

EFFECT OF DIETARY CHROMIUM AND ASCORBIC ACID ON PRODUCTIVE AND REPRODUCTIVE PERFORMANCE OF DOKKI₄ CHICKENS UNDER WINTER CONDITIONS IN EGYPT

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ABSTRACT

The effect of chromium (Cr) and ascorbic acid (AA) as a dietary supplementation on productive and reproductive performance of chickens was studied under winter conditions of Egypt. Total number of 300 (250 hens+50 cocks) Dokki₄ chickens, of 30 weeks old were divided into 5 groups of 5 replicates each (10 hens +1 cock/ replicate). The remaining 25 cocks were divided into 5 groups of 5 cocks each and housed separately for semen evaluation. Birds in the 1st group (Cont.+) were kept in controlled normal temperature (CNT, 25°C) with 64±2% relative humidity (RH) and fed corn-soybean meal basal diet (16.4% crude protein, 2750 kcal ME/kg diet) without any supplementation. Birds in groups from 2 to 5 were exposed to natural low temperature (NLT, 8-18°C) with 65±2%. These groups were fed either basal diet (Cont.-), or the basal diet supplemented either with either 250 ppm AA, 400 ppb Cr or 250 ppm AA plus 400 ppb Cr. Birds in NLT groups decreased ($P \leq 0.05$) relative body weight change (BWC, 8.6%), egg number (12.9%), egg mass (13.5%), laying rate (18.5%), hatchability of fertile eggs (4.6%), sperm motility (5.7%) and response to sheep red blood cells (SRBC's) post-injection at 6-days (13.6%) and at 9-days (25.8%), impaired feed conversion (28.9%) and economic efficiency (EE, 48.1%) and increased feed intake (9.3%), plasma contents of cholesterol (21.3%) and glucose (12.2%) contents and malondialdehyde (MDA) concentration in plasma (105.7%) and in semen (54.1%) compared with control group under CNT. Dietary supplementations alleviated ($P \leq 0.05$) the adverse effects of the NLT. Birds fed basal diet supplemented with AA+Cr under NLT had significantly ($P \leq 0.05$) improved BWC (5.9%), feed conversion (17.1%), egg number (9.5%), egg mass (5.3%), EE (33.6%), plasma cholesterol (10.9%) and glucose (10.2%), semen MDA (25.8%), response to SRBC's post-injection at 6-days (19.8%) and at 9-days (38.3%), sperm motility (6.2%) and hatchability of fertile eggs (5.2%) compared with control under NLT. There was no significant effect on most semen qualities, body temperature, fertility, hatchability (based on total eggs set) and hatched chick weight. It may be concluded that a dietary combination of 250 ppm of AA and 400 ppb of Cr provides positive effect on the productive and reproductive performance and EE of Dokki₄ chickens under NLT.

Keywords: chickens, chromium, ascorbic acid, cold stress, performance, peroxidation

INTRODUCTION

Poultry performance goes down at NLT. An increase in feed intake and a decrease in egg production and feed conversion are believed to be associated with NLT in laying hens (Spinu and Degen, 1993). Environmental cold stress causes a deficiency of AA in poultry (Sahin and Sahin, 2001). Cold conditions cause decreases in plasma concentrations of some vitamins,

minerals and insulin and increase in plasma corticosterone in poultry (Ensminger *et al.*, 1990 and Sigel, 1995). Several studies have shown that AA is an indispensable micronutrient required to maintain the physiological processes of poultry (Sahin *et al.*, 2002). Poultry are known to have the ability to synthesize AA, Pardue and Thaxton (1986) have documented evidence that particular environmental stressors can alert AA utilization or synthesis in avian species. It has been also reported that AA synthesis inadequate under stress conditions such as low or high environmental temperature, humidity and parasitic infestation (Kutlu and Forbes, 1993; Sahin and Kucuk, 2001 and Sahin *et al.*, 2002). Several studies have indicated beneficial effects of AA on the performance of cold-stressed laying hens (Sahin and Sahin, 2001 and Kucuk *et al.*, 2003). Dietary Cr has positively affected the growth rate and feed efficiency of growing poultry (NRC, 1997 and Lien *et al.*, 1999). The beneficial effects of Cr can be observed more efficiently under environmental, dietary and hormonal stresses (Sahin *et al.*, 2001). Supplemental dietary Cr is also recommended by NRC (1997) for animals undergoing environmental stress. In addition, Cr is thought to be essential for activating certain enzymes and for stabilization of proteins and nucleic acids (Okada *et al.*, 1984 and Anderson, 1987). It has been recognized that insulin metabolism influences lipid peroxidation (Gallaher *et al.*, 1997). Cr as an insulin Co-factor is therefore postulated to function as an antioxidant, its deficiency causes disorders of carbohydrates and protein metabolism, reduction in insulin sensitivity in the peripheral tissues as well as a decrease in growth rate (Lindeman, 1996). Chicken and vertebrate spermatozoa display high rates of metabolic activity and consequently reactive O₂ species (ROS) which is believed to increase under stress conditions. They are rich in polyunsaturated fatty acids, which renders them particularly susceptible to oxidation by ROS, especially under stress conditions in humans (Aitken *et al.*, 1989), chickens (Eid *et al.*, 2006), and also in rabbits (Castellini *et al.*, 2000). Over generations, ROS was associated with male infertility (Akiyama, 1999). When hens were inseminated with semen from stressed males, sperm-egg penetration and fertilized egg production decreased when compared to hens inseminated with semen from control males (McDaniel *et al.*, 1995). As a result of, decrease in reproductive performance followed by significant financial losses occurs. It has been suggested that antioxidants reduce the physiological depress to stress in animals (Eid *et al.*, 2003 and 2006). The antioxidant defense against ROS seems to be heavily influenced by nutrition. For this reason, the use of nutraceuticals in human and animal nutrition has considerably increased in the last decades (Young *et al.*, 2000). The oxidative stability of rabbit semen increased in relation to dietary antioxidants like AA (Yousef *et al.*, 2003). Antioxidants can protect against the damaging effect of leukocyte-derived ROS on sperm movement (Baker *et al.*, 1996). Objective of this study was to determine the effect of AA and/or Cr supplementations on the performance of Dokki, chickens reared under winter condition in Egypt.

