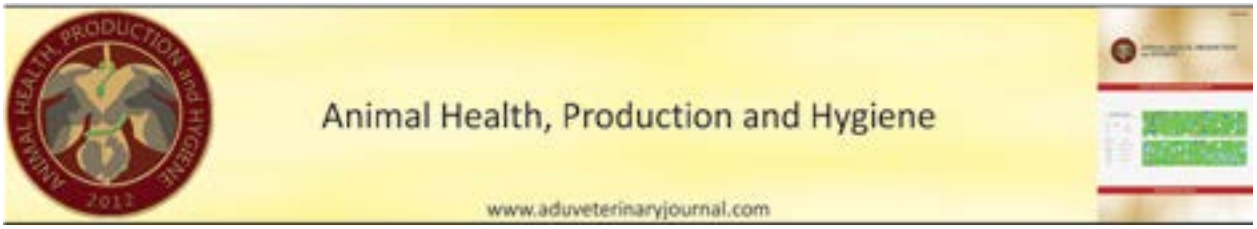

Plasma lactate levels of single and multiple born Chios lambs

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ABSTRACT

Hyperlactemia is closely related to the severity of the disease. Besides, the use of lactate as a biomarker has been increased in human medicine and recently in veterinary medicine as well. The aim of this study was to investigate the plasma lactate level and its relationship with survival in single, twin, triplet and quintuplets born Chios lambs. Blood samples were taken from single, twin, triplet and quintuplets lambs before sucking colostrum within 15 minutes after birth.

Differences in the concentaration of plasma lactate, total protein, albumin and triglyceride were determined and there was significant differences between the groups, however no statistically significant difference was found between the groups in glucose, BUN, urea, creatinine, AST, ALT and cholesterol levels.

In this study, high plasma lactate levels were detected in single, twin, triplet and quintuplets born Chios lambs . Plasma lactate levels are an important predictor for the mortality in an emergency veterinary clinic and there is a correlation between survival and plasma lactate concentration. It can be concluded that the increased number of offsprings produced in one litter and lamb deaths were found to be correlated with the plasma lactate concentrations. Monitoring of plasma lactate concentrations can be recommended for the prognosis lamb survival.

Keywords: Lactate, single and multiple born lambs, Chios breed .

Tek ve Çoklu Doğmuş Sakız Kuzularda Plazma Laktat Düzeyleri

ÖZET

Hiperlaktemi hastalığın ciddiyetiyle yakından ilişkilidir .İnsan hekimliğinde biyobelirteç olarak kullanılan laktat son yıllarda veteriner hekimlikte de yagın olarak kullanılmaya başlamıştır. Bu çalışmanın amacı yenidoğan tek, ikiz, üçüz ve beşiz yavrularda laktat düzeyini ve bunun sağkalım ile ilişkisini belirlemektir. Gebe sakız ırkı koyunlar takip edilmiş ve doğumdan hemen sonra 15 dakika içinde tek, ikiz, üçüz ve beşiz yavrulardan annelerinden kolostrum emmeden kan örnekleri alınmıştır. Plazma laktat total protein, albumin ve trigliserid düzeylerinde gruplar arası fark istatsistiki olarak anlamlı bulunmuş ancak glukoz, BUN, üre, creatinine, AST, ALT ve kolesterol düzeylerinde istatsitiki olarak anlamlı bulunmamıştır. Bu çalışmada tek, ikiz, üçüz ve beşiz yavrularda yüksek laktat düzeyi belirlenmiştir. Laktat düzeyi acil veteriner klinikte mortalitenin önemli bir göstergesidir ve yüksek laktat konsantrasyonu ile sağkalım arasında negative korelasyon vardır. Bu çalışmada çoklu doğumlarda yavru sayısı arttıkça yavru ölümlerinin de arttığı, laktat ölçümü ile monitorizasyonun korele olduğu ve sağkalımın belirlenmesinde laktatın anlamlı olduğu belirlenmiştir.

