






## Measuring and Quantifying the Role of Collagen Crosslinks in Beef Tenderness

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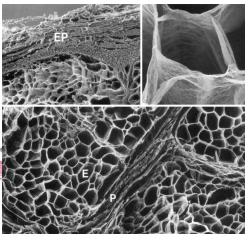
## Contributors to Palatability

- Tenderness
  - Major determinant of eating quality
- Affected by:
  - Myofibrillar proteins
    - Contractile and cytoskeletal
    - Calpain proteolytic system
  - Connective tissue
    - Most abundant protein
    - Soluble/Insoluble







## Collagen Basics

- Layers of Collagen:
  - Epimysium
  - Perimysium
  - Endomysium

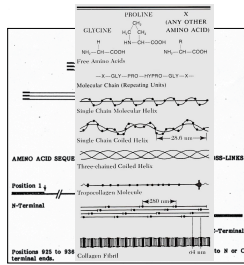


Nishimura et al., 1994






## Collagen Basics

- Structure
  - Long cylindrical protein
  - Suprahelical
  - Diameter (1.5 nm)
- Crosslinks
  - Types






Aberle et al., 2003




## Collagen Basics

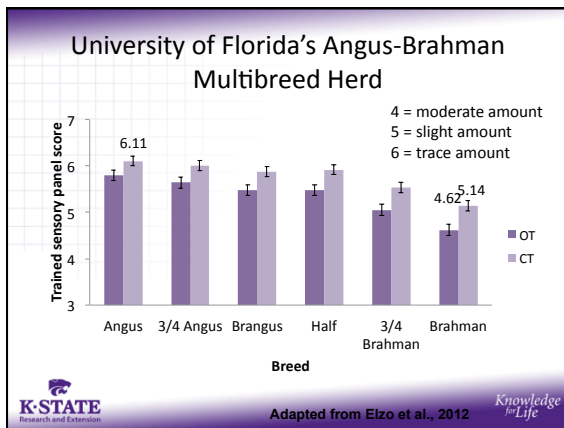
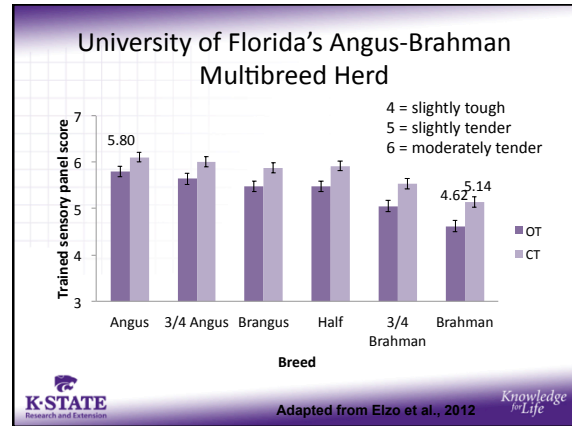
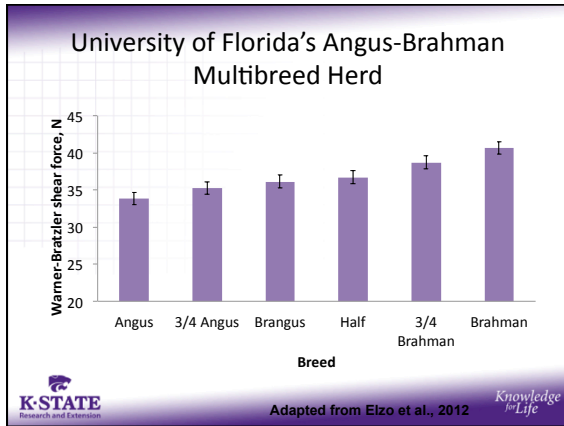
- Determinants of Crosslinking
  - Animal Age
  - Sex
  - Location within carcass
  - Breed?
- How Can We Measure?
  - Traditional/Indirect
  - Molecular Methods

## University of Florida's Angus-Brahman Multibreed Herd

- Continuous genetic composition
- Over 20 years of data
- Unique herd to serve as a model for:
  - Calpain/calpastatin
  - Collagen characteristics



