
ANNALES
UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA
LUBLIN – POLONIA

VOL. XXVIII (2)

SECTIO EE

2010

Department of Biochemistry and Toxicology, University of Life Sciences in Lublin
Akademicka 13, 20-950 Lublin,
e-mail: jerzy.truchlinski@up.lublin.pl

KATARZYNA ROSTEK

**The influence of vitamin E and trolox
on biochemical and hematological indices
in turkey hen's blood**

Wpływ witaminy E i troloxu na wskaźniki biochemiczne i hematologiczne
krwi indyczek

Summary. The study aimed at determining the influence of vitamin E and trolox (vitamin E preparation) as additives to drinking water, on hematological and biochemical indices at turkey hen's blood. The studied material consisted of 6-week-old turkey hens of BIG-6 type maintained since 6th till 16th weeks of their life. Birds were divided into 3 experimental groups: I – control, II – administered with vitamin E addition in the amount of 1.1 ml /kg b.w./daily, III – administered with trolox preparation dissolved in water in the amount of 0.95 mg/kg b.w./daily. Turkey hens were fed *ad libitum* with standard full-dose mixtures (Provimi Polska) according to a program including 5 feeding periods. The following items were determined in blood: activities of aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, as well as total protein, uric acid, triglycerides, total cholesterol, high-density lipoprotein fraction (HDL), and glucose content by means of mono-tests (Cormay). Hematological indices determinations referred to: hematocrite number, hemoglobin content, and leukocytes count. No significant changes in shaping the hematological indices in turkey hens blood were found. The addition of vitamin E and trolox caused the increase of urea and HDL cholesterol fraction levels in blood plasma. The obtained results indicate the opportunity to apply vitamin E and trolox as an antioxidant additive to drinking water for turkey hens.

Keywords: vitamin E, trolox, blood hematological and biochemical indicators, turkey hens

INTRODUCTION

An increased interests in utilizing the natural and synthetic antioxidants, that modulate the animal's metabolic reactions and improve their rearing efficiency, have been

recently observed. Applying antioxidants as additives to a fodder also contributes to the improvement of its quality by saving fat (namely unsaturated essential fatty acids – UFA) against its oxidation.

Due to the fact that supply of appropriate UFA amounts along with antioxidants contained in a fodder, is very important for birds, administration of vitamin E (tocopherol) and – for comparisons – trolox (water-soluble vitamin E equivalent), seemed to be reasonable.

MATERIAL AND METHODS

Studied material comprised of 6-week-old turkey hens of heavy BIG-6 type maintained since 6 till 16 week of life on a straw litter in cages. Study was realized under production conditions with isolated boxes for experimental groups. Birds were randomly divided into 3 experimental groups consisting of 40 turkey hens each. The group I was the control. Birds of group II were administered with liquid vitamin E addition at the amount of 1.1 ml/kg b.w./day. Turkey hens of group III were fed with trolox (6-Methoxy-2,4,7,8-tetramethylchromane-2-carboxylic acid) preparation dissolved in 10 ml of water (0.95 mg/kg b.w./day). In order to determine the daily dose of applied additives, all birds were weighed once a week, and in addition, 10 turkey hens of every group – daily. Results were then averaged.

Biochemical indices

Referring to biochemical assays, activities of the following enzymes were determined in turkey hen's blood plasma: aspartate aminotransferase (liquick Cor-ASAT), alanine aminotransferase (liquick Cor-ALAT), and alkaline phosphatase (liquick Cor-ALP) – by means of kinetic method using test kits (Cormay). Total protein (Liquick Cor-Total protein), uric acid (liquick Cor-UA), triglycerides (liquick Cor-TG), total cholesterol (liquick Cor-Chol), high-density lipoprotein fraction (Cormay HDL), and glucose (liquick Cor-Glucose) contents were also assayed applying colorimetric procedures using diagnostic kits (Cormay).

Hematological indices

In reference to hematological indices, hematocrit value (Ht, micro-hematocrit method), hemoglobin content (Hb, colorimetric method according to Drabkin), and white cells count (WBC, chamber method) were analyzed [Pinkiewicz 1971, Bomski 1989]. Natt-Herrick's liquid was used to dye leukocytes. The percentage of white cells (leukogram) was made by dying blood smears according to Pappenheim's method [Bomski 1989].

