UNIVERSITY OF KRAGUJEVAC

FACULTY OF AGRONOMY ČAČAK

SYMBIOTECH

2nd INTERNATIONAL SYMPOSIUM ON BIOTECHNOLOGY

14–15 March 2024

Faculty of Agronomy in Čačak, University of Kragujevac, Serbia

- PROCEEDINGS -

2nd INTERNATIONAL SYMPOSIUM ON BIOTECHNOLOGY

XXIX Savetovanje o biotehnologiji sa međunarodnim učešćem

- PROCEEDINGS -

ORGANIZER AND PUBLISHER

University of Kragujevac, Serbia
Faculty of Agronomy in Čačak

Honorary Committee

Prof. Dr. Marina Pintar, Slovenia; Prof. Dr. Andrej Bončina, Slovenia; Prof. Dr. Branko Kramberger, Slovenia; Prof. Dr. Tomaž Langerholc, Slovenia; Prof. Dr. Ivica Kisić, Croatia; Dr. Kristina Kljak, Croatia; Prof. Dr. Krunoslav Zmaić, Croatia; Prof. Dr. Zvonko Antunović, Croatia; Prof. Dr. Muhamed Brka, B&H; Prof. Dr. Enisa Omanović-Mikličanin, B&H; Prof. Dr. Ivan Ostojić, B&H; Dr. Adrijana Filipović, B&H; Prof. Dr. Božidarka Marković, Montenegro; Prof. Dr. Sanja Radonjić, Montenegro; Prof. Dr. Vjekoslav Tanaskovik, North Macedonia; Prof. Dr. Ivana Janeska-Stamenkoska, North Macedonia; Prof. Dr. Zlatan Kovačević, B&H; Prof. Dr. Željko Vaško, B&H; Prof. Dr. Dragutin Đukić, Serbia; Prof. Dr. Nedeljko Tica, Serbia; Prof. Dr. Branko Ćupina, Serbia; Prof. Dr. Dušan Živković, Serbia; Prof. Dr. Vladan Bogdanović, Serbia; Dr. Darko Jevremović, Serbia; Dr. Marijana Pešaković, Serbia; Prof. Dr. Cosmin Salasan, Romania;

Technical editors

Prof. Dr Vladimir Kurćubić; Prof. Dr Pavle Mašković; Dr Marija Gavrilović; Dušan Marković, BSc

Print-run: 30

Printed by
MEDIGRAF - Čačak, Aleksandra Savića 42, 32000 Čačak

ISBN 978-86-87611-91-7

Year of publication: 2024

DOI: 10.46793/SBT29.29VD

THE EFFECT OF PROTEASE ENZYME ON THE MEAT CLASSES OF TWO BROILER CHICKEN HYBRIDS

Vladimir Doskovic[1], Snežana Bogosavljević-Bošković[1], Zdenka Škrbić[2], Miloš Lukić[2],Simeon Rakonjac[1], Veselin Petričević[2], Dejan Beuković[3]

Abstract: The aim of the study was to evaluate the effect of three protein levels (with or without protease enzymes) on the weight and percentage yield of each meat class in two genotypes of broiler chickens (fast-growing hybrid Cobb 500 and medium-growing hybrid Master Gris). Each genotype was divided into a control group (C group) fed with a commercial feed mixture and an experimental group fed with a low protein diet of 4% (E-I group) or 6% (E-II group) in relation to the control group, with the addition of 200 mg/kg Ronozyme ProAct (E-I group) or 300mg/kg Ronozyme ProAct (E-II group).

Weight and percentage yield of the meat classes were influenced by the genotype. The fast-growing hybrid Cobb 500 had higher masses of all meat classes and a higher percentage of class I meat, while the percentage of class II and III meat was lower than in the medium-growing hybrid Master Gris ($P<0.05$). Feeding had no effect on the weight and percentage yield of the individual meat classes in either hybrid ($P>0.05$).

Keywords: hybrids of chickens, protease enzyme, meat classes.

Introduction

The quality of poultry meat is the result of complex interactions between genotype, age and sex of the birds and the managment system (Uhlířová et al., 2018). It is well known that nutrition is one of the key factors influencing the quantitative and qualitative characteristics of poultry meat and the profitability of production.