MATERIALS AND METHODS

The present study was conducted at Sakha Animal Production Research Station and Laboratories, APRI, ARC, Egypt during Dec.-Feb. 2007 to study the effects of dietary AA and/or Cr on chicken's performance during winter season. Total number of 300 (250 laying hens+50 cocks) chickens (Egyptian strain, Dokki₄), 30 wk's old were divided into 5 groups of 5 replicates each (10 laying hens+1 cocks/replicate). The remaining 25 cocks were also divided into 5 groups of 5 cocks each and reared separately for semen evaluation. Birds were reared in open sided pens exposed to NLT prevails in Egypt during winter season. Average NLT was 18°C during day time and 8°C during night time with 65±2% RH. The birds were exposed to a light program of 16h of light: 8h of dark /day. Birds were fed, for 12 wk's, either basal (Table 1) diet (1st treatment: control at 25°C, CNT), the basal diet (2nd treatment: control at 8-18°C, NLT), the basal diet supplemented with 250ppm AA (a heat stabilized product produced by Hoffmann-La Roche) (3rd treatment), 400ppb Cr in form of chromium piclonate (CrPi) (Chromax®, Prince Agric. Products) (4th treatment) or 250 ppm AA plus 400 ppb Cr (5th treatment).

Table 1. Composition of the experimental chicken diet (30-42 wk's old)

Ingredient	%
Yellow corn	66.00
Soybean meal (44%)	24.00
Sodium chloride	0.30
Limestone	7.59
Dicalcium phosphate	1.71
Vit. + Min. Mix. ¹	0.30
DL-Methionine	0.10
Calculated analysis²:	%
Crude protein	16.43
Crude fiber	3.20
Ether extract	2.70
Calcium	3.33
Available phosphorus	0.45
Meth.+ Cyst.	0.67
Lysine	0.86

¹Vitamins+Minerals Mixture provided /kg of diet: 6000 IU vit A, 2000 ICU cholecalciferol, 10 IU vit E, 2.5 mg vit K₃, 2.5 mg riboflavin, 12 mg nicotinic acid, 10 mg Ca pantothenate, 300 mg choline chloride, 4 µg cyanocobalamin, 5 mg pyridoxine, 3 mg thiamine, 0.5 mg folic acid, 0.2 mg biotin, Trace mineral (mg/kg of diet): 40 Mn, 40 Zn, 40 Fe, 4 Cu, 0.2 Se, 0.5 I.

²According to Egyptian Feed Composition Tables (2001). Metaolizable energy=2750kcal/kg diet

Productive parameters: Body weight and feed intake were estimated weekly and feed conversion was calculated. Eggs laid and weights were recorded daily. The EE based on feed cost was calculated. At the end of the exp. period, 9 birds /group were slaughtered, blood samples in heparinized tubes were centrifuged at 3000 rpm for 20 min. Plasma cholesterol was estimated (Watson, 1960). Lipid peroxidation as thiobarbituric acid reactive substances (TBARS) was estimated in plasma or semen by method of Placer

et al. (1966) as modified by Matkovic *et al.* (1989). Values of TBARS were expressed in terms of MDA nmol/ml plasma or semen).

Reproductive parameters: Three cocks /group were used for collecting semen, by means of abdominal massage, at the 4th, 8th and 12th.wk of the exp. Semen volume was determined using graded tubes. Sperm concentration ($\times 10^9$ sperm/ml) was determined by a haemocytometer. Immediately after semen collection, sperm motility percentage was measured using a small droplet from each individual placed on a warm slide, covered with a slide and examined for sperm motility microscopically at 400 \times magnification using a stage warmer set at 39°C. Sperm motility was classified as described by Melrose and Laing (1970). Sperm livability was determined (Lake and Stewart, 1978). For fertility and hatchability determination, eggs laid were collected, stored at 15.5 °C dry bulb and 70% RH, incubated at 37.6°C and 55–60% RH, and hatched at 37.3°C and 65-70% RH. Fertility was calculated as number of fertile eggs as relative to total eggs set; meanwhile hatchability was calculated as number of healthy hatched chicks as relative to total or fertile eggs. Data were statistically analyzed using one-way ANOVA (SAS, 1996). Before analysis, all percentages were subjected to logarithmic or arcsine values transformation ($\log_{10}x+1$) to approximate normal distribution. Significant differences among treatment means ($p \leq 0.05$) were separated by Duncan's new multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

