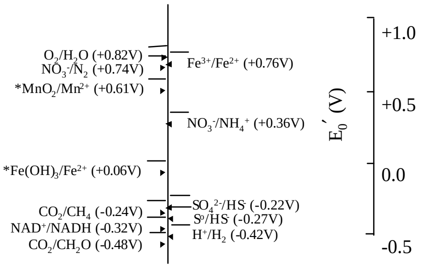
Review of Reduction Potential Concepts & Calculations

- Convention for use of redox tower (e.g. E_0' values) to predict the feasability of coupled half reactions
- Calculation of reduction potentials for coupled half reactions (e.g. $\Delta E_0'$ values)

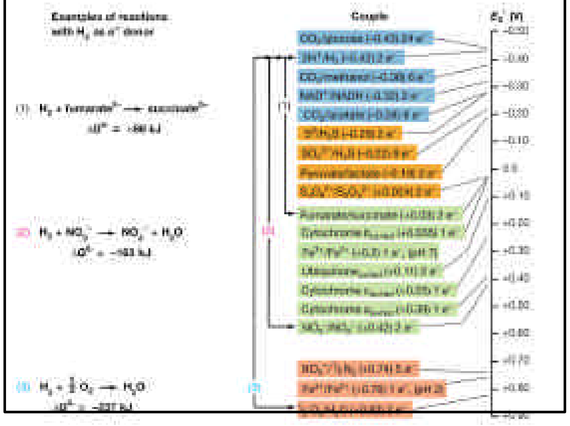
Review of Reduction Potential Concepts

- In order to use reduction potentials (e.g. E_0' values) to assess the potential feasability of coupled redox reaction, all half-reactions must be written as reduction reactions

“Redox Tower” for Microbial Metabolic Reactions



* With $\{Mn^{2+}\}$, $\{Fe^{2+}\}$ = 10^{-6} M



Use of Reduction Potentials as a Gauge for Feasability of Coupled Half Reactions

- Examples of unfeasible:

  1. Oxidation of Fe^{2+} coupled to SO_4^{2-} reduction to HS^-

  $Fe(OH)_3 + e^- \rightarrow Fe^{2+}$ E_0' = +0.06V ($\{Fe^{2+}\}=10^{-6}$ M)

  $0.125SO_4^{2-} + e^- \rightarrow 0.125HS^-$ E_0' = -0.22V

  2. Oxidation of $succinate^{2-}$ coupled reduction of CO_2 to $acetate^-$

  $fumarate^{2-} + 2e^- \rightarrow succinate^{2-}$ E_0' = +0.03V

  $0.5CO_2 + 2e^- \rightarrow 0.5acetate^-$ E_0' = -0.28V

- But what if we think of these coupled reactions as going the other way…?

Use of Reduction Potentials as a Gauge for Feasability of Coupled Half Reactions

- Examples of feasible:

  1. Oxidation of Fe^{2+} coupled NO_3^- reduction to N_2

  $Fe(OH)_3 + e^- \rightarrow Fe^{2+}$ E_0' = +0.06V ($\{Fe^{2+}\}=10^{-6}$ M)

  $0.2NO_3^- + e^- \rightarrow 0.125N_2$ E_0' = +0.74V

  2. Oxidation of $acetate^-$ coupled to SO_4^{2-} reduction to HS^-

  $2CO_2 + 8e^- \rightarrow acetate$ E_0' = -0.28V

  $SO_4^{2-} + 8e^- \rightarrow HS^-$ E_0' = -0.22V

Calculation of Reduction Potentials for Coupled Half Reactions

- The overall reduction potential for two coupled reactions is equal to the difference in the E_0' value for the two half reactions ($\Delta E_0'$)
- Example:

$2H^+ + 2e^- \rightarrow H_2$ $E_0' = -0.42V$

fumarate^{2-} + $2e^- \rightarrow$ succinate^{2-} $E_0' = +0.03V$

$\Delta E_0' = +0.03 - (-0.42) = 0.45V$

Calculation of Reduction Potentials for Coupled Half Reactions

- Consider, however, that the coupled reaction

H_2 + fumarate$^{2-} \rightarrow$ succinate^{2-} + $2H^+$

actually represents the <u>sum</u> of the following two half reactions:

$\boxed{H_2 \rightarrow 2H^+ + 2e^-}$ $\boxed{E_0' = +0.42V}$

$\underline{\text{fumarate}^{2-} + 2e^- \rightarrow \text{succinate}^{2-} \qquad E_0' = +0.03V}$

H_2 + fumarate$^{2-} \rightarrow$ succinate^{2-} + $2H^+$ $\Delta E_0' = +0.42V + (+0.03V)$

$= +0.45V$

This is written as an oxidation reaction, so that the sign of E_0' is reversed relative to when it 's written as a reduction reaction

Calculation of Reduction Potentials for Coupled Half Reactions

- Thus have two ways to compute $\Delta E_0'$ for coupled oxidation-reduction reactions:
  1. Write both half reactions as reduction reactions, and subtract the $\Delta E_0'$ for the putative electron-donating half-reaction from the $\Delta E_0'$ for the putative electron-accepting reaction
  2. Write the putative electron-donating reaction as an oxidation reaction, reverse its sign, and take the sum of the two E_0' values

