The influence of different stress factors on the production and quality of sheep milk

DOI: 10.15567/mljekarstvo.2024.0301

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Received: 25.01.2024. Accepted: 15.06.2024.

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Abstract

Around 10.5 million tonnes of sheep milk are produced worldwide, and around 3.1 million tonnes in Europe. In Europe, sheep milk production has stagnated slightly in recent years and currently accounts for around 1/3 of the global production. Due to its high content of dry matter, milk fat, total protein and casein, sheep milk is an excellent raw material for cheese production. Milk processing and cheese making are not only economically important, but also part of the cultural heritage based on a long tradition, often taking place in rural and less developed areas. Milk production and quality are very complex and are influenced by numerous factors, the most important of which are breed, feeding and body condition, parity and stage of lactation, milking and udder health, and environmental factors. In addition, different types of stress such as heat stress, nutritional stress, weaning stress, shearing stress, stress of regrouping, etc. have a significant impact on the welfare and health of the sheep as well as on the quantity of milk produced, its chemical composition, processing characteristics and the economic efficiency of the flock. In recent years, due to climate change, there has been an increasing number of studies on the effects of heat stress on the reproductive and production characteristics of sheep, as well as on their welfare and health. In addition, heat stress is often associated to the feed and the lack of water. The effects of stress are particularly pronounced in extensive sheep farming and milk production, which are prevalent in sheep farming. The aim of this paper is to describe the effects of the main types of stress on the quantity and chemical composition of sheep milk and on animal welfare.

Keywords: sheep; stress; milk production; chemical composition; welfare

Introduction

Sheep breeding is a very important economic sector in many countries around the world, especially in areas where the possibilities for organising agricultural production and breeding other animal species are limited due to specific geographical, orographic, climatic and vegetation conditions. In addition to their economic and nutritional role, sheep are very important for the preservation of tradition, the maintenance of the landscape and biodiversity. Due to their behavioural characteristics and high adaptability to the environment, sheep have been kept for many centuries for the production of meat, milk, wool and skin (Ruiz-Larranaga, 2018). Although meat (lamb) is the most important sheep product in the world, in many countries the production of sheep milk is significantly more profitable than the production of meat and wool (Bobokulovich and Kasimovna, 2021). The importance of milk production is obvious, as it is a staple food for young animals and has economic and nutritional importance. Sheep milk is an interesting and economically important product, especially in the Mediterranean region (Turkey, Greece, Spain, Italy and France), where around 45 % of the world's sheep milk is produced (Pulina et al., 2018). It is characterised by a significantly higher content of dry matter, milk fat, proteins (casein), minerals and water-soluble vitamins and has a higher energy value than cow's and goat's milk (Balthazar et al., 2017). Due to the high content of the above-mentioned ingredients, especially milk fat, proteins and casein, sheep milk is very suitable for cheese and yoghurt production (Haenlein, 1998). Looking globally, especially in Europe, sheep milk is primarily processed into cheese, many of which are protected by a specific label of origin (Pulina et al. al., 2018).

In the Mediterranean region the climate is characterised by hot summers and cold winters. Even under these conditions, the temperature often deviates from the thermoneutral zone (Curtis, 1983) for sheep (from 5 to 25 °C) and even from that given by Taylor (1992) for adult sheep whose bodies are covered with fleece (from -12 to 32 °C). Temperatures outside the thermoneutral zone can lead to physiological changes and have a negative impact on livestock production (Sejian et al., 2018). Sheep milk production is very demanding and the amount of milk produced and its chemical composition are influenced by a variety of factors, the most important of which are breed (Antunac and Lukač Havranek, 1999; Mioč et al., 2004; Marques et al., 2011), feeding (Ceyhan et al., 2022; Caraba and Caraba, 2023), age of the sheep and lactation order (Gabiña et al., 1993; Libis-Márta et al., 2021), stage of lactation (Antunac et al., 2001; Pavić et al., 2002.; De La Fuente at al., 2009), season (Timlin et al., 2021; Li et al., 2022), physical development and condition (Caraba and Caraba, 2023; Simeonov et al., 2023), housing system (Sevi et al., 1999; Casamassima et al., 2001), milking (Mačuhová et al., 2020; Massouras et al., 2023), etc. and their interactions (De La Fuente at al., 2009; Kawecka et al., 2020; Kasapidou et al., 2021.). Environmental factors

