

# Changes in Selected Blood Minerals, Vitamins and Thyroid Hormones in Barky Ewes during Late Pregnancy, Post-Partum and Early Lactation

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#### Abstract

The trial was carried out to investigate changes in the concentrations of selected blood minerals, vitamins and thyroid hormones in Barky ewes during late pregnancy, *post-partum* and early lactation under stable diet indoor condition. The experiment was carried out on 10 clinically healthy Barky ewes fed on ration composed of concentrate mixture and roughages according to their nutrient requirements during late pregnancy, *post-partum* and early lactation. Jugular blood samples were withdrawn from all ewes during the last 4 weeks of gestation,  $2^{nd}$  week *post-partum* and  $4^{th}$  week of lactation. The plasma concentrations of calcium (Ca), phosphorus (P), magnesium (Mg), copper (Cu), zinc (Zn), selenium (Se), vitamin A, vitamin E,  $\beta$ -carotenes were performed. In addition, serum levels of thyroid hormones (T3 and T4) were determined. Results showed that significant fluctuations (p<0.05) in the plasma levels of Zn, Se, vitamin A, vitamin E and  $\beta$ -carotene and serum T3 and T4 were occurred during late pregnancy, *post-partum* and early lactation.

In conclusion, blood minerals, vitamins and thyroid hormones' profiles can be used to predict *pre-partum* and *post-partum* problems associated with mal-nutrition.

Keywords: Barky ewes, blood minerals, vitamins, T3, T4, feeding

# **INTRODUCTION**

Metabolic profiles have been used to predict pre-partum and post-partum metabolic disorders as well as for diagnosis of metabolic diseases and assessment of the nutritional status of the animals (Radostits et al. 2000 and Khaled, 2001). Pregnancy and lactation are physiological statuses considered to modify metabolism in animals, (Krajničáková et al. 2003 and Iriadam 2007). Blood biochemical profiles carry important role not only during prenatal development but also during neonatal or postnatal period, because the initial period of four weeks after birth of the lamb is critical for its survival if dietary deficiencies remain there by Agrawal et al. (2007). Blood indices may vary depending on factors such as sex, age, weather, stress, season and physical exercise (Kaneko et al., 1999). Mineral and vitamin metabolism have an important role in physiological regulations of pregnancy and lactation. Pregnancy and lactation may resulted in metabolic stress which associated with changes in the mineral and vitamin profiles dependent upon the reproductive status of small ruminants (Antunovi et al. 2002; Yildiz et al. 2005 and Ceylan et al. 2009). Physiological losses of body minerals and vitamins occur during pregnancy and lactation. Therefore, serum concentrations of macro- and trace elements in addition to vitamins represent homeostatic mechanisms that are in a close relationship with nutritional status and hormonal regulation, (NRC,1980 and Krajnicakova et al. 2003). Thyroid hormones play a relatively important role in pregnancy and lactation. Changes in thyroid hormones due to stimulation of both physiological states (pregnancy and milk production) are difficult to distinguish, (Khaled, 2001).

The objective of this study was to investigate changes in selected blood minerals, vitamins and thyroid hormones in Barky ewes during late pregnancy, post-partum and early lactation under indoor stable diet conditions.

#### **MATERIALS & METHODS**

10, clinically healthy Barky ewes were used  $(3.0 \pm 0.4)$  years old), with mean body weight of  $50 \pm 0.7$  Kg, free from internal and external parasites and kept on one farm. All ewes were housed in a barn and fed twice a day (07:00, 19.00) with concentrate mixture, berseem hay and horse bean straw as shown in Tables (1 and 2) and formulated to meet the nutrient requirements of the ewes according to nutrient requirements of sheep, (NRC, 1985) and according to their physiological and productive status: Late pregnancy and post-partum in Table 3 (Fresh matter: 12.11% Crude protein (CP), 10.67 MJ/Kg Digestible energy (DE), 1.0 % Ca and 0.32 % P) and early lactation in Table 4 (Fresh matter: 13.0 % CP, 11.42 MJ/Kg DE, 1.0 % Ca and 0.39 % P). Fresh, clean water was given to the animals ad libitum.

Samples of concentrate mixture, berseem hay and horse bean straw were analyzed (Table 2) for DM, CP, DE, Ca and P according to **A.O.A.C.** (1990).

During the trial, all animals were kept under natural photoperiod and ambient temperature on a farm located in Cairo-Alexandria desert high way.

