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Near-infrared tissue oximetry of beef longissimus muscle for the improvement of meat color and meat color stability

Abstract
Meat color as perceived by consumers serves as a valuable guide for assessing overall quality and wholesomeness of meat. The bright cherry-red color of beef is influenced by tissue oxygen consumption, obstacles to oxygen diffusion, and thickness of the oxymyoglobin layer. The dynamics of meat color depend on several physical properties of muscle including myoglobin redox status and concentration. Physical, chemical, and anatomical differences in muscles cause large variations in color from cut to cut, within a cut, and in cuts made parallel or perpendicular to muscle fibers. Clearly, muscle fiber orientation affects measurements of tenderness and cooking yields; however, variations in myoglobin redox dynamics, oxygen penetration, and color stability due to muscle fiber orientation (parallel or perpendicular) are not well documented. Among the various meat color measurement techniques available, near-infrared (NIR) methods have the advantages of being nondestructive, rapid, inexpensive, and adaptable for online measurements. The NIR tissue oximeter is a relatively new biomedical device that has been used in exercise physiology and in medicine to measure hemoglobin and myoglobin oxygen saturation in brain tissue and cardiac and skeletal muscle. This instrument seems to have promise for use in measuring inherent properties of meat that are related to meat color stability. NIR tissue oximetry may provide continuous real-time measurements of changes in myoglobin oxygen status, thus providing information on tissue oxygenation and hemodynamics. The unique feature of the tissue oximeter is that it uses the theory of photon migration through tissue, allowing for absolute measurement of absorption in, for example, human or animal tissue. If the NIR absorption properties of any chromophore are known, quantitative analysis of color compounds is possible without constant calibration and validation. We are not aware of any research in which NIR tissue oximetry has been used to evaluate color of post-rigor meat. This study was designed to evaluate whether NIR tissue oximetry has promise for measuring meat properties related to meat color. Specific objectives were to determine: (1) effects of parallel vs. perpendicular muscle fiber orientation of meat cuts on NIR measurements, (2) amounts of deoxymyoglobin (DMb), oxymyoglobin (OMb), and total myoglobin (TMb) in the superficial and subsurface layers of beef muscle (longissimus) stored in several packaging formats, and (3) tissue oximeter responses to post-rigor muscle fiber orientation and surface measures of color.

Keywords
Cattlemen's Day, 2009; Report of progress (Kansas State University. Agricultural Experiment Station and Cooperative Extension Service); 1010; Kansas Agricultural Experiment Station contribution ; no. 09-168-S; Beef; Cattle; Near-infrared(NIR); Meat Color Stability

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A. Mohan, M. C. Hunt, T. A. Houser, and T. E. Barstow

Introduction
Meat color as perceived by consumers serves as a valuable guide for assessing overall quality and wholesomeness of meat. The bright cherry-red color of beef is influenced by tissue oxygen consumption, obstacles to oxygen diffusion, and thickness of the oxymyoglobin layer. The dynamics of meat color depend on several physical properties of muscle including myoglobin redox status and concentration. Physical, chemical, and anatomical differences in muscles cause large variations in color from cut to cut, within a cut, and in cuts made parallel or perpendicular to muscle fibers. Clearly, muscle fiber orientation affects measurements of tenderness and cooking yields; however, variations in myoglobin redox dynamics, oxygen penetration, and color stability due to muscle fiber orientation (parallel or perpendicular) are not well documented. Among the various meat color measurement techniques available, near-infrared (NIR) methods have the advantages of being nondestructive, rapid, inexpensive, and adaptable for online measurements.

The NIR tissue oximeter is a relatively new biomedical device that has been used in exercise physiology and in medicine to measure hemoglobin and myoglobin oxygen saturation in brain tissue and cardiac and skeletal muscle. This instrument seems to have promise for use in measuring inherent properties of meat that are related to meat color stability. NIR tissue oximetry may provide continuous real-time measurements of changes in myoglobin oxygen status, thus providing information on tissue oxygenation and hemodynamics. The unique feature of the tissue oximeter is that it uses the theory of photon migration through tissue, allowing for absolute measurement of absorption in, for example, human or animal tissue. If the NIR absorption properties of any chromophore are known, quantitative analysis of color compounds is possible without constant calibration and validation. We are not aware of any research in which NIR tissue oximetry has been used to evaluate color of post-rigor meat.

This study was designed to evaluate whether NIR tissue oximetry has promise for measuring meat properties related to meat color. Specific objectives were to determine: (1) effects of parallel vs. perpendicular muscle fiber orientation of meat cuts on NIR measurements, (2) amounts of deoxymyoglobin (DMb), oxymyoglobin (OMb), and total myoglobin (TMb) in the superficial and subsurface layers of beef muscle (longissimus) stored in several packaging formats, and (3) tissue oximeter responses to post-rigor muscle fiber orientation and surface measures of color.

