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**Original Paper**

# The effect of essential oils on quality and mineral composition of eggshells

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The aim of study was to analyse the dietary effect of pumpkin and flaxseed oils on quality and mineral composition of laying hens eggshell. At 38 weeks of age, Lohmann Brown Lite hens were housed in three-floor cages, divided into three dietary groups (C-control, E1-pumpkin oil (3%), E2-flaxseed oil (3%)). There were housed six hens in one cage. A total 18 hens were monitored. In the control group hens were fed with standard complete feed mixture for laying hens and in the experimental groups by feed mixtures with supplementation of pumpkin or flaxseed oils. Vitamin E was added into feed mixture in the experimental groups. The experiment lasted 52 days. Twelve eggs from each dietary treatment were randomly selected and analyzed. Significant ($P$ <0.05) differences between control and both experimental groups in eggshell strength and eggshell thickness were found. Pumpkin and flaxseed oil supplementations in feed ratio of layers had positive impact on quality of eggshell. As regards the mineral composition of eggshell, significantly ($P$ <0.05) lower contents of magnesium, sodium, potassium and copper in experimental groups compared to the control were observed in eggshell. Significant ($P$ <0.05) differences in content of calcium were detected only between E1 (pumpkin addition) and E2 (flaxseed addition).

**Keywords:** eggshell, feed mixture, flaxseed oil, minerals, pumpkin oil

## 1 Introduction

Eggs are made up of a variety of chemical components, including water, protein, fatty acids, minerals, vitamins, and pigments (Li-Chan and Kim, 2008). Eggs are very good source of vitamins A, D, E a K and whole complex of vitamins of group B also comprising $B_{12}$ which are necessary for well-functioning nervous system. Also includes folic acid, iron, phosphorus, sulphur, potassium, magnesium and from trace elements it is zinc, copper, manganese, bromine and iodine (Nagy et al., 2009). In recent years global consumers are concerned about the livestock and poultry product due to the addition of additives into the feedstuff (European Commission, 2007). Vegetable oils, also called essential oils are aromatic, oily liquids obtained from plant material (flowers, buds, seeds, leaves, twigs, bark, herbs, wood, fruits and roots). It is a complex mixture of different organic molecules – terpenes, alcohols, esters, aldehydes, ketones and phenols. In animals, in particular, promote the secretion of gastric juices, while operating on gut motility and improve the integrity of the intestinal lining (Panda et al., 2009). Pumpkin seeds contain L-tryptophan, n-6 and n-3 fatty acids and very high concentration of vitamin E (Hashemi, 2013). Pumpkin seeds also contain zinc and pumpkin peanuts to 54% oil with digestibility coefficient of 98.2% (Kóňa et al., 2007). Pumpkin seed oil has become a recognized source of phenolic compounds (Andjelkovic et al., 2010). Pumpkin oil has strong antioxidant properties (Stevenson et al., 2007). Flax seed contains about 40% oil, which can be used as part of feed mixture for poultry (Zelenka et al., 2008). Flaxseed oil contains high levels of polyunsaturated fatty acids and these compounds have been reported to reduce the incidence of heart disease and cancer in humans (Zhao et al., 2007). The main goal of this work was to determine the effect of essential oils on qualitative parameters and mineral composition of eggshell.

## 2 Material and methods

The experiment was carried out in accredited testing station of the Department of Poultry Science and Small Animal Husbandry (Faculty of Agrobiology and Food Resources, Slovak University of Agriculture in Nitra). At 38 weeks of age, Lohmann Brown Lite hens were housed in three-floor cages, divided into three diets of groups (C – control, E1 – pumpkin oil, E2 – flaxseed oil). There were

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**Table 1** Nutrient composition of feed mixtures

| Feed mixture components | C | E1 | E2 |
|---|---|---|---|
| Dry matter (%) | 91.12 | 90.88 | 91.4 |
| Crude protein (% of DM) | 17.68 | 17.81 | 17.45 |
| Crude Fat (% of DM) | 6.53 | 7.61 | 7.59 |
| Crude Fibre (% of DM) | 4.09 | 3.71 | 3.78 |
| Ash (% of DM) | 12.01 | 11.49 | 11.26 |
| NFE (% of DM) | 50.81 | 50.26 | 51.32 |
| Organic matter (% of DM) | 79.11 | 79.39 | 80.14 |
| Starch (% of DM) | 35.3 | 35.62 | 35.45 |
| Sugar (% of DM) | 3.9 | 3.84 | 3.94 |
| Ca (mg $kg^{-1}$ DM) | 41,468 | 38,567 | 39,059 |
| P (mg $kg^{-1}$ DM) | 8,855 | 8,436 | 8,060 |

