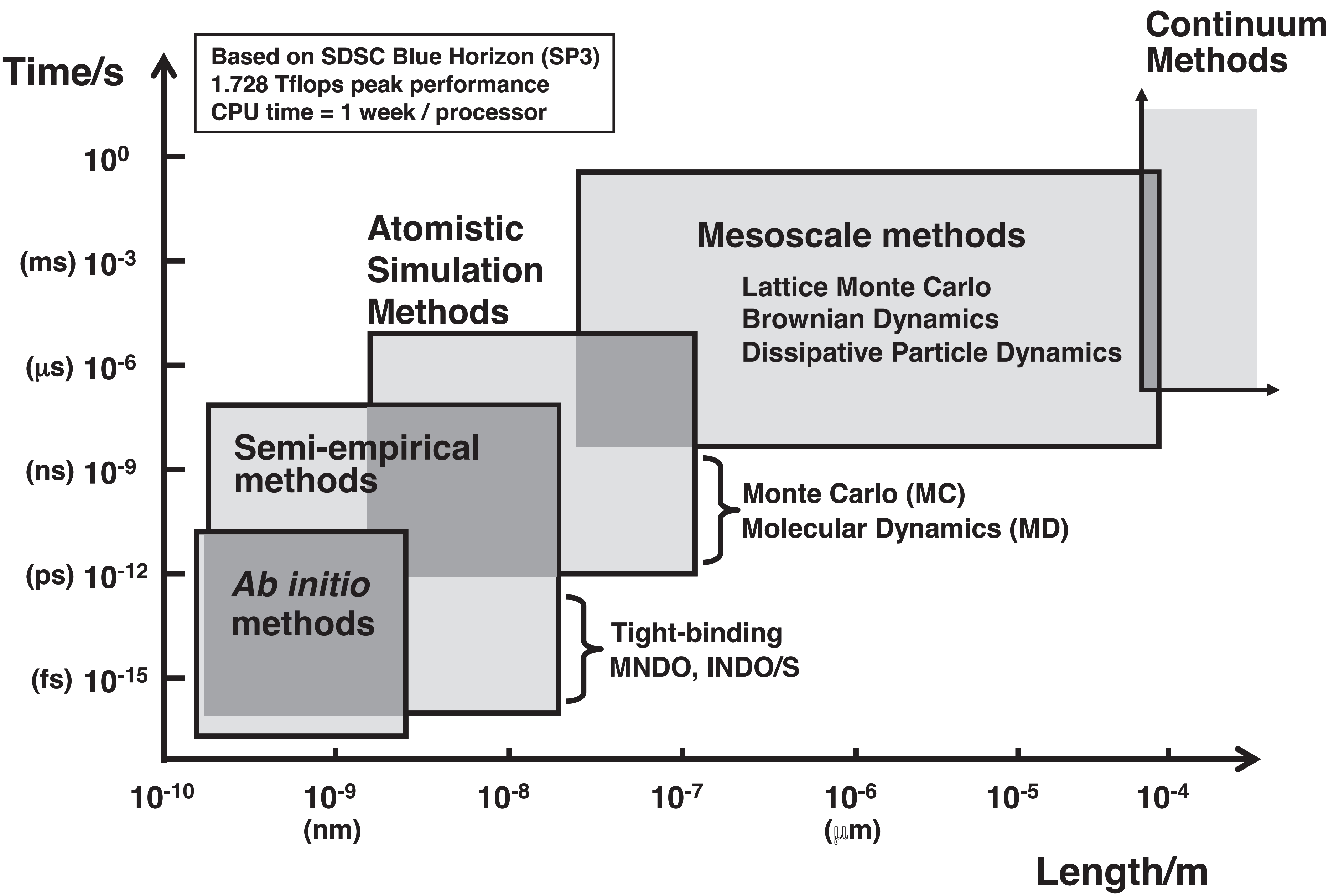
Calculating Binding Rate Constants via Brownian Dynamics

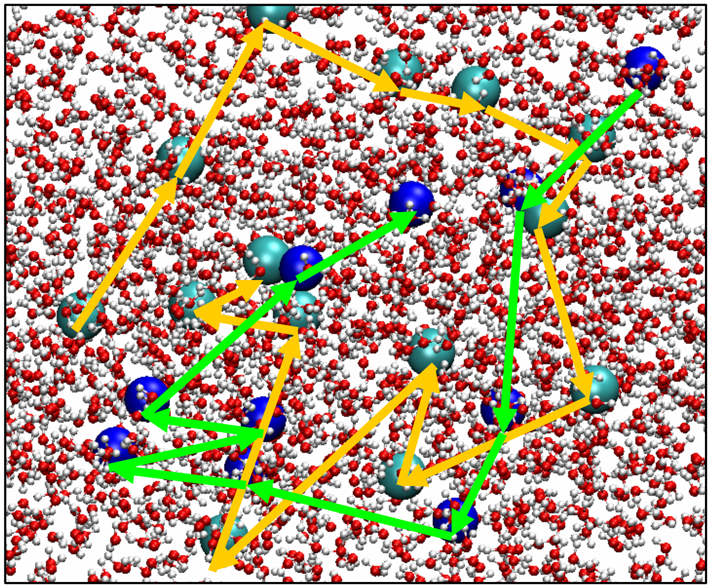
CHEM 430

Multi-Scale Modeling



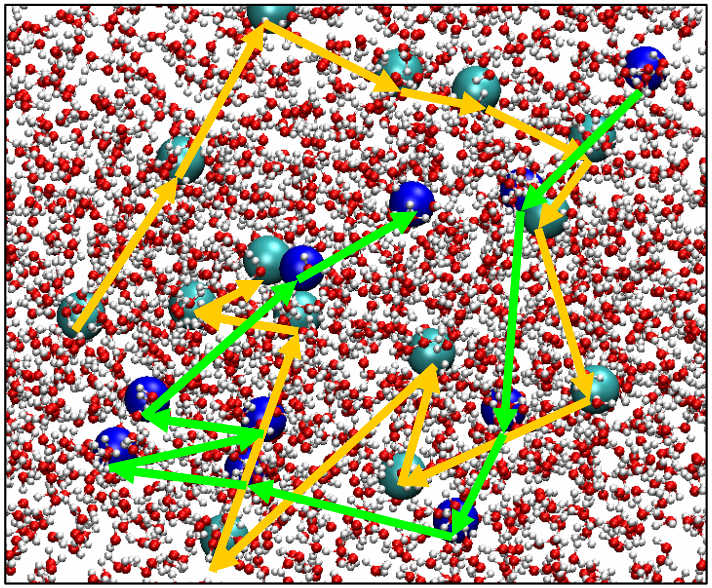
Condensed Phase Kinetics

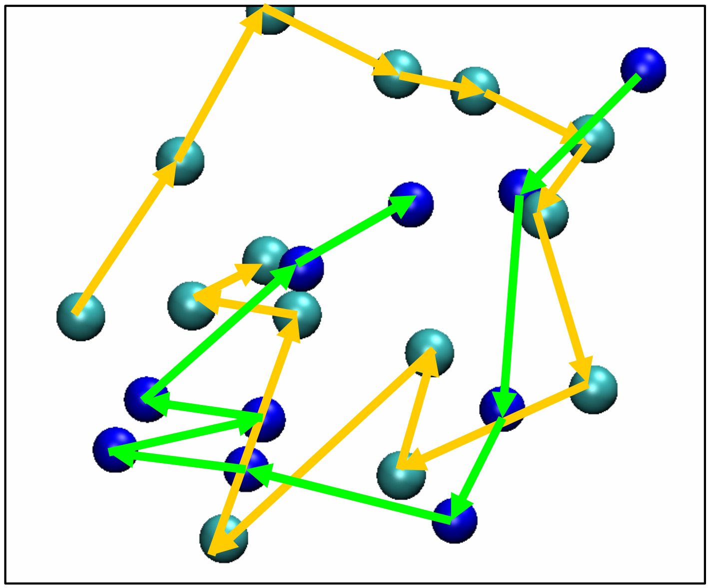
- The kinetic theory of gases involves ballistic motion
- Kinetics in liquids and other condensed phases involves diffusion
- What are the major characteristics?
  - Many molecules interacting at once
  - Short mean free paths
  - No "memory" of momenta



