Effect of the Eucalyptus viminalis leaf extract compound on the amino acid and fatty acid composition of broiler chicken meat

Shamil G. Rakhmatullin, Marina Ya. Kurilkina* and Galimzhan K. Duskaev

Federal Research Center for Biological Systems and Agrotechnologies of the Russian Academy of Sciences, 29, 9 Yanvarya ave., Orenburg 460000, Russia, K_marina4@mail.ru

Abstract. The article presents the results on evaluating the effect of the gamma-octalactone compound isolated from Eucalyptus viminalis leaf extract on the amino acid and fatty acid composition of protein and lipids of broiler chickens' muscle tissue. The object of the study was 7-day-old broiler chickens of the Arbor Acres cross. During the experiment, the control group poultry consumed the main diet (MD); the poultry of the experimental groups received the main diet plus gamma-octalactone at the following dose: group I – 0.05 ml/kg l.w./ day; II – 0.1 ml/kg l.w./ day; III – 0.2 ml/ kg l.w./day. It was found that the experimental compound's introduction into the diets significantly increases protein and fat content, as well as improves the balance of experimental groups' chicken meat for essential amino acids, as evidenced by a significant increase in the content of leucine and isoleucine - 2.27% (P≤0.01), valine - 0.8% (P≤0.05), phenylalanine 0.44 (P≤0.05) % in chickens of the experimental group III in relation to the control. With a relatively stable amount of saturated fatty acids in the experimental groups, compensation for insufficient synthesis of polyunsaturated fatty acids was observed by increased accumulation of monounsaturated fatty acids. However, despite the existing differences in the concentration of unsaturated fatty acids, no significant intergroup changes were observed.

1 Introduction

Every year, there is a growing need to replace synthetic antibiotics in animal husbandry with safer natural compounds to avoid the increasing risk of resistance to antimicrobial preparations. For this reason, there is a consistent trend towards complete or partial rejection of such preparations almost everywhere. In recent years, many researchers working in the field of farm animals feeding have paid attention to biologically active substances formed in plants – phytobiotics. Phytobiotics have been proven to improve the functions of the immune system, have a significant impact on the animal health and increase productive qualities [1-6]. The factors determining the development relevance of highly effective phytobiotics in animal husbandry include that the global phytobiotics' use as an alternative to various antimicrobial drugs will not only protect the health and

* Corresponding author: k_marina4@mail.ru
maximize animal productivity, but will also provide a solution to public health problems including the problem of improving food safety by eliminating the cumulative effects of antibiotics in animal-derived products [7-10]. However, despite a large number of studies stating the growth-stimulating effects of phytobiotics, the exact mechanism of their effect on the qualitative characteristics of broiler chicken meat has yet to be clarified. At the same time, despite the obvious prospects of such studies, the amount of available information on the assessment of individual phytobiotic preparations’ effects on the amino acid and fatty acid compositions of broiler chicken meat is practically absent.

In this regard, the main goal of this study was to study the effect of different compounds' dosages from Eucalyptus viminalis leaf extract on the amino acid and fatty acid composition of broiler chicken meat.

2 Materials and methods

Object of research. Broiler chickens of the Arbor Acres cross.

During the course of research, poultry keeping was carried out in accordance with the instructions and recommendations of Russian Regulations, 1987 (Order No. 755 on 12.08.1977 the USSR Ministry of Health) and "The Guide for Care and Use of Laboratory Animals (National Academy Press Washington. D.C. 1996).

Experimental scheme. For the research, 120 heads of 7-day-old broiler chickens of the Arbor Acres cross were selected; they were divided by the method of analog groups into four groups (n=30): one control and three experimental. The broilers of the control group received the main diet (MD) formed in accordance with the VNITIP standards. In addition to the main diet, broilers of the experimental groups additionally received gamma-octalactone (97%, Sigma-Aldrich, USA) in the following doses: I experimental – 0.05 ml/kg l.w./day; II – 0.1 ml/kg l.w./day; III – 0.2 ml/kg l.w./day.

The experiment was held for 42 days: the preparatory period of 7 days and the accounting period of 35. Poultry had identical feeding and keeping conditions during the experiment. Broilers' feeding and watering was carried out by the group method in accordance with VNITIP recommendations.

The slaughter of the experimental poultry was carried out on the 42nd day of the experiment. Meat samples for laboratory studies were taken from the pectoral chicken muscle.

Equipment and technical means. The research was carried out on the basis of the Test Center using the material and technical means of the Center for Collective Use of Biological Systems and Agrotechnologies of the Russian Academy of Sciences (CCU FSC BSA RAS) (https://ckp-rf.ru/ckp/77384/). The amino acid composition of muscle tissue proteins of experimental broiler chickens was studied by capillary electrophoresis. The fatty acid composition of muscle tissue lipids was determined by gas chromatography.

Statistical processing. Data are expressed as mean values ± standard error of the mean. Statistical analysis was performed using Statistica 10.0 (StatSoft Inc., USA) and Microsoft Excel (Microsoft, USA). Significance of the group differences was estimated using Student’s t-test with p≤0.05 considered as significant.