Nowadays, soybeans are the main source of protein for broiler diets (Tavaniello et al., 2022). However, soybean meal contains anti-nutritional factors such as trypsin inhibitors that reduce nutrient availability and limit the amount of soybean meal that can be included in animal feed (Park et al., 2020). The addition

[1]University of Kragujevac, Faculty of Agronomy, Cara Dušana 34, Čačak, Serbia (vladosko@kg.ac.rs)

[2] Institute for Animal Husbandry, Autoput 16, Poštanski fah 23, 11 080 Belgrade-Zemun, Serbia

[3] University of Novi Sad, Faculty of Agriculture Novi Sad, Trg Dositeja Obradovića 8, Serbia

of enzymes to broiler feed is known for its economic, environmental and nutritional benefits (Jabbar et al., 2021). The addition of exogenous protease improves growth and increases the digestibility of crude protein and metabolizable energy in broiler diets while improving the utilization of amino acids as it cleaves antinutritive factors such as trypsin inhibitor (Ndazigaruye et al., 2019).

Medium-growing broiler genotypes are more resistant to heat stress compared to conventional fast-growing hybrids (Pietrzak et al., 2020), and their meat is also interesting on the market (Devatkal et al., 2019). Research on the use of protease enzymes in feed for broiler chickens has mainly been conducted on fast-growing genotypes (Dosković et al., 2023b; Hafeez et al., 2021; Jabbar et al., 2021; Park et al., 2020;…). However, limited information is available on the effects of different protein levels using protease enzymes on the meat quality of medium-growing chickens kept in an intensive rearing system. The aim of the study was therefore to evaluate the effects of three protein levels (with or without protease enzymes) on the meat quality of fast- and medium-growing chickens, including the interactions between the two factors.

Materials and methods

Two genotypes were used for the experiment: the fast-growing hybrid Cobb 500 and the medium-growing hybrid Master Gris. Each genotype was divided into a control group (100 chickens, group C), which was fed a standard commercial feed mixture, and an experimental group, which received 4 % (100 chickens, group E-I) or 6 % (100 chickens, group E-II) low-protein feed with the addition of 200 mg/kg Ronozyme ProAct (group E-I) or 300 mg/kg Ronozyme ProAct (group E-II) in relation to the control group. The control groups received mixtures with 22% crude protein in the starter phase (0 - 21 days), 19% crude protein in the grower phase (22 - 35 days) and 17% crude protein in the finisher phase (36 - 63 days).

All birds were reared under environmental conditions that meet the requirements for broilers. Feed and water were available to the chickens *ad libitum*.

At the end of the experiment, at 63 days of age, 20 chickens per group (sex ratio 1:1) were selected and slaughtered for meat analysis.

After the carcasses had cooled, they were cut into their basic components: breast, drumsticks, thighs, wings, back and pelvis. These primal cuts were divided into meat class I (breast, drumsticks and thighs), meat class II (wings)

and meat class III (back and plevis). These meat classes were measured and, based on the weight of the meat class and the weight of the ready-to-grill carcass, the proportions of the meat class in the dressed carcass were calculated.

The results were analyzed by Stat Soft Inc Statistica For Windows (Version 7.0., 2006) program. Two-factor (diet treatments and hybrids) analysis of variance and LSD test to compare the treatment means was applied ($P<0.05$).

Results and discussion

The results of the weight of certain classes of meat are presented in Table 1.

Table 1. Weight of different classes of chicken meat on the 63th day of fattening, g

Treatment			Class I (breast, thighs, drumsticks)	Class II (wings)	Class III (back, pelvis)
Hybrids	Groups				
Cobb 500	C	$\bar{X}$	1874.90^{a}	297.39^{ab}	643.74^{a}
		Sd	276.86	46.03	74.74
	E-I	$\bar{X}$	1880.37^{a}	302.24^{a}	642.00^{a}
		Sd	266.21	36.30	80.70
	E-II	$\bar{X}$	1798.79^{a}	297.79^{ab}	625.43^{a}
		Sd	203.18	34.49	71.41
Master Gris	C	$\bar{X}$	1493.27^{b}	277.22^{b}	576.76^{b}
		Sd	197.58	41.05	73.88
	E-I	$\bar{X}$	1454.60^{b}	276.87^{b}	570.66^{b}
		Sd	180.93	35.27	71.88
	E-II	$\bar{X}$	1437.49^{b}	274.41^{b}	550.73^{b}
		Sd	182.71	35.93	67.13

$\bar{X}$-Average, Sd - Standard deviation
Different superscripts (a, b) indicate a significant differences between groups ($P<0.05$)

