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# Influence of amidrazones – 5-oxo-1,2,4-triazine derivative addition on the level of biochemical blood markers and chemical composition of turkey hens tissues

Wpływ dodatku pochodnej amidrazonów – 5-okso-1,2,4-triazyny na poziom wskaźników biochemicznych krwi oraz skład chemiczny tkanek indyczek

Summary. The objective of this study was to determine the effect of 5-oxo-1,2,4- triazine derivative addition to water for turkey hens on the level of their biochemical blood markers and chemical composition of their tissues. The study was conducted on 160 medium-heavy British United Turkey (BUT) 9 hens divided at random and equally into four groups, 40 turkey hens each (two replications  $\times$  20 birds). Since the  $6^{th}$  till the  $16^{th}$  weeks of life, the birds were kept in pens on straw litter. Group I served as a control (T0) and received drinking water without experimental additives. The turkey hens from experimental groups (T15, T30, T45) were administered three various doses of 5-oxo-1,2,4- triazine dissolved in 2 ml of ethanol with drinking water, i.e.: 15 μg kg b.w./day in group T15, 30 µg/kg b.w./day in group T30, and 45 µg/kg b.w./day in group T45. The additives examined were administered to the birds with drinking water for the period of 28 days. Four weeks of amidrazones derivative - 5-oxo-1,2,4- triazine - administration to the birds were followed by a two-week break in supplementation during which the turkey hens of all groups received only pure water for drinking. After this two-week-long period, the additives were again administered to the birds, in doses as at the beginning of the experiment. At the end of the 4<sup>th</sup>, 6<sup>th</sup> and 10<sup>th</sup> weeks of observations, blood was sampled from the wing vein of the birds for biochemical analyses. The biochemical assays were carried out for levels of: AST, ALT, ALP, TP, GLU, CHOL, HDL, TG, and macroelements (K<sup>+</sup>, Na<sup>+</sup>, Ca<sup>+2</sup>, Mg<sup>+2</sup>). On termination of rearing (16<sup>th</sup> week of life), the birds were slaughtered. The chemical composition of meat was determined in samples of breast, thigh, and shank muscles as well as in the liver. The application of various doses of 5-oxo-1,2,4- triazine derivative had no significant effect on the levels of the analyzed biochemical markers of turkey hens blood. An exception was the increased calcium content observed in the 11th week of life. The administration of 5-oxo-1,2,4- triazine to turkey hens from groups T15, T30 and T45 contributed to their better mean body weight gains (by ca. 6.4%) as compared to the control birds. The addition of various doses of 5-oxo-1,2,4- triazine to drinking water was observed to decrease the content of crude fat in breast muscles and in liver, which increases the dietetic value of turkey hens

Key words: turkey hens, blood, tissues, metabolic markers

#### INTRODUCTION

Poultry feeding involves a constant search for additives with immunostimulatory, antioxidative and rearing performance-enhancing properties. In spite of the fact that natural biostimulants are on the top of these additives, still much attention is paid to compounds produced through chemical synthesis. These are often antibiotic-like compounds, like e.g. bacteriophages or compounds with antimycotic and antiviral activity, with promising expected outcomes. Owing to various pharmacological activities of a newly-synthesized derivative of amidrazones – 5-oxo-1,2,4-triazine – a growing interest has been observed in the feasibility of its application as a feed additive for animals. In addition, this compound has been demonstrated not to inhibit the growth of bacteria and fungi constituting the flora of the gastrointestinal tract of man [Modzelewska-Banachiewicz and Szcześniak 2001]. A lack of the toxic effects of 5-oxo-1,2,4-triazine has been shown in the *in vitro* study on kidney cultures of green monkey by Truchliński et al. [2000]. In turn, a study conducted with turkeys where two derivatives of amidrazones were used as feed additives (derivative of 1,2,4-triazole and 5-oxo-1,2,4-triazine) demonstrated satisfactory results including increased levels of immune markers [Sembratowicz et al. 2004, Ognik and Sembratowicz 2009]. Furthermore, markers of the antioxidative system were observed to be stimulated in blood plasma of turkey hens receiving one of the amidrazones derivatives: derivative of 1,2,4-triazole [Ognik et al. 2004].

In view of the above findings, the objective of the undertaken study was to determine the effect of 5-oxo-1,2,4- triazine derivative addition to water for turkey hens on the level of their biochemical blood markers and chemical composition of their tissues.

# MATERIAL AND METHODS

The study was conducted on 160 medium-heavy British United Turkey (BUT) 9 hens divided at random and equally into four groups, 40 turkey hens each (two replications × 20 birds). Since the 6<sup>th</sup> till the 16<sup>th</sup> week of life, the birds were kept in pens on straw litter. The birds were reared under standard zoohygienic conditions, optimal for slaughter turkeys fattening. Over the experimental period, the turkey hens of all groups were receiving all-mash feed mixtures produced by Provimi Polska (Tab. 1), following a programme covering 5 feeding periods. The content of basic nutrients in the feed mixtures corresponded to current recommendations of Nutrient Requirements for Poultry (Normy... 2005). Group I served as a control (TO) and was receiving drinking water without experimental additives. The turkey hens from experimental groups (T15, T30, T45) were administered three various doses of 5-oxo-1,2,4-triazine (methyl ester of 2-[5-oxo-3-(2-pyridyl)-4-phenyl-1,4,5,6-tetrahydro-1,2,4-triazine-6-ylideno] acetic acid)

