### The Relationship Between Some Physico-Chemical, Microbiological Characteristics and Electrical Conductivity of Milk Stored at Different Temperature

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In this research; electrical conductivity ( EC ), pH, lactic acid and total bacteria content of milk were determined. During the storage period at two different temperatures, pH decreased, but lactic acid content and total bacteria increased. The relationship between EC values and these parameters of milk were also investigated. The relation between EC and pH, EC and lactic acid, EC and total bacteria were determined statistically significant (P<0.01). According to the analysis results, a possitive correlation among total bacteria content, lactic acid, and EC values was found, except pH. The results also showed that the electric conductivity can be used for the criteria of milk quality. The presence of lactic acid and pH in milk is important for its quality. Nevertheless, the presence of microorganisms in milk is of great importance for safety of public health.

Keywords: Milk, electrical conductivity, total bacteria, lactic acid, pH

### Farklı Sıcaklıklarda Depolanan Sütlerin Bazı Fiziko-Kimyasal ve Mikrobiyolojik Özellikleri ile Elekteriksel İletkenlik Arasındaki İlişki

Bu çalışma da, iki farklı depolama sıcaklığında bekletilen sütlerin elektriksel iletkenliği (Eİ), laktik asit, pH ve toplam bakteri içeriği arasındaki ilişki belirlenmiştir. Yapılan çalışma sonucunda farklı sıcaklıkta depolanan sütlerin laktik asit ve toplam bakteri içeriği artarken pH değeri ise azalmıştır. Aynı zamanda çalışmada, bu parametrelerin Eİ ile arasındaki ilişki de araştırılmıştır. Elektriksel iletkenlik ile pH, Eİ ile laktik asit, Eİ ile toplam bakteri arasındaki ilişki istatistiksel olarak önemli bulunmuştur (P<0.01). Yapılan iststistiksel değerlendirmeye göre, pH dışında, Eİ ile laktik asit ve toplam bakteri arasında pozitif bir korelasyon tespit edilmiştir. Bu çalışma sonucunda Eİ'nin süt kalite kriteri olarak kullanılabilir olduğunu göstermiştir. Sütün laktik asit ve pH içeri sütün kalitesi için önemli bununla birlikte, mikroorganizma içeriği ise sağlık güvenliği açısından büyük önem taşımaktadır

Anahtar kelimeler: Süt elektriksel iletkenlik, toplam bakteri, laktik asit, pH

#### Introduction

Electrical conductivity measurements have been used extensively in the food industry; for example to detect contaminates in water, and to monitor microbial growth and metabolic activity (Carcia *et al.*, 1995; Curda and Plockova, 1995). The conductivity of milk and dairy products has been studied for more than 40 years to provide values of the fat, water and protein content (Felice *et al.*, 1999; Lawton and Pethig, 1993; Mabrook and Petty, (2002) and to detect mastitis (Nielen *et al.*,1992). Milk has conductive properties because of the existence of charged compounds such as mineral salts. Due to decrease of pH and increase of acidity, minerals of milk are converted from colloidal form to soluble form. (Gelais *et al.*, 1995; Muchetti *et al.*, 1994). The value of EC in milk was increased by the development of various microorganisms and inrease of acidity during the storage period of milk (Sims *et al.*, 1991; Neviani *et al.*, 1992). The average EC of cows' milk is between 5.04 and 5.82 mS cm<sup>-1</sup> (Hamana *et al.*, 1992).

To determine the variety of different microorganisms in food, Thijssen (1991), using "Malthu electrical conductivity method" reported that the EC arose over the limit values depending on the intensity of bacterial metabolism and microorganisms of the genus and species. Proteolytic bacteria counts icreased from  $10^6$  to  $10^8$  CFU mL<sup>-1</sup>, particularly, due to proteolitic activity increased the conductivity in environment (Suhren and Heeschen 1990). To detect bacteria, the impedance systems measure the relative or absolute changes in conductance, capacitance, or impedance, at regular time intervals during the growth of bacteria at a given temperature. The measured electrical signals are then graphically plotted on the ordinate against the incubation times on the abscissa, producing impedance growth curves. Generally, the detection time does not appear reaches the bacteria number until approximately  $10^6$ -  $10^7$  CFU mL<sup>-1</sup> (as determined by the plating method).

