Protein Import & Mitochondrial Assembly

- Partially folded polypeptide
- Positively charged Amphipathic α-helix
- Cleavage of presequence by matrix processing peptidase (MPP)
Targeting of inner membrane proteins

• Single or multi-pass, pre-sequences or multiple internal signals

• They are recognized by mobile chaperones in the inter-membrane space which transfer the protein to a Tim complex

• Mitochondrial genome: inserted via Oxa translocase
Targeting of outer membrane proteins

- Tom complex inserts proteins with $\alpha$-helical transmembrane domains
- SAM complex inserts $\beta$-barrel proteins such as porins
Mitochondrial phospholipids

• PC & PE are synthesized in ER & carried by phospholipid transfer proteins (PS is synthesized from PE)

• The unusual phospholipid, cardiolipin, which contains four fatty acid chains is also synthesized in the mitochondria

• This molecule improves the efficiency of oxidative phosphorylation by restricting proton flow across the membrane
Mitochondrial diseases
General information

• A fertilized human egg carries 2000 copies of the human mitochondrial genome, all but one or two are maternal

• If all of these mitochondrial genomes carried a deleterious mutation would generally not survive

• If a mixed population of both mutant and normal mitochondrial genomes, it will be transmitted to their daughters and sons

• In cases of mitochondrial defects, muscle and nervous tissues are most at risk

• Mitochondria diseases can be classified according to their cause: genetic or biochemical
The biochemical classification of mitochondrial diseases
Steps in Metabolism:
1. Transport of substrates
2. Substrate utilization
3. Krebs cycle
4. Respiratory chain
5. Oxidative phosphorylation
Defects of mitochondrial transport

- interfere with the movement of molecules across the inner mitochondrial membrane, which is tightly regulated by specific translocation systems
Substrate utilization

• Pyruvate dehydrogenase (PDH) deficiency can cause alterations of pyruvate metabolism

• The PDH complex (PDHC) catalyzes the irreversible conversion of pyruvate to acetyl-CoA

• The most devastating phenotype of PDH deficiency presents in the newborn period

• The majority of patients are male with severe metabolic acidosis, elevated lactate in blood or CSF, and associated elevations of pyruvate and alanine
Defects of the Krebs cycle

- Fumarase deficiency is reported with patients having mitochondrial encephalomyopathy
- The enzyme defect has been found in muscle and liver
- Features: excretion of large amounts of fumaric acid and, to a lesser extent, succinic acid in the urine
Abnormalities of the respiratory chain reaction

• Defect in any of the 4 electron chain complexes have been reported
Defects of oxidation-phosphorylation coupling

- The best known example of such a defect is Luft's disease, or nonthyroidal hypermetabolism.
- **Respiratory rate** is at maximal rate even in the absence of ADP, an indication that respiratory control is lost.
- Respiration proceeds at a high rate independently of phosphorylation, and energy is lost as heat, causing hypermetabolism and hyperthermia.
The genetic classification of mitochondrial diseases
Defects of mitochondrial DNA (mtDNA)

• These disorders are associated with dysfunction of the respiratory chain because all 13 subunits encoded by mtDNA are subunits of respiratory chain complexes

• Diseases due to point mutations are transmitted by maternal inheritance
MERRF and others

• One main syndrome is myoclonic epilepsy and ragged red fiber disease (MERRF), which can be caused by a mutation in one of the mitochondrial transfer RNA genes required for synthesis of the mitochondrial proteins responsible for electron transport and production of ATP.

• Other syndromes include
  • Lactic acidosis and stroke-like episodes (MELAS)
  • Leber's hereditary optic neuropathy (LHON)
  • Neurogenic atrophy, ataxia and retinitis pigmentosa (NARP)
Leber's hereditary optic neuropathy (LHON)

- Females (10%) are affected less frequently than males (50%)
- The mutations reduce the efficiency of oxidative phosphorylation and ATP generation

- A rare inherited disease that results in blindness because of degeneration of the optic nerve
- Vision loss is only manifestation, occurs between 15-35
Mutations causing LHON

• 50%: His to Arg (one subunit of complex I)
• 30%: mutations in other subunits or cytochrome b
• A fifth mutation affecting a complex I subunit can cause either LHON or muscular disorders.
• Blindness is the main manifestation
• The low incidence among carriers is because the mixture of mutant and normal mitochondria (heterogeneity)
Defects of nuclear DNA

• The vast majority of mitochondrial proteins are encoded by nuclear DNA.
• All areas of mitochondrial metabolism can be affected.
• The nuclear DNA controls many functions of the mitochondria DNA, including mitochondrial replication.
• Mutations of nuclear genes controlling these functions could cause alterations in the mitochondria DNA.
Peroxisomes
Structural features of peroxisomes

• Small, membrane-enclosed organelles
• They contain enzymes involved in a variety of metabolic reactions, including several aspects of energy metabolism
• They replicate by division
• Most human cells contain 500 peroxisomes
Peroxins

• Chaperones directed for peroxisomes
• Peroxisomal proteins are called
• There are \( \approx 25 \) different peroxins;
• There are 85 genes encoding peroxisomal proteins, most of which are metabolic enzymes
• Internal proteins are synthesized on free ribosomes and then imported into peroxisomes
• Other membrane proteins act as receptors for the import of internal proteins
Function of peroxisomes

• Not all peroxisomes perform the same function

• Oxidation reactions leading to the production of \( \text{H}_2\text{O}_2 \)

• Catalase

• Substrates like uric acid, amino acids, and fatty acids are broken down by oxidative reactions in peroxisomes

• Fatty acids are oxidized in both peroxisomes & mitochondria
Synthesis in peroxisomes

- Cholesterol
- Dolichol
  - made from farnesyl
- Bile acids (liver)
- Plasmalogen
  - important in membranes of heart and brain
• The protein pex3 recruits **pex19** to initiate budding of peroxisome from ER.
  • The new peroxisome fuses with a new or an older one.

• Membrane proteins act as receptors for the import of internal proteins.

• Internal proteins are targeted mostly by peroxisome targeting signal 1 (PTS1) or PTS2 sequences
  • These signals are recognized by cytosolic receptors and proteins are imported via a channel (importomer).
Peroxisome maturation and division

Different proteins are added at different times producing different peroxisomes
Peroxisomal diseases

• Single peroxisomal enzyme deficiencies
  • Defective specific peroxisomal enzymes

• X-linked adrenoleukodystrophy (XALD)
  • Defective transport of very long chain fatty acid (VLCFA) across the peroxisomal membrane
Peroxisomal diseases

• Peroxisomal biogenesis disorders (PBDs)
  • Mutations of PEX genes leading to deficiencies of multiple peroxisomal enzymes
  • Example: Zellweger syndrome
    • Lethal
    • Due to mutations in at least 10 genes such as the receptor of PTS1