Calculation of Reduction Potentials for Coupled Half Reactions

- For the coupled reaction

H_2 + fumarate$^{2-} \rightarrow$ succinate^{2-} + $2H^+$

<u>Approach 1:</u>

$2H^+ + 2e^- \rightarrow H_2$ $E_0' = -0.42V$

fumarate^{2-} + $2e^- \rightarrow$ succinate^{2-} $E_0' = +0.03V$

$\Delta E_0' = +0.03 - (-0.42) = 0.45V$

<u>Approach 2:</u>

$H_2 \rightarrow 2e^- + 2H^+$ $E_0' = +0.42V$

fumarate^{2-} + $2e^- \rightarrow$ succinate^{2-} $E_0' = +0.03V$

$\Delta E_0' = +0.42 + (+0.03) = 0.45V$

A Final Word on the Connection between ΔE and ΔG

- A pair of coupled redox reactions is thermodynamically favorable (i.e. $\Delta G < 0$) only if the reduction potential for the coupled reaction is positive (i.e. $\Delta E > 0$), because…
- $\Delta G^0 = -nF\Delta E^0$

Metabolic Diversity - Part 2

- Classification of organisms in terms of energy metabolism
- Fermentation
- Aerobic respiration
- Anaerobic respiration
- Chemolithotrophy
- Phototrophy

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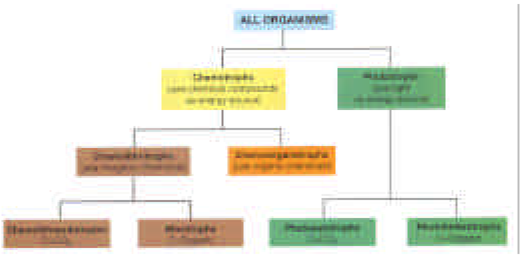
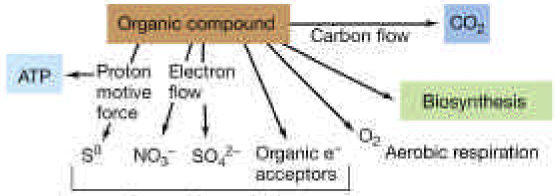
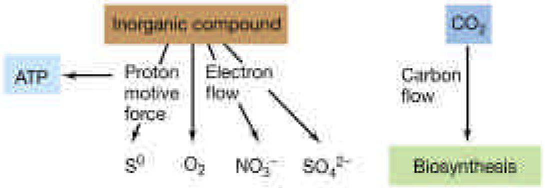
Classification of organisms in terms of energy metabolism



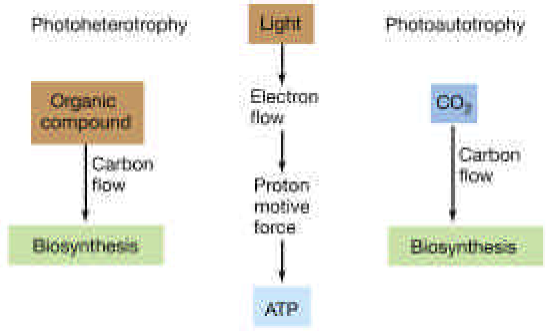
Madigan et al. (2000)



(a) **Chemoorganotrophic metabolism**



(b) **Chemolithotrophic metabolism**



(c) **Phototrophic metabolism**

Metabolic Diversity - Part 2

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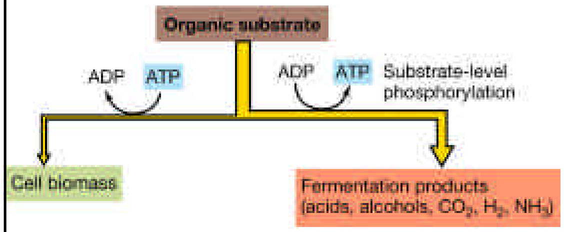


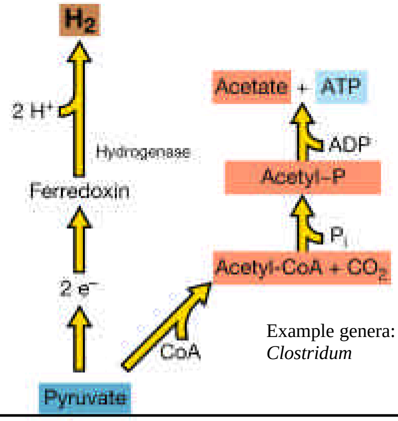
Organic substrate

ADP ATP

ADP ATP Substrate-level phosphorylation

Cell biomass

Fermentation products (acids, alcohols, CO_2, H_2, NH_3)



