INFLUENCE OF GENOTYPE ON LAMB MEAT QUALITY. 2. INTRAMUSCULAR COLLAGEN PROPERTIES

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Background

Meat tenderness, the most important quality attribute for consumers (Love, 1994), originates in structural and biochemical properties of connective tissue and myofibrillar components of muscle. Role of myofibrillar proteins is considered mainly important during the *post mortem* meat tenderization processes (Koohmaraie, 1996). Connective tissue instead, being relatively stable through meat ageing (Nishimura et al., 1998; Geay et al., 2001), is determinant for the so-called background toughness of meat (Kuypers and Kurth, 1995), mostly conditioned by live-animal factors. Intramuscular collagen (IMC), which is the major protein constituent of muscle connective tissue, is stabilized through the synthesis of multivalent crosslinking molecules. Variation in collagen crosslinking leads to variation in the thermal stability of collagen, which has been correlated with changes in eating quality of meat (Bosselmann et al., 1995; McCormick, 1999). Crosslinking patterns in muscle are generally monitored by quantifying concentration of hydroxylysylpyridinoline (HLP), which is the primary, mature and heat-stable crosslink in this tissue (McCormick, 1999). Literature reports a relatively wide range of values for intramuscular collagen amount and crosslinks, varying with a host of conditions related to animal species, breed, sex, age, growth rate and nutrition (McCormick, 1994; 1999; Filetti et al., 2001). However, dated and poorly documented are the informations on sheep. Secondly, a large proportion of the work was performed on *longissimus dorsi* muscle, which is known to be low in connective tissue (McCormick, 1999) and relatively poor predictor of toughness of other muscles in the carcass (Shackelford et al., 1995).

Objective

This study was performed to evaluate IMC amount and maturation in four muscles (different for localization, structure, and/or physiological function) from lambs of Gentile di Puglia (GP) and Merinizzata Italiana (MI) breeds, mainly raised in the center and in the south of Italy for lamb meat production, and of Merinizzata Italiana × Pagliarola (MI×P) cross.

Methods

Thirty-three single male lambs (11 GP, 11 MI, and 11 MI×P) were slaughtered, according to the procedure described by the Italian Scientific Association of Animal Production (A.S.P.A., 1991), at an equal live weight (21.6±0.3 kg) representing an Italian traditional commercial weight. After weaning, lambs were reared under similar conditions and reached the target slaughter weight at a mean age of 93, 71, and 87 d for GP, MI, and MI×P lambs, respectively. *Longissimus dorsi* (LD), *gluteo biceps* (GB), *semimembranosus* (SM), and *semitendinosus* (ST) muscle samples were collected from chilled (2-4°C for 24 h) carcasses, vacuum-packaged and immediately frozen (-40°C) until they were analyzed. Samples were trimmed of fat and epimysium, lyophilized, and then hydrolyzed in 6 N HCl to determine hydroxyproline (Woessner, 1961) and HLP crosslink amounts. IMC concentration was calculated assuming that collagen weighed 7.25 times the measured hydroxyproline weight (Eastoe and Leach, 1958). HLP crosslinks were determined using a modification (Maiorano et al., 1999) of the HPLC procedure developed by Eyre et al. (1984). ANOVA was performed with General Linear Model procedure of SPSS (2000), including the animal effect, with a factorial model where genotype and muscle were the main factors. Differences among unadjusted means were tested by Scheffé's test. Pearson correlation coefficients between the IMC properties and between these and the lamb weight gain were estimated with the same statistical package.

