doi: 10.17221/33/2016-CJFS

Quality of Eggs in Different Production Systems

Guler YENICE[1], Ozgur KAYNAR[2], Mustafa ILERITURK[2], Feryaz HIRA[3] and Armagan HAYIRLI[1]

[1]Department of Animal Nutrition and Nutritional Disorders, Faculty of Veterinary Medicine, Ataturk University, Erzurum, Turkey; [2]Department of Biochemistry, Faculty of Veterinary Medicine, Ataturk University, Erzurum, Turkey; [3]Hiraogullari Poultry Co., Erzincan, Turkey

ABSTRACT

Yenice G., Kaynar O., Ileriturk M., Hira F., Hayirli A. (2016): **Quality of eggs in different production systems**. Czech J. Food Sci., 34: 370–376.

This experiment was carried out to compare morphological egg quality parameters, as well as lipid and protein profiles, of brown eggs laid by chickens reared under different production systems: cage, free-range, and family type. A total of 270 brown eggs were obtained from commercial poultry companies raising Lohmann Brown laying hens in a cage system and free-range unit as well as families possessing hens in their yards. The egg lipid and protein contents, as well as lipid and protein profile, varied among the production systems. However, eggs from the free-range system had similar characteristics to those from the cage system. Quality of eggs from the family type system was quite variable. In conclusion, egg quality can be affected by the production system.

Keywords: egg quality; housing; welfare; HPTLC, SDS-PAGE

The egg is one of the cheapest and most commonly consumed foods for human nutrition. The egg is rich in high-quality protein. Moreover, it is an almost complete vitamin and trace mineral source (Sparks 2006). The external and internal qualities of eggs are very important for consumer health and from a marketing perspective. The egg yield and quality are affected by a number of factors, such as the type of husbandry system (Ozbey & Esen 2007; Radu-Rusu *et al*. 2014). For instance, hens housed in large furnished cages had lower productivity and higher egg quality than those housed in small furnished and conventional cages (Meng *et al*. 2014).

Different husbandry systems are available in laying hen breeding such as free-range, organic, cage, and furnished cage. The cage type is one of the most common husbandry systems, which is a controversial subject among advocates for animal welfare and animal rights. The European Union banned battery cage husbandry of chickens in January 2012 for welfare reasons (Leenstra *et al*. 2014). This experiment was carried out to compare morphological egg quality parameters as well as egg lipid and protein profiles of brown eggs collected from poultry farms with different production systems: cage, free-range, and family type.

MATERIAL AND METHODS

Egg samples. A total of 270 eggs, 90 from each of the three systems, were collected from (1) a poultry farm with cage system, (2) a free-range poultry farm with nest and 2 m^2 yard per bird, and (3) families possessing chickens in their yard in two villages. Eggs from the first two farms were collected from Lohmann Brown hybrid chickens at the age of 40 weeks that were fed conventional feed formulated to meet nutrient recommendations of the NRC (1994). Eggs collected from the family type units were variable in terms of breed, age, and dietary components of the chickens.

All eggs were stored at room temperature for 3 days before performing morphological and biochemical analyses. Morphological characteristics were determined in individual eggs. For biochemical assays, fractions of three eggs were pooled in tubes. Thus, sample sizes per group were 90 and 30 for morphological data and biochemical data, respectively.

Morphological measurements. The egg weight was measured with an electronic balance to the nearest 0.01 g. The egg shape index (%) was calculated by the normal method of (diameter/height) × 100.

doi: 10.17221/33/2016-CJFS

The eggshell strength (kg/cm^2) was measured using a cantilever system by applying increased pressure to the broad pole of the shell using an instrument. The egg yolk diameter, albumen length, and albumen width (mm) were measured with a digital calliper. The albumen and yolk height (mm) was measured using a tripod micrometer.

The yolk (YI) and albumen (AI) indices were calculated using the following formulae as described by Doyon *et al*. (1986):

$$YI = (\text{yolk height}/\text{yolk diameter}) \times 100$$

$$AI = [\text{albumen height}/(\text{albumen length} + \text{albumen width})/2)] \times 100$$

The Haugh Unit (HU) score was calculated using the formula as described by Haugh (1937):

$$HU = 100 \times \log (H + 7.5 - 1.7W^{0.37})$$

where: H – albumen height (mm); W – egg weight (g)

The eggshell thickness was measured after removing the internal membranes of the eggshell, using a micrometer which has the precision of 0.01 mm. Measurements were taken at three regions (middle and two ends) of the shell and then averaged. The yolk colour was determined according to Roche yolk colour fan.

