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Effect of Weaning Age on Thermo-Hematological and Immunocompetence of Barki Lambs in Siwa Oasis, Egypt

M.S. Abdel-Fattah, Y.M. Shaker, A.L.S. Hashem, Ashgan M. Ellamei and Hanan Z. Amer

Department of Animal Physiology, Animal and Poultry Production Division,
Desert Research Center, Cairo, Egypt

Abstract: This study was conducted to examine a system of early weaning (at 60days of age) vs. the common late weaning (at 120 days of age) of Barki lambs. The study lasted 8 months period at Tegzerty Research Station in Siwa Oasis, Egypt belonging to Desert Research Center, Egypt. The study was performed using 70 newborn Barki lambs (28 males and 42 females) with average birth weight recorded (4.12±0.14 and 3.06±0.07 kg vs. 4.11±0.10 and 3.85±0.05 kg) for male and females under early weaning (EW) and late weaning (LW) age, respectively. All lambs were housed with their dams till weaning age (at 60 days or at 120 days). Monthly live body weight (LBW) and different thermoregulatory parameters (rectal temperature, RT; skin temperature, ST; respiration rate, RR and heart rate, HR) were recorded. Also, hematological parameters (erythrocyte count, RBC's; hemoglobin concentration, Hb and packed cell volume, PCV, total leukocytes count, WBC's and their differentiation) were determined. In addition to erythrocyte indices (mean corpuscular volume, MCV, fl; mean corpuscular hemoglobin, MCH, pg and mean corpuscular hemoglobin concentration, MCHC,%) were calculated. A comparative growth trial was conducted for 4 months period from the 5th month till the 8th month of study. The study was performed to evaluate the effect of weaning age and sex on weight gain, thermoregulatory, hematological responses and immunocompetence in Barki lambs. Concerning the effect of weaning age, the results indicated that, early weaned lambs tended to have greater ($P<0.01$) ADG than late-weaned lambs. On the other hand results indicated that, LBW of males tended to be greater ($P<0.001$) than females in both early and late-weaned groups throughout the experimental period. Weaning age had no significant effects on thermoregulatory traits (RT, ST, RR and HR) which more associated with the change in climatic conditions throughout the experimental period. However, under both early and late weaning ages, significant ($P<0.01$) differences between male and female lambs were observed in RT and ST. Hematological parameters (RBC's, Hb, PCV, MCV, MCH and MCHC) were declined significantly ($P<0.01$) with the advancement of animal age. Female lambs had slightly higher ($P<0.01$) values in RBC's, Hb and PCV than males under both the two weaning ages. In respect to immunocompetence, there was no explicit benefit to the adaptive immune system for lambs under two weaning ages. Early weaning did not negatively affect the adaptive immunological competence of lambs as determined by changes in populations of immune cells. On the 1st month after weaning, total WBC's, neutrophil and lymphocyte proportions were increased ($P<0.001$) in EW and LW lambs. Male lambs in both EW and LW age had higher values in total WBC's count and greater ($P<0.001$) neutrophil and lymphocyte concentrations compared with females under two weaning ages.

Key words: Sheep · Weaning age · Weight gain · Thermoregulation · Hematology · Immunity

INTRODUCTION

The most important alteration for the kid after birth is the adaptation to the new physical environment and free-living compulsion from the mother in the neonatal period, the adaptation and development of the organs to the new environment has a vital importance for the continuity of their life [1]. Postnatal maturation for the ventilatory response to O_2 occurs within the first 10 days of life in newborn lambs and is largely due to an increase in sensitivity of the O_2 chemoreceptors and during this period that thermoregulatory, cardiovascular, respiratory

Corresponding Author: M.S. Abdel-Fattah, Department of Animal Physiology, Animal and Poultry Production Division, Desert Research Center, Cairo, Egypt.

and homeostatic mechanisms complete their maturation [2]. Rectal temperature is generally used as a measurement of animal core temperature [3]. Hermoregulatory process is also influenced by birth weight and some reports suggest that lighter lambs at birth have reduced capability to sustain body temperature [4, 5]. Homeothermic animals such as goats, maintain their body heat balance by increasing respiration rate or panting to balance their body core temperature [1]. Heart rate reflects primarily the homeostasis of circulation among with the general metabolic status, that immediately after birth, heart must pump blood at a higher systolic rate into the vascular system that shows high elastic and peripheral resistances which cause a further increase of heart rate. Cardio-respiratory system undergoes prominent changes in neonatal period [6] and it can be influenced by season, day timings, ambient temperature, humidity and exercise [7]. Hematological parameters also pass through a series of changes and are helpful to determine the health and nutritional status of animals [8]. During weaning time in calves, total leukocytes count increased significantly [9]. Early weaning is a relative term, but implies weaning at any time after 14 days of age, but usually before 90 days. Sixty (60) days is a common weaning age in intensively managed sheep operations. Early weaning can be successful to provide lambs drinking water and consuming adequate amounts of dry feed. It allows ewes to return to breeding condition earlier, which is essential for accelerated lambing programs.

The objective of this study was to evaluate the effect of weaning age (60 vs.120 days) and sex on the thermoregulatory, hematological and immunocompetence responses in Barki lambs during pre and post-weaning periods in Siwa Oasis, Egypt.

MATERIALS AND METHODS

Site of the Study: The present study was conducted from March to October 2011 for 8 months (240 days) period in Siwa Research Station (Tegzerty Experimental Farm for Animal Production), which belongs to Desert Research Center, Egypt. It lies at 300 Km southwest of the Mediterranean shoreline and at 60 Km east of the Libyan borders. The ground surface height ranges from 10 to 20 meters below mean Mediterranean Sea level. Siwa Oasis is characterized by desert climate.

