Chem 306
Chapter 21 – Bioenergetics
Lecture Outline III

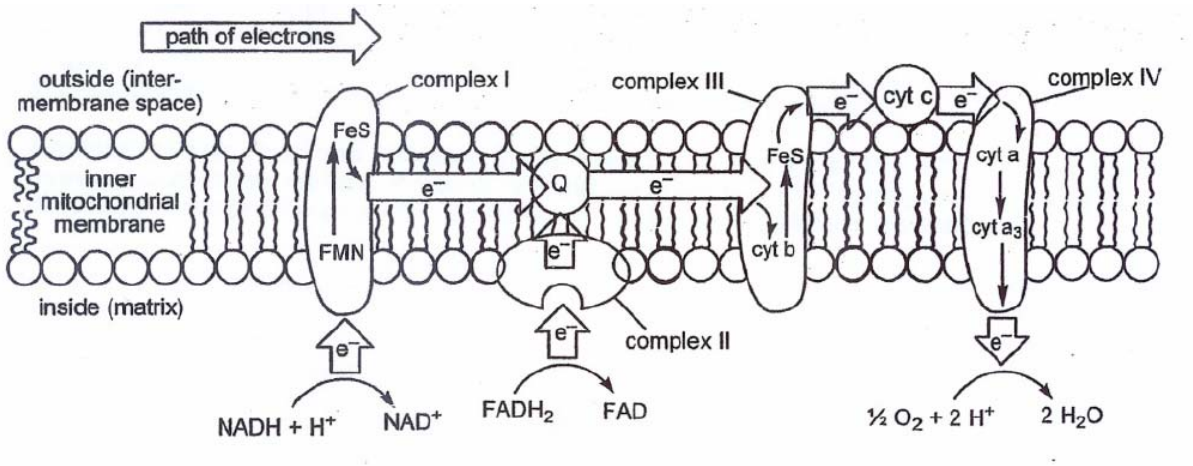
I. HOW IS ATP GENERATED IN THE FINAL STAGE CATABOLISM?

A. OVERVIEW

1. At the end of the citric acid cycle, all six carbons of glucose have been oxidized to CO_2 and a few of the nucleotide phosphates (ATP and GTP). Most of the energy from the oxidation of glucose has been saved in the reduced coenzymes NADH and $FADH_2$.

2. Following the citric acid cycle is the final oxidative pathway known as the electron transport chain.

3. What is the electron transport chain?



4. The e are passed from one electron acceptor to the next **(electron transport).** The final electron acceptor is O_2. The net reaction is $4H^+ + 4\,e + O_2 \rightarrow 2\,H_2O$ + energy. The energy of this reaction is released in small increments.(By itself this reaction is explosive!)

5. Three of the protein complexes in electron transport act as proton (H+) pumps to move protons into the inner membrane space which produces a proton (H+) gradient. Where do the protons come from?

6. The only path back into the matrix for H+ is by way of the enzyme complex ATP synthase. As the H+ pass through this enzyme complex,energy is released and is used to synthesize ATP from ADP and Pi (a process called **oxidative phosphorylation**).

7. As long as oxygen is available for the mitochondrion in the cell, electron transport and oxidative phosphorylation function to produce most of the ATP energy manufactured in the cell.

