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# Growth Performance and Metabolic Profile of Chios Lambs Prevented from Colostrum Intake and Artificially Reared on a Calf Milk Replacer

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**Abstract:** This study was undertaken to investigate the effects of artificial rearing on growth performance and metabolism of lambs as measured by some key blood metabolite parameters. A total of 20 Chios lambs were used. Newborn twin and triplet lambs were divided into 2 groups. The first-born lambs of twins or triplets formed the artificially reared (AR) group. The other lambs formed the ewe-reared (ER) group. Blood samples were taken from lambs of both groups during the first 24 h after birth and at weeks 1, 2, and 3. Effects of rearing method on plasma total protein, albumin, globulin, glucose, urea, total lipids concentrations, survival rate, body weight, and average daily gain (ADG) were assessed. Body weight and ADG were significantly affected by rearing methods ( $P < 0.001$ ). Plasma total protein and globulin concentrations of the lambs in the ER group were significantly higher than in the AR group at 18 and 24 h, and at weeks 1 and 2. Plasma urea concentrations at 18 and 24 h, and plasma total lipid concentrations at 24 h and at week 1 were significantly higher in the ER group than in the AR group. In conclusion, our results indicated that artificial rearing of lambs with calf milk replacer decreased survivability and inhibited growth rates.

**Key Words:** Artificial rearing, newborn lambs, growth performance, metabolic profile

## Kolostrum Alımı Engellenmiş ve Suni Olarak Beslenmiş Sakız Irkı Kuzuların Büyüme Performansı ve Metabolik Profili

**Özet:** Bu çalışma, suni olarak beslemenin kuzuların büyüme performansları ve metabolizmalarına etkilerini bazı anahtar kan metabolitlerin düzeyleri ölçülerek belirlenmesi amacıyla gerçekleştirilmiştir. Hayvan materyali olarak ikiz ve üçüz doğmuş olan 20 adet Sakız ırkı kuzu kullanılmıştır. Yeni doğan kuzular doğumlarının hemen ardından iki gruba ayrılmışlardır. ikiz ve üçüz kuzulardan ilk doğan yavru annesinden ayrılarak suni besleme (AR) grubunu oluşturmuştur. Diğer yavrular annesinin yanında bırakılarak annelerini emmen grubu (ER) oluşturulmuştur. Her iki gruptan da doğumdan sonraki ilk 24 saat boyunca ve 1., 2. ve 3. haftalarda kan örnekleri toplanarak beslenme şeklinin plazma total protein, albumin, globulin, glukoz, üre ve total lipid, konsantrasyonları ile kuzuların yaşama gücü, canlı ağırlık ve günlük ağırlık kazancına (ADG) etkileri belirlenmiştir. Beslenme şeklinin canlı ağırlıklara ve ADG'ye 2. haftadan itibaren  $P < 0.001$  düzeyinde etkili olduğu tespit edilmiştir. ER grubundaki kuzuların plazma total protein ve globulin konsantrasyonlarının AR grubunda yer alan kuzulardan 18. ve 24. saatler ile 1. ve 2. haftalarda önemli düzeyde yüksek olduğu bulunmuştur. ER grubundaki kuzularda doğumdan sonraki 18. ve 24. saatlerde üre ve 24. saat ile 1. haftada total lipid konsantrasyonlarının AR grubundaki kuzulardan önemli düzeyde yüksek olduğu belirlenmiştir. Sonuç olarak doğumdan sonra süt ikame yemi ile suni olarak beslemenin kuzuların yaşama gücünü olumsuz olarak etkilediği ve ideal vücut ağırlığına ulaşmasını engellediği ileri sürülebilir.

**Anahtar Sözcükler:** Suni besleme, yeni doğmuş kuzu, büyüme performansı, metabolik profil

## Introduction

Lambs are born hypo immunocompetent and with a small store of energy for heat production and metabolism (1). The most satisfactory way of providing the newborn

with immunity against disease is to ensure that it gets a large quantity of good quality ewe colostrum in early life (2,3). Basser and Gay (4) found that the mortality rate was lower in calves with a high serum immunoglobulin (IgG) concentration than those with a low serum IgG