Isolation and homogenisation of egg yolk and albumen. The eggshell was broken manually and the albumen was transferred into pre-weighed Falcon tubes. The yolk was washed gently with distilled water and rolled on Whatman filter paper to remove the albumen residue. Then the yolk membrane was punctured with a sterile pipette tip and the yolk transferred into another pre-weighed Falcon tube. The egg component (1 g) was mixed with 2.5 ml of solubilisation buffer (1% SDS, 0.5% Triton X100, 0.5 M Tris HCl, pH 6.8) and then homogenised at 3000 oscillations/min, +4°C, for 1 min using a homogeniser (Tissuelyser LT; Qiagen, Hilden, Germany). The homogenates were aliquoted into Eppendorf tubes and stored at –86°C until biochemical analyses.

Sodium dodecyl sulphate polyacrylamide gel electrophoresis. Protein samples were diluted at 1 : 9 with Sodium Dodecyl Sulphate Polyacrylamide Gel Electrophoresis (SDS-PAGE) denaturing electrophoresis sample buffer and 10 µl of the mixture was loaded into each well. The electrophoresis was carried out in Tris-glycine-SDS running buffer at 20 mA/gel constant current for 90 min using 4% stacking and 10% resolving gel (Laemmli 1970). After completion of running, proteins were treated with Oriole fluorescent stain (Bio-Rad, Herkules, USA). The electrophoretograms were then visualised using the GelDoc XR gel documentation system (Bio-Rad, USA) and were analysed by the ImageLab 5.2 software (Bio-Rad, USA).

High performance thin layer chromatography. An n-hexane/isopropanol (500 µL) mixture at a 3 : 2 ratio (v/v) was added to 1000 µl of yolk homogenates in Eppendorf tubes. After vigorous vortexing the tubes were centrifuged at 5000 *g* at +4°C for 5 min and the upper phases were used for the chromatographic analysis of yolk lipids (Hara & Radin 1978). A standard lipid mixture (cholesteryl oleate, triolein, palmitic acid, cholesterol, glyceryl monooleate, and L-α-phosphatidylcholine) and yolk extracts were spotted on the high performance thin layer chromatography (HPTLC) plates and developed with an *n*-hexane/diethyl ether/formic acid mixture (80 : 20 : 2) (v/v/v). Then the entire plate was dipped in 10% $CuSO_4$ (w/v), in 8% H_3PO_4 (v/v) and lipid classes were visualised by charring the plates at 120°C. Egg yolk lipids were separated into the following classes: cholesterol esters (CE), triacylglycerols (TAG), free fatty acids (FFA), cholesterol (COL), monoacylglycerols (MAG), and phospholipids (PL).

The HPTL chromatograms were analysed as described by Kaynar *et al*. (2013) and results were obtained as percentage of individual lipid class in the total lipid composition.

Determination of triacylglycerol and total protein concentrations. One ml of triacylglycerol (TAG) reagent [4-chlorophenol 3.5 mM, ATP > 0.5 mM, magnesium salt 10 mM, 4-aminophenazone 0.3 mM, microbial glycerol kinase > 250 U/l, microbial glycerol phosphate oxidase > 4500 U/l, horseradish peroxidase > 2000 U/l, microbial lipase > 200.000 U/l, buffer (pH 7.3), sodium azide (0.01%)] was added to 100 µl of yolk samples. After incubating for 30 min, absorbance of yolk samples was read at 505 nm (Fossati & Prencipe 1982). Results were calculated using a standard TAG solution (50 mg/dl) and expressed as gram of TAG per 100 gram of yolk weight (g/100 g).

For the total protein (TP) analysis, albumen and yolk samples (100 µl) were mixed with 0.5 ml of sodium deoxycholate (10%) and 0.5 ml of TCA (10%). The mixtures were incubated at 37°C for 30 min and then centrifuged at 5000 *g* at +4°C for 5 minutes. The precipitates were dissolved in 5.0 ml of 0.1 N NaOH and 5.0 ml of alkaline copper reagent was

doi: 10.17221/33/2016-CJFS

added into the same tubes. After 10 min, 0.5 ml Folin-Ciocalteu reagent was added. Following incubation at room temperature for 30 min, absorbances of albumen and yolk samples were read at 660 nm in a spectrophotometer (μ-Quant; BioTek) against the blank solution (Lowry *et al*. 1951). Results were calculated using a standard protein solution (5 g/dl) and expressed as gram of TP per 100 g of albumen and yolk weight (g/100 g).

