



Proteomic Features Associated with Tenderness of Aged Pork Loins

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Objectives

It is established that tenderness is a fundamental component of meat quality. It is also known that fresh pork loin tenderness does vary. Consumers value quality and are willing to pay a premium for consistently tender pork products. The lack of differentiation in the quality of pork products coupled with the consumer desire for tenderness demonstrates a need for definition of pork quality. Identifying biomarkers to classify tough and tender pork products could be a way to distinguish high quality pork for consumers. Therefore, the objectives of this study were to determine proteolytic and sarcoplasmic proteome differences that contribute to tenderness variation in aged pork loins.

Materials and Methods

Loins ($n = 159$) were collected 1 d postmortem from carcasses of Duroc-sired crossbred commercial pigs and aged for approximately 9 to 11 d. Chops (2.54 cm) were collected and evaluated for purge, cook loss, ultimate pH, visual color and marbling, Hunter L, a, b values, sensory, star probe (kg), and total lipid. Samples were selected for proteomic experiments based on star probe values. Selected samples were within specified ranges for ultimate pH (5.54 to 5.86), marbling score (1.0 to 3.0), and total lipid (1.61 to 3.37%). Samples were classified into either a low star probe (LSP) group ($n = 12$, average star probe = 4.95 kg) or high star probe (HSP) group ($n = 12$, average star probe = 7.75 kg). Proteolytic and muscle fiber type data were collected using SDS-PAGE and Western blot analyses. Data were analyzed with SAS mixed procedure (SAS Inst. Inc., Cary, NC), with a fixed effect of star probe force and random effect of gel. Two-dimensional difference in gel electrophoresis (2D-DIGE) was used to examine sarcoplasmic proteome differences.

Data were analyzed with DeCyder (GE Healthcare), using Student's paired t test. Significantly different protein spots were identified using mass spectrometry.

Results

Calpain-1 was completely autolyzed in both HSP and LSP samples. LSP whole muscle samples had more troponin-T ($P < 0.01$) and desmin ($P < 0.01$) degradation than HSP samples. Both classification groups showed degradation of titin in myofibrillar samples, but select HSP samples also exhibited intact bands of titin. Results from 2D-DIGE showed HSP samples had significantly more abundant metabolic, stress response, and regulatory proteins compared to LSP samples. The stress response protein peroxiredoxin-2 was more abundant in HSP samples as determined by 2D-DIGE ($P \leq 0.01$) and Western blot confirmations ($P = 0.02$). LSP samples showed significantly more degradation of the protein desmin in the 2D-DIGE experiment ($P < 0.01$) and Western blot confirmations ($P < 0.01$).

Conclusion

These results demonstrate extreme proteolytic differences of troponin-T, desmin, and titin influenced measured tenderness of LSP and HSP samples. The protein spot identified as the structural protein desmin in the sarcoplasmic fraction is a novel result and demonstrates degradation influences solubility of this protein. HSP samples had more abundant stress response proteins, such as peroxiredoxin-2, indicating cellular stress may contribute to tougher pork. The protein spots identified as peroxiredoxin-2 and desmin were found at different abundances in the soluble protein fraction of classification groups. Therefore, these proteins have potential to be utilized as biomarkers by examining meat purge to differentiate between tough and tender aged pork products.