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Effect of rearing system on pre-weaning growth, rumen development and its influence on post-weaning performance of lambs

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Abstract

Twenty-four 21-day-old Balouchi male lambs were equally divided into two groups. Twelve subjects were artificially reared (AR), twelve others were ewe reared (ER) and used as control. AR in comparison of ER lambs maintained higher concentrations of blood beta-Hydroxybutyric acid (BHBA); however glucose concentration was not affected by rearing system. Unlike the DNA content and cell size, the RNA concentration and ribosomal capacity (Cs) of AR groups were significantly ($P<0.05$) higher than that of ER lambs. Rearing system did not affect morphologic characteristics of rumen wall except thickness of keratinized layer that was thickest in AR, 20 % less than in ER lambs ($P<0.05$). Stomach weight and capacity in AR animals were significantly ($P=0.05$) higher than ER lambs. Neither post-weaning growth rate nor feed conversion efficiency were affected ($P>0.05$) by rearing method. Also there were no differences with respect to slaughter and dissection data between groups in post-weaning phase. In conclusion, the results of this study showed that naturally rearing system gave rise to developmental and carcass characteristics similar to those observed in artificial raised lambs.

Keywords: sheep, lamb, rearing system, rumen development, growth

Zusammenfassung

Auswirkungen der Aufzuchtmethode von Lämmern auf deren Wachstum vor dem Absetzen, die Pansenentwicklung und auf ihre Schlachtleistung nach dem Absetzen

Einundzwanzig Tage alte Balouchi Lämmer wurden in zwei gleich große Gruppen eingeteilt. Zwölf Tiere wurden ohne das Mutterschaf (AR), zwölf andere zur Kontrolle durch das Mutterschaf (ER) aufgezogen. AR-Lämmer wiesen im Vergleich zu ER-Lämmern zwar eine höhere Konzentration an 3-Hydroxybutansäure (BHBA) im Blut auf, die Glukoseproduktion wurde jedoch nicht durch die Aufzuchtmethode beeinflusst. Im Gegensatz zu DNA-Gehalt und der Zellgröße, war die RNA-Konzentration und die ribosomalen Kapazität (Cs) bei der AR-Gruppe signifikant höher ($P<0,05$) als bei der ER-Gruppe. Die Aufzuchtart hatte keinen Einfluss auf die morphologischen Merkmale der Pansenwand außer auf die Dicke der verhornten Schicht, die bei der AR-Gruppe bis zu 20 % dünner war als bei der ER-Gruppe ($P<0,05$). Magengewicht und -kapazität waren bei der AR-Gruppe signifikant höher ($P=0,05$) als bei der ER-Gruppe. Die Aufzuchtmethode wirkte sich weder das Wachstum nach dem

Absetzen noch die Futterverwertung aus (P>0,05). Es gab auch keine Unterschiede zwischen den Gruppen nach dem Absetzen, was die Schlacht- und Sektionsdaten zeigten. Die Studie zeigte, dass die Aufzucht von Lämmern durch Mutterschafe zu ähnlichen Entwicklungs- und Schlachtkörpermerkmalen führt wie die Aufzucht ohne Mutterschafe.

Schlüsselwörter: Schaf, Lamm, Aufzuchtmethode, Pansenentwicklung, Wachstum,

Introduction

In addition to genotype, animal performance can be affected by the rearing system (Napolitano *et al.* 2002, Osorio *et al.* 2006). Naturally rearing in grass-based sheep production system offers the potential of economic animal production to reduce labour and fixed costs. However, offering grass or grazing as the sole foodstuff to growing lambs cannot support acceptable growth rate. In order to achieve higher weight and quality carcass, necessary nutritional interventions are required in lamb from birth till slaughter (Bhatt *et al.* 2009). Under fat lamb production system al lib creep mixture feeding promote faster rate of growth (average daily gain [ADG] of 140 g, Singh *et al.* 1984) during pre-weaning phase, which could be considered optimum. Similarly, mutton synthetic lambs (Malpura×Dorset and Suffolk) maintained on free suckling and with ad lib creep mixture and cow pea hay feeding attained a growth rate of 208 g/day (Singh & Singh 1987). However, artificial rearing systems base on separation lambs from the ewe at an early age is often associated with reduced animal welfare, as indicated by altered behavioural, endocrine and immune responses (Napolitano *et al.* 1995, Sevi *et al.* 1999). A number of authors demonstrated that the lack of the maternal bond can inhibit the welfare state of the lambs (e.g., Sevi *et al.* 2003) and poor animal welfare can have detrimental effects on performance and meat quality in many animals species as well as in sheep (Napolitano *et al.* 2002).