Statistical analysis. All data were analysed using the PROC GLM procedure of statistical analysis software (SAS 9.4, 2013). Differences in egg quality parameters between the production systems were attained using the *LSD* option at $P < 0.05$.

RESULTS

Table 1 summarises the morphological characteristics of eggs collected from different production systems. Egg weight was the highest (62.53 g) in the cage system, followed by the free-range system (58.14 g) and the lowest (54.02 g) weight was in the family type system ($P < 0.05$). The shape index, eggshell weight, shell thickness and shell stiffness for the cage and free-range systems were similar and greater than those for the family type system ($P < 0.05$). Yolk colour of eggs obtained from the family type system was superior to that obtained from the cage and free-range systems ($P < 0.05$). The cage system was superior to the free-range and family type systems in terms of both yolk index and HU ($P< 0.05$). Albumen index was not different among eggs collected from these chicken production systems.

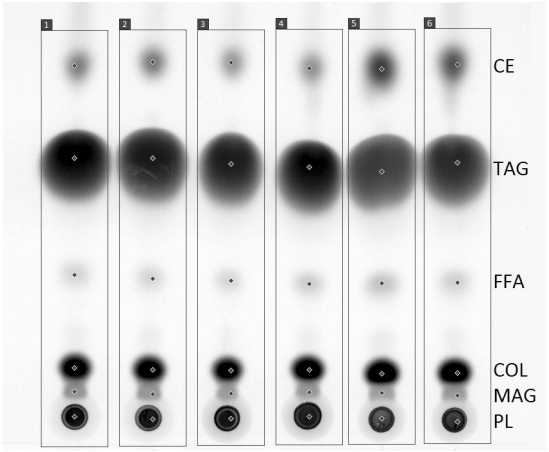

Figure 1. HPTL chromatogram of egg yolk lipids from different production systems

Lane 1–2: free range; Lane 3–4: cage; Lane 5–6: family type; CE – cholesterol esters; TAG – triacylglycerol; FFA – free fatty acids; COL – cholesterol; MAG – monoacylglycerol; PL – phospholipids

Lipid profile. Eggs from the family type housing contained the highest percentages of CE, FFA, and COL in total lipids, while they contained the lowest percentages of MAG and PL as compared to eggs from the cage and free-range systems, respectively (Figures 1 and 2, Table 2). Moreover, yolk triacylglycerol concentrations were in the order of family type > free-range system > cage system (Table 2).

Protein profile. In the egg white, 20 proteins ranging between 8 and 240 kDa molecular weight were determined after electrophoresis. The highest concentration of proteins with 230.0, 200.0, 54.0, 50.4, 22.6, and 19.3 kDa molecular weight was in the free-range system; 157.6 and 103.3 kDa were in

Table 1. Effect of different production systems on the quality parameters of eggs

Parameter	Production system		
	cage	free-range	family type
Egg weight (g)	62.53 ± 0.51^{a}	58.14 ± 0.39^{b}	54.02 ± 0.81^{c}
Shape index (%)	77.74 ± 0.24^{a}	78.01 ± 0.25^{a}	74.55 ± 0.40^{b}
Eggshell weight (g)	7.66 ± 0.07^{a}	7.44 ± 0.09^{a}	6.62 ± 0.11^{b}
Eggshell weight (%)	12.28 ± 0.22^{b}	12.81 ± 0.14^{a}	12.31 ± 0.16^{b}
Shell stiffness (kg/cm^2)	2.70 ± 0.12^{a}	2.85 ± 0.10^{a}	2.13 ± 0.12^{b}
Eggshell thickness (mm)	0.39 ± 0.002^{a}	0.39 ± 0.003^{a}	0.35 ± 0.04^{b}
Yolk colour	10.36 ± 0.09^{b}	10.42 ± 0.07^{b}	11.85 ± 0.21^{a}
Yolk index (%)	41.89 ± 0.26^{a}	40.77 ± 0.22^{b}	40.57 ± 0.38^{b}
Albumen index (%)	6.00 ± 0.16	5.68 ± 0.17	6.05 ± 0.24
Haugh unit	70.10 ± 0.89^{a}	67.81 ± 0.99^{ab}	66.65 ± 1.48^{b}