Experimental Procedures
The longissimus lumborum from three beef loins (USDA Select, A-maturity) were fabricated at 10 days postmortem into steaks about $2 \times 3 \times 4$ in. with the fiber orientation
either perpendicular (PR) or parallel (PL) to a designated muscle surface. Steaks were assigned to four packaging treatments: (1) vacuum packaging (VP), (2) high-oxygen modified atmosphere packaging (HiOx; 80% O₂, 20% CO₂), (3) polyvinyl-chloride film overwrap (PVC; 21,700 cc O₂/m²/24 hour), and (4) HiOx that was converted to PVC after day 2 for subsequent storage in PVC (HiOx-PVC). Steaks were stored in the dark at 36°F for 0, 2, 4, 10, and 15 days, and color of the meat surface was measured after each storage time. Hue angle and chroma were calculated. Tissue oximetry (Figure 1) of the steaks was used to determine the surface and subsurface myoglobin redox status by using an NIR system (OxiplexTS model 96208, ISS Inc., Champaign, IL). The device works by emitting NIR light into tissue at known distances from a collector. This NIR spectrum can penetrate skin, subcutaneous fat/skull, and underlying muscle and is absorbed and scattered within the tissue. Light of two different wavelengths (692 and 834 nm) is used, and the light is modulated at a 110 MHz frequency. Collected light is measured and processed, and the absorption and scattering coefficients of the medium are determined. The assumption is that myoglobin is the only significant absorber of selected wavelengths in muscle and the OMb and DMb concentrations can be quantitated. Data were statistically analyzed by using type-3 tests of fixed effects of the MIXED procedure of SAS. F-test denominator degrees of freedom were estimated by using the Satterthwaite adjustment, and least squares means for significant F-tests were separated by using least significant differences.

**Results and Discussion**

Figure 2 shows a fiber orientation × packaging interaction (P<0.05) of NIR tissue oximeter response for percentages of OMb, DMb, and TMb. Steaks cut perpendicular to the fiber orientation and packaged in HiOx contained 65% OMb, whereas those packaged in HiOx-PVC had 60% OMb (Figure 2A) compared with steaks cut parallel to the fiber orientation (P<0.05). An opposite trend for fiber orientation effects was observed for DMb (Figure 2B). The TMb concentration did not differ (P>0.05) among steaks cut PR and PL, except for steaks packaged in HiOx-PVC (Figure 2C).

There was a fiber orientation × packaging × day interaction (P<0.05) for percentages of OMb and DMb in the four packaging formats (Figure 3). As expected at day 0, OMb percentages of steaks cut PR and PL (Figure 3A) and packaged in VP, PVC, and HiOx and HiOx-PVC were less than 5%, 7%, and 28%, respectively. By day 2, OMb dramatically increased to 78% in HiOx and HiOx-PVC packaged steaks cut either PR or PL, whereas steaks in PVC and VP had 48% and <5% OMb regardless of their fiber orientation. On day 10, OMb level increased to 90% in steaks cut PR and packaged in HiOx but remained the same in steaks cut PR and packaged in HiOx-PVC. However, OMb in steaks cut PL declined to 71% in HiOx and 66% in HiOx-PVC and did not change in steaks cut either PR or PL and packaged in PVC and VP. By day 15, OMb percentage declined further in all aerobic packages and increased slightly in the VP. Levels of DMb (Figure 3B) followed an opposite pattern; however, in general, steaks cut PR had lower percentages of DMb compared with steaks cut PL and packaged in HiOx and HiOx-PVC.

These data clearly demonstrate that fiber orientation affected NIR measurements of myoglobin oxygen status in aerobic packaging formats. Changes in myoglobin redox forms due to fiber orientation, packaging, and postmortem storage altered NIR quantitative measurements of myoglobin pigment forms that were expected from the treatment
combinations. There was a storage day × packaging interaction (P<0.05) of instrumental color (Figure 4). Steaks packaged in HiOx, HiOx-PVC, and PVC decreased (P<0.05) in a* (redness) and chroma (color intensity) as postmortem storage day advanced from day 0 to days 4, 10, and 15 (Figure 4B and 4E). On days 2 and 4, steaks packaged in HiOx and HiOx-PVC had greater redness intensity (P<0.05) than steaks packaged in PVC and VP. However, instrumental b* values decreased (less yellow) for steaks packaged in HiOx, HiOx-PVC, and PVC from days 0, 2, 4, and 10 to day 15 but remained unchanged in VP steaks (Figure 4C).

There were no significant change in muscle lightness (L* values) from days 0, 4, and 10 to day 15 (Figure 4A). Hue angles (overall color) increased for steaks packaged in VP from days 0, 2 and 4 and then decreased from day 10 to 15 (P<0.05) (Figure 4D). Differences among HiOx, HiOx-PVC, PVC, and VP were evident on day 15. These color measurements would be expected considering the redox forms of myoglobin present in the packages.

**Implications**

NIR tissue oximetry measurements have potential for rapid, real-time, and noninvasive assessment of color stability differences between muscles packaged in a variety of packaging formats. However, to obtain a repeatable measurement on post-rigor muscles, fiber orientation, tissue oxygen exposure, and storage time must be controlled.
Figure 2. Fiber orientation × packaging interaction of near-infrared tissue oximeter for percentage of (A) oxymyoglobin, (B) deoxymyoglobin, and (C) total myoglobin.
Figure 3. Packaging × day interaction of tissue oximeter response for (A) oxymyoglobin and (B) deoxymyoglobin.
Figure 4. Storage day × packaging interaction for instrumental surface color.