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Effect of dietary administration of *Mentha piperita* alone and in combination with *Artemisia abrotanum* on growth performance, body composition, physicochemical properties and amino acid composition of meat in *Eimeria*-challenged broilers

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Abstract: This study aimed to evaluate the effects of dietary administration of peppermint (*Mentha piperita*) alone and in combination with southernwood (*Artemisia abrotanum*) on performance, body and carcass composition, physicochemical characteristics, and amino acid composition of breast meat in broilers under *Eimeria* challenge. A total of 160 14-day-old male broilers (Ross 308) were randomly assigned to four dietary groups: the control (C) group was fed a basal diet; the CC group received a grower diet containing the coccidiostat robenidine; the M group was fed the diets with the addition of 20 g peppermint/kg of diet; the MS group was fed the diets with the addition of 10 g peppermint + 10 g of southernwood/kg of diet. The addition of peppermint to the diet increased final live weight compared to the CC group ($P < 0.05$). The highest value of dressing percentage was found in the M group as compared to the CC group ($P < 0.01$) and C group ($P < 0.05$). Compared with the C group, a higher intramuscular fat content was found in the breast meat of broilers in both the M and MS groups ($P < 0.01$; $P < 0.05$, resp.). Concerning the colour of meat, lower values for yellowness b^* and chroma C^* characteristics were observed in the C group compared to the MS group ($P < 0.05$) and mainly to the M group ($P < 0.01$). In spite of the lower crude protein content ($P < 0.05$) in the meat of the MS group, the highest content of all essential amino acids ($P < 0.05$) as well as the total content of all assessed amino acids ($P < 0.01$) were found in the meat of the MS group, resulting in a considerable increase in the nutritional value of the breast meat proteins of these broilers.

Keywords: amino acid; breast muscle; carcass trait; coccidiosis; meat nutrition; phyto-genic additive

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Recently, the targeted use of phytogetic additives in poultry nutrition has been carried out mainly due to their beneficial effects on health, growth performance and carcass yield, or their potential to improve the quality of produced meat and eggs (He et al. 2023). Selected plant products often exhibit anti-inflammatory, antibacterial and anti-protozoal activity, including coccidiostats, and are considered natural alternatives for controlling various poultry diseases, with the advantage that there is usually no risk of resistance (Muthamilselvan et al. 2016; Hussein et al. 2021). Different combinations of several herb species are often used to enhance the effect of administered herbs on the poultry organism (Hussein et al. 2021). Of the present essential oils when using both whole herbs and their parts, the total content of supplemented herbs in the diets is usually up to 2%.

Southernwood (*Artemisia abrotanum*; S herb), together with *Artemisia absinthium*, *Artemisia vulgaris* and *Artemisia cina*, is used as a medicinal plant in natural medicine. Although these plants belong to the same genus, they are different species that often possess specific concentrations of different biologically active substances. In traditional European medicine, S herb is mainly used for liver and biliary tract diseases, parasitic diseases in children and as an antipyretic medication. The main substances contained in S herb that exhibit antibacterial, antifungal, antiparasitic, antioxidant, anticancer, antiallergic and repellent properties include mainly essential oils, coumarins, phenolic acids and flavonoids (Ekiert et al. 2021). In the case of essential oils found in S herb, compounds with monoterpene structure are the dominant fraction. Other fractions include compounds with sesquiterpenoid, diterpenoid, triterpenoid or spiroterpenoid structure and phenylpropanoid derivatives. Of the present essential oils, borneol, camphor, *trans*-sabinyl acetate, 1,8-cineole, piperitone and davanone are generally the most abundant (Ekiert et al. 2021).

Peppermint (*Mentha piperita* L.; M), one of the oldest medicinal herbs, belongs to the family *Lamiaceae* and is characterised by its strongly aromatic leaves (Petricevic et al. 2021). It is beneficial for building the immune system and for its antimicrobial and substantial antioxidant properties (Abdel-Wareth et al. 2019). The chemical compounds of M leaves are mainly menthol, menthone, isomenthone, menthyl acetate, cineole and other active substances such as menthofuran,

isomenthone, limonene, β -pinene, α -pinene, germacrene D, *trans*-sabinene hydrate and pulegone (Beigi et al. 2018). Menthol, the main phenolic constituent in M oil, has significant antibacterial effects. In addition to essential oils, M leaves also contain flavonoids, tannins, triterpenes, bitter substances and other active compounds (Petricevic et al. 2021). Concerning the poultry, M leaves promote growth in the early stage of chicken age, increase their production performance and carcass traits, and improve egg quality (Khempaka et al. 2013; Abdel-Wareth and Lohakare 2014). Moreover, Khempaka et al. (2013) reported that M leaves have beneficial effects on antioxidant activity, abdominal fat deposition and ammonia production in broilers. The active compounds contained in M may play an important role in the inhibition of *Escherichia coli*, *Staphylococcus aureus*, *Salmonella enteritidis* and *Candida albicans* growth, and they can act as a coccidiostat during various stages of the parasitic *Eimeria* life cycle (Muthamilselvan et al. 2016).

