Egyptian Journal of Sheep & Goat Sciences. Vol. 10, (1) P: 41-54, April 2015

CHANGES IN PRODUCTIVE PERFORMANCE, HEMATO-BIOCHEMICAL INDICES, IMMUNE AND ANTIOXIDANT STATUS OF GROWING OSSIMI LAMBS SUBJECTED TO VITAMINS A AND E ADMINISTRATION

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ABSTRACT

This study included 32 growing Ossimi male lambs, averaged 3.5 months of age and 19.08±1.09 kg body weight. They divided into four equal groups (8 lambs each) to determine the effects of vitamins A and E oral administration on growth performance, hematological and biochemical parameters, immune and antioxidant status. The 1st group served as control, the 2nd group (VA) received oral administration of vitamin A at 50,000 IU/head/biweekly, the 3rd group (VE) received vitamin E at 400 mg/head/biweekly, and the 4th group (VA+E) received 50,000 IU vitamin A plus 400 mg vitamin E /head/biweekly. Data showed an increase (P<0.05) in body weight (BW) of lambs received VA and VA+E *vs*. control. Average daily gain (ADG) increased (P<0.05) for lambs received VA and VA+E *vs*. control and VE treatment. ADG was higher (P<0.05) for lambs received VA+E than those received each of VA or VE alone. Feed conversion efficiency (FCE) was improved (P<0.05) for lambs received VA and VA+E *vs*. control and VE treatment. No differences in dry matter intake (DMI) was recorded among the experimental groups. Blood Hb concentrations increased (P<0.05) for lambs received VA, VE and VA+E, while RBCs count and PCV % were increased (P<0.05) only for lambs received VE and VA+E. No significant response of WBCs count among lambs received VA, VE and VA+E *vs.* control. Lymphocytes % increased (P<0.05) with no significant differences in eosinophils, basophils and monocytes % for lambs received VA, VE and VA+E *vs*. control. Lambs treated with VE alone exhibited a decrease (P<0.05) in neutrophils *vs*. lambs of control, VA and VA+E. Serum total protein (TP) and globulin concentrations increased (P<0.05) with VA, VE and VA+E treatments vs. control. Serum TP and albumin concentrations increased (P<0.05) for lambs treated with VA+E treatment when compared to those treated with VA treatment alone. Lambs received VA, VE and VA+E treatments had higher (P<0.05) serum IgG concentrations than the control. VE treatment increased (P<0.05) serum IgG concentrations *vs.* VA treatment. There were significant (P<0.05) increases in serum total antioxidant capacity and glutathione peroxidase (GSH-Px) activity for lambs received VA, VE and VA+E treatments *vs*. control. Lambs of VE treatment had higher (P<0.05) serum GSH-Px activity than those of VA treatment. No significant differences in serum concentrations of glucose, cholesterol, AST and superoxide dismutase activity was noticed due to treatments VA, VE and VA+E vs. control. These results indicate that combination of vitamins A and E exerted beneficial additive effects that improve ADG and physiological responses of lambs. Vitamin A was more effective than vitamin E in enhancing growth performance of lambs, whereas vitamin E had more potent effect on improving immune response and antioxidant status.

Key words: *Vitamin A and E, Growing Lambs, Performance, Physiological responses, Antioxidant status.*

INTRODUCTION

Antioxidant vitamins are vital components for mammalian cell defense against compounds that cause oxidation of cellular molecules. Vitamins A and E are essential, natural occurring and fat-soluble nutrients that are involved in several important biological processes such as immunity, protection against tissue damage, reproduction, growth and development **(Debier and Larondelle, 2005)**. Vitamin A is essential for stimulation of growth, proper development of skeletal tissue, normal reproduction and maintaining the integrity of epithelial tissues **(Pond *et al*., 1995)**. Vitamin E, the most potent antioxidant, works to scavenge free radicals and acts as

terminator of lipid peroxidation **(Liebler, 1993)**. Vitamins A and E are cellular antioxidants, preventing peroxidative damage in cell membranes, and are essential for the well-functioning immune system **(Meglia *et al.*, 2004)**. Animal immune function and health could be impaired by inadequacies of vitamin A and E as antioxidant defense **(Chew, 1987)**. Therefore, the amount of vitamin A needed for immune-enhancement is reported to be higher than the suggested required amounts by NRC **(Nockles and Blair, 1996)**. Supplement with vitamin E at levels above requirements is also associated with variable improvements in sheep performance and immune function **(Rooke *et al.*, 2004)**. Vitamin E is considered synergistic to vitamin A due to its antioxidant activity, which results in a sparing effect upon vitamin A. Therefore, loss of vitamin A from liver stores is accelerated in the presence of vitamin E deficiency, resulted of increased fragility of lysosomal membranes **(Watts, 1991)**. Vitamin E therapy appears to be an effective treatment for hypervitaminosis of vitamin A **(St-Claire *et al.*, 2004)**. In cattle, high dietary levels of vitamin A could depress vitamin E utilization **(Schelling *et al.*, 1995)**. In sheep, however, it was suggested that oversupply of dietary vitamin A does not antagonize vitamin E turnover **(Hidiroglou, 1993)**; and that daily administration of vitamin A, approximately 150 times greater than the daily requirement, were well tolerated by sheep **(Raoofi *et al.*, 2010)**. In rats, it has been suggested that vitamin A might be considered as a potential antioxidant similar to vitamin E in animal nutrition **(Kartha and Krishnamurthy, 1977)**.

