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Iodine-enriched egg production in response to dietary iodine in laying hens

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This study was focused on the impact of dietary iodine on laying hens' performance, iodine yolk, and albumen concentration. A total of 180 layers 32-weeks old layers were distributed into 5 treatments with 4 replicates and 9 hens in each. The diets were supplemented with 0, 5, 10, 15 and 20 mg/kg dietary calcium iodated. Maximum increase in body weight had shown in treatment of 15 mg/kg iodine and treatments of 0 and 20 mg/kg iodine resulted in minimum increase in body weight ($P<0.05$). Egg weight increased by treatments 10 and 15 mg/kg dietary iodine ($P<0.05$). Treatment of 10 mg/kg iodine supplementation resulted in highest egg length and unit surface shell ratio and lowest egg shape index. The lowest numerical value for albumen height was indicated by in treatment 5 mg/kg iodine supplementation. In treatments of 5 and 20 mg/kg iodine supplementation calcium iodated, highest value was obtained in albumen index. Highest yolk height was shown by treatments 5 and 10 mg/kg dietary iodine. The highest yolk index was resulted by treatment 5 mg/kg and lowest value was achieved by treatments 0 and 10 mg/kg dietary iodine. Minimum yolk cholesterol was found by 10 mg/kg dietary iodine ($P<0.05$). No response was observed in treatments in terms of plasma cholesterol ($P>0.05$). Significant increases were appeared among treatments in yolk, albumen, and plasma iodine concentration. It is concluded that iodine egg production and better performance can be obtained by 5 and 10 mg/kg of dietary calcium iodate without any adversely effects.

Key words: Iodine, Yolk, Albumen, Egg, Performance

Introduction

Iodine is an essential micronutrient element for human and animal and it is required for the synthesis of thyroid hormones thyroxine, triiodothyronine and iodinated molecules of the amino acid tyrosine. Although the principal function of thyroxine is the control of cellular oxidation, significant roles are also played in the pituitary gland and gonads. For example, thyroxine in some

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way facilitates the dissipation of juvenile, and the initiation of adult, photorefractoriness in turkeys, and possibly in broiler breeders (Proudman and Siopes, 2002). Iodine deficiency reduces the production of thyroid hormones in humans and animals, leads to morphological and functional change of the thyroid gland and reduction of the formation of thyroxin (Iccidd, 2001).

Iodine supplementation in farm animal can lead to prevention of iodine deficiency in humans. Egg iodine content is variable depending on its intake through a diet including a high amount of iodine. In experiment conducted by Kroupova *et al*. (1999), iodine accumulated in the yolk was more than albumen. Kaufmann *et al*. (1998) stated that iodine intake by 0, 0.5, 1, 2 and 5 mg/kg in diet, did not influence feed efficiency and egg production. They also found that iodine content in egg increased significantly by iodine supplementation. No significant differences were found in body weight, feed consumption, egg yield, egg shell index, shell thickness and egg yolk index, by 0, 3, 6, 12 and 24 mg/ kg^{-1} iodine as dietary calcium iodate (Yalcin *et al*., 2004).

Lichonikova *et al*. (2003) reported that significant effects on Hough units, yolk index and egg shell weight as 6.1 mg/kg iodine compared to 3.6 mg/kg. Christensen *et al*. (1991) stated that a study with two commercial turkey strains, found that both high levels of iodine (0.7 and 4.2 mg / kg in diet) and hen's breed influenced hatchability of turkey eggs. An interaction of 4.2 mg iodine kg^{-1}, with the breeder strain was observed concerning the body weight of embryos. Data in this study may indicate that iodine tolerance in turkey hens can be lower than 4.2 mg iodine kg^{-1}. Numerous measurements have been undertaken to improve the iodine supply to human diets, for example iodized salts (Lind *et al*., 2002; Zimmermann, 2004), supplementation of foods with animal and plant origin iodine (Schöne *et al*., 2003; Zimmermann *et al*., 2005). Other vehicles are reported by Dunn (2003) with addition of iodine in feed of farm animals to increase iodine content in animal products (Kaufmann and Rambeck, 1998; Flachowsky *et al*., 2006; Schöne *et al*., 2006). Iodine transfer from feed into eggs was also much higher (10-20%) than from feed into meat as shown by Richter (1995) and Yalcin *et al*. (2004).

