EFFECT OF VITAMIN A SUPPLEMENTATION ON THE PERFORMANCE AND IMMUNE RESPONSE OF BANDARAH CHICKEN

By

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ABSTRACT: *Sixty four pullets and forty eight cockers (18 wks) from Bandarah local strain were used to investigate the effect of feeding different levels of vitamin A (VA), on layer performance and the immune maturation. Birds randomly chosen from farm flock reared on a conventional growing program from posthatch to 18 wks of age. Birds were vaccinated against Newcastle disease virus (NDV) by Hitchener B_1 at 7 days old, then Lasota vaccine at 18, 28 days old and at 18 wks old they were vaccinated by oil vaccine. Birds at 18 wks of age were weighed, randomly housed in individual cages and allotted to four experimental dietary isonitrogenous and isocaloric Corn-Soybean meal layer diet differed in its content of vitamin A added (0.0, 4000, 12000 and 24000 IU/ kg diet). Feed and water were offered for ad-libitium consumption from 18 to 48 wks of age. The results indicated that increasing VA supplementation increase significantly egg production. Egg weight, egg mass and feed utilization ratio were improved significantly during the later experimental periods by increasing VA supplementation. On the other hand, both of feed intake and body weight were insignificantly affected by VA supplementation at any levels of supplementation. Also, efficiency index of egg production estimated at the end of experimental, was improved significantly ($p< 0.01$) by increasing VA supplementation to layer diets. Supplementation of highest level of VA maximized the economic efficiency (EE) and relative economic efficiency (REE) by 0.40 and 114.3%, respectively, than others level of supplementations. Fertility and hatchability percentage was significantly ($p< 0.01$) increased by increasing of VA supplementation. While, late embryos died (unhitched embryos) percentage decrease significantly ($P< 0.01$) by increasing supplementation of VA. Supplementations of VA at others level had insignificant effect on posthatch chick weights.*

Although, VA supplementation increased significantly ($p< 0.01$) liver weight, at the end of the experimental period (48 wks), spleen, testis and intestinal weights significantly ($p< 0.01$) increased during all experimental periods as VA supplementation increased.

Additions of VA as adjuvant to Newcastle vaccine played an important role to improve the level of immunity, antibodies titer in both serum and yolk. Also, serum antibodies titer of posthatch chicks increased with increase the level of the antibodies titer in there mothers or/and in the eggs laid from layers fed high level of VA. So, we can depend on, and use yolk to measure the antibodies titers, as the immune level of the birds.

The histological study cleared that VA is somewhat safe in high level of 24000 IU/kg but the lowest levels lead to numerous adverse effects that may reflect on the hepatic and intestinal functions as well as on fertility of the affected cases. Also, the affection in spleen may be had adverse effect on the immunity and defensive mechanism of the body.

On conclusion, increasing supplementation of VA up to 24000 IU/kg in layer diets is more safety and increasing the productive and reproductive performance, the immunity of the birds and the economical efficiency.

INTRODUCTION

Vitamin A is a fat-soluble vitamin and essential for many functions such as in growth, development of lymphoid tissues, immune response, the differentiation of epithelial cells, used in the visual cycle, support the viability of the reproductive system and proliferation (Friedman and Sklan, 1989a,b; Friedman *et al.*, 1991; Goss and McBurney, 1992; Bollag, 1996; Squires and Naber, 1993; Brody, 1993; Gerster, 1996, Coskun *et al.*, 1998 and Guven *et al.* 2007). Sklan *et al.* (1994) reported that although the NRC (1994) has provided a recommendation for adequate levels of dietary vitamin A, adequate levels for maximal immune responses of chicken need to be further evaluated. Surai *et al.* (1998) and Lin *et al.* (2002) observed that increasing rate of vitamin A supplementation to layer diets was beneficial to laying performance and immune function, also, markedly increased concentration of vitamin A in the yolk of the hens' eggs and the concentration of vitamin A in the maternal liver was found to be greatly. On the other hand, Coskun *et al.* (1998) indicated that increasing rate of vitamin A supplementation to layer diets had no significant effects on layer performance or immune system under normal conditions.

Egg yolk of avian species contains a wide variety of antibodies raised naturally or by immunization with suitable immunogenic (Olson *et al.*, 1980). Sijtsma *et al.* (1989a, b) reported that the birds marginally vitamin A deficient

had higher severity of disease following experimental Newcastle disease virus (NDV) infection. Egg yolk can be used for diagnosis of various diseases of poultry live stock (Arshad *et al.,* 1996). Catharine Ross (2007) reported that vitamin A (VA, retinol) is essential for normal immune system maturation, but the effect of VA on antibody production, the hallmark of successful vaccination, is still not well understood.

The experiment was designed to investigate the effects of feeding different levels of VA supplementations on layer performance and the immune maturation. Also, the possibility of use yolk NDV antibodies titer levels as indicator to the successful of parents vaccinations against NDV.

MATERIAL AND METHODS

The current study was conducted at El-Sabhia Poultry Research Station, Animal Production Research Institute, Agricultural Research Center. A total of 64 pullets and 48 cockers, eighteen-wks-old, was randomly chosen from the Bandarah farm flock, which was reared on a conventional growing program from posthatch to 18 wks of age. Birds were vaccinated against Newcastle disease virus (NDV) by Hitchener B_1 at 7 days old, then Lasota vaccine at 18, 28 days old and at 18 wks old they were vaccinated by oil vaccine. Birds were weighed and randomly housed in individual cages and allotted to four experimental dietary isonitrogenous and isocaloric Corn-Soybean meal layer diet, of 16 pullets and 12 chokers each. Experimental diets differed in its content of minerals and vitamin premix of vitamin A supplementation (Table1), feed and water were offered for *ad- libitium* consumption from 18 to 48 wks of age.

Experimental dietary treatments :-

Treatment 1: basal diet without added VA (zero dose, ZVA).

