Chapter 9 Part A Lecture Notes: Metabolism – Generation of Energy
“Metabolism is fundamental”

I. Introduction
Metabolism is the total of all chemical reactions in the cell
A. Catabolism = breakdown of complex molecules to simple ones with the release of energy; some of the energy is trapped in the form of ATP
   For heterotrophs: 3 stages Fig 9-1
   1. very complex molecules → constituent parts
   2. constituent parts → simple molecules
   3. simple molecules → energy
B. Anabolism = synthesis of complex molecules from smaller ones with the input of energy from ATP (Chapter 10)
C. Photosynthesis = trapping of light energy and conversion to chemical energy which is then used to reduce CO₂ to organic molecules; note that this is a combination of “catabolism and anabolism” (Chapter 9 Part B and Chapter 10)
D. Amphibolic pathways = pathways that can function both catabolically and anabolically (Fig. 9-2) (Chapter 12)

II. Catabolism of sugars in chemoorganoheterotrophs
A. Very complex molecules → constituent parts
   1. Larger polymers – for bacteria via external hydrolysis to generate di or monosaccharides that can be taken up by the cell and fed into catabolic pathways
      a) starch = a-1,4-glucose digested by amylases
      b) glycogen = a-1,4-glucose and a-1,6-glucose digested by amylases
      c) cellulose = B-1,4-glucose digested by cellulases
      d) pectin = galacturonic acid polymer digested by pectinases
   2. Reserve polymers – for bacteria via phosphorolysis (phosphate attack on bond that joins the two sugars and adds inorganic phosphate to the sugar) to generate di or monosaccharides that can be taken up by the cell and fed into catabolic pathways
      a) Glycogen and starch
      b) PBH: PHB → 3-hydroxybutyrate → acetoacetate → acetyl-CoA → to TCA cycle
   3. Disaccharide cleavage (Fig. 9-15)
      a) Examples
         (1) lactose = glc + gal
         (2) sucrose = glc + fru
         (3) maltose = glc + glc
         (4) cellobiose = B-1,4-diglucose; product of cellulose digestion
      b) two mechanisms (both may fxn in one organism)
         (1) hydrolysis
         (2) phosphorolysis
   4. Monosaccharide interconversions (Fig. 9-15)
      a) Examples: galactose, mannose, fructose
      b) yield products that feed into catabolic pathways
B. Constituent parts \(\rightarrow\) simple molecules

Many routes – we will discuss three below

In all three pathways, ATP is generated via substrate level phosphorylation (the synthesis of ATP from ADP by phosphorylation coupled with the exergonic breakdown of a high-energy organic substrate molecule)

1. Glucose \(\rightarrow\) pyruvate via Embden-Meyerhof pathway (glycolysis)
   a) Main functions
      (1) oxidation of glucose
      (2) generation of ATP
      (3) generation of precursors for biosynthesis (see attached sheet)
   b) Location: cytoplasm of most prokaryotes and mitochondrial matrix cytoplasm of eukaryotes
   c) Independent of oxygen
   d) Details: Fig. 9-3 and Appendix AII-1
      (1) 6 C stage glc \(\rightarrow\) fru1,6P
      (2) 3 C stage G3P \(\rightarrow\) pyruvate
   e) Bottom line reaction:
      \[\text{glucose} + 2\text{ADP} + 2\text{Pi} + 2\text{NAD}^+ \rightarrow 2\text{pyruvate} + 2\text{ATP} + 2\text{NAD}^+ + 2\text{H}^+\]

