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Product Optimization and Chemical and Microbial Properties of Low Fat Cream Formulated with Mastic Gum and Inulin

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Abstract

The objectives of the study were to formulate low fat cream utilizing mastic gum and inulin and to optimize the product for chemical properties and microbial quality with response surface method. Low fat cream (18, 19 and 20% fat concentration) was formulated with addition of mastic gum (in proportions of 0.2, 0.5, and 0.8 wt./vol. %) and inulin (in proportions of 1.2, 1.6, and 2.0 wt./vol. %) together. Chemical properties of samples (pH, protein, fat free dry matter (DM), fat, carbohydrate and energy content) and microbial quality (Total bacterial count, *Coliform, E. coli, Staphylococcus* coagulase positive) were determined during storage. Chemical properties (pH, fat free DM, density and fat) of raw milk and initial cream were determined and they were found suitable for production of low fat cream. At 0.5% gum and 1.6% inulin concentration, fluctuations in pH considerably decreased. Fat free DM, carbohydrate and energy content significantly increased with addition of mastic gum and inulin. Protein significantly increased in sample with 2% inulin ($P < 0.01$). Maximum fat in samples was 20.3% and it was seen at 0.8 and 1.8% of gum-inulin, and at 0.8 and 20% concentration of gum-cream respectively. Presence of mastic gum and inulin inhibited microbial growth in low fat cream. Low fat cream prepared with optimized formula (standardized using Design Expert Software for numerical optimization with concentration of 0.68% mastic gum, 1.88% inulin and 19.8% cream) had excellent quality characteristics and could compare well with commercial low fat cream.

Keywords: *Mastic Gum; Inulin; Low Fat Cream; Chemical Properties; Microbial Quality*

Introduction

Cream is defined as the layer of butter fat on the surface of milk before the milk is homogenized. Cream is basically an emulsion of fat in water. This means that milk fat globules are dispersed as small droplets in a continuous medium of skimmed milk containing protein, lactose, minerals and some vitamins. In many countries, cream is sold in several grades depending on the total butterfat content. Cream can also be dried to a powder for shipment to distant markets. Cream has high levels of saturated fat. Cream produced by cattle grazing on natural pasture often contains some natural carotenoid pigments derived from the plants they eat, which gives the cream a slight yellow tone, hence the name of the yellowish-white color, cream. Cream from goat's milk, or from cows fed indoors on grain or grain-based pellets, is white [1].

There are very limited compounds, which are suitable to be used as a filler in food systems. The resin of pistachios Bene tree is one of them that has been considered in recent years, though with serious limitations. Resin, as a hard and fragile material, can be studied as an organic filler in the food matrices. Additionally, it carries remarkable health properties, which further points at the importance of using this material. Resin, the wild pistachio of Iran, from Bene species (*P. atlantica subsp kurdica* and *mutica*), also known as mastic gum, is obtained by cutting a furrow on the trunk of Bene trees. Mastic gum has many pharmaceutical, food and industrial applications such as in adhesive, nail polish, color, polymeric, pharmaceutics, essence and perfume industry and it is also used in the traditional medicine. Many studies have confirmed the biological properties of Resin Pistachio, namely its anti-bacterial, remedial (especially for digestive diseases), anti-fungal, antioxidant, anti-cancerous, and anti-inflammation properties [2,3]. Another filler used in this study is inulin. Inulin is a group of natural polysaccharides that are present in some plants as a means for storing energy especially in underground roots and stems. It is extracted from the roots of some plants like chicory and artichoke. Investigation on the effect of inulin and polydextrose on the physico-chemical and sensory properties of low fat probiotic yoghurt showed that adding each of these two probiotics as substitute could improve, physical and sensory properties of yoghurt. Also, the number of probiotic bacteria in the total storage period was 8.86 CFU/g which was quite acceptable [4]. In recent years, focus is on development of low fat food products on account of increasing obesity among population and consumer demand. Mancinic., *et al.* [5] stated that viscoelastic properties of low-fat mayonnaise could be improved by the addition of xanthan gum and alginates. There are not many published reports available on the usage and effect of mastic gum as an additive in the formulation of food products, though it has many industrial applications. It is possible to utilize it in dairy products as an additive to improve their quality. Since mastic gum has a low solubility in water, it was reported that it needs a stabilizer in the formulation to make a synthetic protein network in the pizza cheese, hence, stabilized mastic gum can function as a filler in the obtained network [6]. The objectives of present study were to formulate low fat cream using mastic gum and inulin in different proportions and to determine the chemical properties of prepared products for product optimization. In addition, microbial quality of cream was also analyzed in stored samples.

Materials and Methods

Materials

Milk (2.5% fat) and cream (30 % fat) was obtained from Pegah factory, Mashhad, Iran. Sugar and vanilla were purchased from the local supermarket. Inulin powder 10 DP was obtained from Beneo Orafti Company in Tehran, Iran. Mastic gum was obtained from Behin Azma Company in Shiraz, Iran. A commercial pasteurized low fat cream (from Danone Dairy pars factory, Ghazvin, Iran prepared with ingredients: milk, dry milk, gelatin, preservative) was purchased from supermarket and used for comparison. All chemicals needed for microbial analysis, Plate Count Agar medium, Violet Red Bile Agar medium, Lauryl Sulfate Broth medium, Baird-Parker medium, *E. co*li Broth medium, Tryptone water medium and Kovacs indole reagent (Merck, Germany) were purchased from Azma Laban Company in Mashhad, Iran.

Product Development

Preparation of low fat cream

The study design comprised of formulating low fat cream (with 18, 19 and 20% fat) using varying proportions of milk and cream along with mastic gum and inulin derived using Pearson Square is shown in table 1. A total of 20 formulations were prepared with the following process - A known amount of milk was placed in a beaker on a water bath (85^{o}C) and heated to reach 45^{o}C to 50^{o}C followed by addition of inulin and mastic gum powder and stirring well to ensure complete dissolution. The mixture was continued heating for one minute at 75^{o}C, removed from the water bath, cooled to room temperature and mixed in a blender with the calculated amount of cream for a minute (twice for 30s) and placed for 24 hr in the refrigerator at a temperature of 6^{o}C to obtain cream with desired fat [7].

First Phase: Determination of chemical properties of formulated cream model for optimum selection

For selection of optimum model, a total of 20 different formulas of the cream model were produced according to the type and proportion of variables and the RSM (Response surface method) design. The variables were mastic gum, inulin and cream and the amount of each variable is shown in table 1. The effects of the added mastic gum in combination with inulin and cream on chemical properties of the formulated model were examined and the formulation which had best quality characteristics was selected.

