Histological characteristics in *Longissimus thoracis* muscle of Italian and Polish crossbreed horse

Annarita Nicastro¹, Aldo Corriero¹, Francesco Nicastro¹, Davide De Marzo²

¹Dipartimento dell'Emergenza e dei Trapianti di Organi, sezione di Cliniche Veterinarie e Produzioni Animali, Università degli Studi di Bari "Aldo Moro", Bari, Italia - E-mail: francesco.nicastro1@uniba.it; ²Dipartimento di Scienze Agro Ambientali e Territoriali, Bari, Italia

Summary. Owing to high cost, meat horse domestic production does not meet internal market demand, that is why such a livestock is mainly imported from Eastern Europe Countries. Fibre type composition deeply influences post-mortem changes in the conversion of muscle to meat, as well as the product quality. The aim of this study was to investigate histological characteristics of muscle fibres and adypocites related to two different horse genetic types (Abruzzese and Polish), sold in Italy. The two horse genetic types showed little variation in the fiber type of the meat, considering the least square means of all 3 fiber types (red, intermediate and white) found in the samples muscle. The alpha-white fibers were larger than beta-red and alpha-red (P<0.05). Significant differences in distribution were found for most fibre types within breeds. A higher proportion of slow fibers was found in the *Longissimus* muscle of the TPR x Abruzzese (P<0.05). The histological results showed that the two genetic types had small effect on the size of the LD fibers.

Key words: horses, fiber, fat, morphometric

Introduction

Locally produced horse meat in Italy does not meet internal market demand due to for its high costs. Hence, slaughter horses are mainly imported from Eastern Europe Countries. Horse meat consumption is very high in some Italian regions such as Apulia and in the last few years has considerably increased. The fiber type composition deeply influences the post-mortem changes in the conversion of muscle to meat, as well as the product quality. Many recent studies found correlation between fiber type or size, and eating quality. However, the quality characteristics of individual muscles and the factors responsible for these quality traits are not well known (1). Skeletal muscle is composed of different fiber types, which are affected by various factors, such as sex, age, muscle type, breed and hormones (2). Muscle fiber characteristics influence meat quality characteristics such as color, water-holding capacity, marbling and the texture of meat (3). In this regard, it is important to understand the relationship between muscle fiber characteristics and meat quality, and it is also necessary to characterize the muscle fiber in individual horse muscles. Generally, meat quality is determined by the marbling, color, firmness and texture. These quality traits are influenced by muscle fiber characteristics. However, the effects of muscle fiber characteristics on meat quality characteristics have not been studied extensively in horses, although many reports exist in cattle (4, 5), pig (6, 7), poultry (8) and lambs (9). Therefore, the aim of this study was to investigate the relationship between muscle fiber on the Italian and Polish crossbreed horse meat.

Methods

Ten horse for each genetic type (TPR x Abruzzese and TPR x Polish) were slaughtered at the age between 24 and 26 months. After slaughtering the

carcasses were chilled at 3°C for 24-36 h. Samples of Longissimus dorsi (LD) muscle were collected from all animals 4h after slaughter for the histochemical characterization of the muscular fibers. Some pieces of said muscle have been taken, and then immersed in liquid nitrogen for about 10 seconds. Cross section were cut and mounted on spindles before sectioning 15 mm thick using a Reichert-Jung freezing microtome. Serial sections mounted on glass microscope slides were stained with NADH-Tr, myofibrillar ATPase reacted at alkaline pH to differentiate muscle fibre type according to their oxidative and glycolitic capability (10), and hematoxylin according to the procedure outlined by Lillie (12) in order to stain fat cells in the intercellular space. Fibers were classified on the basis of stain reactions using the technique of Ashmore and Doerr (11): beta-red fibers were dark brown, alfa-red fibers were clear in the middle and surrounded by a blue ring, alfa-white fibers were clear. Sections were analysed using an Image Analyzer Vidas by Zeiss to determine fiber diameter. The percentage of each fiber type and its mean surface area was measured in two randomly selected areas on serial sections with an image analysis software program. On average, 200 fibers were analysed in each serial section. All data collected were subjected to analysis of variance and differences between the estimated average, evaluated with the "t" Student (13).

Results and Discussion

The muscle fibers in the LD muscle of the TPR x Abruzzese and TPR x Polish horses were divided into type beta-red, alpha-red and alpha-white (Fig. 1 and 2).