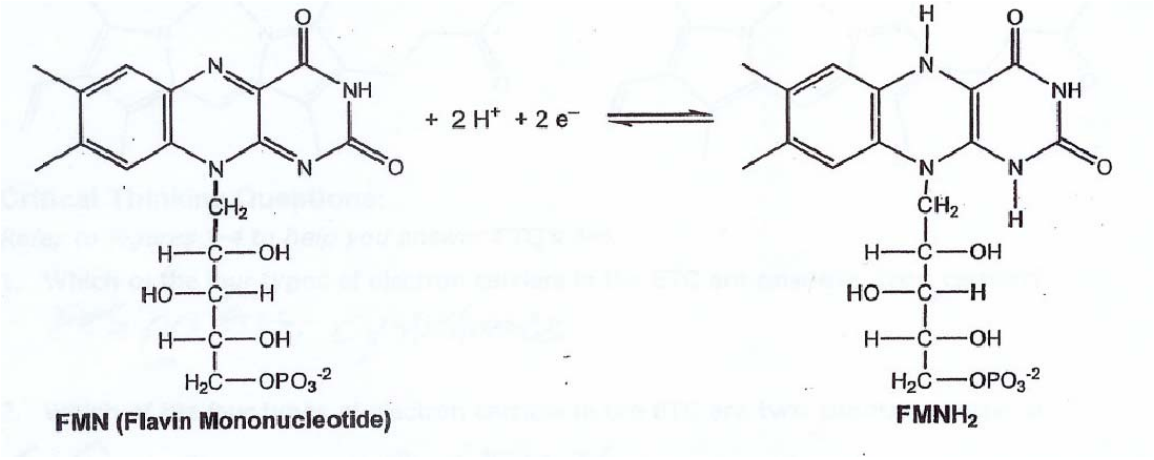
B. SPECIFICS

1. **Electron Carriers**

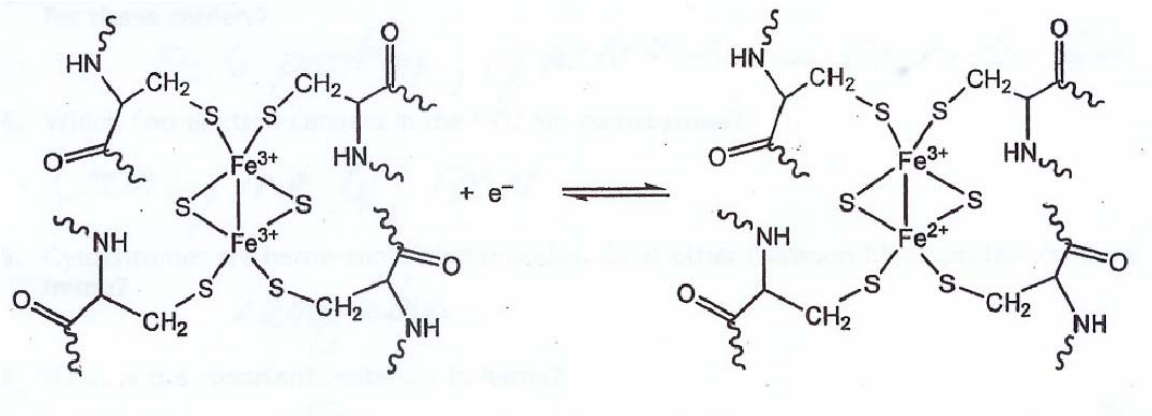
B. The sequence of reactions in electron transport utilizes electron carriers. Each type of electron carrier contains a group or ion that is reduced and then oxidized as the electrons are accepted and then passed on.

B. There are four electron carriers that make up the electron transport system.

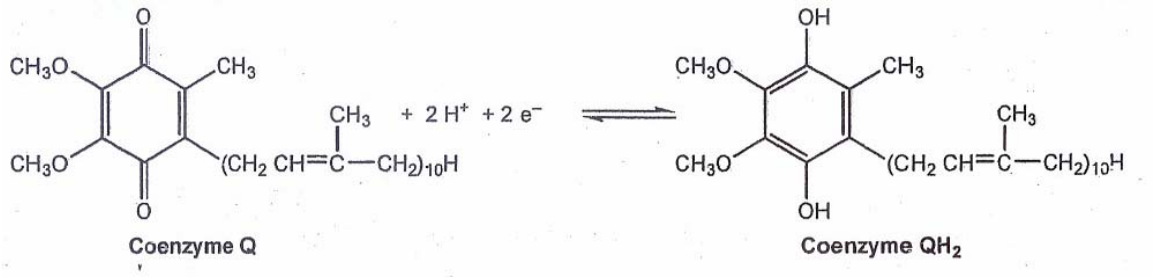
1. **FMN (flavin mononucleotide)** – a coenzyme that is derived from riboflavin (vitamin B2). FMN contains a flavin ring system that is also found in FAD.



