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The effect of astaxanthin and lycopene on the content of fatty acids in the yolks of chicken eggs under different storage regimes

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ABSTRACT
The level of consumers' satisfaction with the quality of edible chicken eggs is determined, in particular, by the attractive appearance of the yolks and their content of biologically active substances that have functional properties. Such compounds include carotenoids astaxanthin and lycopene, which can be deposited in the yolks, provide their pigmentation, and as powerful antioxidants, affect the stability of the fatty acid composition of lipids during egg storage. The aim This study aimed mine the effect of supplements of oil extracts of astaxanthin (10, 20, and 30 mg/kg of feed) or lycopene (20, 40, and 60 mg/kg of feed) on the Dion of young hens on the fatty acid composition of the yolks during eggs storage in temperature conditions 4 ±0.5 °C and 12 ±0.5 °C for 30 days. The experiment used 45 High-Line W36 crossbred laying hens at 24 weeks of age. It was found that the storage temperature of eggs (4 ±0.5 °C and 12 ±0.5 °C) equally affected the fatty acid composition of lipids of egg yolks obtained from laying hens fed lycopene supplements in doses of 20, 40, and 60 mg/kg or astaxanthin in doses of 10, 20 and 30 mg/kg of feed for 30 days. Doses of lycopene from 20 to 60 mg/kg or astaxanthin from 10 to 30 mg/kg in the diet of laying hens contributed to a decrease in egg yolks at both storage temperatures of ω6 PUFA particles: Eicosatetraenoic and 6.9, 12-okadekatrienic acids until their complete disappearance. The addition of astaxanthin to the diet of laying hens reduced and stabilized the ratio of ω3/ω6 PUFA in yolks during egg storage to a greater extent than the addition of lycopene. Storage of lycopene or astaxanthin-enriched edible chicken eggs at 4 ±0.5 °C and 12 ±0.5 °C for 30 days can be used to correct the fatty acid profile of yolk lipids.

Keywords: astaxanthin, lycopene, egg yolks, fatty acids, storage

INTRODUCTION
Chicken eggs are foodstuffs containing essential nutrients and biologically active substances easily digested by humans **[25]**. Such biologically active compounds include carotenoids **[39]** and fatty acids contained in egg yolks **[44]**. One of the criteria determining consumer demand for chicken eggs is the intensity of pigmentation of the yolk. To achieve an attractive color of chicken egg yolks, manufacturers use natural carotenoids that do not possess provitamin activity in animals and humans, but can be deposited in egg yolks, in particular, astaxanthin **[11], [38]** and lycopene **[2], [32]**. Recent studies have shown that carotenoids to some extent, can affect the content and ratio of individual fatty acids in chicken egg yolks **[33]**. Particularly relevant are studies on the development of methods to reduce the ratio of ω6/ω3 fatty acids in the lipid structure of egg yolks **[41]** to the optimal level, which should be in the range of 2:1 − 4:1. It has been proven that ω3 fatty acids play an important role in the body as components of phospholipids that form the structures of cell membranes. In particular, the axenic acid content is high in the retina, brain, and semen **[24]**. In addition to the structural role in cell membranes, fatty acids, which are ω3 and ω6, provide the body with energy and are precursors of eicosanoids, which, as signaling molecules, perform functions in the cardiovascular, respiratory, immune, and endocrine systems **[3]**.

The effect of antioxidants such as astaxanthin and lycopene on the fatty acid profile of chicken egg yolks is also one of the important criteria for assessing their suitability for storage at different temperatures. It is known that the processes of lipid peroxidation that occur in egg yolks during storage **[22]** adversely affect the sensory characteristics of eggs because they impair their taste **[16], [20]**. However, studies on the effect of egg storage on the fatty acid profile of yolks enriched with astaxanthin or lycopene are insufficient in the available literature.

They are mainly devoted to the study of the effectiveness of combining oils with carotenoids of various origins in the diets of laying hens to modify the fatty acid composition of egg yolks **[14]**. Therefore, our study aimed to investigate the effect of astaxanthin and lycopene on the fatty acid content of yolks under different storage regimes of eggs.

Scientific Hypothesis. It was assumed that the enrichment of the diet of laying hens with lycopene or astaxanthin would affect the fatty acid composition of egg yolks depending on the temperature regime of their storage. The ω6/ω3 ratio of polyunsaturated fatty acids in chicken egg yolks undergoes particular changes that depend on non doses of lycopene or astaxanthin in chicken feed during storage regardless of temperature conditions.

MATERIAL AND METHODOLOGY

Samples

All eggs from each group of laying hens were selected for the study from 25 to 31, from 55 to 61, and 85 to 91 days of the experiment.

Chemicals

Supelco 37 Component FAME Mix certified reference material, *Trace*CERT®, in dichloromethane (varied conc.), ampule of 1 mL.
Sodium hydroxide, chemically pure for analysis (Spain).
Sodium chloride, pure for analysis (Germany).
n-Hexan (purity GC 98%), Merck (Germany).
Methanol (HPLC grade), Lot: 1419984, Fisher Scientific UK.
Chloroform for chromatography, Merck (Germany).

Animals and Biological Material

45 laying hens of the High-line W36 cross at 23 weeks were used for the experiment. Laying hens on the principle of groups of analogues were divided into 3 groups of 15 heads in each and kept in cage batteries of 5 heads in each cage. The experiment lasted for 90 days (Table 1). As a source of astaxanthin used 10% oil extract was obtained from the biomass of the alga *Haematococcus Pluvialis* (ALGAE Technologies, Israel). As a source of lycopene, laying hens were fed with a 6% oil extract of lycopene derived from tomatoes (LycoRed, Israel). Laying hens were fed with complete feed, the composition of which is given by **[40]**. Experimental diets were prepared for 4 days, and the feed mixture was mixed and stored in airtight food plastic containers. Watering of laying hens was carried out at will with cup drinkers. Daylight was 16 hours, light − 30 lux, darkness − 8 hours. The air temperature in the room for keeping laying hens was at the level of 21 – 23 °C; relative humidity was in the range of 60 – 62%.

Table 1 Scheme of the experiment.

Group	Diet		
	1 – 30 day	**31 – 60 day**	**61 – 90 day**
Control	Basic diet[1] + 0.33 g/kg of refined sunflower oil	Basic diet[2] + 0.66 g/kg refined sunflower oil	Basic diet[3] + 1.0 g/kg refined sunflower oil
Lycopene diet	Basic diet[1] + 20 mg/kg lycopene (LP20)	Basic diet[2] + 40 mg/kg lycopene (LP40)	Basic diet[3] + 60 mg/kg lycopene (LP60)
Astaxanthin diet	Basic diet[1] + 10 mg/kg astaxanthin (AST10)	Basic diet[2] + 20 mg/kg astaxanthin (AST20)	Basic diet[3] + 30 mg/kg astaxanthin (AST30)

Note: In the basic diet, the same superscripts [1, and 2, 3] show one new content of the refined solar system in the diet.

Instruments

Fatty acid methyl esters were analyzed on a Trace GC Ultra gas chromatograph (USA) using a flame ionization detector (FID) and a 100 m long high-polar capillary column (Supelco, USA). Chromatography conditions: column temperature 140 – 240 °C, detector temperature 260 °C. The sample was added to the chromatography using an automatic dispenser (TriPlus autosampler) in 1 μL. The duration of one analysis on the device was 65 minutes. The fatty acid peaks of egg yolk lipids were identified by comparing them with the time of the release of the peaks of the standard sample Supelco 37 Component FAME Mix, which includes 37 names of fatty acids.