Treatment 2: basal diet + 4000 IU of VA /kg diet (low dose, LVA).

Treatment 3: basal diet +12000 IU of VA /kg diet (commercial dose, CVA).

Treatment 4: basal diet + 24000 IU of VA /kg diet (High dose, HVA).

Body weight (BW) was recorded at 20 wks of age and at the end of experimental period (48 wks of age). Feed intake (FI), egg production (EP), egg weight (EW) were recorded, while feed utilization ratio and egg mass (EM) were calculated every 4 wks intervals throughout the entire experimental period. At the end of experimental, economic efficiency (EE) and relative economic efficiency (REE) were calculated according to input-output analysis data. Also, the efficiency index (EI) of egg production was estimated using the equation suggested by Byerly (1941).

The equation is:- $F1 = (3.56\ W + \Delta\ W + X\ E) / 100.$

Where:-

F1 = calculated food/ bird/day (g). W= mean live-weight of bird (g).

Δ W= live-weight change/100 bird/day (g). X= average egg weight (g).

E= number of egg/100 birds/day.

To get the efficiency index (EI), the calculated food/hen/day (g) was divided by F2 which is the average of observed food consumption/hen/day (g) obtained from the experimental feeding records.

Serum samples were collected from each treatment every 4 wks intervals throughout the entire experimental period and stored at -20 ^{0}C for analyses. At 39 wks, artificial insemination was applied, fertile eggs laid during the 40 wks of age, were collected to estimate egg fertility and hatchability percentages. Hatched chicks were weighed, and then serum samples were collected.

Determination of NDV antibodies titer:

Serum samples from layers at 24, 28, 32, 36 and 40wks of age and from posthatch chicks were used for determination NDV antibodies titer, using methods described by(Liu, 1999), also, yolk samples at 28, 32, 36 and 40 wks of age were used for determination the same analysis.

Carcass organ traits:

At 24, 36 and 48 wks of age 3 males from each treatment were randomly selected and slaughtered for carcass evaluation. Carcass was eviscerated and head and shank were removed, liver, spleen, testis and intestine were dissected from the viscera and weighed. Each portion was expressed as a percentage of life body weight.

Histopatholgic studies:

The collected specimens of the liver, spleen, testis and intestine were fixed in 10% neutral buffered formalin solution, then after washing in tap water passed- through the routine paraffin embedding technique (dehydration in the ascending grades of ethyl alcohol, followed by cleaning in 3 steps of xylol followed by embedding in three changes meted paraffin wax). Paraffin- block of three specimens was prepared then sections of about 5 microns thick were cut and picked on microscopic glass-slide. After dryness, sections were stained with hematoxylin and easin according to the methods of Culling (1983) and then subjected to the light microscopy.

All results were statistically analyzed by General Linear Models (GLM), one way analysis of variance, using SAS software (SAS Institute, 1998). Differences among means were separated using Duncan's multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

Productive Performance:-

Results presented in Table 2 indicated that supplementation of VA at the first periods (24-28 and 28-32wks of age) had insignificant effects on EP at any supplementation levels. Throughout the later experimental periods (32-36 and 36-40, 40-44 and 44-48 wks of age) and for overall main, supplementation of VA had significant ($p< 0.01$) effect on EP. Generally, increasing of VA supplementation significantly increased EP and the highest EP recorded for the birds fed diet supplemented with HVA and CVA. These results are in agreement with the results of Squires and Naber (1993) who observed that egg production of hens fed the unsupplemented diet with vitamin A significantly declined after12 wks. Also, Lin *et al.* (2002) showed that increasing vitamin A supplementation (3000 to 9000 IU/kg) to layer diets had a beneficial effect on rate of laying. On the other hand, Coskun *et al.* (1998) indicated that VA supplementation (0, 4000, 12000 and 24000 IU/kg) to layer diets had no significant effect on egg yield.

Supplementation of VA had insignificant effects on EW (Table, 3) at any levels of supplementation through the first periods (24-28, 28-32, 32-36 and 36-40 wks). While, during the later experimental periods (40-44 and 44-48 wks of age) increasing of VA supplementation significantly ($p< 0.01$) increased EW. However, the overall mean of EW was insignificantly affected by supplementation levels of VA. Egg mass results (Table, 4) tend the same results of EP during the experimental periods and the best results were recorded for the birds fed diet supplemented with HVA and CVA throughout the later experimental periods (32-36 and 36-40, 40-44 and 44-48 wks of age). Also, the overall mean of EM was significantly increased by increasing supplementation of VA, since the average of EW and EP were increased as VA supplementation increased. The same results were recorded by Squires and Naber (1993) they observed that egg weight was consistently changed by vitamin A supplemented. Also, Lin *et al.* (2002) showed that egg weight was significantly increased by the high levels of vitamin A supplementation (6000, 9000 IU/kg) to layer diets.

Feed intake was insignificantly affected by supplementation of VA at any levels during the different periods (Table, 5). These results are in agreement with result of Lin *et al.* (2002) they showed that feed intake was insignificantly affected by vitamin A supplementation to layer diets. Feed utilization ratio during the first periods of the study (24-28 and 28-32wks of age) was

insignificantly affected by VA supplementation at any levels (Table, 6). Increasing supplementation of VA improved significantly ($p< 0.01$) feed utilization ratio during the later periods (32-36 and 36-40, 40-44 and 44-48 wks of age) and for the overall mean of feed utilization ratio. Generally, the best feed utilization ratio recorded for the birds fed diet supplemented with HVA and CVA.

Results in Table (7) indicated that BW at the end of experiment was insignificantly affected by supplementation of VA at any levels. Lin *et al.* (2002) reported that BW was insignificantly affected by vitamin A supplementation to layer diets. Results of efficiency index (EI) were improved significantly ($p< 0.01$) by increasing vitamin A supplementation to layer diets, and the highest EI (74.72%) was recoded for the layers fed HVA (Table, 7).

