

THE EFFECT OF NETTLE FLOUR (*URTICA DIOICA*) IN DIETS FOR BROILER CHICKENS ON PRODUCTIVE PERFORMANCE, LIPID QUALITY AND OXIDATIVE STABILITY OF MEAT

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RESEARCH ARTICLE

Abstract

This experiment was conducted to evaluate the effects of supplementing the diet of broiler chicks with stinging nettle (*Urtica dioica* L) meal on performance, carcass traits, fatty acid (FA) profile and meat cholesterol content and meat antioxidant status. A completely randomized design was used with 120 broilers (Ross-308) divided into two treatments and four replicates (with 15 birds in each replicate) from 1 to 42 days. The treatment groups consisted of: the control group without nettle meal supplements (C) and the experimental group in which the feed was supplemented with nettle meal at a level of 3% (U3). Supplementing the diet with nettle meal at a level of 3% had a negative effect on the productive performance of broilers, the final weight being lower ($p < 0.05$) than in chickens fed with the standard diet. The results showed that the use of nettle flour in feed had significant effects on the carcass traits of broilers ($p < 0.05$). The highest percentage of breast, liver and gizzard (36.42%; 3.49% and 1.64%) and the lowest proportion of abdominal fat (2.95%) were observed in group U3. In addition, the introduction of nettle meal in the feed of broiler chickens led to an increase in the protein and fat content of the breast and a significant reduction in cholesterol levels. The FA profile of breast fat improved by increasing the proportion of FA considered beneficial to human health (C18:3 n-3 and C18:2 n-6). The content of breast meat in lipophilic antioxidants (tocopherols, lutein and zeaxanthin) and the total antioxidant capacity (TAC) measured by the ABTS method (mM Trolox equivalents (TE)/g) were improved by the introduction of nettle meal in chicken feed. The overall results showed that the introduction of 3% nettle meal in the feed of broilers, even though it reduced the final weight of the chickens, had positive effects on the carcass traits, nutritional content and oxidative stability of the meat.

Keywords: fatty acid profile, cholesterol, lipophil antioxidants, antioxidant capacity.

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INTRODUCTION

Recently, there has been an increase in the demands of consumers regarding the quality of agri-food products of animal origin, which must not only be a source of nutrients for human food, but must also provide the consumer with an important intake of bioactive compounds with a positive effect on human health. In this context, nutritionists have paid special attention to medicinal plants, which used in animal feed can determine a significant improvement in the nutritional and sanogenic quality of meat (Mierlita, 2022).

Medicinal plants are accepted in animal feed as possible substitutes for feed antibiotics, being accepted by consumers because they are considered natural sources of bioactive compounds with the effect of promoting animal

production as well as antioxidant effects (Mousavi et al., 2017; Kheiri and et al., 2018; Moula et al., 2019). One of these medicinal plants is nettle (*Urtica dioica* L) (Nasiri et al., 2011; Sharma et al., 2018), which has been less studied in terms of its use in broiler chicken feed and its effect on meat production and quality (Nasiri et al., 2011; Moula et al., 2019).

Nettle (*Urtica dioica* L., family *Urticaceae*) is a wild herbaceous perennial plant, frequently found in the spontaneous flora of our country, being characterized by a high content of nutrients (vitamin C, carotenoids, minerals), as well as tannins, formic acid, salicylic acid, thymol and carvacrol (Gülçin et al., 2004). Nettles have antioxidant, antimicrobial, antifungal and antiviral properties (Upton, 2013). Rutto et al., (2013) and Said et al., (2015) concluded that nettle leaves are rich in proteins, fats, carbohydrates, vitamins, minerals and trace elements (mainly Fe). The protein content

(22 - 30% of DM) and amino acid profile is clearly higher than other leafy plants (Rutto et al., 2013), even than alfalfa flour.