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

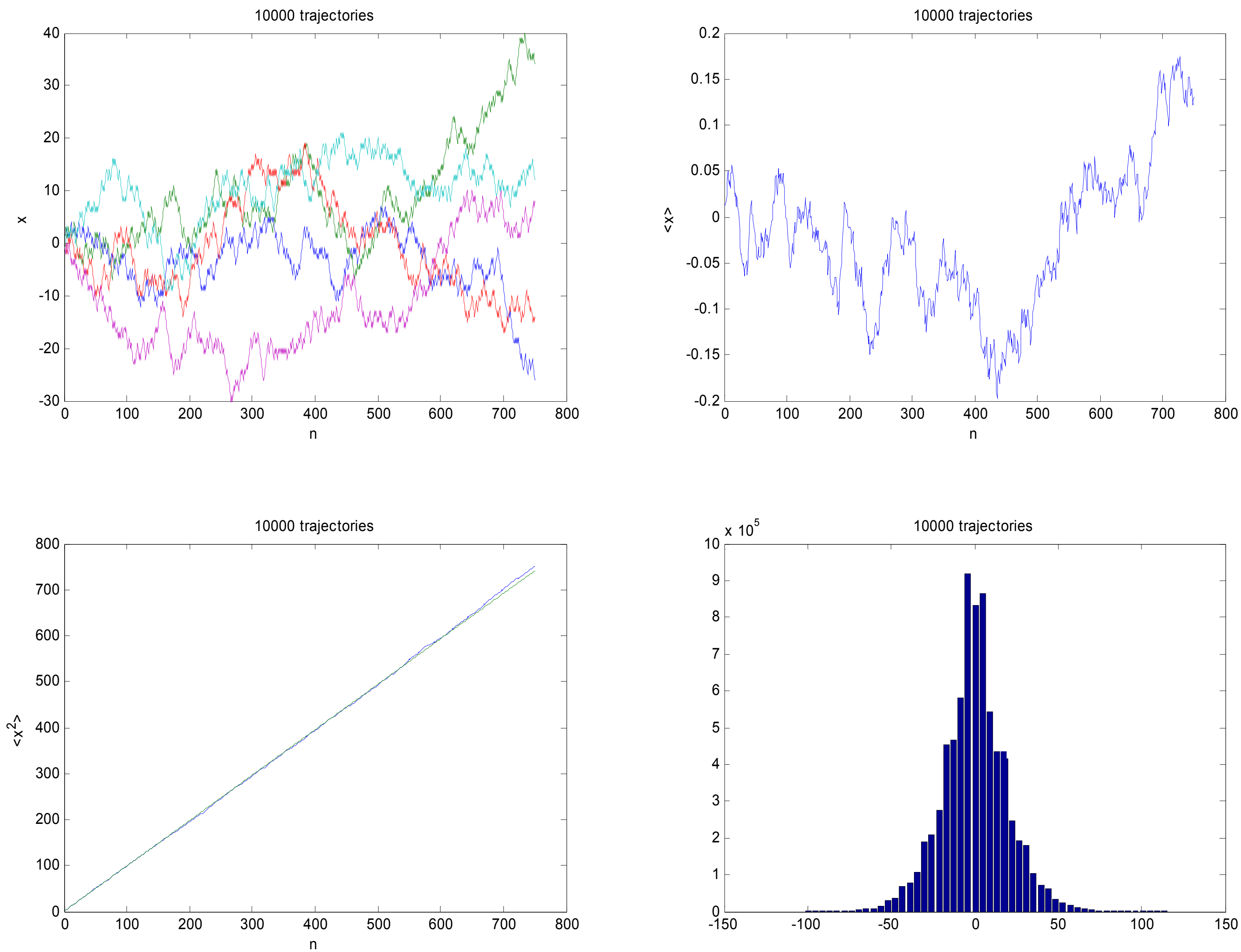
$$P(n_+, N) = \frac{N!}{n_+!(N-n_+)!}\left(\frac{1}{2}\right)^N$$

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

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

$$\langle m^2 \rangle \sim N$$

1D Random Walk



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…

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

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

$$\langle x^2 \rangle_{1\mathrm{D}} = 2Dt$$

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

$$\langle x^2 + y^2 \rangle_{2\mathrm{D}} = 4Dt$$

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

$$\langle x^2 + y^2 + z^2 \rangle_{3\mathrm{D}} = 6Dt$$

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 <*xv*> with particle initially at origin
  - Relate to <x^2> and solve

$$m\left\langle \frac{d}{dt}(xv) \right\rangle = -\zeta\left\langle xv \right\rangle + m\left\langle v^2 \right\rangle$$

$$= -\zeta\left\langle xv \right\rangle + k_B T$$

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

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

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

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!

$$\langle x^2 \rangle = \frac{2k_BT}{\zeta}\left(t - \frac{m}{\zeta}\left(1 - e^{-\zeta t/m}\right)\right)$$

$$\tau = \frac{m}{\zeta}$$

$$\langle x^2 \rangle = \frac{2k_BT}{\zeta}\left(t - \tau\left(1 - e^{-t/\tau}\right)\right)$$

$$\lim_{t \ll \tau} \langle x^2 \rangle = \frac{k_BT}{m} t^2$$

$$= \langle v^2 \rangle^{1/2} t^2$$

$$\lim_{t \gg \tau} \langle x^2 \rangle = \frac{2k_BT}{\zeta} t$$

$$= 2Dt$$

$$D = \frac{k_BT}{\zeta}$$

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

$$E + A \underset{k_{\text{off}}}{\overset{k_{\text{on}}}{\rightleftharpoons}} EA \xrightarrow{k_{\text{cat}}} E + P$$

$$K_d = \frac{k_{\text{off}}}{k_{\text{on}}}, K_M = \frac{k_{\text{off}} + k_{\text{cat}}}{k_{\text{on}}}$$

$$v_{ss} = \frac{k_{\text{cat}} c_E(0) c_S(t)}{K_M + c_S(t)}$$

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

$$E + A \underset{k_{\text{off}}}{\overset{k_{\text{on}}}{\rightleftharpoons}} EA \xrightarrow{k_{\text{cat}}} E + P$$

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

$$v_{ss} \approx \frac{k_{\text{cat}} c_E(0) c_S(t)}{\frac{k_{\text{cat}}}{k_{\text{on}}} + c_S(t)} \approx k_{\text{on}} c_E(0) c_S(t)$$