such as the air temperature in the sheep facilities, relative humidity, wind speed, solar radiation and rainfall should not be ignored as they also have a major influence on milk production (Silanikove, 2000) and, if they deviate too much from the optimum values, can be a source of stress for the ewes. Stress is a reflex reaction of animals in a harsh environment and leads to unfavourable consequences ranging from discomfort to death (Das et al., 2016). Heat stress significantly impairs sheep reproduction and poses a significant risk to the efficiency of meat and milk production under current climate conditions, with the effects increasing as global temperatures rise (van Wettere et al., 2021). One of the most important characteristics of sheep is their resistance and adaptability, which is becoming increasingly important under conditions of global climate change and food and water scarcity. However, it is believed that the resistance of sheep to heat stress without its negative impact on animal welfare, health and productivity is often overestimated (Al-Dawood, 2017).

Stress, especially heat stress, is an increasingly important limiting factor in sheep breeding and modern sheep production in many regions of the world. It has a direct effect on the amount of milk produced and the synthesis of its main components. The Mediterranean is one of the most famous regions in the world for the production of sheep milk and cheese. Unfortunately, a significant increase in temperature is also expected in this area (Pasqui and Di Giuseppe, 2019), which will have a direct impact on this sector. Al-Saiady (2006) states that the influence of heat stress on milk production is more pronounced in older ewes and those with a higher milk yield. Therefore, knowledge and a better understanding of heat stress will contribute to the development of new technologies to mitigate its effects on sheep health and productivity (Schütz, 2022). Cold stress can also affect milk yield in sheep, but this is much less common than in animals that have experienced heat stress (Ravagnolo and Misztal, 2000; Endris and Feki, 2021).

Drought is another factor that can cause stress in sheep breeding as it has a direct impact on the yield and quality of pasture and thus on the production efficiency of sheep in milk production. In addition to nutrition, artificial weaning is one of the most stressful periods in the life of lambs and sheep, with negative consequences on milk production profitability that should not be disregarded. The aim of this paper is therefore to describe the most pronounced types (factors) of stress and their effects on milk yield, welfare and health of sheep.

Number of sheep and milk production in the world and in Europe

Sheep are primarily used for the production of food (meat and milk) with high nutritional value and other economically interesting products (wool, leather, fur, manure, etc.). The

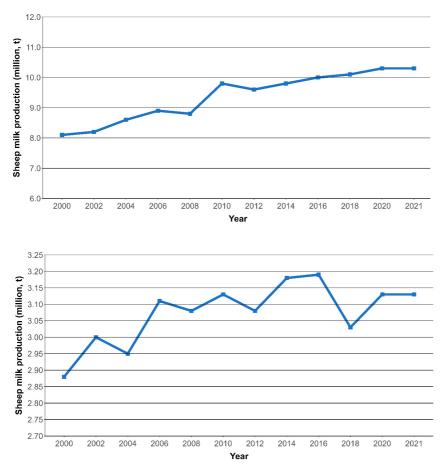


Figure 1. Sheep milk production in the world from 2000 to 2021 (FAOSTAT, 2000-2021)

Figure 2. Sheep milk production in Europe from 2000 to 2021 (FAOSTAT, 2000-2021)

close connection to nature (environment), the preservation of biodiversity and the sustainability of the landscape are also key characteristics of sheep (Zygoyannis, 2014). They are suitable for grazing and maintaining pastures in marginal and inaccessible terrain with sparse vegetation, where they are very effective in converting grazing land into high-value products (meat, milk, wool, etc.). Around 1.28 billion sheep are kept worldwide, producing about 10.4 million tonnes of milk annually (FAOSTAT, 2021). However, the actual production of sheep milk is much higher, as the amount that the lambs suck is not taken into account, and often also the milk that is processed and consumed on the farm. In the last twenty years, the number of sheep worldwide has increased by about 220 million (20.6%), while in Europe the total number of sheep has decreased by 27 million heads or 18 % (FAOSTAT, 2000 and 2021). The largest sheep population and the largest sheep milk production is in Asia, which produces 46.61 % of the world's sheep milk with 44.46 % of sheep. Europe is producing 29.74 % of the world's total milk with only 9.47 % of the world's sheep population. The breeding of dairy sheep is mainly concentrated in the Mediterranean and Black Sea regions, focusing on the Greek or Roman cultural heritage (Caja, 1990), where sheep milk products have been an indispensable part of the human diet for centuries. Over the last twenty years, the production of sheep milk worldwide has increased by about two million tonnes or about 26 % (FAOSTAT, 2000; 2021) and continues to show an upward trend (Figure 1), while in Europe it is about 250,000 tonnes or about 8.6 % higher, with indications of slight stagnation.

