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RELATION BETWEEN SELECTED NUTRIENTS IN THE CHICKEN MEAT DEPENDING ON PHYTOGENIC FEED ADDITIVES

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ABSTRACT

The aim of study was to evaluate the relation between selected nutrients in the breast and thigh muscles after the application of different phytogenic additives in the diet of broiler chickens and between same indicators of meat disregarding additive and parts of carcass, from which muscles originate. We realized an *in vivo* experiment on the Zámostie Company poultry test station with deep litter breeding system. The experiment included 100 pcs of one-day-old hybrid chickens Cobb 500 divided into 2 groups (n = 50): the 1st experimental group with an application of feed additive from chestnut tree and lemon fruit extracts and the 2nd experimental group with an application of feed additive from citrus fruits extract. We used a cereal and soybean basal diet and we divided the fattening period into four phases: starter (1 – 10 days), grower I (11 – 20 days), grower II (21 – 28 days) and finisher (29 – 42 days). We applied a powder form feed mixtures. Nutritive value of feed mixtures was the same in each experimental group during the whole experiment and in accordance with the physiological needs of broiler chickens. We fed the 1st experimental group with a basal diet enriched by feed additive from chestnut tree and lemon fruit extracts (50 g/100 kg). As for the 2nd experimental group, we applied feed additive from citrus fruits extracts through the drinking water (100 mL/100 L). In the 2nd part of our experiment, we compared results obtained from two experimental groups with other four groups of diet. We applied other phytogenic additives to these four groups and we did not take into account the origin of the meat sample. We measured indicators of the chemical composition of protein, fat, water and cholesterol on a sample (50 g) of breast and thigh muscle without skin by the method of FT IR by use of the apparatus Nicolet 6700. Detected relations between nutrients of breast and thigh muscles were defined by correlation coefficient of $-0.6 \leq r \geq +0.6$. When additive with chestnut tree and lemon fruit extracts was used, we detected a negative correlation ($p \leq 0.01$) between protein and cholesterol of breast muscle. In thigh muscle, the negative correlation was observed between protein and energy ($p \leq 0.05$), protein and fat ($p \leq 0.01$) as well as fat and water. The only positive correlation was detected between protein and cholesterol of breast muscle ($p \leq 0.01$), with additive citrus fruits extract. When nutrition and parts of carcass, from which muscles originate, were disregarded, protein of meat increased, energy and fat decreased ($p \leq 0.001$). When fat of meat increased, energy increased ($p > 0.05$) as well, but water decreased ($p \leq 0.05$; $p \leq 0.001$).

Keywords: phytogenic feed additive; breast and thigh muscle; chicken meat; nutrient

INTRODUCTION

Poultry meat is an important source of proteins, but other constituents as fats have an important role in its composition, too. Manipulation in animal feeding **(Kennedy et al., 2005)** or *post mortem* manipulation of carcass body may affect meat quality. In recent years, products containing essential oils derived from several spices and herbs could be used in animal nutrition as feed additives to promote the growth. These phytogenic additives may have more than one mode of action, including improving feed intake and flavour, stimulating the secretion of digestive enzymes, increasing gastric and intestinal motility, endocrine stimulation, antimicrobial, anti-viral, anthelminthic and coccidiostat activities, immune stimulation, and anti-inflammatory and anti-oxidative activity and pigments **(Kirkpinar et al., 2011)**. Earlier published papers **(Smid and Gorris, 1999)** present that essential oils achieved positive performance in antibacterial *in vitro* studies. We need greater concentration of essential oil to achieve the same effect in foods. High levels of fat and/or protein in foodstuffs protect the bacteria from the action of the essential oil in some way **(Tassou, 1995)**.

This short literature review suggests that an application of phytogenic substances based on essential oils have some unanswered questions in food research.

The aim of our study was to evaluate the relation: a) between selected nutrients, energy and water of breast and thigh muscles regarding to application of two different phytogenic additives in the diet of broiler chickens; b) between the same nutrients, energy and water of chicken meat disregarding parts of carcass, from which muscles originate (breast and thigh muscles together).

