

Risk Factors Associated with Passive Immunity, Health, Birth Weight and Growth Performance in Lambs: III- The Relationship among Passive Immunity, Birth Weight, Gender, Birth Type, Parity, Dam's Health, and Lambing Season ^[1]

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Summary

This study was designed to investigate the effect of risk factors such as gender, birth type, parity, dam's health and lambing season associated with passive immunity and birth weight and to also determine interrelationship between passive immunity and birth weight. This study included 301 ewes and 347 lambs born to them on two local Akkaraman crossbred flocks in Kars. Lambs were blood sampled for serum IgG concentration at 24 hours after birth (SlgGC-24) and epidemiological parameters were recorded at birth. Parity, type of birth and gender were significantly associated with birth weight ($R^2=0.339$, $P<0.001$) on multivariable stepwise regression analysis as single born lambs ($P<0.001$), male ($P<0.001$) and lambs born to dams previously lambed ($P<0.001$) were significantly heavier. There was a significant ($P<0.001$) and positive ($R^2=0.136$) linear relationship between birth weight and passive immunity, but only in ill lambs during the neonatal period. In General Linear Model (GLM), lambs born with low birth weight (≤ 3 kg) had significantly lower SlgGC-24 than those born with medium (>3 to ≤ 4) ($P<0.01$) or high (>4 kg) ($P<0.001$) birth weight. Similarly lambs born as a twin and born to unhealthy dams had significantly lower SlgGC-24 ($P<0.05$). In conclusion, some farm management practices and animal characteristics were associated with birth weight and passive immunity and also birth weight had affect on passive immunity. For a productive and profitable farming, producers should take these variables into account and develop appropriate management strategies.

Keywords: Lamb, Serum IgG, Passive immunity, Birth weight, Risk factors

Kuzularda Pasif İmmünite, Sağlık, Doğum Ağırlığı ve Büyüme Performansı İle İlgili Risk Faktörleri: III- Pasif İmmünite, Doğum Ağırlığı, Cinsiyet, Doğum Tipi, Anne Sağlığı, Doğum Sayısı ve Kuzulama Sezonunun Birbiriyle İlişkisi

Özet

Bu çalışma pasif immünite ve doğum ağırlığı üzerine etkili cinsiyet, doğum tipi, kuzulama sezonu, anne doğum sayısı ve sağlığı gibi bazı risk faktörlerinin araştırılması ve ayrıca pasif immünite ve doğum ağırlığının birbiriyle olan ilişkisinin belirlenmesi amacıyla gerçekleştirildi. Çalışma Kars'ta 301 koyun ve bunlardan doğan 347 Akkaraman melezi kuzuyu içeren iki sürüde yürütüldü. Doğumdan 24. saat sonra serum IgG konsantrasyonlarını (SlgGC-24) belirlemek için kan örneği alındı ve doğumda epidemiyolojik parametreler kaydedildi. Çoklu adımsal regresyon analizine göre doğum tipi, cinsiyet ve anne doğum sayısı, doğum ağırlığını önemli seviyede ilişkili faktörler olarak belirlendi ($R^2=0.339$, $P<0.001$). Tek ($P<0.001$), erkek ($P<0.001$) ve daha önce doğum yapmış annelerden doğan ($P<0.001$) kuzuların doğum ağırlığı daha yüksek bulundu. Yalnızca neonatal periyotta hastalık tespit edilen kuzularda pasif immünite ve doğum ağırlığı arasında önemli ($P<0.001$) ve pozitif ($R^2=0.136$) bir linear ilişki olduğu belirlendi. Genel linear modele göre düşük doğum ağırlığı (≤ 3 kg) ile doğan kuzuların SlgGC-24'ları orta (>3 - ≤ 4 kg) veya yüksek (>4 kg) doğanlara göre önemli seviyede (sırasıyla $P<0.01$ ve $P<0.001$) düşük belirlendi. Benzer şekilde ikiz veya hasta annelerden doğan kuzuların SlgGC-24'ları önemli seviyede ($P<0.05$) düşük bulundu. Sonuç olarak bazı çiftlik sevk-idare uygulamaları ve hayvan karakteristikleri doğum ağırlığı ve pasif immünite ile ilişkili bulundu ve ayrıca doğum ağırlığı pasif immünite üzerine etkiledi. Çiftlik verim ve karlılığını arttırmak için üreticilerin bu faktörleri göz önünde tutması ve uygun sevk-idare stratejileri geliştirmesi gerekir.

