MULTIVALENCY

NATURE'S ADJUSTABLE TOOL FOR CHANGING PROTEIN BINDING STRENGTH

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OVERVIEW

- Protein binding
- Multivalency
- Model framework overview
  - Figuring out the model
  - Figuring out effective concentration
  - Applications

UNIVERSITY OF MINNESOTA
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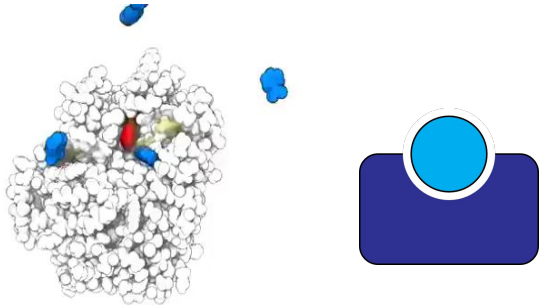


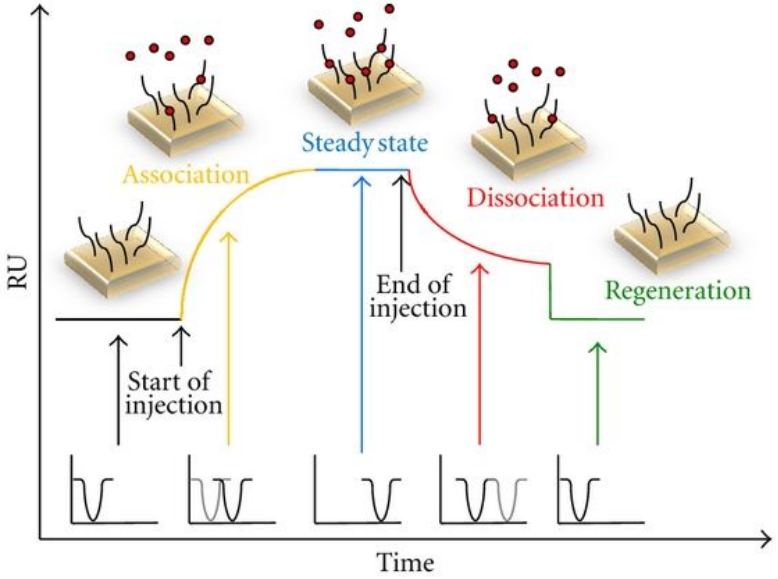
PROTEIN BINDING

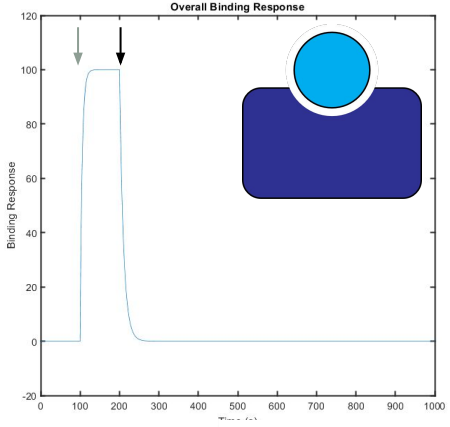
BINDING STRENGTH

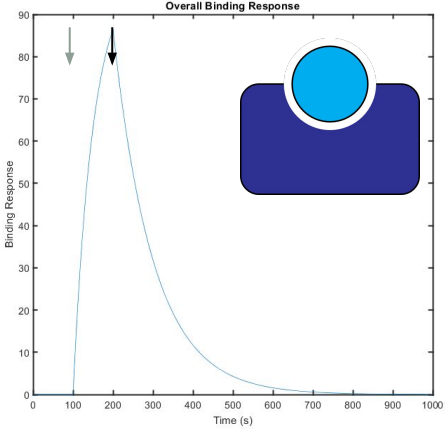
- KD-dissociation constant
  - 1nM, 1mM
- Daily dose
  - Will it be a good pill
- Multivalency can increase it significantly

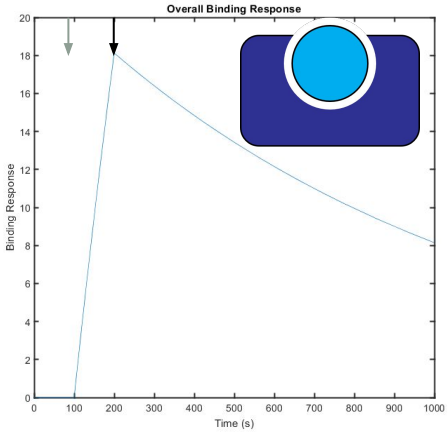
BINDING KINETICS I

- Two phases:





