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IMPACT OF FORMS OF SELENIUM AND SUPPLEMENTAL VITAMIN E ON RABBITS' GROWTH, SLAUGHTER PERFORMANCE AND MUSCLE QUALITY*

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ABSTRACT

Nutrition is vital for raising healthy animals and ensuring higher quality production. To achieve these goals, an appropriate dose and form of antioxidants should be chosen when modulating the animal diet. The aim of this study was to evaluate the impact of selenium forms and supplemental vitamin E on rabbits' growth and slaughter performance as well as muscle quality traits. Forty-five weaned Californian rabbits (42-d-old) were randomly assigned to 3 dietary treatments: standard compound diet (SCD), standard compound diet + 0.5 mg kg⁻¹ organic Se + 100 mg kg⁻¹ vitamin E (OSe+VE), standard compound diet + 0.5 mg kg⁻¹ inorganic Se + 100 mg kg⁻¹ vitamin E (ISe+VE). At the end of the feeding trial, 21 rabbit were randomly selected and euthanized. Slaughter performance was evaluated. *Longissimus dorsi* and hind leg muscle samples were collected post-mortem and further used to determine physicochemical composition, cholesterol, MDA levels in fresh and stored samples, selenium and vitamin E accu-

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mulation, and the profile of fatty acids. OSe+VE and ISe+VE treatments did not affect rabbits' initial and final body weights or slaughter performance ($P \geq 0.05$). However, after evaluating muscle quality traits, our study results showed that OSe+VE and ISe+VE treatments can increase protein levels in the hind leg, while ISe+VE can reduce *Longissimus dorsi* b^* value ($P \leq 0.05$). Experimental treatments improved selenium and vitamin E accumulation in *Longissimus dorsi* and hind leg muscles ($P \leq 0.05$) and increased PUFA ($P \leq 0.05$). The OSe+VE treatment reduced AI, TI indexes, but increased the h/H ratio ($P \leq 0.05$). In conclusion, both Se forms and supplemental vitamin E can be used for growing rabbits without causing any adverse effects on the growth. However, according to rabbits' muscle quality results, the OSe+VE treatment has better impact, leading to higher muscles quality than achieved when SCD and ISe+VE were given.

Keywords: antioxidants, trace minerals, meat quality, accumulation, fatty acids.

INTRODUCTION

Along with the growth of consumption of meat from different species of animals, consumers' interest in higher quality production is rising (POGORZELSKA et al. 2013). High-quality products are often equated to healthy food and rabbit meat presents many beneficial features, which means the rabbit can be theoretically considered as an ideal meat-producing animal. It has a short life cycle, a short gestation period, is particularly prolific and has a high feed conversion efficiency (LEBAS et al. 1986). According to its digestive physiology, rabbits can also ingest cellulose-rich feed and convert about 20% of its protein into edible meat (DALLE ZOTTE 2014). Another essential aspect is that rabbits do not compete with humans for food, making them useful in the sustainable livestock industry. When it comes to the nutritional value, rabbit meat is considered as very healthy food, low in fat, cholesterol and sodium but very rich in proteins (DALLE ZOTTE 2014).

Nutrition plays a vital role in raising a healthy animal and ensuring high quality production. Trace elements along with vitamins are essential nutrients, which act in the same manner in the body of every animal. They are critical in almost every aspect of how a body grows, develops, functions and reproduces (TUFARELLI et al. 2014). For example, selenium is a significant trace mineral and its deficiency has a harmful impact on human and animal wellbeing (KIELISZEK, BŁAŻEJAK 2016). However, it is critical to underline that an appropriate dose and form of selenium should be chosen for animals and humans to reach the best results and to avoid potential toxicity. Various dietary strategies in animal feeding have been developed for providing selenium-enriched meat in order to increase human selenium intake (ZHANG et al. 2010). Animals' diet can be supplemented with two selenium forms: inorganic (sodium selenite and sodium selenate) and organic one (selenium yeast and selenium methionine). However, the metabolism of these two selenium forms in an animal body's is different (SCHRAUZER 2003). For example, the organic form has a more positive impact owing to higher bioavailability

and lesser toxicity compared to the inorganic form, which may be harmful when its dietary concentrations are increased (HASSAN et al. 2015).

Feed can be modified by combining selenium with other additional antioxidants to enhance the action of Se. For example, vitamin E can act as an antioxidant in combination with selenium. Vitamin E has been reported to be an excellent biological antioxidant that protects cells and tissues from free radicals which cause lipoperoxidative harm (KOINARSKI et al. 2005). Selenium works in conjunction with vitamin E, preventing and repairing cellular damage in an animal's body. Therefore, deficiency of these two elements can impair the immune response. Research results indicate that vitamin E supplementation has a beneficial effect on meat quality, and it may be even more effective in combination with selenium by improving the oxidative defence system of cells and tissues (KRSKA et al. 2001).

However, unlike other animal species, information on the effect of diverse selenium forms and supplemental vitamin E in rabbits' diets is limited. Little attention has been paid to how different Se forms, organic and inorganic ones, with additional inclusion of vitamin E in rabbits' diet affect their growth, slaughter performance and meat quality. Thus, the present study was designed to evaluate the impact of different selenium forms and supplemental vitamin E on rabbits' growth, slaughter performance and muscle quality traits.

MATERIAL AND METHODS

Research has been carried out in accordance with Directive 2010/63/EU of the European Parliament and of the Council of 22 September 2010 on the protection of animals used for scientific purposes and compliant with the Commission's recommendation of 18 June 2007 concerning the protection of animals kept for farming purposes.

Experimental design

The trial was carried out on forty-five weaned Californian rabbits, aged 42 d. All 45 rabbits were randomly assigned to 3 treatments ($n=15$ rabbits/treatment) and were fed twice a day with a standard compound diet (inorganic Se, sodium selenite) 0.31 mg kg^{-1} ; vitamin E 50.30 mg kg^{-1}), SCD supplemented with 0.50 mg kg^{-1} organic Se (selenium methionine) and 100 mg kg^{-1} vitamin E (OSe+VE), SCD supplemented with 0.50 mg kg^{-1} inorganic Se (sodium selenite) and 100 mg kg^{-1} vitamin E (ISe+VE). The standard compound diet was formulated and analysed so as to cover the nutrient requirements of growing rabbits, including vitamins and minerals, as recommended by the National Research Council (ARRINGTON et al. 1977) – Table 1. Throughout the study, all the groups received the rations twice a day with an *ad libitum* access to the feed, supplied in the form of pellets.

Table 1

Ingredients and chemical composition of standard compound diet (42-112 day of age)

Ingredients (%)	
Corn	5.00
Oat	18.32
Sunflowers	14.08
Yeast	2.00
Wheat	5.00
Vegetable oil	1.00
Sugar beet slices	10.00
Toxin binder	0.10
Linseed cake	1.00
Vitamin and mineral premix*	3.50
Minced hay	40.00
Total	100
Chemical composition (% , unless stated otherwise)	
Digestive energy (kcal)	2370.70
Metabolism energy (kcal)	2257.20
Crude proteins	16.40
Crude fibre	16.39
Dry matter	89.35
Methionine	0.39
Methionine + cysteine	0.65
Lysine	0.65
Tryptophan	0.20
Linoleic acid	1.04
Calcium [#]	1.29
Phosphorus [#]	0.59
Available phosphorus	0.37
Sodium	0.25
Choline	0.54

[#] analysed value;* Vitamin and mineral premix (per kg of feed): vitamin A 10.08 TV, vitamin D₃ 1.14 TV, vitamin E 50.30 mg, vitamin K₃ 0.99 mg, vitamin B₁ 3.71 mg, vitamin B₂ 2.80 mg, vitamin B₅ 9.80 mg, vitamin B₁₂ 0.01 mg, nicotinic acid 20.40 mg, folic acid 0.22 mg, choline chloride 170.00 mg, Mg 76.28 mg, Fe 317.00 mg, Zn 110.89 mg, Cu 19.16 mg, Co 0.29 mg, I 0.67 mg, Se 0.31 mg (inorganic form).