The largest producers of sheep milk in the world are China (12.13%) and Turkey (10.89%). In China and Turkey, this production has increased by 427,000 tonnes (50.40 %) and 369,000 tonnes (47.70 %) respectively in the last twenty years (FAOSTAT, 2000, 2021). In Europe, the largest producers of sheep milk are Greece, Spain, Italy, Romania and France. Eight European countries (Greece, Spain, Italy, Romania, France, Portugal, Albania and Bulgaria) together produce about 95 % of European sheep milk, with Greece being the largest producer with one third of European sheep milk and the highest average production per ewe (about 131 kg). The importance of this sector in Greece can be seen from the fact that it accounts for 45 % of the total value of livestock production (Pridis et al., 2012). Most of the sheep milk produced in Europe is processed into high-quality cheeses, many of which are protected by PDO (Protected Designation of Origin): Feta, Pecorino Romano, Roquefort, Manchego, Bryndza Podhalańska, Serpa and others (Pulina et al., 2018: Kawecka et al., 2020: Araújo-Rodrigues et al., 2020; Pappa et al., 2023; Garzón et al., 2023). Milk processing and the production of autochthonous cheeses are not only economically important, but also part of the cultural heritage, which is based on a long tradition and is often organised in rural and less developed areas (Freitas and Malcata, 2000). Italy is the leading exporter of sheep milk cheese with a market share of 36 %, followed by France (20 %). On the other hand, the United States and Germany are the main importers, accounting for 42 % and 41 % of the total sheep milk cheese imports respectively (FAOSTAT, 2018). However, despite the long tradition and the production of high-quality cheeses that are recognised and soughtafter on the market, the number of dairy sheep in Europe is declining every year, which is reflected in the production of sheep milk (Figure 2).

The slower increase in sheep milk production in Europe compared to global production mentioned above is the result of a notable decrease in production in Italy for 265,000 tonnes or 35.7 %. On the other hand, sheep milk production increased significantly over the same period in Spain, France, Romania, and Greece, for 42.92 %, 40.90 %, 30.83 % and 28.05 %, respectively.

Heat stress

Climate change is a major threat to the sustainability of livestock farming around the world, particularly in areas with arid climates where sheep (along with goats) are the dominant, most productive, economically profitable and ecologically important domestic species. According to projections by the Intergovernmental Panel on Climate Change (IPCC), global warming will increase temperatures by 1.5 °C to 2 °C in the 21st century. Consequently, the heat stress will probably continue to have a negative impact on the sustainability of livestock production worldwide (Ciliberti et al., 2022), including the reduced pasture productivity, a lack of nutritious feed, a violation of animal welfare and high energy costs for cooling (Ortiz-Colón et al., 2018). As stated by Collier et al. (2017), the thermal environment is the greatest stress factor affecting the efficiency of animal production systems, impacting the development, growth, reproduction and production of all animals. When temperatures fall below 12 °C (lower critical temperature) or rise above 25 to 31 °C (upper critical temperature), thermoregulation mechanisms are seriously impaired and the ability of sheep to maintain homeothermy is reduced. Under these conditions the thermal effects on sheep performance and welfare are most severe. However, the physiological and behavioural adaptations that enable sheep to maintain homeothermy have negative effects on their growth, productivity, welfare and reproduction long before these temperatures are reached (Das et al., 2016). These adaptations include increased heat dissipation and reduced metabolic heat production through peripheral vasodilation and reduced food intake. The prolonged heat stress also leads to endocrine adaptations that further reduce metabolic rate and promote heat dissipation (Silanikove and Koluman, 2015). These changes can further impact production and reproduction by reducing foraging time and/or increasing the distances sheep travel to find food and water. Behavioural responses to heat stress include a reduction or temporal shift in activity to cooler parts of the day and increased use of shade (Al-Dawood, 2017), while the main behavioural reaction of animals are an increase in respiratory rate, rectal temperature and heart rate. Dairy breeds are generally more sensitive to heat stress than meat breeds, and higher producing animals are also more susceptible as they generate more metabolic heat (Das et al., 2016).

