Plant extract and probiotics change elemental status of muscle tissue of broilers

S G Rakhmatullin, G K Duskaev and B H Galiev

Federal Research Centre of Biological Systems and Agrotechnologies RAS, 9 Yanvarya St., 29, Orenburg 460000, Russia

1 E-mail: gduskaev@mail.ru

Abstract. This study was designed to investigate the effect of probiotic (B. adolescentis and L. acidophilus) and plant extract Quercus cortex (EQC) on the elemental status of muscle tissue of broiler chicken. For this, 120 7-day-old broilers were allocated to the following groups: control group – base diet (BD); I – BD + EQC; II – BD + probiotic; III – BD + probiotic + EQC. The following methods were used: atomic emission spectrometry, inductively coupled plasma mass spectrometry. The of pectoral muscle tissue from broilers in group II contained more calcium, iron, copper, manganese, cobalt, chromium, nickel and vanadium (P≤0.05). In the femoral muscles, there was an increased accumulation of magnesium, phosphorus, nickel and cobalt in group II (P≤0.05). The combined supplementation with QC and probiotic leads to an improvement of the changes the elemental status of muscle tissue, depending on its localisation, with minor changes to carcass traits.

1. Introduction
In terms of consumers, broiler meat should have good nutritional characteristics; the chemical composition of muscle tissue is an important element of broiler meat quality [1]. The parameters used to determine the quality of protein include biochemical and amino acid assessments, and biological value estimation [2]. It should be noted that the muscles from different parts of the carcass have different chemical compositions; that of breast (pectoralis major) muscle is different from that of thigh (biceps femoris) muscle [3]. The use of vegetable additives in poultry feed can affect meat quality in different ways. The essential oils of Origanum vulgare L. and Allium sativum are considered to be useful natural additives for improving meat quality (sensory attributes), without changing growth performance parameters [4]. Eucalyptus oil [5] and pomegranate products [6] improve meat composition and the fatty acids profile.

The use of probiotics to improve the quality of broiler meat has also been assessed, with contradictory results [7]. The scientific community has actively discussed the use of non-traditional ingredients for feeding poultry.

The purpose of this study was to determine the effect of EQC and a probiotic preparation, as feed additives, on broiler meat quality assessed as the concentration of the minerals, in the pectoral and femoral muscles.
2. Materials and methods

Poultry maintenance and procedures during the experiments met the requirements of the instructions and recommendations of "The Guide for Care and Use of Laboratory Animals (National Academy Press, Washington, D.C., 1996)".

The preparation of Q. cortex extract included the following steps: placing 50 g of shredded substances into heat-proof ware with 500 ml of hot (70 °C) distilled water, heating in a water bath (30 min), followed filtering. A probiotic preparation based on Bifidobacterium adolescentis B-1-DEP-VGNKI and Lactobacillus acidophilus LH1-DEP-VGNKI (80:1.0 million CFU).

This research was performed using Smena-8 broiler chickens. Acclimatisation lasted 6 days (from day 0 to day 6) and provision of the experimental diets began on day 7 and continued to day 42. For this, 120 broiler chickens were attributed to 4 groups (n = 30), each with 3 replicates of 10 birds. The groups were as follows: reference group – basic diet; experimental group I – BD + EQC at the level of 2.5 ml per kg of body; II – BD + probiotic preparation (1 g / kg feed); III – BD + probiotic + EQC (at the above levels).

The formulation of the experimental diets was based on the recommendations of All-Russian Research and Technological Institute of Poultry. Poultry were decapitated after nembutal and ether anesthesia on day 42.

During slaughter, the average muscle tissues were sampled from each bird, which were used to determine the chemical composition of the body tissues. Homogenised samples were dried at a temperature of 60–70 °C and stored in test tubes with a ground-in lid. The elemental composition of muscle tissue included the definition of 25 chemical elements (Ca, Cu, Fe, Li, Mg, Mn, Ni, As, Cr, K, Na, P, Zn, I, V, Co, Se, Ti, Al, Be, Cd, Pb, Hg, Sn, Sr) by atomic emission spectrometry and mass spectrometry with inductively coupled plasma (Optima 2000 V, Perkin Elmer, USA) and mass spectrometry (Elan 9000, Perkin Elmer, USA) in the Center for Biotic Medicine laboratory. The biosubstrates were ashed using a Multiwave-3000 decomposition microwave system (Anton Paar, Austria).