To ensure optimal health conditions and maximum performance of rabbits, animals were farmed indoors in individual cages. The animals were housed in wire cages with the grid floor (34 × 34 × 61 cm; 1 rabbit per cage) and had free access to individual bowls with clean drinking water and feed. The heating system in the building maintained the temperature of 19±2°C. Housing standards were in accordance with the Council Directive 98/58/EC of 20 July 1998 concerning the protection of animals kept for farming purposes.

Growth and slaughter performance evaluation

All rabbits were weighed individually at the beginning (42 d of age) and at the end of the trial (112 d of age). The average daily weight gain (ADG), daily feed intake (DFI) and feed conversion ratio (FCR) were calculated for each experimental group during the whole feeding trial (42 - 112 d).

At the end of the feeding trial (112 d of age), 21 rabbits ($n=7$ rabbits/treatment) were randomly selected, weighed, starved overnight, and then euthanized in accordance with normal farming practice. Slaughter was carried out at a commercial rabbit farm slaughterhouse in line with the established procedures which comply with the law of the Republic of Lithuania (Order No B1-866 of 31 October 2012 of the Director of the State Food and Veterinary Service on the approval of requirements for the keeping, care and use of animals for scientific and educational purposes).

***Longissimus dorsi* and hind leg muscles samples collection**

The carcasses of rabbits were prepared as reported by BLASCO, OUHAYOUN (2010) and chilled at 4°C for 24 h in a ventilated room. From the reference carcasses, the *Longissimus dorsi* and hind leg muscles were separated. The dissection procedures of warm and chilled carcasses followed the WRSA recommendations (BLASCO, OUHAYOUN 2010). 42 rabbit muscle samples ($n=7$ *Longissimus dorsi*/treatment; $n=7$ hind leg/treatment) were collected post-mortem, minced and stored at -80°C (as fresh meat) and at -18°C (as stored meat) for later malondialdehyde (MDA) analysis. Other meat quality traits (physicochemical indicators, cholesterol levels, Se and vitamin E accumulation, fatty acids profile) were analysed on fresh meat.

Physicochemical assay of muscles

Chemical and physical composition was determined on 42 rabbit muscle samples ($n=7$ *Longissimus dorsi*/treatment; $n=7$ hind leg/treatment). Total protein content was determined by the Kjeldahl method (KING-BRINK, SEBRANEK 1993); fat content – in a Soxtec System by extraction (C. Gerhardt GmbH and Co. KG, Germany) with petrol ether at 40-60°C. Crude fat residue was determined gravimetrically after drying; the ash content was determined by incinerating the samples in a furnace at 600°C (AOAC 2006).

The dry matter content of different rabbit muscles was determined by drying samples at 60°C, after which they were equilibrated to room humidity overnight, milled and passed through a 1-mm sieve and further dried at 105°C to constant weight. Using a Chroma Meter CR-410 Colour Gauge (Konica Minolta, Inc., Osaka, Japan), muscle colour coordinates were defined in the same contrast colour space. Coordinates L^* , a^* , b^* were measured in the light reflectance mode (respectively, the coordinates of brightness, redness, yellowness on the CIE-LAB scale). The standard light source C, whose radiation is close to average daylight, was used for the measurements. Before each measurement, the instrument was calibrated with a light trap and white standard.

Cholesterol and lipid oxidation levels

The cholesterol content in rabbit *Longissimus dorsi* and hind muscles was determined on 42 muscles samples ($n=7$ *Longissimus dorsi*/treatment; $n=7$ hind leg/treatment) according to the method described by POLAK et al. (2008). A high pressure gradient HPLC system Varian ProStar (Varian, Inc., Palo Alto, USA) was used for the cholesterol content determination. Cholesterol separation was performed on a LiChrospher 100 RP-18e (150 × 4.6 mm, 5 µm) chromatography column (Alltech Associates Inc., Deerfield, USA), and the chromatogram was processed at a wavelength of 210 nm. As the mobile phase, a mixture of acetonitrile and 2-propanol (55:45) at a flow rate of 1.0 ml min⁻¹ was used. The chromatography data were summarized and averaged from 7 replicate rabbit muscle samples for each treatment.

An assay of lipid oxidation levels (malondialdehyde (MDA) content) was conducted on 42 fresh muscles samples ($n=7$ *Longissimus dorsi*/treatment; $n=7$ hind leg/treatment) and 42 muscles samples after storage ($n=7$ *Longissimus dorsi*/treatment; $n=7$ hind leg/treatment). The MDA content in the samples was tested at two intervals: 24 h post-mortem and after 3 months following the slaughter; the determinations were carried out by the high performance liquid chromatography method described by MENDES (2009). For this purpose, a high-pressure gradient HPLC system Varian ProStar (Varian, Inc., Palo Alto, USA) with a ProStar 363 fluorescence detector was used. The separation of malondialdehyde-2-thiobarbituric acid (MDA-TBA) was performed on a Gemini C18 (250 × 4.6 mm, 5 µm) chromatographic column (Phenomenex, Inc., Torrance, USA). The mobile phase consisting of 50 mM KH₂PO₄, methanol and acetonitrile at a ratio of 72:17:11 was supplied at a flow rate of 1.0 ml min⁻¹. The MDA-TBA compound was identified and quantified by measuring the fluorescence at E_x 525 nm - E_m 560 nm wavelengths. MDA-TBA was quantified by comparison between the peak area of the MDA-TBA compound in a sample and the peak area of this compound in standard solution.

Accumulation of selenium and vitamin E

To determine the selenium accumulation in *Longissimus dorsi* and hind leg muscles ($n=7$ *Longissimus dorsi*/treatment; $n=7$ hind leg/treatment), samples were digested in a mixture of HNO_3 and H_2O_2 , then placed in Teflon high-pressure vessels and in a Mars 5 microwave oven (CEM Corp., Matthews, USA). After mineralization, Se accumulation was quantified by graphite furnace atomic absorption spectrophotometry. For this purpose, a Thermo Fisher iCE 3000 atomic absorption spectrophotometer with a GFS 35Z graphite furnace with the background correction system based on the Zeeman opto-magnetic effect (Thermo Fisher Scientific Inc., Altrincham, United Kingdom) was used. A standard addition method was applied, as described by NEUGEBAUER et al. (2000). Palladium chloride and magnesium nitrate solutions were used as matrix modifiers.

The accumulation of vitamin E in rabbit muscles ($n=7$ *Longissimus dorsi*/treatment; $n=7$ hind leg/treatment) was determined according to the method of HEWAVITHARANA et al. (2004). To determine vitamin E homologues (α -tocopherol, γ -tocopherol), a high pressure gradient HPLC system Varian ProStar (Varian, Inc., Palo Alto, USA) was used. The separation of vitamin E homologues was performed on a YMC-Pack SIL (250×4.6 mm, $5 \mu\text{m}$) chromatography column (YMC Co., Kyoto, Japan) at a 1.0 ml min^{-1} flow rate. The homologues were identified and the chromatograms were processed at E_x 295 nm - E_m 330 nm wavelengths.

Fatty acids profile

Fatty acids from rabbit muscles ($n=7$ *Longissimus dorsi*/treatment; $n=7$ hind leg/treatment) were extracted and methylated according to the PÉREZ-PALACIOS et al. (2012) method. Fatty acid methyl esters were analysed in a Shimadzu GC-2010 gas chromatography system (Shimadzu corp., Kyoto, Japan) using an Rt-2560 column ($100 \text{ m} \times 0.25 \text{ mm ID}$, $0.20 \mu\text{m}$ film), Restek, Bellefonte, PA, USA. Separated fatty acids were identified by comparison of retention times and MS spectra with the standard Supelco 37 Component FE Mix reagent kit (Supelco Analytical, Thermo Scientific, Bellefonte, PA, USA). The analytical settings were as follows: column temp. was 50°C for 4 min, then increased to 240°C , the carrier gas was helium, and the injector and ion source temperatures were 220°C .

