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Growth rate and metabolic parameters in young turkeys fed diets with different inclusion levels of methionine

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ABSTRACT. The study was conducted on 672 one-day-old female Hybrid Converter turkeys divided into 6 dietary treatments (7 replications per group) which differed in DL-methionine (DL-Met) supplementation levels. The Met content in diets between 1 – 4/5 – 8 weeks of feeding was as follows: 4.1/3.7, 4.7/4.1, 5.6/4.6, 6.1/5.0, 6.8/6.5 and 7.5/7.4 g · kg^{-1}. In groups 3 and 5, dietary Met levels corresponded to those recommended by NRC (1994) and B.U.T. (2012), respectively. The lowest Met content of the diet, without supplemental pure Met, caused a significant decrease in the bird body weight gains. In comparison with group 1, a considerable increase in dietary Met concentrations led to a decrease in the plasma levels of urea ($P = 0.042$), total protein and albumin ($P < 0.001$) in groups 4 – 6, and in the activity of γ-glutamyl transferase in all other groups ($P < 0.001$). The highest glutathione peroxidase (activity and the lowest lipid peroxide concentration in the turkeys' blood were noted in treatments 4 and 5. The lowest and the highest Met content significantly decreased ferric reducing ability of plasma values ($P < 0.001$ vs the other groups). Plasma immunoglobulin A concentrations were significantly ($P < 0.001$) higher in groups 1 and 3 than in the remaining groups. In conclusion, the diet without Met supplementation compromised the growth performance and antioxidant status of turkeys. Diets supplemented with Met according to B.U.T. (2012) recommendations or at levels slightly lower exerted the strongest antioxidant effect.

Introduction

Typical cereal-soyabean meal-based diets, without supplementation of amino acids (AA), have too low biological value of protein relative to the nutrient requirements of poultry. Methionine (Met) is a deficient AA. Its value in cereal and soyabean products is much lower than it is recommended for fast-growing birds (NRC, 1994). The risk of Met deficiency is even greater when the nutritional guidelines of turkey breeding companies (B.U.T., 2012; Hybrid Turkeys, 2013) are taken as a reference point.

Methionine participates in protein synthesis and it is involved in numerous functions including control of oxidative status and synthesis of acute-phase proteins. It may also act as a mediator affecting metabolism and cell functions (Tesseraud et al., 2009;

Jankowski et al., 2014). By participating in the synthesis of intracellular antioxidants, including glutathione and carnitine, sulphur-containing amino acids (SAA) can affect the body's antioxidant status (Fang et al., 2010). Hydrogen sulphide, supplied by SAA, is a precursor of glutathione which alleviates the effects of oxidative stress in cells (Kimura et al., 2010).

Methionine is also involved in the synthesis of immune system proteins, which is another biologically important function of this AA (Fang et al., 2010). In a recent experiment (Bouyeh, 2012) it was observed that, high levels of dietary lysine (Lys) and Met led to increase in blood lymphocytes and decrease in blood heterophils. The content of Met and cysteine (Cys) in the diet can be important in regulating gut growth and intestinal function (Bauchart-Thevret et al., 2009), as well as in stimulating the defensive functions of the poultry gastrointestinal tract (Ruth and Field, 2013).

The previous researches have shown that young broiler chickens do not require more than 5.0 g · kg^{-1} Met for optimum growth (Deng et al., 2007), but higher Met levels are needed to stimulate their immune responses (Bouyeh, 2012). The results of earlier studies (Kubińska et al., 2014, 2015a,b) have not provided a basis for establishing the dietary inclusion levels of Met which could stimulate antioxidant and immune mechanisms in growing chickens and turkeys.

The objective of this study was to verify the hypothesis that increasing dietary levels of Met can improve the turkey growth performance and/or exert antioxidant and immunological effects in the first stage of rearing, from 1 to 8 weeks of age.

Material and Methods

Birds, management and diets

The animal protocol used in this study was approved by the Local Ethics Committee (Olsztyn, Poland), and the study was carried out in accordance with EU Directive 2010/63/EU on the protection of animals used for scientific purposes.