Run	Block	Mastic Gum (%)	Inulin (%)	Cream (%)	pH D(0)	pH D(3)	pH D(6)	pH D(10)	Protein (%)	Fat free dry matter (%)	Fat (%)	Carbohydrate (%)	Energy Content (Kcal.)
1	Block 1	0.2	1.2	18	6.57	6.56	6.55	6.54	2.42	8.2	18.0	4.2	189.38
2	Block 1	0.8	1.2	18	6.58	6.57	6.55	6.54	2.43	8.3	18.1	5.1	194.23
3	Block 1	0.2	2.0	18	6.57	6.56	6.54	6.53	2.44	8.4	18.2	5.8	197.97
4	Block 1	0.8	2.0	18	6.56	6.54	6.52	6.52	2.44	8.5	18.1	9.1	203.89
5	Block 1	0.2	1.2	20	6.55	6.54	6.53	6.52	2.44	8.7	19.0	3.5	199.18
6	Block 1	0.8	1.2	20	6.60	6.60	6.59	6.56	2.42	9.2	20.0	5	211.83
7	Block 1	0.2	2.0	20	6.60	6.58	6.56	6.55	2.45	9.5	20.0	5.9	215.06
8	Block 1	0.8	2.0	20	6.61	6.60	6.58	6.57	2.43	9.7	20.3	10.2	231.82
9	Block 1	0.2	1.6	19	6.58	6.57	6.55	6.54	2.42	8.4	19.1	4.7	201.6
10	Block 1	0.8	1.6	19	6.58	6.57	6.56	6.54	2.41	8.8	20.0	7.8	203.7
11	Block 1	0.5	1.2	19	6.59	6.58	6.57	6.55	2.41	8.4	19.1	4.5	200.84
12	Block 1	0.5	2.0	19	6.59	6.58	6.56	6.55	2.42	9.0	19.1	9.3	217.64
13	Block 1	0.5	1.6	18	6.60	6.57	6.53	6.53	2.44	8.3	18.2	5.2	195.57
14	Block 1	0.5	1.6	20	6.61	6.60	6.55	6.54	2.45	9.3	20.1	5.1	212.36
15	Block 1	0.5	1.6	19	6.60	6.59	6.57	6.55	2.41	8.7	19.3	5.9	208.17
16	Block 1	0.5	1.6	19	6.60	6.59	6.56	6.55	2.42	8.7	19.2	5.6	206.91
17	Block 1	0.5	1.6	19	6.59	6.57	6.56	6.54	2.43	8.6	19.0	6	205.17
18	Block 1	0.5	1.6	19	6.60	6.59	6.57	6.55	2.41	8.6	19.0	6.2	205.47
19	Block 1	0.5	1.6	19	6.60	6.59	6.57	6.55	2.42	8.7	19.2	5.7	207.44
20	Block 1	0.5	1.6	19	6.61	6.60	6.56	6.55	2.42	8.7	19.0	5.7	204.86

Table 1: *Formulation of products with varying proportions of gum, inulin and cream and their chemical properties.*

(D): day.

Optimization of formula: For optimization of formula, Design Expert software was used for numerical optimization. The aim of optimizing the production process of the cream model was to obtain the ideal proportion of ingredients for best quality product with respect to the chemical properties, namely, pH, protein, fat free DM, fat, carbohydrate, and energy content.

Second Phase: Comparison of optimized model with the commercial cream sample

In the second step, the optimized cream model as determined from the first phase was compared with low-fat commercial control samples in terms of chemical properties on 10th day. This evaluation was based on the nutrient content of cream and pH changes over storage duration of 1, 3, 6 and 10 days (D).

Third Phase: Comparison of the optimized model for microbial quality with commercial sample

In the third step, on tenth day the optimized sample from the second stage was compared with the control sample (commercial low-fat cream) in terms of microbial quality for total plate counts, and presence of *Coliform, E. coli* and *Staphylococcus* coagulase positive.

Methods

Chemical analysis

The pH, fat free DM, density and fat of raw milk were determined with the milk analyzer machine (KOMILK TOTAL, Model: M-PRO manufactured in Iran). The acidity of raw milk and cream were determined according to standard AOAC method [8] by titration with an acid of known normality with phenolphthalein indicator (Fetalin) until the pink color was observed.

Chemical analysis for low fat cream samples and optimized sample: The pH was measured using a digital pH Meter (METROHM, Switzerland). The pH meter was alternately calibrated with Buffers 4 and 7 to yield the results with the least error. Protein was determined by Kjeldahl method (N×6.38) and fat by Gerber method according to the general procedures of AOAC [7]. To measure the free fat DM, a 3.0g sample was placed in an oven at 110°C for 3 hr and then transferred to a desiccator, cooled and weighed till constant weight could be obtained [8]. Carbohydrate content was measured by direct acid hydrolysis method. A 4.0g portion of sample was ground well with 10.0 ml ether and filtered to remove fat. The fat free residue was transferred to a beaker with 50 ml cold water and filtered and washed again. The insoluble residue was transferred to a reflux condenser with 200 ml water and 20 ml HCL, heated for 2.5 hr, cooled, neutralized with NaOH and glucose determined in aliquot and converted to CHO by multiplying with 0.925 [7]. The total energy content (as calorie) was calculated by using standard Atwater values of protein = 4.27, carbohydrate = 4.01 and fat = 9.02 kcalories per 100g [9].

Microbial analysis

Total microbial count of cream samples was determined on Plate Count Agar (PCA) culture incubated at 30 ± 1°C for 72h. Cream samples were analyzed for coliform bacteria and *Staphylococcus* coagulase positive. Coliform was determined on Violet Red Bile Agar (VRBA) incubated at 35 ± 1°C for 24 hr and *Staphylococcus* coagulase positive was determined as a surface culture on Baird Parker Agar (BPA) medium incubated at 37 ± 1°C for 24 - 48 hr. The *E. coli* test was conducted in 3 steps. In the first step, the sample was added to Lauryl Sulfate Broth culture and incubated at 37 ± 1°C for 24 - 48 hr. If the samples did not produce gas in the tube, result was negative. If gas was observed, then in the second step, 0.1 ml sample was taken and added to *E. coli* Broth culture and incubated at 44 ± 1°C for 24h. If the samples did not produce gas in the tube, it did not have *E. coli*. If there was gas production, in the third step, 0.1 ml sample was taken and added to the Tryptone water culture and incubated at 37 ± 1°C for 48 hr. Then Kovacs indole reagent is added and a color change to pink or purple indicates presence of *E. coli* [10].

Statistical Analysis

For the first phase, the treatment and determination of the proportions which were based on the central composite design and RSM were determined by using the Design Expert software version 7.0. In the second phase, for optimization of formula, Design Expert software was used for numerical optimizations. The data presented are expressed as mean ± SD. Data was subjected to two way ANOVA by SPSS 11th version and Microsoft Excel, 2010.