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concentration. IgG absorbed from colostrum plays an important role in the serum IgG concentration of newborn lambs (5). The time period between 12 and 36 h of birth is extremely critical for absorption of IgG from colostrum (6). In addition, Solones et al. (7) reported a higher mortality rate and lower serum immunoglobulin concentration in lambs fed artificial colostrum compared to those fed maternal colostrum. Colostrum is an important source of IgG. IgG content of colostrum is 100-fold higher than that of normal milk (8). There is a close relationship between serum total protein concentrations and serum IgG concentrations. Determining concentrations of plasma total proteins is more practical and more economical than radial immunodiffusion and zinc sulphate turbidity tests, which are used for determining the concentration of serum IgG (9). Concentrations of plasma total proteins and globulin can be used as an indicator for monitoring passive transfer and estimating the amount of circulating IgG in newborns. In addition, total protein concentrations in serum and colostrum are important for lamb growth (10).

The capacity of lambs to utilise food can affect their growth performance (11). In particular, energy and protein utilisation can be monitored using the serum levels of some key metabolites such as glucose, urea, and total lipids (12).

This study was carried out to investigate the effect of rearing Chios lambs with calf milk replacer on the passive transfer of IgG growth performance and metabolic profile.

## Materials and Methods

**Animals:** A total of 20 Chios lambs were used and 34 Chios ewes with a high twin rate were used to obtain the lambs. Following a 15-day adaptation period, sponges

containing 60 mg medroxyprogesterone acetate were applied to the ewes intravaginally using special applicators. After 5 days, sponges were removed and 600 IU of 'pregnant mares' serum gonadotropin (folligon, Intervet, The Netherlands) was intramuscularly administered per ewe. After 48 h of the injection, the ewes were placed in boxes with a 1 Chios ram per 10 Chios ewes ratio, for random mating (13). Twenty-four of the pregnant ewes, as diagnosed by ultrasound, were divided into 2 equal groups and placed in 2 separate boxes with similar conditions.

**Feeding and caring of the lambs:** When the ewes started lambing, the first born lambs of twins or triplets were separated from the ewes as soon as the foetal cords were excised. They were then cleaned of foetal membranes and fluid, and housed in isolated and warm boxes for 3 weeks. These lambs constituted the artificial rearing (AR) group. The remaining lambs of the twins and triplets (second and/or third lambs) were allowed to stay with the ewes. These lambs constituted the ewe rearing (ER) group. Ewes and lambs were housed in a separate box prepared for the postnatal period. ER lambs were allowed to suck ewes ad libitum. AR lambs, on the other hand, were fed a commercial calf milk replacer (Joosten-Milk Blue, The Netherlands) every 2 h for the first week, every 4 h for the second week, and every 12 h for the third week. Calf milk replacer was diluted with warm water (1:8 ratio).

Composition of the calf milk replacer is provided in Table 1. The lambs were fed with feeding bottles only during the first 2 weeks, whereas hay and water were added in the third week. Access by personnel was restricted to the isolated boxes to reduce the risk of infection. Personnel were not allowed to enter the boxes while wearing their street clothing. Those personnel

Table 1. The content of calf milk replacer.

Crude protein (%)	20-23	Fe (mg/kg)	60
Fat (%)	15-18	Mg (mg/kg)	300
Crude fibre (%)	1	Vitamin A (IU/kg)	30000
Ash (%)	10	Vitamin D3 (IU/kg)	2000
Humidity (%)	3.5	Vitamin E (mg/kg)	20
Ca (mg/kg)	0.7	Vitamin B1 (mg/kg)	3
P (mg/kg)	0.85	Vitamin B6 (mg/kg)	8
Mn (mg/kg)	8	Vitamin C (mg/kg)	150
Zn (mg/kg)	40	Niacin (mg/kg)	20
Cu (mg/kg)	4	Digestible energy (kcal/kg)	2750

working in the other boxes were not allowed to work in the isolated boxes. Bedding in the isolated boxes was renewed and the boxes were disinfected on a daily basis.

**Collection of blood samples:** Blood samples were taken into plasma tubes containing heparin (Lithium heparin, Venoject, VT-050SHL, Terumo Europe N.V., Belgium) from the jugular veins of the lambs of both groups at 0, 6, 18, and 24 h after birth, and the 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> week. Plasma of the blood samples was removed by centrifugation and transferred into Eppendorf tubes and stored at -20 °C until analysed.

**Biochemical analysis:** Total protein, albumin, glucose, urea, and total lipid concentrations in the plasma samples were determined using commercial test kits (Spinreact, SA-ctra sahare coloma, 7-E-17176 sant esteve de bas-Spain) and a SEAC Ch 100 photometer. Globulin concentrations were calculated from the related data.