In total 672 one-day-old female Hybrid Converter turkeys raised at the Research Laboratory of the Department of Poultry Science, University of Warmia and Mazury in Olsztyn (Poland) were used in this study. The birds were kept in pens on litter in a building with a controlled environment. The temperature and lighting programmes were consistent with the recommendations of Hybrid Turkeys (2013). The birds had free access to feed and water.

Table 1. Composition and nutrient concentrations in basal diets, g · kg^{-1}

Indices	Feeding period, weeks	
	1 – 4	5 – 8
Ingredients		
wheat	286.1	336.7
maize	200.0	150.0
soyabean meal	414.4	420.0
rapeseed	–	25.0
potato protein	40.0	–
soyabean oil	12.5	25.5
sodium sulphate	1.5	1.5
sodium chloride	1.7	1.5
limestone	16.7	15.8
monocalcium phosphate	20.0	15.9
L-lysine HCl	3.4	3.8
L-threonine	0.2	0.8
choline chloride	1.0	1.0
vitamin-mineral premix[1]	2.5	2.5
Nutritional value (analysed)		
ME[2], kcal · kg^{-1}	2750	2850[2]
crude protein	266	255
lysine	17.2	16.5
methionine	4.1	3.7
methionine + cysteine	8.1	7.6
threonine	10.4	10.1
arginine	16.5	16.6

[1] provided per kg of diet; IU: vit. A (all trans-retinol acetate) 13.000, vit. D_3 (cholecalciferol) 5.000; mg: vit. E (all-rac-α-tocopheryl acetate) 100, vit. K_3 4, vit. B_1 4, vit. B_2 15, vit. B_6 5, niacin 110, biotin 0.3, pantothenic acid 28, nicotinic acid 75, folic acid 3, Mn 160, Zn 160, Fe 90, Cu 16, I 3, Se 0.3; [2] ME – calculated metabolizable energy

The birds were randomly divided into 6 experimental groups with 7 replicates of 16 birds each. In group 1, turkeys were fed diets without DL-methionine (DL-Met) supplementation, formulated for weeks 1 – 4 and 5 – 8 (Table 1). In groups 2 – 6, the inclusion rates of DL-Met in diets were as follows: 0.6, 1.5, 2.0, 2.7, 3.4 g · kg^{-1} in the first feeding phase, and 0.4, 0.9, 1.3, 2.8, 3.7 g · kg^{-1} in the second feeding phase. In groups 3 and 5, the total Met content of diets (Table 2) corresponded to the levels recommended by NRC (1994) and B.U.T. (2012), respectively. The final concentrations of amino acids, including Met, in the diets were verified analytically (Table 1). The total sulphur-containing amino acids (SAA) content of the basal diet was 8.1 g · kg^{-1} and 7.6 g · kg^{-1} in the first and second feeding phase, respectively. In diets 2 – 6, the SAA content was increased by the addition of DL-Met.

Growth trial and sample collection

Daily feed intake (DFI), body weight gain (BWG) and feed conversion ratio (FCR) were determined in the periods of 1 – 4 and 5 – 8 weeks.

Table 2. Methionine content of diets in feeding periods of weeks 1 – 4 and 5 – 8

Methionine content	Treatment					
	$M_{4.1/3.7}$	$M_{4.7/4.1}$	$M_{5.6/4.6}$	$M_{6.1/5.0}$	$M_{6.8/6.5}$	$M_{7.5/7.4}$
Met content, g · kg^{-1}						
weeks 1 – 4	4.1	4.7	5.6	6.1	6.8	7.5
weeks 5 – 8	3.7	4.1	4.6	5.0	6.5	7.4
Relative to NRC (1994), %						
weeks 1 – 4	–27	–16	–2	+11	+24	+36
weeks 5 – 8	–20	–10	+2	+11	+44	+55

A pen of 16 birds was considered as an experimental unit. At the end of the trial, seven birds representing the average body weight of each group (one bird from each replication) were selected to determine blood haematological and biochemical parameters, antioxidant capacity and the immune status. Blood samples were collected from the wing vein.

