



An Investigation on Some Compounds Effecting Aroma and Flavour of Strained Yoghurt Produced from Goat Milk

Ebru ŞENEL¹ Zahide KOCABAŞ^{2*} Fatma Şebnem ÖZTEKİN¹ Metin ATAMER¹

Received: December 12, 2008

Accepted: December 29, 2009

Abstract: In this study, acetaldehyde, acetone, pH, titratable acidity, lactic acid, tyrosine and free fatty acids, namely butyric acid (C4), caproic acid(C6), caprylic acid (C8) and capric acid (C10) were taken into consideration affecting the variation in aroma and flavour scores (AFS) of strained yoghurt during the storage period of 60 days. AFS was negatively correlated with acetone ($r=-0.832$, $P<0.01$) and positively correlated with C4 ($r=0.604$, $P<0.05$), C8 ($r=0.638$, $P<0.01$) and C10($r=0.659$, $P<0.01$). The results of regression analysis verified that the independent compounds explain the 91.6% of variation in AFS during the storage period, but not statistically significant ($P>0.05$). Stepwise regression procedure was performed to select the statistically significant compounds affecting AFS. The Stepwise procedure verified that acetaldehyde, acetone and lactic acid should be included in the regression equation ($r^2=0.868$, $P<0.01$). To explore the individual direct effect of these compounds, Path analysis was applied. The results of Path analysis showed that 65.6 % of variation in AFS could be directly explained by acetone ($P<0.01$), which the highest effect on the variation of AFS. The results of this study illustrated that although an increment in AAL compound causes an improvement in aroma and flavour, acetone and lactic acid were responsible for defective aroma and flavour of strained yoghurt.

Key Words: Strained yoghurt, carbonyl compounds, acidity, free fatty acids, regression analysis, Stepwise regression

Keçi Sütünden Yapılan Süzme Yoğurtların Koku ve Lezzetine Etki Eden Bileşenlerin Araştırılması

Öz: Bu çalışmada, asetaldehit, aseton, pH, titrasyon asitliği, laktik asit, tirozin ile butirik asit (C4), kaproik asit (C6), kaprilik (C8) ve kaprik (C10) serbest yağ asitlerinin süzme yoğurdun 60 gün depolanma periyodu boyunca koku ve lezzet puanlarındaki (AFS) değişim üzerine etkisi araştırılmıştır. Hesaplanan korelasyon katsayıları koku ve tat puanları ile aseton arasında ters bir ilişki ($r=-0.832$, $P<0.01$) olmasına karşın C4 ($r=0.604$, $P<0.05$), C8 ($r=0.638$, $P<0.01$) ve C10 ($r=0.659$, $P<0.01$) serbest yağ asitleri ile pozitif ilişkili olduğunu göstermiştir. AFS ile diğer bağımsız bileşenler için uygulanan çoklu regresyon analizi, bağımsız değişkenlerin AFS'deki varyasyonun %91.6'sını açıkladığını fakat istatistik olarak önemli olmadığını göstermiştir. Daha sonra Stepwise regresyon metodu kullanılarak hangi bağımsız değişkenlerin AFS'deki değişim üzerine etkisinin önemli olduğu araştırılmıştır. Stepwise regresyon analizinden elde edilen sonuçlar, AFS'deki varyasyonu açıklamak üzere oluşturulacak regresyon denkleminde asetaldehit, aseton ve laktik asit bileşenlerinin yeterli olacağını göstermiştir ($r^2=0.868$, $P<0.01$). Regresyon denkleminde yer alan bileşenlerin AFS üzerine doğrudan etkisini araştırmak üzere Path analizi uygulanmıştır. Path analizinden elde edilen sonuçlar AFS'deki varyasyonun %65.6'sının aseton ile açıklandığını ($P<0.01$) ve bu bileşenin AFS'deki varyasyon üzerine en etkili bileşen olduğunu göstermiştir. Bu çalışmanın sonuçları asetaldehit düzeyindeki artışın süzme yoğurdun arzu edilen koku ve tat gelişimini sağlamasına karşın aseton ve laktik asit miktarlarındaki artışlar süzme yoğurtta istenmeyen koku ve tat oluşumuna sebep olduğunu göstermiştir.

