

# ARAŞTIRMA MAKALESİ

# Relationships Between Color, Somatic Cell Count and Physicochemical Properties of Raw Milk Collected from Dairy Cattle Farms in Erzurum Province

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#### **Abstract**

In this study, 23 milk samples were taken from different farms in Erzurum province and some physical and chemical properties and conformity to standards were analyzed. The study also aimed to determine possible correlations between the physicochemical properties of milk samples and various color parameters, and to fill the gap in the literature. In the study, the mean values for total dry matter (DM, %), fat (%), non-fat dry matter (NFDM, %), pH, protein (%), fat (%) and somatic cell count (log SCC) of the raw milk samples were found to be 12.22, 8.73, 6.44, 2.93, 3.49 and 5.404, respectively. The average L\*, a\*, b\*, C\* and  $\Delta E$  values of the raw milk samples were determined as 80.62, -56.71, 4.77, 56.92 and 60.141, respectively. The results of statistical analysis showed that there were significant correlations between the color parameters and some physicochemical values. Significant correlations were found between the amount of non-fat dry matter in raw milk samples and the L\*a\*, b\* and  $\Delta E$  values of the samples (r= -0.674, -0.514). The correlations between the fat content of the raw milk samples and the L\*a\*,  $b^*$  and  $\Delta E$  values of the samples were found to be insignificant.

# Erzurum İli Süt Sığırcılığı İşletmelerinden Toplanan Çiğ Sütlerde Renk, Somatik Hücre Sayısı ve Fizikokimyasal Özellikler Arasındaki İlişkiler

#### Özet

Bu çalışmada Erzurum ilinde bulunan süt çiftliklerinden 23 adet çiğ süt örneği alınmış ve örneklerin bazı fiziksel ve kimyasal özellikleri ile standartlara uygunluğu araştırılmıştır. Araştırmada ayrıca süt örneklerinin fizikokimyasal özellikleri ile farklı renk parametreleri arasındaki olası korelasyonları belirlemek ve literatürdeki boşluğu doldurmak amaçlanmıştır. Araştırmada, çiğ süt örneklerinin ortalama kuru madde (%), yağsız kuru madde (%), pH, protein (%), yağ (%) ve somatik hücre sayıları (log SHS) değerleri sırasıyla 12.22, 8.73, 6.44, 2.93, 3.49 ve 5.404 olarak bulunmuştur. Çiğ süt örneklerinin ortalama L\* a\* b\*, C\* ve ΔE değerleri sırasıyla 80.62, -56.71, 4.77, 56.92 ve 60.141 olarak bulunmuştur. İstatistiksel analiz sonuçları renk parametreleri ile bazı fizikokimyasal değerler arasında önemli korelasyonlar olduğunu göstermiştir. Çiğ süt örneklerinin yağsız kurumadde miktarı ile örneklerin L\*a\*, b\* ve ΔE değerleri arasında önemli korelasyonların ise önemsiz olduğu belirlenmiştir.

#### Introduction

Milk can be defined as the miraculous food secreted by the mammary glands of female mammals to nourish and raise their newborns. It contains all the elements that meet the nutritional needs of the offspring in the required quantity, has a whitish color and a unique taste and aroma. Milk is the baby's first food after birth and can fully meet its needs during this time (Patton, 2017). Milk also plays an important role in adult nutrition, as it contains proteins, lactose, vitamins and milk fat (Murphy et al., 2017; Lambrini et al., 2021). Additionally, milk contains lysozyme, lactoperoxidase lactoferrin, system, conjugated linoleic acid (CLA) and various peptides. These compounds are biologically active and act as natural antimicrobials, angiotensin converting enzyme (ACE) inhibitors, antidiabetic agents, anticarcinogenic agents and antiobesity agents. They also have important effects on human nutrition, health and metabolism. (Baran and Adıgüzel, 2020). The relationship between the consumption of milk or dairy products and possible health benefits has been studied at various research (Thorning et al., 2016).

The main constituents of raw milk are relatively constant within a certain limit. Changes give us very essential information about milk quality and animal health (Sengül et al., 2021). The physicochemical and microbiological quality of raw milk is of crucial importance at all stages of dairy products from production to consumption. The quality of raw milk is important from an economic perspective because it affects the price of milk. (Baran and Adıgüzel, 2020). The literature review on the color parameters of raw milk shows that there is not enough research in this area. Although there are many studies on the physicochemical properties and microbiological quality of milk, the studies on the color parameters are insufficient. This study aims to fill this

gap in the literature by providing a comprehensive framework for raw milk and by examining the possible relationship between the physicochemical properties and the color parameters.