Anahtar sözcükler: Kuzu, Serum IgG, Pasif İmmünite, Doğum Ağırlığı, Risk Faktörleri



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INTRODUCTION

Morbidity and mortality are often cited as the most common problems in young lambs and it is a major cause of low productivity of sheep [1-3]. Lambs are born with hypogammaglobulinemia due to syndesmochorial placentation and must be fed colostrum as a source of immunoglobulin-G (IgG) during the neonatal period. This process is named as passive immunity, determined by measuring serum IgG concentration at 24 h after birth (SIgGC-24). Numerous studies in the past three decades correlated neonatal diseases with inadequate serum IgG, or failure of passive transfer of immunity (FPT) in animals and pointed out the importance of IgG in preventing infectious diseases of neonates [4-8]. Recent studies also reported a significantly positive relationship between SIgGC-24 and growth performance in lambs at different stages [9-12]. It is therefore, of paramount importance to investigate the underlying causes of FPT. Only a small number of studies have been conducted to clarify the effect of some environmental factors and animal characteristics on passive immunity in lambs, but these effects were not clearly elucidated [6-8,13-15].

All mammalian species have an 'optimum' birth weight that facilitates uncomplicated natural delivery and improves the rate of neonatal survival [16]. Thus, the birth weight of lambs plays an important role in achieving desirable sheep production as studies determined that birth weight is associated with enhances growth performance and decreased disease incidence [9,17-21]. The birth weight has also been associated with passive immunity [4]. Low birth weight leads to many unfavorable factors including low growth rate especially in the first three months of life, increased risk of morbidity and mortality because these lambs are physically weak to stand and suckle adequate colostrums and less viability at birth that have significant effects on lamb survival and growth performance [9,15,17,22-25]. Two greatest important contributors to problems caused by morbidity, mortality and poor growth performance are low birth weight and FPT [26].

Production and profitability of sheep farms explicitly depends on maintaining healthy lambs with desirable live weight gain. Therefore, it is of paramount importance to identify the cause of diseases and weight loss in lambs and to take appropriate measures accordingly.

The study was designed to determine the relationship between passive immunity and birth weight and also to investigate presumptive effects of gender, type of birth, lambing season, parity and dam's health on passive immunity as well as to investigate the effect of environmental factors and animal characteristics on birth weight in Akkaraman crossbred which constitutes 50% of the total sheep population in Turkey [27].

MATERIAL and METHODS

Animals, Data Collection, Farms Management

This study was carried out in two sheep farms located in the central Kars in North-Eastern Anatolia, Turkey, in 2009. All ewes and lambs were kept under identical feeding and management conditions. Management was typical of North-eastern Anatolian flocks with lambs being born in winter (December to February) or spring (March to May), and being raised intensively. At birth, the lambs were ear-tagged and registered with an individual identification number, and gender, date of birth, parity, and dam's ear tag number and type of birth were recorded for each lamb. The lambs were weighed at birth (before colostrum intake) using a bascule [CASIA DB2-150 kg (± 30 g)]. After this procedure, lambs were allowed to naturally suckle their dams.

Blood Sample Collection, IgG Analysis

Blood samples were collected from all lambs by jugular venipuncture at 24 ± 1 h. Serum was harvested by centrifugation and stored at -20°C until analysis. Passive immune status or serum IgG concentrations 24th h after birth (SIgGC-24) were measured using a commercial ELISA kits (Bio-X Competitive ELISA Kit For Ovine Blood Serum IgG Assay-BIO K 350, Bio-X Diagnostics, Belgium).