Anahtar Kelimeler: Laktat, tek ve çoklu doğum, Sakız ırkı

Introduction

Multiple born lambs have lower survival rates compared to single born lamb. There are several reasons for this: as such low birth weight, unsatisfactory placental nutrition, intrapartum hypoxia, inadequate thermogenesis and insufficient colostrum intake. Lambs with low birth weight, compared to heavier-born lambs, have not only lower surface areas but also lower energy stores (Dwyer, 2003; Mccord et al., 2017). The causes of lamb deaths in the first three days of life have been investigated pathologically and through clinical observation , and it has been reported that the causes of death in single, twin and triple offspring were mainly due to dystocia, starvation and birth position (Kerslake et al., 2005).

Light metabolic and respiratory acidosis occur in newborn lambs and calves in normal births, for which reduced circulation in the placenta during severe labour pains, prolonged parturition and severe acidosis related diseases in mothers are responsible. . . Along with these causes, hypoxia and hypercapnia can occur in the fetus (Vannuchi et al., 2012). The offspring can either die before birth depending on the degree and duration of acidosis or is born with symptoms of asphyxia. A decrease in fetal growth may occur as a result of maternal malnutrition and fetus's failure to obtain sufficient nutrients from the placenta during pregnancy. Inadequate nutrient supply to the fetus can result in low birth weight of offspring. This leads to insufficient thermoregulation of the offspring and postnatal behaviour such as rising to its feet and sucking (Dwyer, 2003).

However, lactate metabolically plays a protective role in providing ongoing cellular energy during tissue hypoperfusion or hypoxia and in reducing acidosis. Therefore, hyperlactatemia has a close relationship with the disease. This is self-protecting response of the organism (Gillespie et al., 2017). Lactate is usually an essential multifaceted metabolic fuel for cellular bioenergetics. Since lactate is used as a biomarker and as a therapeutic endpoint in the diagnosis of human diseases in emergency medicine and intensive care units, it is also essential to improve its use in the field of veterinary medicine. There are studies about the clinical use of lactate, screening, evaluations, risk determination and prognosis in the human medicine . Plasma lactate concentration is a valuable prognostic marker in trauma, sepsis,septic shock, systemic inflammatory response syndrome (SIRS), cardiac arrest, malaria, head trauma, liver failure, and carbon monoxide poisoning (Vincent et al. 2016). An increase in plasma lactate levels in living organisms is closely associated with life-threatening problems, but it is not seen as a cause. For this reason, while normal level of lactate is associated with a healthy organism, high lactate level is associated with poor prognosis.

Since there is a low survival rate due to many reasons in multiple born lambs, this present study aimed to investigate plasma lactate concentration of single or multiple newborn Chios lambs, which has increased use in veterinary medicine in recent years. Chios breeds of sheep were chosen for the material of this study because of their high fertility and high incidence for the production of larger litters. Thus, it can enable the present study to establish a correlation between plasma lactate concentration and offspring survival and growth rate.

Material and Method

Animals

This study was started with the permission of Adnan Menderes University Ethics Committee of Experimental Animals Local Ethics Committee dated 31/05/2018 and numbered 645831/01/2018/71. Newborn lambs of Chios ewes were provided by the Chios Sheep Production Farm of Aydın Metropolitan Municipality. Blood samples were obtained in the blood sampling tubes from *vena jugularis* in newborn lambs which were born between 01.02.2018 and 01.02.2019. In this time period, 12 singles, 6 twins (6x2 = 12), 4 triplets (4x3 = 12) and 1 quintuplet (5x1 = 5) lambs were born. There was no quadruplet litter during the research period in the farm. As soon as the lambs were born, their blood was drawn into vacuum heparinized tubes to separate out the plasma before they sucked colostrum. After that, they were weighed and rectal body temperatures were measured. The blood plasma was separated into 3 different marked tubes per each sample. One tube was kept at +4 °C to measure lactate within latest 6 hours. The other two tubes were stored at -20 °C in order to perform the analyses of glucose, total protein, albumin, urea, creatinine, cholesterol, triglycerides, AST and ALT. After birth, newborn lambs implemented general care procedures under the same conditions. After the birth and blood collection, vitamin E and selenium were administered to newborn lambs. Besides, newborn lambs were vaccinated twice against enterotoxemia when they were one month and 45 days old.

