

Research Article





# Relations between feces, urine, milk and blood fatty acid contents in cattle

#### **Abstract**

The various fatty acids found in the circulation, cells and tissues of animals that have an important place in the feeding of animals are important components of cell membranes related to events that occur in a rich source of energy and cells. Since the fatty acids groups having important functions can not be synthesized by the mammalian cells, they must be taken from the outside of the body. Fatty acids detected in blood, feces, milk and urine can be an indicator to diagnose some metabolic disorders. This study was conducted to investigate the correlation between fecal matter, urine, milk and blood fatty acid contents in Holstein cows by multiple regression analysis.

As a result, milk, blood, stool and urine fatty acid contents were statistically significantly related to each other (p < 0.01).

**Keywords:** fatty acid contents, cattle, relation

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#### Introduction

Fatty acids and their metabolic products have important functions in the animal's body. These functions include being an active energy source, providing resistance to external factors, being the cornerstone of the cell and membrane, and acting as a precursor to hormone-like eicosanoic compounds such as prostaglandins, thromboxanes, and leukotrienes. In addition, hormone-like compounds play a role in the central nervous system, inflammatory diseases and other hormones in the immune system and in the regulation of blood pressure.<sup>1,2</sup>

Polyunsaturated fatty acids (PUFA) have been shown to be effective on many diseases in animal body metabolism. Lack of PUFAs indicates that major metabolic disorders such as chronor heart disease, imbalance in blood and lipid levels, high blood pressure, arthrosclerosis, thrombosis, heart failure, cancer, asthma, febrile diseases and arthritis have arisen.<sup>3–5</sup>

Monocarboxylates (especially propionate acetate, butyrate), termed short chain fatty acids, are the last products of anaerobic microbial fermentation of carbohydrates in the ruminant gastrointestinal tract. Monocarboxylates are absorbed through different parts of the gastrointestinal tract and produce significant amounts of energy in ruminant animals. It also plays a central role in metabolic communication between cellular metabolism and tissues. They are easily passed from the bowel to the blood and hepatic gluconeogenesis is transplanted into the body tissues to be used in lipogenesis and milk synthesis in peripheral tissues. In the investigations carried out it has been reported that the monocarboxylate carrier family in the plasma membrane of the cell is responsible for the transfer of short chain monocarboxylates. Despite the importance of short chain fatty acids (SCFA) as the main energy source in ruminant animals, the mechanism of SCFA transport and absorption is still not fully explored. In their study of Tagang et al.,6 it is stated that monocarboxylate carrier proteins have two main functions, which are facilitating the absorption of SCFA in the gastrointestinal tract and regulating cell pH in skeletal muscles. It has been reported that these tasks are important in terms of physiological homeostasis, animal welfare and productivity.

Fatty acids were first described by Burr in the early 1930s.<sup>7</sup> These authors have identified linoleic acid (C18: 2 n6) and alpha

linolenic acid (C18: 3 n3), important fatty acids in pigs and rats, as essential essential fatty acids for growth, skin structural health and reproduction in their research. Since then, researchers have shown the importance of fatty acids, which are lipid mediator molecules, such as prostaglandins, prostacyclins, thromboxanes, leukotrienes, liposines, and resolvins, which affect cellular function. Fatty acids taken into the body are included in the phospholipids in cell membranes to determine the structural and functional properties of the cells.8 Animals use fatty acids (FA) to grow, to maintain a fertile lactation period, to have a healthy lifestyle, and to increase fertility. In mammals, since cells can not synthesize some fatty acid groups, they must be taken from the outside with feed. Fatty acids are hydrophobic or amphipathic molecules.It has the functions of forming structural components of cell membranes, synthesizing prostaglandins and attaching proteins to cell membranes. Fatty acids are also stored as intracellular triacylglycerides (TAG) in lipid droplets and provide a powerful source of energy when the body needs it.8

Severe negative energy balance in early lactation causes metabolic disturbances in dairy cows. During this period of negative energy balance, the body, fat and muscle proteins function to compensate for the energy deficit. <sup>10</sup>

The widespread mobilization of body reserves significantly increases the concentration of free fatty acids (FFA) in blood plasma in early lactation. Accordingly, FFA's liver uptake, oxidation of the FFA or conversion to very low density lipoproteins exceeds the capacity of the liver. This leads to the accumulation of triacylglycerol (TAG) in the liver, which increases the oxidation of ketone bodies and increases the incidence of ketosis and fatty liver, such as mastitis.