Fertility percentage was significantly ($p< 0.01$) affected by supplementation of VA and the best percentage (100.0%) was recorded for the layer fed CVA and LVA Table (7). That result indicated that VA supplementation at 4000 and 12000 IU/kg diet was sufficient for the best fertility percentage. Late embryos died (unhitched embryos) percentage decrease significantly ($p< 0.01$) by increasing supplementation of VA and the best percentage (2.630%) was recorded for the layer fed HVA. On the other hand, hatchability percentage increased significantly ($p< 0.01$) by increasing supplementation of VA and the best percentage (94.1%) was recorded for the layer fed HVA. Supplementations of VA at any levels had insignificant effect on posthatch chick weights and were ranged between 37.4 and 38.7 g. Squires and Naber (1993) indicated that hatchability of eggs laid by the hens fed the unsupplemented diet with vitamin A was significantly lower at wks 25and 26 than hatchability for those hens receiving dietary vitamin A, while chick weight was not consistently affected by vitamin A supplementation. Coskun *et al.* (1998); Surai *et al.* (1998), Lin *et al.* (2002) and Guven *et al.* (2007) observed that increasing rate of vitamin A supplementation to layer diets was beneficial to laying performance.

The relative economic efficiency (REE) and the economic efficiency (EE) of different formulated diets are shown in Table 8. The results indicated that increasing the level of VA supplementation to layer diets increase the REE and EE. Supplementation of HVA maximized the REE and EE (0.40 and 114.3 %, respectively) than any level of VA supplementations.

Results of some carcass traits are represented in (Table 9). Liver weight at 26 and 34 wks of age were insignificantly affected by VA supplementation. While, increasing VA supplementation in layer diets increased significantly ($p< 0.01$) liver weight, at the end of experiment (48 wks). The increase of liver

weight may be due to VA stored in the liver. So after many wks of experiment livers of the birds fed lower dose of VA are more depletion of VA as compare to those fed highest dose. Also, the histological structure of liver showed that in the case of LVA as well as the cases of ZVA supplementation diffuse changes of moderate to severe degenerations of vacuolar and hydropic changes in most of all the hepatic cells (Fig. 1c and 1d), these results are indication of liver function degeneration and cells damage.

Data of intestinal, testes and spleen weights (Table, 9) indicated that as VA supplementation increased, in the layer diet, their weights increased during the all experimental periods. Spleen is one of the important organs in the living organism, since produce many of immunity cells (eg. T lymphocytes). These results are compatible with the results of serum NDV antibodies titer (Table 9). Also, the histological structure of intestinal, testis and spleen (Fig. 2, 3 and 4) showed active parameters of well developed lymph follicles and vasculatures of spleen, highly active and intact covering epithelium of the intestinal villi and highly active of all the semineferous tubules of testis. Our results are agreement with the results of many investigators, Surai *et al.* (1998) and Lin *et al.* (2002) indicated that increasing rate of vitamin A supplementation to layer diets was beneficial to immune function. Also, Friedman *et al.* (1991); Squires and Naber (1993) and Coskun *et al.* (1998) observed that vitamin A supplementation has a rule of development of lymphoid tissues, immune response, the differentiation of epithelial cells and support the viability of the reproductive system and proliferation. Surai *et al.* (1998) reported that after 3 months, the concentration of vitamin A in the maternal liver was found to be greatly enhanced in proportion to the increasing rates of supplementation with vitamin A. Also, Richter *et al.* (1989) demonstrated that there is a positive relation between the vitamin A supply of hens and the vitamin A content of the liver.

NDV antibodies titer:

The data represented in Tables (10) and (11) showed that addition of HVA gave the highest serum and yolk NDV antibodies titer as compare to the other levels of VA supplementation, through the experimental periods studies (24, 28, 32, 36 and 40 wks of age). The serum antibodies titer was 8.4, 8.5, 9.7, 10.0 and 10.3 with HVA, while it was 7.5, 8.0, 9.0, 9.2 and 9.5 with ZVA during the afromention periods, respectively. On the other hand, yolk antibodies titer was 8.7, 9.5, 10.2 and 10.3 with HVA, while it was 7.9, 8.7, 9.1 and 9.6, respectively, during 28, 32, 36 and 40 wks of age. So the results indicated that addition of VA as adjuvant to Newcastle vaccine play an important role to improve the level of, immunity, antibodies titer in serum and yolk. Also, serum antibodies titer of posthatch chicks increased with increase the level of the antibodies titer in there mothers or/and in the eggs laid from layers fed high

level of VA. However, these results indicated that may be we can depend on, and use yolk to measure the antibodies titers, as the immune level of the birds, since the collection of eggs are more easily than collection of serum, also were save for birds. Results of Sijtsma *et al.* (1989a, b) revealed that increased morbidity is observed in chickens experimentally infected NAD that was fed a diet marginally deficient in vitamin A. Also, Lin *et al.* (2002) observed that increasing rate of vitamin A supplementation to layer diets had a significant effect on NAD antibody titer.

Histopatholgic:-

The hepatic tissue in case of supplement with HVA showed no adverse effect where the hepatic acinose showed normal and intact cells (Fig. 1a), while in case of CVA some small vacualations of mild degenerative changes (Fig. 1b) were seen in some of the hepatic cells. On the other hand, LVA as well as the cases of ZVA showed diffuse changes of moderate to severe degenerations of vacuolar and hydropic changes in most of all the hepatic cells (Fig. 1c and 1d).

The intestinal mucosa in case of HVA showed highly active and intact covering epithelium of the intestinal villi (Fig. 2a) with somewhat thickened villi by edema and cellular reactions. The covering epithelium of the initial villi appeared with excess vacualations after the administration with CVA (Fig. 2b). The intestinal villi following administration with LVA, showed damaged desquamated covering epithelium (Fig. 2c), while in cases of ZVA complete necrosis of most of all the intestinal villi (Fig. 2d) was seen.

