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# Comparative Analysis of Biochemical and Hematological Parameters in Free-Range Pigs of Different Genotypes

Komparativna analiza biokemijskih i hematoloških pokazatelja svinja različitih genotipova iz slobodnog uzgoja

**Đidara, M., Šperanda, M., Margeta, V., Galović, D., Pavlić, M., Prakatur, I.**

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**Fakultet agrobiotehničkih znanosti Osijek, Poljoprivredni institut Osijek**

Faculty of Agrobiotechnical Sciences Osijek, Agricultural Institute Osijek

# COMPARATIVE ANALYSIS OF BIOCHEMICAL AND HEMATOLOGICAL PARAMETERS IN FREE-RANGE PIGS OF DIFFERENT GENOTYPES

Đidara, M.<sup>(1)</sup>, Šperanda, M.<sup>(1)</sup>, Margeta, V.<sup>(1)</sup>, Galović, D.<sup>(1)</sup>, Pavlič, M.<sup>(2)</sup>, Prakatur, I.<sup>(1)</sup>

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## Summary

*This study investigated the biochemical and hematological parameters (at the beginning, after 90 days, and at the end of the experiment) of the pure genotype Black Slavonian (BS) pigs and crossbreeds of Black Slavonian and Duroc (BSxD) pigs, fed the same diet and kept under the same extensive free-range conditions during 200 days of the experiment. In the study, BS and BSxD pigs showed differences in blood protein levels, hinting at dietary disparities. Glucose and triglyceride levels differed between the two groups, affecting energy availability and fat deposition. BS pigs showed higher white blood cell counts, while BSxD pigs exhibited more rapid metabolic activity. Alkaline phosphatase activity declined over time, with variations between the groups. Phosphorus and magnesium concentrations decreased with pig growth, reflecting changes in metabolic demands. Hemoglobin, hematocrit, and red blood cell counts varied between the groups. MCV and MCH values in BSxD pigs indicated rapid red blood cell production. Platelet counts fluctuated with age. This study offers insights into the influence of genetic characteristics and growth rates impact biochemical and hematological parameters of pigs kept under free-range conditions, aiding swine population management.*

**Keywords:** *biochemical parameters, hematological parameters, Black Slavonian, Duroc*

## INTRODUCTION

The Black Slavonian Pig, an indigenous treasure of the Slavonia region in Croatia, stands as a testament to centuries of meticulous breeding and preservation efforts. Renowned for its exceptional meat quality, resilience, and unique genetic heritage, the Black Slavonian Pig breed has played a distinctive role in the cultural and agricultural landscape of its native region. This breed, characterized by its striking ebony coat and robust constitution, reflects not only a rich history but also a testament to the enduring collaboration between nature and human management (Senčić and Samac, 2017).

Duroc is one of the most popular sire breeds in pig breeding. It is excellent in growth rate, body composi-

tion, and it is famous for high intramuscular fat content in meat (Gu et al., 2019). The Duroc pig, a distinct and influential breed within the swine industry, has long captured the attention of researchers, breeders, and producers. Known for its exceptional meat quality, rapid growth rate, and adaptability to various environments, the Duroc breed has played a pivotal role in shaping modern pig production systems. Originating from the United States in the early 19th century, Duroc pigs have undergone decades of selective breeding to enhance their desirable traits, making them a sought-after cho-

(1) Assoc. Prof. Dr. Mislav Đidara, Prof. Dr. Marcela Šperanda, Assoc. Prof. Dr. Vladimir Margeta ([vmargeta@fazos.hr](mailto:vmargeta@fazos.hr)), Assoc. Prof. Dr. Dalida Galović, Assoc. Prof. Dr. Ivana Prakatur – Josip Juraj Strossmayer University of Osijek, Faculty of Agrobiotechnical Sciences Osijek, Vladimira Preloga 1, 31000 Osijek, Croatia, (2) Martina Pavlič – Croatian Agency for Agriculture and Food, Gundulićeva 36b, Osijek, Croatia.

ice for both commercial and research purposes (Gu et al., 2019).

