

# INFLUENCE OF ORGANIC SELENIUM ON HISTOLOGICAL STRUCTURE OF MUSCLE AND QUALITIES OF PORK MEAT\*\*

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\*\* Original scientific paper, presented at 2<sup>nd</sup> International Congress on Animal Husbandry "New Perspectives and Challenges of Sustainable Livestock Farming", Belgrade, 3.-5. October, 2007

**Abstract:** Selenium is one of the most important mineral trace elements, its deficiency has marked effect to utility and health state of animals, and also to health state of humans. Incorporation of Se to muscle tissue depends on dietary level and on selenium form. When we use organic form of selenium, the increase of selenium concentration in muscles is linear. This is important for improvement of pork quality. We compared the influence of selenium (Se) in experimental group with control group of pigs in our experiment. Differences in body weight weren't statistically significant in both groups. The thickness of neck and thickness of spine part was higher in control group. The thickness of fat cells was higher in experimental group in both muscles (39,17 µm, alternatively 36,35 µm,  $P < 0,05$ ). The presence of interstitial tissue in percentage was statistical significant and it was higher in experimental group (3,70 %, alternatively 3,41 %). The average thickness of muscular fibres in muscle triceps brachii /MTB/, muscle longissimus dorsi /MLD/ and muscle biceps femoris /MBF/ was significantly higher at control group ( $P < 0,05$ ). The average thickness of muscular fibres was dominant both groups. The lowest thickness of muscular fibres was in muscle triceps brachii.

**Key words:** pigs, striated muscles, histochemical analysis, selenium

## Introduction

Selenium /Se/ was found by Swedish chemist baron J.J.Berzelius in residues of sulphur acid in 1918. Different syndromes of deficiencies of Se in humans were described in seventies, for example Keshan's disease (*Van Vleet, 1980*).

There were determined syndromes also at animals, like white muscular dystrophy, liver necrosis, heart myopathy and equidative diathesis. There are diseases, which relate to deficient protection against oxidative stress (*Van Vleet, 1980*). *Rotuck et al.* (1973) found that the main role of Se is in antioxidant reactions. There were identified nearly 20 Se-proteins (*Jacques, 2001*). Selenium has important role in immune functions. The selenium deficiency decreases cellular and humoral immune efficiency in humans and animals. It relates to their higher contents in spleen, lymph-nodes and in liver and it relates also to very high GSH-Px activity in lymphocytes (*Rayman, 2000*). Selenium has also potential anticarcinogenic effect, mainly on the basis of statistical and model studies (*Rayman, 2000; Neve, 2000*).

Selenium is one of the most important mineral trace elements, its deficiency has marked effect to utility and health state of animals, and also to health state of humans. *Mahan (1996)* has studied the effect of organic selenium for many years. Incorporation of Se to muscle tissue depends on dietary level and on selenium form. Feeding of inorganic selenium in form of selenite causes only slight increase of selenium concentration in muscles. When we use organic form of selenium, the increase of selenium concentration in muscles is linear. This is important for increase of pork quality. The final product is good way to increase the selenium intake in human nutrition (*Muñoz et al., 1996; Poltársky et al., 1998*).

## Material and methods

The control group consisted of 10 hybrid pigs ( KS – HYB ). These pigs were fed standardized complete feed mixture 0 – 3 and OŠ – 6. The experimental group consisted of 10 butcher hybrid pigs ( SE – HYB). These pigs were fed by OŠ – 3 and OŠ – 6 with addition 0,3 mg.kg<sup>-1</sup> of organic selenium (per os) Sel – Plex.

The plant producing feed mixture ensured the production of standardized feed mixtures. The firm Tekro, s.r.o. Dvory nad Žitavou ensured the production of feed mixtures with Sel – Plex for individual groups. The samples were isolated by necrotic method. We carried out our experiments according to regulations of government of SR. These regulations determine the requirements regarding animal protection which are used in experiments (Collection of laws 289/2003, § 11 ods 1 and 3).

