# Lecture 8
# Mitochondrion and chloroplast

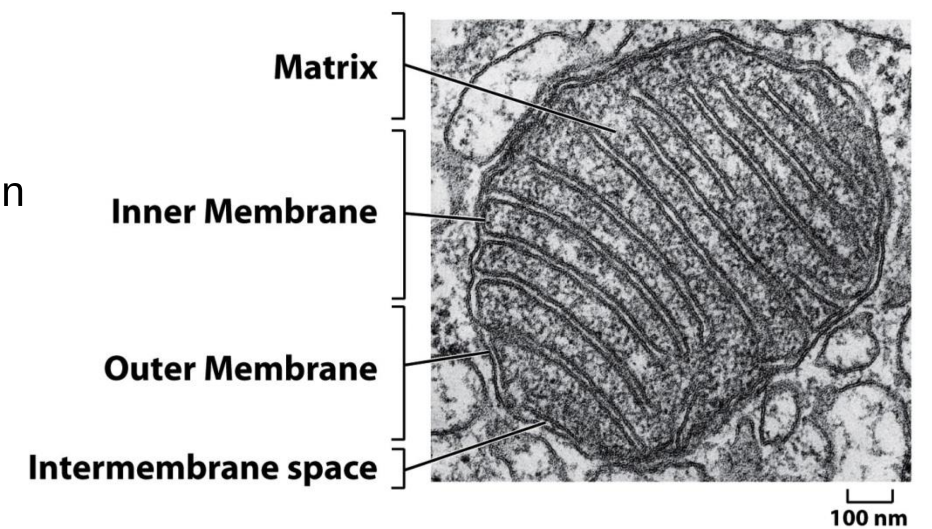
## Outline

I. Mitochondrion structure and its compartment
II. Plastid and chloroplast structure/ function
III. Genetic systems in mitochondria and chloroplasts
IV. Energy conversion---chemiosmotic coupling

# I. mitochondria

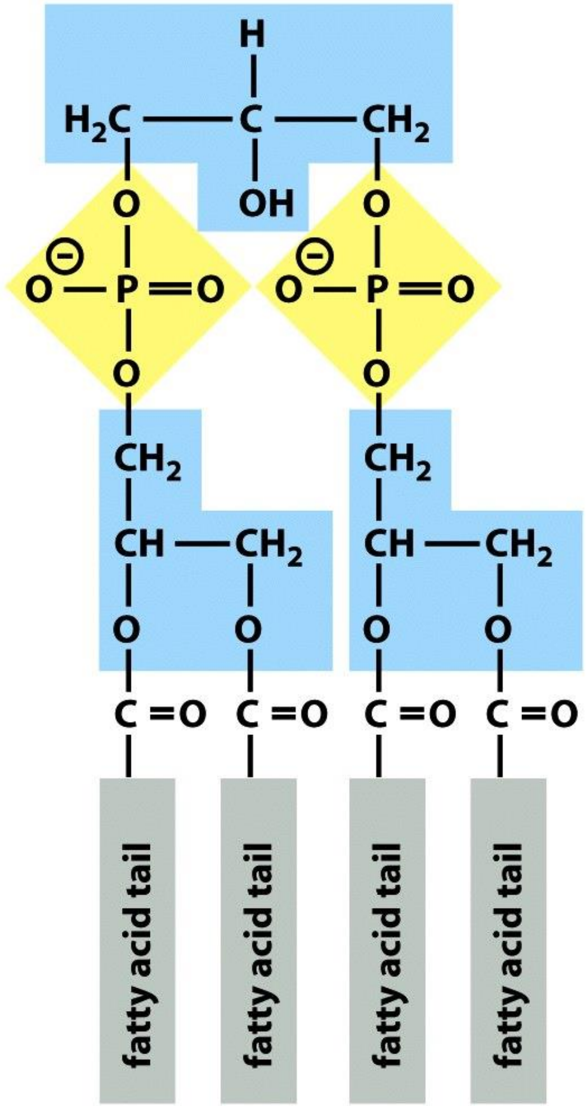
- Outer membrane contains porins and is highly permeable to molecules 5kd or less.
- Inner membrane is highly selective in permeability for molecules and contains the proteins for electron transport proteins and ATP synthase, transporters
- Matrix contains hundreds of enzymes for citric acid cycle, pyruvate oxidation, fatty acid oxidization, mitochondria DNA, tRNA, ribosomes.
- Intermembrane space contains several enzyme to use ATP to phosphorylate other nucleotides

Four compartments in mitochondrion



14.1-mitochondion.mov

# Cardiolipin in inner membrane makes it highly impermeable

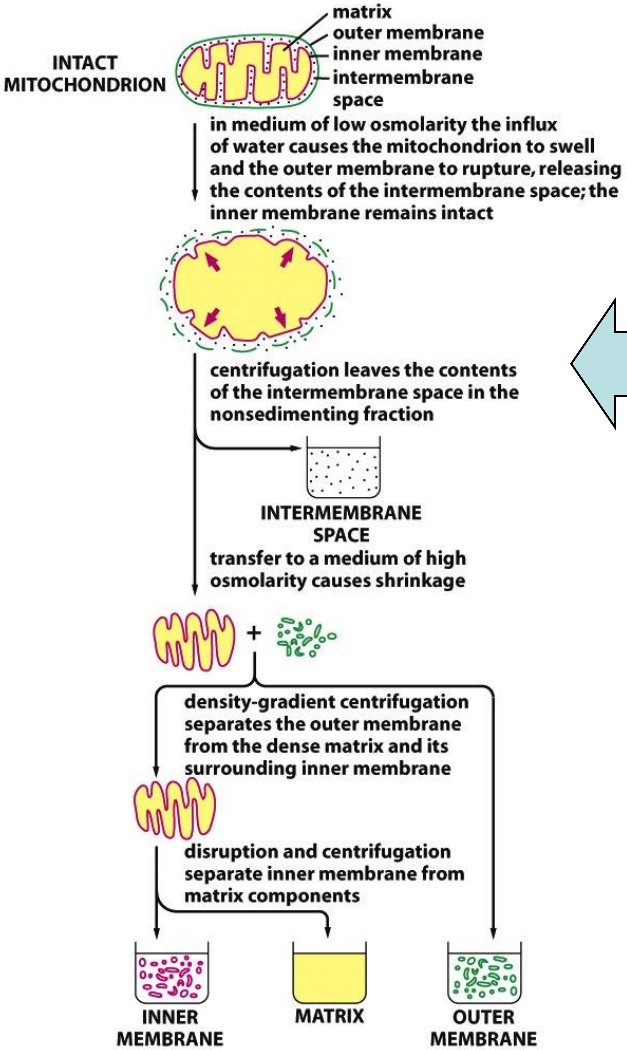



Double phospholipids contains 4 fatty acid chains

It constitutes 20% of total inner membrane.

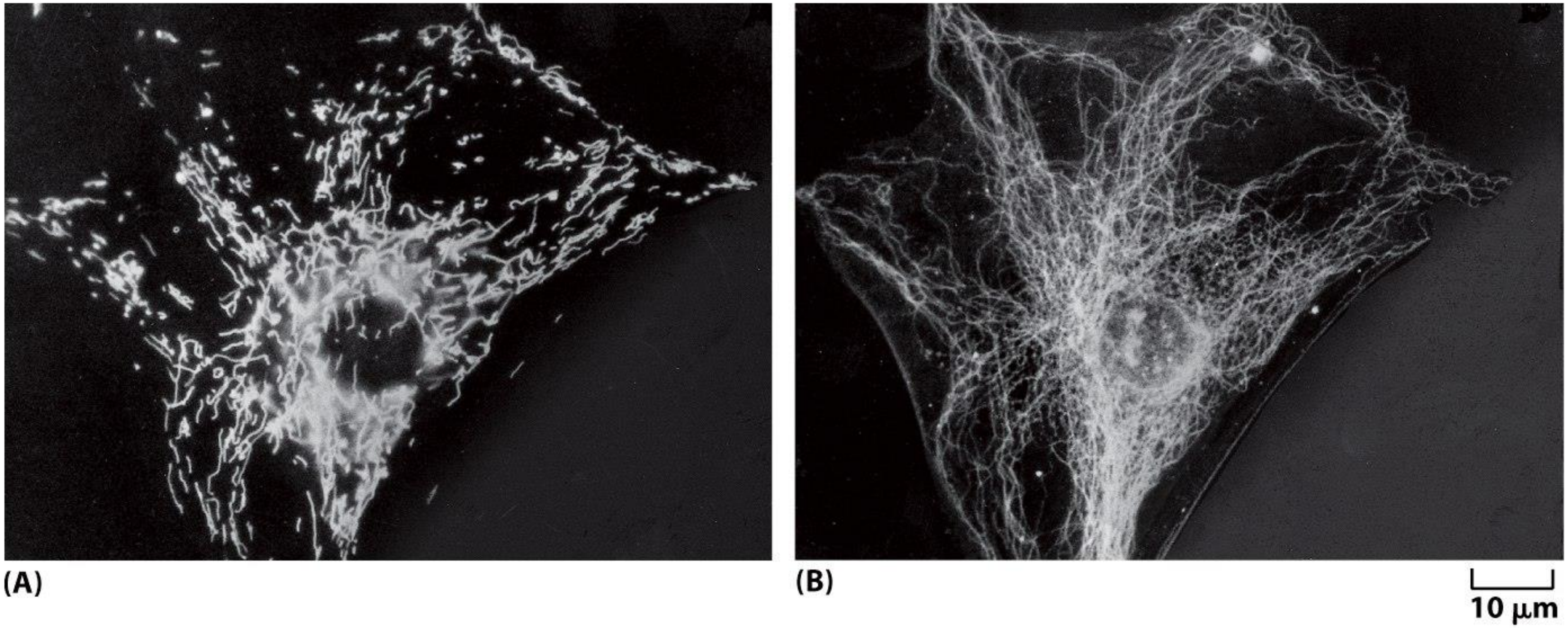
It is synthesized from imported phosphotidylglycerol
In the mitochondria.

# How to fractionate separate compartments for analysis in mitochondria?




Outer membrane is easy to erupt, but inner membrane has many folded cristae, and Is not easy to break down in low Osmotic solution.

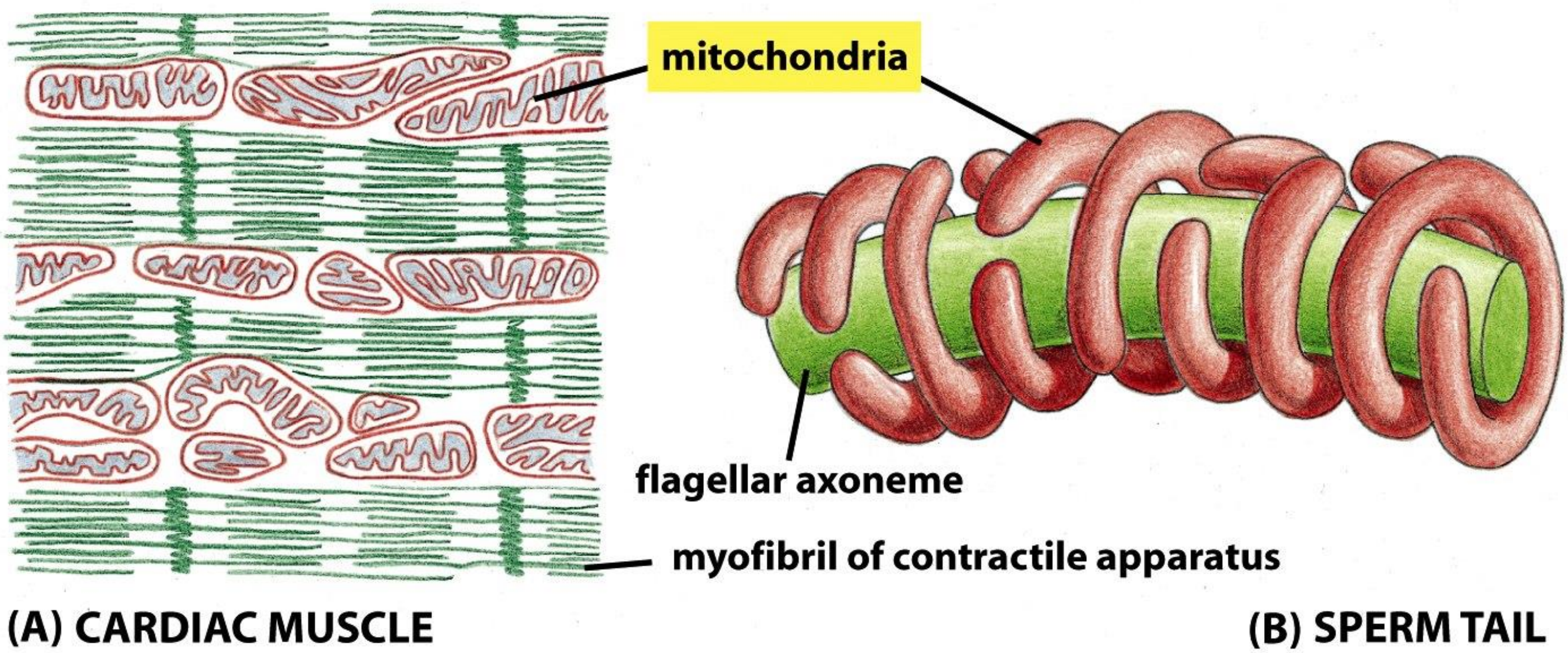
# Mitochondria often associate with microtubules in cells, form moving filaments or chains




Microtubules are important for the orientation and mobility of mitochondria

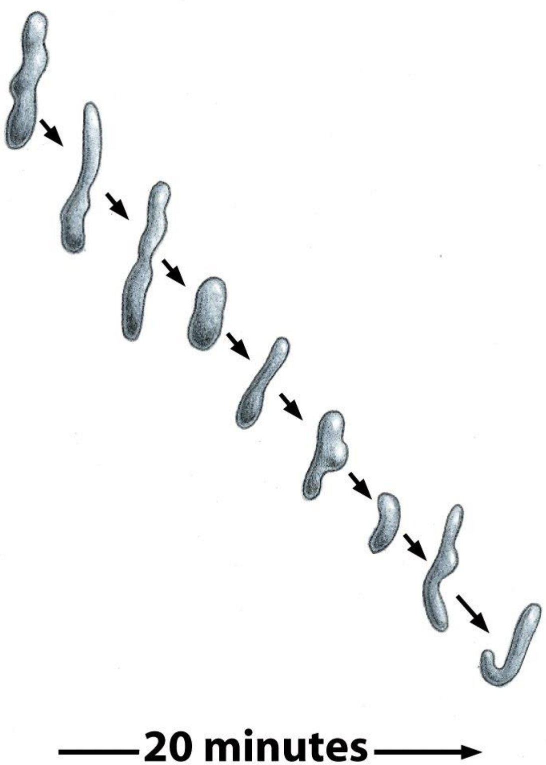
# Mitochondria are mobile but remain fixed in energy requiring sites

