Effects of Dietary Calcium Levels and Estradiol Benzoate on Some Productive Characters in Fayoumi Laying Hens .

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A Total number of 96 Fayoumi hens in their first laying season was used to study the effects of feeding 4 diets containing graded levels of Ca from 1.5 to 3.0% and injected with 2 levels of estradiol (0.5 and 2.0 mg per bird per week) for3 months on egg production and blood parameters . Most of egg characters and blood parameters were affected by the diet. The diet of 1.5 % Ca resulted in the favourable results, since it reduced ($P < 0.01$) feed intake and increased ($P < 0.05$) egg weight, white weight ($P < 0.01$), yolk weight ($P < 0.05$) , red blood cells($P < 0.01$)and white blood cells ($P < 0.01$) . The diet of 3.0 % Ca caused a decrease ($P < 0.01$) in white blood cells. Analysing the data failed to indicate any significant effect of estradiol on egg characters. However, the dose of 2.0 mg of estradiol caused an increase ($P < 0.05$) in the red blood cells, while white blood cells decreased ($P < 0.01$).

The dietary calcium level is of great importance for egg-type hens in which it involved in the regulation of daily feed consumption in laying hens (Tylor, 1970, Meyer et al.,1970 , and Scott et al.,1971) . Thus, the calcium level needs to be adjusted depending on variation in feed

intake for optimum egg production and shell strength (Roland and Harms, 1973) . Food consumption decreased with decreasing the dietary Ca level, despite a specific appetite for Ca which is known to exist in birds (Hughes and Wood-Gush, 1971) . The single most important contributing factor to shell formation is the metabolism of calcium . The regulation of Ca metabolism involves a number of hormonal systems, especially estrogen(Estradiol). Recent studies by Parsons and Combs (1981) indicated that serum Ca^{++} in the laying hens as well as Ca-binding-proteins caused differences in shell strength between lines, but no differences were noted in urinary Ca excretion , rate of transfer of Ca to the shell or in the effect of estradiol. The bound Ca may become free or it may be incorporated into the yolk . Thus, there are three excretions routes for Ca (feces , yolk, shell). Khalifa et al.,(1983) found that synthetic estrogens with high doses ranged from 0.5 mg daily to 3.5 mg. weekly per bird , increased egg production of laying hens. On the other hand, contradictory results were obtained concerning the effects of estradiol on blood analysis (Sturkie, 1954 ; Nirmalan and Robinson, 1972 , and Khalifa and Shebaita, 1984).

The purpose of this study was to examine effects of different dietary Ca levels (ranged from 1.5 to 3.0%) as well as different weekly estradiol injection of low doses on egg production, egg components, efficiency of feed utilization, egg shell quality and some blood parameters .

Materials and Methods

A total number of 96 Fayoumi laying hens at 12 months old was used. The experiment lasted for three months at

the summer season (29.9°C and 47.2 % RH). The hens were divided into four groups according to the different types of diets (Table 1), containing different levels of Ca, which were provided ad lib.. The limitation range of Ca levels from 1.5 to 3.0 % was coincided with the low productivity of native breeds under the conditions of summer months .Also, water was provided ad lib. by means of a continuous flow system . Each group was equally divided into three treatments. The first one served as a control while the two other treatments of estradiol benzoate were injected intramuscularly in the thighs of hens either 0.5 mg or 2.0 mg per bird per week. The control birds were injected with saline solution with the same procedure as the hormonal treatments . Each hen was kept individually in the cage, where the eggs were daily collected . The hens were weighed once a week during the experimental period . The blood samples from each bird were collected weekly in the sterile siliconized tube containing ethylene-diaminetetra-acetic-acid disodium salt (Na_2 EDTA) as an anticoagulant for blood analysis .

The red blood cell count (RBC_s) , white blood cell (WBC_s) , and blood hemoglobin (Hb) using sahli type of clinical hemoglobinometer were determined according to Oser, (1979) . The performance data included egg production (number and weight) , feed consumption , external and internal egg quality . At the end of each month , random sample of eggs from each group and/or hormonal treatments were taken for egg component study (egg white weight , gm (EW) , yolk weight, gm (EY), shell weight , gm (ES) and thickness of shell, mm(EST). The chemical analysis of the internal components of eggs includes protein % , fat % , Ca, mg % and Ca % in egg shell were determined according to the A.O.A.C.(1980). Phosphorus, mg % was determined according to the method described by Jackson (1973) . The data were analysed for statistical method according to Snedecor and Cochran (1968) .

