Skeletal Muscle Fiber-Type Specific Succinate Dehydrogenase Activity Is Not Reduced in Children with Cerebral Palsy

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INTRODUCTION

• Children with cerebral palsy (CP) display increased energy expenditure and decreased physical activity 1,2,3 compared to children with typical development (TD).
• Mitochondrial oxidative metabolism in skeletal muscle is critical during sustained movement. While recent studies report a reduced expression at the mRNA level of metabolism-related proteins, including SDH in children with CP,4,5 no direct measurements of oxidative metabolism have been reported.
• Succinate dehydrogenase (SDH) is a mitochondrial enzyme of the tricarboxylic acid (TCA) cycle and complex II of the electron transport chain. Oxidative capacity varies by fiber type (type1-type2A-type2X) and SDH activity is commonly analyzed as a marker of intrinsic oxidative capacity in skeletal muscle.
• The goal of this study was to directly quantify fiber-type specific SDH activity to evaluate if there are differences between children with CP and TD.

METHODS

• Semitendinosus muscle biopsies were obtained from ten children (5/group, CP mean age=12.2±5.1, 3 male, TD mean age=15±0.7, 4 male)
• Ethical approval provided by UCSD Human Research Protection Program and all subjects & parents consented.
• 10µm cross-sections from flash frozen biopsies using a cryostat at -25°C
• SDH activity (optical density (OD)/min) in cross-sections was measured histochemically as a marker of oxidative activity (Figure 1).
• Images were captured in real-time every 60 seconds for 10 minutes and optical density was calculated for muscle fibers at each time point (Figure 2).
• Type 1 and Type 2A MHC isoforms and the basal lamina were identified by immunohistochemistry to determine fiber-type specific SDH activity (Figure 1,2).
• Myofiber area was measured based on immunolabeling of the basal lamina using Image J in all fibers in which SDH activity was measured (Figure 1).

RESULTS

• Average myofiber areas were 45% smaller in CP vs. TD (2713±2567 µm² vs. 4975±1579 µm²; p<0.05, Figure 3). In children with CP, type 2A and type 1 fiber sizes were not significantly different (2887±3070 µm² vs. 2540±2308 µm²; p=0.37), whereas, in TD children, type 2A fibers were 32% larger than type 1 fibers (5666±1626 µm² vs. 4284±1330 µm²; p<0.05, Figure 3).
• SDH activity per unit area was ~35% lower in Type 2A compared to Type 1 fibers, but no differences were seen between children with CP and TD (Figure 4).

CONCLUSIONS

• SDH activity in Type 1- and 2A skeletal muscle fibers is maintained in children with CP and Type 1 fibers have a higher activity compared to Type 2A fibers.
• Type 1 and type 2A fibers in children with CP are nearly uniform in size, and the average fiber size is decreased in children with CP.
• Further research that focuses on the functional capacity of mitochondria and other components of the electron transport chain in skeletal muscle of children with CP is needed.

ACKNOWLEDGEMENTS

This work was supported by the Department of Veterans Affairs and NIH grants AR061303-01, HD08501, HD08537 and HD44922 to RLL.