Diffusion-Limited Reactions

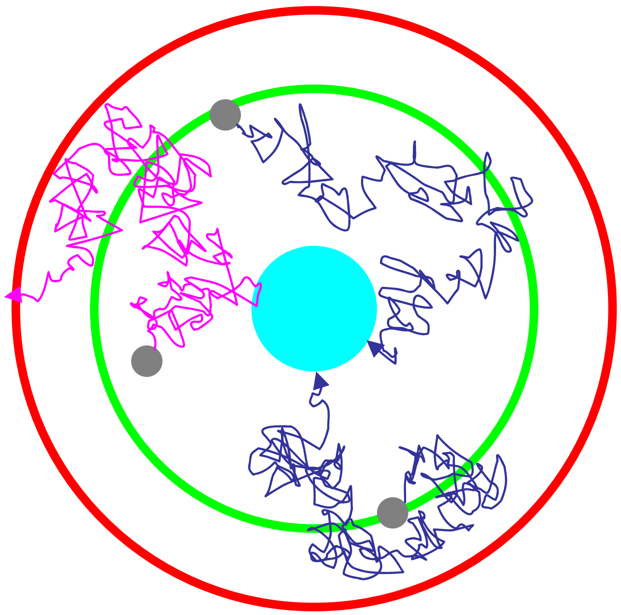
- *Typical* diffusional encounter rate is 10^9 to 10^{10} M^{-1} s^{-1}
  - There are lots of caveats, exceptions, etc.: protein flexibility, electrostatics, limited reaction surface
- Smoluchowski encounter rate:
  - Assumes spherical symmetry
  - Based on solution of PDE
  - No interactions: proportional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

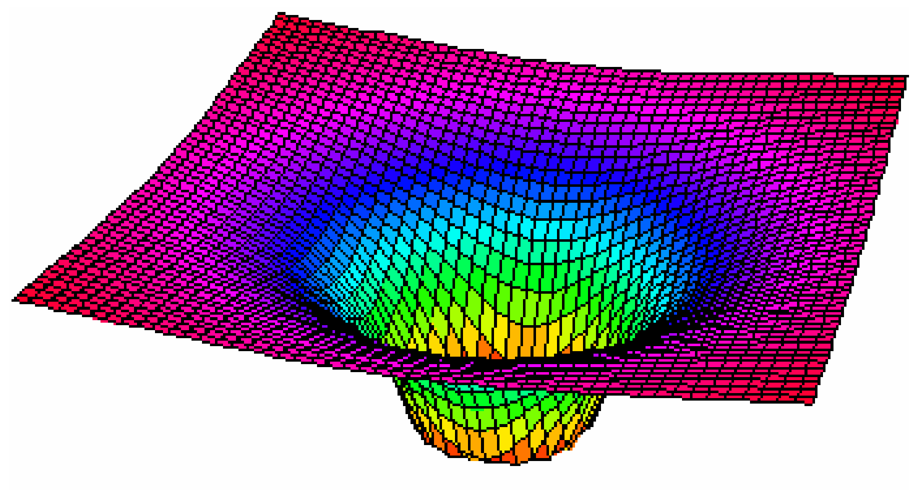
$$k_D(R) = \left[\int_R^\infty \frac{e^{w(r)/k_B T}}{4\pi r^2 D(r)} dr \right]^{-1}$$

$$k_D^0(R) = 4\pi D R$$

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

$$r_i\left(t+\Delta t\right)=r_i\left(t\right)+\Delta t\sum_j \frac{\partial D_{ij}\left(t\right)}{\partial r_j}-\Delta t\sum_j D_{ij}\left(t\right)\frac{\partial W\left(t\right)}{\partial r_j}+R_i\left(\Delta t\right)$$

$$\left\langle R_i\left(\Delta t\right)\right\rangle = 0$$

$$\left\langle R_i\left(\Delta t\right)R_j\left(\Delta t\right)\right\rangle = 2D_{ij}\Delta t$$

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!

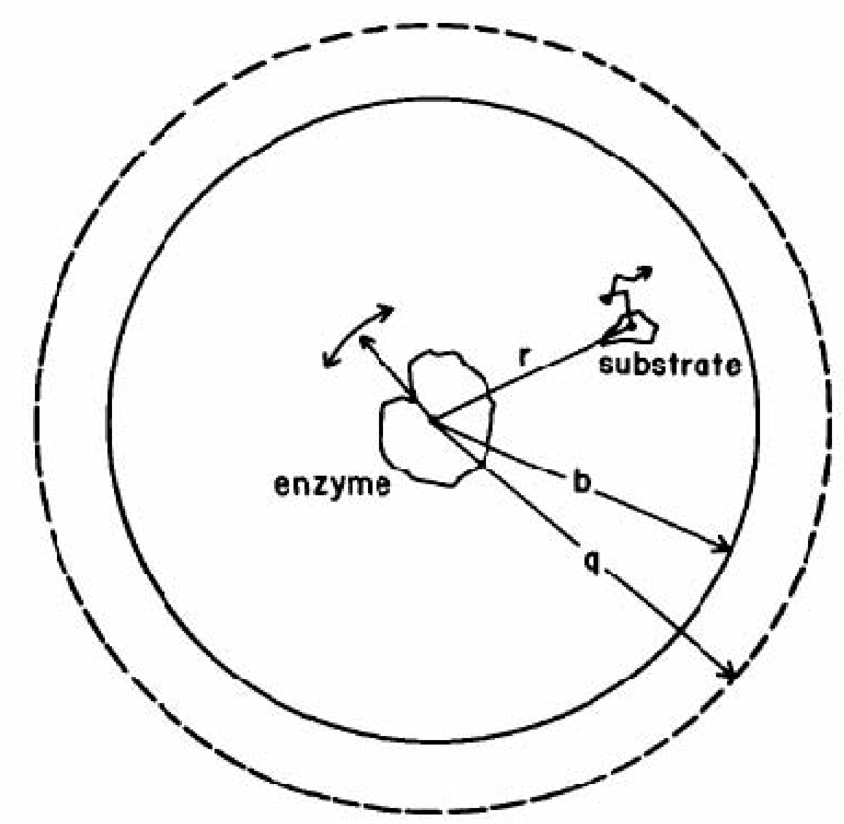
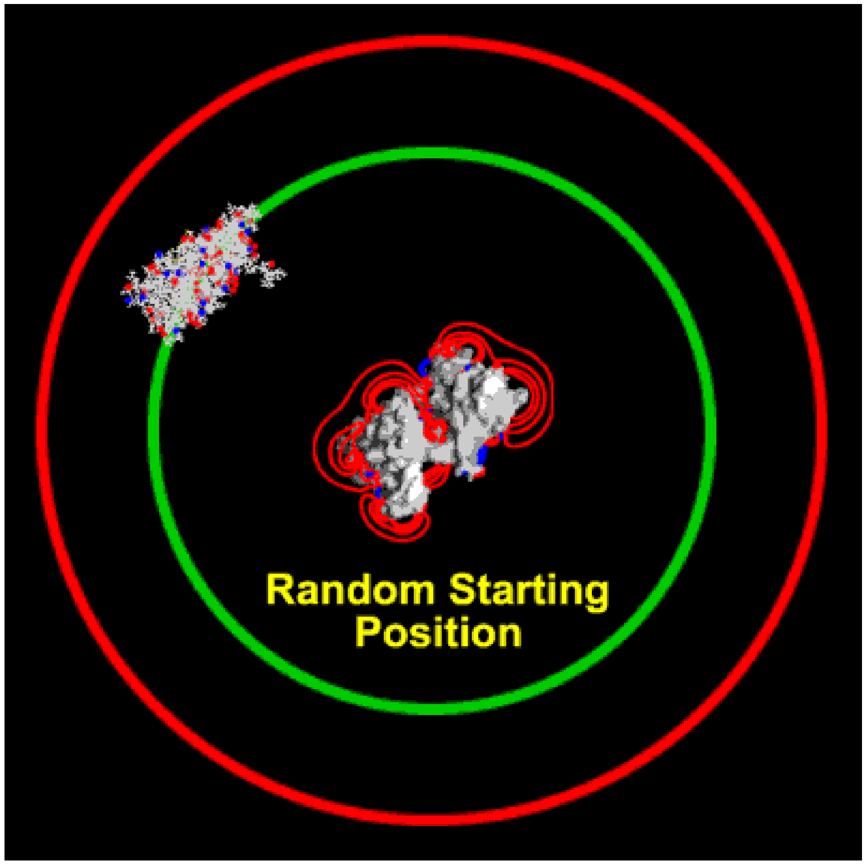