However, the inter-relationships between vitamins A and E are complex and the mechanisms of their interaction are not fully understood. The present study, therefore, focused on some mechanistic aspects through which administration of vitamin A and vitamin E may influence growth performance of growing Ossimi lambs and their physiological reactions related to hematology, serum biochemical profile, immune and antioxidant status.

MATERIAL AND METHODS

Animals and experimental treatments:

Thirty-two of growing Ossimi male lambs average 3.5 months age and 19.08±1.09 kg body weight were used in this study during November 2013 to February 2014 at the Farm of Animal Production Department, Faculty of Agriculture, El-Minia University. The animals were randomly divided into 4 equal groups (8 lambs each) of similar initial body weights. The 1st group served as control, the 2nd group (VA) received oral administration (drenching) of vitamin A (as palmitate) at rate 50,000 IU/head/biweekly, the 3rd group (VE) received vitamin E at rate 400 mg/head/biweekly, and the 4th group (VA+E) received vitamin A at 50,000 IU/head/biweekly plus vitamin E at 400 mg/head/biweekly.

Feeding and management:

Animals were fed on concentrate feed mixture (CFM) and rice straw to cover their nutrient requirements according to live body weight **(NRC, 1985)**. The CFM consisted of 15 % yellow corn, 15 % soybean meal, 30 % sugar beet pulp, 37 % wheat bran, 2.0 % limestone and 1.0 % salt. The calculated feeding value of the CFM was 69.75 % TDN, 17.04 crude protein and 2.53 ME (Mcal/kg). The concentrate contained 1.41 mg/kg DM ß-carotene and 11.97 mg/kg DM vitamin E. The NRC requirements for growing lambs are 69 µg of ß-carotene/kg live weight/day (47 IU of vitamin A/kg live weight/day) and 20-25 mg of vitamin E /lamb/day. The treated animals received 1.41 mg/kg DM of dietary ß-carotene with 50,000 IU/head/biweekly of vitamin A and 11.97 mg/kg DM with 400 mg/head/biweekly of vitamin E. Feeds were offered twice a day at 8 am and 2 pm and mineral blocks and drinking water were available along the experiment. Body weights of lambs were recorded at the start of experiment then biweekly. Feed intake was recorded daily. Averages of daily gain and feed conversion efficiency of lambs were calculated. Parameters were recorded in the morning before animals access to feed or water.

Blood sampling and measurements:

Heparinized blood samples were collected from the jugular vein of each animal at 8.00 am before feeding or drinking. Whole blood samples were analyzed after collection for hemoglobin (Hb), packed cell volume (PCV), red blood cell counts (RBCs) and white blood cell counts (WBCs). The Hb concentration was determined using cyanomethomoglobin method **(Campbell, 1995)**. The PCV was determined using micro-hematocrit tubes with micro-hematocrit centrifuge at 12000 rpm for three minutes. The RBCs and WBCs were counted using the light microscope. Stained blood smears with Lieshman's stain were prepared for the differential WBCs count **(Dacie and Lewis, 1991)**. Non-heparinized blood samples were collected from the jugular vein of each animal and left to clot at room temperature for at least 4 h, then the clots were removed and sera were cleared by centrifugation at 1500×g for 20 min and stored at -20 ºC for later biochemical assay. Serum glucose, total protein, albumin, cholesterol and aspartate transaminase (AST) were determined colorimetrically using Bio-diagnostic product kits (Egypt). Serum globulin concentrations were calculated by difference between total protein and albumin concentrations. Serum immunoglobulin G (IgG) was quantified using ELISA kit supplied by WKEA MED Supplies Corporation. The Elisa micro plate having standards (5 wells) and samples was read at 450 nm using Elisa READER (BIO TEK ELX808), USA. The assay of serum IgG range was 0.7 to 30 µg/ml. Serum total antioxidant capacity (TAC), superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) activities were analyzed colorimetrically by STAT LAB SZSL60-SPECTRUM, using Bio-dignostic kits (Bio-dignostic Company, Egypt). The analyses were performed at Cairo University Research Park (CURP), Faculty of Agriculture, Cairo University.