dissolved in 2 ml of ethanol with drinking water, i.e.: 15 μg/kg b.w./day in group T15, 30 μg/kg b.w./day in group T30, and 45 μg/kg b.w./day in group T45. The additives examined were administered to the birds with drinking water for the period of 28 days. The additives were administered to the birds in the morning hours with a small quantity of water, afterwards the birds were receiving pure water without additives according to daily requirements for drinking water. This mode of administration assured the appropriate dose of the feed additive provided to the birds. Four weeks of amidrazones derivative 5-oxo-1,2,4-triazine – administration to the birds were followed by a two-week break in supplementation during which the turkey hens of all groups were receiving only pure water for drinking. After this two-week break, the additives were again administered to the birds, in doses as at the beginning of the experiment. At the end of the 4th, 6th and 10<sup>th</sup> week of observations, blood was sampled from the wing vein of the birds for biochemical analyses. Monotests by Cormay company and kinetic method were applied to determine activities of the following enzymes in blood plasma of turkey hens: asparagine aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP). Using monotests by Cormay company, blood samples of turkey hens were also analyzed for contents of total protein (TP) glucose (GLU), total cholesterol (CHOL), HDL cholesterol fraction, and triacylglycerols (TG). In addition, blood plasma, meat tissues and liver tissues were determined for contents of selected macroelements (K<sup>+</sup>, Na<sup>+</sup>, Ca<sup>+2</sup>, Mg<sup>+2</sup>) with the atomic absorption spectrometry method using an AAS apparatus.

Once rearing (16<sup>th</sup> week of life) followed by 12-h fasting were completed, the birds (40 birds from each experimental group) were slaughtered. The slaughter and simplified dissection were conducted following recommendations by Faruga and Jankowski [2000]. The chemical composition of meat was determined in samples of breast, thigh, and shank muscles as well as in liver according to AOAC procedures [2000].

Numerical data achieved were subjected to a statistical analysis, using Statistica ver. 5 software, with the one-way analysis of variance (ANOVA) test, at the significance level of 0.05.

### RESULTS AND DISCUSSION

A significant element in the evaluation of feed additives efficacy in animal feeding is the course of metabolic process, which is manifested in, among other things, changes in biochemical blood markers. Values achieved in the reported study for the respective blood markers of turkey hens were presented in Table 2. Data collated therein demonstrated 5-oxo-1,2,4-triazine doses administration to have no effect on glucose (GLU) nor triglycerides levels in blood of turkey hens examined. In turn Krauze [2007b], who was administering a feed additive with a similar chemical structure, i.e. 1,2,4-triazole derivative, to turkey hens with drinking water, recorded a decreased blood level of glucose. It should be emphasized, however, that the values noted in our study for contents of glucose, total protein, cholesterol HDL cholesterol fraction, uric acid, triglycerides, aminotransferases and alkaline phosphatase in blood of the turkey hens receiving 5-oxo-1,2,4-triazine correspond with results of other authors achieved in experiments with turkeys [Krasnodębska-Depta and Koncicki 2000, Sembratowicz 2004], that are adopted as physiological values for that group of animals. Hence, it may be concluded that the addition of 5-oxo-1,2,4-triazine to water did not contribute to the incidence of the pathological

Table 1. Nutrient content of the standard diets Tabela 1. Zawartość pokarmowa standardowych mieszanek

Ingredient – Składnik	Grower II	Grower III	Finisher I	
(Feeding period –	(6–9 week of age (10–12 week of age		,	
okres żywienia)	<ul> <li>6–9 tydzień życia)</li> </ul>	10–12 tydzień życia)	14–16 tydzień życia)	
Maize meal (%)				
Mączka kukurydziana (%)	23.8	35.2	47.4	
Wheat bran (%)	30.0	25.0	25.0	
Otręby pszenne (%)	30.0	25.0	23.0	
Soybean meal 46% protein (%)	38.8	32.7	20.4	
Śruta sojowa 46% białka (%)	30.0	32.7		
Soybean meal 45% protein (%)				
Śruta sojowa 45% białka (%)	-	-	-	
Fish meal 60% (%)				
Mączka rybna 60% (%)	-	-	-	
Fodder chalk (%)				
Kreda pastewna (%)	1.7	1.4	1.5	
Soybean oil (%)		2.0	2.0	
Olej sojowy (%)	2.5	3.0	3.0	
Cytromix Plus <sup>1</sup> (%)	0.2	0.2	0.2	
Farmix <sup>2</sup> (%)	3.0	3.0 2.5 2.5		
	rient composition – S	kładniki odżywcze		
CP (%)	24.5	22.0	17.5	
ME ( kcal kg <sup>-1)</sup>	2913	3007	3129	
Crude fibre (%)				
Popiół surowy (%)	2.72	2.71	2.7	
Lysine (%)				
Lizyna (%)	1.57	1.38	1.17	
Methionine + cysteine (%)				
Metionina + cysteina (%)	0.88	0.79	0.70	
Tryptophan (%)			0.19	
Tryptofan (%)	0.27	0.23		
Arginine (%)				
Arginia (%)	1.50	1.32	0.98	
Calcium (%)				
Wapń (%)	1.17	1.06	0.94	
Phosphorus available (%)				
Fosfor przyswajalny (%)	0.59	0.57	0.47	
Sodium (%)				
Sód (%)	0.15	0.15	0.15	
DOU (70)				

<sup>&</sup>lt;sup>1</sup>Cytromix Plus – citric acid, fumaric acid, phosphoric acid (62%)

 $<sup>^2\</sup>text{Farmix}-\text{mineral}$  and vitamin premix provided the following per kilogram of diet - 433333.0 IU of vitamin A; 133333.0 IU of vitamin  $D_3;$  73.3 mg of vitamin  $K_3;$  100.0 mg of vitamin  $B_1;$  291.7 mg of riboflavin; 175.0 mg of vitamin  $B_6;$  0.9 mg of vitamin  $B_{12};$  58.3 mg of folic acid; 10.5 mg of biotin; 2182.0 mg of niacin; 13333.0 mg of choline; 4 200 mg of calcium pantothenicum; 4 000 mg of Mn; 2 666 mg of Zn; 1 666 mg of Fe; 833 mg of Cu; 26 mg of I; 10 mg of Se; 6,7 mg of Co; 13 g of Ca; 15.5 g of P

