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The effect of feed supplementation with *Salvia officinalis*, *Thymus vulgaris*, and *Rosmarinus officinalis* on the quality of quail meat

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The aim of this study was to determine the effect of a feed mixture containing 0.5% of sage (*Salvia officinalis*), thyme (*Thymus vulgaris*), and rosemary (*Rosmarinus officinalis*) on the quality of quail carcasses and physicochemical properties of their breast muscles. The study was conducted on 80 Japanese quail (40 females and 40 males) divided randomly into four groups of 20 animals each. The control group (I) was given a complete feed mixture without the addition of herbs, while the experimental groups (II, III, and IV) were given feed with 0.5% of powdered sage, thyme, and rosemary, respectively. Quail were weighed before and after slaughter which took place at six weeks of life. Breast and leg muscles were excised from the carcasses in order to determine pH_{24}, free water, cooking loss, meat colour, basic chemical composition, and the levels of selected elements (Na, K, Ca, Mg, P, Na, Se, Cu, Fe). The feed containing rosemary herbs (at 0.5%) had a positive effect on quail leg muscle mass and a tendency towards higher body and breast muscle mass. Feeding herb supplementation resulted in a tendency towards higher free water leakage.

Thyme and sage supplementation resulted in a significantly higher cooking loss ($p \leq 0.05$). All experimental groups showed a decrease in muscle acidity ($p \leq 0.05$) and brighter breast muscle colour compared to the control. Although the addition of herbs to the feed had a positive effect on the chemical composition of quail meat and the levels of macro and microelements, the differences were not statistically significant.

KEY WORDS: quail / medicinal herbs / carcass characteristics / meat quality

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The increasingly popular use of herbs in animal nutrition is associated with their positive effect on the taste and smell of feed, and their ability to act as biocatalysts regulating and accelerating metabolism in the body. Herbs positively influence the secretion of digestive juices, increase appetite and intestinal peristalsis, and improve the absorption of nutrients [Kwiecień *et al*. 2006, Narimani-Rad *et al*. 2011]. The use of herbs also improves the quality of animal products [Cross *et al.* 2007, Genedy and Zeweil 2003, Khaksar *et al*. 2012, Al-Kassie 2009], such as meat, milk, and eggs.

The effectiveness of herbal medicinal raw materials depends on the quality and amounts of biologically active substances contained in them, most notably selenium and antioxidants [Sotek *et al*. 2018].

Taking advantage of antioxidant properties of herbs is important for animal health and the antioxidant stability of animal products (e.g. meat, eggs). For example, the antioxidants contained in rosemary, thyme and sage have been shown to stabilize meat quality [Cuppett and Hall 1998, Craig 1999, Nakatani 2000, Wei and Shibamoto 2007, Gumus *et al*. 2017]. Aksu *et al.* [2014] showed that the use of thyme oil in quail feed positively affected breast muscle pH and improved its colour. Yesilbag *et al*. [2011] showed that the feed supplementation with dried rosemary or rosemary oil decreased breast and leg muscle pH and improved the taste of cooked breast muscle of Ross 308 chickens. In addition, feed containing herbs such as thyme, oregano, mint or rosemary increased their body weight and carcass quality [Yesilbag *et al*. 2011, Narimani-Rad *et al*. 2011].

As reported by Botsoglou *et al*. [2002] and Aksu *et al*. [2014], antioxidants in herbs – including those found in rosemary, sage and thyme – prevent the oxidation of fatty compounds (unsaturated fatty acids, cholesterol) in animal products and increase the antioxidant potential of the consumer's body. The antioxidant properties of rosemary are based on chelating metals and preventing the formation of free radicals [Dorman *et al.* 2003]. Mariutti *et al*. [2011] demonstrated the inhibitory effect of sage on the oxidation of fat and cholesterol in chicken meat.

The aim of the study was to determine the effect of feed mixtures containing 0.5% of *Salvia officinalis*, or 0.5% of *Thymus vulgaris*, or 0.5% of *Rosmarinus officinalis* on the carcass quality and physicochemical properties of breast muscles of quail.