The data were analyzed by least square means analysis of variance using General Linear Models (GLM) procedure of the statistical analysis system (SAS, 2000). The model used to analyze different traits studied for ewes or lambs was as follows:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where: Y_{ij}= ij^{th} Observation, µ = Population mean; T_i = Effect of i^{th} treatments and eij= Random error. Duncan's Multiple Range test was used to detect differences between means of the experimental groups (Duncan, 1955).

RESULTD AND DISCUSSION

Productive performance:

The results in Table (1) illustrate the effect of vitamin A (VA) and/or vitamin E (VE) on growth performance of Ossimi growing lambs. Data showed a significant ($P<0.05$) increase in final body weight (FBW) of lambs received VA and VA+E *vs*. control and the higher FBW was recorded with VA+E treatment (38.76 kg). Average daily gain (ADG) increased ($P<0.05$) by 40.3 and 69.8 % respectively for lambs received VA and VA+E *vs*. control. ADG was higher ($P<0.05$) for lambs received VA+E (219.0 g/d) than those received each of VA (181.0 g/d) or VE (145.0 g/d) alone. Feed conversion efficiency (FCE) was improved ($P<0.05$) by 38.2 and 69.0 % for lambs received VA and VA+E *vs*. control. No significant differences in dry matter intake (DMI) were recorded among the experimental treatments.

The significant improvement in FBW, ADG and FCE for lambs received VA are in line with some earlier studies showed significant beneficial effects of vitamin A that improve productive performance of growing calves **(El-Masry *et al.*, 1998)**, sheep **(Bruns and Webb, 1990; Soliman, 2005)** and buffalo calves **(El-Barody *et al.*, 1993)**. Since vitamin A has major function in metabolism that preserve stability, structural integrity and normal permeability of cell and subcellular membranes, it has positive effects on tissue biosynthesis and growth promotion **(Chew, 1993)**. Generally, vitamin A is supplemented to ruminant especially to those confined to insure their optimum health and maximum productivity **(Alosilla *et al.*, 2007)**. Vitamin A has a role in regulating growth hormone gene expression **(Bedo *et al.*, 1989)**, and energy homeostasis by enhancing uncoupling protein 1 (UCP1) mRNA gene expression and decreasing

serum leptin levels **(Kumar *et al.*, 1999)**. Thus, metabolic disorder, reduced feed efficiency, slowed gains and growth retardation are clinical signs occurred with lack of vitamin A **(Pond *et al.*, 1995)**. Although the effect of vitamin E on lambs' performance was statistically not significant, the averages of FBW, ADG and FCE tended to improve by 4.8, 12.5 and 8.5 % for lambs received VE *vs.* control (Table, 1). Though supplement of vitamin E was beneficial in improving animal' performance of lambs **(Shetaewi *et al.*, 1992)**, buffalo calves **(Amer and Hashem, 2008)** and cattle calves **(Hays *et al.*, 1987 and Galyean *et al.*, 1999)**, other reports showed no significant or limited responses in performance' parameters with supplement of vitamin E on growing male lambs **(Zhao *et al.*, 2013)**, goats **(Yang *et al.*, 2010)** and beef cattle **(Rivera *et al*., 2002)**.

The presented results clearly show that co-administration of VA+E in lambs were more effective and significantly increased their ADG than each of VA or VE alone (Table, 1). In addition, the combined treatment (VA+E) also improved FCE *vs*. control or VE-treatment alone. These results may signify the additive effect of vitamin A and E that enhance lambs' performance. Such positive combined effect of both vitamins was shown in Holstein steer calves received 30,000 IU/d of vitamin A plus 250 IU/d of vitamin E which improved ($P<0.05$) their FBW and ADG and slightly enhance DMI with no significant effect on gain or dietary net energy **(Salinas-Chavira *et al*., 2014)**. Furthermore, a combination of vitamins A and E was more effective than either vitamin alone in reducing heat stress performance in broiler **(Sahin *et al.*, 2001)**. The relationship between vitamin A and vitamin E has been proposed in such a way that vitamin E appears to have an important effect on the utilization and perhaps absorption of vitamin A and that vitamin E protects vitamin A from oxidative breakdown **(Gallo-Tores, 1980)**. In contrast, dietary supplementation of vitamin A had no effect on performance (Feed intake, ADG and FBW) of young lambs **(Arnett *et al.*, 2007)**. Several factors including the extent and duration of treatment, age, phase of growth, species, route of administration and other environmental conditions, likely contribute to the discrepancies in growth performance response to both vitamins supplementation.

Hematological parameters:

The results in Table 2 showed that hematological parameters (Hb, RBCs and PCV) were significantly changed in response to vitamin A and/or vitamin E administration. Blood Hb concentration increased ($P<0.05$) for lambs received VA, VE and VA+E, while RBCs and PCV % were increased ($P<0.05$) for lambs received VE and VA+E than control or VA treatment. This response of hematologic indices may signify a case of active metabolism and biological oxidation on the cellular base for these treated lambs **(Frandson, 1986)** that lead to availability of metabolites required for tissue growth.

