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Research Article

Effect of heat stress on metabolic analysis of sweat in dairy cattle

Efeito do stress térmico na análise metabólica do suor em bovinos de leite

Özgül Anitaş[1*(ORCID 0000-0003-4607-8594)], **Serap Göncü**[1(ORCID 0000-0002-0360-2723)], **Fatma Hepsağ**[2(ORCID 0000-0002-3688-4106)], **Yeşim Özoğul**[3(ORCID0000-0001-7769-9225)]

[1]Faculty of Agriculture, University of Cukurova, Adana, Turkey. *Author for correspondence: ozgulanitas01@gmail.com
[2]Faculty of Applied Sciences, University of Osmaniye Korkut Ata, Kadirli Campus, Osmaniye, Turkey.
[3]Faculty of Fisheries, University of Çukurova, Adana, Turkey.

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ABSTRACT

During heat stress, dairy cattle use a variety of physiological and cellular mechanisms to dissipate heat from their bodies and protect cells from damage. In this process, the change in the fatty acid content of the sweat released from animals contains important findings in terms of regulating body temperature. In this study, the maximum ambient temperature during the day varied between 35 °C and 37 °C, and the relative humidity varied between 37% and 49%. Fatty acid ratios of sweat samples taken from Holstein and Jersey cows under heat stress were determined by gas chromatography. Myristic acid (C14:0), palmitic acid (C16:0), stearic acid (C18:0) and oleic acid (C18:1n9c), fatty acids with the highest ratios in sweat, were recorded. These fatty acids were 9.28%, 26.24%, 11.43%, 26.39% in Holstein cow sweat, respectively; It was detected in Jersey cow sweat at the rates of 10.19%, 23.83%, 27.63%, 20.83%. Total monounsaturated fatty acid content in sweat of Holstein and jersey cows was determined as 26.39% and 20.83%, while total saturated fatty acid content was determined as 46.95% and 61.65%, respectively. The total percentage of fatty acids with the highest percentage detected in sweat under heat stress conditions (myristic acid, palmitic acid, stearic acid and oleic acid) was 73.34% in Holstein cows and 82.48% in Jersey cows. Apart from these, the percentage of other compounds detected in sweat constituted 26.66% in Holstein and 17.52% in Jersey cows. As a result of correlation analysis, a strong positive (r=757) and significant (P<0.01) relationship was found between sweat fatty acids and sweat fatty acids of Holstein and Jersey cows in summer months. It was determined that especially the stearic acid rates (11.43%, 27.63%) in the sweat of Holstein and Jersey. The low level of stearic acid in the sweat of Holstein cows indicates that stearic acid from fat tissue is not used much as an energy source. Jersey cows under heat stress were quite different. Based on these findings, stearic acid detected in the sweat of animals can be determined as a heat stress bioindicator. Differences in the fatty acid ratios in sweat in the study show that the two breeds exhibit different adaptation responses under the same conditions, at the same temperature and humidity.

KEYWORDS: Breed. Fatty acids. GC. Heat stress. Stearic acid. Sweat.

RESUMO

Durante o stress térmico, o gado leiteiro utiliza uma variedade de mecanismos fisiológicos e celulares para dissipar o calor nos seus corpos e proteger as células contra danos. Neste caso, a alteração do teor de ácidos gordos do suor libertado pelos animais contém descobertas importantes em termos de regulação da temperatura corporal. Neste estudo, a temperatura ambiente máxima durante o dia variou entre os 35 °C e os 37 °C, e a humidade relativa variou entre os 37% e os 49%. As proporções de ácidos gordos das amostras de suor retiradas de vacas Holandesas e Jersey sob stress térmico foram determinadas por cromatografia gasosa. Como resultado da análise, o ácido mirístico (C14:0), o ácido palmítico (C16:0), o ácido esteárico (C18:0) e o ácido oleico (C18:1n9c), que foram os ácidos gordos com as maiores proporções no suor , foram comparados em duas raças. Estes ácidos gordos foram 9,28%, 26,24%, 11,43%, 26,39% no suor da vaca holandesa, respetivamente. Foi detectado no suor de vaca Jersey nas taxas de 10,19%, 23,83%, 27,63%, 20,83%. O conteúdo total de ácidos gordos monoinsaturados no suor das vacas holandesas e jersey foi determinado como 26,39% e 20,83%, enquanto o conteúdo total de ácidos gordos saturados foi determinado como 46,95% e 61,65%, respetivamente. A percentagem total de ácidos gordos com maior percentagem detectada no suor em condições de stress térmico (ácido mirístico, ácido palmítico, ácido esteárico e ácido oleico) foi de 73,34% nas vacas Holandesas e de 82,48% nas vacas Jersey. Para além destes, a percentagem de outros compostos detectados no suor constituiu 26,66% nas vacas Holandesas e 17,52% nas vacas Jersey. Como resultado da análise de correlação, foi encontrada uma relação forte positiva (r=757) e significativa (P<0,01) entre os ácidos gordos do suor e os ácidos gordos do suor das vacas Holandesas e Jersey nos meses de Verão. Determinou-se que especialmente as taxas de ácido esteárico (11,43%, 27,63%) no suor das vacas Holandesas e Jersey sob stress térmico eram bastante diferentes. Com base nestas descobertas, o ácido esteárico detetado no suor dos animais pode ser determinado como um bioindicador de stress térmico. As diferenças nas proporções de ácidos gordos no suor no estudo mostram que as duas raças apresentam respostas de adaptação diferentes nas mesmas condições, à mesma temperatura e humidade.