We isolated the samples from m. triceps brachii (MTB), m. longissimus dorsi (MLD) and m. biceps femoris (MBF). The samples were isolated at latest 30 minutes post mortem, and they were cut at size 1x1 cm, marked, wrapped to foil and frozen in liquid nitrogen.

The frozen samples were cut at temperature from -18 °C to -21 °C and the samples were liable to histological and histochemical reactions. The first set of samples was coloured by transparent colours haematoxylin – eosine and toluidine blue, the second set of samples to testing of neutral lipids by oil red „0“. The cuts were incubated to succinatdehydrogenase (SDH) for the determination of muscle fibres types according to method of *Stein and Padykula (1962)*. Three fibres types were identified on the basis of SDH reaction: eta Ret – red, Afa Ret-intermediate and Alfa White – white. We determined the muscle fibres thickness, fat cells thickness and area of different muscles fibres and interstitial tissues in percentage according to *Uhrín and Kulíšek (1989)* by optical microscope OLYMPUS - PROVIS.

We calculated the basic variational – statistical values by statistical program Statgraphics. The results between tested groups were tested by t-test.

## Results and Discussion

**Table 1. Basic traits of slaughtered results**  
**Tabela 1. Osnovne osobine klaničnih rezultata**

<b>Trait/Osobina</b>		<b>x</b>	<b>Min.</b>	<b>Max.</b>	<b>Sx</b>	<b>V %</b>	<b>demonstrativeness</b>
<b>Body weight/ Telesna masa</b>	Control group/ Kontrola	102,05	88,00	113,00	7,67	7,52	P > 0,05 -
	Experimental group/ Eksp.grupa	104,10	92,00	119,50	8,64	8,30	
<b>Part of meat/ Deo mesa</b>	Control group/ Kontrola	53,83	51,86	56,31	1,54	2,86	P < 0,05 +
	Experimental group/ Eksp.grupa	51,96	43,86	56,84	3,78	7,27	
<b>Thickness of neck/ Debljina vrata</b>	Control group/ Kontrola	2,26	1,80	3,30	0,43	19,02	- P > 0,05 -
	Experimental group/ Eksp.grupa	2,65	1,80	4,00	0,66	24,91	
<b>Thickness of spine part/ Debljina kičmenog dela</b>	Control group/ Kontrola	1,51	1,17	2,13	0,34	22,52	P > 0,05 -
	Experimental group/ Eksp.grupa	1,75	1,27	2,97	0,49	28,00	

x – mean, Sx – standard deviation, V % - coefficient of variation,

The average body weight of animal at slaughtering was 102,05 kg in control group and 104,10 kg in experimental group. These differences weren't statistical significant ( P > 0,05). The thickness of neck and spine part was

higher in experimental group compared to control group. The differences weren't statistical significant ( $P > 0,05$ ) (table 1). We determined average fat cells thickness in muscles in both groups within the frame of histological experiments. The average fat cell thickness was 36,35  $\mu\text{m}$  in control group and 39,17  $\mu\text{m}$  in experimental group. These differences were statistical significant ( $P < 0,05$ ). The thickness of fat cells in individual muscles were higher in experimental group. The difference was statistical significant in MTB ( $P < 0,05$ ). The differences weren't significant in MLD ( $P > 0,05$ ). The differences in fat cell thickness in MRF were higher in experimental group ( $P < 0,05$ ) (table 2).

Interstital tissue with fat tissue is very important in term of morphology – function and also in term of evaluation of final product – meat. We detected that values of percentage abundance of tissue in muscles were 3,7 % in experimental group and 3,41 % in control group ( $P > 0,05$ ).

The presence of  $\alpha$  White fibres in percentage was dominant in all muscles in both groups (control group 49,23 % and experimental group 52,34%) ( $P > 0,05$ ).

