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Mitochondrial Function in Oxidative and Glycolytic Bovine Skeletal Muscle Postmortem

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Objectives

Mitochondrial function in *postmortem* muscle is affected by decreasing oxygenation. Functional properties relating to energy production and integrity of mitochondria may influence development of meat quality characteristics. Therefore, the objective was to evaluate changes in mitochondrial function in oxidative and glycolytic muscles during the first 24h postmortem.

Materials and Methods

Steers (n = 6) of primarily Angus (80 to 100%) genetics were harvested at approximately 18.5 mo and 630 kg live weight. Samples from the *longissimus lumborum* (LL) and *diaphragm* (Dia) were collected at 1, 3, and 24h postmortem. Fresh-preserved muscle samples were permeabilized using saponin, and muscle bundles (2–4 mg) were transferred to a high-resolution oxygraph for respiration measurements (oxygen consumption rate, OCR, pmol/sec/mg of tissue). Samples were assessed in duplicate under hyperoxia. First, pyruvate and malate were added to support the TCA cycle and assess leak respiration. Then, ADP was added to support electron flow through complex I. The influence of glutamate on NADH production (complex I) was tested, followed by complex II activation by succinate. Integrity of the mitochondria outer membrane was tested with cytochrome c. Next, an uncoupler (FCCP) was added to force the electron transport system (ETS) to maximum capacity. Citrate synthase (CS) activity (nmol/min/mg tissue) was determined in frozen samples and used as a marker of mitochondria content. Subsequently, respiration data were normalized to CS activity (pmol/sec/U CS) to account for differences in mitochondria content. Coupling efficiency of oxidative phosphorylation was calculated as 1– (Leak/ADP-stimulated oxidative phosphorylation capacity). Raw and normalized OCR were analyzed in a randomized block design, with slaughter date as block and fixed effects of muscle, time, and the interaction. Time was considered a repeated measure.

Results

Muscle type affected (P = 0.0002) leak OCR, with Dia showing a higher rate than LL. After ADP was added, mitochondria from Dia exhibited higher OCR at all times tested and at all steps, with OCR being 4 times higher after FCCP addition. Mitochondrial content, evidenced by greater (P < 0.0001) CS activity in Dia, largely explained differences in OCR between muscles. After OCR was normalized to CS activity, the 1 and 3h postmortem OCR from Dia and LL were similar (P > 0.05). However, at 24h postmortem, OCR after ADP, glutamate, and FCCP additions were greater (P < 0.05) in Dia mitochondria. Time, but not muscle, affected cytochrome c response. At 1h postmortem, cytochrome c increased OCR by 6.6%, supporting that mitochondria outer membrane integrity is not compromised. However, cytochrome c response at 3h postmortem increased 52.4%, indicating outer membrane damage. Coupling efficiency is different between muscles (P = 0.005) with Dia exhibiting greater efficiency.

Conclusion

Despite inherent metabolic differences between the LL and Dia, mitochondria from both muscles are intact and coupled at 1h postmortem. However, by 24h postmortem, functional properties of LL mitochondria are reduced compared to Dia. Declining mitochondrial function may be associated with calcium overload, mitochondrial fragmentation, and protease activation.