The impedance value finally reaches a plateau, where the bacteria has grown to a high concentration of  $10^8$  CFU mL<sup>-1</sup> or greater, and all the resource in the medium have been metabolized to end products (Yang and Bashir, 2008).

The aim of this study is to investigate the relationship among EC, pH, lactic acid and total bacteria counts in milk, depending on different storage periods and temperatures.

### **Material and Methods**

Ninety fresh milk samples collected from healthy cows from Thrace region were transferred in pre- sterilized glass jars of 1000 mL capacity in cold conditions to laboratory and tested for this purpose. The tests were performed at two different temperatures  $(10^{\circ}$ C and 25 °C) with two hours intervals for 8 h storage period.

Titratable acidity was determined as Lactic Acid (LA) (%), using phenolphthalein indicator and 0.1 N NaOH (Anonymous, 2005). Electrical conductivity and pH values of milk samples were determined by using brand electrical conductivity meter (Martini Instruments Code MI806, Serial Number 767252 Romania) which has 0,01- 20,00 mS cm<sup>-1</sup> measurement range and 0,1 mS cm<sup>-1</sup> sensivity. Total bacteria counts of the samples were determined according to the procedure described in Anonymous (1998).

All samples were analyzed in triplicate to verify the statistical significance of all parameters. To compare several groups, analysis of variance (ANOVA) was used. Significant differences between means were determined by Duncan's multiple range tests. The Pearson correlation coefficient (R) and p-value were used to show correlation and their significance. Probability values of P<0.05 and P<0.01 were adopted as the criteria for significant differences (SPSS for Windows, Release 10.01).

### **Results and Discussion**

Changes in physico-chemical and microbiological properties of milk during storage period (SP) at 10 °C and 25°C were given in Table 1. The decrease of pH in milk samples stored at 25°C were higher than the samples stored at 10°C, as it is expected. Electrical conductivity, LA and TB counts increased at both storage temperatures. But increases in these parameters were more clear in the milk samples stored at 25 °C.

Temperature	SP	EC	pН	LA	ТВ	
	( <b>h</b> )	(mS cm <sup>-1</sup> )		(%)	(log CFU mL <sup>-</sup>	
	0	5,497	6,740	0,165	6,794	
	2	5,520	6,730	0,168	7,554	
10°C	4	5,530	6,720	0,172	8,101	
	6	5,571	6,700	0,175	8,572	
	8	5,625	6,680	0,182	8,768	
	Average	5,549	6.710	0,172	7.958	
	0	5,496	6,740	0,165	6,794	
	2	5,547	6,690	0,182	8,583	
25°C	4	5,582	6,570	0,186	8,763	
	6	5,640	6,400	0,198	9,062	
	8	5,779	6,190	0,221	9,153	
	Average	5,609	6,520	0,190	8,471	

Table 1. Physico-chemical and	l microbiological	properties of milk	stored at 10 <sup>6</sup>	$^{\circ}C - 25^{\circ}$	Ϋ́C
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## Effect of storage period on electrical conductivity of milk

conductivity Electrical increased during the SP at two different storage temperatures (Fig. 1). There was a strong relationship between EC and SP both at 10 and 25°C. Increases in EC at 25°C were more than 10°C during the SP. Average EC value was 5,549 mS cm<sup>-1</sup> at 10°C and 5,609 mS cm<sup>-1</sup> at 25°C. Wong (1988) and Peris *et al.*, (1991) found that EC in milk was between 4 and 5 mS cm<sup>-1</sup> at 25° C. The electrical conductivity values of the cow's milk were determined between 5,04 and 5,82 mS cm<sup>-1</sup> by Hamana et al., (1992). These results were similar with ours. The reasons of the differences between our results and literature may be related with different compositional and microbiological characteristics of the milk

samples. According to analysis of varians, the relaltion between EC values in milk, SP and temperature were found statistically significant (P<0.05). At two different temperatures depending on the SP, there was a statistically significant correlation within the changes of EC in milk (P<0.01).Correlation between the EC and the increase of milk storage tempreture was detected by Renda et al. (1975) The equations showed the relationship between EC and SP at 10°C and 25°C are given below. It was also seen from the equations that a rather high correlation was found between EC values and SP at two different temperatures.

 $EC_{10} = 0,0017 SP^2 + 0,0021 SP + 5,5005 (R^2 = 0,99)$ 

 $EC_{25} = 0,0036 SP^2 + 0,0045 SP + 5,5054 (R^2 = 0,98)$ 



Figure 1. Changes at the EC of milk during storage period at  $10 - 25^{\circ}$ C.