Anahtar Kelimeler: Süzme yoğurt, karbonil bileşenleri, asitlik, serbest yağ asitleri, regresyon analizi, path analizi

Introduction

Strained yoghurt, also known as Torba or Süzme yoghurt, is a dairy product that is native to Turkey and Middle East. It is produced from set yoghurt by filling into special cloth bags and removing yoghurt serum. This process extends its shelf life because of decreasing water activity.

Tendency to consume dairy products from goat milk has been recently increasing all over the world different properties of goat milk from other milks. Goat milk has higher concentration of short and medium chain fatty acids and lipoprotein lipase associated with fat phase comparing with cow milk (Agnihotri and

¹ Ankara University, Faculty of Agriculture, Department of Dairy Technology, 06110 Diskapi, Ankara, Turkey

² Ankara University, Faculty of Agriculture, Department of Animal Science, 06110 Diskapi, Ankara, Turkey

Prasad 1993, Alichanidis and Polyachoniadou 1995). It is thought that these properties have determinative effect on development of the characteristic aroma and flavour of dairy products from goat milk.

Lactic acid, being found mainly in serum phase, is also an essential compound influencing the characteristics taste (sharp and acidic) of yoghurt. Although a large amount of serum separated from yoghurt, the lactic acid content in strained yoghurt increases because it becomes more concentrate in product (Atamer et al. 1993).

Tyrosine value is considered amount of total amino acids liberated as tyrosine equivalent by proteolytic activity of starter culture. Generally some flavour defects such as bitterness is usually attributed to the formation of degradation products from proteins. When the degradation products reached up to a threshold value, the flavour of yoghurt can be adversely influenced. (Asperger 1977, Atamer et al. 1993)

Acetaldehyde is one of the major carbonyl compounds influencing characteristic aroma and flavour of yoghurts. The formation of acetaldehyde is governed by many factors. One of these factors is the species of mammals. The characteristic flavour-aroma of set type yoghurt from cow milk becomes more evident at a certain level of acetaldehyde. In literature, there is a wide range of optimum acetaldehyde level suggested for the characteristic aroma-flavour of set type yoghurt from cow milk. For example, Görner et al. (1973) stated that a level of 10–20 mg kg⁻¹ acetaldehyde was required for the optimum flavour-aroma, while Rasic and Kurman (1978) reported that 21–41 mg kg⁻¹ of acetaldehyde was necessary for the typical flavour and aroma in yoghurt. In contrast to this, the level of acetaldehyde in set yoghurt from goat milk was obtained as 4.5–5.5 mg kg⁻¹ by Abrahamsen et al. (1978), 6.2–8.9 mg kg⁻¹ by Rysstand and Abrahamsen (1987), an average of 9.7 mg kg⁻¹ by Yaygın and Mehanna (1988), 1.5–9.0 mg kg⁻¹ by Wojtowski et al. (1995) and 3.19–3.96 mg kg⁻¹ by Karademir et al. (2001). The low level of acetaldehyde could be attributed to the relatively high concentration of glycine in goat milk, which can inhibit enzymes involved in the conversion of threonin to acetaldehyde (Peaker et al. 1981, Tamime and Robinson 1999).

Other compounds such as acetone, butanone–2, diacetyl also contribute to aroma and flavour. The levels of diacetyl content in goat yoghurt have been determined as trace or rather low level (~1 mg kg⁻¹) (Rysstand and Abrahamsen 1987, Yaygın and Mehanna 1988).

Although, there are large numbers of studies on the composition of carbonyl compounds and free fatty acids, and their changes during storage in set type yoghurts, the effect of these compounds on change of aroma and flavour in strained yoghurt has not been clearly determined yet.

The primary aim of this study was to determine the individual direct effect of several compounds on the variation in the aroma and flavour scores of strained yoghurt from goat milk during the storage period. This study also targeted to investigate the relationship between aroma and flavour score, and other compounds of yoghurt. It was also investigated whether all compounds should have been included in the constructed regression equation. This is because some compounds affecting the aroma and flavour scores would be judged unnecessary and could be removed from the regression equation. In this study, acetaldehyde (mg kg⁻¹), acetone (mg kg⁻¹), pH, titratable acidity (°SH), lactic acid (g 100 g⁻¹), tyrosine (mg g⁻¹) and free fatty acids (mg kg⁻¹), namely butyric (C4), caproic (C6), caprylic (C8) and capric (C10) were taken into consideration affecting the variation in aroma and flavour scores (AFS) of strained yoghurt during the storage period.

