formulation, is important in selecting the appropriate pump to transport and design the

required equipment. Furthermore, viscosity is an important factor in the rate of creaming, the mass transfer rate, and the flow conditions of milk and dairy products (Goff, 2008). However, the desirable amount of viscosity in ice cream has not been reported. In general, the results of the previous studies show that by increasing the viscosity, the resistance to melting and softness of the tissue increases and stirring speed decreases (Abdullah *et al.*, 2003). In this study, the results of tissue analysis and melting speeds tests confirms the findings of Abdullah's *et al.* (2003) because increasing the amount of viscosity increases the hardness and resistance to melting of the treatments. In this study, since the only difference among the samples is the concentration of the extract, it can be concluded that the extract of wild rose (*Rosa canina L.*) has increased viscosity.

Moussey *et al.* (2004) reported that the use of gum and an increase in its amount, despite the reduction of water for freezing, makes the ice cream more rigid, because of the increased viscosity. Relatively high hardness in ice cream is desirable (Homayounirad *et al.*, 2005). In this study, sample C had the highest viscosity and hardness in comparison to the other two samples, which was in conformity with the findings of the mentioned research.

The aeration in frozen desserts is directly related to the air entrusted to them during production and because of its impact on the quality of the product, it is of particular importance. Overran of ice cream is important because of its relation to efficiency and its impact on the structure and texture of ice cream (Goff, 2008; Goff and Sahagian, 1996). Some researchers reported the amount of desirable overran in high quality ice cream 25-50%, and some others for hard ice-cream 37.7-71.3 % (Muse and Hartel, 2004). Among the factors that affect the amount of overran we may refer to ingredients, viscosity and type of ice cream maker device in terms of compressor power and blades performance (Akalm *et al.*, 2008; Issariyachaikul, 2008).

Akin (1990) pointed to the problems of increasing overran by more than 40-45 percent in soft and semi-soft ice creams produced in non-continuous ice cream makers.

In this study, the highest amount was observed in the control sample, which was 25.33%. This low aeration may be due to flaw in stirring mechanism, because home ice cream maker has been used. The precise judgment about the ability of the extract on air trapping and maintenance requires more investigations and more advanced aeration systems (Bahramparvar *et al.*, 2008).

Gohari Ardabili *et al.* (2005) carried out a study on the effect of sugar replacement with date saps on the physical and sensory properties of soft ice cream, announced that by increasing

viscosity, the amount of overran decreases. This is in conformity with the findings by Gohari Ardabili *et al.* (2005).

Sugar is the most effective compound in the freezing point, and among the sugars, the lower the molecular weight, the greater its ability to reduce the freezing point. Thus, glucose and fructose monosaccharides reduces the freezing point twice more than sucrose with the same weight (Marshall and Arbuckle, 1996). The reduction in the ice cream temperature has been due to the types of sugar (glucose, fructose) in the extract

Akin *et al.* (2007) stated that the process of melting ice cream is related to the freely movement of molecules, samples C and B had freely of movement lower than control sample, because the viscosity and hardness of these samples were higher than the control sample.

Presence of high amounts of vitamin C, phenolic compounds, flavonoids and carotenoids has introduced fruit of wild rose (*Rosa canina L.*) as an important source of antioxidant compounds (Moure *et al.*, 2001; Von Gadow *et al.*, 1997). Carotenoids are a large group of plant pigments which are available in fruits and leaves of the plants. Carotenoids protect our body against diseases due to its antioxidant properties and prevent the formation of free radicals in human body (Kirakosyan *et al.*, 2004). According to Olsson *et al.* (2004), fruit of wild rose (*Rosa canina L.*) has higher level of antioxidants compared to many other fruits, for example the carotenoids found in fruit of wild rose (*Rosa canina L.*) are 6 to 7 times more than that of blackberry (Olsson *et al.*, 2004). The amount of carotenoids in different species of rose ranges from 189-1192 ( $\mu\text{g} / \text{g}$  dry weight) and the average carotenoids in rose fruits are 651  $\mu\text{g} / \text{g}$  dry weight (Olsson *et al.*, 2005). The phenolic compounds or polyphenols are chemically large and diverse groups which include simple phenolic acids, very large and complex polymers such as tannins and lignin. Pigments like flavonoids are among these compounds (Lila, 2004; Winkel-Shirly, 2002). In all plants, antioxidant activity is related to phenolic and flavonoid compounds (Swetie *et al.*, 2007). The key role of phenolic compounds as removers of free radicals has been reported in several articles (Katalinik *et al.*, 2006; Theriault *et al.*, 2006). Polyphenols have antiviral, antimicrobial properties and high antioxidant abilities (Reyes-Carmona *et al.*, 2005). Fruit of wild rose (*Rosa canina L.*) contains high levels of phenolic compounds that have a beneficial effect on human health (Cinar and Colakoglu, 2005). Moreover, Štajner *et al.* (2014) reported antioxidant activity and swept free radicals in fruits of wild rose (*Rosa canina L.*).



Therefore, anti-microbial properties of the fruit extract of wild rose (*Rosa canina L.*) are related to the function of phenolic compounds. This is evident in the enumeration of microorganisms.

## Conclusion

The results indicated that the hardness of the samples increased by increasing the extract percent. Since the relatively high hardness in ice cream is desirable, adding fruit extract of wild rose (*Rosa canina L.*) to ice cream improves its tissue properties. In addition, by increasing the viscosity and hardness, the melting speed in samples containing extracts was reduced compared with the control sample. Antibacterial properties of fruit extract of wild rose (*Rosa canina L.*) were evident in the total bacterial count. According to these results, the fruit of wild rose (*Rosa canina L.*) is suitable as an antimicrobial agent for patients with safety disorder.

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