Biochemical Analyses

The blood samples were taken to Aydın Adnan Menderes University, Faculty of Veterinary Medicine, Department of Internal Medicine Laboratory. Plasma lactate measurements were made colourimetric with the Eurolyser Solo device (Austria) according to the producer instruction. The final point measurement absorption was 546 nm wavelength. The measurement range of the method is 0.1-15 mmol/L. Plasma glucose, BUN, urea, creatinine, total protein, albumin, triglyceride, cholesterol, AST and ALT level measurements were performed in autoanalyser (Rayto Chemray120, China) using commercial test kits (Archem Diagnostic, Turkey).

Statistical Analysis

The 'SPSS for Windows Version 22 (SPSS Inc, USA) package program was used for all statistical analysis. The level of statistical significance was accepted as $p<005$. Whether the variables showed normal distribution or not was examined by the Shapiro-Wilk test. Average and standard deviation values were specified for descriptive statistics. The difference between groups was determined by one-way variance analysis (One-way ANOVA)which is suitable for normal distribution. Tukey test was performed to determine the group which caused this difference. Non-parametric tests (Kruskal Wallis) were used to show the differences between the groups which were not normally distributed.

Results

The individual data (ear numbers, date of birth, sexes, birth weights, rectal temperatures, lactate levels and postnatal follow-up) of the lambs used in the study are presented in Table 1. Mean plasma lactate, glucose, BUN, urea, creatinine, total protein, albumin, triglyceride, cholesterol, AST and ALT levels of single, twin, triplet and quintuplet groups are presented in Table 2.

According to the present results, 3 of 12 single-born offspring were died when they were 15 days and three months old. There was no mortality in the twin group. Two lambs died in the triplet group within the first month following their birth. There was no, quadruplet deliveries of ewes in the study, and only one quintuplet delivery occurred at the farm. Quintuplet

Table 1 The individual data of the newborn lambs.

Litter size	Animal number	Sex	Birth Weight	Date of Birth	Lactate mmol/L	Rectal Temp.	Status	Date of Death
Single	920	M	5	05.02.2018	8,6	37,7	Alive	
Single	897	F	3,9	08.02.2018	9,4	38,7	Alive	
Single	865	M	6,1	11.02.2018	7,34	37,8	Alive	
Single	864	F	4,5	12.02.2018	14,52	38,2	Alive	
Single	854	M	4,4	18.02.2018	3,44	37,9	Alive	
Single	853	M	4	20.02.2018	4,54	36,5	Alive	
Single	851	M	4,8	20.02.2018	8,74	37,8	Alive	
Single	850	M	4,3	20.02.2018	5,1	38,4	Alive	
Single	846	M	3,3	22.02.2018	>15	37,2	Dead	14.03.2018
Single	841	F	3,8	22.02.2018	3,98	39,1	Dead	16.05.2018
Single	590	F	3,5	31.12.2018	>15	37,6	Dead	09.01.2019
Single	774	F	4,3	07.01.2019	6,94	37,2	Alive	
Twin	914	F	2,7	06.02.2018	4,56	37,8	Alive	
Twin	915	F	3,8	06.02.2018	10,01	38,3	Alive	
Twin	909	F	2,8	06.02.2018	6,44	38,5	Alive	
Twin	910	F	3,6	06.02.2018	4,76	38,2	Alive	
Twin	906	F	3,1	07.02.2018	>15	38,2	Alive	
Twin	905	F	3,6	07.02.2018	4,7	39,1	Alive	
Twin	895	M	4	08.02.2018	8,59	38,6	Alive	
Twin	896	F	3,8	08.02.2018	4,41	38,5	Alive	
Twin	881	M	4,8	09.02.2018	5,86	38,4	Alive	
Twin	882	M	4,5	09.02.2018	6,38	38,5	Alive	
Twin	622	M	5	12.12.2018	2,69	37,4	Alive	
Twin	621	M	4,5	12.12.2018	7,35	37,8	Alive	
Triplet	900	M	4,3	07.02.2018	3,45	37,6	Alive	
Triplet	901	M	3,6	07.02.2018	10,74	38,1	Alive	
Triplet	902	M	2,8	07.02.2018	4,49	37,3	Alive	
Triplet	822	F	2,8	23.02.2018	3,85	37,5	Alive	
Triplet	823	M	3,6	23.02.2018	>15	37,8	Alive	
Triplet	824	M	2,2	23.02.2018	2,58	37,1	Dead	05.03.2018
Triplet	728	F	2,1	07.07.2018	>15	37,5	Dead	18.08.2018
Triplet	727	F	2,6	07.07.2018	10,25	37,6	Alive	
Triplet	726	M	3,6	07.07.2018	6,51	37,4	Alive	
Triplet	582	M	2,6	05.01.2019	4,82	37,6	Alive	
Triplet	583	F	3,7	05.01.2019	5,21	37,4	Alive	
Triplet	584	F	4,6	05.01.2019	7,74	37,2	Alive	
Quintp.	921	F	2	05.02.2018	>15	37,5	Alive	
Quintp.	922	F	1,5	05.02.2018	>15	36,7	Dead	31.05.2018
Quintp.	923	M	2,7	05.02.2018	>15	37,1	Alive	
Quintp.	924	F	1,7	05.02.2018	>15	37,7	Alive	
Quintp.	925	F	2	05.02.2018	12,2	37,2	Alive	