Material and Methods

Materials: Goat milk was obtained from faculty Farm of Animal Science Department and directly transferred to the Faculty Dairy Plant of Dairy Technology Department. Goat milk powder to fortify milk base was produced by Pilot Anyhydro Unit. Mix of “Yogurt 709”. “Yogurt VI” DVS cultures from Wiesby were used as the starter culture.

Production of yoghurt: First, fresh goat's milk was standardized to 3.5% fat and 16% total solids. After heat treatment, inoculation (2% starter culture) and incubation (up to 4.6–4.7 pH), goat's milk yoghurt was refrigerated at 4–5°C for 24 hours. Thereby produced set yoghurt was filled into cloth bag and leaved over a night at 20–22°C to remove the yoghurt serum (draining stage). Strained yoghurts of 250 g, were packed by the containers made from polystyrene, closure of the containers were achieved using polyamide/polyethylene material under vacuum.

Sampling, chemical and sensory analysis: For analysis at the storage period, samples were chosen randomly. Different cups were used for each sample at per period of storage.

Total solids, fat and titratable acidity in strained yoghurt were measured according to Turkish Standards Institution Method TS 1330, (Anonymous 1989). The pH values were measured by using a pH – meter, Model Orion 420 (200 Atlantic Avenue, New Hyde Park, NY 11040, USA) fitted with a standard combined electrode. Lactic acid and tyrosine values were determined spectrophotometrically according to the methods described by Steinholt and Calbert (1960) and Tunail (1978), respectively (Model, Spectronic 20, Milton – Roy Corp., 820 Linden Ave., Rochester, N.Y., 14626, USA).

Carbonyl compounds in strained yoghurt were determined by headspace technique using gas chromatography according to Ulberth (1991). Agilent Model 6890 Series GC System Plus gas chromatography (Agilent Technologies Inc., 395 Page Mill Rd., Palo Alto, CA 54306, USA) fitted with an FID detector was used. Volatiles were separated with a capillary column 30 m x 320 µm id (HP Innowax Polyethylene glycol, Model Agilent 19091N-13). Nominal film thickness for the column was 0.25 µm. Operating conditions for GC analyses were as follows; EPC split-splitless inlet, injection temperature 80°C, splitless, injection volume 1000 µL; oven temperature was raised from 50°C to 70°C at a rate of 4°C per min. and hold 0.5 min., from 70°C to 180°C at a rate of 20 °C and hold 0.2 min., column flow rate 0.7 µL/min⁻¹; detector temperature 250°C; make up gas nitrogen, flow rate for make –up gas 30 µL.min⁻¹, hydrogen flow rate 40 µL.min⁻¹, air flow rate 400 µL. min⁻¹. The temperature of the gas-tight syringe was maintained at 70°C. Crimp top headspace vials with 20 mL capacity were used for GC analyses. Sample of 5 g. were weighted in these vials and closed with crimper. All vials were stored in the freezer -20 °C until they were analyzed on the GC. Before the injection, vials were kept at 70°C/30 min. Then they were kept at room temperature 5 min. and injected to the GC. Quantification of constituents was achieved by means of a computing integrator operated in internal Standard mode. Slope of each standard curve was used as a factor for calculation.

The same gas chromatograph system (Agilent Model 6890 Series GC System Plus) was used in the determination of free fatty acids in strained yoghurt according to the method described by Deeth et al. (1983). Operating conditions in the system, FID detector at 260°C, Capillary column (30m×320 µm id. with 0.25 µm film thickness), Injection mode/volume: Split (1/10) 5 µL at 250°C, flow rates: H₂:Air:N₂ = 33:370:30 ml min⁻¹. Oven temperature: 120°C for 0 min increased to 200°C at a rate of 10°C min⁻¹ then held at 200°C for 2 min and increased to 205°C at a rate of

10°C min⁻¹ and held for 2 min then increased to 210°C at a rate of 10°C min⁻¹ and held for 2 min then increased to 215°C at a rate of 10°C min⁻¹ and held for 3 min and increased to 230°C at a rate of 10°C min⁻¹ held for 3 min.

The scale suggested by Downey 1969 was used in the evaluation of aroma and flavour of strained yoghurt. In this scale, it is given 8 to 10 points if it is accepted as excellent, 5 to 7 points if it is accepted sufficient and lower than 5 points if it is accepted as defective in terms of aroma and flavour. The panel group consisted of experienced 10 panelists from academic staff of Dairy Technology Department The strained yoghurt samples were stored at 4°C for a period of 60 days. Experiment was performed with three replications. All analyses were conducted an interval of 15 days during the storage period.