Based on the information above, the administration of M herb to poultry showed a wide range of positive impacts. However, there is still a certain limitation in the knowledge of its effect on some meat quality indicators. In the case of S herb, its effects on poultry production have not been studied at all in the available literature. So, the aim of the present study was to evaluate the effect of dietary administration of peppermint alone and in combination with southernwood on growth performance, body and carcass composition, physicochemical characteristics, and amino acid profile of breast meat in *Eimeria*-challenged broilers.

MATERIAL AND METHODS

Chicken husbandry and management. The experiment was approved by the Animal Welfare Committee of the University of Veterinary Sciences Brno (Project No. PP10-2023).

A total of 160 14-day-old male broilers (Ross 308) were used in the experiment. The chickens were randomly allocated to four dietary groups (40 birds per group). The chickens were weighed and birds from each group were housed in four floor pens (4 replicates; 10 birds/replicate) bedded with wood shavings. All chickens were housed in the accredited experimental stable of the Department of Animal

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Breeding, Animal Nutrition and Biochemistry, University of Veterinary Sciences Brno, under the same controlled housing conditions that were adjusted according to the Ross 308 guidelines. Throughout the experiment, broilers were exposed to a constant lighting regime of 18 h light and 6 h darkness. The initial ambient housing temperature was 25 °C and it was progressively reduced by 2–3 °C per week until the target housing temperature of 20 °C was reached at 27 days of age. Broilers were stocked at an overall density of 3.14 birds/m². Feed and water were supplied *ad libitum* to chickens. Chickens were observed for any signs of illness and behavioural changes twice a day. Mortality was recorded daily. A two-phase feeding program was used, in which the grower and finisher diets were offered from day 14 to 27 and 28 to 42, respectively. The grower and finisher diets were fed in the form of pellets.

The feeding trial lasted 28 days. On days 21, 28, 35, and 42, each broiler was weighed and the live weight (LW) was recorded, while leftover feed was measured on a pen basis. Thereafter, daily feed intake (DFI) and feed conversion ratio (FCR) were assessed as the growth performance traits. The FCR values were determined based on gain and feed intake adjusted for mortality (feed/gain).

Dietary treatments. The experimental M group was fed the diets with the addition of 20 g M herb per kilogram of basal diet and without the inclusion of a coccidiostat. The experimental MS group was fed the diets with the addition of 10 g M herb + 10 g S herb per kilogram of basal diet and without the coccidiostat. The CC group was fed a grower diet containing the coccidiostat robenidine (33 mg/kg of feed) and without the addition of M and S herbs; the finisher diet did not contain any coccidiostat or any addition of M and S herbs. Chickens in the control (C) group were fed basal grower and finisher diets (Table 1), without any coccidiostat or any addition of M and S herbs. For the experimental M and MS groups, powder obtained from the entire aerial parts of the M and S herbs was incorporated into the diet prior to pellet manufacturing. The chemical composition of diets was determined according to the procedures described by Zapletal et al. (2020). The analysed nutrient compositions of the grower and finisher diets are outlined in Table 2. Until 14 days of age, chickens were fed a commercial starter containing 20.7% of crude protein and 13 MJ of ME/kg and without a coccidiostat.

Eimeria infection. At 14 days of age, control samples of chicken faeces were taken and no *Eimeria* oocysts were detected. Subsequently, at 17 days of age, the chickens in all dietary groups were challenged by a tenfold overdosing of the live vaccine LIVACOX T (Batch No. B108T 26823; Biopharm Co., Prague, Czech Republic) to induce a mild coccidiosis infection as described earlier by Yu et al. (2021) and Zapletal et al. (2025b). Each dose of the inoculum (1 ml) was orally gavaged directly into the crop of each bird. Each dose contained 15 000 live oocysts of *Eimeria acervulina*, *Eimeria tenella*, and *Eimeria maxima*.