Animal welfare has an increasing prevalence and the lack of chronic stress in the animal is one of the preconditions for animal welfare. Especially in scientific research, when sampling for analysis, it is necessary to improve the environment for animals to survive stress and to treat them with the least stress. During the blood, saliva or urine collection, some manipulations of the animal are applied and these applications in free, moving animals are stressful to the practitioner and animal. For this reason, it is very important to have various health indicators in easily releasable samples of animals. In recent years, the bioindicator has been seen as a priority work in this way.



The purpose of this study was to determine the fatty acids in urine, blood, milk and fecal samples taken from animals by Gas Chromatography and to determine the relationship between fatty acids and their importance for cattle breeding.

#### **Material** method

In this study, samples taken from holstein cows with healthy and similar characteristics grown in Cukurova University Faculty of Agriculture Rotating Capital Management, Dairy Cattle Research Unit were used. Experiemantl cows were fed with a total mixing ratio (TMR) of 60: 40 for the coarse feed ratio.

TMR is a mixture of corn silage, alfalfa, wheat straw and concentrate (18% crude protein and 2650kcal / metabolic energy (ME)/kg), and is given at 07.00 and 16.00 in the morning. Experimental animals are provided with an automatic milking system at the central milking place twice a day for 12 hours. Experimental material Milk, blood, feces and urine samples were collected from Holstein cows and fatty acid analyzes were determined by Gas Chromatography in the laboratories of Fisheries Faculty of Cukurova University. Urine samples were obtained by manual stimulation of perineal regions of cows and stool specimens as described by Sankar and Archunan; 11 Blood samples were obtained from Klemm et al. 12 Milk samples were taken from the first milk that accumulated at the tip of the nipple after the milk was milked and the samples were transported to the analysis under proper conditions to preserve the cold chain. Blood samples were collected in the early morning vena jugularis.

Samples were prepared by mixing 10ml aliquots of 10ml sodium citrate in a 30ml injection syringe, sealed with a teflon coated septum. The samples were rapidly frozen and stored at -80°C for analysis by gas chromatography.

Bligh & Dyer<sup>13</sup> method was used for lipid extraction from animal samples. Methyl esters to be formed from the samples were prepared by transmetallation using 2M KOH in methanol and n-hexane; 10mg of extracted oil was dissolved in 2ml of hexane, then 4ml of 2M methanolic KOH were added. The tube was then taken to the hexane layer for GC analysis after centrifugation at 4000rpm for 10 minutes, vortexing for 2 minutes to thoroughly mix the liquor at room temperature.

20ml of the collected samples were sieved using filter paper (60-120 $\mu$ m) and stored frozen at -20 $^{\circ}$ C. <sup>14</sup>

Samples were analyzed by GC Clarus 500 with an automatic sampler (Perkin Elmer, USA) equipped with a flame ionization detector and a silica capillary SGE column (30m x 0.32mm, ID x 0.25µm, BP20 0.25UM, USA) for fatty acid compositions.

After setting the injector and detector temperature to 220°C and 280°C respectively, the oven temperature was kept at 140°C for 5 minutes. At 200°C at 4°C/min and then at 220°C at 1°C/min. The size of the samples was adjusted to 1  $\mu l$  and the carrier gas was run at 16psi, using a 1: 100 split ratio as separation. Two repetitive GC analyzes were performed to determine the fatty acids and the results obtained were expressed as the mean value in% of GC area and  $\pm$  standard deviation.