Statistical Methods: To investigate the relationship between aroma and flavour scores and independent variables, correlation coefficients were calculated. After that, to have more information on the relationship between independent compounds and aroma and flavour scores, multiple regression analysis was applied. Regression analysis also provides information on variation in dependent variable explained by the independent variables. Multiple regression equation is shown in equation 1.

Equation 1;

$$Y_i = \beta_0 + \beta_1 X_{1i} + \beta_2 X_{2i} + \dots + \beta_k X_{ki} + \epsilon_i$$

In the equation, X's represents independent variables and Y is the dependent variable. The β's represents the unknown regression coefficients. The β₀ is the Y intercept of the regression line. The β_k is the slope of the regression line and indicates the change in the mean of Y per unit increase in X_k, when all other independent variables are held constant. The unknown regression coefficients in the model are estimated by using Least Squares Method. The estimated regression coefficients are symbolized by b's. ε_i is the error (Neter et al. 1989, Draper and Smith 1998).

Stepwise regression procedure was used to obtain a satisfactory regression equation. It attempts to achieve an accurate regression equation inserting and removing variables into the equation. This procedure lasts inserting and removing the variables to the equation until an adequate regression equation is achieved. In this procedure, the order of insertion is determined by using partial correlation coefficient as a measure of importance of a variable that is not in the equation yet. This procedure begins forming

regression equation with the variable that is most highly correlated with the response variable. After that, partial correlation coefficients of all variables that are not in the regression equation with the response are calculated. The variable having the highest partial correlation coefficient is chosen as the next variable to enter the equation. When the regression equation is formed, the question arises as to whether or not it is meaningful to embrace the variable in the regression model. The order of addition is determined by using the partial F-test values to select which variable should enter next. The partial F-value is calculated from equation 2.

Equation 2;

$$\text{partial F} = \frac{\text{MSR}(X_k | X_1, \dots, X_{k-1}, X_{k+1}, \dots, X_{p-1})}{\text{MSE}}$$

In the equation, MSR is regression mean square and MSE is the mean square of error. X_k ($k=1,2,\dots,p$) are the independent variables. The partial F-value is compared to a selected or default F to enter value. After a variable has been added, the equation is examined to see if any variable should be deleted (Neter et al. 1989, Draper and Smith 1998, Zar 1999).

After obtaining a satisfactory regression equation, standardized regression coefficients, also known as path coefficients, were calculated to investigate the individual direct effect of independent variables on the variation in aroma and flavour scores. This is because the independent variables are measured in different units and this makes very difficult to compare regression coefficients. The standardized regression coefficient (path coefficient) was calculated using the following equation 3.

Equation 3;

$$P_{y.x_k} = b_k \frac{\sum (X_{ik} - \bar{X}_k)^2}{\sum (Y_i - \bar{Y})^2}$$

The calculated path coefficients seek out the direct effect of an independent variable that is assumed a cause for a response. They also indicate the change in the mean response in units of standard deviations of Y per unit change in the independent variable X_k in units of standard deviations of X_k . Path coefficients are standardized because they are estimated from correlation coefficients. A path coefficient that is written as $P_{y.x_k}$ indicates the path to Y from X_k (Düzgüneş et al. 1987, Neter et al. 1989). All results were statistically analysed for Minitab 15.1 packet programme.

Results

In this study, some carbonyl compounds were studied, but diacetyl, ethanol and 2-butanone were found to be trace amount. Therefore, the compounds of strained yoghurt, namely acetaldehyde, acetone, pH, titratable acidity, lactic acid, tyrosine, C4, C6, C8 and C10, were taken into consideration in the statistical analyses. Even though pH, titratable acidity and lactic acid are the expressions of acidity, they were all included in the analyses because they exemplify the acidity in a different way from each other. The means and standard error of means of all compounds taken into consideration in this study, being calculated over three replications at each time point when measurements were taken, was calculated and are presented in Table 1.

There was an increase in acetaldehyde, acetone, tyrosine and titratable acidity during the first 15 days of storage (Table 1). However, an obvious decrease in pH and free fatty acids were observed during the same period. There were fluctuations in the compounds between 15 and 45 days of storage. After that, there was decrease in acetaldehyde, C4, C8 and C10 during the last 15 days of storage. The first decline in AFS was observed during the first 15 days of storage, which coincided with the rises in the level of acetaldehyde, acetone, tyrosine and titratable acidity. Even though an increase in the aroma and flavour was acquired between 15 and 30 days of storage, a remarkable decrease in acceptability of aroma and flavour scores was noted until the end of storage period. At the 60th days of storage, the aroma and flavour defect was mostly pronounced. That is why the storage of strained yoghurt ended at the end of 60 days.