PALAVRAS-CHAVE: Raça. Ácidos gordos. GC. Stress térmico. Ácido esteárico. Suor.

INTRODUCTION

Heat stress is a condition that negatively affects cattle farms in many parts of the world, causing an increase in the body temperature of animals and thus inducing a number of physiological responses in the body (LIU et al. 2017, GIANNONE et al. 2023). Animals use the energy required for lactation and growth due to their biological condition (SGUÍZZATO et al. 2020), but in addition, heat stress due to increased ambient temperature and humidity causes deterioration of living conditions and decreased quality of life (HABEEB 2020, RONY & ALAMGIR 2023). As a result of heat stress, animals have decreased milk yield, decreased feed intake and reproductive problems (ATRIAN & SHAHRYAR 2012). Among the physiological coping strategies of dairy cows faced with heat stress, increased respiratory rate and respiratory rate as well as sweating play an important role in body temperature regulation. The skin was the primary barrier between the animal body and the external environment and performs essential functions such as prevention of dehydration, regulation of temperature and protection from mechanical, chemical and thermal stress. The most important factor for these functions is sweating. Sweating is initiated by the hypothalamus in response to an increase in body temperature by sending a signal through the sympathetic nervous system. Approximately 99% of sweat is composed of water and 1% of sweat contains mineral substances such as sodium and potassium, urea, protein lactic acid and fatty acids (CHEN et al. 2020). Fatty acids are secreted by apocrine glands and essential for the normal barrier function of the skin. In particular, it has been reported that fatty acids in sweat are released from triglycerides in adipose tissue into sweat through the blood as an energy source in situations such as starvation and stress in animals. Changes in the fatty acid content of sweat released from animals under heat stress conditions are important for the body temperature regulation. Considering the fact that high-producing dairy cows have increased energy requirements, the fatty acids in their bodies play an important role.

In the diet, bloodstream, cells and tissues of living organisms, a variety of fatty acids are found. Fatty acids are a rich source of energy for animals. They are signalling mediators of events in the cell and play various biological roles in the basic structure of membranes. They are structural components of cell membranes, they synthesise prostaglandins and they anchor proteins to the cell membranes. Fatty acids are also stored in the form of intracellular triacylglycerides in lipid droplets and provide a powerful source of energy when the body needs it (ZHANG et al. 2021). Cattle use fatty acids (FA) in their bodies to grow, lactate productively, stay healthy and reproduce.

Research on body temperature regulation responses to hot climates has shown that, in general, acclimatized species were able to maintain higher sweat rates than nonacclimatized species (LEI et al. 2021, TOCHIHARA et al. 2022, SAAT et al. 2005). Consequently, changes in sweat composition can be seen with changes in sweat gland metabolism. However, research on the effect of heat stress on fatty acid content in animal sweat, especially in cattle, is very limited.

Animal welfare is of increasing importance and avoiding chronic stress in the animal is one of the prerequisites for animal welfare. Especially when taking samples for analysis in scientific research, it was necessary to improve the environment of the animals to avoid stress and to treat them in a way that will cause the least stress. During the collection of blood, saliva or urine, the animal was subjected to some manipulation, and in free, mobile animals this can be stressful for the practitioner and the animal. It is therefore of great importance to find various health indicators in the body secretions of animals, such as sweat, which can be easily sampled. In recent years, the term bioindicator has become a priority in studies in this sense.

In recent studies on the determination of fatty acids in cows' sweat, it has been determined that myristic acid (C14:0), palmitic acid (C16:0), stearic acid (C18:0) and oleic acid (C18:1n9c) have the highest ratios in cows' sweat (ANİTAŞ & GÖNCÜ KARAKÖK 2023, SAAT et al. 2005). The chemical structures of these fatty acids are presented in Figure 1.

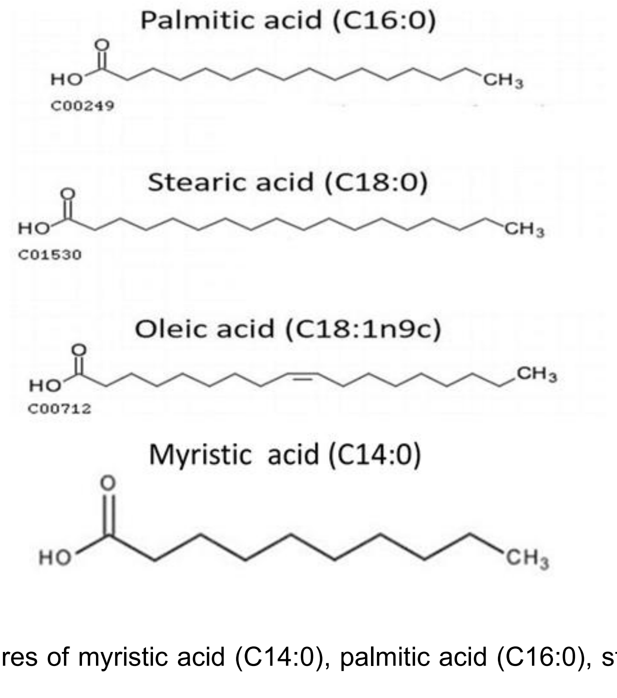

Figure 1. Chemical structures of myristic acid (C14:0), palmitic acid (C16:0), stearic acid (C18:0) and oleic acid (C18:1n9c).