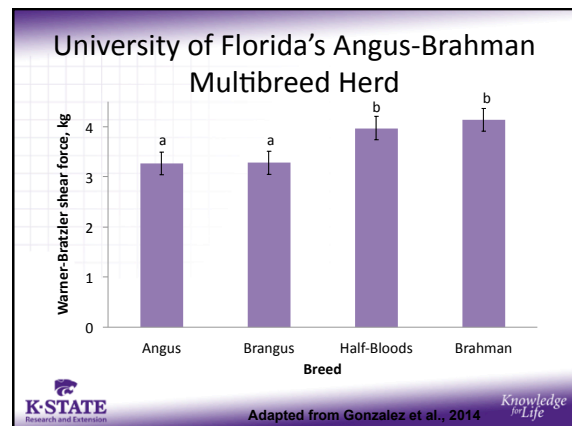
### University of Florida's Angus-Brahman Multibreed Herd

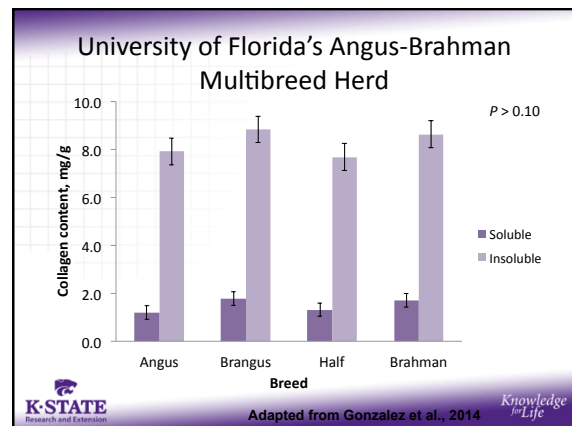
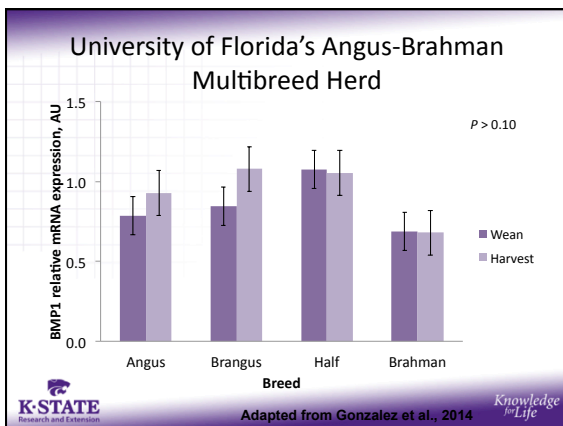
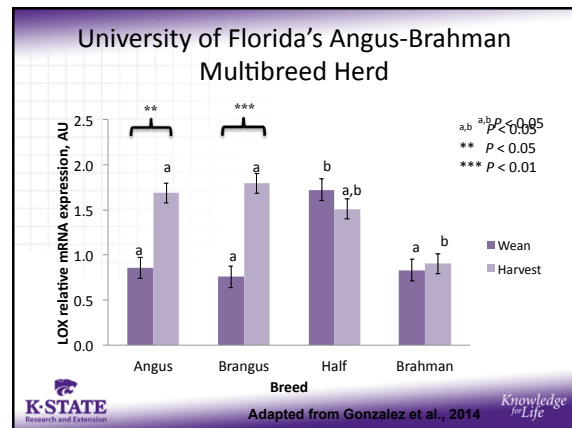
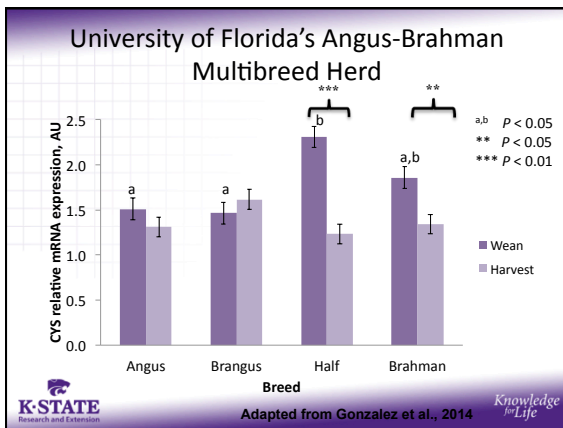
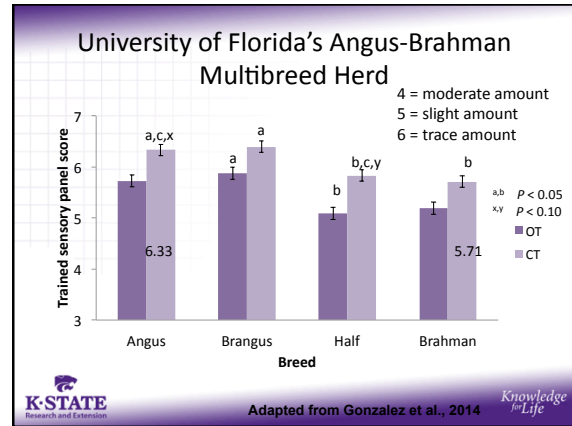
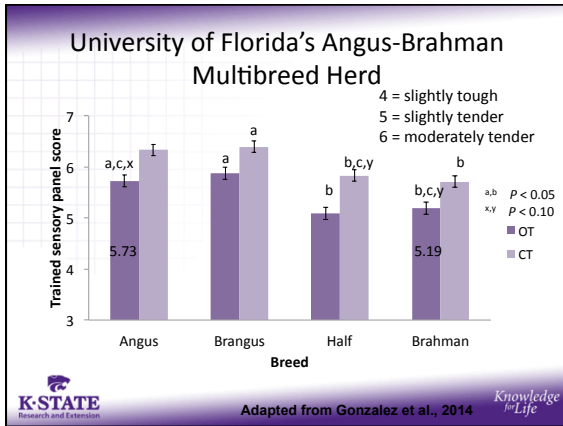
- 13 hd/group biopsied
  - Angus: >26/32
  - Brangus: 20/32
  - Half-Blood: 14-18/32
  - Brahman: 0-9/32
- Enzymatic crosslink pathway
  - Real Time PCR