MATERIAL AND METHODOLOGY

Experiment, broiler chickens, nutrition

We realized an *in vivo* experiment on the Zámostie Company poultry test station with deep litter breeding system. The experiment included 100 pcs of one-day-old hybrid chickens Cobb 500 divided into 2 groups (n = 50): the 1st experimental group with an application of feed additive from chestnut tree and lemon fruit extracts and the 2nd experimental group with an application of feed additive from citrus fruits extract. Phytogenic substances obtained from chestnut tree and lemon fruit extracts created a base of applied feed additive. The feed additive represents a mixture of taste compounds with supporting antioxidative, antimicrobial and cleansing effects in the digestive tract. Citrus fruits extract included extracts obtained from four species of citrus fruits: grapefruit (*Citrus paradisi*), tangerine (*Citrus reticulata blanco*), bergamot (*Citrus aurantium* ss. Bergamia) and sweet orange (*Citrus sinensis*). We used a cereal and soybean basal diet and we divided the fattening period into four phases: starter (1 – 10 days), grower I (11 – 20 days), grower II (21 – 28 days) and finisher (29 – 42 days). We applied a powder form of feed mixtures. Nutritive value of feed mixtures was the same in each experimental group during the whole experiment and in accordance with the physiological needs of broiler chickens. We fed the 1st experimental group with a basal diet enriched by feed additive from chestnut tree and lemon fruit extracts (50 g/100 kg). As for the 2nd experimental group, we applied feed additive from citrus fruits extracts through the drinking water (100 mL/100 L). In the 2nd part of our experiment, we compared results obtained from two experimental groups with other four groups of diet. We applied other phytogenic additives to these four groups and we did not take into account the origin of the meat sample.

Sample analyses

At the end of the experiment (day 42), we randomly selected 6 pcs from each group with an average live body weight of about 1800 g. We performed a slaughtering of chickens by human rapid cut of the carotid artery (*Ateria carotis communis*). We separated breast and thigh muscle from the carcass and we used them for chemical analysis. We measured indicators of the chemical composition of protein, fat, water and cholesterol on a sample (50.0 g) of breast and thigh muscle without skin by the method of FT IR by use of the apparatus Nicolet 6700. We performed a molecular spectroscopy for infrared spectrum of muscle homogenates analyses. The principle of this method is infrared absorption spectrum of the sample passes and there is a change from the rotary vibrating energy conditions of the molecule depending on the changes of the dipole moment of the molecule. Analytical output is the infrared spectrum, which is a graphical representation of the function of the energy dependence, mostly mentioned as a percentage of transmittance (T) or in units of absorbance (A) at a wavelength of the incident radiation. Permeability is defined as ratio of the radiation intensity which has passed through the sample (I) and of the emission intensity of emitted source (Io). Absorbance is defined as a decimal logarithm of 1/T.

Calculation of the energy value of meat according to the measured values of protein and fat, and the corresponding coefficients: 16.75 ×protein +37.68 ×fat (kJ/100 g).

Statistical analyses

We present our results in the form of mean, standard deviation, minimum and maximum values We used Scheffe´s test at the significance level of $\alpha = 0.05$ to compare a difference between groups. We used a Pearson´s correlation coefficient to reflect a degree of relation between two variables of selected chemical indicators of chicken breast muscle, thigh muscle and meat. Pearson´s r reflects the degree of linear relation between the two data sets. Its value is between -1 and +1. A value of +1 means, that there is a perfect positive linear relation between two data sets. A value of -1 means that there is a perfect negative linear relation and a value of 0 means, that there is no linear relation at all between data sets. We mainly focused our attention on the assessment of relation between two variables defined by correlation coefficient $-0.6 \leq r \geq +0.6$. We supplemented our results of correlation coefficient statistical significance at the significance level of $\alpha = 0.05$, 0.01 and 0.001. We used SAS statistical package (**SAS Institute, 1998**) to perform statistical analyses.

RESULTS AND DISCUSSION

Chemical composition of breast and thigh muscles in relation to feed additive and disregarding parts of carcass from which muscles originate (breast and thigh muscles together)

Chemical composition of breast and thigh muscles in relation to feed additive are shown in Table 1 and disregarding parts of carcass from which muscles originate (breast and thigh muscles together) in Table 2.