Material and methods

Birds, environmental condition, feeding

The study was conducted on 120 Japanese (Pharaoh) quail. All the birds were kept in standardised environmental conditions (temperature, air humidity, light programme and feeding conditions). Raising the birds was performed according to the standard technology recommended for this species [Rutkowski 2000]. The birds were fed without any restrictions: in the 1st period with complete starter feed and in the 2nd period with complete finisher feed (Tab. 1 and 2). Throughout the whole experimental period the birds had free access to water. In the second period, 80 quail (40 males and

Table 1. Composition of starter feed in the first rearing period

Category	Ingredient	Amount (%)	Met.En. (MJ)	B.O	Fiber (g)	Calcium (g)	Total Phosforus (g)
Middlings	Corn	28.00	3.85	26.32	8.12	0.11	0.95
Middlings	Wheat	31.29	4.02	37.24	9.07	0.22	1.16
Feed	Soya middlings 5-7% water loss in dry mass, 460 G Total Protein	36.50	3.34	167.90	22.63	1.28	1.97
Mineral feed	NaCl	0.24	0.00	0.00	0.00	0.01	0.00
Mineral feed	Phosphate 1-CA	1.15	0.00	0.00	0.00	2.53	2.65
Mineral feed	Fodder chalk	1.00	0.00	0.00	0.00	3.61	0.00
Feed additive	DL-methionine (technically pure)	0.36	0.00	3.53	0.00	0.00	0.00
Feed additive	L-lysine hydrochloride	0.38	0.80	2.96	0.00	0.00	0.00
Kemin	Kemzyme DRY X	0.08	0.00	0.00	0.00	0.00	0.00
Kemin	Toxfin	0.10	0.00	0.00	0.00	0.00	0.00
Basf	Lutamix DJR	0.50	0.00	0.00	0.00	1.43	0.00
Mineral feed	Sodium bicarbonate	0.20	0.00	0.00	0.00	0.00	0.00
Basf	Natuphos 5% for broiler chickens	0.20	0.05	3.06	0.00	0.90	0.00

Metabolic Energy – 12 MJ, B.O 24%.

Table 2. Composition of finisher feed in the second rearing period

Category	Ingredient	Amount (%)	Met. En.	B.O	Fiber (g)	Calcium (g)	Total Phosforus (g)
Middlings	Barley	15.00	1.78	16.50	7.20	0.06	0.56
Middlings	Corn	25.00	3.44	23.50	7.25	0.10	0.85
Middlings	Wheat	31.76	4.08	37.79	9.21	0.22	1.18
Feed	Soya middlings 3.5-7% water loss in dry mass, 460 G Total Protein	25.00	2.29	115.00	15.50	0.88	1.35
Mineral feed	NaCl	0.22	0.00	0.00	0.00	0.01	0.00
Mineral feed	Phosphate 1-CA	0.70	0.00	0.00	0.00	1.54	1.61
Mineral feed	Fodder chalk	0.80	0.00	0.00	0.00	2.88	1.43
Feed additive	DL-methionine (technically pure)	0.23	0.00	2.25	0.00	0.00	1.43
Feed additive	L-lysine hydrochloride	0.29	0.00	2.26	0.00	0.00	1.43
Kemin	Toxfin	0.10	0.00	0.00	0.00	0.00	1.43
Basf	Lutamix DJR	0.50	0.00	0.00	0.00	1.43	1.43
Mineral feed	Sodium bicarbonate	0.20	0.00	0.00	0.00	0.00	1.43
Basf	Natuphos 5% for broiler chickens	0.20	0.00	3.06	0.00	0.90	0.90

Metabolic Energy – 11, 6 MJ, 20% B.O.

40 females) at the age of 3 weeks (when the sex of birds could be clearly distinguished) were selected and allocated to 4 groups: 3 experimental and control. The birds were kept in separate labelled cages. Each group was randomly divided into 2 subgroups (10 birds each) that constituted the experimental replicates. In the 3rd week of life,

an additional 0.5% of dried powdered rosemary, sage or thyme from Herbapol (5 g/kg) was introduced into the feed mixtures of each respective experimental group. The experiment was conducted up to 6 weeks of age. No deaths were reported during the entire period of rearing.