Productive performance: Data in Table (2) show that NLT significantly ($P \leq 0.05$) reduced final body weight (FBW, 7.6%), RBWC (8.6%), egg number (12.9%), egg mass (13.5%) and laying rate (18.6%), increased feed intake (9.3%) and impaired feed conversion (28.9%) compared to control under CNT. There was a positive ($P \leq 0.05$) effect of AA and/or Cr supplementations on final body weight, RBWC, egg laying rate, egg number, egg weight under NLT. Dietary combination of AA and Cr improved ($P \leq 0.05$) final body weight (6.4%), relative body weight change (5.9%), egg number (9.5%), egg mass (5.3%) and laying rate (16.8%) compared to the control diet at NLT. No significant effect of either NLT or any supplementations was noted on body temperature and egg weight. Stress depressed AA synthesis in poultry (Anderson, 1987), thus may result in marginal Cr and AA deficiency or increased Cr and AA requirements implying that both Cr and AA should be supplemented in such conditions. Both AA and Cr are known to increase the use of corticosteroids released during stress (Pardue and Thaxton, 1986 and Sahin *et al.*, 2001), which had an important role in responding to stress. With respect to the benefits of dietary AA supplementation under stress in terms of better poultry performance, the present results confirmed those of Orban *et al.* (1993) and Kutlu and Forbes (1993). It is a well-known that growth rate and egg production decrease when ambient temperature goes below or above the thermally neutral zone (Ensminger *et al.*, 1990). At temperature degrees above or below the thermally neutral zone, corticosteroid secretion increases in response to stress (Brown and Nestor, 1973). Kutlu and Forbes

(1993) reported that AA reduces the synthesis of corticosteroids hormones in birds. By decreasing synthesis and secretion of corticosteroids, AA alleviates the adverse effects of cold stress and increases poultry performance (Sahin and Sahin 2001). KuKcuk *et al.* (2003) reported that AA supplement improved egg production under cold stress. Also, several researches have documented beneficial effects of AA supplementation on egg production in poultry kept under environmental stress (Orban *et al.*, 1993 and Bains, 1996). Also, Sands and Smith (1999) reported that dietary Cr supplementation increased growth rate in stressed broilers. Lien *et al.* (1999) reported that 1600 ppb/kg or 3200 ppb/kg Cr Pi supplementation to a broiler diet increased feed intake and improved live weight gain. In addition, Stele and Rose (1981) found that an addition of 20 ppm chromium chloride increased weight gain of turkey poults. Moreover, Sahin *et al.* (2001) reported that a supplement of 400 ppb Cr to the diet of laying hens reared under NLT increased egg production and improved feed efficiency. Sahin *et al.* (2002) also reported that the decrease in live weight, feed intake, egg production, and feed efficiency in laying hens reared under cold stress was alleviated by dietary Cr and Zn supplementation. It is obvious that Cr is involved in protein metabolism (Anderson, 1987), and it have a role in nucleic acid metabolism and in stimulation of amino acid incorporation into liver protein *in vitro* (Weser and Koolman, 1969). Okada *et al.* (1984) showed that an interaction of Cr with DNA templates resulted in a significant stimulation of RNA synthesis *in vitro*.

Table (2) Productive performance of Dokki, laying hens reared during winter season as affected by dietary supplemental AA or Cr.

Item	(+) Cont. (25°C)	Low temperature treatments (8-18°C)				P value
		(-) Cont.	+AA 250ppm	+Cr 400ppb	+ AA+Cr	
Initial body weight, g	1503.3 ±8.82	1500.0 ±5.78	1501.7 ±10.14	1508.3 ±10.93	1513.3 ±6.67	0.809
Final body weight, g	1746.7 ^a ±3.33	1613.3 ^d ±6.66	1670.0 ^c ±10.00	1676.7 ^c ±14.53	1716.7 ^b ±8.82	0.001
Body weight change, %	16.19 ^a ±0.66	7.55 ^d ±0.19	11.21 ^c ±0.35	11.16 ^c ±0.38	13.43 ^b ±0.15	0.001
Body Temperature, °C	41.47 ±0.067	40.67 ±0.033	41.50 ±0.058	41.57 ±0.38	41.47 ±0.15	0.166
Egg number /h /12wk	55.00 ^a ±1.154	47.90 ^d ±0.001	49.67 ^{cd} ±0.882	51.00 ^{bc} ±0.577	52.33 ^b ±0.333	0.002
Egg weight, g	49.97 ±0.09	49.69 ±0.01	49.79 ±0.08	49.84 ±0.08	49.76 ±0.133	0.312
Egg mass, kg/h/12wk	2.75 ^a ±0.059	2.38 ^d ±0.001	2.47 ^{cd} ±0.041	2.54 ^{bc} ±0.033	2.60 ^b ±0.022	0.001
Egg Production, %	65.48 ^a ±1.38	53.34 ^d ±0.12	59.13 ^c ±1.05	60.71 ^{bc} ±0.69	62.30 ^b ±0.39	0.001
Feed intake, g/h/d	100.7 ^c ±0.069	110.1 ^a ±0.061	106.8 ^{ab} ±0.68	106.9 ^{ab} ±0.78	102.2 ^{bc} ±3.34	0.013
Feed Conversion	3.08 ^d ±0.06	3.97 ^a ±0.00	3.63 ^b ±0.04	3.53 ^b ±0.06	3.29 ^{cd} ±0.13	0.001

