

THE EFFECT OF GENOTYPE ON MUSCLE FIBRE CHARACTERISTICS OF *M. Longissimus lumborum* OF FATTENERS**

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Abstract: The aim of this study was to compare muscle histochemical composition in *m. longissimus lumborum* between different crossbreed fatteners. The research was carried out on 36 fatteners from three different crossbreed (12 animals in each) as follows: group I [♀Duroc x Hampshire♂], group II [♀Polish Landrace x (Duroc x Hampshire)♂] and group III [♀(Polish Landrace x Polish Large White) x (Duroc x Hampshire)♂].

For histochemical analysis of muscle fibre types the activity of dehydrogenase NADH₂ (diaphorase) was detected using specific histochemical testes. The results of the current histochemical investigations showed that genotypes of fatteners can influence on histochemical composition of the muscle fibre types - especially on percentage and size of muscle fibre. These changes can have some influence on meat consumption quality.

Key words: muscle fibres, histochemistry, *m. longissimus lumborum*, fatteners

Introduction and literature review

Mammalian skeletal muscle are primarily composed of three fibre types (type I – red fibres, type IIA - intermediate fibres and type IIB – white fibres), which differ in metabolic and contractile properties (*Brooke and Kaiser, 1970*). Differences between fibre types may also based on their metabolic characteristics: namely slow-twitch oxidative, fast twitch

oxidative glycolytic and fast-twitch glycolytic fibres (Peter *et al.*, 1972).

The fibre type composition of different skeletal muscles could be one of the most important factors influencing the biochemical events associated with their conversion to meat. Histological and histochemical investigations on pig muscles have revealed relationships between fibre traits and meat quality, such as pH, water-holding capacity, colour and meat tenderness (Essen-Gustavsson and Fjellkner-Modig, 1985; Karlsson *et al.*, 1993; Koch *et al.*, 1995; Ryu and Kim, 2005; Lefaucheur 2006).

The *m. longissimus lumborum* is the most frequently used indicator muscle in meat quality studies in pigs. This muscle contains a higher percentage of type IIB fibres, and has a low oxidative capacity. Muscle metabolism is the summation of the activities of the individual muscle fibres, which comprise the muscle. Some fibre characteristics are mostly determined genetically, while others can be affected by external factors, such as animal's age and sex (Larzul *et al.*, 1997; Wojtysiak *et al.*, 2004), its physical activity, nutrition or intensive selection (Brocks *et al.*, 1998; Klosowska and Fiedler 2003).

Few studies suggest that muscle fibre composition is on one hand affected by growth rate and, on the others, itself affected the carcass lean content. Moreover histochemical profile of muscle fibres is specific for different pig breeds or lines (Larzul *et al.*, 1997; Ruusunen and Puolanne 1997).

Therefore the aim of this study was to compare muscle histochemical profile in *m. longissimus lumborum* between different crossbreed fatteners.

Materials and methods

The research was carried out on 36 fatteners from three different crossbreed (12 animals in each) as follows: group I [♀Duroc x Hampshire♂], group II [♀Polish Landrace x (Duroc x Hampshire)♂] and group III [♀(Polish Landrace x Polish Large White) x (Duroc x Hampshire)♂]. Animals were feed *ad libitum* and slaughtered at 109-kg body weight at commercial slaughterhouses according to routine procedure.

Muscle samples were taken from the right carcass-side from the *m. longissimus lumborum* 20-min post mortem at the 5th lumbar vertebra. They were attached to a piece of cork and immediately frozen in isopentane cooled with liquid nitrogen and stored at -80°C until histochemical analyses were performed. For determination of muscle fibre type frequency and cross-section area, the frozen samples were sectioned at 10-µm thickness at -20°C

in cryostat (Slee MEV, Germany) and stained for dehydrogenase NADH₂ (diaphorase) activity according to the method of Dubovitz et al. (1973). The incubation medium contained nicotinamide adenine dinucleotide (NADH₂) (Sigma Chemical Co, St. Louis, USA) and nitro blue tetrazolium (NBT) (Sigma Chemical Co, St. Louis, USA). After final washes, sections were mounted in glycerine jelly.

