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Broilers responses to dietary wormwood administration under *Eimeria*-challenged conditions

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Abstract: Some herbs can be used as phytogenic additives as they naturally contain specific bioactive phytochemicals that are effective in controlling coccidiosis and some other poultry diseases due to their immunostimulatory effects and they can be also nutritious. The aim of the study was to evaluate the effect of *Artemisia absinthium* (WW) meal inclusion to a diet on the production performance, oocyst output and minerals content of meat in broilers infested by *Eimeria* oocysts. Ross 308 broilers were randomly assigned to four dietary groups. Broilers in the control group were fed basal diets without coccidiostat or WW. Broilers of the R group were fed starter and grower diets supplemented with coccidiostat robenidine. Broilers in the W3 and W6 groups were fed diets containing 3 and 6% of WW in feed, resp. At the age of 16 days, all broilers were infested by oocysts of *Eimeria* spp. to induce a mild coccidiosis infection. The WW meal had no effect on the production performance and carcass traits of broilers ($P > 0.05$). However, the used levels of administered WW meal were not effective in reducing the number of oocysts faecal output in infested broilers. Administration of coccidiostat robenidine prevented the development of *Eimeria* oocysts in the intestine as long as 4 days after the cessation of feeding the medicated diet to broilers ($P < 0.05$). As for minerals of breast meat, broilers of the W3 group displayed a higher Ca level compared to those in the R group ($P < 0.05$). On the contrary, the R group showed the highest Cu level compared to all other dietary groups assessed ($P < 0.01$). It can be concluded that for the coccidiosis control in poultry production, it is advisable to monitor the content of specific bioactive substances with proven anticoccidial activity when using WW products.

Keywords: *Artemisia absinthium*; coccidiosis; chicken; minerals of meat; oocyst output; production

Enteric diseases, including coccidiosis, are the most significant ones in poultry farming from both economic point of view and welfare (M'Sadeq 2023). Coccidiosisis a parasitic disease caused by seven *Eimeria* spp. in poultry that cause considerable damage to the intestinal tract, and which are different in prevalence and pathogenicity (Beski 2023; Khan et al. 2023). *Eimeria* infection can have serious consequences for birds, including symptoms such as intestinal leakage of plasma protein,

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impaired nutrient absorption and growth, tissue damage, increased vulnerability to other infectious gut diseases, higher mortality and so on (Khan et al. 2023; M'Sadeq 2023).

Possibilities to control poultry coccidiosis include anticoccidial chemicals, vaccines and natural products that often contain some herbs. Anticoccidial chemicals, coccidiostats, coccidioides, and ionophores have long been used for decades as a principal tool to control coccidiosis in poultry production. Even though these chemical substances are effective and economically acceptable approach, the hazard of resistance to these substances and public demands for residue-free meat has encouraged development of alternative strategies to control coccidiosis (Chapman et al. 2010; Beski 2023). Vaccination made up of one or more strains of wild-type or attenuated *Eimeria* spp. comprises the other option to control this parasitic disease in poultry farming, whereas in Europe, the use of attenuated *Eimeria* vaccines are only approved. The use of phytogenic products as an alternative to anticoccidial agents can overcome the threats posed by chemical drugs. Such herbs may contain bioactive phytochemicals, which are effective in controlling both avian coccidiosis and some other poultry diseases due to their natural immune-stimulating effects, while being safe and nutritious. In addition, poultry fattened with herbal additives is generally well accepted among current consumers and sometimes can be even therapeutic for consumers of such poultry meat (Srinivasu et al. 2020; El-Shall et al. 2022).

The genus *Artemesia* is of interest because of its broad range of pharmacologic effects, including antioxidant, anti-inflammatory, immune-boosting and antiparasitic properties (Fiamegos et al. 2011; Sharifi-Rad et al. 2022). The plant of *Artemisia absinthium* L., known as wormwood (WW), is used as a stomachic, cholagogue, antiseptic, antispasmodic, carminative, febrifuge and anthelmintic. The numerous bioactive phytochemical substances thus make the WW a potential botanical in-feed additive in animal production (Amin et al. 2022; Zapletal et al. 2024).