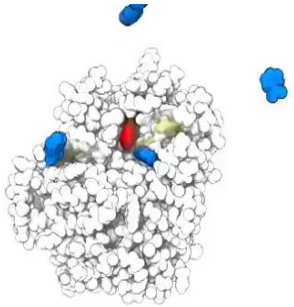




BINDING KINETICS II

- In steady state:

$$k_{on}[R][L] = k_{off}[RL]$$

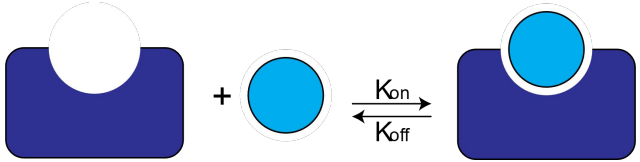


- The speed of transition:

$$v_{association} = k_{on}[R][L]$$
$$v_{dissociation} = k_{off}[RL]$$

- In differential equation form:

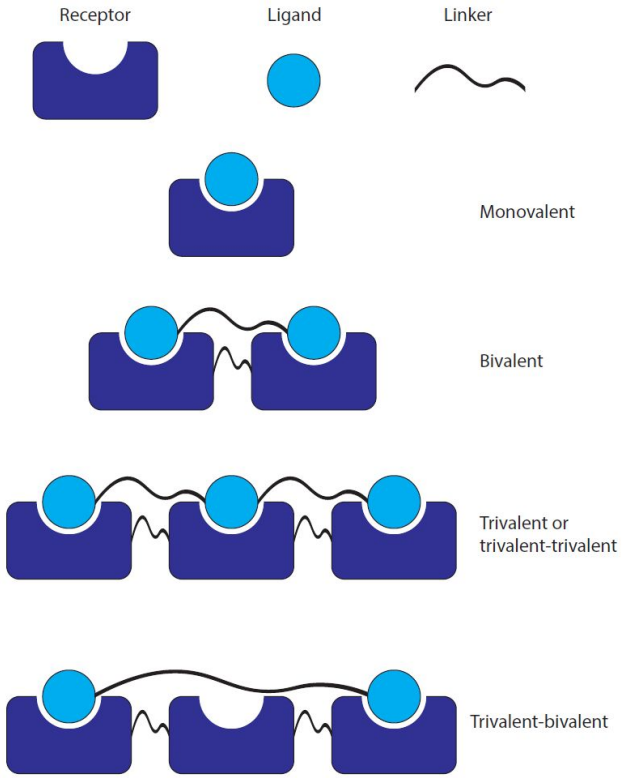
$$\frac{d[R]}{dt} = -k_{on}[R][L] + k_{off}[RL]$$
$$\frac{d[RL]}{dt} = +k_{on}[R][L] - k_{off}[RL]$$



MULTIVALENCY

MULTIVALENCY

- Multivalent molecules
  - Bivalent
  - Trivalent
  - Higher valencies
- Complexity
  - Conformations - states
  - Kinetics
  - Composition – homo, hetero
  - Biological functions

