

SUMMARY

Objective of this study is to evaluate growth pattern of broilers fed diets containing different oils. Data for this study were obtained from an experiment involving 160 unsexed one day old chicks of Ross 308 cross randomly allocated to four dietary treatments ($n=40$) with five replicates in each of them (eight birds per replicate) over 42 days. Feeding experimental diets was introduced in the last 21 days. Composition of experimental diets (C, D1, D2 i D3) was the same, containing different levels of added oils. Control diet contained 6% of sunflower oil. D1, D2 and D3 diets contained added oils in the following combinations: 1% fish oil + 2% linseed oil + 3% rapeseed oil (D1), 1.5% fish oil + 1.5% linseed oil + 3% rapeseed oil (D2) and 2% fish oil + 1% linseed oil + 3% rapeseed oil (D3). All diets were isoenergetic and contained 0.015% vitamin E. Gompertz model fits the data obtained from weekly weighing of birds. The model indicated high accuracy with the determination coefficient of over 99.9%. Predicted mature weight of birds, initial specific growth rate per week, rate of exponential decay of the initial specific growth rate, age of birds at inflection point, weight at the inflection point, and maximum weight gain at inflection point for each dietary treatment were subjected to one-way variance analysis. A positive effect of dietary omega-3 fatty acids on most of examined growing curve parameters has been established.

Key words: broilers, growth curve, n-3 PUFA in diets

INTRODUCTION

Beneficial effects of long-chain n-3 polyunsaturated fatty acids (n-3 PUFA) rich oils on modification of fatty acids profile in meat of broilers are well documented. Many authors emphasize significance of fish, flaxseed and rapeseed oils incorporated in diets on the decreasing of n6/n3 PUFA ratio in broiler meat (Hang et al., 2018; Čengić-Džomba et al., 2016; Škrtić et al., 2009) as well as on the beneficial (Thanabalan and Kiarie, 2021) or adverse (Al-Khalifa et al., 2012) immune responses of birds. On the other hand, relevant literature says little about controversial information (Navidshad, 2009, Elzobier et al., 2016, Duarte et al., 2014) concerning effects of nutrition with different oils on growth characteristics of broilers.

The growth of an animal is a biological phenomenon and can be defined as any change in body size over time (Nariç et al., 2017). Knowing the growth pattern of birds is a useful tool in optimizing the production of broiler meat with respect to responses to genetic selection, environmental change and estimation of nutrients requirements. Thus, growth is biologically determined, although many other factors, including nutrition of animals, can modify some parameters of typically sigmoid pattern growth curve. Gompertz model used in describing growing patterns of living organisms has three very important parameters: an asymptote which denotes mature body weight, specific growth rate of gain, and inflection point which denote maximum growth

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rate on increasing time scale (Caldas et al., 2019). Central point of S-shaped growth curve is inflection point which separates accelerating (starting) and decelerating (finishing) growth phase. Hence, any positive shift of the inflection point of the bird growth curve allows the possibility of a longer period of accelerated growth (and hence better feed conversion ratio) which has a positive effect on production, in general.

If fish oil / corn diets with lower (n-3) : (n-6) PUFA ratios result with greater cell-mediated immunity than the soybean oil / cereal diets (Krover and Klasing., 1997) and if omega-3 PUFAs have significant effects on the intestinal environment and altered intestinal microbiota (Costantini et al., 2017) feeding broilers on oils containing omega-3 PUFAs can be effective means for improving broiler performance. The aim of this study was to examine the effect of added oils rich in n-3 fatty acids on the growth curve characteristics of broilers.

MATERIAL AND METHODS

The investigation was conducted on broiler farm “Brovis” – Visoko, in Bosnia and Herzegovina. A total of 160 unsexed one day old chicks of Ross 308 cross were randomly allocated to four dietary treatments (n=40) with five replicates in each of them (eight birds per replicate) over 42 days. The birds were fed the experimental diets the last 21 days. In the first 21 days, broilers were fed the same diet (starter), while experimental diets were introduced in the last three weeks of the experiment.