Slaughter, body dissection, and meat analysis. At the end of the experiment (day 42), 15 chickens from each dietary group were randomly selected from all replicates for body composition and carcass evaluation. Chickens were weighed before slaughter and then stunned and bled by cutting the jugular vein. Dissection of the chicken body and carcass processing were carried out according

Table 1. Ingredient compositions (%) of basal diets fed broilers

Item	Diet	
	grower	finisher
Wheat	9.38	9.68
Maize	50.0	50.0
Soybean meal	30.5	30.5
Canola oil	4.80	3.50
Monocalcium phosphate	0.70	0.70
Limestone	1.25	1.25
NaCl	0.24	0.24
NaHCO ₃	0.20	0.20
Lysine	0.14	0.14
Methionine	0.23	0.23
Threonine	0.06	0.06
Maize sprouts	2.00	3.00
Vitamin and mineral premix ^a	0.50	0.50
Mastercube [®]	0.50	0.50

^aVitamin and mineral premix provided per kilogram of diet: retinol, 13 500 IU; cholecalciferol, 2 300 IU; alpha-tocopherol, 56.0 mg; menadione, 4.50 mg; thiamine, 4.50 mg; riboflavin, 9.00 mg; pyridoxine, 5.50 mg; cobalamin, 0.02 mg; biotin, 0.22 mg; niacinamid, 70.0 mg; folic acid, 2.20 mg; iron, 68.0 mg; manganese, 110 mg; zinc, 93.0 mg; copper, 12.0 mg; iodine, 1.60 mg; selenium, 0.28 mg; calcium pantothenate, 18.0 mg; butylated hydroxytoluene, 2.80 mg and butylated hydroxyanisole, 0.63 mg

Table 2. Analysed nutrient composition (g/kg) in diets fed broilers as fed basis

Item	Diet			
	C	CC	M	MS
Grower (day 15 to 28)				
Dry matter	905	905	902	903
Crude protein	187	187	189	188
Crude fibre	27.2	27.2	27.0	28.2
Crude fat	70.0	70.0	65.6	66.0
Crude starch	392	392	376	378
Ash	63.2	63.2	63.0	62.7
Calcium	13.2	13.2	12.5	13.0
Inorganic phosphorus	4.90	4.90	4.70	4.70
Metabolisable energy (MJ/kg)	14.0	14.0	13.9	13.8
Finisher (day 29 to 42)				
Dry matter	905	905	901	903
Crude protein	196	196	196	196
Crude fibre	23.4	23.4	26.7	27.9
Crude fat	65.1	65.1	63.7	62.8
Crude starch	385	385	374	377
Ash	63.1	63.1	59.0	57.3
Calcium	12.7	12.7	10.0	9.77
Inorganic phosphorus	5.30	5.30	5.10	4.90
Metabolisable energy (MJ/kg)	14.0	14.0	13.8	13.9

C = control group; CC = group was fed grower diets with coccidiostat robenidine; M = group was fed diets containing 2% of M herb in feed; MS = group was fed diets containing 1% of M herb + 1% of S herb in feed

to procedures outlined by Zapletal et al. (2024). The following components were weighed: skinless neck, gizzard, heart, liver, abdominal fat, carcass with kidneys, both legs (thighs and drumsticks), breast with skin, and finally breast meat. The relative proportion of the respective body components and organs was calculated as their intrinsic weight relative to the slaughter weight (SW) of the chickens. Then the samples of right breast meat with skin were taken and delivered to the laboratory for evaluation of the physical properties of the chicken meat. Finally, the samples of left breast meat were taken, packaged and stored at $-20\text{ }^{\circ}\text{C}$ until the analysis of chemical and amino acid (AA) composition.

To evaluate the proximate chemical composition of breast meat, the dry matter and crude protein (CP) content were determined according to the procedures outlined by Vitula et al. (2011) and the ether extract (EE) and ash content according to the procedures outlined by Gal et al. (2022).

Regarding the physical properties of breast meat, the pH value was measured using a mea-

surement system consisting of a combined needle-tip electrode – TipTrode combination pH puncture electrode (Hamilton, USA). The drip loss test of Honikel (1998) was used to determine the water-holding capacity (WHC). Cooking loss of meat was monitored after heat treatment for 2 h in a water bath (Softcooker Y09; La Felsinea S.R.L., Tiazzola Sul Brenta, Italy) set at $70\text{ }^{\circ}\text{C}$. Chicken meat was weighed before cooking on Boeco BBI-41 laboratory scales (Boeckel & Co GmbH & Co KG, Hamburg, Germany) to the nearest 0.001 g, and immediately vacuum packed into Cryovac[®] CN300 cooking bags of 60 μm in thickness and an OTR (oxygen transmission rate) of $13\text{ cm}^3/\text{m}^2/24\text{ h}/\text{bar}$ at $23\text{ }^{\circ}\text{C}$ and 0% air humidity (Sealed Air Polska Sp. z o.o., Ożarów Mazowiecki, Poland). The packaging was performed using Henkelman Lynx 32 equipment (Henkelman Vacuum Systems, Hertogenbosch, The Netherlands). After heat treatment and removal from the bath, cooling in ice water with the addition of 50% ice followed. After 20 min the meat samples were unpacked, dried with