Figure from: Northrup SH, Allison SA, McCammon JA. *J Chem Phys* **80** (4) 1517-24, 1984.

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*

$$k = \frac{k_D(b)\,\beta}{1-(1-\beta)\Omega}$$

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

$$\Omega = \frac{\int_q^\infty \frac{e^{w(r)/k_B T}}{4\pi D(r) r^2}\,dr}{\int_b^\infty \frac{e^{w(r)/k_B T}}{4\pi D(r) r^2}\,dr}$$

BD for Diffusion-Influenced Reactions

- If only some collisions result in reaction (probability *α*), rate is related to:
  - All of above
  - Reaction probability *α*
  - Probability *Δ* that unsuccessful encounter results in later collision

$$k = \frac{\alpha k_D(b)\left[\dfrac{\beta}{1-(1-\beta)\Omega}\right]}{1-(1-\alpha)\left\{\Delta + (1-\Delta)\left[\dfrac{\beta}{1-(1-\beta)\Omega}\right]\right\}}$$

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

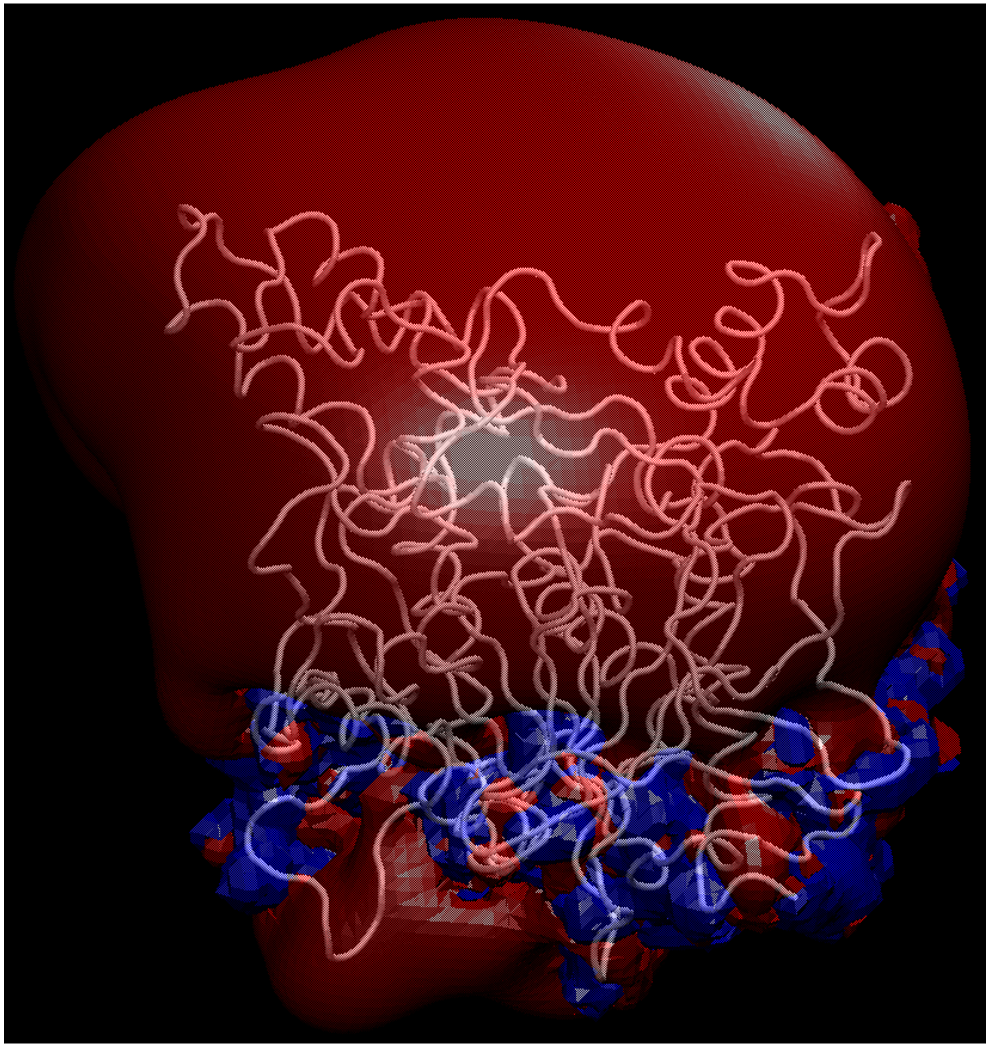
$$\mathbf{F}_i^{\mathrm{lig}} \approx q_i^{\mathrm{lig}} \mathbf{E}^{\mathrm{prot}}$$

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

$$R_{ij} = \begin{cases} a_i + a_j & r_{ij} < a_i + a_j \\ r_{ij} & r_{ij} \geq a_i + a_j \end{cases}$$

