Effect of Mitochondrial Inhibitor on Fresh Meat Color

M. Lawson[1], M. L. Denzer[1], F. Kiyimba[1], G. G. Mafi[1], R. Ramanathan[1]

[1]Department of Animal and Food Sciences, Oklahoma State University, Stillwater, OK 74078

Introduction

- Oxygen consumption (OC) is a muscle property that influences fresh beef color (AMSA, 2012).
- Greater OC results in less bloom and darker meat color, whereas lower OC can produce a bright-red color which is more desirable to the consumer for purchase and consumption.
- In postmortem muscles, mitochondria and oxygen-consuming enzymes are primarily involved in OC (Ramanathan et al., 2020).
- OC is important in characterizing beef color changes (Mancini and Hunt, 2005).
- Although the role of mitochondria in beef color has been reported, limited research has validated the role of mitochondria in meat color. This research investigated the addition of rotenone as a mitochondrial inhibitor.

Methods and Materials

- Chuck Steaks were purchased from Homeland and were coarse ground, mixed with rotenone, vacuum packaged and stored for 24 h to reaction to occur, finely ground, and then packed into quarter pound patties. Control patties are packed on d0. Patties are PVC packaged and placed in retail display case for trials on d0 and d3.
- Surface color, oxygen consumption, metmyoglobin reduction activity (MRA), lipid oxidation, pH readings, and NADH levels were determined.
- OC methodology was based on changes in oxygen myoglobin level when bloom patties were vacuum packaged. Briefly, patties were bloomed for one-hour, vacuum packaged (no continuous oxygen supply), and incubating samples at 30° C for 30 min to promote deoxymyoglobin formation. The HunterLab spectrophotometer readings provide the visible spectra to determine oxymyoglobin, which was calculated as 1 – (K/S 610 ÷ K/S 525).
- MRA methodology using HunterLab spectrophotometer was used to evaluate metmyoglobin production.
- Lipid oxidation methodology used to measure oxidative changes in meat. NADH was determined by enzyme recycling method.
- The assays were replicated three times and the data were analyzed using the Mixed Proc of SAS.

Objective

- The objective of the study was to determine the effect a mitochondrial inhibitor, rotenone, on fresh meat color.
- The purpose of research was to validate the role of mitochondria on meat color (rotenone is a mitochondrial inhibitor and not intended for consumption).

Hypothesis

- We hypothesize that the addition of a mitochondrial inhibitor will inhibit electron flow at complex I and impact meat color.

Results

- Addition of rotenone improved redness of patties (Figure 2 & 4).
- There was no effect of rotenone addition on pH (Figure 5). Addition of rotenone decreased oxygen consumption, probably due to inhibition of complex I activity and utilization of NADH. In support, NADH content was lower in rotenone treated samples.
- Lipid oxidation was lower in rotenone treated samples compared with control (Figure 3).
- Interestingly, MRA increased in rotenone-treated samples.

Discussion and Conclusions

- The current research validates the role of mitochondria in beef color.
- Meat color is determined by reciprocal interaction of biomolecules, mitochondria, and myoglobin.
- In addition, lipid and myoglobin oxidation are inter-related. Primary and secondary oxidation products can enhance myoglobin oxidation. Secondary oxidation products can bind with myoglobin and promote heme release.
- MRA occurs by three different pathways. In the current research, only complex I was blocked with rotenone. Decreased lipid oxidation and other MRA pathways may have increased MRA and redness.
- Future research will determine the role of different mitochondrial inhibitors on beef color.

References & Acknowledgements

The authors would like to thank the Wentz Scholar Program and the Office of Scholar Development and Undergraduate Research for their financial support to conduct this research.

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Methods and Materials

Patty Preparation

- Chuck Steaks coarsely ground on initial day and control patties formed (115g patties)
- Rotenone added in 0%, .1%, and .2% for respective treatments (Control, Rotenone, 2Rotenone)* and stored 24 hours to allow reaction time
- Coarse ground beef with added rotenone finely ground and made into patties
- Patties PVC packaged and put in retail case for trials to be run on d0 and d3

*100mg of rotenone per 100g of ground beef

OCR

Samples were removed from packaging and fresh meat was exposed for a one-hour bloom process

↓

Vacuum packaged

↓

One-hour bloom HunterLab readings

↓

Incubated at 30 °C for 30 min

↓

HunterLab readings taken at 10, 20, and 30 min

↓

Visible Spectra data is then calculated as 1 – (K/S 610 ÷ K/S 525).