Different superscripts among columns differ ($P < 0.05$); all the values are expressed as mean ± SE, $n = 90$ per group

doi: 10.17221/33/2016-CJFS

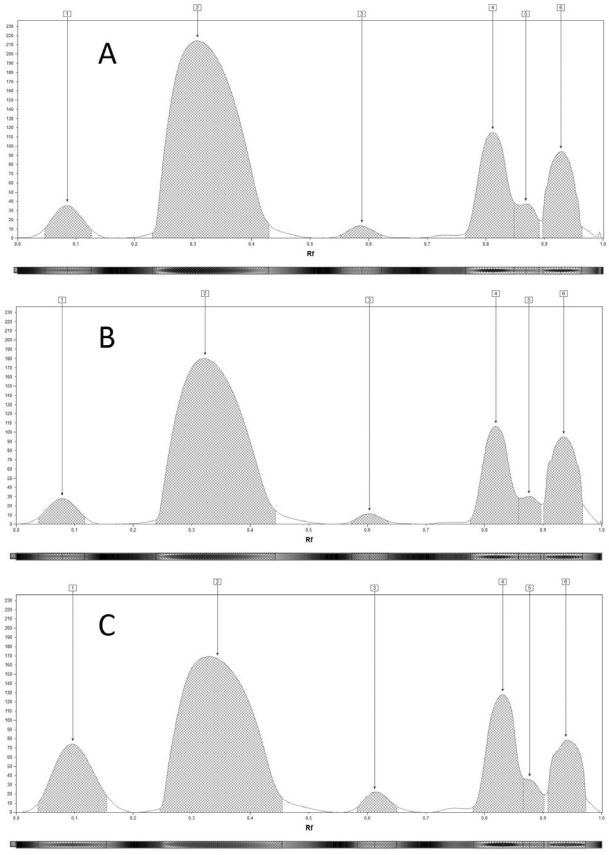

Figure 2. HPTL densitogram of egg yolk lipids from different production systems
Panel A – free range; Panel B – cage; Panel C – family type

cages; another 12 proteins (represent > 87% of total egg white proteins) including especially 59.3 and 33.0 kDa molecular weights that comprise > 50% of total egg white proteins were in the family type husbandry (Figures 3 and 4).

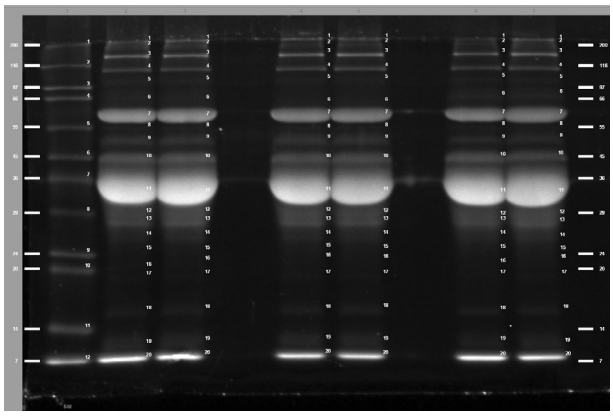

Figure 3. Electrophoretogram of egg white proteins from different production systems
Lane 1: molecular weight marker; Lane 2–3: free range; Lane 4–5: cage; Lane 6–7: family type.

On the other hand, 24 proteins in the range between 7 and 200 kDa molecular weight were determined in egg yolk after electrophoresis. The highest concentration of proteins with 104.0, 66.0, 57.7, 51.3, 45.0, 34.7, 11.2, and 7.0 kDa molecular weight was in the free-range system; 113.7 and 106.0 kDa were in cages; and another 14 proteins (represent > 70% of egg yolk proteins) were in the family type husbandry (Figures 5 and 6).

In parallel with the increase in number of individual proteins, total protein concentrations of yolk and albumen proteins were in the order of the family type > the free-range > the cage systems (Table 2).

