The Role of the Carnitine System in Human Metabolism

Daniel W. Foster, M.D.
Department of Internal Medicine
U.T. Southwestern Medical Center
Gluconeogenesis

Diagram:
- Kidney:
  - Glucose
- Muscle:
  - Glutamine
  - Glucose
  - Amino Acids
  - $\text{CO}_2 + \text{HOH}$
- Liver:
  - Glucose
  - Gluconeogenesis
Ketone Formation
FIGURE 2. The effects of fasting and refeeding on the conversion of oleic acid to ketone bodies in the isolated perfused liver. Rats were fasted for the indicated time and the livers were perfused for 1 h with 0.7 mM oleic acid. The dotted line represents ketone production after refeeding. Major changes in the production of acetoacetate/β-hydroxybutyrate despite fixed levels of fatty acid indicate activation of fatty acid oxidation and ketogenesis. From ref. 7.
FIGURE 5. Ketone body production in the isolated, perfused liver after treatment of animals with glucagon or anti-insulin serum (AIS). Livers removed from the animals shown in Figure 4 were perfused with 0.7 mM oleic acid. Although glucagon did not cause ketosis in vivo, it activated the ketogenic pathway in liver. See text for details. (Reproduced by permission from McGarry, J. D., Wright, P. H., and Foster, D. W.: Hormonal control of ketogenesis: rapid activation of hepatic ketogenic capacity in fed rats by anti-insulin serum and glucagon. J. Clin. Invest. 1975; 55:1202-1209.)
The mitochondrial CPT system

Malonyl-CoA

(Acetyl-CoA carboxylase)

CPT I

CPT II

Acyl-CoA

Carnitine

CoASH

Acylcarnitine

CoASH

Acyl-CoA

β-oxidation

OUTER MEMBRANE

INNER MEMBRANE

MATRIX

TRANSLOCASE
## CPT System Enzymes

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Chromosomal location</th>
</tr>
</thead>
<tbody>
<tr>
<td>CPT-1A (liver)</td>
<td>11 q 13</td>
</tr>
<tr>
<td>CPT-1B (muscle)</td>
<td>22 q 13.3</td>
</tr>
<tr>
<td>CPT-1C (Brain, testis)*</td>
<td>19 q 13.33</td>
</tr>
<tr>
<td>CPT-2 (same all tissues)</td>
<td>1 p 32</td>
</tr>
<tr>
<td>CACT (same all tissues)</td>
<td>3 p 21.31</td>
</tr>
</tbody>
</table>

*Mouse CPT-1C is found on chromosome 7*
<table>
<thead>
<tr>
<th>Tissue</th>
<th>Malonyl-CoA $I_{50}$ (µM)</th>
<th>Carnitine $K_m$ (µM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rat liver</td>
<td>1.7</td>
<td>36</td>
</tr>
<tr>
<td>Human fetal liver</td>
<td>1.6</td>
<td>39</td>
</tr>
<tr>
<td>Rat heart</td>
<td>0.12</td>
<td>167</td>
</tr>
<tr>
<td>Guinea pig liver</td>
<td>0.10</td>
<td>270</td>
</tr>
<tr>
<td>Human skel. muscle</td>
<td>0.025</td>
<td>480</td>
</tr>
<tr>
<td>Rat skel. muscle</td>
<td>0.02</td>
<td>639</td>
</tr>
<tr>
<td>Dog skel. muscle</td>
<td>0.01</td>
<td>660</td>
</tr>
<tr>
<td>Dog heart</td>
<td>0.01</td>
<td>770</td>
</tr>
<tr>
<td>Species</td>
<td>Tissue</td>
<td>Total carnitine content (μmol/gm wet wt)</td>
</tr>
<tr>
<td>----------</td>
<td>-----------------------</td>
<td>----------------------------------------</td>
</tr>
<tr>
<td>Rat*</td>
<td>Liver</td>
<td>0.12 ± 0.01</td>
</tr>
<tr>
<td></td>
<td>Kidney cortex</td>
<td>0.23 ± 0.01</td>
</tr>
<tr>
<td></td>
<td>Gastrocnemius muscle</td>
<td>0.46 ± 0.02</td>
</tr>
<tr>
<td></td>
<td>Heart</td>
<td>0.57 ± 0.04</td>
</tr>
<tr>
<td>Dog†</td>
<td>Quadriceps muscle</td>
<td>2.6</td>
</tr>
<tr>
<td>Human†</td>
<td>Gluteus muscle</td>
<td>3.1</td>
</tr>
<tr>
<td></td>
<td>Erector spinae muscle</td>
<td>3.6</td>
</tr>
<tr>
<td></td>
<td>Quadriceps muscle (a)</td>
<td>2.4</td>
</tr>
<tr>
<td></td>
<td>Quadriceps muscle (b)</td>
<td>2.6</td>
</tr>
<tr>
<td></td>
<td>Pectoralis muscle</td>
<td>2.6</td>
</tr>
</tbody>
</table>