Table 2. Level of biochemical markers in blood of turkey hens receiving the addition of 5-oxido-1,2,4-triazine

Tabela 2. Poziom wskaźników biochemicznych we krwi indyczek otrzymujących dodatek pochodnej 5-okso-1,2,4-triazyny

Parameter	Week of life	Experimental groups – Grupy doświadczalne					
Cecha Tydz. życia		ТО	T15	T30	T45	SEM	
TP g dl <sup>-1</sup>	9	$4.30 \pm 0.5$	$4.52 \pm 0.71$	$4.49 \pm 0.75$	$4.37 \pm 0.38$	0.12	
	11	$5.35 \pm 0.75$	$4.86 \pm 0.85$	$5.12 \pm 0.42$	$5.26 \pm 0.66$	0.14	
	15	$6.16^{a} \pm 0.82$	$5.06^{b} \pm 0.13$	$5.47^{ab} \pm 0.51$	$5.21^{b} \pm 0.83$	0.16	
	$\overline{\mathbf{x}}$	$5.27 \pm 0.69$	$4.81 \pm 0.56$	$5.02 \pm 0.56$	$4.94 \pm 0.62$	0.14	
	9	$12.7 \pm 0.65$	$12.7 \pm 0.61$	$12.8 \pm 0.96$	$12.9 \pm 0.94$	0.19	
GLU	11	$13.2 \pm 1.05$	$13.1 \pm 0.78$	$12.9 \pm 1.06$	$12.8 \pm 1.12$	0.21	
mmol l <sup>-1</sup>	15	$15.4 \pm 0.85$	$14.9 \pm 2.13$	$15.1 \pm 1.31$	$15.2 \pm 1.64$	0.32	
	$\overline{\mathbf{x}}$	$13.76 \pm 0.85$	$13.56 \pm 1.17$	$13.6 \pm 1.11$	$13.63 \pm 1.23$	0.24	
	9	$1.01 \pm 0.14$	$0.88 \pm 0.10$	$0.99 \pm 0.22$	$0.96 \pm 0.15$	0.03	
TG	11	$1.39 \pm 0.18$	$1.45 \pm 0.18$	$1.43 \pm 0.15$	$1.40 \pm 0.14$	0.03	
mmol 1 <sup>-1</sup>	15	$1.34 \pm 0.14$	$1.47 \pm 0.18$	$1.44 \pm 0.21$	$1.46 \pm 0.13$	0.03	
	$\overline{\mathbf{x}}$	$1.24 \pm 0.15$	$1.26 \pm 0.15$	$1.28 \pm 0.19$	$1.27 \pm 0.14$	0.03	
	9	$3.29 \pm 0.39$	$3.28 \pm 0.41$	$3.31 \pm 0.31$	$3.32 \pm 0.37$	0.07	
CHOL	11	$3.35 \pm 0.35$	$3.09 \pm 0.28$	$3.08 \pm 0.26$	$3.15 \pm 0.37$	0.06	
mmol l <sup>-1</sup>	15	$3.43 \pm 0.56$	$3.36 \pm 0.35$	$3.31 \pm 0.44$	$3.38 \pm 0.38$	0.09	
	$\overline{\mathbf{x}}$	$3.35 \pm 0.43$	$3.24 \pm 0.34$	$3.23 \pm 0.33$	$3.28 \pm 0.37$	0.07	
	9	$1.64 \pm 0.25$	$1.84 \pm 0.28$	$1.95 \pm 0.23$	$1.79 \pm 0.20$	0.05	
HDL	11	$1.78 \pm 0.19$	$1.93 \pm 0.22$	$1.96 \pm 0.22$	$1.71 \pm 0.23$	0.05	
mmol 1 <sup>-1</sup>	15	$1.77 \pm 0.17$	$1.89 \pm 0.15$	$1.94 \pm 0.13$	$1.79 \pm 0.14$	0.03	
	$\overline{\mathbf{x}}$	$1.73 \pm 0.20$	$1.88 \pm 0.22$	$1.95 \pm 0.19$	$1.76 \pm 0.19$	0.04	
	9	$0.35^{b} \pm 0.03$	$0.39^{ab} \pm 0.04$	$0.40^{ab} \pm 0.04$	$0.42^{a} \pm 0.03$	0.009	
UA	11	$0.47^{ab} \pm 0.04$	$0.44^{b} \pm 0.03$	$0.43^{b} \pm 0.04$	$0.51^{a} \pm 0.04$	0.01	
mmol l <sup>-1</sup>	15	$0.46^{a} \pm 0.05$	$0.37^{b} \pm 0.05$	$0.36^{b} \pm 0.03$	$0.35^{b} \pm 0.04$	0.01	
	$\overline{\mathbf{x}}$	$0.42 \pm 0.04$	$0.40 \pm 0.04$	$0.39 \pm 0.03$	$0.42 \pm 0.03$	0.009	
	9	190.2 ± 19.7	$205.5 \pm 19.9$	$201.6 \pm 20.7$	199.5 ± 16.4	4.16	
AST	11	$186.3 \pm 10.9$	$185.0 \pm 25.3$	$179.6 \pm 10.9$	$177.5 \pm 18.2$	3.62	
U 1 <sup>-1</sup>	15	190.1 <sup>a</sup> ±14.7	$172.3^{b} \pm 21.1$	$174.6^{b} \pm 11.6$	$165.6^{b} \pm 17.8$	4.36	
	$\overline{\mathbf{x}}$	$188.8 \pm 15.1$	$187.6 \pm 22.1$	$185.2 \pm 14.4$	$180.6 \pm 17.4$	4.04	
	9	$5.42 \pm 0.9$	$4.95 \pm 0.95$	$4.87 \pm 0.61$	$5.28 \pm 0.48$	0.16	
ALT U l <sup>-1</sup>	11	$5.35 \pm 0.72$	$5.29 \pm 0.93$	$5.49 \pm 0.62$	$5.57 \pm 0.84$	0.16	
	15	$6.21^a \pm 0.61$	$6.08^{a} \pm 0.79$	$5.26^{b} \pm 0.43$	$5.56^{ab} \pm 0.47$	0.14	
	$\overline{\mathbf{x}}$	$5.66 \pm 0.74$	$5.44 \pm 0.89$	$5.20 \pm 0.55$	$5.47 \pm 0.59$	0.15	
ALP U 1 <sup>-1</sup>	9	1218 ± 116		$1206.3 \pm 109.6$		22.3	
	11	1271.1 ± 107.4	$1189.3 \pm 88.5$	$1193.1 \pm 88.5$	$1251.5 \pm 113.5$	21.7	
	15	$1173.3 \pm 78.7$	$1189.3 \pm 80.4$	$1193.1 \pm 98.8$	$1251.5 \pm 113.5$	19.4	
	$\overline{\mathbf{x}}$	$1220.7 \pm 100.7$	$1200.7 \pm 94.2$	$1197.5 \pm 98.9$	$1250 \pm 105.5$	21.1	

