Condensed phase kinetics

- The kinetic theory of gases involves ballistic motion
- Kinetics in liquids and other condensed phases involves diffusion
- What are the major characteristics?
  - Many molecules interacting at once
  - Short mean free paths
  - No “memory” of momenta
Brownian motion

• Extreme case of condensed phase dynamics
• No memory of momentum
• “Bath” provides:
  – Random displacements
  – Frictional damping
• Bath action related to molecular collisions
Random walks

- Markov chain: no history dependence
- Consider a discrete time random walk in 1D:
  - Equal probability of moving left or right at each time step
  - Position probability distribution is binomial
  - At a large number of steps, probability becomes Gaussian
  - Mean square displacement grows linearly with time
- These are general features of unbiased random walks
- Can be generalized to continuous time and space

\[
P(n_+, N) = \frac{N!}{n_+!(N-n_+)!} \left(\frac{1}{2}\right)^N
\]

\[
\sim \sqrt{\frac{2}{\pi N}} \exp \left( -\frac{2n_+^2}{N} \right)
\]

\[
\sim \sqrt{\frac{1}{2\pi N}} \exp \left( -\frac{(m+N)^2}{2N} \right)
\]

\[
\langle m^2 \rangle \sim N
\]
1D random walk

10000 trajectories

$\langle x \rangle$

10000 trajectories

$\langle x^2 \rangle$

10000 trajectories

$10^5 x$

10000 trajectories
Diffusion coefficients

• What is the mean squared displacement per unit time?

• For unbiased random walks, it’s pretty simple

• Diffusion coefficients also have microscopic interpretation…

\[ m \to \frac{x, y, z}{l}, N \to \frac{t}{\tau} \]

\[ P_{1D}(x, t) = (4\pi Dt)^{-1/2} \exp\left(-\frac{x^2}{4Dt}\right) \]

\[ \langle x^2 \rangle_{1D} = 2Dt \]

\[ P_{2D}(x, y, t) = (4\pi Dt)^{-1} \exp\left(-\frac{x^2 + y^2}{4Dt}\right) \]

\[ \langle x^2 + y^2 \rangle_{2D} = 4Dt \]

\[ P_{3D}(x, y, z, t) = (4\pi Dt)^{-3/2} \exp\left(-\frac{x^2 + y^2 + z^2}{4Dt}\right) \]

\[ \langle x^2 + y^2 + z^2 \rangle_{2D} = 6Dt \]
Langevin equation

• Newton’s equation with extra terms

• Random force
  – Energy added by bath
  – Mean force is zero

• Viscous drag
  – Energy dissipated by bath
  – Related to velocity decay times

• The viscous drag is related to the diffusion coefficient

\[
m \frac{dv(t)}{dt} = -\zeta v(t) + f(t)
\]

\[
\langle f(t) \rangle = 0
\]

\[
m \frac{d\langle v(t) \rangle}{dt} = -\zeta \langle v(t) \rangle
\]

\[
\langle v(t) \rangle = v(0) e^{-\zeta t/m}
\]
Viscosity and diffusion

- Solve Langevin equation for mean squared position
  - Multiply by $x$ and rearrange
  - Use the fact that the mean squared velocity is $kT$ (Maxwell dist.)
  - Solve for $\langle xv \rangle$ with particle initially at origin
  - Relate to $\langle x^2 \rangle$ and solve

\[
m \langle \frac{d}{dt} (xv) \rangle = -\zeta \langle xv \rangle + m \langle v^2 \rangle = -\zeta \langle xv \rangle + k_B T
\]

\[
\langle xv \rangle = \frac{k_B T}{\zeta} \left( 1 - e^{-\zeta t/m} \right)
\]

\[
\frac{1}{2} \frac{d}{dt} \langle x^2 \rangle = \frac{k_B T}{\zeta} \left( 1 - e^{-\zeta t/m} \right)
\]

\[
\langle x^2 \rangle = \frac{2k_B T}{\zeta} \left( t - \frac{m}{\zeta} \left( 1 - e^{-\zeta t/m} \right) \right)
\]
Viscosity and diffusion