Slaughter parameters

Eighty six-week-old Japanese quail were slaughtered (after 12 h of ante-mortem fasting) by decapitation of the stunned animals with a sharp knife. After bleeding, plucking, and evisceration, the carcasses were kept in a refrigerator at a temperature of about 4°C for 24 hours. The cooled carcasses were weighed and dissected. From each carcass, the separated breast and leg muscles were weighed on the laboratory scales to the nearest 0.01 g. Body weight and carcass weight were used to determine the carcass yield (quotient of carcass weight and body weight). Eighty quail breast muscles were used for physicochemical analyses.

Chemical analysis

The assessment of the chemical composition was based on the determination of the percentage content of basic chemical components: total water, total protein, fat, and ash:

– water content was calculated on the basis of determination of dry matter content by drying the sample at 105°C, after protein denaturation with 96% ethyl alcohol [AOAC, 2003];
– total protein content was determined by the Kjeldahl digestion [AOAC, 2003];
– intramuscular fat content was determined by the Soxhlet extraction [AOAC, 2003];
– ash content was determined by combustion analysis [AOAC, 2003].

The elements in the studied material were determined by emission spectrometry with excitation in inductively coupled argon plasma (ICP OES) using the "Optima 2000 DV" apparatus by Perkin Elmer, after earlier digestion mineralization in the microwave oven by Anton Paar.

Cooking loss

The excised breast muscles were put into 200 ml glass vessels and flooded with 100 ml of water. The sample vessels were placed in a water bath and boiled to 85°C in accordance with the methodology given by Baryłko-Pikielna i Matuszewska [2014]. Then the cooking losses were determined based on the sample weight difference before and after cooking.

Meat colour

The colour was measured with the Mini Scan XE Plus 45/0 with a 31.8 mm measurement area. The camera was standardized according to black and white standards. The applied white standard had the following coordinates: X=78.5, Y=83.3,

and Z=87.8 (for light standard D65 and standard 10º observer). Colour parameters were determined for each sample using the CIE L*a*b* scale and the D65/10° illuminator/observer system [CIE, 1976]. Colour measurements were performed on the internal surface of the raw muscle after refrigeration at 4°C to oxidize myoglobin in the surface muscle layer.

Muscle pH

Meat pH was measured 24 hours after slaughter using a glass electrode (ESAgP-302W type) and the CyberScan 10 pH meter (EUTECH CYBERNETICS PTE LTD) in water extract (distilled water) after 1 hour of extraction in 1:1 meat-to-water proportion.

Free water

Free water was determined by Grau and Hamm method [1953] modified by Pohja and Niinivaara [1957]. It was determined by weighing two 300 mg meat samples on Whatmann 1 tissue paper (to the nearest 0.001 g) and placed between glass plates loaded with a 2 kg weight for 5 minutes. The contours of the pressed meat and drip on the blotting paper were outlined with a chemical pencil. After the blotting paper was dried, the stains were planimetered and then the surface area of the infiltration was calculated from the difference between these surface areas. Then, free water content was calculated by dividing the area of the infiltration in cm^2 by the weight of samples (g).

Statistical analysis

One-way analysis of variance (ANOVA) and the Tukey's test were used to evaluate the influence of herbs on the parameters of carcass (body weight, carcass weight, breast and leg muscle weight, carcass dressing yield) and meat quality (chemical composition, levels of selected pH microelements, colour parameters (L*, a*, b*), free water, cooking loss). Prior to the analysis normality of empirical distribution of these traits was checked using the Shapiro-Wilk test.

Results and discussion

Carcass quality

Table 3 presents the parameters of quail carcass quality. Although there were no statistically significant differences, there was a tendency towards a higher body weight in the quail from experimental groups 3 and 2, where rosemary and sage were added to the feed.