### **Materials and Methods**

#### Materials

The study material consisted of a total 23 raw milk samples collected from dairy farms in Erzurum province. The raw milk samples were filled into sterile glass bottles with a capacity of approximately 300 mL, and transported under cold chain conditions to the laboratories of the Food Department of the Vocational School of Technical Sciences, and subjected to the specified analyses.

### Methods

# **Color Analysis**

The color parameters of the raw milk samples were determined using a **PCE** XXM-20 colorimeter (PCE Instrument, Germany), and the results were expressed using the CIE LAB color space system under LED illumination. In this system, the color is expressed by three coordinates: L\*a\*b\*. The L\* value is displayed on a vertical axis ranging from zero to 100. Zero represent black, which means no light transmission, while 100 is white, which is lightness. The remaining parameters a\* and b\*, are represented as a point within the color space, that is on a coloured plane on which the color flow from red (+a\*) to yellow (+b\*) and from green (-a\*) to blue (-b\*) (Chudy et al., 2020). The line segment obtained by connecting the centre of the vertical axis to a point in space is defined as chroma (C\*). The higher the C\* value, the stronger and clearer the color. The angle formed between the axis +a\* and C\* is called the color tone and is indicated by the letter ho. The color difference ( $\Delta E$ ), the chroma (C\*), the color intensity (h°), the white index (WI) and the yellowness index (YI)

were also calculated using the following equations.  $\Delta E$  was used to define the difference between the color of each sample and the ideal whiteness pattern.

Four different measurements were taken on the milk samples and the average of these 5 values formed the color values of the raw milk (Milovanovic et al., 2020).

$$\Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2}$$
 (1)

$$C^* = \sqrt{((\Delta a^*)^2 + (\Delta b^*)^2}$$
 (2)

$$YI = 142.86. \, b^*. \, L^{*-1}$$
 (3)

$$h = \tan^{-1}(a^*/h^*) \tag{4}$$

$$WI = \sqrt{100 - ((100 - L^*)^2 + a^{*2} + b^{*2})}$$
 (5)

L\* stands for lightness, a\* for the redness b\* for the yellowing, and  $\Delta a = a_0 - a_i$ ,  $\Delta b = b_0 - b_i$ , and  $\Delta L = L_0 - L_i$ . In the study, we have the color parameters of the white standard (L<sub>0</sub>: 100, a<sub>0</sub>:0, b<sub>0</sub>:0)

# Physicochemical analyses

The dry matter (DM) of the raw milk samples was determined by drying the samples at 103 °C until a constant weight obtained. The fat content was determined using the Gerber method according to AOAC (1995). The analysis of lactose, protein, density and ash content of the milk samples was conducted using a milk analyzer (LactoScan MMC, Germany) and the somatic cell count was performed with a DeLeval somatic cell counter (DeLeval DCC, Sweden). The pH value of the raw milk samples was measured using a pH meter (Hanna, Portugal).

### **Statistical Analysis**

All analyses were conducted using SPSS 20.0 for Windows. Descriptive statistics including mean, standard error, minimum and maximum values, variance, and coefficient of variation were calculated for the data obtained in the study. Pearson correlation analysis between pairs of response variables was

performed to determine the significance of the linear relationship.

# **Results and Discussion**

# Color analysis

Objects partially or completely reflect the light energy that fall on them. The reflected waves that reach the observer are recognized by the eyes, and after the signal is transmitted to the brain, a color impression is created (Chudy et al.. 2020). Milk has a white appearance, which results from its physical and chemical structure. The color of raw milk is due to the reflection of light from dispersed fat globule, calcium caseinate and phosphate. Additionally, milk contains two classes of pigments: water-soluble and fat-soluble (Graulet, 2010). The color of raw milk is due to the effects of calcium caseinate, which does not transmit light, and milk fat, which reflects light. Carotenoids (β-carotene, retinol, xanthophylls) and riboflavin (vitamin B2), a water-souble vitamin and contains greenish pigment substances, thus

influence the color, and milk from animals fed with green fodder tends to be yellowish. On the other hand, the type of animal, the breed, the season and the feed it eats also have certain effects on the color the milk. addition. In microorganisms and the diseases they cause can change the color of the milk (Kaygısız et al., 2019; Chudy et al., 2020). In general, consumers' desire for food is initially influenced by appearance.