NFE – nitrogen-free extract, DM – dry matter, C – control group, E1 – pumpkin oil addition, E2 – flaxseed oil addition

six hens in one cage housed. The useful area provided for one laying hen presented 943.2 $cm^2$. Total 18 hens were monitored. The laying hens were kept in the standard bioclimatic and welfare conditions. During experiment, the light regime was 16 hours. The experiment lasted 52 days. In the control group (C), hens were fed with standard complete feed mixture for laying hens, in the experimental groups by feed mixtures supplemented with pumpkin (E1) or flaxseed (E2) oils. Concentration of oils was 3%. Laying hens in all groups received drinking water and feed mixture *ad libitum*. Nutrient composition of feed mixtures is presented in Table 1. At the last week the eggs were collected and processed. Shell thickness was measured at five locations on the egg (sharp, blunt and 3 equator end) using electronic micrometer Digitale Schiebelehre (Mister Tool). Strength of shell was determined using shell strength device with a spiral pressure system. Mineral composition of eggshell was determined by standard laboratory methods and procedures (AOAC, 2000). Content of ash was determined by complete combustion of the sample in a muffle furnace at 550 °C ±25 °C (4–6 hours) to oxidize all organic matter. The contents of mineral nutrients were determined by High Resolution Continuum Source Atomic Absorption Spectrometer ANALYTIK JENA contrAA 700 (Ca, Mg, Na, K, Zn, Cu, Fe, Mn) and 6400 Spectrophotometer (P). Laboratory analyses of minerals were carried out in the Laboratory of Quality and Nutritional Value of Feeds at the Department of Animal Nutrition in Slovak University of Agriculture. Statistical analysis was evaluated by using the statistical programme IBM SPSS 20.0.

## 3 Results and discussion

Eggshell strength is one of the most important external quality parameters of an egg, usually dependent on egg shell proportion and thickness. Differences in egg shell physical parameters are dependent, among other factors, on the rate and extent of mineral deposition in the egg shell (Tůmová et al., 2014). Eggshell strengths of bursting and eggshell thickness are expressed in Table 2. Significant ($P$ <0.05) differences between control and both experimental groups in strength and thickness of shell were found. Average values of shell strength ranged from 21.76 N $cm^{-2}$ in control to 36.44 N $cm^{-2}$ in E1 (pumpkin oil addition). Arpášová et al. (2015) indicated eggshell strength 28.41 N $cm^{-2}$ after adding of oregano oil in diet. After oils supplementation, average eggshell thickness was same in both experimental groups (0.36 mm). Pumpkin and flaxseed oil supplementations in feed ratio of layers had positive impact on quality of eggshell. A very high positive correlation was found in eggshell strength ($r$ = 0.99) and eggshell thickness ($r$ = 1.00) to content of fat in feed mixture. In a previous study (Chetty et al., 2004), it was shown that essential oil supplementation could increase the concentration of calcium in the liver of non-ruminant animals. Vitamin D, which is a fat-soluble vitamin, has a direct effect on calcium absorption. Essential oil is considered to be a good solvent of vitamin D, and thus can improve calcium concentrations in eggshells (Bar et al., 1999). However, in present study significant differences in calcium and phosphorus contents between control and experimental groups in eggshell were not indicated (Table 3). Therefore it is clear that higher content of fat in diets is major factor which caused eggshell quality parameters in this trial. Similar results in comparison with present study were found by Zhang and Kim (2014) that researched effect of dietary olive oil on egg quality parameters of laying hens. Eggshell breaking strength and eggshell thickness were improved in the

**Table 2** Eggshell strength and eggshell thickness

| Group | Statistical parameter | Eggshell strength of bursting | | Eggshell thickness |
|---|---|---|---|---|
| | | mV | N cm$^{-2}$ | mm |
| C | $\bar{x}$ | 0.41$^{a}$ | 21.76$^{a}$ | 0.28$^{a}$ |
| | S.D. | 0.04 | 2.12 | 0.03 |
| | C.V. (%) | 9.76 | 9.74 | 10.71 |
| E1 | $\bar{x}$ | 0.69$^{b}$ | 36.44$^{b}$ | 0.36$^{b}$ |
| | S.D. | 0.18 | 9.47 | 0.02 |
| | C.V. (%) | 26.09 | 25.99 | 5.56 |
| E2 | $\bar{x}$ | 0.67$^{b}$ | 35.49$^{b}$ | 0.36$^{b}$ |
| | S.D. | 0.2 | 10.65 | 0.03 |
| | C.V. (%) | 29.85 | 30.01 | 8.33 |

C – control group, E1 – pumpkin oil addition, E2 – flaxseed oil addition, $\bar{x}$ – mean, S.D. – standard deviation, C.V. – coefficient of variance. Values with different superscript in a column are significant at the 0.05 level