Table 2. Effect of different production systems on the lipid and protein contents of eggs

Parameter	Production system		
	cage	free-range	family type
Albumen total protein (g/100 g)	10.72 ± 0.05^{c}	11.01 ± 0.05^{b}	11.75 ± 0.06^{a}
Yolk total protein (g/100 g)	14.23 ± 0.06^{c}	15.25 ± 0.07^{b}	16.55 ± 0.05^{a}
Yolk triacylglycerol (g/100 g)	20.31 ± 0.18^{c}	21.44 ± 0.14^{b}	23.19 ± 0.17^{a}
Fractions of yolk lipids			
– CE (%)	4.78 ± 0.06^{c}	5.86 ± 0.06^{b}	12.82 ± 0.05^{a}
– TAG (%)	63.14 ± 0.10^{b}	64.26 ± 0.10^{a}	55.00 ± 0.08^{c}
– FFA (%)	1.67 ± 0.04^{b}	1.51 ± 0.02^{c}	2.36 ± 0.03^{a}
– COL (%)	14.86 ± 0.06^{b}	14.96 ± 0.07^{b}	16.70 ± 0.07^{a}
– MAG (%)	2.56 ± 0.03^{b}	2.87 ± 0.03^{a}	2.66 ± 0.03^{b}
– PL (%)	12.99 ± 0.07^{a}	10.54 ± 0.05^{b}	10.46 ± 0.05^{b}

Different superscripts among columns differ ($P < 0.05$); all the values are expressed as mean ± SE, $n = 30$ per group; CE – cholesterol esters; TAG – triacylglycerol; FFA – free fatty acids; COL – cholesterol; MAG – monoacylglycerol; PL – phospholipids.

doi: 10.17221/33/2016-CJFS

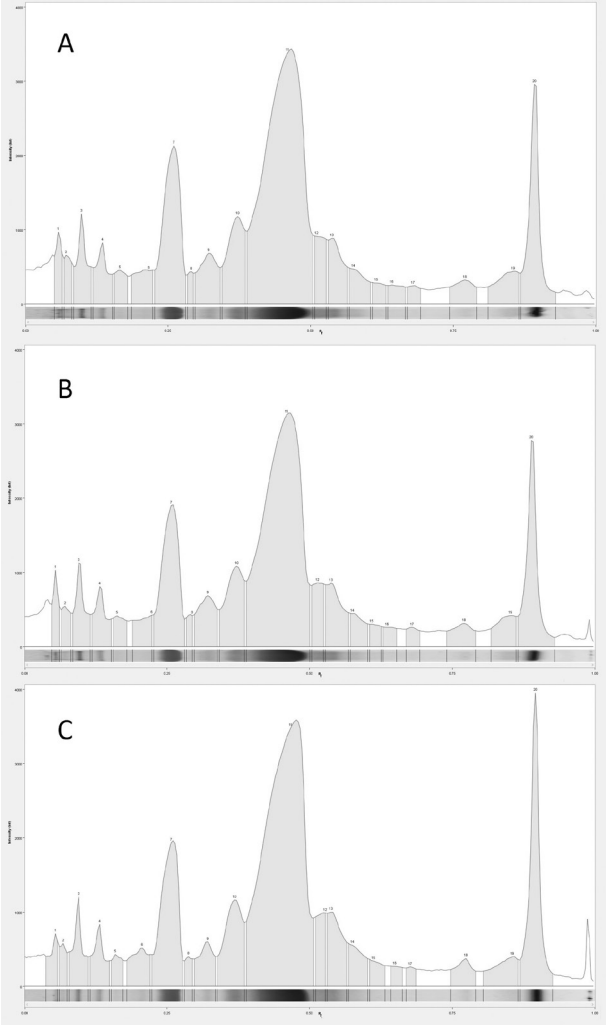

Figure 4. Densitogram of egg white proteins from different production systems
Panel A – free range; Panel B – cage; Panel C – family type

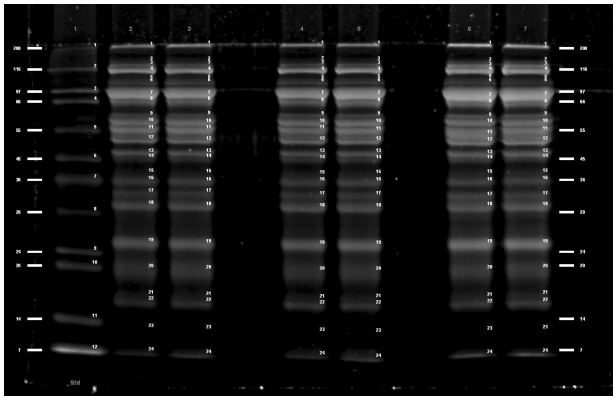

Figure 5. Electrophoretogram of egg yolk proteins from different production systems
Lane 1: molecular weight marker; Lane 2–3: free range; Lane 4–5: cage; Lane 6–7: family type

