Single Molecule Kinetics

• Consider radioactive decay:
  \[ \frac{32}{15}P \rightarrow \frac{32}{16}S^{+1} + e^- + \bar{\nu}_e \]

• Questions:
  – How does one phosphorus molecule know when to decay?
  – Do molecules have watches?
What Does $k$ Mean?

- **One view:** Rate at which radioactive molecules decay (N molecules per second)

- **Valid Alternative:** Probability that one molecule will decay per unit time (N transitions per second)
Algorithmic Thinking

• **Consider:** Ensemble of N radioactive $^{32}$P atoms, rate constant $k$.

• In period $\Delta t$, the likelihood of decay for one atom is:

$$\left( k \frac{\text{transitions}}{s} \right) \left( \Delta t \frac{s}{\text{period}} \right) = k \Delta t \frac{\text{transitions}}{\text{period}}$$

• For each of N molecules, generate a random number $r$ between 0 and 1:
  – If $0 \leq r < k\Delta t$, decay to $^{32}$S$^+$ + e$^-$
  – Otherwise, don’t decay this period
Simulation Results

- Rate constant is $0.06 \text{ s}^{-1}$, time step is 1 s.
- Red line is $e^{-0.06t}$.

(Python simulation code available upon request.)
The Master Equation

- **Paradigm Shift:** Instead of a single rate law, imagine a set of transition probabilities from one state ($^{32}$P) to another ($^{32}$S)
  - Works for more complicated systems, too

- The *master equation* allows us to integrate probabilities and determine population change over time

- Well beyond the scope of this course!
Example: Cholesterol Oxidase

Mechanism:

\[ E \cdot FAD + S \xrightleftharpoons[k_1]{k_{-1}} (E \cdot FAD) \cdot S \xrightarrow[k_2]{E \cdot FADH_2 + P} \]

Source: http://crystal.bcs.uwa.edu.au/px/alice/projects/SCOA.html
**Example: Cholesterol Oxidase**

- Trap enzymes in a rigid matrix, observe individual molecules in a fluorescence microscope

- Measure lifetimes of hundreds of cholesterol oxidase molecules

- Form a histogram: how many molecules had a lifetime of x ms?
Single Molecule Kinetics: Takeaways

• Individual molecules have a distribution of lifetimes

• Average behavior closely follows Michaelis-Menten kinetics

• New insight: If step 1 is slow, is step 2 also slow? *Single molecule dynamics!*

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Final Exam Details

• Tuesday, Dec. 9 from 12:00 – 3:00 pm
  – Location is in our usual classroom
• Total of 200 points (20% of your grade):

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