MRA

Samples submerged in .3% $NaNO_2$ solution for 20 min with fresh cut portion displayed

↓

HunterLab readings taken

↓

Incubated at 30 °C for 2 hours

↓

Final HunterLab readings taken

Figure 1. HunterLab MiniScan XE Plus

Lipid Oxidation

Measure 3g of meat sample

↓

Blend 3g sample with 27mL of TCA in homogenizer for 10 seconds

↓

Pour ground sample into funnel and let filter for 30 minutes

↓

Combine 1mL of filtrate with 1mL of TBA in test tube and vortex

↓

Place glass test tube in 100°C for 10 min and then cool for 5 min

↓

Place 1mL of sample in Spectrometer and measure absorbance at 532nm

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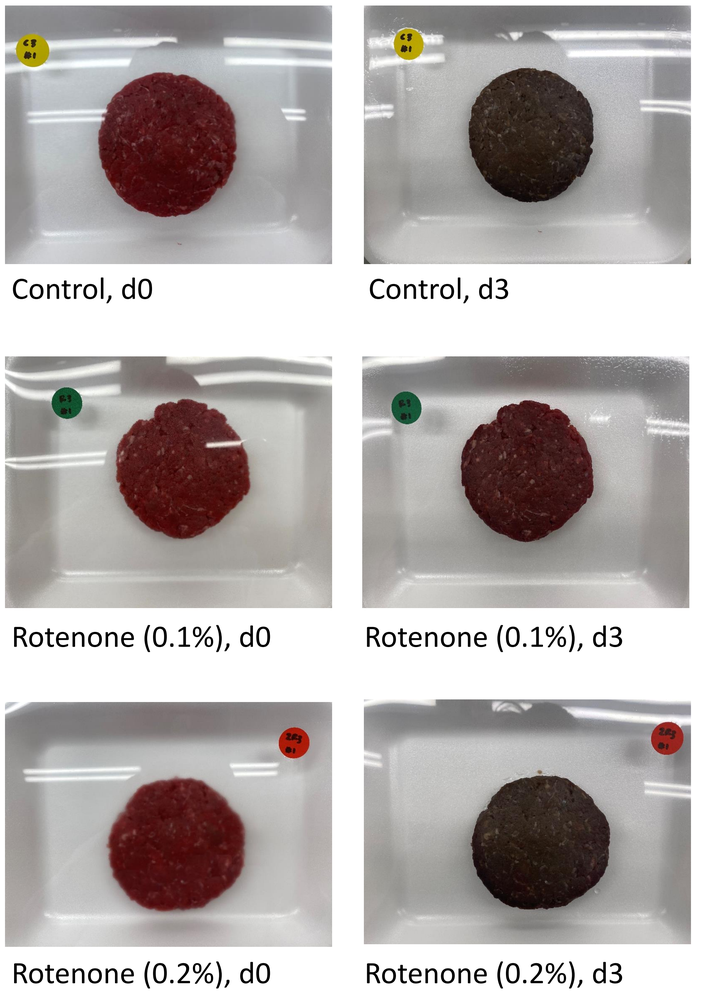
Results