Blood samples, each of 10 ml, were withdrawn via jugular vein puncture from all ewes after morning meal during the last 4 weeks of gestation, 2<sup>nd</sup> week postpartum and 4<sup>th</sup> week of lactation. The blood was collected and processed to provide samples for plasma and serum analysis.

Plasma concentrations of Ca, Mg, Cu, and Zn were analyzed by using the Flame Atomic Absorption Spectrophotometer (HILGER H 1551, ATOMSPEK), while the plasma concentration of selenium was measured by using the Hydrogen generation atomic absorption spectrophotometer (HGAAS). While, plasma inorganic phosphorus was analyzed by the photometric methods using automated analyzer (COBAS MIRA S, ROCHE) and diagnostic kits.

Measurements of plasma vitamins A, E and  $\beta$ -carotene were performed by a method described by **Bouda et al**. (1980) using automated analyzer (Fluorescence spectrophotometer 204, PERKIN - ELMER).

While, quantitative measurements of total circulating Triiodothyronine ( $T_3$ ) and Thyroxine ( $T_4$ ), in blood serum were performed by the solid phase chemiluminescent enzyme immunoassay using IMMULITE automated analyzer and IMMULITE Kits which produced by DPC, Diagnostic products Corporation, Los Angeles, USA.

The data were expressed as Means and Standard errors (SE). The analysis of variance (ANOVA) was used to test the overall significance of differences among the means. Differences at p<0.05 were considered significant. The **SPSS** Statistical Computer Software, SPSS Inc., 2007 version 16.0 was used for statistical analysis.

 Table 1.Physical composition of Concentrate mixture for sheep

 (14 % CP)

Ingredient	Inclusion rate (%)	
Yellow corn, ground	56.70	
Soya bean meal (44 % CP)	7.0	
Linseed meal	10.0	
Wheat bran	15.0	
Rice bran	8.0	
Salt	1.0	
Limestone	2.0	
Sheep premix*	0.30	

\*Composition of sheep premix: vitamin A 12 000 000 IU; vitamin D<sub>3</sub> 3 000 000 IU; vitamin E 30 g; Mn 50 g; Fe 50 g; Zn 50 g; Cu 5 g; I 0.85g; Co 0.15g; Se 0.15 g.

 Table 2. Chemical composition of concentrate mixture, berseem hay and horse bean straw (as fed-basis)

	Concentrate mixture	Berseem Hay	Horse bean Straw
DM %	88.0	90.0	89.0
CP %	14.11	13.4	6.6
DE (MJ/Kg)	13.15	10.67	7.52
Ca %	0.84	2.25	1.0
Р%	0.58	0.26	0.15

Table 3. Physical and	chemical	composition	of the	ration	fed to
late pregnant ewe					

Physical composition/ Ingredient	As fed / Kg	DM / Kg	DM %
Concentrate mixture	0.50	0.44	25.88
Berseem hay	1.00	0.90	52.94
Horse bean straw	0.40	0.36	21.18
Total	1.9	1.7	100
Chemical composition of the diet *			
CP %	12.11		
DE MJ/Kg	10.67		
Ca %	1.0		
P %	0.32		
*			

\*As fed-basis

**Table 4.** Physical and chemical composition of the ration fed to early lactating ewe

Physical composition/ Ingredient	As fed / Kg	DM / Kg	DM %	
Concentrate mixture	1.0	0.88	42.30	
Berseem hay	1.1	0.99	47.60	
Horse bean straw	0.23	0.21	10.10	
Total	2.33	2.08	100	
Chemical composition of the diet *				
СР %	13.0			
DE MJ/Kg	11.42			
Ca %	1.0			
Р%	0.39			

\*As fed-basis

**Table 5.** Plasma concentrations of Macro- and Trace minerals in ewes during late pregnancy, post-partum and early lactation (Mean  $\pm$  SE)

Para	umeter	Late pregnancy	Post-partum	Early lactation
Ca	(mmol.l <sup>-1</sup> )	$2.21 \pm 0.04$	$2.35\pm0.07$	$2.43 \pm 0.08$
Pi	(mmol.l <sup>-1</sup> )	$2.05 \pm 0.09$	$2.02 \pm 0.27$	$1.90 \pm 0.17$
Mg	(mmol.l <sup>-1</sup> )	$0.998 \pm 0.042$	$0.996 \pm 0.045$	$0.933\pm0.03$
Zn	(µmol.l-1)	$16.32 \pm 0.77^{a}$	$14.92 \pm 1.32^{a}$	$13.01 \pm 0.77^{\mathrm{b}}$
Cu	(µmol.l-1)	$13.62 \pm 1.09$	14.31 ± 1.11	$14.05\pm0.62$
Se	(µg.l <sup>-1</sup> )	$113.78 \pm 16.65$ a	$136.39 \pm 26.54$ a	$90.46 \pm 12.82^{b}$

a,b) Means within sub rows with no common superscripts differ significantly (p<0.05).

