Oxidative Stability and Sensory and Functional Properties of Eggs from Laying Hens Fed Supranutritional Doses of Vitamins E and C

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ABSTRACT This study evaluated the effects of two dietary doses of vitamins E and C supplemented separately and together, on the content of vitamin E in the yolk, on the lipid stability of fresh and stored eggs, and on their sensory and functional properties. Hy-Line Brown hens (n = 216) received a basal diet for 8 wk supplemented with 100 or 200 mg DL-α-tocopheryl acetate (E100 or E200, respectively)/kg, 500 or 1,000 mg ascorbic acid (C500 and C1000, respectively)/kg, or 100 mg DL-α-tocopheryl acetate plus 500 mg ascorbic acid (E100+C500)/kg, whereas the control group received no supplementation. Fresh eggs and eggs stored 30, 60, and 90 d at 4 C or stored 28 d at room temperature were analyzed for vitamin E content and TBA-reactive substances (TBARS). We also evaluated functional properties of fresh and cooked eggs and sensory properties of boiled and scrambled eggs. The yolk content of vitamin E depended on the level of dietary addition and decreased after 90 d of storage at 4 C or after 28 d at 25 C. Vitamin supplementation had no effect on fresh or refrigerated eggs, whereas 4 wk of storage at room temperature increased TBARS in the control and the group supplemented with the highest doses of vitamins. Ascorbic acid improved Haugh units and elasticity of albumen gels, whereas cohesiveness and hardness of yolk, albumen and whole-egg gels were not affected by dietary treatment. Panelists were not able to distinguish treated eggs from control eggs.

(*Key words*: vitamins E and C, egg, oxidative stability, functional and sensory properties)

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INTRODUCTION

Egg contains over 11% lipids, mainly concentrated in the yolk (about 33 to 35%), whose fatty acids are more unsaturated than those of most animal lipids (Nys, 2001). The egg is an excellent source of essential fatty acid mainly belonging to the n-6 series (linoleic and arachidonic acids) and also contains moderate amounts of n-3 polyunsaturated fatty acids (PUFA), which are essential for many biological functions (Lesckanich and Noble, 1997). The double bonds of unsaturated fatty acids are particularly sensitive to oxidative deterioration and potentially responsible for the formation of peroxides and off-flavors, changes of taste, texture and color, loss of nutrients, and production of toxic compounds (Eriksson, 1987). However, in view of its high content of susceptible lipid constituents, egg yolk is very resistant to oxidative deterioration during extended refrigerated storage (Pike and Peng, 1985). To stabilize animal products, antioxidants—compounds which act as free radical scavengers—are used in feed mills and food industries, added to the feeds or directly to the foods. Alpha-tocopherol is the most active natural antioxidant used in animal feeding: it exhibits an antioxidant activity at low concentration and a prooxidant activity at high concentration (Chen et al., 1998). The addition of α-tocopherol to hen diets increases the content of vitamin E in the egg yolk in a dose-dependent manner (Jiang et al., 1994, Surai et al., 1997, Meluzzi et al., 2000). Tocopherols may also provide health benefits mainly in preventing cancer and coronary diseases (Diplock, 1991, Knekt et al., 1991), so that the incorporation of vitamin E to the egg may both increase the oxidative stability and provide a source of tocopherols useful for human nutrition and health. Ascorbic acid is a water-soluble vitamin, and it is also an antioxidant by contributing electrons to more oxidized molecules; it can also reduce α-tocopheryl radical (Parcker et al., 1979). As poultry fowls are able to synthesize vitamin C and it is not transferred into the egg, a limited number of studies have been carried out on the role of dietary vitamin C. The research attention was mainly focused on the effects

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Abbreviation Key: E100 = basal diet plus 100 mg dL-α-tocopheryl acetate/kg; E200 = basal diet plus 200 mg dL-α-tocopheryl acetate/kg; C500 = basal diet plus 500 mg ascorbic acid/kg; C1000 = basal diet plus 1,000 mg ascorbic acid/kg; E100+C500 = basal diet plus 100 mg dL-α-tocopheryl acetate and 500 mg ascorbic acid/kg; PUFA = polyunsaturated fatty acids; TBARS = TBA-reactive substances.

of the vitamin on improving the egg-shell quality (Pardue and Thaxton, 1986). Even if vitamin C is neither contained nor transferred to the egg, it could play its antioxidative role in regenerating vitamin E in laying hens. Furthermore it is well known that ascorbic acid is required for the hydroxylation of proline residues necessary for the synthesis of pro-collagen (Weiser et al., 1990), also it could be involved in the synthesis of egg proteins. Several papers are in the literature on the effect of vitamin E on oxidative stability of eggs, but there is a general lack of information about the effects of both vitamins E and C on the quality of chicken eggs. This study aims at evaluating the effects of two dietary doses of vitamins E and C, supplemented alone and together, on the content of vitamin E in the yolk, on lipid stability of fresh and stored eggs and on their sensory and functional properties.