2. **Fe-S** – Fe-S clusters is the name given to a group of iron-sulfur proteins that contain iron-sulfur clusters embedded in the proteins of the electron transport chain. The iron ions in the clusters cycle as Fe 2+ and Fe 3+ as electrons are accepted and lost.

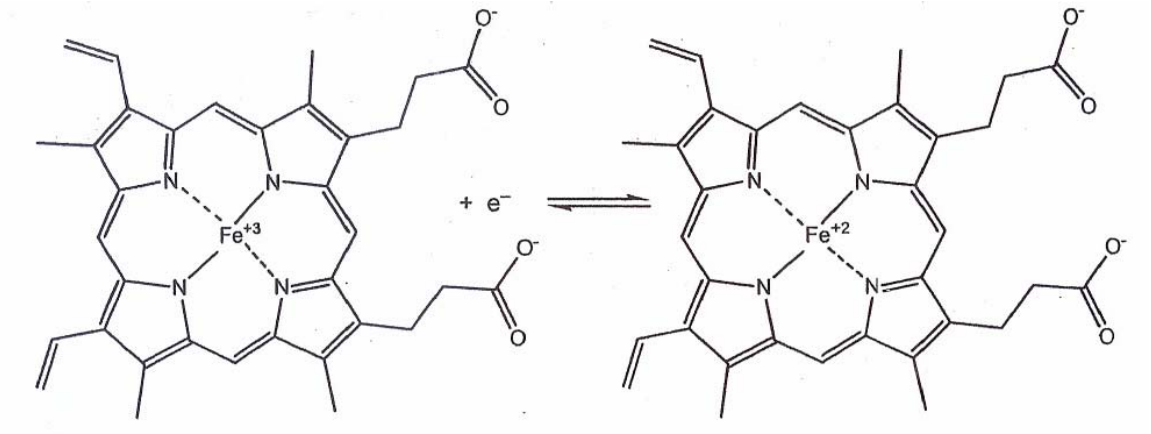


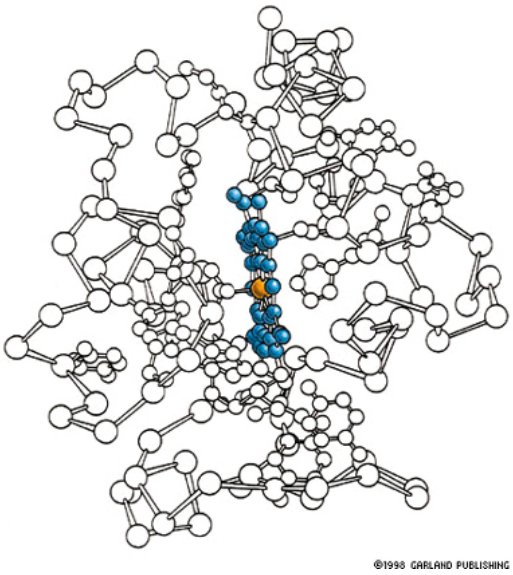
3. **Coenzyme Q (CoQ**), sometimes called ubiquinone, is derived from quinone which is a six carbon cyclic compound with two double bonds and two keto groups attached to a long carbon chain. CoQ is reduced to $CoQH_2$ when the keto groups of quinone accept hydrogen ions and electrons.



4. **Cytochromes** – Cytochromes are proteins that contain an iron ion in a heme group. The different cytochromes are indicated by the letters following the abbreviation for cytochrome (cyt*1*, cyt *b*, etc). In each cytochrome the Fe 3+ accepts a single electron to form Fe 2+, which is oxidized back to Fe 3+ when the electron is passed to the next cytochrome.

$$Fe^{3+} + 1e \rightleftharpoons Fe^{2+}$$





General Cytochrome Structure

**** Questions to ponder ****

1. Is $FMNH_2$ the abbreviation for the oxidized or reduced form of flavin mononucleotide?

2. *Is cyt b (Fe 3+) the abbreviation of the oxidized or reduced form of cytochrome b?*

3. *Identify the following as oxidation or reduction*

$$Q + 2H+ + 2e \rightarrow QH_2$$

$$Fe^{2+} \text{ - S cluster} \rightarrow Fe^{3+} \text{ - S cluster} + e$$

4; Which of the four types of electron carriers in the ETC are one-electron carriers? two-electron carriers?