a, b – values in the same rows with different letters differ significantly at p  $\leq 0.05$  – wartości w wierszach oznaczone różnymi literami różnią się istotnie przy  $\leq 0.05$ ; T0 – control – kontrola; T15 – 15 µg/kg b.w./day – dawka 15 µg/kg m.c./dzień; T30 – 30 µg/kg b.w./day – dawka 30 µg/kg m.c./dzień; T45-45 µg/kg b.w./day – dawka 45 µg/kg m.c./dzień, TP – total protein – białko ogólne, GLU – glucose – glukoza; TG – triglycerides – trilicerydy, CHOL – cholesterol, UA – uric acid – kwas moczowy; AST – asparagine aminotransferase – aminotransferaza asparaginianowa; ALT – alanine aminotransferase – aminotransferaza alaninowa; ALP – alkaline phosphatase – fosfataza zasadowa

condition in turkey hens examined. The analysis of results achieved for total protein content of blood plasma of the turkey hens demonstrated that only in week 15 of life of the birds receiving the addition of 5-oxo-1,2,4-triazine in doses of 15 µg/kg b.w./day and 45  $\mu$ g/kg b.w./day were these values significantly (P  $\leq$  0.05) lower than in the control group. The increased total protein level in blood plasma of turkey hens was reported by Czech et al. [2010] who was adding a mixture of synthetic antioxidants (L100) to the birds' diet. In turn, uric acid is the end product of nitrogen compounds metabolism in birds. The effect of 5-oxo-1,2,4-triazine administration on the level of uric acid was tangible as soon as after 4-week supplementation of the additive (9<sup>th</sup> week of life). In blood plasma of the birds receiving 5-oxo-1,2,4-triazine addition in a dose of 45 μg/kg b.w./day (group T45) the content of uric acid was by 20 % higher than in the control group. The increased level of this parameter in group T45 was also observed in week 11 of birds life, however in respect of the control group the difference noted turned out statistically insignificant. In the 15th week of life, in the groups receiving amidrazones derivative addition, a significant (by 20%) decrease was observed in uric acid level when compared to the control birds. Czech and Ognik [2010], who were administering turkey hens with a mixture of antioxidants (BHT, E310 - propyl gallate, E324 - ethoxyquin, E330 - citric acid), were also reporting an increase in plasma level of uric acid (UA). The activity of asparagine aminotransferase (AST) in the 9<sup>th</sup> and 11<sup>th</sup> week of birds life was at a relatively similar level in the experimental groups. A significant ( $P \le 0.05$ ) suppression of this enzyme's activity was noted already in 15-week turkey hens receiving the derivative (groups: T15, T30, T45). Likewise in the case of asparagine aminotransferase, in the  $9^{th}$  and  $11^{th}$  week of birds life no significant (P  $\leq$  0.05) differences were noted between the groups in the activity of alanine aminotransferase (ALT). A decline in this enzyme's activity was observed already in the 15-week old turkey hens receiving 5-oxo-1,2,4-triazine addition in a dose of 30 μg/kg b.w./day (5.26 U l<sup>-1</sup>), with the decline being statistically significantly different when compared to group T0 (6.21 U l<sup>-1</sup>) and group T15 (6.08 U l<sup>-1</sup>). In turn, Krauze [2007b], noted a significant increase in the activity of both AST and ALT in turkeys receiving 1,2,4-triazole. Many authors report that the increased activity of aminotransferase-group enzymes and alkaline phosphatase are observed in some liver diseases [Dembińska-Kieć and Naskalski 2002].

In the body of turkey hens, macroelements constitute a small weight percentage, however they are vital to particular parts of bird body as well as to physicochemical functions of their cells and tissues. Mineral metabolism in the body of turkeys should be in the state of homeostasis. Fluctuations in that state may induce disorders in the metabolism of mineral compounds [Wertelecki 2003]. Each of the analyzed mineral compounds plays specific functional roles. Levels of most of the macroelements (Na, K, Mg) analyzed in blood plasma of turkey hens (Tab. 3) receiving the addition of the derivative were similar to these recorded in the control birds. In contrast, 5-oxo-1,2,4-triazine addition to water was observed to evoke an increase in the level of calcium, as compared to the control, yet only in the 11<sup>th</sup> week of birds life. Taking into account the entire fattening period, the plasma level of calcium in the case of turkey hens from groups: T15, T30, T45, also turned out to significantly higher than in the control group, by 8.5, 8 and 8.5%, respectively. Calcium is the main constituent of bones, it additionally affects their functions and serves a key role in the regulation of multiple cellular processes [Weisenberg

and Bellorin-Font 1998, Makarski and Makarska 2010]. Plasma levels of calcium noted in the turkey hens receiving 5-oxo-1,2,4-triazine doses were similar to these reported by Krauze [2007b] who was administering various does of 1,2,4-triazole to turkeys. These authors demonstrated also the effect of derivative supplementation on the increased levels of macroelements (Na, K and Mg) in blood plasma of turkey hens, which was not confirmed in our study with 5-oxo-1,2,4-triazine doses.