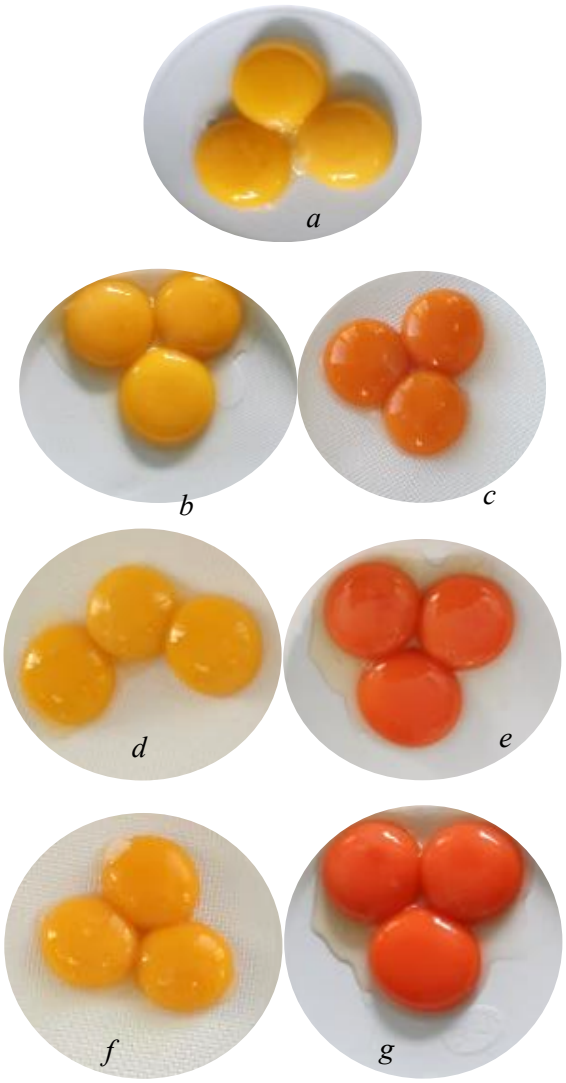

Figure 1 Color of egg yolks of chickens of control group (a), LP20 (b), AST10 (c), LP40 (d), AST20 (e), LP60 (f), AST30 (g).

Laboratory Methods

Lipids were extracted from chicken egg yolks according to the method **[45]**. This was followed by hydrolysis and methylation of fatty acids of chicken egg yolk lipids according to ISO 12966-2:2017.

Description of the Experiment

After weighing and sorting, 9 freshly laid eggs were selected from each group of laying hens to determine the fatty acid composition of the yolks. The remaining eggs from each group of laying hens were divided into two batches and stored: the first batch was stored at a temperature of 4 ±0.5 °C and relative humidity of 80 − 85%, and the second batch − at a temperature of 12 ±0.5 °C and relative humidity 70 − 75% for 30 days. At the end of the storage period, 9 eggs were taken from each group of laying hens, and the fatty acid content of the yolks was examined.

Sample preparation: The proportion of fatty acids in the lipids of chicken egg yolks was calculated by internal normalization, determining their content in percent. The study was performed in 3 parallels. The following fatty acids were determined in chicken egg yolks: dodecanoic (12:0), tetradecanoic (14:0), myristoleic (14:1), pentadecanoic (15:0), hexadecanoic (16:0), trans-3-hexadecene (16:1), heptadecane (17:0), cis-10-heptadecenoic (17:1), octadecane (18:0), oleic (18:1n9c), 9.12 octadecadienoic (18:2n6c), linolenic (18:3n3), 6,9,12-ocadecatriene (18:3n6), eicosan (20:0), cis-11-eicosene (20:1), cis-11,14-eicosadienoic (20:2n6), eicosatetraenoic (20:3n6), arachidonic (20:4n6), 5,8,11,14, 17-eicosapentaenoic (20:5n3), docosan (22:0), 4,7,10,13,16,19-docosahexaenoic (22:6n3).

Statistical analysis

The data obtained in the study were analyzed statistically using the ANOVA program. The normality of data distribution was confirmed using the program R-3.6.3 for Windows **[36]**. The difference between the values in the groups was determined using the Tukey test. The difference was considered significant at $p < 0.05$ (considering the Bonferroni correction).

RESULTS AND DISCUSSION

Storage of chicken eggs for 30 days at a temperature of 4 ±0.5 °C and 12 ±0.5 °C did not affect the ratio in the yolks of saturated fatty acids such as dodecanoic, pentadecanoic, heptadecanoic, eicosenoic, and docosanic. Still, it increased ($p < 0.05$) the proportion of tetradecanoic acid on the background of a decrease ($p < 0.05$) in the content of hexadecanoic and octadecanoic acids compared with freshly laid eggs. Among monounsaturated fatty acids under the above temperature storage conditions of eggs, only the proportion of myristoleic acid in the yolks did not change, while the content of trans-3-hexadecenoic, cis-9-octadecenoic, and cis-11-eicosenoic acids increased ($p < 0.05$) compared with freshly laid eggs. The proportion of cis-10-heptadecenoic acid decreased ($p < 0.05$) in the yolks only when storing eggs at 4 ±0.5 °C compared with freshly laid eggs (Table 2).

Table 2 The content of fatty acids in the yolks of chicken eggs under different storage regimes (% of the total fatty acid content) (control group).

Acid	Egg storage mode			SEM[1]	*p*-value
	Fresh laid	4 ±0.5 °C	12 ±0.5 °C		
Dodecane, 12:0	0.02	0.01	0.01	0.002	0.729
Myristic, 14:0	0.26	0.30*	0.28*	0.006	0.031
Myristoleic, 14:1	0.07	0.06	0.06	0.002	0.729
Pentadecanoic, 15:0	0.06	0.05	0.06	0.002	0.729
Palmitic, 16:0	28.59	27.25*	27.25*	0.155	<0.001
Palmitoleic, 16:1	2.34	2.61*	2.47*,**	0.054	0.024
Heptadecanoic, 17:0	0.19	0.18	0.18	0.002	0.729
Cis-10-heptadecenoic, 17:1	0.05	0.03*	0.04	0.004	0.064
Stearic, 18:0	12.58	9.94*	10.15*	0427	<0.001
Oleic, 18:1n9c	34.92	37.79*	37.29*,**	0.465	<0.001
Linoleic, 18:2n6c	15.90	17.27*	16.99*,**	0.220	<0.001
Gamma-linolenic, 18:3n6	0.01	0.01	0.01	0.002	1.000
Linolenic, 18:3n3	0.45	0.22*	0.22*	0.040	<0.001
Arachic, 20:0	0.14	0.16	0.15	0.004	0.256
Cis-11-eicosenoic, 20:1	0.24	0.47*	0.45*,**	0.038	<0.001
Cis-11,14-eicosadienoic, 20:2n6	0.17	0.15*	0.14*	0.004	0.050
Eicosatetraenoic, 20:3n6	0.22	0.20*	0.20*	0.004	0.009
Arachidonic, 20:4n6	2.70	2.76	2.67**	0.029	0.944
5,8,11,14,17-eicosapentaenoic, 20:5n3	ND	ND	ND	ND	ND
Behenic, 22:0	0.06	0.04	0.04	0.003	0.047
4,7,10,13,16,19-docosahexaenoic, 22:6n3	1.04	0.94*	0.94*	0.018	0.003
Σ SFA	41.90	38.44*	38.53*	0.574	<0.001
Σ UFA	58.10	62.52*	61.09*,**	0.700	<0.001
Σ MUFA	37.61	40.96*	40.31*,**	0.541	0.011
Σ PUFA	20.49	21.56	21.16	0.178	<0.001
Σ ω3 PUFA	1.49	1.16*	1.15*	0.056	<0.001
Σ ω6 PUFA	19.00	20.39*	20.00*	0.222	<0.001
ω3/ω6 PUFA	12.76	17.57*	17.34*	0.789	<0.001

Note: * – $p < 0.05$ compared with freshly laid eggs, ** − $p < 0.05$ compared with data for storage of eggs at 4 ±0.5 °C using Tukey test with Bonferroni correction; SEM[1] − standard error of the mean (n = 9).