Climatic factors: temperature, relative humidity, solar radiation and wind speed have a direct influence on the availability and quality of feed, welfare, disease incidence and production efficiency of the animals (Joy et al., 2020). Climatic changes with high summer temperatures and large temperature fluctuations are considered negative factors for the production efficiency of sheep (Joy et al., 2020). Therefore, heat stress is one of the limiting factors for milk production in hot climates (Johnson et al., 1962), which is particularly difficult to avoid in the prevailing extensive sheep production systems. Considering that extensive production systems are based on grazing, climate change and rising temperatures will affect vegetation growth and water availability (Nardone et al., 2010). Therefore, under the conditions of global climate changes, the selection of a breed and a suitable breeding system is a very important strategy for sustainable livestock production in the future (Baumgard et al., 2012). In the Mediterranean region, most sheep graze on pastures in summer and are exposed to direct sunlight and high air temperatures, which is reflected in the amount of feed and water consumed and the quantity and quality of their products (Shilja et al., 2016). High environmental temperatures affect the productivity of lactating sheep by increasing their energy requirements for maintenance by 7 to 25 %, partly due to accelerated respiration (Sevi and Caroprese, 2012). However, the mechanisms by which heat stress negatively affects the metabolism and synthesis of milk in sheep are not well defined (Mehaba et al., 2021). Under heat stress, the combination of reduced food intake and higher energy requirements stimulates the mobilisation of body reserves from adipose tissue and body proteins to provide amino acids for protein synthesis and carbon sources for gluconeogenesis (Rhoads et al., 2011). Studies on the influence of heat stress and prolonged drought on milk production in sheep and their chemical composition are not standardised, which makes it difficult to interpret and compare their results. The studies on this topic largely agree on the reduction in total milk production, a lower proportion of protein (especially casein) and fat in milk and an increase in the proportion of saturated fatty acids with a decrease in oleic, vaccinic, linoleic and linolenic acids as well as a significant reduction in the coagulability of milk (Sevi et al., 2002; Sevi, 2003; Nudda et al., 2005; Salama et al., 2014). This directly affects the quantity and quality of the cheese, which are reduced (Sevi and Caroprese, 2012). In addition, Finocchiaro et al. (2005) reported

a lower total milk yield, but without significant effects on the fat and protein content of the milk. The reduction in milk production can be up to 20 % when the air temperature in the sheep housing space rises above 35 °C (Sevi et al., 2001a). In contrast, Mehaba et al. (2021), for example, found that heat stress in sheep during lactation had no effect on total milk production, but decreased the fat and protein content of milk and increased the number of somatic cell count (SCC). It has also been found that feed intake in ewes decreased by 11 % and the rectal temperature increased by 0.77 °C, as did respiratory rate per minute by 90 and water consumption by 28 % (Mehaba et al., 2021). The content of milk fat, protein and the number of somatic cells have a significant influence on the processing properties of the milk, the firmness of the curd and the yield and quality of the cheese produced (Gonzalez-Ronquilloa et al., 2021).

High temperatures have been shown to have a negative effect on dairy traits in sheep, with the negative effects of temperature combined with high humidity being even more pronounced and further affecting sheep welfare and production efficiency. The combined effects of air temperature and humidity in conjunction with the degree of thermal stress are usually represented as a single in the temperature-humidity index (THI). Different animal species react differently to ambient temperature and humidity. This has led to the development of various approaches and formulae for calculating the temperature-humidity index, of which we have selected the one intended for sheep proposed by Marai et al (2001) expressed in degrees Celsius. The equation for calculating THI is:

THI = db °C - {(0.31 - 0.31 RH) (db °C - 14.4)}

where db °C is the dry bulb temperature (°C) and RH is the relative humidity (RH%)/100. The values of THI obtained indicate the following: < 22.2 = no heat stress; 22.2 to < 23.3 = moderate heat stress: 23.3 to < 25.6 = severe heat stress and 25.6 and above = extremely severe heat stress (Marai et al., 2001). Although the THI classes listed differ depending on the study and calculation formula, it is difficult to compare the absolute values of their results.

In general, it was found that the production efficiency of sheep decreases with increasing THI. For example, Finocchiaro et al. (2005) found that sheep of the Valle del Belice breed are affected by heat stress from a THI of 23, leading to a decrease in production yields. In addition, Sevi et al. (2001a) reported that the Comisana dairy sheep breed is affected by heat stress when THI is above 27, while the effects of heat stress were noticeable in Omani and Australian Merino sheep when THI was >32 (Srikandakumar et al., 2003). In addition, Finocchiaro et al. (2005) stated that yields tended to vary more between temperatures within the relative humidity category than between relative humidity categories within the same temperature class. They also indicated that high temperature is more important than high relative humidity when considering the relationship between heat stress and milk yield.

