Examining the Impact of Nitrate and Nitrite Treatment on Fatty Acid Oxidation in C2C12 Mouse Muscle Cells

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Background

- **What is nitrate and nitrite**
  - Inorganic molecule
  - Nitric Oxide and Cardiovascular Health

- **Where do people get it?**
  - Leafy green vegetables (Spinach, beets)
  - Cured meats
  - Exercise Supplements

- **Why do people care about it?**
  - Cancer
  - Nitrate consumption makes exercise more efficient in sedentary people
    - Decreased oxygen consumption during exercise
    - Increase endurance
Exercise and Nitrate

- Nitrate decreased oxygen cost of exercise but nitrite increased oxygen cost of exercise. How is this occurring?

- No differences between nitrate and nitrite treatment in mitochondrial abundance or efficiency.
Hypothesis

- Fatty acid oxidation is the utilization of fats to produce energy
- Acyl Carnitines
- Metabolomics found differential amounts of fats and acyl carnitines suggesting difference in fatty acid oxidation that may be driving the differences in oxygen consumption

- Test the hypothesis that nitrate and nitrite differentially affect fatty acid oxidation
Method

- Used differentiated C2C12 mouse muscle cells to test the hypothesis
- Treated with increasing doses of nitrate or nitrite for 3 days
- Measured fatty acid oxidation using a radio-labeled fat (palmitate)
Healthy Differentiated Muscle Cells

Control

Nitrate 50 µM

Nitrite 500 nM

Nitrate 500 µM

Nitrite 1.5 µM
Nitrate and Nitrate did not Affect Uptake of Fat in Muscle

- Palmitate in media was decreased when cells were present
- Muscle cells took up the palmitate equally
Nitrate and Nitrite Did Not Change Fatty Acid Oxidation

End Product of Fatty Acid Oxidation is CO₂
Conclusions

- No detectable difference in fatty acid oxidation with nitrate or nitrite treatment in C2C12 cells

- What does this mean
  - Still need to do more research
  - Tissue culture compared to whole organism
  - Alternate hypothesis
    - Differential usage of Type I and Type II muscles
Current Work
- Other potential mechanisms to be interrogated in the lab
  - Changes in hormones related to a stressed state
  - Changes in gene expression that drive differential utilization of metabolic fuels

So What?
- Nitrate has been proven to lower the oxygen cost of exercise in sedentary people
- You can get it from small spinach salad, a couple of beets, beet juice, dietary supplements
- This may also have protective effects for cardiovascular health
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