The average amount of each fatty acid was used to calculate the total of saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids. Lipid quality indices, atherogenic index (AI) and thrombogenicity index (TI), were calculated according to ULBRICHT, SOUTHGATE (1991):

$$\text{AI} = [\text{C12:0} + (4 \times \text{C14:0}) + \text{C16:0}] / [\Sigma\text{-PUFA } n\text{-6} + \Sigma\text{-PUFA } n\text{-3} + \Sigma\text{-MUFA}].$$

$$\text{TI} = [\text{C14:0} + \text{C16:0} + \text{C18:0}] / [(0.5 \times \Sigma\text{-MUFA}) + (0.5 \times \Sigma\text{-PUFA } n\text{-6}) + (3 \times \Sigma\text{-PUFA } n\text{-3}) + (\Sigma\text{-PUFA } n\text{-3} / \Sigma\text{-PUFA } n\text{-6})].$$

The hypocholesterolemic/hypercholesterolemic (h/H) ratio was calculated according to FERNÁNDEZ et al. (2007):

$h/H = (C18:1\ n-9 + C18:2\ n-6 + C20:4\ n-6 + C18:3\ n-3 + C20:5\ n-3 + C22:5\ n-3 + C22:6\ n-3) / (C14:0 + C16:0)$.

Statistical analysis

Data analysis was performed in SPSS for Windows, version 25.0 (IBM SPSS Inc., IL., USA, 2017). One-way analysis of variance (ANOVA) post-hoc test (Fisher-LSD) was conducted to detect differences among groups. A *P* value of less than 0.05 ($P \leq 0.05$) was considered statistically significant.

RESULTS

Growth and slaughter performance

The impact of the different forms of Se and supplemental vitamin E on rabbits' growth performance is presented in Table 2. Neither Se forms nor higher vitamin E levels (OSe+VE and ISe+VE, respectively) showed any significant impact on rabbits' initial and final body weights ($P \geq 0.05$). During the whole feeding trial (42 - 112 d), the highest average daily gain (ADG) and feed conversion ratio (FCR) were observed in OSe+VE treatment ($P \leq 0.05$). Rabbits fed with organic Se (OSe+VE) consumed more feed and had significantly higher daily feed intake (DFI) compared to ISe+VE and SCD treatments ($P \leq 0.05$).

Table 2
The impact of the forms of Se and supplemental vitamin E on rabbits' growth performance (42 - 112 day of age; $n=45$)

Parameters	SCD	OSe+VE	ISe+VE	SEM	<i>P</i> value
Initial body weight (g)	839.49	839.43	838.08	21.89	.951
Final body weight (g)	2434.31	2474.58	2447.22	27.82	.162
ADG (g)	22.72 ^a	23.40 ^b	22.99 ^{ab}	0.26	.011
DFI, (g)	70.53 ^a	71.88 ^b	71.11 ^a	0.38	.001
FCR	3.51 ^a	3.69 ^b	3.57 ^{ab}	0.08	.027

SCD – standard compound diet; OSe+VE – SCD + 0.5 mg kg⁻¹ organic Se + 100 mg vitamin E; ISe+VE – SCD + 0.5 mg kg⁻¹ inorganic Se + 100 mg vitamin E;

^{a,b} Means in a row with different superscripts differ ($P \leq 0.05$);

SEM – standard error of means;

ADG – average daily gain; DFI – daily feed intake; FCR – feed conversion ratio.

After evaluating slaughter performance, it emerged that the different Se forms and supplemental vitamin E did not have any significant impact the slaughter performance traits ($P \geq 0.05$) – Table 3.

Table 3

The impact of the forms of Se and supplemental vitamin E on rabbits' slaughter performance (112 day of age; $n=21$)

Parameters	SCD	OSe+VE	ISe+VE	SEM	<i>P</i> value
Pre-slaughter weight (g)	2505.18	2507.80	2506.40	57.03	.964
Warm carcass (g)	1397.66	1423.80	1403.80	40.91	.535
Chilled carcass (g)	1364.77	1390.14	1375.58	54.97	.653
Hind part (%)	34.27	36.17	33.50	2.53	.312
Fore part (%)	24.85	25.67	24.91	1.08	.463
Loin (%)	14.96	15.22	14.63	0.68	.404
Hind leg muscles (%)	20.71	21.65	21.04	0.88	.310
Perirenal fat (%)	1.88	2.09	1.92	0.22	.373
Carcass yield (%)	73.02	73.96	72.84	1.32	.413

SCD – standard compound diet; OSe+VE – SCD + 0.5 mg kg⁻¹ organic Se + 100 mg vitamin E; ISe+VE – SCD + 0.5 mg kg⁻¹ inorganic Se + 100 mg vitamin E; SEM – standard error of means.

Chemical and physical composition of muscles

No significant influence of the Se forms and vitamin E supplementation were found after evaluating the chemical composition of rabbits' muscles (protein, fat, ash) ($P \geq 0.05$) (Table 4). However, a significantly higher protein content was found in hind leg muscle samples in OSe+VE and ISe+VE treatments ($P \leq 0.05$) compared to SCD. Likewise, the dry matter of both muscles did not show any significant differences between the dietary treatments ($P \geq 0.05$). No significant differences were found among the dietary treatments in L^* (brightness) and a^* (redness) values of rabbits *Longissimus dorsi* and hind leg muscles ($P \geq 0.05$). Excluding rabbits fed with the ISe+VE diet, *Longissimus dorsi* muscle colour was significantly less yellowish according to b^* coordinate and compared to SCD ($P \leq 0.05$).

Cholesterol and lipid oxidation levels

Comparing cholesterol levels in different rabbits' muscles types, higher cholesterol levels were determined in hind leg muscle than in *Longissimus dorsi* (Table 4). Cholesterol levels in *Longissimus dorsi* muscles tended to slightly increase, in contrast to hind leg muscles, in which they slightly decreased. However, the cholesterol data obtained in our study are not statistically significant ($P \geq 0.05$).

Table 4 shows lipid oxidation levels (malondialdehyde MDA content) in fresh and stored (3 months at -18°C) rabbit muscle samples after the diet of rabbits was enriched with organic and inorganic Se and supplemental vitamin E. However, no significant differences between MDA levels in the separate treatments were observed ($P \geq 0.05$). Comparing different Se forms,

Table 4

The impact of the forms of Se and supplemental vitamin E on rabbits' *Longissimus dorsi* and hind leg muscle quality traits

Parameters	SCD	OSe+Ve	ISe+VE	SEM	<i>P</i> value
	<i>Longissimus dorsi</i> muscles				
Protein (%)	21.37	20.92	21.72	0.62	.222
Fat (%)	2.30	2.81	2.00	0.41	.069
Ash (%)	1.39	1.47	1.40	0.10	.490
DM (%)	25.72	26.40	25.12	0.82	.145
L* value	56.58	56.68	57.48	1.13	.440
a* value	11.18	11.61	10.95	0.57	.267
b* value	5.91 ^a	5.15 ^{ab}	4.93 ^b	0.40	.029
Cholesterol (mg 100 g ⁻¹)	45.33	45.87	46.34	1.44	.498
MDA (µmol kg ⁻¹)					
Fresh	0.73	0.66	0.79	0.07	.111
After 3 months	1.09	0.97	1.15	0.18	.329
	Hind leg muscles				
Protein (%)	20.88 ^{ab}	20.54 ^a	21.52 ^b	0.35	.015
Fat (%)	3.63	3.73	4.06	0.35	.243
Ash (%)	1.22	1.16	1.04	0.09	.078
DM (%)	26.10	25.48	26.81	0.69	.078
L* value	55.44	55.54	55.81	2.20	.870
a* value	12.96	13.24	12.37	1.54	.582
b* value	4.02	4.04	4.12	0.63	.879
Cholesterol (mg 100 g ⁻¹)	56.17	55.58	56.12	2.55	.820
MDA (µmol kg ⁻¹)					
Fresh	1.38	1.39	1.44	0.19	.762
After 3 months	1.81	1.83	2.21	0.25	.128

SCD – standard compound diet; OSe+VE – SCD + 0.5 mg kg⁻¹ organic Se + 100 mg vitamin E; ISe+VE – SCD + 0.5 mg kg⁻¹ inorganic Se + 100 mg vitamin E;

^{a,b} Means in a row with different superscripts differ ($P \leq 0.05$);

SEM – standard error of means;

DM – dry matter, MDA – malondialdehyde.

the lowest MDA levels were found in the organic form treated (OSe+VE) fresh and stored *Longissimus dorsi* muscles, compared to SCD or ISe+VE. More precisely, MDA levels in fresh and stored *Longissimus dorsi* muscles were about 10% and 11% lower in the OSe+VE compared to the SCD treatment ($P \geq 0.05$). However, the lowest MDA levels in fresh and stored hind leg muscle samples were observed in SCD, slightly higher in OSe+VE and the highest levels were achieved in the ISe+VE treatment ($P \geq 0.05$).