Two examples: cardiac muscle and flagella of sperm cells

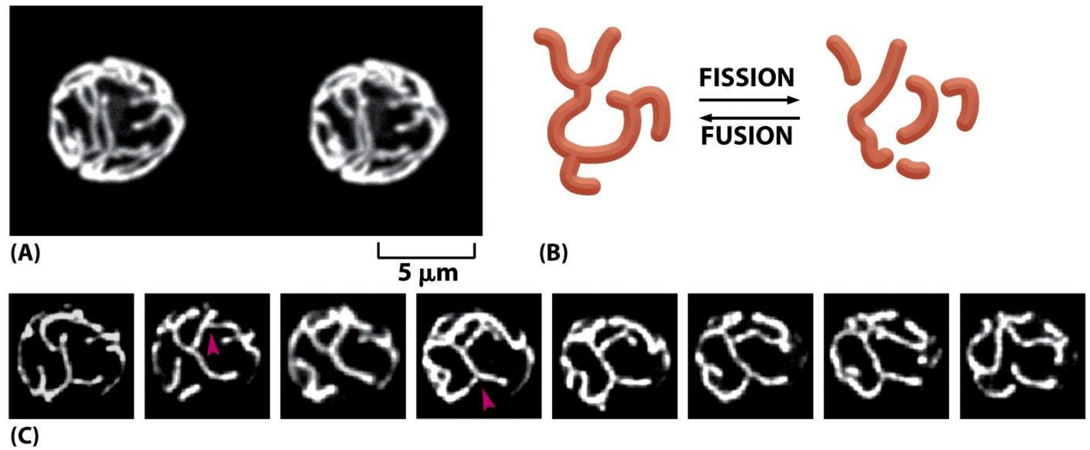


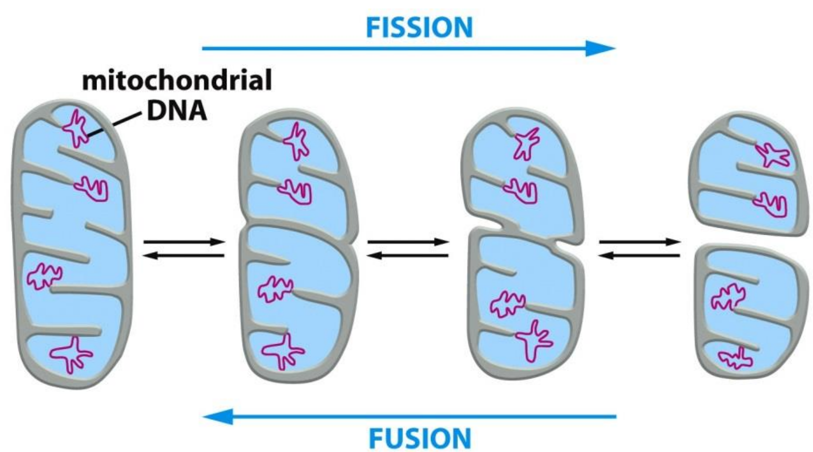
# Plasticity of mitochondria

Shape change of a mitochondria in a living cell during 20 minutes

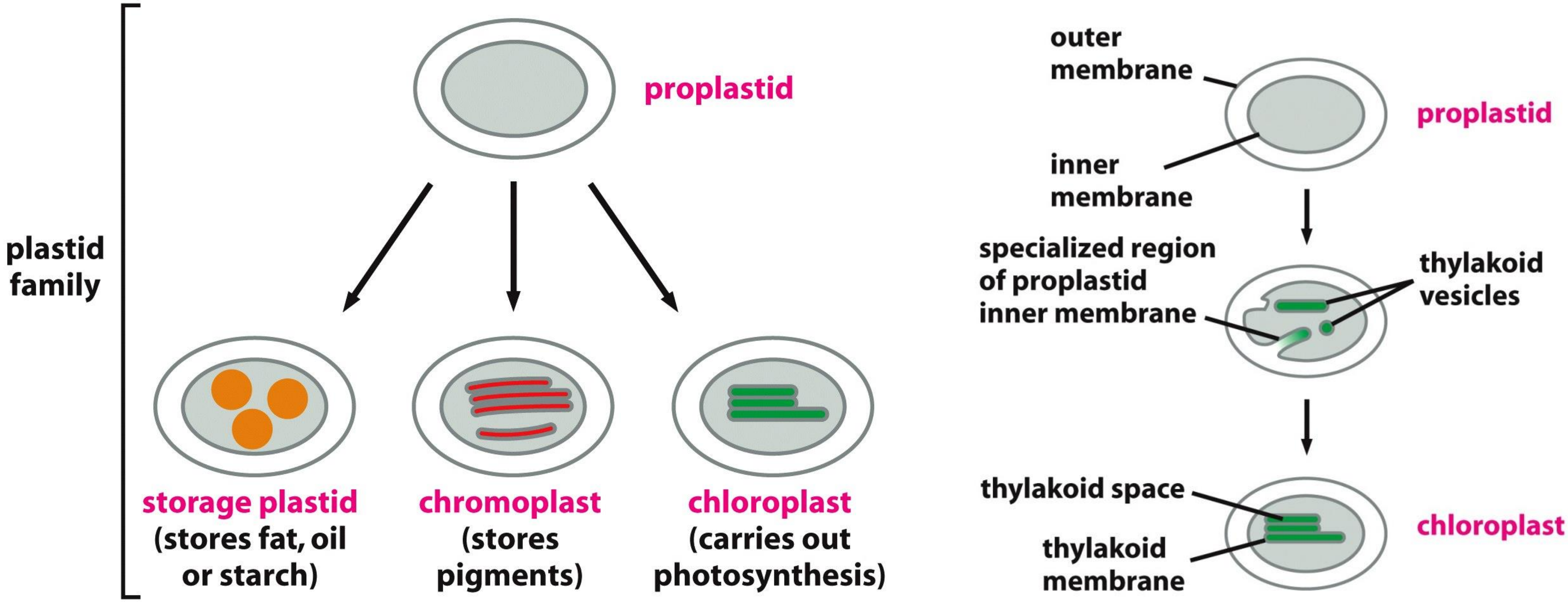



Mitochondria are dynamic and fuse and separate constantly



# II. Plastids and evolution of chloroplast




This is dependent on the different environment
In differentiated cells

# Plastid diversity

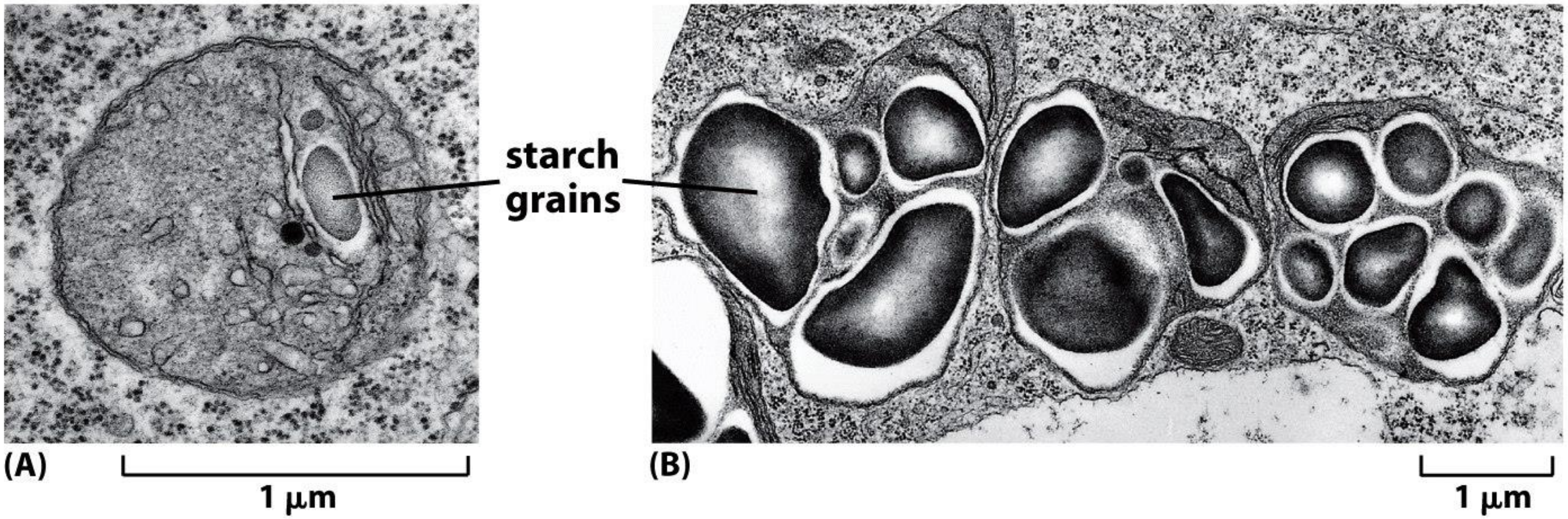
- In one plant species, all plastids share the same genome of their own.
- They are:

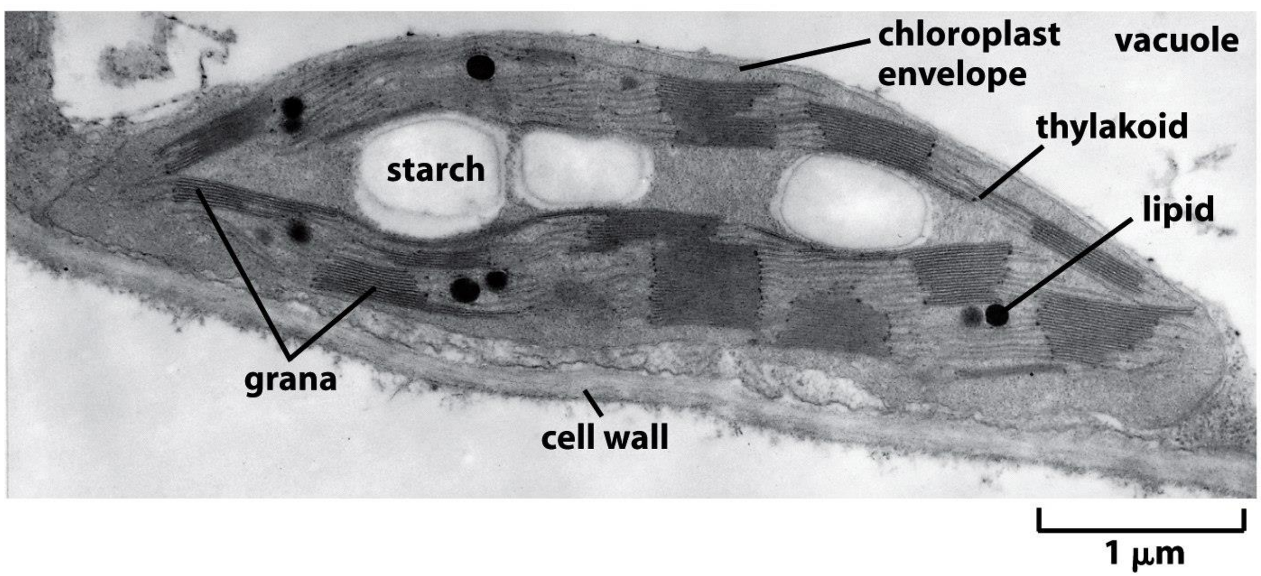
 1. most notable sites for photosynthesis

 2. storage of materials

 3. provide compartments for Purine/Pyrimidine synthesis, amino acid synthesis, fatty acid synthesis in plants

# Plastids, chloroplast

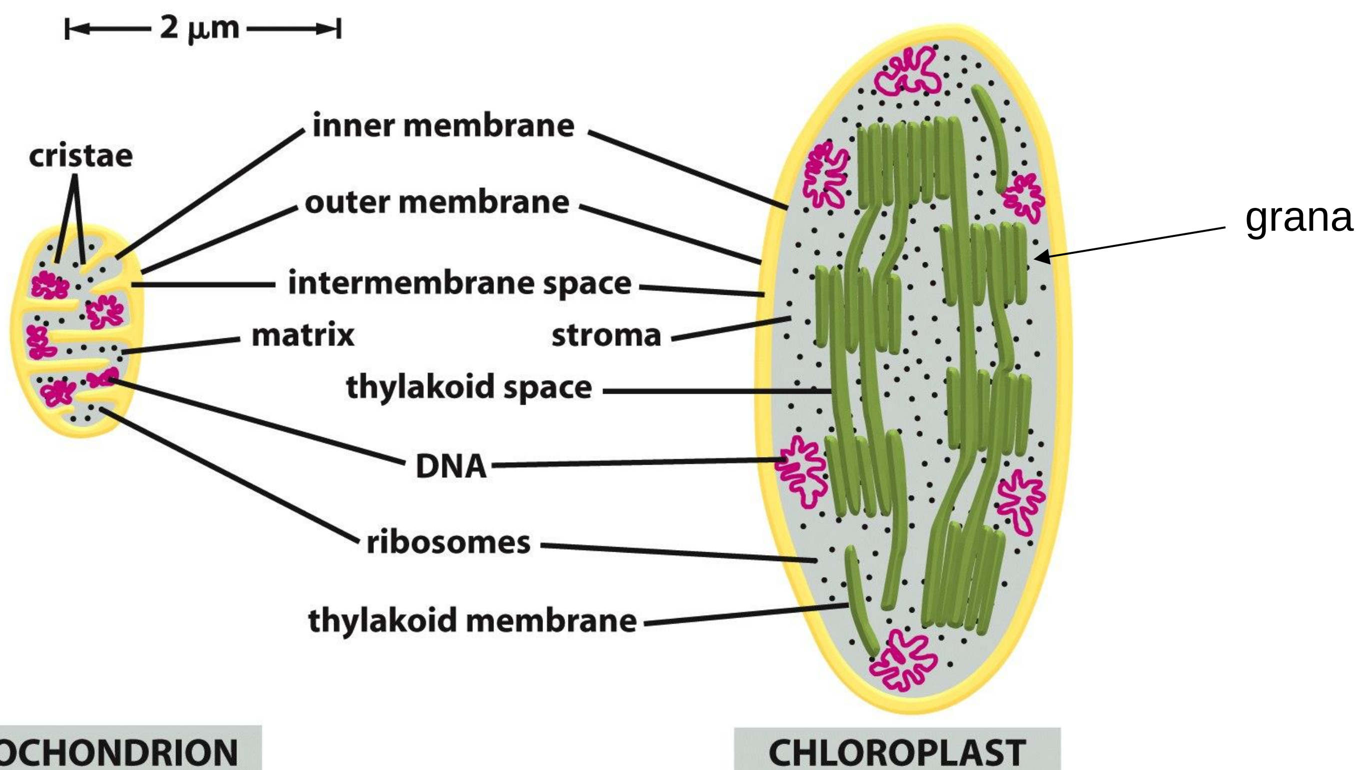


# Chloroplast compartments

Outer membrane: highly permeable
Inner membrane:  much less permeable, localization of membrane transport proteins
Stroma: sites of metabolic enzymes, chloroplast ribosomes, RNAs, chloroplast DNA.
Thylakoid membrane:  sites of electron-transport chains, photosynthetic light-capturing systems, ATP synthase
Thylakoid space: interconnected.

Both contains multiple copies of their genome DNA molecules.