The correlation coefficients that were calculated to inspect the degree of relationships among compounds are given in Table 2. AFS was negatively correlated with acetone ($P<0.01$) and positively correlated with C4 ($P<0.05$), C8 ($P<0.01$) and C10 ($P<0.01$) (Table 2). This indicates that an increase in amount of acetone could cause a decrease in suitability of aroma and flavour. According to the results, an improvement in aroma and flavour can be accounted for by an increase in the levels of C4, C8 and C10 due to the positive correlations between them. The calculated correlation coefficients also showed that the pH, titratable acidity and lactic acid had adverse effect on AFS due to the negative correlation between AFS and them, which is statistically insignificant.

Table 1. Mean and standard error of mean of all compounds studied at the interval of 15 days

Compounds	1 st day	15 th day	30 th day	45 th day	60 th day
	$\bar{X} \pm S_{\bar{x}}$	$\bar{X} \pm S_{\bar{x}}$	$\bar{X} \pm S_{\bar{x}}$	$\bar{X} \pm S_{\bar{x}}$	$\bar{X} \pm S_{\bar{x}}$
AAL (mg kg ⁻¹)	8.363 ± 0.693	8.540 ± 0.000	10.090 ± 3.650	8.510 ± 3.830	6.300 ± 1.240
ACE (mg kg ⁻¹)	3.010 ± 0.314	10.110 ± 0.419	10.41 ± 1.310	11.507 ± 0.534	11.713 ± 0.140
pH	3.890 ± 0.040	3.787 ± 0.007	3.870 ± 0.006	3.993 ± 0.015	3.900 ± 0.038
TITA (°SH)	108.920 ± 1.250	119.060 ± 2.940	117.120 ± 1.510	116.810 ± 2.230	118.890 ± 3.510
LACA (g 100 g ⁻¹)	1.386 ± 0,013	1.374 ± 0,003	1.379 ± 0.037	1.361 ± 0,000	1.434 ± 0.037
TYRO (mg g ⁻¹)	0.224 ± 0,005	0.300 ± 0,051	0.247 ± 0.008	0.247 ± 0.003	0.260 ± 0.004
C4 (mg kg ⁻¹)	22.730 ± 4,940	4.977 ± 0.746	5.987 ± 0.296	8.990 ± 2.930	5.830 ± 1.480
C6 (mg kg ⁻¹)	7.243 ± 0.918	6.447 ± 0.392	6.127 ± 0.147	6.867 ± 0.541	6.853 ± 0.411
C8 (mg kg ⁻¹)	18.340 ± 1.200	5.983 ± 0.321	6.590 ± 0.414	9.800 ± 1.720	6.493 ± 0.800
C10 (mg kg ⁻¹)	54.550 ± 2.840	14.673 ± 0.559	16.530 ± 1.380	21.660 ± 4.240	16.930 ± 2.450
AFS	7.733 ± 0.067	5.733 ± 0.067	6.067 ± 0.176	5.133 ± 0.481	4.000 ± 0.529

Table 2. Correlation coefficients among the compounds taken into consideration

	AAL	ACE	pH	TITA	LACA	TYRO	C4	C6	C8	C10
AAL	1.000									
ACE	0.115	1.000								
pH	0.132	0.138	1.000							
TITA	-0.422	0.596*	-0.350	1.000						
LACA	0.199	0.185	0.089	-0.237	1.000					
TYRO	-0.139	0.300	-0.397	0.583*	-0.127	1.000				
C4	-0.169	-0.790**	-0.044	-0.456	-0.101	-0.338	1.000			
C6	-0.398	-0.248	-0.202	0.137	-0.103	0.128	0.680**	1.000		
C8	-0.094	-0.830**	0.233	-0.649**	-0.014	-0.465	0.832**	0.346	1.000	
C10	-0.022	-0.868**	0.151	-0.691**	0.033	-0.429	0.835**	0.322	0.976**	1.000
AFS	0.173	-0.832**	-0.212	-0.511	-0.412	-0.254	0.604	0.073	0.638**	0.659**

*P<0.05, **P<0.01

Multiple regression analysis was applied to the data to obtain more information on the relationship between aroma and flavour score and the other independent compounds. Regression analysis also provides information on how much variation in aroma and flavour score can be clarified by the other compounds considered as independent variables during the storage period. The results of the multiple regression analysis were presented in Table 3.