Crossbreeding has long been a strategy in swine production aimed at harnessing the strengths and compensating for the weaknesses of different pig breeds (Kadirvel et al., 2023). In this pursuit, the fusion of the Black Slavonian Pig, an indigenous pig from Croatia, and the Duroc pig, globally acclaimed for its growth and meat quality, is a promising avenue for innovation in the swine industry. The combination of these two distinct genetic lineages offers the potential to create a unique breed with enhanced attributes, bringing together the heritage of the Black Slavonian Pig's resilience and local adaptation with the Duroc's renowned productivity and meat excellence. This research aimed to investigate and compare the biochemical and hematological parameters of two distinct pig populations: the first being of pure Black Slavonian genotype, and the second consisting of a crossbreed between Black Slavonian and Duroc breeds. Both groups were raised under identical extensive free-range conditions. This study tried to explain potential variations in these parameters between the purebred and crossbred pigs, providing valuable insights into how genetic backgrounds may influence the health and physiological profiles of swine reared in similar environmental settings.

## MATERIAL AND METHODS

### Animals and Diets

The experiment was approved by the Local Ethical Review Committee for Animal Experiments at the Faculty of Agrobiotechnical Sciences Osijek, Croatia (protocol no. 2198-94-02-23-02). The nutritional experiment was carried out at a private pig farm. The study was conducted on 30 Black Slavonian breed piglets (average weight  $26.9 \pm 2$  kg) and 30 piglets of crossbreed (female Black Slavonian x male Duroc, average weight  $30.1 \pm 1.8$  kg), with an average age of  $90 \pm 3$  days at the onset of experiment, trying to keep sex ratio 1:1. Piglets were placed separately in a divided free range, 0.5 hectare fenced area, and were fed two times a day with a diet of 1.2 kg/piglet/day until day 90, and then with a diet of 2.2 kg/pig/day (Table 1), with the addition of 3 kg of fresh alfalfa during the season and 1.5 kg of dried alfalfa off the season and with constant *ad libitum* water access. Animals were individually weighted on days 90 and 200 of the experiment.

The contents of basic nutrients in cereal grains and diets were determined according to Official Methods of Analysis of AOAC International (AOAC, 1995) (dry matter (DM, AOAC: 934.01); crude protein (CP, Kjeldahl method, AOAC: 984.13); crude ash (CA, AOAC: 942.05); ether extract (EE, Soxhlet method, AOAC: 920.39A) with the use of a BUCHI Extraction System B-811 (BÜCHI, Flawil, Switzerland); crude fiber (CF, Henneberg and Stohmann method, AOAC: 978.10).

**Table 1. Ingredients and chemical composition of the concentrate part of the diet for growing pigs during the experimental period**

Tablica 1. Sastav i kemijski sastav koncentriranoga dijela obroka svinja u porastu tijekom pokusnoga razdoblja.

	Day 0 – 90 / Dan 0 – 90	Day 90 – 200 / Dan 90 – 200
<i>Ingredients of the concentrate, %</i>		
Corn / kukuruz	50	50
Barley / ječam	15	15
Wheat / pšenica	10	10
Toasted soybean / tostirana soja	24,5	20
Premix / premiks Domino SZ®	0.5	0.5
Wheat bran / pšenično brašno	-	4.5
NEL MJ/kg DM	12.96	12.70
<i>Chemical composition</i>		
Dry matter / suha tvar, %	89.1	89.2
Crude fibre / sirova vlaknina	5.20	5.50
Crude protein / sirovi protein	15.7	13.4
Crude ash / pepeo	7.1	7.2
Ether Extract / masti	2.4	2.1

### Hematological, and Biochemical Parameters

On the zero day (pigs aged 90 days) and on the 90th (pigs aged 180 days) and the 200th day (pigs aged 290 days) of the experiment, 10 animals were randomly chosen from each group. Blood from the jugular vein was taken by the Vacutainer system in lithium heparin anti-coagulant tubes (Becton Dickinson, Plymouth, England, UK). The samples were centrifuged (1.500 g/10 min. at 4°C), and the plasma was separated and frozen at -80 °C till the analyses. The samples for hematological analyses were taken into the Ca-EDTA tubes (Becton Dickinson, Plymouth, England, UK) and analyzed within two hours on automated hematology analyzer Poch-100iV Diff (Sysmex, Japan). Plasma was analyzed on a clinical chemistry analyzer Beckman Coulter AU680 (Beckman Coulter, Germany), using Beckman Coulter commercial kits.