TABLE 1 : Percentage composition and proximate analysis of the experimental laying dietes .

Ingredients	Percentage			
	Diet 1 %	Diet 2 %	Diet 3 %	Diet 4 %
Yellow corn, crushed	65.0	63.5	62.0	60.5
Soybean meal (44% C.P.)	14.0	14.0	14.0	14.0
Dec. cotton seed meal	4.0	4.0	4.0	4.0
Wheat bran	4.0	4.0	4.0	4.0
Fish meal (72% C.P.)	7.0	7.0	7.0	7.0
Oyster shell	3.5	5.0	6.5	8.0
Bone meal	2.0	2.0	2.0	2.0
Salt (Sodium chloride)	0.5	0.5	0.5	0.5
Total	100	100	100	100
Moisture	10.35	10.52	10.42	10.37
Crude protein	19.03	18.90	18.77	18.63
Ether extract	4.05	3.99	3.97	3.93
Crude ash	7.25	8.76	10.10	11.49
Crude fibre	3.98	3.97	3.95	3.91
Nitrogen - free extract	55.34	53.86	52.79	51.67
Calcium	1.50	2.00	2.50	3.00
Total phosphorus	0.66	0.66	0.66	0.66
Kilocalories M.E./ kg. diet	2910	2860	2815	2780
C/ P ratio	152.92	151.32	149.97	149.43
Ca / P ratio	2.27	3.03	3.79	4.55

Each ton of the diet was supplemented with 2 kg. of a vitamin and mineral mixture prepared according to the levels reported by Crawford et al.(1969) .

Results and Discussion

1. Egg characteristics :

Table 2 shows the mean of body weight, egg number, egg weight and mass in grams as influenced by Ca levels and estradiol doses. The data revealed insignificant difference (as summerized in Table 7) for the characters under study with respect to either Ca levels or hormonal treatments . However, a significant difference ($P < 0.05$) was observed in egg weight between Ca levels .

The dietary Ca level of 1.5 % increased egg weight than the other levels . This finding was confirmed by Bolden and Jensen, (1985) who stated that egg weight of Single Comb White Leghorn birds was significantly increased by decreased dietary Ca from 3.5 to 2.0 %, the physiological nature of which is not clearly understood . It appears that there is a considerable variation from experiment to experiment in the quantitative requirements of calcium for optimum performance of laying hens. No significant differences in production or in egg number and body weight occurred between dietary Ca levels , however, diet of 1.5 % Ca had the higher values for these traits than the other levels. On the other hand, the data (Table 2) failed to show any significant differences in the above-mentioned characters between different doses of hormone . The data obtained in the present study may be as a result of low injection in weekly doses of estradiol (0.5 or 2.0 mg) . Khalifa et al., (1983) using Fayoumi layers pointed out that the daily estradiol doses of 0.5 mg/bird improved egg characteristics than the weekly doses of 3.5 mg/bird. This improvement by estradiol treatment is in accordance with Prahov (1964),

TABLE 2 . The effects of Ca levels and/or estradiol doses on body weight and egg characters of Fayoumi laying hens .

Classification	No.	Body weight gm Mean ± SE	Egg number gm Mean ± SE	Egg weight gm Mean ± SE
Overall mean	96	1443 ± 20	40.4 ± 1.3	44.2 ± 0.26
Ca levels				
a. 1.5%	24	1491 ± 52	40.5 ± 3.2	45.5 ± 0.57a*
b. 2.0%	24	1480 ± 29	41.9 ± 2.3	43.6 ± 0.59b
c. 2.5%	24	1438 ± 43	39.3 ± 2.7	43.7 ± 0.35b
d. 3.0%	24	1360 ± 33	39.8 ± 2.4	44.1 ± 0.53ab
Estradiol doses				
a. Control	32	1401 ± 32	43.0 ± 2.4	44.3 ± 0.44
b. 0.5 mg	32	1446 ± 29	41.6 ± 2.3	44.5 ± 0.58
c. 2.0 mg	32	1480 ± 44	36.5 ± 2.0	43.9 ± 0.38

* Mean values bearing the common letter are not significantly different.

and Sharaf et al.,(1966) who explained this phenomenon by the physiological effect of estrogen upon the ovary and the oviduct which causing their activation and enhancing ovulatory processes . It could be recommended from the results obtained in this study that for improving egg number and egg weight, the hormone should be taken in daily doses .

2. Egg components :

The mean ± S E values of egg components (EW , EY , ES and EST) are shown in Table (3). Results indicate that all egg component characteristics were significantly influenced by Ca level, while estradiol did not affect the egg components (as summarized in Table 7) .