Table 3. The results of multiple regression analysis

Predictor	b ± S _b	R ²	F _(10,4) value
Constant	41.270 ± 27.050		
Acetaldehyde	0.125 ± 0.080		
Acetone	-0.242 ± 0.222		
pH	-4.899 ± 4.304		
Titrateable acidity	-0.008 ± 0.091		
Lactic acid	-9.666 ± 5.965	0.916	4.36
Tyrosine	0.207 ± 7.015		
C4	0.023 ± 0.102		
C6	-0.279 ± 0.594		
C8	0.259 ± 0.231		
C10	-0.071 ± 0.07		

b, regression coefficient, S_b, standard error of regression coefficient

The regression equation for AFS of strained yoghurt that was formed using the estimated coefficients in Table 3 is given in Equation 4.

Equation 4;
 $AFS = 41.3 + 0.125 AA - 0.242 AC - 4.90 pH - 0.0083 TA - 9.67 LA + 0.21 TYR + 0.023 C4 - 0.279 C6 + 0.259 C8 - 0.0706 C10$

(AA: acetaldehyde, AC: acetone, TA: titrateable acidity, LA: lactic acid; TYR: tyrosine)

As seen in the regression equation, acetaldehyde, tyrosine, C4 and C8 had positive effects on the AFS, which means that an increment in these compounds caused an improvement in aroma and flavour. However, the remaining compounds negatively related to the AFS, which means that these compounds were responsible for decline in satisfactory aroma and flavour of strained yoghurt.

The results of regression analysis verified that the independent compounds explained the 91.6% of variation in AFS during the storage period. Although a practically significant variation in AFS was elucidated

by the independent compounds, the results of analysis of variance confirmed that the constructed regression equation was not statistically significant ($P > 0.05$). This problem was encountered for the reason that the number of observation was too small to inspect the effect of ten independent compounds on the variation in AFS during the storage period.

The results of Stepwise procedure verified that acetaldehyde, acetone and lactic acid should be included in the regression equation. The results of the regression analysis are given in Table 4.

After applying Stepwise regression, the regression equation for AFS was formed using the estimated coefficients in Table 4 and is given in equation 5.

Equation 5;
 $AFS = 21.8 + 0.117 AA - 0.315 AC - 10.139 LA$
 (AA: acetaldehyde, AC: acetone, LA: lactic acid)

The obtained regression equation for AFS demonstrated that 0.117 acetaldehyde can be interpreted as an increase of 0.117 units in the mean AFS with one unit increase in acetaldehyde when the acetone and lactic acid were constant. Moreover, a unit increase in acetone caused a decrease in 0.315 units in the mean AFS when the acetaldehyde and lactic acid were held constant even though a decline of 10.139 units in the mean of AFS was resulted from a unit increase in lactic acid when acetaldehyde and acetone were constant. In other word, Acetaldehyde was positively related to the AFS, which means that an increment in this compound caused an improvement in aroma and flavour. Nonetheless, acetone and lactic acid were negatively related to the AFS, which means that these compounds were responsible for defective aroma and flavour of strained yoghurt.

However, as mentioned before, the primary purpose of this study was to explore the individual direct effect of the compounds included in the regression equation. Therefore, path coefficients were also calculated to determine individual direct effects of compounds included in the regression equation.

The path diagram is given in Figure 1. The calculated direct effects of each compound, path coefficients (P), on aroma and flavour scores are exhibited in Table 5. The squared path coefficient is the individual determination coefficient, which means how much variation in AFS can be explained directly by a single independent compound (Table 5).

Table 4. The results of multiple regression analysis excluding acetaldehyde, acetone, lactic acid

Predictor	Coefficients \pm Standard error	R ²	F _(4,10) -value
Constant	21.765 \pm 4.781		
Acetaldehyde	0.117 \pm 0.040	0.867	23.83**
Acetone	-0.315 \pm 0.044		
Lactic acid	-10.139 \pm 3.520		

* $P < 0.01$

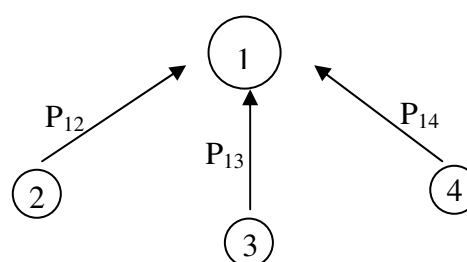


Figure 1. Path diagram exhibiting the direct effects of compounds on aroma and flavour.