Among the polyunsaturated fatty acids, which belong to ω 3, there was a decrease ($p < 0.05$) in the content of linolenic and 4,7,10,13,16,19-docosahexaenoic in yolks compared to freshly laid eggs, and 5, 8,11,14,17-eicosapentaenoic acid was not detected at all in the yolks of freshly laid eggs, and different storage temperatures.

Storage of chicken eggs at temperatures of 4 ±0.5 °C and 12 ±0.5 °C for 30 days did not affect the ratio in the yolks of 6,9,12-octadecatrienoic and 5,8,11,14-eicosatetraenoic acids but contributed to the redistribution of other ω6 PUFA compared to freshly laid eggs. This was expressed in an increase ($p < 0.05$) in the proportion of linoleic

and a decrease (p <0.05) in the level of cis-11,14-eicosadienoic and cis-8,11,14-eicosatrienoic acids in the yolks compared to freshly laid eggs. Such changes in the storage process of edible eggs caused a decrease in FA SFA due to an increase (p <0.05) Σ UFA in the yolks compared to freshly laid eggs. Under these conditions, Σ UFA in chicken egg yolks was mainly increased (p <0.05) due to the proportion of monounsaturated fatty acids, while Σ PUFA remained stable at both storage temperatures of chicken eggs. Among polyunsaturated fatty acids in chicken egg yolks during storage decreased (p <0.05) Σ ω3 PUFA relative to Σ ω6 PUFA, which in turn increased (p <0.05) the ratio ω3/ω6 PUFA.

Table 3 The effect of lycopene at a dose of 20 mg/kg of feed on the fatty acid content in chicken egg yolks under different storage regimes (% of the total fatty acid content).

Acid	Egg storage mode			SEM[1]	*p*-value
	Freshly laid	4 ±0.5 ºC	12 ±0.5 ºC		
Lauric, 12:0	0.02	0.01	0.01	0.002	0.729
Myristic, 14:0	0.23	0.29*	0.28*	0.010	<0.001
Myristoleic, 14:1	0.04	0.06*	0.05*	0.004	0.050
Pentadecanoic, 15:0	0.07	0.05	0.06	0.003	0.171
Palmitic, 16:0	26.70	27.67*	27.43*	0.162	0.009
Palmitoleic, 16:1	2.06	2.28*	2.25*	0.037	<0.001
Heptadecanoic, 17:0	0.19	0.18	0.17*	0.005	<0. 001
Cis-10-heptadecenoic, 17:1	0.07	0.05*	0.05*	0.004	0.008
Stearic, 18:0	11.84	11.23*	11.23*	0.105	<0.001
Oleic, 18:1n9c	34.68	36.75*	37.01*	0.389	<0.001
Linoleic, 18:2n6c	16.26	15.63*	15.53*,**	0.123	<0.001
6,9,12-octadecatrienoic, 18:3n6	0.02	0.16*	0.15*	0.023	<0.001
Linolenic, 18:3n3	0.20	0.40*	0.39*	0.033	<0.001
Arachidic, 20:0	0.16	0.02*	0.02*	0.023	<0.001
Cis-11-eicosenoic, 20:1	0.45	0.39*	0.39*	0.010	<0.001
Cis-11,14-eicosadienoic, 20:2n6	0.17	0.07*	0.06*	0.017	<0.001
Eicosatetraenoic, 20:3n6	0.26	0.24	0.22	0.007	0.181
Arachidonic, 20:4n6	4.66	3.26*	3.53*	0.236	0.010
5,8,11,14,17-eicosapentaenoic, 20:5n3	1.86	1.21*	1.14*	0.115	<0.001
Behenic, 22:0	0.09	0.07*	0.06*	0.004	0.005
4,7,10,13,16,19-docosahexaenoic, 22:6n3	ND	ND	ND	ND	ND
Σ SFA	39.29	39.54	39.26	0.081	0.359
Σ UFA	60.71	60.49	60.78	0.075	0.272
Σ MUFA	37.29	39.53*	39.75*,**	0.410	0.045
Σ PUFA	23.42	20.96*	21.03*	0.415	0.127
Σ ω3 PUFA	2.06	1.61*	1.53*	0.083	<0.001
Σ ω6 PUFA	21.36	19.35*	19.50*	0.336	<0.001
ω3/ω6 PUFA	10.37	12.00*	12.72	0.356	<0.001

Note: * – p <0.05 compared with freshly laid eggs, ** − p <0.05 compared with data for storage of eggs in conditions of 4 ±0.5 °C using Tukey test with Bonferroni correction; SEM[1] – standard error of the mean (n = 9).

Feeding of laying hens with the addition of lycopene at a dose of 20 mg/kg of feed significantly affected the fatty acid composition of the yolks at different temperatures of egg storage. The content of saturated fatty acids, such as dodecanoic and pentadecanoic, did not change compared to freshly laid eggs' data (Table 3). The proportions of tetradecanoic and hexadecanoic acids increased (p <0.05) due to a decrease (p <0.05) in the particles of octadecanoic, eicosanoic, and behenic acids in the structure of yolk lipids compared to freshly laid eggs. As for the content of heptadecanoic acid, its level probably decreased (p <0.05) in chicken egg yolks only when stored at 12 ±0.5 °C for 30 days compared to freshly laid eggs. Storage of eggs obtained from laying hens treated with lycopene at a dose of 20 mg/kg of feed, caused almost the same redistribution of monounsaturated acids in the structure of yolk lipids in both modes of storage. It was found that the content of myristoleic, trans-3-hexadecenoic, and cis-9-octadecenoic acids increased (p <0.05), and the proportion of cis-10-heptadecenoic and cis-11-eicosenoic acids decreased (p <0.05) in the yolks compared to freshly laid eggs.

Among ω3 PUFA, which were detected in egg yolks with the use of lycopene laying hens at a dose of 20 mg/kg of feed, the content of 9,12,15-octadecanoic acid increased (p <0.05) against a background of decreasing (p <0.05) level 5, 8,11,14,17-eicosapentaenoic acid at both storage temperatures compared to freshly laid eggs. The peak of

4,7,10,13,16,19-docosahexaenoic acid was not detected on the chromatograms of the yolks of freshly laid eggs and during their storage at different temperatures. The addition of lycopene at a dose of 20 mg/kg of feed for laying hens did not affect the content of only cis-8,11,14-eicosatrienoic acid in egg yolks during storage, while the proportion of the remaining ω 6 PUFA was redistributed as follows: content 9, 12-octadecadienoic, cis-11,14-eicosadienoic and 5,8,11,14-eicosatetraenoic acids decreased (p <0.05), and the level of 6,9,12-octadecanoic acid increased (p <0.05) compared to freshly laid eggs. Increasing the dose of lycopene to 40 mg/kg of feed for laying hens did not affect the ratio of saturated acids such as dodecanoic and pentadecanoic. In contrast, the proportions of tetradecanoic, heptadecanoic, eicosanoic, and behenic acids increased (p <0.05), and the proportion of hexadecane p <0.05) in the yolks at both temperatures of egg storage. Among the monounsaturated fatty acids that are part of the structure of yolk lipids, there was a decrease (p <0.05) only the proportion of cis-11-eicosenoic acid. In contrast, the proportion of trans-3-hexadecenoic, cis-10-heptadecenoic, and cis-9-octadecenoic acids increased (p <0.05) compared with freshly laid eggs (Table 4).

