Native beef MMP-2 may contribute to postmortem collagen degradation in extended aged beef

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Introduction: Connective tissue plays a significant role in beef tenderness and is thought to contribute a fixed amount of background toughness to beef (Bailey and Light, 1989), but many past research have demonstrated weakening of connective tissue structure during postmortem aging of meat from various livestock species (Liu et al., 1994; Nishimura et al., 1995; Nishimura et al., 2008). Collagenase type matrix metalloproteinases (MMPs) are a family of calcium and zinc dependent proteases that is responsible for the proteolytic turnover and cleavage of intramuscular connective tissue (Löffek et al., 2011). Our lab has recently demonstrated native beef MMP-2 activity in beef through collagen zymography. However, the role of MMP-2 in the beef tenderization process is not well defined. Therefore, the objectives of this study were 1) to determine if the native beef MMP-2 can contribute to connective tissue degradation in a simulated standard industry postmortem aging condition and 2) to explore approaches to improve the native beef collagenase activity.

Materials and methods: Longissimus lumborum, gastrocnemius and gluteus medius were collected and fabricated from 10 USDA Choice beef carcasses (n=30) at 3 d postmortem. For the first objective, forty 10% polyacrylamide gels containing 0.5 mg/ml bovine type I collagen were casted and run with non-reducing SDS-PAGE containing 30 ug of sarcoplasmic proteins from the beef samples described above. The 40 gels were incubated at 4 ± 2 °C in a MMP zymogram developing buffer containing 5 mM CaCl2 adjusted to a pH of 5.7 and subjected to 4 storage periods: 1) 48 h; 2) 21 d; 3) 42 d and 4) 63 d. For the second objective, 40 collagen polyacrylamide gels were casted, loaded and run as described previously, but incubated at 37 °C for 36 h in 4 different developing buffers: 1) 5 mM CaCl2 only; 2) 5 mM CaCl2 +20 µM of ZnCl2; 3) 5 mM CaCl2 +50 µM of ZnCl2; 4) 5 mM CaCl2 +100 µM of ZnCl2.

Results: No detectable native beef MMP-2 activity was found in 48 h, 21 d nor 42 d gels under simulated industry meat storage condition. However, noticeable native beef MMP-2 activity was found in all three muscle extracts in 63 d gels, which gastrocnemius had greater MMP2 activity than gluteus medius ($P < 0.05$) and tended to have greater MMP2 activity than longissimus lumborum ($P = 0.07$). Furthermore, the addition of 20 µM of ZnCl2 in the developing buffer increased the native beef MMP-2 activity by 9 folds, while the addition of 50 and 100 µM of ZnCl2 increased native beef MMP-2 activity by 6 and 4 folds compared to 5 mM CaCl2 only, respectively ($P < 0.05$).

Conclusion: Our aforementioned study confirmed that native beef MMP-2 can contribute to collagen degradation in extended aged beef (42 d +), and a small increase in intracellular zinc concentration may significantly improve this collagenase activity during aging.

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Literature:

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