- Two time scales for mean squared displacement
  - Related to “collisional time”
  - At short times, the motion is ballistic
  - At long times, the motion is Brownian
    - Velocity doesn’t matter
    - RMSD can be related to diffusion coefficient
- The friction and diffusion coefficient are related!

\[
\langle x^2 \rangle = \frac{2k_B T}{\zeta} \left( t - \frac{m}{\zeta} (1 - e^{-\zeta t/m}) \right)
\]

\[
\tau = \frac{m}{\zeta}
\]

\[
\langle x^2 \rangle = \frac{2k_B T}{\zeta} \left( t - \tau \left( 1 - e^{-t/\tau} \right) \right)
\]

\[
\lim_{t \approx \tau} \langle x^2 \rangle = \frac{k_B T}{m} t^2 = \langle v^2 \rangle^{1/2} t^2
\]

\[
\lim_{t \gg \tau} \langle x^2 \rangle = \frac{2k_B T}{\zeta} t = 2Dt
\]

\[
D = \frac{k_B T}{\zeta}
\]
The Stokes-Einstein relationship

- Stokes law provides an expression for the viscous drag around various simple shapes
- This can be combined with the MSD derivation above
- The final relationship is called the Stokes-Einstein relation

\[ \zeta = 6\pi \eta a \]

\[ D = \frac{k_B T}{\zeta} = \frac{k_B T}{6\pi \eta a} \]
Mass action kinetics

- The rate of change in concentration or probability
- Describes a variety of phenomena: chemical reactions, binding events, etc.
- Relates changes in concentrations with time to (powers of) species concentrations
- Assumes large numbers of species
- Ignores fluctuations due to small copy numbers

\[ aA + bB \underset{k_2}{\overset{k_1}{\rightleftharpoons}} P \]

\[ \frac{dc_P(t)}{dt} = k_1 c_A^a(t) c_B^b(t) - k_2 c_P(t) \]
Michaelis-Menten kinetics

• Three basic reactions
  – Substrate-enzyme association
  – Substrate-enzyme dissociation
  – Catalysis and product-enzyme dissociation

• Steady-state assumption used below

\[
E + A \underset{k_{\text{off}}}{\overset{k_{\text{on}}}{\rightleftharpoons}} EA \overset{k_{\text{cat}}}{\rightarrow} E + P
\]

\[
K_d = \frac{k_{\text{off}}}{k_{\text{on}}}, \quad K_M = \frac{k_{\text{off}} + k_{\text{cat}}}{k_{\text{on}}}
\]

\[
\nu_{ss} = \frac{k_{\text{cat}} c_E (0) c_S (t)}{K_M + c_S (t)}
\]
What is a diffusion-limited reaction?

- Consider a reaction where the chemical step is *instantaneous*
  - All reactions which form ES complex go to products
  - The rate-limiting aspect of the reaction is binding to the enzyme
- Assume low substrate concentrations

\[
E + A \xrightleftharpoons[k_{off}]{k_{on}} EA \xrightarrow{k_{cat}} E + P
\]

\[
k_{cat} \gg k_{off}, \quad c_S(t) \ll \frac{k_{cat}}{k_{on}}, \quad K_M \approx \frac{k_{cat}}{k_{on}}
\]

\[
\nu_{ss} \approx \frac{k_{cat} c_E(0) c_S(t)}{k_{cat} + k_{on} c_S(t)} \approx k_{on} c_E(0) c_S(t)
\]
Diffusion-limited reactions

- **Typical** diffusional encounter rate is $10^9$ to $10^{10} \, \text{M}^{-1} \, \text{s}^{-1}$
  - There are lots of caveats, exceptions, etc.: protein flexibility, electrostatics, limited reaction surface
- **Smoluchowski encounter rate:**
  - Assumes spherical symmetry
  - Based on solution of PDE
  - No interactions: proportional to sum of diffusion coefficients and separation
  - Interactions: related to integral of potential
- Thought to represent evolutionary pressure
- Examples
  - Superoxide dismutase
  - Acetylcholinesterase
  - Barstar-barnase

\[
k_D(R) = \left[ \int_R^{\infty} \frac{e^{w(r)/k_B T}}{4\pi r^2 D(r)} \, dr \right]^{-1}
\]
\[
k_D^0(R) = 4\pi DR
\]
Methods for diffusional encounter simulations