Meteorological Parameters: Meteorological data including ambient temperature (AT, °C) and relative humidity (RH, %) were recorded using Hygro-thermometer during measurement of the thermo-regulatory traits. Data were recorded once weekly at 07:00 am. A temperature-humidity index (THI) is a single value representing the combined effects of air temperature and humidity. The mean temperature-humidity index (THI) was calculated using the following equation:

$$THI = 0.8 \times AT°C + (RH, \%) \times (AT°C - 14.4)/100) + 46.4 \ [10].$$

Mean ± SE of monthly ambient temperature (AT, °C), relative humidity (RH, %) and temperature-humidity index (THI) throughout the experimental period are presented in Table 1. THI is commonly used as an indicator of the degree of climatic stress on animals where a THI of 72 and below is considered as no heat stress, 73-77 as mild heat stress, 78-79 as moderate and >80 as severe [11]. Accordingly, animals were under a mild heat stress during the 2^{nd} and 3^{rd} month (April and May) while were under severe heat stress from the 4^{th} till the 6^{th} month (from June to August) and returned to moderate stress during the 7^{th} and 8^{th} months (September and October) of the experimental period.

Estrus Synchronization of Ewes: Breeding ewes were oestrus synchronized perior to mating using two doses (10 days apart) of 1ml intra-muscular Estrumate (cloprostenol; prostaglandin synthetic analogue, PGF2α, Imperial chemical Industries limited, UK); Each ml of this analogue contains 263 µg Cloprostenol Sodium BP (vet) equivalent to 250µg Cloprostenol. Mating was carried out naturally using highly fertile Barki rams.

Live Body Weight (LBW, kg): Seventy (70) lambs were used from birth till 240 days of age. The birth weight was taken within 12 hr after birth and when lambs were dry. Thereafter, all lambs were weighed monthly up to 8 months (240 days) of age at 08:00 am by using an aviary weighing–machine to the nearest 100 grams. The average daily weight gain (ADG) in grams per day was calculated.

Experimental Design and Animal Management: Seventy lambs (28 rams and 42 ewes) and their dams were divided into two groups/sex (14 ram vs. 21 ewe lambs) in 4 pens and fed on dam's milk till reaching to weaning age (60 vs.120 days). At weaning age, dams were separated from their lambs without any contact between them. Weanling rams were separated from weanling ewes to prevent ewe lambs from breeding too early. Fattening period for 4 months was performed from the 5^{th} month till the 8^{th} month of age. Weaned lambs were group-fed on

Table 1: Mean ± SE of monthly climatic data (at 07:00 am) throughout the study

Month	AT (°C)	RH (%)	THI
March	24.65±0.55	46.23±1.30	70.86±0.93
April	27.92±0.60	41.66±1.30	74.40±0.95
May	29.70±0.50	40.88±1.40	76.41±0.95
June	34.88±0.42	35.66±1.30	81.61±0.86
July	35.32±0.50	35.22±1.50	82.02±1.00
August	36.53±0.40	35.64±1.20	83.51±0.80
September	32.41±0.60	40.33±1.00	79.59±0.80
October	32.88±0.50	39.66±0.83	80.03±0.66

AT= ambient temperature, RH= relative humidity, THI= temperature humidity index

Table 2: The chemical composition on DM basis (%) of CFM and Alfalfa hay

Item	DM%	OM	CP	CF	EE	NFE	Ash
CFM	89.25	91.52	16.31	11.56	3.42	60.23	8.48
Alfalfa hay	90.42	87.36	12.73	29.46	1.39	43.78	12.64

CFM= concentrate feed mixture, DM= dry matter; OM= organic matter; CP= crude protein;
CF= crude fiber; EE= ether extract; NFE= nitrogen free extract

ground concentrate mixture and chopped alfalfa hay during the first month following weaning process, then complete pelleted concentrate and alfalfa hay were used up to 240 days of age. Supplementation with concentrate was at 2.5% of body weight per day. Average daily supplement were adjusted according to monthly body weight changes of the ram and ewe lambs according to Kearl [12]. Drinking water was available *ad lib* to all groups twice daily in the morning (at 10:00 hr) and at the evening (at 16:00 hr). A sample of the concentrate ration and the alfalfa hay were analyzed according to A.O.A.C [13]. The chemical composition of commercial concentrate feed mixture and alfalfa hay used during post-weaning period is presented in Table 2.

Thermoregulatory Parameters: Rectal temperature (RT, °C) was measured by using a clinical thermometer which was inserted about 6-7 cm, into the animal rectum for one minute. Skin temperature (ST,°C) was measured to the nearest of 0.1°C using a YSI 408 Banjo surface probe from the middle to side position of the animal and read out with the YSI 46 Tele-thermometer. Respiration rate (RR, r.p.m) was measured in respirations per minute, by counting flank movements per minute. Heart rate (HR, b.p.m) was measured in beats per minute by using a clinical stethoscop from the jointing point of left-front leg and body. All thermoregulatory parameters were measured at 7:00 am monthly.

Blood Sampling: Monthly, just before offering feed and water approximately 10 ml of blood was taken from jugular vein of each animal in test tubes containing Lithium heparin as anticoagulant till 8 months of age. Hemoglobin concentration (Hb, g/dl) was estimated in blood according to Drabkin and Austin [14]. The packed cells volume (PCV %) was estimated by the use of the microhematocrit method according to Cheryl *et al.* [15]. The blood samples were packed in heparinized capillary tubes sealed at one end and then the tubes were centrifuged in a microhematocrit centrifuge at 12.000 revolutions per minute for 5 minutes and read on a micro capillary tube reader, this packed cell volumes (PCV %) was expressed on percentage of RBC's per volume of blood. Erythrocytes count (RBC's x 10^6 cells/µl) was made by diluting whole blood, 1:200 in physiological saline solution (0.9% NaCl) and employing an improved neubauer counting chamber certified cover glass (Thom's hemocytometer) cited by Cheryl *et al.* [15].

Erythrocyte Indices: Red blood cells are responsible for gas exchange, carrying oxygen and carbon dioxide in their heme structures. The mean corpuscular volume (MCV, fl), mean corpuscular hemoglobin (MCH,pg) and mean corpuscular hemoglobin concentration (MCHC,%) are characteristics of the RBC's indicating average cell size, average cell hemoglobin content and average cell hemoglobin concentration, respectively and they were calculated.