Although recent studies have confirmed a number of different beneficial effects of dietary administered *Artemisia* spp. herbs on the chicken organism (Abdullah and Al-Barwary 2020; Wang et al. 2024; Zapletal et al. 2024), there are still quite limited insights into the effect of use of wormwood herb as a dietary supplement on *Eimeria* oocysts output and production performance of broilers after coccidiosis infection. Thus, this study aimed to evaluate the effect of wormwood meal inclusion to a diet on the production performance, faecal oocyst output, carcass traits and minerals content of breast meat in fast-growing broilers challenged with *Eimeria* spp.

MATERIAL AND METHODS

The experiment was approved by the Animal Welfare Committee of University of Veterinary Sciences Brno (Approval No. PP9-2022).

Broiler husbandry

Feeding trial was performed on 208 one-day-old male Ross 308 chicks, which were randomly assigned to four treatments, each with four replicate pens and thirteen birds per pen in the accredited stable of the Department of Animal Breeding, Animal Nutrition and Biochemistry, University of Veterinary Sciences Brno. Broilers of a specific dietary group were housed in one enclosed room, which was divided into 4 equally sized pens (replicates). All broilers were housed in floor pens bedded with wood shavings and under the same controlled housing conditions that were adjusted according to the Ross 308 guidelines (Aviagen 2018). Feed and water were supplied *ad libitum* for broilers.

Broilers were observed for any signs of illness and behavioural changes twice a day. The trial lasted for 42 days. During the trial period, starter diets were fed between days 1–14, grower diets between days 15–28, and finisher diets between days 29–42. The starter diet was fed in the form of crumbled pellets and the grower and finisher diets were fed in the form of pellets. On days 7, 14, 21, 28, 35 and 42, individual birds were weighed and leftover feed was weighed on pen-basis. Then, average daily gain (ADG), daily feed intake (DFI), European production efficiency factor (EPEF) and European broiler index (EBI) were evaluated as the main performance traits. EPEF and EBI were calculated according to the following formula stated earlier by Tyl et al. (2024) and Zaremba et al. (2024), respectively:

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$$EPEF = (FLW \times VIA)/(LFP \times FCR) \times 100 \quad (1)$$
$$EBI = (VIA \times ADG)/(FCR \times 10) \quad (2)$$

where:
FLW – final live weight (kg);
VIA – viability (%);
LFP – length of fattening period (days);
FCR – feed conversion ratio (kg/kg);
ADG – average daily gain (g/bird/day).

Dietary treatments

Broilers in the control (C) group were fed basal diets without coccidiostat or WW herb. Broilers of the R group were fed starter and grower basal diets supplemented with coccidiostat robenidine (33 mg/kg of feed); the finisher diet for the R group didn't contain coccidiostat. The broilers in the W3 and W6 groups were fed diets containing 3 and 6% of WW in feed, respectively. The W3 and W6 diets didn't contain any commercial coccidiostat. On dry matter basis, the meal from whole aerial parts of WW (*Artemisia absinthium*) included to W3 and W6 diets contained: 11.4% of crude protein, 33.5% of crude fibre, 2.2% of crude fat, 8.1% of ash, 0.52% of total polyphenols and 0.23% of total flavonoids. For all dietary groups, diets were designed as isoenergetic and isonitrogenous. Table 1, 2 and 3 show the composition of diets used, when Mastercube® is a pellet binder composed of a synergistic blend of polysaccharide gums, starch and mineral hardener.

Eimeria infestation and oocysts counting

At the age of 16 days, broilers in all dietary groups were infested by tenfold overdosing of live vaccine LIVACOX T (Biopharm Co., Prague, Czech Republic) to induce a mild coccidiosis infection according to procedure earlier used by Yu et al. (2021). Inoculum dose of 1 ml was orally gavaged into the crop to each broiler. Inoculum dose contained 15 000 live oocysts of *E. acervulina*, *E. tenella* and *E. maxima*.

Collective faeces per pen base were gathered on days 6, 10, 13, 17, 20, 24, and 27 post infection (DPI). For the examination of faecal samples for oocysts per one gram of faeces (OPG), the apparently freshest broiler faeces in each pen were always collected to make the examination representative, and the person sampling the pens always was provided with clean shoe covers when entering each enclosed room. The freshest faeces were collected from 5 locations in the pen, with four samples collected from the corners and the fifth sample obtained from the centre of the pen to contain at least 15 g of faeces without bedding. These samples were mixed into one and the McMaster technique was further applied.

