Biological Degradation

- Biodegradation involves the transformation of organic pollutants
- Enzyme-catalyzed reactions
- Inorganic pollutants (e.g. Fe$^{2+}$, Pb$^{2+}$, NO$_3^-$, NH$_4^+$) may also be used by microorganisms and plants
- For oxidation/reduction reaction
- Generally do not remove inorganic pollutant from environment

Biological Degradation

- Biological transformations
  - Can accelerate by several orders of magnitude the rate of transformation of a pollutant
  - Enzymes lower the activation energy required for a reaction
  - Provide a surface where pollutant and oxidant can meet

Biological Degradation

- The organic pollutant may be degraded
  - As a substrate (i.e. a good food source), usually as a carbon source
  - Via cometabolism (i.e. the bugs generally prefer something else but have the enzymes to partially degrade the pollutant)
A major issue in biodegradation is “bioavailability”:
- Is the pollutant available to the microorganisms for degradation?
- Is it dissolved?
- Can they use a sorbed pollutant?

Microbes have enzymes inside and outside (exoenzymes) of the cell wall
- Not all microbes carry the necessary enzymes for degradation of all pollutants
- Pollutants with unusual chemical structures will not be easily degraded

Some enzymes are very specific to a particular chemical and do not serve to degrade/transform other molecules
- Other enzymes are non-specific and result in the “chance” transformation of pollutants - this is the explanation for cometabolism

Example: degradation of TCE
- Methanothrops (methane eaters) have an enzyme, denominated Methane Monooxygenase (MMO), which is quite non-specific
- MMO is also used by bacteria to degrade most hydrocarbons
In general, one microorganism will not be responsible for the complete degradation of the pollutant to CO$_2$, H$_2$O (and Cl$^-$, NH$_4^+$, etc.). Microorganisms cooperate to make the transformation, each one providing their enzymatic capabilities and making use of the carbon source.

Example of cooperative degradation:

- Inhibition:
  - In some cases, the presence of a pollutant may inhibit the activity of an enzyme.
  - Sometimes pollutants “compete” for the enzyme (competitive inhibition) so that one pollutant is degraded before the other.

Transformation does not always produce simpler molecules.

Some intermediate products may be quite a bit more toxic than the original pollutant (e.g., TCE -> PCE -> Vinyl Chloride).
Factors affecting biodegradation of a pollutant in the environment:
- pollutant structure
- environmental factors
- availability (pollutant, nutrients, O₂)
- microbial numbers (concentration of cells)

Pollutant Structure
- Pollutants that are not commonly found in the environment (e.g., artificially derived like pesticides, PCBs, dioxins) are more resistant to biodegradation
- Pollutant structure affects solubility, sorption and volatility, which determine bioavailability
- Branches in the carbon chain and certain functional groups affect the rate of biodegradation
- Example:
  - linear alkylbenzylsulfonates (ABS) degrade fast
  - nonlinear ABS degrade very slow

Environmental Factors
- pH
- soil texture and permeability
- soil moisture
- temperature
- redox potential (aerobic or anaerobic)
- available organic matter and nitrogen

Availability
- The pollutant must usually be dissolved for transformation
- It has to diffuse to the microbes and in some cases through external polymeric matrices that protect the microbes against changes in soil moisture
Availability

- Other nutrients (N, P) and micronutrients (Fe, Ni, Cr, etc) must also be available for cell growth.
- Nutrients may be delivered to the cells via injection of an aqueous solution.
- In waste water treatment, the addition of micronutrients is of significance.

Aerobic Conditions

- Surface soil and vadose zone are typically aerobic.
- Content of O₂ can decrease significantly as one goes further down.
- O₂ depletion via microbial activity.
- Used for degrading soil organic matter.

Oxidation-Reduction

Contaminant + \( X_{\text{ox}} \) → Products + \( X_{\text{red}} \)

Capture of Energy by Terminal e-acceptor (\( X_{\text{ox}} \)):

\[ 0_2 > \text{Fe}^{3+} > \text{NO}_3^- > \text{SO}_4^{2-} > \text{CO}_2 \]

Redox Conditions

- Aerobic Conditions (high oxygen availability).
- Anaerobic Conditions (no oxygen availability).

Redox Sequence
Aerobic Conditions

- In general, aerobic degradation is much faster than anaerobic degradation.
- Hydrocarbons are typically easily degraded under aerobic conditions.
- If the hydrocarbon has an oxygen atom in the structure, then it can generally degrade anaerobically.

Redox Conditions

Chlorinated hydrocarbons are typically stable or only slowly degradable under aerobic conditions.