Parameter Cecha	Week of life	Expe	CEM			
	Tydzień życia	ТО	T15	Т30	T45	SEM
	9	$119.6 \pm 3.51$	$118.3 \pm 5.90$	$116.7 \pm 3.17$	$117.7 \pm 3.74$	0.98
Na	11	$116.7 \pm 10.4$	$115.3 \pm 4.82$	$115.1 \pm 3.78$	$114.7 \pm 7.09$	1.58
mmol 1 <sup>-1</sup>	15	$114.4 \pm 3.05$	$112.2 \pm 2.64$	$113.1 \pm 5.69$	$118.0 \pm 8.96$	1.38
	$\overline{\mathbf{x}}$	$116.9 \pm 5.65$	$115.2 \pm 4.45$	$114.9 \pm 4.21$	$116.8 \pm 6.59$	1.31
K mmol l <sup>-1</sup>	9	$3.66 \pm 0.25$	$3.76 \pm 0.34$	$3.73 \pm 0.33$	$3.71 \pm 0.29$	0.07
	11	$3.94 \pm 0.23$	$3.80 \pm 0.37$	$3.86 \pm 0.25$	$3.86 \pm 0.22$	0.06
	15	$3.91 \pm 0.38$	$3.81 \pm 0.36$	$3.86 \pm 0.22$	$4.0 \pm 0.08$	0.07
	$\overline{\mathbf{x}}$	$3.83 \pm 0.28$	$3.79 \pm 0.35$	$3.81 \pm 0.26$	$3.85 \pm 0.19$	0.06
Ca mmol 1 <sup>-1</sup>	9	$3.77 \pm 0.31$	$3.72 \pm 0.44$	$3.77 \pm 0.24$	$3.70 \pm 0.41$	0.08
	11	$3.05^{b} \pm 0.36$	$3.87^{a} \pm 0.09$	$3.75^{a} \pm 0.12$	$3.77^{a} \pm 0.19$	0.09
	15	$3.52 \pm 0.19$	$3.6 \pm 0.54$	$3.65 \pm 0.28$	$3.72 \pm 0.18$	0.07
	$\overline{\mathbf{x}}$	$3.44^{b} \pm 0.28$	$3.73^{a} \pm 0.35$	$3.72^{a} \pm 0.21$	$3.73^{a} \pm 0.26$	0.08
Mg mmol 1 <sup>-1</sup>	9	$0.71 \pm 0.21$	$0.53 \pm 0.12$	$0.78 \pm 0.11$	$0.80 \pm 0.07$	0.03
	11	$0.78 \pm 0.05$	$0.79 \pm 0.08$	$0.81 \pm 0.08$	$0.82 \pm 0.03$	0.01
	15	$0.74 \pm 0.05$	$0.80 \pm 0.05$	$0.79 \pm 0.05$	$0.81 \pm 0.05$	0.01
	$\overline{\mathbf{x}}$	$0.74 \pm 0.10$	$0.70 \pm 0.08$	$0.79 \pm 0.08$	$0.81 \pm 0.05$	0.01

Table 3. Content of macroelements in blood plasma of turkey hens Tabela 3. Zawartość makroelementów w osoczu krwi indyczek

Table 4 collates data on bioelements analyzed in meat tissues and liver of the slaughter turkey hens. No statistically significant differences were noted between the groups in terms of sodium, potassium and calcium levels in turkey hens tissues. Only in the case of magnesium, a statistically significantly lower level of this element was recorded in group T30 (14.9 g kg<sup>-1</sup>), compared to the control birds (17.8 g kg<sup>-1</sup>). Magnesium affects the activation of many intracellular enzymes, e.g. phosphatase. In addition, likewise calcium, it is a constituents of blood plasma and occurs in links with functional proteins. Truchliński *et al.* [2006], when administering a 1,2,4-triazole derivative to turkey hens, noted increased levels of magnesium in their liver, blood and feathers. The increase in this element's level in breast and leg muscles was also demonstrated by Makarski and Makarska [2010] while adding a copper bioplex to drinking water for the

a, b – values in the same rows with different letters differ significantly at p  $\leq 0.05$  – wartości w wierszach oznaczone różnymi literami różnią się istotnie przy  $\leq 0.05$ ; T0 – control – kontrola; T15 – 15  $\mu$ g/kg b.w./day – dawka 15  $\mu$ g/kg m.c./dzień; T30 – 30  $\mu$ g/kg b.w./day – dawka 30  $\mu$ g/kg m.c./dzień; T45-45  $\mu$ g/kg b.w./day – dawka 45  $\mu$ g/kg m.c./dzień

birds. A higher, as compared to control, level of this element in meat tissues and liver was additionally reported by Krauze [2007a] who was administering different doses of 1,2,4-triazole derivative to turkey hens.

Table 4. Content of macroelements in tissues of turkey hens Tabela 4. Zawartość makroelementów w tkankach indyczek