The use of nettle meal in the feed of broilers had positive effects on performance, carcass traits and on biochemical and immune blood parameters (Nasiri et al., 2011; Hashemi et al., 2018). In one experiment, the addition of 2% nettle to the diet of broilers increased their body weight (Kwiecien and Mieczan, 2009). In pigs, the use of nettle extract had positive effects on meat quality, improving oxidative stability and polyunsaturated/saturated fatty acid ratio (Hanczakowska et al. 2007). Research by Avci et al., (2006) demonstrated that mice fed a diet supplemented with nettle meal showed significant reductions in triglycerides and total cholesterol content in blood plasma. Also Grigorova et al. (2022) demonstrated that supplementing the diet of laying hens with nettle (0.3-0.5% of diet) determined a significant improvement in yolk pigmentation, a decrease in the cholesterol concentration in the yolk as well as in blood glucose and total serum cholesterol content.

The aim of this study was to investigate the effects of supplementing the diet of broiler chicks with stinging nettle (*Urtica dioica* L) meal on performance, carcass traits, fatty acid (FA) profile and meat cholesterol content and meat antioxidant status.

MATERIAL AND METHOD

Preparation of nettle flour (Urtica dioica)

The nettle plants were harvested from the spontaneous flora in May 2023. Drying of the plants was done in the shade to prevent the loss of vitamins and other volatile nutrients. To prevent the nettle plants from moulding, they were turned daily from one side to the other. After drying, the nettle plants were ground with a laboratory grinder PC-KSW 1021 N (Clatronic, Ost 40, Germany). Nettle meal was introduced into the structure of diets for broilers in the proportion of 3.0% (% of feed weight) by replacing soybean meal and corn.

Chickens, diets and their management

This experiment was carried out in a completely randomized design using a total of 120 broiler chickens (Ross-308) allocated to 2 treatments and 4 replicates (with 10 birds in each pen) from 1 to 42 days and included (Table 1). The treatment groups consisted of: the control group without nettle meal supplement (C) and the experimental group in which the feed was supplemented with nettle meal at a level of 3% (U3).

Table 1.

Experimental design of the trial of supplementing the diet of broiler chicks with nettle (*Urtica dioica* L) meal.

Diets (treatment)	Inclusion rate of nettle meal (% of diet)			Replicates	Chickens per replicate	Total chickens per treatment
	Starter	Grower	Finisher			
Control (C)	0	0	0	4	15	60
Experimental (U3)	3.00	3.00	3.00	4	15	60
Total	-	-	-	8	-	120

Diets were formulated based on NRC (1994) recommendations for energy and nutrient content (Table 2).

Diets and water were provided ad libitum. The lighting schedule during the experimental period consisted of a period of 23 h light and 1 h dark throughout the experimental period. The medium temperature was gradually decreased from 33°C to 25°C on day 21 and then kept constant. On day 42, two birds per replicate were randomly selected and slaughtered, and live weight, carcass weight, thigh muscle, breast muscle, liver, gizzard fat and percentage of carcass parts to carcass weight were determined gravimetrically. Breast samples were taken to determine the

proximate chemical composition, antioxidant compounds, FA profile, cholesterol concentration and antioxidant activity. Until chemical analyses, the samples were stored in plastic zipper bags at -20°C.

Chemical Analysis

Nettle flour samples for laboratory analyzes were taken in compliance with the SR EN ISO 6498:2012 standard. Thus, three laboratory samples were constituted which were kept in jars with rubbed stoppers until the chemical analyses.

The crude chemical composition of nettle flour was performed in accordance with the AOAC International (2005) technique,

determining: dry matter (DM) (Method 934.01); crude ash by calcination (Met. 942.05); crude protein by the Kjeldahl method (Met. 954.01); crude fat (ether extract - EE) by the Soxhlet method (Met. 920.39); crude cellulose by acid hydrolysis followed by basic hydrolysis of the sample (Met. 978.10) and organic matter by difference (OM = 100 - crude ash).

Lutein and zeaxanthin were determined by the HPLC technique, using a Perkin Elmer

200 equipped with a UV detector (445 nm). Tocopherols were determined using a Finnigan Surveyor Plus HPLC equipped with a PDA-UV detector at a wavelength of 292 nm.