MATERIALS AND METHODS

Animal Care and Dietary Treatments

Hy-Line Brown hens (n = 216; 44-wk-old) were randomly housed in laying cages (four birds per cage) in a windowed poultry house at a light regimen of 16 h light:8 h dark. Six groups of 36 hens (nine replicates per group) were randomly assigned to each six dietary treatments. Two groups received a corn-soybean basal diet that met the National Research Council (1994) requirements, supplemented with 100 or 200 mg DL-α-tocopheryl acetate[2] (E100 and E200, respectively)/kg, two groups received the basal diet supplemented with 500 or 1,000 mg ascorbic acid[3] (C500 and C1000, respectively)/kg, the fifth group received a supplementation of 100 mg DL-α-tocopheryl acetate plus 500 mg ascorbic acid (E100+C500)/kg, and the remaining group (control) had no supplementation. Feed and water were provided ad libitum. The trial lasted 8 wk.

Vitamin Analysis

The content of vitamin E was determined on fresh eggs laid after 5 wk of dietary treatment and on eggs stored 30, 60, and 90 d at 4 C as well as on eggs stored 28 d at room temperature (25 C). Five pools of four yolks each per group were prepared at the scheduled time and stored at –40 C until analysis. One gram of egg yolk was extracted and saponified with 30 mL of ethanol:KOH 50% (1:1, vol/vol) and kept overnight under nitrogen gas at room temperature in the dark. Hexane (20 mL) plus butylated hydroxytoulene (1 g/L) and 20 mL of KH_2PO_4 were added to the flask and carefully mixed for 5 min. After 1 h of rest, 5 mL of the upper organic solvent layer were drawn and evaporated with nitrogen gas. The dried material was recovered with 1 mL of ethanol, and 10 μL were injected into an HPLC series 1090,[4] fitted with a Machery-Nagel (C 18-5) column. Samples were eluted with a solution of methanol/water 97:3 (vol/vol) and run isocratically at a flow of 1.5 mL/min. α-tocopherol was read at a wavelength of 292 nm and quantitatively measured using a solution of α-tocopherol[5] as external standard. As the egg does not contain ascorbic acid, no vitamin C analysis was performed in egg matrix.

Measurement of Lipid Oxidation

The lipid oxidation of the yolk was assessed at the same scheduled time of the vitamin E analysis using the same pools. To quantify malondialdehyde and other TBA-reactive substances (TBARS), the TBARS method was performed according to Tarladgis et al. (1960), Rhee (1978), and Marshall et al. (1994). Twenty grams of yolk was carefully homogenized with 30 mL double distilled water and 10 mL of a 0.5% propyl gallate/ethylenediamminetetraacetic acid solution. Of this dispersion 30 g were added to 78 mL double distilled water and 2.5 mL 4N HCl and distilled. Of distillate 4 mL were combined with 4 mL of a 0.02 M solution of 2-thiobarbituric acid in glacial acetic acid and boiled 35 min; then samples were cooled 10 min in cold water and optical density was read at 538 nm on a Jasco 7800 spectrophotometer[6]. The absorbance values were multiplied by 7.8 to correct for incomplete recovery.

Egg Quality and Egg Functional Properties

After 4 wk of dietary treatment, 30 eggs per group stored 1 d at room temperature (20 to 22 C) were individually weighed to calculate the Haugh index following the relation:

$$\text{Haugh Units} = 100 \cdot \log (H - 1.7 \cdot P^{0.37} + 7.57)$$

where H = height of spread egg white in mm and P = egg weight in grams. Two weeks later, eggs were collected to evaluate their functional properties.