Since heat stress is an event that causes an increase in the body temperature of animals and thus evokes some physiological reactions in the body, sweating plays an important role in the physiological coping strategies of dairy cows under this condition. This study was conducted to determine whether there are differences in physiological coping strategies by determining the ratios of myristic acid (C14:0), palmitic acid (C16:0), stearic acid (C18:0) and oleic acid (C18:1n9c) in sweat samples taken under heat stress from Holstein and Jersey breed cows housed in the same region and under the same conditions by gas chromatography.

MATERIALS AND METHODS

Experimental animals and area:

In this study, Holstein and Jersey cows in Cukurova University Faculty of Agriculture Dairy Research and Production Unit were used. For the study, sweat samples were obtained from 4 Holstein and 4 Jersey cows (total n: 8). Healthy cows with similar characteristics (Weight 600-670 kg, body condition score between 2.5 and 3, 3-5 years old, having given birth at least once, no reproductive problems, postpartum 45-60 days) were used. It was determined by the enterprise veterinarian as a result of anamnesis information and clinical examinations that there was no disease in the animals used in the study.

Sweat samples from animals were collected without harming the animals in accordance with the Animal Welfare Rules. There are 150 dairy cows on the farm, 40 of which are healthy and have similar characteristics. In order to determine the number of animals to be used in the study, it was taken into account that the smallest sample size to represent this population was 5% at the 5% confidence interval. Thus, since the number of dairy cows to be used in the research was determined as 10% (=40/0.10) of the total cows, it was sufficient to take samples from 4 cows.

The cows on the farm were fed a total mixed ration (TMR) (ratio of concentrate to roughage = 60:40). The TMR consisted of corn silage, lucerne and concentrate (18% crude protein and 2,650 kcal/metabolisable energy (ME) per kg) and was fed at 7 am and 4 pm.

Measuring of a temperature-humidity index

Adana was a province located in the eastern Mediterranean part of the Mediterranean Region in southern Turkey, with an altitude of 23 m above sea level and a latitude range of 35-38 latitude and longitude range of 34-46 east. During the study period, the maximum ambient temperature ranged between 35 °C and 37 °C during the day and the relative humidity was between 37% and 49% (Table 1).

The temperature humidity index (THI) was calculated according to the following equation 1.

$$THI = [(0.8x\ Temperature\ °C) + ((\%\ Relative\ Humidity\ /100)\ x\ (Temperature\ °C - 14{,}4)) + 46.4)] \quad \text{Eq. 1}$$

(BROUCEK et al. 2006).

The THI value for dairy cattle will be evaluated as indicated in MORAN's (2005) study (THI 72-78 was moderate stress, 79-89 was severe stress and 90-98 was very severe stress).

THI values calculated during the weeks of the experiment are given in Table 1.

Table 1. Ambient temperature (°C), relative humidity (%) and THI values.

	Ambient temperature (°C)			Relative humidity (%)			THI		
	Average (x̄± Sx)	Minimum	Maximum	Average (x̄± Sx)	Minimum	Maximum	Average (x̄± Sx)	Minimum	Maximum
Week 1	35.86 ± 0.69	35	37	44.86 ± 3.02	40	49	84.39 ± 1.58	81.06	85.94
Week 2	34.86 ± 0.69	34	36	49.57 ± 3.51	46	56	83.92 ± 1.34	81.06	85.14
Week 3	36.29 ± 0.49	36	37	42.00 ± 3.42	37	46	84.50 ± 0.51	83.84	85.14
General	35.67 ± 0.62	34	37	45.48 ± 3.31	37	46	84.27 ± 1.14	81.06	85.94

THI values were calculated in the study. For this purpose, indoor temperature (°C) and relative humidity (%) values were determined using HOBO Data Loggers placed in the chambers of each experimental group (Figure 2).

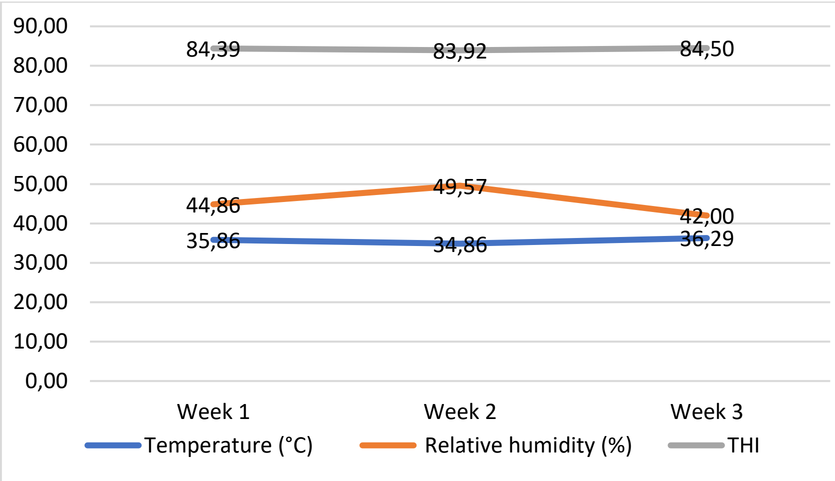

Figure 2. Temperature, relative humidity and THI values on the days of the experiment.