Statistical analysis

Achieved numerical data were subjected to statistical analysis with a help of Statistica ver. 5 software. The single-factorial variance analysis ANOVA was applied at the significance level 0.05.

RESULTS AND DISCUSSION

As it can be seen in Tables 1 and 2, no significant differences referring to uric acid, protein, triglycerides, lactic dehydrogenase, alanine aminotransferase, and aspartate aminotransferase levels at blood plasma between turkey hen experimental groups, were found. Achieved values are similar to those observed in earlier studies [Krasnodębska-Depta and Koncicki 2000].

Table 1. Biochemical indices levels in blood plasma of 9-week-old (4th observation week) turkey hens administered with vitamin E and trolox additives

Tabela 1. Poziom wskaźników biochemicznych w osoczu krwi 9-tygodniowych indyczek (4 tydzień obserwacji) otrzymujących do wody dodatek witaminy E i troloxu

Specification Wyszczególnienie	Control Kontrola	Trolox Trolox	Vitamin E Witamina E
Urea, mmol l ⁻¹ Mocznik	3.39 ^b ± 0.25	8.26 ± 0.59	8.73 ^a ± 0.36
Uric acid, mmol l ⁻¹ Kwas moczowy	0.19 ± 0.02	0.20 ± 0.01	0.15 ± 0.03
Protein, g dl ⁻¹ Białko	7.60 ± 1.66	9.30 ± 1.66	8.27 ± 1.33
Cholesterol, mmol l ⁻¹	3.56 ± 0.19	3.59 ± 0.18	3.46 ± 0.04
Glucose, mmol l ⁻¹ Glukoza	12.16 ± 6.24	25.61 ^a ± 7.57	17.5 ^{ab} ± 8.94
HDL, mmol l ⁻¹ Fracja cholesterolowa HDL	1.24 ^b ± 0.18	4.47 ^a ± 0.40	3.34 ^a ± 0.10
Triglycerides, mmol l ⁻¹ Triglicerydy	0.87 ± 1.10	0.92 ± 0.20	0.83 ± 0.10
ALP, U l ⁻¹ Fosfataza zasadowa	81.11 ± 11.07	73.66 ± 17.01	96.81 ± 11.86
ALT, U l ⁻¹ Aminotransferaza alaninowa	6.06 ± 1.58	5.03 ± 1.80	5.18 ± 0.11
AST, U l ⁻¹ Aminotransferaza asparaginianowa	145 ± 30.0	141 ± 25.7	138 ± 28.0

a, b – p < 0.05 statistically significant difference

a, b – p < 0,05 różnice statystycznie istotne

± standard deviation

± odchylenie standardowe

Considerable differences ($p \leq 0.05$) arose between urea content; however, only at the 9th week of turkey hen's life (4th observation week) (Tab. 1). Birds given with vitamin E and trolox addition, showed much higher urea level than those from control group (3.39 mmol l⁻¹) amounting to 8.26 mmol l⁻¹ (for trolox) and 8.73 mmol l⁻¹ (for vitamin E), respectively. Urea is a final product of nitrogen transformations of proteins. The urea concentration in blood is a resultant of a production occurring in a liver, as well as kidney excretion.

Table 2. Biochemical indices levels in blood plasma of 11-week-old (6th observation week) turkey hens administered with vitamin E and trolox additives.

Tabela 2. Poziom wskaźników biochemicznych we krwi 11-tygodniowych indyczek (6 tydzień obserwacji) otrzymujących do wody dodatek witaminy E i troloxu

Specification Wyszczególnienie	Control Kontrola	Trolox Trolox	Vitamin E Witamina E
Urea, mmol l ⁻¹ Mocznik	7.00 ± 0.36	6.43 ± 1.04	7.06 ± 0.34
Uric acid, mmol l ⁻¹ Kwas moczowy	0.71 ± 0.04	0.79 ± 0.03	0.76 ± 0.05
Protein, g dl ⁻¹ Białko	5.54 ± 0.69	5.22 ± 0.24	6.39 ± 0.30
Cholesterol, mmol l ⁻¹ Cholesterol	2.62 ± 0.60	2.34 ± 0.25	2.18 ± 0.27
Glucose, mmol l ⁻¹ Glukoza	16.2 ± 0.55	19.4 ± 0.14	15.2 ± 0.10
HDL, mmol l ⁻¹ Frakcja cholesterolowa HDL	1.07 ± 0.34	1.04 ± 42.94	1.03 ± 0.12
Triglycerides, mmol l ⁻¹ Triglicerydy	2.36 ± 0.79	2.62 ± 0.95	2.70 ± 0.53
ALP, U l ⁻¹ Fosfataza zasadowa	57.25 ± 19.56	59.94 ± 16.61	56.97 ± 14.27
ALT, U l ⁻¹ Aminotransferaza alaninowa	6.21 ± 1.23	6.32 ± 1.25	6.34 ± 0.93
AST, U l ⁻¹ Aminotransferaza asparaginianowa	134 ^b ± 35.0	148 ^a ± 33.0	137 ^{ab} ± 37.8