In two experimental groups, values of protein content of breast muscles are relatively similar 23.83 and 24.09 g/100 g, respectively. We observed slightly larger difference between the protein content of breast muscle compared to protein content of thigh muscle depending on the type of phytogenic substances in feed mixtures. We did not observe a statistically significant difference ($p > 0.05$) between groups. Similar values of protein content in breast muscles found **Haščík et al., (2012),** but in the hybrid combination of the chickens Ross 308.

We recorded the protein content of 22.73 g/100 g in the group with a feed additive from chestnut tree, and lemon fruit extracts compared with 22.15 g/100 g for the group with the feed additive from citrus fruits extract in the thigh muscles. We observed statistically significant difference ($p \leq 0.05$) between protein content of thigh muscle of the 1st group and the breast muscle of the 2nd group (Table 1). Table 2 presents that, the average protein content of chicken meat was 23.21 g/100 g disregarding parts of carcass, from which muscles originate. The type of phytogenic supplements did not affect the energy value of breast muscle and thigh muscle.

The energy values of breast samples were 435.69 kJ/100 g and 437.26 kJ/100 g in the 1st experimental group and the 2nd experimental group, respectively. Slightly lower energy value of broiler chicken breast muscles Cobb 500 found **Angelovičová and Semivanova (2013)**. We

Table 1 Nutrients, energy and water in the chicken breast and thigh muscles in relation to feed additive.

Variable	n	Breast muscle Mean	*SD*	Min.	Max.	Thigh muscle Mean	*SD*	Min.	Max.
Feed additive from chestnut tree and lemon fruit extracts									
Protein (g/100 g)	6	23.83	0.49	22.90	24.21	22.73	0.92	21.69	24.40
Energy (kJ/100 g)	6	435.69	6.03	428.72	444.50	458.13	15.82	438.17	474.04
Fat (g/100 g)	6	0.69	0.24	0.46	1.02	1.39	0.60	0.35	2.00
Water (g/100 g)	6	74.85	0.35	74.29	75.32	74.33	0.44	73.77	74.83
Cholesterol (g/100 g)	6	0.23	0.07	0.17	0.34	0.32	0.09	0.22	0.45
Feed additive from citrus fruits extract									
Protein (g/100 g)	6	24.09	0.42	23.57	24.63	22.15	0.49	21.58	22.92
Energy (kJ/100 g)	6	437.26	11.21	424.86	453.78	459.26	12.55	440.66	472.53
Fat (g/100 g)	6	0.69	0.42	0.39	1.50	1.21	0.07	1.09	1.29
Water (g/100 g)	6	74.73	0.24	74.39	75.07	74.09	0.56	73.62	74.92
Cholesterol (g/100 g)	6	0.25	0.11	0.12	0.36	0.36	0.06	0.31	0.44

Legend: n – the number of samples, *SD* – standard deviation, Min. – the minimum value, Max. – the maximum value.

Table 2 Nutrients, energy and water in the chicken meat disregarding parts of carcass from which muscles originate (breast and thigh muscles together).

Variable	n	Mean	*SD*	Min.	Max.
Protein (g/100 g)	72	23.21	0.91	21.58	24.71
Energy (kJ/100 g)	72	448.35	18.42	415.73	492.78
Fat (g/100 g)	72	1.05	0.49	0.21	2.10
Water (g/100 g)	72	74.46	0.59	72.52	75.32
Cholesterol (g/100 g)	72	0.29	0.08	0.10	0.45

Legend: n – the number of samples, *SD* – standard deviation, Min. – the minimum value, Max. – the maximum value.

detected almost the same energy value of thigh muscle in two experimental groups. The energy value of breast muscle and thigh muscle of two experimental groups was not statistically significant (p >0.05), while calculated energy value disregarding parts of carcass, from which muscle originates (Table 2) was 448.35 kJ/100 g.