Denli *et al.* [2004] recorded a positive and statistically significant effect of thyme and sage oil on body weight gain in quail. Also Franciosini *et al.* [2016] reported improved body weight in broiler chickens up to 36 days of age by using oregano and oregano combined with rosemary in feed. However, there are also reports such as of Bülbül *et al.* [2015] where the supplementation with sage and laurel oil did not

Table 3. Basic characteristics of quail carcass quality depending on the herb species in diet

Item	Group							
	control		*Salvia* 0.5%		*Thymus* 0.5%		*Rosmarinus* 0.5%	
	mean	SD	mean	SD	mean	SD	mean	SD
Body weight (g)	189.80	20.18	195.88	16.49	189.93	22.34	206.35	23.43
Carcass weight (g)	123.30	11.17	122.64	12.49	123.15	20.09	132.51	14.38
Carcass dressing yield percentage (%)	65.13	3.17	62.64	4.42	65.03	6.65	64.32	2.91
Breast muscle weight (g)	34.49	5.01	34.05	3.97	33.83	5.38	37.48	6.86
Leg muscle weight (g)	26.15[a]	3.13	25.98[a]	2.27	25.36[a]	2.36	27.74[b]	9.65

[ab]Within a row means bearing different superscript differ significantly at $p \leq 0.05$.

affect the body weight (as well as the parameters of quail carcasses). The reported increases in body weight in poultry may result from an improved taste of feed which then contributes to higher feed consumption by the animals [Windisch *et al*. 2008].

There was no effect of herbs used on quail carcass weight, carcass yield, and breast muscle weight (Tab. 3). Only in group 3 did the use of rosemary in the feed have a significant effect ($p<0.05$) on the weight of quail leg muscles. This confirms the results of studies such as Narimani-Rad *et al*. [2011] where mixtures of medicinal plants (thyme – *Thymyus vulgaris*, oregano – *Oreganum vulgare*, and peppermint – *Mentha x piperita)* used in nutrition had a statistically significantly positive effect ($p \leq 0.05$) on carcass yield and quality. On the other hand, Norouzi *et al*. [2015] found no effect of rosemary and yarrow supplementation on body weight and carcass quality in broiler chickens.

Of the three supplemented herbs, rosemary had the greatest effect on carcass yield and quality; quail from this group were significantly characterized by the highest femoral muscle weight, as well the highest body weight and breast muscle weight.

These results are consistent with the positive effect of herbal preparations in poultry diets and their positive influence on body weight and carcass quality [Bampidis *et al*. 2005, Kwiecień *et al*. 2006, Mohammed and Abbas 2009, Toghyani *et al*. 2010, Gumus *et al*. 2017].

According to Hippenstiel *et al*. [2011], this is related to the antibacterial effect of essential oils, beneficial for the bacterial flora in the digestive track, which is associated with the increased production of digestive enzymes and improved digestion. In addition, Mathlouthi *et al*. [2012] argue that the beneficial effect of active herbal substances on the carcass dressing yield of broiler chickens results from their positive effect on the processes of digestion, nutrient absorption and the immune system.

pH

The characteristics of quail breast muscles are presented in Table 4. The control group had a statistically significantly less acidity of breast muscles than the

Table 4. Characteristics of quail breast muscle quality depending on the herb species in diet

Item	Group							
	control		*Salvia* 0.5%		*Thymus* 0.5%		*Rosmarinus* 0.5%	
	mean	SD	mean	SD	mean	SD	mean	SD
pH_{24}	6.09[a]	0.06	5.90[b]	0.15	5.84[b]	0.20	5.90[b]	0.19
L*	36.84	1.38	39.87	2.71	37.63	3.54	38.04	3.32
a*	10.87[ab]	0.76	10.66[a]	1.05	11.65[b]	1.78	10.85[ab]	1.47
b*	12.55	1.46	13.15	1.97	11.54	2.56	11.70	2.21
Cooking loss (%)	38.06[ac]	1.68	39.51[c]	1.79	40.20[b]	1.16	38.98[abc]	1.79
Free water (%)	2.68	2.29	4.74	3.91	4.26	2.13	3.74	0.91

[abc]Within a row means bearing different superscript differ significantly at $p \leq 0.05$.

experimental ones. A decrease in muscle pH resulting from rosemary supplementation was observed in broiler chickens by Yesilbag *et al*. [2011], which was attributed by those authors to a decreased level of microbiological contamination and delayed meat spoilage. However, some studies provide opposite results. Aksu *et al*. [2014] recorded a significant increase in the pH of breast muscles of quail regardless of the size of the supplementation with thyme feed oil. Similarly, an increase in the pH of broiler chicken breast muscles was observed following supplementation with various herbs [Lipiński *et al*. 2019]. Other researchers, such as Chang-Song *et al*. [2017] and Kirkpinar *et al.* [2014], recorded no influence of herbs on muscle pH in broiler chickens.