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filter paper and reweighed. Instrumental texture measurement (Warner–Bratzler shear force test; W–B) of meat was carried out after sous vide cooking of meat at 70 °C for 2 hours. Colour of the fresh meat samples was measured using a Konica Minolta CM-5 spectrophotometer (Konica Minolta, Japan) according to the CIE $L^*a^*b^*$ system and CIE $L^*C^*h^*$ system.

The analysis used for determination of AA content in meat was based on the methods applied by Strakova et al. (2025) using the Automatic Amino Acid Analyser AAA 400 (Ingos a.s., Prague, Czech Republic). The AA analysis was used to measure the following essential AA (EAA) – lysine (Lys), methionine (Met), isoleucine (Ile), leucine (Leu), threonine (Thr), valine (Val), histidine (His), arginine (Arg), phenylalanine (Phe) and the following non-essential AA (NEAA) – asparagine (Asn), serine (Ser), glutamine (Gln), proline (Pro), glycine (Gly), alanine (Ala), cysteine (Cys), tyrosine (Tyr).

Statistical analysis. All statistical procedures were performed using STATISTICA CZ software, v10 (StatSoft Inc., Tulsa, USA). The arithmetic mean and standard error of the mean (SEM) were determined for most of the traits evaluated.

The pen was used as an experimental unit. The Shapiro–Wilk test was used to test the normality of the distribution of data in the evaluated dietary groups of chickens. With the exception of mortality, the GLM procedure was used to evaluate the effect of diet on the evaluated traits, with diet included as a fixed effect and pen as a random effect. Subsequently, for live weight (LW), Tukey's HSD post-hoc test was used to test the significance between dietary treatment groups. For DFI, FCR, carcass traits and meat quality traits, Tukey's post-hoc test was then used to test the significance between dietary treatment groups. A chi-square test with Yates correction was used to evaluate the effect of diets on the mortality of chickens during the experimental period.

Statistical significance was set at $P < 0.05$.

RESULTS AND DISCUSSION

Growth performance and feed conversion. The impacts of dietary inclusion of the investigated M and S herbs on LW, DFI, FCR and mortality of male broilers are presented in Table 3. Significant differences in LW between dietary groups were observed

at 21, 35 and 42 days of age ($P < 0.01$; $P < 0.05$). Specifically, at 21 days of age, significantly higher LW was found in the C group compared to the CC and MS groups ($P < 0.05$). However, at 42 days of age, the highest LW was observed in the M group, which was significantly different from the CC group ($P < 0.05$). This finding is in general agreement with Abdel-Wareth et al. (2019), who found that the LW of broilers at 21 and 35 days of age increased with the increasing dietary addition of peppermint leaves (ranging from 0.5% to 1.5%) and also with the findings of Petricevic et al. (2021), who found a gradual increase in LW of 42-day-old male broilers when 0.4 and 0.6% of powder from dried whole peppermint was added to the diets. In the MS group in the present study, its final LW did not differ from the other dietary groups assessed ($P > 0.05$).

Apart from the period between 14 and 20 days of age of broilers ($P < 0.05$), there were no significant differences in DFI values between the evaluated dietary groups in the present study. In the period between 14 and 20 days of age, a higher DFI value was found in the C group compared to the CC group ($P < 0.05$). This finding is not in agreement with the results of Zapletal et al. (2025a), who found an increase in DFI values in *Eimeria*-infected broilers fed a diet with a coccidiostat compared to a control group between 21 and 27 days of age.

Regarding feed conversion within the respective periods evaluated in the present study, there was no significant difference in FCR between dietary groups ($P > 0.05$). A trend towards improved feed efficiency was observed in the MS group (FCR = 1.37) compared to the CC group (1.57; $P = 0.088$) between 28 and 34 days of age. Abdel-Wareth et al. (2019) observed an increase in DFI and a simultaneous decrease in FCR in broilers fed diets with 1 and 1.5% addition of peppermint powder from day 21 of age compared to the control group. These authors attributed the afore-mentioned fact to the content of menthol and other active compounds (e.g. cineole, citral, geraniol, linalool) in peppermint, which should promote appetite and improve digestion and nutrient absorption. Reda et al. (2025) reported that dietary supplementation of essential oils from peppermint and cloves led to the increased activity of digestive enzymes (amylase, lipase, and protease) in broilers, which act as digestive stimulants. These essential oils administered in the diet to a certain level thus improve the efficiency of nutrient hydrolysis through a number of physiologi-