Laboratory analysis

Amino acids were analysed by HPLC (Beckman 6300, Beckman Instruments Corp., Palo Alto, CA, USA) after 24 h of hydrolysis with 6 N hydrochloric acid at 120 °C, using the methodology recommended by NFIA (1991). For the analysis of SAA, performic acid oxidation treatment was applied to prevent cysteine destruction. Phenol was added to prevent partial destruction of phenylalanine, histidine and arginine.

Immediately after collection, blood samples were aliquoted into test tubes containing heparin as an anticoagulant. The samples were centrifuged at 3 000 *g* for 10 min. The obtained plasma was stored at −20 °C until analysis.

The content of haemoglobin (Hb) and haematocrit (Ht) was determined using an automatic haematology analyser (Abacuss Junior Vet, Diatron, Hungary).

The content of triacyloglycerols (TAG), total cholesterol (TC), uric acid (UA), plasma total protein (TP), albumin (ALB), creatinine (CREAT), and the activities of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), creatine kinase (CK), lactate dehydrogenase (LDH) and gamma-glutamyl transferase (GGT) were measured using an automatic biochemical analyser (Plasma Diagnostic Instruments Horiba, Kyoto, Japan).

As described previously (Ognik and Wertelecki, 2012), the following indicators of the redox status were determined in the turkey blood plasma or erythrocytes: the concentrations of vitamin C, lipid peroxide (LOOH), the sum of reduced glutathione and oxidized glutathione (GSH + GSSG), the activities of catalase (CAT), superoxide dismutase (SOD) and glutathione peroxidase (GPx), and the ferric reducing ability of plasma (FRAP).

The turkey immune system response was determined based on the plasma levels of immunoglobulin A (IgA) and interleukin (IL)-6 using the Bigenet UMV340 blood cell reader, USCN Life Science Inc. kits (IL-6 and IgA) (Plasma Diagnostic Instruments Horiba, Kyoto, Japan).

Statistical analysis

One-way analysis of variances (ANOVA) was performed with the use of STATISTICA ver. 10.0 PL software (StatSoft Polska Sp z o.o., Kraków, Poland). The significance of differences between means in groups was determined by Duncan's test.

Results

All diets were formulated using the same basal diet and contained the same amounts of nutrients except Met content (Table 1). In the experimental diets, Met levels ranged from 4.1 to 7.5 g · kg^{-1} in weeks 1 – 4, and from 3.7 to 7.4 g · kg^{-1} in weeks 5 – 8 (Table 2).

Different dietary levels of Met had a significant influence on the turkey growth performance (Table 3). In the first feeding phase, significant differences in daily feed intake (DFI) were noted between treatment $M_{4.1/3.7}$ vs all other treatments, and between treatments $M_{4.7/4.1}$ and $M_{7.5/7.4}$ ($P \leq 0.001$). Similar differences were observed in body weight gain (BWG) ($P \leq 0.001$), whereas significant differences in feed conversion ratio (FCR) were found only between $M_{4.7/4.1}$ vs the other treatments ($P = 0.001$). In the second feeding phase and over the entire experimental period, DFI and BWG were significantly lower in $M_{4.7/4.1}$ than in the remaining treatments ($P \leq 0.001$), whereas no differences in FCR were found between the compared treatments.

The dietary treatments had no significant effect on the levels of haematocrit, haemoglobin, total cholesterol, triacylglycerols and uric acid or the activity of ALT and CK but they affected other examined blood biochemical parameters (Table 4). As compared with the lowest level of dietary Met (treatment $M_{4.1/3.7}$), the dose of 6.1/5.0 g · kg^{-1} or higher (treatments $M_{6.1/5.0}$ – $M_{7.5/7.4}$) decreased the plasma concentrations of urea ($P \leq 0.05$), total protein ($P \leq 0.001$) and albumin ($P \leq 0.001$). In regard to other biochemical parameters of blood plasma, no differences were found between turkeys

Table 3. Growth performance of turkeys fed diets with different methionine contents[1]