5. *Which two-electron carriers are proteins? What metal ion cofactor is required for these carriers?*

6. *Which two-electron carriers are coenzymes?*

7. *Cytochromes are heme-containing proteins. What other common blood protein contains heme?*

2. Electron Transport

A. Along and embedded in the inner membrane of the mitochondria are protein complexes containing the enzymes and electron carriers required for electron transport.

B. Two electron carriers, CoQ and cyt *c*, are not firmly attached to the membrane – they are mobile electron carriers shuttling electrons between the protein complexes that are bound to the membrane.

C. There are four distinct protein complexes containing enzymes and electron carriers needed for electron transport.

1. The four protein complexes needed for electron transport are:
   Complex I – NADH Dehydrogenase
   Complex II – Succinate Dehydrogenase
   Complex III – Coenzyme Q – Cytochrome c Reductase
   Complex IV – Cytochrome c Oxidase

2. ***Complex I – NADH Dehydrogenase***

   a. Contains the e carriers FMN and Fe-S clusters.

   b. All of the NADH generated in the cell transfers H+ and e to FMN. The reduced $FMNH_2$ forms while NADH is reoxidized to NAD+ which returns to oxidative pathways such as the citric acid cycle. The e are transferred from $FMNH_2$ to the Fe – S clusters and then to CoQ.

   c. Check out the reactions for Complex I!

$$NADH + H+ + FMN \rightarrow NAD + FMNH_2$$

$$FMNH_2 + Q \rightarrow QH_2 + FMN$$

Overall Rxn: $NADH + H+ + Q \rightarrow QH_2 + NAD+$

3. ***Complex II – Succinate Dehydrogenase***

   a. This complex is specifically used when $FADH_2$ is generated by the conversion of succinate to fumarate in the citric acid cycle. The electrons from FADH2 are transferred to Coenzyme Q to yield QH_2.

   b. The overall reaction at Complex II

$$FADH_2 + Q \rightarrow FAD + QH_2$$

4. ***Complex III – Coenzyme Q – Cytochrome c Reductase***

   a. The mobile carrier QH_2 transfers the electrons it has collected from NADH and $FADH_2$ to an Fe – S cluster and then to cytochrome b, the first cytochrome in complex III.

$$QH_2 + 2 \text{ cyt } b \text{ (Fe 3+)} \rightarrow Q + 2\text{cyt } b \text{ (Fe 2+)} + 2H+$$

   b. From cyt *b*, the electron is transferred to an Fe – S cluster and then to cyt c . Each time a Fe 3+ ion accepts an e , it is reduced to Fe 2+, and then oxidized back to Fe 3+ as the e is passed on to the next e acceptor.

   c. The mobile e carrier cyt *c* moves the e to Complex IV

5. ***Complex IV – Cytochrome c Oxidase***

At Complex IV, electrons are transferred between 3 cytochromes (*c, a,* and a_3). In the final step of electron transport, electrons and H+ combine with O_2 to form water.