Anaerobic Conditions

- Under anaerobic conditions, hydrocarbons are quite stable (e.g. petroleum reservoirs).
- Chlorinated hydrocarbons are more easily degraded anaerobically.
- Different organisms cooperate to degrade the pollutants anaerobically.

All of these are slow reactions.
### Rates of Biodegradation

- **Biodegradation rate is typically diffusion-limited when**
  - pollutant is a good source of carbon for the microbes
  - microbes have the enzymes to degrade pollutant
  - The degradation rate may be limited by advection or diffusion of other nutrients (O₂, NH₄⁺, etc.)

**Michaelis-Menten Enzyme Kinetics**
- Degradation is not limited by pollutant transport
- There is no increase in number of microorganisms or enzyme concentrations
  - low pollutant concentration
  - pollutant not easily metabolized

If bioavailability is an issue, the rate may be controlled by diffusion of the pollutant from the bulk water to inside the cell:

$$\text{Rate} = \text{Flux} = \frac{D}{z} \left( C_{\text{bulk}} - C_{\text{cell}} \right)$$
Rates of Biodegradation

Michaelis-Menten Enzyme Kinetics

Think of it just as a chemical transformation accelerated by the presence of catalysts (enzymes) which have a constant concentration.

The microbes usually have another source of food (substrate) and use the pollutant by chance (cometabolism).

Rates of Biodegradation

Michaelis-Menten Enzyme Kinetics are modeled using considering the rate of pollutant removal:

\[
\text{Rate} = \frac{dC}{dt} = -\frac{V_{\text{max}} C}{K_{\text{MM}} + C}
\]

\(V_{\text{max}}\) = fastest possible degradation rate

\(K_{\text{MM}}\) = concentration of substrate at which degradation is half of \(V_{\text{max}}\)

A note of caution:

Remember the assumptions (pollutant concentration is not limited by desorption or diffusion, low pollutant concentration, no growth of microbes due to pollutant)

\(V_{\text{max}}\) depends on the concentration of microbes, so it must be determined case by case.

---

**TABLE**

<table>
<thead>
<tr>
<th>Substance</th>
<th>(V_{\text{max}})</th>
<th>(K_{\text{MM}})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toluene in water</td>
<td>1.0 M</td>
<td>1.0 M</td>
</tr>
<tr>
<td>Hexachlorobenzene</td>
<td>0.04 M</td>
<td>0.03 M</td>
</tr>
<tr>
<td>Hexachlorobenzene in solution</td>
<td>0.04 M</td>
<td>0.03 M</td>
</tr>
<tr>
<td>Nonchlorinated alkyl substituted</td>
<td>0.04 M</td>
<td>0.03 M</td>
</tr>
</tbody>
</table>

Source: Schwarzenbach et al., 1993
Rates of Biodegradation

- **Monod Kinetics**
  - The population of microbes increases because the pollutant supports the growth of microorganisms.
  - The concentration of microbes $[B]$ changes as a function of time, based on a growth rate, $\mu$ (in units of $h^{-1}$).

\[
\frac{d[B]}{dt} = \mu [B]
\]

$[B] = [B]_0 \exp(\mu t)$

The rate of growth, $\mu$, is a function of the availability of food (pollutant).

There is a maximum cell growth rate, $\mu_{\text{max}}$, which corresponds to the situation when the pollutant is present in excess.

\[
\mu = \frac{\mu_{\text{max}} C}{K_M + C}
\]

$K_M$ is the concentration where $\mu = 1/2 \mu_{\text{max}}$.

The rate of degradation of the pollutant depends on the growth rate of the microorganisms as well as the yield, $Y$, which measures the effectiveness of the pollutant in producing cells.

\[
\frac{dC}{dt} = -\frac{\mu_{\text{max}}[B] C}{Y (K_M + C)}
\]

Source: Schwarzenbach et al., 1993
Biodegradation of Metals

- Metals are used by the microbes for their biologic processes, but are not used like carbon as a major building block.
- Metals are not accumulated in microbes to a large extent.

Biodegradation of Metals

- High concentration of metals can interfere with microbial processes, and can result in changes in pH.
- pH also determines the bioavailability of metals, which may be dissolved at a given pH but precipitate at another pH.

Biodegradation of Metals

- Some metals are used as part of the enzymatic machinery of the microbes.
- Other metals are used as part of the microbial oxidation/reduction process: the oxidized metals may be more soluble and thus once excreted can transport along with groundwater.

Biodegradation of Metals

- Some metal transformations result in increased toxicity and/or lipophilicity of the organometallic pollutant.

Biodegradation of Metals

- Once the organometallic pollutant is lipophilic, it can then bioaccumulate up the food chain.
- Some bacteria do accumulate metals, like uranium, by incorporating them into their polymeric matrices.