Nutritional stress

Although sheep are spread all over the world, they are the predominant domestic species in arid and semiarid areas where rainfall is a fundamental factor in the growth of vegetation (pasture) and the yield of food for sheep. Poor quality pasture is low in nitrogen (protein) and high in crude fibre, which limits food intake and digestibility and is the cause of animal malnutrition (Smith, 2002). An optimal diet can help animals adapt to poor environmental conditions by providing them with sufficient energy in times of heat stress or other negative environmental factors (Sejian et al., 2014). Feeding has a direct influence on the quantity and chemical composition of sheep milk (protein content, milk fat content and fatty acid composition), milk flavour (especially in highproducing animals) and the yield, quality and flavour of cheese (Pulina et al., 2006; Tüfekci and Sejian, 2023). For this reason, the relationship between feeding and milk quality is often evaluated by the technological and coagulation characteristics of milk, which are strongly influenced by the amount of milk fat and protein and the number of somatic cells (SCC) in sheep milk (Pulina et al., 2006). Sheep grazing on quality pastures have a positive influence on the flavour, antioxidant properties and fatty acid profile of cheese (de Renobales et al., 2012; Nudda et al., 2020). As most sheep are kept on pasture during the growing season, the animals are often exposed to nutritional stress due to fluctuations in the quantity and quality of pasture as a result of soil quality and various meteorological changes. The botanical composition of pastures can influence the quality, flavour and safety of sheep milk, and the above characteristics can be improved by changing the structure and guality of sheep ration (Pulina et al., 2006). The amount of energy in the sheep ration or energy balance is the most important factor influencing the fat content of sheep milk, especially in the first third of lactation when the energy balance is negative (Bocquier and Caja, 1993). This relationship between energy balance and milk fat content is most pronounced in sheep with a high milking capacity, while it is much less pronounced in animals with a low milking capacity (Cannas and Avondo, 2002). A negative energy balance leads to a strong mobilisation of body fat tissue, which is confirmed by the increased concentration of long-chain fatty acids (LCFA) in the blood, the proportion of which decreases with increasing body condition (Pulina et al., 2006). Diet influences the presence of conjugated linoleic acid (CLA) and other fatty acids (FA) in sheep milk. Acetate, which is produced by the fermentation of fibres in the rumen of sheep, is the starting material for the composition of milk fat. It is estimated that 81 to 83 % of the variation in the amount and composition of milk fat is related to the amount and form of neutral detergent fibre (NDF) and acid detergent fibre (ADF) in the ration (Suton, 1989). A reduction in NDF below the minimum level have a negative effect on the formation of acetate and the development of microflora in the rumen, which has a direct effect on the reduction of fat content in milk (Grbeša and Samaržija,

1994). Mele et al. (2005) concluded that the amount of neutral detergent fibres only correlates positively with milk fat content when milk production is greater than 1 kg/ day. The condition of sheep has a significant influence on the fatty acid composition of sheep milk (Table 1). Table 1 shows that the milk of sheep in better physical condition contains more short- and medium-chain fatty acids (C4:0, C6:0, C8:0, C10:0, C12:0 and C14:0), and less long-chain fatty acids (C16:0, C16:1, C18:0, C18:1 and C18:2). The highest proportion of saturated fatty acids (SFA=70.52 % and 66 %), followed by monounsaturated fatty acids (MUFA=19.5% and 28 %) and polyunsaturated fatty acids (PUFA=4.19 % and 6 %) was found in the milk of Lacaune (Antunović et al., 2024) and Chura (Sánchez et al., 2010) sheep, respectively. Although various authors report different fatty acid compositions of sheep milk in different sheep breeds, the main reason for these differences is attributed to the influence of feeding and not to the breed, which is confirmed by Tsiplakou et al. (2008).

Feeding also influences the presence of toxins and other undesirable substances in milk and dairy products, which has a direct impact on food safety (Pulina et al., 2006). Nutritional stress and the lack of some vitamins in the ration of sheep can significantly affect the number of somatic cells (SCC) in sheep milk, which is reflected in the yield and quality of cheese (Nudda et al., 2023).