$$4H^+ + 4\,e + O_2 \rightarrow 2\,H_2O + \text{energy}$$

**** Questions to ponder ****

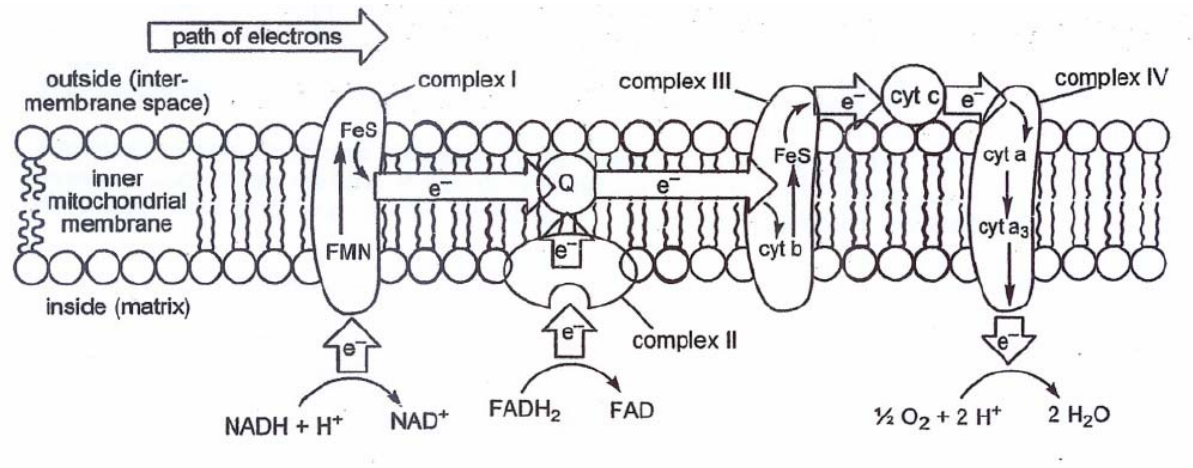
1. *What reduced coenzymes provide the electrons for electron transport?*

2. *What happens to the energy level as electrons are passed along the electron transport chain?*

3. *How are electrons carried from complex I to complex II? From complex III to complex IV?*

4. *How is NADH oxidized in electron transport? $FADH_2$?*

Pathway of Electrons in Electron Transport



3. Oxidation Phosphorylation and ATP

a. We have seen that energy is generated when the electrons from the oxidation of substrates flow through the electron transport chain.

b. This energy is coupled with the production of ATP for the cell creating the process called **oxidative phosphorylation**.

c. The energy from electron transport is linked to a proton gradient that drives the synthesis of ATP (This **chemiosmotic theory** was proposed by Peter Mitchell - Nobel Prize in 1978.)

1. Three of the complexes (I, III, and IV) extend through the inner membrane with one end of each complex in the matrix and the other end in the intermembrane space.

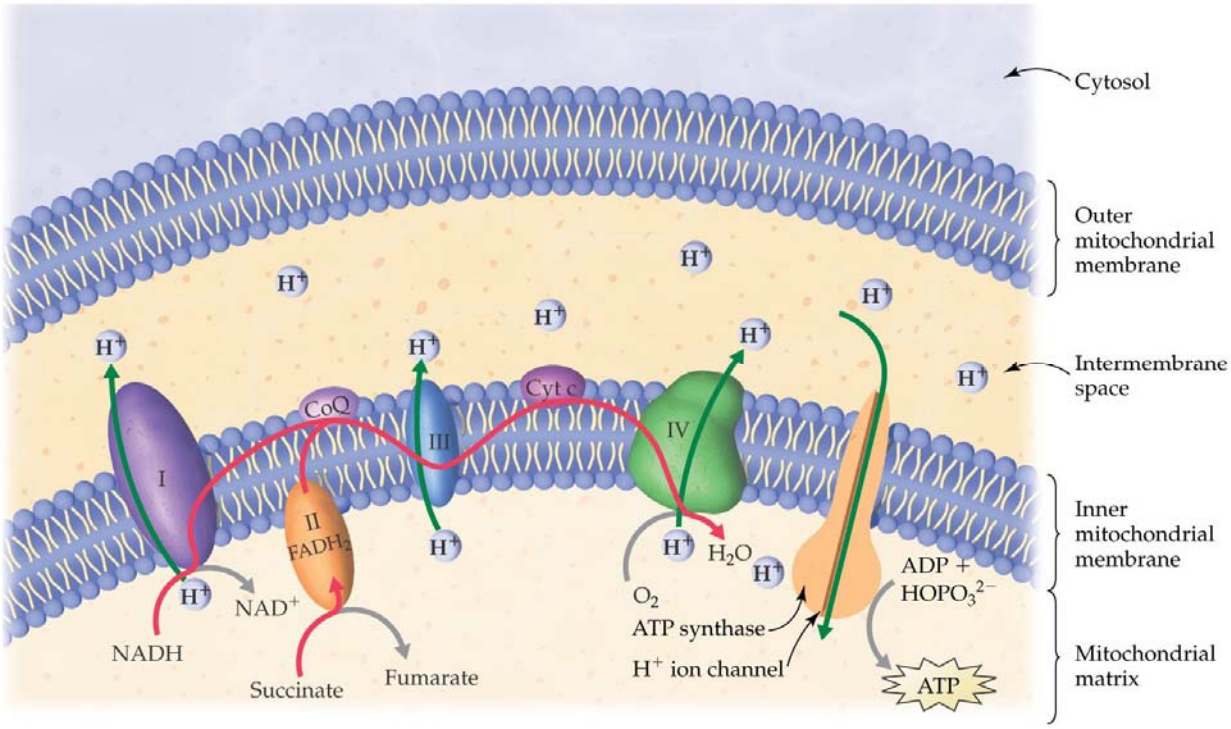
2. Each of these complexes acts as a proton pump by pushing protons (H+) out of the matrix and into the intermembrane space. This increase in H+ in the intermembrane space creates a proton gradient which is the result of the charge difference and the H+ concentration gradient. Another term for the proton gradient in an electrochemical gradient.