The firmness of the yolk membrane was evaluated on 10 eggs per group stored 1 d at room temperature. A special tool (a perforated metal capsule) connected to an Instron Universal Testing Machine 4301[7] equipped with a 100 N load cell at a crosshead speed of 1.67 mm/s was used. For technical reasons the functional characteristics of yolk were evaluated in the groups supplemented with vitamin E and in the control groups too, whereas in groups supplemented with vitamin C, only the characteristics of albumen were tested. Furthermore, functional properties of whole egg gels were evaluated in all groups. Six pools of four eggs each were used to prepare emulsions, and their stability was calculated by measuring

[2]Rovimix E—50 supplied by Hoffmann La Roche, Ltd., Basel, Switzerland.
[3]Rovimix C—EC supplied by Hoffmann La Roche, Ltd., Basel, Switzerland.
[4]Hewlett-Packard, Avondale, PA.
[5]α-Tocopherol (T-3251) supplied by Sigma Chemical Co., St. Louis, MO.
[6]Jasco Corporation. 2967-5 Ishikawa-Cho Hachioji, Tokyo, Japan.
[7]UTM—Universal Testing Machine, Instron International Ltd., High Wycombe, England.

the relative drainage at 60 and 120 min, as reported in Scholtyssek (1995). The foam stability and the foaming ability of the albumen were evaluated according to Scholtyssek (1995). Whole egg, yolk, and albumen coagulums were obtained by heating them separately 35 min in a water bath at 80 C and stored at 4 C until texture profile analysis according to Woodward and Cotterill (1986, 1987) and Shafer et al. (1998). Textural characteristics of the gels (seven samples per group) were evaluated by a dynamometric double compression test (50% compression) using the Instron equipment and a load cell of 100 N according to Woodward and Cotterill (1986, 1987). Hardness, cohesiveness, and elasticity of the samples were determined according to Bourne (1978).

Sensory Evaluation

Sensory properties were assayed by using eggs laid 8 wk after the beginning of the trial and stored 15 d at 4 C. Eggs were boiled by placing in water at room temperature. Then the water was raised to the boiling point, and the eggs were kept in the boiling water for 7 min. Eggs were then cooled to an external temperature of about 40 C and peeled; yolks were separated, divided into four pieces, and stored at room temperature in airtight containers. Scrambled eggs were prepared by using homogenates of yolk and albumen (70 mL albumen and 30 mL yolk) cooked 4 min with continuous mixing in a frying pan on an electric plate. Neither salt nor oil was added. Scrambled eggs were cooled at room temperature. A triangular test (Porretta, 1992) was performed by employing 24 untrained panelists but usual consumers of eggs. According to the test, each panelist was presented with three pieces of samples of which two were identical (the same sample) and the other was different. A randomized design allowed the sampling of eggs in any order. Each panelist was asked to recognize the different sample. In each taste session the following comparisons were made: control vs. E 200; control vs. C1000; control vs. E100+C500.

Statistical Analysis

All data were statistically analyzed by ANOVA using the General Linear Model procedures of the SAS Institute (SAS Institute, 1985), considering as fixed effect the vitamin supplementation. As for vitamin E and TBARS data, also the duration of egg storage was considered as a fixed effect. Percentage values were transformed into arc sin of their square root prior to the statistical analysis. When significant effects were found, mean values were separated by using the Student Newman-Keuls test.

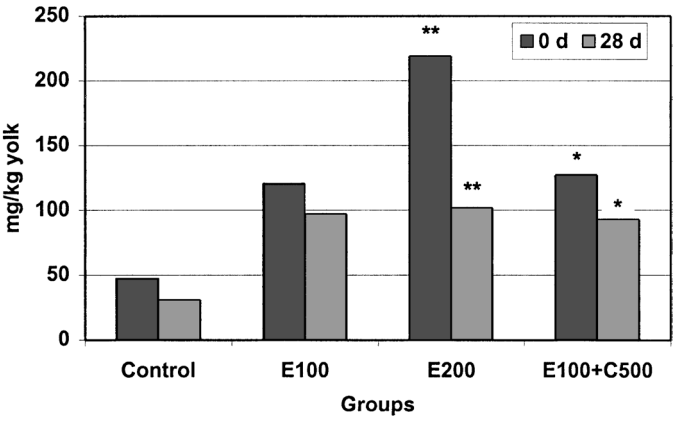

FIGURE 1. α-Tocopherol content of fresh eggs and eggs stored 28 d at room temperature. Asterisks denote values that differed within a given dietary treatment (**$P < 0.01$; *$P < 0.05$). E100 = basal diet plus 100 mg DL-α-tocopheryl acetate/kg; E200 = basal diet plus 200 mg DL-α-tocopheryl acetate/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