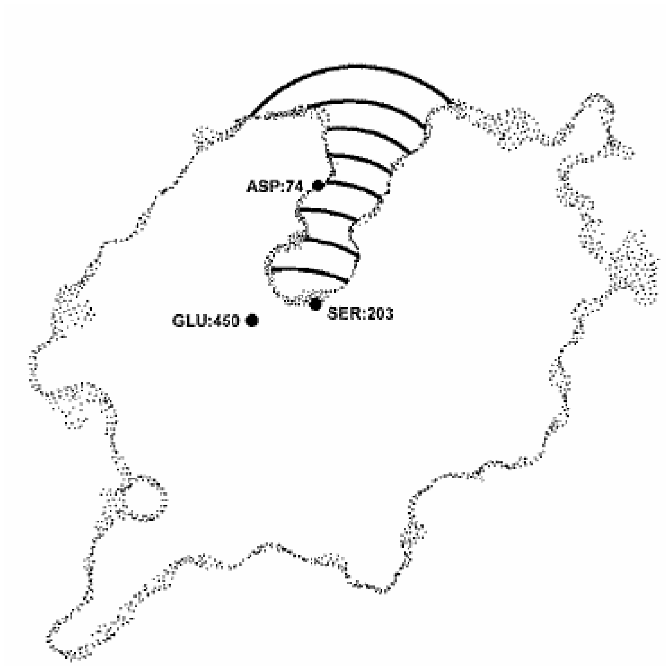
Application to Acetylcholinesterase

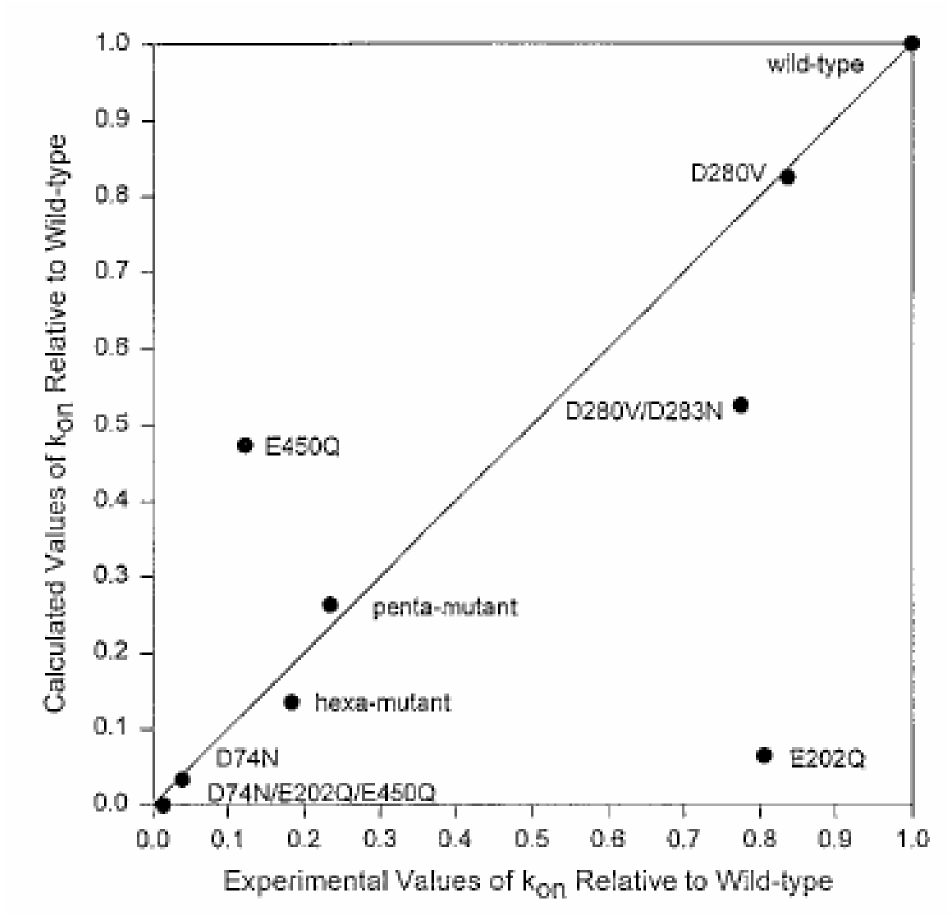
- Hydrolytic enzyme in neuromuscular junction
- Subject of extensive computational (BD) and experimental study
- Properties:
  - Diffusion-limited catalysis
  - Long, narrow active site gorge
  - Significant electrostatic influences



AChE/TMA Binding

- Binding of neurotransmitter-like molecule to acetylcholinesterase
- Diffusion-controlled binding
- Big dependence on [NaCl]
- Sensitivity to charged residue mutations

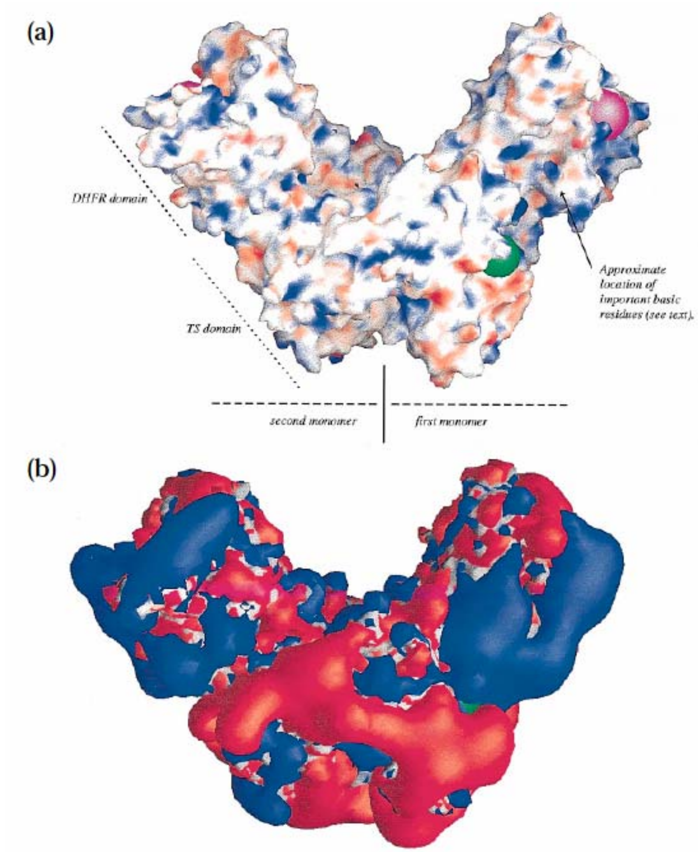




Figures from: Tara S, et al. *Biopolymers* **46** (7) 465-74, 1998.

DHFR-TS Substrate Channeling

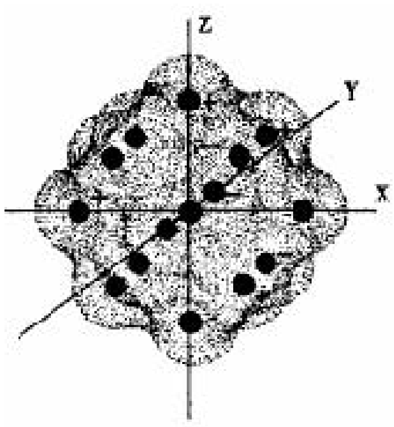
- Bifunctional enzyme: thymidylate synthase produces dihydrofolate used by dihydrofolate reductase
- Electrostatic steering between active sites enhances efficiency
  - Reduces diffusional broadening
  - Does not “direct” between sites