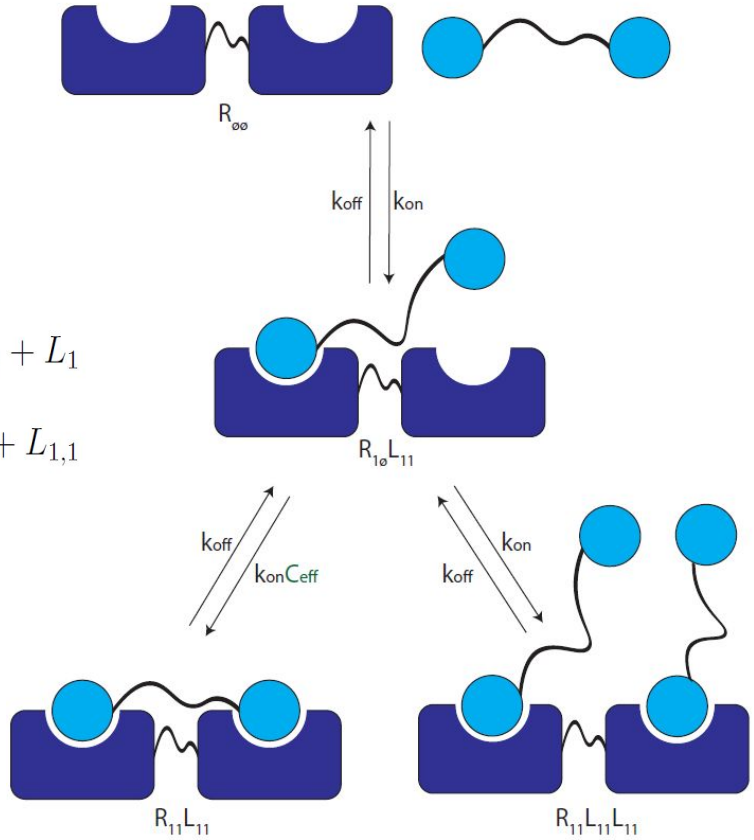


BIVALENT EXAMPLE

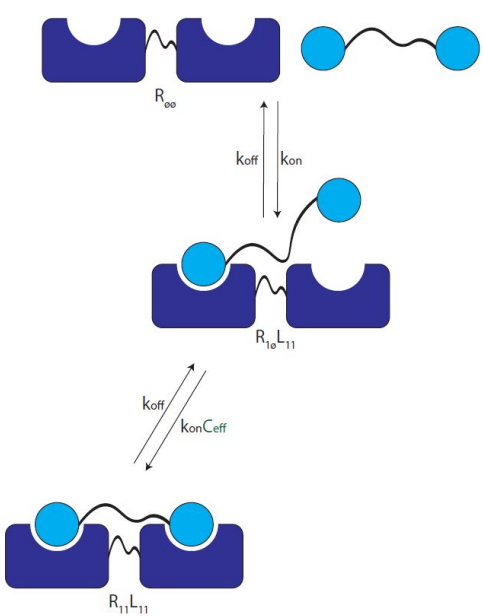
$$Monovalent: \quad R_1L_1 \underset{dissociation}{\overset{association}{\rightleftharpoons}} R_\varnothing + L_1$$

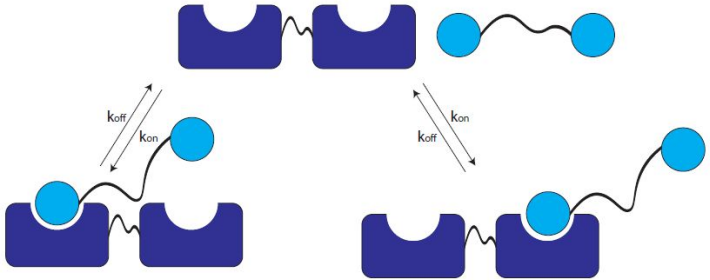
$$Multiple: \quad R_{1,1}L_1L_1 \underset{dissociation}{\overset{association}{\rightleftharpoons}} R_{1,\varnothing}L_1 + L_1 \underset{dissociation}{\overset{association}{\rightleftharpoons}} R_{\varnothing,\varnothing} + L_1$$

$$Multivalent: \quad R_{1,1}L_{1,1} \underset{dissociation}{\overset{association}{\rightleftharpoons}} R_{1,\varnothing}L_{1,1} \underset{dissociation}{\overset{association}{\rightleftharpoons}} R_{\varnothing,\varnothing} + L_{1,1}$$