The dietary Ca level of 1.5 % significantly increased white weight (E W) and yolk weight (E Y) than the other levels . Meanwhile, the diet of 3.0 % Ca level caused an increase ($P < 0.01$) in shell thickness (EST) than the other Ca levels . It appears that the increase in the EW (albumen) of the diet of 1.5 % Ca may be due to its concomitant increase in egg weight than the other Ca levels . In this respect, Fletcher et al., (1983) reported that eggs of increasing weight are known to have additional proportions of albumen at the expense of yolk and shell at a consistent rate throughout each day . Yolk formation proceeds at a constant rate throughout the day and largely occurs during the 6 days before ovulation for any one yolk . Shell formation is phasic and is restricted to the interval when an ovum is present in the uterus . The inability of hens to increase the amount of shell with egg size has attributed to calcium logistics . This finding was in agreement with

TABLE 3 : The effects of Ca levels and/or estradiol doses on egg components of Fayoumi laying hens .

Classification	White weight (EW) , gm Mean ± S.E.	Yolk weight (EY) , gm Mean ± SE	Shell weight (ES) , gm Mean ± SE	Shell thickness (EST) , mm. Mean ± SE
Overall mean	21.9 ± 0.33	15.6 ± 0.16	5.06 ± 0.07	0.40 ± 0.003
Ca levels				
a. 1.5 %	24.1 ± 0.53 a*	16.1 ± 0.27 a	5.23 + 0.20 a	0.40 ± 0.004 b
b. 2.0 %	20.6 ± 0.84 b	15.5 ± 0.24 a	4.61 ± 0.16 b	0.37 ± 0.009 c
c. 2.5 %	21.3 ± 0.66 b	16.0 ± 0.32 a	5.30 ± 0.14 a	0.40 ± 0.007 b
d. 3.0 %	21.6 ± 0.56 b	14.7 ± 0.46 b	5.11 ± 0.14 a	0.43 ± 0.006 a
Estradiol doses				
a. Control	22.1 ± 0.59	16.0 ± 0.35	5.07 ± 0.10	0.40 ± 0.007
b 0.5 mg	21.8 ± 0.64	15.5 ± 0.30	4.94 ± 0.10	0.40 ± 0.006
c. 2.0 mg	21.8 ± 0.60	15.2 ± 0.23	5.18 ± 0.16	0.41 ± 0.008

* Mean values bearing the common letter are not significantly different .

Mongin and Sauveur, (1974) who reported that hens offered extradietary calcium have been shown to preferentially consume these supplements during shell deposition and improve shell quality . However, the dietary calcium level in excess of 3.0 % increased shell thickness (Cox and Balloun, 1968). No significant differences were observed between hormone doses for egg component , however, the shell weight and thickness tended to be slightly increased by 2.0 mg dose of estradiol(Table 3).Steroid hormones as estradiol have been implicated in the regulation of Ca and P metabolism in the laying hen through several modes of action which may affect egg shell quality (Curl et al., 1985) .

Table 4 represents the effects of the diets containing different Ca levels and hormonal doses on egg characteristics including protein % , fat % , Ca, mg % and P, mg % for egg yolk , egg white and egg meat (yolk + white) besides Ca % in egg shell . It could be observed from the present data that the previous parameters as percentages did not affected by neither dietary calcium levels nor hormonal treatments . However, the said parameters calculated as quantity showed some variations among Ca level diets and/or hormonal doses . The diet of 1.5 % Ca caused an increase in protein, fat , Ca and P of the egg components , account for, higher egg weight of this diet . On the other hand, the control treatment revealed slight increase for the previous parameters than the different doses of hormone. However, P in egg yolk (mg.) increased by hormonal treatment .

It could be concluded from the present results that estradiol improved the absorption of phosphorus for egg yolk .

TABLE 4 . Effect of the different dietary Ca levels and hormonal doses on egg components .

