Notes 12/5

Wednesday, December 05, 2007
10:02 AM

FINAL SLIDES

Oxidative Phosphorylation

Dec. 5, 2007

Oxidative Phosphorylation

Dec. 5, 2007

1

Announcements

- You may only take the final for the section you are enrolled in
- Friday
  - Last day of lecture
  - Lecture evaluations

Announcements

- You may only take the final for the section you are enrolled in
- Friday
  - Last day of lecture
  - Lecture evaluations

2

Overview

- FAQ
- Electron transport chain
- Oxidative Phosphorylation
- ATP Synthase

Overview

- FAQ
- Electron transport chain
- Oxidative phosphorylation
- ATP Synthase

3

OAA can be converted into PEP, so why can't this [acetylCoA] be used for gluconeogenesis in vertebrates?

- OAA + GTP <--> PEP + CO_2 + GDP

Slide 10 from Nov. 30th lecture

AcetylCoa must go through turn of TCA cycle to become OAA and carbons are lost so it isn't a conversion to PEP.

Glyoxolate pathway can (not in humans though)

OAA can be converted into PEP, so why can't this [acetylCoA] be used for gluconeogenesis in vertebrates?

- OAA + GTP <--> PEP + CO_2 + GDP

Slide 10 from Nov. 30th lecture

4

FAD and the TCA Cycle enzymes

- FAD is covalently linked to succinate dehydrogenase
- FAD is NOT covalently linked to succinyl-CoA
- Please revise your Nov. 27th lecture notes (slide 20)

FAD and the TCA Cycle enzymes

- FAD is covalently linked to succinate dehydrogenase
- FAD is NOT covalently linked to succinyl-CoA
- Please revise your Nov. 27th lecture notes (slide 20)

5

Electron Transport Chain

- Chain of redox-active cofactors
- Why use a chain?
  - More energetically efficient to pass electrons through bonds rather than physically through space
- Transferring electrons through bonds rather than space is more efficient
- Electron tunneling is a term used to describe this process

Electron Transport Chain

- Chain of redox-active cofactors
- Why use a chain?
- Transferring electrons through bonds rather than space is more efficient
- Electron tunneling is a term used to describe this process

6

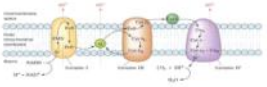
Complex I (NADH-Coenzyme Q Reductase)

*Missing picture of complexes I, III, and IV

- Passes electrons from NADH to CoQ (coenzyme Q)
- 850 kD (largest complex)
- Flavin mononucleotide (FMN) redox-active prosthetic group
- Iron-sulfur clusters
  - 6 to 7 of them
  - Redox active
  - Help in electron-transport

Iron is used because it has multiple oxidation states Fe^{2+} and Fe^{3+}

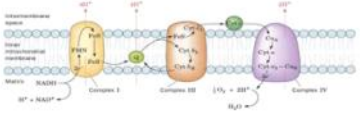
Complex I (NADH-Coenzyme Q Reductase)



- Passes electrons from NADH to CoQ
- 850 kD (largest complex)
- Flavin mononucleotide (FMN) is a redox-active prosthetic group
- Iron-sulfur clusters
  - 6 to 7 of them
  - Redox-active groups
  - Help in electron-transport

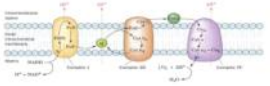
7

The Coenzymes of Complex I are FMN and CoQ



- FMN and CoQ
  - Each have 3 oxidation states
  - Can transfer 1e- or 2 e-
  - Each form stable free radicals

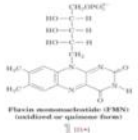
The Coenzymes of Complex I are FMN and CoQ



- FMN and CoQ
  - Each have 3 oxidation states
  - Can transfer 1e- or 2 e-
  - Each form stable free radicals

8

Figure 22-17a
Oxidation states of the coenzymes of complex I. (*a*) FMN.



- Looks a bit like FAD
- Different because FAD has AMP group connected to phosphate group while FMN doesn't have this

Figure 22-17a
Oxidation states of the coenzymes of complex I. (*a*) FMN.