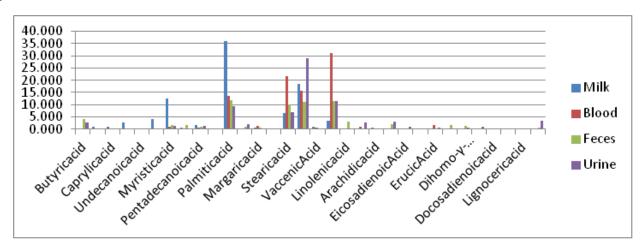
#### Statistical analysis

The statistical analysis of the obtained data was performed using the one-way analysis of variance (One-WayAnova) and the SPSS package program. In addition, correlation analysis, multiple regression analysis, whether there is any relation between dependent variables and independent variables, and the level, direction and significance levels of the existing relationship have also been tried to be determined by SPSS package program. The scatter graphs were also used for the visual expression of the analysis results. After applying variance analysis (ANOVA) to the obtained data, multiple comparison analysis was performed with P < 0.05 significance level.

## Findings and discussion

The amounts of fatty acids detected in milk, blood, faeces and urine of cows in study are shown in Table 1 and the changes are given in Graph 1.

When Table 1 and Graph 1 are examined, it is seen that the animals have high levels of capric acid, lauric acid, myristic acid, palmitic acid, stearic acid, oleic acid and linoleic acid in the milk. In these fatty acids, palmitic acid is the most abundant in milk. Butyric acid is not present in milk, although it is in urine and in faeces.



**Graph I** Comparison of fatty acids in milk, blood, feces and urine of cows.

Table I Fatty acids and percentages detected in milk, blood, feces and urine of cows

Name of Fatty Acids	Percentage of fatty acids in milk	Percentage of fatty acids in blood	Percentage of fatty acids in feces	Percentage of fatty acids in the urine 2,70	
Butyricacid	0,00	0,00	4,25		
Caproicacid	0,86	0,08	0,30	0,34	
Caprylicacid	0,81	0,07	0,00	0,07	
Capricacid	2,75	0,16	0,00	0,00	
Undecanoicacid	0,11	0,00	0,00	0,00	
Laurikacid	3,97	0,26	0,21	0,00	
Myristicacid	12,56	1,13	1,68	1,46	
Myristoleicacid	0,54	0,33	1,59	0,00	
Pentadecanoicacid	1,51	0,49	1,00	1,15	
Methylpentadecanoate	0,21	0,30	0,43	0,00	
Palmiticacid	36,05	13,45	11,66	9,45	
Palmitoleicacid	0,36	0,32	0,91	2,05	
Margaricacid	0,61	1,22	0,49	0,14	
Heptadecenoicacid	0,07	0,09	0,00	0,00	
Stearicacid	6,69	21,64	10,07	6,79	
Oleicacid	18,33	15,75	11,14	28,92	
VaccenicAcid	0,87	0,48	0,00	0,00	
Linoleicacid	3,52	31,01	11,41	11,52	
Linolenicacid	0,06	0,34	3,07	0,22	
Alpha LinolenicAcid	0,24	0,96	0,36	2,74	
Arachidicacid	0,64	0,21	0,33	0,35	
Eicosanoicacid	0,09	0,17	2,10	3,07	
EicosadienoicAcid	0,06	0,09	0,00	1,12	
Arachidonicacid	0,06	0,16	0,33	0,00	
ErucicAcid	0,23	1,74	0,00	0,44	
BehenicAcid	0,04	0,00	1,82	0,14	
Dihomo-γ-linolenicacid	0,03	0,11	1,42	0,44	
EicosapentaenoicAcid (EPA)	0,02	0,15	0,00	1,07	
Docosadienoicacid	0,08	0,00	0,00	0,00	
NervonicAcid	0,06	0,00	0,00	0,00	
Lignocericacid	0,03	0,13	0,33	0,26	
Docosahexaenoicacid	0,05	0,08	0,54	3,58	

Blood palmitic acid, stearic acid, oleic acid and linoleic acid at high levels. There are no butyric acid, undecanoic acid, behenic acid, docosadienoic acid and nervonic acid, although linoleic acid is the highest among these fatty acids.

