Three pork muscles exhibited different *in vitro* protein digestibility

Xiaoyu Zou, Guanghong Zhou, Li Li, Chong Wang, Chunbao Li*

Key Laboratory of Meat Processing and Quality Control, MOE; Key Laboratory of Meat Processing, MOA; Jiangsu Synergetic Innovation Center of Meat Processing and Quality Control; Nanjing Agricultural University; Nanjing, Jiangsu 210095, P. R. China

*Corresponding author email: chunbao.li@njau.edu.cn

Abstract –Porcine trapezius, longissimus dorsi and biceps femoris muscles have been shown to contain different percentages of oxidative type IIA and glycolytic type IIB muscle fibers. Little is known about the effect of fiber type on protein digestion. In this study, we compared the in vitro digestion attributes of trapezius, longissimus dorsi and biceps femoris muscles. These muscles were in vitro digested by pepsin alone or followed by trypsin. The in vitro digestibility and particle size were measured. The results indicated that longissimus dorsi muscle had the highest digestibility under both pepsin and trypsin treatments (P<0.05). In non-digested samples, biceps femoris muscle showed the greatest particle size (P<0.05), but enzymatic digestion decreased particles to the similar size for all the three muscles (P>0.05). Fiber types could be the key factor to cause the differences in protein composition and their susceptibility to digestion.

Key words –Pork, In vitro digestion, Particle size, fiber type.

I. INTRODUCTION

Meat is an important source of high quality protein, vitamins, and minerals [1]. Previous studies indicated that pork and veal cuts had different protein content and amino acid profile in cooked meat [2]. In addition, different cuts are composed of different types of muscle fibers and different amounts of connective tissues, in terms of metabolic enzymes and collagen, which may cause different eating quality, especially of tenderness [3]. In practice, different cuts could be recommended to cook in different ways, for example, striploins and tenderloins are more suitable for roasting, while cuts from legs are suitable for mincing and long-term cooking [4]. For the same cut, minced beef was found to be more rapidly digested and absorbed than beef steak in older men [5]. This could be attributed to the cooking temperature effect [6]. However, it is still less known about the effect of meat cuts on protein digestibility at the same cooking conditions. In meat, there are more than 1000 proteins that can be classified into three types, i.e., myofibrillar proteins, sarcoplasmic proteins and stromal proteins [7]. However, little is known about the digestibility and the digestion products of proteins from different cuts. In this context, the objective of this study to compare the in vitro digestion attributes of pork proteins from trapezius, longissimus dorsi and biceps femoris muscles by measuring in vitro digestibility and particle sizes.

II. MATERIALS AND METHODS

Sampling and cooking procedure

Three pork cuts, including neck (m. trapezius), loin (m. longissimus dorsi) and outside (m. biceps femoris) were obtained at 24 h post-mortem from 8 Huai black pigs (a native pig breed) with ultimate pH values of 5.54±0.07, 5.44±0.05 and 5.43±0.05 respectively. All visible fat and epimysial connective tissue were removed. Pork muscles were cut vertically into 2cm-thick pieces (weights: 50 to 65 g each). All samples were packed in plastic pouches and cooked in a 72℃ water bath (Crystal Industries, USA) for about 30 min. The center temperature was tracked by a thermal probe (Pt 100, Testo AG, Germany). When the center temperature reached 70℃, meat samples were taken out and chilled in air to room temperature.

In vitro digestion

Cooked meat samples were in vitro digested according to Wen et al. [6] with some modification.

Measurement of particle sizes

The homogenization and in vitro digestion of cooked meat samples were performed as described above. Particle sizes of homogenates were measured before and after digestion by an integrated-laser light scattering instrument (Mastersizer 3000, Malvern, Worcestershire, UK). Five parameters were achieved, including: (1) D[4,3], which represents the mean diameter in volume; (2) D[3,2], which shows the mean diameter in surface; (3) Dx (50), which shows the average size of 50% of the sample particles that have a lower size; (4) Dx (10), which represents the average size of 10% of the sample particles that have a lower size; and (5) Dx (90), which represents the average size of 90% of the sample particles that have a lower size.