Total antioxidant capacity (TAC) was measured by the phosphomolybdenum method at an absorbance of 695 nm. Results were expressed as mM Trolox equivalents (TE)/g breast. All samples were analyzed in triplicate.

Table 2

Ingredient composition and nutritive value of the diets (% as fed-basis).	Starter (1-10 d)		Grower (11-35 d)		Finisher (36-42 d)	
	C	U3	C	U3	C	U3
	Ingredients(%)					
Maize	48.43	45.66	48.76	46.50	52.56	50.27
Wheat	8.00	8.00	10.00	10.00	10.00	10.00
Soybean meal	37.00	36.10	33.80	32.60	29.80	28.60
Nettle meal (<i>Urtica dioica</i>)	-	3.00	-	3.00	-	3.00
Sunflower oil	2.80	3.40	4.00	4.40	4.50	4.80
Ground limestone	1.14	1.14	1.15	1.15	1.20	1.20
Dicalcium phosphate	1.30	1.30	1.20	1.20	1.14	1.14
Salt	0.30	0.30	0.30	0.30	0.30	0.30
L-Lysine-HCl	0.30	0.34	0.18	0.21	-	0.08
DL-Methionine	0.23	0.26	0.11	0.14	-	0.11
Vitamin-mineral premix	0.50	0.50	0.50	0.50	0.50	0.50
Calculated nutrients						
Metabolizable Energy, kcal/kg	3000.0	3012.0	3119	3106	3150	3164
Crude protein (%)	22.00	22.06	20.52	20.46	19.00	18.97
Lysine (%)	1.30	1.30	1.15	1.15	1.05	1.05
Methionine (%)	0.55	0.55	0.52	0.52	0.48	0.48
Ca (%)	0.90	0.90	0.85	0.85	0.82	0.82
P (%)	0.42	0.42	0.40	0.40	0.37	0.37

Statistical Analysis

Data obtained were subjected to ANOVA using the General Linear Model (GLM) procedure of SAS (2005). Significant differences among treatment means were evaluated using Tukey's multiple range tests. Statements of statistical significance are based on $P < 0.05$.

RESULTS AND DISCUSSIONS

Nutrient and energy contents of stinging nettle meal and treatment diets

Nettle flour (*Urtica dioica*) is characterized by a relatively high protein content, which is on average 24.48% (% of DM) with variation limits between 21.37% and 27.51% respectively (table 3). The fat content is relatively low (1.52 - 3.72%) but it has a high crude cellulose content (9.88 - 15.63%), which could limit the use of nettle meal in large quantities in broiler feed, especially in starter feed (Mierlita, 2022).

Table 3

Chemical composition of nettle powder (% of DM)			
Items	Mean	Min.	Max.
Dry matter (DM)	88.82	87.45	90.83
Organic matter (OM)	86.58	82.79	90.30
Crude protein (CP)	24.48	21.37	27.51
Crude fat (EE)	2.87	1.52	3.72
Crude fibre (CF)	13.32	9.88	15.63
Gross energy (kcal/kg)	3257	3018	3503
Crude ash	13.42	9.70	17.21

The CP content of nettle flour found in the current study was comparable to the values reported by Cross (2007), Tozer (2007), and Mierlita (2022), respectively, but lower than those found by Bekele et al. (2015) and Liu (2007) who reported CP levels of 26.13% and 29.40% respectively. Bekele et al. (2015) reported an EE content of nettle flour twice as high as the value we found in this study (5.8% vs. 2.87%), probably due to the cultivar and time of harvesting nettles.

Effect of dietary treatment on growth performance and carcass traits

The effects of supplementing the diet with nettle meal on the performance of broilers are summarized in Tables 4 and 5. The use of nettle meal in broiler feed significantly reduced ($p < 0.05$) final body mass and average daily

weight gain compared to the diet standard. However, no significant effects were observed on feed intake and feed conversion in broilers ($p > 0.05$). The results obtained in this study are not in agreement with those previously reported by Nasiri et al. (2011) and Kwiecien and Mieczan (2009), who demonstrated that the addition of 1.5% and 2% nettle meal respectively in the diet of broilers did not affect the productive performance of the chickens or even had positive effects on their weight gain. This difference between the results of the present experiment and those reported above may be the result of different causes, such as the different level of nettle meal in the chicken feed, nettle variety or farm management.