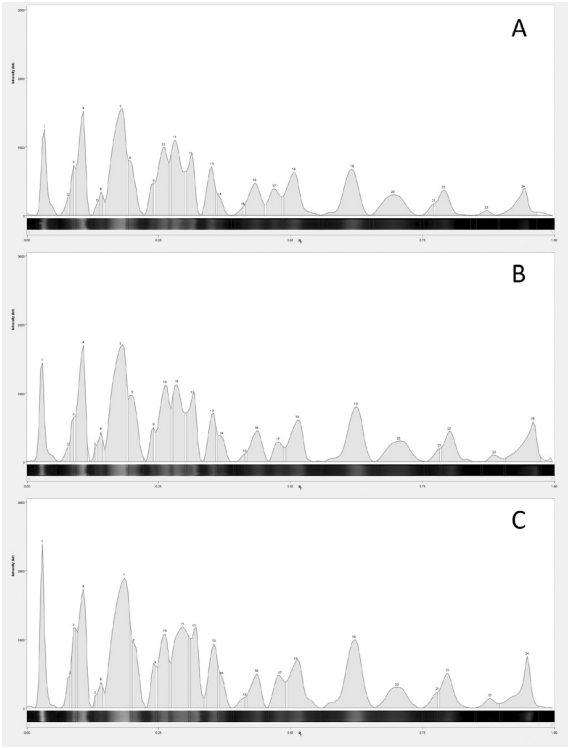

Figure 6. Densitogram of egg yolk proteins from different production systems
Panel A – free range; Panel B – cage; Panel C – family type

DISCUSSION

All egg quality parameters differed by the husbandry system, except for albumen index. The deepest yolk colour was observed in the family type system. Shell breaking strength, eggshell thickness, shape index, yolk colour in conventional and free-range systems were similar but significantly ($P < 0.05$) different from that of the family type system. Similarly, Sekeroglu *et al.* (2010) reported that there was no difference between free range and cage systems. In agreement with a report by Pištěková *et al.* (2006), a significant difference in egg weights among the husbandry systems was observed ($P < 0.05$). Conversely, Sekeroglu *et al.* (2008) reported that weights of eggs were not affected by production system in a similar study.

Eggshell quality (breaking strength, weight, and thickness) could be related to the Ca level of the shell. It is generally accepted that dietary Ca supplementation should play an important role in maintaining the good eggshell quality (Arpášová *et al.* 2010). High variability in eggshell quality from the family type system could be related to a diet which is uncontrollable.

doi: 10.17221/33/2016-CJFS

Welfare is critical for poultry production (Blokhuis 1994). The cages, while the most feasible system for hen housing, negatively influence the welfare of hens (Appleby 1993; Craig & Swanson 1994), because hens are not able to express natural behaviours including nesting, roosting, and scratching (Nicol 1987; Baxter 1994). When a bird is first exposed to unfavourable environmental conditions, physiological stress occurs and the hypothalamic-pituitary-adrenal cortical system is activated by the action of catecholamines. Secretion of the adrenocorticotropic hormone triggers the release of corticosterone (CS) from the adrenal cortex (Romero & Butler 2007; Virden & Kidd 2009), which reduces the synthesis and increases the degradation of proteins in skeletal tissues (Puvadolpirod & Thaxton 2000) and increases the rate of gluconeogenesis (Virden & Kidd 2009). Moreover, CS assists catecholamines to initiate lipolysis and increase fat deposition (Virden & Kidd 2009). The rate of free fatty acid release from adipose tissue is affected by many hormones that influence either the rate of esterification or the rate of lipolysis. Insulin inhibits the release of free fatty acids from adipose tissue, which is followed by a fall in circulating plasma free fatty acids. It enhances lipogenesis and the synthesis of acylglycerol. Besides, epinephrine, norepinephrine, adrenocorticotropic hormone (ACTH), and corticosterone accelerate the release of free fatty acids from adipose tissue and raise the plasma free fatty acid concentration by increasing the rate of lipolysis of the triacylglycerol stores. Glucocorticoids also promote lipolysis via synthesis of new lipase protein (Murray 2009). These data indicate that the blood chemistry of birds is tightly associated with animal welfare. Thus, stress factors in relation to the production system alter blood protein and lipid levels of hens (Puvadolpirod & Thaxton 2000; Virden & Kidd 2009).