treatment with addition olive oil (5% in feed mixture) as compared to the control. In the study Arpášová et al. (2014), who added oregano essential oil and into diets of layers, significant increase was recorded in eggshell thickness after oregano oil addition (1 ml kg$^{-1}$) compared to the control. Higher eggshell strength was found in experimental group (oregano oil) in comparison with control group ($P$ >0.05). In contrast, Cherian et al. (2009) studied effect of *Camelina sativa* (oilseed crop) in the diet of layer birds on egg parameters. Eggshell thickness of experimental eggs (5, 10 and 15% addtition of *Camela* meal) was lower compared to the control eggs. However, differences among groups were not significant ($P$ >0.05). Shakoor et al. (2002), Shakoor et al. (2003) and Kucukersan et al. (2010) also showed non-significant changes of egg quality characters in laying hens fed diet supplemented with oils. Arpášová et al. (2015) who researched influence of different doses of essential oils reported insignificant differences in eggshell strength and eggshell thickness. Eggshell strength was lower after thyme oil addition in diet (0.5 and 1 ml kg$^{-1}$) and higher after oregano oil addition in diet (1 ml kg$^{-1}$) compared to the control. Values of eggshell strength ranged from 27.17 (thyme oil addition) to 28.4 N cm$^{-2}$ (oregano oil addition). As regards eggshell thickness higher doses of thyme and oregano oils (1 ml kg$^{-1}$) in feed mixture had positive effect ($P$ >0.05). Similar findings to previous study found Botsoglou et al. (2005). Park et al. (2015) presented eggshell thickness of layers in the range from 0.37 to 0.40 mm. Tůmová et al. (2014) indicated average eggshell thickness of Lohmann Brown laying hens 0.31 mm.

The eggshell consists of 95% minerals, primarily calcium and others including phosphorus and magnesium (Sugino et al., 1997). Eggshells contain trace amounts

**Table 3** Minerals composition of eggshell

| Group | | Ca | P | Mg | Na | K | Cu | Zn |
|---|---|---|---|---|---|---|---|---|
| | | g kg$^{-1}$ | | | | | mg kg$^{-1}$ | |
| C | $\bar{x}$ | 373.17ab | 1.31 | 4.62a | 1.12a | 0.47a | 2.14a | 34.43 |
| | S.D. | 13.93 | 0.21 | 0.35 | 0.08 | 0.03 | 0.28 | 3.09 |
| | C.V. (%) | 3.73 | 16.38 | 7.48 | 7.18 | 7.47 | 13.18 | 8.93 |
| E1 | $\bar{x}$ | 343.83a | 1.35 | 3.31b | 0.81b | 0.34b | 1.67b | 34.88 |
| | S.D. | 30.35 | 0.076 | 0.27 | 0.04 | 0.03 | 0.26 | 9.47 |
| | C.V. (%) | 8.83 | 5.63 | 7.39 | 5.47 | 8.68 | 14.95 | 27.15 |
| E2 | $\bar{x}$ | 389.17b | 1.20 | 3.30b | 0.82b | 0.35b | 1.75ab | 41.10 |
| | S.D. | 37.23 | 0.10 | 0.14 | 0.03 | 0.02 | 0.17 | 16.60 |
| | C.V. (%) | 9.57 | 8.49 | 4.14 | 3.30 | 5.87 | 9.70 | 40.38 |

C – control group, E1 – pumpkin oil addition, E2 – flaxseed oil addition, $\bar{x}$ – mean, S.D. – standard deviation, C.V. – coefficient of variance. Values with different superscript in a column are significant at the 0.05 level

of micro elements, i.e. magnesium, boron, copper, iron, manganese, molybdenum, sulphur, silicon and zinc. It is probably the best natural source of calcium and it is about 90% absorbable (Nys et al., 1999; Li-Chan and Kim, 2008).

Mineral composition of eggshell is summarized in Table 3. Significantly ($P$ <0.05) lower contents of magnesium, sodium, potassium and copper in experimental groups compared to the control were found in eggshell. Significant ($P$ <0.05) differences in content of calcium were detected only between E1 (pumpkin addition) and E2 (flaxseed addition). Tendency of a highest ($P$ > 0.05) concentration of phosphorus after pumpkin oil supplementation and zinc after flaxseed oil supplementation were observed in eggshell.

The eggs produced by hens fed the control treatment (without experimental oil) were significantly different only from those derived from hens fed the diet supplemented with 2.5% canola oil + 2.5% soybean oil, which presented lower mineral content (Faitarone et al., 2013). The effect of dietary plant extracts added into feed mixture of layers on the content of phosphorus, calcium and magnesium in eggshell were not significant (Lokaewmanee et al., 2014). Herkeľ et al. (2016) also reported that phytogenic additive including essential oil may significantly affect mineral composition in poultry products. Feeding olive oil along with conjugated linoleic acid completely prevented changes in the mineral content of eggs (Aydin et al., 2001). Average contents of Ca 344 g $kg^{-1}$, P 1.17 g $kg^{-1}$, Mg 3.32 g $kg^{-1}$ and Zn 4.82 g $kg^{-1}$ were observed in egg shell of Lohmann Brown laying hens (Tůmová et al., 2014).

## 4 Conclusions

Dietary intake of flaxseed and pumpkin oils in feed mixture positively affected quality of eggshell. Strength of bursting of shell and thickness of eggshell were markedly ($P$ <0.05) higher in both experimental groups compared to the control. The effect of essential oil supplementation in diets of laying hens influenced also mineral composition in eggshell. Evidently ($P$ <0.05) lower contents of magnesium, sodium and potassium in E1 (pumpkin oil addition) and E2 (flaxseed oil addition) and copper in E1 in comparison with control group were found in eggshell. The amounts of other minerals were relatively balanced among groups and were not considerably affected of oil addition in diet.

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