RESULTS AND DISCUSSION

Vitamin E Content

The α-tocopherol content of fresh and stored eggs is given in Table 1. At each sampling time, the yolk concentration of vitamin E depended on the dose in hen diets ($P < 0.01$). When vitamin E was administered in the presence of a high dose of ascorbic acid (C500+E100), its levels in the yolk were 5 to 8 mg/g yolk higher than the ones observed when the same dose of vitamin E (E100) was administered alone, but these differences were not statistically significant. Jiang et al. (1994) observed that egg-yolk α-tocopherol content increased linearly as dietary DL-α-tocopheryl acetate increased. Surai et al. (1997) showed a very fast transfer of vitamin E from the diet to the egg yolk of hens fed up to 20,000 ppm vitamin E. The experiments of Gebert et al. (1998) and Meluzzi et al.

TABLE 1. α-Tocopherol content of fresh eggs and of eggs stored at 4 C (mg/kg yolk)

Groups[1]	0 d	30 d	60 d	90 d	SEM
Control	47.2^{C}	32.4^{C}	33.6^{C}	30.6^{C}	7.00
E100	$120.6^{B,X}$	$107.1^{B,XY}$	$81.4^{B,XY}$	$73.0^{BC,Y}$	8.90
E200	$219.1^{A,x}$	$200.1^{A,x}$	$174.7^{A,xy}$	$124.4^{A,y}$	18.57
E100+C500	$127.3^{B,X}$	$112.5^{B,XY}$	$89.3^{B,XY}$	$81.9^{B,Y}$	8.01
SEM	12.14	12.81	10.95	10.28	

$^{A-C}$Means in a column without a common superscript differ significantly ($P < 0.01$).

$^{X-Z}$Means in a row without a common superscript differ significantly ($P < 0.01$).

$^{x-z}$Means in a row without a common superscript differ significantly ($P < 0.05$).

[1]E100 = basal diet plus 100 mg DL-α-tocopheryl acetate/kg; E200 = basal diet plus 200 mg DL-α-tocopheryl acetate/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

TABLE 2. Effect of DL-α-tocopheryl acetate and ascorbic acid supplementations on TBA-reactive substances of fresh eggs and eggs stored at 4 C (mg malondialdehyde/kg yolk)

Groups[1]	0 d	30 d	60 d	90 d	SEM
Control	0.506[b]	0.620[ab]	0.837[ab]	1.009[a]	0.13
E100	0.422	0.555	0.577	0.571	0.79
E200	0.527	0.589	0.588	0.779	0.14
C500	0.517	0.791	0.632	0.743	0.11
C1000	0.513	0.565	0.697	0.689	0.12
E100+C500	0.795	0.902	0.899	0.815	0.69
SEM	0.10	0.11	0.09	0.13	

[a,b]Means in a row without a common superscript differ significantly ($P < 0.05$).

[1]E100 = basal diet plus 100 mg DL-α-tocopheryl acetate/kg; E200 = basal diet plus 200 mg DL-α-tocopheryl acetate/kg; C500 = basal diet plus 500 mg ascorbic acid/kg; C1000 = basal diet plus 1,000 mg ascorbic acid/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

(2000) are in accordance with the previous authors. The content of yolk vitamin E during the storage at 4 C (Table 1) remained quite stable until 60 d of storage; afterward it significantly declined in all the supplemented groups (about 40%). In the control group the vitamin E content quickly declined, about 32% after 30 d of storage, and remained almost stable thereafter. These data agree with those of Cherian et al. (1996b), who observed no effect of a 40-d storage on α-tocopherol content of eggs. In contrast, Gebert et al. (1998) observed that a storage of up to 6 mo at 4 C did not significantly modify the content of yolk vitamin E. Table eggs kept at room temperature (25 C) resulted in a fall of vitamin E content after only 4 wk confirming the importance of low temperature in nutrient preservation of foods (Figure 1).