We obtained interesting results of the fat content of breast muscles in our experiment. We detected the same value of 0.69 g/100 g in the group with feed additive from chestnut tree and lemon fruit extracts, and in the group with feed additive from citrus fruits extract. Fat content values of thigh muscle samples were higher than values of breast muscle samples. In the group with feed additive from chestnut tree and lemon fruit extracts, we detected an average value 1.39 g/100 g of fat in thigh muscle, and 1.21 g/100 g of fat in the group with feed additive from citrus fruits extracts. A difference of fat content was not statistically significant (p >0.05) between groups. The value of fat content in chicken meat was 1.5 g/100 g, when we disregarded the part of carcass, from which the muscle originates and feed additive. Moisture content results in breast samples were almost the same, i.e. 74.85 g/100 g and 74.73 g. /100 g in the 1st experimental group and the 2nd experimental group, respectively.

We did not find statistically significant difference (p >0.05) between experimental groups. Similar values of dry matter of chicken breast muscle of Cobb 500 found **Medveď and Angelovičová (2010).**

Other study with the same hybrid of chickens – Cobb 500 presents lower water content values of 73.73 and 73.29 g/100 g. Authors used commercial feed and they slaughtered chickens of experimental groups at the age of 42 days **(Suchý et al., 2002)**.

Our results of detected water content in the thigh samples were 74.33 g/100 g and 74.09 g/100 g in the 1st experimental group and 2nd experimental group, respectively. The conclusions of other studies **(Al-Sultan, 2003; Latshaw and Moritz, 2009)** confirmed decreased water content of the thigh muscles compared with breast muscle. We obtained same results. Water content of chicken meat was 74.46 g/100 g disregarding parts of carcass, from which the muscles originates and feed additive.

The image of meat and meat products is relatively negative due to their content of fat and saturated fatty acids, cholesterol, sodium and any other substances (e.g. nitrosamines) that somehow can be involved in most prevalent diseases of Western societies like cardiovascular diseases and *diabetes mellitus* **Micha et al., 2010)** and cancer **(Ferguson, 2010)**. Our results of cholesterol content were almost the same in relation to the type of used phytogenic substances. The cholesterol content was slightly increased in the group with feed additive from chestnut tree and lemon fruit extracts compared to group with feed additive from citrus fruits extract. We did not observe statistically significant difference between two experimental groups (p >0.05). The cholesterol content of breast muscles was 0.23 g/100 g in the 1st experimental group and 0.25 g/100 g in the 2nd experimental group. The cholesterol content of thigh muscles was 0.32 g/100 g (the 1st experimental group) and 0.36 g/100 g (the 2nd experimental group). The cholesterol content of chicken meat was 0.29 g/100 g when we disregarded parts of carcass from which the muscles originate.

Within this context, the poultry meat has maintained its identity and a higher value compared to other species for several reasons. Indeed, worldwide poultry meat production and consumption have increased rapidly and, in many parts of the world, it is assumed, per capita consumption of poultry meat will continue to grow

(Cavani, 2009). Relatively low and competitive prices compared to other meats, the absence of cultural or religious obstacles, and dietary and nutritional properties are the main factors explaining poultry meat's attractiveness **(Valceschini, 2006)**.

Relation between selected nutrients, energy and water of chicken breast and thigh muscles depending on feed additive and disregarding parts of carcass from which the muscles originate (breast and thigh muscles together)

A positive correlation coefficient indicates that an increase in the first variable would correspond to an increase in the second variable, thus implying a direct relation between the variables. A negative correlation indicates an inverse relation where as one variable increases, the second variable decreases.

Relation between selected nutrients, energy and water of chicken breast and thigh muscles in the 1st experimental group with feed additive from chestnut tree and lemon fruit extracts

Relation between selected nutrients, energy and water of chicken breast muscle in the 1st experimental group

Table 3 presents a correlation coefficient (r) between nutrients, energy and water of breast muscles in the 1st experimental group.

Combined essential oils have additive, synergistic, and antagonistic effects **(Burt, 2004)**. Many commercial products on the market have one or more combined essential oils. Utilization of any feed additive is justified due to the larger beneficial effect compared to the cost of the product. In our experiment, feed additive from chestnut tree and lemon fruit extracts influenced the correlative relation between certain nutrients, energy and water of breast muscle. We detected the relation with correlation coefficient of -0.6 ≤ r ≥ +0.6 between protein and water, between protein and cholesterol and between energy and water. All relations with correlation coefficient value of -0.6 ≤ r ≥ +0.6 were not statistically significant. A positive correlation without statistically significant difference (*p >0.05*) was between protein and water. We observe statistically significant difference (*p* ≤0.01) between protein and cholesterol. When protein content of breast muscle increased, cholesterol content decreased. When energy value of breast muscle increased, water content decreased, without statistically significant differences (*p* >0.05).