Table 4

The effect of nettle meal (*Urtica dioica* L) supplementation of broiler chicks' diet on the evolution of body mass and average daily weight gain.

	Body weight (BW)				Body weight gain (BWG)			
	C	U3	SEM	p-value	C	U3	SEM	p-value
1 d	42.31	41.93	0.093	0.521	-	-	-	-
1-10 d	486.33	481.37	3.965	0.614	44.40	43.94	0.108	0.289
11-28 d	1559.57	1516.32	22.73	0.048	59.62	57.49	0.859	0.042
29-35 d	2203.81	2156.31	19.86	0.077	92.03	91.43	1.631	0.069
36-42 d	3021.76	2942.16	23.58	0.036	116.85	112.26	1.914	0.048
1-42 d	3021.76	2942.16	23.58	0.036	70.94	69.06	1.321	0.048

Table 5

The effect of nettle meal (*Urtica dioica* L) supplementation of broiler chicks' diet on the feed intake (FI) and feed conversion ratio (FCR).

	Daily feed intake (g/chick)		FCR (kg feed/kg BWG)	
	C	U3	C	U3
1-10 d	43.60	44.12	1.082	1.104
11-28 d	72.37	69.96	1.314	1.317
29-35 d	145.40	146.75	1.681	1.708
36-42 d	206.12	204.65	1.764	1.823
1-42 d	99.98	99.05	1.609	1.634

The effects of the introduction of nettle meal in the feed on the carcass traits of broilers are summarized in Table 6. It can be seen that the highest breast percentage (36.42%) was observed in the U3 group that was additionally fed with nettle meal. The presence of antioxidants and phenolic substance in nettle may be the main reason for the improvement of breast percentage of broiler carcasses (Nasiri et al., 2011). Harmful bacteria present in the gastrointestinal tract of chickens can cause the breakdown of amino acids and

therefore reduce their absorption, as the antimicrobial substances present in nettle can reduce the harmful bacterial populations in the gastrointestinal tract and improve the levels of absorbed amino acids (Lee et al., 2003 ; Gülçin et al., 2004; Nasiri et al., 2011; Mierlita, 2022). In addition, nettle meal has been shown to stimulate pancreatic secretions (Lee et al. 2003) increasing the digestibility and availability of feed nutrients, and therefore improving carcass traits. also, increasing the percentage of liver by using nettle meal can

have positive effects by increasing bile secretion on nutrient digestion. With increased amounts of amino acids absorbed, organs such as the breast and thigh attract more growth. The lowest carcasses and breast percentages were recorded in the control group. The results

obtained in this study regarding carcass traits are in agreement with the study of Modiry et al. (2010), Nobakht et al. (2010) and who reported that the presence of nettle meal in the feed improved the carcass traits of broilers.

Table 6

The effect of nettle meal (*Urtica dioica* L) supplementation of broiler chicks' diet on carcass development

Carcass traits	C		U3		SEM	p
	g	%	g	%		
Carcass		72.14		72.81	0.785	0.471
Abdominal fat		4.28		2.95	0.037	0.025
Thigh muscle		24.83		25.29	0.614	0.048
Breast muscle		33.80		36.42	0.672	0.084
Liver		2.87		3.49	0.176	0.038
Gizzard		1.48		1.64	0.731	0.047

Effect of Dietary Treatments on Nutritional Composition of Breast Muscle

The results regarding the nutritional composition of the breast muscle samples are presented in Table 7. The highest concentration of crude protein ($p < 0.001$) and fat ($p < 0.05$) was found in the chickens fed additionally with nettle meal (group U3). This result is beneficial because a higher protein content meets the demands of consumers who need high-quality and easily degradable proteins.