Table 1: Effects of vitamin A and vitamin E administration on productive performance of growing Ossimi lambs (mean ± SEM).

	Treatments					
Parameters	**Control**	**VA**	**VE**	**VA+E**	**SEM**	**Sig.**
IBW (kg)	19.08	19.08	19.08	19.08	1.09	NS
FBW (kg)	30.65 c	35.38 ab	32.11 bc	38.76 a	1.47	*
ADG (g/day)	129.0 c	181.0 b	145.0 c	219.0 a	0.008	*
DMI (kg/head/day)	1.14	1.20	1.18	1.22	0.045	NS
FCE (kg feed/kg gain)	9.2 a	6.66 b	8.49 a	5.54 b	0.59	*

a,b,c means within the same row having different superscripts significantly different (* $P<0.05$), NS = not significant

VA = Vitamin A, VE = Vitamin E, VA+E = Vitamin A plus vitamin E.

IBM = Initial body weight, FBW = Final body weight, ADG= Average daily gain,

DMI = Dry matter intake, FCE = Feed conversion efficiency.

Table 2: Effects of vitamin A and vitamin E administration on hematology and differential leucocytes count parameters of growing Ossimi lambs (mean ± SEM).

Parameters	Treatments				SEM	Sig.
	Control	VA	VE	VA+E		
Hb (g/dl)	10.04 c	11.58 a	12.72 b	12.15 ab	0.30	*
RBCs ($x10^6/mm^3$)	9.0 b	9.6 b	11.4 a	11.1 a	0.21	*
PCV (%)	29.0 b	30.1 b	35.7 a	33. 8 a	1.04	*
WBCs ($x10^3/mm^3$)	7.22	7.24	7.30	7.44	0.20	NS
Neutrophils (%)	30.66 a	28.1 a	25.0 b	27.8 ab	2.40	*
Eosinophils (%)	4.06	4.18	4.04	4.1	0.15	NS
Basophils (%)	0.55	0.52	0.51	0.5	0.02	NS
Lymphocytes (%)	58.8 b	63.1 a	66.2 a	63.3 a	3.77	*
Monocytes (%)	4.00	4.10	4.15	4.0	0.11	NS

a,b,c means within the same row having different superscripts significantly different (* P<0.05). NS = not significant

VA = Vitamin A, VE = Vitamin E, VA+E = Vitamin A plus vitamin E.

Some studies reinforce the present results showing similar response of increasing blood Hb due to vitamin A administration in sheep **(Soliman, 2005)**, goats **(Yang *et al*., 2010)** and Holstein dairy calves **(Moosavian *et al*., 2010)**. Vitamin A appear to be involved in the pathogenesis of anemia through diverse biological mechanisms via enhancing growth and differentiation of erythrocyte progenitor cells, potentiates immune system to infection, reduces the anemia of infection and requiring it for Fe metabolism and mobilization **(Semba and Bloem, 2002)**. Moreover, studies in rats have shown that Fe deficiency alters plasma and liver levels of vitamin A. So, vitamin A deficiency may associated with altered Fe metabolism, including reduced plasma Fe and sometimes anemia; and this effect does not appear to be caused by increased RBC destruction **(Pond *et al.*, 1995)**. Indeed, evidences in dairy calves treated with vitamin A and/or Fe indicated that the relationship between vitamin A and Fe remains to be unclear **(Moosavian *et al*., 2010)**. The significant increase in Hb, RBCs and PCV for lambs treated with VE has similar response, for these parameters, to vitamin E supplement found in coarse-wool lambs (Shetaewi *et al*., 1992). Hematological responses to vitamin E could mediate its enhancing for erythropoiesis and decreasing the premature erythrocyte hemolysis by reducing the fragility of erythrocytes **(Jiliani and Iqbal, 2011)**.

As shown in table 2, no significant response of WBCs count was observed among lambs received VA, VE and VA+E compared to control. The WBCs profile showed a marked increase (P<0.05) in lymphocytes (%) concomitant with no significant differences in eosinophils, basophils and monocytes percentages for lambs received VA, VE and VA+E *vs*. lambs of control, but they did not show significant differences among the treated groups. However, lambs treated with VE alone exhibited a decrease (P<0.05) in neutrophils *vs*. lambs of control, VA and VA+E. The insignificant change in WBCs count in lambs treated with VA agrees with similar response found in sheep **(Soliman, 2005)**, goats **(Yang *et al*., 2010)** and dairy calves **(Moosavian *et al*., 2010)**. The response of significant increase in lymphocyte percentages for lambs treated with VA, VE and VA+E may be considered a useful response that improve their immune function, disease resistance and general health. The increase in blood lymphocyte populations may be a good indicator of an immune response **(Qureshi *et al*., 2001)**. The presented results are consistent with some studies dealt with the effect of vitamins A or E on lymphocyte counts. For instance, lymphocyte populations were significantly increased in response to either injection of vitamin E in calves **(Reddy *et al*., 1986)** and in Awassi rams **(Ali *et al*., 2009)**, or dietary supplementation of vitamin A in goats **(Yang *et al*., 2010)**, indicating that both

vitamins could improve the immune function of these animals. Vitamin E can affect lymphocyte production in the bone marrow of ruminants **(Hogan *et al.*, 1993)**. Vitamin A also plays a central role in the development and differentiation of WBCs, such as lymphocytes, which play critical roles in immune response **(Semba, 1998)**.