Clinical Examination

Clinical examination were performed as previously defined by the authors [2]. The health status of the lambs was monitored on farms by visits made on a daily basis during the neonatal period. Throughout the study period, ewes determined to have disease (mastitis, pneumonia, enteritis, pregnancy toxemia etc.) were categorized as ill and recorded with their ear tag number.

Statistical Analysis

The present study included 301 Akkaraman crossbreeds and 347 lambs born to them. However, lambs whose parameters (birth weight, gender, type of birth, health status, passive immunity or serum IgG level and birth date for lambs; parity and health status of dams) were not recorded were excluded from the study thus leaving 322 lambs to be included in this study.

Study consisted of two sections. In first section, the affect of gender, type of birth, lambing season, parity and dam health on the birth weight was studied. Mean \pm SD (Range) values for each parameter was calculated. Parity was categorized as 1, 2, 3 and ≥ 4 . The Tukey HSD test was used to identify differences in birth weight in lambs grouped according to parity. Independent Samples T test was used to identify variations in birth weight according to gender, type of birth, lambing season and health of the dam. Multivariable stepwise linear regression analysis (MSRA)

was used to evaluate the association between type of birth, gender, health of dams and lambing season (considered as categorical independent variables), and parity (considered as continuous independent variables) and birth weight (continuous dependent variables). The linear regression model with the all potential independent variables considered in the study defined as follows: $Y = \alpha + \beta_1 x_1 + \beta_2 x_2 + \beta_3 x_3 + \beta_4 x_4 + \beta_5 x_5 + \epsilon$ Where Y denotes birth weight, x_1 is the type of birth (single=0, twin=1), x_2 is the gender (male=0, female=1), x_3 is the parity (1=1, 2=2, 3=3, $\geq 4=4$), x_4 is the lambing season [Spring (March, April, May)=0, Winter (December, January, February)=1], x_5 is the health of the dam (healthy=0, ill=1), α is the y-intercept, and $\beta_1, \beta_2, \beta_3, \beta_4$ and β_5 are the regression coefficients and ϵ , indicates the random part of the theoretical model. To identify the best models in the stepwise technique, the coefficients of determination (R^2) were used. The coefficient of determination was multiplied by 100 and expressed as a percentage to indicate the total variation in Y explained by the selected independent variables.

In second section the effect of birth weight, gender, type of birth, lambing season, parity and dam's health on passive immune status or SlgGC-24. Simple regression model was used to evaluate relationship between SlgGC-24 and birth weight. The methods of multivariable and simple regression have been described previously in detail [11,28]. The General Linear Model (GLM) procedure of SPSS was used to evaluate the association between some animals or management factors and SlgGC-24. The fixed effects considered in the model were: birth weight (≤ 3 kg=1, >3 to ≤ 4 kg=2, >4 kg=3), gender (female=1 and male=2), parity (1, 2, 3, ≥ 4), type of birth (twin=1 and single=2), health status in dams (ill=1 and healthy=2) and lambing season (winter=1, spring=2). The significant differences between fixed items were tested using Duncan test and results were expressed as least square means (LSM \pm SE) for the GLM procedure. The computer program SPSS (SPSS, version 16.0, SPSS Inc, Chicago, IL) was used for all analyses and values of $P < 0.05$ were considered to be significant. The program Origin 6 was used to obtain scatter diagram illustrations (Origin 6 Copyright© 1991-1999 Microcol TM, Software, Inc) in regression analysis.

RESULTS

Risk Factors Associated with Birth Weight

Birth weight ranged from 2.260 to 5.900 g (4.037 \pm 674 g). Variations in birth weight based on various factors have been given in Table 1. Single born lambs were significantly ($P < 0.001$) heavier at birth than twin born lambs. Males were significantly ($P < 0.001$) heavier than females at birth. Lambs born to dams with any illnesses had significantly ($P < 0.001$) lower birth weight than lambs born to healthy dams. Lambs born in winter had insignificant higher birth weight than those born in the spring season ($P = 0.08$).