The testis following administration with HVA appeared highly active with normal spermatogenesis where most of all the semineferous tubules appeared bluged with mature spermatazooal contents (Fig. 3a). Some groups of immature stages of spermatagonial cells appeared with the Lumina of some semineferous tubules (Fig. 3b), in the testis of cases CVA. The testis in cases of LVA, showed inactive spermatogenesis with less mature spermatagonial cells and excess of multinucleated spermatocytic giant cells (Fig. 3c). The semineferous tubules in cases of ZVA appeared with damaged spermatagonial cells, less or no spermatazooal contents with their lumina appeared filled with an excess of necrotic debris (Fig. 3d).

In cases of HVA, spleen appeared with normal and active parameters of well developed lymph follicles and vasculatures (Fig. 4a). Spleen in cases of CVA, showed less activity, where few numbers of mature or well developed lymph follicles were seen in addition to the appearance of some immature newly formed vasculatures and germinal centers (Fig. 4b). In cases of LVA, excess numbers of small newly formed vasculature and follicles appeared mixed with some other dispersed lymphocytic elements (Fig. 4c). The spleen in

cases of ZVA showed congestion with expansion of the germinal center of the present lymph follicles (Fig. 4d).

The obtained results for the microscopic changes revealed occurrence of a variable common histological change in the epithelial elements in hepatic cells, covering epithelium of the intestinal villi as well as spermatogenic cells of testis. These detected changes were correlated with the levels of administration as the HVA lead to presence of nearly normal and active histological parameters, while the decreases in the levels of supplement with VA lead to the appearance of some degenerative changes and less active histological parameters in these organs. These adverse effects reached its peak in case of ZVA, since, sever degeneration and necrosis was detected in the hepatic cells, covering epithelium of the intestinal villi and spermatogenic and spermatagonial cells of the semineferous tubules. On the other hand, splenic changes were also indicative for the occurrence of some adverse effects related to the low levels of VA, on structure and histological parameters of the spleen. Coskun *et al.* (1998) reported that lymphoid organs were normal in histological structure for chickens fed diets based on white, corn and soybean meal or sunflower oil meal supplemented and unsupplemented with vitamin A.

The histological study showed that VA is somewhat safe and had good results at the highest level (24000IU/kg diet) but the lowest levels lead to numerous adverse effects that may reflect on the hepatic and intestinal functions as well as on fertility of the affected cases. Also, the affection in spleen may be had adverse effect on the immunity and defensive mechanism of the body.

On conclusion, increasing supplementation of VA up to 24000 IU/kg in layer diets is more safety and increasing the productive and reproductive performance and the immunity of the birds.

Table (1): Composition* and the nutritive value of the basal diets.

Feed stuffs	%	Energy and nutrient content	
Yellow corn (8.5%CP.)	630.0	ME (kcal/kg)	2728
Soybean meal (43% CP)	270.0	Crude protein (%calculated)	16.97
Di-Calciume phosphate	15.0	Ca %	3.0
Limestone	76.0	P % (available).	0.41
Premix**	3.0	Fiber (%)	2.88
Dl-Methionine	1.0	Ether extract (%)	2.70
NaCl	5.0	Methionine	0.35
Total	1000.0	TSAA (%)	0.64
Cost / Ton (L.E.)	1980	Lysine	0.68

*As recommendation of Anim. Prod. Res. Inst., Agric Res. Center, Minis of Agric,

** four premix were used for each treatments separately and its composition of 3 kg is similarly in: Vit D_3 2,000,000; Vit E 10,000 mg, Vit K_3 1,000 mg, Vit B_1 1,000 mg, Vit B_2 4,000 mg, Vit B_6 1,500 mg, Vit B_{12} 10 mg; Niacin 20,000 mg; Pantotenic acid 10,000 mg, Folic acid 1,000 mg, Biotin 50 mg, Choline chloride 500, 000 mg, Cu 3,000 mg, Iodine 300 mg, Fe 30,000 mg; Mn 40,000 mg, Zn 45,000 mg, Selenium 100 mg. Each premix were defer in its content of vitamin A, which was supplied by Zero, 4000000, 12000000 24000000IU, respectively.

Table (2) Effect of VA levels on egg production (Egg/hen/day)during experimental periods

Age (wks)	VA supplementation (1000 IU/kg diet)				Significant
	24	12	4	zero	
24-27	0.65±0.11	0.67±0.06	0.58±0.06	0.65±0.09	NS
28-31	0.69±0.06	0.69±0.08	0.66±0.05	0.70±0.05	NS
32-35	0.64 ± 0.02^{A}	0.64 ± 0.02^{A}	0.59 ± 0.03^{B}	0.44 ± 0.06^{C}	**
36-39	0.66 ± 0.01^{A}	0.61 ± 0.01^{AB}	0.57 ± 0.01^{B}	0.58 ± 0.01^{B}	**
40-43	0.61 ± 0.01^{A}	0.61 ± 0.01^{A}	0.55 ± 0.01^{B}	0.55 ± 0.01^{B}	**
44-48	0.66 ± 0.02^{A}	0.68 ± 0.01^{A}	0.66 ± 0.02^{A}	0.60 ± 0.01^{B}	**
overall	0.65 ± 0.02^{A}	0.65 ± 0.02^{A}	0.60 ± 0.01^{B}	0.59 ± 0.02^{B}	**

Means within the same row with different superscript are significantly different
NS = not significant *** = Significantly at 0.01.*

Table(3) Effect of VA on egg weight (g)during experimental periods

Age (wks)	VA supplementation (1000 IU/kg diet)				Significant
	24	12	4	zero	
24-27	43.9±0.8	47.6±1.1	46.5±0.9	45.7±2.0	NS
28-31	45.9±0.7	49.1±1.1	46.0±0.6	47.6±1.7	NS
32-35	48.4±1.5	49.9±0.6	49.6±0.1	49.6±1.6	NS
36-39	50.6±0.3	51.3±0.4	51.6±0.1	51.2±0.5	NS
40-43	52.6 ± 1.2^{A}	57.7 ± 0.3^{A}	49.4 ± 0.8^{B}	50.3 ± 0.9^{B}	**
44-48	52.5 ± 0.4^{A}	52.6 ± 0.4^{A}	50.8 ± 04^{B}	50.9 ± 0.7^{B}	**
overall	49.0±0.4	50.6±0.3	48.9±0.3	49.2±0.7	NS