Selenium and vitamin E accumulation in muscles

Table 5 presents the impact of the forms of Se and supplemental vitamin E on the accumulation of Se and vitamin E homologues in rabbits' *Longissimus dorsi* and hind leg muscles. The diet supplemented with the organic Se form (OSe+VE) significantly increased the Se content in *Longissimus dorsi* and hind leg muscles ($P \leq 0.05$). The ISe+VE treatment also tended to increase Se levels in both muscles, but the accumulation was slightly lower compared to the organic form treatment ($P \leq 0.05$).

During our study, the accumulation of vitamin E homologues (α -tocopherol; γ -tocopherol) in rabbit muscles was determined (Table 5). The highest concentration of α -tocopherol was found in *Longissimus dorsi* muscles from the ISe+VE treatment ($P \leq 0.05$); slightly less accumulated in the OSe+VE treatment, and the lowest accumulation was in SCD ($P \leq 0.05$). However, contrary results were received after evaluating hind leg muscles: the highest accumulation of α -tocopherol was observed in the organic selenium treated group (OSe+VE), and lower in SCD ($P \leq 0.05$). A similar trend was reflected by the γ -tocopherol accumulation results: the highest concentration of this homologue was in ISe+VE *Longissimus dorsi* samples, and lower in the SCD and OSe+VE treatments ($P \leq 0.05$). More γ -tocopherol was observed in hind leg muscle samples from the OSe+VE treatment and less – from the SCD treatment ($P \leq 0.05$).

Table 5

The impact of the forms of Se and supplemental vitamin E on accumulation of Se and vitamin E homologues in rabbit *Longissimus dorsi* and hind leg muscles

Parameters	SCD	OSe+VE	ISe+VE	SEM	<i>P</i> value
Se ($\mu\text{g g}^{-1}$)					
<i>Longissimus dorsi</i>	0.34 ^a	0.52 ^b	0.42 ^c	0.02	.000
Hind leg	0.33 ^a	0.42 ^b	0.36 ^a	0.02	.002
α -tocopherol (mg kg^{-1})					
<i>Longissimus dorsi</i>	2.70 ^a	5.31 ^b	6.39 ^b	0.58	.001
Hind leg	2.52 ^a	5.11 ^b	4.09 ^b	0.73	.000
γ -tocopherol (mg kg^{-1})					
<i>Longissimus dorsi</i>	0.29 ^a	0.49 ^b	0.68 ^c	0.06	.015
Hind leg	0.39 ^a	0.55 ^b	0.49 ^{ab}	0.07	.000

SCD – standard compound diet; OSe+VE – SCD + 0.5 mg kg^{-1} organic Se + 100 mg vitamin E; ISe+VE – SCD + 0.5 mg kg^{-1} inorganic Se + 100 mg vitamin E;

^{a,b,c} Means in a row with different superscripts differ ($P \leq 0.05$);

SEM – standard error of means.

Fatty acid profile

Our study results show that some significant effects of the different Se forms and supplemental vitamin E in diets were observed on the profiles of fatty acids in rabbit muscles (Table 6). The proportions of lauric (12:0),

The impact of the forms of Se and supplemental vitamin E on fatty acids profiles in rabbits' *Longissimus dorsi* and hind leg muscles

Item (%)	SCD	OSe+VE	ISe+VE	SEM	<i>P</i> value
	<i>Longissimus dorsi</i> muscles				
	saturated				
1	2	3	4	5	6
C14:1 <i>n</i> -7	0.19	0.13	0.12	0.04	.083
C15:1 <i>cis</i> -10	0.46 ^a	0.00 ^b	0.04 ^c	0.01	.000
C16:1 <i>cis</i> -9 <i>n</i> -9	2.56 ^a	2.51 ^a	3.55 ^b	0.24	.001
C17:1 <i>n</i> -9	0.16 ^{ab}	0.08 ^a	0.23 ^b	0.04	.002
C18:1 <i>cis</i> -9 <i>n</i> -9	26.29	25.69	26.39	0.61	.275
C18:1 <i>trans</i> -9 <i>n</i> -9	1.77 ^a	1.50 ^b	1.34 ^b	0.08	.000
C18:2 <i>n</i> -6	23.04 ^a	24.64 ^b	22.94 ^a	0.55	.010
C18:3 <i>n</i> -6	4.11	3.98	4.16	0.31	.560
C20:1 <i>n</i> -9	0.31 ^a	0.35 ^a	0.23 ^b	0.03	.001
C20:2 <i>n</i> -6	2.19 ^a	0.88 ^b	0.37 ^b	0.42	.001
C20:3 <i>n</i> -3	2.13 ^a	2.72 ^b	2.42 ^c	0.09	.000
C20:4 <i>n</i> -6	0.17 ^a	1.04 ^b	0.88 ^b	0.28	.010
C20:5 <i>n</i> -3	0.12 ^a	0.21 ^b	0.18 ^c	0.01	.000
C22:4 <i>n</i> -6	0.09	0.05	0.09	0.05	.440
C22:5 <i>n</i> -3	0.18 ^a	1.13 ^b	1.18 ^b	0.20	.000
C22:6 <i>n</i> -3	0.14 ^a	0.77 ^b	0.69 ^b	0.08	.000
	Unsaturated				
C4:0	0.06	0.04	0.06	0.02	.199
C6:0	0.02	0.12	0.02	0.06	.121
C8:0	0.01	0.10	0.01	0.05	.099
C10:0	0.05 ^a	0.10 ^{ab}	0.14 ^b	0.03	.015
C12:0	0.14 ^a	0.08 ^b	0.11 ^{ab}	0.02	.011
C14:0	2.38	2.06	2.16	0.17	.082
C15:0	0.45 ^a	0.36 ^b	0.42 ^a	0.02	.001
C16:0	23.49	22.67	22.67	0.45	.091
C17:0	0.44	0.42	0.43	0.02	.323
C18:0	9.07 ^a	8.40 ^b	9.17 ^a	0.27	.014
vSFA	36.11 ^a	34.33 ^b	35.19 ^{ab}	0.59	.011
Σ-MUFA	31.73 ^{ab}	30.25 ^a	31.91 ^b	0.75	.046
Σ-PUFA	32.16 ^a	35.41 ^b	32.90 ^a	0.56	.000
Σ-PUFA/Σ-SFA	0.89 ^a	1.03 ^b	0.94 ^a	0.02	.000
Σ- <i>n</i> -6	29.49 ^{ab}	30.59 ^a	28.54 ^b	0.54	.003