Results and Discussion

The results of the study are compiled in tables 2-5 and figures 1-7.

Chemical Properties of Product

Chemical properties of raw milk and cream

The raw milk and cream used for preparing low fat cream should be of good quality. Milk should be collected in safe condition from healthy livestock and transferred to the factory in the shortest possible time. The quality of raw milk and cream used for producing low-fat cream in present study were subjected to chemical tests as shown in table 2. All the quality parameters analyzed for raw milk and cream were in the standard range [11,12].

Sample	Moisture (%)	Dry matter (g/100)	Density (g/cm^3)	Fat (%)	Acidity (%)	pH
Milk	89.5	10.5	1.0295	2.5	0.144	6.67
Standard range [11,12]	89.2 - 89.8	At least 8	1.029 - 1.030	1.8 - 3.0	0.14 - 0.16	6.6 - 6.8
Cream	65.0	5.0	-----	30.0	0.12	6.62
Standard range [11,12]	50-70	4.5-6	-----	18 - 35	0.09 - 0.15	6.5 - 6.8

Table 2: *Chemical composition of milk and cream.*

The quadratic model for pH on 0, 3rd, 6th and 10th day (Table 3); and for protein, fat free DM, fat, carbohydrate and energy for the formulated cream sample was statistically significant at a confidence level of 99% (P < 0.01) and lack of fit value was not significant (P > 0.05). [Real values for all chemical properties are included in table 1]. A description of individual quality parameters based on the response surface curves drawn by the Design Expert software is presented below.

Changes in pH over storage

The changes observed in pH of cream samples due to interaction between changing concentrations of gum and inulin through the storage period are depicted in 3-D (Figure 1). As can be seen at the fixed concentration of 19% cream, by increasing the concentration of gum and inulin to moderate levels (0.5% of gum and 1.6% inulin) the pH of samples increased. However, on further increase of gum to 0.8% the pH of the sample decreased, which is probably due to the significant antagonistic effect of AB phrase in ANOVA table 3. Similar effect was observed up to 10th day of storage. The maximum and minimum pH observed in cream samples was between 6.61 to 6.52, which shows that the fluctuations in pH were very low through the period of storage. The effect of gum was larger than the effect of inulin in all cases. On the first and third day of storage, the maximum pH in samples was observed at concentration of 0.5% gum and 1.6% inulin and the minimum pH in the samples was seen at concentration of 0.2% gum and 1.2% inulin. On sixth and tenth day, the concentrations of gum and inulin for maximum pH were 0.8 and 1.2%; and 0.8 and 2.0% respectively. For the minimum pH, the concentrations remained similar as seen earlier with one exception of inulin at 1.4% on tenth day. According to results, addition of mastic gum and inulin increased the protein level in low-fat cream. As mastic gum has buffering properties [13], it lead to considerable reduction in fluctuations in pH during shelf life of cream. This shows the effectiveness of using mastic gum and inulin in low fat cream.

Variables	pH (Storage days)				Protein	Fat free DM	Fat	CHO	Energy
	0 (D)	3 (D)	6 (D)	10 (D)					
Model	6.60**	6.59**	6.56**	6.55**	2.42**	8.66**	19.20**	5.85**	206.01**
A-mastic	6.000*	7.000*	7.000**	5.000*	-4.000^{ns}	0.13**	0.22**	1.31**	4.23**
B-inulin	4.000ns	1.000ns	-3.000ns	1.000ns	6.000**	0.23**	0.15^{ns}	1.70**	7.09**
C-cream	9.000**	0.012**	0.012**	8.000**	2.000^{ns}	0.47**	0.88**	0.030^{ns}	8.92**
AB	-7.500**	-8.750*	-7.500*	-3.750	-1.250^{ns}	-0.037^{ns}	-0.11^{ns}	0.65**	0.65^{ns}
AC	7.500**	0.011**	0.013**	8.750**	-6.250**	0.063*	0.16^{ns}	0.20*	2.33*
BC	0.010**	8.750*	7.500*	8.750**	-1.250^{ns}	0.11^{ns}	0.14^{ns}	0.25^{ns}	2.20**
A^2	-0.018**	-0.015*	-1.818**	-3.636*	-5.000^{ns}	-0.050^{ns}	0.21^{ns}	0.40*	-2.87**
B^2	-8.182ns	-4.545*	8.182**	6.364*	-5.000^{ns}	0.050^{ns}	-0.24^{ns}	0.55**	3.72**
C^2	6.818ns	4.545*	-0.017**	-8.636*	0.025**	0.15**	-0.19^{ns}	-0.70**	-1.56^{ns}
Residual	-------	--------	--------	-------	-------	-------	-------	-------	-------
Lack of Fit	ns	ns	ns	ns	ns	ns	ns	ns	ns
Pure Error	--------	--------	--------	-------	--------	--------	--------	--------	--------
Cor Total	---------	--------	--------	-------	--------	--------	---------	---------	---------
CV %	0.100	0.13	0.1	0.077	0.24	0.83	1.15	3.87	1.01
R^2	0.9216	0.8917	0.9203	0.9103	0.8978	0.9838	0.9511	0.9899	0.9742
R^2_{adj}	0.8510	0.7941	0.8485	0.8295	0.8059	0.9692	0.9070	0.9808	0.9510
Press	2.791	2.256	2.517	1.383	9.083	0.49	3.07	2.81	282.53
Ad-P	13.220	10.891	14.742	0.077	10.853	32.477	16.144	42.310	27.686

Table 3: *Analysis of variance (ANOVA) of quadratic polynomial models for pH during storage and chemical composition (Coefficient estimate).*

(P ≤ 0.05), (P ≤ 0.01)**, ns: not significant at 95% level. (D): Days; DM: Dry Matter.*