H_2

$2\ H^+$

Hydrogenase

Ferredoxin

$2\ e^-$

Pyruvate

CoA

Acetyl-CoA + CO_2

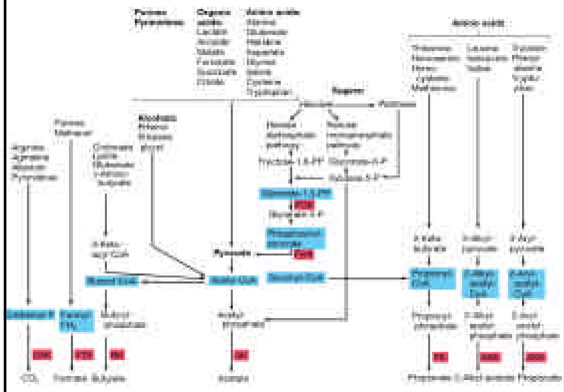
P_i

Acetyl-P

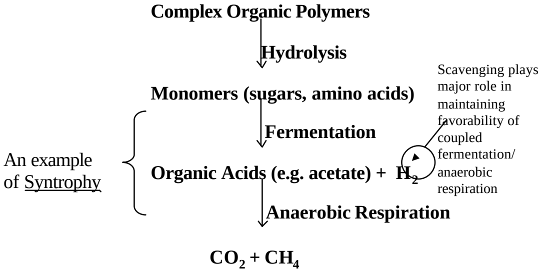
ADP

Acetate + ATP

Example genera: *Clostridium*



Anaerobic Microbial Foodchain



Complex Organic Polymers

Hydrolysis

Monomers (sugars, amino acids)

Fermentation

Organic Acids (e.g. acetate) + H_2

Anaerobic Respiration

$CO_2 + CH_4$

An example of Syntrophy

Scavenging plays major role in maintaining favorability of coupled fermentation/ anaerobic respiration

Importance of H_2 & Acetate Scavenging in Anaerobic Microbial Food Chain

- Typical mixture of products from hexoses fermentation (e.g. by *Clostridium*):
  - Butyrate ($CH_3CH_2CH_2COO^-$)
  - Propionate ($CH_3CH_2COO^-$)
  - Acetate (CH_3COO^-)
  - H_2
  - CO_2

Importance of H_2 & Acetate Scavenging in Anaerobic Microb Food Chain - Contd

- Although organic acids such as butyrate and propionate may be oxidized directly by some anaerobic respiratory bacteria, they cannot be metabolized by methanogens (which utilize only H_2/CO_2 and acetate)
- Complete degradation of complex organic carbon under methanogenic conditions requires the activity of organic acid-oxidizing, H_2-producing bacteria such as *Syntrophomonas*

Importance of H_2 & Acetate Scavenging in Anaerobic Microb Food Chain - Contd

- Consider degradation of butyrate:
  $\text{Butyrate}^- + 2H_2O \rightarrow 2\text{Acetate}^- + 2H_2 + H^+$
  $\Delta G_0' = +48$ kJ/mol
  - reaction is unfavorable under standard conditions
- But if acetate is scavenged to ≈ 10 µM and H_2 to 10^{-5} atm (equivalent to dissolved H_2 concentration of ca. 10 nM):
  $\Delta G' = -68$ kJ/mol

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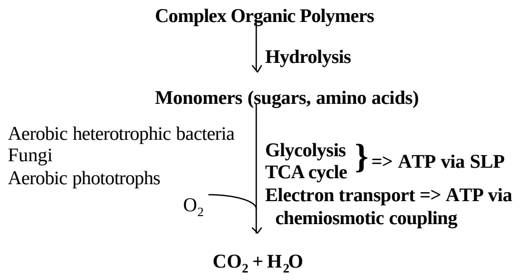
Aerobic Microbial Metabolism


Complex Organic Polymers

Hydrolysis

Monomers (sugars, amino acids)

Aerobic heterotrophic bacteria
Fungi
Aerobic phototrophs

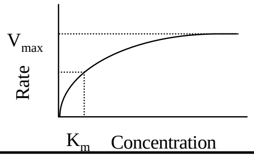
Glycolysis
TCA cycle } **=> ATP via SLP**
Electron transport => ATP via chemiosmotic coupling

O_2

$CO_2 + H_2O$

Characterisitics of Aerobic Microorganisms

- Single organism can oxidize wide variety of organic compounds
- Can express alternative terminal oxidases => enhanced scavenging of O_2

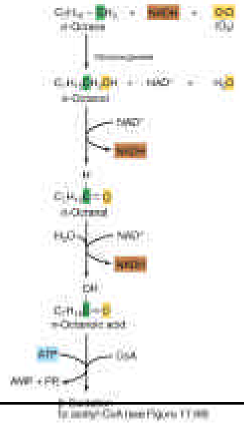


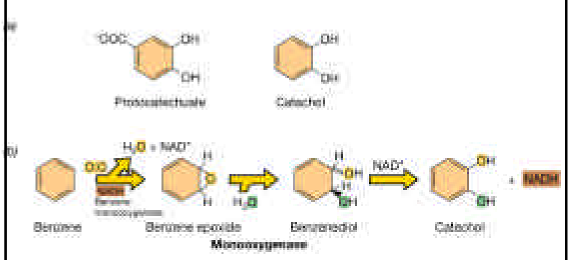
$$V = V_{max}\frac{C}{K_m + C}$$

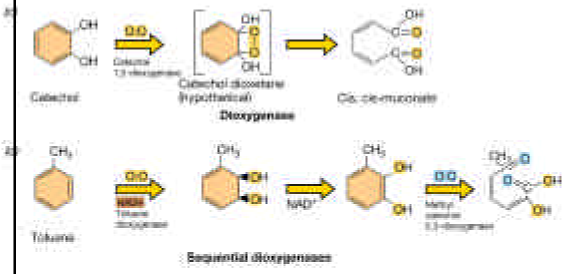
K_m for O_2 generally ≤ 1 µM

Characterisitics of Aerobic Microorganisms - Contd

- Can use O_2 or H_2O_2 as reactants in which one or more O atoms are incorporated into an organic carbon molecule
- Enzymes involved = oxygenases (mono- or di-) and peroxidases
- Permits oxidation of aliphatic and aromatic hydrocarbons as well as lignin