Numerical data were processed by the one-way analysis of variance (ANOVA) and the results were presented as mean values for groups and standard errors of the mean (StatSoft Inc. 2009). All results are presented as means and the variability in data was expressed as pooled SEM. The significance of difference among the groups was assessed using Tukey’s test. Statistical significance was considered when P<0.05

3. Results and discussion

In our experiment, birds fed with probiotics had a higher amount of muscle tissue and a higher content of calcium (P=0.024) and phosphorus in their pectoral and femoral muscles (Tables 1 and 2). Alfaig et al. [8] have shown that birds that receive probiotics better retain minerals, especially phosphorus, calcium and nitrogen, and the ratio of effective protein. A higher ratio of effective protein may subsequently contribute to an increase in meat yield. It has been previously shown [9] that the addition of probiotics increases the absolute and relative mass of the pectoral muscles.

| Element | Control | I | II | III | SEM | P-value |
|---------|---------|---|----|-----|-----|---------|
| Ca      | 0.163   | 0.208 | 0.492 | 0.194 | 0.10 | 0.024   |
| K       | 7.368   | 7.826 | 7.825 | 6.765 | 0.67 | 0.152   |
| Mg      | 0.979   | 1.045 | 1.091 | 0.960 | 0.10 | 0.101   |
| Na      | 1.578   | 1.553 | 1.252 | 1.550 | 0.15 | 0.103   |
| P       | 7.721   | 8.242 | 8.618 | 7.701 | 0.75 | 0.181   |

Table 1. Effect of EQC and probiotics on concentration of chemical elements in pectoral muscles, ‘µg g’.
In addition, the addition of probiotic preparations to the diet of poultry contributed to a higher accumulation of substances such as iron, copper, manganese, cobalt, chromium, nickel and vanadium in the pectoral muscles. At the same time, a significant accumulation of magnesium, nickel and cobalt was observed in the femoral muscles.

Polyphenols have different properties and are able to bind a number of metals [10]. This probably leads to their decrease in the more active femoral muscles of birds in the first group (EQC; calcium, iron, copper, cobalt, chromium, vanadium) with an increase in calcium (P=0.024) and cobalt (P=0.001) in the less active pectoral muscles. Other authors also identified an increase in calcium in pectoral muscles with *Boswellia serrata* resin [11]. The manifestation of the iron-binding ability of polyphenols was also traced, the iron content in the pectoral and femoral (P=0.094-0.098) muscles decreased. This is consistent with the results of multiple studies on monogastric animals [12-14].

Table 2. Effect of EQC and probiotics on concentration of chemical elements in femoral muscles, ‘µg g’.

| Element | Control | I | II | III | SEM | P-value |
|---------|---------|---|----|-----|-----|---------|
| **Macroelements** | | | | | | |
| Ca | 0.262 | 0.193 | 3.63 | 0.27 | 0.15 | 0.042 |
| K | 9.419 | 9.699 | 10.708 | 11.751 | 0.97 | 0.103 |
| Mg | 1.112 | 1.099 | 1.361 | 1.289 | 0.10 | 0.012 |
| Na | 4.042 | 3.558 | 3.922 | 4.79 | 0.35 | 0.124 |
| P | 6.483 | 6.561 | 8.888 | 7.593 | 0.67 | 0.051 |
| **Essential microelements** | | | | | | |
| Zn | 68.81 | 55.360 | 62.99 | 57.55 | 6.08 | 0.231 |
| Fe | 56.67 | 33.94 | 38.08 | 38.44 | 4.12 | 0.094 |
| Cu | 3.87 | 2.18 | 2.45 | 3.31 | 0.25 | 0.047 |
| Mn | 0.64 | 0.52 | 0.78 | 0.66 | 0.10 | 0.101 |
| I | 0.13 | 0.16 | 0.12 | 0.15 | 0.00 | 0.001 |
| Se | 0.51 | 0.66 | 0.52 | 0.60 | 0.10 | 0.102 |
| Cr | 0.16 | 0.12 | 0.12 | 0.11 | 0.00 | 0.001 |
| Co | 0.009 | 0.007 | 0.01 | 0.007 | 0.00 | 0.001 |
| **Conditionally essential microelements** | | | | | | |
| Si | 4.81 | 4.37 | 5.05 | 7.06 | 0.50 | 0.081 |
| B | 0.51 | 0.55 | 0.55 | 0.62 | 0.10 | 0.103 |
Group III in terms of the content of mineral elements in the pectoral muscle was close to that of the control group, whereas iodine decreased (P=0.033). Polyphenols probably leveled the holding capacity of probiotics, affecting the elemental composition of the muscle tissue. The element content in the femoral muscles of chickens in this group differed slightly; silicon and nickel increased, whereas a decrease in chromium, cobalt and vanadium was observed (P=0.001).

The results of a recent analysis of variance indicated the presence of associations between mineral elements and amino acids in the muscle tissue of chickens [15] and this fact could have a significant impact on the composition of these substances in the muscle tissue of the broilers in our experiment.

4. Conclusion
The combined supplementation with EQC and probiotic leads to an improvement of the changes the elemental status of muscle tissue (regarding calcium, iron, copper, manganese, cobalt, chromium, nickel and vanadium), depending on its localisation (pectoral or femoral muscles).

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