**Statistical analyses:** The data were analysed using SPSS (version 11.5). Effects of rearing methods, litter size, and gender on body weight and ADG were determined using the General Linear Model (GLM). Survival rates were analysed by frequency ( $\chi^2$ ) analysis. The plasma total protein, albumin, globulin, glucose, urea, and total lipid concentrations of the ER and AR groups were compared using independent-sample t test. All data were expressed as mean  $\pm$  SE.

## Results

Lamb survival rates are presented in Table 2. The lamb survival rate in the ER group was higher than that in the AR group. The survival rate of single lambs was higher than that of twins or triplets, but the difference was not significant. No clear difference was observed between twins and triplets. The least square means of body weights and average daily gain (ADG) of the lambs are given in Table 3. From the second week onward, the effect of rearing method on body weight gain was significant ( $P < 0.001$ ), as the body weight of the ER lambs was significantly higher than the AR lambs. ADG in the ER group was significantly ( $P < 0.001$ ) higher than that of the AR group. The effects of litter size and gender on body weight and ADG were not significant, throughout the entire study.

Plasma total protein, albumin, globulin, glucose, urea, and total lipids concentrations are provided in Table 4. Total protein and globulin concentrations in the ER group were significantly higher than in the AR group at 18 h and 24 h, and at the 1<sup>st</sup> and 2<sup>nd</sup> week after birth ( $P < 0.001$ ,  $P < 0.001$ ,  $P < 0.01$ , and  $P < 0.01$ , respectively). Plasma albumin and glucose concentrations were not affected by rearing method. Plasma urea concentrations in ER lambs were significantly higher than in AR lambs at 18 and 24 h after birth ( $P < 0.001$  and  $P < 0.001$ ,

Table 2. The survival rate (%) in artificially reared and ewe-reared lambs.

	Total lambs n	Live lambs n	Dead lambs n	Lamb survival %
Rearing method				
AR <sup>1</sup>	24	15	9	62.5
ER <sup>2</sup>	23	21	2	91.3
				Ns <sup>3</sup>
Litter size				
Single	13	12	1	92.3
Twin	19	13	6	68.4
Triplet	15	11	4	73.3
				Ns
Rearing method x Litter size				
AR-Single	5	4	1	80.0
AR-Twin	9	5	4	55.6
AR-Triplet	10	6	4	60.0
				Ns
ER-Single	8	8	0	100
ER-Twin	10	8	2	80
ER-Triplet	5	5	0	100
				Ns

<sup>1</sup>Artificially reared group, <sup>2</sup>Ewe-reared group, <sup>3</sup>Not significant

Table 3. Least squares means ( $\pm$  SE) for body weight and ADG in artificially and ewe-reared lambs.

Variables	n	Birth Weight	BW <sup>1</sup> at 1 Week (kg)	BW at 2 Weeks (kg)	BW at 3 Weeks (kg)	ADG at Birth to 3 Weeks (g)
Rearing method		Ns <sup>2</sup>	Ns	***	***	***
ER <sup>3</sup>	10	3.14 $\pm$ 0.14	4.38 $\pm$ 0.46	6.44 $\pm$ 0.34	7.78 $\pm$ 0.32	265.23 $\pm$ 22.72
AR <sup>4</sup>	10	3.17 $\pm$ 0.15	3.94 $\pm$ 0.28	4.53 $\pm$ 0.31	5.51 $\pm$ 0.29	135.67 $\pm$ 20.78
Litter size		Ns	Ns	Ns	Ns	Ns
Twin	14	3.28 $\pm$ 0.13	4.57 $\pm$ 0.25	5.62 $\pm$ 0.28	6.87 $\pm$ 0.26	210.75 $\pm$ 18.76
Triplet	6	3.03 $\pm$ 0.16	3.72 $\pm$ 0.31	5.35 $\pm$ 0.35	6.42 $\pm$ 0.33	190.15 $\pm$ 23.10
Lamb sex		Ns	Ns	Ns	Ns	Ns
Female	12	3.20 $\pm$ 0.16	3.77 $\pm$ 0.33	5.42 $\pm$ 0.37	6.75 $\pm$ 0.34	198.56 $\pm$ 24.37
Male	8	3.10 $\pm$ 0.13	4.55 $\pm$ 0.26	5.55 $\pm$ 0.29	6.54 $\pm$ 0.93	202.34 $\pm$ 19.45
Overall mean	20	3.18 $\pm$ 0.58	4.20 $\pm$ 1.77	5.58 $\pm$ 0.18	6.70 $\pm$ 0.21	202.42 $\pm$ 30.69
Rearing method X Litter size		Ns	Ns	Ns	Ns	Ns
Rearing method X Lamb Sex		Ns	Ns	Ns	Ns	Ns