Metabolic Diversity - Part 2

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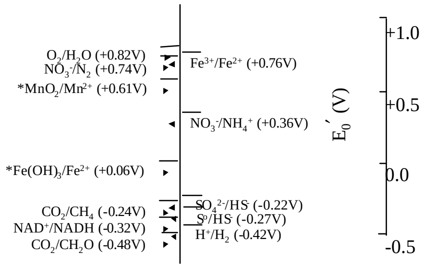
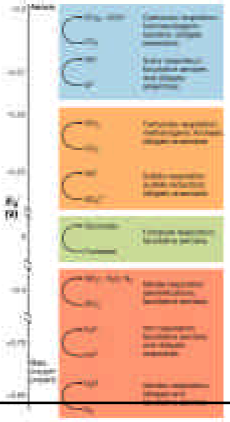
“Redox Tower” for Microbial Metabolic Reactions



* With $\{Mn^{2+}\}, \{Fe^{2+}\} = 10^{-6}$ M



Free Energy of Organic Carbon Oxidation Reactions

Reaction	$\Delta G_0'$ (kJ/mol CH_2O)
$CH_2O + O_2 \rightarrow HCO_3^- + H^+$	-513
$CH_2O + 0.8NO_3^- \rightarrow 0.4N_2 + HCO_3^- + 0.4H_2O + 0.2H^+$	-471
$CH_2O + 0.5NO_3^- + 0.5H_2O \rightarrow 0.5NH_4^+ + HCO_3^-$	-323
$CH_2O + 2MnO_2 + 3H^+ \rightarrow 2Mn^{2+} + HCO_3^- + 2H_2O$	-406*
$CH_2O + 4Fe(OH)_3 + 7H^+ \rightarrow 4Fe^{2+} + HCO_3^- + 10H_2O$	-203*
$CH_2O + 0.5SO_4^{2-} \rightarrow 0.5HS^- + HCO_3^- + 0.5H^+$	-99
$CH_2O + 0.5H_2O \rightarrow 0.5CH_4 + 0.5HCO_3^- + 0.5H^+$	-83
$CH_2O \rightarrow 0.5CH_3COO^- + 0.5H^+$	-75

* $\{Mn^{2+}\}, \{Fe^{2+}\} = 1$ µM

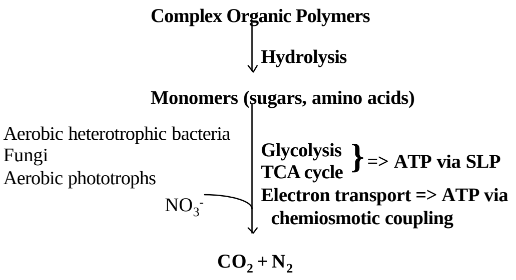
Assimilatory vs. Dissimiliatory Reduction

- Assimilatory: reduction of inorganic compounds (e.g. NO_3^-, Mn(IV) and Fe(III) oxides, SO_4^{2-}, and CO_2) for acquisition of cellular nutrients
- Dissimilatory: reduction of inorganic compounds as electron acceptors in energy metabolism (of anaerobic respiratory bacteria)

Dissimilatory Nitrate Reduction

- Denitrification: reduction of NO_3^- to N_2:
  - Carried out by facultative anaerobes, which substitute NO_3^- for O_2 as the terminal electron acceptor
  - Requires alteration of one or more electron tranport chain components, including the terminal reductase)
  - Denitrifiers oxidize complex organic carbon compounds directly to CO_2
- Diversity of denitrifiers is extremely high – second only to aerobic heterotrophs

Denitrification



Complex Organic Polymers

Hydrolysis

Monomers (sugars, amino acids)

Aerobic heterotrophic bacteria
Fungi
Aerobic phototrophs

Glycolysis
TCA cycle } **=> ATP via SLP**

NO_3^-

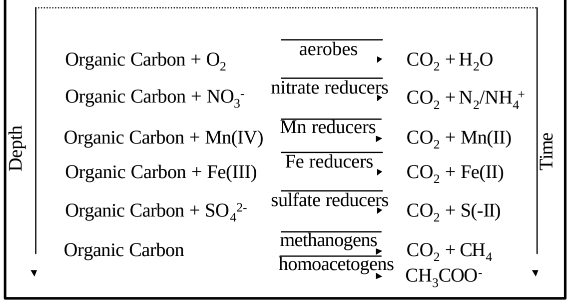
Electron transport => ATP via chemiosmotic coupling

$CO_2 + N_2$

Dissimilatory Nitrate Reduction - Contd

- Dissimilatory NO_3^- reduction to NH_4^+:
  - Carried out by fermentative bacteria as (relatively) minor sink for electrons during primarily fermentative metabolism
  - Also carried out by certain specialized anaerobic respiratory bacteria which reduce oxidized metals such as Mn(IV) and Fe(III)

Distribution of Microbial Respiratory Processes in Aquatic Sediments



Organic Carbon + O_2 $\xrightarrow{\text{aerobes}}$ $CO_2 + H_2O$

Organic Carbon + NO_3^- $\xrightarrow{\text{nitrate reducers}}$ $CO_2 + N_2/NH_4^+$