During the last few years, quality of iodine nutrition in some European countries has developed (Lind *et al*., 2002; Thamm *et al*., 2007) but there are still questions regarding the methods of iodine supplementation. However, no analysis are carried out on iodine content of the feed. Since differences observed were small and not always significant, a clear overall picture could not be obtained. The attention to limitations on the consumption of iodized salt in children, elderly people and residents of regions with limited access to marine foods reach in iodine makes it necessary to add it to human food as in iodized eggs. These reflections (various methods of iodine supplementation, amount iodine content in diet and iodine original sources) and conflicting results by several researchers are reasons for arranging the test of iodine in the diet of laying hens in various regions of Iran with different condition such as in Hamedan province which cold environment and height altitude (1850 m) for enriched egg production.

Materials and methods

A total of 180 White Leghorn layer (W36 Hy- line) 32 weeks of age were chosen and they were randomly distributed into 5 treatments with 4 replicates and 9 hens per each. A sixteen-hour light regime was adopted during the whole experiment and climatic conditions (temperature, moisture) were monitored.

Feed and drinking water were provided *ad libitum* at all times. The composition of the feed and the nutrient contents (formulated by NRC recommended (1994)) are shown in Table 1. The basal diet iodine (I_2) was measured (0.824 mg/kg). Diets were supplemented by calcium iodated Ca $(IO_3)_2$, with 0, 5, 10, 15 and 20 mg/kg iodine content. Therefore dietary iodine rations (I_2) content were calculated in 0.824, 2.014, 3.204, 4.394, and 5.584 mg/kg (I_2) (Table1). The experiment was conducted for 10 weeks. During the experimental period, the eggs were collected daily and weighed individually. Egg production was calculated on a hen-day basis. Feed consumption was evaluated biweekly.

In five series during the experiment, 6 eggs from each replicate were collected randomly and during a 16h period, weighed and analysed for egg quality characteristics. The yolk height, albumen height, yolk index, albumen index, and Haugh units were recorded. A total of 60 eggs (12 eggs from each treatment) were collected randomly at the end of the experiment for iodine and cholesterol measurement. The iodine concentration in egg yolk and albumen was determined by a spectrophotometer method (440 nm) using alkaline ashing based on the Sandell-Kolthoff reaction (Bednar *et al*., 1964). The principle of the assessment was the reduction of Ce^{4+} to Ce^{3+} in the presence of As^{3+}, and the catalytic effect of iodine. Mineralization took place in an alkaline medium at 600°C. In the present method, organic sample fades away as the result of high temperature and iodine released from the organic compound attaches to alcoholic group. The resulting product is not soluble in acid and thus the iodine content can be determined chromatographically. In this method, iodine catalyzes a reductive oxidation reaction. Total iodine, i.e. both inorganic and protein-bound iodine, was measured in this way. Yolk cholesterol was determined with modifications using the cholesterol assay kit (Pars Azmoun, Karaj, Iran, CHAD-PAP 2009).The kit contained cholesterol assay and standard cholesterol solution, used for calibrate curve.

All data were analyzed using the GLM procedure (SAS Institute, 2004). Variables with significant F tests ($P< 0.05$) were compared using Duncan's multiple range tests. Moreover, an orthogonal polynomial regression analysis was performed for the data.