2. Glucose \(\rightarrow\) pyruvate via pentose phosphate pathway (or the hexose monophosphate pathway)
   a) Main functions:
      (1) intermediates can be used to produce ATP
         (a) via feeding of intermediates into glycolysis
         (b) via conversion of NADPH to NADH
      (2) generation of NADPH which serves as an energy source for reduction of molecules during biosynthesis
      (3) biosynthesis of 4 and 5 C sugars which are precursors to ribose and deoxyribose and aromatic amino acid
   b) Location: cytoplasm of some prokaryotes
   c) Independent of oxygen
   d) Details: Fig. 9-4 and Appendix AII-2
      (1) Stage 1
         (a) oxidation-decarboxylation reaction
         (b) \(3\text{Glc6P} + 6\text{NADP} + 3\text{H}_2\text{O} \rightarrow \rightarrow \)
         \[3\text{ribulose5P} + 3\text{CO}_2 + 6\text{NADPH} + 6\text{H}^+\]
      (2) Stage 2 = Isomerization to make precursors for stage 3
      (3) Stage 3 (Fig. 9-5)
         (a) sugar rearrangements via
            (i) transketolase - transfers a 2 C fragment from ketose to aldose
            (ii) transaldolase - transfers a 3 C fragment from ketose to aldose
         (b) \(2\text{xyulose5P} + \text{ribose 5P} \rightarrow 2\text{fru6P} + 3\text{glyceraldehyde3P}\)
e) Bottom line reaction
(1) Intermediates such as glc6P and glyceraldehyde 3P can be fed into glycolysis
(2) Depends on number of turns of the cycle - complete oxidation of glc6P:
\[
\text{Glucose-6-P} + 12\text{NADPH} + 7\text{H}_2\text{O} \rightarrow 6\text{CO}_2 + 12\text{NADPH} + 12\text{H}^+ + \text{Pi}
\]

f) Similar reaction in the Calvin cycle

3. Glucose $\rightarrow$ pyruvate via Entner-Doudoroff pathway
   a) Alternative pathway found in a few bacteria
   b) Details: Fig 9-6 and Appendix AII-3
   c) Bottom line: 1ATP (substrate level phosphorylation) + 1 NADH + 1 NADPH per glucose

C. Simple molecules $\rightarrow$ energy: aerobic respiration
Energy yielding processes in which organic or inorganic substances are electron donors and oxygen is electron acceptor; substrate is metabolized with the involvement of an exogenous oxidizing agent.

\[
\text{Reduced cpd} \rightarrow \text{Oxidized Intermediate} \rightarrow \text{Reduced external electron acceptor}
\]

Composed of 4 main parts for chemoheterotrophs: (1) pre-TCA (2) TCA (3) ETC and (4) oxidative phosphorylation

1. Pre-TCA
   yield per glucose = 2 NADH + H^+

\[
\text{Pyruvate} + \text{CoA} \rightarrow \text{Acetyl CoA} + \text{CO}_2
\]
2. **Tricarboxylic acid cycle (TCA or Krebs Cycle)**
   a) **Functions**
      (1) Oxidation of acetyl CoA to 2 CO₂ to generate stored energy in the form of NADH + H⁺, FADH₂, GTP
      (2) Supplies carbon skeletons for biosynthesis (see attached)
b) **Location** = cytoplasm of most proks and mitochondrial matrix of euks
c) **TCA cycle is widely distributed**; some organisms that do not use the TCA cycle to generate energy still have TCA cycle enzymes
d) **Does not require oxygen**
e) **Details** (Fig. 9-7 and Appendix II-4)
   (1) Attachment of acetyl-CoA to oxaloacetate to form citrate
   (2) Oxidation/ decarboxylations that yield CO₂ and NADH + H⁺,
      (a) #1 isocitrate → a-ketoglutarate
      (b) #2 a-ketoglutarate → succinyl CoA
   (3) succinyl CoA → → → oxaloacetate
      (a) yields GTP
      (b) yields FADH₂
      (c) yields NADH + H⁺
f) **Bottom line:** yield per glucose (2 acetyl CoA)
   (1) 6 NADH + H⁺
   (2) 2 FADH₂
   (3) 2 GTP

3. **Electron transport chains (ETC)**
   a) **Function**
      (1) ETC can generate a proton motive force (PMF) by oxidation of NADH + H⁺ and FADH₂ → PMF is then used as an energy source to make ATP
      (2) ETC can also work in reverse to generate NADH + H⁺ at the expense of PMF
b) **Proton motive force (PMF)**
   (1) An energized state of a membrane due to the unequal distribution of protons
   (2) composed of both chemical (delta pH) and an electrical (difference in charge) energy
   (3) generated by electron transport chain (but is affected by other cellular activities that alter proton concentration and charge distribution)
c) **Location:** in the plasma membrane for proks and the mitochondrial inner membrane for euks.
d) Components of ETC (electron carriers)

* Note that there is not one universal set of components in the ETC. The makeup varies depending on the organism and the conditions in which an organism is growing.