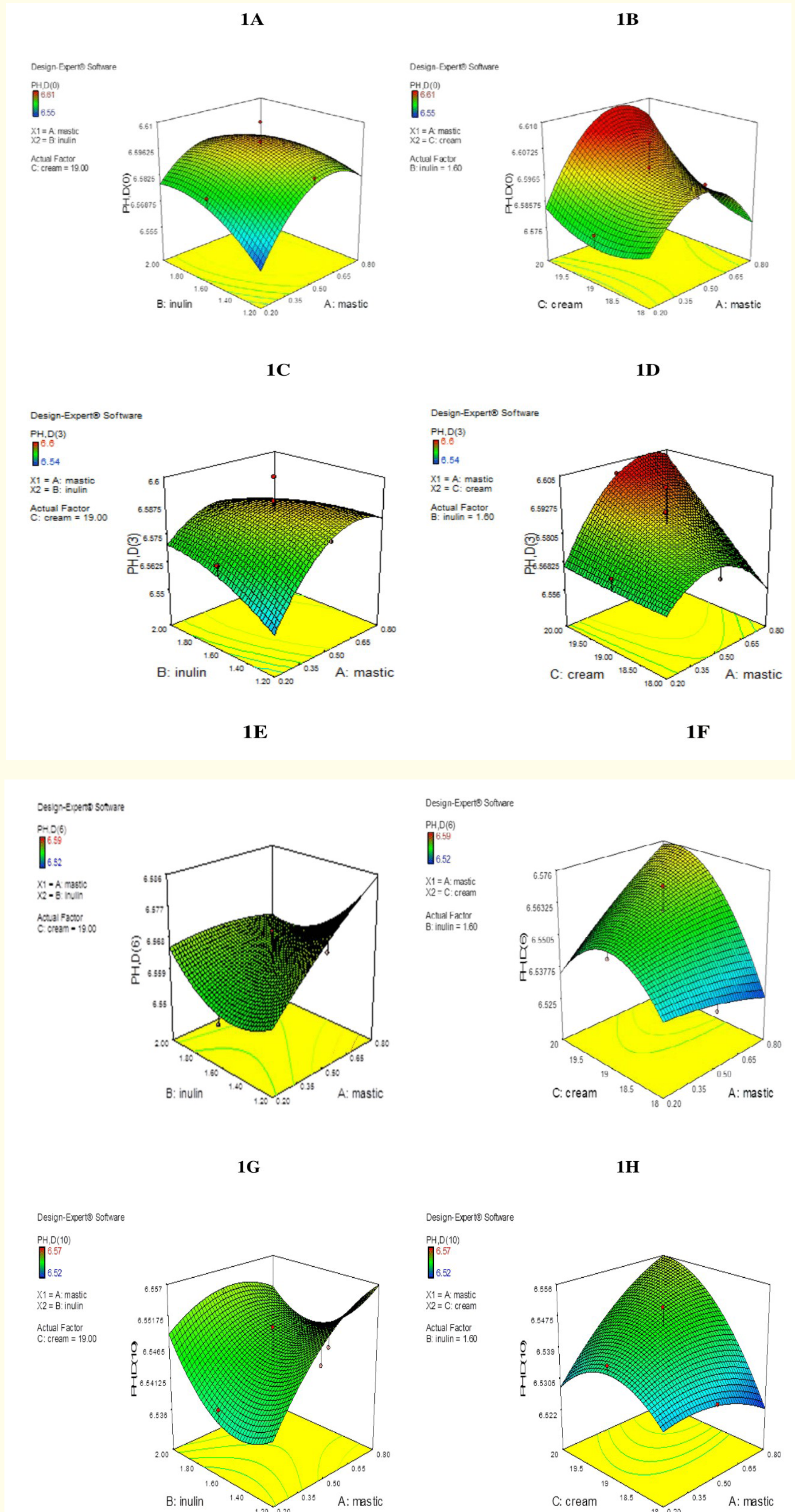

Figure 1: *Change in PH as a function of interaction between gum – cream and gum –inulin on 0(D): A and B, 3(D) C and D, 6(D) E and F, 10(D): G and H [D: days].*

Protein

Proteins in milk cream are chains of amino acid molecules that are connected by peptide bonds and comprise 3.3% of total protein. There are two distinct types of proteins in milk, casein and whey. Casein contains phosphorus and coagulates or precipitates at pH 4.6. Casein micelle granules are maintained as a colloidal suspension in milk and stabilize emulsion in milk products. Whey proteins are ingredients with unique functional properties for use in foods. They bind water in cream and yogurt, give structural stability and improve textural quality of dairy products [14]. As seen in table 3, linear expression for protein was significant only for inulin concentration ($P < 0.01$) and among quadratic expressions only cream concentration was significant ($P < 0.01$). Interaction between independent variables of gum-cream was significant ($P < 0.01$), as seen in 3-D charts (Figure 2). The average protein value was 2.43%. As shown in figure 2, at a fixed cream concentration of 19%, with increase in gum concentration, protein in sample decreased; though increasing inulin concentration raised it again and at a fixed concentration of inulin (1.6%), and low concentrations of cream, protein in samples increased with increasing gum concentration. However, with increase in cream concentration, increasing gum concentration caused reduction in protein. Data indicates that inulin had the greatest impact on the protein. In cream, protein level is one of the main factors influencing energy content. In low fat cream the protein level is less than full fat cream. Maximum protein value in the samples was obtained at 0.2 and 2% concentration of gum-inulin, and 0.2 and 20% of gum-cream. Minimum protein was obtained at 0.2 and 1.2% concentration of gum-inulin and at 0.8 and 19% of gum-cream respectively. Grindrod and Nickerson [15] investigated reactions of gums with skim milk and purified proteins. The results showed that some gums cause no reaction with milk proteins, however, some cause a strong protein-gum interaction such as carrageenan, locust bean gum, carboxymethylcellulose, pectin, alginate and modified starch. Hydrocolloids are not the only polymer type that are available in dairy products. Hydrocolloids, along with milk proteins (casein and whey proteins), dissolve in the aqueous phase and form a protein-polysaccharide-water polymer. Polymeric interaction often leads to the formation of complex phase or phase separation [16]. Results showed that the linear expression of inulin was significant and inulin affected the cream protein. Inulin, a polysaccharide forms a complex phase. Reaction of inulin with milk protein lead to formation of polymer that affects the textural properties of the cream.

Variable		Desirable formula software prediction	Desirable formula production	Control sample
Mastic		0.68 [a]	0.680 [a]	---------
Inulin		1.88 [a]	1.88 [a]	---------
Cream		19.8 [a]	19.8 [a]	---------
pH	D (0)	6.61272[a]	6.623±0.001[a]	6.60±0.0005[a]
	D (3)	6.60077 [a]	6.601±0.0002[a]	6.590±0.0007[a]
	D (6)	6.57320 [a]	6.592±0.002[a]	6.562±0.001[a]
	D (10)	6.56056 [a]	6.589±0.001[a]	6.55±0.0006[a]
Protein		2.42994[a]	2.431±0.003[a]	2.453±0.002[a]
Free Fat DM		9.46120 [a]	10.495±0.002[a]	11.1±0.1[a]
Fat		20.0967 [a]	20.009±0.04[a]	20.8±0.02[a]
Carbohydrate		8.4638 [a]	8.553±0.003[a]	10.413±0.03[b]
Energy		225.572 [a]	225.158±0.03[a]	239.846±0.05[b]
Desirability		0.829	-------------	--------------

Table 4: *Optimization of product formula for desirable quality.*

The mean value with different letters are significantly different ($P \leq 0.05$). D: Days.