<sup>1</sup>Body weight, <sup>2</sup>Not significant, <sup>3</sup>Artificially reared group, <sup>4</sup>Ewe-reared group, \*\*\* P < 0.001

Table 4. Plasma total protein, albumin, globulin, glucose, urea, and total lipid concentrations in artificially and ewe-reared lambs.

		0 h	6 h	18 h	24 h	1 week	2 weeks	3 week
T. Protein (g/dl)	ER <sup>1</sup>	5.84 $\pm$ 0.38	7.77 $\pm$ 0.75	10.21 $\pm$ 0.67***	9.61 $\pm$ 0.87***	9.27 $\pm$ 0.90**	7.41 $\pm$ 0.27**	7.29 $\pm$ 0.44
	AR <sup>2</sup>	6.95 $\pm$ 0.62	7.43 $\pm$ 0.43	5.91 $\pm$ 0.52	5.75 $\pm$ 0.31	6.43 $\pm$ 0.64	5.17 $\pm$ 0.28	6.28 $\pm$ 0.57
Albumin (g/dl)	ER	3.54 $\pm$ 0.33	3.45 $\pm$ 0.33	3.67 $\pm$ 0.25	3.59 $\pm$ 0.22	3.33 $\pm$ 0.19	3.49 $\pm$ 0.44	3.96 $\pm$ 0.24
	AR	4.19 $\pm$ 0.39	4.16 $\pm$ 0.42	2.74 $\pm$ 0.16	3.03 $\pm$ 0.15	3.42 $\pm$ 0.16	3.20 $\pm$ 0.16	3.69 $\pm$ 0.29
Globulin (g/dl)	ER	2.30 $\pm$ 0.18	4.32 $\pm$ 0.32	6.54 $\pm$ 0.58***	5.96 $\pm$ 0.44***	5.94 $\pm$ 0.33**	3.92 $\pm$ 0.49**	3.33 $\pm$ 0.23
	AR	2.76 $\pm$ 0.21	3.27 $\pm$ 0.32	3.23 $\pm$ 0.27	2.72 $\pm$ 0.16	3.01 $\pm$ 0.33	1.97 $\pm$ 0.10	2.59 $\pm$ 0.21
Glucose (mg/dl)	ER	11.22 $\pm$ 1.64	28.09 $\pm$ 8.88	51.20 $\pm$ 8.46	57.71 $\pm$ 6.10	60.23 $\pm$ 5.00	80.64 $\pm$ 6.71	102.65 $\pm$ 12.64
	AR	16.23 $\pm$ 3.18	20.35 $\pm$ 6.43	48.59 $\pm$ 7.73	54.50 $\pm$ 7.73	61.69 $\pm$ 7.35	64.42 $\pm$ 7.03	80.90 $\pm$ 9.78
Urea (mg/dl)	ER	41.94 $\pm$ 4.30	42.85 $\pm$ 2.81	59.90 $\pm$ 4.43***	70.98 $\pm$ 2.89***	54.14 $\pm$ 2.15	44.10 $\pm$ 3.90	42.07 $\pm$ 4.98
	AR	43.82 $\pm$ 3.22	36.57 $\pm$ 2.31	29.26 $\pm$ 1.63	32.30 $\pm$ 2.78	44.77 $\pm$ 4.26	35.08 $\pm$ 1.92	44.30 $\pm$ 2.76
T. Lipid (mg/dl)	ER	126.41 $\pm$ 9.08	214.78 $\pm$ 20.31	183.63 $\pm$ 23.82	255.54 $\pm$ 43.71***	249.68 $\pm$ 43.62**	218.11 $\pm$ 38.81	255.17 $\pm$ 34.75
	AR	131.37 $\pm$ 20.65	124.25 $\pm$ 25.12	109.92 $\pm$ 26.06	99.81 $\pm$ 12.85	127.34 $\pm$ 64.51	151.67 $\pm$ 22.65	206.63 $\pm$ 25.58

<sup>1</sup>Artificially reared group (n = 10), <sup>2</sup>Ewe-reared group (n = 10), \*\*P < 0.01, \*\*\*P < 0.001

respectively). Plasma urea concentration in the ER group increased after birth and reached a peak concentration at 24 h. At week 1, urea concentration of the ER group showed a rapid decrease and approached that of the AR group. Plasma total lipid concentrations of the ER group at 24 h (P < 0.001) and first week (P < 0.01) were significantly higher than those of the AR group.