There are several studies comparing performance and welfare of naturally and artificially reared lambs of local breeds (e.g. Napolitano *et al.* 2006, Bhatt *et al.* 2009), but there is no sufficient data about the effect of rearing system on pre-weaning rumen development and post-weaning performance of lambs. Therefore, present experiment aim is to asses the effect of different pre-weaning feeding regimes on rumen development and growth of lambs and its influence on post-weaning performance and growth.

Materials and methods

Twenty-four 21-day-old Balouchi male lambs were divided into two groups of 12 in each. Ewe reared (ER) animals were raised exclusively on maternal milk. Throughout the course of the experiment ER lambs were kept with their dams. Dams were supplemented with hay and concentrate (0.5 kg alfalfa and 0.3 kg concentrate including 40 % corn, 20 % soybean meal, 20 % beet pulp and 20 % wheat bran) and at second day *postpartum* were allowed to graze at pasture for 6 h a day. Artificially reared (AR) lambs were removed from the ewes and housed in a separate straw-bedded pen and fed a starter diet containing 15 % alfalfa (Table 1). Lambs in groups 2 were allowed to be with their dams for 30 min twice daily and weaned when they were 9 weeks of age.

The animals were weighed at week 0 and then once a week before morning feeding. Blood samples were taken from the jugular vein at the beginning of the experiment (day 0) and the end of each week (up to week 9). The serum was separated after centrifugation at 1 800 g for 10 min and stored at −18 °C until analysis.

Table 1
Ingredients composition of diets fed to lambs during different experimental phases, % diet DM

	Diets*	
	Pre-weaning	Post-weaning
Feed		
Corn	58	40
Soybean meal	21	5
Alfalfa	15	33.5
Beet Molasses	4	-
Barley silage	-	20
Vit-Min Supplementation	0.4	0.3
White Salt	0.2	0.2
Limestone	1.4	1
Chemical Composition		
ME, kcal/kg	3.02	2.50
CP, %	19.95	14.05
NDF, %	15	29.74
ADF, %	8.4	19.74
Ca, %	0.81	0.82
P, %	0.40	0.48

*pre-weaning diet fed to AR and post-weaning diet fed to ER and AR lambs

Eight animals (4 per treatment) were slaughtered at 63 days of age. The entire digestive tract was removed, the ruminal compartment separated (the reticulo-rumen remained together), emptied, washed clean, drained of excess water and weighed. Samples (approximately 1 cm^2) were collected from the dorsal, ventral, caudal dorsal and caudal ventral blind sacs and pillar and atrium ruminis areas. Tissue samples were obtained within 30 min of death, placed in individual containers and fixed immediately in a 10 % formaldehyde solution for subsequent measurements. In the lab tissue samples dehydrated in a series of ethanol solutions from 70 to 100 %. The material was sectioned with an automatic microtome, at 6 µm thickness, stained with hematoxylin mixture and Eosin. The material was observed under a light microscope (Olympus BX-51) at 20× and 40×. Digital images of stained sections were taken using an Olympus BX-51 camera (DP 11) and measurements were made using image analysis computer software (DP2-BSW Version 1.3 from Olympus). Papillary height was defined as the distance from the tip to the base of the papillae and papillary width was defined as the average width of the base, middle and tip of the papillae. Papillae length and width were used to estimate surface area per cm^2 of each rumen section. Papillae denseness was determined using digital images, from Scanning Electron Microscopy (SEM, VEGA TESCAN, Czech Republic). Surface area of papillae per surface area of each ruminal section was presented as the surface area ratio (SAR) and calculated using the following methodology (Hill *et al.* 2005). Papillae were considered to be cylindrical in shape with one closed end. Therefore, equation 1 was used to calculate lateral area of papillae, based on the surface of a cylinder plus the area of a circle.

Equation 2 was used to calculate the average SAR of each section of the rumen by multiplying the average surface area of the papillae in each section by the average denseness or number of papillae per unit area in that section:

$$\textit{Surface Area of Papillae (cm}^2\textit{)} = 2 \times r \times \pi \times L + \pi \times r^2 \quad (1)$$

where r is the radius in cm, L is the length in cm.

$$\textit{SAR} = \textit{(average surface area of papillae in section X)} \times \textit{(average papillae denseness in section X)} \quad (2)$$

where *X* can be caudal, ventral, or dorsal.

For extract of total RNA and DNA the Trizol RNA Prep 100 kit and Accuprep Genomic DNA Extraction Kit; Cat No: K-3032 were used, respectively. Ribosomal capacity (the capacity for protein synthesis) and cell size was calculated as the ratio of RNA to protein and protein to DNA, respectively (Tesseraud *et al.* 1996).