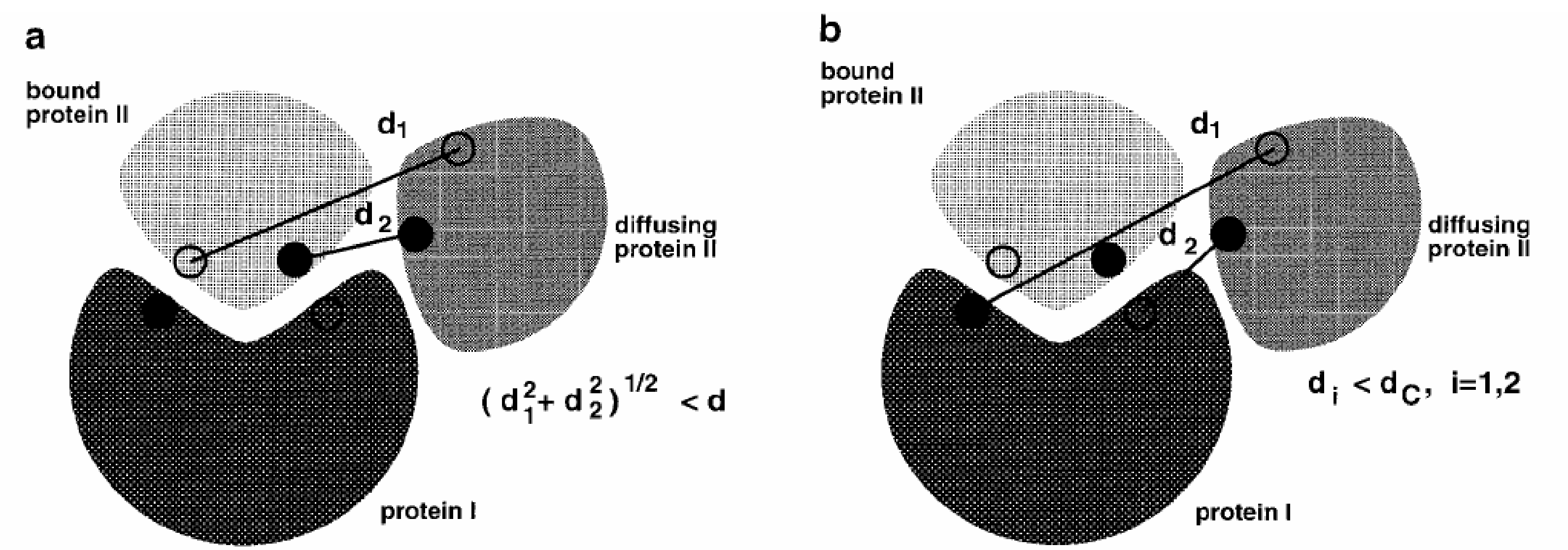
Figures from: Elcock AH, et al. *J Mol Biol* **262**, 370-4, 1996.

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

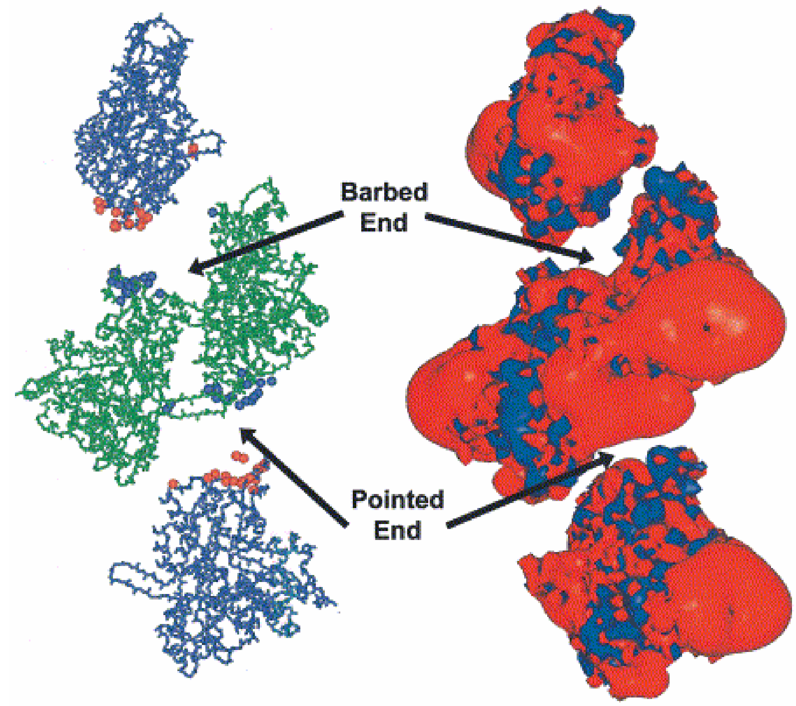


Figures from: Gabdoulline RR, Wade RC. *J Phys Chem* **100** 3868-78, 1996 and ibid. *Methods* **14** 329-41, 1998.



Actin Polymerization

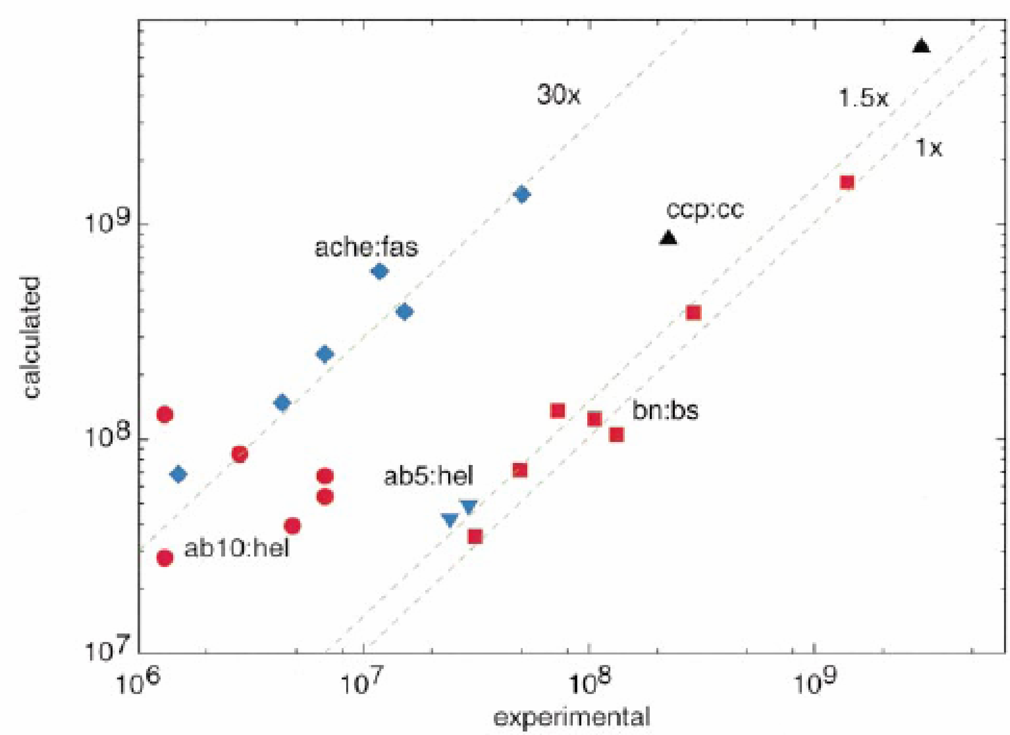
- BD simulations of actin polymerization
- Reproduced experimental observation of faster polymerization at “barbed” filament end
- Implicated electrostatics in faster binding to barbed end
- Also observed effect of ADF-cofilin on polymerization