Appearance, including color, plays an important role in food selection and preference. Beyond impact on overall food acceptance, the possible relationship between color parameters and other food characteristics should not be overlooked. It is known that the color of food is related to taste or quality characteristics (Jaros and Rohm, 2001). Table 1 gives an overview of the color parameters of the raw milk samples.

Table 1. Physicochemical, textural, and color properties of y raw milk samples

	N	Mean	Std. Error	Variance	Minimum	Maximum	Coefficient of variation	
Physicochemica								
DM (%)	23	12.22	0.103	0.246	11.12	12.92	4.05	
NFDM (%)	23	8.73	0.104	0.252	7.81	9.74	5.75	
Fat (%)	23	3.49	0.963	0.214	2.17	4.30	13.25	
Protein (%)	23	2.93	0.036	0.029	2.62	3.29	5.84	
Lactose (%)	23	4.16	0.051	0.060	3.72	4.69	5.89	
Ash (%)	23	0.73	0.089	0.002	0.65	0.81	5.93	
Density	23	1.03	0.002	0.001	1.03	1.031	0.12	
рН	23	6.64	0.042	0.042	6.00	6.87	3.09	
SCC (Log)	23	5.40	0.071	0.116	4.54	5.99	6.29	
Color propertie	es							
L*	23	80.62	0.434	4.349	77.41	85.53	2.59	
a*	23	-56.71	1.035	24.767	-67.46	<b>-</b> 44.20	-8.78	
b*	23	4.77	0.171	0.674	2.55	6.05	17.22	
C*	23	56.92	1.028	24.339	44.48	67.58	8.67	
ΔΕ	23	60.14	1.093	27.486 46.90		71.26	8.72	
Н	23	-0.09	0.003	0.001	-0.12	-0.04	-21.99	
WI	23	39.86	1.093	27.486	28.74	53.10	13.15	
YI	23	8.44	0.306	2.167	4.50	10.88	17.45	

The L\* values measured for lightness showed a variation range of 77.41-85.53, the mean value was found to be 80.62. The a\* values of the milk samples, which correspond to the position on the red-green axis, varied between -44.20 and - 67.46, the average a\* value was -56.71. The b\* values varied between 2.55 and 6.05, the average value was determined to be 4.77. As can be seen from the results, the color of the raw milk samples lies in the second quadrant of the coordinate system with the axes a\* and b\*, which indicates a yellowish color. Examination of the color properties of the raw milk samples shows that relative color stability is achieved. The b\* values confirm the presence of a yellowish color in the milk samples. Variations in the a\* and b\* values can be attributed to differences in the feedstuffs provided to the animals. The  $C^*$  and  $\Delta E$  values were high and were close to each other for all raw milk samples. These results show that all milk samples have an attractive degree of whiteness. The low coefficient of variation also confirms these results. Gürbüz et al., (2023) stated that color affects the quality, freshness, expected taste, commercial value and consumer acceptance of dairy products.

Table 2 summarizes the Pearson correlation coefficients (r) and probabilities (p) between the response variables of the milk samples. Negative correlations (r=-0.549, p=0.0007) and (r=-0.668, p=0.000) were found between the L\* value and DM and between L\* value and the amount of protein and lactose, respectively. This result shows that the

brightness decreases as the total DM content of milk increases. Additionally, negative correlations (r=-0.513, p=0.012) and (r=-0.515, p=0.001) were also found between the a\* value and the amount of protein and the a\*value and the amount of lactose respectively. These results indicate that the yellow coloration intensifies with increasing DM content. However, the yellowish color is usually more pronounced in high-fat dairy products such as butter and full-fat cheese, as the carotenoid pigments are fat-soluble, and therefore dependent on fat concentration. Positive correlations (r=0.516, p=0.012) and (r=0.518, p=0.011) were found between the C\* value and the protein and lactose content. These results suggest that the C\* value should also correlates with the total DM content; however, a significant correlation was found only between the C\* value and the NFDM. The higher the C\* value, indicates a stronger and more visible the color. The C\* value is centered at 0, and brighter tones increase as values move away from the center. The negative correlation between milk fat content and the C\* value weakened the correlation with the total DM. The storage of raw milk under cold conditions could have caused this result due to the turbidity of the milk fat. Generally, there are few studies on the parameters L\*, a\*, b\* parameters of raw milk. Scibisz et al., (2019) reported that C\* values were related to acidity in yogurts. However, in the present study, the correlation between the C\* value and pH was found to be low and not statistically significant (Table 2) (r=-0.199, p=0.363).