lambs had lower live weight and higher lactate level than all other lambs, but only one lamb died in this group in the third month after the birth. Of the 41 lambs included in the study, six lambs died between 15 days and three months old of age. . Among them, lamb number 841 was treated for general condition disorder and weakness, and lamb number 824 was treated for respiratory disease, but both of them died. The other four lambs (ear-numbered 846, 590, 922, 728) were recorded as sudden death without receiving any treatment.

According to the results, differences in metabolic parameters between single and multiple born lambs were evaluated. Statistical differences in plasma lactate, total protein, albumin and triglyceride levels were determined between the groups, while glucose, BUN, urea, creatinine, AST, ALT and cholesterol levels were not different statistically between the groups.

Average plasma lactate levels were 6.05 ± 4.1 mmol / L, 6.70 ± 3.6 mmol / L, 12.47 ± 4.3 mmol / L, 14,44 ± 1.25 mmol / L in single, twin, triplet and quintuplet born lambs respectively. As the number of offspring in one litter increased, the average lactate levels of the offspring also increased, and the highest level was identified in quintuplets. The mean lactate levels of triplet and quintuplets groups were higher (p <0,001) compared to single and twin offspring groups.

Total protein level was 4.69 ± 0.29 g / dL, 4.08 ± 0.19 g / dL, 4.21 ± 0.25 g / dL and 4.07 ± 0,16 g / dL in single, twin, triplet and quintuplet born lambs respectively. The lowest level was in the quintuple offspring group. Similarly, serum albumin level was 3.19 ± 0.12 g / dL, 3.04 ± 0.11 g /dL, 3.07 ± 0.12 g / dL and 2.95 ± 0.09 g / dL in single, twin, triplet andquintuplet born lambs and the mean albumin level of the quintuplet offspring group was lower than the other groups. The difference between total protein and albumin levels were found to be significant (p <0.001). Triglyceride levels were 57.91 ± 20.89 mg / dL, 51.53 ± 16.36 mg / dL, 53.50 ± 23.93 mg / dL and 72.50 ± 7.63mg / dL in single, twin, triplet and quintuplet born lambs respectively, and the level in the quintuplet offspring was higher than all other groups. No statistically significant difference was found between the groups in terms of glucose, BUN, urea, creatinine, cholesterol, AST and ALT concentrations.

Discussion

The neonatal period requires extra physiological, morphological and behavioural changes to ensure successful adaptation to the extrauterine environment. In some cases this adaptation is slowly shaped and there exist differences between species (Piccione et al., 2007; Piccione et al., 2008). Newborns are under metabolically unstable conditions. Therefore, newborns, especially in the first week of life, are vulnerable to diseasesand this condition can result in a high mortality rate (Piccione et al., 2010). One of the most important problems in cattle breeding is neonatal deaths which occur at an approximate rate of 15%, which is also one of the reasons for economic losses in small ruminant husbandary (Dwyer, 2008). Efforts to prevent neonatal deaths should be the main aim of veterinary neonatology studies. For these reasons, clinical follow-up of newborn lambs may provide an early diagnosis in determining adaptation failure. With all these clinical follow-ups, it may be possible to establish corrective procedures in cases such as inability to correct the acid-base balance and failure to provide thermal or metabolic hemostasis, which causes mortality.