Table 1. Rations ingredient and calculated nutritional levels in treatments

Composition (g/kg)	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5
maize	480	480	480	480	480
Soybean meal	180	180	180	180	180
gluten	52.6	52.6	52.6	52.6	52.6
Wheat	150	150	150	150	150
Soybean oil	30	30	30	30	30
Dicalcium Phosphate [1]	13.6	13.6	13.6	13.6	13.6
Oyster shell [2]	84.8	84.8	84.8	84.8	84.8
NaCl [3]	3	3	3	3	3
Mineral premix [4]	2.5	2.5	2.5	2.5	2.5
vitamin premix [5]	2.5	2.5	2.5	2.5	2.5
Dl-Methionine	1	1	1	1	1
$Ca(IO_3)_2$ [6]	0.0000	0.0005	0.001	0.0015	0.0020
Metabolisable energy (MJ)	12.1417	12.1417	12.1417	12.1417	12.1417
Crude Protein (%)	16.72	16.72	16.72	16.72	16.72
Calcium [7](%)	3.6	3.6	3.6	3.6	3.6
Available Phosphorus [7] (%)	0.36	0.36	0.36	0.36	0.36
Sodium [7] (%)	0.17	0.17	0.17	0.17	0.17
Methionine (%)	0.35	0.35	0.35	0.35	0.35
Lysine (%)	0.76	0.76	0.76	0.76	0.76

[1]Contains of 18.7% phosphorus and 22% calcium.[2] Contain of 38% calcium. [3] Contain of 39% sodium.[4] Mineral premix in 1 kg contain: Mn (oxide) 64 gr, Zn (oxide) 44 gr, Fe (sulphate) 100 gr, Cu (sulphate) 16 gr, I (calcium iodated) 0.5 gr, Co 0.2 gr, Se (1%) 0.8 gr.[5] Vitamin premix in 1 kg contain: vitamin A 7.2 gr, Vitamin D3 7 gr, Vitamin E (Alfatocopherol) 14.4 gr, Vitamin K3 1.6 gr, Thiamine 0.72 gr, riboflavin 3.3 gr, Pantothenic acid 12 gr, niacin 12.16 gr, Pyridoxine 6.2 mg, Cobalamine 0.6 gr, Biotin 0.2 gr, Colin- Chloride 440 mg.[6] Contain of 65.21% I and 34.78% Ca.[7] Calculated by tables of NRC (1994).

Results

There were no significant differences among the treatments in feed consumption, feed conversion ratio, egg production, egg mass, broken and soft-shelled-eggs (Table 2). No differences were observed on initial and final body weight in treatments (Fig.1). In contrast maximum increased in body weight has shown by treatment of 15 mg/kg iodine as of Ca $(IO3)_2$. While minimum increased in body weight was indicated in treatments 0 and 20 mg/kg iodine supplementation as of Ca $(IO3)_2$ (mg/kg) (Fig.2). There were no significant differences in ratio egg production to protein intake in treatments (Fig.3). Egg weight increased by treatments of 10 and 15 mg/kg supplementary iodine as of Ca $(IO3)_2$ (Table 2).

There were no significant differences among treatments in terms of egg width, egg surface area, yolk diameter, shell weight and shell ratio (Table 3). Highest values for egg length and unit surface shell weight and the lowest value for egg shape index were obtained by 10 mg/kg iodine as of Ca $(IO3)_2$. The lowest value for albumen height was indicated by 5 mg/kg iodine supplementation. Highest value was found in albumen index by both treatments 5 and 20 mg/kg iodine supplementation. Highest yolk height was achieved by both treatments 5 and 10 mg/kg iodine supplementation in contrast lowest recorded was shown by treatments 0 and 20 mg/kg iodine supplementation in this respect. The highest yolk index and Haugh unit were observed in treatment 5mg/kg iodine supplementation. Yolk index and Haugh unit were appeared in lowest rate by treatments 0 and 10 mg/kg iodine supplementation. While Specific gravity in treatment 10mg/kg iodine indicated significant decreased compared to other treatments (Table 3). Minimum yolk cholesterol was shown by treatments receiving 10mg/kg iodine supplementation. No respond were shown among treatments in plasma cholesterol (Fig.4). Significant increases were seen among treatments in yolk, albumen and plasma iodine concentration (Table 4).

