Non-genetic, Exposure-related Mitochondrial Disease ("Mitochondrial Toxicity")

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Mitochondrial Disease

• Multi-organ clinical presentation
• Metabolic phenotype
• Progressive

• Etiology
  – Genetic
  – Somatogenic/acquired (toxicity)
Mitochondrial Disease

- Etiology
  1. Genetic
     - mutations to nuclear (nDNA) &/or mitochondrial DNA (mtDNA)
     - Heritable – X-chromosome
Mitochondrial Electron Transport Chain

<table>
<thead>
<tr>
<th>Complex</th>
<th>nDNA</th>
<th>mtDNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>35</td>
<td>7</td>
</tr>
<tr>
<td>II</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>III</td>
<td>10</td>
<td>1</td>
</tr>
<tr>
<td>IV</td>
<td>10</td>
<td>3</td>
</tr>
<tr>
<td>V</td>
<td>12</td>
<td>2</td>
</tr>
</tbody>
</table>

Genetic Mitochondria Disease Foundations
Mitochondrial Disease

• Etiology

  2. Somatogenic/acquired
     “Mitochondrial Toxicity”

• Pharmaceutical

• Environmental

• Occupational
Mito Action Broadcasts
(*drug-induced mitochondrial disease*)

- May 1, 2009
  Dr. James Dykens, Pfizer Pharmaceuticals
  “Drug Toxicity and Pharmaceuticals”

- June 4, 2010
  Dr. Katherine Sims from Massachusetts General Hospital
  “Mitochondrial Toxicity”
Multiple Targets for Drug-induced Mitochondrial Toxicity

(Wallace, K.B. Curr Med Chem. 2015 May 13)
Chemical Targeting to Mitochondria

Drugs & chemicals are not distributed equally throughout the cell

- Cardiolipin
  ![Cardiolipin](http://figshare.com/articles/_Structure_of_Cardiolipin_/658399)

- Electrical membrane potential
Mitochondrial Electron Transport Chain

Complex: I
- NADH dehydrogenase
- Intermembrane space
- Inner mitochondrial membrane
- Mitochondrial matrix
- e-
- H+
- H2O
- ADP
- ATP
- NADH
- NAD+

Transmembrane potentials:
- ~ 1 pH unit alkaline matrix
- ~ 180 mV negative matrix
Positively charged chemicals are drawn from the cell contents into and accumulate in mitochondria
Pharmaceutical Uncouplers of Mitochondrial Respiration:

Amphetamines, doxorubicin, Flufenamic acid, Diflunisal, Tolfenamic acid, Mefenamic acid, Diclofenac, Indomethacin, Naproxen, Nimuesulide
Chemical Inhibitors of Mitochondrial Electron Transport

- Rotenone
- Menadione
- Benzoquinone
- Cyanide
  - CO
  - H₂S

Chemical Uncouplers of Mitochondrial Respiration
- Pentachlorophenol
- Heavy metals: Pb²⁺, As²⁺, Cd²⁺, Hg²⁺
- Paraquat
- perfluorosulfonamide
The Bigger Picture of “Mitochondrial Toxicity”

- Substrate delivery
- Molecular organization
Aspirin, Ibuprofen, Valproic acid
Chemical-induced Mitochondrionopathies

**Metabolism**
- Glucose → Pyruvate
  - Iodoacetate
  - Dichloroacetate
  - Lactic acid
- Fatty acids → β oxidation
- Acetyl CoA → TCA Cycle
  - NAD⁺ → NADH
- NAD⁺ → NADH
- Fluoroacetate, Fluoroacetamide
  - Valproic Acid, Salicylates
- Oxidative phosphorylation
  - ADP → ATP
  - [H⁺]
  - Phenols, Amides, Amphiphilic Cations, Quinones

**Genetic Mechanisms**
- mtDNA replication
- Nucleoside analogues
- Antibiotics

**Protein Translocation**
- Heat Shock Proteins (HSPs)
- Protease inhibitors

**Summary Update**
Molecular Targets for Mitochondrial Toxicity

From the mitochondrial or nuclear genome to protein incorporated into the mitochondrial structure

- Gene translation
- Protein synthesis
- Protein translocation and assembly
Chemical-induced Mitochondrionopathies

**METABOLISM**

- Glucose → Pyruvate → Acetyl CoA → TCA Cycle
- NAD⁺ → NADH
- β oxidation

**OXIDATIVE PHOSPHORYLATION**

- Fatty acids → β oxidation → NAD⁺ → NADH → ATP

**GENETIC MECHANISMS**

- mtDNA replication
- Transcription
- Translation

**PROTEASE INHIBITORS**

**NUCLEOSIDE ANALOGUES**

**ANTIBIOTICS**

**PHENOLS, AMIDES, AMPHIPHILIC CATIONS, QUINONES**

**PROTEIN TRANSLOCALIZATION**

- HSPs
- INHIBITORS
nDNA mutations
Cytosolic ribosomes
Precursor processing

Depolarize membrane potential
Protease inhibitors
Chemical-induced Mitochondrionopathies