Table 1. Compositions of starter diets fed broilers from day 1 to 14 as fed basis

Item	Diet			
	C	R	W3	W6
Ingredients (%)				
Wheat	7.96	7.96	7.96	7.96
Maize	46.9	46.9	46.9	46.9
Soybean meal	34.2	34.2	34.2	34.2
Canola oil	0.94	0.94	0.94	0.94
Monocalcium phosphate	0.94	0.94	0.94	0.94
Limestone	1.41	1.41	1.41	1.41
NaCl	0.23	0.23	0.23	0.23
$NaHCO_3$	0.19	0.19	0.19	0.19
Lysine	0.19	0.19	0.19	0.19
Methionine	0.26	0.26	0.26	0.26
Threonine	0.07	0.07	0.07	0.07
Mineral and vitamin premix	0.47	0.47	0.47	0.47
Mastercube®	0.28	0.28	0.28	0.28
Sunflower hulls	0.90	0.90	0.45	0.00
Sunflower seed meal	0.30	0.30	0.15	0.00
Alfalfa pellets	2.70	2.70	1.35	0.00
Apple pomace	2.10	2.10	1.05	0.00
Wormwood meal	0.00	0.00	3.00	6.00
Robenidine (mg/kg of feed)	33.0	0.00	0.00	0.00
Analysed nutrients (%)				
Crude protein	20.5	20.6	20.6	20.4
Crude fibre	3.6	3.5	3.7	4.0
Crude fat	3.1	3.1	3.5	3.3
Crude starch	35.6	35.4	34.5	34.1
Ash	5.9	5.9	5.9	5.8
Calcium	0.80	0.81	0.89	0.92
Inorganic phosphorus	0.59	0.57	0.56	0.55
Metabolisable energy (MJ/kg)	12.8	12.8	12.9	12.7

C = a control group; R = a group was fed medicated starter and grower diets with coccidiostat robenidine; W3 = a group was fed diets containing 3% of WW meal in feed; W6 = a group was fed diets containing 6% of WW meal in feed

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All of the samples were prepared directly after collecting. Exactly 3 g of faeces were mixed with 42 ml of water. The solution was sieved trough a mesh and transferred into conical tubes (15 ml). These tubes were centrifuged (2 000 rpm) for 3 minutes. Later all the supernatant was poured off and saturated salt solution was added. The tube was vortexed for 10 seconds. An aliquot of the faecal sample was transferred into a McMaster chamber and all the *Eimeria* oocysts were counted manually in both parts of the chamber. The sum was multiplied by a dilution factor (50) to obtain the final number of OPG value.

Table 2. Compositions of grower diets fed broilers from day 15 to 28 as fed basis

Item	Diet			
	C	R	W3	W6
Ingredients (%)				
Wheat	8.77	8.77	8.77	8.77
Maize	46.8	46.8	46.8	46.8
Soybean meal	28.5	28.5	28.5	28.5
Canola oil	4.49	4.49	4.49	4.49
Monocalcium phosphate	0.65	0.65	0.65	0.65
Limestone	1.17	1.17	1.17	1.17
NaCl	0.22	0.22	0.22	0.22
$NaHCO_3$	0.19	0.19	0.19	0.19
Lysine	0.13	0.13	0.13	0.13
Methionine	0.22	0.22	0.22	0.22
Threonine	0.06	0.06	0.06	0.06
Maize sprouts	1.87	1.87	1.87	1.87
Mineral and vitamin premix	0.47	0.47	0.47	0.47
Mastercube®	0.47	0.47	0.47	0.47
Sunflower hulls	0.90	0.90	0.45	0.00
Sunflower seed meal	0.30	0.30	0.15	0.00
Alfalfa pellets	2.70	2.70	1.35	0.00
Apple pomace	2.10	2.10	1.05	0.00
Wormwood meal	0.00	0.00	3.00	6.00
Robenidine (mg/kg of feed)	33.0	0.00	0.00	0.00
Analysed nutrients (%)				
Crude protein	19.0	19.0	19.1	19.1
Crude fibre	3.3	3.3	3.2	3.2
Crude fat	8.0	8.0	8.0	8.1
Crude starch	35.2	35.2	35.5	35.1
Ash	5.4	5.5	5.4	5.3
Calcium	0.87	0.89	0.83	0.91
Inorganic phosphorus	0.48	0.49	0.48	0.47
Metabolisable energy (MJ/kg)	14.0	13.9	14.0	14.0