Experimental diets

Composition of experimental diets (C, D1, D2 i D3) was the same and contained different levels of sunflower, linseed and fish oils (1%, 1.5%, 2%), and fixed level of rapeseed oil (3%), so total amount of added oils in each diet was 6%. The control diet contained 6% sunflower oil. All diets were isoenergetic and contained 0,015% vitamin E (Table 1).

Fatty acids composition of experimental diets is shown in Table 2.

Feed intake measuring was done by subtracting residual feed from the total supplied feed. Body weight (BW) and feed intake were recorded weekly in groups of eight birds (replicate).

Table 1 Ingredients and nutritional value of experimental diets

Tablica 1. Sastav i hranjiva vrijednost pokusnih obroka

Ingredients / Krmiva	Experimental diets / Pokusni obroci %			
	C	D1	D2	D3
Corn	41.00	41.00	41.00	41.00
Wheat	17.00	17.00	17.00	17.00
Soybean meal	22.00	22.00	22.00	22.00
Sunflower meal	10.35	10.35	10.35	10.35
Sunflower oil	6.00	-	-	-
Rapeseed oil	-	3.00	3.00	3.00
Linseed oil	-	2.00	1.50	1.00
Fish oil	-	1.00	1.50	2.00
Limestone	1.20	1.20	1.20	1.20
Dicalcium phosphate	1.40	1.40	1.40	1.40
NaCl	0.235	0.235	0.235	0.235
Methionine	0.14	0.14	0.14	0.14
Lysine	0.16	0.16	0.16	0.16
Vit/Min, Premix*	0.50	0.50	0.50	0.50
Vitamin E	0.015	0.015	0.015	0.015
Nutrient composition / Kemijski sastav				
Crude protein, %	19.51			
Ether extract, %	8.35			
Crude fibre, %	4.78			
Ash, %	5.52			
ME, MJ/kg	13.06			
Ca, %	0.91			
P, %	0.70			
Lysine, %	1.01			
Methionine + cystine, %	0.73			

*in 1 kg: vitamin A 2400000 IJ, vitamin D3 480000 IJ, vitamin E 6000 mg, vitamin K3 500 mg, vitamin B1 300 mg, vitamin B2 1200 mg, nicotinamid 7000 mg, calcium pantothenate 2000 mg, vitamin B6 800 mg, vitamin B12 3000µg, folic acid 150 mg, biotin 10 mg, cholinechlorid 80000 mg, methionine 100000 mg, J 200 mg, Mn 16000 mg, Zn 10000 mg, Co 20 mg, Fe 6000 mg, Cu 1000 mg, Se 30 mg, antioxidant 20000 mg, flavophosphopol 600 mg, coccidiostatic 20000 mg.

C = feeding a mixture containing 6% sunflower oil; D1 = feeding a mixture containing 1% fish oil + 2% linseed oil + 3% rapeseed oil, D2 = feeding a mixture containing 1.5% fish oil + 1.5% linseed oil + 3% rapeseed oil, D3 = feeding a mixture containing 2% fish oil + 1% linseed oil + 3% rapeseed oil.

C = hranidba smjesom sa 6 % suncokretovog ulja; D1 = hranidba sa smjesom koja sadrži 1 % ribljeg ulja + 2 % lanenog ulja + 3 % repičinog ulja; D2 = hranidba sa smjesom koja sadrži 1,5 % ribljeg ulja + 1,5 % lanenog ulja + 3 % repičinog ulja, D3 = hranidba sa smjesom koja sadrži 2 % ribljeg ulja + 1 % lanenog ulja + 3 % repičinog ulja.

