BRAIN BIOENERGETICS AND OXIDATIVE STRESS IN SEPSIS

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Neuropsychological effects of Critical Illness

Acute brain dysfunction

Cognitive (delirium)
Autonomic
Immune

Long-term consequences

Cognitive decline
Psychological impairment
Long term cognitive impairment after critical illness

- **Association Between Acute Care and Critical Illness Hospitalization and Cognitive Function in Older Adults** - Ehlenbach, W. J. et al. JAMA 2010;303:763-770
  - 2.3x likelihood of cognitive decline after critical illness hospitalization compared with those who had no hospitalization

- **Long-term cognitive impairment and functional disability among survivors of severe sepsis**. Iwashyna, TJ et al. JAMA. 2010;304(16):1787-1794
  - Severe sepsis in this older population was independently associated with substantial and persistent new cognitive impairment and functional disability among survivors.
Infection - Systemic inflammation

“Cytokines storm”

BBB dysfunction

Neuroinflammation → Mitochondrial dysfunction

Bioenergetic Imbalance

Neuronal injury

Delirium, Dementia and long-term poor outcome

Old age
Anti-Ch
CNS infection
Dementia
Sepsis
- CLP
- LPS
- Fecal Peritonitis
- Pneumonia

Control
- Sham, vehicle

**Histopathology:**
- Glia activation
- Oxidative stress
- Neuronal injury (NeuN, Fluorojade)

**Neurological and cognitive assessment:**
- Severity score (acute)
- Spatial memory (Open field, Object recognition, Water maze)
- Aversive memory (inhibitory avoidance task)
- Depression (forced swimming, tail suspension)
- Anxiety (elevated maze)

**Biochemistry:**
- Oxidative stress (HNE, NT)
- Apoptosis (TUNEL, caspases)
- Synaptic proteins
Systemic inflammation in experimental sepsis:

Tissue hypoxia, bioenergetic imbalance (↑AMP/ATP ratio)
Cognitive testing after *P. Aeruginosa* pneumonia

Freezing - 13 dias

Freezing - 50 dias
Brain Metabolism

Although the brain represents only 2% of the body weight:

- It receives 15% of the cardiac output,
- 20% of total body oxygen consumption.
- 25% of total body glucose utilization.
Brain Metabolism

• **Glucose oxidation**: provides more than 90% of the energy needed.

• **Oxidation of non-glucose substrates**: ketones/lactate during prolonged fasting; not in everyday life.

• Brain function almost totally dependent on a continuous supply of glucose and oxygen from the arterial circulation.
Astrocyte-neuron lactate shuttle model
Brain Metabolism

• **Glycogen**---stored exclusively in glial cells (astrocytes). Metabolize to lactate that can be taken up and used as fuel by neurons.

• Low content in brain (~3 mmol/kg). Unable to sustain brain metabolism for more than 4 to 5 minutes.
Aerobic metabolism increase the energy efficiency between nutrient oxidation and ATP synthesis.

<table>
<thead>
<tr>
<th>TABLE 19–5</th>
<th>ATP Yield from Complete Oxidation of Glucose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Process</td>
<td>Direct product</td>
</tr>
<tr>
<td>Glycolysis</td>
<td>2 NADH (cytosolic) 2 ATP</td>
</tr>
<tr>
<td>Pyruvate oxidation (two per glucose)</td>
<td>2 NADH (mitochondrial matrix)</td>
</tr>
<tr>
<td>Acetyl-CoA oxidation in citric acid cycle (two per glucose)</td>
<td>6 NADH (mitochondrial matrix) 2 FADH₂ 2 ATP or 2 GTP</td>
</tr>
<tr>
<td>Total yield per glucose</td>
<td></td>
</tr>
</tbody>
</table>

*The number depends on which shuttle system transfers reducing equivalents into the mitochondrion.