Least squares means for longissimus fiber type diameter and percentage are presented in figures 4 and 5. A clear difference in fiber type composition was observed among the Abruzzese and Polish breeds horse muscle. The LD muscle showed higher diameter (μ m) for all three fiber types in Abruzzese horses (beta-Red 8,90 vs 6,28 P < 0.01; Alfa-Red 9,54 vs 8,14 P < 0.05; and alfa-White 9,57 vs 9,00). The alfa-White fibers are the largest of the three types. These results were in agreement with Cassens and Cooper (14), and Rosser (15) in cattle. Fiber type proportions for by each fiber type in Ld muscle, are summarized in Figure 5. On the average, Ld



Figure 1. Section of LD muscle fibre type stained with ATP



Figure 2. Section of LD muscle fibre type stained with NADH



Figure 3. Section of LD intramuscular fat stained with Hematoxylin



Figure 4. Diameter of muscle fibers in the LD muscle from horse of two genetic types (μm)



Figure 5. Distribution of muscle fibers in the LD muscle from horse of two genetic types (%)

muscle contained 25% Beta-Red type, 47% Alpha-Red type and 28% alpha-White fibers respectively in Polish horses. Fiber type proportion between Abruzzese and Polish horses was similar except for alpha-White (32% vs 28% P < 0.05) with higher percent in the Abruzzese crossbreed. The diameter of the adipocytes was larger in Abruzzese horses (P<0.05), while the amount and the size of adypocites in the Polish horse proved to be lower and smaller than the other genetic type. These results were in agreement with Henckel et al. (16) who reported that the intracellular fat content was positively correlated with the frequency of alpha-white fibres.

Conclusions

The results about the histological characteristics of the two genetic types evidenced little effect on the size of fiber type, while the amount and the size of adypocites proved to be lower in the LD muscle of the TPR x Polish horse meat.

References

- Ashmore, C. R., Tompkins, G., & Doerr, D. (1972). Postnatal development of muscle fiber types in domestic animals. Journal of Animal Science, 34, 37-41.
- Karlsson, A. H., Klont, R. E., & Fernandez, X. (1999). Skeletal muscle fibres as factors for pork quality. Livestock Production Science, 60, 255–269.
- Totland, G. K., Kryvi, H., & Slinde, E. (1988). Composition of muscle fiber types and connective tissue in bovine M. semitendinosus and its relation to tenderness. Meat Science, 23, 303–315.
- 4. Ozawa, S., Mitsuhashi, T., Mitsumoto, M., Matsumoto, S., Itoh, N., & Itagaki, K. (2000). The characteristics of muscle fiber types of longissimus thoracis muscle and their influences on the quantity and quality of meat from Japanese Black steers. Meat Science, 54, 65–70.
- Nicastro F., A. Pagone, F. Pinto, Y.L. Xiong, R. Gallo L. Zezza (2003). Effect of diet enriched with omega –3 on beef meat quality: Histological characteristics. Internat. Congress of Meat Science and Technology. Campinas, Brasile, August, 31 – September 5.
- Rehfeldt, C., Fiedler, I., & Stickland, N. C. (2004). Number and size of muscle fibres in relation to meat production. In M. F. W. te Pes, M. E. Everts, & H. P. Haagsman (Eds.), Muscle Development of Livestock Animals (pp. 1–38). Wallingford, UK: CABI Publishing.
- Ryu, Y. C., & Kim, B. C. (2005). The relationship between muscle fiber characteristics, postmortem metabolic rate, and meat quality of pig longissimus dorsi muscle. Meat Science, 71, 351–357.
- Kim, G. D., Jeong, J. Y., Moon, S. H., Hwang, Y. H., Park, G. B., & Joo, S. T. (2008). Effects of muscle fibre type on meat characteristics of chicken and duck breast muscle. Proceedings of 54th International Congress of Meat Science and Technology (pp. 124). South Africa: Capetown.
- Nicastro, F., R. Gallo, L. Zezza (2007). Organic lamb of the Apulian Region. 2.Bio-histochemistry characteristics of longissimus dorsi muscle. 53st International Congress if Meat Science and Technology August 5-10, 2007 – Beijing – China
- Nicastro F. (1989). An improved procedure for identifying fibre types in ovine muscles. Meat Science, 24: 73-78
- Ashmore C.R., Doerr L. (1971) Comparative aspects of muscles fiber types in different species. Experimental Neurology, 31: 408-418
- 12. Lillie R.D. Histopathologic technique and pratical histochemistry. (1965), McGraw-Hill Book Co, New York.
- 13. Sas (2002). SAS/STAT Software for PC. Cary, NC: SAS Institute Inc.
- 14. Cassens R. G., Cooper C. C. (1971). Red and White mus-

cle. Advances in Food Research 19, 1 – 74.

- Rosser B.W.C., Norris B. J., Nemeth P. M. (1992). Metabolic capacity of individual fibres from different anatomic locations. Journal of Histochemistry and Cytochemistry 40 (6), 819 – 825.
- 16. Henckel, P., Oksbjerg, N., Erlandsen, E., Barton-Gade, P. and Bejerholm, C. (1997). Histo and biochemical characteristics of the Longissimus dorsi muscle in pigs and their relationships to performance and meat quality. Meat Science. 1997, 47: 311-321.

Correspondence: Nicastro Francesco Via G. Amendola 165/A, Bari, Italia, fax: 0805442837 tel: 0805442837 E-mail: francesco.nicastro1@uniba.it