In this study, the average daytime temperature was 35,67 °C, relative humidity was 45.48% and THI value was 84.27 during the experimental week. Table 1 shows that the THI values were between 79-89 (MORAN 2005) during the three weeks of the study and therefore the animals were in stressful environments.

Data and sweat sample collection:

About an hour before to the trial, the animals were brought to the animal processing facilities unit in order to collect sweat samples. Animals were given sweat samples in accordance with KENNEDY's (2011) study methodology. The animals' nose regions were washed with water and patted dry with a paper towel prior to sampling. The animal's snout was used to collect sweat samples, which were then placed in 20 ml glass containers and kept in the freezer at -20°C until the analysis day (KUMAR et al. 2000). The samples were taken out of the freezer and allowed to defrost in the refrigerator the day prior to examination. The fatty acid content of was examined using gas chromatography (GC) in the laboratories of Çukurova University Faculty of Fisheries.

Analysis of Samples Using GC

The method used by BLIGH & DYER (1959) for lipid extraction analysis in sweat samples of Holstein and Jersey animals was applied. As per ICHIHARA et al. (1996), 10 mg of extracted oil was diluted in 2 ml of hexane, and 4 ml of 2M methanol-KOH was added to produce methyl esters from sweat analysis through trans-methylation with 2M KOH in methanol and n-hexane. After two minutes at ambient temperature, the tube was vortexed to fully combine the liquid. The sample was moved to a hexane layer for GC analysis following centrifugation at 4000 rpm for 10 minutes.

The fatty acid profile (30 m × 0.32 mm ID 0.25 lm BP20 0.25 UM, USA) was analyzed using a Clarus 500 GC autosampler (Perkin-Elmer, USA) fitted with a flame ionization detector and fused silica capillary SGE column. For five minutes, the oven's temperature was maintained at 140 °C. Next, it was raised to 200°C at a rate of 2 °C per minute, and it was maintained there at 220 °C at a rate of 1 °C per minute. The injector was operated at 220 °C, while the detector was operated at 280 °C. A single µl of material was used, and the carrier gas was regulated at 16 ps. A 1:100 injection ratio was used. Fatty acid peaks were found by comparing the FAME retention time with the FAME mixture's standard component. The findings are shown as means ± standard error in % GC area after three repeated GC analyses.

Statistical analyses:

Statistical analyses were performed using SPSS (Statistical Package Program for Social Science) 20.0 package program by determining fatty acids from sweat taken from animals. Data were expressed as mean ± standard deviation (SD). After variance analysis (ANOVA) was applied to the collected data, correlation analysis was applied to determine the relationship between breed and fatty acids using the significance level of $P < 0.05$. Pearson Correlation Coefficient provides information about the strength and direction of the linear relationship between two variables specified by measurement. It takes values between -1 and +1. The strength of the relationship decreases as it approaches 0, and the strength of the relationship increases as it approaches +1. The confidence interval in the analyses was determined as 95% (significance level $P < 0.05$).

RESULTS

In the study, fatty acids were analysed using gas chromatography on sweat samples collected from Holstein and Jersey cows subjected to heat stress in summer conditions. As a result of the analysis, the highest percentages of fatty acids detected in Holstein and Jersey sweat are shown in Table 2.

Table 2. Sweat fatty acid composition of Jersey and Holstein cows.

Fatty acids	Sweat fatty acids (%)						P-value
	Holstein cows			Jersey cows			
	Minimum	Maximum	Average ($\bar{x}$± Sx)	Minimum	Maximum	Average ($\bar{x}$±Sx)	
Myristic acid (C14:0)	8.76	9.80	9.28 ± 0.74	8.16	12.21	10.19 ± 2.86	0.000*
Palmitic acid (C16:0)	26.29	26.19	26.24 ± 0.07	23.65	24.00	23.83 ± 0.25	0.000*
Stearic acid (C18:0)	10.99	11.86	11.43 ± 0.62	28.75	26.50	27.63 ± 1.59	0.448
Oleic acid (C18:1n9c)	28.33	24.45	26.39 ± 2.74	20.38	21.28	20.83 ± 0.64	0.464

The ratio are shown as the mean ± standard deviation (SD) and * indicate statistically significant differences ($P < 0.05$).

The rates of the 4 fatty acids with the highest percentage in the sweat of animals under heat stress in summer, myristic acid (C14:0), palmitic acid (C16:0), stearic acid (C18:0) and oleic acid (C18:1n9c), were shown in Table 2. Among these fatty acids, myristic acid, palmitic acid and stearic acid were saturated fatty acids. Oleic acids were monounsaturated fatty acids. In Table 1, the total saturated fatty acid rate in sweat was determined as 46.95% in the sweat of Holstein cows and 61.65% in the sweat of Jersey cows. In addition, it was determined that the saturated fatty acid ratios were higher in the sweat of Jersey cows, and the monounsaturated fatty acid ratio was higher in the sweat of Holstein cows (Figure 3).