a, b – p < 0.05 statistically significant difference

a, b – p < 0,05 różnice statystycznie istotne

± standard deviation

± odchylenie standardowe

The increase of glucose level at the 9th week of life was recorded at turkey hens administered with trolox (25.61 mmol l⁻¹). In reference to the control (12.16 mmol l⁻¹), the difference appeared to be statistically significant (p ≤ 0.05). Glucose is a principal energetic substrate for an organism and food (sucrose, starch), glycogen reserves in a liver, and synthesis reactions are its main sources. Glucose concentration in blood results from the following processes: glycogenesis, glycogenolysis, as well as gluconeogenesis and glycolysis.

Data presented in Table 1 reveal that additives, such as trolox or vitamin E, dissolved in drinking water, contributed to the significant increase of HDL cholesterol level (4.47 and 3.34 mmol l⁻¹, respectively) as compared to the control (1.24 mmol l⁻¹). Considerable differences in HDL cholesterol contents could be observed at the 9th week of turkey hen's life (after 4-week administration of additives). A significant increase of that item was also recorded at the end of experiment (15th week of life). When comparing to the control (1.04 mmol l⁻¹), the changes were as follows: 1.43 mmol l⁻¹ for trolox and 1.34 mmol l⁻¹ for vitamin E.

Table 3. Biochemical indices levels in blood plasma of 15-week-old (10th observation week) turkey hens administered with vitamin E and trolox additives

Tabela 3. Poziom wskaźników biochemicznych w osoczu krwi 15-tygodniowych indyczek (10 tydzień obserwacji) otrzymujących do wody dodatek witaminy E i troloxu

Specification Wyszczególnienie	Control Kontrola	Trolox Trolox	Vitamin E Witamina E
Urea, mmol l ⁻¹ Mocznik	7.20 ± 0.15	7.06 ± 0.14	6.97 ± 0.21
Uric acid, mmol l ⁻¹ Kwas moczowy	0.68 ± 0.01	0.52 ± 0.03	0.68 ± 0.02
Protein, g dl ⁻¹ Białko	3.86 ± 0.25	4.21 ± 0.26	3.86 ± 0.54
Cholesterol, mmol l ⁻¹ Cholesterol	3.23 ± 0.45	3.29 ± 0.48	3.06 ± 0.15
Glucose, mmol l ⁻¹ Glukoza	18.15 ± 2.9	16.93 ± 0.96	17.53 ± 2.16
ALP, mmol l ⁻¹ Fosfataza zasadowa	1.04 ^b ± 0.16	1.43 ^a ± 0.42	1.34 ^a ± 0.42
Triglycerides, mmol l ⁻¹ Triglicerydy	1.45 ± 0.86	1.59 ± 1.77	1.45 ± 1.80
ALP, U l ⁻¹ Fosfataza zasadowa	58.4 ± 12.4	60.3 ± 11.4	57.5 ± 10.8
ALT, U l ⁻¹ Aminotransferaza alaninowa	6.49 ± 0.75	6.91 ± 1.16	6.82 ± 0.46
AST, U l ⁻¹ Aminotransferaza asparaginianowa	132 ± 26.1	139 ± 31.0	137 ± 28.2

a, b – p < 0.05 statistically significant difference

a, b – p < 0,05 różnice statystycznie istotne

± standard deviation

± odchylenie standardowe

Table 4. Hematological indices levels in blood plasma of 9-week-old (4th observation week) turkey hens administered with vitamin E and trolox additives

Tabela 4. Poziom wskaźników hematologicznych we krwi 9-tygodniowych indyczek (4 tydzień obserwacji) otrzymujących do wody dodatek witaminy E i troloxu