Weaning stress

In the intensive sheep milk production, it is becoming increasingly common practice to separate the lambs from the ewe immediately after birth or after suckling the colostrum. This provides more milk for processing (market) and reduces the cost of rearing the lambs. The earlier separation of the lambs from the ewes contributes significantly to higher milk production, as sheep produce around 25 to 30 % of the total amount of milk in the first two months of lactation (Dikmen et al., 2007). In such systems, lambs must be fed with other dairy feeds, of which milk replacer for lambs is the most commonly used (Napolitano et al., 2008). On the other hand, in extensive production the lambs stay longer with the ewes and suckle, so that the quantity of milk milked is significantly lower and the costs for feeding the lambs are higher, which significantly reduces profitability (Gargouri et al., 1993; McKusick et al., 2001). As a possible compromise between the two extremes, which balances the breeders' requirements for higher milk production and lower costs in lamb rearing, more and more breeders are opting for a mixed system in flocks with high-yielding ewes. In this approach, the lambs remain with the ewes and suckle for a period of time after milking. They are then separated a few hours before the next milking. The said mixed system fits with the fact that in the first month of lactation the daily amount of milk produced increases, while the demand from lambs is relatively low, so that the amount of milk produced is sufficient for the lambs and the commercial demand (Sevi et al., 2009). In practice, this is usually achieved by separating the lambs from the ewes in the evening so that they can be together again after the morning milking. McKusick et al. (2001) found no difference in the commercial milk yield of sheep whose lambs were separated 24 hours after lambing and those where the lambs remained and suckled. They explained this by the fact that suckling and physical contact stimulate milk secretion. The mixed system is economically superior in the production of milk and lambs, but the residual milk, where the lambs are in contact with the sheep during the milking period, contains significantly less milk fat (Fuertes et al., 1998; McKusick et al., 2001), which can have a negative effect on cheese production (Reguena et al., 1999). This phenomenon is a consequence of physiological stress,

Table 1. Fatty acid composition of milk fat (g/100 g of FA) of Sarda ewes with different energy balances and body weight variations (Rossi and Pulina, 1991)

Fatty acids	Body weight change						
	-3.8 kg/week	-1.1. kg/week	+1.5 kg/week				
C4:0	2.21	2.49	3.31				
C6:0	0.84	1.29	2.81				
C8:0	0.65	1.09	2.87				
C10:0	1.52	2.70	5.62				
C12:0	1.10	1.88	4.07				
C14:0	3.43	6.96	9.84				
C16:0	24.15	24.67	22.86				
C16:1	1.57	1.56	1.50				
C18:0	13.58	10.93	7.14				
C18:1	28.47	21.52	16.91				
C18:2	6.47	5.86	5.42				
C18:3	0.65	0.27	0.31				

which leads to a decrease in the secretion of oxytocin, the hormone responsible for the release of milk (Barreta et al., 2020). Gimpl and Fahrenholz (2001) claimed that the separation of lambs triggers stress in ewes, which is reflected in increased cortisol levels in the blood. The increased cortisol level reduces the secretion of oxytocin, which influences the milk outflow from the udder. Ewes with lambs "constrict" the milk during milking to retain more of it for their lambs, and this milk has a different composition (McKusick et al., 2001). Early weaning disrupts the bond between ewes and lambs and causes behavioural, physiological and immunological changes in lambs and ewes with negative consequences for health and welfare which directly affect lamb growth (Karakus, 2014; Freitas-de-Melo et al., 2022). The separation of lambs and ewes can be the cause of mastitis, as observed in cows (Ungerfeld et al., 2015). Borys et al. (2014) state that milk from sheep milked 12 hours after lambs weaning contained significantly less dry matter (15.05:16.48 g/100 g), protein (4.82:5.18 g/100 g) and milk fat (4.84:6.05 g/100 g) and significantly more lactose (5.02:4.78 g/100 g) than milk from sheep milked two weeks after lamb weaning.