**Table 2. Thickness of fat cells in  $\mu\text{m}$**

**Tabela 2. Debljina čelija masti u  $\mu\text{m}$**

<b>EXPERIMENTAL GROUP/OGLEDNA GRUPA</b>					
<b>Trait/Osobina</b>	<b>x</b>	<b>Min.</b>	<b>Max.</b>	<b>Sx</b>	<b>V %</b>
<b>MTB</b>	39,70	26,00	54,00	7,38	18,60
<b>MLD</b>	41,10	24,00	56,00	8,02	19,52
<b>MRF</b>	36,70	24,00	50,00	7,01	19,09
<b>Total Ø</b>	39,17	24,00	56,00	1,84	4,69
<b>CONTROL GROUP/KONTROLNA GRUPA</b>					
<b>MTB</b>	34,85	20,00	54,00	9,28	26,64
<b>MLD</b>	38,50	20,00	54,00	9,23	23,97
<b>MRF</b>	35,70	24,00	52,00	7,12	19,95
<b>Total Ø</b>	36,35	20,00	54,00	1,56	4,29

The presence of  $\beta$  Red fibres was equal in three muscles in experimental group – average 27,14 %. The presence of  $\beta$  Red fibres was lower in control group - 22,83% ( $P < 0,05$ ). The presence of  $\beta$  Red fibres was lowest in MLD in control group (17,68%).  $\alpha$  Red fibres were dominant in control group - 24,53% in comparison to experimental group – 16,83%. The difference was significant. ( $P < 0,005$ ) (table 3).

**Table 3. Area of different muscles fibres and interstitial tissues in percentage**  
**Tabela 3. Površina različitih mišića i intersticijalnog tkiva u procentima**

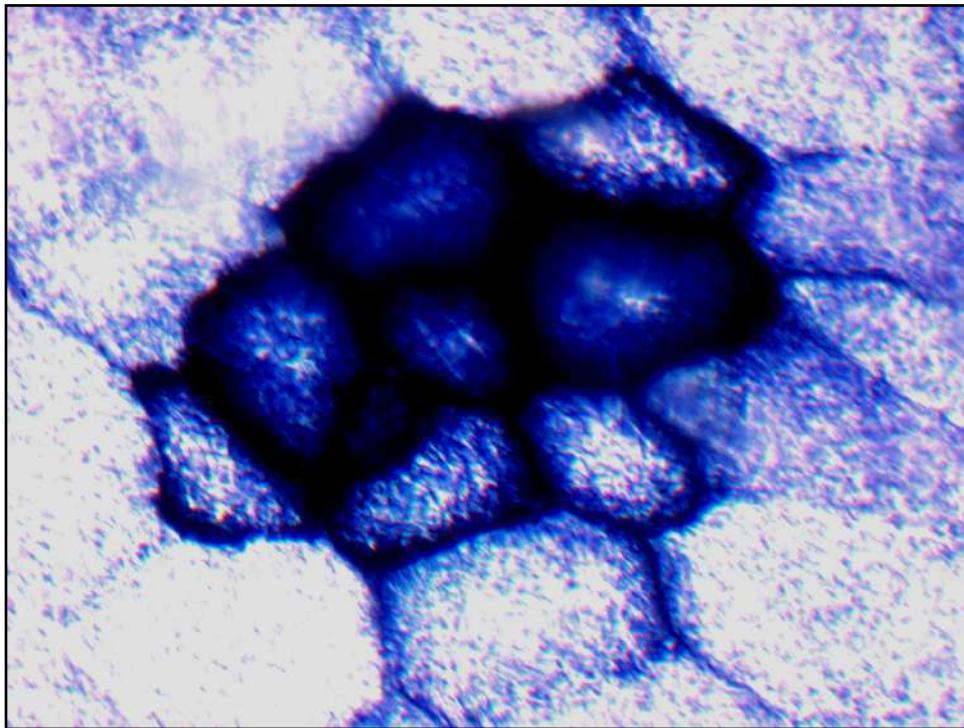
<b>EXPERIMENTAL GROUP/OGLEDNA GRUPA</b>				
<b>Trait</b>	<b>IV</b>	<b>β Red</b>	<b>α Red</b>	<b>α White</b>
<b>MTB</b>	3,61	28,73	13,26	54,40
<b>MLD</b>	3,58	25,06	15,44	55,92
<b>MRF</b>	3,90	27,63	21,78	46,69
<b>Total</b>	3,70	27,14	16,83	52,34
<b>Ø</b>				
<b>CONTROL GROUP/KONTROLNA GRUPA</b>				
<b>MTB</b>	2,92	26,12	15,07	55,89
<b>MLD</b>	3,51	17,68	27,01	51,80
<b>MRF</b>	3,80	24,68	31,52	40,00
<b>Total</b>	3,41	22,83	24,53	49,23
<b>Ø</b>				