Total Leukocytes Count and Differentiation: In the 1st, 3rd, 5th and 8th months, part of the whole blood samples was intended for total leukocytes (WBC's x 10^3 cells/µl) count in blood diluted 1:50 in Turk's solution by means of a Neubauer's hemocytometer cited by Cheryl *et al.* [15]. Blood smears were prepared on microscope slides and fixed with methyl alcohol, then stained with Wright's stain. The differential distribution of leukocytes was determined by counting 100 cells per slide using the method described by Cheryl *et al.* [15] and expressed as a percentage.

Statistical Analysis: Statistical analysis was conducted using the general linear model (GLM) procedures of SAS [16]. A repeated measurement model was used. Distributed Duncan's tests [17] were used to compare the treatment means.

RESULTS AND DISCUSSION

Growth Performance: Average body weight ± SE of male and female lambs at birth, 4 and 8 months of age (fattening period) and average daily gain (ADG) under early (60 days) or late (120 days) weaning age are presented in Table 3. The results indicated that, lamb sex had highly significant effect (P<0.01) on birth weight. Babar *et al.* [18] stated that male lambs generally stay slightly longer in mother's womb than females and hence heavier at birth. Regarding the effect of weaning age, the results in Table 3 showed that early and late-weaned lambs had similar LBW at the age of 4 months (at the beginning of fattening period) in males (21.28±0.11 and 21.43±0.24 kg for early and late weaning, respectively), while female lambs were the heavier by about 5.12 % in late weaning compared with early weaning (18.94±0.07 and 19.91±0.07 kg for EW and LW, respectively). However, early weaned lambs (males and females) had the higher (P<0.001) final LBW and ADG compared with late weaned lambs (36.95±0.28 and 33.56±0.18 vs. 34.31±0.19 and 32.58±0.09 kg, for LBW) and (130.58 and 121.83 vs. 107.33 and 105.58 kg, for ADG). Abbas *et al.* [19] explained the superiority of early weaned lambs in body weight post-weaning could be due to differences in rumen development or rumen capacity and increasing solid feed consumption compared to normal or late weaned lambs.

Similar results were obtained by Schichowski *et al.* [20] reported that lambs weaned at 8 weeks of age had greater ADG compared with lambs weaned at 16 weeks of age. Also, Alhadrami *et al.* [21] and Abou Ward *et al.* [22] indicated that, age at weaning affected (P<0.009) average daily gain and feed conversion ratio in favor of the early weaned group lambs (8 weeks) compared with the late-weaned lambs (12 weeks). Previous studies were obtained by Khoury *et al.* [23] who found that on early weaned calves, especially female buffalo calves showed lower gains at weaning and at subsequent age up to six months as compared to late weaned ones. Caneque *et al.* [24] reported that, the effect of weaning age was less marked in lambs separated from their dams at 65 days of age. While, Samy [25] found that, similar final LBW for both the two ages weaning groups (8 or 12 weeks). In contrast, Bonsma and Engela [26] found that lambs weaned at 8 weeks of age had the same growth as unweaned controls. Also, Bosman and Bonsma [27] confirmed this result on lambs.

With respect to the effect of sex, the results in Table 3 indicated that, in both early and late-weaned lambs, males were more efficient in ADG than females. Saddiqi *et al.* [28] reported differences between male and female lambs in birth weight; males were slightly heavier (0.46 kg) than females. The difference in ADG between male and females may be due to the high birth and weaning weights for male lambs and subsequently the anabolic role of testosterone in male which had potential's to improve feed intake, feed conversion and stimulate growth compared with female lambs. Similar results were reported by Suliman [29] who indicated that male lambs were heavier than females at different ages from 4 to 12 months of age. Fahmy *et al.* [30] found that sex effect on body weight increased with advance in age due to the increased secretion of sex hormones with advance of age. Abbas *et al.* [19] reported that male lambs had significantly (P<0.01) higher average daily gain than females from 6 to 9 months and significant (P<0.05) from birth to 12 months of age. Previous study performed by Ramzan *et al.* [31] compared between 12 Beetal and Barbari kids (6 males and 6 females) to 120 days of fattening period and found that, male were heavier than femal kids by 10.8% in the Beetal and 11.4% in the Barbari breed.

Concerning the effect of age on growth performance, the results in Table 3 indicated that LBW (kg) and ADG (g/day) increased significantly (P<0.001) on early and late-weaned lambs by the advancement of age but early-weaned lambs recorded the higher (P<0.001) values. In accordance, Saddiqi *et al.* [28] reported that, linear regression model between live weight and post-natal day's shows a steady increase in live weight with the increase of animal's age. Similar results were obtained by Abbas *et al.* [19] and Fahmy *et al.* [30], who found that sex effect on body weight increased with advance in age. In contrast, the response to stress separation by both cows and calves increased when calves were separated at later rather than earlier ages; also, calves separated at the later age gained more live weight [32, 33]. It was explained that weaning age is an important factor which might affect the growth performance of lambs, while the animal sex

Table 3: Means ± SE of growth performance in Barki lambs under early (EW) and late (LW) weaning age in Siwa Oasis

	Weaning age			
	EW		LW	
Item	Males	Females	Males	Females
No. of animals	14	21	14	21
Birth Weight (BW,kg)	4.12 ± 0.14^{a}	3.06 ± 0.07^{c}	4.11 ± 0.10^{a}	3.85 ± 0.05^{b}
Fattening period:				
Initial weight, kg	21.28 ± 0.11^{b}	18.94 ± 0.07^{d}	21.43 ± 0.24^{a}	19.91 ± 0.07^{c}
Average, kg	20.11 ± 0.09^{b}		20.67 ± 0.09^{a}	
Final weight, kg	36.95 ± 0.28^{a}	33.56 ± 0.18^{c}	34.31 ± 0.19^{b}	32.58 ± 0.09^{d}
Average, kg	35.25 ± 0.09^{a}		33.44 ± 0.09^{b}	
ADG (g/d)	130.58	121.83	107.33	105.58

EW=early weaning (at 60 days of age); LW = late weaning (at 120 days of age); ADG = average daily gain; ^{a}=P<0.05