Table 3. Live weight, daily feed intake (DFI), feed conversion ratio (FCR) and mortality of male broilers in relation to diet

Item	Diet				SEM	P-value
	C	CC	M	MS		
Chickens housed	40	40	40	40		
Live weight (g)						
At 14 day	378	377	382	379	2.34	0.876
At 21 day	667 ^a	627 ^b	638 ^{ab}	627 ^b	4.68	0.007
At 28 day	1 028	1 007	997	990	9.42	0.527
At 35 day	1 572	1 482	1 613	1 616	19.2	0.046
At 42 day	2 283 ^{ab}	2 126 ^b	2 387 ^a	2 272 ^{ab}	28.1	0.011
DFI (g/bird/day)						
14 to 20 day	60.6 ^a	52.9 ^b	59.5 ^{ab}	56.3 ^{ab}	1.01	0.029
21 to 27 day	87.7	83.7	89.1	86.8	0.87	0.182
28 to 34 day	110	103	124	123	3.46	0.086
35 to 42 day	169	162	183	168	4.36	0.500
14 to 42 day	107	100	114	109	1.87	0.093
FCR (g/g)						
14 to 20 day	1.47	1.49	1.63	1.59	0.03	0.090
21 to 27 day	1.72	1.54	1.74	1.68	0.03	0.184
28 to 34 day	1.42	1.57	1.42	1.37	0.03	0.088
35 to 42 day	1.75	1.79	1.75	1.90	0.05	0.727
14 to 42 day	1.59	1.60	1.63	1.63	0.02	0.783
Mortality (n)	2	6	1	2	–	0.156

^{a,b}Means within a row with different superscript letters differ ($P < 0.05$)

C = control group; CC = group was fed grower diets with coccidiostat robenidine; DFI = daily feed intake; FCR = feed conversion ratio; M = group was fed diets containing 2% of M herb in feed; MS = group was fed diets containing 1% of M herb + 1% of S herb in feed

cal mechanisms (Brenes and Roura 2010), which generally has a positive effect on the performance of fattened poultry. However, excessive levels of essential oils can reduce the effectiveness of nutrient hydrolysis due to bioactive compound saturation or antimicrobial effects on the gut microbiota, which regulates the enzyme activity (Brenes and Roura 2010). Bai et al. (2023) observed that the inclusion of peppermint and its products at some concentrations in the diet alters the caecal microbiota in chickens, with a beneficial effect on decreasing the abundance of potentially pathogenic bacteria. The microbial function prediction analysis in the above-mentioned study revealed that the favourable structure of the caecal microbiota was mainly enhanced by fatty acid degradation, fatty acid metabolism, amino sugar metabolism, nucleotide sugar metabolism, and other specific pathways.

The mortality of broilers over the entire experimental period in the present study did not differ

significantly between dietary groups ($P > 0.05$). Despite the absence of statistically significant differences between the experimental groups, the highest mortality was recorded in the CC group, whereas the lowest mortality was observed in the M group. In a study by Zapletal et al. (2025a), a similar level of broiler mortality was found when investigating the effect of 3 and 6% inclusion of wormwood (*Artemisia absinthium*) in diets used for broilers exposed to *Eimeria* infection.

Body and carcass composition. The administration of phytogetic additives in the present study had a significant effect on some indicators of body composition in 42-day-old broilers (Table 4). A significant difference was observed in SW and dressing-out percentage between the evaluated dietary groups ($P < 0.01$). As for dressing-out percentage, the highest value was found in the M group, which differed significantly from both the CC group ($P < 0.01$) and C group ($P < 0.05$).

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Table 4. Body composition (% of slaughter weight) of 42-day-old male broilers depending on diet

Trait	Diet				SEM	P-value
	C	CC	M	MS		
SW (g)	2 372 ^{AB,bc}	2 233 ^{B,c}	2 623 ^{A,a}	2 460 ^{AB,ab}	33.3	0.001
Dressing-out	67.7 ^{AB,b}	66.5 ^B	70.3 ^{A,c}	69.8 ^{A,ab}	0.37	0.001
Legs	20.8	20.5	21.7	21.7	0.19	0.050
Breast with skin	20.6	21.5	21.6	21.4	0.24	0.360
Breast meat	19.1	20.0	20.1	19.8	0.23	0.393
Neck without skin	1.03	1.10	1.02	1.08	0.02	0.429
Gizzard	0.75	0.64	0.72	0.81	0.03	0.063
Liver	2.45	2.50	2.21	2.24	0.04	0.023
Heart	0.59 ^{AB}	0.63 ^A	0.54 ^B	0.54 ^B	0.01	0.002
Abdominal fat	0.85 ^{AB}	0.73 ^B	0.99 ^A	0.90 ^{AB}	0.03	0.010