The other lambs (8 animals in each group) were maintained under uniform feeding, fattening diet (Table 1), for 12 weeks and performance and carcass characteristics were determined.

Because blood parameters and rumen morphological characteristics were measured over the time and area, a repeated measures approach using ANOVA with mixed linear models in SAS 9.1 (SAS 2004) was used. The means were compared by the Duncan test.

Results and discussion

Pre-weaning phase

Blood Metabolites

As shown in Table 2, glucose did not differ significantly ($P>0.05$) between groups however was higher in artificial reared in comparison with the ewe reared lambs. Plasma glucose concentration declined with advancing the age ($P=0.13$). The concentrations were typical of non-ruminants at the beginning (96.4 mg/dl) but reached to a level of 63.5 mg/dl by weeks 9 of age (Figure 1). Plasma glucose concentrations reached to the typical level of mature ruminants post-weaning and did not differ afterward between treatments.

Table 2
Effects of rearing system on blood metabolites

Measurement	Group			Effect	
	ER	AR	SEM	Group	Time
Glucose, mg/dl	74.02	84.99	21.23	0.90	0.13
BHBA, mmol/l	0.27[b]	0.40[a]	0.03	0.001	0.001

ER: ewe reared lambs, AR: Artificial reared lambs, Values in the same row with different superscripts are significantly different.

Plasma concentrations of BHBA were significantly ($P=0.001$) affected by the group. Artificial reared in comparison of ewe reared lambs maintained higher concentrations of blood BHBA (0.27 and 0.40 mmol/l for ER and AR, respectively). It was suggested that increasing in blood BHBA were closely related to the availability of fermentable starter. Concentration of blood

BHBA in artificial reared animals likely reflected high production of butyrate in the rumen with subsequent metabolism to it by ruminal epithelium. Butyrate is a ketogenic acid in young animals (Sutton *et al.* 1963). Lambs in ewe rearing system maintained lower levels of BHBA, probably due to marginal ruminal fermentation of ingested forage, milk entering the rumen via backflow, incomplete esophageal groove closure, or lower tract fermentation of digesta (Quigley *et al.* 1991).

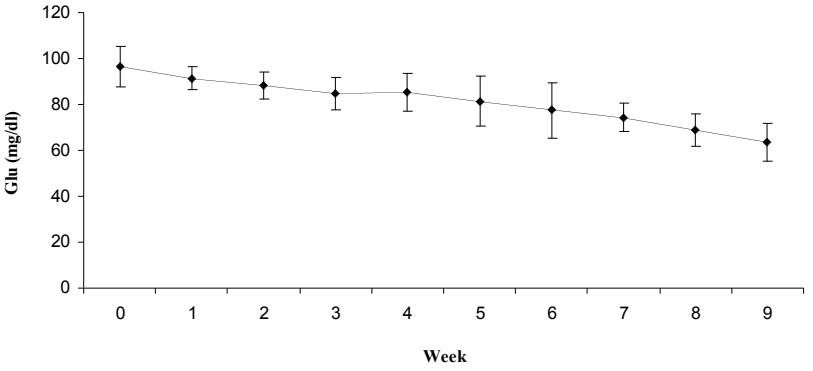

Figure 1
Changes in plasma glucose concentration of lambs over time

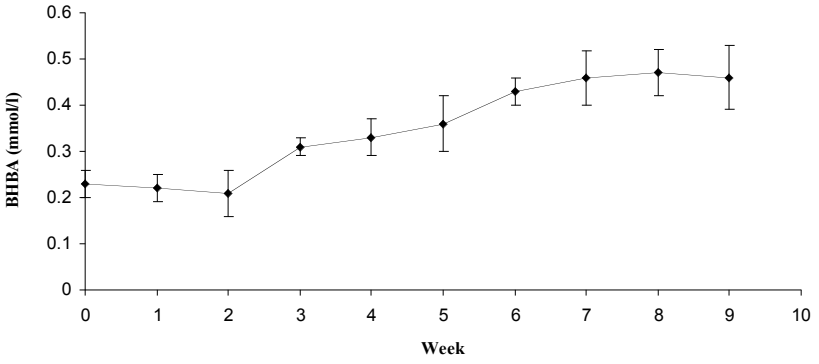

Figure 2
Changes in plasma BHBA concentration of lambs over time

Blood BHBA increased significantly with advancing the age of lambs (P=0.001) and increased markedly from wk 3 to 9 of age (Figure 2). Induction of ketogenesis by ruminal mucosa may result from exposure to dry feed consumption. This suggestion is in agreement with the findings of Greenwood *et al.* (1997) and Coverdale *et al.* (2004), who reported that metabolism of butyrate by ruminal wall of young ruminants increased with age advancement, and presumably, dry feed intake. Sutton *et al.* (1963) reported that metabolic activity of ruminal epithelium is induced by presence of ruminal VFA, especially butyrate. Early grain feeding and consequent VFA production possibly causes an increase in metabolic activity of ruminal epithelium, thereby, increasing production of a larger amount of BHBA per unit feed intake.