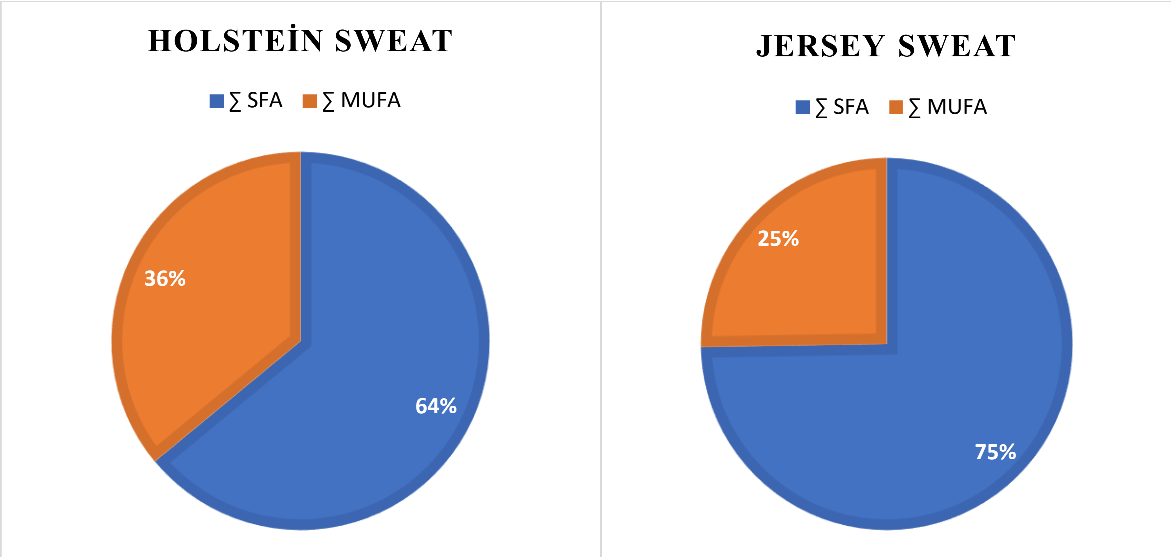

Figure 3. ∑SFA and ∑MUFA ratios in Holstein and Jersey sweat under heat stress.

The total percentage of fatty acids with the highest percentage detected in sweat, especially in cases of heat stress, was 73.34% in Holstein cows and 82.48% in Jerseys. Apart from myristic acid, palmitic acid, stearic acid and oleic acid, other compounds detected in sweat constitute 26.66% in Holstein and 17.52% in Jersey (Figure 4). The 4 fatty acids detected in sweat in Table 2 constitute more than 3/4 of the total fatty acids (Figure 4).

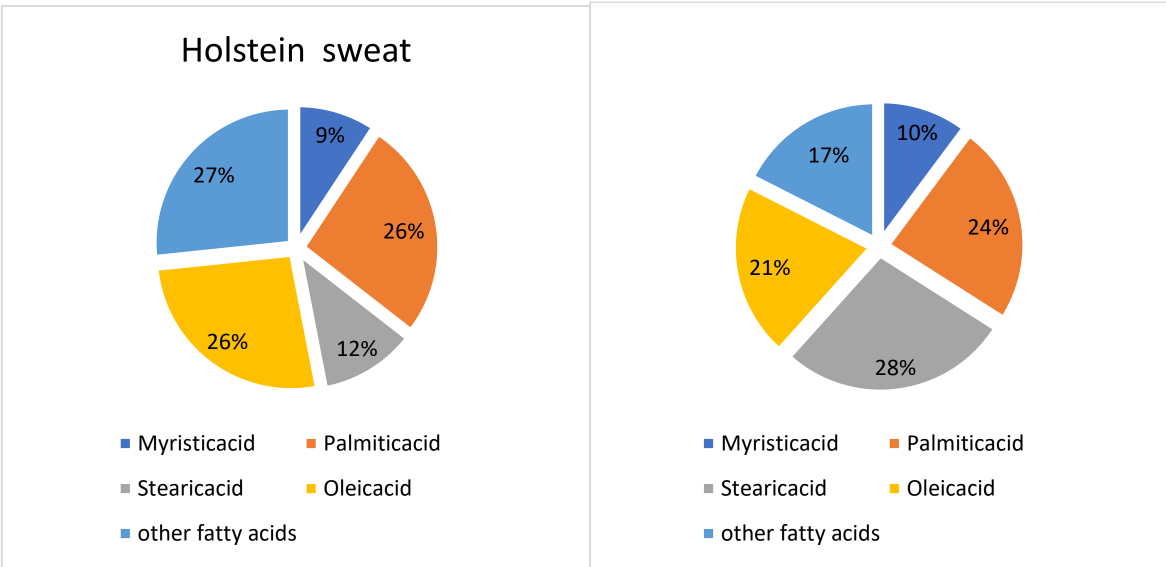

Figure 4. Fatty acid ratios of Holstein and Jersey sweat under heat stress.

Table 2 shows that the fatty acid with the highest rate in the sweat of Holstein cows was oleic acid, and in the sweat of Jersey cows it was stearic acid. Palmitic acid and oleic acid ratios were detected to be higher in Holstein cows, and myristic acid and stearic acid ratios were detected to be higher in Jersey cows (Figure 5).