### Statistical Analysis

Descriptive statistics for plasma biochemical parameters were performed using TIBCO Statistica (2020). The assumption of normality was checked using the Shapiro-Wilk test. Biochemical and hematological parameters normally distributed were represented by means and SEM (Tables 2 and 3). Variables not normally distributed were subjected to  $\log^{10}$  transformation to obtain a normal distribution of values prior to statistical analysis and are represented by medians and the associated interquartile range (Tables 2 and 3). For the evaluation of the effect of genetic group and days in the experiment on the variability of biochemical and hematological parameters of individual animals, the following fixed statistical model was used:

$$y_{ijk} = \mu + T_i + D_j + T_i D_j + e_{ijk},$$

where:

$y_{ijk}$  = estimated trait (biochemical parameter in plasma);

$\mu$  = intercept;

$T_i$  = fixed effect of genetic group  $i$  (groups = BS and BSD);

$D_j$  = fixed effect of day  $j$  ( $j = 0, 90,$  and  $200$  days of the experiment);

$e_{ijk}$  = residual.

The significance of the differences between the analyzed traits due to the fixed effect of genetic group and time, as well as any interactions, was tested by *post hoc* Tuckey's test at the level of  $P < 0.05$ .

### RESULTS AND DISCUSSION

On the 90<sup>th</sup> day of the experiment, average weight of the BS genotype was  $61.70 \pm 6$  kg and of the BSxD genotype  $72.2 \pm 5$  kg and on day 200 average weight of the BS genotype was  $128.40 \pm 15$  kg and of the BSxD genotype  $143.1 \pm 13$  kg. All biochemical and hematological parameters determined are presented in Table 2 and Table 3.

Total protein and albumin are critical indicators of nutritional health. Total protein measures overall protein levels, reflecting their dietary intake and overall health. Albumin, a specific protein component, evaluates liver function and protein synthesis. Irrespectively of a genotype, both total protein and albumin concentration were higher ( $P < 0.05$ ) in the second and third sampling compared to the first. This could indicate better protein digestion and amino-acid utilization from feed in older pigs. Significant ( $P < 0.05$ ) interaction between the day of sampling and genotype with lower ( $P < 0.05$ ) values of both parameters in plasma of BSxD crossbreed could indicate that the dietary level of protein was not sufficient to cover protein needs, unlike in BS which had slower growth rate compared to BSxD. Zaitsev et al. (2021) found a total protein value of 69.95 g/L and albumin value of 36.66 g/L in Duroc breed male pigs at day 175 of fattening compared to 68.4 g/L and 27.5 g/L on the 90th day in BSxD crossbred. Albumins are better markers of protein availability than total serum proteins. Urea concentrations in the second and third sampling were higher ( $P < 0.05$ ) in the plasma of BSxD which might suggest a lower biological value of protein supplied in feedstuff (Chmielowiec-Korzeniowska and Babicz, 2009) than required for the BSxD genotype.

**Table 2. Biochemical parameters of the Black Slavonian and the Black Slavonian x Duroc crossbreed pigs on the zero day, 90th day, and the 200th day of the experiment.**  
*Tablica 2. Biokemijski pokazatelji crne slavonske svinje i križanaca crne slavonske svinje i pasmine durok 0., 90. i 200. dana pokusa.*

