

## ENRICHED EGGS WITH VITAMIN E AND SELENIUM

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### Abstract

*The common techniques of enriching egg with vitamins and minerals by using large amount of them in the feed become very usefull procedure for producing designer eggs. Enriching egg yolk with vitamin E, in egg producing industry, is very popular because of its antioxidative characteristics in humans. But, there is not much investigations for transferring of vitamin E in egg yolk during the second laying cycle of production using old (80 weeks) molted layers. These investigations were important for egg producing farms because of importance of keeping egg quality and improving functional characteristics of them. For this purpose the basic feed with 30 mg of vitamin E/kg and 0.3 mg selenium/kg was used as a control group. The hens from the experimental groups were feed with higher content of vitamin E in two combinations: 1. – 100 mg vitamin E/kg feed and selenium content of 0.3, 0.38 and 0.46 mg/kg and 2. – 230 mg vitamin E/kg feed and selenium content of 0.3, 0.38 and 0.46 mg/kg. Vitamin E concentration in the yolk was increasing linnearly from 1.62 mg/yolk in the control group, on 2.90 mg/yolk in experimental group of hens where the content of vitamin E was enriched to 100 mg vitamin E/kg feed and on 5.58 mg vitamin E/yolk when the amount of vitamin E in feed was increased to 230 mg/kg. The higher content of vitamin E in hen's feed show trend of increasing the egg production and intensity of laying. The similar trend was noticed with increasing of selenium in the feed also.*

**Key words:** enriched eggs, vitamin E, selenium

### INTRODUCTION

In the common practice for enriching of egg yolk with vitamins and minerals during the procedure of producing designer eggs the important place takes enriching with vitamin E and selenium. Enriching with these vitamin and mineral the producers expect positive effects on the antioxidant capacity of egg yolk because vitamin E and selenium have an important role in the antioxidative processes. Also it is expected that their increased amount in feed and their transfer in egg yolk initiate increasing the amount of glutathione peroxides content in yolk which is one of the most effective antioxidative enzyme (SeGSH-Px) [10]. The experiment with the broiler parents fed with higher content of vitamin E (40, 100, 200 mg/kg feed), and selenium (0.2 and 0.4 mg/kg feed) and their combination increased of vitamin E and Se content in eggs, and also noticed an increasing of concentration of glutathione in chicken liver. In the other investigations was

noticed higher vitamin E concentration in eggs from hens fed with enriched vitamin E in feed as: - in ordinary eggs it was found 2 mg vitamin E/100g yolk, but in enriched eggs, 8 mg [8]. From this and the other investigations it was found the possibility of transfer the mentioned vitamin in yolk and the production of designed eggs enriched with vitamin E. Those types of eggs belong to the category of functional food product in human nutriton. All of above mentioned, gives an idea that transferring of vitamin E and the some other supstances from feed to yolk is possible and gives an opportunity for production of designer eggs.

Many authors did a lot of investigations using vitamin E in the diet of layers with the aim to improve the egg quality [7], to improve the antioxidant system [11], to correct lipid oxidation [3], but all of them were working with realitively young hens in the first faze of laying.

Working with older hens after artificial molting (80 weeks and older) where the immune and antioxidative system could be collapsed is extremely rare.

But that type of layers old and molted become very common in our region and the data for transferring of antioxidants from the feed to the eggs are rare. Because this type of egg production become frequent, it was made an decision to examine the possibility of enriching them with vitamin E, using large amount of it and supplementing the feed with selenium.

## MATERIAL AND METHOD

With the aim to answer the established issue it was performed an experiment, in farm condition, with molted laying hens (Hisex Brown) on 80 weeks of age. The procedure of molting was done in the period of 72-78 weeks of age. During the one month experiment the hens were at the peak of laying. In the experiment were involved 70 hens. They were divided in 3 groups: 10 in the control group (30 mg vitamin E/kg and 0.3 mg selenium/kg feed), 30 in first experimental group (100 mg vitamin E/kg feed), and 30 in second experimental group (230 mg vitamin E/kg feed). First and second experimental groups were divided in 3 subgroups (10 hens per subgroup) treated with 0.3; 0.38 and 0.46 mg selenium /kg feed. The hens were accommodated 2 in each cage.