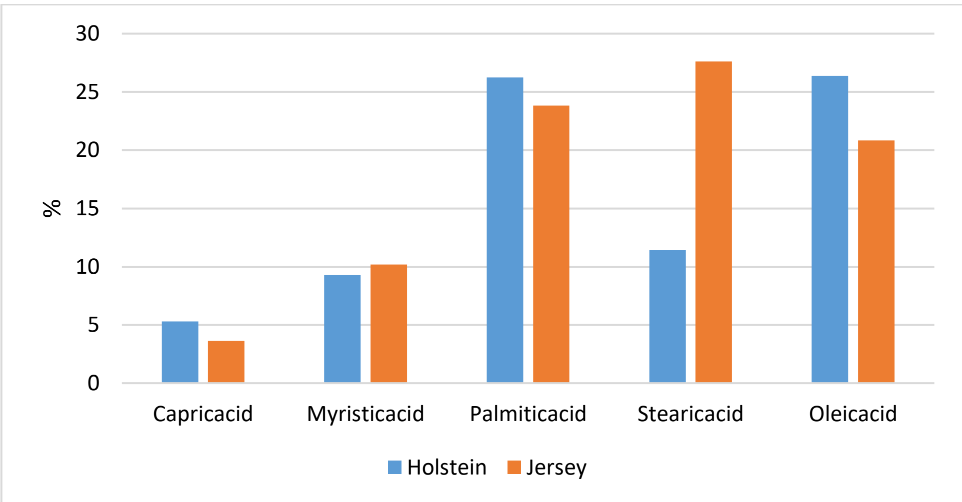

Figure 5. Sweat fatty acid composition of Jersey and Holstein.

When these 4 fatty acids were examined, Table 2 shows that the highest difference in percentage rates between the Holstein and Jersey was in stearic acid.

The retention times of fatty acids detected on Holstein and Jersey cows in the chromatogram were shown in Figure 6.

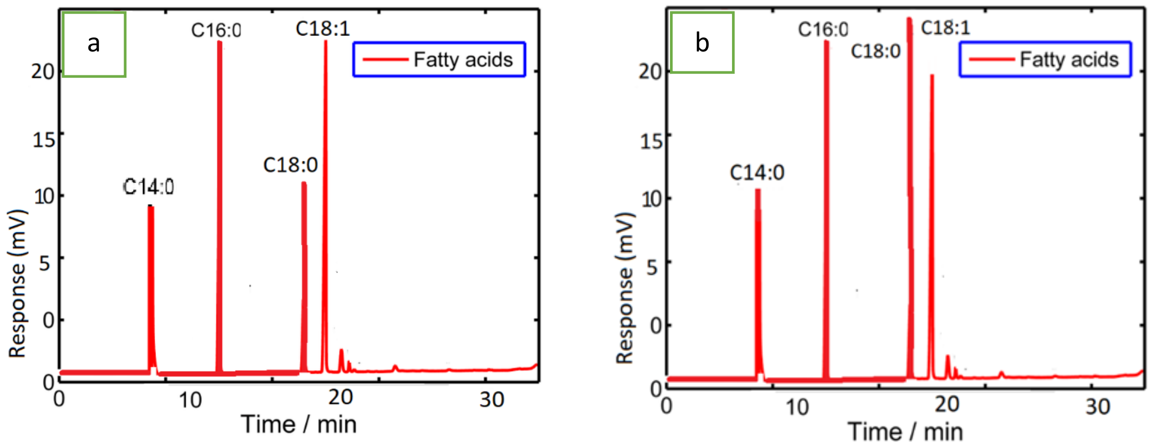

Figure 6. Identification chromatogram of Holstein (a) and Jersey (b) sweat fatty acids.

When the retention times of the samples after being injected into the device were examined in Figure 6, it was seen that the fatty acids detected in the sweat of both Holstein and Jersey cows were similar. Myristic acid (C14:0) reached its peak at the 9th minute, palmitic acid (C16:0) at the 14th minute, stearic acid (C18:0) at the 18th minute and oleic acid (C18:1n9c) at the 19th minute.

In the study, the relationship between sweat fatty acids of Holstein and Jersey cows in summer months was determined by correlation analysis. The results of the correlation analysis were shown in Table 3.

Table 3. Correlation between sweat fatty acids of Holstein and Jersey cows in summer months.

Breeds- Fatty acids	**Pearson r**	**0.757****
	P	0.000

**r: Correlation is significant at the 0.01 level (2-tailed). P significance

As a result of correlation analysis in the study, a strong positive (r=757) and significant (P <0.01) relationship was found between sweat fatty acids of Holstein and Jersey breed cows in summer months (Table 3). This correlation was shown in Figure 7.