From the data analysis, it can be concluded that there are differences in the weight of certain meat classes between the studied genotypes, so that the fast-growing Cobb 500 chickens had a higher weight in all meat classes than the medium-sized Master Gris chickens ($P<0.05$). The chickens from the feed treatments of both hybrids had similar weights in the meat classes, from which we conclude that the formulations used in the complete mixtures for broilers had no effect on the weight of the different meat classes ($P>0.05$). The fact that the reduced crude protein content with or without the enzyme protease has no effect on the composition of the carcass is consistent with the observations of Dosković et al. (2023a), Chodová et al. (2021), Ndazigaruye et al. (2019).

The percentages of certain chicken meat classes in the dressed carcass of slaughtered chickens are shown in Table 2.

Table 2. The percentage of the different chicken meat classes in the dressed carcass on 63 day of fattening, %

Treatment			Class I (breast, thighs, drumsticks)	Class II (wings)	Class III (back, pelvis)
Hybrids	Groups				
Cobb 500	C	$\bar{X}$	63.80[a]	10.14[b]	22.03[b]
		Sd	2.42	0.89	1.50
	E-I	$\bar{X}$	64.07[a]	10.35[b]	21.94[b]
		Sd	2.26	0.71	1.23
	E-II	$\bar{X}$	63.40[a]	10.51[b]	22.06[b]
		Sd	1.87	0.80	1.20
Master Gris	C	$\bar{X}$	60.97[b]	11.30[a]	23.57[a]
		Sd	0.97	0.55	0.73
	E-I	$\bar{X}$	60.46[b]	11.51[a]	23.73[a]
		Sd	0.98	0.53	0.75
	E-II	$\bar{X}$	60.76[b]	11.60[a]	23.30[a]
		Sd	1.19	0.46	0.83

$\bar{X}$ - Average, Sd - Standard deviation
Different superscripts (a, b) indicate a significant difference between groups (P<0.05)

The genotype had a significant effect on the investigated carcass traits (P<0.05). Compared to Cobb 500 chickens, Master Gris chickens had a lower proportion of breast, drumstics and thighs, i.e. class I meat, and a slightly higher proportion of wings (class II meat) and back and plevis (class III meat) (P<0.05). Kreuzer et al. (2020) state that the genotype has the greatest influence on the composition of the carcass, while Chodová et al. (2021) point out that fast-growing chickens have the highest proportion of breast (P<0.001), while medium-growing chickens are in the middle between fast and slow-growing genotypes.

The addition of exogenous protease to the feed at a concentration of 200 mg/kg (E-I group) or 300 mg/kg feed (E-II group) with a reduction in crude protein content of 4 or 6 % compared to the control feed (C group) had no significant effect on the proportion of all meat classes (P>0.05). There were also no significant interactions observed between the diet and the genotype on the percentage of all meat classes (P>0.05). Duque-Ramírez et al. (2023) also found that there were no significant differences in the inclusion of proteases in the diet (P>0.05) in relation to the partial yield of carcass cuts of meat classes I and II 42 days of fattening. Dosković et al. (2023a) found that only in female chickens were there differences in the

percentage yield of class I and III meat between the E-I (200mg Ronozyme ProAct/kg feed) and E-II (300mg Ronozyme ProAct/kg feed) feeding treatments ($P<0.05$) in chickens aged 49 days, while there were no differences between the standard feeding treatment (C group) and the E-I group or E-II group in the carcass quality parameters examined ($P<0.05$).

Conclusion

The present study focused on the comparison of carcass quality parameters such as mass and percentage of each meat class of medium growth genotypes - Master Gris (reared in an intensive feeding system, characteristic of commercial fast growing chickens) with the hybrid Cobb 500. In addition, the application of protease enzymes with the reduction of crude protein content in two hybrids with different growth intensity and their effect on these chicken carcass quality parameters was analyzed.

The results showed that the genotypes studied differed in weight and percentage yields of the meat classes. The fast-growing hybrid Cobb 500 had higher masses of all meat classes and a higher percentage of class I meat, while the percentage of class II and III meat was lower than in the medium-growing hybrid Master Gris ($P<0.05$). For both hybrids, the feed had no significant effect on the weight and percentage yield of the individual meat classes ($P>0.05$). The proposed feed formulations for broilers, which provided for a 4% reduction in crude protein content, i.e. 6% at each fattening stage (starter, grower, finisher) with the addition of 200 mg/kg feed or 300 mg/kg feed protease enzymes (Ronozyme ProAct) compared to the standard feed, did not cause any changes in the weight and percentage yield of the meat classes ($P>0.05$).