Indices	Treatment[2]						SEM	*P*
	$M_{4.1/3.7}$	$M_{4.7/4.1}$	$M_{5.6/4.6}$	$M_{6.1/5.0}$	$M_{6.8/6.5}$	$M_{7.5/7.4}$		
Daily feed intake, g per bird								
weeks: 1 – 4	51.1^{c}	58.5^{b}	60.1^{ab}	59.9^{ab}	60.2^{ab}	61.96^{a}	0.610	0.001
5 – 8	160.3^{b}	187.2^{a}	188.2^{a}	182.0^{a}	190.4^{a}	189.1^{a}	2.075	0.001
1 – 8	105.3^{b}	122.9^{a}	124.2^{a}	120.6^{a}	125.3^{a}	125.5^{a}	1.304	0.001
Body weight gain, kg per bird								
weeks: 1 – 4	0.99^{c}	1.18^{b}	1.20^{ab}	1.22^{ab}	1.21^{ab}	1.24^{a}	0.014	<0.001
5 – 8	2.54^{b}	2.95^{a}	2.93^{a}	2.85^{a}	2.98^{a}	2.95^{a}	0.029	<0.001
1 – 8	3.53^{b}	4.13^{a}	4.13^{a}	4.07^{a}	4.20^{a}	4.19^{a}	0.040	<0.001
Feed conversion ratio, kg · kg^{-1} of BW[3]								
weeks: 1 – 4	1.45^{a}	1.39^{b}	1.40^{b}	1.39^{b}	1.39^{b}	1.40^{b}	0.005	0.001
5 – 8	1.73	1.72	1.80	1.76	1.71	1.72	0.013	0.379
1 – 8	1.65	1.62	1.68	1.65	1.62	1.63	0.009	0.348

[1]data representing mean values of 7 pens per treatment; [2]see Table 2; [3] BW – body weight; [abc] – means with different superscripts within a row are significantly different at $P < 0.05$; SEM – standard error of mean

Table 4. Haematological and biochemical parameters in the blood of turkeys[1]

Indices	Treatment[2]						SEM	*P*
	$M_{4.1/3.7}$	$M_{4.7/4.1}$	$M_{5.6/4.6}$	$M_{6.1/5.0}$	$M_{6.8/6.5}$	$M_{7.5/7.4}$		
Content in the blood								
Ht, %	34.1	32.7	33.3	32.5	31.5	32.7	0.433	0.655
Hb, g · dl^{-1}	8.93	8.36	9.45	8.34	8.28	8.57	0.154	0.194
Content in the blood plasma								
TC, mmol · l^{-1}	2.66	2.54	2.74	2.52	2.46	2.38	0.060	0.135
TAG, mmol · l^{-1}	0.737	0.663	0.920	0.871	0.837	0.876	0.045	0.158
UREA, mmol · l^{-1}	0.783^{a}	0.503^{ab}	0.581^{ab}	0.453^{b}	0.50^{b}	0.462^{b}	0.042	0.042
TP, g · dl^{-1}	37.7^{a}	34.3^{abc}	35.3^{ab}	32.1^{bcd}	29.9^{cd}	28.2^{d}	0.812	0.001
ALB, g · dl^{-1}	205^{a}	188^{bc}	201^{ab}	177^{c}	175^{c}	172^{c}	2.863	0.001
CREAT, µmol · l^{-1}	7.46^{a}	5.21^{ab}	4.40^{ab}	3.57^{b}	5.84^{ab}	6.13^{ab}	0.430	0.019
UA, µmol · l^{-1}	267	300	329	260	261	267	11.32	0.127
Activity of enzymes in the blood plasma, U · l^{-1}								
AST	249^{a}	201^{bc}	234^{ab}	191^{c}	208^{ab}	147^{d}	7.347	<0.001
ALT	9.23	8.79	6.69	7.39	8.01	7.47	0.420	0.137
ALP	1471^{ab}	1318^{b}	1772^{a}	1287^{b}	1521^{ab}	1174^{b}	52.05	<0.001
CK	1813	1998	2599	2079	1433	1876	113.6	0.080
LDH	648^{ab}	656^{ab}	762^{a}	661^{ab}	742^{ab}	564^{b}	20.19	0.009
GGT	6.64^{a}	3.04^{b}	3.60^{b}	2.26^{b}	3.20^{b}	3.04^{b}	0.359	<0.001