In the faeces the rates of butyric acid, palmitic acid, stearic acid, oleic acid, linoleic acid and linoleic acid are high. The highest ratio of fatty acids is palmitic acid, stearic acid, oleic acid and linoleic acid. ecause the percentages of these acids are very close to each other. There are no capric acid, caprylic acid, undecanoic acid, vaxenic acid,

heptadecanoic acid, eicosyenoic acid, erucic acid, EPA, doosadienoic acid and nervonic acid in the faeces.

Palmitic acid, stearic acid, oleic acid, linoleic acid are high in the urine. Oleic acid is the fatty acid with the highest rate in the urine. There are no capric acid, undecanoic acid, lauric acid, myristoleic acid, methylpentadecanoate, heptadecanoic acid, vaksenic acid, arachidonic acid, doosadienoic acid and nervonic acid in the urine.

The ratio of  $\Sigma$ SFA,  $\Sigma$ MUFA and  $\Sigma$ PUFA of fatty acids in milk, blood, feces and urine is given in Table 2.

Table 2  $\Sigma \text{SFA}, \Sigma \text{MUFA}$  and  $\Sigma \text{PUFA}$  ratios of fatty acids in milk, blood, feces and urine

	Percentage of fatty acids (%)								
Fatty Acids	Milk	Blood	Feces	Urine					
C10:0	2,75	0,16	0,00	0,00					
C11:0	0,11	0,00	0,00	0,00					
C12:0	3,97	0,26	0,21	0,00					
C13:0	0,00	0,00	0,00	0,00					
C14:0	0,54	0,33	1,59	0,00					
C15:0	1,51	0,49	1,00	1,15					
C16:0	36,05	13,45	11,66	9,45					
C17:0	0,61	1,22	0,49	0,14					
C18:0	6,69	21,64	10,07	6,79					
C20:0	0,64	0,21	0,33	0,35					
C21:0	0,00	0,00	0,00	0,00					
C22:0	0,04	0,00	1,82	0,14					
C23:0	0,00	0,00	0,00	0,00					
C24:0	0,03	0,13	0,33	0,26					
∑SFA	52,94	37,89	27,5	18,28					
C14:1	0,54	0,33	1,59	0,00					
C15:1	0,21	0,30	0,43	0,00					
C16:1	0,36	0,32	0,91	2,05					
C17:1	0,07	0,09	0,00	0,00					
C18:1n7c	0,87	0,48	0,00	0,00					
C18:1n9c	18,33	15,75	11,14	28,92					
C20:1	0,09	0,17	2,10	3,07					
∑MUFA	20,47	17,44	16,17	34,04					
C18:2n6	3,52	31,01	11,41	11,52					
C18:3n6	0,06	0,34	3,07	0,22					
C18:3n3	0,24	0,96	0,36	2,74					
C20:2 cis	0,06	0,09	0,00	1,12					
C20:3 n6	0,03	0,11	1,42	0,44					
C20:4 n6	0,06	0,16	0,33	0,00					
C20:5n3	0,02	0,15	0,00	1,07					
C22:6 n3	0,05	0,08	0,54	3,58					
∑PUFA	4,04	32,9	17,13	20,69					
MUFA/SFA	0.386	0,460	0,588	1,862					
PUFA/SFA	0,076	0,868	0,622	1,131					
PUFA/MUFA	0,197	1,886	1,059	0,607					
∑n6	3,67	31,62	16,23	12,18					
$\sum$ n3	0,31	1,19	0,9	7,39					
n6/n3	11,83	26,57	18,03	1,64					

Kovac et al. <sup>15</sup> analyzed the fatty acids found in the blood of animals and found the results as  $\Sigma SFA = 42,67$ ,  $\Sigma MUFA = 16,86$ ,  $\Sigma n6 = 33,97$  and  $\Sigma n3 = 4,50$ . When the results of the analysis are compared with those of Table 2,  $\Sigma SFA$ ,  $\Sigma n6$  and  $\Sigma n3$  values are high and  $\Sigma MUFA$  values are low.