Means within the same row with different superscript are significantly different
NS = not significant *** = Significantly at 0.01.*

Table(4) Effect of VA levels on egg mass (g/hen/day)during experimental periods

Age (wks)	VA supplementation (1000 IU/kg diet)				Significant
	24	12	4	zero	
24-27	28.5±1.2	32.2±1.7	26.7±2.2	29.7±1.8	NS
28-31	31.5±1.4	33.4±1.2	30.2±2.2	33.2±1.7	NS
32-35	30.9±0.9 ^{A}	31.9±0.4 ^{A}	29.2±1.5 ^{A}	21.9±1.1 ^{B}	**
36-39	33.3±0.2 ^{A}	31.5±1.7 ^{B}	29.2±0.2 ^{B}	29.9±0.7 ^{B}	**
40-43	32.3±1.3 ^{A}	32.3±0.3 ^{A}	27.0±1.0 ^{B}	27.4±0.6 ^{C}	**
44-48	34.7±0.9 ^{A}	35.8±0.5 ^{A}	33.3±1.1 ^{B}	30.5±0.4 ^{C}	**
overall	37.1±0.8 ^{A}	32.8±1.1 ^{AB}	29.3±0.4 ^{B}	28.8±1.1 ^{B}	**

Means within the same row with different superscript are significantly different
NS = not significant *** = Significantly at 0.01.*

Table(5) Effect of VA levels on feed intake(g/hen/day)during experimental periods

Age (wks)	VA supplementation (1000 IU/kg diet)				Significant
	24	12	4	zero	
24-27	121.8±7.5	124.9±0.9	121.8±7.4	128.0±6.9	NS
28-31	112.1±8.7	132.8±11.1	123.5±4.2	133.8±10.7	NS
32-35	110.5±3.3	117.1±1.7	113.1±3.1	110.7±4.7	NS
36-39	112.6±2.3	111.8±0.9	110.2±0.7	110.7±4.7	NS
40-43	121.1±0.3	119.7±0.4	120.2±0.7	118.9±0.3	NS
44-48	117.1±0.6	119.6±0.6	120.1±0.5	119.0±0.3	NS
overall	115.9±2.9	120.7±2.1	118.1±2.1	120.9±2.6	NS

Means within the same row with different superscript are significantly different
NS = not significant

Table(6) Effect of VA levels on feed utilization ratio(kg feed/kg egg) during experimental periods

Age (wks)	VA supplementation (1000 IU/kg diet)				Significant
	24	12	4	zero	
24-27	4.58±0.8	4.01±0.4	4.65±0.4	6.31±4.0	NS
28-31	3.66±0.4	4.00±0.3	4.15±0.3	4.09±0.3	NS
32-35	3.59±0.2 ^{D}	3.67±0.1 ^{C}	3.91±0.3 ^{A}	5.80±1.0 ^{B}	**
36-39	3.38±0.1 ^{C}	3.71±0.4 ^{AB}	3.77±0.1 ^{A}	3.72±0.2 ^{AB}	**
40-43	3.76±0.0 ^{B}	3.68±0.1 ^{B}	4.47±0.2 ^{A}	4.33±0.1 ^{A}	**
44-48	3.38±0.1 ^{C}	3.33±0.1 ^{C}	3.62±0.1 ^{B}	3.88±0.1 ^{A}	**
overall	3.72±0.2 ^{C}	3.73±0.1 ^{C}	4.09±0.1 ^{B}	4.69±0.6 ^{A}	**

Means within the same row with different superscript are significantly different
NS = not significant *** = Significantly at 0.01.*

Table(7) Effect of VA levels on hen body weight(g), chicks weight (g), efficiency index of egg production(%) and some hatchability treats

Parameters	VA supplementation (1000 IU/kg diet)				Sig
	24	12	4	zero	
Initial body weight(g)	1478.5±65.6	1503.8±21.9	1520.5±51.9	1440.9±60.9	NS
Final body weight(g)	1567.5±68.6	1567.5±33.8	1566.3±36.6	1618.7±90.5	NS
Efficiency index (%)	74.72 ± 0.4^{A}	73.32 ± 0.5^{A}	71.60 ± 0.3^{B}	69.93 ± 0.6^{B}	**
Fertility (%)	97.8 ± 1.2^{B}	100.0 ± 0.0^{A}	100.0 ± 0.0^{A}	97.7 ± 1.2^{B}	**
Late embryos died (%)	2.63 ± 1.5^{B}	7.88 ± 2.2^{A}	7.08 ± 1.7^{A}	6.44 ± 0.9^{A}	**
Hatchability (%)	94.1 ± 0.9^{A}	92.1 ± 2.2^{B}	92.9 ± 1.6^{B}	91.29 ± 0.2^{B}	*
Posthatch chicks weights(g)	37.4±0.6	38.7±1.6	37.7±1.5	37.4±0.8	NS

Means within the same row with different superscript are significantly different
*NS = not significant * = Significantly at 0.05 ** = Significantly at 0.01.*

Table (8): Economic efficiency as affected by graded levels of VA at the end of experimental period.