 Table 2. Pearson correlation coefficients and probabilities of raw milk samples between physiochemical and color properties

	NFDM	Fat	Protein	Lactose	Ash	Density	Hd	Log	L*	a*	b*	C*	ΔΕ	h	WI	YI
DM (%)	0.569	0.454	0.559	0.550	0.586	0.300	0.324	0.188	-0.549	-0.295	0.295	0.300	0.338	-0.025	-0.338	0.379
p-value	0.005	0.030	0.006	0.007	0.003	0.165	0.131	0.390	0.007	0.171	0.172	0.165	0.115	0.911	0.115	0.075
NFDM (%)	1.000	-0.475	0.999	0.999	0.998	0.481	0.105	-0.071	-0.674	-0.510	0.075	0.514	0.544	0.164	-0.544	0.176
p-value		0.022	0.000	0.000	0.000	0.020	0.633	0.746	0.000	0.013	0.735	0.012	0.007	0.454	0.007	0.421
Fat (%)		1.000	-0.485	-0.494	-0.455	-0.200	0.233	0.279	0.143	0.237	0.234	-0.236	-0.228	-0.204	0.228	0.214
p-value			0.019	0.017	0.029	0.360	0.284	0.197	0.515	0.277	0.282	0.279	0.295	0.349	0.295	0.326
Protein (%)			1.000	1.000	0.997	0.497	0.095	-0.082	-0.668	-0.513	0.069	0.516	0.546	0.168	-0.546	0.170
p-value				0.000	0.000	0.016	0.666	0.710	0.000	0.012	0.754	0.012	0.007	0.443	0.007	0.439
Lactose (%)				1.000	0.996	0.506	0.087	-0.090	-0.665	-0.515	0.066	0.518	0.547	0.170	-0.547	0.166
p-value					0.000	0.014	0.694	0.684	0.001	0.012	0.763	0.011	0.007	0.438	0.007	0.448
Ash (%)					1.000	0.464	0.117	-0.080	-0.686	-0.507	0.096	0.510	0.543	0.148	-0.543	0.200
p-value						0.026	0.594	0.716	0.000	0.014	0.662	0.013	0.007	0.500	0.007	0.361
Density						1.000	-0.021	-0.303	-0.061	-0.144	0.166	0.146	0.138	-0.069	-0.138	0.170
p-value							0.925	0.159	0.781	0.513	0.450	0.506	0.531	0.756	0.531	0.437
pH							1.000	-0.068	-0.004	0.199	0.061	-0.199	-0.177	-0.132	0.177	0.064
p-value								0.759	0.985	0.362	0.782	0.363	0.420	0.549	0.420	0.773
SCC (Log)								1.000	-0.035	-0.116	-0.226	0.113	0.106	0.278	-0.106	-0.213
p-value									0.874	0.598	0.299	0.608	0.631	0.200	0.631	0.328
L*									1.000	0.826	0.009	-0.830	-0.868	-0.393	0.868	-0.143
p-value										0.000	0.969	0.000	0.000	0.064	0.000	0.516
a*										1.000	0.371	-1.000	-0.997	-0.731	0.997	0.243
p-value											0.081	0.000	0.000	0.000	0.000	0.263
b* ,											1.000	-0.361	-0.322	-0.898	0.322	0.988
p-value												0.091	0.134	0.000	0.134	0.000
C*												1.000	0.997	0.723	-0.997	-0.232
p-value													0.000	0.000	0.000	0.286
$\Delta E$													1.000	0.695	-1.000	-0.189
p-value														0.000	0.000	0.389
h														1.000	-0.695	-0.828
p-value															0.000	0.000
WI															1.000	0.189
p-value																0.389

L\*: lightness, scale 0 (black) to 100 (white); a\*: redness, + a\* (red) to – a\* (green); b\*: yellowness, + b\* (yellow) to – b\* (blue); ΔΕ: color difference; C\*: chroma; YI: yellowing index