Shearing stress

Shearing is a necessary and common procedure in sheep farming and is usually carried out once a year to improve production efficiency and sheep welfare. In many areas, the timing of shearing is mainly adapted to the weather conditions, especially the air temperature. Shearing alters the boundary of the animals thermoneutral zone, increases the lower critical temperature and stimulates an adaptive response to maintain body homeostasis (Symonds et al., 1988). During shearing, the animals are exposed to numerous procedures: catching, pulling, separation from the herd, turning, leg tethering, close human contact, noise, vibration and buzzing of the shearing machine, as well as possible injury to the skin and muscles, which can directly affect animal welfare and production performance (Sanger et al., 2011; Arfuso et al., 2022). Shearing during mid- to late-pregnancy increases voluntary feed intake (Parker et al., 1991) and milk yield without affecting milk composition (Sphor et al., 2011). In addition, shearing may enhance beneficial maternal and newborn behaviors that facilitate bonding and hence lamb survival at birth (Banchero et al., 2010). Shearing, especially that not adapted to the environmental temperature, affects thermoregulation and certain physiological changes, which is confirmed by the increased concentration of cortisol in blood plasma (Piccione et al., 2008; Carcangiu et al., 2008). When shearing is carried out at lower air temperatures, cold stress occurs, body heat loss and feed consumption are increased, and production efficiency of the animals is reduced (Panaretto, 1968). Negative effects of shearing on milk secretion have been observed when lactating sheep are exposed to cold in the first days after shearing, as they spend most of their energy on

their own thermoregulation. Thomson et al. (1981) found significantly lower milk production and increased levels of total nitrogen in the milk of sheep kept at a temperature of 1 °C after shearing. Shearing influences the amount of the dry matter intake, and thus the quantity and chemical composition of the milk. Feed consumption after shearing depends strongly on the breed and is about 5 % higher in Lacaune sheep (Elhadi et al., 2019) and 10 % higher in Latxa sheep (Ruíz et al., 2008), which is explained by the increased energy requirement after shearing. As a result of the increased consumption after shearing, Lacaune sheep produced 10 % more milk than unshorn sheep (Elhadi et al., 2019). However, the authors state that shearing had no significant effect on the content of the main components of the milk.

In addition to the aforementioned changes in the amount of milk produced after shearing, some studies report a change in the chemical composition of the milk, particularly the proportion of milk fat. For example, a higher fat content after shearing of 9 % was observed in Sarda sheep (Rassu et al., 2009), 14 % in Tsigai sheep (Aleksiev and Gerchev, 2009) and up to 28 % in Suffolk crossbred sheep (McBride and Cristoperson, 1984). Aleksiev and Gerchev (2009) also found a significantly higher percentage of dry matter and casein in sheep milk milked on the second day after shearing (19.37:18.26 %, 4.23:3.81 %). Furthermore, Rassu et al. (2009) found not only an increased fat content in sheep milk after shearing, but also an altered profile of fatty acids with an increased proportion of C8, C10, C12 and C16 fatty acids. The mentioned change in the composition of fatty acids is not a general rule, considering that Elhadi et al. (2019) did not determine the influence of shearing on the fatty acid profile in sheep milk.

Stress of regrouping

Ideally, animals live in socially stable groups in which they experience cohesion and security as group members, coordinate their behavioural repertoire, show social, affiliative behaviour (Mellor, 2015), cooperation and prosocial behaviour (Rault, 2019). In addition, the feeling of belonging to a group seems to have a buffering effect on the perception of stress (Špinka, 2012). Many conventional management practices, especially in intensive production systems aimed at higher profitability and easier implementation of the technological process, require the mixing of already formed groups, undermining their integrity and hierarchy, which can be stressful for the animals (Fraser and Broom, 1990). Regrouping disrupts the social order of the group and leads to social stress for both the newcomers and the existing members of the group. Under such conditions, new members are usually met with aggression (Fraser et al., 1995), and low-ranking animals are more affected, especially if the regrouping leads to crowding and competition for food and water. This affects the production efficiency of the group in both qualitative

	Chios Day before and after regruping ¹			Chios and Karaguniko Day before and after regruping ¹				
Examined parameter								
	-1	1	4	13	-1	1	4	13
Daily milk yield (mL)	1194	1169	1159	1173	1288	1177	1212	1167
Milk fat (%)	5.20	5.16	4.63	4.66	4.83	4.83	4.69	4.61
Milk protein (%)	4.97	5.01	5.11	5.00	5.13	5.23	5.28	5.25
Milk lactose (%)	4.68	4.69	4.69	4.63	4.70	4.62	4.61	4.65
Milk total solids-not-fat (%)	10.20	10.24	10.33	10.16	10.38	10.40	10.54	10.46

Table 2. Effect of regrouping on milk yield and composition in Chios and Karaguniko ewes (adapted from Papakitsos et al., 2023)