^{abcd} Means within each row with different superscripts are significantly different (p<0.05). AA = ascorbic acid, Cr = chromium, feed conversion = g feed / g egg mass

Malondialdehyde, plasma constituents and immune response to SRBS's
Results in Table (3) showed that NLT increased ($P \leq 0.05$) plasma cholesterol (21.3%) and glucose (12.2%) contents and MDA concentrations in plasma (105.7%) and in semen (54.1%) and decreased response to SRBC's post-injection at 6-days (13.6%) and at 9-days (25.8%) compared to control under CNT. Cholesterol, glucose and MDA concentrations were lower ($P \leq 0.05$) in the groups supplemented with AA and /or Cr than the control group under CNT. Post-injection responses to SRBC's at 6- and 9-days were higher ($P \leq 0.05$) in the groups supplemented with AA and AA+Cr than the control group under NLT. Results cited herein confirmed those of Kucuk *et al.* (2003) and Metwally (2004). Generally, supplementing AA decreased plasma contents of glucose and cholesterol. These results might be due to decreased corticosterone (Catabolic) and increased insulin (anabolic) concentrations upon AA supplementations. The Cr stimulates and regulates the action of insulin (Mowat, 1994) which is involved in anabolic processes (Colgan, 1993). Through increasing the effectiveness of insulin, Cr indirectly potentiates AA transportation (Mann and Newton, 1975 and Seaborn *et al.*, 1994). Also, Cr was shown to be a protective factor against heart disease by achieving a regression of cholesterol induced arteriosclerosis (Abraham *et al.*, 1991).

Table 3. MDA, plasma constituents and immune response to SRBC's of Dokki₄ hens during winter season as affected by dietary AA or Cr.

Item	(+) Cont. (25°C)	Low temperature treatments (8-18°C)				P value
		(-) Cont.	+AA 250ppm	+Cr 400ppb	+ AA +Cr	
Plasma cholesterol, mg/dl	153.67 ^a ±2.33	186.33 ^a ±1.86	173.67 ^{bc} ±1.86	175.33 ^b ±2.91	166.00 ^c ±3.06	0.001
Plasma glucose, mg/dl	229.00 ^c ±1.00	257.00 ^a ±1.53	239.33 ^b ±2.040	235.00 ^{bc} ±2.089	230.07 ^c ±2.85	0.001
Plasma MDA, nmol/ml	1.23 ^c ±0.15	2.53 ^a ±0.15	1.83 ^b ±0.09	1.85 ^b ±0.08	1.53 ^{ab} ±0.15	0.001
Semen MDA, nmol/ml	4.23 ^c ±0.15	6.52 ^a ±0.13	5.43 ^b ±0.14	5.39 ^b ±0.11	4.84 ^c ±0.11	0.001
Post-injection responses to SRBC's at:						
3- days	5.12	4.54	4.28	4.18	4.57	0.105
	0.13	0.15	0.11	0.13	0.14	
6- days	7.35 ^{ab}	6.35 ^c	7.36 ^{ab}	6.60 ^{bc}	7.61 ^a	0.001
	0.12	0.10	0.10	0.13	0.11	
9- days	4.85 ^a	3.60 ^b	4.85 ^a	3.85 ^b	4.98 ^a	0.001
	0.11	0.12	0.13	0.11	0.10	

^{abc} Means within each row with different superscripts are significantly different ($p \leq 0.05$).
MDA= Malondialdehyde, AA =ascorbic acid, Cr = chromium

It is well known that stress increases MDA level as a lipid peroxidation indicator (Halliwell and Gutteridge, 1989 and Sahin *et al.*, 2002). Antioxidant systems (glutathione peroxidase, superoxide dismutase and vitamins: E, C, A) are important in scavenging free radicals and their metabolic products, as well as in maintaining normal cellular physiology restoring depletion of various antioxidants in stressed poultry (Halliwell and Gutteridge, 1989). In the present study, semen MDA was decreased with

dietary AA and/or Cr supplementation (Table 3). These results confirmed those of Gursu *et al.* (2004) and Sahin *et al.* (2002). The presence of high concentrations of polyunsaturated fatty acid (Ravie and Lake, 1985) within the lipid fraction necessitates the presence of an efficient antioxidant system to protect against peroxidative damage and possible associated sperm dysfunction. Because the actual mechanism of lipid peroxide formation by fowl spermatozoa remains unidentified, it is not yet known whether the presence or absence of some factors such as antioxidants could suppress the production of high concentrations of lipid peroxides and subsequently enhance the sperm quality under stress conditions (Aitken *et al.*, 1989). They observed also that Lipid content of the seminal and spermatozoa make it a good target for free radical attacks under oxidative stress. This assumption is supported by the elevation of MDA under cold condition.

Table (4) Semen characteristics of Dokki₄ cocks reared during winter season as affected by dietary supplemental AA or Cr.