VA.in diet	Egg production Egg/hen/day (24-48wk)	Egg price(1) (L.E)	Total feed consumption (24-48wk)kg	Total feed cost (2) (L.E)	Net return (3)	EE (4)	REE (5) %
HVA	0.65	54.6	19.471	38.94	15.66	0.40	114.30
CVA	0.65	54.6	20.278	40.56	14.04	0.35	100.00
LVA	0.60	50.4	19.841	39.68	10.72	0.27	77.14
ZVA	0.59	49.56	20.311	40.62	8.97	0.22	62.86

1-Price of 30 eggs = 15 L.E. 2-Total cost = Total feed intake x Feed cost(2.0 LE/kg diet).
3-Net return = Egg price – Total cost. 4-Economic Efficiency = Net return / Total cost
5-Relative Economic Efficiency (REE).

Table (9) Effect of VA levels on some carcass organs (g/100 BW) during experimental periods

organ	Age (wk)	VA supplementation (1000 IU/kg diet)				Sig.
		24	12	4	zero	
Liver	24	1.33±0.1	1.36±0.1	1.39±0.2	1.40±0.1	NS
	34	1.39±0.04	1.40±0.04	1.35±0.05	1.33±0.05	NS
	48	1.69 ± 0.11^{A}	1.66 ± 0.12^{A}	1.37 ± 0.10^{B}	1.28 ± 0.11^{B}	**
Intestinal	24	5.26 ± 0.39^{A}	4.78 ± 0.41^{B}	4.71 ± 0.42^{B}	4.70 ± 0.40^{B}	**
	34	4.09 ± 0.05^{A}	4.29 ± 0.04^{A}	3.30 ± 0.04^{B}	3.27 ± 0.05^{B}	**
	48	4.33 ± 0.12^{A}	4.59 ± 0.11^{A}	3.90 ± 0.11^{B}	3.87 ± 0.12^{B}	**
Spleen	24	0.19 ± 0.01^{A}	0.14 ± 0.01^{B}	0.14 ± 0.01^{B}	0.15 ± 0.01^{B}	**
	34	0.17 ± 0.01^{A}	0.13 ± 0.01^{A}	0.10 ± 0.1^{B}	$0.09 \pm .01^{B}$	**
	48	0.15 ± 0.01^{A}	0.16 ± 0.01^{A}	0.12 ± 0.01^{B}	0.06 ± 0.01^{C}	*
Testis	24	1.94 ± 0.01^{A}	1.76 ± 0.01^{B}	1.24 ± 0.01^{C}	1.77 ± 0.02^{B}	**
	34	2.19 ± 0.02^{A}	1.86 ± 0.02^{B}	1.48 ± 0.01^{C}	0.94 ± 0.02^{D}	**
	48	1.83 ± 0.2^{A}	1.68 ± 0.3^{B}	1.38 ± 0.2^{C}	0.89 ± 0.3^{D}	**

Means within the same row with different superscript are significantly different
*NS = not significant * = Significantly at 0.05 ** = Significantly at 0.01.*

Table (10) Effect of VA levels on serum NSD titer of hens and posthatch chicks .

Biard	Age (wks)	VA supplementation(1000 IU/kg diet)				Sig.
		24	12	4	zero	
Hen	24	8.40 ± 002^{A}	8.00 ± 0.01^{B}	7.90 ± 0.01^{C}	7.50 ± 0.01^{D}	**
Hen	28	8.50±0.01	8.30±0.01	8.20±0.01	8.00±0.01	NS
Hen	32	9.70 ± 0.01^{D}	9.50 ± 0.01^{A}	9.20 ± 0.01^{B}	9.00 ± 0.01^{C}	**
Hen	36	10.00 ± 0.01^{A}	9.80 ± 0.01^{B}	9.50 ± 0.01^{C}	9.20 ± 0.01^{D}	*
Hen	40	10.30 ± 0.01^{A}	10.00 ± 0.01^{B}	9.70 ± 0.01^{C}	9.50 ± 0.01^{C}	*
Chicks	1 day	6.50 ± 0.01^{A}	6.30 ± 0.01^{B}	6.00 ± 0.01^{C}	6.00 ± 0.01^{C}	*

Means within the same row with different superscript are significantly different
*NS = not significant * = Significantly at 0.05 ** = Significantly at 0.01.*

Table (11) Effect of VA levels on egg NSD titer

Age (wks)	VA supplementation (1000 IU/kg diet)				Sig
	24	12	4	zero	
28	8.70 ± 0.01^{A}	8.50 ± 0.01^{B}	8.00 ± 0.01^{C}	7.90 ± 0.01^{C}	*
32	9.50 ± 0.01^{A}	9.20 ± 0.01^{B}	8.90 ± 0.01^{C}	8.70 ± 0.01^{C}	*
36	10.20 ± 0.01^{A}	9.80 ± 0.01^{B}	9.50 ± 0.01^{C}	9.10 ± 0.01^{D}	**
40	10.30 ± 0.01^{A}	10.10 ± 0.01^{A}	9.80 ± 0.01^{B}	9.60 ± 0.01^{B}	*

Means within the same row with different superscript are significantly different
** = Significantly at 0.05 ** = Significantly at 0.01.*

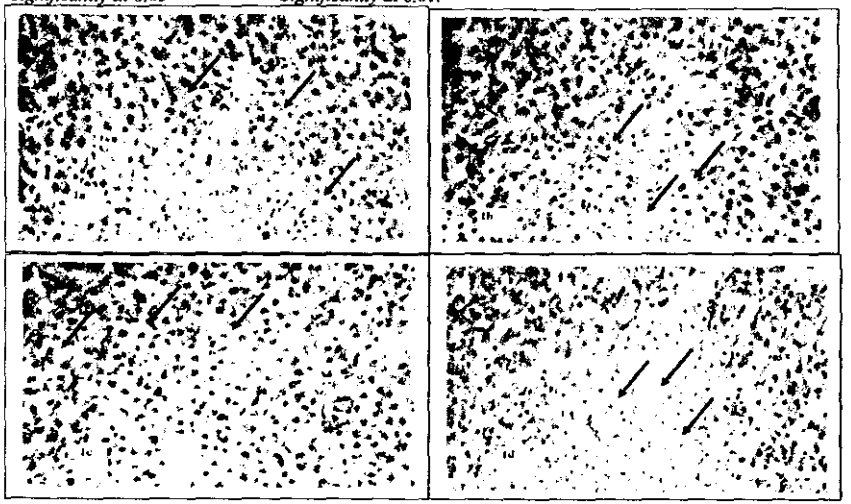

Fig. 1a: liver (HVA supplementation), showing normal hepatic acinose (arrows) with intact cell. (H. and E. X 250).