Classification	Experimental diets				Hormonal doses		
	1.5% Ca	2.0% Ca	2.5% Ca	3.0% Ca	0	0.5 mg	2.0 mg
Egg yolk :							
Protein %(1)	16.70	16.70	16.64	16.65	16.64	16.67	16.71
Protein ,gm(2)	2.69	2.59	2.66	2.45	2.66	2.58	2.54
Fat %	32.20	32.21	32.22	32.25	32.20	32.25	32.21
Fat, gm	5.18	4.99	5.16	4.74	5.15	5.00	4.90
Ca, mg %	162	164	165	163	163	164	164
Ca, mg	26.10	25.42	26.40	23.96	26.08	25.42	24.93
P , mg %	592	593	592	594	592	592	593
P , mg	95.3	91.92	94.72	87.32	94.72	91.76	90.14
Egg White :							
Protein %	11.01	10.93	10.88	10.91	10.98	10.96	10.93
Protein, gm	2.65	2.25	2.32	2.36	2.43	2.39	2.38
Ca , mg %	13	12	13	13	13	13	12
Ca mg	3.13	2.47	2.77	2.81	2.87	2.83	2.62
P, mg %	19	17	17	17	17	18	18
P mg	4.58	3.50	3.62	3.67	3.76	3.92	3.92
Egg meat :							
Protein %	13.47	13.58	13.52	13.60	13.54	13.55	13.53
Protein , gm	5.41	4.90	5.04	4.94	5.16	5.05	5.01
Fat %	11.99	11.95	11.92	11.99	12.00	11.97	11.92
Fat, gm	4.82	4.31	4.45	4.35	4.57	4.46	4.41
Ca , mg %	60	60	60	60	61	59	60
Ca, mg	24.12	21.66	22.38	21.78	23.23	22.01	22.20
P , mg %	219	219	220	219	220	220	218
P,mg	88.4	79.06	82.06	79.50	83.82	82.06	80.66
Egg shell :							
Ca %	36.00	36.05	36.01	36.09	36.03	36.07	36.06
Ca. gm	1.88	1.66	1.91	1.84	1.83	1.78	1.87

(1) calculated as percentage .

(2) Calculated as quantity .

3. Blood parameters :

The blood parameters (RBCs , WBCs , Hb and MCH) for either Ca levels or hormonal doses are given in Table 5 , and the significance of these parameters was summarized in Table 7 . The diet of 3.0 % Ca level decreased ($P < 0.01$) white blood cells than the other Ca levels . On the other hand , the diet of 2.5% Ca caused a decrease ($P < 0.01$) in red blood cells than the other Ca levels . The maximum hemoglobin was obtained by feeding the diets of 2.5 and 3.0% Ca followed by the 1.5 % Ca while the least hemoglobin value was obtained for the diet of 2.0 % Ca . The mean corpuscular hemoglobin (MCH) increased ($P < 0.01$) with the increase of dietary Ca level from 1.5 to 2.5 and 2.5 to 3.0 % . It seems that the dietary Ca levels are more related to plasma Ca or P rather than the said blood parameters in this study . The estradiol treatment of 2.0 mg increased ($P < 0.05$) the red blood cells than the other treatments . The white cells decreased ($P < 0.01$) followed by the untreated birds and the highest white blood cells were observed in the estradiol of 0.5 mg. The dose of 0.5 mg of the hormone caused a decrease ($P<0.01$) in hemoglobin , while the decrease ($P < 0.01$) in MCH was observed in the dose of 2.0 mg of the hormone . The reduction in the red cells count of the untreated birds and the hormone of 0.5 mg may be due to the formation of egg component . In this study , egg production of both untreated group and hormone of 0.5 mg were higher than the hormone of 2.0 mg, however , these differences were not significant . Domm and Taber (1946) had the same findings since they observed in chickens that the period of highest egg production coincides with the lowest red cells count. Sturkie and Textor (1960) found that female castrates

TABLE 5 : The effects of Ca levels and/or estradiol doses on blood parameters of Fayoumi laying hens .

Classification	RBCs mils/ c.mm Mean ± SE	WBCs (10^3)/c.mm Mean ± SE	Hemoglobin (Hb)gm% Mean ± SE	**MCH (pg) Mean ± SE
Overall mean	2.40 ± 0.06	37.5 ± 1.92	10.96 ± 0.10	49.0 ± 1.3
Ca levels				
a. 1.5%	2.61 ± 0.09 *a	46.7 ± 5.20 a	10.80 ± 0.09 b	42.3 ± 1.2 b
b. 2.0%	2.40 ± 0.10 a	40.0 ± 3.41 b	9.83 ± 0.12 c	43.2 ± 2.3 b
c. 2.5%	2.00 ± 0.07 b	40.0 ± 5.90 b	11.53 ± 0.34 a	58.6 ± 2.1 a
d. 3.0%	2.60 ± 0.19 a	23.3 ± 2.60 c	11.67 ± 0.19 a	52.0 ± 4.2 a
Estradiol doses				
a. Control	2.25 ± 0.09 b	35.0 ± 2.98 b	11.33 ± 0.33 a	54.6 ± 3.4 a
b. 0.5 mg	2.36 ± 0.16 ab	52.5 ± 5.29 a	10.38 ± 0.06 b	48.9 ± 2.4 ab
c. 2.0 mg	2.60 ± 0.06 a	25.0 ± 1.56 c	11.18 ± 0.14 a	43.6 ± 1.0 b

* Mean within a classification within a column followed by the same letter do not differ significantly, otherwise they do differ signnificantly at $P < 0.05$.

