

Effect of *Psycrotroph* Bacteria on Physical and Chemical Properties of Traditional White and Ultrafiltration Cheese

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The dominant factor of stored raw milk in low temperature are *Psycrotroph* bacteria and these bacteria as referred as to microorganisms that can survive and grow at 7 °C or lower. *Psycrotroph* bacteria is not heat resistance and pasteurization destroy them, but their proteolytic and lypolytic enzymes are heat resistance. These enzymes save their activities in pasteurization temperature and also UHT processing. In this study, effect of *Psycrotroph* bacteria on physical and chemical properties on ultrafiltration and white cheese was investigated. The results showed that number of *Psycrotroph* bacteria on white and ultrafiltration cheese was significant, so with increasing *Psycrotroph* bacteria the dry matter per cent, per cent of fat and firmness of texture was reduced. In spite of this, moisture of curd and softness of cheese texture increased by multiplying *Psycrotroph* bacteria. These changes in the ultrafiltration cheese were more than white cheese.

Key Words: *Psycrotroph* bacteria, Physical and chemical properties, Traditional white, Ultrafiltration cheese.

INTRODUCTION

Cheese is a condensed milk that its main of dry matter is protein (mainly casein) and fat which produced after coagulation and release of liquid and soluble matter of milk is called whey protein. Cheese ripening are including cheese storage at low temperature 0-16 °C for 2 to 3 min that its physical, chemical and bacteriological properties was changed and the taste, aroma and texture of curd was prepared. The most important changes in cheese ripening are biochemical change, change in aroma, taste and texture and the main biochemical changes that happened during ripening are include glycolysis, lypolysis and proteolysis¹⁻⁴.

These reactions cooperate with catabolic reaction such as deamination, decarboxylation, desulfurization and esterification. Initially, lactose changes into lactic acid, propionic acid, acetic acid and CO₂. Lactic acid was used by bacteria in the second step and the decomposition of nitrogen materials begin. This process was carried out by yeasts and molds and therefore the main parts of proteinous materials change from insoluble to soluble. In spite of this, fat hydrolyzed by lypolytic enzymes and

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free fatty acids and glycerols produced. These changes were more in the Roquefort cheese. The chief biochemical changes for the period of cheese ripening are protein hydrolysis which is the main factor of inducing texture, aroma in cheese. Decreasing pH or increasing acidity cause from lactose fermentation and production of amino acid and fatty acid by proteolysis and lypolysis during ripening. In the first month, production of amino acid continues strongly, but the main factor of pH decreasing was related to lactic acid production⁴⁻⁸.

Rheology of cheese is the study of deformation and flow, when affected by force. The rheological properties of cheese are properties which determine their reaction to force such as pressure and shear which is used in production or its consumption. These properties are including inherent properties such as elasticity and viscosity that determine by applying fixed force on the cheese sample under the experimental conditions. The relation between force and deformation may be described by different rheological phrase such as firmness coefficient⁹.

The behaviour of cheese as affected by force can be described by some phrase such as firmness, toughness, elasticity, tenderness and cohesiveness. Cheese rheology is a function of composition, physico-chemical state of components and curd particles. Physico-chemical properties are including agglomeration of fat, hydrolysis and the hydratability of p-casien. Rheological properties of cheese mainly changed by type and age¹⁰.

The investigation showed that *Psycrotroph* bacteria including negative gram rod bacteria that mainly relates to *Pseudomonas*, *Aromobacter*, *Aeromonas*, *Coliforms* and *Flavobacteriums* which dominate for change the properties and spoilage of milk at low temperature. pH increasing in the period of cheese storage cause by proteolysis and reach to 5 had some disadvantages, because the optimum pH for proteolytic activities of enzyme is 5.5-5.8.

EXPERIMENTAL

All the chemicals used in these experiments were Merk. The experiment was performed in east Azerbaijan of Pegah milk factory. The effect of bacteria on cheese was determined by dry matter and fat which was measured by Gerber method and texture was measured by instron. Incoming raw milk after some quality experiments (acidity, fat, density, pH, moisture content and temperature) was accepted.

Milk sample: Raw milk was prepared from winter season milk that used in the cheese making factory in the Pegah of east Azerbaijan of Iran. Rennet used in production of ultrafiltration and white cheese was from of fromase. The starter was mix of thermophile starter type yoghurt 709, yoghurt 231 and mesophile starter type G₃ mix 6, G₃ mix 7 that prepared in the ratio of 7 to 1, respectively.