Molecular characteristics

Molecular characteristics of ruminal tissue samples are shown in Table 3. Neither DNA content (P=0.77) and nor cell size (P=0.87) were affected by rearing methods. Unlike the DNA content and cell size, RNA content and ribosomal capacity (Cs) of AR group significantly (P<0.05) higher than that of ER lambs.

Table 3
Effects of rearing system on molecular indices of ruminal tissue

Measurement	Group			
	ER	AR	SEM	*P*-value
RNA, µg/g	2 631.6[b]	3 359.9[a]	286.92	0.03
DNA, µg/g	156.40	132.60	39.87	0.77
Pr, mg/g	410.0	406.87	19.89	0.82
Cs, $\times 10^{-3}$	6.41[b]	8.25[a]	0.60	0.01
Cell Size, $\times 10^{3}$	2.92	3.07	0.65	0.87

ER: ewe reared lambs, AR: artificial reared lambs, Values in the same row with different superscripts are significantly different.

Although no data was found in molecular approaches of ruminal development studies but Estornell *et al.* (1994) indicated that addition of a supplementary energy source to a well-balanced diet improve growth and protein synthesis in growing rats. This change could be attributed to an increase in the ribosomal activity for protein synthesis in stratum basal layer of epithelium. It seems that high production in butyrate and propionate, increases energy source for epithelial cells and led to higher cell division, as Sakata & Tamate (1979) showed that butyric acid infused directly into the rumen of sheep resulted in a stimulation of mitotic indices. However Baldwin *et al.* (2004) demonstrated factors other than direct action by nutrients cannot be eliminated as possible agents controlling ruminal epithelial proliferation although clear primary candidates have not yet been delineated conclusively.

Ruminal morphologic characteristics

Rearing system did not affect morphologic characteristics of rumen wall except thickness of keratinized layer that was thickest, 20 % in AR than ER lambs (P<0.05) (Table 4). With agreement with our results, Roth *et al.* (2009) showed no differences in papilla length between calves weaned by the concentrate-dependent and conventional methods in any of the rumen areas. Although earlier studies showed that solid food intake was positively correlated with papillae growth (e.g., Tamate *et al.* 1962), we found any influence of starter feed consumption on rumen development in our data. This might be explained by the fact that all our animals received solid food of appropriate quality, which could have been of sufficient quantity to initiate rumen development. However, more rapid fermentation in AR lambs caused to thickest keratinized layer in these animals. In a study on male goats, Mgasa *et al.* (1994) verified that animals fed supplementary concentrate ration showed faulty keratinization (dyskeratosis) of the stratum corneum in most parts of the rumen. Cells of the stratum appeared rounded and vacuolated, and some without nucleus. Their work showed that goats on concentrates had much more pronounced development of rumen papillae and

keratinization of the stratum corneum compared to those on green forage, indicating that the structure of the forestomach is probably under the influence of the diet. In this experiment, differences observed in muscular layer and rumen wall thickness were not significant (P>0.05) among the treatments but animals fed starter diet exhibited thickest muscular layer and rumen wall. The papilla density and SAR were did not differ among groups (P>0.05).

Table 4
Effects of rearing system on morphological characteristics of rumen

Measurement	Group		SEM	Effect	
	ER	AR		Group	Sac
Papillae height, µm	1 398.59	1 539.43	66.44	0.37	<0.0001
Papillae width, µm	219.73	301.13	23.67	0.18	<0.0001
Epithelium, µm	62.09	63.16	5.52	0.20	0.01
Keratinized layer, µm	12.13[a]	8.95[b]	0.63	0.01	0.0006
Muscular layer, µm	757.15	1 034.60	94.73	0.16	<0.0001
Rumen Wall, µm	1 094.20	1 190.22	91.03	0.57	<0.0001
Papilla Density, No/cm^2	111.52	108.77	0.95	0.11	0.17
SAR, cm^2	1.153	1.159	0.002	0.17	<0.0001

ER: ewe reared lambs, AR: artificial reared lambs, Values in the same row with different superscripts are significantly different.