Table 2 Fatty acids composition of experimental diets, g/100 g of total fatty acids

Tablica 2. Sastav masnih kiselina u pokusnim obrocima, g/100 g ukupnih masnih kiselina

Fatty acids / Masne kiseline	Experimental diets / Pokusni obroci			
	C	D1	D2	D3
C14:0	0.09	0.58	0.89	1.29
C16:0	7.22	8.23	8.55	9.38
C16:1n-9	0.08	0.77	1.08	1.65
C18:0	2.95	2.74	2.44	2.40
C18:1n-9	24.58	34.05	35.56	36.20
C18:2n-6	60.62	34.06	29.99	27.51
C18:3n-3	1.34	14.79	13.99	11.62
C20:2n-6	0.09	0.24	0.33	0.54
C20:2n-9	0.66	0.37	0.35	0.26
EPA	0.15	0.83	1.36	1.94
DPA	0.08	0.30	0.50	0.75
DHA	0.06	0.86	1.48	2.20
∑ n3	1.63	16.78	17.33	16.51
∑ n6	60.71	34.30	30.32	28.05
n6/n3	37.25	2.04	1.75	1.70
SFA	10.26	11.55	11.88	13.07
MUFA	24.66	34.82	36.64	37.85
PUFA	63.00	51.45	48.00	44.82
SFA/MUFA	0.42	0.33	0.32	0.34
SFA/PUFA	0.16	0.22	0.25	0.29

C = feeding a mixture containing 6% sunflower oil; D1 = feeding a mixture containing 1% fish oil + 2% linseed oil + 3% rapeseed oil, D2 = feeding a mixture containing 1.5% fish oil + 1.5% linseed oil + 3% rapeseed oil, D3 = feeding a mixture containing 2% fish oil + 1% linseed oil + 3% rapeseed oil.

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Crude protein, ether extract, crude fibre, ash, Ca and P in feed were determined by standard laboratory methods while energy, lysine and methionine + cystine content was calculated.

The fatty acids content in feed was determined by gas chromatography. Extraction of lipids was performed on 0.5 g samples according to Folch et al. (1957). Lipids were dissolved in heptane and methylated using Na-methoxide and methanolic

HCl 3N (Supelco, PA, USA). Methyl esters of fatty acids were analyzed using a Carlo Erba 8130 gas chromatograph equipped by 100m capillary column (CP-sil 88 WCOT, 100-mx 0.25 mm, Chrompack, Middelburg, The Netherlands). Starting temperature was 70°C, and after 4 minutes the temperature was increased to 13 degrees per minute until reaching 175°C and this temperature was maintained 27 minutes after which the temperature was further increased to 4°C per minute until reaching 215°C and maintained 31 minutes. The temperature was again increased to 4°C per minute until reaching the final temperature of 225°C. The flow of gas was 1.62 ml / minute, with 1: 34 ratio in the flame-ionizing detector. Fatty acids were converted to amounts of fatty acids expressed g/100g fatty acids. A cod liver oil was used as standard for every tenth sample.

Data analysis

Gompertz model was fit to the data. The Gompertz growth curve is described by the following equation:

$$W_t = \beta_A \exp(-\beta_1 \exp(-\beta_2 t))$$

where: W_t = is the expected value as a function of t, β_A = asymptotic body weight of mature birds, β_1 = initial specific growth rate per week, β_2 = rate of exponential decay of the initial specific growth rate, t = time in weeks.

Age of birds at inflection point (AIP), weight at the inflection point (WIP) and maximum weight gain at inflection point were calculated as:

$$AIP = (\ln \beta_1) / \beta_2 \quad WIP = \beta_A / e \quad MWG = \beta_2 WIP$$

Adjusted determination coefficient (R^2_{adj}) was used as goodness of fit criteria of used model.

Statistical analysis

The obtained results were subjected to one-way analysis of variance (ANOVA) using statistical package program SPSS (v.21.0). Statistically significant effects were further analysed and means were compared using LSD test. Statistical significance was considered at $P < 0.05$.