Statistical analysis

One-way analysis of variance and Duncan's multiple-range test were performed to test the differences in digestibility and particle sizes with the SAS program (SAS Institute Inc, USA).

III. RESULTS AND DISCUSSION

As shown in Figure 1, significant differences in protein digestibility were observed among three cuts in both pepsin and trypsin digestion conditions ($P<0.05$). After pepsin digestion, the neck (m. trapezius) showed much lower digestibility than the loin (m. longissimus dorsi) and the outside (m. biceps femoris) ($P<0.05$, 62.27%, 79.19% and 76.65%, respectively). The latter two cuts did not show significant difference in digestibility ($P>0.05$). After two-step (pepsin and trypsin) digestion, the digestibility values of the three cuts increased greatly ($P<0.05$). The loin samples had the greatest values of the digestibility ($P<0.05$, 92.22%), and the neck and the outside samples did not differ ($P>0.05$, 85.23% vs. 88.07%). The differences in digestibility could be attributed to different protein composition. According to Ruusunen and Puolanne (2004), the light muscles have higher area percentages of glycolytic type IIB fibers and the dark muscles contained higher area percentages of type I fibers and oxidative type IIA [8].

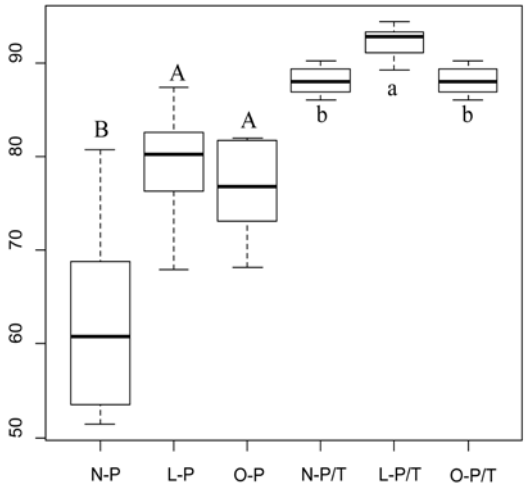

Figure 1 in vitro digestibility of three cuts after pepsin and trypsin digestion

N, L and O, i.e., the neck, the loin and the outside;P and T ,i.e., pepsin and trypsin;

A, B, averages of pepsin-treated digestibility differ significantly with different uppercases ($P<0.05$);

a,b averages of pepsin-and-trypsin-treated digestibility differ significantly with different lowercases ($P<0.05$).

Particle size reflects the extent of myofibrillar fragmentation and has a relationship to eating quality (e.g. tenderness) [9]. It may also reflect the extent of breakdown of meat during chewing and subsequent digestibility. In this study,

homogenization resulted in the greatest values of particle size, i.e., Dx(50), D[3,2], D[4,3], Dx(10) and Dx(90) for m. biceps femoris ($P<0.05$, Table 1), but the values were not significant between m. trapezius and m. longissimus dorsi ($P>0.05$). However, there was no significant difference in any variables for particle size between any two cuts after pepsin digestion ($P>0.05$, Table 1). After two-step digestion, only the D [4,3] values of m. trapezius were smaller than those of the other two muscles ($P<0.05$). This indicates that enzymatic digestion may overwhelm the muscle-specific difference in digestibility.

It is notable that Particle sizes of muscles decreased greatly from mouth to stomach. This could be ascribed to pepsin digestion, in which native protein molecules were broken down into small peptides and free amino acids. The values of D[3,2], D[4,3], Dx(10), Dx(50) and Dx(90) were further decreased from stomach to intestine just because of the degradation of protein or peptides into smaller ones under trypsin digestion.