Tissue Tkanka	Group	Macroelements – Makroelementy				
	Grupa	Na, g kg <sup>-1</sup>	K, g kg <sup>-1</sup>	Ca, g kg <sup>-1</sup>	Mg, g kg <sup>-1</sup>	
Breast muscles	ТО	$0.90 \pm 0.08$	$1.71 \pm 0.30$	$0.14 \pm 0.01$	$17.8^{a} \pm 0.98$	
	T15	$1.86 \pm 0.19$	$1.70 \pm 0.19$	$0.14 \pm 0.03$	$15.6^{ab} \pm 1.69$	
	T30	$1.80 \pm 0.18$	$1.74 \pm 0.08$	$0.14 \pm 0.02$	$14.9^{b} \pm 2.05$	
Mięśnie	T45	$1.77 \pm 0.22$	$1.67 \pm 0.26$	$0.15 \pm 0.02$	$16.5^{ab} \pm 1.08$	
piersiowe	SEM	0.043	0.051	0.005	0.44	
Thigh	TO	$1.78 \pm 0.12$	$1.67 \pm 0.16$	$0.16 \pm 0.03$	$15.0 \pm 1.12$	
	T15	$1.70 \pm 0.23$	$1.63 \pm 0.28$	$0.15 \pm 0.02$	$14.0 \pm 0.35$	
muscles Micánia	T30	$1.67 \pm 0.28$	$1.56 \pm 0.22$	$0.16 \pm 0.01$	$14.1 \pm 0.38$	
Mięśnie udowe	T45	$1.76 \pm 0.26$	$1.55 \pm 0.06$	$0.15 \pm 0.02$	$14.7 \pm 1.04$	
	SEM	0.054	0.045	0.005	0.21	
Shank	TO	$1.69 \pm 0.14$	$1.80 \pm 0.21$	$0.32 \pm 0.09$	$14.6 \pm 0.47$	
muscles	T15	$1.64 \pm 0.10$	$1.82 \pm 0.12$	$0.28 \pm 0.01$	14.5± 1.03	
	T30	$1.60 \pm 0.14$	$1.77 \pm 0.11$	$0.30 \pm 0.04$	$14.7 \pm 1.18$	
Mięśnie	T45	$1.61 \pm 0.10$	$1.78 \pm 0.17$	$0.33 \pm 0.02$	$14.8 \pm 0.78$	
podudzia	SEM	0.029	0.037	0.009	0.20	
	TO	$1.72 \pm 0.14$	$1.87 \pm 0.28$	$0.19 \pm 0.01$	$16.9 \pm 1.31$	
Liver Wątroba	T15	$1.65 \pm 0.12$	$1.85 \pm 0.08$	$0.22 \pm 0.03$	$16.7 \pm 1.09$	
	T30	$1.78 \pm 0.28$	$1.86 \pm 0.28$	$0.21 \pm 0.03$	$17.3 \pm 1.52$	
	T45	$1.76 \pm 0.14$	$1.78 \pm 0.24$	$0.19 \pm 0.03$	$17.3 \pm 1.47$	
	SEM	0.043	0.053	0.007	0.31	

a, b – values in the same rows with different letters differ significantly at p  $\leq 0.05~$  – wartości w wierszach oznaczone różnymi literami różnią się istotnie przy  $\leq 0.05;$  T0 – control – kontrola; T15 – 15  $\mu g/kg$  b.w./day – dawka 15  $\mu g/kg$  m.c./dzień; T30 – 30  $\mu g/kg$  b.w./day – dawka 30  $\mu g/kg$  m.c./dzień; T45-45  $\mu g/kg$  b.w./day – dawka 45  $\mu g/kg$  m.c./dzień

Data presented in Table 5 indicate that the turkey hens from groups T15 and T30 achieved better body weight gains, as compared to the control birds, in the 9<sup>th</sup>-11<sup>th</sup> week of life, whereas these from group T45 (by ca 3.2 %) – already in the last week of life. Considering the entire rearing period, the turkey hens from groups T15, T30, and T45 were observed to achieve better body weight gains, by ca. 7.7%, 6.3%, 5.4%, respectively, as compared to the control birds. Due to changes in chemical properties likely to occur upon the influence of multiple factors, including the nutritional ones, in liver and breast muscles, these tissues are often subject of studies to both biochemists and dietitians [Makarski *et al.* 2006]. In our study, however, the administration of various doses of an amidrazones derivative – 5-oxo-1,2,4-triazine, was observed not to affect the percentage of dry matter nor crude protein (Tab. 5). The percentage of fat in breast muscles and liver of the turkey hens receiving the tested additive (groups T15, T30 and T45) was substantially lower than in the control group. The reduced level of crude fat in breast and

Table 5. Rearing performance and chemical composition of meat of turkey hens receiving the addition of 5-oxo-1,2,4-triazine derivative

Tabela 5. Wydajność rzeźna i skład chemiczny mięśni indyczek otrzymujących dodatek pochodnej 5-okso-1,2,4-triazyny