Item	(+) Cont. (25°C)	Low temperature treatments (8-18°C)				P value
		(-) Cont.	+AA 250ppm	+Cr 400ppb	+ (AA+Cr)	
Semen volume, ml	0.360 ±0.012	0.350 ±0.012	0.363 ±0.012	0.357 ±0.003	0.383 ±0.003	0.111
Concentration of sperm, 10 ⁹ /ml	2.19 ±0.106	2.18 ±0.107	2.20 ±0.173	2.27 ±0.145	2.40 ±0.058	0.651
Sperm motility, %	85.57 ^a ±1.18	80.67 ^b ±0.166	83.50 ^{ab} ±2.760	84.00 ^{ab} ±0.577	85.66 ^a ±1.202	0.048
Life sperm, %	86.00 ±1.16	86.00 ±1.16	88.00 ±1.16	88.70 ±1.33	90.00 ±2.57	0.166
Abnormal sperm, %	4.00 ±2.31	4.00 ±2.31	3.00 ±0.90	2.60 ±1.54	2.00 ±0.16	0.836

^{abc} Means within each row with different superscripts are significantly different (p≤0.05).
AA =ascorbic acid, Cr = chromium

Semen characteristics: Effects of AA, and/or Cr on semen volume, sperm cell concentration, and percentages of motile, life and abnormal spermatozoa under the NLT are illustrated in Table (4). There were no significant effects of NLT on all semen characteristics except for sperm motility % were decreased (5.73%) compared with the control under CNT. Only dietary supplemented with AA+Cr significantly enhanced sperm motility comparing to the control under NLT. On the other hand, this combination completely recovered sperm motility to the same level of the control under CNT. It is of interest to note that the decline in sperm motility was associated with accumulation of the MDA in semen. The combination between AA and Cr decreased MDA in the seminal because of its antioxidative properties and as a result of this; sperm motility was enhanced under cold stress. This is in accordance with the report of Surai *et al.* (1997), who showed that enhancement of the antioxidant capacity of semen could present a major opportunity for improving male fertility. This was clear in the present data too. The combination between AA and Cr protects sperm cell membrane from free radical attacks under cold stress, which is translated into enhanced motility and insignificant reduction in dead sperm %, in addition to the reduced MDA value under cold stress.

Fertility and hatchability: Data concerning the effect AA and/or Cr under NLT on reproductive traits of laying hens are presented in Table (5). No significant effects of either NLT or dietary supplements were found on fertility, hatchability (based on total eggs set) and chick weight at hatch. Significant decrease (4.6%, $P=0.044$) were found in hatchability (based on fertile eggs set) by NLT. Dietary supplemented with AA+Cr showed higher hatchability (based on fertile eggs set) compared to the control group at NLT and equaled with that of the control at CNT. This significant effect may refer to the combined antioxidative effect of AA+Cr. However, reverse reports (Whitehead and Keller, 2003 and McDaniel *et al.*, 2004) did not find any positive effect for improving fertility or hatchability due to AA.

Table 5. Reproductive performance of Dokki₄ laying hens reared during winter season as affected by dietary supplemental AA or Cr.

Item	(+) Cont. (25°C)	Low temperature treatments (8-18°C)				P value
		(-) Cont.	+AA 250ppm	+Cr 400ppb	+ (AA+Cr)	
Eggs fertility, %	88.00 ±1.16	88.00 ±1.16	88.00 ±1.51	88.50 ±1.25	90.00 ±1.73	0.734
Hatchability of total eggs set, %	75.40 ±1.26	74.40 ±1.24	76.93 ±1.05	76.53 ±0.87	77.27 ±1.59	0.766
Hatchability of fertile eggs, %	87.01 ^a ±0.84	83.00 ^c ±1.15	85.20 ^b ±0.87	85.00 ^b ±0.58	87.33 ^a ±0.49	0.044
Hatched chicks weight, g	35.20 ±0.82	35.10 ±0.83	35.86 ±0.82	35.53 ±0.48	35.90 ±0.23	0.804

^{abc} Means within each row with different superscripts are significantly different ($p \leq 0.05$).
AA = ascorbic acid, Cr = chromium

Economic efficiency (EE): Results in Table (6) indicated that laying hens exposed to NLT (8-18°C) for 12 weeks impaired EE (48.1%). Dietary supplementations improved EE values by 33.6, 21.3 and 10.2% for groups fed basal diet with AA+Cr, AA and Cr, respectively, as compared with that of the control group which exposed to NLT. This improvement could be due to reduce the amount of feed intake required to produce one unit of egg mass.

Table 6. Economic efficiency of Dokki₄ laying hens reared during winter season as affected dietary supplemental AA or Cr.

Items	(+) Cont. (25°C)	Low temperature treatments (8 -18°C)			
		(-) Cont.	+AA 250ppm	+Cr 400ppb	+ (AA+Cr)
Price / kg feed, LE	1.500	1.500	1.515	1.600	1.615
Total feed cost / h, LE	12.690	13.875	13.590	14.368	13.873
Total revenue / h, LE*	22.00	19.16	19.87	20.4	20.93
Net revenue / h, LE	9.31	5.285	6.28	6.032	7.057
EE, %	73.37	38.09	46.21	41.98	50.87
Relative EE, %*	100	51.91	62.78	57.22	69.33

* EE (Economic efficiency) = [total revenue (number of newly healthy hatched chicks × its price (1.20 LE) + (useless eggs for incubation × its price (0.30 LE)) per hen – total feed cost (total feed intake × its price, LE/hen) + total feed cost] × 100.