Vitamin E was added in the basal diet as  $\alpha$ -tocopheryl acetate. Selenium was added as mineral selenium in the basic feed, but in first and second experimental group was supplemented (enriched) by selenium yeast with concentration of 800 mg Se/kg. Enriching the basal feed with selenium from Se yeast was 0.08 and 0.16 mg/kg, but the total selenium was 0.38 and 0.46 mg/kg.

The composition of the basic feed (Table 1) in which was added different amount of vitamin E as  $\alpha$ -TA mixed in 2 different vitamin-mineral compositions (premises) shown in Table 2. Selenium yeast was added separately in amount of 100 and 200 grams per tone of basal feed.

Table 1. Composition of the basic feed

Ingredient	%
Maize	54.72
Soybean meal	22.50
Sunflower meal (28%)	5.00
Maize gluten	2.00
Sunflower oil, crude	2.88
Synthetic methionine	0.07
Choline chloride (60%)	0.11
Potassium carbonate	0.31
Sodium bi carbonate	0.40
Bentonal	0.30
Mono calcium phosphate	1.25
Calcium carbonate	9.79
Salt	0.17
Premix	0.50
Total	100.00
ME, Kcal/kg	2750
Crude protein, %	17.8
Lysine, %	0.91
Methionine, %	0.36
Methionine + cistine, %	0.69
Threonine, %	0.63
Tryptophane, %	0.19
Arginine, %	1.12
Calcium, %	4.00
Phosphorus, total, %	0.62
Phosphorus, available, %	0.37
Potassium, %	0.82
Sodium, %	0.21
Chlorine, %	0.17
Electrolyte balance, mEq/kg	249
Selenium mg/kg	0.30

Table 2. Basic composition of vitamin-mineral additives (premises) calculated in kg feed

	Type of premix	
	Premix 1	Premix 2
	BPLX-211	BPLX-212
Vitamin A, IU/kg	15 000	15 000
Vitamin D <sub>3</sub> , IU/kg	3 500	3 500
Vitamin E, mg/kg	30	230
Vitamin K <sub>3</sub> , mg/kg	3	3
Vitamin B <sub>1</sub> , mg/kg	4	4
Vitamin B <sub>2</sub> , mg/kg	6	6
Vitamin B <sub>6</sub> , mg/kg	5	5
Biotin, mg/kg	0.2	0.2
Folic acid, mg/kg	1	1
Niacin, mg/kg	40	40
Pantotenic acid, mg/kg	12	12
Vitamin B <sub>12</sub> , mcg/kg	10	10
Cu, mg/kg	9	9
Mn, mg/kg	100	100
Zn, mg/kg	80	80
Fe, mg/kg	60	60
Se, mg/kg	0.3	0.3
J, mg/kg	0.25	0.25
Carophyl red, mg/kg	30	30
Antioxidant	+++	+++

In the data of Table 2 it is presented that premix 1 was designed to supply 30 mg of vitamin E in kg feed, and the premix 2 to supply 230 mg/kg. With the aim to provide 100 mg, and 230 mg vitamin E in kg feed in the experimental feed was used the different addition of the both premixes. Premix 1 was used in the control feed where it was supplied, 0.3 mg selenium in kg, as in the first combinations of experimental groups where it also was supplied 0.3 mg selenium/kg feed. Enlarging the selenium content in feed on the higher level then 0.3 mg/kg it was done with adding the selenium yeast product named "kvasel", in amount of 0.1 g and 0.2 g in kg feed in experimental groups where the selenium contents was 0.38 and 0.46 mg/kg.

During the realization of this experiment the following productive performances were monitored: - live weight, laying intensity, egg mass (measuring the daily egg production once a week), vitamin E content in yolk at the 10<sup>th</sup>, 20<sup>th</sup> and 30<sup>th</sup> day of the treatment measured in g yolk and presented in 100 g of yolk and one yolk.

For establishing the vitamin E concentration in the egg yolk it was used 6 yolks for every sample, in fresh condition, homogenized and then saponified with stirring in an alcoholic solution of potassium hydroxide. The analytes were then extracted with hexane and washed with water. The organic faze was removed by evaporation and the residue was dissolved in methanol, filtered and then injected into the chromatographic system. The mobile phase used was a 2.5 mM acetic acid-sodium acetate buffer. The flow rate was 1.0 ml/min [2]. The High Performance Liquid Chromatography (HPLC) system used for analysis was Perkin Elmer. The results were expressed in mg/100 g yolk, and in one yolk. Obtained results from the investigation was statistically tested [9].