1) NADH dehydrogenases
   a) Mitochondrial (hydrogen carriers)
      i) 40 protein subunits
      ii) 4 FMN
      iii) Ubiquinones
      iv) Nonheme proteins (Fe-S)
   b) Bacterial
      i) Many have less protein subunits
      ii) Otherwise similar
2) flavoproteins (hydrogen carriers)
3) cytochromes (electron carriers)
4) non heme iron proteins – usually Fe-S proteins (electron carriers)
5) oxidases - special cytochromes that donate their electrons to oxygen
6) quinones (hydrogen carriers)

e) General details (coupling redox reactions to proton translocation):
   Protons are translocated to the periplasm (or mitochondrial inner membrane space) by several mechanisms during redox reactions in ETC

1) Loops
   a) based on the fact that both hydrogen and e- carriers are involved in redox reactions and alternate in the chain
   b) 2 hydrogens are taken up by hydrogen carrier \( \rightarrow \) electrons are passed to electron carrier at the outer surface and protons are released at the outer surface \( \rightarrow \) electron carrier will pass 2e- to hydrogen carrier which will then pick up to 2 protons to make hydrogens \( \rightarrow \)
2) Proton pumping – electron carriers actually pump protons outside during redox reactions (i.e NADH dehydrogenase)
3) Q cycles
   a) reduced quinones carries 2 protons and 1 electron across the membrane releasing the protons on the external face of the membrane \( \rightarrow \) becomes oxidized to semiquinone \( \rightarrow \) semiquinone picks up 2 more protons and the other electron \( \rightarrow \) repeat

f) Details of eukaryotic electron transport in mitochondria (see attached)

Details of prokaryotic electron transport (see attached)

h) Bottom line depends on the exact ETC
   1) H/O = number of protons that are translocated though all the available sites in the ETC per O reduced; varies depending on ETC
   2) H/O about 10 in mitochondria; usually less for bacteria
4. **Oxidative phosphorylation**
   a) Function: Use of the PMF generated by the ETC to make ATP using ATP synthase
   b) Location: in the plasma membrane for proks and the mitochondrial inner membrane for euks
   c) Details- see attached
      (1) Energy may be required for release of ATP (not for actual synthesis)
      (2) When protons move down the gradient through ATP synthase, a conformational change in the F1 subunits causes releases ATP from the high energy/high affinity site
   d) Bottom line: $H/P = \#$ of protons required to flow down ATP synthase to generate 1 ATP = measured between 2-4

5. Net energy per NADH + H+ varies among organism
   \[ P/O = \text{number of ATP produced per O reduced} = P/O = (P/H)*(H/O) \]

D. Simple molecules $\rightarrow$ energy: **Anaerobic respiration**

Energy yielding processes in which organic or inorganic substances are electron donors and an external substance (either inorganic or organic but not oxygen) is electron acceptor; substrate is metabolized with the involvement of an exogenous oxidizing agent.

1. Similar to above for ETC except terminal electron acceptors can be NO₃, SO₄, CO₂, fumarate, etc.
2. N sources as electron acceptors (Fig. 40-9)
   a) NO₃ as electron acceptor
      (1) Pseudomonads, Alkaligenes, some enteric bacteria
      (2) Dissimilatory nitrate reduction
      \[ \text{NO}_3^- + 2\text{e}^- + 2\text{H}^+ \rightarrow \text{NO}_2 + \text{H}_2\text{O} \]
      (3) Reaction is usually coupled with others because limited energy generates and nitrite is toxic thus denitrification
   b) Denitrification
      (1) reduction of nitrate to gaseous products; depletes soil of essential N
      (2) see attached
      (3) bottom line: \[ 2 \text{NO}_3^- + 10\text{e}^- + 12\text{H}^+ \rightarrow \text{N}_2 + 6\text{H}_2\text{O} \]
3. SO₄
4. CO₂
   a) Methanogens (Ch. 20)
   b) Cofactors that carry the electrons are distinct from those described above
      \[ 4\text{H}_2 + \text{CO}_2 \rightarrow \text{CH}_4 + 2\text{H}_2\text{O} \]
5. fumarate
6. Usually energy yield is less for all these, but niches that they can inhabit gives them a competitive advantage
E. Simple molecules $\rightarrow$ energy: Fermentative catabolism