The pH range recorded in the experimental groups in our study (5.84-5.90) is typical for breast muscles of quail [Jakubowska *et al*. 2013, Karamucki *et al*. 2013a].

Colour

In the breast muscle of quail that received sage, thyme and rosemary in the feed, a tendency towards a lighter meat color (L*) was shown compared to those from the control group (Tab. 4).

Herbal supplementation resulted in the increased brightness of breast muscles than control, although not statistically significantly (Tab. 4), which is consistent with the reports of other authors [Aksu *et al*. 2014, Yesilbag *et al*. 2011, Chang-Song *et al*. 2017]. Quail fed with thyme were characterized by statistically significantly higher redness of breast muscle compared to the group supplemented with sage. An increased proportion in redness was also observed by Kirkpinar *et al*. [2014] following feed supplementation with oregano combined with garlic, as well as garlic alone. In addition to redness, the proportion of yellowness ranged from 11.54 to 13.15 (Tab. 4).

According to Simitzis *et al*. [2008] the addition of oregano essential oils in sheep's diet may have indirectly resulted in the colour change of meat, probably by reducing the oxidation of haemoglobin and modifying the mechanisms that activate the distribution of pigment in animal tissues. Aksu *et al*. [2014], showed a dose-dependent and positive effect of thyme essential oil on Japanese quail meat; at a dose of 400 mg/kg it improved oxidative stability and colour parameters.

Finally, the experimental groups also showed tendency to a brighter meat colour. According to literature data [Qiao *et al*. 2000, Warner *et al*. 1997], higher muscle brightness is often accompanied by decreased protein solubility and an increased degree of protein denaturation, especially sarcoplasmic proteins.

The tendency to a lighter colour of breast muscles of quail receiving herbal supplementation may have been related to changes in meat structure caused by a pH decrease, evidenced by higher cooking losses and a tendency to higher free water (Tab. 4).

Changes in the meat structure affect the degree of light absorption by meat [Karamucki *et al*. 2013b]. Our results are somewhat in agreement with the findings of Kirkpinar *et al*. [2014] who noted a significant ($p \leq 0.05$) effect of diet with oregano and garlic supplementation on the brightness of breast muscles of broiler chickens. However, Nieto *et al*. [2010] found no effect of rosemary leaf distillate in pregnant sheep's diet on the colour of lamb meat after slaughter.

A significant increase in meat brightness was observed only after 14 and 21 days of storage in the lamb group where the dose of rosemary in mothers' feed was 20%.

Free water

Free water in the breast muscles of quail ranged, and herbal supplementation of feed resulted in higher free water, although the differences were statistically insignificant. The highest cooking losses were recorded following sage and thyme supplementation in comparison to the control group (Tab. 4), which is consistent with the findings of Nasir and Grashorn [2010], who found higher cooking losses in groups receiving supplementation with *Echinacea purpurea* and *Nigella sativa* as feed additives in broiler chickens, compared to the control. WHC is a very important feature of quail meat as it is intended for direct culinary processing. Meat with high WHC retains better juiciness and tenderness after heat treatment. However, in our experiment, herbal supplementation did not reduce the cooking loss and instead, it resulted in significantly higher cooking losses. This is consistent with the results of Gardzielewska *et al*. [2003], where the addition of Digestarom herbal preparation deteriorated WHC in chicken breast muscles, and resulted in a high free water leakage and a very high thaw loss. On the other hand, Park *et al*. [2015] showed that dried oregano powder reduced cooking loss in the breast muscles of ducks. Similarly, Kołodziej-Skalska *et al*. [2011] concluded that a mixture of extracts from *Origanum* spp., *Cinnamomum* spp., and *Capsicum* in pig feed reduced cooking loss and increased the ability to hold water in pork.