	Day 0 / Dan 0		Day 90 / Dan 90		Day 200 / Dan 200		P-value Genotype / P-vrijednost Genotip	P-value Day / P-vrijednost Dan	P-value D x G / P-vrijednost D x G
	BS	BSxD	BS	BSxD	BS	BSxD			
Total protein, g/L / Ukupni proteini, (g/L)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	63.2 (2.9)	67.5 (2.5)	74.3 (1.4)	68.4 (2.5)	79.1* (1.5)	64.8* (1.8)	<b>0.0003</b>	
Albumin, g/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	24.4 (1.6)	28.1 (2.3)	35.4* (1.2)	27.5* (1.2)	37.2* (1.6)	26.2* (1.5)	<b>0.0001</b>	
Triglycerides, mmol/L / Trigliceridi, mmol/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	0.65 (0.62-1.09) <sup>y</sup>	0.78 (2.85-3.23) <sup>y</sup>	0.81 (0.78-0.87) <sup>y</sup>	0.50 (0.4-0.66) <sup>y</sup>	0.96 (0.63-1.12) <sup>y</sup>	0.82 (0.57-1.1) <sup>y</sup>	0.1229	
Cholesterol, mmol/L / Kolesterol, mmol/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	2.47 (0.06)	2.63 (0.16)	2.87 (0.10)	2.79 (0.17)	3.49 (0.11)	3.06 (0.10)	0.0638	
Glucose, mmol/L / Glukoza, mmol/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	5.25 (4.96-6.1) <sup>y</sup>	5.78 (4.64-6.76) <sup>y</sup>	5.63* (5.12-5.82) <sup>y</sup>	4.32* (3.89-4.8) <sup>y</sup>	5.73* (5.56-5.97) <sup>y</sup>	4.4* (4.11-4.47) <sup>y</sup>	<b>0.0025</b>	
AST, U/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	48.9 (39.3-54.1) <sup>y</sup>	47.5 (39.3-58.4) <sup>y</sup>	50.9 (45.8-59.1)	44.2 (37.6-50.8) <sup>y</sup>	42.6 (30.2-55.1) <sup>y</sup>	34.9 (29.4-39.2) <sup>y</sup>	0.2382	
ALT, U/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	57.1* (4.6)	100.34* (6.2)	53.2* (2.5)	90.5* (3.8)	53.2 (1.9)	58.5 (3.1)	<b>0.0001</b>	
ALP, U/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	159.2 (114.7-172.1) <sup>y</sup>	194.1 (152.3-244.5) <sup>y</sup>	136.8 (120.2-147.2) <sup>y</sup>	164.9 (109.2-194.8) <sup>y</sup>	130.7* (93.1-160) <sup>y</sup>	85.3* (77.2-88.3) <sup>y</sup>	<b>0.0022</b>	
GGT, U/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	55.9 (5.6)	49.9 (3.4)	36.2* (3.0)	63.0* (5.1)	39.0 (7.1)	51.2 (5.4)	<b>0.0089</b>	
Urea, mmol/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	5.92* (5.74-6.97) <sup>y</sup>	3.69* (3.15-4.14) <sup>y</sup>	3.25* (2.86-3.35) <sup>y</sup>	5.48* (5.1-6.65) <sup>y</sup>	3.91* (3.56-4.29) <sup>y</sup>	6.2* (5.5-6.92) <sup>y</sup>	<b>0.0001</b>	
Ca, mmol/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	2.55 (2.47-2.61) <sup>y</sup>	2.55 (2.41-2.72) <sup>y</sup>	2.63 (2.56-2.69) <sup>y</sup>	2.45 (2.3-2.51) <sup>y</sup>	2.72 (2.63-2.8)	2.52 (2.36-2.71) <sup>y</sup>	0.6367	
P, mmol/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	2.69 (0.09)	3.00 (0.09)	2.15* (0.12)	3.45* (0.13)	2.56 (0.05)	2.65 (0.04)	<b>0.0001</b>	
Mg, mmol/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	0.713 (0.70-0.81) <sup>y</sup>	0.745 (0.70-0.80) <sup>y</sup>	0.838* (0.80-0.85) <sup>y</sup>	0.994* (0.90-1.05) <sup>y</sup>	0.990* (0.91-1.10) <sup>y</sup>	0.727* (0.68-0.75) <sup>y</sup>	<b>0.0001</b>	
Fe, μmol/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	18.6 (1.1)	22.0 (1.8)	20.9 (1.8)	21.5 (1.1)	29.5* (1.4)	14.9* (1.8)	<b>0.0061</b>	

BS, Black Slavonian genotype; BSxD Black Slavonian and Duroc crossbreed; SEM, standard error of mean; IQR interquartile range; ALT, alanine aminotransferase; AST, aspartate aminotransferase; ALP, alkaline phosphatase; GGT, γ-glutamyl transferase; \*values significantly (P < 0.05) differ between groups at a given day / značajna (P < 0.05) razlika vrijednosti između skupina u određenome danu

BS, crna slavonska; BSxD križanci crne slavonske i duroka; SEM, standardna greška srednje vrijednosti; IQR međukvartilni raspon; ALT, alanin aminotransferaza; AST, aspartat aminotransferaza; ALP, alkalna fosfataza; GGT, γ-glutamil transferaza,\*

**Table 3. Hematological parameters of Black Slavonian and Black Slavonian x Duroc crossbreed pigs at day 0, 90 and 200 of the experiment.**  
 Tablica 3. Hematološki pokazatelji crne slavonske svinje i križanaca crne slavonske svinje i pasmine durok 0., 90. i 200. dana pokusa.