^{a-c}Means within a row with different superscript letters differ ($P < 0.05$); ^{A,B}Means within a row with different superscript letters differ ($P < 0.01$)

C = control group; CC = group was fed grower diets with coccidiostat robenidine; M = group was fed diets containing 2% of M herb in feed; MS = group was fed diets containing 1% of M herb + 1% of S herb in feed; SW = slaughter weight

A significant increase in dressing percentage was also observed in the MS group compared to the CC group ($P < 0.01$). Similarly, Asadi et al. (2017) reported a higher dressing percentage in broilers fed diets with peppermint at the 0.4% and 0.6% level as compared to the control group and broilers fed diets with lower doses of peppermint. In addition, a positive effect on carcass characteristics was observed by Wang et al. (2024) in 42-day-old broilers, when diets were supplemented with 1, 3, and 5% of fermented *Artemisia argyi*.

A significant difference was also detected in the leg proportion in the present study ($P < 0.05$), with a trend towards higher values in the herb-supplemented groups (M, MS). Regarding the abdominal fat, a significantly higher proportion in the M group compared to the CC group was observed ($P < 0.01$). This finding is consistent with the results of Ocak et al. (2008), who reported an increased proportion of abdominal fat in broilers fed a diet supplemented with 0.2% of peppermint leaves. They relate the increased abdominal fat deposition to the higher growth rate of broilers, which in turn may lead to the greater body fat accumulation (Tumova and Teimouri 2010). Besides, the percentage of abdominal fat in broilers of the C group in the present study is slightly higher than that found by Tyl et al. (2025) in 35-day-old male Ross 308 broilers.

A lower heart proportion was recorded in the M and MS groups compared to the CC group in the present study ($P < 0.01$; Table 4). This difference

reflects the higher dressing percentage in the M and MS groups. Further, a trend towards the lower liver proportion was noted in the M and MS groups compared to the CC and C groups ($P < 0.05$), which is in agreement with Asadi et al. (2017). Although there was no significant difference ($P = 0.063$), the lowest proportion of gizzard in the present study was observed in the CC group compared to the other dietary groups assessed.

Physicochemical properties and amino acid profile of meat. Regarding the proximate chemical composition of breast meat in the present study (Table 5), the dietary effect was confirmed ($P < 0.01$) on the CP and EE content. Compared to the C group, the M and MS groups showed lower CP content ($P < 0.05$). In contrast, the C group showed significantly lower EE content compared to the MS group ($P < 0.05$) and especially to the M group ($P < 0.01$). These findings may be related to the intrinsic content and composition of biologically active compounds naturally occurring in both studied herbs (Ekiert et al. 2021; Anyaoku et al. 2023).

As for the physical properties of breast meat in the present study (Table 5), a dietary effect was found on the yellowness b^* and chroma C^* characteristics ($P < 0.01$). Specifically, the lowest values for yellowness b^* and chroma C^* characteristics were found in the C group, where their levels were significantly lower compared to the MS group ($P < 0.05$) and especially to the M group

Table 5. Proximate chemical composition (g/kg on basis of fresh meat) and physical properties of male broiler breast meat depending on diet

Trait	Diet				SEM	P-value
	C	CC	M	MS		
Chemical traits						
Dry matter	237	240	240	238	1.13	0.843
Crude protein	217 ^a	214 ^{ab}	207 ^b	209 ^b	1.28	0.007
Ether extract	14.7 ^{B,b}	19.1 ^{AB,ab}	22.7 ^{A,a}	20.6 ^{AB,a}	0.87	0.003
Ash	11.3	11.1	11.3	11.4	0.05	0.137
Physical traits						
WHC (%)	1.66	1.37	1.48	1.17	0.08	0.185
Cooking loss (%)	21.3	22.7	23.9	22.8	0.36	0.06
WB (N)	13.0	10.9	12.6	12.8	0.34	0.082
pH	5.80	5.76	5.79	5.77	0.02	0.821
<i>L</i> [*]	54.0	55.3	55.3	55.4	0.38	0.473
<i>a</i> [*]	-0.66	-0.02	0.12	-0.19	0.13	0.184
<i>b</i> [*]	10.6 ^{B,b}	12.0 ^{AB,ab}	13.2 ^A	12.3 ^{AB,a}	0.26	0.001
<i>C</i> [*]	10.6 ^{B,b}	12.1 ^{AB,ab}	13.2 ^A	12.3 ^{AB,a}	0.25	0.001
<i>h</i> [*]	93.6	90.3	89.4	91.5	0.64	0.123