Organic Carbon + Mn(IV) $\xrightarrow{\text{Mn reducers}}$ CO_2 + Mn(II)

Organic Carbon + Fe(III) $\xrightarrow{\text{Fe reducers}}$ CO_2 + Fe(II)

Organic Carbon + SO_4^{2-} $\xrightarrow{\text{sulfate reducers}}$ CO_2 + S(-II)

Organic Carbon $\xrightarrow{\text{methanogens}}$ $CO_2 + CH_4$

$\xrightarrow{\text{homoacetogens}}$ CH_3COO^-

Dissimilatory Mn(IV) and Fe(III) Reduction

- Carried out by strict and facultative anaerobes which oxidize organic acids and H_2 (i.e. end-products of hydrolysis & fermentation)
- Both soluble and solid-phase forms are utilized
- Reactivity (particle size, surface area, presence of surface-bound Mn(II)/Fe(II)) of solid-phases exerts major impact on reducibility
- Physiology and biochemistry poorly understood compared to other anaerobic respiratory bacteria (but is improving rapidly)

Dissimilatory Sulfate Reduction

- Carried out by mainly strict anaerobes (although some SRB can utilized O_2!) which oxidize organic acids and H_2
- Partially reduced S compounds such as $S_2O_3^{2-}$ (thiosulfate), SO_3^{2-} (sulfite), and S^o (elemental sulfur) can also be reduced to HS^-
- Many SRB can also utilized NO_3^-, Mn(IV), Fe(III), and other oxidized metals as electron acceptors (mainly for E generation only)
- Some H_2 oxidizers are autotrophic

Methanogenesis

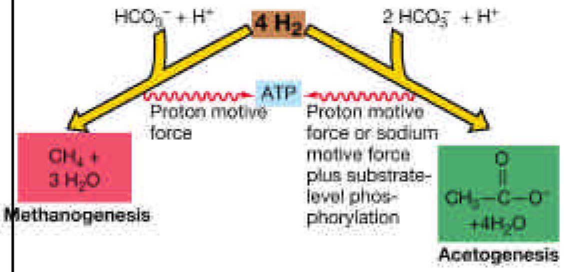
- Carried out by strictly anaerobic Archaea call methanogens
- Two major pathways:
  - $H_2 + CO_2 \rightarrow CH_4$ (CO_2 reduction)
  - $CH_3COO^- \rightarrow CH_4 + CO_2$ (acetoclastis)
- Specialized biochemistry, although ATP generation is by chemiosmotic coupling
- H_2 oxidizers are autotrophic

Acetogenesis

- Organsims involved are referred to as homoacetogens, which catalyze the reaction

  $4H_2 + 2CO_2 \rightarrow CH_3COO^- + 2H_2O + H^+$

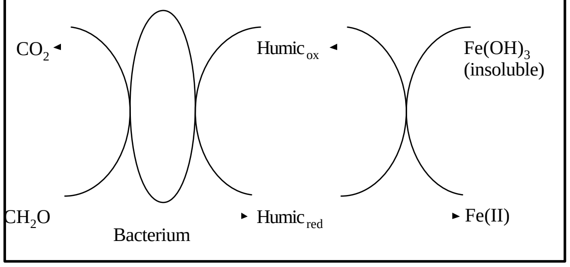
  $\Delta G_0' = -105$ kJ/mol
- Process utilizes the acetyl-CoA pathway
- Acetogens take over for methanogens in environments where conditions are unfavorable for methanogenesis (e.g. cold, low pH) => acetate accumulation in northern peatlands



Other Electron Acceptors for Anaerobic Respiration

- $Cl(V)O_3^-$ (chlorate) $\rightarrow Cl(-I)^-$
- $Cr(VI)O_4^{2-}$ (chromate) $\rightarrow Cr(III)(OH)_3(s)$ (Cr hydroxide)
- $Co(III)\text{-}EDTA^- \rightarrow Co(II)\text{-}EDTA^{2-}$
- $Se(VI)O_4^{2-}$ (selenate) $\rightarrow Se(IV)O_3^{2-}$ (selenite)
- $Se(IV)O_3^{2-}$ (selenite) $\rightarrow Se(0)(s)$ (elemental selenium)
- $As(V)O_4^{3-}$ (arsenate) $\rightarrow As(III)O_3^{2-}$ (arsenite)
- $UO_2(VI)(CO_3)_2^{2-}$ (uranyl carbonate) $\rightarrow$ $U(IV)O_2(s)$ (uraninite) + $2HCO_3^-$
- $Tc(VI)O_4^-$ (pertechnetate) $\rightarrow TcO_2(s)$ (Tc dioxide)
- $Au(III)^{3+}$ (gold ion) $\rightarrow Au(0)(s)$ (elemental gold)
- $\text{Humic(quinone)}_{ox} \rightarrow \text{Humic(quinone)}_{red}$
- $R\text{-}Cl^- \rightarrow R\text{-}H + Cl^-$ (e.g. $\underset{\text{Chlorobenzoate}}{C_7H_4O_2Cl^-} \rightarrow \underset{\text{Benzoate}}{C_7H_5O_2^-}$)

Model for Promotion of Solid-Phase Fe(III) Reduction via Bacterial Humics Reduction