Tissue	Experimental groups – Grupy doświadczalne				SEM		
Tkanka	T0 T15		T30 T45		SEM		
Body weight gains, kg – Przyrosty masy ciała, kg							
5–9 week – 5–9 tydzień	1.83	2.09	2.07	2.08	0.009		
9–11 week – 9–11 tydzień	1.25	1.47	1.49	1.25	0.012		
11–15 week – 11–15 tydzień	3.01	3.04	2.94	3.11	0.025		
5–15 week – 5–15 tydzień	6.09	6.60	6.50	6.44	0.11		
Dry matter, % – Sucha masa, %							
Breast muscles	$26.4 \pm 0.96$	$26.2 \pm 0.77$	$25.9 \pm 0.57$	$25.2 \pm 0.99$	0.22		
Mięśnie piersiowe	20.4 ± 0.70	20.2 ± 0.77	23.7 ± 0.37	23.2 ± 0.77			
Thigh muscles	$24.9 \pm 0.48$	$24.6 \pm 0.51$	$24.5 \pm 0.48$	$24.4 \pm 0.57$	0.18		
Mięśnie udowe	21.7 ± 0.10	21.0 ± 0.51	21.3 ± 0.10	21.1 ± 0.57	0.10		
Shank muscles	$24.2 \pm 0.45$	$24.1 \pm 0.73$	$24.5 \pm 0.60$	$24.1 \pm 0.66$	0.14		
Mięśnie podudzia							
Liver – Wątroba	$31.3 \pm 0.90$	$30.02 \pm 0.22$	$30.3 \pm 0.76$	$30.0 \pm 0.96$	0.21		
	Crude protein,	% – Białko sur	owe, %				
Breast muscles	$25.8 \pm 0.56$	$25.5 \pm 0.22$	$24.6 \pm 0.14$	$25.4 \pm 0.69$	0.13		
Mięśnie piersiowe	23.6 ± 0.50	23.3 ± 0.22	24.0 ± 0.14	23.4 ± 0.07	0.13		
Thigh muscles	$22.6 \pm 0.62$	$21.7 \pm 0.75$	$21.6 \pm 0.73$	$21.7 \pm 0.69$	0.20		
Mięśnie udowe	22.0 ± 0.02						
Shank muscles	$22.0 \pm 0.74$	$21.7 \pm 0.94$	$22.0 \pm 0.63$	$21.0 \pm 0.68$	0.19		
Mięśnie podudzia							
Liver – Wątroba	$17.9 \pm 0.92$	$18.4 \pm 0.59$	$18.6 \pm 0.69$	$18.5 \pm 0.91$	0.23		
	Crude fat, %	<ul> <li>Tłuszcz suro</li> </ul>	wy, %				
Breast muscles	$0.92^{a} \pm 0.04$	$0.68^{b} \pm 0.04$	$0.69^{b} \pm 0.01$	$0.68^{b} \pm 0.03$	0.027		
Mięśnie piersiowe	0.72 ± 0.04	0.08 ± 0.04	0.07 ± 0.01	0.08 ± 0.03	0.027		
Thigh muscles	$1.72 \pm 0.20$	$1.88 \pm 0.08$	$1.81 \pm 0.07$	$1.91 \pm 0.10$	0.034		
Mięśnie udowe	1.72 = 0.20	1.00 ± 0.00	1.01 = 0.07	1.51 = 0.10	0.05		
Shank muscles	$2.40 \pm 0.23$	$2.75 \pm 0.32$	$2.76 \pm 0.11$	$2.66 \pm 0.25$	0.065		
Mięśnie podudzia							
Liver – Wątroba	$3.70^{a} \pm 0.28$	$2.85^{c} \pm 0.21$	$3.22^{b} \pm 0.20$	$3.02^{bc} \pm 0.20$	0.096		
Ash, % – Popiół, %							
Breast muscles	$3.26 \pm 0.29$	$2.75 \pm 0.51$	$2.77 \pm 0.44$	$2.80 \pm 0.35$	0.10		
Mięśnie piersiowe	3.20 = 0.29	2.00 ± 0.55	0.10				
Thigh muscles	$2.00 \pm 0.34$	$2.19 \pm 0.07$	$1.95 \pm 0.48$	$1.88 \pm 0.29$	0.080		
Mięśnie udowe		2.17 ± 0.07					
Shank muscles	$1.96 \pm 0.58$	$1.59 \pm 0.13$	$1.64 \pm 0.25$	$1.51 \pm 0.15$	0.086		
Mięśnie podudzia							
Liver – Wątroba	$6.14^{a} \pm 0.17$	$4.61^{c} \pm 0.55$	$5.18^{b} \pm 0.35$	$5.01^{bc} \pm 0.22$	0.160		

a, b, c – values in the same rows with different letters differ significantly at p  $\leq 0.05~$  – wartości w wierszach oznaczone różnymi literami różnią się istotnie przy  $\leq 0.05;$  T0 – control – kontrola; T15 – 15  $\mu g/kg$  b.w./day – dawka 15  $\mu g/kg$  m.c./dzień; T30 – 30  $\mu g/kg$  b.w./day – dawka 30  $\mu g/kg$  m.c./dzień; T45-45  $\mu g/kg$  b.w./day – dawka 45  $\mu g/kg$  m.c./dzień

leg muscles of chickens receiving the addition of sodium salt of n-butyric acid, prebiotics and probiotics in a feed mixture, was also reported by Pietrzak *et al.* [2009], who found the results achieved as positive from the dietetic point of view. Sembratowicz [2004] also demonstrated a decrease in the percentage of crude fat in the analyzed meat tissues upon the administration of plant immunomodulators to turkey hens. In the livers of the turkey hens from groups T15, T30, T45 analyses demonstrated also a significantly lower level of crude ash.

#### CONCLUSIONS

- 1. The application of various doses of 5-oxo-1,2,4-triazine derivative had no significant effect on the levels of the analyzed biochemical markers of turkey hens blood. An exception was an increase in calcium content observed in the 11<sup>th</sup> week of life.
- 2. The administration of 5-oxo-1,2,4-triazine to turkey hens from groups T15, T30 and T45 contributed to their better mean body weight gains, by ca. 6.4%, as compared to the control birds.
- 3. The addition of various doses of 5-oxo-1,2,4-triazine to drinking water was observed to decrease the content of crude fat in breast muscles and in liver, which increases the dietetic value of turkey hens meat.

#### REFERENCES

- AOAC, 2000. Official Methods of Analysis. Association of Official Analytical Chemist. EUA.
- Czech A., Ognik K., 2010. Wpływ dodatku mieszaniny syntetycznych przeciwutleniaczy na wskaźniki metabolizmu u indyczek. Zesz. Prob. Post. Nauk Roln., 544, 53–62.
- Czech A., Ognik K., Chachaj R., 2010. Efficiency of synthetic antioxidants mixture additive in turkey hens' diet. Annales UMCS, sec. EE, Zootechnica, 28, 4, 1–7.
- Dembińska-Kieć A., Naskalski J.W., 2002. Diagnostyka laboratoryjna z elementami biochemii klinicznej. Urban & Partner, Wrocław, 997.
- Faruga A., Jankowski J., 2000. Indyki hodowla i użytkowanie. PWRiL, Warszawa.
- Krasnodębska-Depta A., Koncicki A., 2000. Fizjologiczne wartości wybranych wskaźników biochemicznych w surowicy krwi kurcząt brojlerów. Med. Wet., 56, 7, 456–460.
- Krauze M., 2007. Influence of differential rations of 1,2,4-triasole derivative on the level of chosen blood indicators and rearing efficiency of slaughter turkey-hens. Annales UMCS, sec. DD, Medicina Veterinaria, 62, 1, 8–14.
- Krauze M., 2007a. Evaluation of elements composition in turkeys' tissues after intake N³-amidrasones. Pol. J. Environ. Stud. 16, 3A, 160–162.
- Makarski B., Zadura A., Kwiecień M., 2006. The effect of Cu-lysine chelate In turkeys diets on the results of slaughter analysis, chemical comopsition and the fatty AIDS profile tissues. Acta Sci. Pol., Zootechnica, 5, 2, 57–66.
- Makarski B., Makarska E., 2010. Effect of addition of Ca, Cu, Fe, Zn and lactic acid to drinking water of these elements in muscles of slaughter turkeys. J. Elemetol., 15, 1, 119–129.