FIGURING OUT ALL THE STATES

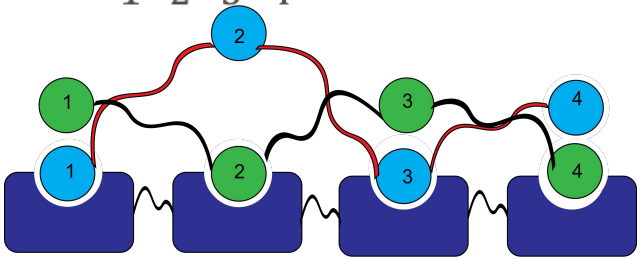




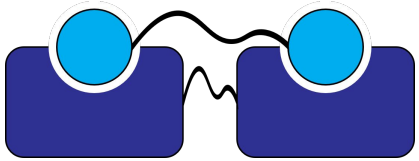
IDENTIFYING STATES AND TRANSITIONS

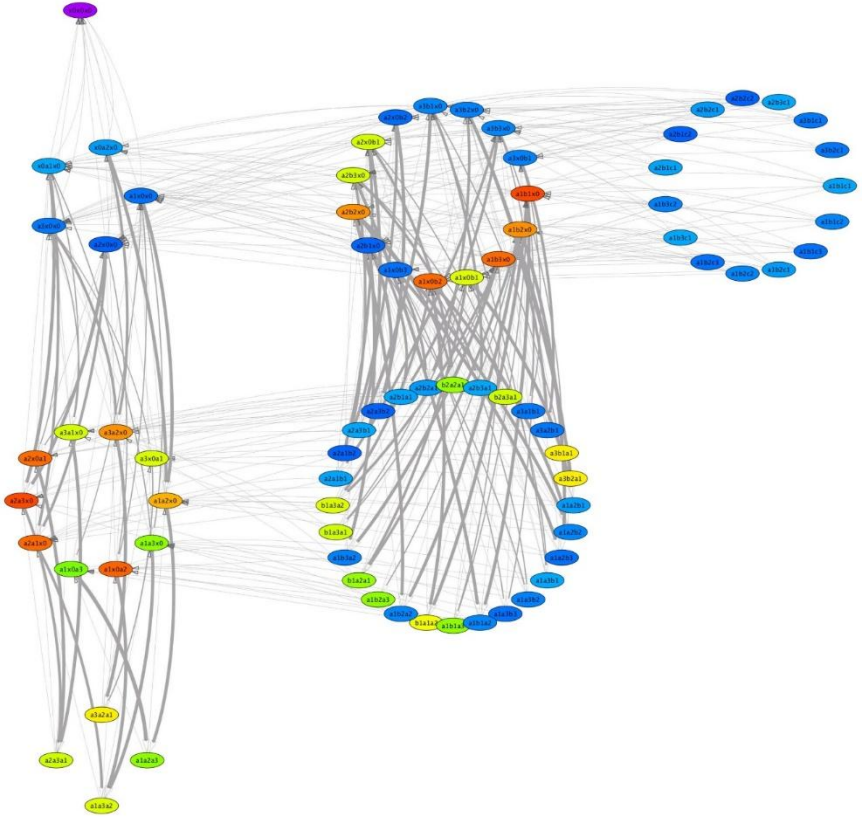
- Increasing complexity
- Notation of states:

$$R_{L_1^1 L_2^2 L_3^1 L_4^2} L_{1234} L_{1234}$$



$$R_{L_1^1 L_2^1} L_{12} \mid R_{L_1^1 L_1^1} L_{11} \mid R_{12}$$



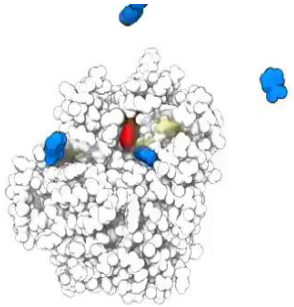


DYNAMIC NETWORK

BINDING KINETICS II

- In steady state:

$$k_{on}[R][L] = k_{off}[RL]$$



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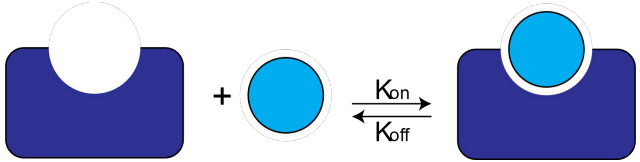


ODES (OPTIONAL)

- Write an ode based on text
- You have n=10000 tasks
- Each task have 10% chance each hour to finish
- When will 95% of the tasks finish
- $\frac{dn}{dt} = -0.9n$
- Due to overheating each hour your computation is 1% slower
- $\frac{dh}{dt} = 0.01$, $\frac{dn}{dt} = -(0.9 + h)n$
- Calculate n(t) = 500
  - Math
  - Numerical

EXAMPLE

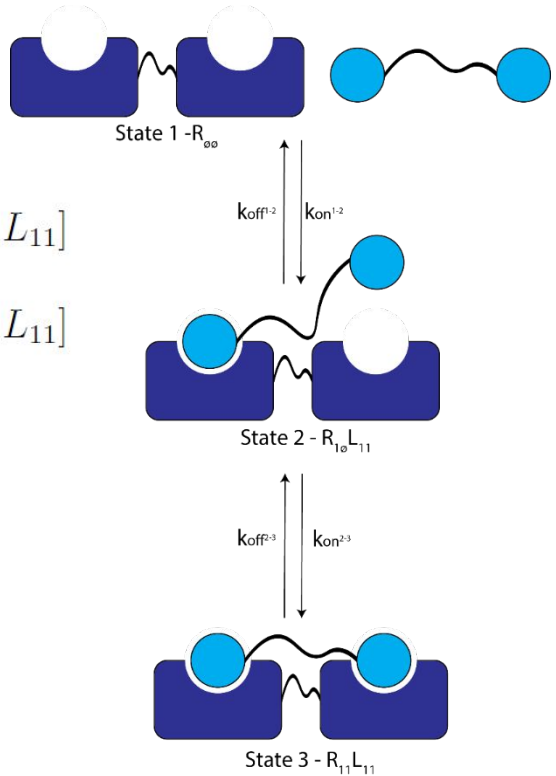
$$\frac{d[R_{\varnothing\varnothing}]}{dt} = -2k_{on}^{1-2}[R_{\varnothing\varnothing}][L_{11}] + k_{off}^{1-2}[R_{1\varnothing}L_{11}]$$