Energy yielding processes in which molecules within the catabolic pathway serve as both electron donors and acceptors; substrate is metabolized without the involvement of an exogenous oxidizing agent

1. Glycolysis is first step $\rightarrow$ then classified by end products
2. Location = cytoplasm
3. Occurs in the absence of oxygen
   a) For organisms that break down glucose as above, the lack of an external electron acceptor results in the accumulation of reduced NADH + H+, which will eventually result in the depletion of NAD+.
   b) Thus, inhibition of pyruvate dehydrogenase allows the use of pyruvate derivatives as electron acceptors in redox reaction to regenerate oxidized NAD+.
4. Types (Fig. 9-14)
   a) Lactic acid
      (1) Homolactic (Fig. 9-14-1)
         (a) Product: only lactic acid
         (b) Starts with glycolysis and then reduces pyruvate to lactic acid
         (c) Lactococcus, Lactobacillus, Streptococcus, Bacillus
      (2) heterolactic (Fig 23-14) –
         (a) product: lactic acid plus others
         (b) starts with pentose phosphate pathway $\rightarrow$ uses G3P to feed into the end of glycolysis as above to generate lactic acid; also bi-product acetyl phosphate can be converted to acetyl CoA and used to make acetaldehyde and ethanol as for (b)
         (c) Leuconostocs
   b) Straight alcoholic (Fig. 9-14-2)
      (1) product: ethanol
      (2) decarboxylation of pyruvate to generate acetaldehyde
      (3) reduction of acetaldehyde to generate ethanol
      (3) mostly yeast, some bacteria
c) Propionic (Fig. 9-14-3)
   (1) product: propionate
   (2) Pyruvate $\rightarrow$ propionate
   (3) Propionic acid bacteria – swiss cheese

d) Formic acid (Fig. 9-14-5 and additional paths)
   Products always include formate (and other acids)
   (1) mixed acid- products are formate plus acetate, lactate, succinate
   (2) butanediol- products are formate, lactate, and 2,3-butanediol

e) Homoacetate
   (1) Acetate is the only product
   (2) Some Clostridium

5. Actual generation of energy
   a) Most important = substrate level phosphorylation
   b) Much rarer
      (1) decarboxylases that are proton pumps to generate a PMF (see attached)
      (2) syntrophy – coupling of reaction that at standard conditions would not be energetically favorable with reaction that removes products to drive reaction (see attached)

6. Anaerobic food chain (Fig. 40-7)
   a) Important for the regeneration of gaseous carbon
   b) CHO $\rightarrow$ organic acids, alcohol, acetate, H$_2$, CO$_2$ by fermenters
   c) H$_2$, CO$_2$ $\rightarrow$ CH$_4$, CO$_2$ by methanogens
   d) CO$_2$ $\rightarrow$ organic material by phototrophs
III. Catabolism of lipids in chemoorganoheterotrophs
   A. Lipid structure (Fig. 9-16)
   B. Very complex → constituents
      For bacteria: lipases break triglycerols into glycerol and fatty acids
   C. Constituents → simple molecules → energy
      1. Glycerol is oxidized to dihydroxyacetone phosphate → into glycolysis
      2. Fatty acids are degraded to acetyl CoA via β-oxidation path (Fig. 9-17)
         1 turn (2 C removes) produces:
            a) acetyl CoA → fed into TCA cycle
            b) 1 NADH + H+ and 1 FADH₂ → for ETC

IV. Catabolism of proteins in chemorganoheterotrophs
   A. Amino acid / protein structure (Appendix I-14-16)
   B. Very complex → constituents to the cell
      For bacteria: secreted proteases break down proteins into amino acids that are transported into cell
   C. Constituents → simple molecules → energy
      1. Deamination of amino acids to generate organic acids (frequently via transamination Fig. 9-18 and attached) → Organic acid can be converted into an intermediate for use in the TCA cycle
      2. Amino acids oxidases - flavoproteins that oxidize amino acids → e- to ETC
      3. Amino acid dehydrogenases