Table 7

Nutritional composition of broiler breast muscle (%)

	Treatments		SEM	p-value
	C	U3		
Dry matter	26.06	28.46	0.417	< 0.001
Crude protein	21.94	23.93	0.138	< 0.001
Crude fat	2.28	2.56	0.031	0.047
Ash	1.26	1.29	0.013	0.078

The concentration of cholesterol in the fat of the breast meat samples was lower in chickens fed additional nettle meal compared to those fed the standard diet (fig. 1). We believe that the high concentrations of flavonoids in nettle flour, through the strong antioxidant activity of flavonoids, contribute to the regulation of lipid metabolism and the modulation of cholesterol concentration in meat. However, few studies have been done on the effects of some medicinal plants used in poultry diet on cholesterol concentration in chicken meat (Vlaicu et al., 2021).

Addition of nettle meal to the broiler diet affected the fatty acid profile of breast fat (Fig. 2). Compared to the control group, broilers fed the nettle meal diet had a higher meat content

of linoleic acid (LA, C18:2 n-6) and α -linolenic acid (ALA, C18 :3n-3).

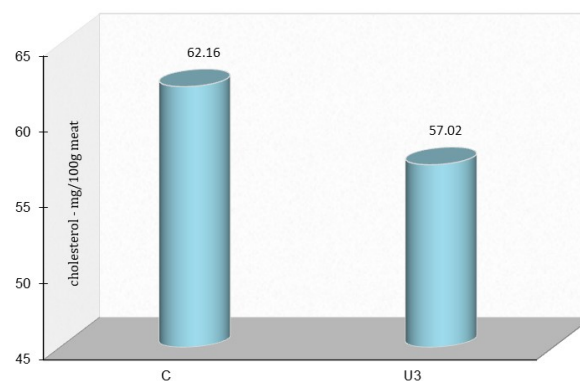


Figure. 1. **Effect of nettle meal (*Urtica dioica* L) supplement in broiler chicken diet on breast meat cholesterol content.**

Since the basic ingredients of the diet used in this study were corn and soybean meal, which are generally rich in oleic (C18:1 n-9) and linoleic (C18:2 n-6) acids, these FAs were the more abundant acids in the analyzed meat samples (Stanisic et al., 2023). Previous research reported that nettle leaves contain PUFA (polyunsaturated FA), of which α -linolenic acid is the most abundant, followed by palmitic and linoleic acids (Rutto et al., 2013), which supports the results of this study. In addition, the results of this study are in agreement with previous reports by Dukic Stojcic et al., (2016), who concluded that supplementing broiler diets with nettle meal increases the n-3 PUFA of breast muscle. On the other hand, these results were not confirmed in the studies carried out by Stanisic et al. (2023).

As shown in Figure 3, nettle meal supplementation increased the total

tocopherols, lutein and zeaxanthin content of breast meat in group U3 compared to group C. Also, total antioxidant capacity (TAC) was higher in group U3 compared to group C.

These results show that the antioxidant action of breast meat can be increased by supplementing the broiler diet with nettle meal. Nettle meal contains various compounds, mostly polyphenols that have antioxidant

activities, so it can improve the quality of broiler meat. Some authors (Franz et al., 2010; Loetscher et al., 2013; Ahmed et al., 2018), reported that some medicinal plants (rosemary, sage, oregano and thyme) have the potential to improve the oxidative stability of beef broiler because of their antioxidant properties.