	Day 0 / Dan 0		Day 90 / Dan 90		Day 200 / Dan 200		P-value Day / P-vrijednost Dan	P-value Genotype / P-vrijednost Genotip	P-value D x G / P-vrijednost D x G	
	BS	BSxD	BS	BSxD	BS	BSxD				
WBC, x10 <sup>9</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	21.2(17.6-23.5) <sup>y</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	16.9* (14.3-22.8) <sup>y</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	12.3 (12-16.7) <sup>y</sup>	14.6 (13-17.1) <sup>y</sup>	0.0001	0.3237	0.0182
RBC, x10 <sup>12</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	7.7 (0.3)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	6.9 (0.1)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	8.5 (0.2)	7.0 (0.1)	0.0586	0.0001	0.2432
Hgb, g/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	117.5 (105-116) <sup>y</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	125.5 (123-131) <sup>y</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	144.5* (139-149) <sup>y</sup>	119* (115-127) <sup>y</sup>	0.0001	0.0166	0.0001
HCT	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	0.38 (0.01)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	0.40 (0.01)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	0.39 (0.01)	0.39* (0.01)	0.0065	0.0328	0.0142
MCV, fl	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	55.7* (53.7-57.5) <sup>y</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	57.8* (55.1-59.5) <sup>y</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	52.9 (51.7-54.6) <sup>y</sup>	55.75 (52.9-57.1) <sup>y</sup>	0.1605	0.0001	0.0047
MCH, pg	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	16.6* (16.2-17.4) <sup>y</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	18.2* (17.7-19) <sup>y</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	16.6 (16.3-17.2) <sup>y</sup>	17.2 (15.8-17.8) <sup>y</sup>	0.0001	0.0001	0.0018
MCHC, g/L	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	299.8 (2.2)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	317.2 (2.0)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	316.7 (1.7)	308.3 (3.5)	0.0001	0.6381	0.0286
PLT, x10 <sup>6</sup>	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	511.8*s (41.2)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	433.8 (60.0)	Mean (SEM) / %Median (IQR) srednja vrijednost (SEM) / %medijan (IQR)	402.3 (22.5)	457.8 (19.1)	0.0001	0.1493	0.0056

BS, Black Slavonian genotype; BSxD Black Slavonian and Duroc crossbreed; SEM, standard error of mean; IQR, interquartile range; WBC, total white blood cells; RBC, total red blood; Hgb, hemoglobin; HCT, hematocrit; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; PLT, total platelet number; \*values significantly (P<0.05) differ between groups at a given day  
 BS, crna slavonska; BSxD križanci crne slavonske i duroka; SEM, standardna greška srednje vrijednosti; IQR međukvartilni raspon; WBC, broj leukocita; RBC, broj eritrocita; Hgb, hemoglobin; HCT, hematocrit; MCV, prosječni volumen eritrocita; MCH, količina hemoglobina u eritrocitima; MCHC, koncentracija hemoglobina u eritrocitima; PLT, broj trombocita; \* značajna (P<0,05) razlika vrijednosti između skupina u određenoj danu

Similar to the protein content, glucose concentration was lower ( $P < 0.05$ ) in the plasma of BSxD at second and third sampling, suggesting a lower energy availability from diet for the crossbreed pigs with a higher growth rate. Glucose is a critical clinical chemistry parameter in pigs, reflecting their metabolic state. Glucose is essential as a primary source of energy for rapid weight gain. During the periods of high-calorie intake, glucose provides an immediate energy source for the growth and fat deposition. Proper glucose metabolism ensures efficient weight gain and meat quality. For the Duroc breed, Zaitsev et al. (2021) established a blood glucose value of 5.33 mmol/L. The overall reduction in glucose concentration with age, which was determined ( $P < 0.05$ ) in our study, is also described in the literature (Zaitsev et al., 2021). Glucose concentration in pigs generally decreases with age. This phenomenon is attributed to the shift in metabolic demands. Young pigs require higher glucose levels for rapid growth, but as they mature, their metabolism becomes more efficient, necessitating less glucose. This age-related decline in glucose concentration is a natural aspect of their physiological development.