Table 4: Means ± SE of rectal temperature (°C), skin temperature (°C), respiration rate (rpm) and heart rate (bpm) for Barki lambs under early (EW) and late (LW) weaning age in Siwa Oasis

Treatment		Early Weaning (EW)				Late Weaning (LW)			
Age	Sex	RT	ST	RR	HR	RT	ST	RR	HR
1 Month	M	39.49^{a}	34.50^{b}	70.64^{a}	94.64^{b}	39.44^{a}	34.60^{b}	70.71^{b}	91.33^{a}
	F	39.36^{b}	34.50^{b}	67.41^{b}	95.33^{b}	39.26^{b}	34.90^{b}	70.71^{b}	89.55^{b}
2 Month	M	39.54^{a}	34.53^{b}	72.22^{a}	92.64^{a}	39.61^{a}	34.48^{a}	69.79^{b}	88.29^{a}
	F	39.46^{a}	34.58^{b}	68.44^{b}	90.10^{b}	39.45^{b}	34.2^{b}	68.67^{b}	85.44^{b}
3 Months	M	39.19^{*}	34.33^{b}	69.44^{*}	92.10^{*}	39.53^{a}	34.53^{a}	68.07^{b}	90.79^{a}
	F	39.09^{*}	34.32^{b}	65.22^{*}	90.24^{*}	39.30^{b}	34.51^{b}	68.67^{b}	86.44^{b}
4 Months	M	39.51^{a}	35.55^{a}	74.04^{a}	96.89^{a}	39.69^{a}	34.47^{b}	72.86^{a}	90.79^{a}
	F	39.39^{a}	35.39^{a}	75.05^{a}	94.29^{a}	39.56^{a}	34.49^{b}	69.62^{a}	88.44^{a}
5 Months	M	39.74^{a}	35.56^{a}	75.64^{a}	95.64^{a}	39.32^{*}	34.34^{b}	73.55^{*}	92.11^{*}
	F	39.64^{a}	35.60^{a}	73.80^{a}	91.71^{a}	39.13^{*}	34.15^{b}	72.30^{*}	91.44^{*}
6 Months	M	39.83^{a}	35.63^{a}	78.50^{a}	97.21^{a}	39.71^{a}	35.62^{a}	82.33^{a}	96.33^{a}
	F	39.84^{a}	35.64^{a}	77.40^{a}	98.71^{a}	39.56^{a}	35.34^{a}	77.22^{a}	93.55^{a}
7 Months	M	39.56^{a}	35.45^{a}	68.86^{a}	80.64^{a}	39.60^{a}	35.32^{a}	73.00^{a}	81.22^{a}
	F	39.48^{a}	35.40^{a}	66.38^{a}	73.24^{a}	39.37^{a}	34.53^{a}	68.66^{a}	78.29^{a}
8 Months	M	39.46^{a}	35.51^{a}	65.00^{a}	76.43^{a}	39.58^{a}	35.13^{a}	68.55^{a}	80.33^{a}
	F	39.23^{a}	34.48^{a}	62.57^{a}	71.10^{a}	39.36^{a}	34.41^{a}	65.66^{a}	79.05^{a}
±SE	M	0.01	0.03	0.40	1.11	0.01	0.03	0.40	1.11
	F	0.01	0.02	0.32	0.91	0.01	0.02	0.32	0.91
Sex mean	M	39.54^{*}	35.22^{*}	71.79^{*}	90.77^{*}	39.56^{*}	34.81^{*}	72.36^{*}	88.9^{*}
	F	39.44	35.03	69.53	88.09	39.37	34.57	70.19	86.53
Overall mean		39.49^{NS}	35.12^{NS}	70.66^{NS}	89.43^{NS}	39.46	34.69	71.27	87.71

M= males; F= females; SE= standard error; ^{a} = P<0.05; *= P<0.01; ^{NS}= Non-significant; RT = rectal temperature;
ST = skin temperature; RR = respiration rate; HR = heart rate; rpm = respiration per minute; bpm= beats per minute

seems to be of secondary importance. Therefore, this study may be suggested that weaning lambs as early as (60 days of age) led to develop the capability of such lambs at an earlier age. Caneque *et al.* [24], Doney and Peart [34] and Cardellino and Benson [35] reported that from day 75, milk production of ewes begins to drop and the rate of growth in weaned lambs decreases somewhat, as solid feed intake.

Thermoregulatory Response

Rectal (RT, °C) and Skin (ST, °C) Temperatures: Mean values ± SE of monthly rectal and skin temperatures of both male and female lambs under early and late weaning age are presented in Table 4 throughout the experimental period. The results in Table 4 revealed that there was significant (P<0.01) fall of RT and ST in all lambs at the 3rd and 5th month for early and late weaning age, respectively.

For early-weaned lambs, the rate of change recorded (-0.76 and –0.49 % vs. –0.68 and –0.52 %) for male and females, respectively. The corresponding values for late-weaned lambs were (-0.30 and –0.75 % vs.-0.33 and -2.15 %) for male and females, respectively. This decline in RT and ST may be referred to the negative effect of weaning stress on appetite of animals which may cause a decrease in DMI and subsequently heat production. In contract, Lynch *et al.* [36] reported that RT increased (P<0.05) in abrupt weaned beef calves on d_3, d_7 and d_{14} compared with pre-weaning baseline. Also, Duff and Galyean [37] found that small increase in RT of calves at post-weaning and this increase was not of clinical significance. Brody [38] stated that the change in heat production with age is related to many factors, including puberty, weaning, changes in growth rate and stabilization of the neuro-endocrine homeothermic system. Concerning the effect of weaning age, statistical analysis indicated that weaning age had no significant effect (P>0.05) on RT and ST.