## Discussion

Sufficient colostrum intake in the first days following birth plays an essential role in survival rate, resistance to infections, and ADG of newborn lambs (2). Emsen et al. (11) reported that the survival rate of Awassi lambs fed on ewe milk and calf milk replacer was 75% and 85%, respectively. They explained this result by the Awassi

lambs' adaptation to calf milk replacer. The reason why the lamb survival rate in the present study is different from those of Awassi lambs is that Awassi lambs sucked colostrum during an initial stay with the ewes of 2-3 days. In the present study, separation of newborn lambs from their mothers occurred immediately after birth, which resulted in the lower survival rate. Colostrum is the only IgG source for newborns (8). There is a close relationship between IgG concentration in colostrum and that in blood (14). IgG plays an important role in building passive immunity and acquiring resistance to infectious illnesses (10). Argüello et al. (15) reported that animals with low concentrations of blood IgG had relatively higher mortality rates.

Emsen et al. (11) also studied the effect of artificial rearing on the body weight of Awassi lambs. They reported that the body weight in their ER group was significantly higher than those of the AR group in the 2<sup>nd</sup> and 4<sup>th</sup> weeks after birth, while ADG was not different. In this study, the reason why body weight and ADG in the ER group were higher than those in the AR group could also be explained by the immediate separation of the lambs from ewes, with no colostrum intake.

Chen et al. (6) reported that plasma protein concentrations reached a peak concentration within 24 h in lambs that sucked colostrum and then appreciably decreased. Likewise, Argüello et al. (15) reported that IgG absorption reached a maximum concentration between 18 and 48 h after birth. In the present study, artificial rearing did not significantly affect total protein and globulin concentrations at 0 and 6 h, and the 3<sup>rd</sup> and 4<sup>th</sup> weeks. However, significant effects were determined at 18 and 24 h, and the 1<sup>st</sup> and 2<sup>nd</sup> weeks. We can explain these results by the higher absorption of IgG between 18 h and the second week compared to other sampling intervals.

The lack of differences in plasma glucose concentrations depends on the gluconeogenesis processes of AR lambs. Fonnesbeck and Symons (16), and

Berschauer et al. (17) reported that blood urea concentration was a useful indicator of the status of protein metabolism and that changes in urea concentrations were a reflection of changes in both dietary nitrogen metabolism and in protein metabolism. The reason why urea concentration was higher in the ER lambs of the present study within their first 24 h was probably due to the richer protein content of colostrum compared to calf milk replacer, indicating increased protein utilisation in the ER group. The decrease of urea in the ER group after the first week could have been due to the conversion of colostrum to normal milk in the ewes (18).

Composition of the colostrum is subjected to change after birth. Lipid content of colostrum has been reported to be higher 6 and 24 h after birth than at 1 h (18). Total lipid concentrations in our ER group were higher than in the AR group at 6 and 24 h after birth. This difference can be explained by the fact that fat concentration continuously increased in colostrum, while it remained constant in the calf milk replacer. Mersmann and McNeil (19) reported that fatty acids in blood were the source of triglyceride synthesis in adipose tissue. Lower total lipid concentration found in the AR lambs of the present study resulted in a smaller accumulation of fat in their adipose tissue. This also explains the differences in body weights between the groups (Table 3). At week 3, total lipid concentration of AR lambs approached that of ER lambs. We can speculate that this was because, in addition to milk or calf milk replacer, the lambs were allowed to have hay and concentrated feed so that rumen functions would start to develop.

Results indicated that the lack of maternal colostrum and milk had detrimental effects on lamb survival and growth rates; thus, it cannot be suggested as a valid rearing method. Colostrum intake by newborn lambs immediately after birth and the following weeks significantly increased plasma IgG concentration, which affects resistance to infectious diseases as well as survivability.

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