[1]data representing mean values of 7 birds per treatment; [2]see Table 2; Ht – haematocrit, Hb – haemoglobin, TC – total cholesterol; TAG – triacyloglycerols; UREA – urea, TP – total protein, ALB – albumin, CREAT – creatinine, UA – uric acid, AST – aminotransferase; ALT – alanine aminotransferase; ALP – alkaline phosphatase; CK – creatine kinase; LDH – lactate dehydrogenase; GGT – gamma-glutamyl transferase; [abc] – means with different superscripts within a row are significantly different at $P < 0.05$; SEM – standard error of mean

receiving the lowest level of dietary Met and the level recommended by NRC (treatments $M_{4.1/3.7}$ and $M_{5.6/4.6}$, respectively). In comparison with treatment $M_{5.6/4.6}$, the highest level of Met in treatment $M_{7.5/7.4}$ decreased the activity of AST, ALP (both $P \leq 0.001$) and LDH ($P \leq 0.01$).

Turkeys fed diets with a medium level of Met in treatments $M_{5.6/4.6}$ and $M_{6.1/5.0}$ had the highest plasma vitamin C concentrations and significant differences were noted in relation to groups fed diets with the lowest and highest Met content (treatments $M_{4.1/3.7}$ and $M_{7.5/7.4}$, respectively) (Table 5). There was no difference between $M_{6.1/5.0}$ and $M_{5.6/4.6}$. The CAT activity was the lowest in the blood plasma of turkeys from treatment group $M_{5.6/4.6}$ with a statistically significant difference in comparison with all other groups except $M_{6.1/5.0}$. No significant differences in SOD activity were found between treatments, whereas GPx activity was significantly higher ($P \leq 0.001$) in treatments $M_{6.1/5.0}$ and $M_{6.8/6.5}$

Table 5. Redox status in the blood of turkeys[1]

Indices	Treatment[2]						SEM	*P*
	$M_{4.1/3.7}$	$M_{4.7/4.1}$	$M_{5.6/4.6}$	$M_{6.1/5.0}$	$M_{6.8/6.5}$	$M_{7.5/7.4}$		
Content in the blood plasma, μmol · l^{-1}								
vit. C	4.71^{c}	4.89^{ab}	4.92^{a}	4.96^{a}	4.82^{abc}	4.77^{c}	0.017	<0.001
GSH+GSSG	0.315	0.360	0.345	0.342	0.404	0.357	0.011	0.324
FRAP	722^{b}	931^{a}	1132^{a}	1125^{a}	1001^{a}	677^{b}	39.13	<0.001
LOOH	23.2^{ab}	24.9^{a}	21.3^{a}	16.6^{c}	12.9^{d}	18.9^{bc}	0.757	<0.001
Activity of enzymes in the blood erythrocytes, U · gHb^{-1}								
CAT	736^{a}	683^{a}	543^{b}	638^{ab}	694^{a}	719^{a}	16.39	0.004
SOD	247	233	218	211	194	201	6.029	0.095
GPx	135^{b}	161^{b}	152^{b}	214^{a}	229^{a}	155^{b}	6.425	<0.001

[1]data representing mean values of 7 birds per treatment; [2]see Table 2; Hb – haemoglobin, CAT – catalase; SOD – superoxide dismutase; GPx – glutathione peroxidase; GSH+GSSG – total glutathione; LOOH – peroxides; FRAP – ferric reducing ability of plasma; ^{abc} – means with different superscripts within a row are significantly different at $P < 0.05$; SEM – standard error of mean

vs the other treatments. Significant differences were observed selected groups with respect to the plasma concentrations of LOOH. Significantly lower LOOH concentrations were noted in treatments $M_{6.1/5.0}$ and $M_{6.8/6}$ vs treatments $M_{4.1/3.7}$ – $M_{5.6/4.6.}$, and in treatment $M_{7.5/7.4}$ vs treatments $M_{4.7/4.1}$ and $M_{5.6/4.6}$ ($P \leq 0.001$). Turkeys from treatment groups $M_{4.1/3.7}$ and $M_{7.5/7.4}$ were characterized by similar FRAP values, which were significantly ($P \leq 0.001$) lower than in the remaining groups.