$$\frac{d[R_{1\varnothing}L_{11}]}{dt} = +2k_{on}^{1-2}[R_{\varnothing\varnothing}][L_{11}] - k_{off}^{1-2}[R_{1\varnothing}L_{11}] \quad -k_{on}^{2-3}[R_{1\varnothing}] + 2k_{off}^{2-3}[R_{11}L_{11}]$$

$$\frac{d[R_{11}L_{11}]}{dt} = \quad +k_{on}^{2-3}[R_{1\varnothing}] - 2k_{off}^{2-3}[R_{11}L_{11}]$$

$$\begin{vmatrix} \frac{d[R_{\varnothing\varnothing}]}{dt} \\ \frac{d[R_{1\varnothing}L_{11}]}{dt} \\ \frac{d[R_{11}L_{11}]}{dt} \end{vmatrix} = \begin{vmatrix} -2k_{on}^{1-2}[L_{11}] & +k_{off}^{1-2} & \\ +2k_{on}^{1-2}[L_{11}] & -k_{off}^{1-2} - k_{on}^{2-3} & +2k_{off}^{2-3} \\ & +k_{on}^{2-3} & -2k_{off}^{2-3} \end{vmatrix} \begin{vmatrix} [R_{\varnothing\varnothing}] \\ [R_{1\varnothing}L_{11}] \\ [R_{11}L_{11}] \end{vmatrix}$$

$$\frac{d\boldsymbol{x}(t)}{dt} = \boldsymbol{A}(t)\boldsymbol{x}(t)$$

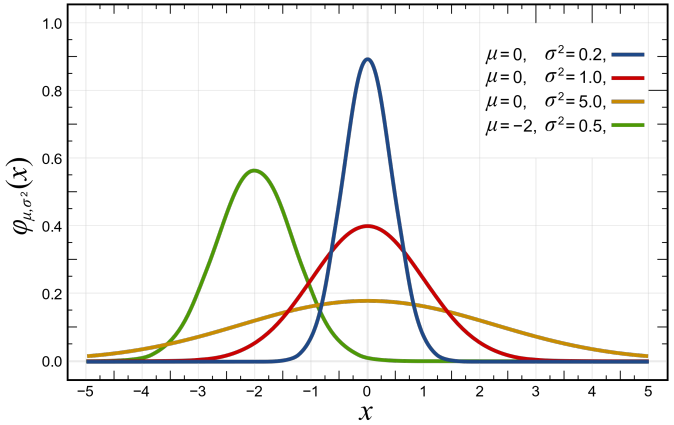


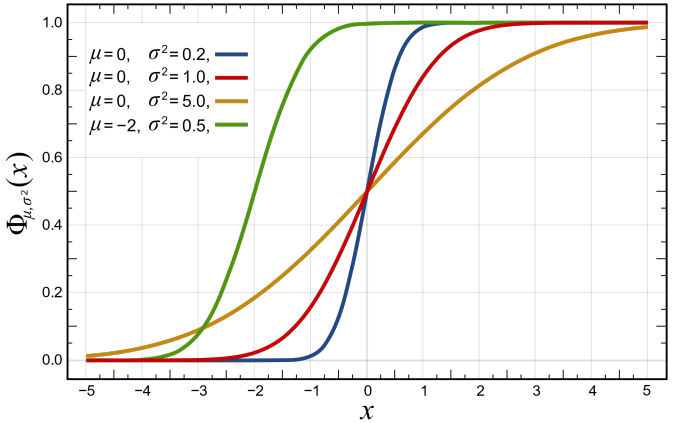
EFFECTIVE CONCENTRATION

INTEGRALS

- PDF – probability density function
- Integral of a PDF
- Integral the product of two PDF

- Bonus: relation between integration and derivation





EFFECTIVE VALUES

for i in [1:1000]