Table 4 The effect of lycopene at a dose of 40 mg/kg of feed on the fatty acid content in chicken egg yolks for different storage modes (% of the total fatty acid content).

Acid	Egg storage mode			SEM[1]	*p*-value
	Freshly laid	4 ±0.5 ºC	12 ±0.5 ºC		
Lauric, 12:0	0.02	0.01	0.01	0.002	0.125
Myristic, 14:0	0.31	0.39*	0.38*	0.013	<0.001
Myristoleic, 14:1	0.03	0.02	0.03	0.002	0.178
Pentadecanoic, 15:0	0.02	0.01	0.02	0.002	0.178
Palmitic, 16:0	26.48	21.48*	21.30*	0.852	<0.001
Palmitoleic, 16:1	2.09	2.93*	2.93*	0.140	<0.001
Heptadecanoic, 17:0	0.15	0.20*	0.19*	0.008	<0.001
Cis-10-heptadecenoic, 17:1	0.01	0.07*	0.06*	0.009	<0.001
Stearic, 18:0	12.19	10.28*	10.13*	0.344	<0.001
Oleic, 18:1n9c	36.46	40.64*	40.24*	0.668	<0.001
Linoleic, 18:2n6c	17.29	19.67*	19.63*	0.396	<0.001
Gamma-linolenic, 18:3n6	ND	ND	ND	ND	-
Linolenic, 18:3n3	0.50	0.73*	0.71*	0.037	<0.001
Arachidica, 20:0	0.15	0.18*	0.18*	0.006	0.011
Cis-11-eicosenoic, 20:1	0.09	0.06*	0.05*	0.006	<0.001
Cis-11,14-eicosadienoic, 20:2n6	0.16	0.11*	0.10*	0.010	<0.001
Eicosatetraenoic, 20:3n6	ND	ND	ND	ND	-
Arachidonic, 20:4n6	2.20	2.50*	2.42*	0.051	0.001
5,8,11,14,17-eicosapentaenoic, 20:5n3	0.27	0.24*	0.24*	0.006	0.045
Behenic, 22:0	0.21	0.26*	0.25*	0.009	0.002
4,7,10,13,16,19-docosahexaenoic, 22:6n3	1.37	1.13	1.13	0.053	0.138
Σ SFA	39.53	32.81*	32.47*	1.159	<0.001
Σ UFA	60.47	68.08*	67.53*	1.230	<0.001
Σ MUFA	38.69	43.72*	43.31*	0.809	<0.001
Σ PUFA	21.79	24.36*	24.14*	0.424	<0.001
Σ ω3 PUFA	2.14	2.09	2.08	0.040	0.837
Σ ω6 PUFA	19.65	22.27*	22.15*	0.432	<0.001
ω3/ω6 PUFA	9.24	10.64	10.67	0.280	0.025

Note: * – p <0.05 compared with freshly laid eggs, ** − p <0.05 compared with data for storage of eggs in conditions of 4 ±0.5 ºC using Tukey test with Bonferroni correction; SEM[1] – standard error of the mean (n = 9).

Feeding laying hens with lycopene at a dose of 40 mg/kg of feed changed the ratio in the yolks of eggs of polyunsaturated fatty acids belonging to ω 3, in particular, increased (p <0.05) the proportion of Linolenic, but decreased (p <0.05) the content of 5,8,11,14,17-eicosapentaenoic acid compared to freshly laid eggs. At the same time, 6,9,12-octadecatrienoic acid disappeared from chicken egg yolks, and 4,7,10,13,16,19-docosahexaenoic acid appeared which did not depend on the temperature of their storage. Lycopene at a dose of 40 mg/kg in the diet of laying hens changed the ratio Σ SFA in favour (p <0.05) Σ UFA in lipids of egg yolks. This was due to both an increase (p <0.05) in the particles of Σ MUFA and Σ PUFA in the lipids of chicken egg yolks. It should be noted that the increase (p <0.05) in the level of Σ PUFA, in this case, was characterized by an increase (p <0.05) in the proportion of Σ ω6 PUFA in both storage regimes, but it did not affect the ratio ω3/ω6 PUFA in

chicken egg yolks. A further increase in the dose of lycopene to 60 mg/kg in the diet of laying hens contributed to an increase (p <0.05) in the content of most saturated fatty acids in yolks in both egg storage regimes, except for dodecanoic and heptadecanoic, the level of which did not change and tetradecanoic acid, decreased (p <0.05) compared with freshly laid eggs (Table 5). This dose of lycopene did not affect the level of such monounsaturated fatty acids as myristoleic and cis-10-heptadecenoic with a simultaneous decrease (p <0.05) in the proportion of trans-3-hexadecenoic and increase (p <0.05) cis-11-eicosenoic acids in yolks in both storage modes compared to freshly laid eggs. Among ω3 fatty acids in the yolks of eggs of laying hens fed lycopene at a dose of 60 mg/kg of feed, a decrease (p <0.05) in the proportion of 5,8,11,14,17-eicosapentaenoic and 4,7,10,13,16,19-docosahexaenoic on the background of increasing (p <0.05) share of linolenicoic acid in both storage modes compared to freshly laid eggs. Under such conditions, a redistribution of ω 6 fatty acid particles in chicken egg yolks was detected, which was characterized by the disappearance of 6,9,12-ocadecatrienic and cis-8,11,14-eicosatric acid peaks on the chromatograms and a decrease (p <0.05) in the 5.8 particles, 11,14-eicosatetraenoic with a simultaneous increase (p <0.05) in the level of Linoleic and cis-11,14-eicosadienoic acids in both storage modes compared to freshly laid eggs. The use of lycopene at a dose of 60 mg/kg of feed for laying hens thus contributed to an increase (p <0.05) Σ SFA relative to Σ UFA in egg yolks in both storage modes. The decrease (p <0.05) in the proportion of Σ UFA in chicken egg yolks under the influence of lycopene, in this case, occurred only due to Σ MUFA. The increase (p <0.05) in the proportion of U PUFA in yolks was observed in both storage regimes of chicken eggs, and it was due to an increase (p <0.05) in the content of Σ ω6 PUFA, which ultimately led to an increase (p <0.05) in the coefficient ω3/ω6 PUFA in the structure of lipids.

Table 5 The effect of lycopene at a dose of 60 mg/kg of feed on the content of fatty acids in chicken egg yolks under different storage regimes (% of the total fatty acid content).