• Discrete methods
  – Langevin dynamics
  – Brownian dynamics
  – Monte Carlo

• Continuum methods
  – Fokker-Planck
  – Smoluchowski equation
Discrete simulations of binding events

• Brownian dynamics
• Use as normal dynamics methods
  – Integrate stochastic equations of motion
  – Average: configurations, thermodynamics, etc. (nothing that depends on viscosities!)
• Use as encounter simulation method
First-order BD integration

- Calculate
  - Diffusion coefficient gradient
  - Potential of mean force gradient
  - Random displacement
- Works for large time steps provided the gradients don’t change (much)
- Position components can be x, y, z – or separate particle coordinates
- Coupling between particle diffusion components: hydrodynamic interactions

\[
\begin{align*}
  r_i(t + \Delta t) &= r_i(t) + \Delta t \sum_j \frac{\partial D_{ij}(t)}{\partial r_j} - \Delta t \sum_j D_{ij}(t) \frac{\partial W(t)}{\partial r_j} + R_i(\Delta t) \\
  \langle R_i(\Delta t) \rangle &= 0 \\
  \langle R_i(\Delta t) R_j(\Delta t) \rangle &= 2D_{ij}\Delta t
\end{align*}
\]
BD for encounter rate calculation

• Assumptions:
  – Low enzyme and substrate concentrations (no enzyme-enzyme or substrate-substrate interactions)
  – Diffusion control
  – Implicit solvent

• Basic idea: what is the probability that two molecules started at distance $b$ will encounter one another rather than wandering off to infinity?
BD for encounter rate calculation

• BD trajectory:
  – Start two molecules at a separation $b$ where the potential is centrosymmetric
  – Integrate BD equation of motion until
    • Molecules satisfy reaction criteria
    • Molecules exceed separation distance $q$
    • A maximum number of steps are taken

• Perform multiple BD trajectories:
  – Accumulate collision frequencies
  – Statistics are noisy; multiple runs needed!

Figure from: Northrup SH, Allison SA, McCammon JA. *J Chem Phys* 80 (4) 1517-24, 1984.
BD for encounter rate calculation
BD for diffusion-limited reactions

• Collision frequencies can be transformed into rates
• Think: flux through reactive site!
• If all collisions result in reaction (diffusion-limited), rate is related to:
  – Rate of diffusion to separation \( b \) (can use Smoluchowski formula)
  – Collision frequency
  – Probability that trajectories leaving \( q \) returns to \( b \)

\[
k = \frac{k_D(b)\beta}{1 - (1 - \beta)\Omega}
\]

\[
k_D(b) = \left[ \int_b^\infty \frac{e^{w(r)/k_BT}}{4\pi D(r)r^2} dr \right]^{-1}
\]

\[
\Omega = \frac{\int_q^\infty \frac{e^{w(r)/k_BT}}{4\pi D(r)r^2} dr}{\int_b^\infty \frac{e^{w(r)/k_BT}}{4\pi D(r)r^2} dr}
\]
BD for diffusion-influenced reactions

• If only some collisions result in reaction (probability $\alpha$), rate is related to:
  – All of above
  – Reaction probability $\alpha$
  – Probability $\Delta$ that unsuccessful encounter results in later collision

\[
k = \frac{\alpha k_D(b)}{1 - (1 - \alpha) \left\{ \Delta + (1 - \Delta) \left[ \frac{\beta}{1 - (1 - \beta) \Omega} \right] \right\}}
\]
Interactions in BD calculations

• Forces
  – Long-range influences only
  – Electrostatics: approximate charge-field calculations
    • Poisson-Boltzmann calculation for protein, charge model for ligand
    • No desolvation
    • Little “internal dielectric” screening (some effective charge methods)