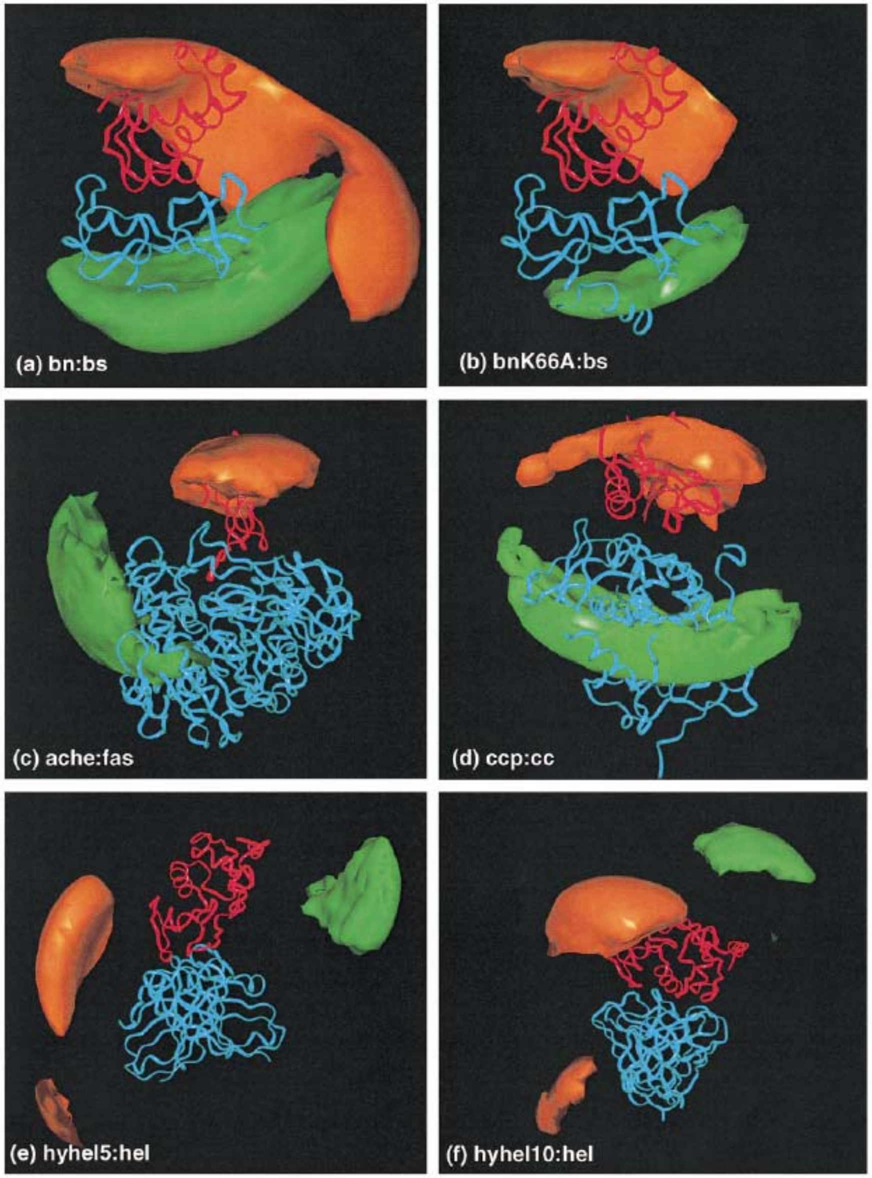


Figures from: Sept D, Elcock AH, McCammon JA. *J Mol Biol* **294** (5) 1181-9, 1999.

Lots of Protein-Protein Association Rates

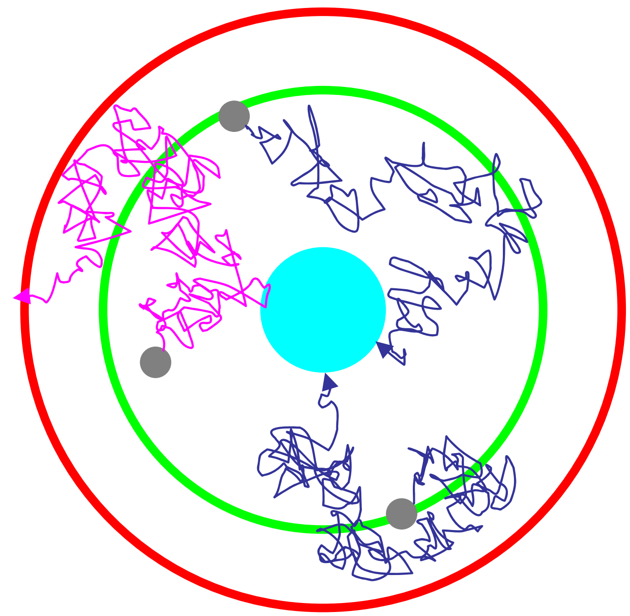
- Systems studied:
  - Barstar-barnase
  - AChE-Fas2
  - Cyt C peroxidase-Cyt C
  - HyHEL antibodies and lysozyme
- Agreement with experiment is good
- Antibody/lysozyme and AChE-Fas2 rates overestimated: not diffusion-limited?
- Electrostatics aren't always helpful!
- Figures from: Gabdoulline RR, Wade RC. *J Mol Biol* **306** 1139-55, 2001.