RESULTS AND DISCUSSION

Average weekly body weight of birds fed each experimental diet shown in Table 3. Mean body weight was slightly below values stated in Manual of performance objectives of Ross®PM3 Broiler.

At 21st day of the experiment body weights of broilers were similar ($P>0.005$). With inclusion of different oils in diets the body weight of broilers changed differently in each experimental group. At 42-days, broilers from the control group were slightly lighter compared to D1 group. Body weights of broilers at the end of previous week (35 days) show a different pattern; differences between control and D3 group were statistically significant (Table 2). Thus, combination of different oils in experimental diets might have caused these differences.

Analysis of growth curve parameters (Table 4) confirms previous statement. Gompertz growth curve generally shows sigmoid pattern (Figure 1) characterized by point of inflection somewhere after the middle of the growing period which divides accelerated growth phase (weight gain is more pronounced) which lasts from hatching to AIP and after that, declining growth rate until reaching mature body weight.

Adjusted determination coefficient (R^2) as one of goodness of fit criteria in all curves was a number higher than 0.999 (Table 3) indicating excellent fit of the data suggesting that Gompertz model could

be rather accurate in the depiction of the field data. Similar R^2 value of Gompertz model in describing broilers growth has been reported by Koncagul & Cadirci (2010), Caldas et al., (2019) and Demuner et al., (2017).

Predicted mature body weight in the control group, and D3 Gompertz curve model equations is lower than in the other two diets. Also, decay specific growth rate (β_2) of broilers fed D3 and control diets was statistically higher compared to others.

Age of birds at inflection points of the curve is of great interest because it shows the age when birds have maximum growth rate. AIP in this experiment ranged from 23.36 to 28.39 days (Table 3) indicating that birds fed D1 and D3 diets reached their maximum weight gain later in growing period, and ultimately had greater mature body weight.

A very strong correlation coefficient (Table 5) found to exist between the age of bird at inflection point and the body weight of mature birds indicating that body weight of chicks at AIP has an influence on mature body weight.

It seems that adding oils rich in PUFA to the diet affects performance of broilers. A positive effect of omega-3 fatty acids in diets of broilers (Elzobier et al., 2016) and turkey broilers (Lalev et al., 2021) on body weight of birds has been established. Fish, linseed and rapeseed oils containing omega 3 fatty acids (eicosapentaenoic-EPA) and docosa-

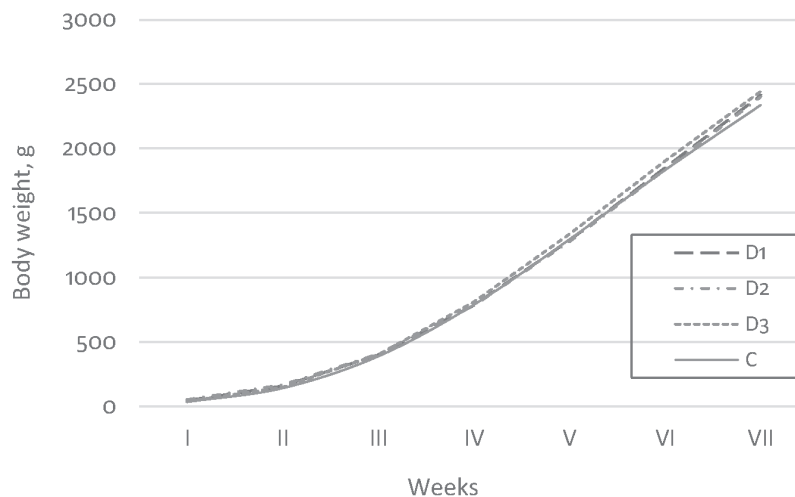


Figure 1 Gompertz growth curves of broilers fed different oils

Grafikon 1. Krivulja rasta brojlera, po Gompertz modelu, hranjenih različitim pokusnim smjesama

Table 3 Weekly body weights of broilers fed different experimental diets

Tablica 3. Tjelesna masa brojlera po tjednima hranjenih različitim pokusnim smjesama

