Reaction Rate (Kinetics)
- Importance of metabolism in detoxification or activation of a toxicant depends on
  - Capability of detox enzymes for catalyzing the reaction
    - Dependent on ability of toxicant to form a complex with the detox enzyme
  - Rate of reaction
    - Faster reactions will increase the rate of elimination of the toxicant
    - Less re-circulating to blood and tissues

Reaction Rate
- Described by mathematical functions known as rate laws
  - Describe the relationship between time and the concentration of the toxicant
  - Two commonly used functions
    - Power rate law
      - First-order rate equation
    - Hyperbolic kinetics
    - Michaelis-Menten Kinetics

Reaction Kinetics
**Rate Law** = a mathematical function or differential equation describing the turnover rate of a compound as a function of the concentration.

**Power Rate Law**
\[ \text{Rate} = -\frac{d[C]}{dT} = kC^n \]

First Order when \( n = 1 \)
\[ \frac{d[C]}{dt} = -k[C] \]  
* Differential eq.

or
\[ [C] = [C]_0 \cdot e^{-kt} \]  
* Integrated eq.

Linearization of First-Order Function
\[ \ln[C] = -kt + \ln[C]_0 \]

\[ T_{1/2} = \frac{\ln 2}{k} = 0.693/k \]

\( K = \text{rate constant (1/day)} \)
Hyperbolic Kinetics

\[
\text{Rate} = \frac{-dC}{dT} = \frac{k_1 C}{k_2 + C}
\]

Enzyme or surface catalyzed.
Rate slows down as [C] increases.

Concentration of Chemical, [C] or [S]

**Double Reciprocal Plot for Calculation of Vmax & Km**

\[
\frac{1}{V} = \frac{1}{V_{\text{max}}} + \frac{K_m}{V_{\text{max}}}
\]

Lineweaver-Burk Plot
Slope: \( \frac{K_m}{V_{\text{max}}} \)
Intercept: \( -\frac{1}{K_m} \)
Intercept: \( \frac{1}{V_{\text{max}}} \)

**Practical Application of Michaelis-Menton Kinetics**

Disappearance of toxicant speeds up as concentration decreases below the level associated with Vmax or enzyme saturation. However, at sufficiently low concentration, metabolism might slow again because of necessity for complexation before reaction moves toward the product side.

**Elimination**

- Excretion
  - Metabolic reactions result in more water soluble compounds, facilitating excretion
  - Reduction in amount of parent compound available to target sites
  - Clearance
    - Volume of blood (or plasma) cleared of chemical per unit time
  - Routes
    - Expired air, saliva, bile, feces, urine, milk, hair
  - Extent less important than rate
    - All compounds eventually 100% excreted

**Excretion**

The Ultimate Fate
Insects have Malpighian tubules that function analogously to kidneys.

Activity & Total Dosage

Mix/load/apply

- 17.8 µg/kg
- 6.8 µg/kg
- 4.9 µg/kg

Area Under the Curve (AUC)
Gives Information About Whole Body Dose

Adapted from Nash et al. ‘82

Elimination rate can differ among different development stages.


Corn Rootworm Adult

Corn Rootworm Larva

Metabolism and Excretion Influences Selectivity

Corn Rootworm Hours After Dosing with Isofenphos

<table>
<thead>
<tr>
<th>Life Stage</th>
<th>1</th>
<th>2</th>
<th>4</th>
<th>8</th>
<th>24</th>
</tr>
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<tbody>
<tr>
<td>Adult</td>
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<tr>
<td>External Rinse</td>
<td>17</td>
<td>11</td>
<td>9</td>
<td>5</td>
<td>1</td>
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<tr>
<td>Internal Extract</td>
<td>71</td>
<td>65</td>
<td>54</td>
<td>44</td>
<td>16</td>
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<tr>
<td>Container Rinse</td>
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<td>4</td>
<td>10</td>
<td>18</td>
<td>35</td>
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<tr>
<td>Container Parent</td>
<td>3</td>
<td>2</td>
<td>3</td>
<td>2</td>
<td>1</td>
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</table>