(1. AFS, 2. Acetaldehyde, 3. Acetone, 4. Lactic acid)

Table 5. The direct effects (path coefficients) of acetaldehyde, acetone, lactic acid on AFS

Compounds	Path coefficients	P ²
Acetaldehyde	0.332 [*]	0.110
Acetone	-0.810 ^{**}	0.656
Lactic acid	-0.328 [*]	0.108

* $P < 0.05$ ** $P < 0.01$

Acetone had the highest effect on the variation of aroma-flavour scores of strained yoghurt (Table 5). The 65.6 % of variation in AFS of strained yoghurt during the storage period could be directly explained by acetone ($P < 0.01$). Acetaldehyde and lactic acid had statistically significant direct effects on the variation in AFS.

Discussion

The descriptive statistics calculated in this study showed that there was an increase in acetaldehyde, acetone, tyrosine and titratable acidity, but a decrease in pH and free fatty acids during the first 15 days of storage, which concurred with a noticeable decline in AFS (Table 1).

AFS was negatively correlated with acetone ($P < 0.01$) and positively correlated with C4 ($P < 0.05$), C8 and C10 ($P < 0.01$). This indicated that an increase in amount of ACE led to an unacceptable aroma and

flavour. According to the results, an improvement in aroma and flavour could be accounted for by an increase in the levels of C4, C8 and C10 (Table 2). It could be suggested that C4, C8 and C10 play an important role in development of typical aroma and flavour of strained yoghurt.

When the multiple regression analysis was applied to attain information on the relationship between AFS and the independent compounds, the results of this analysis verified that 91.6% of variation AFS was elucidated by the independent variables. However, the results of hypothesis control proved that the calculated determination coefficient and the regression equation formed were not statistically significant. This problem is generally encountered when the number of observation is rather small to study with many independent variables. Therefore, it should be borne in mind that while investigating the relationship between a dependent variable and several independent variables, one should take as many observations as possible.

Stepwise regression procedure was apply to answer the question as to whether or not it was possible to form a satisfactory regression equation which accurately explains the variation in AFS by the independent variables taken into consideration. The results of this procedure emphasized that the changes in variation of AFS could be satisfactorily accounted for by acetaldehyde, acetone and lactic acid (Table 4).

After calculating a reasonable regression equation, the individual effect of acetaldehyde, acetone and lactic acid was scrutinized by calculating standardized regression (path) coefficients. The calculated path coefficients showed that acetone has the most significant adverse direct effect on AFS ($P < 0.01$). Moreover, acetaldehyde and lactic acid had also statistically significant individual direct effect on AFS (Table 5).

The amount of acetaldehyde changed between 6.3 and 10.09 mg kg⁻¹ during storage period (Table 1), which was in accordance with the suggested levels for the optimum aroma and flavour (Abrahamsen et. al. 1978, Rysstand and Abrahamsen 1987, Yaygın and Mehanna 1988). Hence, the results of this study showed that if the amount of acetaldehyde is in a certain range, it positively affects the aroma and flavour of strained yoghurt.

In this study, lactic acid levels were much higher than that reported in the literature. The fact that there was a negative direct effect of lactic acid on AFS could be resulted from higher level of lactic acid.

The findings of this study verified that the changes in variation of AFS of strained yoghurt resulted from the changes in acetaldehyde, acetone and lactic acid. AFS of strained yoghurt was positively affected by the increase in acetaldehyde although acetone and lactic acid had negative effect on characteristic aroma and flavour of it.