Chios sheep are fed and cultivated in Turkey because of their high fertility and delivery of multiple lambs in a litter. However, multiple lambs lead to low survival rate due to various reasons such as low birth weight, hypersensitivity to placental insufficiency, intrapartum hypoxia, inadequate thermogenic mechanism and insufficient colostrum intake. While normal lactate levels at lambing are associated with better survival rate, however high lactate levels are associated with poor prognosis. The most effective cause of this may be lactic acidosis developing due to hyperlactatemia). Metabolic acidosis is evident in small ruminants within 10-15 minutes after birth. Increased lactic acid is one of the causes of metabolic acidosis. Acidosis, hypercapnia and hypoxia in newborns stimulate carotid chemoreceptors showing an increase in O_2 sensitivity and affect the respiratory centre, which causes an increase in tidal volume and respiratory rate. This may be responsible for the significant increase in pO_2 in the days after birth so that it may provide an improvement in respiratory function due to increased gas diffusion capacity through vascularization of the alveolar-capillary membrane and lung capillary (Piccione et al, 2006). Furthermore, the cause of acidosis after birth is explained by decreased blood circulation in the placenta, prolonged delivery process and severe acidosis of ewes. Therefore, hypoxia and hypercapnia occur in the fetus (Sahal et al., 1994). Prolonged deliveries, changes in offspring position and conditions that require support for delivery are defined as difficult births. It may cause acute hypoxia and elevated plasma lactate due to cord compression (Mellor and Stanford, 2004). Utero-placental blood flow may be decreased because number of placentomes are lower in lambing withtwo or more offspring than single one (Kenyon et al., 2007; Vonnahme et al., 2008). Heat production of lambs with placental insufficiency is also limited, and a negative correlation was found between heat production and plasma lactate levels in the first 6-8 hours of life (Kerslake et al., 2010). Since the capacity of the placenta to carry oxygen and nutrients depends on the surface area of the placenta, high lactate concentration can be formed in multiple deliveries because it affects the number of placentomes and uteroplacental blood flow (Kerslake et al., 2010).

Glucose is the main energy substrate for fetal and placental metabolism in all mammalian species (Brolio et al., 2010).

Table 2 Average concentration of plasma biochemical parameters in lamb litters (mean, and standard deviation).

Groups	Lactate mmol/L $X \pm S_x$	Glucose mg/dL $X \pm S_x$	BUN mg/dL $X \pm S_x$	Urea mg/dL $X \pm S_x$	Creatinin mg/dL $X \pm S_x$	Total protein g/dL $X \pm S_x$	Albumin g/dL $X \pm S_x$	Tryglyceride mg/dL $X \pm S_x$	Cholesterol mg/dL $X \pm S_x$	AST IU $X \pm S_x$	ALT IU $X \pm S_x$
Single n=12	6,05±4,1[b]	53,50±20,25	14,60±2,70	31,25±5,80	1,35±0,38	4,69±0,29[a]	3,19±0,12[a]	57,91±20,89[ab]	34,16±6,73	23,41±4,62	3,33±1,65
Twin n=12	6,70±3,6[b]	34,76±11,51	13,76±2,80	29,16±5,99	1,53±0,58	4,08±0,19[b]	3,04±0,11[b]	51,53±16,36[b]	29,00±5,30	19,38±4,07	2,69±0,63
Triplet n=18	12,47±4,3[a]	45,72±30,19	14,24±3,77	30,85±8,44	1,53±0,48	4,21±0,25[b]	3,07±0,12[ab]	53,50±23,93[ab]	34,33±11,04	23,16±3,91	3,83±2,85
Quintuplets n=5	14,44±1,25[a]	41,33±13,45	15,88±0,73	34,00±1,58	1,86±0,27	4,07±0,16[b]	2,95±0,09[b]	72,50±7,63[a]	33,80±1,64	20,20±3,27	2,60±0,54
p	***	NS	NS	ND	NS	***	***	**	NS	NS	NS