3 Results

Data analysis on the chemical composition of broiler chickens' muscle tissue indicates that the maximum amount of nutrients was synthesized in the muscles of poultry in the experimental group III. In terms of protein content, they were 3.5% (P<0.05) higher than
the control group. Also, there was a tendency to increase the mass fraction of fat ($P \leq 0.001$) and dry matter in the body of this experimental group's broilers (Table 1).

Table 1. The chemical composition of muscles of experimental broiler chickens ($X \pm Sx$), % (age – 42 days, $n=30$).

| Index      | Group                  |
|------------|------------------------|
|            | control | I experimental | II experimental | III experimental |
|            | $M$     | $m$        | $M$     | $m$        | $M$     | $m$        | $M$     | $m$        |
| Dry matter | 22.5    | 0.70       | 23.2    | 0.59       | 23.50   | 0.66       | 23.70   | 0.55       |
| Protein    | 19.0    | 0.80       | 20.1    | 0.62       | 21.5    | 0.98       | 22.5    | 0.95*      |
| Fat        | 1.28    | 0.03       | 1.81    | 0.05***    | 1.38    | 0.06       | 1.82    | 0.04***    |
| Ash        | 0.99    | 0.04       | 0.98    | 0.06       | 0.99    | 0.05       | 0.98    | 0.04       |

Note: * – $P \leq 0.05$; *** – $P \leq 0.001$ compared with the control group

As a result of the conducted studies, certain differences in the concentration of amino acids in the muscle tissue of experimental poultry were revealed (Table 2).

Table 2. Amino acid composition of muscles of experimental broiler chickens ($X \pm Sx$), % (age – 42 days, $n=30$).

| Name of amino acid | Group                  |
|-------------------|------------------------|
|                   | control | I experimental | II experimental | III experimental |
|                   | $M$     | $m$        | $M$     | $m$        | $M$     | $m$        | $M$     | $m$        |
| Arginine          | 5.00    | 0.25       | 6.43    | 0.23***    | 5.78    | 0.24*      | 5.77    | 0.31*      |
| Lysine            | 6.03    | 0.27       | 6.68    | 0.20*      | 6.99    | 0.28**     | 7.08    | 0.31**     |
| Tyrosine          | 2.71    | 0.23       | 2.86    | 0.20       | 2.95    | 0.22       | 2.80    | 0.24       |
| Phenylalanine     | 2.30    | 0.21       | 2.77    | 0.19       | 2.71    | 0.24       | 2.74    | 0.14*      |
| Histidine         | 1.80    | 0.25       | 1.96    | 0.18       | 2.18    | 0.20       | 2.14    | 0.21       |
| Leucine+Isoleucine| 7.93    | 0.22       | 9.44    | 0.21***    | 9.31    | 0.23***    | 10.20   | 0.27***    |
| Methionine        | 1.65    | 0.16       | 1.81    | 0.21       | 1.81    | 0.24       | 1.87    | 0.17       |
| Valine            | 3.54    | 0.23       | 4.33    | 0.26*      | 4.23    | 0.19*      | 4.34    | 0.28*      |
| Proline           | 2.64    | 0.20       | 3.17    | 0.22*      | 3.05    | 0.24       | 3.18    | 0.28       |
| Threonine         | 3.05    | 0.24       | 3.69    | 0.27*      | 3.63    | 0.23*      | 3.76    | 0.25*      |
| Serine            | 2.35    | 0.25       | 2.74    | 0.22       | 2.72    | 0.20       | 3.08    | 0.24*      |
| Alanin            | 4.38    | 0.27       | 5.23    | 0.21**     | 5.45    | 0.25**     | 5.56    | 0.28**     |
| Glycine           | 3.18    | 0.23       | 3.74    | 0.20*      | 3.77    | 0.24*      | 4.42    | 0.22***     |

Note: * – $P \leq 0.05$; ** – $P \leq 0.01$; *** – $P \leq 0.001$ compared with the control group

Thus, a predominance over the control was observed in terms of essential amino acids content in the muscle tissue of experimental groups' chickens. In particular, it was found that the arginine content in poultry meat of the experimental groups increased in relation to
the control by 0.77 (P≤0.05) -1.43% (P≤0.001), lysine – by 0.65 (P≤0.05) -1.05% (P≤0.01), phenylalanine – by 0.44 (P≤0.05) %, leucine and isoleucine – by 1.38-2.27% (P≤0.01), valine – by 0.69–0.8% (P≤0.05), threonine – by 0.58-0.71% (P≤0.05).

In terms of the content of interchangeable amino acids, the advantage of the experimental groups over their control peers was also revealed in terms of: serine – by 0.73% (P≤0.05), alanine – by 0.85-1.8% (P≤0.01), glycine – by 0.56 (P≤0.05)-1.24% (P≤0.001), respectively. The content of the remaining amino acids in the experimental groups tended to increase, namely, tyrosine, proline, histidine, methionine. At the same time, the chickens of the experimental group III had an advantage in the content of almost all the amino acids studied.