In the current study, VA and VA+E treatments had no significant effect on neutrophils %, while VE treatment significantly reduced neutrophils % with no significant differences in the % of monocytes, acidophils or basophils (Table, 2). Similar results were noticed in Awassi rams when neutrophils % were decreased with the injection of vitamin E **(Ali *et al.*, 2009)**. However, neither phagocytic index nor neutrophils % was affected by vitamin E injection in cattle **(Hogan *et al.*, 1992)**. In *in vitro* study on growing calves, **Eicher *et al.* (1994)** reported that neutrophil phagocytosis improved with vitamins A and E compared to vitamin E alone. Indeed, differences in neutrophils response affected by many factors including supplement levels of vitamin E, mode of administration, selenium status and animal species used. Monitoring the hematological indices in sheep gives a clear picture of their nutritional and health status before the changes are visible on the animal **(Antunovic *et al.*, 2009)**.

Serum biochemical parameters:

Data in Table 3, showed an increase ($P<0.05$) in serum total protein (TP) and globulin concentrations with VA, VE and VA+E treatments vs. control while serum TP and albumin concentrations increased ($P<0.05$) for lambs treated with VA+E compared to those treated with VA alone. This result may revealed that administration of vitamin A and/or vitamin E at dosage used in this study improved protein synthesis and metabolism. This response may be account for the trend towards improved growth performance (FBW, ADG and FCE) for these treated lambs. The present results agree with comparable responses reported by **El-Shahat and Abdel-Monem (2011)** who demonstrated that dietary supplementation of vitamin E significantly improved levels of serum TP, albumin and globulin concentrations in Baladi sheep. Elevated plasma globulin was observed on lambs received VA as shown in Table 3. **El-Masry *et al.* (1998)** noticed similar trend of increasing globulin fractions for growing calves supplemented with vitamin A, but they failed to show a significant increase in plasma TP or albumin. In Ossimi sheep, supplement with vitamin A significantly increased plasma TP and albumin, however, globulin was not affected **(Soliman, 2005)**. In cattle, feeding vitamin E (4400 IU/d) increased serum albumin fraction but did not affect different fractions of globulin **(Rahmani *et al.*, 2014)**. Similar responses were detected in buffaloes **(Helal *et al.*, 2009)**. In rabbits, plasma TP concentration was not significantly affected by vitamin E deficiency, but albumin levels were lower and globulin levels were higher **(Diehl and Delincee, 1986)**. In the current study, a significant increase in serum albumin concentrations was observed for lambs received VE and VA+E treatments compared to those received VA alone (Tables, 3). This response may be physiologically useful in controlling the osmotic pressure and flow of water between blood and tissue fluids **(Kobeisy *et al.*, 1997)**. Furthermore, the significant increases in globulin and immunoglobulin G concentrations as well as lymphocytes % of lambs treated with VA and/or VE (Tables, 2) may support the findings that both vitamins could enhance the animal ‘immune function **(Smith and Hays, 1987; Shinde *et al.*, 2007; Yang *et al.*, 2010)**.

Data also show that the other serum metabolite concentrations such as glucose, cholesterol and aspartate transaminase (AST) activity were not significantly differed due to treatments with VA, VE and VA+E (Tables, 3). These results are in agreement with other reports showed no significant changes in serum cholesterol, triglyceride concentrations or the sum of the two lipid fractions in sheep fed vitamin E alone **(Njeru *et al.*, 1994)**. Over-supplementation of vitamin A had no effect on serum TP, albumin and globulin in dairy calves (Moosavian *et al.*, 2010). Similar findings were obtained with feeding vitamin E that did not affect serum glucose, cholesterol and AST concentrations in rats **(Jang *et al.*, 1999)**.