In the above-mentioned studies, Baer<sup>16</sup> said that the reason for the variation in fatty acid ratios is due to factors such as lactation, seasonal changes, nutrition, genetic variation, and changes in the energy state of the cattle such as cattle somatotrophy.

The result of the correlation analysis to determine the relationship between fatty acid contents detected in milk, feces, blood and urine specimens is given in Table 3. As a result of the correlation analysis, statistically significant (p<0.01) relation between milk, feces, blood and urine fatty acids was found (Table 3).

Table 3 Relationship between fatty acids in milk, feces, blood and urine

	n	R	P
Milk-blood	32	0,487**	0,005
Milk-feces	32	0,706**	0,000
Milk-urine	32	0,607**	0,000
Blood-feces	32	0,899**	0,000
Blood-urine	32	0,701**	0,000
Feces-urine	32	0,804**	0,000

<sup>\*\*</sup>Correlation is important at 0.01 level (according to r value)

As a result of the correlation analysis to determine the relationship between fatty acid contents detected in milk, feces, blood and urine samples. There was a moderate correlation between milk and blood, and this correlation was statistically significant (p<0.01) (r 0.487, p=0.005). There is a high level of correlation between milk and feces and this relationship is statistically significant (p<0.01) (r=0.706, p=0.000). It can be said that there is a moderate relationship between milk and urine, and this relationship is statistically significant (r=0.607, p=0.000). There is also a high correlation between blood and feces and blood and urine, and this relationship is statistically significant (p<0.01) (r=0.899, p=0.000 and r=0.701, p=0.000, respectively). It was also found that there was a high level of correlation between faeces and urine (r=0.804, p=0.000) and statistically significant (p<0.01).

Table 2 shows that there is a positive correlation between levels of fatty acids detected in milk, feces, blood and urine.

The results of multiple regression analysis to determine the level of relationships between levels of fatty acids detected in milk, feces, blood, and urine are given in the table.

According to the table above, the ANOVA test was significant. The P value is 0,000 (p<0.01). That is, the regression model is meaningful as a whole.

The results of the multiple linear regression analysis to determine the relationship between fatty acid contents detected in milk, faeces, blood, and urine specimens are shown in Table 4.

When we look at the values in the table, the  $R^2$  value is calculated as 0,612 and the corrected  $R^2$ =0,569 in the dependent variable column and it is determined that there is a moderate correlation in the positive direction. The feces, urine and blood, which are independent variables, can account for 57% of the dependent variable, milk. The dependent

variable kanda, corrected R<sup>2</sup>=0.835, indicates that there is a high level of correlation in the positive direction and that the independent variables milk, feces, urine can accommodate 83% of the change in the dependent variable.

When the independent variable is feces, corrected  $R^2$ =0.909, it is seen that there is a very high level of correlation in the positive direction and that the independent variables milk, blood and urine can cover 91% of the change in the faeces. Adjusted  $R^2$ =0.613 in the dependent variable, urine is found to be moderate and corresponds to 61% of the independent variables milk, blood, feces.

The coefficients and significance values of the regression model obtained as a result of the analysis applied to the fatty acids are given in Table 5. When dependent variable milk is selected, the coefficient of the constant term is calculated as -0.697 and the value of p is specified as 0,504. In this case, the fixed term does not make

sense. The coefficient of the regression model of the blood is -0,765. The significance level of the T test was calculated as p=0,010 and p<0,05, which is statistically significant. However, as can be seen, the conversion rates of the fatty acids in the blood are inversely correlated with milk. Because the regression model coefficient is negative. Outcome regression model coefficient was calculated as 2,696 and p=0.00 and milk and feces were statistically significant (p<0,05).