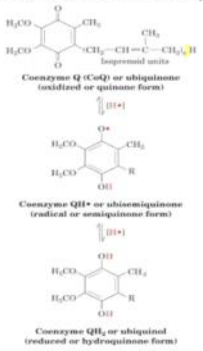
9

Figure 22-17b
Oxidation states of the coenzymes of complex I. (*b*) CoQ.

- Hydrophobic tail
- Mammals n = 10
- Others n = 6 or 8

10

Figure 22-17b
Oxidation states of the coenzymes of complex I. (*b*) CoQ.

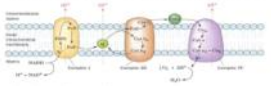


CoQ aka ubiquinone (named because it is ubiquitous)

- Hydrophobic tail (soluble in inner mitochondrial membrane)
- Mammals n = 10
- Others n = 6 or 8

- Reason they have multiple oxidation states is because radicals are stable.

Complex II (Succinate-Coenzyme Q Reductase)



- Complex II not shown above
- 127 kD (smallest complex)
- Passes electrons from succinate ($FADH_2$) to CoQ
- Associated with proton pumping
- Contains succinate dehydrogenase & other small hydrophobic subunits
- Doesn't involve enough energy for ATP production

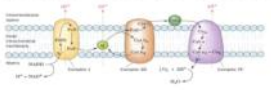
11

Complex II (Succinate-Coenzyme Q Reductase)

*Missing picture (complex II not shown in picture though)

- 127 kD (smallest complex)
- Passes electrons from succinate ($FADH_2$) to CoQ
- Contains succinate dehydrogenase & other small hydrophobic subunits
- Doesn't involve enough energy for ATP production
- Associated with proton pumping

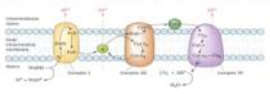
The Coenzymes of Complex II are FAD and Fe-S clusters



- FAD is covalently bound to succinate DH
- Two Fe-S clusters
- Cytochrome b_{560} is also present

12

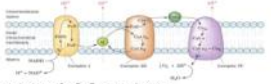
The Coenzymes of Complex II are FAD and Fe-S clusters



- FAD is covalently bound
- Two Fe-S clusters
- Cytochrome b_{560} is also present

Function to transfer electrons (because Fe has multiple oxidation states)

Complex III (Coenzyme Q-Cytochrome c Reductase)



- Passes electrons for CoQ to cytochrome c
- Transfer of electrons from Q to Complex III is associated with proton pumping (discuss later if there's time)
  - Redox loop mechanism or
  - proton pump mechanism
- Coenzymes
  - b-cytochromes
  - one cytochrome c_1
  - Fe-S clusters

13

Complex III (coenzyme Q-Cytrochrome c Reductase)

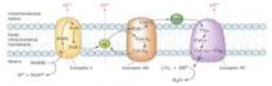
- Passes electrons for CoQ to cytochrome c
- Transfer of electrons of Q to Complex III is associated with proton pumping (discuss later if there's time)
  - Redox loop mechanism or
    - Thought that there is a method that protons bound to coenzyme q are the ones that get through mitochondria membrane and are pumped into of inner membrane space
  - Proton pump mechanism
    - Protons go through complex III itself. Somehow complex III changes shape and protons pump through complex III itself
  - Just know that protons are somehow transferred into inner membrane space.
- Coenzymes
  - b-cytochroms
  - One cytochrome c_1
  - Fe-S clusters

What is a cytochrome?

- An electron transport heme protein
- Found in most organisms
- Heme alternates between Fe (II) and Fe (III) during e- transport
- Cytochrome identity can be based on absorption spectrum
- Outer membrane surface protein or transmembrane protein
- Three types: cytochromes a, b, and c

14

Complex III (Coenzyme Q-Cytochrome c Reductase)



- Passes electrons for CoQ to cytochrome c
- Coenzymes
  - b-cytochromes
  - one cytochrome c_1
  - Fe-S clusters

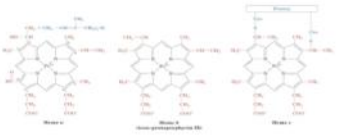


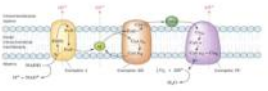
Figure 22-22a Porphyrin rings in cytochromes. (*a*) Chemical structures.