Concerning the effect of sex, the results in Table 4 indicated that males were had the higher values (P<0.05) in the mean of RT and ST compared with the values of female lambs under both early and late weaning age. For early-weaned lambs, the values were (39.54 and 35.13°C vs. 39.46 and 34.86°C) for RT and ST, respectively. The differences between sexes were (0.08 and 0.27 °C) higher for RT and ST, respectively. The corresponding values for late-weaned lambs were (39.6 and 34.92 °C vs. 39.37 and 34.56 °C) for RT and ST, respectively. The differences between sexes were (0.18 and 0.36 °C) higher for RT and ST, respectively. Sahoo and Mishra [39] stated that, by their nature, males are heavier than females, in addition males have greater muscle mass and higher feeding level would produce more heat than females. Kasa *et al.* [40] reported that there were sex differences in RT and ST with 0.2 °C for Saanen goat which males had the highest values compared with females and therefore males were less tolerant than females. Also, Ocak *et al.* [1] reported significant differences between male and female kids in RT while no correlation between growth and thermo-physiological parameters.

The measured climatic data (Table 1) shows that lambs were under severe heat stress during the 4^{th}, 5^{th} and 6^{th} month of age (June, July and August months, respectively), as indicated by temperature humidity index (THI). This was reflected on significant (P<0.01) higher RT and ST in all lambs (Table 4) under early and late weaning age during the 4^{th}, 5^{th} and 6^{th} month of age, when heat dissipation was lower than heat gain and therefore, thermal balance could not be maintained. These results are in agreement with those obtained by Abdoun *et al.* [41] in camels and sheep.

On the other hand, the mammalian skin is an important pathway for heat exchange between the body surface and the environment. Therefore the observed elevation in ST of male and femal lambs during these months can be attributed to the elevation in climatic conditions. Marai and Habeeb [42] reported that the elevation of ST in Buffalo's can be attributed to the exposure to heat stress which alters the blood flow and redistributes it to the surface. Also, McManus *et al.* [43] reported that the increase in ST in Brazilian sheep under hot summer conditions is attributed to the exposure to heat stress, which has been reported to cause vasodilation of skin capillary bed and consequently increase the blood flow to the skin surface to facilitate heat dissipation. In accordance, Marai *et al.* [44] reported an increase in RT and ST in Egyptian Suffolk rams with the increase in ambient temperature. The results showed that RT and ST of early-weaned lambs (both male and females) had two highest values in the 5^{th} month (39.74 and 35.56°C vs. 39.64 and 35.6°C for males and females, respectively) and in the 6^{th} month (39.83 and 35.63°C vs. 39.84 and 35.64°C for males and females, respectively), while late-weaned lambs reached highest value in the 6^{th} month only with averages (39.71 and 35.62°C vs. 39.56 and 35.34°C for males and females, respectively). Similar results obtained by Abdoun *et al.* [41] who found that RT and ST was significantly (P<0.05) increased in sheep and camels during summer season. Alamer and Al-Hozab [45] reported that the increase of the body core temperature and rectal temperature has been considered as good indicators to the level of heat stress upon animals.

Cardio-Respiratory Response: Mean values ± SE of monthly respiration and heart rates of both male and female lambs under early and late weaning age are presented in Table 5. Values for RR and HR in the present study fall within the range of recently published data [46], who determined HR in sheep using a stethoscope. Concerning the effect of weaning age, statistical analysis indicated that weaning age had no significant effect (P>0.05) on RR and HR. The overall mean of RR and HR recorded 70.66 and 89.43 vs. 71.27rpm and 877.71bpm for EW and LW lambs, respectively. Concerning the effect of sex, the results in Table 4 indicated that males had the higher values (P<0.05) in the mean of RR and HR compared with the values of female lambs under both early and late weaning age. For early-weaned lambs, the

Table 5: Means ± SE of blood erythrocytes count (x10^{6}cells/µl), hemoglobin (g/dl) and packed cell volume (%) in early (EW) and late (LW) weaned Barki lambs in Siwa Oasis

Treatment		Early Weaning (EW)			Late Weaning (LW)		
Age	Sex	RBC's	Hb	PCV	RBC's	Hb	PCV
1 Month	M	10.35^b	10.37^b	28.66^b	10.47^a	10.57^a	29.19^a
	F	10.59^a	10.75^a	29.68^a	10.35^b	10.40^b	28.34^b
2 Month	M	11.50^a	11.45^a	30.96^a	11.45^a	11.33^b	30.38^b
	F	11.26^b	11.23^b	30.34^b	11.30^b	11.43^b	30.29^b
3 Months	M	10.14**	9.98**	31.66**	11.37^b	11.30^b	30.77^b
	F	10.41**	10.30**	31.98**	11.50^a	11.70^a	31.56^a
4 Months	M	10.35^b	10.29^b	28.34^b	11.21^b	11.36^b	30.94^b
	F	10.86^a	10.52^a	29.95^a	11.68^a	11.83^a	31.22^a
5 Months	M	10.44^b	10.37^a	28.88^b	9.90**	9.75**	31.99**
	F	10.29^b	10.29^b	28.63^b	10.34**	10.31**	32.33**
6 Months	M	10.16^a	10.21^a	28.67^a	9.71^b	9.64^a	25.76^a
	F	10.28^a	9.89^a	27.46^a	10.05^a	9.69^a	27.65^a
7 Months	M	9.93^a	9.75^a	27.12^a	9.81^a	9.76^a	26.62^a
	F	9.48^a	9.71^a	26.91^a	9.48^a	9.45^a	26.18^a
8 Months	M	9.79^a	9.32^a	26.76^a	9.84^a	9.37^a	26.68^a
	F	9.87^a	9.39^a	27.12^a	9.40^a	9.36^a	26.28^a
±SE	M	0.05	0.05	0.13	0.05	0.05	0.13
	F	0.04	0.04	0.11	0.04	0.04	0.11
Sex mean	M	10.33^b	10.22^b	28.88^b	10.47^b	10.40^b	29.04^b
	F	10.38^a	10.26^a	29.01^a	10.52^a	10.52^a	29.23^a
Overall mean		10.35	10.24	28.94	10.49*	10.46*	29.13*

M= males; F= females; SE= standard error; ^{a}= P<0.05; *= P<0.01; ** = P<0.001; ^{NS}= non-significant

RBC's = red blood cells; Hb = hemoglobin; PCV = packed cell volume

values were (71.97 and 90.77 vs. 69.53 rpm and 88.09 bpm) for RR and HR, respectively. The differences between sexes were (3.51 and 3.04 %) higher for RR and HR, respectively. The corresponding values for late-weaned lambs were (72.36 and 88.9 vs. 70.19 rpm and 86.53 bpm) for RT and ST, respectively. The differences between sexes were (3.10 and 2.74 %) higher for RT and ST, respectively.