C = a control group; R = a group was fed medicated starter and grower diets with coccidiostat robenidine; W3 = a group was fed diets containing 3% of WW meal in feed; W6 = a group was fed diets containing 6% of WW meal in feed

Table 3. Compositions of finisher diets fed broilers from day 29 to 42 as fed basis

Item	Diet			
	C	R	W3	W6
Ingredients (%)				
Wheat	9.05	9.05	9.05	9.05
Maize	46.8	46.8	46.8	46.8
Soybean meal	28.5	28.5	28.5	28.5
Canola oil	3.27	3.27	3.27	3.27
Monocalcium phosphate	0.65	0.65	0.65	0.65
Limestone	1.17	1.17	1.17	1.17
NaCl	0.22	0.22	0.22	0.22
$NaHCO_3$	0.19	0.19	0.19	0.19
Lysine	0.13	0.13	0.13	0.13
Methionine	0.22	0.22	0.22	0.22
Threonine	0.06	0.06	0.06	0.06
Maize sprouts	2.81	2.81	2.81	2.81
Mineral and vitamin premix	0.47	0.47	0.47	0.47
Mastercube®	0.47	0.47	0.47	0.47
Sunflower hulls	0.90	0.90	0.45	0.00
Sunflower seed meal	0.30	0.30	0.15	0.00
Alfalfa pellets	2.70	2.70	1.35	0.00
Apple pomace	2.10	2.10	1.05	0.00
Wormwood meal	0.00	0.00	3.00	6.00
Analysed nutrients (%)				
Crude protein	18.8	18.7	18.7	18.9
Crude fibre	4.1	4.1	3.7	4.1
Crude fat	6.3	6.3	6.4	6.2
Crude starch	35.5	35.4	34.7	33.9
Ash	5.2	5.2	5.1	5.0
Calcium	0.78	0.76	0.70	0.71
Inorganic phosphorus	0.45	0.46	0.48	0.49
Metabolisable energy (MJ/kg)	13.5	13.5	13.5	13.4

C = a control group; R = a group was fed medicated starter and grower diets with coccidiostat robenidine; W3 = a group was fed diets containing 3% of WW meal in feed; W6 = a group was fed diets containing 6% of WW meal in feed

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Slaughter and meat analysis

At the end of the trial, eight chickens (two birds/replicate) per a dietary group were randomly selected for carcass characteristics evaluation. The birds were stunned and exsanguinated by cutting the jugular vein. Slaughter and carcass processing were carried out according to procedures described by Zapletal et al. (2024). Breast meat samples were taken, packed and stored at –20 °C until an analysis of their mineral composition was carried out. The content of mineral elements was determined according to methods used by Gal et al. (2022).

Statistical analysis

The arithmetic mean and standard error of mean (SEM) were determined for all evaluated traits. Normality analysis of data distribution was performed by a Shapiro-Wilk test; normality was not found in OPG values. Data showing normal distribution were processed by a GLM procedure, whereas the diet was included as a fixed effect and pen was included as a random effect. Tukey's post-hoc test was used when significant effects were found. The pen was used as an experimental unit, in accordance with the procedure used by Tumova et al. (2021). To determine differences in OPG values among dietary groups concerned, the Kruskal-Wallis test was used. Thereafter, multiple comparisons of mean ranks was used when significant effects were found. The significance was considered at the $P < 0.05$ level. All statistical procedures were performed by the STATISTICA CZ v10 software.

RESULTS AND DISCUSSION

Broiler performance

The dietary effect of WW herb on the ADG, DFI, EPEF and EBI is shown in Table 4. The results of this study showed that the inclusion of WW to the diet had no effect ($P > 0.05$) on ADG, DFI, EPEF and EBI values in broilers fattened up to 42 days of age.