The frequency and cross-section area (CSA) of fibre types were quantified with an image analysis system Multi Scan v.14.02. A minimum of 200 fibres was examined from each cross section. Additionally, the relative area (RA) occupied by each fibre type was calculated from the corresponding numerical percentages and mean cross-section area (CSA).

Data was examined by ANOVA and tested for differences by Tuckey test. A confidence level of $P < 0.05$ was chosen to indicate statistical significance.

Results and Discussion

Microstructure of *m. longissimus lumborum* from three different crossbreeds of fatteners is illustrated in photos 1-3. In all sections red fibres occurred in characteristic "nests" formed from 4 to 8 fibres with a great number of formazan granules occurring regularly over the whole area of the fibre. Intermediate fibres were found singly or in very small groups close to the red fibres. White fibres had only a small number of formazan granules.

The histometrical parameters of fibre types are presented in table 1. There were significant differences in both the percentage of fibre types, and their cross-sectional area (CSA), between the examined groups of fatteners. Muscles from group III had a higher percentage of white fibres and a lower percentage of red fibres, than muscles from group II and I. On the other hand, genotypes had no effect on fibre type relative areas (RA). Additionally, present study showed that white and intermediate fibres had the greatest cross-sectional area in the group III. On the contrary, the smallest one was observed in group I. Likewise, the greatest cross-sectional area of red fibres was found in group III compared with group I and II, between that no significant differences were observed.

Differences in fibre types composition observed in present study between three different crossbreeds of fatteners appear similar to those reported earlier by *Karlsson et al. (1993)* and *Larzul et al. (1997)*, in which authors have demonstrated that muscle fibre composition and size is specific for different pig breeds or lines. *Ruusunen and Puolanne (1997)* found that

the muscles of Hampshire are more oxidative than those of Landrace or Yorkshire pigs.

Pig muscles with many oxidative fibres tolerate stressful conditions better during transport, resulting in a slower fall in pH after slaughter. Muscle fibres affect postmortem changes in muscle due to the differences in their glycolytic or oxidative capacity. After slaughter, the pH value of light muscle decreases more rapidly and to a lower ultimate value than that of dark muscle because light muscle contains more glycogen and more glycolytic enzymes.

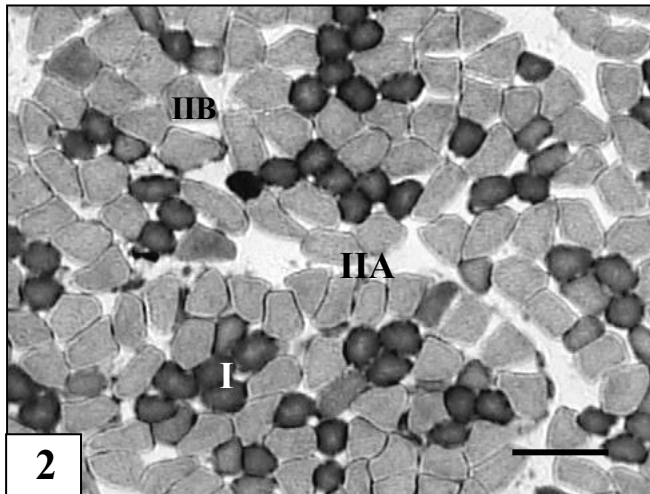
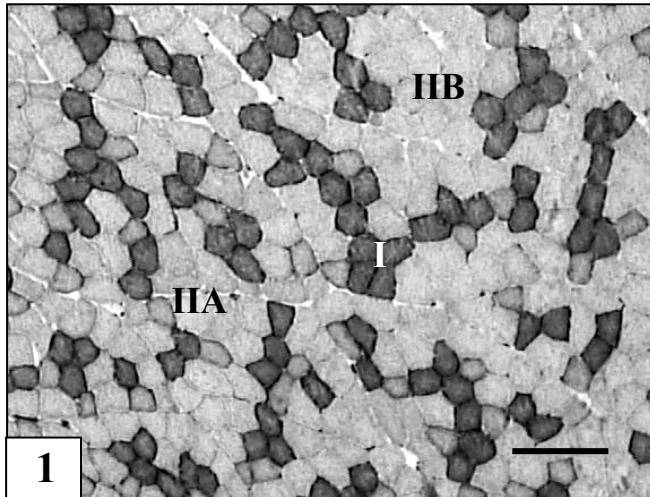
Table 1. Percentages, cross-sectional area (CSA) and relative areas (RA) of red (I), intermediate (IIA) and white (IIB) muscle fibres in *m. longissimus lumborum* of three examined genotypes group of fatteners.