Significantly ($P \leq 0.001$) higher IgA concentrations were noted in treatments $M_{4.1/3.7}$ and $M_{5.6/4.6}$ vs the other groups. The dietary treatments had no influence on IL-6 concentrations in the turkey blood plasma (Table 6).

Table 6. Concentrations of immunoglobulin A and interleukin 6 in the blood of turkeys[1]

Indices	Treatment[2]						SEM	*P*
	$M_{4.1/3.7}$	$M_{4.7/4.1}$	$M_{5.6/4.6}$	$M_{6.1/5.0}$	$M_{6.8/6.5}$	$M_{7.5/7.4}$		
IgA, μg · ml^{-1}	2.37^{a}	2.01^{b}	2.24^{a}	1.93^{b}	1.87^{b}	2.04^{b}	1.005	<0.001
IL-6, pg · ml^{-1}	20.8	18.2	19.0	16.8	17.8	23.2	0.036	0.114

[1]data representing mean values of 7 birds per treatment; [2]see Table 2; IgA – immuno-globulin A, IL-6 – interleukin 6; ^{ab} means with different superscripts within a row are significantly different at $P < 0.05$; SEM – standard error of mean

Discussion

Dietary Met content and the growth performance of turkeys. In many studies investigating the effects of diets with graded Met levels, the points of reference were NRC (1994) recommendations (Deng et al., 2007). In some experiments (Shen et al., 2015), diets without supplemental DL-Met contained approximately 3.0 g · kg^{-1} of this amino acid, i.e. less than 60% of the levels recommended by NRC (1994) in the first feeding phase. In our study, Met deficiency in the basal diet was much lower, at approximately 27% and 20% when compared to NRC (1994) recommendations for the first and second feeding phase, respectively. In groups with higher dietary Met levels, the content of this amino acid in turkey diets in the first feeding phase was higher than that recommended by NRC (1994), but it matched or was close to B.U.T. (2012) guidelines (groups 5 and 6, respectively).

Numerous experiments with broiler chickens have shown that deficiencies in essential amino acids, particularly Met, considerably decrease feed intake (Deng et al., 2007; Rubin et al., 2007; Hadinia et al., 2014). In our study, lower feed intake was noted only in turkeys fed diet not supplemented with DL-Met. Similarly to experiments with chickens (Rubin et al., 2007; Bouyeh, 2012), the diet without DL-Met deteriorated feed conversion, but only in the first month of feeding. In the previous experiments with turkeys, diets without DL-Met supplementation had no adverse effect on the growth performance of four-week-old birds (Kubińska et al., 2014), but they decreased the BWG of eight-week-old birds (Kubińska et al., 2015b).

In our study, similarly to earlier experiments with chickens (Deng et al., 2007; Rubin et al., 2007; Bouyeh, 2012) and turkeys (Kubińska et al., 2015b), the diet containing no supplemental DL-Met contributed to a decrease in BWG, in comparison with birds fed diets supplemented with Met at levels being in line with NRC (1994) recommendations. Dietary Met concentrations higher than the levels recommended by NRC (1994) and B.U.T. (2012) did not improve the turkey BWG. In a study by

Lemme et al. (2005), diets with graded Met levels, at 90%, 100% and 120% of the SAA requirements of turkeys, contributed to a linear increase in BWG, with no improvement in feed conversion. Our findings indicate that increasing dietary Met concentrations above NRC (1994) recommendations does not significantly improve the growth performance of young turkeys (1 to 8 weeks of age).