Table 3 Correlation coefficient (r) between nutrients, energy and water of breast muscles in the 1st experimental group.

Variable	Energy	Fat	Water	Cholesterol
Protein	-0.27	-0.42	0.60	-0.95++
Energy		-0.12	-0.63	0.01
Fat			-0.47	0.43
Water				-0.36

Legend: Numerical data – the correlation coefficient (r) between two variables.
++: value with superscript mark is significantly different (*p* ≤0.01).

We did not find any literature information about effects of essential oils on relations between nutrients of chicken meat, parts of carcass, respectively. Essential oils have antimicrobial, antifungal and antioxidant effects. The effects of several type of essential oils, their combinations, or a combination with other substances might be related to the relations between nutrients of chicken meat, breast and thigh muscles. Citrus species of various origins have been assessed for their phenolic constituents and antioxidant activities **(Guimarães et al., 2009)**. Citrus fruits, citrus fruit extracts and citrus flavonoids exhibit a wide range of promising biological properties including anti-atherogenic, anti-inflammatory and antitumor activity, inhibition of blood clots and strong antioxidant activity **(Middleton and Kandaswami, 1994)**.

Extracts of citrus fruit (e.g. lemon, orange, and grape fruit) are among the most studied natural antimicrobials for food applications. Extracts of citrus fruit effectively decrease the growth of bacteria. Limonoids obtained from *Citrus limon* showed good antibacterial and antifungal activity **(Corbo et al., 2008)**. There are several citrus species, i.e. *Citrus limon* (lemon), *Citrus aurantium* (bitter orange), *Citrus limetta* (sweet lemon), *Citrus jambhiri* (rough lemon) and *Citrus paradise* (grapefruit) **(Al-Ani et al., 2009)**.

Three types of flavonoids occur in citrus fruit, i.e. flavanones (including 3-hydroxyflavanones), flavones (including 3-hydroxyflavones) and anthocyanins **(Horowitz and Gentili, 1977)**.

Eight tested limonoids, i.e. nomilin, limonin, deacetylnomilin, limonol, obacunone, deoxylimonin, isoobacunoic acid and ichangin stimulated the detoxifying enzyme, glutathione S-transferase **(Lam and Hasegawa, 1989)**. Glutathione S-transferase enzymes are one of the major enzyme systems responsible for the detoxification of xenobiotics **(Chasseaud, 1979)**.

Vitamin C and bioflavonoids. Bioflavonoids are a class of water-soluble plant pigments. Vitamin C-rich fruits and vegetables, especially citrus fruits, are often rich sources of bioflavonoids as well. Two small published studies examined the effect of bioflavonoids on the bioavailability of ascorbic acid. In one study, synthetic ascorbic acid given in a natural citrus extract containing bioflavonoids (in the ratio of bioflavonoids to ascorbic acid of 4:1), proteins, and carbohydrates, was more slowly absorbed and 35% more bioavailable than synthetic ascorbic acid alone, based on plasma levels of ascorbate over time and 24-hour urinary excretion of ascorbate. In the other study, there was no difference in the bio-availability of 500 mg of synthetic ascorbic acid and that of a commercially available vitamin C preparation with added bioflavonoids, where the ratio of bioflavonoids to ascorbic acid was 0.05:1 **(Higdon, 2001)**.

Relation between selected nutrients, energy and water of chicken thigh muscles in the 1st experimental group

Table 4 presents a correlation coefficient (r) between nutrients, energy and water of thigh muscles in the 1st experimental group.

Table 4 Correlation coefficient (r) between nutrients, energy and water of thigh muscles in the 1st experimental group.