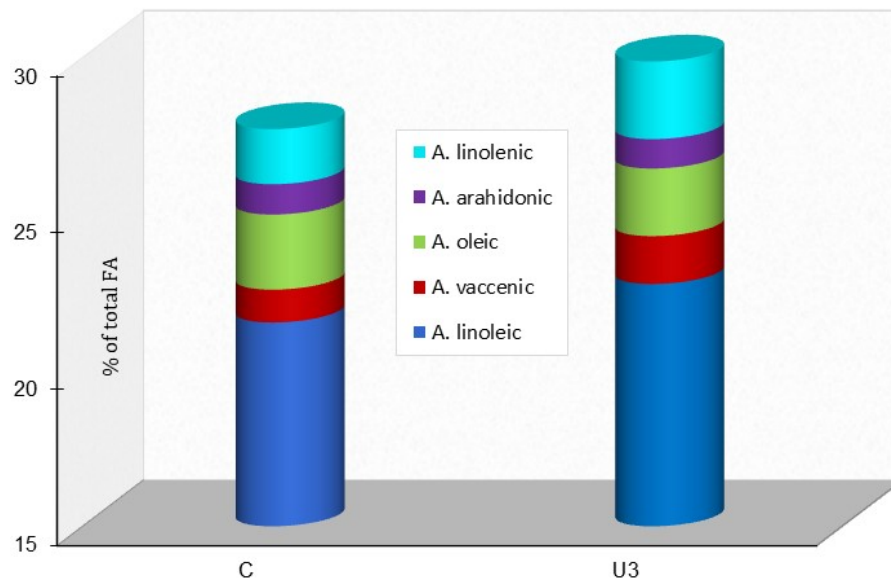


Figure 2. Effect of nettle meal (*Urtica dioica* L) supplement in the broiler chicken diet on the content of major FA in breast meat.

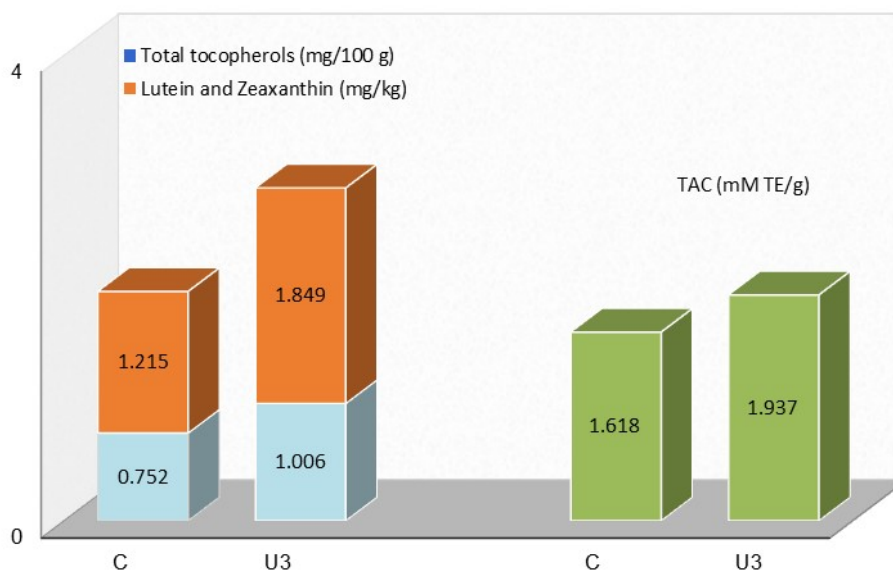


Figure 3. Effect of stinging nettle meal (*Urtica dioica* L) supplement in the diet of broiler chicks on the antioxidant content and antioxidant activity of breast meat.

CONCLUSIONS

Supplementing the diet with nettle meal at a level of 3% had a negative effect on the productive performance of broilers, the final weight being lower ($p < 0.05$) than in chickens fed with the standard diet. The results showed that the use of nettle flour in feed had significant effects on the carcass traits of broilers ($p < 0.05$). The highest percentage of breast, liver and gizzard (36.42%; 3.49% and 1.64%) and the lowest proportion of abdominal fat (2.95%) were observed in group U3. In addition, the introduction of nettle meal in the feed of broiler chickens led to an increase in the protein and fat content of the breast and a significant reduction in cholesterol levels. The FA profile of breast fat improved by increasing the proportion of FA considered beneficial to human health (C18:3 n-3 and C18:2 n-6). The content of breast meat in lipophilic antioxidants (tocopherols, lutein and zeaxanthin) and the total antioxidant capacity (TAC) measured by the ABTS method (mM Trolox equivalents (TE)/g) were improved by the introduction of nettle meal in chicken feed. The overall results showed that the introduction of 3% nettle meal in the feed of broilers, even though it reduced the final weight of the chickens, had positive effects on the carcass traits, nutritional content and oxidative stability of the meat.

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