Fig.1b: liver (CVA supplementation), showing mild affection of hepatic cells by tiny or small vacillations (arrows). (H. and E. X 250).

Fig.1c: liver (LVA supplementation), showing moderate affection of hepatic cells with vacuolar degeneration (arrows). (H. and E. X 250).

Fig.1d: liver (ZVA supplementation), showing moderate affection of hepatic cells with vacuolar degeneration (arrows). (H. and E. X 250).

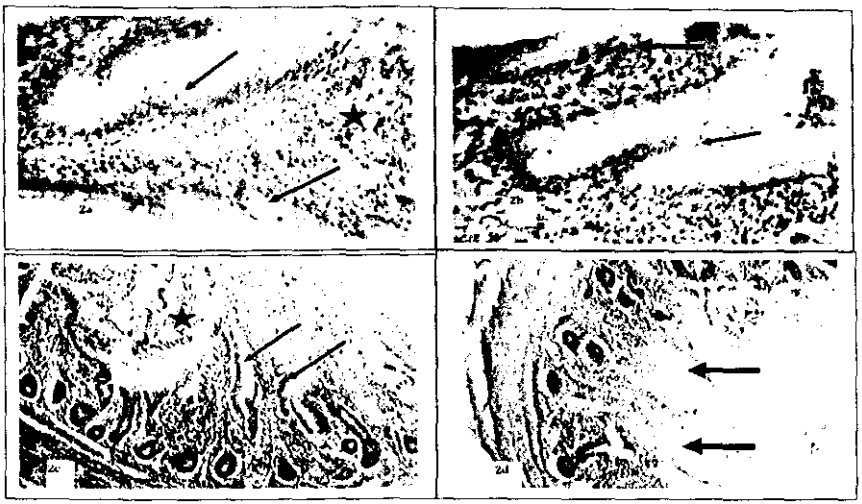

Fig. 2a: Intestine (HVA supplementation), showing intact covering epithelium of villi (arrows) with thickening by an excess of cellular aggregations (black asterisk). (H. and E. X250).

Fig.2b: Intestine (CVA supplementation), showing persistent and intact covering epithelium of villi but with an excess of cellular vacualations (arrows). (H. and E. X 400).

Fig.2c: Intestine (LVA supplementation), showing degenerated (arrows) and desquamated (black asterisk) covering epithelium. (H. and E. X 250).

Fig.2d: Intestine (ZVA supplementation), showing complete necrosis in most of all the intestinal villi (arrows). (H. and E. X 250).

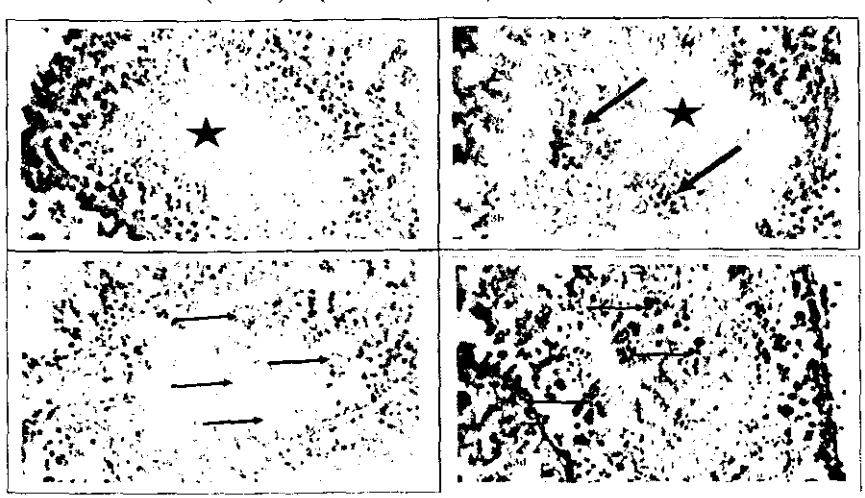

Fig. 3a: Testis (HVA supplementation), showing normal spermatogenesis with an excess of mature spermatazooal contents (black asterisk) bluged the lumen of semineferous tubule. (H. and E. X400).

Fig.3b: Testis (CVA supplementation), showing one semineferous tubule bluged with excess of mature spermatazooal contents (black asterisk) mixed with other groups of immature spermatagonial cells (arrows). (H. and E. X 400).

Fig.3c: Testis (LVA supplementation), showing less spermatogenesis with numerous multinucleated spermatocytic giant cells (arrows) inside the lumen of one semineferous tubule. (H. and E. X 400).

Fig.3d: Testis (ZVA supplementation), showing damaged semineferous tubule with an excess of necrosis spermatazooal cells (arrows). (H. and E. X 400).

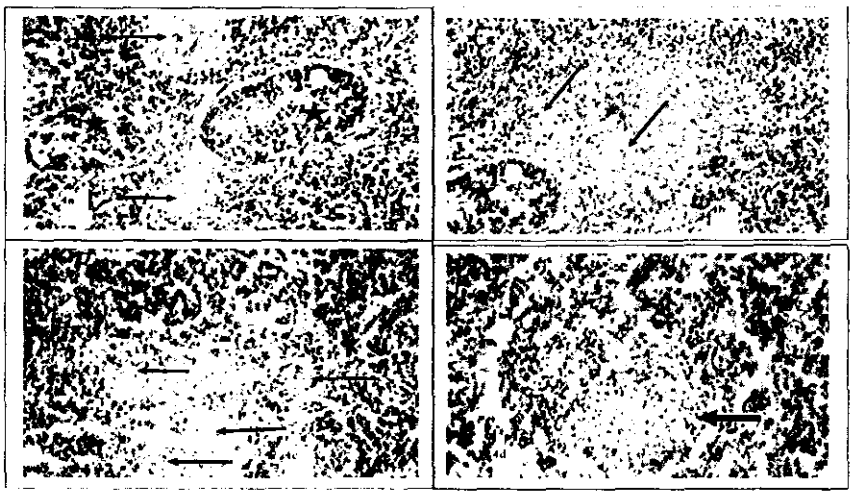

Fig. 4a: Spleen (HVA supplementation), showing two well developed lymph follicles (black asterisk) in addition to thick walled arterioles (arrows). (H. and E. X400).