## RESULTS AND DISCUSSIONS

From the presented data (Tab. 3) it can be seen that the increasing of vitamin E in feed mixture does not influence significant changes on hen's live weight at the end of one month experimental period ( $P>0.05$ ). Supplementing the diet instead vitamin E groups (100 mg/kg) and (230 mg/kg) with

Se-yeast doesn't cause changes in hens live weight either ( $P>0.05$ ).

The selenium yeast supplementation in the feed cause the significant increasing of the egg production. Differences of egg production were significant between the group of 0.30 mg Se/kg feed and 0.38 mg Se/kg, and 0.30 mg Se/kg feed and 0.46 mg Se/kg of feed ( $P<0.01$ ).

Analysis of variants between control group (30 mg vitamin E and 0.30 mg Se/kg feed); vitamin E group (100 mg vitamin E and 0.30 mg Se/kg feed); and vitamin E group (230 mg vitamin E and 0.30 mg Se/kg feed) doesn't have differences between egg production intensity which means that vitamin E supplementation level from 30 mg to 230 mg per kg feed doesn't increase egg production ( $P>0.05$ ). Similar discussion on egg productivity was found in the study [11], but in the other study referenced as [4] and [1] the optimum productivity was noticed when it was given 250 mg vitamin E/kg feed in stress condition because vitamin E increases productivity by preventing cell damage of liver, which is important for egg yolk protein synthesis. Our experiment was designed in normal temperature conditions from 20 to 25°C in the air.

Egg production is affected by level of selenium in the diet [6] from 64.9 % to 72.3% when Se level was enriched from 0.1 to 0.3 % in the feed of broiler breeders.

Yolk  $\alpha$ -tocopherol (Vitamin E) was increasing linearly with the level of supplemented vitamin E in the feed at the 10<sup>th</sup> day of experiment its level on the peak was depending of the concentration in the feed. The lowest level was found in the yolk of control group and the highest at the experimental group (230 mg/kg feed). The differences are significant at the level of  $P<0.01$ . Similar results by same tendency was reported in reference [7].

The hens fed with enriched feed with vitamin E it was found 8 mg vitamin E in 100 g egg content, but in the ordinary eggs the content was 2 mg [8]. These results are hardly comparative with ours because in our experiment was analyzed egg yolk which takes 30 % of whole egg.

The other authors [5] presented that vitamin E content in ordinary eggs was 1.32

mg, and in the enriched was 3.76 mg. These results can be compared with ours because the vitamin E is contained only in yolk, which content in ordinary eggs of our experiment was 1.62 mg, but in the experimental groups it was 2.90 mg and 5.58 mg.

Results from our investigations are presented graphically and clearly demonstrate the tendencies of increasing the vitamin E content in yolk (Figure 1).

Figure 1 show that the vitamin E content in egg yolk is directly depending of concentration of the same vitamin in the feed which was used for hen nutrition. This tendency confidently shows that it is possible to design the quality of table eggs by adding the higher amounts of vitamin E in hen's feed. On this way we may to expect improvement the antioxidative capacity of eggs because of the enlarging the content of this vitamin and selenium, and may to be expected also the increasing of antioxidative enzyme glutathione peroxidase [10].

## CONCLUSIONS

On the basis of the results obtain from the experiment with aged molted laying hens in second egg laying cycle fed with higher concentration of vitamin E in feed mixture (30, 100, 230 mg/kg) it can be concluded the following:

1. The higher vitamin E content in the feed for laying hens enables vitamin E enriched egg production which concentrations in the yolk was 8.4, 14.87 and 29.37 mg/100g yolk, and 1.62, 2.90 and 5.58 mg vitamin E per one yolk, depending to there increasing in the feed.

2. Production of designed eggs – enriched egg yolk with higher contents of vitamin E is possible in farm conditions and it can be produced as an functional products which can satisfied daily requirements of human population on the level up to 50% if the daily consumption is one egg.