** MCH (Pg) = $\frac{10 \times \text{Hb gm \%}}{\text{RBCs (mils)/c.mm}}$

of birds (poulards) have the same number of red cells as normal females , indicating that estrogen (estradiol) has no effect on erythropoiesis . It seems , in this study , that hemoglobin and MCH were higher ($P < 0.01$) in the untreated birds rather than the hormonal treatments .

4. Feed consumption :

It appears as shown in Tables (6 and 7) that there are an increase ($P < 0.01$) in feed intake/ hen / day with increasing the dietory Ca levels , however, this trend is not pronounced in the diet of 3.0 % Ca. The Ca intake increased ($P < o.ol$) with increasing the Ca level of different diets . Feed consumption decreased with decreasing concentration of calcium in the diet since the diet of 1.5 % Ca decreased ($P < 0.01$) feed intake , Ca intake , and improved feed effeciency thant the other Ca levels.

Similar results were obtained by Gilbert et al., (1981) who indicated that feed consumption decreased as dietary calcium level decreased . Feed consumption in our study , is strongly affected by the poorer egg production at summer months which lead to decrease growth of ovarian follicles and concequently results in a reduced feed requirement as a result of reduction in hormonal secretion of the ovary . Injecting estradiol weekly had no significant effect on feed intake. This finding is in agreement with reports of Libby et al., (1953) .

TABLE 6. Feed intake , Ca intake and feed efficiency for different treatments .

Classification	Feed intake gm/bird/day	Ca intake gm/bird/day	* Feed efficiency
Overall mean	109.0	2.47	5.57
Ca levels			
a. 1.5%	102.7	1.54	5.10
b. 2.0%	108.6	2.17	5.42
c. 2.5%	117.6	2.94	6.22
d. 3.0%	107.1	3.21	5.54
Estradiol dose			
a. control	108.5	2.47	5.18
b. 0.5 mg	108.6	2.47	5.34
c. 2.0 mg	108.6	2.47	6.18

* Feed efficiency = $\frac{\text{Feed intake (kg)}}{\text{Egg mass (kg.)}}$

TABLE 7 . Significance of Ca levels and estradiol doses for characters under study .

Characters	Ca level	Estradiol
Body weight	NS	NS
Egg number	NS	NS
Egg weight	*	NS
Egg mass	NS	NS
White weight	**	NS
Yolk weight	*	NS
Shell weight	**	NS
Shell thickness	**	NS
Feed intake	**	NS
Ca intake	**	NS
Red blood cells	**	*
White blood cells	**	**
Hemoglobin (Hb)	**	**
MCH	**	**

NS : Not significant
** : significant at $P < 0.01$
* : significant at $P < 0.05$.

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تأثير كالسيوم العليقه وبنزوات الاستراديول على بعض الصفات الانتاجيه للدجاج الفيومى

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استخدم عدد ٩٦ دجاجه فيومى بياضه لدراسة تأثير أربع مستويات من كالسيوم العليقه (١ر٥ ، ٢ر٥ ، ٣ر٥ ٪) مع مستويين من الاستراديول من طريق الحقن (٠ر ، ٢ر ملليجم لكل طائر أسبوعيا) لمدة ٣ أشهر على أنتاج البيض ومكونات الدم . أعطت العليقه المحتويه على ١ر٥ كالسيوم أفضل النتائج حيث أنخفضت معنويا كمية الغذاء المستهلك وتزاد معنويا وزن البيض ، ووزن بياض البيض ، ووزن الصفار ، وعدد كرات الدم الحمراء وكرات الدم البيضاء . أما العليقه المحتويه على ٣ر٥ ٪ كالسيوم فقد سببت أنخفاضا فى عدد كرات الدم البيضاء . ولم يكن لحقن الاستراديول بالمعدلات المذكورة أى تأثير على المواصفات الانتاجيه للطيور أو مكونات البيض ومع ذلك فأن حقن الاستراديول بمعدل ٢ ملليجم أسبوعيا أدى الى زيادة معنويه فى عدد كرات الدم الحمراء وأنخفاضا معنويا فى عدد كرات الدم البيضاء .