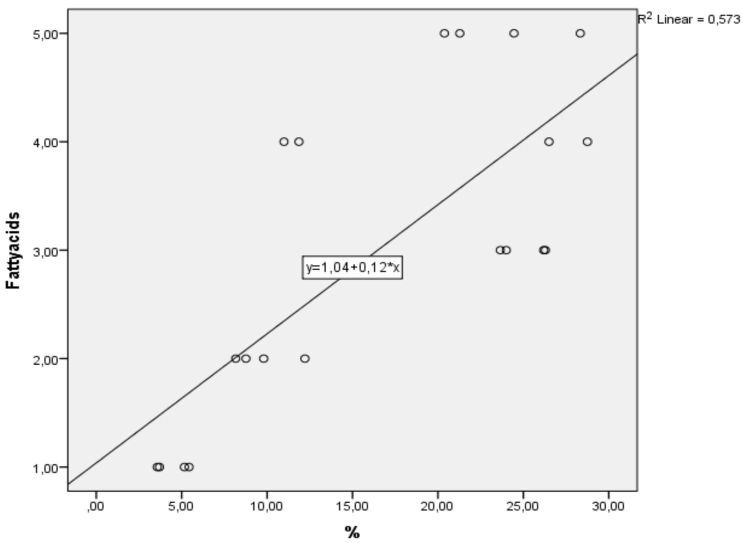

Figure 7. Correlation graph between sweat fatty acids of Holstein and Jersey cows in summer.

DISCUSSION

Sweating was the most important body temperature regulation mechanism. This mechanism reduces body temperature, but, some substances are also expelled from the body (ATRIAN & SHAHRYAR 2012). SRIKANDAKUMAR & JOHNSON (2004) stated that blood pH, blood pCO_2 (kPa), bicarbonate (HCO_3, mmol/L) and plasma chloride (Cl-, mmol/L) levels of Holstein and Jersey cows change under heat stress. The fact that fatty acid ratios in sweat at different levels among breeds in our study was consistent with the study of SRIKANDAKUMAR & JOHNSON (2004). Since it was known that fatty acids released into sweat through the blood (CHEN et al. 2020), the variation in fatty acid ratios between Holstein and Jersey in Table 2, and especially the differences in fatty acids such as stearic acid, showed that breeds respond differently to heat stress.

The body's compensatory physiological responses to heat stress include increases in metabolism, blood flow to the skin, and sweating rate (ESTEVAO et al. 2024). During this period, the body's thermoregulation mechanisms are also negatively affected (PÉRIARD et al. 2022). One of these mechanisms is that the physicochemical properties of fatty acids, which evoke some physiological reactions in the body and are used for various tasks, are affected by high temperatures (ESTEVAO et al. 2024). When the fatty acid ratios of Holstein and Jersey cows housed under the same conditions were examined in the study, it was seen that the fatty acid ratios detected in the sweat of Jersey cows, especially stearic acid, were released at high levels in sweat, as stated by ESTEVAO et al. (2024), and the thermoregulation mechanisms in the body of Jersey cows were more affected than those of Holstein cows.

Studies have shown that oleic acid (C18:1n9c) from the MUFA group has the highest environmental impact and is less heritable compared to other fatty acid traits (DAURIA et al. 2022). In addition, C18:1 is supported by previous findings that it shows the highest changes in variance components at high THI values (TESSARI et al. 2020). Recent studies have determined that C18:1 should be used as a biomarker for early diagnosis of metabolic diseases such as negative energy balance in plasma and other body fluids (CHURAKOV et al. 2021, TESSARI et al. 2020). In the current study, the different sweat oleic acid ratios of Holstein and Jersey cows indicate that their responses to the negative energy balance in the body during heat stress are different.

NUNOME et al. (2010) showed that total FA concentrations in sweat also increased with increasing fasting duration and that FAs in sweat released into sweat via the blood from the triacylglyceride (TG) of adipose tissue as an energy source during longer fasting periods. Therefore, in our study, the FA concentration, especially the stearic acid ratio, in the sweat of Holstein and Jersey cows can be considered a promising indicator of TG levels in the blood. In Table 2, the fact that the rate of stearic acid in the sweat of Holstein cows was lower than that of Jersey cows shows that triacylglycerides from the fat tissue of Holstein cows were not used much as an energy source in case of heat stress (NUNOME et al. 2010).

DELGADO-POVEDANO et al. (2018)'s finding that they detected the most saturated fatty acids in post-exercise sweat was comparable with our study. When the fatty acids with the highest percentage in sweat were examined in our study, it was determined that myristic acid, palmitic acid and stearic acid (C18:0) were saturated fatty acids, and only oleic acid was unsaturated fatty acid (Table 2). Additionally, it can be seen in Table 2 that the percentage of total saturated fatty acids was higher than that of unsaturated fatty acids. Since heat stress is an event that causes the body temperature of animals to increase and therefore evokes some physiological reactions in the body, sweating plays an important role in the physiological coping strategies of dairy cows under this situation. When total saturated fatty acid levels were examined in Holstein and Jersey housed under the same conditions in the same region, the higher percentage in Jersey cows showed that there were differences in the physiological coping strategies of both breeds (Table 2).

It has been determined that monounsaturated fatty acids (MUFA) can change plasma lipids and lipoprotein composition and therefore reduce inflammation, oxidative stress and improve blood pressure (PENG et al. 2011, BERMUDEZ et al. 2011). Since the oleic acid detected in sweat in our study from the MUFA group, its ability to regulate blood pressure and reduce oxidative stress, especially in animals under heat stress, is important. Table 2 shows that oleic acid (C18:1n9c) was detected at a rate of 26.39% in Holstein cows and 20.83% in Jersey. The study shows that Holstein cows use more oleic acid in their bodies during heat stress (Figure 4). Fatty acids in the tissues of animals were stored as triglycerides. Especially in energy-requiring situations such as hunger and stress, fatty acids are released into the blood from triglycerides in the fat tissue as an energy source (NUNOME et al. 2010). In the study, it was observed that Jersey cows (82.48%) used more fatty acids in their tissues than Holstein cows (73.34%) under heat stress (Table 2).