When the blood was selected as the dependent variable, the blood regression model coefficients B=-0,292 and p=0,008 were found and the relationship between blood and milk was found significant. (P<0.05). Since B carries a negative value, the fatty acid change rates in the milk have an inversely proportional relationship with blood. The relation of blood to stool was also significant (p=0,000, p<0,05). Another way to estimate the proportion of blood fatty acids, which are dependent variables, is that fatty acids found in milk and feces contribute significantly to the model.

Table 4 Results of multiple regression analysis

The dependent variable	R	$\mathbb{R}^2$	Adjusted R <sup>2</sup>	df	F	p
Milk	,782	,612	,569	3	14,205	0,000
Blood	,923	,851	,835	3	53,470	0,000
Feces	,958	,918	,909	3	104,208	0,000
Urine	,806	,650	,613	3	17,340	0,000

Table 5 Coefficients and significance value of regression model

The dependent variable- Milk	В	p	The dependent variable- Blood	В	p	The dependent variable Urine	В	p	The dependent variable Feces	В	P
Fixed term	-,697	,504	Fixed term	-,846	,167	Fixed term	-,145	,844	Fixed term	,445	,046
Blood	-,765	,010	Milk	-,292	,008	Milk	,044	,751	Milk	,142	,000
Feces	2,696	,000	Feces	2,259	,000	Feces	1,277	,029	Blood	,312	,000
Urine	,082	,757	Urine	-,037	,816	Blood	-,053	,816	Urine	,125	,029

When the feces was selected as the dependent variable, the regression model coefficient of feces was B=0,445 and p<0,05 in p, milk, blood and urine, so the relation of feces with milk, blood and urine was found significant. It has been determined that the change in fatty acids in milk, blood, and urine did not significantly predict fatty acids in the feces.

When the dependent variable urine was selected, the urine regression model coefficient B=-0,144 was found and it was found that the urine had an inverse relationship with the feces (P<0.05). The table indicates that changes in the fecal fatty acids in the stool will contribute significantly to the change in the fecal matter due to the reflection of the urine in the reverse direction.

To meet the energy needs of animals with a negative energy balance, adipose tissues act and increase plasma non-unesterified fatty acids (NEFA) concentrations due to lipolysis.<sup>17</sup> Increases in feeding (energy) uptake increase the need for precursors (acetate, butyrate) for milk oils (C: 6-C: 16), which are mainly expressed in the mammary gland.<sup>18</sup> According to this information, Miettinen and Huhtanen, in their study of 1989, found that there is an increasing positive correlation between chain length of fatty acids up to chain length C16 and ME-balance in milk.<sup>19</sup> A high positive correlation between C16: 0 (palmitic acid) and ME-balance was associated with the expression of a large proportion of palmitic acid in the mammary glands. By dropping the metabolic energy, body oils are actuated

and the proportions of palmitoleic and C18 fatty acids in the milk increase. The reason for this is that the triglycerides of body fat stores in cattle are mostly fatty acids in the form of palmitic, stearic and oleic acids. They point out that palmitic acid is partly due to desaturation activity in the mammary gland, which converts palmitoleic acid and stearic acid to oleic acid, and that these results may be a good indicator of the fatty acid composition of milk fat in the dairy cows.

Kamila et al.<sup>20</sup> noted that the high presence of unesterified fatty acids (NEFA) is a sign of the consumption of lipid reserves to balance the disproportion between the energy provided by feed ration and the energy required for milk production, and is associated with a higher incidence of metabolic disease later in life. Geishauser et al.<sup>21</sup> reported that  $\beta$ -hydroxybutyric acid (BHBA) concentrations in the first week after birth may be indicative of subclinical ketosis. Jorjong et al.<sup>22</sup> reported that the increase in the concentration of C18: 1 cis-9 (oleic acid = milk fat) in the second week of lactation may be a symptom that can be detected early for the risk of developing high blood levels of NEFA, and based on this knowledge, Kamila et al.<sup>20</sup> noted that the increase in oleic acid found in milk during early stages of lactation in high yielding cows could be applied as a biomarker for the early detection of NEFA and BHBA at high levels of calm.