*: p<0.05 ** : p<0.01 ***:p<0.001 NS :Not significant

Anaerobic glycolysis is a process that uses glucose to produce lactate in an oxygen-free environment (Barroso et al., 2006). Lactate plays a key role as a marker of fetal and neonatal distress in both human and veterinary obstetrics (Armstrong et al., 2006), and it is one of the main components of metabolic acidosis (Borruto et al., 2008). It has been reported in many studies that hyperlactatemia occur physiologically in newborns (Peternelli-Silva , 2012 and 2013; Regazzi, 2015). The increase in lactate levels in newborns can be observed physiologically. It takes 24 hours for lactate to return to normal levels. Hyperlactatemia after birth is associated with placental production of lactate. Hyperlactemia in neonatal animals and its gradual reduction in 24 hours was demonstrated for sheep by Peternelli-Silva et al. (2013) and Castagnetti et al (2010), and for horses by Cruz (2014). According to a research by Peternelli-Silva et al. (2018) in which lactate levels in ewes at lambing, in placenta and newborn lambs were examined, the lactate level was 3.09 mmol/L in ewes at lambing, 5.78 mmol/L in placenta, and in newborn offspring the maximum level was 5.98 mmol/L at lambing and the minimum level was 3.80 mmol/L at 24 hours after lambing. Şahal et al (1994) reported the lactate concentration in newborn lambs as 4.84 mmol/L. A study conducted by Peternelli-Silva et al. (2013) on 18 sheep (9 Suffolk and 9 hybrid sheep) and 20 healthy newborn lambs, average lactate level in newborn lambs was 3 mmol/L. Kerslake et al. (2010) reported postnatal plasma lactate concentration in single, twin and triplet lambs as 7.9±1.15, 7.7±0.54 and 7.6±0.57 mmol / L, respectively. The same researcher pointed out that glucose concentrations were lower in twin and triplet groups.

In the present study, lactate values in newborn lambs were obtained between 6.05 and 14.44 mmol / L . The highest lactate concentrations were determined in multiple born offspring groups (triplets, quintuplets). In some studies, lactate levels in newborn lambs were lower than our findings (Şahal et al. 1994; Peternelli-Silva et al. 2013; Partidge, 2017), and in others, they were higher (Kerslake, 2010; Aridos et al. 2017). The reason for the differences may be the effect of breed or the method of analysis. However, in all of these studies, lactate values in newborn lambs are higher than sheep's reference values, and this means hyperlactatemia develops mostly in newborns. It was concluded in the present study that the sudden death pattern in 4 lambs after birth whose birth lactate concentrations were very high, was associated with the inability to compensate the lactic acidosis. This condition has been interpreted as the cause of high lactate concentrations in multiple offspring with low birth weight, especially in quintuplets.

In the present study, blood glucose levels were examined and no statistical difference was found between offspring groups, but there was a tendency to be lower in single offspring than the multiple born offspring. Similarly, Hannock et al (2012) determined the birth weight and blood sugar levels in twin offspring to be lower than single offspring. This study determined that birth weights of multiple born lambs were lower than single born offspring, and the lowest birth weights were detected in the quintuplets group. This is due to the mother's inability to feed the fetus because there is more than one offspring in the uterus. As a matter of fact, in multi-fetus pregnancies, decrease of blood glucose and increase of plasma free fatty acids and beta-hydroxybutyrate levels are indicative of this inadequacy (Hancnock et al. 2012). The same researchers reported that the blood glucose levels of twin-born offspring were lower than those of single offspring.

Vannucchi et al. (2012) reported in a clinical and hemogasometric study that blood glucose levels of new born Santa Ines lambs were 30 mg/dL at and after the birth. Peternelli-Silva et al. (2018) found out that the average blood glucose of lambs was 46.86 mg / dL in 60 minutes after lambing. The same researchers also reported that glucose levels increased to normal levels in 24 hours. In the present study, blood glucose concentrations were similar with this studies, and the highest value was 53.50 ± 20.25 mg / dL in single born offspring, 34.76 ± 11.51 mg / dL in twins, 45.72 ± 30.19 mg / dL in triplets and 41.33 ± 13.45 mg / dL in quintuplets. Although there was no statistically significant difference between the groups, low glucose levels in multiple born lambs were interpreted as the mother's inability to feed the fetus sufficiently. Moreover, it can suggested that there is a correlation between high lactate concentration and low glucose.