The most important role has average thickness of muscle fibres in MTB, MLD and MRF at morphometrical evaluation of adversely stripped skeletal muscular tissue.

The average thickness of muscular fibres was 76,63 µm in experimental group and 79,65 µm in control group. This difference was statistical significant in favour of control group ( $P < 0,05$ ). The highest average thickness of muscular fibres was in MRF (experimental group 84,72 µm and control group 86,61 µm). The lowest average thickness of muscular fibres was in MTB in both groups. The average thickness of muscular fibres in MLD had median values (table 4).

**Table 4. Diameter of muscles fibres in investigated muscles in µm**  
**Table 4. Prečnik mišićnih tkiva u ispitivanim mišićima u µm**

<b>EXPERIMENTAL GROUP/ OGLEDNA GRUPA</b>					
<b>Trait</b>	<b>x</b>	<b>Min.</b>	<b>Max.</b>	<b>Sx</b>	<b>V %</b>
<b>MTB</b>	69,4	59,36	96,78	1,7	2,45
<b>MLD</b>	75,77	59,98	102,95	1,46	1,93
<b>MRF</b>	84,72	65,00	106,35	2,01	2,37
<b>Total Ø</b>	76,63	59,36	106,35	2,01	2,37
<b>CONTROL GROUP/ KONTROLNA GRUPA</b>					
<b>MTB</b>	73,30	62,15	102,00	2,74	3,74
<b>MLD</b>	79,03	59,36	103,25	3,05	3,86
<b>MRF</b>	86,61	63,36	120,95	6,04	6,96
<b>Total Ø</b>	79,65	59,36	120,95	3,94	4,85



**Picture 1. MRF (m. rectus femoris).  $\alpha$ R- $\alpha$ RED,  $\beta$ R- $\beta$ RED,  $\alpha$ W- $\alpha$ WHITE. SDH. enl. 200x**  
**Slika 1. MRF (m. rectus femoris).  $\alpha$ R- $\alpha$ RED,  $\beta$ R- $\beta$ RED,  $\alpha$ W- $\alpha$ WHITE. SDH. enl. 200x**

## Conclusion

We compared influence of selenium (Se) in experimental group with control group of pigs in our experiment. Differences in body weight weren't statistically significant in both groups. The thickness of neck and spine part was higher at control group. The thickness of fat cells was higher at experimental group in both muscles ( $39,17 \mu\text{m}$ , resp.  $36,35 \mu\text{m}$ ,  $P < 0,05$ ). The presence of interstitial tissue in percentage was statistical significant and it was higher at experimental group (3,70 %, resp. 3,41 %). The average thickness of muscular fibres in MTB, MLD and MRF was significant higher at control group ( $P < 0,05$ ). The average thickness of muscular fibres was dominant in both groups. The lowest thickness of muscular fibres was in MTB.