3. To equalize the pH between the intermembrane space and the matrix, there is a tendency for the protons to return to the matrix. However, protons cannot return to the matrix through the inner membrane.

4. The only way that H+ can return to the matrix is to pass through a protein complex called **ATP synthase**. As the protons flow through ATP synthase, energy generated from the proton gradient is used to drive the ATP synthesis. This is the coupling of the energy from electron transport to the synthesis of ATP from ADP and Pi.

$$ADP + Pi + \text{energy} \xrightleftharpoons{\text{ATP synthase}} ATP$$

The Mitochondrial Electron-Transport Chain and ATP Synthase



Animations of the proton gradient and electron transport -

http://vcell.ndsu.nodak.edu/animations/

http://vcell.ndsu.nodak.edu/animations/atpgradient/movie.htm

http://vcell.ndsu.nodak.edu/animations/etc/movie.htm

http://highered.mcgraw-hill.com/sites/0072437316/student_view0/chapter9/animations.html#

http://www.youtube.com/watch?v=QHmdJtiaNYg&mode=related&search=

C. Substances That are Respiratory Chain Inhibitors

Respiratory Chain Inhibitors	Site/Mode of Action
Rotenone (a fish poison isolated from plants)	Binds to complex I and blocks the transfer of electrons from Fe–S clusters to ubiquinone (Q)
Carboxin	Binds to complex II and blocks the transfer of electrons from $FADH_2$ to ubiquinone (Q)
Antimycin A (an antibiotic)	Binds to complex III and blocks the transfer of electrons from ubiquinol (QH_2) to Fe–S clusters
Cyanide (one of the most potent human toxins)	Blocks electron flow by binding to Fe^{3+} within cytochromes of complex IV
Carbon monoxide (a common air pollutant found in auto exhaust)	Blocks electron flow by binding to Fe^{2+} within cytochromes of complex IV
Oligomycin (an antibiotic)	Blocks the flow of H^+ through ATP synthase