### University of Florida's Angus-Brahman Multibreed Herd

- Meat analysis
  - Warner-Bratzler shear force
  - Trained sensory panel
  - Soluble/Insoluble collagen content
- Statistical Analysis
  - Randomized complete design with repeated measures



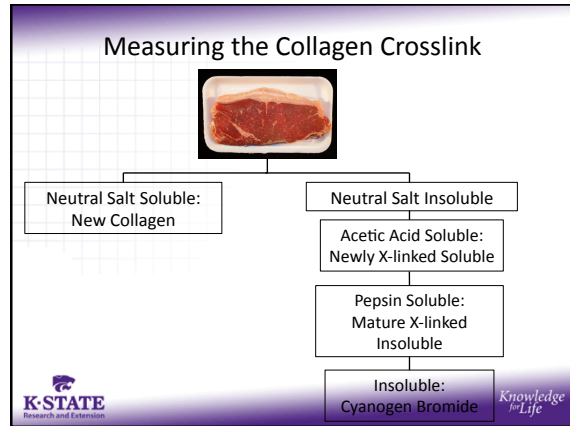
Adapted from Gonzalez et al., 2014





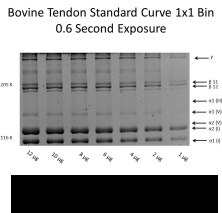


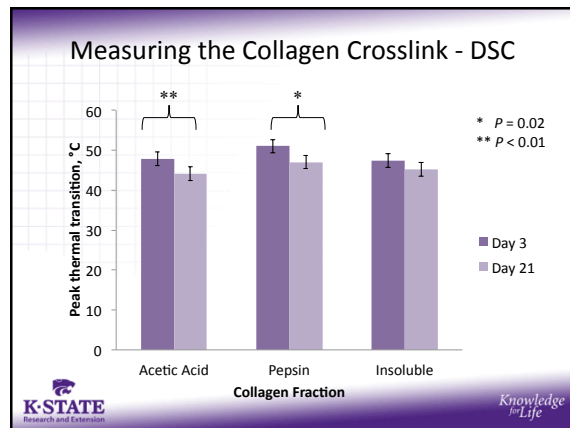
### University of Florida's Angus-Brahman Multibreed Herd

- Conclusions:
  - Multibreed herd will yield meat that is divergent in tenderness and connective tissue.
  - Cystatin C and lysyl oxidase mRNA expression indicates that breeds exhibit differential crosslink pattern.
  - No difference in soluble and insoluble collagen content utilizing traditional methods.
    - Are we measuring the correct attribute of collagen?