Lipid Oxidation

In Table 2 the results of yolk TBARS analysis are shown. At any sampling time (0, 30, 60, and 90 d), no differences emerged among supplemented groups and control even if slightly higher concentrations of oxidative products were detected in control eggs and in E100+C500 eggs. In literature there is no agreement on this matter. Gebert et al. (1998) reported that elevated concentration of vitamin E (100 to 200 ppm) increased TBARS number in accordance with Chen et al. (1998), who postulated that vitamin E acted as a prooxidant at the yolk concentration of 75 ppm or above that corresponded to 120 ppm in the diet. Conversely, Cherian et al. (1996a) and Galobart et al. (2001a,b) detected a clear antioxidant effect of dietary α-

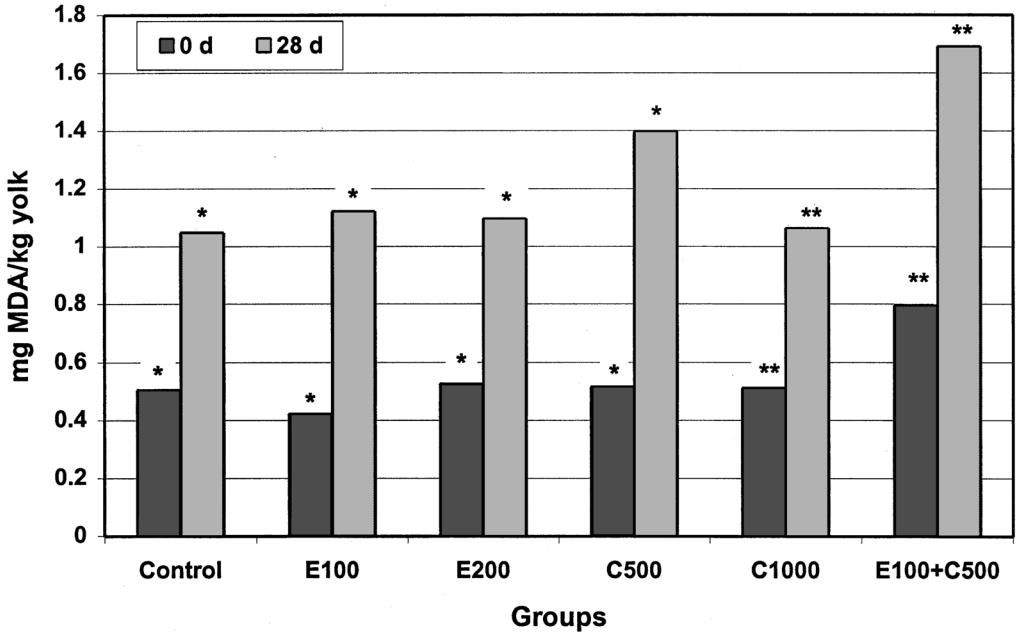

FIGURE 2. Effect of DL-α-tocopheryl acetate and ascorbic acid supplementation on TBA-reactive substances (TBARS) of fresh eggs and eggs stored 28 d at room temperature. Asterisks denote values that differed within a given dietary treatment (**$P < 0.01$; *$P < 0.05$). MDA = malondialdehyde. E100 = basal diet plus 100 mg DL-α-tocopheryl acetate/kg; E200 = basal diet plus 200 mg DL-α-tocopheryl acetate/kg; C500 = basal diet plus 500 mg ascorbic acid/kg; C1000 = basal diet plus 1,000 mg ascorbic acid/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

TABLE 3. Egg weight, Haugh index, and firmness of yolk membrane

Groups[1]	Egg weight (g)	Haugh index (U)	Firmness of yolk membrane (n)
Control	61.1	90.7[ab]	2.93
E100	60.7	88.1[b]	3.28
E200	61.7	89.7[ab]	2.98
C500	61.3	91.6[ab]	3.04
C1000	60.9	93.9[a]	3.21
E100+C500	61.9	88.6[b]	3.11
SEM	0.74	1.33	0.15

[a,b]Means in a column without a common superscript differ significantly ($P < 0.05$).