# Mitochondria and chloroplast DNA in different cells

**Table 14–2 Relative Amounts of Organelle DNA in Some Cells and Tissues**

| ORGANISM | TISSUE OR CELL TYPE | DNA MOLECULES PER ORGANELLE | ORGANELLES PER CELL | ORGANELLE DNA AS PERCENTAGE OF TOTAL CELLULAR DNA |
|---|---|---|---|---|
| **MITOCHONDRIAL DNA** | | | | |
| Rat | liver | 5–10 | 1000 | 1 |
| Yeast* | vegetative | 2–50 | 1–50 | 15 |
| Frog | egg | 5–10 | $10^7$ | 99 |
| **CHLOROPLAST DNA** | | | | |
| *Chlamydomonas* | vegetative | 80 | 1 | 7 |
| Maize | leaves | 0–300** | 20–40 | 0–15** |

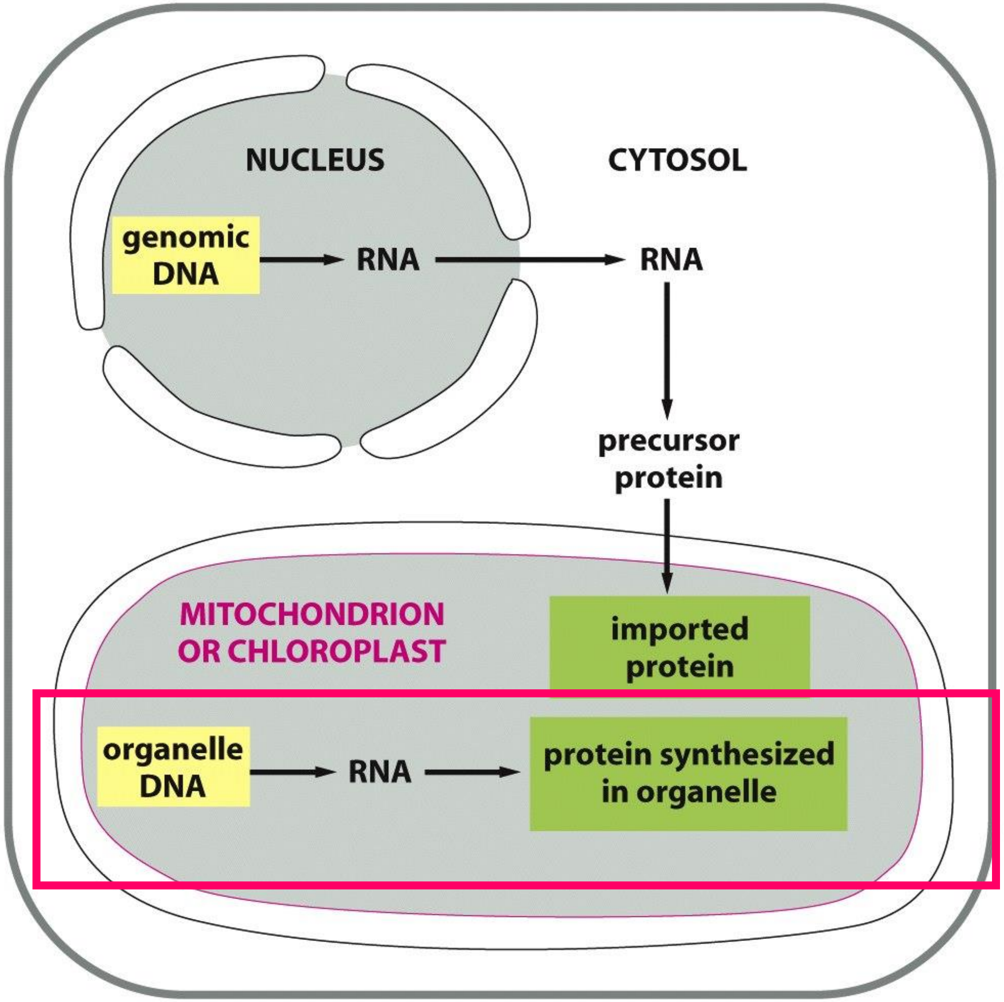
*The large variation in the number and size of mitochondria per cell in yeasts is due to mitochondrial fusion and fission.
**In maize, the amount of chloroplast DNA drops precipitously in mature leaves, after cell division ceases: the chloroplast DNA is degraded and stable mRNAs persist to provide for protein synthesis.

The number of both chloroplast and mitochondrion can vary depending on physiological conditions and needs of the cells, they are dynamic.

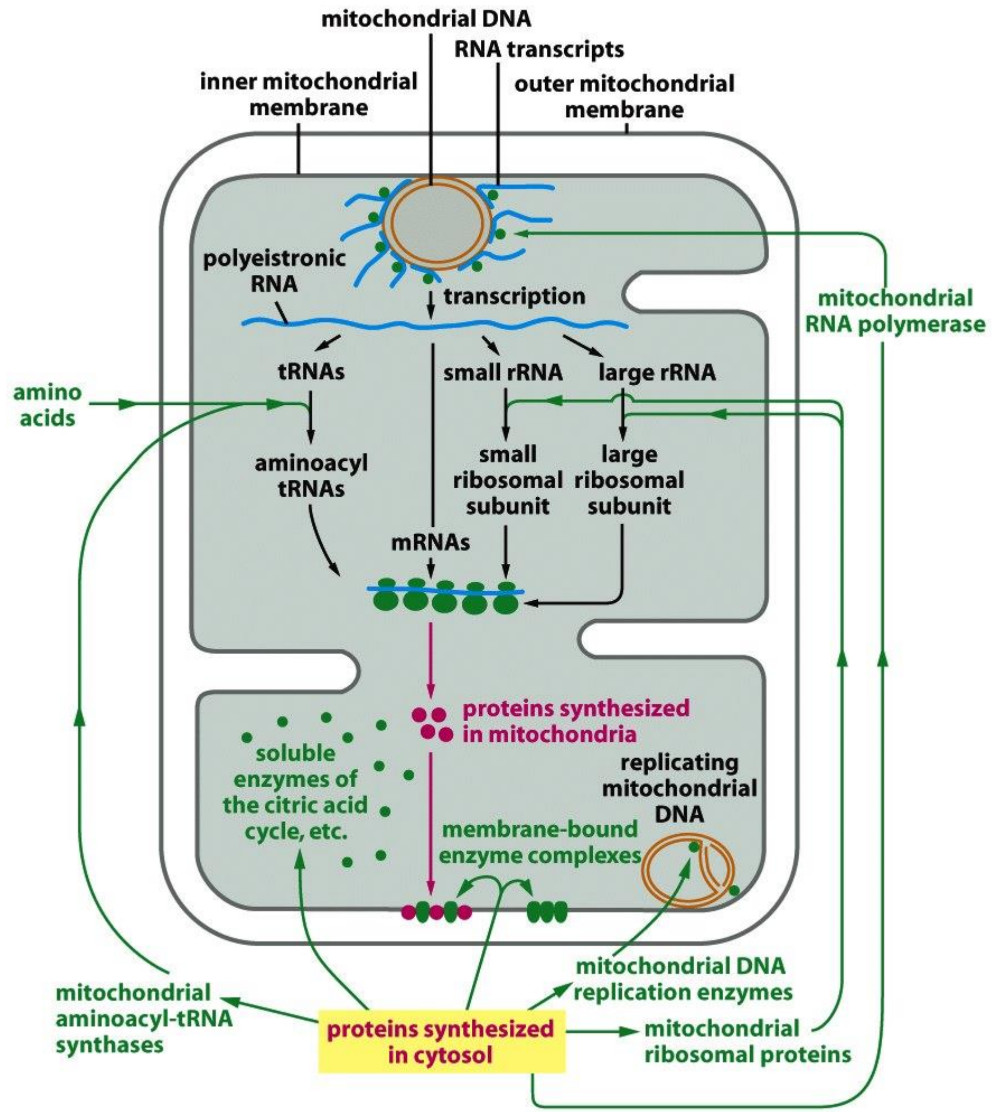
# III. Genetic systems of mitochondrion and plastid

- Genetic system of mitochondria and chloroplasts
- Origin of mitochondria and chloroplast genomes
- Non-mendelian inheritance
- Diseases associated with mitochondria defects.

# Mitochondrion and chloroplast contain their own complete genetic systems



# Coordinated protein synthesis between nucleus and mitochondria



# Animal mitochondria contain the simplest genetic systems known

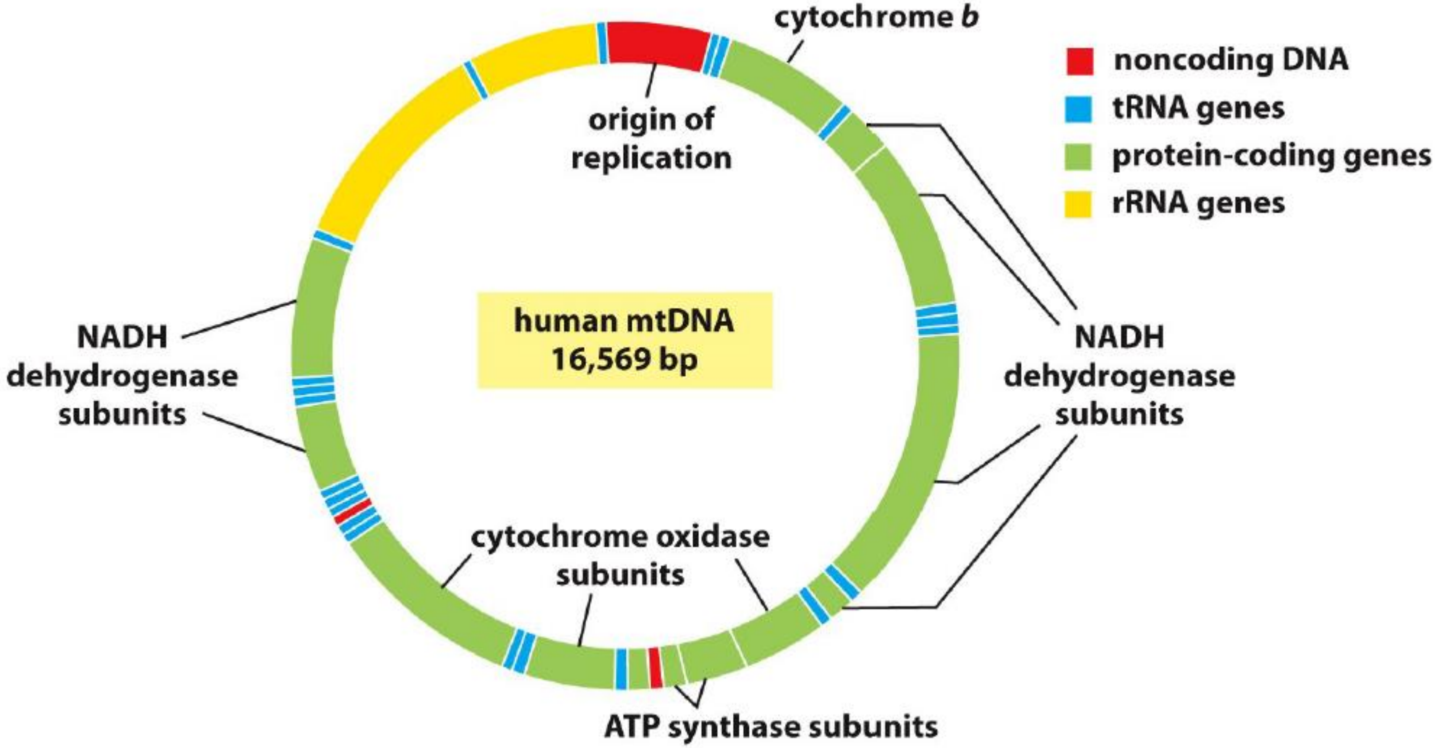
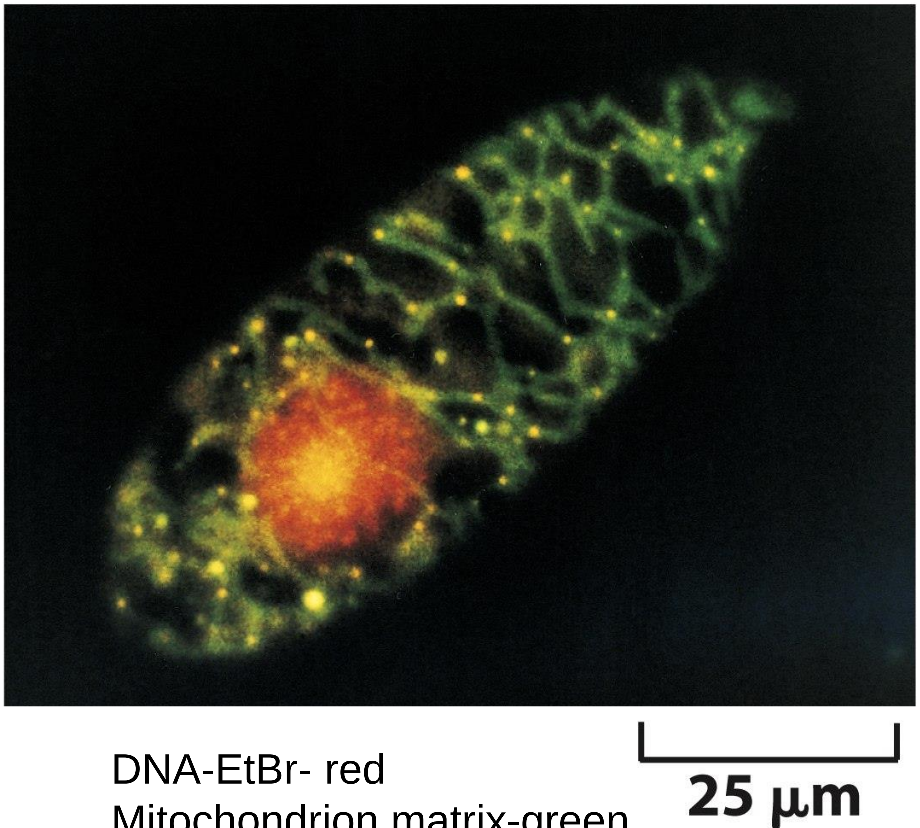



Figure 14-65 Molecular Biology of the Cell 6e (© Garland Science 2015)

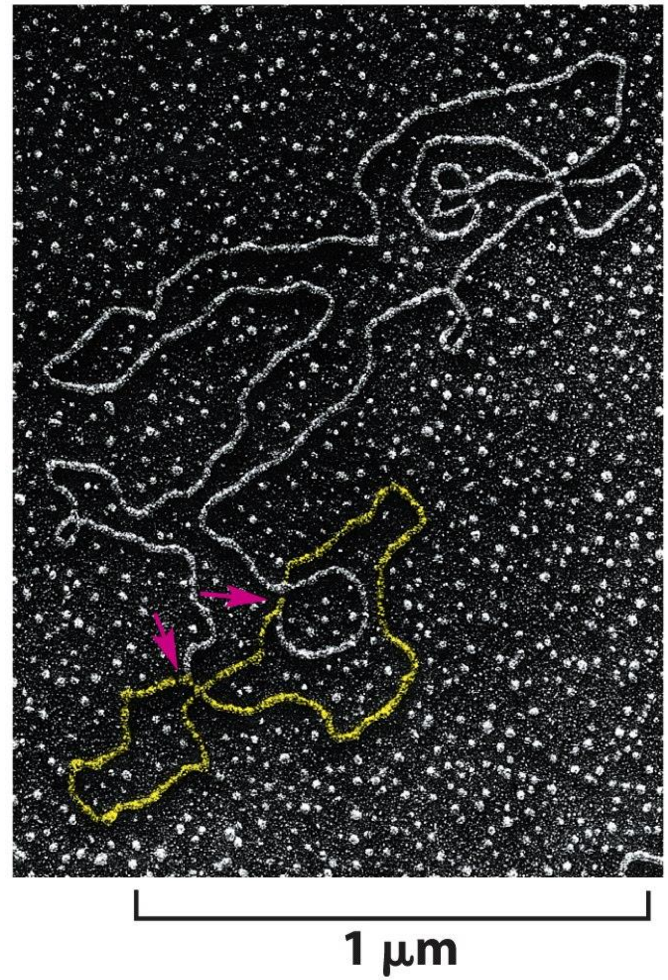
# Chloroplast and mitochondrion protein synthesis machinery

- Chloroplast more resembles present-day bacteria in its protein synthesis machinery
- Both chloroplast and mitochondrion use N-formyl-Met as the initiation amino acid in protein synthesis.
- Protein synthesis in both chloroplast and mitochondrion can be inhibited by antibiotics.--- side effect of antibiotics in eukaryotic organisms?