As for growth intensity, almost identical ADG values were found for the entire fattening period in broilers fed starter and grower mixture with the coccidiostat robedinine (79.5 g/bird) and in broilers fed 3% of WW in the diet (78.3 g/bird). The results of this study are in agreement with the findings reported by Zapletal et al. (2024) for female broilers fed diets containing 5 and 10% of WW between 21 and 42 days of age. By contrast, Wang et al. (2024) found a lower ADG value in broilers fed diets supplemented with *Artemisia argyi* and fermented *Artemisia argyi* between days 1 and 14 of age compared to the control group. However, in the final phase of fattening, these authors observed a significant increase in ADG in broilers fed a diet with 3% of fermented *Artemisia argyi*, which they attributed to an increase in probiotics in the intestines that promote digestion and nutrient absorption. Besides that, the levels of ADG values found in this study are generally higher than those found by Zapletal et al. (2024) in *Eimeria*

Table 4. Effect of diet on broilers performance

Item	Diet				SEM	*P*-value
	C	R	W3	W6		
ADG (g)						
1 to 6 day	19.0	19.5	19.5	18.7	0.17	0.407
7 to 13 day	32.8	36.5	35.9	34.8	0.60	0.101
14 to 20 day	59.4	62.0	63.0	57.7	0.86	0.119
21 to 27 day	86.8	84.3	88.1	86.5	0.90	0.567
28 to 34 day	124	128	122	119	1.3	0.104
35 to 42 day	141	147	142	137	2.7	0.367
1 to 42 day	77.3	79.5	78.3	74.9	4.55	0.987
DFI (g/bird)						
1 to 6 day	24.6	26.0	25.3	24.8	0.38	0.669
7 to 13 day	56.9	58.4	56.0	55.8	1.10	0.822
14 to 20 day	86.7	92.5	90.1	85.0	1.10	0.077
21 to 27 day	137^{ab}	153^{a}	139^{ab}	136^{b}	2.4	0.025
28 to 34 day	176	175	176	167	1.5	0.117
35 to 42 day	234	237	223	215	6.0	0.673
1 to 42 day	119	123	118	114	1.4	0.065
EPEF	500	522	531	497	6.7	0.270
EBI	493	515	524	494	6.4	0.315

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

ADG = average daily gain; C = a control group; DFI = daily feed intake; EBI = European broiler index; EPEF = European production efficiency factor; R = a group was fed medicated starter and grower diets with coccidiostat robenidine; W3 = a group was fed diets containing 3% of WW meal in feed; W6 = a group was fed diets containing 6% of WW meal in feed

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non-challenged female broilers between 21 and 42 days of age.

Regarding feed intake of broilers in this study, a significantly different value for DFI among the tested diets was found in the period between 21 and 27 days of age only. Specifically, broilers of the R group showed a significantly higher feed intake compared to those of the W6 group ($P < 0.05$). The DFI values in the other particular fattening periods, as well as for the entire fattening period assessed, did not differ among the dietary groups ($P > 0.05$). The findings of this study are in general agreement with the results of study carried out by Zapletal et al. (2024) and also with results of study conducted by Kostadinovic et al. (2015), in which the supplementation of wormwood leaf meal in the amounts of 10 to 20% to the basal diet was monitored for 42 days of broiler fattening.

In this study, the inclusion of WW herb in the diet had no significant effect on the resulting EPEF value ($P > 0.05$), with the highest value being obtained in the W3 group. The EPEF value indicates the economic effect of broiler production, where the higher the value of this indicator, the more profitable the production (Aviagen 2018). The EPEF values in this study were higher than those found by Zhang et al. (2021) in fattening Arbor Acres male broilers up to 42 days of age (410 to 451), and also than those found by Tyl et al. (2024) in fattening broilers up to 35 days of age, which is the current length of fattening for fast-growing broilers in the Czech Republic. As for another production efficiency indicator, the European broiler index (EBI), there was no significant effect of diets on its value ($P > 0.05$) in this study; while the highest EBI value was again found in the W3 group (524). In addition, the EBI values found in this study were considerably higher than those found by Alizadeh-Ghamsari et al. (2023) in the case of fattening Ross 308 male broilers up to 42 days of age (324 to 362).