cont. Table 6

1	2	3	4	5	6
Σ - <i>n</i> -3	2.56 ^a	4.82 ^b	4.46 ^b	0.28	.000
Σ - <i>n</i> -6/ Σ - <i>n</i> -3	11.53 ^a	6.41 ^b	6.48 ^b	0.49	.000
AI	0.52 ^a	0.47 ^b	0.48 ^{ab}	0.02	.029
TI	0.91 ^a	0.74 ^b	0.78 ^b	0.03	.000
h/H	2.18 ^a	2.44 ^b	2.38 ^b	0.08	.006
	Hind leg muscles				
	Saturated				
C14:1 <i>n</i> -7	0.15 ^a	0.12 ^a	0.29 ^b	0.03	.000
C15:1 <i>cis</i> -10	0.52 ^a	0.14 ^b	0.04 ^b	0.09	.000
C16:1 <i>cis</i> -9 <i>n</i> -9	1.42	1.53	1.56	0.10	.178
C17:1 <i>n</i> -9	0.22	0.12	0.12	0.05	.071
C18:1 <i>cis</i> -9 <i>n</i> -9	26.23 ^a	23.17 ^b	25.97 ^a	0.69	.001
C18:1 <i>trans</i> -9 <i>n</i> -9	1.83 ^a	1.31 ^b	1.36 ^b	0.15	.005
C18:2 <i>n</i> -6	25.62	26.06	25.35	0.67	.317
C18:3 <i>n</i> -6	2.91 ^a	5.12 ^b	4.34 ^c	0.28	.000
C20:1 <i>n</i> -9	0.48 ^a	0.36 ^b	0.39 ^b	0.03	.001
C20:2 <i>n</i> -6	0.24	0.24	0.25	0.02	.619
C20:3 <i>n</i> -3	0.13	0.35	0.18	0.11	.066
C20:4 <i>n</i> -6	0.40	0.13	0.08	0.16	.067
C20:5 <i>n</i> -3	1.15 ^a	2.10 ^b	1.72 ^{ab}	0.30	.009
C22:4 <i>n</i> -6	0.09 ^a	1.01 ^b	0.26 ^a	0.17	.000
C22:5 <i>n</i> -3	0.33	0.66	0.37	0.21	.141
C22:6 <i>n</i> -3	0.65	0.68	0.66	0.24	.908
	Unsaturated				
C4:0	0.06	0.11	0.13	0.06	.256
C6:0	0.01 ^a	0.01 ^a	0.13 ^b	0.02	.001
C8:0	0.01 ^a	0.01 ^a	0.12 ^b	0.01	.000
C10:0	0.06 ^a	0.18 ^a	0.49 ^b	0.07	.000
C12:0	0.26 ^a	0.42 ^a	1.05 ^b	0.12	.000
C14:0	1.96	2.14	2.01	0.25	.483
C15:0	0.50	0.42	0.35	0.08	.082
C16:0	25.64 ^a	24.14 ^b	24.49 ^b	0.31	.000
C17:0	0.82	0.88	0.85	0.07	.421
C18:0	8.30	8.61	7.45	0.72	.133
Σ -SFA	37.62	36.92	37.07	0.74	.362
Σ -MUFA	30.86 ^a	26.75 ^b	29.72 ^a	0.69	.000
Σ -PUFA	31.52 ^a	36.34 ^b	33.21 ^c	0.75	.000

cont. Table 6

1	2	3	4	5	6
Σ -PUFA/ Σ -SFA	0.84 ^a	0.99 ^b	0.90 ^a	0.03	.001
Σ -n-6	28.99 ^a	32.55 ^b	30.55 ^a	0.73	.000
Σ -n-3	2.26 ^a	3.79 ^b	2.93 ^{ab}	0.60	.025
Σ -n-6/ Σ -n-3	13.88 ^a	9.40 ^b	10.97 ^{ab}	2.05	.050
AI	0.54	0.53	0.53	0.02	.348
TI	0.98 ^a	0.86 ^b	0.87 ^{ab}	0.05	.041
h/H	2.09 ^a	2.26 ^b	2.22 ^b	0.05	.006

SCD – standard compound diet; OSe+VE – SCD + 0.5 mg kg⁻¹ organic Se + 100 mg vitamin E; ISe+VE – SCD + 0.5 mg kg⁻¹ inorganic Se + 100 mg vitamin E;

^{a,b,c} Means in a row with different superscripts differ ($P \leq 0.05$);

SEM – standard error of means;

SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids; AI – atherogenic index; TI – thrombogenicity index; h/H – hypocholesterolemic/hypercholesterolemic ratio.

pentadecanoic (15:0) and stearic (18:0) acids in the OSe+VE treatment *Longissimus dorsi* muscles significantly decreased, compared to the SCD and ISe+VE treatments ($P \leq 0.05$). On the contrary, the dietary ISe+VE supplementation increased the lauric (12:0) acid concentration in the hind leg muscles compared to SCD and OSe+VE ($P \leq 0.05$). Dietary ISe+VE and OSe+VE supplementations decreased the palmitic (16:0) acid content in the hind leg muscles ($P \leq 0.05$). Other changes between individual monounsaturated fatty acids (MUFA) were observed. Pentadecanoic (15:1) and elaidic [18:1 (trans-9 n-9)] acids decreased in samples from all treatments ($P \leq 0.05$). The composition of palmitoleic [16:1 (n-7)] and heptadecenoic [17:1 (n-9)] acids increased in hind leg muscles under the influence of the ISe+VE treatment ($P \leq 0.05$). Among MUFA, oleic [18:1 (cis-9 n-9)] acid underwent the biggest decrease in hind leg muscles from the ISe+VE treatment ($P \leq 0.05$).

The main differences between treatments were observed among polyunsaturated fatty acids (PUFA) in rabbits' *Longissimus dorsi* muscles (Table 6). The proportion of PUFA (except eicosadienoic [20:2 (n-6)] acid) in ISe+VE and OSe+VE treated rabbits' *Longissimus dorsi* muscles significantly increased compared to SCD ($P \leq 0.05$). In particular, the compositions of arachidonic [20:4 (n-6)], docosapentaenoic [22:5 (n-3)] and docosahexaenoic [22:6 (n-3)] acids significantly increased in *Longissimus dorsi* muscles from both experimental treatments ($P \leq 0.05$).

During our study, a positive trend was obtained for both especially important ratios – Σ -PUFA/ Σ -SFA and Σ -n-6/ Σ -n-3 (Table 6). Therefore, the appropriately increasing amount of PUFA in rabbits' muscles positively changed the above ratios in *Longissimus dorsi* and hind leg muscles of rabbits fed OSe+VE ($P \leq 0.05$). The inclusion of organic Se (OSe+VE) lowered AI and TI indexes, whereas the ratio between the hypocholesterolemic and

hypercholesterolemic fatty acids (h/H) was significantly higher in rabbits' *Longissimus dorsi* and hind leg muscles compared to SCD ($P \leq 0.05$).

DISCUSSION

Growth and slaughter performance

Both selenium forms and higher vitamin E levels in diets did not show any significant impact on rabbits' initial and final body weights. However, our study results are in accordance with other research (DOKOUPILOVÁ et al. 2007, MAROUNEK et al. 2009), where no significant effects of Se and higher vitamin E levels on rabbits' body weight was observed. The highest average daily gain and feed conversion ratio were observed under the organic Se treatment. Therefore, rabbits fed with the organic Se enriched diet consumed more feed and had significantly higher daily feed intake compared to rabbits from the other treatments. Similar results were reported by EBEID et al. (2013) who claimed that organic Se and vitamin E addition in rabbits' feed resulted in a higher average daily gain and daily feed intake. However, EL-KHOLY et al. (2019) obtained the opposite results: they reported that using various forms of Se with vitamin E in rabbits' diet lowered the growth performance parameters described above.

According to the results of our research, the addition of inorganic and organic selenium neither depressed nor increased the rabbits' growth performance. The reason may be the fact that organic Se has high absorption, so its sources could pass through the intestinal membrane and enter blood by active transport. Selenium assists in normal cell growth and has an important role in modulating the action of transcription factors and cell signalling systems (KIELISZEK, BŁAŻEJAK 2016). Furthermore, Se improves the metabolism of the thyroid hormones, which is essential for normal growth and metabolism because selenoenzymes modulate or control many phases of thyroid hormone metabolism (MEHDI, DUFRASNE 2016).

One of the most important properties of rabbit meat are pre-slaughter and carcass weights, yield (when whole carcasses are sold), and percentage of parts by weight of the carcass (when carcasses portions are sold). However, our study results show that the different Se forms and supplemental vitamin E did not have significant impact on any slaughter performance traits. Same as in our experiment, HASSAN et al. (2015) did not establish any effect of Se on pre-slaughter and carcass weight of rabbits. The ineffective feed supplementation with different forms of Se was a finding that agrees with other research, where no significant differences in traits of rabbit and broiler carcass were found (MAROUNEK et al. 2009, BAKHSHALINEJAD et al. 2019).