Figure 2: Pictorial representation of the effects of rotenone on color



Control, d0 | Control, d3

Rotenone (0.1%), d0 | Rotenone (0.1%), d3

Rotenone (0.2%), d0 | Rotenone (0.2%), d3

Figure 3: Effects of rotenone and storage time on lipid oxidation

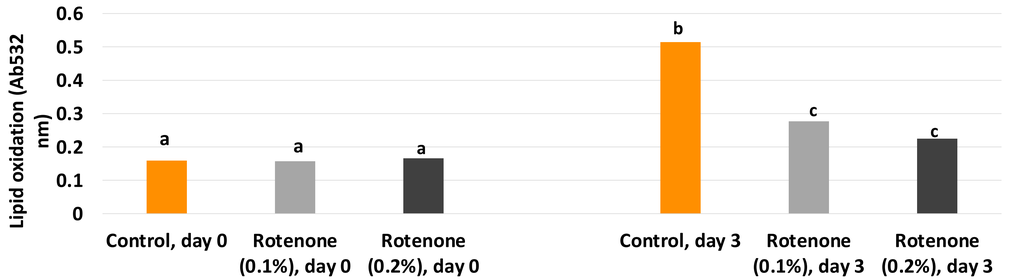


Figure 4: Effects of rotenone on redness during three day storage

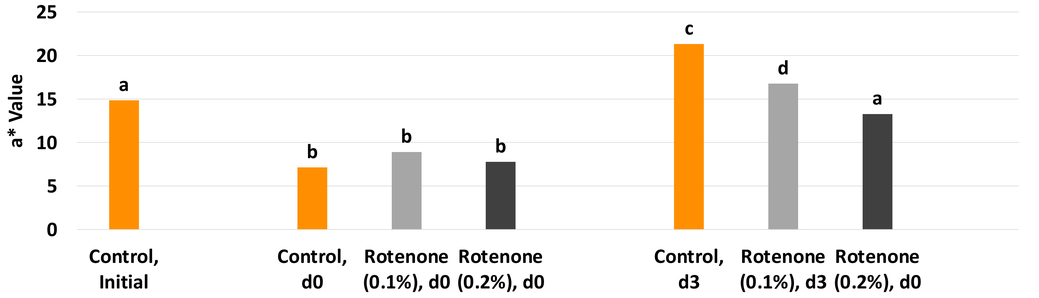


Figure 5: Effects of rotenone addition on pH and NADH content. Least squares mean with different letters indicate difference (P < 0.05).

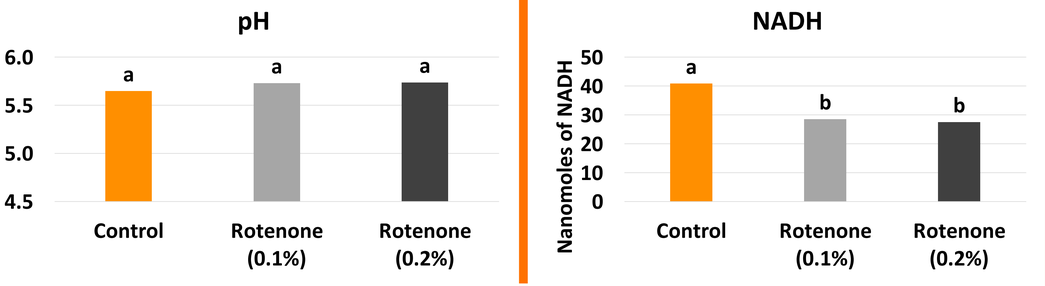


Figure 6: Effects of rotenone addition on metmyoglobin reducing activity. Least squares mean with different letters indicate difference (P < 0.05).

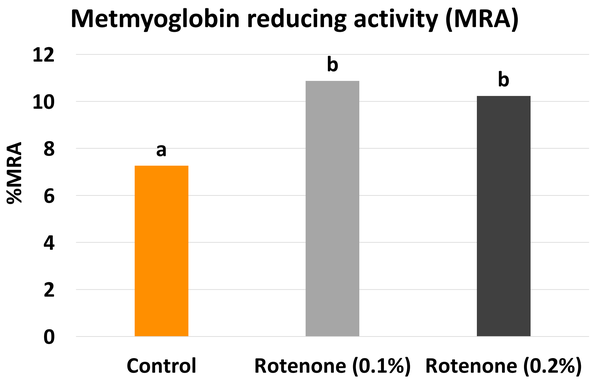
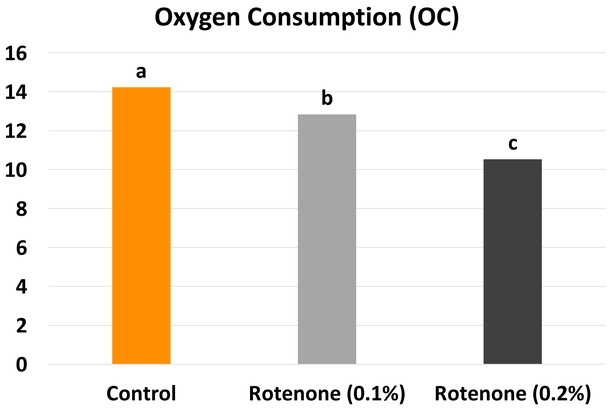


Figure 7: Effects of rotenone addition on oxygen consumption. Least squares mean with different letters indicate difference (P < 0.05).



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