Weeks / Tjedni	Diet / Obrok			
	C	D1	D2	D3
0	40.83±0.17 ^b	39.83±0.79 ^a	40.85±0.25 ^b	40.65±0.43 ^b
1	154.23±4.42 ^b	158.68±6.29 ^{ab}	162.05±1.69 ^a	157.83±4.12 ^{ab}
2	385.86±5.52 ^b	406.72±5.98 ^a	406.50±10.03 ^a	395.00±14.60 ^{ab}
3	818.13±8.07 ^a	819.69±9.27 ^a	819.69±7.86 ^a	829.06±7.10 ^a
4	1300.89±68.92 ^a	1262.81±37.45 ^a	1244.38±12.35 ^a	1296.29±28.15 ^a
5	1845.18±35.15 ^b	1895.18±38.26 ^{ab}	1884.38±58.81 ^{ab}	1921.47±7.79 ^a
6	2361.88±23.97 ^b	2445.27±44.06 ^a	2423.75±83.80 ^{ab}	2403.04±27.46 ^{ab}

a, b Data with different letters in superscript in each row are significantly different from each other ($P < 0.05$)

C = feeding a mixture containing 6 % sunflower oil; D1 = feeding a mixture containing 1% fish oil + 2% linseed oil + 3% rapeseed oil, D2 = feeding a mixture containing 1.5% fish oil + 1.5% linseed oil + 3% rapeseed oil, D3 = feeding a mixture containing 2% fish oil + 1% linseed oil + 3% rapeseed oil.

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Table 4 Growth curve parameters of broilers fed different experimental diets

Tablica 4. Parametri krivulja rasta brojlera hranjenih različitim pokusnim smjesama

	Diet/Obrok			
	C	D1	D2	D3
β_A	4197.01±511.75 ^b	4987.22±247.43 ^a	5170.63±498.08 ^a	4267.53±269.44 ^b
β_1	4.86±0.19 ^a	4.72±0.21 ^a	4.65±0.14 ^a	4.88±0.12 ^a
β_2	0.36±0.04 ^a	0.31±0.01 ^b	0.30±0.01 ^b	0.36±0.01 ^a
AIP	23.57±2.94 ^b	27.31±1.18 ^a	28.39±1.03 ^a	23.36±0.94 ^b
WIP	1822.74±222.25 ^b	2165.92±107.46 ^a	2245.58±216.31 ^a	1853.36±117.01 ^b
MWG	92.14±3.05 ^b	96.56±5.18 ^a	96.46±6.97 ^a	94.66±3.22 ^{ab}
R ²	1.000	0.999	0.999	0.999

a, b Data with different letters in superscript in each row are significantly different from each other ($P < 0.05$)

C = feeding a mixture containing 6 % sunflower oil; D1 = feeding a mixture containing 1% fish oil + 2% linseed oil + 3% rapeseed oil, D2 = feeding a mixture containing 1.5% fish oil + 1.5% linseed oil + 3% rapeseed oil, D3 = feeding a mixture containing 2% fish oil + 1% linseed oil + 3% rapeseed oil.

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Table 5 Estimated correlations of Gompertz growth curve parameters

Tablica 5. Koorelativne veze parametara krivulje rasta po Gompertz modelu

	β_A	β_1	β_2	AIP	WIP	MWG
β_A	1					
β_1	-0.982**	1				
β_2	-0.998**	-0.987**	1			
AIP	1.000**	-0.982**	-0.998**	1		
WIP	0.996**	-0.992**	-0.999**	0.996**	1	
MWG	0.882*	-0.786	-0.854	0.882*	0.833	1

** Correlation is significant at the 0.01 level (2-tailed).

*Correlation is significant at the 0.05 level (2-tailed).

hexaenoic acid-DHA), which are essential nutrients necessary for health and normal body functions as well as for immune response, have positive effect on metabolic processes in bodies of birds. In the long term, EPA/DHA has beneficial anti-inflammatory effect and helps restore damaged intestinal microflora of rats (Pusceddu et al., 2015), which could result in improved utilization of nutrients from diets.