Table 2. The effect of iodine supplementation on performance of laying hens

Treatment: Iodine Supplementation levels as of Ca $(IO3)_2$ (mg/kg)							**Polynomial analysis P-value**			
	0	5	10	15	20	SEM	treat	L	Q	C
Feed consumption[1]	91.11	88.22	88.57	90.53	88.72	1.583	0.782	0.696	0.616	0.278
Egg production[2]	75.03	75.59	75.17	76.52	71.62	2.051	0.475	0.379	0.513	0.428
Egg weight[3]	60.87 [c]	61.36[bc]	62.98[a]	62.11[ab]	61.04[c]	0.321	0.002	0.308	0.000	0.215
FCR[4]	1.98	1.91	1.94	1.91	2.02	0.038	0.257	0.326	0.498	0.065
Egg mass[1]	45.67	46.47	46.10	47.56	43.71	1.170	0.264	0.454	0.108	0.282
Soft-shelled- eggs[2]	0.21	0.20	0.42	0.15	0.38	0.104	0.377	0.475	0.904	0.475
Broken eggs[2]	1.62	1.71	2.15	2.15	2.22	0.521	0.717	0.127	0.641	0.759

The same letter in a raw means not significant difference at $\alpha<0.05$. SEM= Standard Error Mean. 1(g/hen). 2 %, 3(g), 4: FCR (kg feed/kg egg). L: linear Q: quadratic C: cubic.

Table 3. The effect of iodine supplementation on egg traits of laying hens

Treatment: Iodine Supplementation levels as of Ca $(IO3)_2$ (mg/kg)							Polynomial analysis P-value			
	0	5	10	15	20	SEM	Treat	L	Q	C
Egg length (mm)	57.84^{ab}	57.23^{b}	59.01^{a}	57.21^{b}	57.29^{b}	0.428	0.007	0.837	0.453	0.714
Egg width (mm)	43.93	43.68	43.46	43.38	43.39	0.210	0.606	0.440	0.589	0.297
Shape Index (%)	76.00^{ab}	77.15^{a}	74.79^{b}	76.26^{a}	^{ab} 75.23	0.511	0.014	0.849	0.508	0.324
Egg Surface Area (cm^2)	89.11	87.58	89.91	87.51	87.59	0.854	0.188	0.276	0.653	0.576
Albumen height (mm)	4.75^{ab}	4.94^{a}	4.57^{b}	4.75^{ab}	4.79^{ab}	0.101	0.020	0.165	0.683	0.637
Albumen Index (%)	14.62^{ab}	15.71^{a}	14.1^{b}	14.71^{ab}	15.51^{a}	0.397	0.001	0.381	0.924	0.024
Yolk height (mm)	17.41^{b}	18.21^{a}	18.29^{a}	17.99^{ab}	17.51^{b}	0.206	0.004	0.082	0.260	0.672
Yolk diameter (mm)	45.06	44.73	44.73	45.77	45.27	0.277	0.526	0.788	0.896	0.870
Yolk Index (%)	39.22	41.3^{a}	40.23^{ab}	40.54^{ab}	39.64^{ab}	0.531	0.011	0.697	0.555	0.805
Specific gravity (%)	1.076^{a}	1.076^{ab}	1.073^{b}	1.075^{ab}	1.075^{ab}	0.009	0.021	0.751	0.437	0.546
Shell weight (gr)	5.54	5.52	5.52	5.58	5.42	0.091	0.905	0.612	0.620	0.585
Shell Ratio (%)	8.73	8.56	8.45	8.64	8.56	0.123	0.614	0.551	0.300	0.457
Unit surface shell weight (mg/cm^2)	0.70^{ab}	0.69^{b}	0.70^{a}	0.69^{b}	0.69^{b}	0.002	0.018	0.352	0.325	0.853
Haugh Unit (%)	82.38^{ab}	85.15^{a}	79.53^{b}	85.55^{ab}	83.29^{ab}	1.27	0.049	0.817	0.966	0.461

The same letter in a raw means not significant difference at $\alpha<0.05$. SEM= Standard Error Mean. L: linear Q: quadratic C: cubic

Table 4. Iodine concentration in yolk, albumen and plasma

Treatment: Iodine Supplementation levels as of Ca $(IO3)_2$ (mg/kg)							Polynomial analysis P-value			
	0	5	10	15	20	SEM	Treat	L	Q	C
Yolk iodine*	0.22^{e}	0.34^{d}	0.58^{c}	0.80^{b}	1.13^{a}	0.011	<0.0001	<0.0001	<0.0001	0.0227
Albumen iodine*	0.019^{e}	0.052^{d}	0.075^{c}	0.091^{b}	0.126^{a}	0.003	<0.0001	<0.0001	0.0458	0.0014
Plasma iodine**	0.332^{e}	3.01^{d}	6.11^{c}	8.17^{b}	9.4^{d}	0.097	<0.0001	<0.0001	0.6384	0.9540