Variable	Energy	Fat	Water	Cholesterol
Protein	-0.83+	-0.95++	0.70	-0.25
Energy		0.75	-0.64	0.35
Fat			-0.82+	0.10
Water				-0.15

Legend: Numerical data – the correlation coefficient (r) between two variables,
+, ++: value with superscript mark is significantly different ($p \leq 0.05$, $p \leq 0.01$).

Extracts of chestnut tissue display a strong antimicrobial activity against many plant pathogens, which is probably associated with antimicrobial compounds such as flavonol glycoside and several terpenoid substances **(Hao et al., 2012)**. The results presented in study **(Blaiotta et al., 2013)** indicate that chestnut extracts can greatly improve the tolerance of lactobacilli to simulated gastric and bile juice. Chestnut extracts exhibited a surprising effect in improving the tolerance to gastric transit of lactobacilli. The study confirmed that scoparone and scopoletin isolated from chestnut inner shell extract have antioxidant effects, and scopoletin has relatively higher antioxidant capacity than scoparone in an oxidative stress-induced in vitro system.

Chestnut inner shell extract including scoparone and scopoletin as main compounds has the ability to protect against damage due to oxidative stressors including tert-butyl hydroperoxide, carbon tetrachloride (CCl_4), and high-fat diet, by preventing reactive oxygen species generation, decrease of antioxidant enzyme activity, and inhibiting malondialdehydu production. The chestnut inner shell extract might be useful as a natural ingredient for the prevention of oxidative damage in liver cells and tissues **(Noh et al., 2010)**.

In our experiment, the influence of feed additive from chestnut tree and lemon fruit extracts on relation between certain nutrients, energy and water of the thigh muscles was higher compared to breast muscle. We detected a relation with correlation coefficient of $-0.6 \leq r \geq +0.6$ between protein and energy, protein and fat, protein and water, energy and fat, energy and water as well as fat and water. All these correlation relations were not statistically significant. When energy and fat of thigh muscles increased, protein content decreased. These relations were statistically significant ($p \leq 0.05$, $p \leq 0.01$, respectively). When fat content of the thigh muscles increased, water content decreased, with statistically significant difference ($p \leq 0.05$).

Many authors have concluded that essential oils exhibit greater antimicrobial activity than other major components taken together. This could means that either the minor components are critical to the antimicrobial activity or that synergistic effects may occur **(Burt, 2004)**. The major components reflect the biological properties of essential oils, but minor components can modulate their activity, for example the cell penetration, hydrophobicity and fixation on membranes **(Bakkali et al., 2008)**. The composition of essential oil results in interactions between the components that both qualitatively and quantitatively change their evaporation rates **(Saiyasombati and Kasting, 2003)**.

Relation between selected nutrients, energy and water of chicken breast and thigh muscles in the 2nd experimental group with feed additive from citrus fruits extract

Relation between selected nutrients, energy and water of chicken breast muscle in the 2nd experimental group

Table 5 presents a correlation coefficient (r) between nutrients, energy and water of breast muscles in the 2nd experimental group.

We could consider citrus essential oils as suitable alternatives to chemical additives for use in the food industry, attending to the needs for safety and satisfying the demand of consumers for natural components **(Viuda-Martos et al., 2008)**. Since the major component of citrus essential oils is limonene, the chemical, physical and biological properties of this compound greatly affect the properties of the citrus essential oils **(Bakkali et al., 2008)**. For this reason, we can find documented antimicrobial effect of citrus essential oils attributed to the essential oil or to limonene as well, as its main component. Biodegradation essential oils, and in particular limonene; mechanism by which essential oils inhibit anaerobic digestion is not yet understood **(Ruiz and Flotats, 2014)**.

Table 5 Correlation coefficient (r) between nutrients, energy and water of breast muscles in the 2nd experimental group.

Variable	Energy	Fat	Water	Cholesterol
Protein	-0.08	-0.51	-0.40	-0.50
Energy		-0.62	0.08	0.86+
Fat			0.37	-0.40
Water				0.32

Legend: Numerical data – the correlation coefficient (r) between two variables,
+: Value with superscript mark is significantly different ($p \leq 0.05$).