traits	group I $\bar{x} \pm SE$	group II $\bar{x} \pm SE$	group III $\bar{x} \pm SE$
Percentages of muscle fibres [%]			
IIB	57,27 ± 1,20a	56,79 ± 2,19a	65,48 ± 1,34b
IIA	12,99 ± 0,98a	15,18 ± 1,41a	13,03 ± 0,68a
I	29,74 ± 0,76a	28,03 ± 1,25a	21,49 ± 0,86b
Cross-section area (CSA) [μm^2]			
IIB	5001,3 ± 100,52a	5984,71 ± 143,91b	6597,55 ± 122,29c
IIA	2915,84 ± 60,06a	3445,02 ± 92,69b	3894,22 ± 88,49c
I	3292,39 ± 72,97	3513,98 ± 93,90a	5713,69 ± 74,77b
Relative area [%]			
IIB	67,84 ± 1,16a	69,27 ± 1,23a	71,34 ± 1,39a
IIA	8,97 ± 0,39a	10,60 ± 0,81a	8,38 ± 0,57a
I	23,19 ± 0,76a	20,07 ± 0,98a	20,28 ± 0,84

a, b, c - means (in rows) with different superscripts differ significantly at $p < 0,05$.

According to *Essen-Gustavsson and Fjelkner-Modig (1985)* muscle oxidative capacity affects meat quality by improving the sensory properties of meat. Moreover, evidence from the literature (*Karlsson et al., 1993; Koch et al., 1995*) indicates that muscle characteristics and particularly fibre type frequency may be an important source of variation in eating quality. However, the influence of fibre type on meat tenderness is not fully understood. Studies in pigs (*Karlsson et al., 1993*) and cattle (*O'Halloran et al., 1997*) have shown that the frequency of fast glycolytic fibres (white fibres - type IIB) is negatively correlated with toughness. This was indirectly supported by a positive correlation between the proportion of slow oxidative

fibres (red fibres - type I) and toughness in study on bulls (*Renand et al., 2001*). Moreover, a positive relationship between the frequency or percentage area of red fibres and sensory tenderness and negative relationship between the frequency of white fibres and tenderness has been reported in cattle (*Maltin et al., 1998*). Other studies suggest that differences between the same muscles may only be evident in specific breeds of any one