15

What is a cytochrome?

- An electron transport heme proteins
- Found in most organisms
- Heme alternates between Fe (II) and Fe (III) during e- transport Cytochromes have heme
- Cytochrome identity can be based on absorption spectrum Name cytochrome (chrome refers to color)
- Outer membrane surface protein or transmembrane protein
- 3 types: cytochroms a, b, and c

Cytochrome c



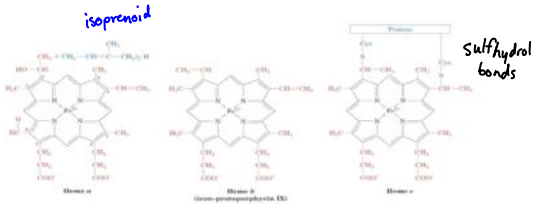
- Peripheral membrane protein
- Loosely bound to outer surface of inner mitochondrial membrane
- Shuttle electrons between Cyt c_1 and complex IV

16

Cytochromes a, b, c get their names from the porphyrin rings they have

(3 isoprenoids = farnesil)

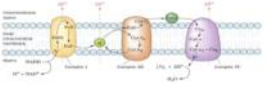
isoprenoid



Sulfhydrol bonds

hemoglobin & myoglobin

Complex IV (Cytochrome c Oxidase)

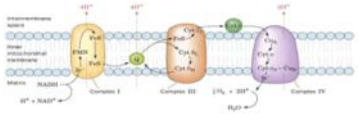


- A.k.a. COX
- Transfers electrons from Cyt c to O_2
- 4Cytochrome $c^{2+} + 4H^+ + O_2 \rightarrow 4$Cytochrome $c^{3+} + 2H_2O$
- Coenzymes = cyt a, cyt a_3, two Cu ion centers
- 2 protons pumped out per 1/2 O_2; 4 protons pumped out per O_2

17

Figure 22-22a Porphyrin rings in cytochromes. (*a*) Chemical structures.

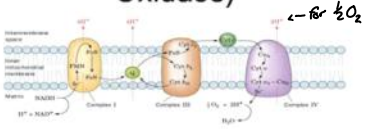
Cytochrome c



- Peripheral membrane protein
- Shuttle electrons between Cyt c_1 and complex IV
- Loosely bound to outer surface of inner mitochondrial membrane

Complex IV (Cytochrome c Oxidase)

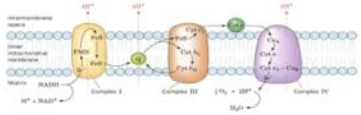
AKA: COX



- Transfers electrons from Cyt c to O_2
- Coenzymes = cyt a, cyt a_3, ~~Cu ions~~ 2 Cu ions centers

- Terminal complex in electron transport chain (O_2 is very good oxidizing agent and is terminal)
- $4 Cytochrome\ c^{2+} + 4H^+ + O_2 \rightarrow 4 Cytochrome\ c^{3+} + 2H_2O$
  - O_2 is reduced with 4 electrons
- 2 copper containing subunits (a and b)
- 2 cytrochrome units
- 2 protons pumped out per 1/2 O_2; 4 protons pumped out per O_2

What happens to the energy of electron transfer?

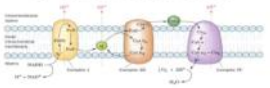


- The energy is conserved in a proton gradient
- ATP synthesis is then possible by proton flow through specific protein channels
- Chemiosmotic theory helps describe this

Slide 19: Missing: Oxidative Phosphorylation
- Endergonic (requires energy) synthesis of ATP from ADP and P_i in mitochondria
- Catalyzed by ATP synthase (Complex X)
- Energy coupling makes this posible

Slide 20 Missing: What is energy Coupling?
- Several hypotheses
  - Conformational coupling hypothesis (Paul Boyer of UCLA)
  - Chemiosmotic hypothesis (Peter Mitchel)

What happens to the energy of electron transfer?