• Diffusion coefficients
  – Should include rotation, translation, and configuration changes
  – No hydrodynamic interactions
    • Probably OK for small ligands
    • Stokes-Einstein isotropic diffusion coefficients
    • Coefficients do not depend on distance or configuration
  – Hydrodynamic interactions
    • Include water-mediated effects
    • Oseen and other (approximate) analytic forms
    • Configuration- and distance-dependent

\[ \mathbf{F}_{i}^{\text{lig}} \approx q_{i}^{\text{lig}} \mathbf{E}^{\text{prot}} \]

\[ D_{ij}^{\alpha\beta} \approx \frac{k_B T}{c \pi \eta} \left( \frac{\delta_{ij}}{a_i} \mathbf{I} + \frac{1 - \delta_{ij}}{2R_{ij}} \left( \mathbf{I} + \frac{\mathbf{r}_{ij}}{r_{ij}^2} \right) \right) \]

\[ R_{ij} = \begin{cases} a_i + a_j & r_{ij} < a_i + a_j \\ r_{ij} & r_{ij} \geq a_i + a_j \end{cases} \]
Application to acetylcholinesterase

- Hydrolytic enzyme in neuromuscular junction
- Subject of extensive computational (BD) and experimental study
- Properties:
  - Diffusion-limited catalysis
  - Long, narrow active site gorge
  - Significant electrostatic influences
AChE/TMA binding

- Binding of neurotransmitter-like molecule to acetylcholinesterase
- Diffusion-controlled binding
- Significant dependence on [NaCl]
- Sensitivity to charged residues

DHFR-TS substrate channeling

• Bifunctional enzyme: thymidylate synthase produces dihydrofolate used by dihydrofolate reductase

• Electrostatic steering between active sites enhances efficiency
  – Reduces diffusional broadening
  – Does not “direct” between sites

Protein-protein encounter

- Same basic procedure as before
- Reaction criteria are harder to evaluate – often a variable in the simulation
- Effective charge method
  - Use full electrostatic grid for one molecule
  - Use a smaller number of charges: termini and charged residues
- Usually neglect:
  - Desolvation terms
  - Hydrodynamic interactions (with exceptions)
  - Ion relaxation
  - Flexibility
  - Substrate-substrate interactions

Actin polymerization

- BD simulations of actin polymerization
- Reproduced experimental observation of faster polymerization at “barbed” filament end
- Implicated electrostatics in faster binding to barbed end
- Also observed effect of ADF-cofilin on polymerization

Lots of protein-protein association rates

- Systems studied:
  - Barstar-barnase
  - AChE-Fas2
  - Cyt C peroxidase-Cyt C
  - HyHEL antibodies and lysozyme
- Agreement with experiment is good
- Antibody/lysozyme and AChE-Fas2 rates overestimated: not diffusion-limited?
- Electrostatics aren’t always helpful!
Continuum diffusion simulation methods

- **Discrete methods**
  - Solve stochastic ODEs
  - Provide atomic problem resolution
  - Facilitate integration of stochastic phenomena
  - Software: MCell, UHBD, etc.

- **Continuum methods**
  - Solve deterministic PDEs
  - Bridge larger length scales
  - Facilitate integration of continuum mechanics phenomena
  - Software: SMOL
Continuum diffusion motivation

• Demonstrate:
  – Accurate description of enzyme binding kinetics (steady-state and time-dependent)
  – Simulation of synapse electrophysiology
  – Extreme adaptivity of methods to bridge length scales

• Long-term goals:
  – Integrate continuum and discrete methods
  – More complete description of cellular-scale processes
Smoluchowski equation

\[
\frac{\partial \rho(x)}{\partial t} = \nabla \cdot J(x) = \nabla \cdot D(x) \left[ \nabla \rho(x) \right] + \beta \rho(x) \nabla W(x)
\]

Concentration change over time

- Flux
- Diffusion term
- Drift term

- \( \rho(x) = \bar{\rho} \)
- "Bulk" boundary condition

- \( \rho(x) = 0 \)
- Reactive boundary condition

- \( n(x) \cdot J(x) = 0 \)
- Reflective boundary condition

The observable: the time-dependent rate constant

\[ k(t) = \int J(s) \cdot n(s) ds \]
Advances and outlook

• Receptor flexibility
• Detailed binding mechanisms
• Imperfect reactivity; calculate “re-entrant” trajectories

Camphor release pathway from cytochrome P450 from Guallar lab.

Advances and outlook

- Cellular simulations
- Proteomics-scale interactions
- Crowded environments