Procedures: Dry mater of cheese was measured by moisture analyzer at 125 °C. Fat content of cheese was measured by Gerber method. The firmness of cheese was determined by instron universal model 1140 in the Laboratory of Faculty of Food Science, Tabriz University. In the firmness experiments of cheese, the pressure and

shear tests was conducted. For each sample of cheese, sample of 2 cm thickness was prepared of sample and shear resistance was measured. Load cell was 50 Nyotons and used a punch with 15 cm length and 0.11 cm diameter for putting into sample. The velocity of penetration of punch was 20 cm/min and velocity of chart was also 20. After calibrating the apparatus with zero and five, the maximum required force for cutting the sample was calculated by plotted curve. The cheese samples were prepared in the dimension 2 cm × 2 cm × 2 cm for pressure test to evaluating the firmness. Then, the special punch for this experiment was selected and the apparatus arranged to press the 75 % of initial sample length. All of calibration was also carried out for this apparatus.

The required force for pressing the sample was calculated by plotted graph of apparatus. Shear resistance graphs (s) for both type of cheese was calculated and the required force for pressing was earned. Total plate count (TPC) of raw milk was determined according to Iran standard number 356. Amount of *Psycrotroph* bacteria was measured according to Iran standard number 3451. The regression of coefficient (R^2) for cheese properties was determined by SPSS, Excel and Quattropro software. Normal distribution of data was used by the known method^{1,5}.

RESULTS AND DISCUSSION

The dry matter of traditional white cheese was measured after 2nd and 4th weeks and 6 months. Some initially studies confirmed the normalization of data for dry matter. There was a significant meaning between the *Psycrotroph* bacteria and dry matter of cheese in the period of ripening at 1 % possibility level. As the *Psycrotroph* bacteria increased, the dry matter of white cheese decreased. In comparison between 2nd and 4th week, the average of dry matter in the 4th week was lower than 2nd week and also for 4th and 6th week, the average of 6th week was lower than 4th week. It means as the time passes, the dry matter of cheese with multiplying *Psycrotroph* bacteria will be decreased. On the other hand, there was a interaction between amount of *Psycrotroph* bacteria and time of ripening for basis of dry matter¹¹.

The dry matter data of ultrafiltration cheese was showed the normalization for three periods of time according to primary examinations. There was a significant meaning between the *Psycrotroph* bacteria and dry matter of cheese in three periods of ripening at 1 % possibility level. As the *Psycrotroph* bacteria increased, the dry matter of ultrafiltration cheese decreased. There was a significant meaning difference among the periods of ripening with regard to dry matter of ultrafiltration cheese at 1 % possibility level. The average of dry matter was decreased, as time passed. In fact, the average of dry matter in the 6th week was lower than 4th week and lower than 2nd week. *Psycrotroph* bacteria decomposes the proteins and lipids by production of proteolytic and lypolytic enzymes and as a result, the dry matter will be reduced by time. The results of some researches about the reduction of dry matter of cheese during ripening, microbial factors such as microbial quality of raw milk

was effective and make to reduce the dry matter through proteolysis in the period of ripening^{10,11}.

One of the main changes in dry matter of cheese is absorption of water by proteins. If the polar groups are more in the latex of protein, water absorption is more and therefore the dry matter diminished. Proteolysis released the amino acids, peptides, amino and carboxyl groups and therefore increases the solubility and water adsorption of proteins. As the proteolysis was stronger, the dry matter of cheese was reduced due to the more adsorption. The results was given in this study was compliance with the results of some similar studies. A comparison between the dry matter changes in white and ultrafiltration cheese during ripening (2 weeks, 1 month and 45 days) was carried out by t-student. There was a significant difference between the dry matter of white and ultrafiltration cheese in three periods of ripening. As can be seen, the average of dry matter in the ultrafiltration cheese was lower than white cheese and this was reduced by time. The lowest amount was the 6th week. For evaluation of fat changes during ripening, the fat content of white cheese was measured in 2nd, 4th and 6th weeks. The primary studies showed the normalization of data. The fat content of white cheese was lessened after 2nd weeks, as the *Psycrotroph* bacteria increased. The t-student was used for comparison between the changes of fat content of white cheese during ripening. There was a significant different between the fat content of white cheese. The fat content was reduced by time. It means, the fat content of cheese was lessened as the *Psycrotroph* bacteria increased. The effect of *Psycrotroph* bacteria on the fat content of ultrafiltration cheese was measured during ripening in the interval of 2nd, 4th and 6th weeks. At first, the normalization of data was confirmed by Smirnoff and Colmogroff. The results showed that fat content in the 6th week was lower than 4th and less than 2nd week. Some studies about the reduction of fat by multiplying *Psycrotroph* bacteria showed that *Psycrotroph* bacteria was so important during ripening and cause to reduce fat by lypolysis. The lipase enzyme was induced by *Psycrotroph* bacteria was remained after pasteurization and make to severe lypolysis in the dairy products such as butter and cheese. Also another study showed that in cheddar cheese which has 10^7 cfu/mL *Psycrotroph* bacteria, the cheese will be tender after 4 months. It is known fact that the improper aroma was induced in the raw milk which had 5×10^5 cfu/mL *Psycrotroph* bacteria before heat treatment, although this aroma was not present in the raw milk. Thus, improper aroma was caused by direct lipase action on fat of raw milk. In fact by some process such as stirring the fat globules were damaged and natural milk lipases were better act and lypolysis occurred. The t-student was used for comparison between the changes of fat content of white cheese and ultrafiltration cheese during ripening. The average data showed that the reduction of fat in ultrafiltration cheese was more than white cheese and as time progress, the fat lessened more.