Faecal oocyst output

As for oocyst shedding in faeces of infected broilers in this study, inclusion of both 3 and 6% WW meal to the diet was not an effective strategy to reduce the number of OPG (Table 5). Contrary to our expectation, the chosen levels of dietary administered WW meal even led to the increase in shed oocysts in comparison with the C group up to 17th DPI ($P > 0.05$). The findings of this study are not in agreement with the findings of Remmal et al. (2011), who found a considerable destructive effect of applied essential oil (EO) derived from *A. absinthium* on *Eimeria* oocysts under in vitro conditions, nor with the findings of Kostadinovic et al. (2016), who confirmed a positive effect of dietary administered EO from *A. absinthium* on *Eimeria* oocyst output in broiler faeces from 3rd to 9th DPI compared to a positive control group without any targeted treatment. Moreover, in the study of Kostadinovic et al. (2016), in broilers treated with EO from *A. absinthium*, faecal values of OPG were always significantly higher compared to those found in broilers fed diet with coccidicide salinomycin (60 mg/kg) throughout the monitoring period. Although a positive effect of the administered leaf powder of *A. annua*, a different *Artemisia* species, on reduced faecal OPG and health in *Eimeria tenella* infested poultry was recently reported (El-Shall et al. 2022), the mere dietary inclusion of meal from *A. absinthium* in this study did not lead to the expected reduction in the number of oocysts faecal outputs. The intrinsic composition of some biologically active compounds contained in WW can be greatly influenced by a number of factors, with the geographical origin of the herb, soil type and altitude of cultivation, harvest time, and length and method of storage being among the significant ones (Amin et al. 2022). In this regard, direct

Table 5. Oocyst count per 1 gram (OPG) in faeces in relation to diet

DPI	Age (days)	Diet				SEM	*P*-value
		C	R	W3	W6		
6	21	150^{ab}	0^{b}	425^{a}	437^{a}	174.9	0.013
10	25	212^{ab}	0^{b}	1 013^{a}	938^{a}	326.8	0.005
13	28	5 187^{ab}	0^{b}	13 038^{a}	8 025^{a}	4 738.1	0.020
17	32	3 075^{ab}	0^{b}	7 300^{ab}	12 638^{a}	3 977.4	0.023
20	35	1 650	137	2 325	1 575	1 002.0	0.068
24	39	400	225	137	337	114.3	0.316
27	42	175	837	200	187	104.8	0.144

[a,b]Means within a row with different superscript letters differ ($P < 0.05$)

C = a control group; DPI = days post infection; R = a group was fed medicated starter and grower diets with coccidiostat robenidine; W3 = a group was fed diets containing 3% of WW meal in feed; W6 = a group was fed diets containing 6% of WW meal in feed

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application of the derived EO from *A. absinthium*, ideally with validated contents of β-thujone, 1-8 cineole and *p*-cymene (Remmal et al. 2011; Muthamilselvan et al. 2016), seems to be a more reliable tool for controlling coccidiosis in poultry production.

In case of the diets tested in this study, only supplementation with the coccidiostat robenidine at dose of 33 mg/kg starter and grower was an effective strategy to eradicate infested attenuated *Eimeria* spp. ($P < 0.05$); no shed oocysts were detected in any of the faeces samples examined up to 17th DPI. Moreover, dietary administration of robenidine successfully prevented the development of *Eimeria* oocysts in the broiler intestine for 4 days after the cessation of feeding the medicated grower.

The highest value for faecal OPG in the W3 group (13 038) was detected at 13 DPI as was that of the C group (5 187), while the peak OPG count in the W6 group (12 638) was detected at 17 DPI. Subsequently, broilers fed the tested WW-supplemented diets showed a decrease in oocyst shedding, with a minimum value at the end of fattening. Conversely, in broilers of the R group, there was an increase in oocyst faecal output from day 35 of age due to absence of coccidiostat in the finisher diet, with a maximum OPG value (837) at the end of the trial period. This growing trend corresponded to some extent to the situation with a gradual increase in the number of oocysts shed in the WW groups after the experimental infestation with *Eimeria* spp.