# Visualization of mitochondria DNA

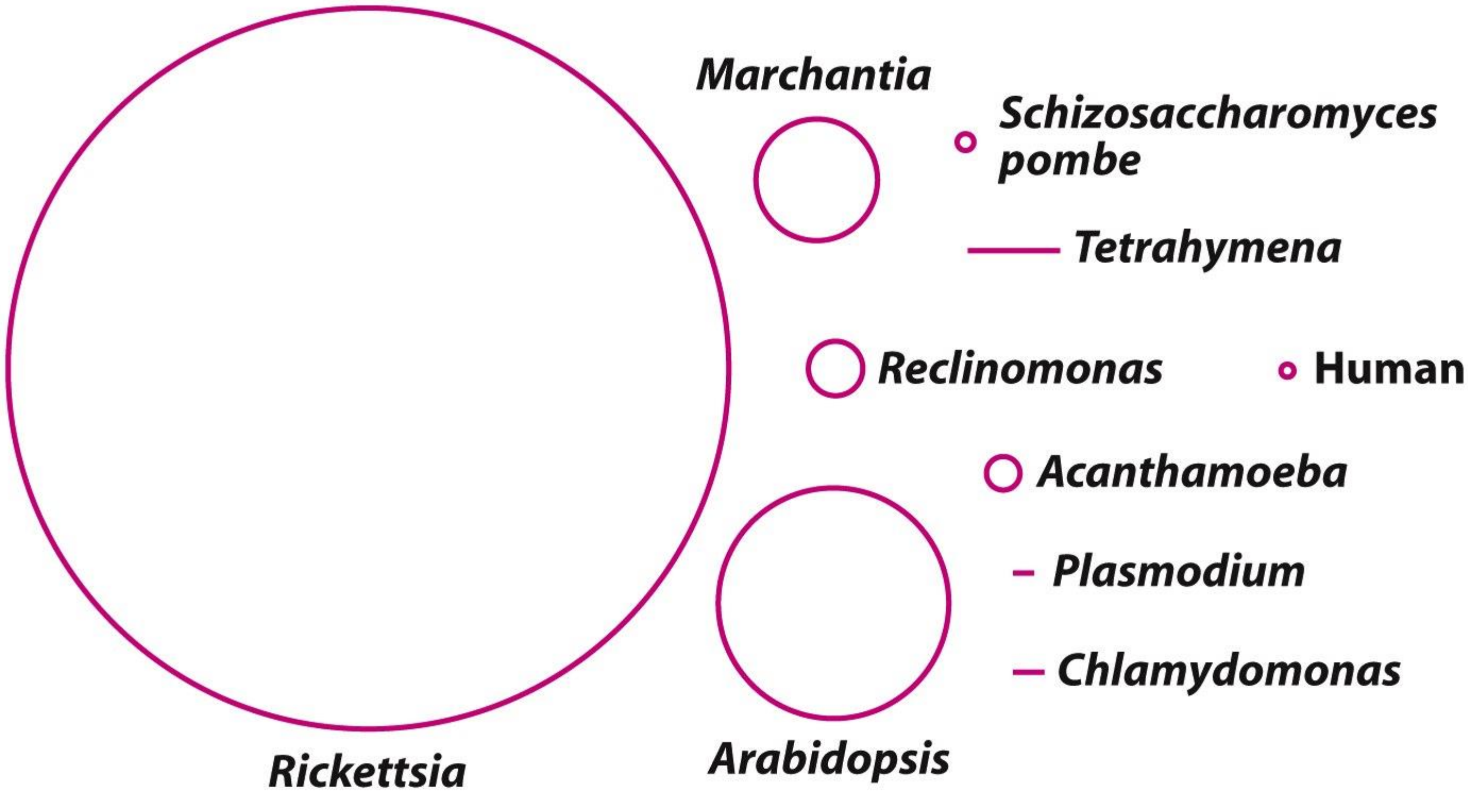



DNA-EtBr- red
Mitochondrion matrix-green



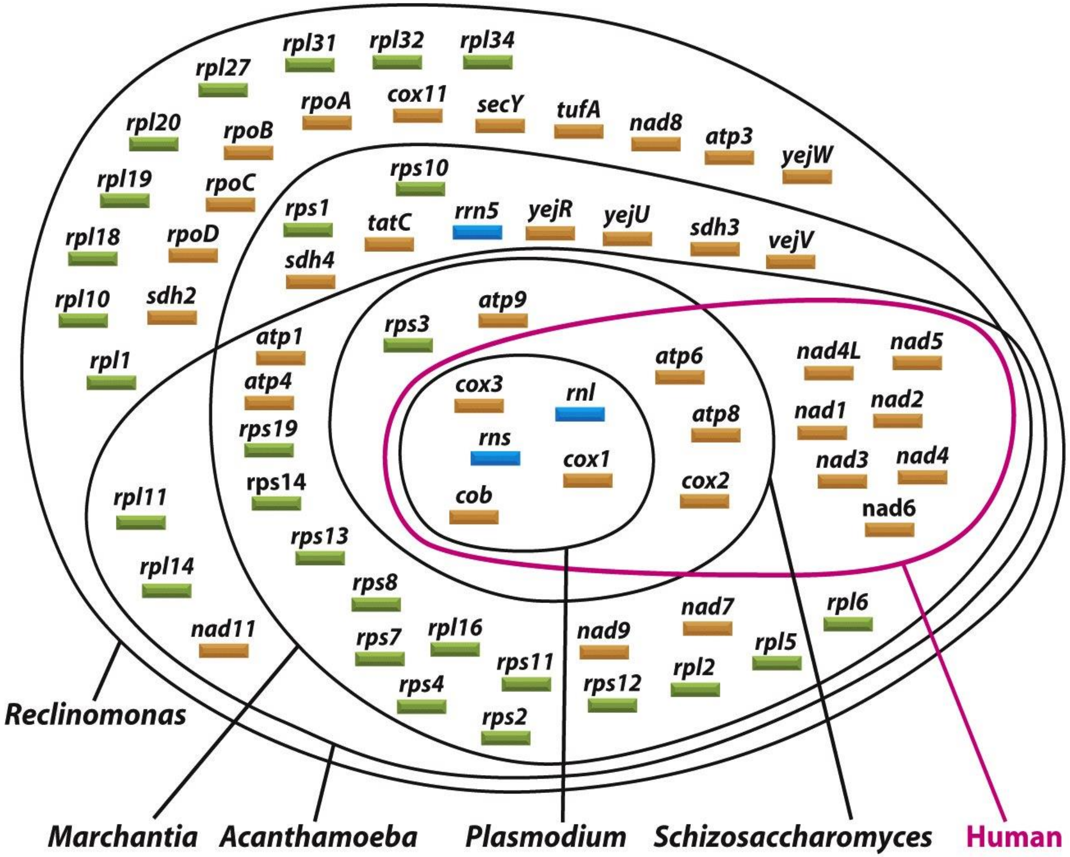

Circular mitochondria DNA is
Replicating, replication occurs
In a cell cycle dependent manner

# Various sizes of mitochondria genomes




Generally range from 6000 bp to 300,000bp, for mammals, ~16500bp.
Some chloroplasts and mitochondria genomes are in linear form.

# Gene transfer occurs after symbiosis events




Evidence to support this:

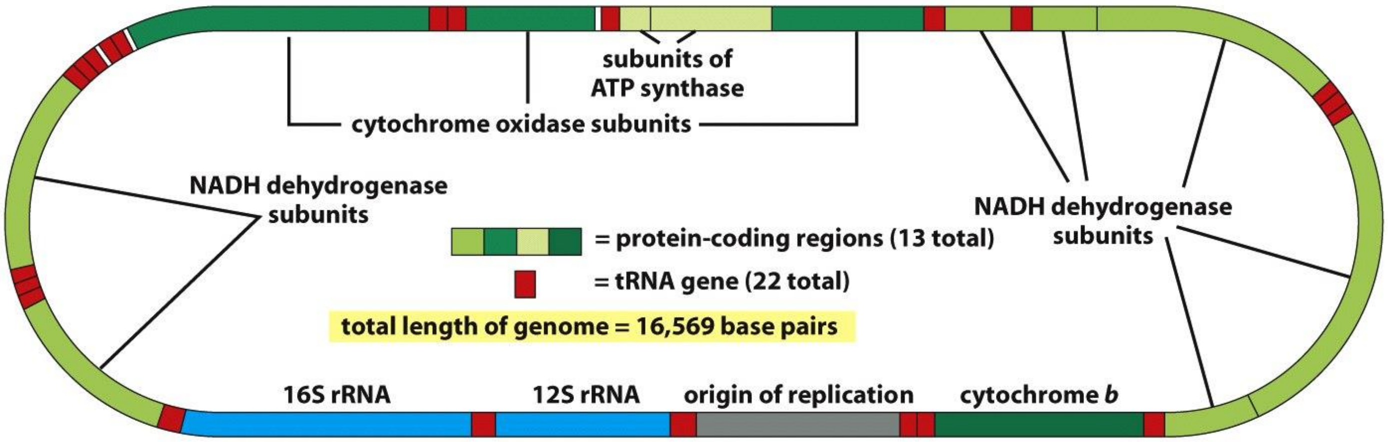
1. Most of mitochondria and chloroplast proteins are encoded by nuclear genes.

2. SOD in mitochondria more resembles that of bacteria than cytosolic SOD

3. Sequential reduction in mitochondria encoding genes during evolution

# Human mitochondria genome has its distinct property

- Dense gene packing
- Relaxed codon usage( 22tRNA versus >30tRNA in cytosol)
- Variant genetic code




10X higher nucleotide substitution rate due to lower DNA replication fidelity and less efficient DNA repair.

# Transcription in mitochondria

- Occurs from one promoter but synthesis on both direction with both DNA strands as templates.
- mRNA has no 5’-cap but has poly A tail
- Some plant and fungi mitochondria RNA has intron, as well as for some plant chloroplast, introns are spliced out in a RNA –mediated catalysis or facilitated by proteins.

# Variant genetic code in mitochondria

**Table 14–3 Some Differences Between the "Universal" Code and Mitochondrial Genetic Codes***

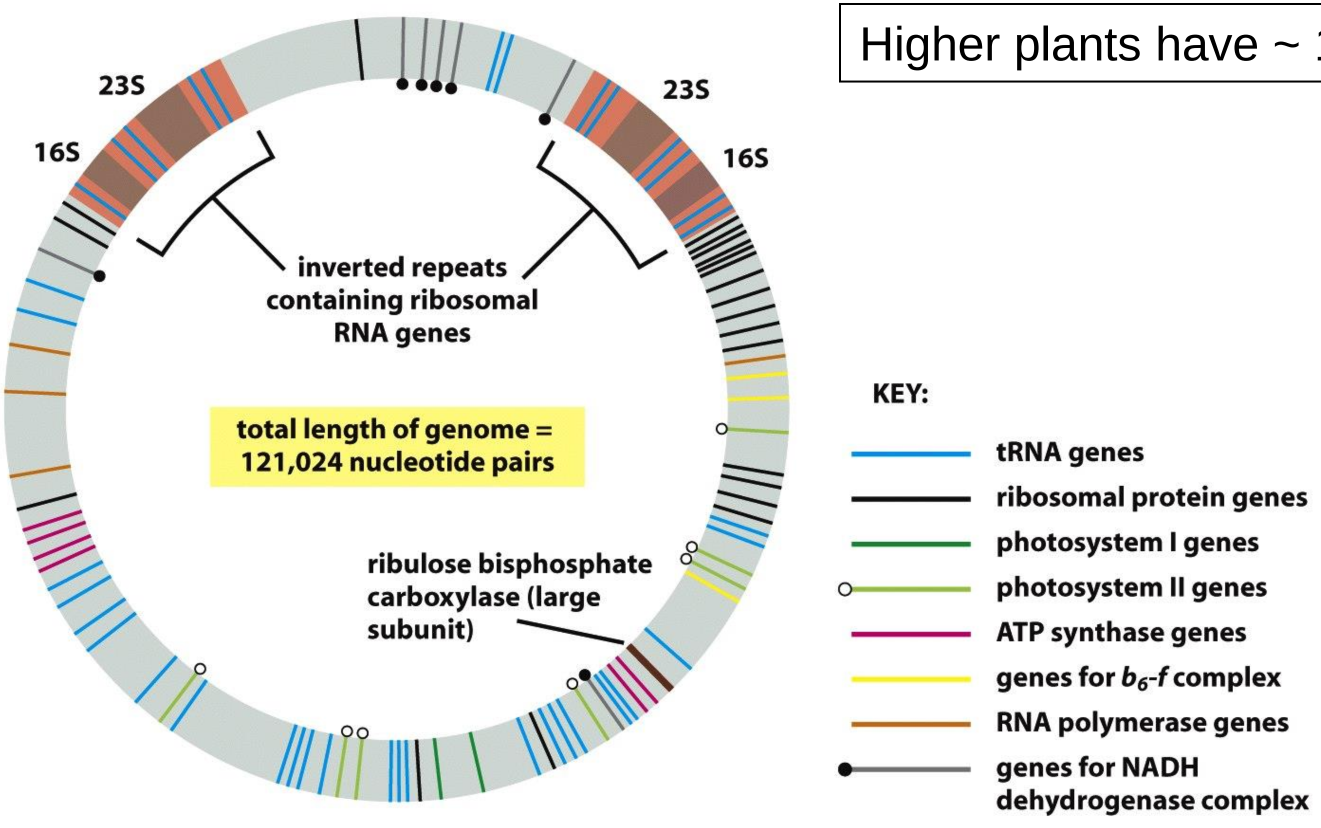
| | | MITOCHONDRIAL CODES | | | |
|---|---|---|---|---|---|
| CODON | "UNIVERSAL" CODE | MAMMALS | INVERTEBRATES | YEASTS | PLANTS |
| UGA | STOP | *Trp* | *Trp* | *Trp* | STOP |
| AUA | Ile | *Met* | *Met* | *Met* | Ile |
| CUA | Leu | Leu | Leu | *Thr* | Leu |
| AGA, AGG | Arg | *STOP* | *Ser* | Arg | Arg |

