Marbling Texture Has No Effect on Collagen Characteristics

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Introduction
Currently, 119 different branded beef programs are governed by the U.S. Department of Agriculture (USDA)—Agricultural Marketing Service. While marbling texture is not officially considered when USDA quality grades (USDA, 1997) are determined, 75% of branded beef programs require carcasses to have fine or medium textured marbling to meet set standards (USDA, 2017). While there are a multitude of factors that can contribute to beef eating experience, marbling is often thought to play a key role. Moody et al. (1970) reported fine marbled beef was more tender than coarser marbled beef, and proposed perimysial connective tissue as the likely cause for the observed difference. Aside from the extent of postmortem proteolysis and muscle fiber structure, collagen is a major influencer of tenderness (Koohmaraie et al., 2002). Differences in tenderness between muscles can occur, in part, due to background effects related to the amount of connective tissue and/or solubility of collagen (Smith and Carpenter, 1974). To date, the Moody et al. (1970) theory has not been adequately evaluated. Therefore, the objective of this study was to determine the effects of marbling texture on collagen traits and adipocyte cross-sectional area.

Experimental Procedures
Beef strip loins (n = 117) from three USDA quality grades [Top Choice (Modest and Moderate marbling), Low Choice, and Select] and three marbling textures (fine, medium, and coarse), were selected using visual appraisal. To fit the criteria for one of three marbling textures, 75% of the marbling in the ribeye had to meet the USDA standard for fine, medium, or coarse textured marbling. After selection, strip loins were taken to the Kansas State University Meat Laboratory, Manhattan, KS, and fabricated into 1-in steaks. Four marbling flecks and the surrounding meat (0.6 in$^3$) were then taken from the medial, central, and lateral portion for adipocyte histochemical analysis. The remaining portion of the steak was vacuum packaged, aged 21 days, homogenized using a Waring blender (Waring Products Division; Hartford, CT), and stored at -112°F for collagen analysis. Each marbling fleck was cryosectioned and subjected to Masson’s trichrome staining for perimysial collagen and adipocyte staining. Photomicrographs were taken and the cross-sectional area of a minimum of 200 adipocytes were measured and perimysium connective tissue thickness was measured every 10 μm (Figure 1). Insoluble, soluble, and total collagen content was determined using the
methods of Gonzalez et al. (2014). Perimysial collagen was extracted from the meat, freeze dried, and analyzed using a differential scanning calorimeter (Shimadzu Scientific Instruments, Kyoto, Japan) to determine peak melting temperature. Data were analyzed as a completely randomized design with a 3 × 3 factorial arrangement.

**Results and Discussion**

There were no marbling texture × quality grade interactions (P>0.05) for all traits studied. All three marbling textures (fine, medium, and coarse) contained a similar (P>0.05) amount of soluble and insoluble collagen (Table 1). Additionally, each marbling texture had a similar (P>0.05) amount of total collagen. All three quality grades had a similar (P>0.05) amount of soluble, insoluble, and total collagen.

Incidentally, a similar trend was shown when evaluating the effects of marbling texture and quality grade on perimysial thickness, and peak thermal transition temperature. Quality grade did not have an effect (P>0.05) on perimysial thickness. Furthermore, marbling texture had no effect (P>0.05) on peak thermal transition temperature. Lastly, all quality grades contained a similar (P>0.05) perimysial thermal melting temperature, thus showing that quality grade had no effect (P>0.05) on peak thermal melting temperature.

Marbling texture and quality grade impacted (P<0.05) adipocyte cross sectional area (Figure 2). Coarse marbled steaks contained larger (P<0.05) adipocytes than fine marbled steaks. Adipocytes of medium marbled steaks were similar (P>0.05) in size when compared to adipocytes of fine and coarse marbled steaks. While adipocytes of Top and Low Choice steaks were similar (P>0.05) in size, their adipocytes were larger (P<0.05) than adipocytes of Select steaks.

**Implications**

These results indicate that marbling texture has no effect on collagen traits and any potential tenderness differences among beef varying in marbling texture are not related to these traits. However, both quality grade and marbling texture category impacted adipocyte cross sectional area.

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Table 1. Least squares means of collagen characteristics of beef strip loin steaks of three marbling textures and three quality grade treatments

| Treatment          | Soluble collagen, mg/g | Insoluble collagen, mg/g | Total collagen, mg/g | Perimysial peak transitional temperature, °F | Perimysial thickness, µm |
|--------------------|------------------------|---------------------------|----------------------|---------------------------------------------|--------------------------|
| Marbling texture   |                        |                           |                      |                                             |                          |
| Coarse             | 1.48                   | 9.49                      | 10.98                | 119.97                                      | 43.23                    |
| Medium             | 1.74                   | 9.81                      | 11.56                | 129.33                                      | 46.26                    |
| Fine               | 1.72                   | 9.92                      | 11.64                | 226.78                                      | 41.44                    |
| Standard error of  | 0.12                   | 0.34                      | 0.38                 | 36.90                                       | 2.20                     |
| the least squares  |                        |                           |                      |                                             |                          |
| mean¹              |                        |                           |                      |                                             |                          |
| P-value            | 0.27                   | 0.65                      | 0.41                 | 0.17                                        | 0.31                     |
| Quality grade      |                        |                           |                      |                                             |                          |
| Top Choice²        | 1.50                   | 9.86                      | 11.36                | 122.52                                      | 44.56                    |
| Low Choice         | 1.77                   | 9.61                      | 11.37                | 118.11                                      | 42.54                    |
| Select             | 1.67                   | 9.77                      | 11.44                | 125.44                                      | 43.84                    |
| Standard error of  | 0.12                   | 0.34                      | 0.38                 | 36.90                                       | 2.20                     |
| the least squares  |                        |                           |                      |                                             |                          |
| mean¹              |                        |                           |                      |                                             |                          |
| P-value            | 0.31                   | 0.87                      | 0.99                 | 0.57                                        | 0.81                     |
| Texture × QG       |                        |                           |                      |                                             |                          |
| P-value            | 0.19                   | 0.28                      | 0.12                 | 0.47                                        | 0.36                     |

¹Standard error (largest) of the least squares means in the same main effect (marbling texture or quality grade).
²USDA marbling score of Modest⁰⁰-Moderate¹⁰⁰.
Figure 1. Representative photomicrograph of Masson’s trichrome staining of bovine longissimus lumborum intramuscular adipose and muscle tissue a) Photomicrograph utilized for intramuscular adipocyte cross-sectional area analysis b) Photomicrograph utilized for perimysium thickness measures (A = perimysium; B = marbling adipocyte; C = muscle cell); scale bars = 100 µm.
Figure 2. Least squares means of adipocyte cross-sectional area of beef strip loin steaks of
a) three marbling textures and b) three quality grade treatments
Top Choice = marbling score of Modest$^{00}$ to Moderate$^{100}$. Means within a panel without a
common superscript differ (P<0.05).