^{a,b}Means within a row with different superscript letters differ ($P < 0.05$); ^{A,B}Means within a row with different superscript letters differ ($P < 0.01$)

a^{*} = redness; *b*^{*} = yellowness; C = control group; *C*^{*} = chroma; CC = group was fed grower diets with coccidiostat robenidine; *h*^{*} = hue angle; *L*^{*} = lightness; M = group was fed diets containing 2% of M herb in feed; MS = group was fed diets containing 1% of M herb + 1% of S herb in feed; WB = Warner–Bratzler shear force; WHC = water holding capacity

($P < 0.01$). This finding is consistent with the results of Aminzade et al. (2012), who also found an increase in yellowness *b*^{*} parameter in the breast meat of Japanese quails, which were fed a diet with supplementation of 1.5% and 3.0% peppermint. Meat colour is related to pigment (myoglobin) content and can be affected by many factors, including also the nutritional status (Honikel 1998). Peppermint and southernwood are known to contain anthocyanins, carotenoids, chlorophyll and flavonoids, among others (Ekiert et al. 2021; Anyaoku et al. 2023), and as shown by the results of the present study, the dietary inclusion of these herbs can markedly influence the pigment content in poultry meat. In addition, in the present study a trend towards a higher level of cooking loss was observed in the M group compared to the C group ($P = 0.06$), which is not in agreement with findings of Abdel-Wareth et al. (2019).

Despite the CP content being lower ($P < 0.05$) in the breast meat of broilers in the MS group compared to the C group in the present study, the meat of broilers in the MS group was found to have the

highest content of both all EAA ($P < 0.05$) and all NEAA ($P < 0.01$), and at the same time the total content of all AA ($P < 0.01$; Table 6). In the case of individual EAA, dietary treatments affected the contents of Thr, Val, and Arg ($P < 0.01$) as well as the contents of Lys and His ($P < 0.05$). As for individual NEAA, dietary treatments affected the contents of Ser, Gln, Pro, and Ala ($P < 0.01$) and furthermore the contents of Asn, Gly, and Cys ($P < 0.05$). Of the total number of AA that were found to be significantly different in relation to the dietary effect, 11 of them had the highest level in the MS group. Among the other AA, only His content was significantly higher in the CC group compared to the M group ($P < 0.05$). Significantly lower total contents of the evaluated EAA were found in the C and M groups compared to the MS group ($P < 0.05$). Total content of all evaluated NEAA in the MS group was significantly higher in comparison with all other evaluated dietary groups of broilers ($P < 0.01$). It can be concluded that dietary inclusion of the southernwood herb affected protein synthesis in *Eimeria*-challenged broilers. This finding is in general agreement with

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Table 6. Amino acids content (g/100 g of fresh meat) in male broiler breast meat depending on diet

Trait	Diet				SEM	P-value
	C	CC	M	MS		
Lys	1.67 ^b	1.79 ^{ab}	1.70 ^{ab}	1.82 ^a	0.02	0.015
Met	0.43	0.43	0.42	0.33	0.02	0.052
Ile	0.99	1.05	1.00	1.07	0.02	0.069
Leu	1.50	1.56	1.49	1.57	0.02	0.069
Thr	0.81 ^{AB,b}	0.78 ^B	0.83 ^{AB,b}	0.90 ^{A,a}	0.01	0.001
Val	1.01 ^b	1.08 ^{ab}	1.01 ^b	1.12 ^a	0.02	0.004
His	1.07 ^{ab}	1.14 ^a	1.04 ^b	1.13 ^{ab}	0.01	0.018
Arg	1.18 ^B	1.17 ^B	1.28 ^B	1.49 ^A	0.03	0.001
Phe	0.76	0.78	0.76	0.81	0.01	0.076
Suma EAA	9.40 ^b	9.77 ^{ab}	9.52 ^b	10.25 ^a	0.11	0.012
Asn	1.87 ^{ab}	1.83 ^b	1.88 ^{ab}	2.04 ^a	0.03	0.022
Ser	0.68 ^B	0.58 ^C	0.71 ^{AB,b}	0.79 ^{A,a}	0.02	<0.001
Gln	3.03 ^B	3.05 ^{AB,b}	3.11 ^{AB,ab}	3.30 ^{A,a}	0.03	0.005
Pro	0.63 ^{B,b}	0.70 ^{B,ab}	0.73 ^{B,a}	0.85 ^A	0.02	0.001
Gly	0.85 ^b	0.87 ^{ab}	0.86 ^{ab}	0.91 ^a	0.01	0.023
Ala	1.12 ^B	1.15 ^B	1.16 ^B	1.26 ^A	0.02	0.001
Cys	0.16 ^b	0.15 ^b	0.16 ^{ab}	0.19 ^a	0.01	0.013
Tyr	0.63	0.60	0.62	0.64	0.01	0.237
Suma NEAA	8.97 ^B	8.94 ^B	9.23 ^B	9.98 ^A	0.11	0.001
Total AA	18.4 ^B	18.7 ^{AB,b}	18.8 ^{AB,b}	20.2 ^{A,a}	0.22	0.002