The chemical content of the egg is directly linked to the blood chemistry of hen. Egg yolk lipids such as glycerides, phospholipids, cholesterol – cholesterol esters, and other lipids (Christie & Moore 1972b) are transported from the blood (Christie & Moore 1972a, b) by blood lipoproteins – VLDL (Burley *et al*. 1984). Yolk proteins (vitellogenin, livetins) are synthesised by the liver (Wallace 1985).

Egg nutrients are transported to developing follicles via the vascular system (Moran 1987) and the present study questioned if eggs differed in morphological properties and lipid protein profile according to the production systems that are, in turn, related to welfare. The highest egg yolk and albumen protein concentrations and the highest egg yolk total lipid concentrations were determined in eggs of family type hens while the lowest values were in cage hens. When the lipid profiles of egg yolk were analysed, the highest percentage of FFA, with the lowest percentage of TAG and PL, were determined in family type eggs. Moreover, the highest FFA/TAG, MAG/TAG ratios with the highest total triacylglycerol concentration were in family type eggs. When considering only the lipid profile, the increase in the percentages of lipolysis end-products including FFA and MAG, with a decrease in the percentage of TAG, suggested a stress related lipolysis profile in family type eggs. However, when the lipid profiles were evaluated with the concentration of TAG, it was considered that the changes in lipid profiles were related to the physical activity of animals rather than to stress. Besides, despite the balanced diet, the poorest eggs were in cages when considering the total lipid and protein concentrations of all groups.

CONCLUSSION

In summary, egg lipid and protein contents, as well as lipid and protein profile, varied among the production systems (Figures 1–6 and Table 2). However, contrary to expectations, eggs from the free-range system had similar characteristics to those from the cage system. Therefore, the free-range system does not always represent the better welfare because of cannibalism, lack of feed, diseases, parasites etc., which can affect the quality of eggs. On the other hand, the best egg in terms of the nutritional value was from family-type chickens living in their natural environment. The most striking egg quality parameter in eggs collected from different production system was yolk colour, which in part could also be due to variations in breed and age of hens and diets consumed. However, as only one farm was sampled for each production system, the data should be used with caution.

References

Appleby M.C. (1993): Should cages for laying hens be banned or modified? Animal Welfare, 2: 67–80.

Arpášová H., Halaj M., Halaj P. (2010): Eggshell quality and calcium utilization in feed of hens in repeated laying cycles. Czech Journal of Animal Science, 55: 66–74.

Baxter M.R. (1994): The welfare problems of laying hens in battery cages. Veterinary Record, 134: 614–619.

doi: 10.17221/33/2016-CJFS

Blokhuis H.J. (1994): Intensive production units and welfare: domestic fowl. Revue Scientifique et Technique (International Office of Epizootics), 13: 67–78.

Burley R.W., Sleigh R.W., Shenstone F.S. (1984): Lipoproteins from the blood and egg yolk of the hen. the transfer of very-low-density lipoprotein to egg yolk and possible changes to apoprotein B. European Journal of Biochemistry, 142: 171–176.

Christie W.W., Moore J.H. (1972a): The lipid components of the plasma, liver and ovarian follicles in the domestic chicken. Comparative Biochemistry and Physiology - Part B: Biochemistry & Molecular Biology, 41: 287–295.

Christie W.W., Moore J.H. (1972b): The lipid composition and triglyceride structure of eggs from several avian species. Comparative Biochemistry and Physiology - Part B: Biochemistry & Molecular Biology, 41: 297–306.

Craig J.V., Swanson J.C. (1994): Welfare perspectives on hens kept for egg production. Poultry Science, 73: 921–938.

Doyon G., Bernier-Cardou M., Hamilton R.M.G., Castaigne F., Randall C.J. (1986): Egg quality. 2. Albumen quality of eggs from five commercial strains of white leghorn hens during one year of lay. Poultry Science, 65: 63–66.

Fossati P.L., Prencipe L. (1982): Serum triglycerides determined colorimetrically with an enzyme that produces hydrogen peroxide. Clinical Chemistry, 28: 2077–2080.