The same letter in a raw means not significant difference at $\alpha<0.05$. SEM= Standard Error Mean. *: µg/g; **mg/L L: linear Q: quadratic C: cubic

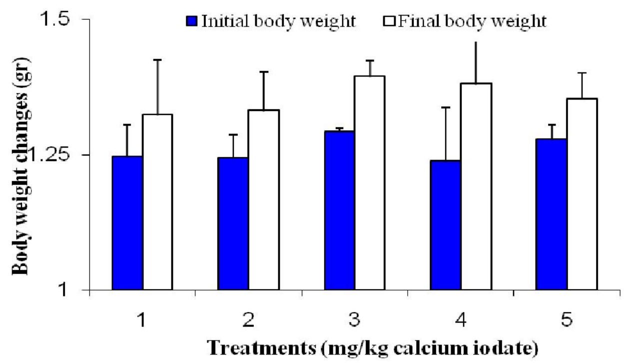

Fig .1. Effect of iodine supplementation on body weight changes
Fig.1. (1: 0% calcium iodate; 2: 0.005% calcium iodate; 3: 0.010% calcium iodate; 4:0.015 calcium ; 5:0.020% calcium iodate)

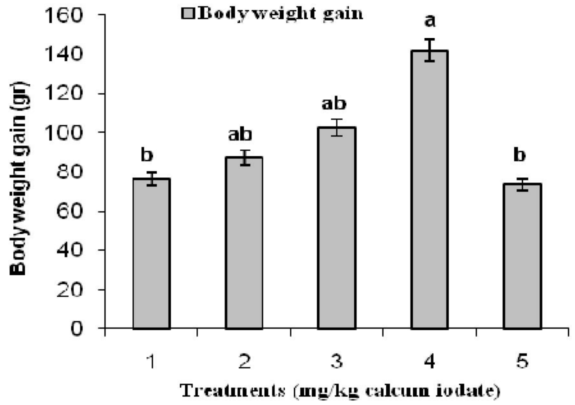

Fig.2. Effect of supplementation of iodine on body weight gain
Fig.2. (1: 0% calcium iodate; 2: 0.005% calcium iodate; 3: 0.010% calcium iodate; 4: 0.015% calcium iodate; 5: 0.020% calcium iodate

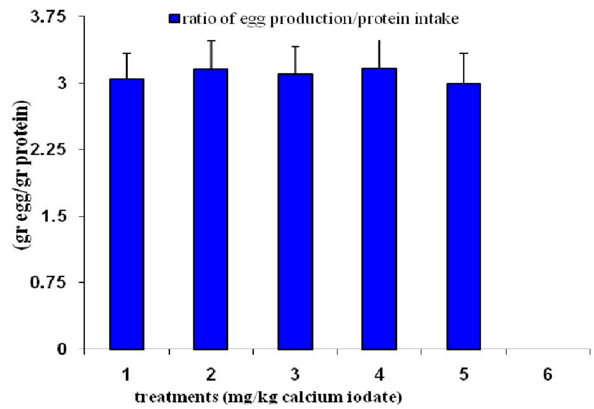

Fig. 3. effect of iodine supplementation on egg production/ protein intake.
Fig.3. 1: 0% $Ca(IO3)2$; 2: 0.005% $Ca(IO3)2$; 3: 0.010% $Ca(IO3)2$; 4:0.015% $Ca(IO3)2$; 5:0.020% $Ca(IO3)2$