The study results of the fatty acid composition of experimental broiler chickens' muscle tissue showed that the highest content of saturated fatty acids was found in the experimental groups - 33.8–35.8%. At the same time, there was a significant superiority of the III experimental group, which amounted to 2.2% relative to the control (Table 3).

Table 3. Fatty acid composition of muscles of experimental broiler chickens (X±Sx), % (age – 42 days, n=30).

| Name of fatty acid       | control | I experimental | II experimental | III experimental |
|-------------------------|---------|----------------|----------------|-----------------|
|                         | M       | m              | M              | m               | M               | m               |
| Saturated               |         |                |                |                 |                 |
| Palmitic (C16:0)        | 22.5    | 0.60           | 22.8           | 0.74            | 23.8            | 0.68            | 23.6            | 0.27*           |
| Stearin(C18:0)          | 10.9    | 0.25           | 10.9           | 0.30            | 11.8            | 0.33*           | 12.1            | 0.30**          |
| Arachinoic (C20:0)      | 0.20    | 0.004          | 0.10           | 0.005***        | 0.20            | 0.02            | 0.10            | 0.008***        |
| Monounsaturated         |         |                |                |                 |                 |
| Palmitoleic (C16:1)     | 1.90    | 0.04           | 2.20           | 0.05***         | 2.20            | 0.09**          | 2.80            | 0.05***         |
| Oleic (C18:1)           | 36.8    | 0.98           | 39.3           | 1.20            | 35.3            | 1.10            | 37.9            | 1.02            |
| Polyunsaturated         |         |                |                |                 |                 |
| Linoleic (C18:2)        | 24.8    | 0.86           | 22.7           | 0.97            | 23.9            | 0.60            | 21.4            | 1.86            |
| Linolenic (C18:3)       | 2.90    | 0.07           | 2.00           | 0.55            | 2.80            | 0.06            | 2.10            | 0.48            |

Note: * – P≤0.05; ** – P≤0.01; *** – P≤0.001 compared with the control group

When analyzing the lipid composition for individual fatty acids, some intergroup features were also recorded. Thus, the concentration of palmitic acid in the experimental groups varied in the range of 22.8-23.8%. The concentration of stearic acid in the context of the studied groups fluctuated in the range of 10.9-12.1%. At the same time, chickens of the experimental group III differed in their maximum content, surpassing control peers by 1.1 (P <0.05) and 1.2% (P<0.01). However, at the maximum level of palmitic and stearic fatty acids in the experimental groups, the content of arachinic acid was minimal in relation to the control (P≤0.001).

As a result of the analysis, it was found that the content of monounsaturated fatty acids in the experimental groups was in the range of 37.5-41.5% exceeding the control by 2-2.8%. This advantage was initially due to the high concentration of palmitoleic acid 2.20-2.80% (P≤0.001). For example, chickens of the experimental group III prevailed in its
concentration over the control by 0.9% (P≤0.001). There were no significant intergroup differences in the content of oleic acid.

Polyunsaturated fatty acids accounted for about 23.5-26.7%, with minimum values in the meat of the experimental group III and maximum values in the control group. When analyzing the concentration of linoleic and linolenic acids, no significant intergroup differences were observed.

### 4 Discussion

The data obtained in this experiment show that the inclusion of gamma-octalactone isolated from Eucalyptus viminalis leaf extract in the main diet of experimental groups' chickens contributed to the improvement of the poultry meat qualitative indicators, as evidenced by the study results on the amino acid composition, as well as the ratio of saturated, polyunsaturated, and monounsaturated fatty acids. The explanation of this fact is possible considering previously conducted studies, in which it was found that substances isolated from Eucalyptus viminalis plants exhibit broad antimicrobial and anti-inflammatory activity due to pharmacologically active compounds (macrocarpal A and B, lactones and its derivatives) [11].

The ability of lactones to suppress oxidative stress is well-known [12]. In this regard, the probable action mechanism that affected the biochemical composition of broiler muscles in the experiment may be the anti-inflammatory and antioxidant properties of the substance. This fact can be confirmed by the results of previous studies. It is known that Forsythia suspensa extract weakens oxidative damage to the pectoral muscles of broilers [13]. Adding 80 mg/kg of natural capsaicin extract to the broiler diet can improve the digestibility of nutrients and the quality of meat [14]. Improving the meat quality is possible by reducing the metabolism of muscle glycolysis and improving the oxidative status of muscles due to dietary supplements with chitosan oligosaccharides [15]. Feeding 2% blueberry extract increased the concentration of various fatty acids in breasts and thighs of broiler chickens [16].

### 5 Conclusion

Thus, the research results indicate that the muscle tissue obtained from broiler chickens of all experimental groups was characterized by high biological value. At the same time, the most optimal ratio of amino acids in the muscles was observed in the group of chickens receiving gamma-octalactone in the diet at a dosage of 0.2 ml/kg l.w./day.

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