Acid	Egg storage mode			SEM[1]	*p*-value
	Freshly laid	4 ±0.5 ºC	12 ±0.5 ºC		
Lauric, 12:0	0.02	0.01	0.01	0.001	0.078
Myristic, 14:0	0.33	0.23*	0.23*	0.017	<0.001
Myristoleic, 14:1	0.03	0.02	0.02	0.002	0.125
Pentadecanoic, 15:0	0.02	0.05*	0.04*	0.005	<0.001
Palmitic, 16:0	26.40	28.23*	28.25*	0.311	<0.001
Palmitoleic, 16:1	2.77	2.11*	2.11*	0.110	<0.001
Heptadecanoic, 17:0	0.11	0.13	0.13	0.004	0.078
Cis-10-heptadecenoic, 17:1	0.02	0.02	0.01	0.002	0.729
Stearic, 18:0	10.90	11.70*	11.70*	0.136	<0.001
Oleic, 18:1n9c	38.80	36.41*	36.48*	0.395	<0.001
Linoleic, 18:2n6c	12.82	14.36*	14.31*	0.255	<0.001
Gamma-linolenic, 18:3n6	ND	ND	ND	ND	-
Linolenic, 18:3n3	0.35	0.41*	0.41*	0.010	<0.001
Arachidic, 20:0	0.12	0.14*	0.14*	0.005	0.049
Cis-11-eicosenoic, 20:1	0.02	0.06*	0.06*	0.007	<0.001
Cis-11,14-eicosadienoic, 20:2n6	0.12	0.13*	0.13*	0.002	0.031
Eicosatetraenoic, 20:3n6	ND	ND	ND	ND	-
Arachidonic, 20:4n6	5.12	4.39*	4.39*	0.124	<0.001
5,8,11,14,17-eicosapentaenoic, 20:5n3	0.17	0.10*	0.09*	0.025	<0.001
Behenic, 22:0	0.16	0.20*	0.19*	0.007	<0.001
4,7,10,13,16,19-docosahexaenoic, 22:6n3	1.68	1.28*	1.25*	0.071	<0.001
Σ SFA	38.05	40.68*	40.69*	0.443	<0.001
Σ UFA	61.95	59.32*	59.31*	0.443	<0.001
Σ MUFA	41.63	38.64*	38.67*	0.498	<0.001
Σ PUFA	20.33	20.67*	20.63*	0.065	0.023
Σ ω3 PUFA	2.27	1.84*	1.81*	0.076	<0.001
Σ ω6 PUFA	18.05	18.88*	18.82*	0.139	<0.001
ω3/ω6 PUFA	7.95	10.28*	10.40*	0.405	<0.001

Note: * – p <0.05 compared with freshly laid eggs, ** − p <0.05 compared with data for storage of eggs in conditions of 4 ±0.5 ºC using Tukey test with Bonferroni correction; SEM[1] – standard error of the mean (n = 9).

Feeding laying hens with astaxanthin at a dose of 10 mg/kg of feed did not affect the content of saturated fatty acids such as dodecanoic and pentadecanoic, but reduced (p <0.05) the proportion of octadecanoic, eicosanoic, and behenic acids on the background of increasing (p <0.05) acid in the yolks in both storage modes compared to freshly laid eggs. As for heptadecanoic acid, its level increased (p <0.05) in chicken egg yolks only when stored at 4 ±0.5 °C compared to freshly laid eggs (Table 6). Astaxanthin supplementation in the diet of laying hens at the above dose increased (p <0.05) the proportion of all monounsaturated fatty acids, except for cis-10-heptadecenoic, the level of which was stable in the yolks in both egg storage regimes.

The use of astaxanthin at a dose of 10 mg/kg of feed for laying hens contributed to changes in the ratio of individual ω3 PUFA, namely an increase (p <0.05) in the proportion of Linolenicoic acid. In contrast, the level of 5,8,11,14, 17-eicosapentaenoic and 4,7,10,13,16,19-docosahexaenoic acids decreased (p <0.05) in yolks under both storage regimes compared to freshly laid eggs. An even greater effect of astaxanthin at the above dose was found on the level of ω 6 PUFA, which was characterized by the disappearance on the chromatogram of the peak of 6,9,12-octadecatrienoic acid with a simultaneous decrease (p <0.05) in the proportion of cis-11,14-eicosadienoic, cis-8, 11,14-eicosatrienoic, and 5,8,11,14-eicosatetraenoic acids in the yolks under both storage modes compared to freshly laid eggs. Thus, astaxanthin at a dose of 10 mg/kg of feed for laying hens contributed to a decrease (p <0.05) in the proportion of Σ SFA in favour of Σ UFA. This was due to an increase in the proportion of Σ MUFA in the lipid structure of chicken egg yolks. Despite the low content of astaxanthin in the diet of laying hens, it also contributed to a decrease (p <0.05) in the proportion of Σ PUFA, which is due to a decrease (p <0.05) in both part ω3 PUFA and Σ ω6 PUFA in chicken egg yolks during storage in both temperature regimes. As a result of this redistribution of unsaturated fatty acids in chicken egg yolks under the influence of astaxanthin increased (p <0.05) the coefficient ω3/ω6 PUFA compared to freshly laid eggs.

Table 6 The effect of astaxanthin at a dose of 10 mg/kg of feed on the content of fatty acids in chicken egg yolks under different storage regimes (% of the total fatty acid content).

Acid	Egg storage mode			SEM[1]	p-value
	Freshly laid	4 ±0.5 ºC	12 ±0.5 ºC		
Lauric, 12:0	0.02	0.03	0.02	0.002	0.179
Myristic, 14:0	0.23	0.29*	0.29*	0.010	<0.001
Myristoleic, 14:1	0.03	0.06*	0.06*	0.005	<0.001
Pentadecanoic, 15:0	0.07	0.07	0.06	0.002	0.729
Palmitic, 16:0	27.00	27.50*	27.53*	0.089	<0.001
Palmitoleic, 16:1	2.03	2.13*	2.13*	0.017	<0.001
Heptadecanoic, 17:0	0.20	0.22*	0.21	0.004	<0.001
Cis-10-heptadecenoic, 17:1	0.06	0.06	0.06	0.002	1.000
Stearic, 18:0	12.88	10.64*	10.61*	0.380	<0.001
Oleic, 18:1n9c	32.57	35.40*	35.39*	0.471	<0.001
Linoleic, 18:2n6c	16.67	17.96*	17.98*	0.219	<0.001
Gamma-linolenic, 18:3n6	ND	ND	ND	ND	-
Linolenic, 18:3n3	0.47	0.49*	0.49*	0.005	0.011
Arachidic, 20:0	0.05	0.01*	0.01*	0.006	<0.001
Cis-11-eicosenoic, 20:1	0.42	0.50*	0.50*	0.014	<0.001
Cis-11,14-eicosadienoic, 20:2n6	0.26	0.15*	0.15*	0.019	<0.001
Eicosatetraenoic, 20:3n6	0.29	0.27*	0.27*	0.004	0.079
Arachidonic, 20:4n6	4.79	2.84*	2.84*	0.325	<0.001
5,8,11,14,17-eicosapentaenoic, 20:5n3	0.21	0.18*	0.18*	0.006	<0.001
Behenic, 22:0	0.09	0.06*	0.06*	0.005	<0.001
4,7,10,13,16,19-docosahexaenoic, 22:6n3	1.69	1.17*	1.17*	0.086	<0.001
Σ SFA	40.52	38.81*	38.79*	0.289	<0.001
Σ UFA	59.48	61.19*	61.21*	0.289	<0.001
Σ MUFA	35.11	38.14*	38.13*	0.505	<0.001
Σ PUFA	24.37	23.22*	23.08*	0.215	<0.001
Σ ω3 PUFA	2.36	1.84*	1.84*	0.087	<0.001
Σ ω6 PUFA	22.01	21.21*	21.23*	0.139	0.002
ω3/ω6 PUFA	9.31	11.53*	11.52*	0.370	<0.001

Note: * – p <0.05 compared with freshly laid eggs, ** − p <0.05 compared with data for storage of eggs in conditions of 4 ±0.5 °C using Tukey test with Bonferroni correction; SEM[1] – standard error of the mean (n = 9).