Serum immune and antioxidant status:

The results showed that lambs received VA, VE and VA+E treatments had higher (P<0.05) serum immunoglobulin G (IgG) concentrations by 17.0, 34.4 and 22.3 %, respectively than the control (Table, 4). Lambs received VE treatment had higher (P<0.05) serum IgG concentrations by 15.0 % than those received VA treatment. Serum IgG concentration of lambs received the combined treatment of VA+E was not significantly differed compared with each of VA or VE alone. Earlier reports reviewed the essential role of vitamin A in immune function. According to **Smith and Hays (1987)**, with respect to immunity, supplement of vitamin A enhanced the delay of hypersensitivity, cytotoxic activity, and graft-versus-host responses to antigens. In addition, serum Igs responses and plaque-forming cell numbers were increased by vitamin A supplement. Immune enhancement accompanied by elevated T-lymphocyte numbers and interleukin-2 production. Conversely, vitamin A deficiency was associated with depressed immunity. Delayed hypersensitivity to a contact allergen, natural killer cell activity, and mitogen responses were depressed during vitamin A deficiency. Similarly, decreased serum antibody responses occurred in animals with reduced serum vitamin A concentrations. In goats, the improved antioxidant status together with the enhanced immune function, by vitamin A supplementation, indicated that vitamin A might serve as an antioxidant to protect the immune cells against oxidant stressors and thereby maintain optimum immune function **(Yang *et al*., 2010)**. So, the observed significant increases in serum IgG concentrations and lymphocytes percentage of lambs treated with VA in the present study (Tables 4 and 2) suggest that VA supplementation could enhance the immune response of lambs. On the other hand, VE was more potent to increase serum IgG concentration especially when compared to the effect of VA (Table, 4). Vitamin E has been implicated in stimulation of serum antibody synthesis, particularly IgG antibodies **(Tengerdy, 1980)**. Vitamin E at high doses had increased serum IgG concentrations in lambs **(Gentry *et al*., 1991)** and in ewes **(Anugu *et al*., 2013)**. In addition, linear increase in serum IgG titers was noted with vitamin E supplement in beef cattle **(Rivera *et al*., 2002)**. Vitamin E can increase the viscosity of phagocyte cell membranes, leading to improved phagocytosis of foreign bodies, and may also increase production of IgG **(St-Laurent *et al*., 1990)**. Furthermore, the protective effects of vitamin E on animal health may be involved with its role on reduction of glucocorticoids, which are known to be immunosuppressive **(Golub and Gershwin, 1985)**. Metabolites concentration in serum represents a buffering state for metabolic synthesis and catabolism end products (Swenson, 1984). It could also be noticed that changes in blood hematological and biochemical parameters, in the present study, were within the normal physiological range of sheep as previously documented **(Duncan and Prasse, 1986)**.

Table 3: Effects of vitamin A and vitamin E administration on serum biochemical constituents of growing Ossimi lambs (mean ± SEM).

	Treatments					
Parameters	**Control**	**VA**	**VE**	**VA+E**	**SEM**	**Sig.**
Glucose (mg/dl)	47.4	54.7	58.5	56.2	6.3	NS
Total protein (g/dl)	6.30 c	7.54 b	7.90 a	8.15 a	0.24	*
Albumin (g/dl)	3.10 b	3.05 b	3.88 a	3.85 a	0.13	*
Globulin (g/dl)	3.34 b	4.49 a	4.19 a	4.30 a	0.09	*
Cholesterol (mg/dl)	66.5	67.9	64.7	70.0	4.3	NS
AST (U/L)	99.2	111.5	101.8	104.6	8.7	NS

a,b,c means within the same row having different superscripts significantly different (* P<0.05). NS = not significant

VA = Vitamin A, VE = Vitamin E, VA+E = Vitamin A plus vitamin E.

AST = aspartate transaminase.

Table 4: Effects of vitamin A and vitamin E administration on immunoglobulin G and antioxidant enzymes activity of growing Ossimi lambs (mean ± SEM).

	Treatments					
Parameters	**Control**	**VA**	**VE**	**VA+E**	**SEM**	**Sig.**
IgG (µg/ml)	16.6 c	19.4 b	22.3 a	20.3 ab	1.3	*
TAC (mM/L)	0.620 b	0.833 a	0.777 a	0.755 a	0.043	*
SOD (U/ml)	230.0	236.7	231.9	240.0	4.60	NS
GSH-Px (mU/ml)	4.70 c	6.71 b	8.25 a	7.44 ab	0.618	*

a,b,c means within the same row having different superscripts significantly different (* P<0.05). NS = not significant

VA = Vitamin A, VE = Vitamin E, VA+E = Vitamin A plus vitamin E.

IgG= Immunoglobulin G, TAC= Total antioxidant capacity, SOD= Superoxide dismutase, GSH-Px = Glutathione peroxidise