******Red* **italics indicate that the code differs from the "Universal" code.**

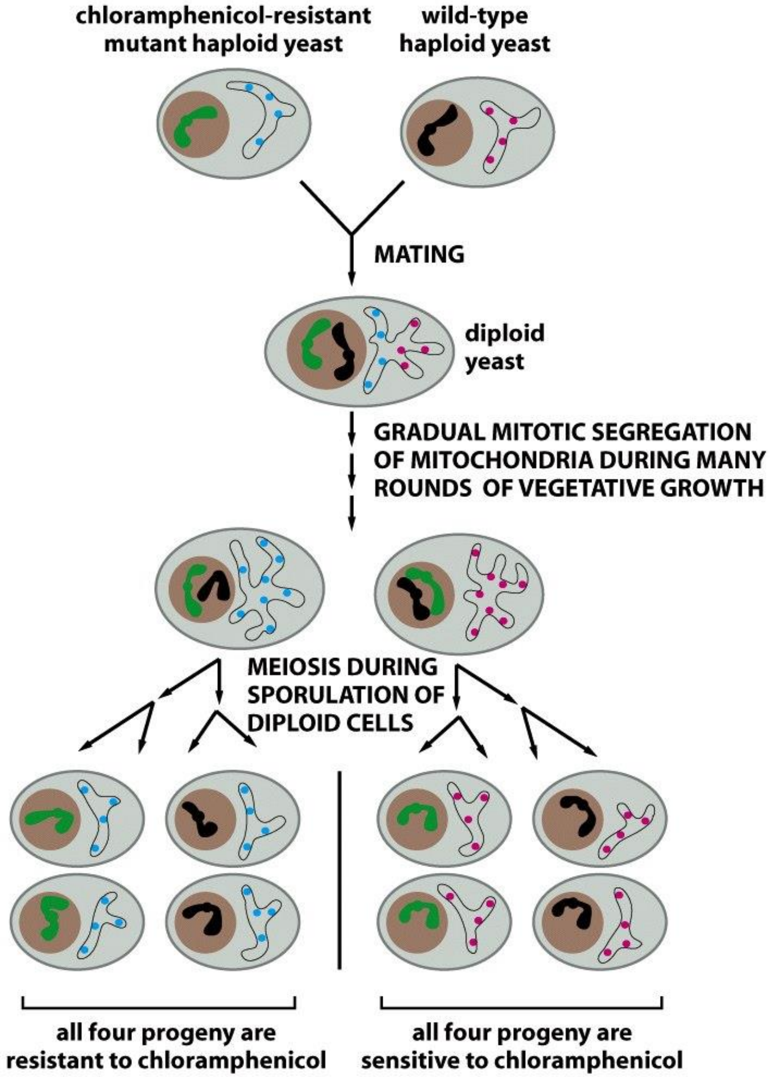
How to explain mitochondria genetic code is different from universal genetic code?

# Genes in chloroplast genomes, all are highly similar

Higher plants have ~ 120 genes



# Mitochondrial genes are inherited by a non-mendelian mechanism



# Non-mendelian inheritance---
# cytoplasmic inheritance

- When diploid cells that have segregated their mitochondrial genomes in this way undergo meiosis to form four haploid daughter cells, each of the four daughters receives the same mitochondrial genes.

- The defination of mitotic segregation:

  One daugher cell with more inheritance of one allele would enrich for this allele in the subsequent mitotic divisions.

# Organelle genes are maternally inherited in many organisms

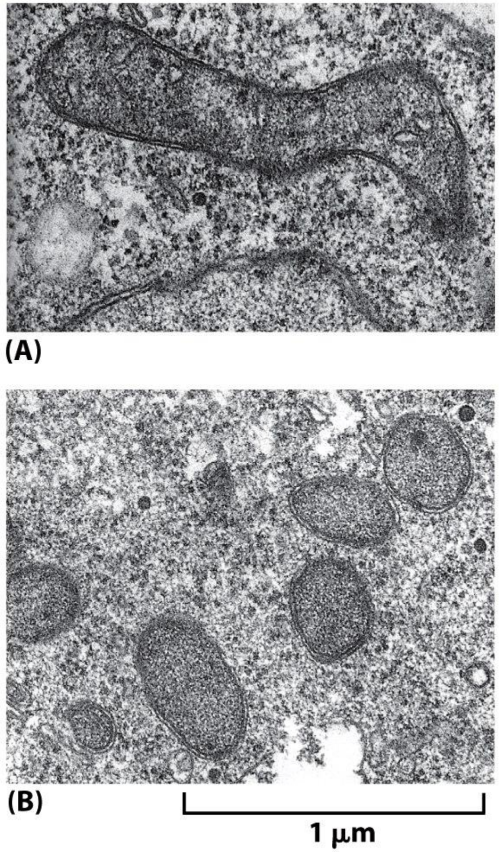
- Human zygote has most maternal mitochondria.
- 2/3 of higher plants have maternal chloroplasts.
- Inheritance in yeast is bi-parental.

# Myoclonic epilepsy and ragged red fiber disease (MERRF)

- Mutation in one of the mitochondria tRNA gene, a defect in making electron transport protein which results in defects in ATP synthesis: most notably on muscle and nerve cells. Muscle weakness, epilepsy, etc.

# Petite mutants for mitochondria and chloroplasts

- Mutants with deletion of majority of mitochondria or chloroplasts DNA




These mutants have defect in synthesizing ATP
and lack inner membrane cristae.

However, mitochondria has normal outer and inner membrane
And can even perform DNA replication, etc.

Supporting that nucleus is sufficent to support mitochondria
Biogenesis .

# Different lipid origins in mitochondria and chloroplast

- Mitochondria: most imported from ER
- Chloroplast: most synthesized on its own

# Mitochondria and aging

- High rate of superoxide due to error in electron transfer to oxygen

- High percentage of damage on mitochondria and in turn on the cells

# IV Energy Conversion

- Energy conversion in mitochondria
- Energy conversion in chloroplast

# Electronic transfer in battery

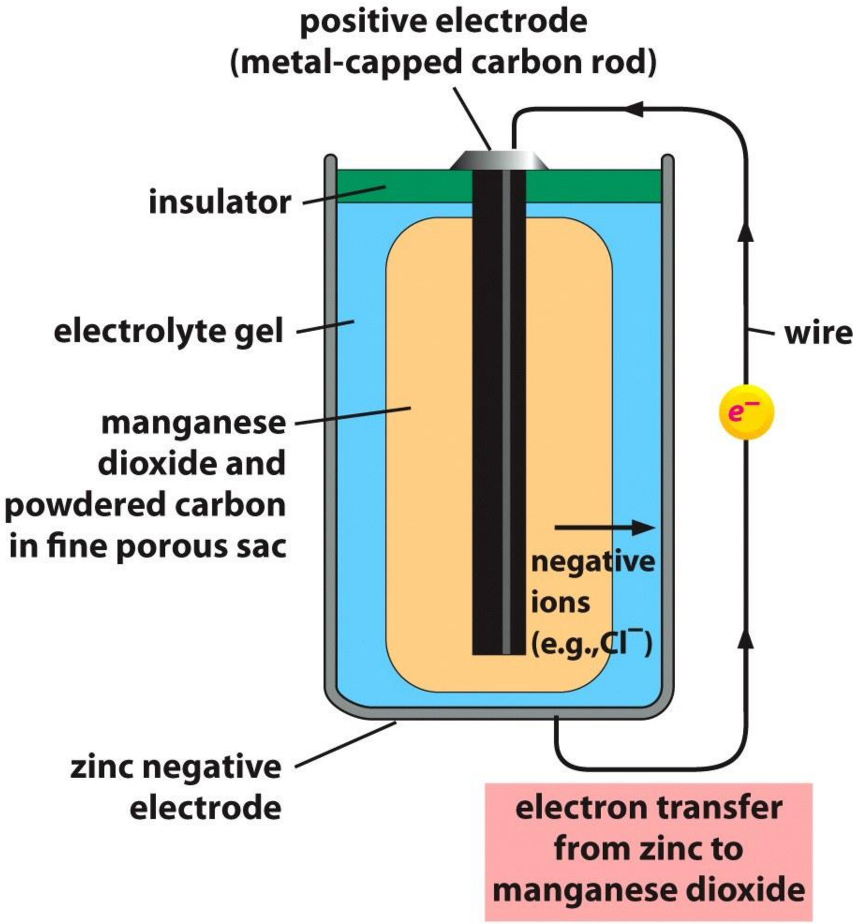



Figure 14-2a Essential Cell Biology 3/e (© Garland Science 2010)

# Electronic energy  can be consumed  into heat

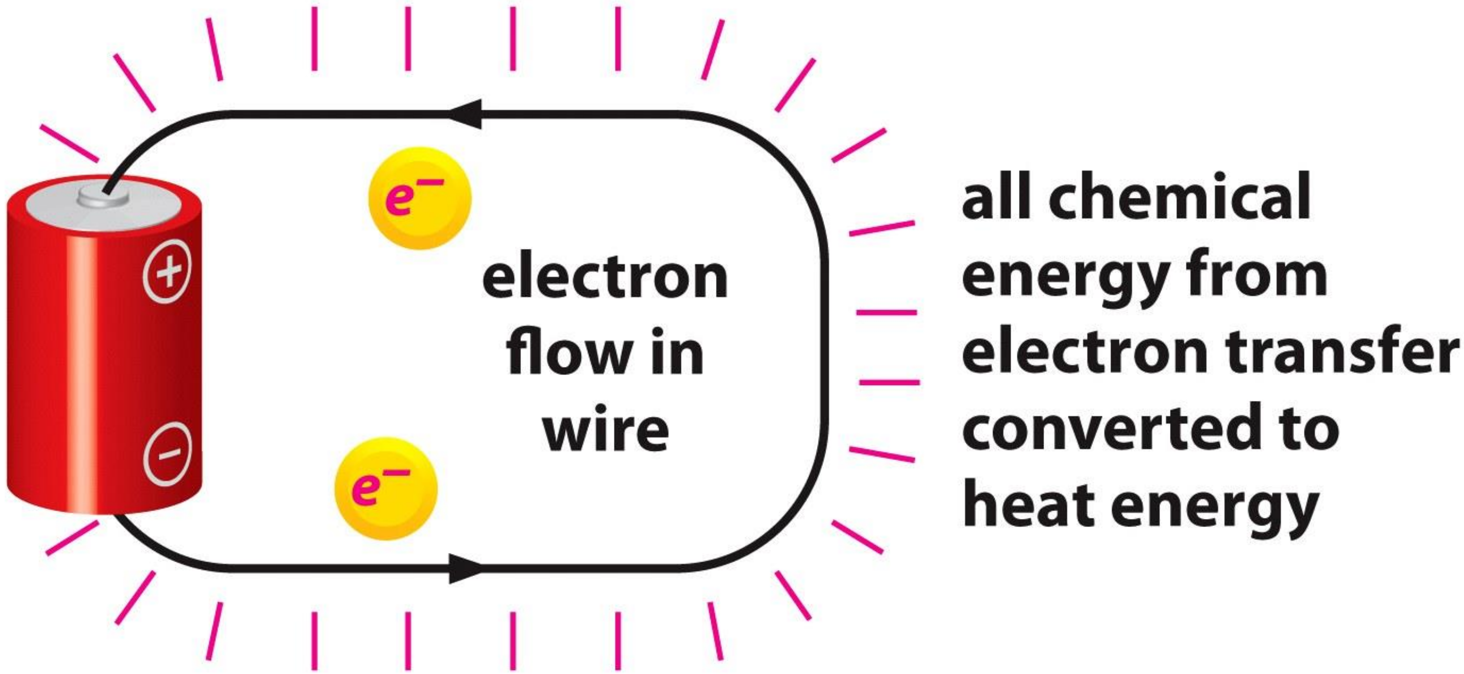
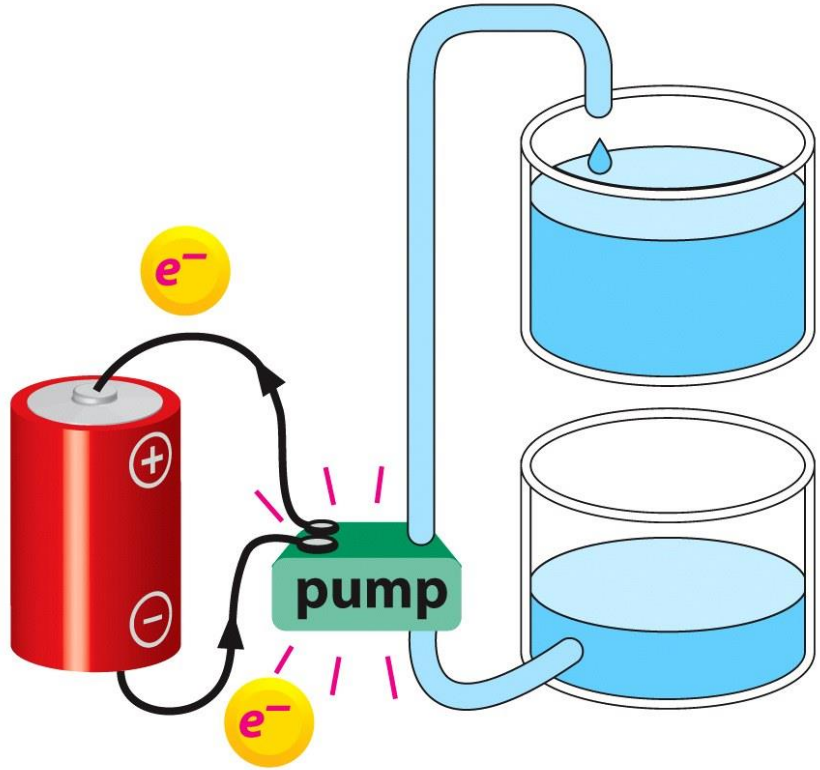



Figure 14-2b Essential Cell Biology 3/e (© Garland Science 2010)