Chemical and physical composition of muscles

For consumers, important meat quality indicators are meat chemical composition, dry matter content and colour. Variation in meat quality depends on the sex and age of an animal, muscle metabolic type, ratio of nutrients and their amount used in the animal's diet. In human nutrition, rabbit meat is ideal for all groups of consumers: it is rich in proteins, B vitamins and minerals, but low in sodium, fat and cholesterol. The rabbit meat energy content (603 kJ 100 g⁻¹ in loin meat and 899 kJ 100 g⁻¹ in foreleg meat, average carcass value) is mostly attributable to proteins (DALLE ZOTTE 2014).

Nevertheless, no significant influence of the two Se forms and vitamin E supplementation were found after evaluating the chemical composition of rabbits' muscles, except one parameter, namely a significantly higher protein content was found in both Se and vitamin E treated hind leg muscle samples. Likewise, chemical composition, dry matter of *Longissimus dorsi* and hind leg muscles did not show any significant differences between treatments. Our study results are similar to the ones reported by DOKOUILOVÁ et al. (2007), who indicated that selenium yeast (inorganic form) supplementation did not have any effect on the dry matter and fat content of rabbit loin meat. MAROUNEK et al. (2009) reported that rabbits' dietary supplementation with different Se forms had no impact on muscle dry matter, protein and fat either. However, contradictory results were reported by other scientists, who claimed that rabbit meat chemical composition was significantly affected by dietary supplementation of various forms of Se with vitamin E (HASSAN et al. 2019). Since the results of other scientists and our study differ, we cannot determine any pattern of meat chemical composition changes when rabbits' diets are supplemented with different forms of Se and with vitamin E.

Among all animal species used for meat production, rabbits produce meat usually characterised by the highest meat brightness. Along with poultry, rabbit meat is qualified as white meat. After slaughter, oxyhaemoglobin in the meat is oxidized to metmyoglobin, which gives the meat a grey tone. Therefore, the balance of these compounds causes discoloration, and it is the antioxidant properties of vitamin E that inhibit the change in oxyhaemoglobin. However, no significant differences were found among dietary treatments in brightness (L*) and redness (a*) coordinates of rabbits' muscles. Excluding rabbits fed with the inorganic Se form, the colour of *Longissimus dorsi* muscles was significantly less yellowish according to the b* coordinate. A similar trend was observed by CALVO et al. (2017), when dietary supplementation with Se-enriched yeast (organic form) resulted in higher b* values in pigs' muscles compared to those from the SeS (inorganic form) treated group. Dietary manipulation like this can have further effects on other quality parameters of the carcass and retail cuts, for example, meat and fat colour, which are important for consumers when deciding to purchase a product. Therefore, additional vitamin E used in this study should theoretically

affect the quality of the meat, as it is used to increase the stability and intensity of the meat colour. However, our study results show no such trend, which agrees with other research, where no significant effects on the colour index of rabbit muscles were found after supplementing the animals diets with vitamin E (CORINO et al. 1999).

Cholesterol and lipid oxidation levels

Rabbit meat has an excellent nutritional value and dietary properties as this is lean meat rich in protein, high in polyunsaturated fatty acids, and very low in cholesterol. Cholesterol levels in rabbit tissues depend primarily on genetic and environmental factors. However, dietary feeds and supplemented additives can also affect cholesterol deposition in tissues (DALLE ZOTTE 2014). In our study, cholesterol levels in *Longissimus dorsi* muscles tended to increase slightly, unlike in hind leg muscles, where they slightly decreased. Our results are contradictory to those reported by SALEH et al. (2013), who found that a diet supplemented with organic selenium can decrease cholesterol levels in rabbit muscles. But since no significant differences were observed between treatments in our study, the mechanism of changes in cholesterol levels in rabbit muscles remains unclear.

The fact that rabbit meat is high in unsaturated fatty acids makes it more sensitive to oxidative processes. Oxidation can cause modifications of lipids and proteins in muscles. Therefore, it can affect the organoleptic and nutritional properties of meat, which can be reflected by economic losses and health disorders (INSANI et al. 2008). Moreover, Se inclusion in rabbits' nutrition helps to ensure lipid oxidative stability and guarantee high meat quality (DOKOUPILOVÁ et al. 2007). Both, Se and vitamin E are essential and highly efficient antioxidants, which helps to protect rabbits against lipid and protein oxidation (EBEID et al. 2013). Nevertheless, during our study, no significant differences in MDA levels between the treatments were observed. However, when comparing the different Se forms, the lowest MDA levels were determined in organic Se form treated fresh and stored *Longissimus dorsi* muscles, suggesting that the organic selenium and vitamin E combination has a positive effect on cell preservation and lipid oxidation due to their synergistic relationship. This can also be explained by the fact that organic Se builds up Se reserves in body tissues, primarily in muscles, in the form of selenium methionine, which could improve the antioxidant defence even during stress conditions (SURAI et al. 2016). MDA levels were measured in rabbits' hind leg muscles as well, which is more sensitive to oxidative impairment due to its higher fat content. The lowest MDA levels were observed in the treatment with standard compound diet, and the highest levels appeared in fresh and stored hind leg muscle samples from the inorganic selenium treatment. Results from an analysis of hind leg muscle samples are in line with other studies, where it was demonstrated that Se supplementation cannot affect the oxidative stability of rabbit meat

(DOKOUPILOVÁ et al. 2007, MAROUNEK et al. 2009). According to CALVO et al. (2017), the oxidative status of samples is related to the storage time. The MDA levels were much higher in muscles samples from the ISe+VE treatment compared to the organic Se supplemented group. This finding agrees with our results where organic Se provided superior antioxidant activity and was more effective in delaying post-mortem oxidation reactions than inorganic selenium. However, it is important to note that differences in the MDA results obtained during our study were not significant.

Selenium and vitamin E accumulation in muscles

The diet supplemented with the organic Se form significantly increased the Se content in *Longissimus dorsi* and hind leg muscles. While the inorganic Se treatment also tended to increase Se levels in both muscles, the Se accumulation was slightly lower. The Se deposition in tissues was consistent with the concept that the organic Se form tends to be deposited more readily than the inorganic one in slowly metabolising tissues, such as loin and thigh muscles (SCHRAUZER 2003). The same trend was reflected in our study: organic Se influenced and improved selenium accumulation in rabbit muscles, thus being higher than when the inorganic Se form was fed. Similar results were reported by other scientists, who indicated that organic Se or organic Se and vitamin E combination are more effective than inorganic selenium in terms of Se deposition in rabbit muscles (MAROUNEK et al. 2009, EBEID et al. 2013, AMER et al. 2019). It is likely that organic sources of Se can be absorbed by active transport and non-specifically incorporated into proteins, which is preferentially absorbed and utilized by the body, unlike the inorganic form (SCHRAUZER 2003).

During our study, accumulation of vitamin E homologues (α -tocopherol; γ -tocopherol) in rabbit muscles was determined. The highest concentration of α -tocopherol was determined in *Longissimus dorsi* muscles under the inorganic Se and additional vitamin E treatment. Nevertheless, contradictory results were received after evaluating hind leg muscles: the highest accumulation of α -tocopherol was observed in the organic Se treated group. A similar trend became apparent after determining the accumulation of γ -tocopherol homologue: the highest content was in the inorganic Se treated *Longissimus dorsi* samples, but greater amount of this homologue was observed in hind leg muscles in the organic Se form and additional vitamin E treatment. Our study results are in line with the data provided from research including similar experiments with rabbits and antioxidant additives (EBEID et al. 2013).

The accumulation of selenium and vitamin E in rabbit muscles was determined considering the synergistic relationship that exists between these substances to protect against cellular damage caused by reactive oxygen species. This synergism may directly enhance meat quality (SURAĪ, DVORSKA 2002). Interestingly, a combination of organic selenium dose of 0.5 mg kg⁻¹

and vitamin E dose of 100 mg kg⁻¹ in a rabbits' diet increased the vitamin E and Se deposition in hind leg muscles. Production of such enriched rabbit meat could be considered as a way to incorporate these vital antioxidants in the human diet. Our results apparently indicate that Se and vitamin E supplementation accelerated the accumulation of both elements in rabbit muscles. This agrees with the opinion of SURAI, DVORSKA (2002), who elucidated the fact that dietary supplementation with organic Se significantly increased the vitamin E concentration in broiler chicken breast muscles. Thus, the synergistic relationship between Se and vitamin E was confirmed and this synergism might enhance meat quality characteristics, which is also revealed in our study.