Table 19-5
Lehninger Principles of Biochemistry, Fifth Edition
© 2008 W.H. Freeman and Company
Mitochondrial dysfunction

**Functional changes**

Inhibition of Krebs Cycle Enzymes

Disturb on mitochondrial respiratory chain function

Dissipation of Mitochondrial Membrane Potential ($\Delta \Psi_m$)

Electron transport blockade

Reduction of ATP Synthesis
Sepsis induces brain mitochondrial dysfunction

Increased $O_2$ consumption non-associated to ATP synthesis

Decreased PHOSPHOX efficiency

Decreased mitochondrial content

Proton leak and mitochondrial depolarization

**Critical Care Medicine** 2008 Jun;36(6):1925-32.
Sumary of brain mitochondrial dysfunctions during sepsis

↓ Cytochromes content

↑ proton leak

↓ membrane potential

↓ BIOENERGETIC EFFICIENCY (ADP:O)
INCREASE IN 18-FDG UPTAKE IN LPS-INDUCED SEPSIS

Control

LPS 2h

LPS 6h

LPS 24h

BRAIN 18-FDG UPTAKE

* P < 0.05  n=19
Endotoxemia increase the 14C-glucose uptake in brain

Control

LPS 2h

μCi/g

Control  LPS 2h
INCREASED BRAIN GLUCOSE UPTAKE IN EXPERIMENTAL SEPSIS

2-NBDG uptake in brain slices

Hexokinase activity (24h)
High metabolic rate

Mitochondrial Dysfunction

Dependence on glucose uptake

SEPSIS
Limited O₂ and glucose delivery (hypotension, hypoxemia, Hypoglycemia)

Tissue ischemia and organ dysfunction

Delirium
Long-term cognitive impairment
4-HNE staining in the corpus callosum (cc) and around ventricles (v)
Around the central ventricle

control

Sepsis 6h
Mitochondrial Hydrogen Peroxide Generation in Septic Brain Is Reduced

A

B

25 pmols

H$_2$O$_2$

2.5 min.

FCCP 5 μM

oligo 1 μg/mL

ADP 1 mM

suc 10 mM

sham

CLP

0

10

20

H$_2$O$_2$ (pmols/min/mg ptn)

basal

suc 10 mM

ADP 1 mM

Oligo 1 μg/mL

FCCP 5 μM

sham

CLP

*
Metabolic coupling between glucose and superoxide production:

Apocynin treatment prevents oxidative stress and astrogliosis in the hippocampus early after sepsis.

Oxidative stress in the hippocampus is associated with Nox2 expression after sepsis induction.

The role of Nox2-derived ROS in the development of cognitive impairment after sepsis

Apocynin treatment prevents cognitive impairment after sepsis
WT treated with Apocynin

CD11b

GFAP

NOX2 deficient (gp91^--^--)

CD11b

GFAP

Hernandes et al. Journal of Neuroinflammation 2014, 11:36
http://www.jneuroinflammation.com/content/11/1/36
THERAPEUTIC TARGETS IN SEPTIC ENCEPHALOPATHY:

APOCYNIN

AMPK  $\uparrow$  AMP

ATP

BBB damage

Microglia activation

Neuronal damage

oxidative stress

$O_2^-$

$H_2O_2$

$ONOO^-$

Glutamate $Ca^{2+}$

NMDA-R

NO

$PI3K/p38$

iNOS

NfkB

ppp

glucose

NADPH + $O_2$

glucose

glycolysis

mitochondrial dysfunctions

swelling, mPTP, proton leak, uncoupling

inflammation

bioenergetic failure

apoptosis

low NAD

cytokines
Concluding remarks:

- Sepsis causes brain tissue hypoxia and bioenergetic imbalance
- There is mitochondrial dysfunction and compensatory increase in glucose uptake
- Deregulation in metabolic adaptation pathways lead to oxidative stress and organ dysfunction
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AMPK and NO signaling to brain metabolic adaptation in sepsis:

**AMPK and NO signaling**

- **AMPK** (40 KDa)
- **iNOS** (130 KDa)
- **Cyclophilin** (18 KDa)

**Graphs**

- **AMPK** expression over time (0h, 6h, 24h, 3 days)
- **iNOS** expression over time (0h, 6h, 24h, 3 days)

**Diagrams**

- **NO**
- **OXIDATIVE STRESS**
- **ROS**
- **AMPK**
- **AMP/ATP**
- **Glucose uptake**
- **ATP**

**Legend**

- **Load control**