Hara A., Radin N.S. (1978): Lipid extraction of tissues with a low-toxicity solvent. Analytical Biochemistry, 90: 420–426.

Haugh R.R. (1937): The haugh unit for measuring egg quality. U.S. Egg Poultry Magazine, 43: 552–555.

Kaynar O., Ileriturk M., Hayirli A. (2013): Evaluation of computational modifications in hptlc with gel analysis software and flatbed scanner for lipid separation. JPC - Journal of Planar Chromatography - Modern TLC, 26: 202–208.

Laemmli U.K. (1970): Cleavage of structural proteins during the assemble head of bacteriophage T4. Nature, 227: 680–685.

Leenstra F., Maurer V., Galea F., Bestman M., Amsler-Kepalaite Z., Visscher J., van Krimpen M. (2014): Laying hen performance in different housing systems; why do they differ and how to close the gap? Results of discussions with groups of farmers in the Netherlands, Switzerland and France, benchmarking and model calculations. European Poultry Science, 78: 1–10.

Lowry O.H., Roserbrough N.J., Farm A.L., Randal R.J. (1951): Protein measurement with folin phenol reagent. Journal of Biological Chemistry, 193: 265–275.

Meng F., Chen D., Li X., Li J., Bao J. (2014): Effects of large or small furnished cages on performance, welfare and egg quality of laying hens. Animal Production Science, 55: 793–798.

Moran Jr E.T. (1987): Protein requirement, egg formation and the hen's ovulatory cycle. The Journal of Nutrition, 117: 612–618.

Murray R. K. (2009): Harper's Illustrated Biochemistry. New York, McGraw-Hill Medical and London, McGraw-Hill.

Nicol C.J. (1987): Behavioural responses of laying hens following a period of spatial restriction. Animal Behaviour, 35: 1709–1719.

NRC (1994): Nutrient Requirements of Poultry. 9th Ed. Washington DC, National Academy Press.

Ozbey O., Esen F. (2007): The effects of different breeding systems on egg productivity and egg quality characteristics of rock partridges. Poultry Science, 86: 782–785.

Pištěkova V., Hovorka M., Večerek V., Straková E., Suchý P. (2006): The quality comparison of eggs laid by laying hens kept in battery cages and in a deep litter system. Czech Journal of Animal Science, 51: 318–325.

Puvadolpirod S., Thaxton J.P. (2000): Model of physiological stress in chickens. 4. Digestion and metabolism. Poultry Science, 79: 383–390.

Radu-Rusu R.M., Usturoi M.G., Leahu A., Amariei S., Radu-Rusu C.G., Vacaru-Opris I. (2014): Chemical features, cholesterol and energy content of table hen eggs from conventional and alternative farming systems. South African Journal of Animal Science, 44: 33–42.

Romero M.L., Butler L.K. (2007): Endocrinology of stress. International Journal of Comparative Psychology, 20: 89–95.

Sekeroglu A., Sarica M., Demir E., Ulutas Z., Tilki M., Saatci M. (2008): The effects of housing system and storage length on the quality of eggs produced by two lines of laying hens. Archiv für Geflügelkunde, 72: 106–109.

Sekeroglu A., Sarica M., Demir E., Ulutas Z., Tilki M., Saatci M., Omed H. (2010): Effects of different housing systems on some performance traits and egg qualities of laying hens. Journal of Animal and Veterinary Advances, 9: 1739–1744.

Sparks N.H.C. (2006): The hen's egg – is its role in human nutrition changing? World's Poultry Science Journal, 62: 308–315.

Virden W.S., Kidd M.T. (2009): Physiological stress in broilers: ramifications on nutrient digestibility and responses. Journal of Applied Poultry Research, 18: 338–347.

Wallace R.A. (1985): Vitellogenesis and oocyte growth in nonmammalian vertebrates. In: Browder L.W. (ed.): Developmental Biology. Vol. 1. New York, Plenum Press: 127–177.

Received: 2016–01–29
Accepted after corrections: 2016–07–13

Corresponding author:
Ass Prof Guler Yenice, Ataturk University, Faculty of Veterinary Medicine, Department of Animal Nutrition and Nutritional Disorders, Erzurum, Turkey; E-mail: gulerata@atauni.edu.tr, gulerata2003@hotmail.com