Other Electron Acceptors for Anaerobic Respiration - Contd

- Fumarate → succinate
- Glycine → acetate
- Dimethyl sulfoxide (DMSO) → Dimethyl sulfide
- Trimethylamine-N-oxide (TMAO) → Trimethylamine (TMA) (=> smelly fish)
- $RCl_x \rightarrow RH_x + xCl^-$
  - Carbon tetrachloride
  - Trichloroethylene
  - Tetrachlorethylene

Chemorganotrophy in Archaea

- Thermophilic and/or acidophilic chemoorganotrophs are found in the Archaea
  - Fermenters
  - Aerobic respirers
  - Fe(III) and humic substance reducers
  - S^o and SO_4^{2-} reducers
  - See Chapter 14 in Madigan et al. (2000) for details; also Lovley et al. 2000. Chem. Geol. 169:289-298 on Fe(III) & humic reducers

Metabolic Diversity – Part 2

- Classification of organisms in terms of energy metabolism
- Fermentation
- Aerobic respiration
- Anaerobic respiration
- ➡ Chemolithotrophy
- Phototrophy

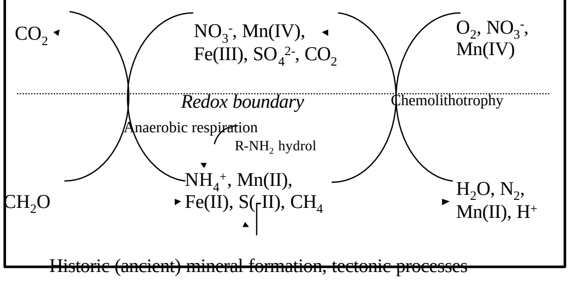
Chemolithotrophy

- Energy obtained from oxidation of inorganic compounds
- Most chemolithotrophic microorganism are autotrophic
- Energy generation generally occurs via electron transport
- Reducing power for CO2 fixation comes either directly from the reduced inorganic compound or reverse electron transport

Major Chemolithotrophic Processes

- H_2 oxidation
- Reduced sulfur oxidation & disproportionation
- Ferrous iron oxidation
- Ammonium oxidation (nitrification)
- Methane oxidation (methanotrophy)

Organotrophic-Lithotrophic Redox Cycling



Bacterial H_2 oxidation

- Carried out by all major respiratory groups of bacteria and archaea
- Many H_2 oxidizers are autotrophic
- Reducing power for CO_2 fixation can come directly from H_2 due to low E_0' of H^+/H_2 redox couple (-0.42V) compared to the $NADP^+/NADPH$ couple (-0.32V)

Bacterial Reduced S Oxidation

- Major reactions:

	$\Delta G_0'$ (kJ/mol)
$HS^- + 2O_2 \rightarrow SO_4^{2-} + H^+$	-798
$HS^- + 0.5O_2 + H^+ \rightarrow S^o + H_2O$	-209
$S^o + H_2O + 1.5O_2 \rightarrow SO_4^{2-} + 2H^+$	-587
$S_2O_3^{2-} + H_2O + 2O_2 \rightarrow 2SO_4^{2-} + 2H^+$	-813

- *NO_3^- and Mn(IV) can also serve as oxidants for enzymatic oxidation
- Fe(III) reacts spontaneously with HS^- but does not function as enzymatic oxidant

* Note recent discovery of giant marine S bacteria (*Thioploca*) which accumulate NO_3^- from overlying water for subsurface HS^- oxidation

Bacterial Reduced S Oxidation - Contd

- Sulfide-oxidizing bacteria (e.g. *Beggiatoa*, *Thiomicrospira*) are generally microaerophiles which live in opposing gradients of HS^- and O_2, where they can compete effectively with abiotic oxidation
- Enzymatic catalysis can enhance kinetics of HS^- oxidation by factor of 1000

Bacterial Reduced S Oxidation - Contd

- Many sulfide-oxidizing bacteria are autotrophic
- Reducing power for CO_2 fixation comes from reverse electron transport, in which ATP is used to drive electron flow backward through the electron transport chain in order to allow for reduction of NAD^+ or $NADP^+$ to NADH or NADPH
- Reverse electron transport is required since the E_0' of the SO_4^{2-}/HS^- and S^o/HS^- couples (-0.24V and -0.27V, respectively) are less negative than the NAD+/NADH and NADP+/NADPH couples (both ca. -0.32V)

Bacterial Reduced S Oxidation - Contd

- FeS (iron monosulfide, formed via the reaction $Fe^{2+} + HS^- \rightarrow FeS + H^+$) can be oxidized enzymatically with O_2, NO_3^-, and Mn(IV)
- FeS_2 (pyrite, formed via various reaction pathways; the dominant iron sulfide mineral in sedimentary environments) is probably not oxidized enzymatically
- FeS_2 oxidation is driven indirectly via Fe^{2+} oxidation

Bacterial Reduced S Disproportionation

- Process represents a type of inorganic fermentation
- First such reaction was discovered by accident during attempts to cultivate HS^- oxidizing bacteria:

$S_2O_3^{2-} + H_2O \rightarrow SO_4^{2-} + H_2S \quad \Delta G_0' = -22$ kJ/mol

- If have supply of oxidant to regenerate $S_2O_3^{2-}$ from partial oxidation of H_2S (e.g. Mn(IV) and Fe(III) oxides), can oxidize large quantities of H_2S through this pathway