Lower ( $P < 0.05$ ) triglycerides in the plasma of BSxD pigs during the whole experiment might also suggest a lower energy availability. In fattening pigs, triglycerides play a vital role as an energy reserve (Liu et al., 2015). When pigs consume more calories than they immediately need, the excess energy is stored in the adipose tissue in the form of triglycerides. These stored triglycerides can be mobilized and converted back into energy when food is scarce or during periods of increased metabolic demand. Monitoring triglyceride levels helps to assess the efficiency of fat deposition and overall nutritional management in fattening pigs, to ensure they reach the desired weight and quality for the market.

Under restricted feeding in the 24th week of age, high lean growth pigs had higher basal levels of WBC (Clapperton et al., 2006). This phenomenon might be a reaction to their rapid muscle development and increased metabolic activity. The body's immune system recognizes this as a natural stress, leading to a rise in white blood cells. This was also the case in our study, as the genetics of the BS breed is generally associated with high daily adipose tissue deposition and here determined a lower WBC count on days 90 and 200 compared to the BSxD.

Higher ( $P < 0.05$ ) activity of ALT was determined at first and second sampling, and GGT at second sampling in the BSxD genotype. The higher activity of ALT (Alanine Aminotransferase) and GGT (Gamma-Glutamyl Transferase) in faster-growing pigs can be attributed to their increased metabolic rate and rapid muscle development (Kapelanski et al., 2004). These enzymes are primarily produced in the liver and are involved in various metabolic processes, including protein synthesis and energy production. In faster-growing pigs, the liver works more efficiently to meet the increased energy and protein demands required for their rapid growth. Therefore, the elevated ALT and GGT levels in

the blood often have a normal physiological response to the increased metabolic activity associated with rapid growth (Kapelanski et al., 2004).

Alkaline phosphatase (ALP) in fattening pigs serves as an enzyme that reflects both bone health and liver function (Sousa et al., 2011). The ALP activity tends to be higher in young pigs, especially piglets, as their bones are actively growing. As pigs grow older and their bones mature, the ALP levels typically decrease. In our study, we found a decrease ( $P < 0.05$ ) in the ALP activity over time which is in correlation with previous research (Tymczyna et al., 2012). At the last sampling (day 200), a significantly lower ( $P < 0.05$ ) ALP activity was found in the BSxD genotype compared to the BS, which probably means that these pigs finished their intensive bone growth phase faster than the BS pigs, reflected in a lower ALP activity. The trends of the ALP activities reversed at the first and second sampling when the BSxD genotype had faster growth.

Similar to the ALP activity, the P concentration in our study was significantly ( $P < 0.05$ ) lower in the third sampling compared to the first two. This indicates that in young fattening pigs, especially piglets, blood phosphorus levels are generally higher. This is because they require more phosphorus for bone growth and development (Tymczyna et al., 2012). The significantly ( $P < 0.05$ ) higher P concentration on the day 90 of our study was found in the BSxD group probably reflecting higher overall body growth in this group.

Although the magnesium blood concentration is typically higher in younger fattening pigs, dietary magnesium intake plays a significant role in blood magnesium levels (Katalin et al., 2004). Properly balanced diets are essential to maintaining adequate magnesium levels. Dietary magnesium imbalances can lead to fluctuations in blood concentrations. In our study, we found a lower ( $P < 0.05$ ) concentration of Mg at the third sampling in the BSxD compared to the BS genotype, which is in agreement with the fact that in fattening pigs the metabolic rate decreases towards the end of their rapid growth phase, resulting in lower blood concentrations of magnesium (Zang et al., 2014). This is because they no longer require as much magnesium for rapid muscle and bone development. Their metabolic processes become more balanced through dietary intake, leading to lower magnesium levels in the blood.

Significantly ( $P < 0.05$ ) lower blood hemoglobin and hematocrit levels were determined on the 200th day in the BSxD group when compared to the BS genotype. When fattening pigs complete their growth and reached their desired weight, their blood hemoglobin and hematocrit levels often decrease (Zhang et al., 2022). This is because their metabolic demands decrease, as they no longer need to support rapid muscle and bone development (Lindholm-Perry et al., 2021). In contrast, in our study, a slower-growing BS genotype still gaining weight on the 200th day could have the higher hemoglobin and hematocrit levels due to the ongoing need for oxygen transport to the growing tissues.



The overall RBC count was lower ( $P < 0.05$ ) in the BSxD genotype when compared to the BS, which could reflect a faster growth of the BSxD group. The pigs that experience a faster growth often have a lower red blood cell count. This occurs because their rapid muscle development increases the blood volume, while the production of red blood cells may not keep pace. As a result, the concentration of red blood cells in the bloodstream decreases (Lindholm-Perry et al., 2021).