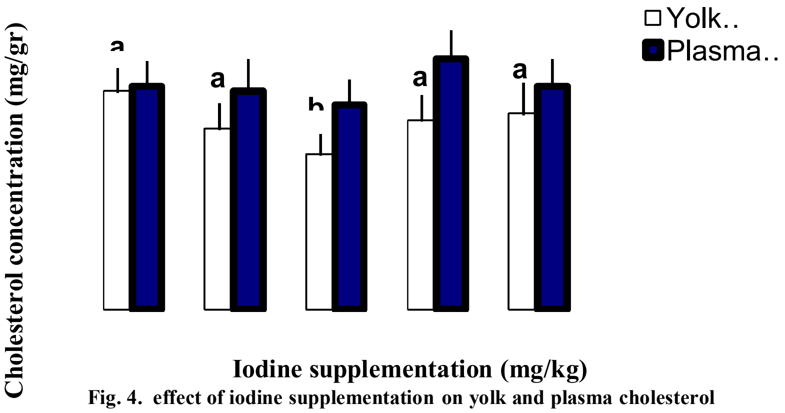

Fig. 4. effect of iodine supplementation on yolk and plasma cholesterol
Fig. 4. (1: 0% Ca(IO3)2; 2: 0.005% Ca(IO3)2; 3: 0.010% Ca(IO3)2; 4:0.015%Ca(IO3)2; 5:0.020% Ca(IO3)2

Discussion

The high levels of dietary iodine did not have any effect on the feed intake and feed efficiency. There were no significant differences in ratio egg production to protein intake in treatments (Fig. 3). In agreement by the present study some researchers reported that iodine supplementation as of Ca $(IO3)_2$ had no significant effect on feed consumption or feed efficiency (Christensen and Ort, 1990; Christensen *et al.*1991; Kaufmann *et al.*, 1998; Lichonikova and Zeman, 2004 and Yalcin *et al.*, 2004). No differences were reported on initial and final body weight in treatments (Fig.1). Some studies have noted different results in this respect (Christensen and ort, 1990; Yalcin *et al.*, 2004. It seems that dietary iodine has no effect in the starting time of the period.

Although iodine supplementation had no significant effect on production of eggs and egg mass, a sharp increase in production was observed in layers by treatment 15 mg/kg iodine as of Ca $(IO3)_2$. Kaufmann *et al.* (1998) stated that mean values of egg production of laying hens were not affected by amounts of iodine to provide 0.5, 1.0, 2.0 and 5 mg/kg. Also Yalcin *et al.* (2004) have noted that no reaction was found in this trait by 0, 3, 6, 12 and 24 mg/kg iodine supplementation as of Ca $(IO3)_2$. Lichonikova and Zeman, (2004) confirmed that higher level of iodine did not have any effect on egg production by 3.57 and 6.07 mg/kg iodine supplementation as of Ca $(IO3)_2$. All these conflicting results were shown by present study. The present study was in agreement with the previous results.

Higher egg weight in this experiment have shown by treatment s of 10 and 15 mg/kg iodine as of Ca $(IO3)_2$ treatments (P<0.05; Table 2). Yalcin *et al*. (2004) have observed also higher egg weight in treatments of 0, 3 and 6 mg/kg iodine supplementation. In the study of Christensen and Ort (1990, 1991), lower egg weights have been reported in turkey diet by 35 and 350 mg/kg iodine. But diets with 2.1 and 3.5 mg/kg iodine had no differences on this factor. Also Christensen *et al*., (1991) have stated that egg weight increased by iodine supplementation in 3.5 mg/kg. They reported an increased in egg weight only in British United Turkeys but no differences in egg weight in Nicholas Turkeys. The present study was in agreement and also disagreement with these results again these conflicting achieved was monitored. No differences were indicated on egg mass in treatments, except the high rate in treatment by 15 mg/kg iodine in this case. Lichonikova and Zeman, (2004), confirmed this result. With respect to broken and soft-shelled-eggs there were no significant differences among treatments. With internal traits of egg, there was no significant differences between treatments in terms of egg width, egg surface area, yolk diameter, shell weight and shell ratio (Table 3). Lichonikova *et al*. (2003) have found significant effects on Haugh units, yolk index and eggshell weight at 6.1 mg iodine kg-1 feed compared to 3.6 mg kg.