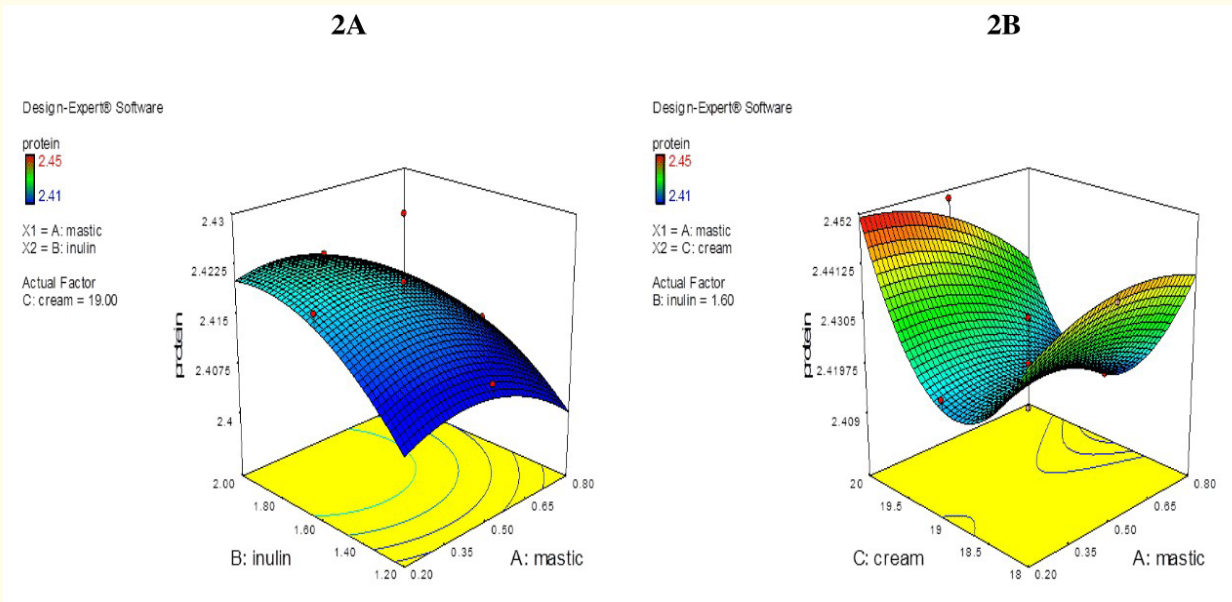

Figure 2: *Change in protein as a function of interaction between A: Gum - cream and B: Gum - inulin.*

Fat free DM

For fat, all linear expressions of independent variables became significant at 99% level ($P < 0.01$) and among quadratic expressions, only quadratic expression of cream concentration was significant at level of 99% ($P < 0.01$) (Table 3). Expressions related to interaction between independent variables of inulin-cream at 99% ($P < 0.01$) and interaction among independent variables of gum-cream at ($P < 0.05$) was significant. As per ANOVA data, median was equal to 8.73. As shown in figure 3, at fixed cream concentration of 19%, fat free DM in samples decreased as mastic gum concentration increased; but increase in inulin lead to increase in fat free DM. At fixed concentration of inulin (1.6%) and low concentrations of cream, with increase in gum concentration, fat free DM in samples increased. On the other hand, increase in cream concentration by 19% caused decrease in fat free DM, but with cream concentration exceeding this amount, fat free DM increased. Maximum fat free DM in samples was produced at 0.2 and 2% concentration of gum-inulin and at 0.2 and 20% concentration of gum-cream. Minimum fat free DM in cream samples was produced at 0.2 and 1.2% level of gum-inulin and at 0.8 and 19% level of gum-cream, respectively. According to the Iran National Standard No.191 [12] the fat free DM range in cream is between 8-9.6%. Cream texture does not have a desirable consistency in less than 8.0% range of fat fee DM. On the other hand, if the dry matter is more than the standard, it has a negative effect on the taste of the cream, and also the cream loses the creamy texture. The linear expressions of mastic gum and inulin were significant indicating that the addition of mastic gum and inulin affected fat free DM content in low-fat cream by increasing it within the standard range (Table 4). These results are in agreement with the findings of Alakali., *et al.* [17] who reported that addition of stabilizers (gelatin, carboxyl methyl cellulose and corn starch) increased the solid contents of thermized yoghurt. At high level of mastic gum and inulin, the creamy texture was reduced and the texture was excessively rigid, which may affect the organoleptic properties of low fat cream adversely. Therefore, a moderate percentage of gum has a more favorable effect on the product. A similar trend was reported by Routray and Mishra [18] who reported that the increase in total solids content by 5% lead to a doubling of the product's viscosity. Also, increase of the SNF (solids not fat) content increased the consistency and rigidity, and reduced the syneresis in low fat yogurt.

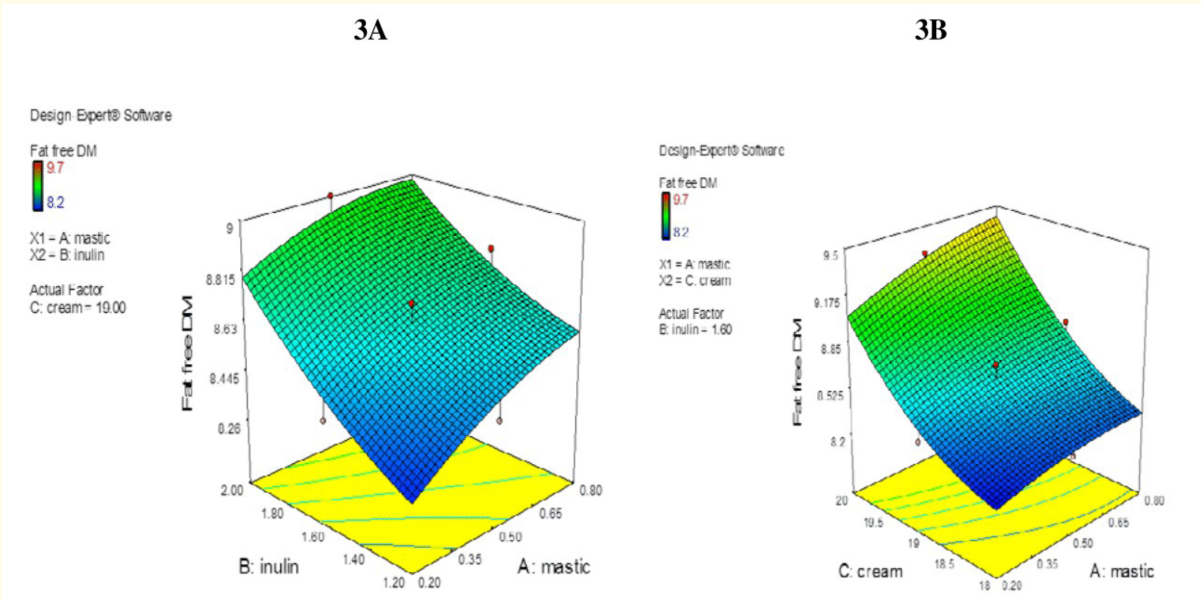

Figure 2: *Change in protein as a function of interaction between A: Gum - cream and B: Gum - inulin.*