Parameter Late pregnancy Post-partum Early lactation  $3.30\pm0.14^{\,a}$  $3.85\pm0.14^{\,a}$ Vitamin A (µmol.l-1)  $2.80 \pm 0.35^{b}$ Vitamin E (µmol.l-1)  $7.10\pm0.22^{\,\mathrm{a}}$  $6.45 \pm 0.43^{a}$  $6.02 \pm 0.18^{b}$ β-carotene ( $\mu$ mol.l<sup>-1</sup>)  $1.03 \pm 0.06^{a}$  $1.07 \pm 0.12^{a}$  $0.43 \pm 0.09^{b}$ 

Table 6. Plasma concentrations of vitamins A, E and  $\beta$ -carotene

in ewes during late pregnancy, post-partum and early lactation

(Mean  $\pm$  SE)

significantly (p<0.05).

a,b) Means within sub rows with no common superscripts differ

 
 Table 7. Serum concentrations of Thyroid hormones in ewes
 during late pregnancy, post-partum and early lactation (Mean

Parameter	Late pregnancy	Post-partum	Early lactation
T3 (nmol.l <sup>-1</sup> )	$1.76 \pm 0.129^{b}$	$2.22\pm0.37^{b}$	$3.24\pm0.25^{\text{ a}}$
T4 (nmol.l <sup>-1</sup> )	$70.38 \pm 4.93$ <sup>b</sup>	74.50 ± 11.74 <sup>b</sup>	$118.13 \pm 11.10^{a}$
T4:T3	39.98	33.56	36.46

a,b) Means within sub rows with no common superscripts differ significantly (p<0.05).

### **RESULTS & DISCUSSIONS**

Minerals are the essential nutrients bearing a significant role in the animal reproduction, because their excess or deficiency produces detrimental effect on the performance of livestock (Underwood, 1981). So, the blood mineral profile can be used to predict pre-partum and post-partum problems associated with mineral deficiencies.

Changes in the plasma levels of selected macro and traceelements in ewes during late pregnancy, post-partum and early lactation were illustrated in table 5. The results revealed that the concentrations of Ca, P, Mg and Cu in the plasma of ewes remained within the normal physiological ranges during different stages of reproduction as reported by Baumgartner & Pernthaner (1994) when sheep were kept indoors and fed hay with concentrate mixture supplement. However, significant (p<0.05) fluctuations in plasma Zn and Se levels were occurred during pre-partum and post-partum periods. Plasma Zn and Se levels were significantly (p<0.05) increased in late pregnancy and post-partum compared to early lactation. This could be related to the increase in the accumulation rate of Zn in foetus (William et al. 1972). Moreover, other reports (Egan, 1972; Masters & Fels, 1980) have shown that pregnancy resulted in depletion of Zn in grazing ewes. The marked decrease in serum Zn level in late pregnancy in Desert ewes could be partly related to haemodilution (Elnageeb & Abdelatif, 2010). Se increases the ewe's fecundity and improves health and increases body weight of the neonatal lambs (Horton & Mc carthy 1986).

Selenium is transported to the lambs along two pathways: via the placenta during the foetal stage, and with the colostrum at the neonatal phase. Selenium, even at low concentration in the ewe's body, is efficiently passed on to the foetus (Koller et al. 1984).

Many factors associated with diet composition can affect mineral utilization and bioavailability, the provision of salt lick can improve the mineral status of ewes particularly when exposed to nutritional and physiological stress,(Elnageeb & Abdelatif, 2010).

Significant (p<0.05) fluctuations in the plasma levels of vitamins A and E and  $\beta$ -carotene in ewes during late pregnancy, post-partum and early lactation were showed in the Table 6. The plasma vitamin A was significantly (p<0.05) decreased during last month of pregnancy compared to post-partum and early lactation. The decrease resulted from the utilization of the vitamin A for the colostrum and milk synthesis accordingly to the growing of the foetus, (Chawla and Kaur, 2004; Daniel et al. 1991). It was determined that plasma vitamin A levels during last month of pregnancy were similar to findings of other researchers (Bouda et al. 1980; Vlcek et al. 1980 and Kara et al. 2001).