Table 1 Particle sizes of muscle homogenates before and after digestions (means ± standard deviations)

	D [3,2]	D [4,3]	Dx (10)	Dx (50)	Dx (90)
			Before digestion		
N	41.19±8.27b	195.63±32.77b	17.45±6.46b	164.25±39.01b	420.75±54.73b
L	39.59±4.77b	219.38±31.78b	15.39±3.39b	187.50±31.61b	469.00±64.46b
O	71.85±23.79a	344.88±115.06a	46.01±24.10a	305.63±106.42a	698.00±221.09a
			After pepsin digestion		
N-P	12.89±2.45a	58.92±17.84a	5.12±0.83a	20.42±7.47a	132.22±28.92a
L-P	14.49±2.39a	45.36±15.74a	6.29±1.26a	19.02±5.32a	103.22±48.32a
O-P	14.09±1.53a	48.26±19.10a	6.00±1.45a	23.91±6.37a	151.2±79.91a
			After pepsin and trypsin digestion		
N-P/T	7.64±2.76a	13.26±3.81b	3.84±1.07a	7.25±0.81a	33.03±12.80a
L-P/T	7.06±1.12a	20.70±7.58a	3.66±0.59a	7.77±1.53a	44.75±12.66a
O-P/T	7.77±2.07a	19.03±4.74a	3.97±0.82a	7.95±1.95a	35.27±9.03a

N, L and O, i.e., the neck, the loin and the outside; P and T ,i.e., pepsin and trypsin;
a,b Means with different superscripts differ significantly ($P<0.05$).

IV. CONCLUSION

In this study, we compared in vitro protein digestibility and digestion products among three pork cuts. Longissimus dorsi showed the highest degree of protein digestion, moderate particle size and protein intensities after pepsin and trysin treatments.

ACKNOWLEDGEMENTS

This work was funded by 31530054 (NSFC), CARS-36-11 (MOA), CX(15)1006, and KYCYL201502. We thank Dr. Chen Dai for his technical assistance.

REFERENCES

1. P.M. de Castro Cardoso Pereira and A.F. dos Reis Baltazar Vicente. Meat nutritional composition and nutritive role in the human diet. Meat Science 93 (2013) 586-592.
2. H. Greenfield, J. Arcot, J.A. Barnes, J. Cunningham, P. Adorno, T. Stobaus, R.K. Tume, S.L. Beilken, W.J. Muller. Nutrient composition of Australian retail pork cuts 2005/2006. Food Chemistry 117 (2009) 721–730.
3. C.B. Li, G.H. Zhou and X.L. Xu. Comparisons of meat quality characteristics and intramuscular connective tissue between beef longissimus dorsi and semitendinosus muscles. Journal of Muscle Foods, 2007, 18, 143-161.
4. Industry-wide Cooperative Meat Identification Standards Committee. Uniform Retail Meat Identity Standards Manual. Available: http://beefretail.org/urmis.aspx.
5. B. Pennings, B.B.L. Groen, J.W. van Dijk, A. de Lange, A. Kiskini, M. Kuklinski, J.M.G. Senden, L.J.C. van Loon. Minced beef is more rapidly digested and absorbed compared with beef steak, resulting in greater postprandial protein retention in older men. American Journal of Clinical Nutrition, 2013,98: 121-128.
6. Wen, S. Y., Zhou, G. H., Li, L., Xu, X. L., Yu, X. B., Bai, Y., & Li, C. B. (2015). Effect of Cooking on in vitro digestion of pork proteins: A peptidomic perspective. Journal of Agriculture and Food Chemistry, 63, 250-261.
7. R.A. Lawrie, Chemical and biochemical constitution of muscle, pp75-127. In: Lawrie's Meat Science (Seventh Edition). Editors: R.A. Lawrie and D.A. Ledward. Woodhead Publishing Limited. 2006, ISBN: 978-1-84569-159-2.
8. M. Ruusunen, E. Puolanne. Histochemical properties of fiber types in muscles of wild and domestic pigs and the effect of growth rate on muscle fiber properties. Meat Science, 2004, 67: 533–539.
9. L.U. Karumendu, R. van de Ven, M.J. Kerr, M. Lanza, D.L. Hopkins. Particle size analysis of lamb meat: Effect of homogenization speed, comparison with myofibrillar fragmentation index and its relationship with shear force. 2009, 82: 425–431.