Larvae

<table>
<thead>
<tr>
<th>Life Stage</th>
<th>1</th>
<th>2</th>
<th>4</th>
<th>8</th>
<th>24</th>
</tr>
</thead>
<tbody>
<tr>
<td>External Rinse</td>
<td>7</td>
<td>2</td>
<td>1</td>
<td>0.4</td>
<td>0.2</td>
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<tr>
<td>Internal Extract</td>
<td>59</td>
<td>46</td>
<td>38</td>
<td>21</td>
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<td>9</td>
<td>13</td>
<td>19</td>
<td>33</td>
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<tr>
<td>Container Parent</td>
<td>9</td>
<td>11</td>
<td>14</td>
<td>21</td>
<td>23</td>
</tr>
</tbody>
</table>

Storage

- Type of elimination mechanism
  - Influenced by rate of metabolism and hydrophobicity
    - Example: DDT
      - Slow metabolic rate
      - High Kow
  - Temporary mechanism in that chemical is released from storage sites
  - Equilibrium between adipose tissue and blood
  - Of concern for very recalcitrant compounds
  - Concept of bioconcentration factor (ecotox.)
  - Ratio of matrix (or food) concentration to concentration in an organism
Case Study

- Uptake, metabolism, and elimination of two hydrophobic contaminants by midge larvae
  - Experiment from Lydy et al. (2000) Arch. Environ. Contam. Toxicol. 38:163
  - Exposed midge larvae in water to either DDE or 2-chlorobiphenyl (2-CB)

![Images of midge larvae]

**DDE**
- WS: ~40 µg/L
- Log Kow: ~5.7
- Nominal Water Concentration: 7.3 µg/L

**2-chlorobiphenyl**
- WS: ~5900 µg/L
- Log Kow: ~4.49
- Nominal Water Concentration: 5.1 µg/L

**Two Compartment Toxicokinetic Model**

- $C_p$: [parent compound in animal] $k_u$: Uptake clearance coefficient
- $C_m$: [metabolites in animal] $k_{ep}$: Parent elimination rate constant
- $K_u$: uptake clearance coefficient

**Toxicokinetic Functions**

**DDE**
- $C_a = k_u * C_w * t$
- $C_a$: total DDE in midge
- $C_w$: DDE concentration in water
- $K_u$: uptake clearance coefficient

**2-chlorobiphenyl**
- $dC_{tot}/dt = (k_{ep}C_p) - (k_{em}C_m)$
- $C_{tot}$: total 2-CB residues in midge larvae

![Graphs of DDE and 2-CB uptake and elimination over time](https://example.com/dde_2-cb_graphs.png)
Modeling Results (Lydy et al. 2000)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>DDE</th>
<th>2-CB</th>
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</thead>
<tbody>
<tr>
<td>$K_u$ (mL/g/h)</td>
<td>84.1</td>
<td>66.0</td>
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<tr>
<td>$K_{soa}$ (h$^{-1}$)</td>
<td>0.100</td>
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<tr>
<td>$K_{adm}$ (h$^{-1}$)</td>
<td>0.073</td>
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</tr>
<tr>
<td>$K_m$ (h$^{-1}$)</td>
<td>0.031</td>
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<td>BCF</td>
<td>504</td>
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<tr>
<td>T1/2 (days)</td>
<td>5.7</td>
<td></td>
</tr>
</tbody>
</table>

Toxicokinetics & Plants

- All mechanisms & processes same as in animals
- However, must consider dose transfer from the environment (ditto if considering aquatic and soil dwelling animals)
- Other parameters important:
  - Soil sorption coefficient ($K_{oc}$)
  - Air:water transfer coefficient ($K_w$)

Conceptual Model of Relationships

- External Dose/Exposure
  - Internal Dose/Exposure
    - Tissue Dose/Exposure
    - Cellular/Subcellular Dose
- Adverse Effects/Pathology
  - Late Responses
  - Early Responses
  - Cellular/Subcellular Interactions
  - Toxicodynamics