Specification Wyszczególnienie	Control Kontrola	Trolox Trolox	Vitamin E Witamina E
Hb, mmol l ⁻¹ Hemoglobina	10.2 ± 0.75	10.7 ± 0.48	10.6 ± 0.54
Ht, l l ⁻¹ Hematokryt	31.0 ± 5.42	34.0 ± 3.25	34.0 ± 4.25
WBC, 10 ⁹ l ⁻¹ Białe ciała krwi	30.2 ± 3.02	30.2 ± 2.08	29.5 ± 5.06

± standard deviation

± odchylenie standardowe

Table 5. Hematological indices levels in blood plasma of 11-week-old (6th observation week) turkey hens administered with vitamin E and trolox additives

Tabela 5. Poziom wskaźników hematologicznych w osoczu krwi 11-tygodniowych indyczek (6 tydzień obserwacji) otrzymujących do wody dodatek witaminy E i troloxu

Specification Wyszczególnienie	Control Kontrola	Trolox Trolox	Vitamin E Witamina E
Hb, mmol l ⁻¹ Hemoglobina	9.25 ± 0.28	10.2 ± 0.31	9.29 ± 0.75
Ht, l l ⁻¹ Hematokryt	34.9 ± 1.62	31.0 ± 2.85	33.0 ± 2.85
WBC 10 ⁹ l ⁻¹ Białe ciała krwi	31.0 ± 5.04	29.8 ± 3.81	30.6 ± 4.02

± standard deviation

± odchylenie standardowe

Table 6. Hematological indices levels in blood plasma of 15-week-old (10th observation week) turkey hens administered with vitamin E and trolox additives

Tabela 6. Poziom wskaźników hematologicznych w osoczu krwi 15-tygodniowych indyczek (10 tydzień obserwacji) otrzymujących do wody dodatek witaminy E i troloxu

Specification Wyszczególnienie	Control Kontrola	Trolox Trolox	Vitamin E Witamina E
Hb, mmol l ⁻¹ Hemoglobina	9.88 ± 0.83	10.03 ± 0.66	10.48 ± 0.58
Ht, l l ⁻¹ Hematokryt	34.5 ± 3.02	32.9 ± 4.20	34.2 ± 2.31
WBC, 10 ⁹ l ⁻¹ Białe ciała krwi	28.5 ± 4.25	30.1 ± 2.42	29.9 ± 3.52

± standard deviation

± odchylenie standardowe

Despite of recorded differences between turkey hen groups, achieved results of urea, glucose, and HDL cholesterol contents appeared to be similar to those reported by other authors [Vasicek *et al.* 1991, Koncicki *et al.* 1999, Krasnodębska-Depta and Koncicki 2000, Sembratowicz 2004].

Available literature references provide with many papers on vitamin C application as an additive to bird's fodder. On the contrary, there are few works upon the influence of vitamin E as a fodder additive for turkey hens. Both vitamin C and vitamin E are anti-oxidants, thus their effects on the HDL cholesterol fraction level may be similar as well.

Studies performed by Badzian *et al.* [2001] upon chickens administered with soybean oil and vitamin C revealed a significant decrease of LDL cholesterol fraction, while HDL cholesterol fraction increase was observed only when only ascorbic acid was given. The decrease of the total cholesterol level in turkey hen's blood plasma was reported by Faruga and Jankowski [1996] who applied herbal mixture Melemix. Sembratowicz [2004] also made similar observations when fed turkey hens with water extract of birdweed (*Polygonum aviculare*).

Available literature supplies only information on vitamin E application to reduce the stress at animals. Knapik-Czajka [1998] found that vitamin E has protective effects on adrenal cortex and enhances the ability of fighting against stresses through inhibiting the catabolic action of glycocorticosteroids. A slight reduction in the stress reaction at transported birds expressing as a bit lower corticosterone level, was reported by Wójcik *et al.* [2001], who given vitamin E and vitamin A to broiler-chickens; it led to the decrease of MDA level in blood plasma.

It is difficult to univocally conclude on the trolox influences on biochemical indices at turkey hen's blood plasma due to no apparent trends within achieved results and only incidental increase of some of them.

No significant differences between experimental groups were recorded in reference to biochemical and hematological assays of studied birds' blood plasma (Tab. 3–6).