The measured climatic data (Table 1) shows that lambs were under severe heat stress during the 4th, 5th and 6th month of age (June, July and August months, respectively), which was accompanied with higher RR and HR (P<0.01) in all lambs (Table 4) under early and late weaning age, where heat dissipation was lower than heat gain and therefore, thermal balance could not be maintained. Alamer and Al-Hozab [45] stated that RR can be used as an indicator of heat stress and to estimate the adverse effects of environmental temperature. Moreover, Silanikove [47] suggested that RR was a practical and reliable measure of heat load. Therefore, the observed elevation in both RR and HR in both EW and LW lambs during the 4th, 5th and 6th months of age, indicate that lambs were exposed to warm climatic conditions. Finally, thermoregulatory parameters dynamics were related with the climatic conditions which represent the main factor depending on their dynamics and not associated with weaning age.

Hematological Responses: Blood picture of animal might be influenced by certain factors such as nutrition, management, breed of animal, sex, age, diseases and stress factors [48]. Mean values ± SE of erythrocytes count (RBC's X 10^6 cells/µl), hemoglobin (Hb, g/dl) and packed cell volume (PCV, %) in early and late-weaned lambs are presented in Table 5.

Our values for hematological parameters fell within the range of recently published by Jones and Allison [49] who reported that for hematological parameters in sheep are: RBC's 9-15 x 10^{6}cells/µl; PCV, 27-45%; Hb, 9-15g/dl; MCV, 28-40fL; MCH, 8-12pg and MCHC, 31-34%. As shown in Table (5) regardless the effect of sex, no differences in values for RBC's, Hb and PCV in both EW and LW groups during the 1st and 2nd month of experimental period. At the 3rd month with EW lambs values for RBC's count and HB concentration decreased in all lambs compared with LW lambs. Dealing with the effect of weaning stress, as shown in Table (5) in both early (in the 3rd month) and late-weaned (in the 5th month) lambs, weaning stress resulted a significant ($P<0.001$) decrease in both sexes for RBC's count and Hb concentration but a slight increase (($P<0.001$) in PCV percentage. For early-weaned lambs, the percentage of change in RBC's and Hb and PCV were – 2.03, -3.76 and 10.50 % vs. -1.70, - 4.20 and 11.58 % for male and females, respectively. The corresponding values for late-weaned lambs were – 5.44, - 7.76 and 9.59 % vs. – 0.97, - 0.86 and 14.08 % for male and females, respectively. In contrast, blood hemoglobin concentration increased in EW but not in control cows, with significant differences apparent after day 30 of weaning process [50]. The results indicated that, Hb concentration and PCV percentage were related to the changes observed in RBC's count for all lambs throughout the experimental period. Similar results were reported by Ullrey *et al.* [51]. With respect to the effect of sex, animal sex is also believed to have a significant effect on RBC's, Hb and PCV values, the results in Table 5 indicated that the sex mean of RBC's, Hb and PCV was low ($P<0.05$) in male animals than females. This observation is in contrast with values obtained for Red Sokoto goats in Nigeria [52] in which male animals have higher values than females. In accordance, Tibbo *et al.* [53] found that female goats had significantly ($P<0.5$) higher RBC's count, Ht, WBC's count, lymphocytes and eosinophils than male goat. Similar results were obtained by Saddiqi *et al.* [28] on Kajli lambs. Opara *et al.* [54] who reported that, there was significantly ($p<0.05$) higher percentage of PCV, Hb and RBC in female West African Dwarf (WAD) goats than the males.

Concerning the effect of age, from values presented in Table 5, it is noticed that, with the increase of age there is a tendency to decrease RBC's, Hb and PCV. The rate of change for early-weaned lambs were (-5.41, -10.12 and -6.63 % vs.-4.64,-9.45 and -5.37 %) in male and females, respectively. The corresponding values for late-weaned lambs recorded (-6.02, -1.35 and -8.60 % vs. -9.20, - 10.00 and -7.27 %) in male and females, respectively. Previously, Tambuwal *et al.* [52] reported that values of Hb concentration in Red Sokoto goats are adjusted according to oxygen carrying capacity of the blood and hence its level can vary according to different age groups. Similar results were obtained by Opara *et al.* [54] who reported that age of West African Dwarf (WAD) goats was observed to have a significant effect on parameters like Hb, RBC and MCHC values. Also, age had a significant effect on the hematological parameters in various animal species [48].

Erythrocyte Indices Responses: Table 6 shows the mean values ±SE of erythrocyte elements (MCV, MCH and MCHC) in early (EW) and late (LW) weaned lambs throughout the experimental period. The results indicated that MCV increased significantly ($P<0.01$) only on the first month after weaning process (at the 3rd and 5th month of age for EW and LW lambs, respectively), the rate of increase in MCV recorded 12.75 and 9.60 % vs. 15.80 and 12.93 % for male and femal lambs under early and late weaning ages, respectively. This increase in MCV explains the pronounced decrease on MCH and MCHC. The increase in MCV led to a big red cell size and evidenced the decrease in number of RBC's of lambs during the first month after weaning process (Table 5). Holman and Dew [55] reported that there were inverse relationship between MCV and erythrocyte count (RBC's) values in goat. The highest increase in MCV after one month of weaning process for LW lambs compared with EW lambs, may be attributable to variations in climatic changes (Table 1), where increasing AT and THI which reached to 35.32°C and 82.02, respectively may increase water intake and reduce feed intake to minimize the rise in body temperature and subsequently more absorption of water into the erythrocyte occurred (increase size of the erythrocyte) which led to the decrease in both MCH and MCHC. Schneider *et al.* [56] observed that heat stressed dairy cows consumed less feed (13.6 vs.19.4 kg/day) and more water intakes (86.0 vs. 81.9 l/day) than cows in a thermal neutral environment. It would, thus, be logical to suggest that weaned lambs rapidly learned to drink water after weaning. These results are in agreement with those reported by Jain [57] and Lane and Albrecht [58]. Previous study conducted on calves by Lynch *et al.* [59] found that RBC's number and PCV percentage decreased ($P<0.05$) by 6% on d_7 to d_{21} and by 3% on d_{14} and d_{21}, respectively, compared with pre-weaning baseline, while there was no change in Hb concentration post-weaning. It is apparent (Table 6); the overall means of MCV, MCH and MCHC were similar. Statistical analysis indicated that weaning age had no significant effect ($P>0.05$) on erythrocyte elements.