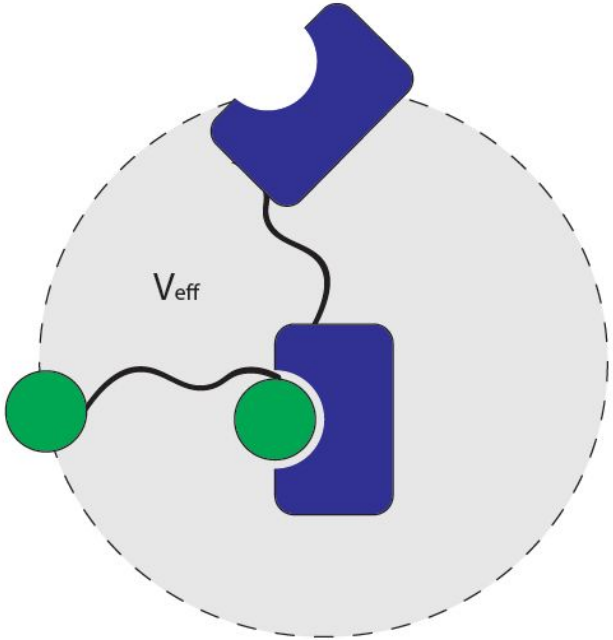
- x = rnorm(mean=10, sd=2)
- y = rnorm(mean=7, sd=2)
- If x-y < 0.01
  - counter +=1

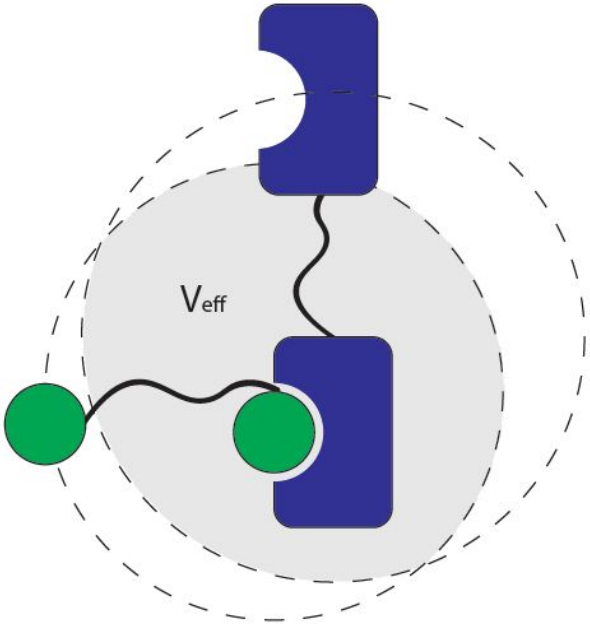
for i in [1:1000]

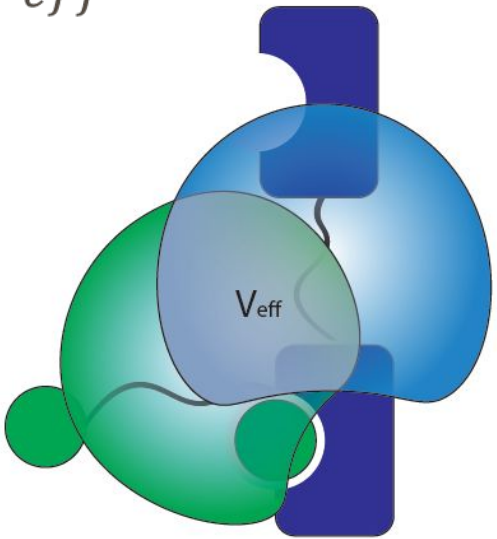
- x = rnorm(mean=10, sd=2)
- for i in [1:eff]
  - z = runif(0,20)
  - If x-y < 0.01
    - counter +=1

EFFECTIVE VOLUME OR CONCENTRATION

- $v_{association} = k_{on}[R][L]$ -> $k_{on}[R]C_{eff}$



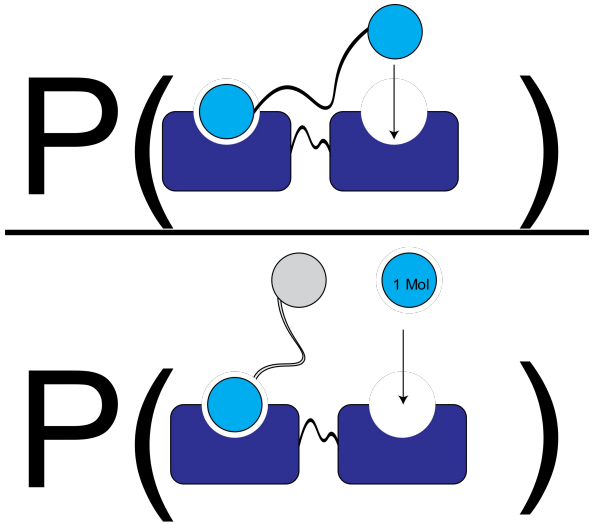




EFFECTIVE CONCENTRATION

- Effective Concentration:

$$P(binding|RL) = P\left(binding\middle|R, L_{C_{eff}}\right)$$

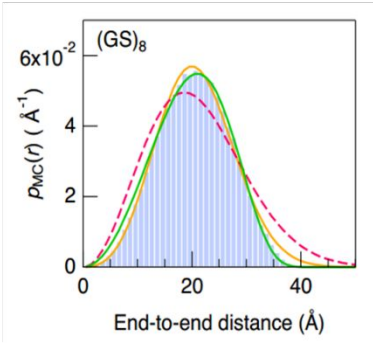


- Binding or collision probability:

$$\int f_L(x) f_R(x) dx$$

- Linker end-to-end PDF

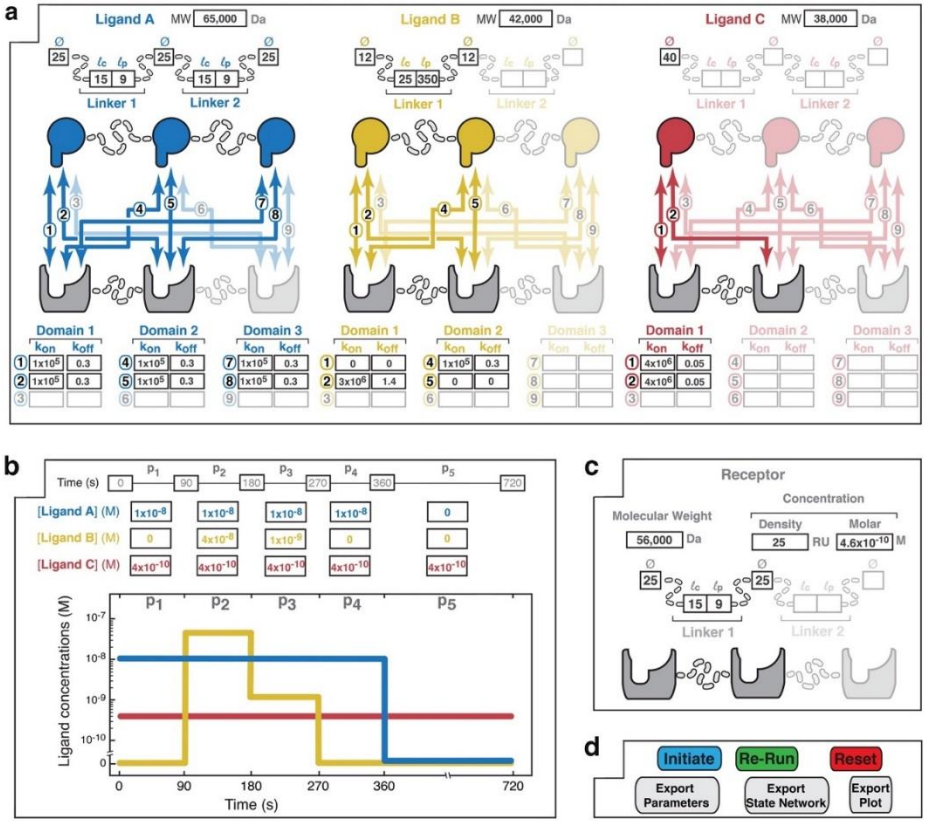
$$PDF(x, y, z, Lp, Lc)\ \alpha\ \frac{1}{\left(1 - \frac{x^2 + y^2 + z^2}{L_c^2}\right)^{\frac{2}{9}}} e^{\left(-\frac{9L_c}{8L_p}\frac{1}{1-\frac{x^2+y^2+z^2}{L_c^2}}\right)}$$