## Uticaj organskog selenia na histološku strukturu mišića i kvalitet svinjskog mesa

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### Rezime

Selen je jedan od najvažnijih mineralnih elemenata u tragovima, njegov nedostatak ima uticaj na zdravstveno stanje životinja i zdravstveno stanje ljudi. Inkorporacija Se u mišićno tkivo zavisi od nivoa selenia u obroku i njegove forme. Kada se koristi organski oblik selenia, povećanje koncentracije selenia u mišićima je linearno. Ovo je veoma važno za poboljšanje kvaliteta svinjskog mesa. Poredili smo uticaj selenia (Se) u oglednoj grupi sa kontrolnom grupom svinja u našem ogledu. Razlike u telesnoj masi nisu bile statistički signifikantne u obe grupe. Debljina vrata i kičmenog dela je bila veća u kontrolnoj grupi. Debljina ćelija masti je bila veća u oglednoj grupi kod oba mišića ( $39,17 \mu\text{m}$ , odnosno  $36,35 \mu\text{m}$ ,  $P < 0,05$ ). Prisustvo intersticijalnog tkiva u procentima je bilo statistički signifikantno i veće u oglednoj grupi (3,70 %, respektivno 3,41 %). Prosečna debljina mišićnih tkiva u mišiću triceps brachii /MTB/, mišiću longissimus dorsi /MLD/ i mišiću biceps femoris /MBF/ je bila signifikantno veća u kontrolnoj grupi ( $P < 0,05$ ). Prosečna debljina mišićnih tkiva je bila dominantna u obe grupe. Najmanja debljina mišićnih vlakana je bila u mišiću triceps brachii.

**Ključne reči:** svinje, mišići, histohemijska analiza, selen

### References

- BOBČEK, B., et al. (2004). Effects of dietary organic selenium supplementation on selenium content, antioxidative status of muscles and meat quality of pigs. Czech J. Anim. Sci., 49, 2004, p. 411 – 417.
- JACQUES, K. A. (2001): Selenium metabolism in animals. The relationship between dietary selenium from and physiological response. In: LYONS, T. P.: JACQUES, K. A (eds.), Science and technology in the feed industry – Proceedings of Alltech 's 17th annual symposium. Nottingham: University Press, 2001, p. 319 – 348.
- MAHAN, D. C., CLINE, T. R., RICHERT, B. (1999): Effect of dietary levels of selenium-enriched zeast and sodium selenite as selenium sources fed to growing-finishing pigs on performance, tissue selenium, serum glutathione

- peroxidase activity, car-cass characteristics and loin quality. In: *Anim. Sci.*, vol. 77, 1999, p. 2172 – 2179.
- MAHAN, D. C., KIM, Y. Y. (1996).: Effect of inorganic or organic selenium at two dietary levels on reproductive performance and tissue selenium concentrations in first parity gilts and their progeny. *J. Anim. Sci.*, 74, 1996, p. 2967 – 2974.
- MUÑOZ, A. M., GARINDO, D., GRANADOS, M. V. (1997): Effect of selenium yeast and vitamins C and E on pork meat exudation. In: *Biotechnology in the feed industry – the 14th annual symposium*, 1997, p. 1 – 29.
- NEVE, J. (2000): New indices for assessment of trace element status and requirement, with a special focus on selenium. In: Roussel, A. M., Favier, A., Anderson, R. A., eds. *Trace elements in man and animals 10: Proceedings of tenth international symposium on trace elements in man and animals*. New York: Pleunum Press, 2000, p. 317 – 322.
- RAYMAN, M. G. (2000).: The importance of selenium to human health. *Lancet*, 15 (356), 2000, p. 233 – 241.
- ROTRUCK, J. T., POPE, A. L., GANTHER, H. E., SWANSON, A. B., HAFEMAN, D. G., HOKSTRA, W. G. (1973): Selenium – biochemical role as a component of glutathione peroxidase. In: *Science*, vol. 179, 1973, p. 588 – 590.
- STEIN, J. M., PADYKULA, H. A. (1962). Histochemical classification of individual skeletal fibres of rat. In: Amer. In: *J. Anato.*, vol. 110, 1962, p. 103 – 123.
- VAN VLEET, J. F (1980): Current knowledge of selenium-vitamin E deficiency in domestic animals. In: *J. Amer. Veter. Med. Assoc.*, vol. 176, 1980, p. 321 – 325.
- UHRÍN, V., KULÍŠEK, V. (1980). (Výskumný ústav chovu a šľachtenia hydiny, Ivanka pri Dunaji): Využitie morfometrických metód pre stanovenie hrúbkby svalových vláken. *Živočíšna Výroba*, 25, 1980 p. 935 – 942.