In the present study, the magnitude of the increases of performance was greater with a combination of AA and Cr rather than with each of them separately. These results revealed additive effects, indicating that AA and Cr work together or act synergistically. Similarly, Seaborn *et al.* (1994) found an interaction between Cr and AA on bone and brain Mn retention and distribution in guinea pigs, and stated that dietary Cr may influence AA metabolism via protecting ascorbate from oxidative destruction. In addition, insulin is known to play a role in AA transportation in RBC's, and glucose competitively inhibits AA transport (Mann and Newton, 1975). Through increasing the effectiveness of insuline, Cr indirectly promotes the AA transportation (Seaborn *et al.*, 1994). Based on the present results, it may be conclude that dietary supplementation with a combination of 250 ppm AA and 400 ppb of Cr provided the highest positive effect on the performance and EE of local Dokki₄ chickens under winter condition in Egypt.

REFERENCES

- Abraham, A.S.; B.A. Brools and U. Eylath (1991). Chromium and cholesterol-induced atherosclerosis in rabbits. *Ann Nutr. Metab*, 35:203-215.
- Aitken, R.J.; J.S. larkson and S. Fishel (1989). Generation of reactive oxygen species, lipid peroxidation, and human sperm function. *Biology of Reproduction*, 40:183-197.
- Akiyama, M. (1999). *In vivo* scavenging effect of ethylcysteine on reactive oxygen species in human semen. *Nippon Hinyokika Gakkai*. 90:421.
- Anderson, R.A. (1987). Chromium. *Trace Elements in Human and Animal Nutrition*. Academic Press, New York. 225-244.
- Bains, B.S. (1996). Role of vitamin C in stress management. *World Poult. Sci*, 12:4-12.
- Baker, H.W.; J. Brindle; D.S. Irvine and R.J. Aitken (1996). Protective effect of antioxidants on the impairment of sperm motility by activated polymorph nuclear leukocytes. *Fertil. Steril.*, 65: 411-419.
- Brown, K.L. and K.E. Nestor (1973). Physiological response of turkey selected for high and low adrenal response to cold stress. *Poult. Sci.*, 52: 1948.
- Castellini, C.; P. Lattaioli; M. Moroni and A. Minelli (2000). Effect of seminal plasma characteris on fertility of rabbit spermatozoa. *Anim. Sci.*,63: 275.
- Colgan, M. (1993). Chromium boosts insulin efficiency. In: *Optimum Sports Nutrition*. New York : Advanced Research Press, PP:313
- Duncan, D.B. (1955). Multiple range and multiple F-test *Biometric*, 11:1-42.
- Egyptian Feed Composition Tables for Animal and Poultry Feedstuffs (2001). Technical bulletin 1, central Lab. Feed and Food, ARC, Egypt.
- Eid, Y.; T. Ebeid and H. Younis (2006). Vitamin E supplement reduces dexame-thasone induced oxidative stress in chicken semen. *Br.Poult., Sci.*,47:350
- Eid, Y.; A. Ohtsuka and K. Hayashi (2003). Tea polyphenols reduce glucocorticoid induced growth inhibition and oxidative stress in broiler chickens. *Brit. Poult. Sci.*, 44: 127-132.

- Ensminger, M.E.; J.E. Oldfield and W.W. Heinemann (1990). Feeds and Nutrition. The Ensminger Publishing Company, USA, pp. 593-666.
- Gallaher, D.C.; A.S. Sallany; D.W. Shoeman and J.M. Olson (1997). Diabetes increase excretion of urinary malonaldehyd conjugate in rat. *Lipid* 28:663
- Gursu, M.F.; M. Onderci; F. Gulcu and k. Sahin (2004). Effects of vitamin C and folic acid supplementation on serum paraoxonase activity and metabolites induced by heat stress in vivo. *Nutr. Res.*, 24:157.
- Halliwell, B. and J.M.C. Gutteridge (1989). *Free Radicals in Biology and Medicine*. 2nd ed. Oxford University Press, New York.
- Kucuk, O.; N. Sahin; K. Sahin; M.F. Gursu; F. Gulcu; M. Ozcelik and M. Issi (2003). Egg production, Egg quality and lipid peroxidation status in laying hens maintained at a low ambient temperature (6°C) and fed a vitamin C and vitamin E supplemented diet. *Vet. Med. Czech*, 48:33.
- Kutlu, H. R. and J.M. Forbes (1993). Changes in growth and blood paramenters in heat – stressed broiler chicks in response to dietary ascorbic acid. *Livest. Product Sci.*, 36:335-350.
- Lake, P.E. and J.M. Stewart (1978). Artificial insemination in poultry in: Ministry of Agric., Fisheries and Food Bulletin No.213, (London, H.M. Stationery).
- Lien, T.F.; Y.M. Horng and S. Yangkh (1999). Performance, serum characteristics, carcass traits and lipid metabolism of broilers as effected by supplement of chromium picolinate. *Brit. Poult. Sci.*, 40:357.
- Lindeman, M. D. (1996). Organic chromium – the missing link in farm animal nutrition. In: Lyons T. P., Jacques K. A. (eds.) : *Biotechnology in the feed Industry: Proceedings of Alltech's 12th Annual symposium*. Nottingham Univ. Press, Nottingham, UK., pp:299-314.
- Mann, G.V. and P. Newton (1975). The membrane transport of ascorbic acid. Second conference on vitamin C. *Ann. NY Acad. Sci.*, 258: 243.
- Matkovics, B.; I. Szabo and I.S. Varga (1989). Determination of enzyme activities in lipid peroxidation and glutathione pathways (in Hungarian). *Laboratiumi Diagnosztika*, 15:284-249.
- McDaniel, C.D.; J.E. Hood and H.M. Parker (2004). An attempt at alleviating heat stress infertility in male broiler breeder chickens with dietary ascorbic acid. *international J. of Poult. Sci.*, 3: 593-602
- Melrose, D. R. and J.A. Laing (1970). *Fertility and Infertility in the Domestic Animals*, 2nd ed. (London, Bailliere Tindall).
- Metwally, M.A. (2004). Effect of dietary vitamin C on the performance of Dandarawi hens subjected to cold stress. *Egypt. Poult. Sci.*, 24: 465.
- Mowat, D.N. (1994). Organic chromium. A new nutrient for stressed animals. *Biotechnology in the feed Industry: Proceedings of Alltech's tenth animal symposium*, Lyons, TP and Jacques KA (Ede). Nottingham University Press, Nottingham, UK, pp: 275-282.
- National Research Council, NRC (1997). The role of chromium in animal nutrition. National Academy Press, Washington, D.C.
- Okada, S.; H. Tsukada and H. Ohba (1984). Enhancement of nucleolar RNA synthesis by chromium (III) in regenerating rat liver. *Biochem.*, 21, 113.