Blood biochemical parameters. In a recent experiment performed on young turkeys (Kubińska et al., 2014) fed diets with a low, medium and high Met content (4.5, 6.0 and 7.1 g · kg^{-1}, respectively) during the first four weeks of age no significant differences were noted in the serum biochemical parameters of birds. In a few studies investigating the blood biochemical parameters of chickens fed Met-supplemented diets, dietary Met concentrations had various effects on serum total protein content. Hadinia et al. (2014) demonstrated that increased dietary Met levels led to an increase in globulin concentrations, causing no changes in total protein levels in broiler chickens. Similar results were reported by Adeyemo et al. (2010), whereas Attia et al. (2005) found that Met-supplemented diets increased serum total protein and albumin levels in the slow-growing chickens. The results of the cited studies do not explain the trend noted in our experiment where the highest plasma levels of total protein, albumin, urea and creatinine were determined in the group of turkeys fed diets with the lowest Met content. This could be the result of the unbalanced amino acid composition of protein in diets not supplemented with DL-Met. The BWG of turkeys in this group was lower than in the other groups although they received the same amount of dietary protein. Less protein was utilized for weight gain, which led to an increased accumulation of nitrogen compounds in the blood of those birds, including synthesized urea. Enhanced urea synthesis puts a burden on the liver, as manifested by elevated activity levels of livers enzymes – AST and GGT. Such an interpretation is consistent with the findings of other authors (Donsbough et al., 2010) who demonstrated that dietary supplementation with Met reduced serum urea concentrations in broiler chickens. According to Donsbough et al. (2010), the serum concentrations of urea, ammonia and uric acid are good indicators of amino acid utilization in broiler chicken diets. In our study, dietary Met levels had no effect on the plasma concentrations of uric acid, and differences in the activities of selected enzymes (ALP and LDH) did not correspond to differences in the Met content in turkey diets.

Blood redox and immunological status. Recent research has revealed that dietary supplementation of Met can alleviate oxidative stress in broiler chickens (Swennen et al., 2011; Chen et al., 2013; Shen et al., 2015). In our earlier study on 9- to 16-week-old turkeys (Jankowski et al., 2016), a diet with the highest Met content did not improve the antioxidant or immune status of birds. The results of our study, in which younger turkeys were involved, are inconclusive and ambiguous, but suggest that diets supplemented with Met at levels that matched or were slightly lower than B.U.T. (2012) recommendations exerted antioxidant effects, whereas diets with higher Met concentrations had prooxidative effects. The above was confirmed by higher GPx activity and lower LOOH concentrations, accompanied by high plasma FRAP values in turkeys from groups 4 and 5, and less desirable values of those indicators in group 6. Both the highest and lowest dietary Met levels exerted similar effects on many parameters (concentrations of vitamin C and LOOH, activity of CAT and GPx, FRAP values). Ingenbleek (2011) demonstrated that dietary Met deficiency induced oxidative stress in vegetarian subjects consuming deficient amounts of Met. In another experiment (Castellano et al., 2015), a decrease in blood FRAP values was noted in piglets fed Met-deficient diets.

The GSH/GSSG ratio in the liver is an important indicator of oxidative stress (Serru et al., 2001). Other authors (Chen et al., 2013) reported that the GSH/GSSG ratio increased with increasing inclusion levels of dietary Met in broiler chickens. Such a correlation was not observed in the blood plasma of turkeys in our study.

Dietary Met has been found to be a major contributor to the synthesis of immune system proteins, including IgA – the most abundant immunoglobulin (Wu et al., 2012). A similar function may be performed by vitamin E, the main antioxidant in feed. Muir et al. (2002) reported an increase in the serum concentrations of IgA and improved chicken immunity, which resulted from increased production of IgA in the intestinal tract, stimulated by the addition of vitamin E to the diet. In the present experiment, higher dietary Met levels did not lead to an increase in IgA concentrations in the turkey blood plasma. An increase in the antioxidant capacity of blood plasma, observed in selected groups, had no influence on IgA levels.

Conclusions

Diets not supplemented with DL-methionine, with total sulphur-containing amino acids content (8.1 and 7.6 g · kg^{-1}) and low Met content (4.1 and 3.7 g · kg^{-1}) in the first and second feeding phase,

respectively, decrease the body weight gains and deteriorate the metabolic parameters of turkeys, including the plasma antioxidant status. A slight increase in dietary Met levels (4.7 g · kg^{-1} in weeks 1 – 4 and 4.1 g · kg^{-1} in weeks 5 – 8, respectively) is sufficient to increase the growth rate of turkeys, whereas higher levels of Met (6.8 and 6.5 g · kg^{-1}, respectively) are required to increase the plasma concentrations of immunoglobulins and to reduce oxidative stress.

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