* Values are means ± SEM for six animals.
† Values are from a single determination in different subjects.
FATTY ACID SYNTHESIS AND OXIDATION IN LIVER

Glucose → Glycerol-P → Fatty acyl-CoA → Triglyceride → VLDL → Plasma FFA

Pyruvate → Acetyl-CoA → Citrate → Ketone bodies

Carnitine → Fatty acid → Malonyl-CoA → ACC → Acetyl-CoA

FAS → VLDL
Triglyceride synthesis

Fed

Fatty acyl CoA

β-oxidation

Fasted
(+) Decanoylcarnitine (DC) and Fat Metabolism in Perfused Rat Liver

1 – $^{14}$C oleic acid metabolism (30 minutes) % recovered

<table>
<thead>
<tr>
<th></th>
<th>Ketones</th>
<th>Liver Lipids</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>1.4</td>
<td>31.4</td>
<td>97.5</td>
</tr>
<tr>
<td>Fasted*</td>
<td>15.6</td>
<td>14.2</td>
<td>89.5</td>
</tr>
<tr>
<td>Fasted (+DC)*</td>
<td>0.9</td>
<td>27.9</td>
<td>92.5</td>
</tr>
</tbody>
</table>

*Fasted 24 hours
MALONYL CoA REGULATION

acetyl CoA

acetyl CoA carboxylase

Malonyl CoA

fatty acid oxidation

( - )

( + )

fatty acid synthesis

malonyl CoA decarboxylase

acetyl CoA
AMPK AND ACTIVATION OF FATTY ACID OXIDATION

Glucagon → AMPKK → Fatty acyl-CoA

AMPK (inactive) → AMPK-P (active)

Acetyl-CoA → Malonyl-CoA

ACC (active) → ACC-P (inactive)

Fatty acyl-CoA → Fatty acyl-carnitine

Liver → Ketoacids

β-oxidation

CPT I

Non-hepatic tissues → CO₂ + HOH

CPT II

FADS
OTHER ACTIVATORS OF FATTY ACID $\beta$-OXIDATION

(1) Peroxisome proliferator-activated receptor-\(\gamma\) coactivator 1-\(\alpha\)

(2) Stearoyl-CoA desaturase
PEROXISOME PROLIFERATOR-ACTIVATED RECEPTOR-γ COACTIVATOR 1 α (PGC-1α)

- Glucagon
- Cyclic AMP
- A kinase
- CREB

PGC-1α synthesis and activation

- Mitochondrial biogenesis
- Activation of fatty acid β-oxidation
- Activation of gluconeogenesis
- Heart development
FIGURE 16. The regulation of glycolysis and gluconeogenesis by fructose-2,6-bisphosphate (F-2,6-P₂). F-2,6-P₂ activates phosphofructokinase, sustaining glycolysis, and deactivates fructose-1,6-bisphosphatase, inhibiting gluconeogenesis. See text for details. PEP stands for phosphoenolpyruvate.
STEAROYL-CoA DESATURASE (SCD-1) DEFICIENCY

- Leptin high fat diet decreased FA and TG synthesis
- Weight loss increased food intake
- Uncoupled mitochondria (?)
- Genetic (asebia mice)