### Measuring the Collagen Crosslink

- SDS-PAGE/Western Blotting
  - Pepsin soluble fraction
  - Measures quantity
- Differential Scanning Calorimetry
  - All 3 fractions
  - Measures thermal transition



### Current Research and Future Directions

- Important Questions:
  - What is the pattern of enzymatic crosslinking?
    - AFRI submission
  - Can we develop a more reliable measurement of collagen crosslinks?
  - What impact does extended aging elicit on collagen crosslinks?

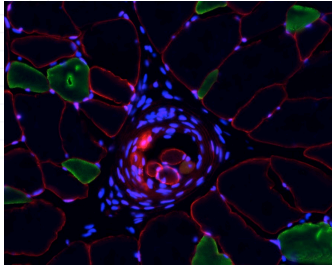



### Acknowledgements

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- Virginia Tech
  - Dr. Sally Johnson
- Beef Cattle Research Center
  - Dr. Jim Drouillard

### Questions



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**Simple correlation coefficients of tenderness traits with calpastatin activity, total and insoluble collagen content of Brahman cattle**

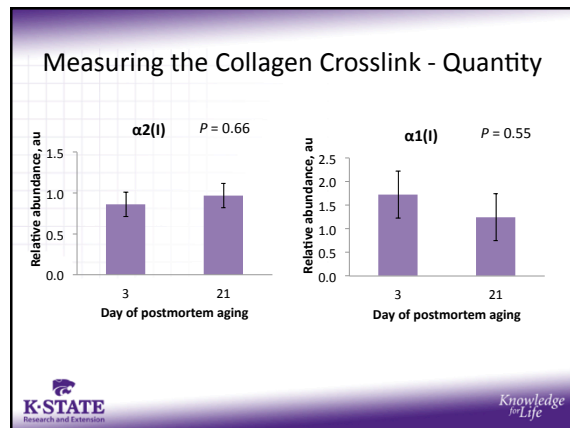
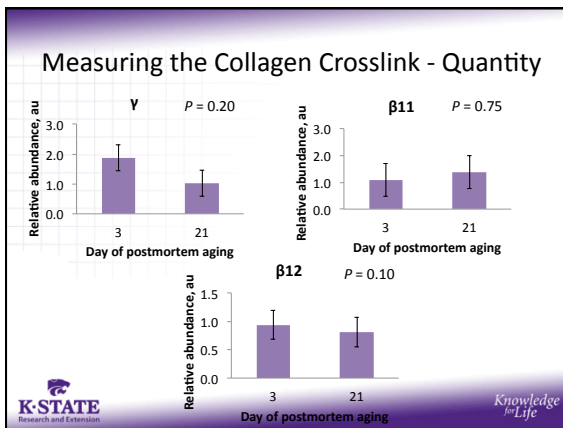
|   | Warner-Bratzler shear force (N) |        |        | OT <sup>1</sup> |
|---|---------------------------------|--------|--------|-----------------|
|   | 7 day                           | 14 day | 21 day |                 |
| <b>Collagen, mg/g muscle</b>              |                                 |        |        |                 |
| Total                                     | 0.66                            | 0.56   | 0.82   | -0.36           |
| Insoluble                                 | 0.66                            | 0.57   | 0.83   | -0.37           |
| Calpastatin <sup>2</sup> , units/g muscle | -0.12                           | -0.05  | -0.06  | -0.01           |

<sup>1</sup>Sensory panel overall tenderness.  
<sup>2</sup>Calpastatin activity measured at 24 hours.

Adapted from Riley et al., 2005

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\*\*Note - I am not endorsing or condemning one breed of cattle over another.

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### Outline

- Collagen Contribution to Palatability
- Collagen Basics
- University of Florida Study
- Measuring Collagen Crosslinks
- Conclusions and Future Directions

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