Results and discussion

Table 1 shows the overall effects of genotype and muscle on IMC properties. Genotype clearly influenced ($P = 0.0001$) all studied variables: muscles of GP lambs had the lowest ($P < 0.001$) IMC amount, as well as the highest ($P < 0.001$) HLP concentration and HLP/IMC ratio. No differences ($P > 0.05$) were found between MI and MI×P lambs. Under similar rearing and feeding conditions, as in this study, differences in collagen amount and maturation mainly depend by animal-related factors: genotype, age, and growth rate. Our findings are consistent with previous literature, which reports a marked genetic influence on collagen content and thermal stability in lamb (Heinze et al., 1986), beef (Boccard et al., 1979; Campo et al., 2000) and pig (Lebret et al., 2001). Originally, the maturity of IMC has been evaluated by measuring collagen heat-solubility, whereas progressively more sophisticated technologies, such as the quantification of HLP heat-stable crosslinks, were later employed (Harper et al., 1999; McCormick, 1999). Also animal age plays a significant and complex role in muscle metabolism and structure as well as the toughness of meat. Age variation in meat toughness is not considered to be due to modifications in myofibrillar component but strictly related to changes in collagen amount and maturation (McCormick, 1994; Harper, 1999). Although there were only slight differences of age among lambs in our study, MI and MI×P lambs grew faster than GP lambs (250±7 and 227±6 vs 198±9 g/d, respectively; $P < 0.05$) and reached target slaughter weight earlier than GP lambs. Growth rate-dependent shifts in muscle collagen amount and/or crosslinking have been reported (McCormick, 1994; Bosselmann et al., 1995; Harper, 1999; Maiorano et al., 2000a, b). In fact, slaughtering animals after a period of rapid growth is generally thought to produce meat with collagen characteristics conducive to tenderness (McCormick, 1994). Newly synthesized collagen indeed adds to the existing collagen in such a way that the overall toughness properties of connective tissue are lowered (Harper, 1999). This mechanism was first proposed by Etherington (1987) who concluded that, during rapid growth, newly synthesized collagen dilute the older and is less crosslinked than the pre-existing collagen. The Etherington's hypothesis is supported by results of simple correlation analysis showed in Table 2. The IMC amount showed a positive correlation with average daily weight gain of lambs ($r = 0.314$; $P < 0.01$), whereas HLP muscle concentration and HLP/IMC ratio inversely related to weight gain ($r = -0.342$ and -0.508, respectively; $P < 0.001$). Moreover, muscle content of IMC significantly correlated to that of HLP crosslinks ($r = 0.302$; $P < 0.01$) as well as to HLP/IMC ratio ($r = -0.203$; $P < 0.05$). A marked muscle effect also influenced ($P = 0.0001$) IMC properties. The LD muscle had lower amounts of collagen and HLP crosslinks ($P < 0.01$ and $P < 0.001$, respectively) than the SM, GB and ST muscles. Moreover, IMC was less mature in the dorsal muscle (as indicated by the lower HLP/IMC ratio, $P < 0.01$) than in the other ones. Among muscles of pelvic limb, GB had a higher ($P < 0.01$) IMC concentration than ST, with the SM being intermediate, whereas no differences ($P > 0.05$) were found in HLP muscle content. The ratio of HLP to collagen was higher ($P < 0.01$) in ST than in SM muscle, with GB showing the intermediate value. These findings closely agree with the largest part of literature, that indicates real differences in both the concentration and thermal stability of IMC among different skeletal muscles of rat (Palokangas et al., 1992), sheep (Heinze et al., 1986; Maiorano et al., 2000a, b), goat (Maiorano et al., 2001), and bovine (McCormick, 1999). Above-mentioned differences in muscle collagen amount and, above all, in the extent of IMC crosslinking are considered to be closely related to the physiological function of muscle. Generally, in fact, locomotor muscles possess more crosslinks than postural muscles (Palokangas et al., 1992; Zimmerman et al., 1993; Bosselmann et al., 1995). Our results are consistent with the conclusions of Kuypers and Kurth (1995) and Harper (1999) that variation in IMC properties with muscle type and function lead to the well-known differences in background toughness among meat cuts originating from different areas of the carcass. In this study, however, marked genotype × muscle interactions were also

detected on IMC amount (P = 0.005), HLP muscle concentration (P = 0.002), and IMC maturity (P = 0.0001). This indicates that there are significant within-muscle differences in IMC properties due to lamb genotype, as showed in Figure 1.

Conclusions

Results of the present study evidence that lambs of different genotype, slaughtered at a similar live weight, exhibit differences in IMC amount and maturation leading to a variability in toughness of lamb meat production. Specifically, GP lambs produce a meat that could be tougher than that from the faster-growing purebred and crossbred MI lambs. Moreover, it is confirmed that the well-known differences in background toughness among meat cuts originating from different areas of the carcass correspond to strong variations in muscle collagen content and crosslinking in spite of substantial within-muscle differences due to lamb genotype.