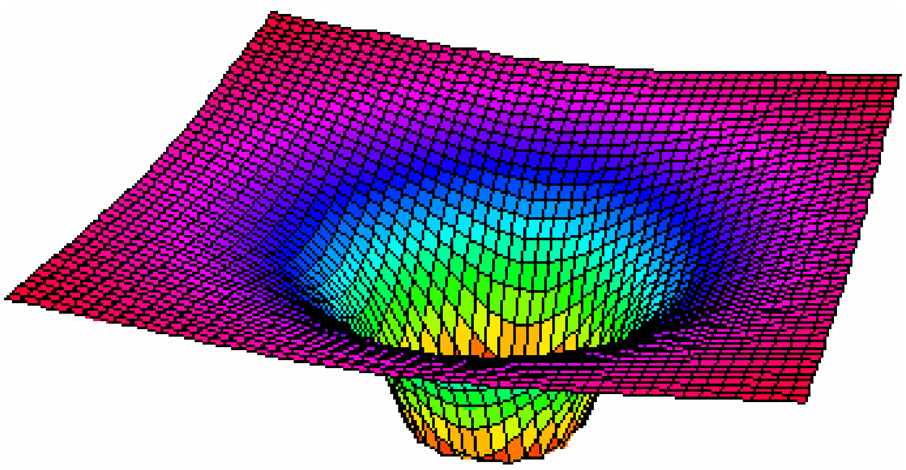




Continuum Diffusion Simulation Methods

- 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

Concentration change over time

Diffusion term

Drift term

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

Flux

Diffusion coefficient

External potential

$$\rho(x) = \bar{\rho}$$

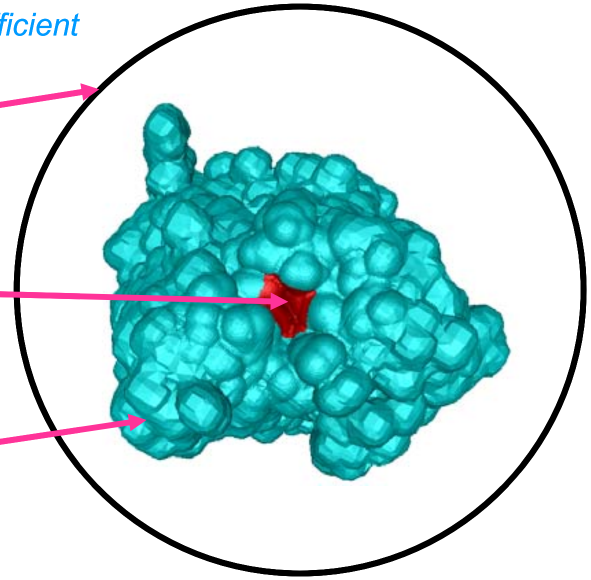
"Bulk" boundary condition

$$\rho(x) = 0$$

Reactive boundary condition

$$n(x) \cdot J(x) = 0$$

Reflective boundary condition

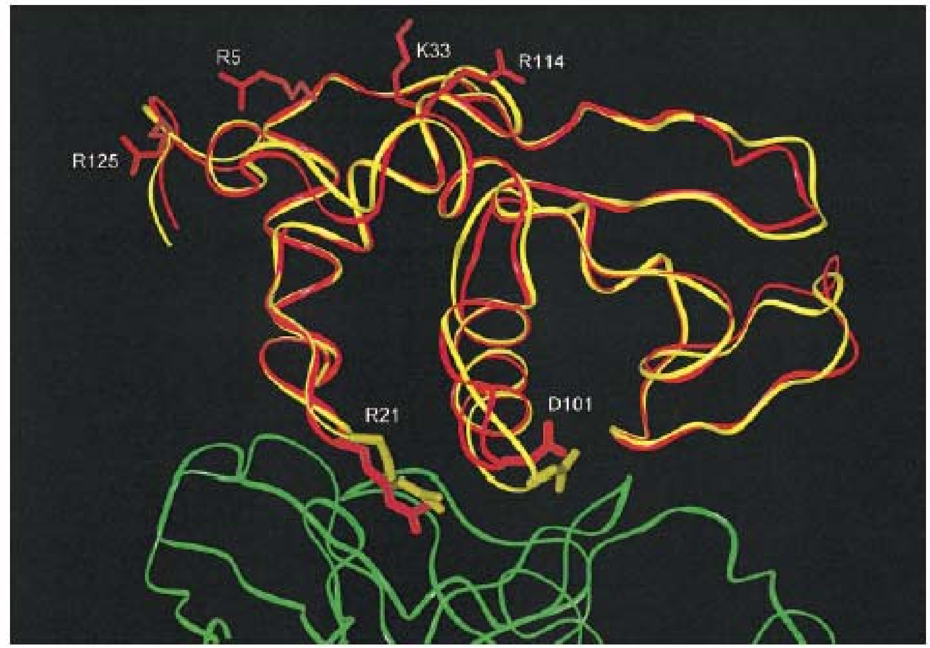


$$k(t) = \oint J(s) \cdot n(s) ds$$

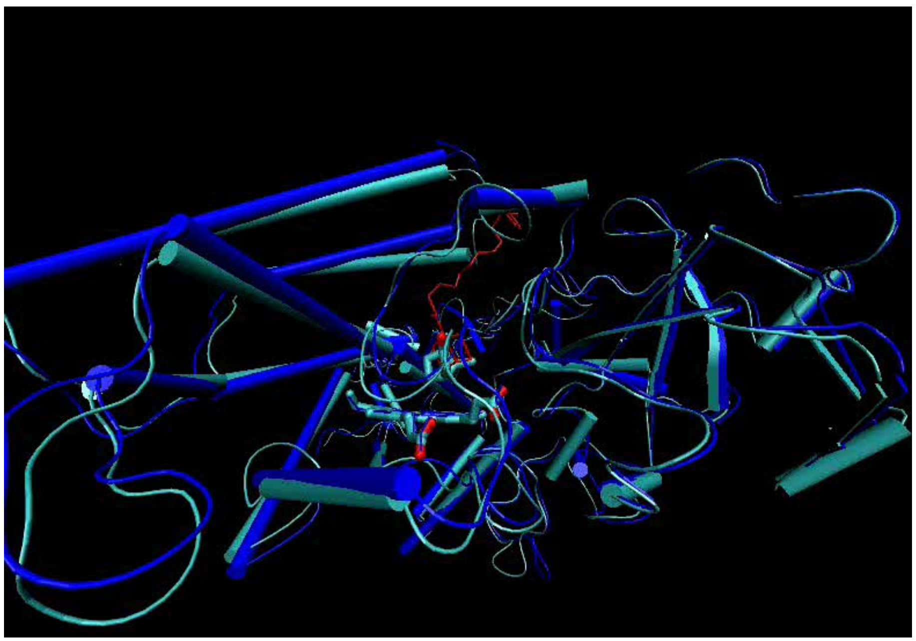
The observable: the time-dependent rate constant

Advances and Outlook

- Receptor flexibility
- Detailed binding mechanisms
- Imperfect reactivity; calculate "re-entrant" trajectories



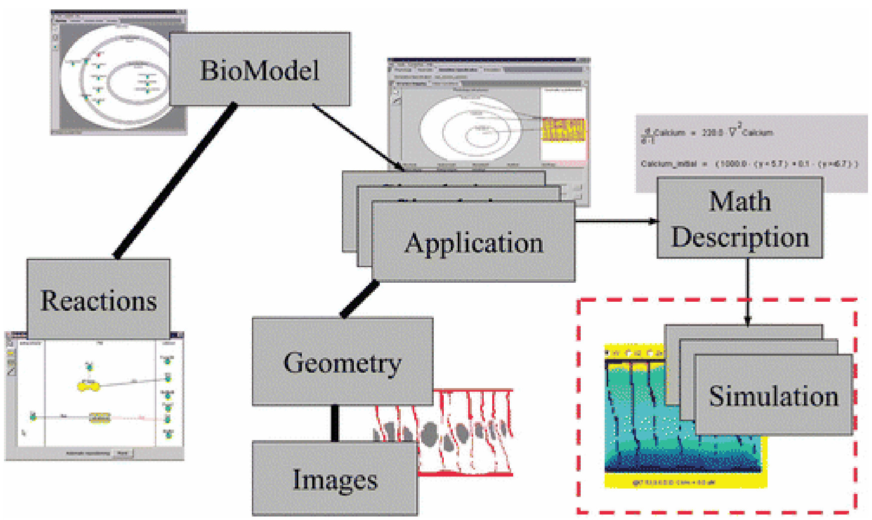
HEL-antibody flexibility in lysozyme binding from Gabdoulline RR, Wade RC. *J Mol Biol* **306** 1139-55, 2001.



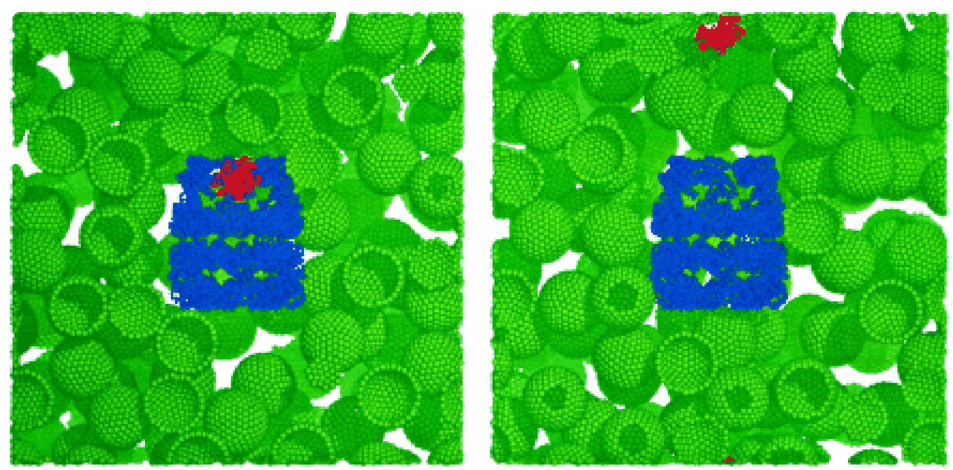
Camphor release pathway from cytochrome P450 from Guallar lab.

Advances and Outlook

- Cellular simulations
- Proteomics-scale interactions
- Crowded environments



Virtual Cell schematic from Slepchenko BM, Schaff JC, Carson JH, Loew LM. *Annu Rev Biophys Biomol Struct* **31** 423-41, 2002.



Crowding and GroEL simulation from Elcock AH. *PNAS* **100** (5) 2340-4, 2003.