Table 3. Concentration of vitamin E in egg yolk from laying hens fed with different concentration of vitamin E and Se in the feed

Indicator	Vitamin E						
	30 mg/kg	100 mg/kg			230 mg/kg		
	Selenium mg/kg						
	0.3	0.3	0.38	0.46	0.3	0.38	0.46
Number of experimental hens	10	10	10	10	10	10	10
Hen's age, weeks	80	80	80	80	80	80	80
Live weight of hens							
- at the beginning, kg	2.18	2.28	2.24	2.23	2.25	2.24	2.28
- at the end, kg	2.26	2.18	2.28	2.40	2.27	2.26	2.23
Egg production							
- intensity, %	82.00	78.00	94.00	96.00	84.00	94.00	92.00
- average egg mass, g	70.97	71.64	72.72	74.18	73.45	69.96	72.63
Vitamin E in the yolk							
- 10 <sup>th</sup> day, mg/100g	-	16.3	16.5	-	21.1	28.4	30.1
- 20 <sup>th</sup> day, mg/100g	6.70	14.7	15.6	13.4	32.4	28.0	31.4
- 30 <sup>th</sup> day, mg/100g	10.20	11.7	15.3	15.9	31.1	25.1	36.6
Average content, mg/100g (vit. E groups)	8.40	14.87			29.37		
Vitamin E in one yolk							
- 10 <sup>th</sup> day, mg/yolk	-	3.21	3.20	-	4.03	5.37	5.73
- 20 <sup>th</sup> day, mg/yolk	1.29	2.89	3.03	2.60	6.18	5.29	5.98
- 30 <sup>th</sup> day, mg/yolk	1.96	2.30	2.97	3.08	5.93	4.74	6.97
Average content, mg/yolk (vit. E groups)	1.62	2.90			5.58		
Yolk mass, g	19.24	19.68	19.42	19.38	19.07	18.89	19.05

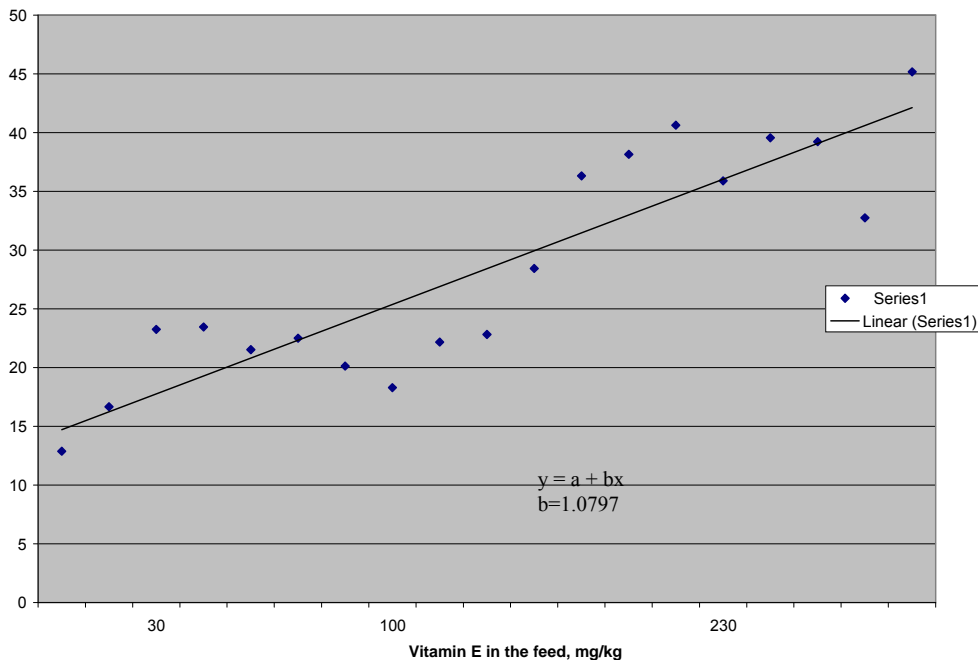


Figure 1. Content of vitamin E in 100g yolk

3. The content of vitamin E in hen's feed had not tendencies of increasing the laying intensity in normal environmental condition, but the higher amount of selenium increase the egg production.

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