- The energy is conserved in a proton gradient
- ATP synthesis is then possible by proton flow through specific protein channels

18

Oxidative Phosphorylation

- Endergonic synthesis of ATP from ADP and P_i in mitochondria
- Catalyzed by ATP synthase (Complex V)
- Energy coupling makes this all possible

19

What is energy coupling?

- Several hypotheses
  - Conformational coupling hypothesis (Paul Boyer of UCLA)
  - Chemiosmotic hypothesis (Peter Mitchell)

20

Chemiosmotic model was proposed by Peter Mitchell

- Paradigm for the chemical mechanism that couples proton flux with phosphorylation
- Electrochemical energy across mitochondrial membrane derived from
  - a.k.a. proton motive force
    - Chemical gradient (difference in [H+])
    - Electrical gradient (separation of charges)
  - drives ATP synthesis

21

Evidence that support chemiosmotic hypothesis

- Oxidative phosphorylation requires intact inner mitochodrial membrane
- Inner mitochondrial membrane is impermeable to ions
- Measurable electrochemical gradient across the mitochondrial membrane is seen
- Increasing inner mitochondrial membrane permeability inhibits ATP synthesis
- Increasing the acidity of intermembrane space stimulates ATP synthesis

22

Chemiosmotic model was proposed by Peter Mitchell

- Paradigm for the chemical mechanism that couples proton flux with phosphorylation
- Electrochemical energy across mitochondrial membrane derived from
  - a.k.a. proton motive force
    - Chemical gradient (difference in [H+])
    - Electrical gradient (separation of charges)

    Matrix and intermitochondrial space
  - drives ATP synthesis

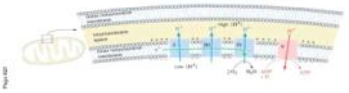
ATP Synthase (Complex V)

- Involved in ATP production
- Inner mitochondrial transmembrane complex
- Multisubunit structure

23

- Slide 22 Missing: Evidence that support chemiosmotic hypothesis
  - Oxidative phosphorylation requires intact innre mitochondrial membrane
  - Inner mitochondrial membrane is impermeable to ions
  - Mesearable electrochemical gradient across the mitochondrial membrane is seen
  - Increasing inner mitochondrial membrane permeability inhibits ATP synthesis
  - Increasing the acidity of intermembrane space stimulates ATP syntesis.

Figure 22-29 Coupling of electron transport (*green arrow*) and ATP synthesis.



24

ATP Synthase (COMPLEX V)

- Involved in ATP production
- Inner mitochondrial complex
- Multisubunit structure

Figure 22-43 Model of the *E. coli* F_1F_0–ATPase.

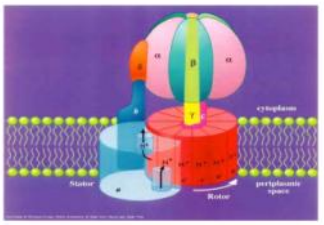
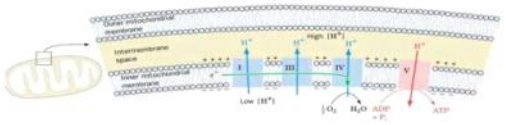


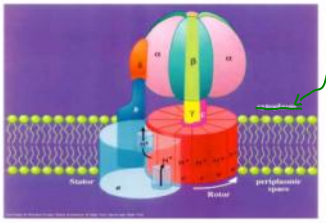
Figure 22-29 Coupling of electron transport (*green arrow*) and ATP synthesis.



- Inner membrane space is more acidic than matrix
- Electrochemical gradient (more positive charge in inner membrane space than matrix)

Figure 22-43 Model of the *E. coli* F_1F_0–ATPase.

Complex 5



Lolipop structure

Mechanically turns like rotor