- Modzelewska-Banachiewicz B, Szcześniak Z., 2001. The influence of 1,2,4-triazole and 5-oxo-1,2,4-triazyne derivatives on some species of the human digestive tract microflora. Annales UMCS, sec. DDD, Pharmacia, 14, 1, 9–11.
- Normy żywienia drobiu, 2005. Wyd. IV zm. i uzup. PAN IFiZZ Jabłonna.
- Ognik K., Sembratowicz I., 2009. Influence of different rates of new synthetised 5-oxo-1,2,4-triazyne derivative on some immunological and hematological indices of blood of turkey hens. J. Apel. Anim. Res., 36, 235–237.
- Ognik K., Sembratowicz I., Modzelewska-Banachiewicz B., 2004. Stężenie wybranych pierwiastków i aktywność enzymów antyoksydacyjnych we krwi indyczek otrzymujących Echinovit C oraz pochodną 1,2,4-triazolu. J. Elementol., 9, 3, 445–449.
- Pietrzak D., Mroczek J., Garbaczewska A., Florowski T., Riedel J., 2009. Wpływ wybranych dodatków do paszy o działaniu przeciwbakteryjnym na jakość mięsa i tłuszczu kurcząt. Med. Wet., 65, 4, 268–271.
- Sembratowicz I., 2004. Wpływ wyciągów ziołowych na wybrane wskaźniki krwi i efekty odchowu indyczek. Annales UMCS, sec. EE, Zootechnica, 22, 43, 325–332.
- Truchliński J., Krauze M., Modzelewska-Banachiewicz B., 2006. Wpływ dodatku czosnku i pochodnej 1,2,4-triazolu na poziom wybranych elementów mineralnych w surowicy krwi i tkankach indyczek. Annales UMCS, sec. EE, Zootechnica, 24, 47, 349–355.
- Weisenger J., Bellorin-Font E., 1998. Magnesium and phosphorus. Lancet, 352, 391.
- Wertelecki T., 2003. Wybrane makro- i mikroelementy w żywieniu drobiu. Najważniejsze mikroelementy. Indyk Pol., 5, 3, 4–19.

Streszczenie. Celem podjętych badań było określenie wpływu dodatku pochodnej amidrazonów – 5-okso-1,2,4-triazyny do wody dla indyczek na poziom wskaźników biochemicznych krwi oraz skład chemiczny tkanek. Badania przeprowadzono na 160 indyczkach typu średniociężkiego British United Turkey (BUT) 9 podzielonych losowo i równomiernie na cztery grupy. Każda grupa liczyła po 40 indyczek (dwa powtórzenia, w każdym po 20 sztuk). Ptaki utrzymywano od 6 do 16 tygodnia życia w kojcach, na ściółce ze słomy. Indyczki z grupy I (kontrolnej – T0) otrzymywały do picia wodę bez dodatków doświadczalnych. Indyczki z grup doświadczalnych (T15, T30, T45) otrzymywały do wody trzy różne dawki rozpuszczonej w 2 ml etanolu 5-okso-1,2,4--triazyny. W eksperymencie zastosowano następujący sposób dawkowania 5-okso-1,2,4-triazyny: 15 μg/kg m.c./dzień dla grupy T15, 30 μg/kg m.c./dzień dla grupy T30 oraz 45 μg/kg m.c./dzień dla grupy T45. Po 28 dniach wprowadzona została dwutygodniowa przerwa w suplementacji, podczas której indyczki wszystkich grup otrzymywały jedynie czystą wodę. Po tym czasie zwierzęta ponownie otrzymywały do wody te same dodatki w identycznych dawkach jak na początku eksperymentu. Pod koniec 4, 6 i 10 tygodnia obserwacji z żyły skrzydłowej ptaków pobrano krew do badań biochemicznych, w których oznaczono: AST, ALT, ALP, TP, GLU, CHOL, HDL, TG, makroelementy (K<sup>+</sup>, Na<sup>+</sup>, Ca<sup>+2</sup>, Mg<sup>+2</sup>). Po zakończonym odchowie (16 tydzień życia) przeprowadzono ubój. Określono skład chemiczny miesa w próbkach mieśni piersiowych, udowych, podudzia i w wątrobie. Zastosowanie dodatku różnych poziomów pochodnej 5-okso-1,2,4-triazyny nie wpłynęło istotnie na poziom badanych wskaźników biochemicznych krwi indyczek. Wyjątek stanowił wzrost zawartości wapnia w 11 tygodniu życia. Dodatek 5-okso-1,2,4 triazyny do wody dla indyczek z grup T15, T30 oraz T45 przyczynił się do uzyskania lepszych o ok. 6,4% średnich przyrostów w stosunku do indyczek z grupy kontrolnej Dodatek różnych poziomów 5-okso-1,2,4-triazyny do wody pitnej spowodował obniżenie tłuszczu surowego w mięśniach piersiowych i wątrobie, co podniosło walory dietetyczne mięsa indyczek.

Slowa kluczowe: indyczki, krew, tkanki, wskaźniki metaboliczne