LOFTEN et al. (2014) stated in their study that stearic acid (C18:0) did not accumulate in the tissues of cows with negative energy balance and that cows metabolized stearic acid for energy in the liver and muscles. They also stated that a large amount of stearic acid was secreted through milk, both as stearic acid and oleic acid (C18:1n9c). The change in sweat fatty acid ratios released from animals in cases of heat stress was important for the animal's body temperature regulation (LOFTEN et al. 2014). In the study, stearic acid, which was detected especially in the sweat of Jersey cows, was excreted from the body at higher rates (27.63%) than in the sweat of Holstein cows (11.43%). In this case, it was shown in Table 2 that the Jersey metabolizes more stearic acid for energy in the liver and muscles as a body temperature regulation during heat stress.

Although one of the most important known roles of sweating is to dissipate increased body temperature, it also has other homeostatic functions. (BAKER 2019). For example, sweat glands, similar to the kidney system, also play an important role in excretory functions responsible for clearing excess micronutrients, metabolic wastes and toxic substances in the body (BAKER 2019). Since it was known that fatty acids necessary for the body were released from triglycerides in adipose tissues during heat stress, the higher levels of fatty acids (especially stearic acid) in the sweat of Jersey cows in the study indicate that they were used at a higher rate in adipose tissue and the metabolic wastes were excreted through sweat (Table 2).

The ability to regulate increased body temperature is an evolutionary adaptation that allows mammals to maintain (at least to some extent) their biological functions despite environmental temperature fluctuations. (SILANIKOVE 2000). Environmental temperatures have risen by 0.2 to 0.6 °C since 2000 and were expected to continue to rise by a further 5.8 °C by the end of the century (IPCC 2007); it was therefore predicted that more and more cows will be exposed to heat stress and it will become increasingly important to exploit the animal's natural ability to regulate heat and its morphological differences. For this reason, the adaptation performance of Holstein and Jersey cows raised in Adana region, which was very hot and humid in the summer season (Table 1), is important. Stearic acid (C18:0) was found 11.43% in the sweat of Holstein cows and 27.63% in Jersey cows, indicating that the two breeds exhibit different adaptation responses under the same conditions at the same temperature and humidity (Table 2).

High environmental temperatures may affect rumen microorganisms that synthesize B vitamins, amino acids and fatty acids, on which ruminant nutrition depends to a great extent. In the present data, a strong positive and significant ($P <0.01$) relationship was found between sweat fatty acids in Holstein and Jersey cows in summer months (Table 3), indicating that heat stress affects rumen microorganisms that synthesize fatty acids in both breeds (DUFFY et al. 2019).

CONCLUSION

The highest rate fatty acid compositions of sweat from Jersey and Holstein dairy cattle were recorded under summer conditions. Changes in fatty acid ratios in the sweat of Holstein and Jersey cows indicate that the animal breeds respond differently to heat stress. Fatty acids in sweat reach through blood from triacylglycerides in adipose tissue as an energy source during prolonged stress. Oleic acid (C18:1n9c), which has important functions such as regulating blood pressure and reducing oxidative stress especially in heat stressed animals, was found to be higher rate in Holstein cows. In addition, stearic acid (C18:0), which was metabolized for energy in the liver and muscles and released through sweat under heat stress, was excreted at very high rates in the sweat of Jersey cows, indicating that it is important for the ability to regulate body temperature of the animal. Stearic acid ratios detected in the sweat of Holstein and Jersey cows can be used in other studies as a marker to determine heat stress conditions.

AUTHOR CONTRIBUTIONS

Conceptualization, methodology, and formal analysis, Özgül ANİTAŞ, Yeşim ÖZOĞUL, Fatma HEPSAĞ and Serap GÖNCÜ; software and validation, Yeşim Özoğul and Özgül ANİTAŞ; investigation, Özgül ANİTAŞ, Serap GÖNCÜ and Fatma HEPSAĞ; resources and data curation, Fatma HEPSAĞ and Yeşim Özoğul; writing-original draft preparation, Özgül ANİTAŞ, Yeşim Özoğul and Serap GÖNCÜ; writing-review and editing, Yeşim ÖZOĞUL, Fatma HEPSAĞ and Özgül ANİTAŞ; visualization, Yeşim ÖZOĞUL and Serap GÖNCÜ; supervision, Özgül ANİTAŞ, Fatma HEPSAĞ and Serap GÖNCÜ; project administration, Serap GÖNCÜ, Özgül ANİTAŞ and Yeşim ÖZOĞUL; funding acquisition, Serap GÖNCÜ Yeşim ÖZOĞUL, Özgül ANİTAŞ and Fatma HEPSAĞ. All authors have read and agreed to the published version of the manuscript.

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DATA AVAILABILITY STATEMENT
The data can be made available under request.

CONFLICTS OF INTEREST
The authors declare no conflict of interest.

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