With respect to the effect of sex, the observed sex means (Table 6) of MCV, MCH and MCHC indicated that there were no significant differences between male and female lambs under EW and LW age in these parameters.

With respect to the effect of age, the results indicated that mean values of MCH and MCHC decreased with the advancement of age but this reduction was relatively more for MCHC. Similar results were obtained by Saddiqi *et al.* [28] in newly born Kajli lambs. Also, Opara *et al.* [54] found that age was observed to have a significant effect on MCHC values of goat.

Table 6: Means ± SE of mean corpuscular volume (fl), mean corpuscular hemoglobin (pg) and mean corpuscular hemoglobin concentration (%) in early (EW) and late (LW) weaned Barki lambs in Siwa Oasis

Treatment		Early Weaning (EW)			Late Weaning (LW)		
Age	Sex	MCV	MCH	MCHC	MCV	MCH	MCHC
1 Month	M	27.75^{a}	10.03^{b}	36.26^{b}	27.91^{a}	10.10^{b}	36.24^{b}
	F	28.08^{b}	10.18^{a}	36.28^{b}	27.45^{b}	10.07^{b}	36.76^{a}
2 Month	M	26.96^{b}	9.98^{b}	36.99^{a}	26.54^{b}	9.90^{b}	37.37^{a}
	F	27.04^{b}	10.01^{a}	37.07^{a}	29.91^{a}	10.14^{a}	37.80^{a}
3 Months	M	31.29*	9.85*	31.54*	27.10^{b}	9.99^{b}	36.90^{b}
	F	30.77*	9.21*	29.91*	27.54^{a}	10.20^{a}	37.10^{b}
4 Months	M	28.15^{a}	10.60^{a}	37.89^{a}	27.72^{a}	10.20^{b}	36.79^{a}
	F	27.66^{a}	10.70^{a}	38.81^{a}	26.77^{a}	10.10^{b}	37.92^{a}
5 Months	M	27.70^{a}	10.77^{a}	38.92^{a}	32.32*	9.85*	30.47*
	F	27.93^{a}	10.03^{a}	36.06^{a}	31.00*	9.88*	31.88*
6 Months	M	28.35^{a}	10.86^{a}	38.43^{a}	26.59^{a}	9.97^{a}	37.52^{a}
	F	26.76^{a}	9.63^{a}	36.09^{a}	27.59^{a}	9.67^{a}	35.07^{a}
7 Months	M	27.39^{a}	9.84^{a}	36.03^{a}	27.19^{a}	9.97^{a}	36.72^{a}
	F	28.47^{a}	10.28^{a}	36.16^{a}	27.64^{a}	9.99^{a}	36.20^{a}
8 Months	M	27.41^{a}	9.55^{a}	34.87^{a}	27.19^{a}	9.55^{a}	35.15^{a}
	F	28.46^{a}	9.86^{a}	34.65^{a}	28.07^{a}	10.00^{a}	35.68^{a}
±SE	M	0.18	0.06	0.21	0.18	0.06	0.21
	F	0.15	0.05	0.17	0.15	0.05	0.17
Sex mean	M	28.12^{NS}	10.18^{NS}	36.40^{NS}	27.82^{NS}	9.94^{NS}	35.89^{NS}
	F	28.15	9.99	35.63	28.25	10.01	36.05
Overall mean		28.13^{NS}	10.08^{NS}	36.01^{NS}	28.03	9.97	35.97

M= males; F= females; SE= standard error; ^{a}= P<0.05; *= P<0.01; ^{NS}= Non-significant
MCV = mean corpuscular volume; MCH = mean corpuscular hemoglobin;
MCHC = mean corpuscular hemoglobin concentration

Table 7: Means ± SE of total leukocytes count (x10^3cells/µl), neutrophils (%), lymphocytes (%), monocytes (%), eosinophils (%) and basophils (%) for Barki lambs under early (EW) and late (LW) weaning age in Siwa Oasis