- Orban, J.I.; D.A. Roland; K. Cummins and R.T. Lovell (1993). Influence of large doses of ascorbic acid on performance, plasma Ca, bone characteristics and egg shell quality in broiler and leghorn hens. *Poult. Sci.*, 72: 691.
- Pardue, S.L. and J.P. Thaxton (1986). Ascorbic acid in poultry. A Review. *World's Poult. Sci.*, 42:107-130.
- Placer, Z.A.; L.L. Cushman and B.C. Jahnson (1966). Estimation of products of lipid peroxidation in biochemical systems. *Anol. Biochem.*, 16:329.
- Ravie, O. and P.E. Lake (1985). The phospholipid-bound fatty acids of fowl and turkey spermatozoa. *Anim. Reprod. Sci.*, 9:189.
- Sahin, K.; O. Kucuk; N. Sohin and O. Ozbey (2001). Effect of dietary chromium picolinate supplementation on egg production, egg quality and serum concentrations of insulin, corticosterone and some metabolites of Japanese quails. *Nutrition Res.*, 21:1315-1321.
- Sahin, K. and O. Kucuk (2001). Effect of vitamin C and vitamin E on performance, digestion of nutrients, and carcass characteristics of Japanese quails reared under chronic heat stress (34° C). *J. Anim. Physiol. Anim. Nutr.*, 85 : 335-342.
- Sahin, K. and N. Sahin (2001). Optimal dietary concentration of vitamin C and Chromium picolinate for alleviating the effect of low ambient temperature (6°C) on egg production, some egg characteristics, and nutrient digestibility in laying hens. *Vet. Med. Czech*, 46:229-236.
- Sahin, K.; O. Kucuk; N. Sahin and M. Sari (2002). Effects of vitamin C and vitamin E on lipid peroxidation status, serum hormone, metabolite, and mineral concentration of Japanese quail reared under heat stress (34°C). *Int. J. vitam. Nutr. Res.*, 72: 91-100.
- Sands, J.S. and M.O. Smith (1999). Broilers in heat stress conditions : effects of dietary manganese and chromium picolinate supplementation. *J. Appl. Poult. Res.*, 8: 280-287.
- SAS Institute (1996). SAS User's Guide. Statistics Version 7. Inc., Cary, Nc.
- Seaborn, C. D.; N. Cheng; B. Adeleye; F. Owens and B.J. Stoecker (1994). Chromium and chronic ascorbic acid depletion effects on tissue ascorbate, manganese, and 14 C retention from 14 C – ascorbate in guinea pigs. *Biol. Trac Elem. Res.*, 41: 274-285.
- Siegel, H. S. (1995). Stress, strains and resistance. *Br. Poult. Sci.*, 36:3.
- Spinu, M., and A.A. Degen (1993). Effect of cold stress on performance and immune response of White leghorn hens. *Br. Poult. Sci.*, 34:177.
- Stele, N.C. and R.W. Rose (1981). Effect of trivalent chromium on hepatic lipogenesis by the turkey poultry. *Poult. Sci.*, 60: 617-622.
- Surai, P.F.; G. Wishart; B. Speake; R. Noble; A. Macpherson; N. Sparks; I. Ionov and I. Kostyuk (1997). Effects of vitamin E and selenium in the cocker's diet on lipid peroxidation in spermatozoa. *Brit. Poult. Sci.*, 38:S54
- Watson, D. (1960). Determination of cholesterol. *Clin. Chem. Acta.*, 5 :637.
- Weser, U. and U.J. Koolman (1969) Untersuchungen proteinbiosynthese in Rattenleber-zellerkernen. *Hoppe Seyler's, physiol. Chem.*, 350:1273
- Whitehead, C.C. and T. Keller (2003). An update on ascorbic acid in poultry *World's Poultry Science Journal*, 59: 161–184.

- Young, L.E.; K.D. Sinclair and I. Wilmut (2000). Large offspring syndrome in cattle and sheep. Rev. Reprod., 3:155-63.
- Yousef, M.I.; G.A. Abdallah and K.I. Kamel (2003). Effect of ascorbic acid and vitamin E supplementation on semen quality and biochemical parameters of male rabbits. Anim. Reprod. Sci., 76: 99-111.