[1]E100 = basal diet plus 100 mg DL-α-tocopheryl acetate/kg; E200 = basal diet plus 200 mg DL-α-tocopheryl acetate/kg; C500 = basal diet plus 500 mg ascorbic acid/kg; C1000 = basal diet plus 1,000 mg ascorbic acid/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

tocopheryl acetate on n-3 PUFA-enriched eggs. The former authors did not find any change in TBARS values in eggs not enriched with n-3 PUFA, confirming our results. During storage, TBARS were almost stable in all treated groups, whereas in the long-term (90 d) the oxidative products of control eggs significantly increased. The oxidation products increased in all groups after a 4-wk storage at room temperature (Figure 2). The differences appeared remarkable also in presence of high doses of vitamins E and C, confirming the significant effect of temperature (25 C) in lipid stability.

Egg Quality and Egg Functional Properties

Haugh index evaluated on fresh eggs collected after 4 wk of dietary treatment ranged around 90 HU with the highest value (93.9 HU) recorded in group C1000. This value appeared significantly higher than those of groups supplemented with 100 ppm of vitamin E alone or in the presence of vitamin C. No data were found in literature supporting this result; however Benabdeljelil and Jensen (1990) reported that the dietary addition of ascorbic acid restored the interior quality of eggs counteracting the detrimental effects on albumen resulting from 10 ppm of dietary vanadium. No differences emerged between the groups supplemented with the two dosages of vitamin E and the control too (Table 3). This result agrees with those of Cherian et al. (1996b), who observed no effects of dietary added tocopherols on the Haugh units of fresh and stored eggs. Even if the mechanism by which vitamin C improves albumen firmness is unknown, it could be postulated that as ascorbic acid molecule is a cofactor in pro-collagen synthesis, it might also be involved in the albumen protein synthesis and, probably, in albumen firmness. Egg weight was unaffected by a dietary treatment in accordance with Gebert et al. (1998) and Meluzzi et al. (2000). Firmness of yolk membrane, an important trait for industrial separation of yolk and albumen, was not significantly affected by the vitamin supplementation, although higher figures were observed in all treated groups in comparison with control (Table 3). Emulsifying

TABLE 4. Effect of DL-α-tocopheryl acetate supplementation on functional properties of yolk

Groups[3]	Emulsion relative drainage[1] (%)	Emulsion stability[2] (%)	Gel hardness (n)	Gel cohesiveness	Gel elasticity (%)
Control	27.42	5.42	30.17	0.57	92.43[A]
E100	25.75	4.75	33.00	0.57	87.82[B]
E200	24.50	5.25	33.35	0.57	87.43[B]
E100+C500	22.25	4.92	33.86	0.56	89.96[B]
SEM	0.02	0.01	1.58	0.08	0.01[4]

[A,B]Means in a column without a common superscript differ significantly ($P < 0.01$).

[1](Drainage volume at 120 min × 100)/emulsion volume.

[2]Relative drainage at 120 min – relative drainage at 60 min.

[3]E100 = basal diet plus 100 mg DL-α-tocopheryl acetate/kg; E200 = basal diet plus 200 mg DL-α-tocopheryl acetate/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

[4]SEM of values transformed into archsine of the root square of the percentage.

TABLE 5. Effect of ascorbic acid supplementation on functional properties of albumen

Groups[3]	Foam stability[1]	Foaming ability[2]	Gel hardness (N)	Gel cohesiveness	Gel elasticity (%)
Control	0.52[b]	8.23	13.23	0.50	87.74[b]
C500	0.50[b]	8.39	16.72	0.50	91.10[a]
C1000	0.51[b]	8.75	13.96	0.48	89.51[ab]
E100+C500	0.62[a]	8.59	15.64	0.49	90.74[a]
SEM	0.03	0.21	0.95	0.01	0.01[4]

[a,b]Means in a column without a common superscript differ significantly ($P < 0.05$).

[1](Volume of albumen – drainage at 60 min)/volume of albumen.

[2](Weight of 100 mL albumen – weight of 100 mL foam)/ weight 100 mL foam.

[3]C500 = basal diet plus 500 mg ascorbic acid/kg; C1000 = basal diet plus 1,000 mg ascorbic acid/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

[4]SEM of values transformed into archsine of the root square of the percentage.

TABLE 6. Effect α-tocopheryl acetate and ascorbic acid supplementation on functional properties of whole egg

Groups[1]	Gel hardness (n)	Gel cohesiveness	Gel elasticity (%)
Control	20.07	0.53	92.06^{A}
E100	19.93	0.54	91.31^{AB}
E200	19.85	0.54	90.25^{ABC}
C500	18.27	0.53	89.28^{BC}
C1000	18.18	0.54	88.57^{C}
E100+C500	18.98	0.55	90.04^{ABC}
SEM	0.60	0.01	0.009[2]

$^{A–C}$Means in a column without a common superscript differ significantly ($P < 0.01$).