The coefficient of regression for fat in the 2nd week of white cheese was -0.114 and it means by multiplying 1000000 *Psycrotroph* bacteria population, the cheese

fat was lowered to 0.114 %. But for ultrafiltration cheese, this data was 0.189 % (this comparison between the 4th and 6th weeks of two type of cheese was conducted).

The effect of *Psycrotroph* bacteria on reduction of fat during ripening in the ultrafiltration cheese was less than the white cheese. The firmness of texture of white and ultrafiltration cheese was evaluated by pressure and sliding experiments. The normalization of data was confirmed by primary method. There was a significant difference between *Psycrotroph* bacteria and texture of cheese in white cheese and in three periods of ripening. As the psycrotroph bacteria was increased, the firmness of cheese was diminished in the 2nd, 4th and 6th weeks and this reduction by time increased. The t-student was used for comparison between the changes of texture during ripening in the three periods of ripening. As time progressed and amount of *Psycrotroph* bacteria was increased, the texture of cheese was softened.

When the amount of *Psycrotroph* bacteria of raw milk which is used for cheese making was less than 8 millions per mL in pressure test and 9 millions per mL in sliding test, there was not significant meaning difference between the firmness of 2nd and 4th week cheese. Whereas, the amount of *Psycrotroph* bacteria reached to 8 millions or more and 9 millions or more in pressure or sliding test, respectively, the firmness of cheese after 4 week was significantly lower than 2nd week. There was a significant loss on the firmness of cheese texture between 4th week and 6th week of cheese ripening which prepared from raw milk had 5 millions *Psycrotroph* bacteria or more (pressure and sliding test) was seen. There was a significant correlation between psycrotroph bacteria and texture of cheese by use of pressure or sliding test during 2nd weeks, 1 month and 45 days in the ultrafiltration cheese. As the cheese aged, the firmness of texture was lessened and till the softness of texture was more sensible after 6 months. Proteolysis released the polar groups such as amino acids, peptides, amino and carboxyl groups and therefore increases the solubility and water adsorption of proteins and how much the proteolysis was stronger, due to the adsorption of water, the dry matter of cheese reduced more and the texture will be softer. According to the previous reports on the cheddar cheese, some factors such as moisture content, pH and casein proteolysis had the great effects on texture and the high population of *Psycrotroph* bacteria of raw milk can cause to increase the moisture of curd and therefore reduce the firmness.

Increasing the proteolytic enzymes was a destruction factor in protein latex and more proetolysis and more dehydration, soluble compound production and penetration of them to whey and reduces the dry matter and finally diminishes the cheese firmness. There was not a significant difference between firmness of white cheese and ultrafiltration cheese in all of experiments^{12,13}.

Conclusion

In general, the firmness of ultrafiltration cheese was significantly softer than white cheese in all the periods. The results showed that the texture of ultrafiltration and white cheese were softer as time progress and this reduction

of firmness was more in the ultrafiltration cheese until in the samples which had high *Psycrotroph* bacteria after 6th week, the texture of cheese so soft and completely destroyed. By multiplying *Psycrotroph* bacteria, the texture of cheese will be softener. The coefficient of regression showed that the texture of ultrafiltration cheese is more softener than white cheese. Any of these coefficients showed that by multiplying 1000000 *Psycrotroph* bacteria the texture of cheese will be softener¹⁴.

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