Bacterial Reduced S Disproportionation - Contd

- Other major reaction:
  $4S^o + 4H_2O \rightarrow 3H_2S + SO_4^{2-} + 2H^+$ $\Delta G_0' = +41$ kJ/mol
  On its one, an unfavorable reaction, but…
- When coupled to reaction of H_2S with MnO_2 or $Fe(OH)_3$, it becomes favorable:
  $3S^o + 2H_2O + MnO_2 \rightarrow 2H_2S + SO_4^{2-} + Mn^{2+}$
  $\Delta G_0' = -94$ kJ/mol
  $3S^o + 2Fe(OH)_3 \rightarrow 2FeS + SO_4^{2-} + 2H_2O + 2H^+$
  $\Delta G_0' = -86$ kJ/mol

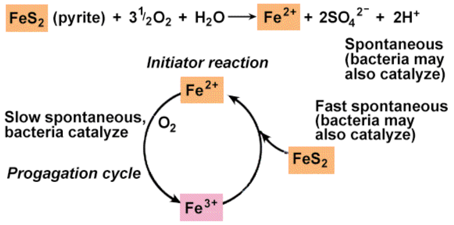
Bacterial Reduced S Disproportionation - Contd

- Recent studies indicate that $S_2O_3^{2-}$ and S^o disproportionation coupled to redox cycling of Mn and Fe are likely to be major pathways for sulfide oxidation in marine sediments

Bacterial Fe(II) Oxidation

- Fundamental distinction between acidic and circumneutral pH environments
  - At pH < 2, Fe^{2+} does not react spontaneously with O_2; thus energy from the reaction
    $Fe^{2+} + 0.25O_2 + H^+ \rightarrow Fe^{3+} + 0.5H_2O$
    $\Delta G_0 = -49$ kJ/mol
    is freely available to bacteria
  - Bacterial Fe^{2+} oxidation (e.g. by *Thiobacillus ferrooxidans*) is critical process in formation of acid mine drainage

Key Role of Fe(II)-Oxidizing Bacteria in Pyrite Oxidation



Bacterial Fe(II) Oxidation - Contd

- Fundamental distinction between acidic and circumneutral pH environments
  - At circumneutral pH, Fe^{2+} reacts spontaneously with O_2*; thus Fe(II) oxidizing bacteria must compete with abiotic reaction for energy from the reaction
    $Fe^{2+} + 0.25O_2 + 2.5H^+ \rightarrow Fe(OH)_3 + 2H^+$
    $\Delta G_0 = -113$ kJ/mol
  - As in HS^- oxidation, bacteria involved are microaerophiles which locate themselves in opposing gradients of Fe^{2+} and O_2 in vicinity of a redox interface environment

*half-life of Fe^{2+} in well-oxygenated water is < 5 min

Bacterial Fe(II) Oxidation - Contd

- Many Fe(II)-oxidizing bacteria, both acidophilic and neutrophilic, are autotrophic
- Reducing power for CO_2 fixation must come from reverse electron transport, since the E_0' of the Fe^{2+}/Fe^{3+} and $Fe^{2+}/Fe(OH)_3$ couples (+0.76 and +0.06V, respectively) are much higher than the NAD+/NADH and NADP+/NADPH couples
- Biochemistry/genetics known only for *Thiobacillus ferrooxidans*

Bacterial Mn(II) Oxidation

- Mn^{2+} does not react spontaneously with O_2; at pH < 8.5; thus energy from the reaction

  $Mn^{2+} + 0.5O_2 + H_2O \rightarrow MnO_2 + 2H^+$

  $\Delta G_0' = -81$ kJ/mol

  is freely available to bacteria in typical aquatic environments
- Mn(II) oxidation is carried-out by a wide variety of heterotrophic organisms
- No evidence for E generation or autotrophy
- Purpose for Mn(II) oxidation is unknown; possiblities include protection from grazing (by Mn(IV) oxide encrustation) and sequestration of nutrients sorbing to Mn(IV) oxides

Bacterial NH_4^+ Oxidation

- NH_4^+ can be produced during dissimilatory NO_3^- reduction, and is also released during overall process of organic matter decomposition in both aerobic and anaerobic environments; also present in some hydrothermal vent fluids
- Major oxidant for NH_4+ at Earth surface temperature is O_2
- Conversion of NH_4^+ to NO_3^- through reaction with O_2 is known as nitrification

Bacterial NH_4^+ Oxidation - Contd

- Two major steps in nitrification $\Delta G_0'$ (kJ/mol)

  1. $NH_4^+ + 1.5O_2 \rightarrow NO_2^- + 2H^+ + H_2O$ -275
  2. $NO_2^- + 0.5O_2 \rightarrow NO_3^-$ -74
- Step 1 is catalyzed by and *Nitrosomonas* (classical) and *Nitromonas* (recently identified)
- Step 2 is catalyzed by *Nitrobacter*
- Steps 1 and 2 involve ammonium oxidase and nitrite oxidase enzymes, respectively
- Both groups are autotrophic, and diversity within them is very low, e.g. compared to NO_3^- reducers

Bacterial NH_4^+ Oxidation - Contd

- New, important mechanism for enzymatic NH_4^+ oxidation recently identified, called *anammox* to refer to anaerobic ammonium oxidation