Increasing the dose of astaxanthin in the diet of laying hens to 20 mg/kg did not affect the ratio in the yolks of saturated fatty acids such as dodecanoic and pentadecanoic. However, it increased (p <0.05) the proportion of hexadecanoic, heptadecanoic, and eicosanoic, and behenic acids in the background 0.05) particles of tetradecanoic and octadecanoic acids in both modes of storage of eggs compared with freshly laid (Table 7). Enrichment of the diet of laying hens with astaxanthin in the specified dose did not affect the content of such monounsaturated fatty acids as cis-9-myristic and cis-10-heptadecenoic but increased (p <0.05) the level of palmitoleic, oleic, and cis-11-nicotine in both modes of storage of eggs compared with freshly laid. The content of individual ω3 PUFA in egg yolks was affected by astaxanthin at a dose of 20 mg/kg, depending on the storage temperature. Thus, the content of Linolenicoic acid in the yolks increased (p <0.05) at a temperature of 4 ±0.5 °C, but at a temperature of 12 ± 0.5 °C, it decreased (p <0.05) compared to freshly laid eggs. The proportion of 5,8,11,14,17-eicosapentaenoic acid in chicken egg yolks under the influence of astaxanthin in the above dose decreased (p <0.05), while the level of 4,7,10,13,16,19-docosahexaenoic acid the additive was not affected. Even more, pronounced changes in the composition of ω6 PUFA were found in chicken egg yolks during storage at different temperatures under the influence of astaxanthin. They were characterized by the disappearance on the chromatogram of the peaks of 6,9,12-octadecatrienoic and cis-8,11,14-eicosatrienoic acids in the yolks of both freshly laid eggs and their storage at different temperatures. At the same time, against the background of a decrease (p <0.05) in the content of linoleic increase (p <0.05), the level of cis-11,14-eicosadienoic fatty acids in egg yolks in both modes of their storage compared to freshly laid. In this case, the ratio of Σ SFA and Σ UFA remained stable in the yolks of chicken eggs in both modes of storage. However, the use of astaxanthin in laying hens at a dose of 20 mg/kg of feed caused a redistribution in the structure of the UFA particles Σ PUFA and Σ MUFA in favour (p <0.05) of the latter. The proportion of Σ PUFA decreased (p <0.05) due to a decrease (p <0.05) in the level of Σ ω 6 PUFA in the lipid structure of chicken egg yolks, although the ratio ω3 / ω6 PUFA remained unchanged.

Table 7 The effect of astaxanthin at a dose of 20 mg/kg of feed on the content of fatty acids in chicken egg yolks under different storage regimes (% of the total fatty acid content).

Acid	Egg storage mode			SEM[1]	p-value
	Freshly laid	4 ±0.5 °C	12 ±0.5 °C		
Lauric, 12:0	**0.02**	**0.02**	0.02	0.003	0.070
Myristic, 14:0	0.30	0.27*	0.27*	0.007	<0.001
Myristoleic, 14:1	0.03	0.05	0.05	0.003	0.072
Pentadecanoic, 15:0	0.03	0.02	0.02	0.002	0.125
Palmitic, 16:0	26.23	28.37*	28.14*	0.357	<0.001
Palmitoleic, 16:1	2.11	2.60*	2.59*	0.084	0.000
Heptadecanoic, 17:0	0.15	0.18*	0.18*	0.006	<0.001
Cis-10-heptadecenoic, 17:1	0.02	0.02	0.02	0.001	0.629
Stearic, 18:0	12.23	10.12*	10.21*,**	0.347	<0.001
Oleic, 18:1n9c	36.80	38.01*	38.21*	0.265	0.043
Linoleic, 18:2n6c	17.28	15.32*	15.49*	0.334	<0.001
Gamma-linolenic, 18:3n6	ND	ND	ND	ND	-
Linolenic, 18:3n3	0.51	0.53*	0.50*,**	0.006	0.005
Arachidic, 20:0	0.15	0.21*	0.20*	0.009	<0.001
Cis-11-eicosenoic, 20:1	0.09	0.21*	0.19*	0.019	<0.001
Cis-11,14-eicosadienoic, 20:2n6	0.16	0.19*	0.18*,**	0.005	<0.001
Eicosatetraenoic, 20:3n6	ND	ND	ND	ND	-
Arachidonic, 20:4n6	2.21	2.26*	2.21**	0.012	0.002
5,8,11,14,17-eicosapentaenoic, 20:5n3	0.27	0.22*	0.20*,**	0.011	<0.001
Behenic, 22:0	0.21	0.25*	0.22**	0.007	0.007
4,7,10,13,16,19-docosahexaenoic, 22:6n3	1.21	1.20	1.11	0.042	0.200
Σ SFA	39.32	39.42	39.25	0.098	0.834
Σ UFA	60.68	60.58	60.75	0.098	0.834
Σ MUFA	39.05	40.88*	41.06*	0.352	0.004
Σ PUFA	21.63	19.70*	19.68*	0348	0.003
Σ ω3 PUFA	1.99	1.94	1.81	0.049	0.347
Σ ω6 PUFA	19.64	17.76*	17.88*	0.326	0.002
ω3/ω6 PUFA	9.95	9.19	9.90	0.235	0.389

Note: * – p <0.05 compared with freshly laid eggs, ** − p <0.05 compared with data for storage of eggs in conditions of 4 ±0.5 ºC using Tukey test with Bonferroni correction; SEM[1] – standard error of the mean (n = 9).

Feeding laying hens with astaxanthin at a dose of 30 mg/kg of feed had an even stronger effect on the fatty acid profile of yolk lipids, both freshly laid and at different egg storage regimes compared with doses of 10 and 20 mg/kg (Table 8). It was found that this increase in astaxanthin content in the diet of laying hens caused the disappearance on the chromatogram of the peaks of two saturated fatty acids: dodecanoic and tetradecanoic, and reduced (p <0.05) the proportion of hexadecanoic and octadecanoic acids but did not affect the content of pentadic acids in the yolks under different storage modes of eggs compared to freshly laid. Among the monounsaturated acids of chicken egg yolks was also detected under the influence of astaxanthin disappearance on the chromatogram of the peak of myristoleic acid and decrease (p <0.05) the level of cis-11-eicosenoic acid on the background of increasing (p <0.05) content of palmitoleic, cis-10-heptadecenoic and cis-9-octadecenoic acids in both modes of storage of eggs compared with freshly laid. Changes in the level of ω3 PUFA in egg yolks of laying hens treated with astaxanthin at a dose of 30 mg/kg were characterized only by a decrease (p <0.05) in the proportion of 5,8,11,14,17-eicosapentaenoic acid, while the content of other acids changed significantly did not give in.

Table 8 The effect of astaxanthin at a dose of 30 mg/kg of feed on the fatty acid content in chicken egg yolks for different storage modes (% of the total fatty acid content).