Chemical composition of meat

Our results (Tab. 5), which show no influence of herbal supplementation on the basic chemical composition of meat, are consistent with other reports, which showed no effect of herbs in feed on dry matter, total protein and fat levels in poultry muscles [Kirkpinar *et al*.2014, Gardzielewska *et al*. 2003, Jakubowska *et al.* 2013, Rossi *et al*. 2013].

Table 5. Correlation coefficients between natural logarithm from the actual number of 1 mL somatic cells for milk samples obtained during different periods

Milk sampling time period	Day 1 to 7	Day 8 to 14	Day 15 to 21
Day 8 to 14	0.72		-

Mineral content

Although the experimental groups of quail had an increased proportion of K, P, Ca, Cu, and Se in breast muscles compared to the control, the differences were not statistically significant (Tab. 6). Some reports do show a significant influence of herbs on the content of mineral components in poultry blood or meat. In a study where broiler chickens were given 0.25 or 0.5% of rosemary in their diet, a statistically significant increase ($p \leq 0.05$) was observed in meat levels of Mg, Ca, and Na [Jameel 2019]. Also in the research on turkeys, where a 1% herbal mixture was introduced (yarrow, marigold, ribbon, hawthorn fruit, horsetail, horsetail, nettle, and chokeberry pomace), an increase in Cu retention and a decrease in K retention in the breast and thigh muscles of turkeys were observed. The addition of those herbs also increased the content of Cu, Zn, C, and Mg and decreased Fe in the whole blood of turkeys [Makarski and Polonis 2001]. Finally, Gumus *et al*. [2017] showed that thyme oil supplementation (*Thymus vulgaris*) in Japanese quail significantly increased Mg concentration in blood serum, whereas Ca, Fe, and P levels did not change.

Table 6. Levels of selected macro and microelements in quail meat depending on the herb species in diet

Item	Group							
	control		*Salvia* 0.5%		*Thymus* 0.5%		*Rosmarinus* 0.5%	
	mean	SD	mean	SD	mean	SD	mean	SD
K (g/kg)	2.79	0.15	2.87	0.14	2.82	0.08	2.83	0.09
Mg (g/kg)	0.334	0.016	0.335	0.013	0.341	0.008	0.343	0.015
P (g/kg)	2.71	0.14	2.76	0.10	2.80	0.09	2.78	0.13
Na (g/kg)	0.672	0.059	0.668	0.042	0.680	0.036	0.687	0.049
Ca (g/kg)	0.124	0.023	0.136	0.022	0.132	0.042	0.131	0.029
Cu (mg/kg)	2.19	0.43	2.32	0.39	2.33	0.30	2.21	0.28
Fe (mg/kg)	26.7	4.0	27.3	3.8	27.4	4.3	24.6	4.2
Se (mg/kg)	0.285	0.105	0.280	0.152	0.315	0.111	0.357	0.138

Conclusions

The present study shows that the addition of powdered herbs to quail feed had a positive effects on some of the carcass and meat quality parameters. Supplementation with rosemary statistically significantly increased leg muscle weight, as well as

resulted in a tendency to a higher body weight and breast muscle weight. The addition of 0.5% of rosemary, thyme and sage to quail feed statistically significantly decreased the pH of pectoral muscles. At the same time, a tendency for higher free water was observed in all experimental groups, and statistically significantly higher cooking losses in the groups supplemented with thyme and sage. In addition, statistically significantly higher redness (a*) of breast muscle was observed in the group of quail receiving feed supplemented with thyme. However, none of the supplements had any statistically significant effect on muscle lightness (L*), yellowness (b*), basic chemical composition, and the content of selected macro- and microelements in the meat, although in the experimental groups there was a tendency to elevated levels of these elements compared to control.

The obtained results suggest that further studies are needed to determine the optimal level of supplementation of quail feed with rosemary, sage and thyme, as well as a mixture of these three herbs.

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