Fat

Milk contains a total of approximately 3.4% fat, which is comprised of complex fatty acids [19]. The triglycerides of milk fat are in the form of globules. The milk protein is surrounded by triglycerides globules and phospholipid membrane that stabilizes the globules in the serum (water) phase of milk [17]. Functionally, fat affects the textural properties, melting point, viscosity, crystallinity and spreadability of many foods. In food, fats act as a structural component and cannot be eliminated without finding a fat replacer. In addition, fat is a substantial solvent for most hydrophilic compounds which are effective in providing flavor. As a result, structure, texture and taste of foods can change with modified fat content. Therefore, the lack of fat causes a change in the distribution of effective molecules responsible for taste of the product [20]. Fat replacers are macromolecules that are used to provide all or part of the fat function in a food product and fat substitutes contribute less calories than fat [21]. Linear expressions for gum-cream at 99% ($P < 0.01$) were significant, but none of quadratic expressions were significant and linear state could be seen in 3-D charts (Figure 4). Expressions related to interaction between independent variables was also not significant. Considering variance analysis table, data median was equal to 19.10. As shown in figure 4, at fixed cream concentration of 19%, with increase in gum and inulin, fat increased and at a fixed inulin concentration (1.6%), fat increased with increase in gum concentration and fixed cream concentration. Maximum fat in samples was produced at 0.8 and 1.8% of gum-inulin, and at 0.8 and 20% concentration of gum-cream respectively. Minimum fat in cream samples was produced at 0.2 and 1.2% concentration of gum-inulin and at 0.2 and 18% concentration of gum-cream respectively. The linear expression of gum was significant showing the effectiveness of mastic gum on fat content of low fat cream. It seems that mastic gum would be able to provide some of the functional properties of fat in a low fat cream. These results are in agreement with previous studies of Radi., *et al*. [7] who showed that on using sweetener stevioside as fat replacer for cream production, textural properties could be improved.

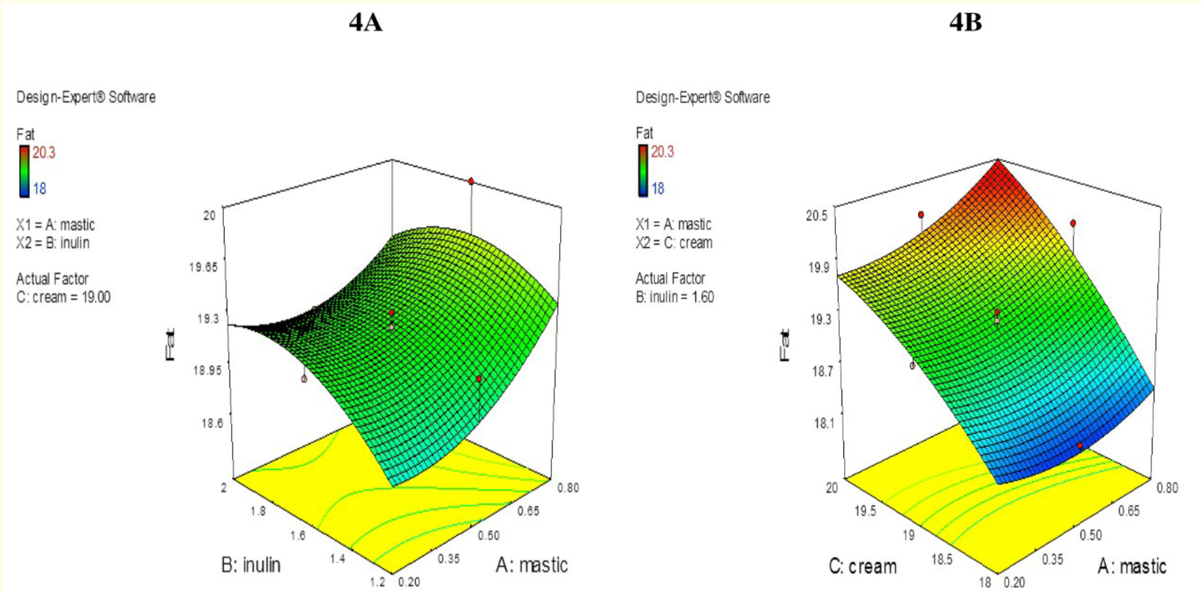

Figure 4: *Change in fat as a function of interaction between A: Gum - cream and B: Gum - inulin.*

Carbohydrate

For the carbohydrate content of cream, linear expressions of gum-inulin and quadratic expressions of inulin-cream were significant at level of 99% ($P < 0.01$). Also, quadratic expressions of gum concentration were also significant at level of 95% ($P < 0.05$) and accordingly, 3-D charts exhibited the curve. Expressions related to interaction between gum-inulin (99%, $P < 0.01$) and between cream-gum (95%, $P < 0.05$) were significant. Also considering variance analysis table, the data median was equal to 5.97. As shown in figure 5, at fixed cream concentration of 19%, carbohydrate content increased with increasing gum-inulin concentration. At fixed inulin concentration (1.6%), with increase in gum concentration, carbohydrate content increased in samples. On analyzing the effect of cream in medium concentration (19%), an increase in carbohydrate content was seen, however on exceeding a critical limit, there was a decrease. Milk contains approximately 4.9% carbohydrate content, which is predominately lactose. It is a disaccharide of glucose and galactose [22]. Mastic gum has approximately 87.9% total carbohydrate content, whereas inulin contains almost 86.4%. As shown in table 3, the linear expressions of gum and inulin were significant indicating that the presence of carbohydrate content in inulin and gum impacted the amount of carbohydrate content in low fat cream. The findings are in agreement with the reported addition of stevioside sugar in cream which has a significant impact on carbohydrate content of cream [8]. Inulin is the complex carbohydrate content (dietary fiber) and it is essential for regulating the body. In the food system, the carbohydrate content present in inulin can bind with water and provide a creamy texture and mouthfeel in a low fat food as a fat replacer [23]. Maximum carbohydrate content in samples was produced at 0.2% and 1.2% concentration of gum-inulin and 0.8 and 19.0% concentration of gum-cream. The minimum was produced at 0.2 and 1.2% concentration of gum-inulin and at 0.2 and 20% concentration of gum-cream, respectively.