Data showed a significant (P<0.05) increase in serum total antioxidant capacity (TAC) (P<0.05) by 34.4, 25.3 and 21.8 % for lambs received VA, VE and VA+E treatments *vs*. control, respectively (Table, 4). No significant differences in serum superoxide dismutase (SOD) activity noticed among the experimental treatment groups. There were significant (P<0.05) increases in serum glutathione peroxidase (GSH-Px) activity estimated by 42.8, 75.5 and 58.3 % for lambs received VA, VE and VA+E treatments *vs*. control respectively. Lambs received VE treatment had higher (P<0.05) serum GSH-Px (8.25 mU/ml) activity by 23.0 % than those received VA treatment (6.71 mU/ml). Studies focused that effect of vitamins E and A on serum antioxidant enzymes activities in sheep is limited. The present results agree with some reports on the antioxidant effects of vitamin A by **Burton and Ingold (1984)** and **Yang *et al.* (2010)**. In goats, dietary supplementation of vitamin A at level 2000-3000 IU/kg DM has been shown to increase (P<0.05) serum TAC and GSH-Px activities. However, the higher level of vitamin A at 5000 IU/kg DM had no effect on those enzyme activities in serum as reported by **Yang *et al.* (2010)**. They also found that, regardless of level of supplementation, vitamin A had no effect on serum SOD activity. The enhancing effect of vitamin A on increasing serum GSH-Px activity is likely due to the oxygen scavenging and lipo-peroxyl radical quenching function of vitamin A **(Stahl *et al*., 1997)**. Vitamin A acts as a powerful free radical scavenger and considers the most effective naturally occurring quencher of singlet oxygen and other free radicals **(Whittaker *et al*., 1996; Dugas *et al*., 1999)**. Regarding vitamin E, it has more potent effect to increase serum GSH-Px activity especially when compared to the effect of vitamin A (Table, 4). Vitamin E is a powerful antioxidant for body defense against oxidative stress **(Ibrahim *et al*., 1997)**. The enhancing effect of vitamin E on antioxidant enzymes activities has been shown in some investigations. In a study on goats, adding vitamin E (80 IU/ kid/d) can increase serum TAC and activities of serum SOD and GSH-Px **(Hong *et al*., 2010)**. In male buffalo calves, blood GSH-px activity but not SOD, was significantly increased in response to vitamin E supplementation **(Shinde *et al*., 2008)**. Hepatic SOD activity was not affected by vitamin E supplement, while GSH-px activity was significantly increased in rats. Enhanced GSH-px activity with vitamin E might aid hepatic enzymes to eliminate active oxygen in organs **(Jang *et al*., 1999)**. Vitamin E may have a controlling effect on oxidative stress through modulation IL-2 mRNA expression of SOD **(Das *et al*., 2012)**. As vitamin E dosage is concerned, both excess of dietary vitamin E and vitamin E deficiency can significantly depress the activity of hepatic and plasma GSH-px activity **(Yang *et al*., 1976)**. The protective effect of vitamin E on lipid peroxidation may not due to alteration of antioxidant enzyme activity but mainly mediated through its chain-breaking antioxidant enzyme activity **(Mantha *et al*., 1993)**.

In the current study, the significant increases in serum TAC and GSH-Px activities with combined the treatment of VA+E were comparable to that of VA or VE alone (Table, 4). It could be noticed that lambs received VA+E treatment had higher ($P<0.05$) serum GSH-Px activity by 58.3 % than control and tended to be higher by 10.9 % than lambs treated with VA. This observation could display a trend of possible beneficial effects of combined VA with VE at the level of antioxidant status. In this respect, in accordance to **Watts (1991)**, vitamin E may considered synergistic to vitamin A due to its antioxidant activity, which results in a sparing effect upon vitamin A. Likewise, loss of vitamin A from liver stores is accelerated in the presence of vitamin E deficiency as a result of increased fragility of lysosomal membranes. In addition, it has been found out that dietary supplementation of vitamin A markedly lowered *in vitro* lipid peroxidation and that vitamin E supplementation along with vitamin A still further reduced the *in vitro* lipid peroxidation of the tissues, suggesting that vitamin A also might be considered as a potential antioxidant similar to vitamin E in animal nutrition **(Kartha and Krishnamurthy, 1977)**.

CONCLUSION

Based on the present results, it could be concluded that vitamins A and E combination may exerted beneficial additive effect to improve ADG and physiological responses of growing lambs. Vitamin A is more effective than vitamin E in enhancing growth performance of lambs (FBW, ADG and FCE), whereas vitamin E had more potent effect that increase serum GSH-Px activity and IgG concentration thus improve their antioxidant status and immune response.

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CHANGES IN PRODUCTIVE PERFORMANCE, HEMATO-BIOCHEMICAL INDICES, IMMUNE AND ANTIOXIDANT STATUS OF GROWING OSSIMI LAMBS SUBJECTED TO VITAMINS A AND E ADMINISTRATION

الملخص العربى

التغيرات فى الأداء الإنتاجى، المؤشرات الهيماتولوجية والبيوكيميائية للدم وحالة المناعة ومضادات الأكسدة فى حملان الأوسيمى النامية نتيجة إعطاء فيتامين ه وفيتامين أ