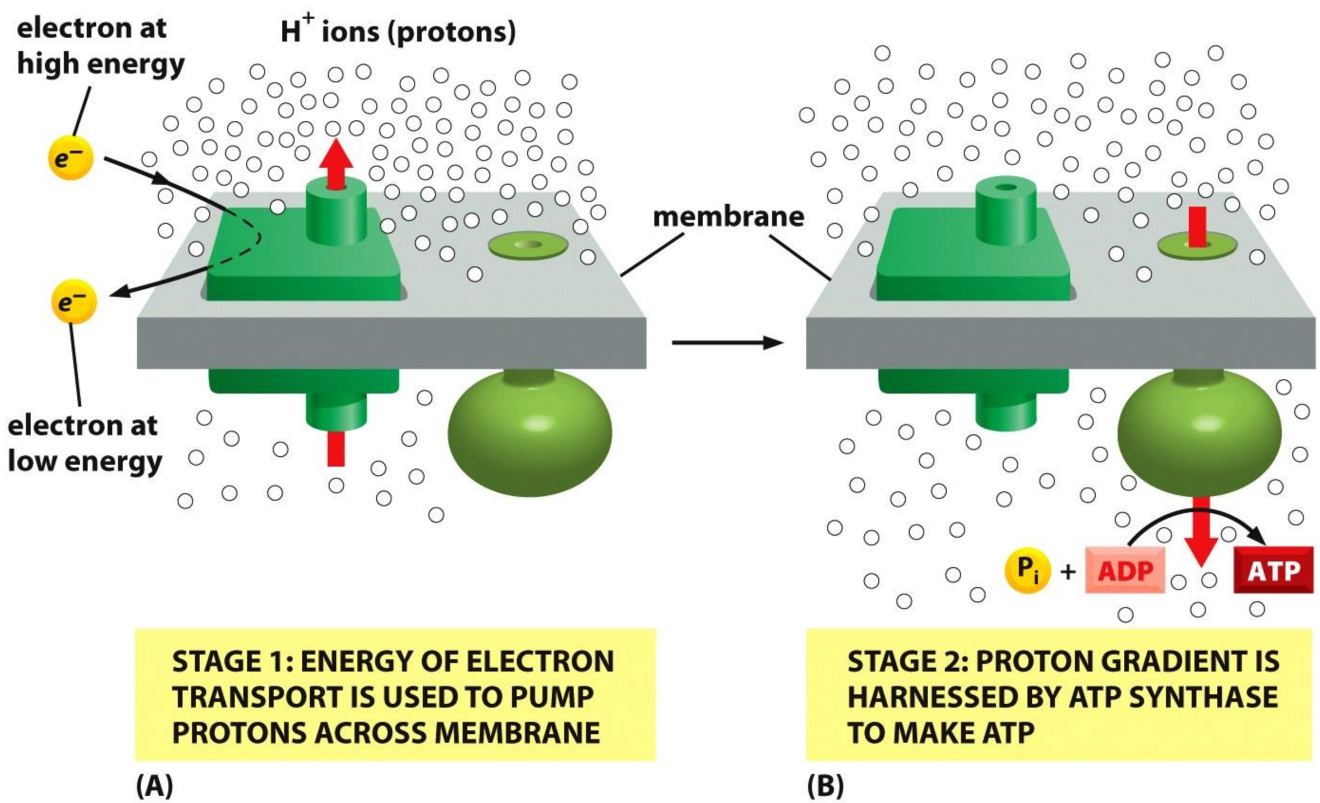
# Electronic flow converts into other forms of energy




chemical energy from electron transfer converted to the potential energy stored in a difference in water levels; less energy is therefore lost as heat energy

Figure 14-2c Essential Cell Biology 3/e (© Garland Science 2010)

# Systems to provide energy for lives

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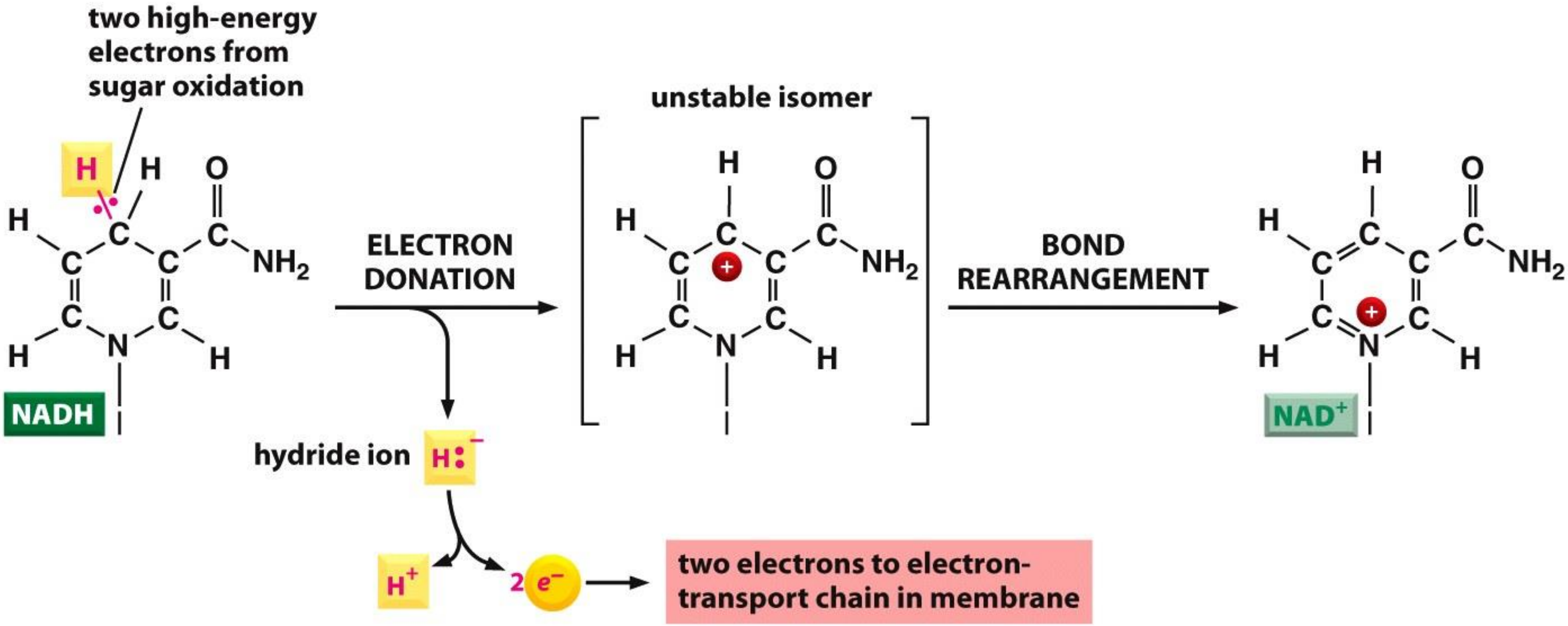
# How is electron generated?

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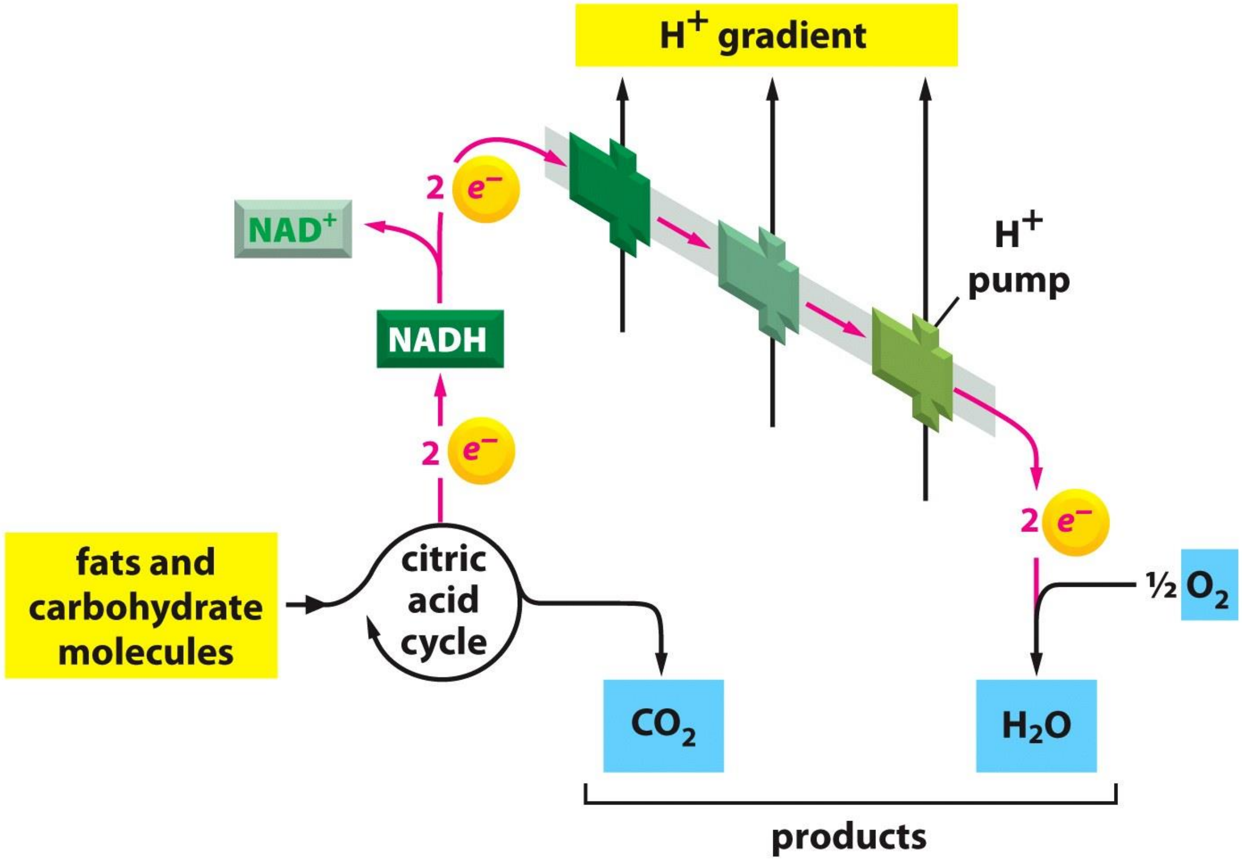
NADH and FADH2 carry high every electrons

# Electrons are transferred by the electron transfer chain (ETC)

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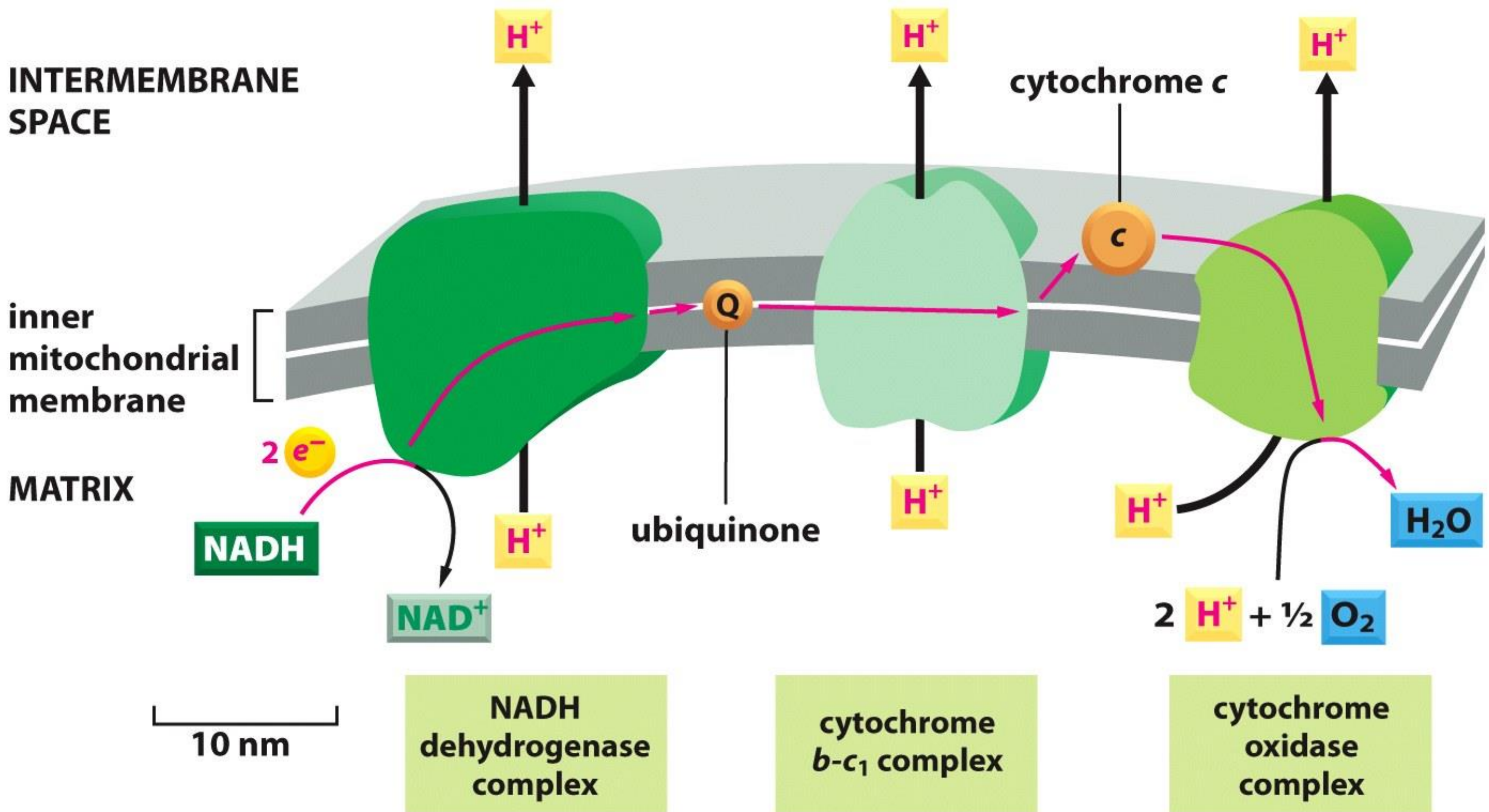
All happens in the inner membrane of mitochondria

# Three respiratory complexes

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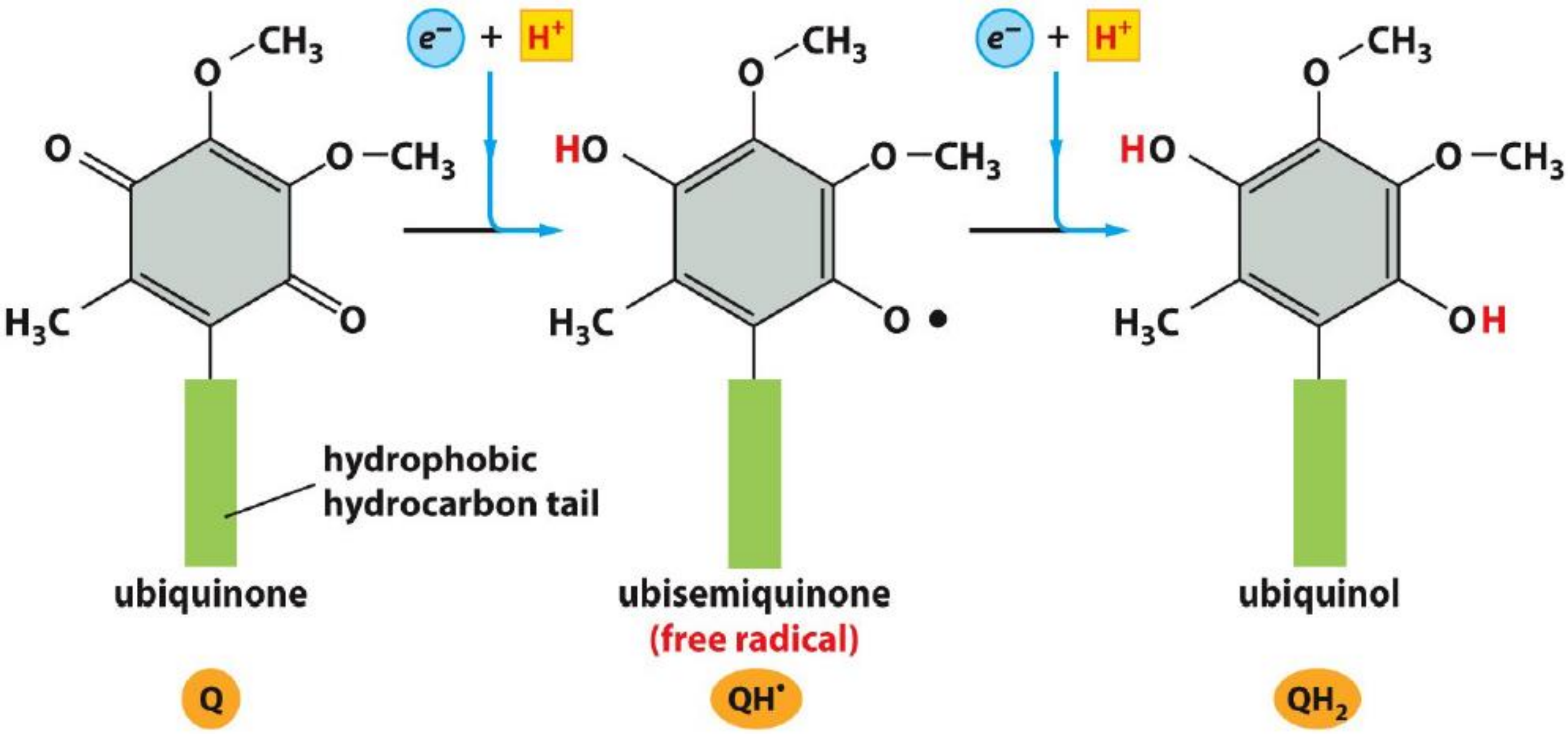
# The Q cycle