Fig.4b: Spleen (CVA supplementation), showing one of the well developed lymph follicles (black asterisk) with other newly formed germinal centers (arrows). (H. and E. X400).

Fig.4c: Spleen (LVA supplementation), showing excess of small ill-developed germinal centers (arrows) with surrounding area of dispersed lymphocytic elements (white asterisk). (H. and E. X400).

Fig.4d: Spleen (ZVA supplementation), showing one ill-developed lymph follicle with expanded germinal center (arrows) surrounded by areas of congestion (white asterisk). (H. and E. X400).

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الملخص العربي

تاثير اضافة فيتامين أ على الأداء الانتاجى والاستجابة المناعية لدجاج البندرة

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أستخدم عدد ٦٤ دجاجة و٤٨ ديك من سلالة البندرة المحلية ، لدراسة تأثير التغذية على عدد من مستويات فيتامين أ وذلك على صفات الأداء والجهاز المناعى.

تم اختيار الطيور عشوائيا من قطيع المحطة الذى عومل وربى على البرنامج المتبع عادة وذلك من عمر يوم الى عمر ١٨ أسبوع. وتم تحصين الطيور ضد مرض النيوكاسل بواسطة لقاح هتشنر عند عمر ٧ أيام وبلقاح لاسوتا عند عمر ١٨ و٢٨ يوم وباللقاح الزيتى عند عمر ١٨ أسبوع من العمر. عند عمر ١٨ أسبوع تم وزن الطيور وتقسيمها عشوائيا وتسكينها فى اقفاص فردية ومعاملتها باربع معاملات غذائية متماثلة فى محتواها من البروتين والطاقة وتعتمدفى تكوينها على الذرة وكسب فول الصويا وتختلف فى محتوى خليط الفيتامينات من فيتامين أ المضاف (صفر و٤٠٠٠ و ١٢٠٠٠ و٢٤٠٠٠وحدة دولية/كجم علف). تم تقديم العلف والماء بصورة حرة للطيور خلال فترة الدراسة(١٨ – ٤٨ أسبوع).

أوضحت النتائج أن زيادة الأمداد من فيتامين أ يؤدى الى زيادة معنوية فىانتاج البيض و وزن وكتلة البيض وأن الكفاءة التحويلية للعلف تتحسن معنويا خلال الفترات المتأخرة من الدراسة. ومن جهة أخرى فأن كلامن كمية العلف المأكول ووزن الجسم لم يتأثرا معنويا بزيادة الأمداد من فيتامين أ عند اى مستوى من الفيتامين. كذلك فان دليل كفاءة انتاج البيض المقدر عند نهاية الدراسة يتحسن معنويا بزيادة الأمداد بفيتامين أ. أستخدام المستوى المرتفع من فيتامين أ يعظم من الكفاءة الأقتصادية و الكفاءة الأقتصادية النسبية (٠.٤٠ و ١١٤.٣% على التوالى) لأستخدام العلف مقارنة بالمستويات الأخرى من الفيتامين.

النسبة المئوية للخصوبة والتفريخ تزداد معنويا بزيادة الأمداد من فيتامين أ بينما النسبة المئوية للكتاكيت الكابسة تنخفض معنويا. زيادة الأمداد من فيتامين أ لم يكن لها تأثير معنويا على وزن الكتاكيت الفاقسة.

زيادة الأمداد من فيتامين أ يزيد معنويا وزن الكبد وذلك فى الفترة الأخيرة للدراسة(٤٨ اسبوع). وزن الطحال و الخصيتين و الأمعاء يزداد معنويا بزيادة الأمداد من فيتامين أ طوال فترة الدراسة.

أضافة فيتامين أ كعامل محفز ومنشط عند التحصين ضد مرض النيوكاسل فأنه يقوم بدور هام فى تحسين ورفع المناعة ومستوى الأجسام المضادة بسيرم الدم وكذلك فى صفار البيض. كذلك فأن مستوى الأجسام المضادة بسيرم دم الكتاكيت الفاقسة يزداد وذلك بزيادة مستوى الأجسام المضادة للأمهات وكذلك للبيض المنتج من الدجاجات التى غذيت على مستوى مرتفع من فيتامين أ. لذلك يمكن القول انة يمكن الأعتماد على أستخدام وقياس مستوى الأجسام المضادة فى صفار البيض كمقياس لمستوى المناعة للطيور.

أوضحت الدراسة التشريحية ان أستخدام مستوى ٢٤٠٠٠ وحدة دولية من فيتامين أ /كج علف يكون آمنا أما المستويات المنخفضة أو عدم وجود الفيتامين بالعلف يؤدى الى عديد من التأثيرات السيئة والتى تنعكس على وظائف الكبد والأمعاء وكذلك الخصوبة. ومن جهة أخرى فأن التأثير الضار على الطحال يؤثر على النظام المناعى للجسم.

لذلك يمكن استنتاج أن استخدام مستوى ٢٤٠٠٠ وحدة دولية من فيتامين أ /كجم علف فى أعلاف الدجاج البياض يكون أكثر أمنا و يؤدى الى زيادة معدلات الأداء الأنتاجى والتناسلى والمناعى للطيور و الكفاءة الأقتصادية.