The antimicrobial activity of terpenes and terpenoids (cyclic hydrocarbons) is due mainly to their interaction with the cell membrane **(Bakkali et al., 2008)**. We detected an influence of feed additive from citrus fruits extract only on relations between energy and fat, as well as energy and cholesterol. A correlation coefficient of relation between energy and fat, as well as energy and cholesterol was $-0.6 \leq r \geq +0.6$. An interesting result is the relation between energy and fat. When fat content of breast muscles increased, energy content decreased. The difference was not statistically significant ($p > 0.05$). We detected statistically significant difference ($p \leq 0.01$) of positive correlation between energy and cholesterol.

Relation between selected nutrients, energy and water of chicken thigh muscles in the 2nd experimental group

Table 6 presents a correlation coefficient (r) between nutrients, energy and water of thigh muscles in the 2nd experimental group.

Table 6 Correlation coefficient (r) between nutrients, energy and water of thigh muscles in the 2nd experimental group.

Variable	Energy	Fat	Water	Cholesterol
Protein	-0.41	0.04	-0.27	-0.30
Energy		0.66	-0.25	0.42
Fat			0.05	-0.14
Water				-0.73

Legend: Numerical data – the correlation coefficient (r) between two variables.

The influence of feed additive from citrus fruits extract on relation between some nutrients, energy and water of thigh muscles was lower compared to the effects of feed additive from chestnut tree and lemon fruit extracts. We found a positive correlation with correlation coefficient of -0.6 ≤ r ≥ +0.6 between energy and fat of thigh muscles. The difference was not statistically significant (p >0.05). Citrus essential oils are a complex mixture of volatile compounds that show, among other properties, antifungal activity by reducing or totally inhibiting fungal growth in a dose-response manner **(Sharma and Tripathi, 2006)**. This activity is a resalt of a single major compound or synergistic or antagonistic effect of various compounds **(Deba et al., 2007)**.

Relation between selected nutrients, energy and water of chicken meat disregarding parts of carcass, from which muscles originate

Table 7 presents a correlation coefficient (r) between nutrients, energy and water of chicken meat disregarding parts of carcass, from which muscles originate (breast and thigh muscles together).

Table 7 Correlation coefficient (r) between nutrients, energy and water of chicken meat disregarding parts of carcass from which the muscles originate (breast and thigh muscles together).

Variable	Energy	Fat	Water	Cholesterol
Protein	-0.64^{+++}	-0.75^{+++}	0.42^{+++}	-0.37^{+++}
Energy		0.70^{+++}	-0.67^{+++}	0.39^{+++}
Fat			-0.59^{+++}	0.26^{+}
Water				-0.30^{++}

Legend: Numerical data – the correlation coefficient (r) between two variables,
+, ++, +++: Value with superscript mark is significantly different (p ≤0.05, p ≤0.01, p ≤0.001).

In our experiment, disregarding nutrition and parts of carcass, from which muscles originate, we found a statistically significant relation with correlation coefficient of -0.6 ≤ r ≥ +0.6 between protein and energy, protein and fat, as well as energy and fat, energy and water, fat and water of chicken meat. When protein content of chicken meat increased, energy content and fat content decreased with statistically significant differences (p ≤0.001). These relations are relatively comparable to the relations between protein, energy and water of thigh muscles in the 1st experimental group. When fat content of chicken meat increased, energy content increased as well without statistically significant difference (p >0.05), but water content decreased with statistically significant difference (p ≤0.001). When energy content of chicken meat increased, water content decreased with statistically significant difference (p ≤0.001). We observed a relation with correlation coefficient of -0.6 ≤ r ≥ +0.6 between cholesterol and protein, cholesterol and fat, cholesterol and energy, cholesterol and water, as well as protein and water. The differences were statistically significant (p ≤0.001, p ≤0.01 and p ≤0.05, respectively). These relations are relatively comparable to relations between cholesterol, energy and water of thigh muscles in the 1st experimental group.

CONCLUSION

We can confirm based on a statistical evaluation of the results of the experiment that:

a) broiler chicken nutrition is one of the major factors which must be taken into account in the production of chicken meat;

b) various additives of phytogenic substances in the feed mixtures for broiler chicken differently affected relations between protein and cholesterol in breast muscle, and between protein and energy, between protein and fat, and the fat and water in thigh muscle.

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