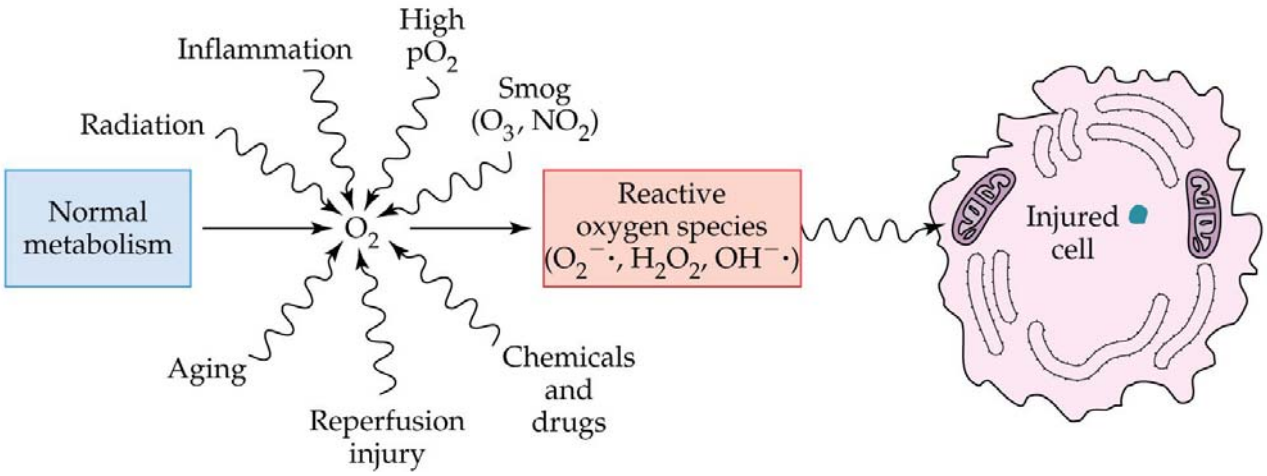
II. WHAT ARE THE HARMFUL BY-PRODUCTS PRODUCED FROM OXYGEN, AND WHAT PROTECTS AGAINST THEM?

Biochemical reactions that consume oxygen do not always produce water. Sometimes the products are reactive species such as

1. free radicals

2. hydrogen peroxide, H_2O_2

Conditions that can enhance the production of free radicals and H_2O_2 are shown below.



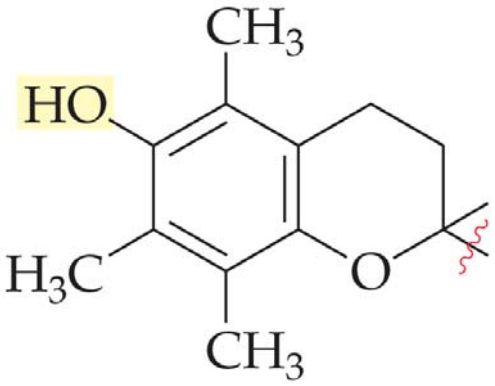
Protection from the reactive species above is provided by several enzyme systems including superoxide dismutase and catalase.

$$2\,O_2^-\cdot + 2\,H^+ \xrightarrow{\text{Superoxide dismutase}} H_2O_2 + O_2$$

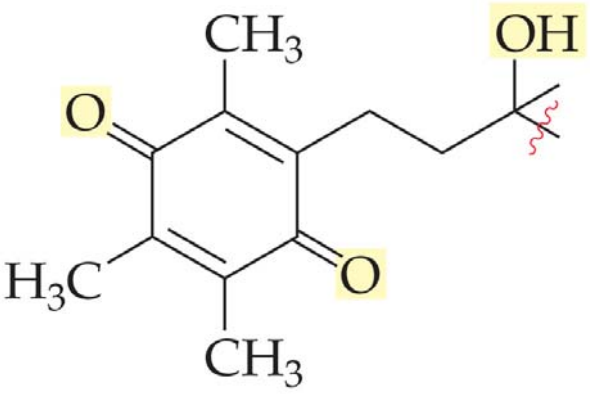
Superoxide ion — Hydrogen peroxide

$$2\,H_2O_2 \xrightarrow{\text{Catalase}} 2\,H_2O + O_2$$

Antioxidant vitamins such as vitamin C, E ,and A provide protection from free radicals.



Vitamin E



Product of vitamin E reacting as an antioxidant