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أستخدم فى هذه الدراسة عدد ٣٢ حمل أوسيمى نامية بمتوسط وزن ١٩.٠٨ ± ١.٠٩ كجم وثلاثة شهور ونصف من العمر وذلك بهدف تقييم تأثير إعطاء فيتامين ه وفيتامين أ على الأداء الإنتاجى، المؤشرات الهيماتولوجية والبيوكيميائية للدم ، حالة المناعة ومضادات الأكسدة. قسمت الحيوانات عشوائياً إلى أربعة مجموعات متساوية (٨ حملان في كل منها). المجموعة الأولى للمقارنة (كنترول)، المجموعة الثانية (VA) أعطيت فيتامين أ بمعدل ٥٠.٠٠٠ وحدة دولية/رأس/كل أسبوعين ، والمجموعة الثالثة (VE) أعطيت فيتامين ه بمعدل 400 ملجم /رأس/كل أسبوعين ، والمجموعة الرابعة (VA+E) أعطيت فيتامين أ بمعدل ٥٠.٠٠٠ وحدة دولية + فيتامين ه بمعدل 400 ملجم /رأس/كل أسبوعين.

وقد أظهرت النتائج ما يلى :- سجلت حملان المعاملات (VA) ، (VA+E) قيم أعلى معنوياً لمتوسطات وزن الجسم النهائى مقارنة بحملان الكنترول والمعاملة (VE). زادت متوسطات معدل الزيادة اليومية فى الوزن معنوياً لحملان المعاملة (VA) ، (VA+E) مقارنة بالكنترول وحملان المعاملة (VE). كما كان هناك زيادة معنوية فى متوسطات معدل الزيادة اليومية فى الوزن لحملان المعاملة المشتركة (VA+E) عند مقارنتها بحملان كل من المعاملة (VE) والمعاملة (VA) منفردتين. لم تلاحظ أية تغييرات معنوية في إجمالي استهلاك المادة الجافة لحملان المعاملات مقارنة بالكنترول ، بينما حدث تحسين معنوى (P <0.05) فى معدلات تحويل الغذاء حملان المعاملات (VA) ، (VA+E) مقارنة بحملان المعاملة (VE) والكنترول.

- أظهرت حملان المعاملات (VA) ،(VE)، (VA+E) زيادة معنوية فى تركيز هيموجلوبين الدم ، بينما زاد تركيز الهيموجلوبين وعدد كرات الدم الحمراء والمكونات الخلوية للدم لحملان المعاملات (VE) ، (VA+E) مقارنة بحملان بالكنترول. لم تلاحظ أية تغييرات معنوية في العدد الكلى لكريات الدم البيضاء وكذلك نسبة الكريات حمضية الصبغ ، والكريات قاعدية الصبغ والكريات الأحادية نتيجة للمعاملات بينما أظهرت حملان المعاملات (VA) ،(VE)، (VA+E) زيادة معنوية فى نسبة الكريات الليمفاوية مقارنة بالكنترول. أظهرت حملان المعاملة (VE) إنخفاض معنوى فى نسبة الكريات المتعادلة مقارنة بحملان الكنترول والمعاملات (VA) ، (VA+E).

- أظهرت حملان المعاملات (VA) ،(VE)، (VA+E) زيادة معنوية فى تركيزات السيرم من البروتين الكلي والجلوبيولين مقارنة بالكنترول ، بينما زادت معنوياً تركيزات السيرم من البروتين الكلي والألبيومين لحملان المعاملة المشتركة (VA+VE) مقارنة بحملان المعاملة (VA). أظهرت حملان المعاملات (VA) ،(VE)، (VA+E) زيادة معنوية فى تركيزات السيرم من الإمينوجلوبيولين (IgG) مقارنة بالكنترول فيما زاد تركيز السيرم من الإمينوجلوبيولين (IgG) فى حملان المعاملة (VE) مقارنة بحملان المعاملة (VA). كان هناك زيادة معنوية فى إجمالى القدرة المضادة للأكسدة (TAC) ونشاط إنزيم الجلوتاثيون بيروكسيديز (GSH-Px) فى السيرم لحملان المعاملات (VA) ،(VE)، (VA+E) مقارنة بالكنترول ، فيما زاد نشاط إنزيم (GSH-Px) فى السيرم معنوياً لحملان المعاملة (VE) مقارنة بحملان المعاملة (VA). لم تلاحظ أية تغييرات معنوية في تركيزات سيرم الدم من الجلوكوز ، الكوليستيرول وكذلك فى مستويات نشاط أنزيم (AST) ونشاط إنزيم (SOD) فى حملان المعاملات (VA) ،(VE)، (VA+E) مقارنة بالكنترول.

بناءَ على النتائج المقدمة، فإن إعطاء فيتامين ه وفيتامين أ للحملان النامية يمكن أن يؤدى إلى تأثيرات مفيدة مشتركة لهذه الفيتامينات لتحسين معدل الزيادة اليومية فى الوزن وكذلك الإستجابات الفسيولوجية لهذه الحملان. وقد كان فيتامين أ أكثر فاعلية لتعزيز أداء النمو بينما كان لفيتامين ه تأثير أكثر فاعلية لتعزيز الإستجابة المناعية وحالة مضادات الأكسدة.