At the first two sampling periods, the MCV and MCH were higher ( $P < 0.05$ ) in the BSxD genotype when compared to the BS group. During the initial growing period, the pigs with a faster growth exhibited the higher MCV and MCH values (Albers et al., 2022). This is a result of their accelerated red blood-cell production to support rapid muscle development. The MCV measures the size of red blood cells, and the MCH quantifies the amount of hemoglobin in each cell (Lindholm-Perry et al., 2021). The faster-growing pigs often have larger and more hemoglobin-rich red blood cells, which is an adaptive response to meet the increased oxygen-carrying demands of their growing tissues.

Significantly ( $P < 0.05$ ) higher PLT number was determined at first sampling in the BS genotype when compared to the BSxD. During the initial growing period, the pigs with a slower growth often exhibit higher platelet levels. This may occur as a compensatory response to the potential factors affecting their growth rate, such as dietary deficiencies or health challenges. Disregarding a genotype, the highest ( $P < 0.05$ ) PLT number was at the first sampling (Day 0). This is in correlation with Pliszczak-Król et al. (2016), who also established the highest PLT number in 4-week-old piglets. Lombardi et al. (2005) have documented even more elevated PLT counts, ranging from 753 to  $785 \times 10^9/L$  in the piglets aged three to eight weeks. During the initial growing period, the pigs often exhibit higher platelet levels. This is a part of their natural growth and development process. Platelets play a crucial role in blood clotting and wound healing, which are essential during the periods of rapid growth, when the tissues are expanding and are potentially prone to injury.

## CONCLUSIONS

The results of this study emphasize the importance of considering genotype, age, and growth rate when assessing the porcine health and metabolism. The plasma glucose concentration in the BSxD was significantly lower ( $P < 0.05$ ) during the second and the third sampling, indicating a reduced energy availability from the diet for crossbred pigs with an increased growth rate. The BS pigs displayed the elevated white blood cell counts, whereas the BSxD pigs demonstrated a swifter metabolic activity. The alkaline phosphatase activity experienced a gradual decrease over time, exhibiting distinctions between the groups. Phosphorus and magnesium levels diminished as the pigs grew, mirroring the shifts in metabolic requirements. Hemoglobin, hematocrit, and the red blood cell counts exhibited variances

between the sets. The MCV and MCH measurements in the BSxD pigs suggested prompt red blood cell production. The trends observed provide valuable insights for optimizing nutritional management and ensuring the welfare of pigs in extensive free-range systems. Further research could investigate the strategies to address protein and energy requirements in the crossbred pigs to effectively support their higher growth rates.

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## KOMPARATIVNA ANALIZA BIOKEMIJSKIH I HEMATOLOŠKIH POKAZATELJA SVINJA RAZLIČITIH GENOTIPOVA IZ SLOBODNOGA UZGOJA

### SAŽETAK

**U ovome istraživanju utvrđeni su biokemijski i hematološki pokazatelji svinja različitoga genotipa crne slavonske (BS) i križanaca crne slavonske svinje i duroka (BSxD), hranjenih istim obrokom i držanih u istim uvjetima slobodnoga uzgoja. Istraživanjem su utvrđene veće koncentraciji ukupnih bjelančevina i glukoze u plazmi crne slavonske svinje, što je najvjerojatnije pokazatelj većih hranidbenih potreba križanaca. Broj leukocita bio je veći u krvi crne slavonske svinje. Aktivnost alkalne fosfataze u plazmi smanjivala se tijekom trajanja pokusa, a utvrđene su i razlike između skupina. Koncentracija fosfora i magnezija opadala je u skladu s rastom svinja, odražavajući promjenu metaboličkih zahtjeva. Hemoglobin, hematokrit i broj eritrocita varirali su između skupina. Vrijednosti MCV-a i MCH-a kod križanaca ukazivale su na brzu proizvodnju eritrocita. Opisane razlike u biokemijskim i hematološkim pokazateljima svinja različitih genotipova u uvjetima slobodnoga uzgoja koristan su alat za bolje praćenje zdravlja i poboljšanje proizvodnih pokazatelja.**

**Ključne riječi:** biokemijski pokazatelji, hematološki pokazatelji, crna slavonska, durok

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