Acknowledgement

This study was funded by the Ministry of Science, Technological Development and Innovation of Republic of Serbia, No. 451-03-66/2024-03/200088 and 451-03-66/2024-03/ 200022.

References

Chodová D., Tůmová E., Ketta M. (2021). The response of fast-, medium- and slow-growing chickens to a low protein diet. Czech Journal of Animal Science 66 (3), 97-105.

Devatkal S.K., Naveena B.M., Kotaiah T. (2019). Quality, composition, and consumer evaluation of meat from slow-growing broilers relative to commercial broilers. Poultry Science 98, 6177-6186.

Dosković V., Bogosavljević-Bošković S., Škrbić Z., Milošević B., Lukić M., Rakonjac S., Petričević V. (2023a): Effect of protease added in food and sex on chicken meat clasess. 1st International Symposium on Biotechnolgy, Čačak 17-18 March, Proceedings: 223-229, doi: 10.46793/SBT28.223D

Dosković V., Bogosavljević-Bošković S., Škrbić Z., Lukić M., Milošević B., Rakonjac S., Petričević V., Bjedov S. (2023b). The effects of protease and sex on tissue composition in major primal cuts of broilers. Contemporary Agriculture 72 (4), 165-169, doi: 10.2478/Contagri-2023-0021.

Duque-Ramírez C.F., Javierre J.A., Peñuela-Sierra L.M., Diaz-Vargas M. (2023). Effect of exogenus protease on performance, nutrient digestibility, intestinal histomorphometric, meat quality characteristics, carcass yield in broilers fed low protein diets. Tropical Animal Health Production 55 (3), 190, doi: 10.1007/s11250-023-03562-y

Hafeez A., Iqbal S., Sikandar A., Din S., Khan I., Ashraf S., Khan R.U., Tufarelli V., Laudadio V. (2021). Feeding of phytobiotics and exogenous protease in broilers: comparative effect on nutrient digestibility, bone strength and gut morphology. Agriculture 11, 228, doi: 10.3390/ agriculture11030228

Jabbar A., Tahir M., Alhidary I.A., Abdelrahman M.A., Albadani H., Khan R.U., Selvaggi M., Laudadio V., Tufarelli V. (2021). Impact of microbial protease enzyme and dietary crude protein levels on growth and nutrients digestibility in broilers over 15–28 days. Animals 11, 2499, https://doi.org/10.3390/ ani11092499

Kreuzer M., Muller S., Mazzolini L., Messikommer R.E., Gangnat I.D.M. (2020). Are dual-purpose and male layer chickens more resilient against a low-protein-low-soybean diet than slowgrowing broilers? British Poultry Science 61 (1), 33-42.

Ndazigaruye G., Kim D.H., Kang C.W., Kang K.R., Joo Y.J., Lee S.R., Lee K.W. (2019). Effects of low-protein diets and exogenous protease on growth performance, carcass traits, intestinal morphology, cecal volatile fatty acids and serum parameters in broilers. Animals 9 (5), 226.

Park J.H., Lee S.I., Kim I.H. (2020). The effect of protease on growth performance, nutrient digestibility, and expression of growth-related genes and amino acid transporters in broilers. Journal of Animal Science and Technology 62 (5), 614-627.

Pietrzak E., Dunislawska A., Siwek M., Zampiga M., Sirri F., Meluzzi A., Tavaniello S., Maiorano G., Slawinska A. (2020). Splenic gene expression

signatures in slow-growing chickens stimulated in ovo with galactooligosaccharides and challenged with heat. Animals 10, 474.

Tavaniello S., Fatica A., Palazzo M., Zejnelhoxha S., Wu M., Marco L.D., Salimei E., Maiorano G. (2022). Carcass and meat quality traits of medium-growing broiler chickens fed soybean or pea bean and raised under semi-intensive conditions. Animals 12, 2849, doi: 10.3390/ani12202849

Uhlířová L., Tůmová E., Chodová D., Vlčková J., Ketta M., Volek Z., Skřivanová V. (2018). The effect of age, genotype and sex on carcass traits, meat quality and sensory attributes of geese. Asian-Australasian Journal of Animal Sciences 31, 421-428.