Plasma levels of vitamin E and  $\beta$ -carotene were significantly increased in late pregnancy and *post-partum* compared to early lactation. It was demonstrated in this study that the plasma vitamin E and B-carotene levels were similar to the results obtained by other researchers (Yildiz et al. 2005; Aksakal et al. 1995; Kara et al. 2001 and lynch 1983).

The reduced concentrations of antioxidants such as vitamin A, E and  $\beta$ -carotene affect the immune system and phagocytic activity of cells and results in an increase in the incidence of mastitis and puerperal diseases in pregnancy, delivery, and postpartum periods.

Mean concentrations of serum thyroid hormones T3, T4 and T4:T3 in ewes during late pregnancy, post-partum and early lactation were illustrated in Table 7. Serum levels of T3 and T4 were significantly declined in last month of pregnancy and post-partum compared to early lactation. Decrease in thyroid hormones around parturition due to alterations in cardiac output and increased blood volume, (Illek et al. 1998; Dalvi et al. 1995 and Khaled, 2001).

A significant increase (p<0.05) in serum levels of T3 and T4 during early lactation might be due to thyroid hormones are galactopoietics and may play an important role in regulation of lactation and stimulate the basic metabolic rate via the metabolism of carbohydrates, lipids and proteins, (Kaneko, 1999).

Pregnancy, lactation and feeding strongly influence the dynamics of plasma thyroid hormone concentrations, low and high amounts of dietary protein in the diets significantly decrease plasma concentration of thyroid hormones T<sub>4</sub> and T<sub>2</sub>, (Tiirats, 1995, 1997).

## CONCLUSION

Reproductive status and nutrition have to taken into consideration for correct interpretation of blood mineral, vitamin and thyroid hormone values for sheep. The blood minerals, vitamins and thyroid hormones' profiles can be used to predict pre-partum and post-partum problems associated with mal-nutrition.

± SE)

### REFERENCES

- Agrawal, S.; R. Kumar and K.B. Sharma.(2007): Studies on metabolic profile of ewes during gestation and possible relation with neonatal mortality. Indian J. Small Ruminants, 13: 192-195.
- [2] Aksakal, M.; Karakilcik, A.Z.; Kalkan, C.; Çay, M. and Naziroglu, M. (1995): Levels of β-carotene and vitamin E at various stages of reproductivity in cows. Turk J Vet Anim. Sci, 19, 59-64.
- [3] Antunovic, Z.; Sencic, D.; Šperada, M. and LIKER, B.( 2002): Influence of the season and the reproductive status of ewes on blood parameters. Small Ruminant Research 45: 39-44.
- [4] A.O.A.C. (1990): Official methods of analysis (15th Ed). Association of Official Analytical Chemists ,Washington, DC.
- [5] Baumgartner, W. and Pernthaner, A. (1994): Influence of age, season and pregnancy upon blood parameters in Austrian Karakul sheep. Small Ruminant Research 13:147-151.
- [6] Bouda, J.; Jagos, P.; Dvorak, VI. and Hamsik, V.1(980): Vitamin A and carotene metabolism in cows and their calves fed from buckets. Acta Vet Brno 49: 45-52.
- [7] Ceylan, E.; Tanritanir, P. and Dede, D. (2009): Changes in some macro-minerals and biochemical parameters in female healthy Siirt Hair goats before and after parturition. J. Animal and Veteinary advances, 8 (3): 530-533
- [8] Chawla, R. and Kaur, H. (2004): Plasma antioxidant vitamin status of periparturient cows supplemented with α-tocopherol and β-carotene. Anim.Feed Sci Technol. 114: 279- 285.
- [9] Dalvi, S.H.; Deshmukh, B.; Marti, T. and Talvelkar, B. A. (1995): Concentration of blood serum thyroid hormones during late pregnancy, parturation and early lactation of cross bred cows. Indian J. Anim. Sci. 65: 15 -19
- [10] Daniel, L.R.; Chew, B.P.; Tanaka, T.S. and Tjoelker, L.W. (1991): β- carotene and vitamin A effects on bovine phagocyte function in vitro during peripartum period. J. Dairy Sci 74, 124-128.
- [11] Egan, A.R. (1972): Reproductive responses to supplemental zinc and manganese in grazing Dorset Horn ewes. Australian J. Experimental Agriculture and animal Husb., 12: 131-135.
- [11] Elnageeb, M.E. and Abdelatif, M. (2010): The minerals profile in Desert sheep (*Ovis aries*): Effects of pregnancy, lactation and dietary supplementation. American-Eurasian J. Agric. & Environ. Sci., 7 (1):18-30.
- [12] Horton, S.A. and McCarthy, F.D. (1986): Use of injectable vitamin E and selenium-vitamin E emulsion in ewes and suckling lambs to prevent nutritional muscular dystrophy. J. Anim. Sci., 62: 497-508.
- [13] Illek, J.; Khaled, N. F.; Sedlakova, D. and Sindelar, M. (1998): Concentration of thyroid hormones in dairy goats during lactation. In: 6<sup>th</sup> International symposium on the milking of Small Ruminants, Sept 26- Oct 1, Athens, Greece.
- [14] Iriadam, M. (2007): Variation in certain haematological and biochemical parameters during the peri-partum period in Kilis does. Small Ruminant Research 73: 54-57.
- [15] Kaneko, J. J.; J.W. Harvey and L.B. Michael, (1999):