[1]E100 = basal diet plus 100 mg DL-α-tocopheryl acetate/kg; E200 = basal diet plus 200 mg DL-α-tocopheryl acetate/kg; C500 = basal diet plus 500 mg ascorbic acid/kg; C1000 = basal diet plus 1,000 mg ascorbic acid/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

[2]SEM of values transformed into archsine of the root square of the percentage.

properties of yolk did not show any substantial variation due to the supplementation with vitamin E, whereas elasticity of yolk gels, evaluated through the texture profile analysis, was significantly decreased by the dietary treatment ($P < 0.01$). However the addition of vitamin C restored this trait ($P < 0.05$) showing an ameliorating effect of the vitamin (Table 4). The elasticity of albumen gel too was improved by ascorbic acid supplementation, particularly when the dose of 500 ppm was used alone or in the presence of vitamin E (Table 5). Considering texture profile analysis of whole-egg gels, again vitamin E exerted a lowering effect on elasticity, as observed for the yolk gels; the same effect was also observed in groups supplemented with ascorbic acid differently from what was observed in albumen gels. These results show that vitamin C has a different effect either on whole egg or on albumen. Hardness and cohesiveness of the three types of gels were not affected by dietary treatment of hens (Tables 4, 5, and 6). Foam stability of albumen was significantly improved by vitamin E when administered together with vitamin C (Table 5). These results are the first contribution to the understanding of functional properties of eggs obtained from hens fed vitamin-supplemented diets.

Sensory Evaluation

The statistical analyses of data concerning sensory quality of eggs showed no differences between eggs from groups E200, C1000, E100+C500 vs. control, neither for scrambled nor for boiled eggs, as only 4 to 7 panelists out of 24 recognized the different sample (= positive response) (Table 7). Leeson et al. (1998) recorded a decline in egg-yolk flavor and overall egg acceptability when a higher level of vitamin E (100 ppm vs 10 ppm) was used in conjunction with 20% dietary flaxseed. The above-mentioned authors also found indications of preference for ordinary eggs from hens fed diets containing 10 rather than 100 ppm vitamin E, and they ascribed this result to the prooxidant instead of antioxidant action of high doses of vitamin E, as also suggested by Fennema (1987). Other

TABLE 7. Results of the sensory test on boiled and scrambled eggs

	Boiled eggs		Scrambled eggs	
Groups[1]	Positive responses	Probability	Positive responses	Probability
E200	4/24	NS	5/24	NS
C1000	7/24	NS	6/24	NS
E100+C500	6/24	NS	6/24	NS

[1]E200 = basal diet plus 200 mg DL-α-tocopheryl acetate/kg; C1000 = basal diet plus 1000 mg ascorbic acid/kg; E100+C500 = basal diet plus 100 mg DL-α-tocopheryl acetate and 500 mg ascorbic acid/kg.

authors, using in laying-hen diet up to 400 IU vitamin E per kilogram feed (Jiang et al., 1994) or exceptionally up to 20,000 IU (Surai et al., 1997), did not describe any effect of the vitamin on the sensory properties of eggs.

In conclusion, supranutritional administrations of vitamin E and C increased the yolk content of vitamin E, and this increase was quite stable during long-term storage. These results support the hypothesis that since the shell egg is a closed system, little exposed to oxygen and oxidant agents, thereby little involved in oxidative reactions, vitamin E is not consumed in preventing oxidative processes. When ordinary table eggs were stored in refrigerated conditions, the development of oxidative products was negligible. On the contrary, the storage at room temperature increased TBARS and reduced the content of vitamin E presumably consumed in oxidative processes. Vitamin C did not interfere in the yolk deposition of vitamin E and, as expected, its antioxidative role in regenerating vitamin E observed in beef meat (Schaefer et al., 1995) did not emerge. This trial has also suggested that vitamins are able to modify functional properties of egg and particularly ascorbic acid can improve the quality of albumen, i.e., elasticity of gel, whereas the organoleptic traits of cooked eggs were not affected by vitamin treatments. However the role of vitamin C in laying-hen feeding is controversial, and further research is needed in this field.

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