## The relationship between EC and pH during storage period at 10 - 25 °C

The pH decreased while EC increased during SP at 10 and 25 °C. Decreased in pH at 10 °C were slower than at 25 °C (Figure 2). Due to the microbial growth rate, the decrease in pH in milk at low temperatures is expected slowly. Increase in temperature in milk together with the storage period causes increases in phosphate ion content which has a lower molar conductivity and pH decreased rapidly (Muchetti *et al.*, 1994). In the homolactic fermentation which arises during the storage period, at the same time, by means of increasing of soluble salt fractions in colloidal salts, depending on pH, EC increases (Lanzanova *et al.* 1993).

As a result of reduction in pH, calcium and phosphorus bonds were released from the structure of colloidal milk protein. Colloidal calcium and phosphorus between pH 4,9-5,0 is completely soluble and this makes EC increase. Depending on the storage period at each temperature, pH changes were significant (P<0.05). Correlation was determined between pH and EC (P<0.01). Relationship between EC and pH values were also shown at the Figure 2. The correlation equations between EC and pH at 10 and 25°C are given below:

$$pH_{10} = -0,4744 \ EC_{10} + 9,3461 \ (R^2 = 0,9859)$$
  
 $pH_{25} = -2,0459 \ EC_{25} + 17,993 \ (R^2 = 0,9729)$ 



Figure 2 The relationship between EC and pH during storage period at 10 - 25°C

# The relationship between EC and LA contents of the milk samples during storage period at $10 - 25^{\circ}C$

Lactic acid content increased during SP at both storage temperatures (10 °C -  $25^{\circ}$ C). There was a positive relationship between LA contents and EC values (Fig. 3). This positive correlation was also determined by Lanzanova *et al.*, (1992) and Neviani (1990). As can be expected, increase in LA content at 10°C was less than 25°C. EC

increased with increase in lactate ions (Latrille *et al.*, 1992). According to variance analysis, depending on temperature and storage period of milk, changes occured in LA content (P<0.05) was found significant. There is a positive correlation between the development of EC and LA in milk (P<0.01). The correlation equations between LA and EC values at 10 and  $25^{\circ}$ C are given below:

 $LA_{10} = 0,1286 \ EC_{10} - 0,5409; \ (R^2 = 0,97)$ 

 $LA_{25} = 0,1896 EC_{25} - 0,8728; (R^2 = 0,98)$ 



Figure 3 The relationship between EC and LA content during storage period at 10- 25°C

# The relationship between EC and the TB count during storage period at 10 - $25^{\circ}C$

Total bacteria content increased during the SP at both storage temperatures (10 and 25°C). Increases in TB count at 10°C were less than 25°C (Figure 4). Bacterial content of milk with enhanced metabolic activity due to the increased EC of milk is increased and at the same time the change was determined by Gelais et al. (1995) and Muchetti et al. (1994). The changes in bacterial contamination (time t:0...28 hours) were determined by Daunoras (2006). The correlation between TB counts and the EC

change was indicated by (Daunoras, 2007). Increase of storage period and temperature was found significant in the development of TB content of milk (P<0.01). Statistically significant correlation was also determined. According to the correlation coefficients, a high relationship was found between TB counts and EC values.

The relationship equations between TB and EC values at 10 and 25°C are given below:

 $TB_{10} = -186 EC_{10}^{2} + 2083, 6 EC_{10} - 5826, 4; (R^{2} = 0,98)$ 

 $TB_{25} = -63,123 EC_{25}^{2} + 719 EC_{25} - 2037,9; (R^{2}) = 0,91)$ 



Figure 4 The relationship between EC and TB counts during storage period at 10 - 25 °C

### Conclusion

In order to follow the changes of some physical and chemical properties in milk after milking, EC measurements can be thought an alternative way. Temperature and storage period are important factors affecting EC and some physico- chemical properties of milk. In this study, depending on storage period and temperature, pH of milk decreased and EC increased because of the production of LA by fermentation. But during the storage period, other determined parameters such as LA content and TB counts increased except pH. It is possible to estimate some physical, chemical and microbiological properties of milk by using the relationships between EC and these properties of milk even without analysis at a short time. We found strong and significant releationships between EC and these properties in this study. By the help of these results, the devices which measure these properties can be developed and used in milk collection centers.

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