Clinical Biochemistry of Domestic Animals. Academic Press, Sandiego California, USA.

- [16] Kara, H.; Karatas, F.; Servi, K.; Akar, Y. and Konar V. (2001): Investigation of levels plasma adrenalin, noradrenalin, vitamins A and E at various of pregnancy periods in Holstein cattle. F U J of Health Sci, 15: 171-174.
- [17] Khaled, N.F. (2001): Interactions between nutrition, blood metabolic profile and milk composition in dairy goat. Ph.D thesis, VFU, Brno, Czech Repu. Pp. 227
- [18] Koller, L.D.; Whitbeck, G.A. and South, P.J. (1984):Trans-placental transfer and colostrum concentrations of selenium in beef cattle. Am. J. Vet. Res. 45: 2507-2510.
- [19] Krajnicakova, M.; Kovac, G.; Kostecky, M.; Valocky, I. and Maracek, I. (2003): Selected clinico-biochemical parameters in the puerperal period of goats. Bulletin of Veterinary. Inst. Pulawy, 47: 177-182.
- [20] Lynch, G.P. (1983): Changes of tocopherols in blood serum of cows fed hay or silage. J. Dairy Sci, 66, 1461-1465.
- [21] Masters, D.G. and Fels, H. E. (1980): Effect of zinc supplementation on the reproductive performance of grazing merino ewes. Biology of trace Element Res., 2: 281-290.
- [22] NRC, (1980): Mineral Tolerances of Domestic Animals. National Research Council, National Academy of Science, Washington, DC.
- [23] NRC, (1985): Nutrient requirements of sheep. Sixth Revised edition, National Academy Press, 2101 Constitution Avenue, NW, Washington, DC 20418.
- [24] Radostts, O.M.; C.C. Gay, D.C. Blood and K.W. Hinchelife. (2000): Veterinary Medicine. 9th Edn., Harcourt Publishers Ltd., London, pp: 1417-1420.
- [25] SPSS 16.0, (2007): SPSS Statistical Computer Software, SPSS Inc., Chicago, IL, USA.
- [26] Tiirats, T. (1995): The blood plasma concentration of thyroxine (T<sub>4</sub>), triiodothyronine (T<sub>3</sub>), cortisol and insulin in cows at low, normal and high dietary protein intake. In.: Proc. of IX <sup>th</sup> Int. Conf. on production diseases in farm animals, Sept. 11-14; Free Univ. of Berlin , Fac. of Vet. Med. Ed. By H. Martens, Stuttgart: Enke.
- [27] Tiirats, T.(1997): Thyroxine, triiodothyronine and reverse- triiodothyronine concentrations in blood plasma in relation to lactational stage, milk yield, energy and dietary intake in Estonian dairy cows. Acta Vet. Scand. 38: 339 -348.
- [28] Underwood, E. J. (1981): The Mineral Nutrition of Livestock. Common wealth Agricultural Bureau.London, UK.
- [29] Vlcek, Z.; Nedbalkova, J.; Svobodova, R. and Chlupac, J. (1980): Vitamin A in the blood serum, liver, corpora lutea, placentomas and fetal fluids of cows and in the blood serum and liver of their fetuses in the latter third of pregnancy. Acta Vet Brno, 49: 37-44.
- [30] Williams, J.W.; Beutler, A.; Reselev, J and Rundels, R.W. (1972): Haematology, Mc Grawhill. New York, London, pp: 100 – 124.
- [31] Yildiz, A.; Balikci, E. and Gurdogan, F. (2005): Serum mineral levels at pregnancy and postpartum in single and twin pregnant sheep. Biology of Trace Element Res., 107(3): 247-254.