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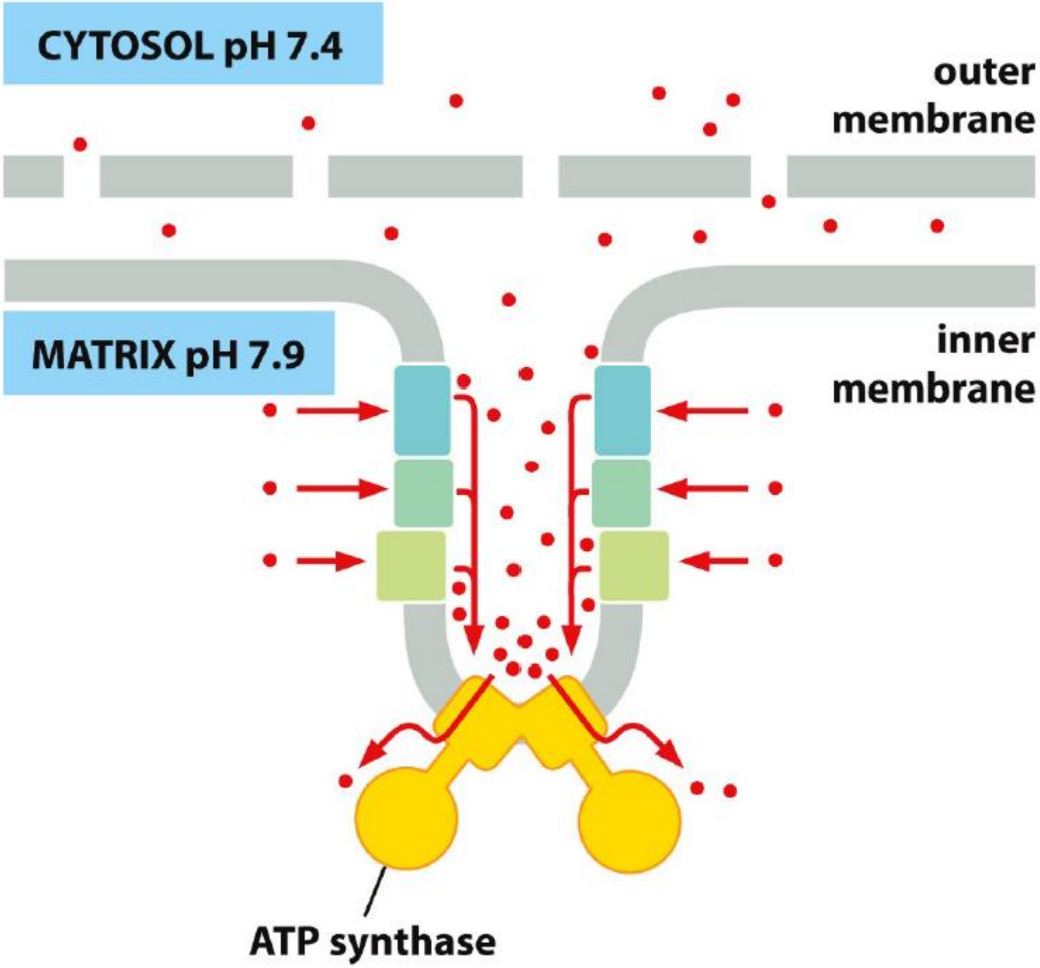
- New discovery revealed that the Q-cycle might not exist…

# Proton is pumped back coupled with ATP synthesis

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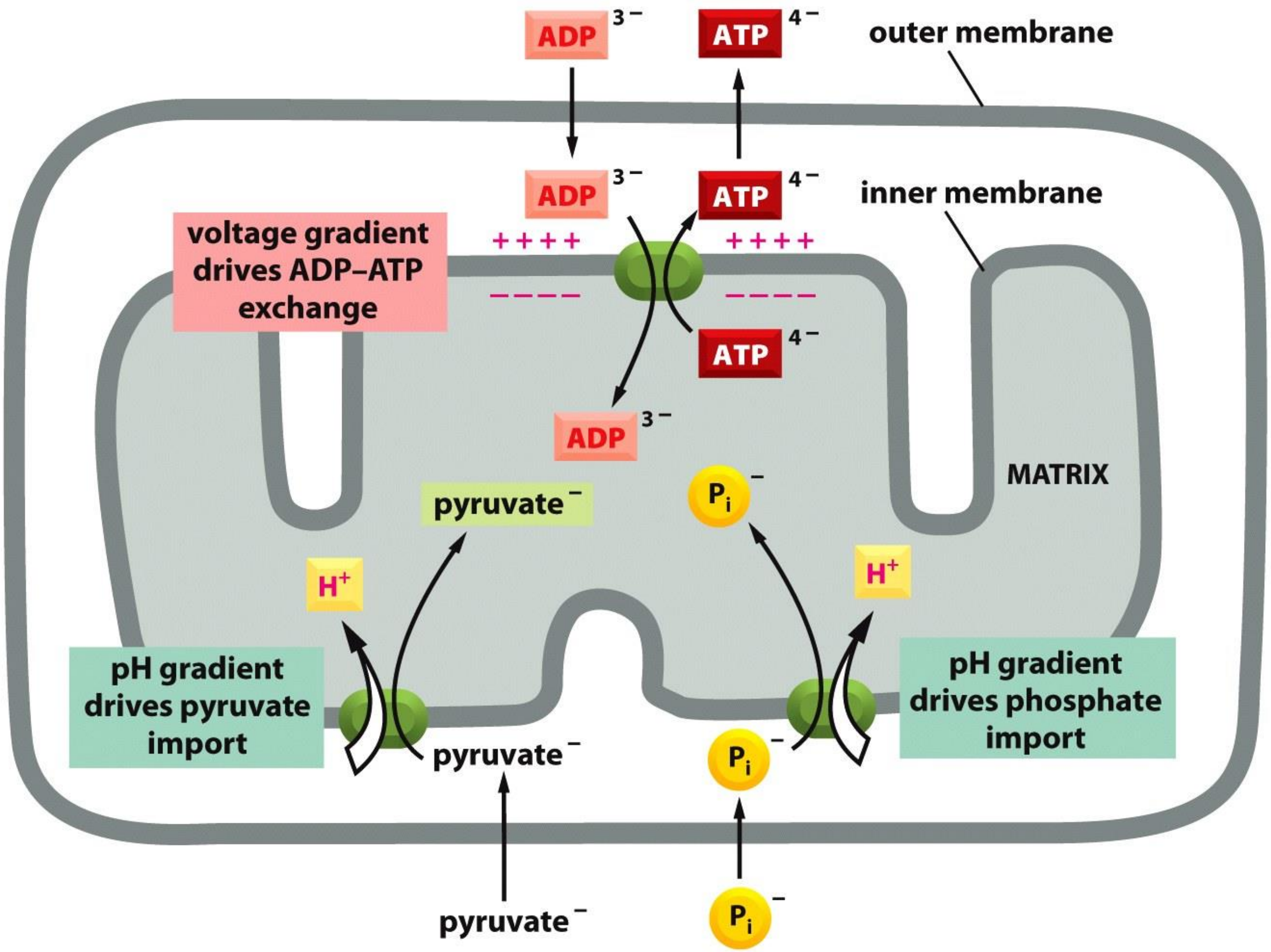
# ADP and Pi are transported in by electrochemical potential and proton gradient

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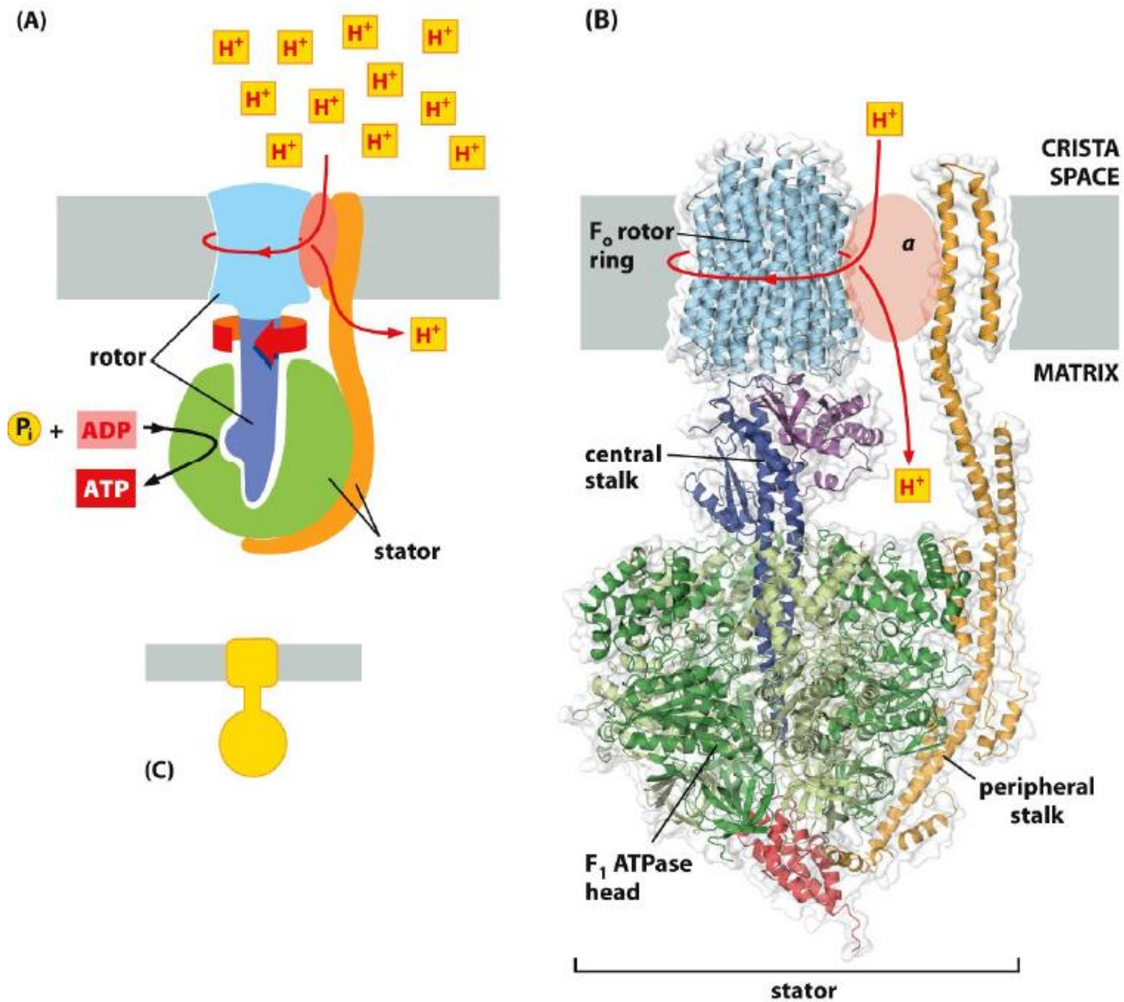
# ATP synthesis

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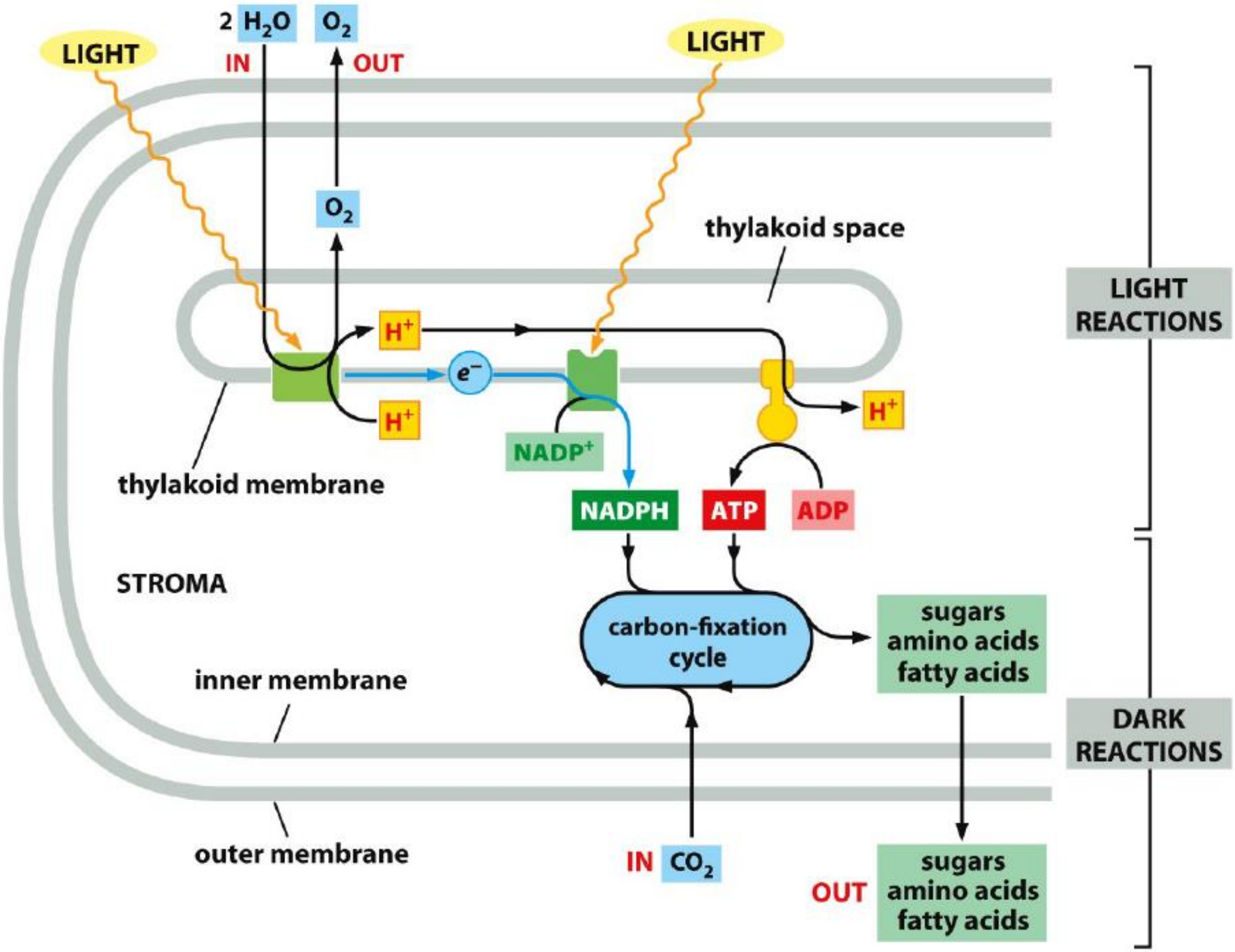
# Chloroplast capture energy from sunlight and use it to fix carbon

