Intestine-specific Loss of Cpt2, Required for Long-chain Fatty Acid Oxidation, Alters Systemic Energy Metabolism and Glucose Homeostasis

Mitchell Lavarias
Intestine Has a Critical Role in Systemic Metabolism

Diet

Brain

Liver

Intestine

Pancreas

Insulin

Muscle

Glucose

Amino Acids

Lipids
Fasting Induces Gene Expression of Intestinal Cpt2, which is Necessary for Fatty Acid Oxidation
Ketogenic Feeding Induces Intestinal Cpt2 Gene Expression

Intestine Cpt2 Expression

Relative Expression

Chow Fed
Chow Fasted
Ketogenic

0 2 4 6 8 10

a
b

b
Carnitine Palmitoyltransferase 2 (CPT2) in Long-chain Fatty Acid Oxidation
Intestinal Cpt2 is Necessary for Long-chain Fatty Acid Oxidation

Data collected in collaboration with Rashpal Dhillon
Cpt2\(^{\text{IKO}}\) Mice have Lower Glucose and Increased Triglycerides upon Fasting
Fasting and Cpt2 Deficiency Alter Intestinal Carnitine Species Concentration

Intestine:

- **L-Palmitoylcarnitine (C16:0)**
  - Fed: 1
  - Fasted: 4

- **L-Octanoylcarnitine (C8:0)**
  - Fed: 1
  - Fasted: 3

- **L-Carnitine**
  - Fed: 1.5
  - Fasted: 2

Data collected in collaboration with Spencer Haws
Cpt2^IKO^ Mice Maintain Intestinal Acetyl-CoA, but have Reduced Pyruvate and α-Ketoglutarate

Intestine:

Data collected in collaboration with Spencer Haws
Using a Ketogenic Diet to Simulate the Fasted State

Ketogenic Diet

- Fat: 90.5%
- Protein: 9.2%
- Carbs: 0.3%
Loss of Intestinal Cpt2 Reduces Body Weight on a Ketogenic Diet
Loss of Intestinal Cpt2 Reduces Lean Mass on a Ketogenic Diet

- Cpt2^{ff}
- Cpt2^{IKO}

**Body Weight (g)**

Weeks on Ketogenic Diet

- Cpt2^{ff}
- Cpt2^{IKO}

**Body Weight Change (g)**

- Cpt2^{ff}
- Cpt2^{IKO}

**Lean Mass (g)**

Weeks on Ketogenic Diet

- Cpt2^{ff}
- Cpt2^{IKO}

Lean Mass Change (g)

- Cpt2^{ff}
- Cpt2^{IKO}

* indicates statistical significance.
Loss of Intestinal Cpt2 Results in Hypoglycemia and Hyperketonemia on Ketogenic Diet

**Blood Glucose**

<table>
<thead>
<tr>
<th>Weeks on Ketogenic Diet</th>
<th>Cpt2^{ff}</th>
<th>Cpt2^{IKO}</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>150</td>
<td>120</td>
</tr>
<tr>
<td>1</td>
<td>160</td>
<td>110</td>
</tr>
<tr>
<td>2</td>
<td>170</td>
<td>130</td>
</tr>
<tr>
<td>3</td>
<td>180</td>
<td>150</td>
</tr>
<tr>
<td>4</td>
<td>190</td>
<td>170</td>
</tr>
<tr>
<td>5</td>
<td>200</td>
<td>180</td>
</tr>
</tbody>
</table>

**Ketone Bodies**

<table>
<thead>
<tr>
<th>Weeks on Ketogenic Diet</th>
<th>Cpt2^{ff}</th>
<th>Cpt2^{IKO}</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>1</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>2</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>3</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>4</td>
<td>4</td>
<td>5</td>
</tr>
</tbody>
</table>

* denotes significant difference.
Intestinal Cpt2 Protects Mice from Ketogenic Diet Induced Fatty Liver
Loss of Intestinal Cpt2 in Mice Fed a Ketogenic Diet Reduces Liver Amino Acids

Liver:

Data collected in collaboration with Spencer Haws
Loss of Intestinal Cpt2 in Mice Fed a Ketogenic Diet Increases Intestine Amino Acids

Intestine:

Data collected in collaboration with Spencer Haws
Loss of Intestinal Fatty Acid Oxidation in Mice Fed a Ketogenic Reduces Liver, but not Intestinal TCA Cycle Intermediates

Data collected in collaboration with Spencer Haws
Supplementation of Casein and Medium-chain Triglycerides into a Ketogenic Diet

- Ketogenic Diet:
  - Fat: 90.5%
  - Protein: 9.2%
  - Carbs: 0.3%

- Ketogenic Diet Supplemented with Casein:
  - Fat: 81.4%
  - Protein: 18.3%
  - Carbs: 0.3%

- Ketogenic Diet with Medium-chain Triglycerides (MCKeto):
  - LCT: 45.25%
  - MCT: 45.25%
  - Fat: 90.5%
  - Protein: 9.2%
  - Carbs: 0.3%
Supplementation of Casein Protects Against Lean Mass Loss in Cpt2\textsuperscript{IKO} Mice Fed a Ketogenic Diet
Loss in Lean Mass is Specific to Long-Chain Fatty Acid Ketogenic Diet in Cpt2\textsuperscript{IKO} Mice
One Day of Long-chain Ketogenic Feeding is Sufficient to Elicit Changes in Blood Metabolites in Cpt2\(^{\text{IKO}}\) Mice
Intestinal Cpt2 Modulates Systemic Energy Metabolism and Nutrient Availability

- Reduced Lean Mass
- Hepatic Steatosis
- Amino Acids

Ketogenic Diet

- Cpt2\textsuperscript{IKO}
- Fatty Acid Oxidation
- Amino Acids
- Glucose
- Ketone Bodies
Acknowledgements

Yen Lab:
Eric Yen
David Nelson
Mei-I Yen
Madison Ray
Lauren Tancer
Peilin Wang
Lingfeng Hu
Shaza Abdul Rahim

Collaborators:
Michael Wolfgang (Johns Hopkins)

Denu Lab:
• Spencer Haws (UW-Madison)
• Rashpal Dhillon (UW-Madison)

Melanie Gillingham (OHSU)

Funding:
MANTP:
NIH Grant T32
DK007662