  $NH_4^+ + NO_2^- \rightarrow 2N_2 + 2H_2O$ $\Delta G_0' = -358$ kJ/mol
- Discovered as a result of lack of proper N mass balance during sewage sludge degradation
- Currently being exploited for removal of N from sewage and other wastestreams
  - Concept: bleed in just enough O_2 to catalyze NH_4^+ oxidation to NO_2^- and then let annamox take care of the rest

Bacterial CH_4 Oxidation

- Organisms involved are referred to as methanotrophs; below to wider group of organisms known as methylotrophs which utilized a variety of C_1 compounds (e.g. methanol, formate, carbon monoxide)
- Catalyze the reaction

  $CH_4 + 2O_2 \rightarrow CO_2 + 2H_2O$ $\Delta G_0' = -818$ kJ/mol
- Initial step requires enzyme methane monooxygenase; similar to ammonium monooxygenase => potential for cross reactivity

Bacterial CH_4 Oxidation - Contd

- Methanotrophs are not autotrophic, since they assimilate a reduced C_1 compound
- Two classes of methanotrophs are known:
  - Type I: use ribulose monophosphate pathway for C_1 assimilation
  - Type II: use serine pathway for C_1 assimilation
- Methanotrophs can degrade certain chlorinated aliphatic compounds (e.g. trichloroethylene) via process known as cometabolism; here methane monooxygenase can attack C-Cl bonds, ultimately yielding CO_2 and Cl^-

Chemolithotrophy in Archaea

- Thermophilic and/or acidophilic chemolithotrophs are found in the Archaea
  - Electron donors: H_2, S^o, Fe(II)
  - Electron acceptors: O_2, NO_3^-, S^o, SO_4^{2-}, CO_2
  - See Chapter 13 in Madigan et al. (2000) for details

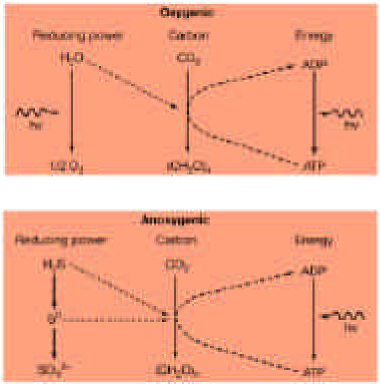
Bacterial Metabolism Part 2 – Metabolic Diversity

- Classification of organisms in terms of energy metabolism
- Fermentation
- Aerobic respiration
- Anaerobic respiration
- Chemolithotrophy

➡ Phototrophy

Photosynthesis

- Light reactions: conservation of light energy in chemical energy (ATP)
- Dark reactions: chemical energy and stored electrons (NADH, NADPH) are used to reduce CO_2 to organic compounds
- Oxygenic (O_2-producing) vs. anoxygenic (no O_2 production)



Light drives oxidation of H_2O in oxygenic photosynthesis

Phototrophy

- Oxygenic photosynthesis:

  $CO_2 + H_2O$ + light energy → $CH_2O + O_2$
  - Green plants, microalgae
  - Provides energy for all biogeochemical cycles
  - Production of alkalinity through CO_2 consumption can lead to precipitation of carbonate minerals

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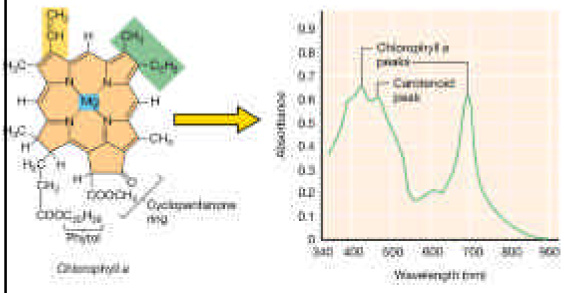
Phototrophy

- Anoxygenic photosynthesis

  CO_2 + (H_2S* or Fe(II)) + light energy → CH_2O + SO_4^{2-}/S^o or $Fe(OH)_3$
  - Organisms: Purple and green photosynthetic bacteria
  - Habitats: stratified lakes and microbial mats
  - Production of alkalinity through CO_2 consumption can lead to precipitation of carbonate minerals

* Discovery of potential for utilization of electrons from H_2S in anoxygenic photosynthesis was a key step toward understanding biochemistry of oxygenic photosynthesis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CH
H
CH3
C2H5
H3C
H
Mg
N
H
H3C
CH3
H
H3C
H
CH2
H
O
COOCH3
Cyclopentanone ring
COOC20H39
Phytol
Chlorophyll a
Chlorophyll a peaks
Carotenoid peaks
Absorbance
0.9
0.8
0.7
0.6
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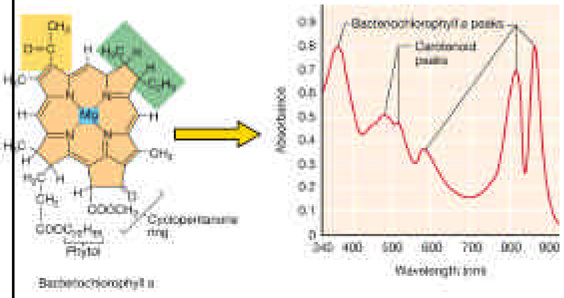
Wavelength (nm)


CH3
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COOCH3
Cyclopentanone ring
COOC20H39
Phytol
Bacteriochlorophyll a
Bacteriochlorophyll a peaks
Carotenoid peaks
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Wavelength (nm)