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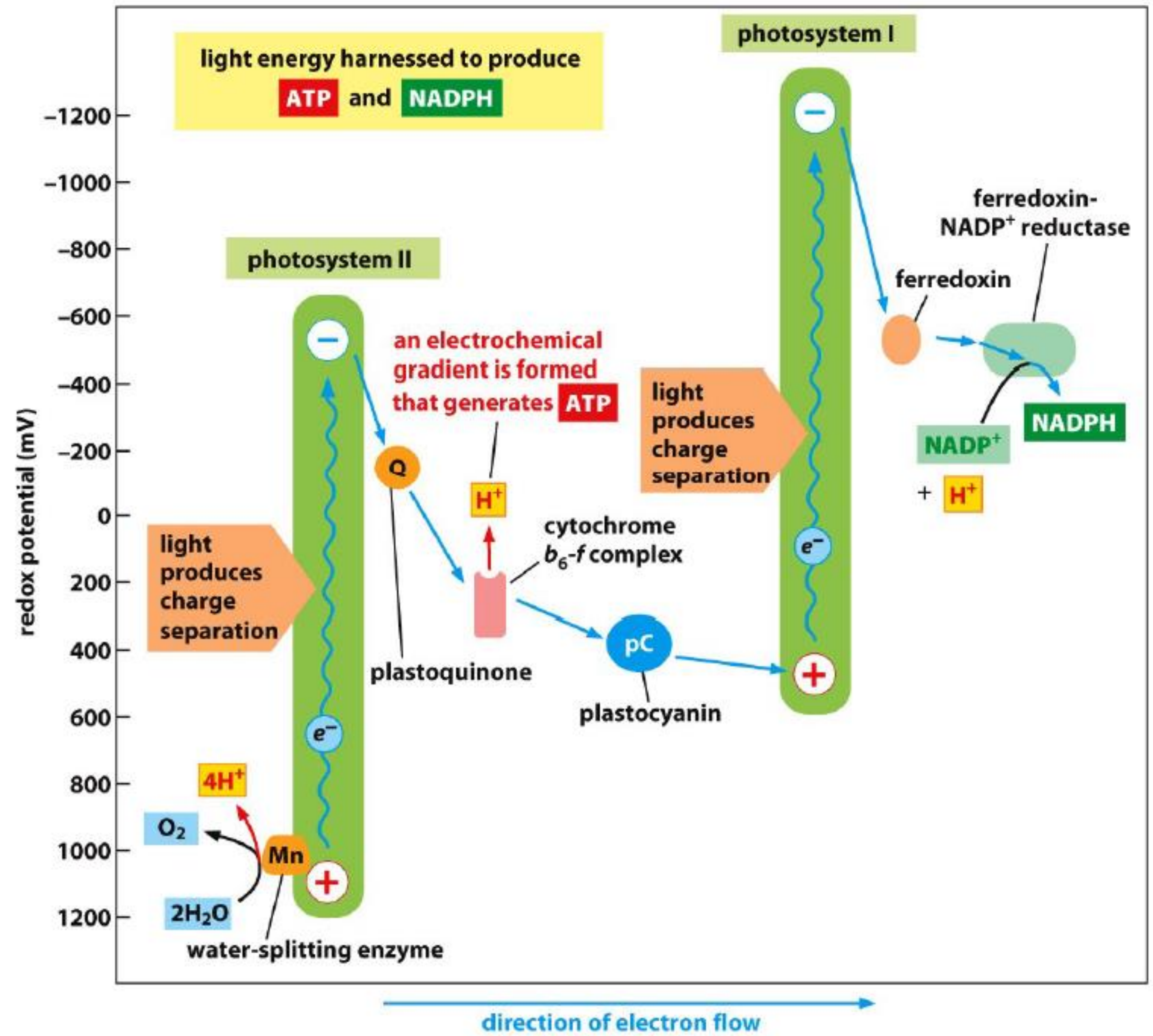
# Thylakoid membrane contains two different photosystems

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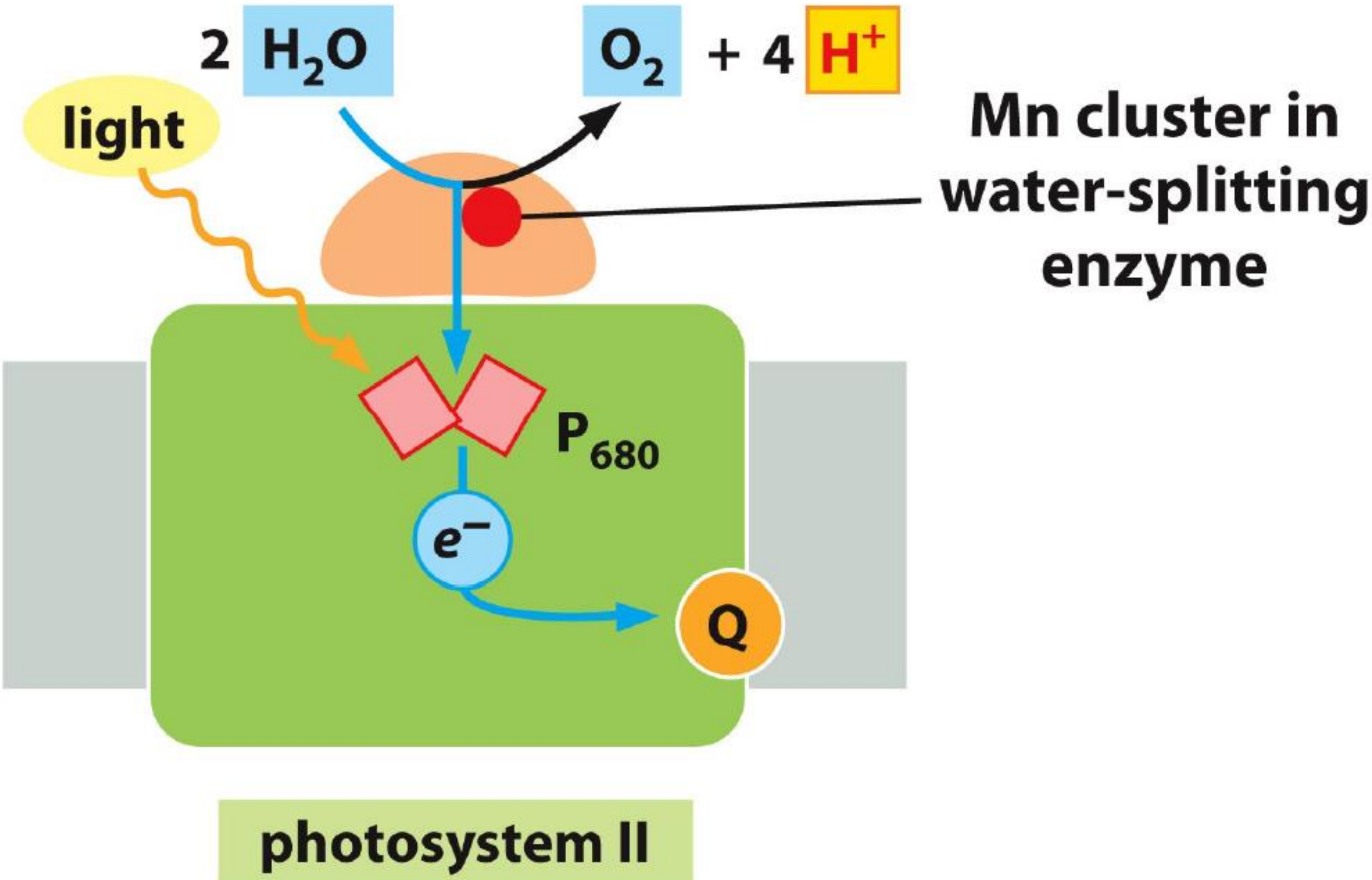
# PS II uses Mangnese cluster to withdraw electrons from water

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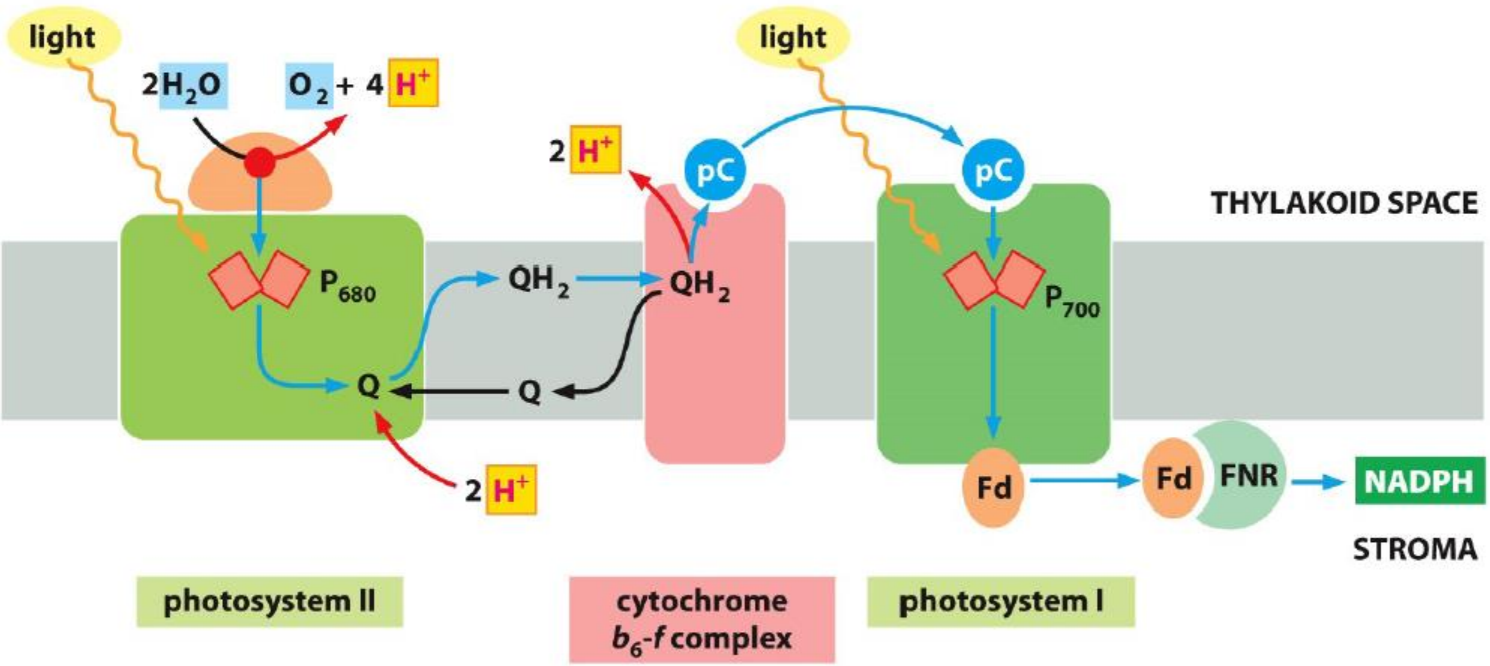
# Cytochrome *b6-f* system connects PS II to PS I

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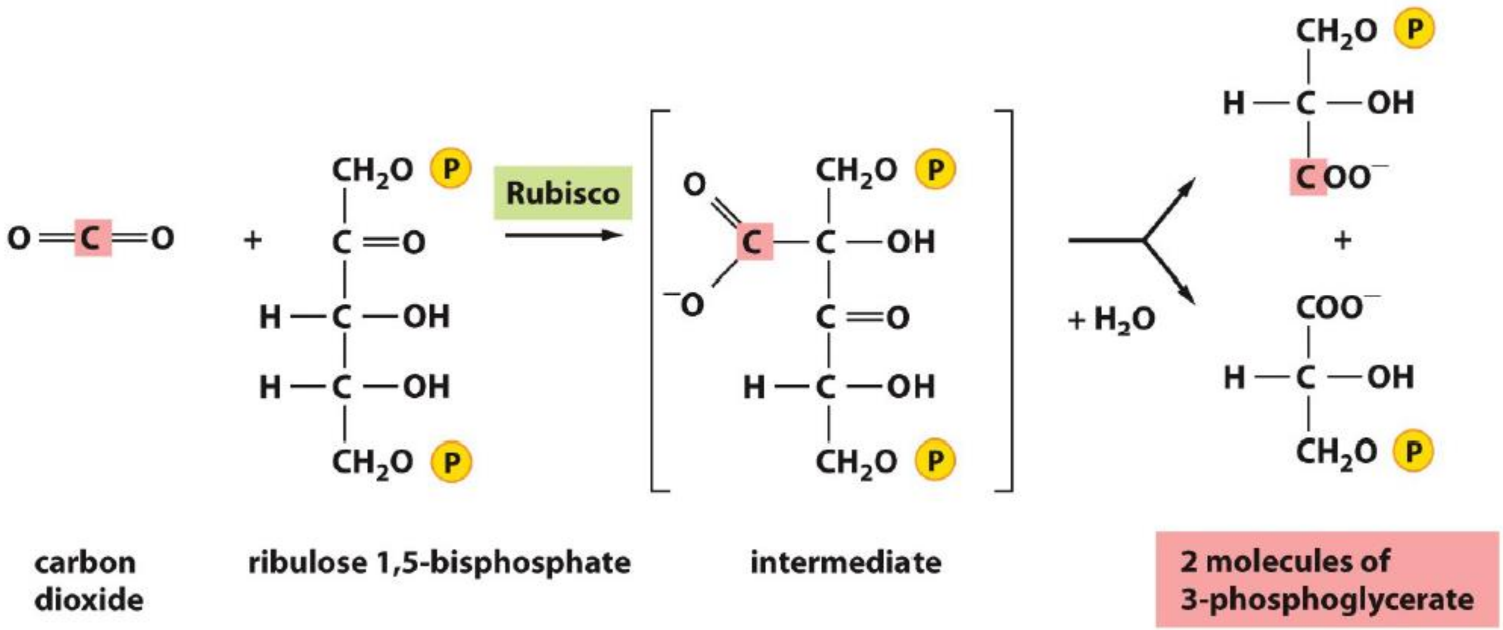
# ATP and NADPH are used to convert CO2 into sugar

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