**METABOLISM**

- Glucose → IODOACETATE
- Pyruvate → DICHLOORACETATE
- LACTIC ACID
- NADH
- NAD+

**OXIDATIVE PHOSPHORYLATION**

- Fatty acids
- β oxidation
- Acetyl CoA
- NAD+
- NADH

**TCA Cycle**

- VALPROIC ACID
- SALICYLATES
- FLUOROACETATE
- FLUOROACETAMIDE
- ROTENONE
- MALONYL CoA
- ANTIMYCIN A
- CYANIDE
- PHENOLS, AMIDES
- AMPHIPHILIC CATIONS
- QUINONES

**GENETIC MECHANISMS**

- mtDNA
- transcription
- replication
- NUCLEOSIDE ANALOGUES
- PROTEASE INHIBITORS
- PROTEIN TRANSLOCALIZATION

**ANTIBIOTICS**

- translation
- [H+]
- ADP
- ATP
- HSPs

**NAD+**
mtDNA replication and translation
HOW NRTIs WORK

1. HIV REVERSE TRANSCRIPTASE
   The HIV reverse transcriptase enzyme uses the HIV RNA chain as a template to synthesize a DNA copy using nucleotides in the host T-cell.

2. NRTIs
   NRTIs are small molecule drugs that are very similar to the host cell nucleotides, and reverse transcriptase incorporates them into the new HIV DNA chain as if they were the endogenous nucleotides.

   Natural state nucleotide
   - Phosphate groups (two of them are released on insertion)
   - OH group (Point of insertion for next nucleotide)

   NRTI
   - OH group is missing in the NRTI, so the next nucleotide can’t be inserted

3. DNA CHAIN TERMINATION
   The difference between NRTIs and the endogenous nucleotides is that the NRTIs do not possess the chemical group necessary to allow for continued synthesis of the DNA chain. Consequently, once the NRTI is inserted into the DNA chain it is impossible for the reverse transcriptase to add any further nucleotides, resulting in termination of the DNA chain and interruption of the HIV replication process.

https://www.flickr.com/photos/5winfographics/9034866105
Examples of NRTIs

- Racemic 2′,3′-dideoxythymidine (3′-dDTP, 3′-dDTPC)
- AYX-754 [3′-dDTPC]
- SFD-754
- Aminoribavirine
- DPC-817 [3′-dDTPC]
- Dextroribavirine
- Reverset
- ACH-126443 [β-L-Fd4C]
- Eluvorabine
- MVV-310 [FdC(T)4d, FLT]
- Alvudidine
- Diaminopurine
- dioxolane (DAPD)
- Amlofoxarin
- Etravirine [TMC125, R165535]
- Rilpivirine [TMC278, R778474]
- Dapivirine [TMC115, R147581]
Mitochondrial Protein Synthesis Inhibitors

Antibiotics
Chloramphenicol
Tetracycline
Linezolid

http://www.hvatinn.is/frodeiksmolar/hvad-er-rna/
OXIDATIVE PHOSPHORYLATION

**METABOLISM**

- **Glucose** → **Pyruvate** → **LACTIC ACID**
  - **IODOACETATE** → **NADH**
  - **DICHLOROACETATE** → **NADH**

**TCA Cycle**

- **Acetyl CoA** → **β oxidation**
  - **NADH** → **NAD+**

**OXIDATIVE PHOSPHORYLATION**

- **Fatty acids** → **NADH** → **ATP**
  - **[H+]**
  - **[H+]**
  - **MALONYL CoA**
  - **ANTIMYCIN A**
  - **CYANIDE**

**GENETIC MECHANISMS**

- **mtDNA** → **transcription**
  - **NUCLEOSIDE ANALOGUES**
  - **ANTIBIOTICS**
  - **translation**

**PROTEIN TRANSLOCALIZATION**

- **HSPs**
  - **PROTEASE INHIBITORS**
  - **ANTIMYCIN A**

**FAEY acids**

- **FLUOROACETATE**
  - **FLUOROACETAMIDE**
  - **VALPROIC ACID**
  - **SALICYLATES**
  - **ROTENONE**
  - **MALONYL CoA**

**PHENOLS, AMIDES**

- **AMPHIPHILIC CATIONS**
  - **QUINONES**

**H+**
Other “uncharacterized” Environmental Mitochondrial Toxicities

- Cigarette smoke
- Air pollution, particulates
- Poly aromatic hydrocarbons (PAHs)
- Herbicides 2,4-dichlorophenoxyacetic acid (2,4-D), dinoseb
- mtDNA genotoxicants, mutagens
- etc.
Reports of demonstrated mitochondrial toxicity represent a “hazard” that may or may not be a “real” risk under normal or intended exposure conditions.

“hazard” versus “risk”
Extended Readings

Mitochondrial Disease v. Mitochondrial Toxicity

Genetic v. Environmental

Thank you