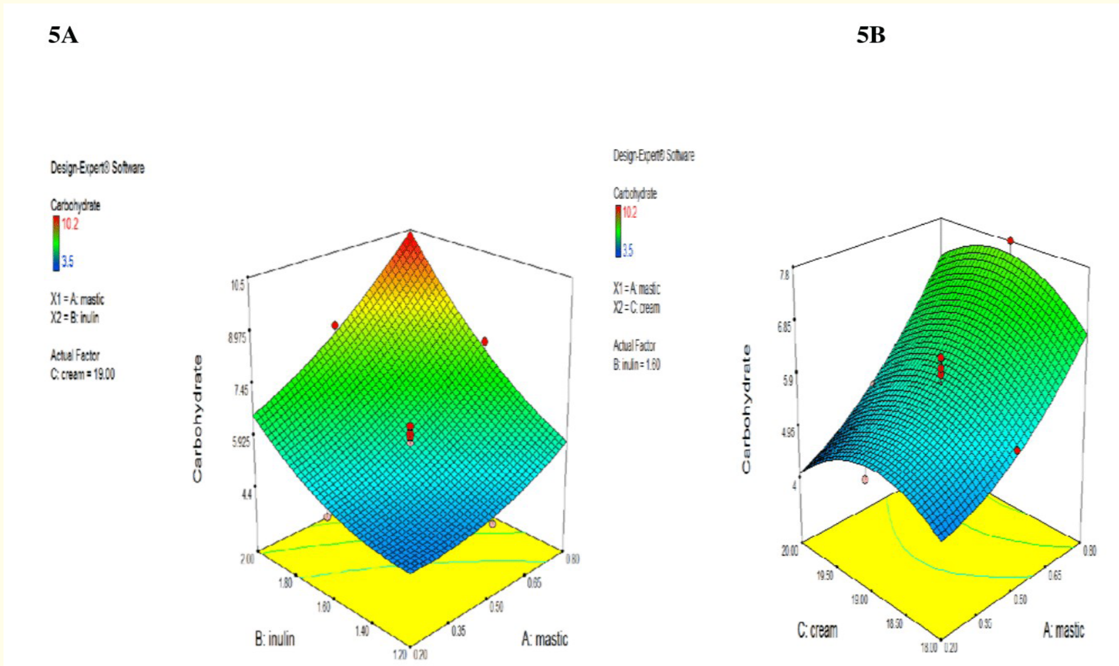

Figure 5: *Change in carbohydrate as a function of interaction between A: Gum - cream and B: Gum - inulin.*

Energy content

For the energy content of cream samples, all linear expressions of independent variables, i.e., inulin, gum and cream were significant and among quadratic expressions, only inulin and gum were significant, both at level of 99% ($P < 0.01$). Expressions related to interaction between independent variables of inulin-cream at a P-value of 99% ($P < 0.01$) and gum-cream at 95% ($P < 0.05$) were significant. As per the variance analysis table, the data median was equal to 205.65. As shown in figure 6, at fixed cream concentration of 19%, energy content in samples decreased as gum concentration increased; however, it increased with increasing inulin concentration. Energy content is calculated by the total sum of protein, carbohydrate and fat. The amount of mastic gum, inulin and cream affected the energy content. Energy content of low fat cream was significantly lower than full fat cream on account of reduction in fat content. Maximum energy content in samples was seen at 0.8 and 20% concentration of gum and inulin and 0.8 and 20% of gum and cream. Minimum energy content in cream samples was produced at 0.2 and 18% concentration of gum-inulin, and 0.2 and 18% of gum-cream, respectively.

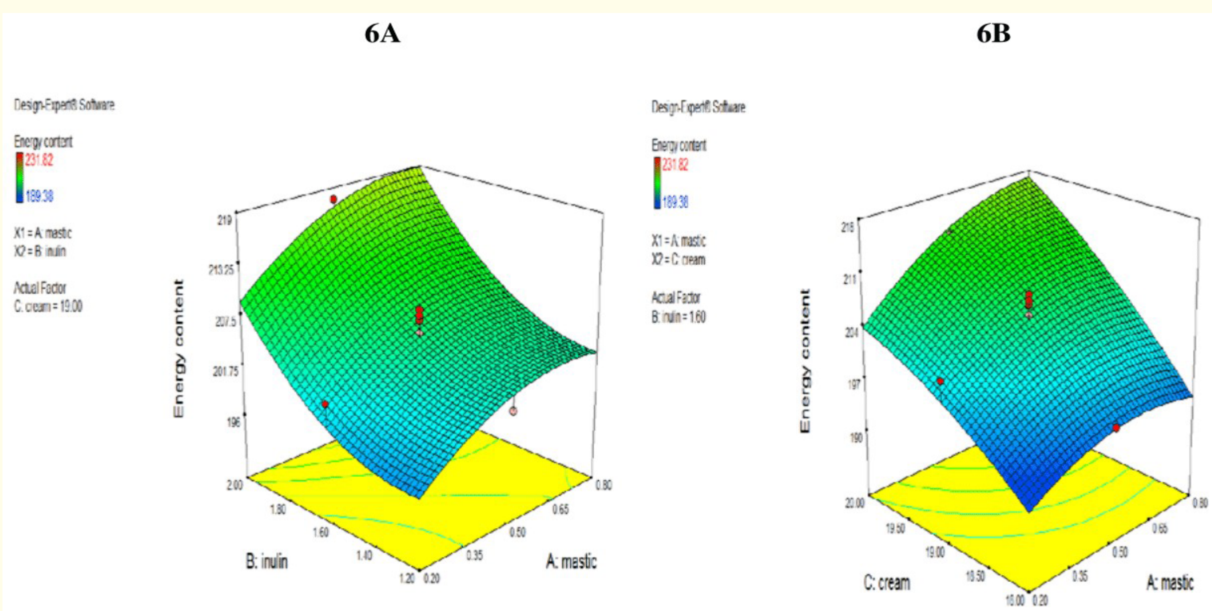

Figure 6: *Change in energy content as a function of interaction between A: Gum - cream and B: Gum - inulin.*

Optimization of product formula

For optimizing product formulation, the target range is selected after analyzing the variance of the data by selecting the variables for maximization or minimization of desired response in the obtained domain. In present study inulin, pH and protein were chosen in the maximum range, and corresponding range values for mastic gum, cream, fat free DM, fat, carbohydrate and energy content were considered. The upper, lower, and the range of each of these variables were determined. Finally, an optimized model for the production of cream using the combination of variables was obtained and the data is presented for desirable formula software prediction in table 4.