# Physicochemical properties

The color of the milk can be an indicator of some physicochemical changes especially during the storage period (Milovanoviç et al., 2020). Basic physicochemical analyzes of raw milk samples were performed to obtain data that can support the color parameters. The results of the physicochemical analyzes are shown in Table 1. The dry matter content of the raw milk samples ranged from 11.12% to 12.92%, with a mean value of 12.22%. The DM values determined in the study are similar to those reported by Açık and Özdemir (2022) and are consistent with the average dry matter content of milk samples collected in Erzurum (13.82%). The table 2 shows that the degree of whiteness in raw milk is influenced by dry matter components such as proteins and lactose content. Additionally, there was a high significant correlation between the NFDM and L\* values of milk samples (r=-0.674, p=0.0001). The mean fat content of the milk samples was determined to be 3.49%, with the minimum and maximum values ranging from 2.17% to 4.30% (Table 1). The coefficient of variation for fat content was found to be quite high. This shows that the milk samples are not homogeneous in terms of fat content. The fat content determined in the study is compatible with the findings of Akın et al., (2016). The correlations between fat content of the milk samples and all color parameters were found to be insignificant (Table 2). The protein content of the raw milk samples ranged from 2.62% to 3.29%, with a mean value of 2.932%. Since low protein content in milk leads to quality problems in dairy products, high protein content is a desirable characteristic. The whiteness in milk is caused by the presence of colloidal particles such as casein micelles, which can scatter light in the visible spectrum. According to the Turkish Food Codex Communiqué on Raw and Heat-Processed Drinking Milk (Communiqué No: 2009) raw cow's milk must contain at least 2.80% protein. The protein values determined in the study (2.93%) were lower than the average protein content of the raw milks from Erzurum reported by Diler and Baran (2014) and Açık and Özdemir (2022) (3.11%- 3.26%, respectively). Significant correlations were found between the protein content and the color parameters L\*  $a^*$ ,  $C^*$  and  $\Delta E$  value. The lactose content of the raw milk samples ranged from 3.72% to 4.69%, with a mean value of The lactose content values 4.161%. determined in the study are similar to those of Açık and Özdemir (2022) and are compatible with the average lactose amount of milk samples collected in (4.97%).Also. significant Erzurum correlations were found between the lactose content and the color parameters of L\* a\*, C\* and  $\Delta E$ .

The pH of the raw milk samples varied between 6.0 and 6.87, the mean value was determined to be 6.64. A pH below 6.4 indicates high microbial growth, which can compromise the quality of raw milk, and suggest that may coagulate when heated. The coefficient of variation of pH values was found to be high. However, no significant positive or negative correlations were found between pH and color parameters L\* a\*, C\* and  $\Delta E$ . The mean pH value determined in the study were similar mean pH value (6.64) of the raw milks collected from Kastamonu of Özdemir and Kahyaoğlu (2020). Similar results were found by Tuncer et al., (2016) and Gayretli (2013).

The SCC of the raw milk samples ranged from 4.54 log cells/ml to 5.99 log cells/ml with a mean value of 5.404 log cells/ml. Milk from dairy cows contains somatic cells (SC). SCC is the main indicator of milk quality. SCC used as a diagnostic indicator of mastitis should be less than 100.000 (5 log) cells/ml in milk from a healthy animal. SCC is as indicator of raw milk quality and is used to monitor the incidence of mastitis in cow and is a

general indicator of the hygienic conditions of milk production on farms (Hunt et al., 2013; Kaskous, 2021). It was found that 18 (78%) of the analyzed raw milk samples were above the limit value determined for SCC (<5log cells/ml). The mean pH value determined in the study were similar mean pH value (6.64) of the raw milks collected in Erzurum by Acık and Özdemir (2022). Similar results were found by Tuncer et al., (2016) and Gayretli (2013). Eighteen (78%) of the analyzed raw milk samples were found to contain SC values above the established limits for SCC ( $\leq$  5 log cells/ml). No significant positive negative correlations were found between SCC and any of the color parameters.

#### **Conclusions**

In Turkey, the number of large farms producing milk and dairy products is increasing. However, there are still many small family farms that may not prioritize milk quality and hygiene regulations. In this study, the physicochemical instrumental color parameters of raw milk samples from different dairy farms in Erzurum province and the correlations between the results were investigated. It investigated whether also properties of the milk samples were sufficient considering the Turkish Food Codex Communiqué on Raw and Heat-Processed Drinking Milk. The evaluation of results revealed strong and significant correlations between the color parameters of the raw milk and the dry matter and nonfat dry matter content of the samples. Significant correlations were also observed between the color parameters of the raw milk and the protein and lactose content of the samples. However, the correlations between the color parameters of the raw milk and the fat content and pH of the milk proved to be non-significant. This is an unexpected, surprising result and shows that there is still a lack of information on the correlations between color analysis and milk quality. The correlations between the

SCC values and the color parameters of the milk samples were found to be statistically insignificant.

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