Any significant influence of applied experimental agents, in a form of drinking water additives, on majority of studied biochemical, namely hematological indicators was not found. However, taking into account some increase of HDL cholesterol fraction and urea levels, it can be supposed that their administration may bring positive effects manifesting as a stimulation of antioxidation mechanisms.

CONCLUSIONS

1. Administration of vitamin E (tocopherol) and trolox to drinking water for turkey hens did not cause any significant changes in shaping the hematological indices at their blood plasma.

2. Considerable increase of urea and HDL cholesterol fraction level, was recorded in turkey hens fed with vitamin E and trolox additives.

3. Achieved results indicate the possibility of applying vitamin E and trolox as antioxidants added to drinking water for turkey hens. However, these findings need to be verified in further studies comprising larger bird populations and wider assays spectrum.

REFERENCES

- Badzian B., Nagórska-Stasiak B., Kowalczyk M., 2001. Wpływ oleju sojowego i witaminy C na poziom lipidów w surowicy krwi kurcząt. *Med. Wet.*, 57, 12, 921–923.
- Bomski J., 1989. Podstawowe laboratoryjne badania hematologiczne. PZWŁ, Warszawa.
- Faruga A., Jankowski J., 1996. 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
- Knapik-Czajka M., 1998. Rola wybranych antyoksydantów pokarmowych w ochronie frakcji LDL przed utlenieniem. *Bromat. Chem. Toksykol.*, 2, 93–98.
- Koncicki A., Krasnodębska-Depta A., Guiro S., 1999. Wskaźniki hematologiczne i biochemiczne krwi w przebiegu histomonadozy indyków. *Med. Wet.*, 55, 10, 647–677.
- Pinkiewicz E. (red.), 1971. Diagnostyka laboratoryjna chorób zwierząt. WSR Lublin.
- Sembratowicz I., 2004. Wpływ wyciągów ziołowych na wybrane wskaźniki krwi i efekty odchowu indyczek. *Annales UMCS, sec. EE, Zootechnica*, 22, 325–332.

- Wójcik A., Sowińska J., Iwańczuk-Czernik, 2001. Zastosowanie u indyków średniociężkich witaminy E i selenu przed obrotem przedubojowym w celu łagodzenia stresu transportowego. *Folia Univ. Agric. Stetin., Zootechnica* 42, 169–174.
- Vasicek V.L., Schwendenwein L., Voll S., 1991. Chemische Blutuntersuchung zur Erstellung von Richtwerten bei Mastputen verschiedenen Alters. *Dt. Tierärztl. Wschr.*, 98, 126–129.

Streszczenie. Celem badań było określenie wpływu witaminy E i troloxu (preparatu witaminy E) dodawanych do wody na wskaźniki hematologiczne i biochemiczne krwi indyczek. Materiał badawczy stanowiły sześciotygodniowe indyczki typu BIG-6 utrzymywane od 6 do 16 tygodnia życia. Zwierzęta podzielono na 3 grupy doświadczalne. I – kontrola, II – z dodatkiem witaminy E w ilości 1,1 ml /kg m.c./dzień, III – z dodatkiem rozpuszczonego w 10 ml wody preparatu trolox w ilości 0,95 mg/kg m.c./dzień. Indyczki otrzymywały *ad libitum* standardowe mieszanki pełnoporcjowe firmy Provimi Polska zgodnie z programem obejmującym 5 okresów żywieniowych. Oznaczono we krwi aktywność aminotransferazy asparaginianowej, aminotransferazy alaninowej, fosfatazy zasadowej, poziom białka całkowitego, kwasu moczowego, triglicerydów cholesterolu całkowitego, frakcję lipoproteinową o wysokiej gęstości (HDL) oraz zawartość glukozy przy użyciu monoteistów firmy Cormay. Oznaczono również poziom kortykosteronu metodą radio-kompensacyjną. Oznaczenia wskaźników hematologicznych dotyczyły: liczby hematokrytowej, zawartości hemoglobiny oraz liczby krwinek białych. Nie stwierdzono istotnych zmian w kształtowaniu się wskaźników hematologicznych krwi indyczek. Dodatek witaminy E i troloxu spowodował wzrost zawartości mocznika oraz frakcji HDL cholesterolu w osoczu krwi. Wyniki wskazują na możliwość zastosowania witaminy E i troloxu jako dodatku antyoksydacyjnego do wody dla indyczek.

Słowa kluczowe: witamina E, trolox, wskaźniki hematologiczne i biochemiczne krwi, indyczki