Pertinent literature

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Table 1. Effects of genotype and muscle on IMC properties of GP, MI and MI×P lambs (mean values ± standard errors)

	IMC µg/mg(1)	HLP µg/mg(1)	HLP/IMC mol/mol
Genotype			
GP	$20.8^{A} \pm 0.6$	$4.8^{A} \pm 0.2$	$0.160^{A} \pm 0.004$
MI	$25.5^{B} \pm 1.0$	$3.4^{B} \pm 0.2$	$0.094^{B} \pm 0.003$
MI×P	$25.1^{B} \pm 0.7$	$3.2^{B} \pm 0.3$	$0.086^{B} \pm 0.007$
Muscle			
Longissimus dorsi	$19.6^{a} \pm 0.7$	$2.6^{A} \pm 0.1$	$0.096^{a} \pm 0.006$
Semimembranosus	$24.6^{bc} \pm 0.6$	$4.2^{B} \pm 0.3$	$0.120^{b} \pm 0.009$
Gluteo biceps	$26.2^{b} \pm 1.1$	$4.7^{B} \pm 0.3$	$0.130^{bc} \pm 0.008$
Semitendinosus	$22.6^{c} \pm 0.8$	$4.4^{B} \pm 0.3$	$0.142^{c} \pm 0.012$
Significance			
Genotype (G)	0.0001	0.0001	0.0001
Muscle (M)	0.0001	0.0001	0.0001
G × M	0.005	0.002	0.0001

(1)Of lyophilized muscle

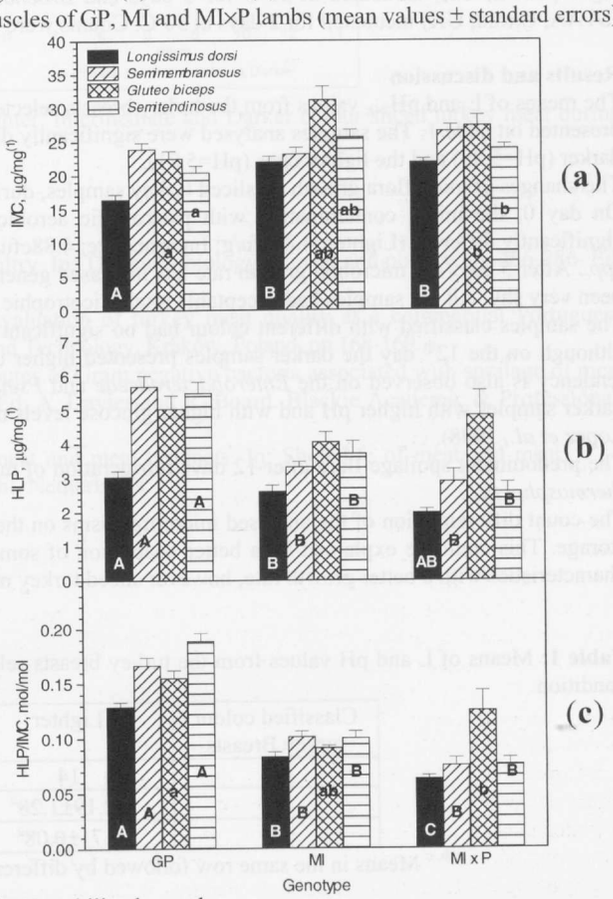
Different superscripts, within a column, stand for significant differences (a, b, c: P < 0.01; A, B: P < 0.001)

Table 2. Simple correlation coefficients between IMC properties and between these and lamb weight gain

	Weight gain	IMC	HLP
IMC	0.314**		
HLP	–0.342***	0.302**	
HLP/IMC	–0.508***	–0.203*	0.851***

*P < 0.05; **P < 0.01; ***P < 0.001

Figure 1. IMC (**a**) and HLP (**b**) amounts and HLP/IMC ratio (**c**) in muscles of GP, MI and MI×P lambs (mean values ± standard errors)



(1)Of lyophilized muscle

Different letters, within the same muscle, stand for significant differences (a, b: P < 0.01; A, B, C: P < 0.001)