Amount of added fish oil appears to play a role as higher concentrations of fish oil can be immunosuppressive (Krover and Klasing, 1997) or have adverse effect on ability to digest it. Fish oil has more 4-fold saturated long-chain fatty acids (>14 C atoms) than sunflower, rapeseed and linseed oil which are less digestible than fats rich in medium-chain fatty acids or unsaturated fatty acids (Smink, et al., 2008). This can explain lower predicted mature weight and earlier AIP in Gompertz nonlinear model of broilers fed D3 diet compared to D1 and D2 diets (Table 3), but still higher values compared to control diets. Navidshad (2009) reported decreasing final body weight and daily weight gain of broiler fed fish oil; however, Panda et al., (2016) did not find any adverse effect of incorporating fish oil in broilers' diets at concentrations of 2% and 3% on broiler's performances. Despite presence of linatine (pyridoxine antagonist) and anti-nutrient cyanogenic glycosides in the linseed meal adverse effect of including linseed oil on broilers performances (comparing to control group) were not observed. Reason for this is that by extraction over 90% of total cyanogenic compounds in ground linseed are reduced (Varga and Diosady, 1994). Also, a negative effect of including rapeseed oil into the diets should not be significant. There are reports that levels of full-fat canola or '00' rapeseed reaching 100 or even 150 g/kg diet did not result in a significant decline of broilers performances (Pietras et al., 2000).

CONCLUSIONS

Gompertz model can be very useful tool in describing pattern of broilers growth. The value of AIP varied depending on the nutritive composition of diets. Broilers fed oils rich in n-3 PUFA have higher values of age at inflection point as well as higher predicted mature weights. These differences may become more pronounced during prolonged broiler fattening (extensive system), which may also affect the economic valorisation of broiler fattening.

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SAŽETAK

Cilj ovog istraživanja je bio procijeniti obrazac rasta broilera hranjenih obrocima koji sadrže različita ulja. Podaci za ovu studiju dobiveni su iz pokusa koji je uključivao 160 neseksiranih jednodnevnih pilića Ross 308 provenijencije nasumično raspoređenih u četiri hranidbena tretmana ($n=40$) s pet ponavljanja u svakom od njih (osam ptica po ponavljanju) tijekom 42 dana. Hranjenje pokusnim obrocima uvedeno je u posljednjih 21 dana. Struktura pokusnih obroka (C, D1, D2 i D3) bila je ista ali s različitim količinama dodanih ulja. Kontrolni obrok sadržavao je 6 % suncokretovog ulja. D1, D2 i D3 obroci sadržavali su dodana ulja u sljedećim kombinacijama: 1 % ribljeg ulja + 2 % lanenog ulja + 3 % ulja repice (D1), 1,5 % ribljeg ulja + 1,5 % lanenog ulja + 3 % ulja repice (D2) i 2 % ribljeg ulja + 1 % lanenog ulja + 3 % ulja repice (D3). Svi su obroci bili izoenergetski i sadržavali su 0,015 % vitamina E. Gompertzov model je korišten za opisivanje rasta ptica, a na temelju podataka dobivenih iz tjednog vaganja. Model je pokazao visoku točnost s koeficijentom determinacije od preko 99,9 %. Asimptotska tjelesna masa odraslih ptica, specifična stopa rasta po tjednu, stopa eksponencijalnog opadanja početne specifične stope rasta, starost ptica u točki infleksije, tjelesna masa i maksimalni prirast u točki infleksije za svaki hranidbeni tretman podvrgnuti su analizi varijance. Utvrđen je pozitivan učinak omega-3 masnih kiselina na većinu ispitivanih parametara krivulje rasta.

Ključne riječi: brojleri, krivulja rasta, n-3 PUFA u obrocima