Acid	Egg storage mode			SEM[1]	p-value
	Freshly laid	4 ±0.5 ºC	12 ± 0.5 ºC		
Lauric, 12:0	**0.01**	ND	ND	0.004	-
Myristic, 14:0	0.30	ND	ND	0.051	-
Myristoleic, 14:1	ND	ND	ND	ND	-
Pentadecanoic, 15:0	0.03	0.02	0.01	0.003	0.068
Palmitic, 16:0	26.49	24.31*	24.31*	0.365	<0.001
Palmitoleic, 16:1	2.09	3.28*	3.28*	0.200	<0.001
Heptadecanoic, 17:0	0.15	0.16	0.16	0.004	0.369
Cis-10-heptadecenoic, 17:1	0.02	0.05*	0.06*	0.007	<0.001
Stearic, 18:0	12.19	8.57*	8.57*	0.613	<0.001
Oleic, 18:1n9c	36.47	42.19*	42.07*	0.946	<0.001
Linoleic, 18:2n6c	17.29	15.49*	15.75*	0.312	0.013
Gamma-linolenic, 18:3n6	ND	ND	ND	ND	-
Linolenic, 18:3n3	0.51	0.47	0.46	0.008	<0.001
Arachidic, 20:0	0.15	0.17	0.16	0.003	0.086
Cis-11-eicosenoic, 20:1	0.08	0.02*	0.02*,**	0.010	<0.001
Cis-11,14-eicosadienoic, 20:2n6	0.16	0.17	0.16	0.002	0.178
Eicosatetraenoic, 20:3n6	ND	ND	ND	ND	-
Arachidonic, 20:4n6	2.21	3.17*	3.09*	0.155	<0.001
5,8,11,14,17-eicosapentaenoic, 20:5n3	0.27	0.21*	0.20*	0.054	<0.001
Behenic, 22:0	0.21	0.22	0.21	0.003	0.236
4,7,10,13,16,19-docosahexaenoic, 22:6n3	1.35	1.51	1.50	0.039	0.400
Σ SFA	39.53	33.44*	33.42*	1.025	<0.001
Σ UFA	60.47	66.56*	66.58*	1.025	<0.001
Σ MUFA	38.69	45.55*	45.42*	1.135	0.001
Σ PUFA	21.78	21.02	21.16	0.195	0.258
Σ ω3 PUFA	2.13	2.19	2.16	0.051	0.907
Σ ω6 PUFA	19.65	18.83	19.00	0.187	0.163
ω3/ω6 PUFA	9.35	8.60	8.82	0.243	0.491

Note: * – p <0.05 compared with freshly laid eggs, ** − p <0.05 compared with data for storage of eggs in conditions of 4 ±0.5 ºC using Tukey test with Bonferroni correction; SEM[1] – standard error of the mean (n = 9).

Astaxanthin at a dose of 30 mg/kg of feed had a more pronounced effect on the level of ω6 PUFA in egg yolks, which was characterized by the disappearance on the chromatogram of the peaks of 6,9,12-ocadecatrienic and cis-8,11,14-eicosatrienoic acids, both fresh and eggs that were stored at both temperatures. At the same time, a decrease (p <0.05) in the proportion of linoleic acid and an increase (p <0.05) in the proportion of 5,8,11,14-eicosatetraenoic acids in chicken egg yolks were observed under both storage regimes compared to freshly laid eggs. Such changes in the ratio of individual fatty acids in chicken egg yolks, in turn, caused a decrease (p <0.05)

in the proportion of Σ UFA in favour of Σ SFA. The reason for such changes in balance was an increase (p <0.05) in the proportion of U MUFA in the lipid structure of chicken egg yolks in both storage regimes compared to freshly laid eggs. It should also be noted that the feeding of laying hens astaxanthin at a dose of 30 mg/kg of feed did not affect the balance of Σ PUFA, in particular, Σ ω3 PUFA and Σ ω6 PUFA in the yolks at different egg storage regimes.

Our research results show that the storage temperatures of chicken eggs 4 ±0.5 °C and 12 ±0.5 °C, do not differ significantly in terms of the effect on the ratio of fatty acids in the yolks. Similar results were obtained in a study by **[13]**, who did not show a significant difference in the ratio of α-linolenic acid, arachidonic and docosahexaenoic acids in the yolks for egg storage from 4 °C to 20 °C for 6 weeks.

It is known that during the storage of edible eggs in the yolks, there are changes characteristic of the oxidation of lipids and certain fatty acids and, consequently, a decrease in their absolute content **[29]**. Lipid oxidation is a process that affects the stability of egg yolk during storage. This can change the quality of table eggs and lead to a deterioration in taste, aroma, and color and the formation of toxic substances in eggs **[37], [43]**. One of the factors that characterize the intensity of lipid oxidation is the level of malonic dialdehyde in egg yolks **[34]**. The concentration of malonic dialdehyde in egg yolks increased after storage at 4 °C for 60 and 90 days. Enrichment of chicken egg yolks ω 3 and ω6 with fatty acids also stimulated the formation of malonic dialdehyde when stored at 4 °C for 30 and 60 days **[37]**, which also indicates the process of lipid oxidation in yolks. The same fact was confirmed in studies by **[8]**, **[10]**, who reported a decrease in the total content of ω 3 fatty acids in the yolks of eggs of laying hens fed fish oil or olive leaves, after 60 days of storage at 4 °C.

This destruction of fatty acids in the yolks in our experiment occurred during the storage of eggs for 30 days at 4 ±0.5 °C and 12 ±0.5 °C. It contributed to a decrease in the proportion of FA SFA mainly due to the two main saturated fatty acids − hexadecanoic and octadecane. Nevertheless, the proportion of Σ UFA in egg yolks increased mainly due to Σ MUFA, which to some extent normalized the ratio of Σ SFA to Σ MUFA, which became desirable for food lipids and reached the limit of 1:1 **[29]**. As can be seen from the table. 2, among Σ UFA during storage of table eggs, there is a destruction of ω3 PUFA, while the share of ω6 PUFA against this background increases slightly, which increases the shift ω3/ω6 PUFA by almost 5 units in both modes of egg storage. However, 5,8,11,14,17-eicosapentaenoic acid in the yolks of freshly laid eggs of laying hens of the control group was not detected, which is probably due to the low intensity of its de novo synthesis from precursors in the body, as well as its oxidation.

Prevention of auto-oxidation of fatty acids in chicken egg yolks can be achieved by adding antioxidants to chicken feed, such as vitamin E **[7]**, sources of natural flavonoids **[19]**, and carotenoids **[21]**.

In our experiment, feeding laying hens with lycopene supplements significantly affected egg yolks' fatty acid profile during both storage regimes. At the same time, most of the fatty acids that were part of the structure of egg yolk lipids changed. Given the fact that all fatty acids of chicken egg yolk, which were stored at different temperatures, could be exposed only to exogenous influences, we can only talk about changing their ratio from the standpoint of resistance to oxidation at different doses of lycopene in the diet of laying hens, and hence in the eggs themselves **[39]**. Thus, feeding laying hens a supplement of lycopene at a dose of 20 mg/kg of feed helped to improve the preservation in egg yolks of one of the main saturated acids − hexadecanoic, but this did not apply to octadecanoic acid, the proportion of which did not affect the balance Σ SFA under their storage. The proportion of Σ MUFA increased, and Σ PUFA decreased in egg yolks due to Σ ω3 PUFA and Σ ω6 PUFA.