In a study on newborn lambs, total protein levels of healthy lambs were reported to be between 5.9 g / dL and 6.3 g / dL (Nozarian et al., 2010). Lepherd et al. (2009) reported concentrations of total protein and albumin 5.7 g / dL and 3.4 g / dL in lambs respectively. Bornez et al. (2009) determined total protein levels in newborn lambs between 5.7 and 6.4 g / dL. Total protein and albumin levels obtained in the present study are consistent with the studies conducted on lambs previously. However, total protein and albumin levels were found to be higher in single-born offspring than multi-born offspring, and the lowest level was found in the quintuple offspring group. The difference between groups was statistically significant. In the literature, no research was encountered comparing the total protein and albumin levels of multiple offspring and single offspring. The results of the present study can be interpreted that low total protein and albumin levels of the offspring, especially of quintuplets, could be a consequence of low birth weights and insufficient placental nutrition.

In this study, no statistically significant difference was found between single and multiple new born offspring in terms of plasma BUN and urea levels. The average BUN levels in new born lambs were found to be nonsignificant between groups and the min and max concentrations were 13.76 ± 2.80 and 15.88 ± 0.73 mg / dL. Vanucchi et al (2012) found out 14.60 ± 3.54 mg / dL BUN level in hour zero and 16.20 ± 3.23 mg / dL in the 60th minute in their research on newborn lambs (bu cümle anlaşılmıyor !!!!!), and they reported that this BUN level was within the reference ranges which is 10-26 mg / dL and also confirmed by Lester et al. (2009). This study determined that triglyceride concentration in newborn quintuplets lambs was statistically higher than that of the other groups. Atakişi et al (2013) interpreted that the increasing concentration of triglycerides after birth was due to the usage and conversion of energy sources for the increased energy requirements. Indeed, low glucose levels are an indicator of this fact. In this study, it is believed that the high triglyceride concentration in quintuplets was due to the efforts of lambs with low birth weight to produce more energy in order to provide thermoregulation and metabolic hemostasis. In the present study, no difference was found between groups in terms of cholesterol concentrations. Since cholesterol is not transmitted to the offspring through the placenta, the offspring receive large molecules (such as immunoglobulin, fat-soluble vitamins, and cholesterol) for growing up through the colostrum and mother's milk. Therefore, blood cholesterol levels of mother and postnatal lambs are related to lipid levels in the feed taken. The lower level of post-natal cholesterol level than that of a healthy adult was interpreted as normal. There was no significant difference between the groups in terms of concentrations of AST and ALT enzymes, which are indicative for liver functions, and it was in-

terpreted that these lambs had a similar state of liver functions immediately after the birth.

Lactic acidosis occurs when plasma lactate level exceeds 4-5 mmol / L (optimum range: 0.5-1.5 mmol / L). However, previous studies showed that birth and especially multiple lambs births may cause lactic acidosis (Vannucchi et al., 2012; Silva et al., 2013). Six out of the 41 studied newborn lambs died, and the lactate levels of 4 dead lambs (% 66.6) were determined to be 15 mmol / L at lambing. Based on the present results, low birth weight and high lactate concentration can be an important indicator for offspring survival.

When the number of offspring in a litter increases, the lactate levels is also increasing, and this creates a risk for lactic acidosis. This situation applies to all single, twin, triplet and quintuplet births of Chios sheep. Therefore, early intervention and treatment of lactic acidosis in newborn lambs are essential. Based on the data of the present study, clinical and laboratory monitoring of the offspring of this breed which is characterised by high fertility and multiple offspring will be important to increase the survival rate.

The possibility to run lactate analysis in the field and on farms is increasing because of the development of fast test kits. The development of practical individual patient-side devices areeasy-to-use and provide cheap analysis. In light of the study, lactate analysis is considered an usefull test in postpartum monitoring. The routine application of lactate analysis as an important marker in controlling and preventing newborn lamb deaths due to lactic asidosis, which poses a major problem in herd and farm management. It is thought that the result of the present study will economically be beneficial for the farms if they can be implemented into practice and used effectively in farm management.

Conflict of interest: The authors declare that they have no conflict of interest.

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