Table 5
Effects of rearing system on non-carcass organs weight and capacity

Measurement	Group		SEM	P-Value
	ER	AR		
Empty Body Weight, EBW, kg	15.725	15.675	0.61	0.39
Stomach Weight, SW, %EBW	3.14[b]	3.81[a]	0.12	0.05
Rumen Weight, %EBW	2.34	2.55	0.08	0.10
Rumen Weight, %SW	67.96	66.92	1.68	0.63
Omasum Weight, %EBW	0.56	0.58	0.05	0.67
Omasum Weight, %SW	16.15	15.21	1.34	0.79
Stomach Capacity, SC, g	3 352.5[b]	5 097.0[a]	106.98	0.0007
Rumen Capacity, %SC	89.5	92.21	0.72	0.07
Omasum Capacity, % SC	7.95[a]	4.7[b]	0.51	0.01
Heart Weight, %EBW	0.42	0.49	0.03	0.17
Liver Weight, %EBW	1.69	1.76	0.19	0.82
Lungs Weight, %EBW	0.89	1.15	0.14	0.29
Kidneys Weight, %EBW	0.38	0.42	0.03	0.39

ER: ewe reared lambs, AR: artificial reared lambs, Values in the same row with different superscripts are significantly different.

Table 5 shows the effect of rearing methods on non-carcass characteristics. Empty body weight (EBW) were similar for two groups of lambs but stomach weight (% EBW) and capacity in artificial reared animals were significantly (P=0.05) higher than ewe reared lambs. It has been suggested that ruminal and intestinal mass development is usually faster for lambs reared under natural conditions (De la Fuente *et al.* 1998). Our findings do not agree, since digestive tract weight was greater for artificially than for naturally reared lambs. However, feeding frequency or meal size could moderate gastrointestinal development (Kristensen *et al.* 2007), which could explain the differences between naturally and artificially reared lambs as average meal size would be

expected to be greater for the latter. Omasum capacity of ewe reared lambs was higher than other groups (P=0.01) probably because more intake of milk. The other non-carcass organs weight and capacity were not significantly (P>0.05) affected by rearing methods.

Post-weaning phase

Mean values of dry matter intake, daily body weight gain and feed conversion efficiency are shown in Table 6. Neither growth rate nor feed conversion efficiency were affected (P>0.05) by rearing method. Other authors reported similar growth rates for artificially or naturally reared lambs, although this depends on breed and feeding regime (Napolitano *et al.* 2002). There were no differences with respect to slaughter and dissection data (Table 7) between groups in post-weaning phase.

In conclusion, the results of this study show that naturally rearing systems gave rise to developmental and carcass characteristics similar to those observed in artificial raised lambs.

Table 6
Dry matter intake and body weight gain of the different group's lambs in post-weaning phase

Measurement	Group		SEM	Effect	
	ER	AR		Group	Time
Dry matter intake, g/day	1 528.7	1 540.6	26.0	0.87	<0.001
Body weight gain, g/day	227.60	252.81	19.4	0.49	<0.001
Feed conversion efficiency	7.50	7.45	0.67	0.92	<0.001

ER: ewe reared lambs, AR: artificial reared lambs, Values in the same row with different superscripts are significantly different.

Table 7
Slaughter and dissection data of the experimental lambs in post-weaning phase

Measurement	Group		SEM	*P*-Value
	ER	AR		
Slaughter weight, kg	44.43	43.10	1.29	0.48
Empty body weight (EBW), kg	39.55	38.31	1.15	0.45
Hot carcass weight, kg	21.98	21.40	0.87	0.67
Cold carcass weight (CCW), kg	21.60	21.09	0.86	0.70
Dressing percentage, %	54.60	55.02	0.70	0.15
Internal organs and body parts, % of EBW				
Heart	0.44	0.48	0.03	0.28
Liver	2.20	2.30	0.09	0.44
Lungs	1.35	1.28	0.15	0.81
Kidneys	0.34	0.33	0.03	0.87
Omental and mesenteric fat	2.09	2.24	0.26	0.23
Fat-tail	5.63	4.97	0.85	0.51
Head	5.41	5.17	0.15	0.17
Feet	2.86	2.71	0.12	0.21
Dissection data				
Loin, % of CCW	18.50	19.10	0.46	0.17
Pelvic Limb, % of CCW	31.40	31.70	0.75	0.66
Brisket, % of CCW	18.76	18.62	0.68	0.20
Neck, % of CCW	12.03	11.85	0.95	0.35
Shoulder, % of CCW	19.20	18.70	1.30	0.42

ER: ewe reared lambs, AR: artificial reared lambs, Values in the same row with different superscripts are significantly different.

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