Treatment		Early Weaning (EW)					Late Weaning (LW)				
Age (months)		1	3	5	8	Average ±SE	1	3	5	8	Average ±SE
Males	WBC's	10.76^{a}	11.33^{a}	10.61^{a}	11.66^{a}	11.1±0.08*	10.69^{a}	10.88^{a}	11.50^{b}	11.57^{a}	11.2±0.08*
	Neut,%	64.07^{a}	64.2^{a}	62.14^{a}	64.80^{a}	63.8±0.23*	62.57^{b}	63.50^{b}	63.93^{a}	63.71^{b}	63.43±0.23*
	Lym,%	22.86^{b}	23.7^{b}	25.21^{a}	22.21^{b}	23.49±0.21	24.93^{a}	23.00^{a}	25.86^{a}	24.00^{b}	24.50±0.21*
	Mon,%	5.71^{a}	5.14^{b}	5.36^{b}	5.50^{a}	5.43±0.13^{NS}	4.79^{b}	6.00^{a}	4.43^{b}	5.36^{a}	5.14±0.13^{NS}
	Eos,%	6.93^{b}	6.64^{b}	7.07^{b}	7.14^{a}	6.90±0.19^{NS}	7.36^{a}	7.14^{NS}	5.36^{b}	6.79^{a}	6.70±0.19^{NS}
	Bas,%	0.43^{b}	0.21^{b}	0.21^{b}	0.43^{b}	0.32±0.01	0.36^{b}	0.29^{b}	0.43^{a}	0.36^{a}	0.36±0.01^{NS}
Females	WBC's	10.67^{b}	11.17^{b}	10.53^{b}	11.59^{b}	10.99±007	10.57^{b}	10.70^{b}	11.54^{b}	11.28^{b}	11.02±0.07
	Neut,%	61.81^{b}	63.57^{b}	62.0^{b}	63.67^{b}	62.7±0.19	63.0^{a}	64.71^{a}	61.67^{b}	63.90^{a}	63.32±0.19
	Lym,%	25.38^{a}	24.19^{a}	25.0^{b}	24.38^{a}	24.74±0.17*	24.3^{b}	22.29^{b}	25.62^{b}	25.14^{a}	24.34±0.17
	Mon,%	5.33^{b}	5.33^{a}	5.67^{a}	5.33^{b}	5.41±0.11	4.86^{a}	5.62^{b}	5.48^{a}	4.81b	5.2±0.11
	Eos,%	6.90^{b}	6.67^{a}	6.9^{a}	6.71^{b}	6.79±0.16	7.24^{b}	7.05^{b}	6.86^{a}	5.86^{b}	6.75±0.16
	Bas,%	0.57^{a}	0.24^{b}	0.43^{a}	0.43^{b}	0.42±0.03*	0.52^{a}	0.33^{a}	0.38^{b}	0.33^{b}	0.40±0.03

SE= standard error; *= P<0.01; ^{a}=P<0.05; ^{NS}= non-significant; WBC's = leukocytes; Neut = neutrophils; Lym = lymphocytes;
Mon = monocytes; Eos = eosinophils; Bas = basophils

Total WBC's and Differential Counts: Weaning process has been regarded as influencing immunity via stress-related mechanisms [60]. The results of the total leukocytes count values and their differential percentages of Barki lambs under early and late weaning ages are presented in Table 7. The present results indicated that, weaning age had no significant (P=0.61) effect on total circulating leukocyte count and the subsets percentages. In accordance, Blanco *et al.* [61] found no effect of age at weaning (90 vs. 120 days) or breed of calves on neutrophil and lymphocyte proportion but had minor effect on the pattern of change in WBC's count of the youngest calves (EW) over time. In both early and late weaned lambs, weaning process was a stressful experience as evidenced by the increase ($P<0.001$) in total WBC's count, this increase was largely due to the increase ($P<0.001$) of neutrophil and lymphocyte proportions at the 1st month after weaning process while monocyte and eosinophil proportions decreased compared with pre-weaning values. Similarly, Blanco *et al.* [61] weaning increased total WBC's more clearly in EW calves at 6hr, but concentrations declined at 168h after weaning. Previously, Hulbert *et al.* [62] found that, circulating neutrophils values were higher among early weaned calves (weaned at 23.7±2.3 d of age) than conventionally weaned calves (weaned at 44.7±2.3 d of age). Hickey *et al.* [63] reported a similar response to weaning in 7-month-old calves. These changes in leucocyte population are associated with acute stress [64] and may increase the susceptibility to disease and reflect a reduced capacity of calves to cope with stressors of the feedlot environment. However, no differences were found on the WBC's and their differential count due to age at weaning.

Regardless the effect of sex, the overall means of total WBC's count and neutrophil proportion were 11.05±0.05 and 63.3±0.15 vs. 11.10±0.05 $x10^3$cells/µl and 63.4±0.15 % for lambs under EW and LW ages, respectively. Similarly, Smith *et al.* [65] and Bueno *et al.* [66] stated that there was no immunosupression or increased predisposition to illness associated with age at weaning. Specifically, EW calves had an increase in total WBC numbers and the neutrophil to lymphocyte ratio 48 h after weaning. On the contrary, Pollock *et al.* [67] reported that age at weaning affected cellular-mediated immune responses in calves weaned at 5, 9 and 13 weeks of age, early weaning effects are essentially nutritional and all leucocyte parameters returned to baseline concentrations by 168 h after weaning. Recently, Davis *et al.* [68] reported that age of weaning had no affect on lymphocyte proliferation of pigs. Previous studies have documented clear changes in circulating numbers of neutrophils and lymphocytes, attenuated proliferation of T and B lymphocytes and modified expression of surface adhesion and antigen presenting molecules on leukocytes subjected to stress hormones *in vivo* and *in vitro* [63,69,70]. The hypothesis of this study was that the abrupt weaning of calves is a management stressor, which will increase the physiological measures of stress hormones, resulting in attenuation of immune function.

In respect of the effect of sex, the results in Table 7 indicated that early and late weaned male lambs had higher ($P<0.001$) values in WBC's count and neutrophil proportion and reached higher values at the end of experimental period compared with female lambs. The averages of total WBC's count and neutrophil proportion at the end of study for EW lambs were 11.66 and 64.80 vs.11.59x103cells/µl and 63.67 % for male and females, respectively. The corresponding values for LW lambs were 11.57 and 63.71 vs. 11.28 $x10^3$cells/µl and 63.90 % for male and females, respectively. Similarly, Opara *et al.* [54] found that, sex was observed to have a significant effect on the WBC's count, lymphocyte and neutrophil values of West African Dwarf (WAD) goats. The male (WAD) goats had increased total WBC's and lymphocyte values compared to the female animals, where as the females had increased neutrophil values compared to the male animals. This finding is similar to observation reported for Red Sokoto goats by Tambuwal *et al.* [52].

CONCLUSION

In conclusion, our findings showed that early weaning system improved growth performance compared with late weaning even though the non- significant effect of weaning age on the thermo-hematological parameters on all lambs. In this study weaning process was the most effective on hematological and immune competence on the 1st month after weaning in both EW and LW lambs (long-term effect). Late weaned lambs displayed better values in most thermo-hematological traits and immune competence compared with early weaned lambs, suggesting that late weaning process may give lambs advantages to cope with stress better.

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