Microbial count (CFU g^{-1})	Desirable formulation	Control sample	Standard
Total count of microorganisms	893^{b}	1364^{c}	2×10^{4a}
Coliform	2^{b}	6^{c}	10^{a}
Escherichia coli	Negative	Negative	Negative
Staphylococcus coagulase positive	Negative	Negative	Negative

Table 5: *Microbial quality: Comparison between Desirable formulation, Control sample and Standard.*
The mean value with different letters are significantly different (P ≤ 0.05).

The results showed that optimum cream produced less fluctuations of pH than control cream, which can be explained on the basis of increased buffering capacity in optimized cream due to the presence of mastic-inulin in the formulated sample. Also, there was no significant difference in the formulated cream and the predictive model. As shown in table 4, for all variables there was no significant difference between optimized cream and prediction model. Between commercial cream and optimized sample, there were no significant differences in parameters of pH, protein, fat free DM and fat. However, the carbohydrate content and energy content in optimized sample was considerably lower in comparison to commercial cream. The higher carbohydrate content and energy content in the commercial sample can be explained on the basis of added additives such as gelatin and dry milk which are used for better texture of product.

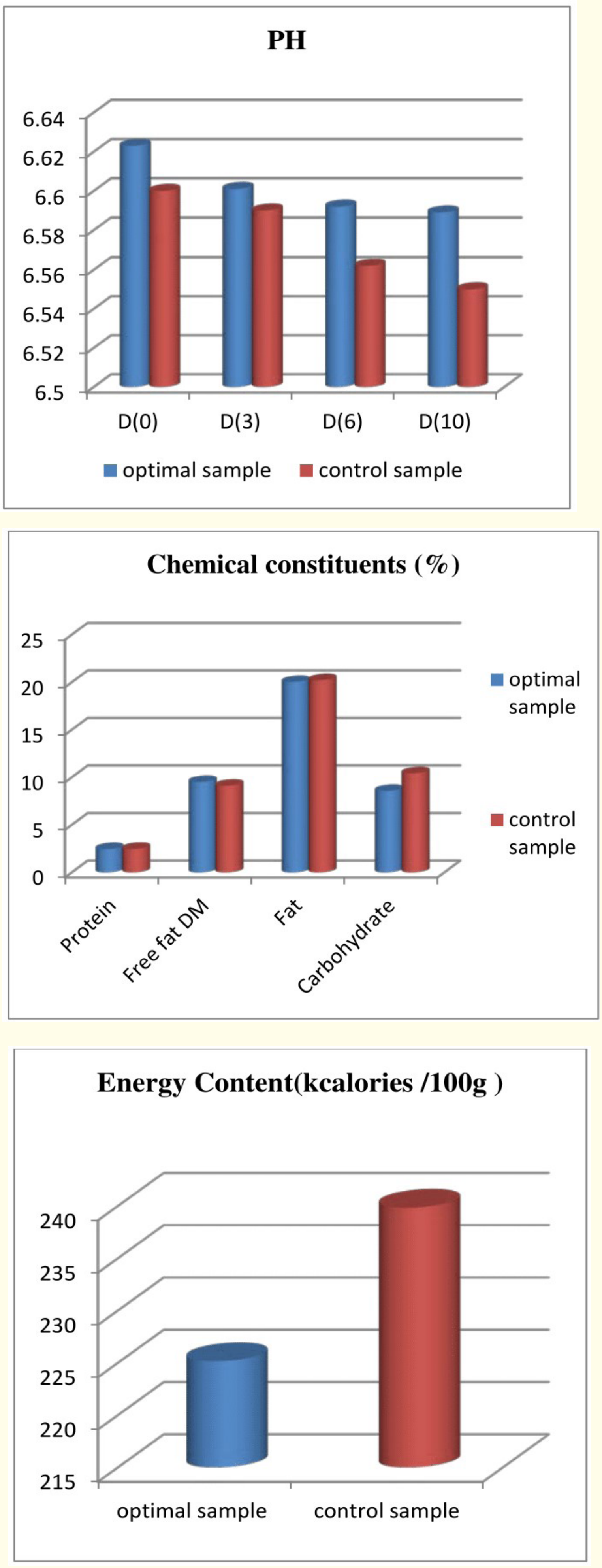

Figure 7: *Comparison of chemical properties between optimized experimental product and commercial control product [D: days].*

Microbial Quality

Cream can be subjected to microbial spoilage during storage with microorganisms such as lactic acid bacteria, which develop gas, *Coliform, E. coli* and test positive for *Staphylococcus* coagulase. The growth of microorganisms in the cream may cause undesirable flavors, off-color, bitterness and undesirable texture [24]. In industry for dairy products, artificial preservative such as potassium sorbate, calcium propionate, sodium benzoate, natamycin and nisin are used to control microorganisms. Many adverse side effects of using such chemicals have been mentioned [25]. In present study, mastic gum and inulin was used to inhibit microbial growth in cream samples. The National Standard of Iran proposes the maximum total count for microorganisms in cream as 20,000 CFU/g [10]. The microbial count of the control commercial sample (1364 CFU/g) and the optimized production sample (893 CFU/g) was in acceptable range. The *Coliforms* were also within the prescribed range and *E. coli* and *Staphylococcus* coagulase positive were negative.

The obtained microbial results showed that the control sample and the optimized sample were within the accepted standards (Table 5). Triplepenes and tryptopenides in the gum, which simulate the properties of steroid compounds are said to have antimicrobial properties. In addition to the antimicrobial properties, mastic gum has also been shown to have antifungal and antioxidant properties [26]. Inulin can also increase the antimicrobial activity by boosting the growth of probiotics and increasing the production of antimicrobial compounds such as acetic acid, lactic acid, benzoic acid, and various types of bacteriocin compounds. Based on the results, the effectiveness of the inulin and gum composition for controlling and reducing the microorganisms in optimized cream is proved.

Conclusion

Consumer demand for low-fat dairy products has increased due to lower calories and prevention of obesity, cardiovascular diseases such as atherosclerosis and cancer. Therefore, accordingly, hydrocolloid can be used as a fat replacer to create desirable properties in low-fat dairy products. Research studies have focused to find the best combination of compounds and their most suitable proportion for formulating low-fat dairy products with acceptable organoleptic properties. In this study mastic gum and inulin have been used in low fat cream and they do not have an adverse effect on the chemical properties. They tend to enhance the positive quality characteristics of formulated products. On the other hand, mastic gum and inulin could control the microbial growth in cream and these can be used for preservative effect.

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