^{a,b}Means within a row with different superscript letters differ ($P < 0.05$); ^{A-C}Means within a row with different superscript letters differ ($P < 0.01$)

AA = amino acids; Ala = alanine; Arg = arginine; Asn = asparagine; C = a control group; CC = a group was fed grower diets with coccidiostat robenidine; Cys = cysteine; EAA = essential amino acids; Gln = glutamine; Gly = glycine; His = histidine; Ile = isoleucine; Leu = leucine; Lys = lysine; M = a group was fed diets containing 2% of M herb in feed; Met = methionine; MS = a group was fed diets containing 1% of M herb + 1% of S herb in feed; NEAA = non-essential amino acids; Phe = phenylalanine; Pro = proline; Ser = serine; Thr = threonine; Tyr = tyrosine; Val = valine

the results of recent studies by Zapletal et al. (2024), and also with a study by Zapletal et al. (2025a) in *Eimeria*-challenged broilers. It appears that dietary inclusion of herbs of the genus *Artemisia* can significantly alter the amino acid profile of the *pectoralis* muscle of chickens, and the potential dietary use of selected species of these herbs could thus purposely increase the nutritional value of chicken meat proteins. Similarly, dietary supplementation of some phytogetic additives was associated with an increase in the content of some AA in poultry meat and eggs (Gui et al. 2025) and in pork (Lin et al. 2020). This beneficial effect on the quality of animal products was attributed to the increased abundance of beneficial bacteria in the jejunum (Gui et al. 2025) and improved digestion and absorption of nutrients in the small intestine (Lin et al. 2020). An interesting

finding in the present study is a markedly higher Ser content ($P < 0.001$) in the meat of broilers fed a diet with combined supplementation of M and S, especially in relation to broilers fed the grower diet with a coccidiostat. This finding is somewhat consistent with the results of the study by Zapletal et al. (2025a) in the case of dietary administration of wormwood.

Regarding the dosage of phytogetic feed additives, it should be emphasised that mild levels can positively stimulate performance by improving nutrient utilisation and promoting overall health. However, higher doses can lead to reduced feed intake, malabsorption, and potential toxicity (Obianwuna et al. 2024). It is known that the use of different levels of phytogetic additives in chickens can affect feed or water palatability, with higher

levels generally leading to reduced intake (Dilawar et al. 2021; Ravi et al. 2025). Based on the findings of previous studies evaluating various dietary concentrations of *Artemisia* spp. herbs (Zapletal et al. 2024; Zapletal et al. 2025a), it can be concluded that specific doses substantially altered the performance, health status of fattened broilers, and also the quality of their meat. To specify the limits for the routine use of the studied herbs in commercial poultry production, it would be useful to conduct further studies focused on the form of their possible technological processing and application, dietary exposure, period of administration during fattening, etc.

CONCLUSION

In conclusion, in *Eimeria*-challenged broilers, the addition of 2% peppermint powder to the diet led to an increase in final live weight compared to broilers fed the grower diet with a coccidiostat, which was however associated with a higher deposition of both abdominal fat and fat in the breast meat of these chickens. Dietary administration of peppermint alone and in combination with southernwood resulted in a favourably higher dressing percentage, which was related to a higher proportion of legs compared to the group fed the grower diet with a coccidiostat. Furthermore, the improvement in dressing percentage in broilers fed diets with supplemented herbs was associated with a lower proportion of heart and liver.

As for the colour of breast meat, higher values of yellowness b^* and chroma C^* characteristics were observed in broilers fed diets supplemented with herbs compared to the control group. In spite of the fact that a lower crude protein content was detected in the meat of broilers fed a diet with the combined administration of peppermint and southernwood, the highest content of all essential amino acids as well as the total content of all assessed amino acids were found in their meat, resulting in a considerable increase in the nutritional value of breast meat proteins of these broilers.

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Conflict of interest

The authors declare no conflict of interest.

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