Stefańska et al.<sup>23</sup> conducted this study to investigate the effect of ruminal fluid pH on blood, urine, faeces and milk biochemical indices and to determine which may be useful for the diagnosis of

subacute ruminal acidosis. As a result of the study, they reported that ruminal fluid pH showed a positive correlation (p≤0.01) with the stamina concentrations of SFA and blood pH values. Reduction in the absorption rate of ruminal essential fatty acids in animals with subacute renalacidosis causes ruminal pH to fall for two reasons: Ruminal essential fatty acids are the accumulation and decrease of bicarbonate input and pH from the blood stream. Decrease in the pH of the ruminal fluid was found to be the resultant increase in IGF-I, free fatty acid levels, and decrease in rectal temperature and blood pH values. These data indicate that these values may be used as indicators in physiological changes and may help in the diagnosis of subacute ruminal acidosis syndrome when serial measurements are made.

The energy metabolism of dairy cows during the transition period is closely related to lipid metabolism. The best indicator of negative energy balance and the degree of mobility of lipids at transition from body fat reserves is due to increased free fatty acid (SFA) concentrations. In this period there is an increase in blood SFA concentrations bound to albumin and carried to the liver. 24,25 Djokovic et al.26 reported significantly higher concentrations of free fatty acids in the blood of ketosis cows (p<0.01) compared to healthy cows before and after calving. There was a significant positive correlation (r=0.51; p<0.05) between the free fatty acids in the liver and the lipid content in the liver and a negative correlation (r =-0.64; p<0.05) between free fatty acids and triacylglycerol concentrations in the ketosis cows stated. When they compared the results of the study with the results of other authors,24 they showed that the increase in the concentration of free fatty acids in the blood clearly showed that the lipid content in the liver cells was increased. Liver lesions that appear as a consequence of lipomobilization are usually observed in highly productive cows at the first stage of lactation. The development of these hepatic lesions increases the likelihood of diseases such as ketosis, mastitis, metritis, hypocalcemia, abomasum depression and placenta retention.<sup>27</sup>

In this study, the ratio of  $\Sigma$ SFA was found high in milk, feces and stool as shown in Table 2. As a result of the regression analysis made in Table 4, it is found that the dependent variable  $R^2$ , which is corrected  $R^2$  =0.835, has a high level of correlation in the positive direction and that the independent variables milk, feces, urine can meet 83% of the variation in the dependent variable. According to the regression analysis (Table 5), the relation of feces with milk, blood and urine was significant and it was found that the change of fatty acids in milk, blood and urine did not significantly predict fatty acids in feces.<sup>28</sup>

## **Results**

The various fatty acids found in the blood circulation, cells and tissues of animals are a rich source of energy for animals and important components of events occurring in the cell. It has been determined that there is a significant relationship between fecal matter, milk, blood and urine fat contents in this study which is carried out to investigate the correlation between fecal matter, urine, milk and blood fatty acid contents in holstein cows and multiple regression analysis. When the fatty acid compositions of milk, feces urine and blood, and SFA,  $\Sigma$ MUFA and  $\Sigma$ PUFA ratios were examined, it was found that the ratio of  $\Sigma$ SFA to milk, blood and faeces was high and  $\Sigma$ MUFA ratio was high in urine. As a result of the correlation analysis, statistically significant (p <0.01) relationship between milk, feces, blood and urine fatty acids was found.

There are studies that report that the fatty acid composition of milk fat may be a good indicator of energy status in dairy cows. It is understood that the sample showing close similarity and correlation with blood fatty acids from blood, feces and urine can be used at a high level in the prediction of negative energy according to the regression model.

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None.

### **Conflicts of interest**

Author declares there is no conflict of interest.

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