¹Day-1: 1 day before, and days 1, 4 and 13: 1st, 4th and 13th day after regrouping

and quantitative terms, as has been demonstrated in cattle (Torres-Cardona et al., 2014), goats (Fernandez et al., 2007) and sheep (Sevi et al., 2001b). Regrouping therefore not only has a negative impact on the health and welfare of the animals, but also affects their production and can also jeopardise the farmer's economic profit. In order to achieve higher profits in intensive milk production, the regrouping of dairy animals is a common practice. In most cases, dairy ewes are regrouped according to age, milk production and body condition. As Papakitsos et al. (2023) found, average, maximum and minimum heart rate, number of vocalisations and flight distance were significantly increased on the first day after regrouping, indicating great emotional distress in the regrouped ewes. This was observed to a similar extent regardless of whether the mixed animals belonged to only one (Chios) or two (Chios and Karaguniko) breeds. Table 2 shows the influence of regrouping the sheep on the production and chemical composition of the milk.

Milk yield and composition were generally not significantly affected by regrouping, only milk fat content (%) appeared to be significantly lower in the Chios breed after day 4. In addition, milk yield was significantly lower as a result of regrouping on day 1 when Chios and Karaguniko ewes were mixed together, while the decrease in milk fat content followed after the third day (Papakitsos et al., 2023; Table 2). The reduction in milk fat content as

a result of regrouping was also demonstrated by Sevi et al. (2001b) in Comisana ewes. The influence of regrouping on behaviour, welfare, health and milk production has not been studied as closely in sheep as in other ruminants. In cows, for example, a significant influence of regrouping on behaviour, lying time, feeding and rumination (von Keyserlingk et al., 2008) as well as milk production (Torres-Cordona et al., 2014) was found. On goat farms, it is recommended not to regroup animals or add new individuals to a group, as this disrupts the process of individual recognition (Miranda-de la Lama and Mattiello, 2010), increases aggression (Andersen et al., 2007).

Conclusion

An important part of sheep farming is milk production where the animals are often under severe stress, which endangers their health, affects their welfare and has a negative impact on the quantity, chemical composition and processing properties of the milk. Therefore, recognising the sources of stress and their mutual interaction as well as understanding the effects of stress on dairy animals is crucial for the profitable production of quality milk without compromising animal welfare.

Utjecaj stresa na proizvodnju i kvalitetu ovčjeg mlijeka

Sažetak

Ukupno se u svijetu uzgaja oko 1,28 milijardi ovaca koje godišnje proizvedu oko 10,5 milijuna tona ovčjeg mlijeka, od toga je u Europi 121,6 milijuna ovaca (9,47 %), s ukupnom proizvodnjom mlijeka od oko 3,1 milijun tona. U Europi je posljednjih godina zamjetna umjerena stagnacija proizvodnje ovčjeg mlijeka i trenutačno je na razini oko 1/3 svjetske proizvodnje. Ovčje mlijeko je zbog visokog sadržaja suhe tvari, mliječne masti, ukupnih bjelančevina i kazeina izvrsna sirovina za proizvodnju sira. Prerada mlijeka i proizvodnja sira nije samo gospodarski važna, nego je i dio kulturnog nasljeđa, utemeljena na dugoj tradiciji te često organizirana na ruralnim i manje razvijenim područjima. Proizvodnja i kvaliteta mlijeka je pod utjecajem brojnih čimbenika od kojih su najvažniji pasmina, hranidba i tjelesna kondicija, redoslijed i stadij laktacije, mužnja i zdravlje vimena te čimbenici okoliša. Uz navedeno, različite vrste stresa kao što su toplinski, hranidbeni, stres odbića, striža, stres pregrupiranja i dr., imaju značajan utjecaj na dobrobit i zdravlje ovaca te na količinu proizvedenoga ovčjeg mlijeka, njegov kemijski sastav, preradbene odlike te gospodarsku učinkovitost stada. U posljednje vrijeme, usljed klimatskih promjena, sve su brojnija istraživanja utjecaja toplinskog stresa na reprodukcijske i proizvodne odlike ovaca te na njihovu dobrobit i zdravlje. Uz to, toplinski je stres često povezan s nedostatkom hrane i vode. Utjecaj stresa osobito je izražen u ekstenzivnom sustavu uzgoja ovaca i proizvodnje mlijeka koji je u ovčarskoj proizvodnji dominantan. Cilj ovog rada je opisati utjecaj najvažnijih izvora stresa na količinu i kemijski sastav proizvedenoga mlijeka, te na dobrobit i zdravlje ovaca.

Ključne riječi: ovca; stres; proizvodnja mlijeka; kemijski sastav; dobrobit

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