In research conducted by Lichonikova and Zeman (2004), treatment receiving 6.07 mg/kg iodine had higher shell weight than treatment receiving 3.57 mg/kg. In a different study, Christensen and Ort, (1990) stated that there were not significant differences in albumen index and haugh unit between turkeys fed with diets containing 2.1 mg of iodine in potassium iodide combined. In research conducted by Yalcin *et al*. (2004), treatment receiving 24 mg/kg calcium iodated had lower egg albumen index and Haugh unit than treatments by 0, 3, 6 and 12 mg/kg supplementary iodine. They also stated that iodine supplementation in diets did not have any significant effects on egg shape index and egg yolk index.

In this study, iodine supplementation as of Ca $(IO3)_2$ had no any effect on cholesterol concentration in plasma, but yolk cholesterol in treatment receiving 10mg/kg iodine supplementation as of Ca $(IO3)_2$ was lower than other treatments (Fig. 3). A regression of data from Perry *et al*. (1989, 1990) have shown that cholesterol in plasma increased by 0.3 mmol/l for each 100 mg/kg increase in dietary iodine concentration. Yalcin *et al*. (2004) have found that there were no significant differences between treatments in terms of cholesterol in yolk. Inconsistent results in this case can be due to differences in the amounts of iodine supplementation, the combined amount of mineral supplement in diet or differences in methods of measuring cholesterol.

Increased albumen and yolk iodine is proportional to level of supplementary iodine in diet ($P<0.05$, Table 4). In agreement with present study, some researchers reported similar results (Kaufmann *et al*., 1998; He *et al*., 2000 and Dobrzanski *et al*., 2001). Supplementing the diet with 5 mg iodine kg-1 increased the iodine content in the egg yolk from 7 to 50 µg iodine egg-1 (Kaufmann and Rambeck, 1998). Souci *et al*., (2000) have reported those iodine average concentrations of 97, 75-158 and 68 µg kg-1 total egg, yolk and egg white, respectively.

Iodine contents of 0.8, 2.9, 5.2, 11.1 and 21.5 mg / kg layer diet resulted in 48, 107, 180, 290 and 511 µg iodine kg-1 egg albumen, and 443, 664, 1122, 1953 and 3352 µg iodine Kg-1 egg yolk, respectively (Yalçin *et al*., 2004). Nakajima *et al*. (1980) did not found any differences in iodine accumulation in eggs during 23 days following a repeated iodine supplementation in feeds (iodized oil from safflower seeds, sea algae and KI). They recorded a sharp increase of yolk iodine concentrations on day 4 following supplementation of KI to the layers. The maximum values, which were found on day 9 to 11, were 60-70 times higher than iodine yolk concentration in the control group. After termination of iodine supplementation, a sharp drop was recorded on day 5, and on day 11 iodine yolk concentration was the similar in the control. Kroupova *et al*. (1999) mentioned that 8-week application of 3.5 mg I/Kg of feed mixture resulted to increase of yolk iodine concentrations to 18597± 1655 µg iodine kg-1 of yolk. Different response to higher iodine doses is affected by different usability of iodine source (Herzig and Suchy, 1996 and Bobek, 1998) and also in the strain (Rys *et al*., 1997) and by the presence of antinutritious substance which have an impact on iodine resorption (Garwin *et al*., 1992 and Garber *et al*., 1993). The level of iodine in egg yolk was higher than its level in albumen. The present study was in agreement with the previous results (Vadujal, 1996; Yalcin *et al*., 2004 and Travnicek *et al*., 2006).

The plasma iodine increased with high levels of iodine consumption as of $Ca(IO3)_2$. Cao *et al*., (1999) have shown that in laying hens fed with diet containing 50 and 100 mg/kg iodine, plasma iodine increased by 1.35 and 2.01 mg/l, respectively. Travnicek *et al*. (2000) fed broilers with diets containing 3.5 and 11.0 mg/kg iodine and recorded 4.9 and 6.5 mg/l plasma iodine, respectively.

Conclusion

The results of this study indicated that supplementation of iodine in the levels of 5 to 10 mg/kg in diet of laying hens (White Leghorn) can supply a portion of iodine requirement. This finding could make a more clear results comparing to the others previous conflicting achieved, particularly in those regions with no access to sea and marine food sources. Further investigation need to be carried out in terms of more details of iodine effects in diet, meat, plasma, egg yolk and fecal on poultry industry production and human requirement.

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