Table 6. Carcass traits of broilers within dietary groups

Trait (g)	Diet				SEM	*P*-value
	C	R	W3	W6		
Carcass	2 542	2 571	2 409	2 427	48.2	0.597
Legs meat	553	523	526	521	10.9	0.757
Breast meat	807	825	741	742	19.1	0.483
Abdominal fat	35.8	38.1	32.5	33.9	1.68	0.654

C = a control group; R = a group was fed medicated starter and grower diets with coccidiostat robenidine; W3 = a group was fed diets containing 3% of WW meal in feed; W6 = a group was fed diets containing 6% of WW meal in feed

Carcass and minerals in meat

The average weight of carcass, thigh muscle, breast muscle and abdominal fat was not affected by the diets assessed in this study ($P > 0.05$; Table 6). This finding is in overall agreement with the results of the study conducted by Zapletal et al. (2024) on female broilers. In addition, Wang et al. (2024) found that the fermented *Artemisia argyi*, a different *Artemisia* species, positively affected carcass traits of broilers, and dietary supplementation with 3% of fermented *Artemisia argyi* was determined to be the most beneficial in this regard.

The effect of dietary WW on the content of minerals in chicken meat has not yet been studied in the available literature. In the case of the mineral macronutrient levels of breast meat in this study, a dietary effect was found only on the Ca level ($P < 0.05$;

Table 7. Minerals content (mg/100 g) of breast meat in relation to diet (on basis of fresh meat)

Trait	Diet				SEM	*P*-value
	C	R	W3	W6		
P	207	204	205	207	3.1	0.986
Ca	12.9^{ab}	11.1^{b}	15.6^{a}	14.5^{a,b}	0.54	0.017
K	364	364	364	362	2.8	0.991
Na	57.4	67.4	59.1	56.5	1.82	0.128
Mg	25.1	25.3	24.8	24.6	0.26	0.841
Cu	0.023^{B}	0.038^{A,a}	0.025^{AB,b}	0.024^{AB,b}	0.002	0.005
Fe	1.10	1.11	0.841	0.664	0.086	0.186
Mn	0.081	0.104	0.111	0.072	0.015	0.773
Zn	0.600	0.597	0.584	0.638	0.010	0.166

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

C = a control group; R = a group was fed medicated starter and grower diets with coccidiostat robenidine; W3 = a group was fed diets containing 3% of WW meal in feed; W6 = a group was fed diets containing 6% of WW meal in feed

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Table 7); the levels of P, K, Na and Mg did not differ among the dietary groups assessed ($P > 0.05$). Meat of chickens fed the W3 diet was found to have a significantly higher Ca level compared to the R group ($P < 0.05$). A non-significantly higher Ca level was also observed in the W6 group than in the R and C groups ($P > 0.05$).

As for the mineral microelements in the breast meat of broilers in this study, a significant dietary effect was found only for the Cu level ($P < 0.01$; Table 7); Fe, Mn and Zn levels did not differ among the dietary groups assessed ($P > 0.05$). The Cu level was highest in the R group, and it was significantly higher in comparison with both the C group ($P < 0.01$) and the W3 and W6 groups ($P < 0.05$). Huang et al. (2022) found that infecting chickens with *Eimeria tenela* impaired Cu and Zn levels in their bodies, specifically reducing their levels in liver and serum, whereas the administration of the anticoccidial agent berberine to *Eimeria*-infested chickens led to a significant increase in the serum Cu level. These authors suggest that administration of the above mentioned anticoccidial agent aided in maintaining Cu homeostasis in the chicken body through the mRNA expression of their transport genes. This fact could be somewhat related to the finding of this study, where the highest meat Cu level was found in broilers fed the starter and grower feed with the coccidiostat robedinine, which successfully prevented the development of oocysts until the age of 32 days in broilers and may have contributed to the higher Cu deposition in their meat later on. In addition, Huang et al. (2022) add that the administration of Cu and Zn would have a positive effect on resistance of chickens to coccidiosis.

CONCLUSION

Dietary inclusion of WW meal throughout the fattening period had no negative effect on the performance and carcass traits of broilers. However, contrary to our expectation, the chosen levels of dietary administered WW were not effective in reducing the number of *Eimeria* oocysts shed in infested broilers. On the basis of the results obtained, it can be concluded that for the coccidiosis control in poultry production, it is advisable to monitor the content of specific bioactive substances with proven anticoccidial activity when using WW products. Moreover, dietary administration of coccidiostat robenidine prevented the development of *Eimeria* oocysts in the broiler intestine for 4 days after the cessation of feeding the medicated grower diet. Besides that, the higher copper content in breast meat of broilers fed medicated starter and grower diets suggests that the coccidiostat robenidine may markedly interfere with copper metabolism in *Eimeria*-challenged chickens.

Conflict of interest

The authors declare no conflict of interest.

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