Profile of fatty acids

Most rabbit meat quality research conducted in recent years has focused on incorporating bioactive compounds in a diet for the benefit to human health. Moreover, rabbit meat consumption could become a good source of these bioactive compounds for consumers, since manipulation of rabbits' nutrition is very effective in increasing levels of n-3 polyunsaturated fatty acids (PUFA). Several scientists have reported that the profile of meat fatty acids can be modified by oxidative processes and that the oxidative stability of lipids is closely associated with composition of fatty acids (MONIN et al. 2003). The susceptibility of unsaturated fatty acids to oxidation is related to the percentage of unsaturation of fatty acids because polyunsaturated fatty acids are more receptive to oxidation than monounsaturated ones. Selenium and vitamin E are major antioxidants that protect tissues from oxidative damage by depositing themselves in cell membranes, where they protect PUFAs from oxidation (KASAPIDOU et al. 2009). The mentioned effect of these antioxidants transfers into the tissues, including muscles, and the result can be improved by regulating the form and doses of these substances in a diet.

Our study results show some significant effects of the different forms of Se and supplemental vitamin E in diets were observed on the profile of fatty acids in rabbit muscles. The main differences between the treatments were observed among PUFAs in rabbit *Longissimus dorsi* muscles. The proportions of PUFAs were significantly increased in *Longissimus dorsi* muscles of rabbits fed the diets with both Se forms and with additional vitamin E. This can be explained by the fact that Se and α -tocopherol may protect PUFAs from oxidation, or they may promote fatty acid synthesis by a positive effect on the activity of the enzymes Δ 4-, Δ 5-, Δ 6- desaturases and elongases involved in the processes leading to the formation of long-chain polyunsaturated fatty acids, LC-PUFA (ALAGAWANY et al. 2019). KRALIK et al. (2012) reported that the n-3 PUFA ratio in breast muscles increased when broiler diets were supplemented with Se. Moreover, according to SURAI, SPARKS (2000), the proportion of n-3 PUFA in different chicken tissues was higher when the diet was supplemented with vitamin E.

During our study, a positive trend was obtained for two especially important ratios, i.e. $\Sigma\text{PUFA}/\Sigma\text{SFA}$ and $\Sigma(n-6)/\Sigma(n-3)$. Therefore, increasing the appropriate amount of PUFAs in rabbit muscles positively changed the $\Sigma\text{PUFA}/\Sigma\text{SFA}$ and the $\Sigma(n-6)/\Sigma(n-3)$ ratios in rabbit *Longissimus dorsi* and hind leg muscles under the organic Se form treatment. Likewise, the dietary inclusion of organic Se lowered AI and TI indexes, whereas the ratio between the hypocholesterolemic and hypercholesterolemic fatty acids (h/H) was significantly higher in rabbit *Longissimus dorsi* and hind leg muscles. According to TURAN et al. (2007), nutritional quality indexes can indicate the potential of a tested sample for plaque aggregation. In other words, low AI and TI values indicate high quantities of anti-atherogenic fatty acids in oil or fat. In our present study, the h/H ratio was higher in rabbit *Longissimus dorsi* and hind leg muscles under the organic Se and supplemental vitamin E treatment in comparison to the standard compound diet. To sum up, our results prove that rabbit meat satisfies the recommendations for a healthier diet: AI and TI should be as low as possible, while the h/H ratio should be high.

CONCLUSIONS

The present findings demonstrated that both selenium forms and supplemental vitamin E can be used for rearing rabbits without causing any adverse effects on the growth, owing to their positive effect on the physico-chemical composition of rabbit muscles. However, it can also be concluded that different selenium forms and vitamin E combinations did not play a direct role in improving rabbits' growth results and slaughter efficiency. Nevertheless, the synergism between these substances reflected in our study indicates that such enriched rabbit meat can be considered as a useful source of these vital antioxidants in the human diet. This is supported by the findings related to the deposition of Se and vitamin E homologues in rabbit muscles. To recapitulate, higher meat quality was achieved under the organic Se and supplemental vitamin E combined treatment, which resulted in higher Se accumulation in both muscles, higher accumulation of vitamin E homologues in hind leg muscles, improved fatty acid profiles, the lowest AI and TI indexes, and the highest h/H ratios in both rabbits' muscles.

Conflict of interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

REFERENCES

- ALAGAWANY M., ELNESR S.S., FARAG M.R., ABD EL-HACK M.E., KHAFAGA A.F., TAHA A.E., TIWARI R., YATOO M.I., BHATT P., KHURANA S.K., DHAMA K. 2019. *Omega-3 and omega-6 fatty acids in poultry nutrition: Effect on production performance and health*. *Animals*, 9(8): 573. DOI: 10.3390/ani9080573
- AMER S.A., OMAR A.E., ABD EL-HACK M.E. 2019. *Effects of selenium- and chromium- enriched diets on growth performance, lipid profile, and mineral concentration in different tissues of growing rabbits*. *Biol. Trace Element Res.*, 187(1): 92-99. DOI: 10.1007/s12011-018-1356-4
- Association of Official Analytical Chemists International 2005. *Official Methods of Analysis*. AOAC 2006, Gaithersburg, Maryland, 18th ed.
- ARRINGTON L.R., CHEEKE P.R., LEBAS F., LEBAS S. 1977. *Nutrient requirements of rabbits*. Second revised edition. National Research Council (NRC), Washington DC.
- BAKSHSHALINEJAD R., HASSANABADI A., SWICK R.A. 2019. *Dietary sources and levels of selenium supplements affect growth performance, carcass yield, meat quality and tissue selenium deposition in broilers*. *Anim. Nutr.*, 5(3): 256-263. DOI: 10.1016/j.aninu.2019.03.003
- BLASCO A., OUHAYOUN J. 2010. *Harmonization of criteria and terminology in rabbit meat research. Revised proposal*. *World Rabbit Sci.*, 4(2): 10-16.
- CALVO L., TOLDRÁ F., RODRÍGUEZ A.I., LÓPEZ-BOTE C., REY A.I. 2017. *Effect of dietary selenium source (organic vs. mineral) and muscle pH on meat quality characteristics of pigs*. *Food Sci. Nutr.*, 5(1): 94-102. DOI: 10.1002/fsn3.368
- CORINO C., PASTORELLI G., PANTALEO L., ORIANI G., SALVATORI G. 1999. *Improvement of color and lipid stability of rabbit meat by dietary supplementation with vitamin E*. *Meat Sci.*, 52(3): 285-289. DOI: 10.1016/S0309-1740(99)00004-2
- DALLE ZOTTE A. 2014. *Rabbit farming for meat purposes*. *Animal Frontiers*, 4(4): 62-67. DOI: 10.2527/af.2014-0035
- DOKOUPILOVÁ A., MAROUNEK M., SKŘIVANOVÁ V., BŘEZINA P. 2007. *Selenium content in tissues and meat quality in rabbits fed selenium yeast*. *Czech J. Anim. Sci.*, 52(6): 165-169.
- EBEID T.A., ZEWEIL H.S., BASYONY M.M., DOSOKY W.M., BADRY H. 2013. *Fortification of rabbit diets with vitamin E or selenium affects growth performance, lipid peroxidation, oxidative status and immune response in growing rabbits*. *Livest. Sci.*, 155(2-3): 323-331. DOI: 10.1016/j.livsci.2013.05.011
- EL-KHOLY K.H., TAG EL-DEEN H.T., ABD-EL-LATEIF A.I., MEKAOUY A.I. 2019. *Effects of dietary selenium sources on metabolic, enzymatic and immunoglobulin serum profiles in growing rabbits*. *Pak. J. Nutr.*, 18(5): 430-436. DOI: 10.3923/pjn.2019.430.436
- FERNÁNDEZ M., ORDÓÑEZ J.A., CAMBERO I., SANTOS C., PIN C., HOZ L. 2007. *Fatty acid compositions of selected varieties of Spanish ham related to their nutritional implications*. *Food Chem.*, 101(1): 107-112. DOI: 10.1016/j.foodchem.2006.01.006
- HASSAN F.A., ABDEL-AZEEM N.M., ABDEL-RAHMAN S.M., AMIN H.F., ABDEL-MAWLA L.F. 2019. *Effect of dietary organic selenium supplementation on growth performance, carcass characteristics and antioxidative status of growing rabbits*. *World's Vet. J.*, 9(1): 16-25. DOI: 10.36380/wvet.2019.vv9j1
- HASSAN F.A., HOBALLAH E.M., BASYONY M.M., EL-MEDANY S. 2015. *Effect of dietary selenium enriched micro-algae supplementation on growth performance and anti-oxidative status of rabbits under high ambient temperature in summer season*. *EJNF*, 18(2): 229-244. DOI: 10.21608/ejnf.2015.105111
- HEWAVITHARANA A.K., LANARI M.C., BECU C. 2004. *Simultaneous determination of vitamin E homologs in chicken meat by liquid chromatography with fluorescence detection*. *J. Chromatogr. A.*, 1025(2): 313-317. DOI: 10.1016/j.chroma.2003.10.052
- INSANI E.M., EYHERABIDE A., GRIGIONI G., SANCHO A.M., PENSEL N.A., DESCALZO A.M. 2008. *Oxidative stability and its relationship with natural antioxidants during refrigerated retail*