C16:0 SCD-1 C16:1
C18:0 SCD-1 C18:1
OTHER REGULATORS OF LIPOGENESIS

1. Insulin-induced gene 1 and 2 (Insig 1 and 2)

2. Carbohydrate-responsive element binding protein (ChREBP)
Fig. 2. Effect of carnitine on ketogenesis from oleic acid in perfused livers from fed rats. Livers were perfused with non-circulating medium containing 0.7 mM oleic acid and the output of acetocacetate and β-hydroxybutyrate was determined every 5 min. The symbols used in panels A and B are as follows: (○), livers from fed animals; (×), livers from starved animals; (△), livers from fed animals in which L-carnitine was infused at a concentration of 0.5 mM from the 15-min time point. Values represent means ± SEM for the number of livers shown in parentheses.
Figure 1. Relationship between the concentration of carnitine and the rate of olate oxidation in homogenates of rat tissues. The indicated quantities of tissue were incubated as described under “Methods.” Values are means ± SEM for three experiments with each tissue.
<table>
<thead>
<tr>
<th>Treatment</th>
<th>Ketone production from oleate, μmol/100 g body wt per 30 min</th>
<th>Free carnitine, nmol/g wet wt of liver</th>
<th>Total carnitine, nmol/g wet wt of liver</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fed (8)</td>
<td>26 ± 3</td>
<td>40 ± 5</td>
<td>102 ± 10</td>
</tr>
<tr>
<td>Fed, glucagon 3 hr (6)</td>
<td>87 ± 5</td>
<td>68 ± 8</td>
<td>220 ± 13</td>
</tr>
<tr>
<td>Fasted (6)</td>
<td>118 ± 8</td>
<td>70 ± 5</td>
<td>228 ± 13</td>
</tr>
<tr>
<td>Alloxan diabetic (6)</td>
<td>192 ± 10</td>
<td>172 ± 12</td>
<td>416 ± 6</td>
</tr>
</tbody>
</table>
FUNDAMENTAL THESSES

1. If too little fat is oxidized, life is threatened.

2. If too much fat is oxidized, life is threatened.
SYSTEMIC CARNITINE DEFICIENCY

1. Fasting hypoglycemia
2. No or limited fasting ketosis
3. Elevated plasma NH$_3$
4. Hepatic encephalopathy with “flap” and seizures
5. Multiorgan triglyceride storage
6. Muscle weakness and rhabdomyolysis
7. Progressive cardiomyopathy
8. Carnitine low in plasma and tissues
9. Gene defect: mutated carnitine transporter (OCTN$_2$)
CEREBRAL THROMBOSIS IN DKA

W.G., a 21 y/o BM with known insulin-dependent diabetes mellitus, was admitted in diabetic ketoacidosis. Admission hemoglobin 20.3 g/dl, hematocrit 60.8%, WBC 21,200. Ethanol negative. Trace salicylate. Lactate 2.0 mM, amylase 182 (nl <110), lipase 798 (nl <208), pH 7.15, pO₂ 99 mm.
CEREBRAL THROMBOSIS IN DKA

<table>
<thead>
<tr>
<th>Time</th>
<th>Glucose</th>
<th>Na</th>
<th>K</th>
<th>HCO₃</th>
<th>Cl</th>
<th>Creat</th>
<th>Gap</th>
<th>State</th>
</tr>
</thead>
<tbody>
<tr>
<td>2130</td>
<td>1457</td>
<td>140</td>
<td>6.5</td>
<td>8</td>
<td>96</td>
<td>6.2</td>
<td>36</td>
<td>Drowsy</td>
</tr>
<tr>
<td>0130</td>
<td>554</td>
<td>158</td>
<td>2.9</td>
<td>17</td>
<td>128</td>
<td>4.1</td>
<td>13</td>
<td>Drowsy</td>
</tr>
<tr>
<td>0800</td>
<td>295</td>
<td>145</td>
<td>3.9</td>
<td>18</td>
<td>115</td>
<td>2.6</td>
<td>12</td>
<td>Drowsy</td>
</tr>
</tbody>
</table>

1130 Unresponsive. Right facial paralysis, right hemiparesis. Head CT negative. Spinal tap unremarkable.

Next day - Left hemiparesis, dilated right pupil. CT-stroke, edema, shift. Died 72 hours after admission.
ACUTE CORONARY OCCLUSION

INCIDENCE OF VT AND VF

PVC FREQUENCY

FAT OXIDATION/CPT SYSTEM

1. Diabetic ketoacidosis
2. Hypoglycemia/Reye syndrome
3. Insulin secretion
4. Insulin resistance/pathogenesis of NIDDM
5. Primary muscle disease
6. Sudden death in coronary artery disease
7. Control of feeding signals in hypothalamus
8. Sperm development and motility
9. Therapy of obesity
METABOLIC FUTURE FOR THE CARNITINE /CPT SYSTEM

1. Treatment of non-alcoholic steatohepatitis
2. Treatment of lipotoxicity in heart
3. Treatment of type 2 diabetes mellitus and insulin resistance
4. Treatment of obesity
Wild-type

ACS Transgenic Untreated

ACS Transgenic Leptin Treated

Heart TG (mg/g)

<table>
<thead>
<tr>
<th></th>
<th>Wild-type</th>
<th>ACS Transgenic Untreated</th>
<th>ACS Transgenic Leptin Treated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart TG (mg/g)</td>
<td>3.0</td>
<td>1.5</td>
<td>0.0</td>
</tr>
</tbody>
</table>