Birth weight in lambs born to primiparous ewes was significantly lower than those born to second ($P < 0.01$), third ($P < 0.001$) and fourth or higher ($P < 0.001$) parity ewes.

Type of birth, parity and gender had significant effect on birth weight on MSRA. These three independent variables explained 34% ($R^2 = 0.339$) of the variation in birth weight in final model. High dam's parity had lambs with great birth weight while twin and female lambs had lower birth weight ($P < 0.001$) (Table 2).

Relationship Between Birth Weight and Passive Immunity in Lambs

Relationship between SlgGC-24 and birth weight (BW), determined on simple regression model is given in Table 3. There was a significant ($P < 0.001$) but weak ($R^2 = 0.076$) linear relationship between SlgGC-24 (mg/ml) and BW in lambs that contracted disease during the neonatal period. However, there was no relationship between SlgGC-24 and BW in lambs that were healthy during the neonatal period (Table 3).

Table 1. Variations in birth weight according to factors analysed in lambs

Tablo 1. Kuzularda analiz edilen faktörlere bağlı doğum ağırlığındaki değişiklikler

Factor	Group	n	Birth Weight (g)
Type of Birth	Twin	84	3505 \pm 436*
	Single	238	4224 \pm 643
Sex	Male	173	4157 \pm 670*
	Female	149	3896 \pm 652
Lambing Season	Winter	98	3938 \pm 696
	Spring	224	4079 \pm 661
Dam Health	Ill	17	3442 \pm 619*
	Healthy	305	4069 \pm 662
Parity	1	54	3650 \pm 652 ^a
	2	137	4048 \pm 662 ^b
	3	90	4138 \pm 582 ^b
	≥ 4	41	4284 \pm 740 ^b

* $P < 0.001$

Table 2. Risk factors associated with birth weight on multiple regression analyses*

Tablo 2. Doğum ağırlığı ile ilişkili risk faktörleri üzerine adimsal regresyon modelleri*

Model	Formulas	R ²	P
Onset	BW = 4224 - (719 x type of birth)	0.220	<0.001
1	BW = 3676.9 - (781.3 x type of birth) + (238.1 x parity)	0.321	<0.001
Final	BW = 3764.8 - (759.5 x type of birth) + (233.6 x parity) - (179.5 x gender)	0.339	<0.001

* **Independents:** Type of birth, gender, lambing season, dam's parity and health, **BW (Dependent):** Birth weight

Table 3. Simple regression models between SlgGC-24 and birth weight in neonatal lambs**Tablo 3.** Neonatal Kuzularda SlgGC-24 ve doğum ağırlığı arasındaki basit regresyon modelleri

N	Formulas	R ²	P
322	SlgGC-24 ^a = 8.024 + (0.003 x BW)	0.034	<0.01
269	SlgGC-24 ^b = 21.12 + (0.001 x BW)	0.001	0.53
53	SlgGC-24 ^c = (0.006 x BW) - 9.017	0.136	<0.001

^a General (without any evaluation of clinical examination), ^b healthy, ^c ill
SlgGC-24: Serum IgG concentrations 24th h after birth

Table 4. Risk factors associated with passive immunity (SlgGC-24) in lambs**Tablo 4.** Kuzularda pasif immunité ile ilişkili (SlgGC-24) risk faktörleri

Factor	Group	N	SlgGC-24 (mg/dl)	F	P
BW (g)	Low	27	1246±242 ^a	3.17	0.04
	Medium	122	1858±167 ^b		
	High	173	1837±173 ^b		
Type of Birth	Twin	84	1466±168*	4.51	0.03
	Single	238	1829±173		
Gender	Male	173	1567±160	1.55	0.21
	Female	149	1727±160		
Lambing Season	Winter	98	1512±176	2.99	0.08
	Spring	224	1782±158		
Dam Healthy Status	Ill	17	1333±277*	4.55	0.03
	Healthy	305	1962±102		
Parity	1	54	1553±206	1.37	0.25
	2	137	1557±165		
	3	90	1864±192		
	≥4	41	1614±221		