The use of lycopene at a dose of 40 mg/kg of feed for laying hens prevented the oxidation of the main saturated fatty acids of egg yolks, as well as individual ω3 PUFA and ω6 PUFA, but at a dose of 60 mg/kg ultimately contributed to a shift in the ratio of ω3 PUFA/ω6 PUFA in favour of the latter. With increasing lycopene dose to 40 and 60 mg/kg of feed, changes in the profile of fatty acids in chicken egg yolks during both storage regimes intensified, which was expressed in the disappearance of peaks of two ω6 PUFA − Gamma-linolenic and cis-8,11,14-eicosatriene, which are not the main, while the proportion of Linoleic, which is the main ω6 PUFA in the yolks, increased. This may be due to both the effect of lycopene on the activity of the synthesis of these fatty acids in the body **[4]** and the antioxidant protection process against their destruction during storage **[5]**.

As shown in the **[31]**, the inclusion of antioxidants in the diet of laying hens, including sources of lycopene (dried tomatoes, red peppers) may affect the oxidative stability of egg yolk lipids during storage. The diet with flaxseed (4.5%) in combination with sweet red pepper and a mixture of tomatoes (2%) caused higher oxidative stability of lipids than the diet with flaxseed. Stating eggs reduced the yolks' antioxidant activity but did not reduce the stability of lipids to oxidation. In another study **[1]** it was shown that the concentrations of lycopene, β-carotene, lutein, and vitamin A in serum and egg yolk increased against the background of reduced concentrations of malonic dialdehyde when used in the diet of laying hens tomato powder at a dose of 5 and 10 g/kg of feed.

In general, as can be seen from the data obtained in our experiment, a clear pattern of the effect of lycopene dose on the fatty acid profile of chicken egg yolks during storage is not observed. Still, changes in the ratio of fatty acids in both saturated and unsaturated series almost did not differ at both temperatures.

The use of astaxanthin in the diet of laying hens is due to its ability to color the yolks and its antioxidant properties. It is known that the mechanism of action of astaxanthin is its ability to absorb free radicals by autooxidation, followed by degradation of this carotenoid **[17], [26]**. In addition, astaxanthin *H. Pluvialis* increases the activity of antioxidant enzymes such as superoxide dismutase, glutathione peroxidase, and total antioxidant capacity against the background of reduced levels of malonic dialdehyde in blood plasma, liver of laying hens, and egg yolk **[18]**.

The use in our experiment in feeding laying hens astaxanthin *H. Pluvialis* dose-dependently affected the fatty acid profile in the lipids of egg yolks, both freshly laid and stored at different temperatures (Tables 5 − 7). Among the saturated fatty acids of the yolks during egg storage, there was a redistribution as follows: the proportion of hexadecanoic acid increased in the structure of egg yolk lipids only until the dose of astaxanthin 20 mg/kg of feed, and at 30 mg/kg its value probably decreased, while the share of the second of the main saturated acids of egg yolks − octadecane was reduced for all doses of astaxanthin in the diet of laying hens. During the same period, the main monounsaturated acid of chicken egg yolks oleic showed an increase in the proportion in the structure of lipids. In egg yolks during storage, the addition of astaxanthin at a dose of 10 mg/kg in the diet of laying hens increased the proportion of basic ω6 PUFA linoleic. In contrast, in higher doses, there was a decrease in its share in the structure of yolk lipids. In addition, a dose of astaxanthin 10 mg/kg caused the disappearance of the peak on the yolk chromatogram of only one ω6 PUFA gamma-linolenic. In comparison, a dose of astaxanthin 20 mg/kg in the diet of laying hens also disappeared eicosatetraenoic. A dose of 30 mg/kg of diet caused the disappearance of three more short-chain acids, dodecanoic and tetradecanoic, and myristoleic during egg storage. These changes in the fatty acid composition of yolks in both egg storage regimes may be due to lipolysis and lipid oxidation **[43]**. During egg storage, lipids can be hydrolyzed by endogenous enzymes in the yolk with the release of free fatty acids, and the process of lipid peroxidation can promote the formation and accumulation of hydroperoxides and secondary oxidation products.

Our data on the fluctuations of Σ MUFA in yolks during egg storage using lycopene or astaxanthin supplements in the diet of laying hens are consistent with the data of **[35]** on the distribution of fatty acids in yolks enriched with ω3 PUFA when stored under 4 °C.

The disappearance and redistribution of several saturated and unsaturated fatty acids in the yolks of freshly laid chicken eggs in our experiment may indicate the effect of different doses of astaxanthin on desaturase Δ6 activity in chicken tissues, as well as a complex mechanism of competition for the same enzyme essential polyunsaturated fats carbon chain extensions **[12], [27]**. In turn, the disappearance of certain saturated fatty acids in the yolks under the influence of different doses of astaxanthin during the storage of chicken eggs requires further research. It is difficult to compare our research results with other scientists' data because there is very little such information in the literature **[23]**. Most researchers have used sources of various oils to modify the fatty acid composition of egg yolk lipids **[9], [30], [42]**.

As can be seen from our research results, storage of edible chicken eggs enriched with lycopene or astaxanthin **[39]**, at both temperatures, helps to achieve a ratio of Σ SFA to Σ MUFA within 1:1, which corresponds to physiological parameters for humans **[29]**.

Our research results show that the storage of edible eggs obtained from laying hens that were not fed carotenoid supplements helps to increase the ratio of ω3/ω6 PUFA from 1:12.76 to 1:17.57, 17.34 depending on the temperature, which is not desirable for consumers' health. At the same time, the feeding of laying hens to the addition of lycopene at a dose of 60 mg/kg of feed can provide this ratio in freshly laid eggs at 1:7.95, and during the storage period − 1:10.28, 1:10.40 (Tables 2 − 4). Even better in this regard is the addition of astaxanthin in the diet of laying hens: starting with a dose of 20 mg/kg of feed, you can achieve a ratio of ω3/ω6 PUFA in the yolks of freshly laid eggs at 1:9.95 y, and at a dose of 30 mg/kg, respectively 1:9.35. It is important to maintain this ratio for both modes of egg storage (Tables 5 – 7). The researchers agree that the optimal ratio of ω3/ω6 PUFA should not exceed 1:2 − 1:4. This is because a nutritional imbalance in polyunsaturated fatty acids (excess ω6 and deficiency ω3) is a major cause of many chronic diseases, including cardiovascular disease, cancer, inflammatory diseases, autoimmune diseases, and many physiological disorders in humans **[6]**.

CONCLUSION

Temperature storage regimes of 4 ±0.5 °C and 12 ±0.5 °C eggs equally affect the fatty acid composition of egg yolks obtained from hens fed supplements of lycopene at doses of 30, 40, and 60 mg/kg or astaxanthin at doses of 10, 20, and 30 mg/kg of compound feed for 30 days, compared to freshly laid eggs. An increasing dose of lycopene from 20 to 60 mg/kg or astaxanthin from 10 to 30 mg/kg in the diet of laying hens in the yolks of freshly laid eggs, as well as at both temperatures of their storage decreases the proportion of ω6 PUFA: cis-8,11, 14-eicosatrienoic, and 6,9,12-ocadecatrienic acids until their complete disappearance. The addition of astaxanthin to the diet of laying hens is characterized by a much stronger effect on the ratio of saturated and unsaturated fatty acids in chicken egg yolks during storage than the addition of lycopene. Feeding astaxanthin to laying hens reduces and stabilizes the ω3/ω6 PUFA ratio in yolks during egg storage to a greater extent than adding lycopene to the diet of laying hens. The obtained research results can be the basis for the choice of storage regime of carotenoid-enriched edible chicken eggs, taking into account the correction of the fatty acid profile of yolk lipids.

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