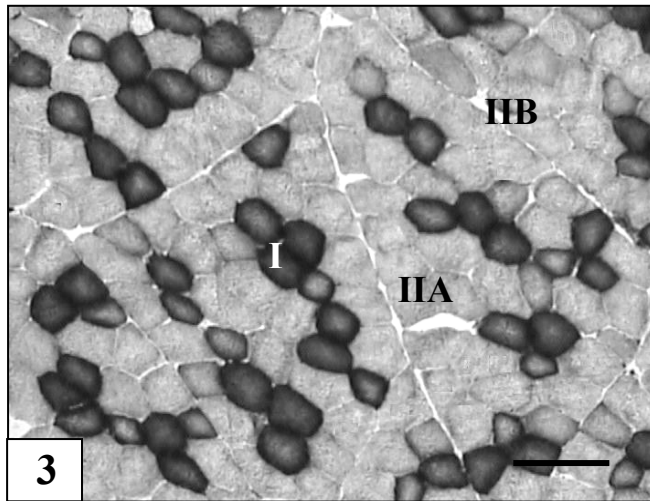


Fig. 1-3. Cross section of *m. longissimus lumborum* in fatteners – (1) group I [♀Duroc x Hampshire♂], (2) group II [♀Polish Landrace x (Duroc x Hampshire)♂] and (3) group III [♀(Polish Landrace x Polish Large White) x (Duroc x Hampshire)♂]. Diaphorase reaction activity: I-red fibres, IIA- intermediate fibres, IIB- white fibres. Scale bar 200µm.

species. It has been shown that the shear force of *longissimus* muscle differ significantly in Duroc, but not in Berkshire and Large White pigs (Chang *et al.*, 2003).

Additionally, variation in fibre type composition may effect on colour of meat. Meat colour is the major factor limiting the quality and acceptability of meat and meat products. The right colour of meat can be conditioned by the ferrous oxymyoglobin (oxyMb) - Philips *et al.* (2001), which is directly connected with the percentage and size of the muscle fibre types (Warriss *et al.*, 1990).

Conclusion

The obtained results indicated that:

- Muscles from group III had a higher percentage of white fibres and a lower percentage of red fibres, than muscles from group II and I.
- Genotypes had no effect on fibre type relative areas (RA).

- White and intermediate fibres had the greatest cross-sectional area in the group III, in contrast, the smallest one was observed in group I.
- The greatest cross-sectional area of red fibres was found in group III compared with group I and II, between that no significant differences were observed.

UTICAJ GENOTIPA NA KARAKTERISTIKE MIŠIĆNOG VLAKNA *M. longissimus lumborum* TOVLJENIKA

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Rezime

Cilj ovog istraživanja je bio poređenje histohemijskog sadržaja mišića *m. longissimus lumborum* kod tovljenika meleza različitih rasa. Istraživanje je izvedeno na 36 tovljenika tri različita meleza (12 životinja u svakoj grupi): grupa I [♀durok x hempšir♂], grupa II [♀poljski landras x (durok x hempšir♂)] i grupa III [♀(poljski landras x poljska bela velika) x (durok x hempšir♂)]. U roku od 20 minutak post mortem uzorci mišića su uzeti za histohemijsku analizu sa *m. longissimus lumborum* na desnoj polutki. Radi razlikovanja tipova mišićnih vlakana: tip I (crvena), tip IIA (srednja), i tip IIB (bela) delovi su bojeni za aktivnost dehidrogenaze NADH₂ (diaforaza). Ocenjivan je procenat mišićnih vlakana, oblast poprečnog preseka (CSA) i relativna ovlast (RA). Dobijeni rezultati ukazuju da je genotip tovljenika imao uticaj na sastav mišićnog vlakna – posebno na procenat i veličinu mišićnog vlakna. Najveći poprečni presek belih i srednjih vlakana je utvrđen kod tovljenika grupe III, dok je u grupi I utvrđena najmanja oblast poprečnog preseka. Osim toga, veća oblast poprečnog preseka crvenih vlakana je utvrđena kod grupe III u poređenju sa grupom I i II, između kojih nisu utvrđene signifikantne razlike. Takođe, *m. longissimus lumborum* kod tovljenika iz grupe III su imali viši procenat belih tkiva i niži procenat crvenih tkiva, nego mišići tovljenika iz grupa II i I. S druge strane, genotipovi tovljenika nisu uticali na relativne oblasti tipova vlakana (RA). Razlike u karakteristikama mišićnog tkiva između ispitivanih meleza tovljenika su imale uticaj na kvalitet mesa sa aspekta konzumacije.

Ključne reči: mišićna tkiva, histohemija, *m. longissimus lumborum*, tovljenici

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