SlgGC-24: Serum IgG concentrations 24th h after birth

Risk Factor for Passive Immunity in Lambs

Factors affecting passive immunity in lambs are presented in Table 4. Of the variables, birth weight, type of birth and health of the dam had a significant effect on the SlgGC-24 of the lambs. The overall least squares mean of SlgGC-24 obtained was 1.647±148 mg/dl. Twin lambs had significantly (P<0.05) lower SlgGC-24 than single lambs. Lambs born with low birth weight (≤3 kg) had significantly lower SlgGC-24 than those born with medium (>3 to ≤4) (P<0.01) or high (>4 kg) (P<0.001) birth weight. SlgGC-24 was significantly lower in lambs born to diseased dams than in lambs born to healthy dams (P<0.05). Parity and gender had no effect on SlgGC-24. A lower SlgGC-24 was determined in lambs born in the winter but this association was not significant (P=0.08).

DISCUSSION

This study presented the factors influenced birth

weight and passive immunity and described the relationship between them.

Risk Factors Associated with Birth Weight

Parity, type of birth and gender were significant sources of variation for birth weight in the present study. Primiparous mothers produced lighter lambs when compared to experienced ewes as previously reported [16-21,23,24,29-33]. This may be attribute to that the reproductive organs of primiparous ewes are less developed and less able to bear large fetuses, so the dam's physiology limits the fetal size [30-33] and these ewes may be in the process development, so both the fetus and the dam might compete for nutrients thus consequently negatively influencing birth weight [20,30-32]. Contradicting results between parity and birth weight have also been reported [4,14,34-37]. In this study, lambs born to healthy dams had greater birth weight than those born to sick dams (Table 1). This may be because unhealthy dams provide inadequate nutrition to the fetus, and thus negatively effected birth weight. However, the condition of the dam's health was not a significant source of variation for birth weight as there were only seventeen sick dams in this study versus 305 healthy dams.

Female lambs had lower birth weight than their male counterparts in this study. This findings is in agreement with previous studies [20-24,29,31-33,36-38]. These results have been attributed to the differences in the rate of skeletal development as well as differences in chromosomal structure during the prenatal growth period [34] as the presence of a Y-chromosome and the products of SRY gene activation have gender-specific effects on fetal growth, and therefore males apparently grow faster in utero than respective females [16,32]. However, opposite results have also been reported that birth weight did not differ between male and female lambs [4,14,18,30,39].

Single born lambs were heavier than their twin born counterparts at birth in this study. These results were in parallel with previous studies [20-22,29-34,36-39]. Birth weight declines as the litter size increases due to limited uterus and carnuncles space to gestate offspring and insufficient nutrient provided for the development of all the fetuses [16,30].

Lambs born in winter were insignificantly lighter than those born in spring as previously reported [22,32,34]. In winter, the need for nutrition and energy increases due to cold stress and the quality and amount of food sources deteriorates, so insufficient diet may affect fetal growth and consequently cause low birth weight as reported by Sušić et al. [22].

In this study the variable with the highest influence was found to be birth type, whereas parity and gender were found to have secondary influence in MSRA analyses. However, in the present study, a large proportion of the

variation in birth weight (approximately 65%) was not explained by the variation in type of birth, gender and parity, but was attributable to some other factor. These include, but are not limited to, management intervention, the farming production system, prenatal nutrition of ewes, sufficient placentation, average weight and number of cotyledons as well as dam's body condition score and live weight gain^[11,16,23-26,40]. The amount of variation attributable to farm management procedures, the production system and the ewe's gestational nutrition and breed was minimized in this study because all lambs were taken from two farms that have similar management practices and reared Akkaraman crossbreeds.