تأثير الكروم وحمض الاسكوربيك فى العلف على الأداء الإنتاجى والتناسلى لدجاج نقى، تحت ظروف الشتاء فى مصر

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اجرى هذا البحث بغرض دراسته تأثير اضافة الكروم وحمض الاسكوربيك فى العلف على الأداء الإنتاجى والتناسلى لدجاج نقى، تحت ظروف الشتاء (ديسمبر - فبراير) فى مصر بمحطه بحوث الانتاج الحيوانى بسخا- معهد بحوث الانتاج الحيوانى. استخدم فى هذه الدراسه عدد ٣٠٠ طائر (٢٥٠ دجاجة بياضه + ٥٠ ديك) من سلالة ندى، عمر ٣٠ اسبوع وتم تقسيمها الى ٥ مجاميع بكل مجموعة ٥ مكررات وكل مكررة (١٠ دجاجات + ١ ديك) أما ٢٥ ديك المتبقية قسمت أيضا الى ٥ مجاميع وكل مجموعة ٥ ديوك تم تسكينها فرديا لجمع السائل المنوى وتقييمه. وتم وضع المجموعة الاولى فى حجرة التحكم الحرارى عند درجة الحرارة العادية ٢٥°م ورطوبه نسبيه ٦٤±٢% (كنترول ايجابى) وتم تغذيتها على علف الاساس بدون اضافات و الذى تم تكوينه على اساس الذره الصفراء وكسب فول الصويا ليحتوى على ١٦,٤% بروتين خام و ٢٧٥٠ كيلو كالورى طاقه ممتله/كجم علف. وتم تعريض المجاميع من ٢ الى ٥ لدرجة الحرارة المنخفضة الطبيعىه خلال فصل الشتاء (متوسط درجة الحرارة العظمى نهارا ١٨°م، والذنيا ليلا ٨°م) مع رطوبة نسبية ٦٥±٢% وتم تغذية تلك المجاميع الاربع اما على علف الاساس بدون اضافات (كنترول سالب تحت ظروف الحرارة المنخفضه) أو علف الاساس مضافا اليه ٢٥٠ ملجرام حامض الاسكوربيك/كجم علف أو ٤٠٠ ميكروجرام كروم (من بيكولينات الكروم)/كجم علف أو ٢٥٠ ملجرام حامض الاسكوربيك + ٤٠٠ ميكروجرام كروم/كجم علف. ادى انخفاض درجة الحرارة خلال فتره التجربه الى انخفاض جوهريا فى كل من معدل التغير فى وزن الجسم وعدد البيض الناتج وكتلة البيض ومعدل الانتاج ونسبه الفقس على اساس البيض المخصب وحركه الحيوانات المنويه والاستجابه المناعيه ضد كرات الدم الحمراء للغم بعد الحقن بسته ايام وتسعه ايام وذلك بمقدار: ٨,٦، ١٢,٩، ١٣,٥، ١٨,٦، ٤,٦ و ٥,٧ و ١٣,٦، ٢٥,٨، ٤٨,١% على التوالي وتدهورا فى كل من كفاءه تحويل العلف الى بيض والكفاءه الاقتصاديه بمقدار ٢٨,٩، ٢٨,١ و ٤٨,١% على التوالي وزياده فى كل من العلف الماكول ومحتوى بلازما الدم من الكلسترول والجلوكوز وتركيز المالبونالدهيد فى كل من بلازما الدم والسائل المنوى وذلك بمقدار: ٩,٣، ٢١,٣ و ١٢,٢ و ١٠,٧ و ٥٤,١% على التوالي وذلك بالمقارنه مع مجموعه الكنترول الايجابى المرياه فى درجة الحرارة العادية. الاضافات الغذائيه تسببت فى تخفيف او تلاشى اثار الحرارة المنخفضه على اداء الدجاج. سجلت الطيور المغذاه على علف الاساس مضافا اليه حامض الاسكوربيك مع الكروم افضل الاضافات وحسنت جوهريا اثار الحرارة المنخفضه فى كل من معدل التغير فى وزن الجسم والكفاءه الغذائيه والكفاءه الاقتصاديه وعدد البيض الناتج وكتله البيض ومحتوى بلازما الدم من الكلسترول والجلوكوز وتركيز المالبونالدهيد فى السائل المنوى والاستجابه المناعيه بعد ٦ ايام و ٩ ايام وحركه الحيوانات المنويه ونسبه الفقس على اساس البيض المخصب وذلك بمقدار ٥,٩، ١٧,١ و ٣٣,٦ و ٩,٥ و ٥,٣ و ١٠,٩ و ١٠,٢ و ٢٥,٨ و ١٩,٨ و ٣٨,٣ و ٦,٢ و ٥,٢% على التوالي وذلك بالمقارنه مع مجموعه الكنترول السلبى المرياه فى درجة الحرارة المنخفضه. لا توجد اختلافات معنويه بين المجموعات فى معظم صفات السائل المنوى ودرجه حراره الجسم وخصوبه البيض ونسبه الفقس على اساس البيض الكلى ووزن الكتاكيت الفاقسه. وتخلص هذه الدراره الى ان اضافة كل من حامض الاسكوربيك بمعدل ٢٥٠ ملجرام والكروم بمعدل ٤٠٠ ميكروجرام معا لكل كجم علف اعطت تأثير ايجابيا على الأداء الإنتاجى والتناسلى والكفاءه الاقتصاديه لدجاج سلالة ندى، المرياه تحت ظروف درجة الحرارة المنخفضة خلال فصل الشتاء فى مصر.