References

- Abrahamsen, R. K., A. Svensen and G. Tufto. 1978. Some bacteriological activities during the incubation of yoghurt from goat's and cow's milk. XX. International Dairy Congress Paris – France.
- Agnihotri, M. K. and V. S. S. Prasad. 1993. Biochemistry and processing of goat milk and milk products. *Small Ruminant Research* 12, 151 – 170.
- Alichanidis, E. and A. Polyachoniadou. 1995. Special features of dairy products from ewe and goat milk from the physicochemical and organoleptic point of view. In: *Production and Utilization of Ewe and Goat Milk*. IDF. 41 Square Vergote, Brussels.
- Anonymous 1989. Yoğurt Standardı. TS 1330 Türk Standartları Enstitüsü, Ankara.
- Asperger, H. 1977. Applicability of analytic methods for assesment of yoghurt quality. *Dairy Science Abstracts* 3, 594.
- Atamer, M., M. Yildirim and O. Dağlıoğlu. 1993. Set ve Süzme yoğurtlarının depolama sürecindeki tat-aroma değişimi üzerine asitlik gelişimi, lipoliz, oksidasyon ve proteolizin etkisi. *Doğa. Türk Veteriner ve Hayvancılık Dergisi* 17: 49-53.
- Deeth, H. C., C. H. Fitz – Gerald and A. J. Snow. 1983. A gas chromatographic method for the quantitative determination of free fatty acids in milk and milk products. *New Zealand Journal of Dairy Science and Technology* 18: 13-20.
- Downey, W. K. 1969. Lipid oxidation as a source of off-flavour development during the storage of dairy products. *Journal of the Society of Dairy Technology* 22: 154-162.
- Draper, N. R. and H. Smith. 1998. *Applied Regression Analysis*. Third Edition. John Willey ve Sons, Inc., New York.
- Düzgüneş, O., T. Kesici, O. Kavuncu and F. Gürbüz. 1987. *Araştırma ve Deneme Metodları (İstatistik Metodlar-II)*. Ankara Üniversitesi, Ziraat Fakültesi Yayınları:1021, ders Kitabı: pp 295, Ankara.
- Görner, F., M. Palo and M. Seginova. 1973. Aroma compounds in cultured milks. *Dairy Science Abstracts* 35: 3173.
- Karademir, E., M. Atamer, B. Tamuçay and Ş. Yaman. 2001. Some properties of goat's milk yoghurts made by different fortification methods. *Milchwissenschaft* 57: 261-263.
- Neter, J., W. Wasserman and M. H. Kutner. 1989. *Applied Linear Regression Models*. Richard D. Irwin, Inc. Boston, USA.

- Peaker, M., A. Faulkner and D. R. Blatchford. 1981. Changes in the milk citrate concentration during lactation in the goat. *Journal of Dairy Research* 48: 357 - 361.
- Rasic, J. L. and J. A. Kurman. 1978. *Yoghurt. Scientific Grounds, Technology, Manufacture and Preparations. Vol. I. Fermented Fresh Milk Products. Technical Dairy Publishing House. Copenhagen – Denmark.*
- Rysstand, G. and R. K. Abrahamsen. 1987. Formation of volatile aroma compounds and carbon dioxide in yoghurt starter grown in cows and goats milk. *Journal of Dairy Research* 54: 257 – 266.
- Steinholt, T. K. and H. E. Calbert. 1960. A rapid colorimetric for the determination of lactic acid in milk and milk products. *Milchwissenschaft* 15: 7-11.
- Tamime A. Y. and R. K. Robinson. 1999. *Yoghurt Science and Technology. Second Edition. Woodhead Publishing Limited. Cambridge England.*
- Tunail, N. 1978. A study on some important physiological properties of lactic acid bacteria isolated from White brined cheese used as starter. Associate Profesorship Thesis, Ankara.
- Ulberth, F. 1991. Headspace gas chromatographic estimation of some yoghurt volatiles. *Journal of the Association of Official Analytical Chemists* 74: 630-634.
- Wojtowski, J., R. Dankow, E. Kozal and J. Wojciechowski. 1995. Influence of bacterial cultures on quality of yoghurt from cow, sheep, goat and buffalo milk. XXI. International Dairy Congress. Brief Communications. Volume I.. Moscow.(pp 294).
- Yaygın, H. and N. M. Mehanna. 1988. A comparative study on some volatile flavour components of yoghurt made from milk of different species. *Indian Journal of Dairy Science* 41(4):432 - 436.
- Zar, J. H. 1999. *Biostatistical Analysis (Fourth Edition). Prentice-Hall Inc., Simon ve Schuster Viacom Company, New Jersey, USA.*

Correspondence Address:

Zahide KOCABAŞ
Ankara University, Faculty of Agriculture
Department of Animal Science
06110 Diskapi, Ankara, Turkey
Tel: 90-312-5961501
E-mail: kocabas@agri.ankara.edu.tr