Low birth weight decreases yield and profitability in sheep farms due to its negative effect on growth performance and passive immunity and consequently increased predisposition to diseases^[4,6,11,17,41]. Therefore, efforts should be made to develop management practices that increase birth weight such as supplementation of ration with enough concentrated feed and protein in the third trimester^[23,24,42].

Relationship Between Passive Immunity and Birth Weight

There was a significant but poor positive linear relationship between birth weight and SIgGC-24 in sick lamb in neonatal period. This may be due to the fact that lambs born with low birth weight might have not received colostrum due to physical weakness and abnormal behavior at birth, leading to weak relationship with their mother, poor suckling reflex and less vigority^[19,26,40,43]. Birth weight had a significant effect on passive immunity since lambs with a birth weight of ≤ 3 kg had a significantly lower SIgGC-24 than did lambs with a birth weight of >3 to ≤ 4 or >4 kg on GLM in this study. Our study also determined that based on simple regression analysis of variables from 322 lambs without grouping them according to clinical examination, there was a positive linear relationship between birth weight and passive immunity. These results have been confirmed by previous studies^[4,5,24,44]. On the other hand, we did not detect a significant relationship between birth weight and SIgGC-24 in healthy lambs during the neonatal period in our study. This is consistent with the results of other studies in healthy calves^[45], lambs^[11,13], and kids^[46]. However, the influence of birth weight on passive transfer of IgG in neonatal ruminants has not yet been fully elucidated. Massimini et al.^[11], hypothesized that the negative relationship between birth weight and the acquisition of passive immunity^[48,49] in newborn lambs could be an indirect link that reflects the effects of other important physiological factors, such as duration of gestation and hormonal status at birth but the potential influences of these independent variables were not evaluated in this study.

Other Factors Effect on Passive Immunity

Gender had no effect on passive immunity in this study but SIgGC-24 of male lambs was slightly higher than females. This is similar to previous reports^[6-8,12-15,44,50]. Twin lambs had lower SIgGC-24 than single lambs in the present study, which concurs with previous findings^[8,44]. This may be attributed to that twin lambs have to compete for colostrums and are physically weak and with low birth weight thus being unable to suckle a sufficient amount of colostrum, leading to low IgG levels in their serum. On the other hand, it has been reported that ewes with twins have higher colostrum production than ewes with single lambs but immunoglobulin concentrations in the lambs fall significantly in proportion with the litter size^[6,7].

Passive immunity is reported to be affected by the lambing season^[15]. Cold-stressed newborn ruminants may have a slower rate of intestinal immunoglobulin absorption^[51], and may also be reluctant to stand and suckle voluntarily^[6,43,52] consequently, an increased risk of failure of passive transfer is expected in the period from December to February, which is also the coldest period of the year in Kars. Nevertheless, winter born lambs had insignificant ($P=0.08$) lower SIgGC-24 in our study as lambs born indoors, this might have reduced the potential impact of cold stress^[50].

The level of IgG in lambs born to primiparous ewes was observed to be insignificantly lower than in those born to ewes giving birth previously in this study. These results are in line with some earlier studies in lambs^[4,6,12,13,44]. Primiparous ewes may have a lower volume, concentration, or quality (low IgG level) of colostrum and poorer maternal ability than do mature ewes^[7,23,43,53] and lambs born to primiparous ewes are with low birth weight and weak to stand to suckle as was the case in this study^[7,23,53]. On the other hand, dam's health had a significant effect on passive immunity in our study. Healthy dams produce good quality colostrum, which also influences the absorption of immunoglobulin from the intestines, and such dams also have good maternal ability^[4,23,26,40,43,53] as might have been the case in our study.

In conclusion, the present study identified some important environmental factors and animal characteristics affecting passive immunity and birth weight in lambs in such details in the world and for the first time in Turkey. These results indicated that for proper lambing management, consideration should be given to maximizing the health of the flock and supplementing feed ratio of the ewes during gestation that are primiparous, carrying twin and with ill-health. Consequently, sheep farmers need to ensure that each lamb remains healthy and gains sufficient live weight in order to increase productivity and profitability.

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