- display of beef produced in Argentina.* Meat Sci., 79(3): 444-452. DOI: 10.1016/j.meatsci.2007.10.017
- KASAPIDOU E., ENSER M., WOOD J.D., RICHARDSON R.I., WILKINSON R.G., SINCLAIR L.A. 2009. *Influence of vitamin E supplementation and basal diet on the vitamin E status, performance and tissue fatty acid concentration in lambs.* Animal, 3(4): 516-526. DOI: 10.1017/S1751731108003820
- KIELISZEK M., BŁĄZEJAK S. 2016. *Current knowledge on the importance of selenium in food for living organisms: a review.* Molecules, 21(5): 609. DOI: 10.3390/molecules21050609
- KING-BRINK M., SEBRANEK J.G. 1993. *Combustion method for determination of crude protein in meat and meat products: collaborative study.* J. AOAC Int., 76(4): 787-793.
- KOINARSKI V., GEORGIEVA N., GADJEVA V., PETKOV P. 2005. *Antioxidant status of broiler chickens, infected with Eimeria acervulina.* Rev. Med. Vet., 156(10): 498-502.
- KRALIK Z., KRALIK G., GRČEVIĆ M., SUCHÝ P., STRAKOVÁ E. 2012. *Effects of increased content of organic selenium in feed on the selenium content and fatty acid profile in broiler breast muscle.* Acta Vet. Brno, 81(1): 31-35. DOI: 10.2754/avb201281010031
- KRSKA P., LAHUCKY R., KÜCHENMEISTER U., NÜRNBERG K., PALANSKA O., BAHELKA I., KUHN G., ENDER K. 2001. *Effects of dietary organic selenium and vitamin E supplementation on post-mortem oxidative deterioration in muscles of pigs.* Arch.Tierz., 44(2): 193-201.
- LEBAS F., COUDERT P., ROUVIER R., DE ROCHEMBAU H. 1986. *The rabbit: husbandry, health and production.* FAO Animal Production and Health Series No 21.
- MAROUNEK M., DOKOUPILOVÁ A., VOLEK Z., HOZA I. 2009. *Quality of meat and selenium content in tissues of rabbits fed diets supplemented with sodium selenite, selenized yeast and selenized algae.* World Rabbit Sci., 17(4): 207-212. DOI: 10.4995/wrs.2009.645
- MEHDI Y., DUFRASNE I. 2016. *Selenium in cattle: a review.* Molecules, 21(4): 545. DOI: 10.3390/molecules21040545
- MENDES R., CARDOSO C., PESTANA C. 2009. *Measurement of malondialdehyde in fish: A comparison study between HPLC methods and the traditional spectrophotometric test.* Food Chem., 112(4): 1038-1045. DOI: 10.1016/j.foodchem.2008.06.052
- MONIN G., HORTÓS M., DIAZ I., ROCK E., GARCIA-REGUEIRO J.A. 2003. *Lipolysis and lipid oxidation during chilled storage of meat from Large White and Pietrain pigs.* Meat Sci., 64(1): 7-12. DOI: 10.1016/s0309-1740(02)00130-4
- NEUGEBAUER E.A., SANS CARTIER G.L., WAKEFORD B.J. 2000. *Methods for the determination of metals in wildlife tissues using various atomic absorption spectrophotometry techniques.* Canadian Wildlife Service Environmental Conservation Branch, Canada.
- PÉREZ-PALACIOS T., RUIZ J., FERREIRA I.M.P.L.V.O., PETISCA C., ANTEQUERA T. 2012. *Effect of solvent to sample ratio on total lipid extracted and fatty acid composition in meat products within different fat content.* Meat Sci., 91(3): 369-373. DOI: 10.1016/j.meatsci.2012.02.021
- POGORZELSKA J., MICIŃSKI J., OSTOJA H., KOWALSKI I.M., SZAREK J., STRZYŻEWSKA E. 2013. *Quality traits of meat from young Limousin, Charolais and Hereford bulls.* Pak. Vet. J., 33(1): 65-68.
- POLAK T., RAJAR A., GAŠPERLIN L., ŽLENDER B. 2008. *Cholesterol concentration and fatty acid profile of red deer (Cervus elaphus) meat.* Meat Sci., 80(3): 864-869. DOI: 10.1016/j.meatsci.2008.04.005
- SALEH A.A., EBEID T.A., EID Y.Z. 2013. *The effect of dietary linseed oil and organic selenium on growth performance and muscle fatty acids in growing rabbits.* Pak. Vet. J., 33(4): 450-454.
- SCHRAUZER G.N. 2003. *The nutritional significance, metabolism and toxicology of selenomethionine.* Adv. Food Nutr. Res., 47: 73-112. DOI: 10.1016/s1043-4526(03)47002-2
- SURAI P.F., DVORSKA J.E. 2002. *Effects of selenium and vitamin E content of the diet on lipid peroxidation in breast muscle tissue of broiler hens during storage.* APSS, 14: 187-192.
- SURAI P.F., FISININ V.I., KARADAS F. 2016. *Antioxidant systems in chick embryo development. Part 1. Vitamin E, carotenoids and selenium.* Anim. Nutr., 2(1): 1-11. DOI: 10.1016/j.aninu.2016.01.001

-
- SURAI P.F., SPARKS N.H.C. 2000. *Tissue-specific fatty acid and α -tocopherol profiles in male chickens depending on dietary tuna oil and vitamin E provision*. Poultry Sci., 79(8): 1132-1142. DOI: 10.1093/ps/79.8.1132
- TUFARELLI V., PETRERA F., KHAN R., LAUDADIO V. 2014. *Erratum to: Vitamin and trace element supplementation in grazing dairy ewe during the dry season: effect on milk yield, composition, and clotting aptitude*. Trop. Anim. Health Prod., 46(6): 1103.
- TURAN H., SÓNMEZ G., KAYA Y. 2007. *Fatty acid profile and proximate composition of the thornback ray (*Raja clavata*, L. 1758) from the Sinop coast in the Black Sea*. J. Fish. Sci., 1(2): 97-103. DOI: 10.3153/jfscom.2007012
- ULBRICHT T.L.V., SOUTHGATE D.A.T. 1991. *Coronary disease seven dietary factors*. Lancet, 338(8733): 985-992. DOI: 10.1016/0140-6736(91)91846-m.
- ZHANG W., XIAO S., SAMARAWEEERA H., LEE E.J., AHN D.U. 2010. *Improving functional value of meat products*. Meat Sci., 86(1): 15-31. DOI: 10.1016/j.meatsci.2010.04.018