USING THE MODLE

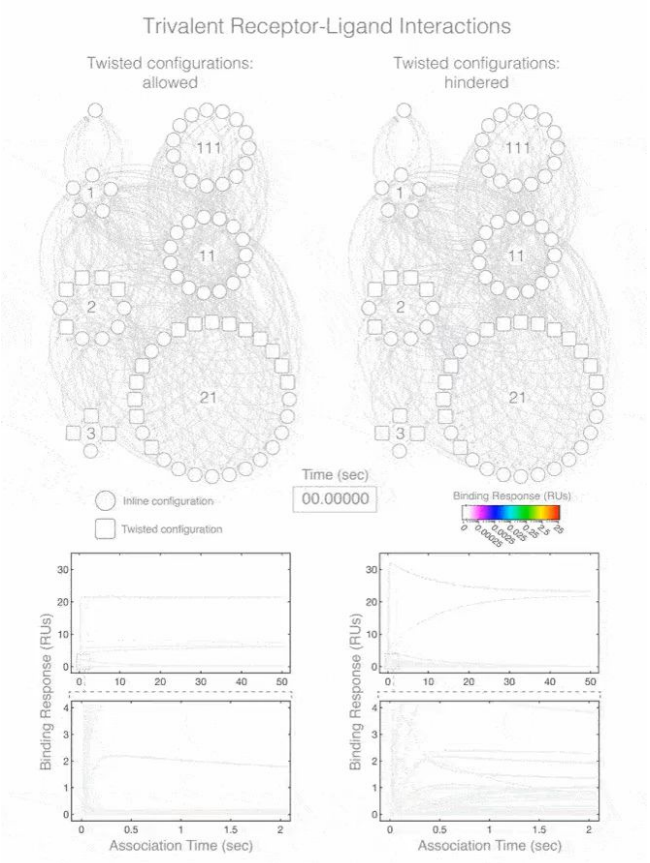
MVSIM

- Ligand and receptor parameters
  - Topology
  - Monovalent rate constants
- Ligand concentration
  - Three ligand, multiple steps
- Output
  - SPR or concentration curves
  - Microstates
  - Network representation



MICROSTATES OF A COMPLEX SYSTEM

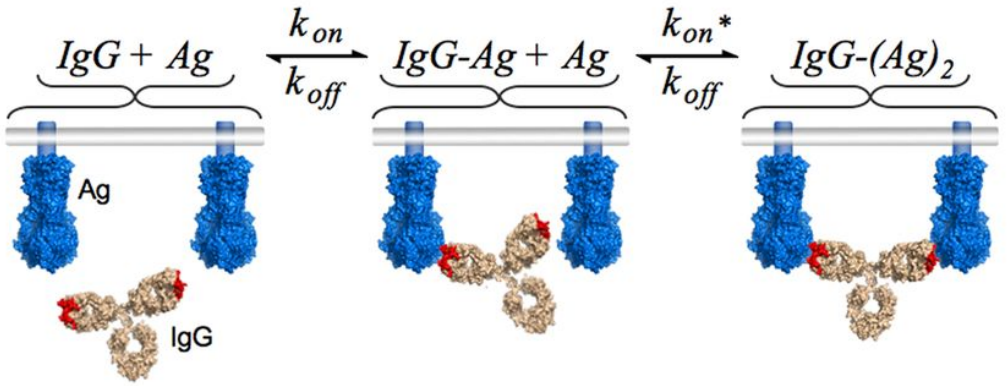
- Solved by ODE15s solver
- Can predict avidity, specificity
- Explanation for uncanonical behaviors
- Applicable to a wide range of multivalent interaction
- Support for protein engineering

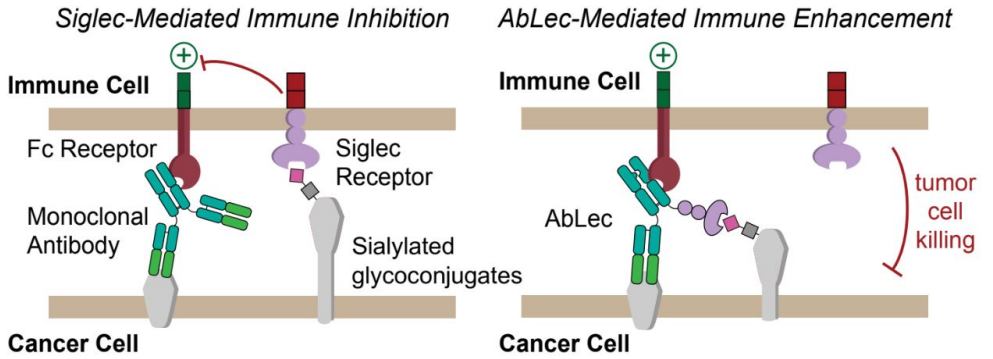


APPLICATION EXAMPLE – ANTIBODY DESIGN

- Bispecific antibodies
- Surface antigen concentration
- Specific targeting

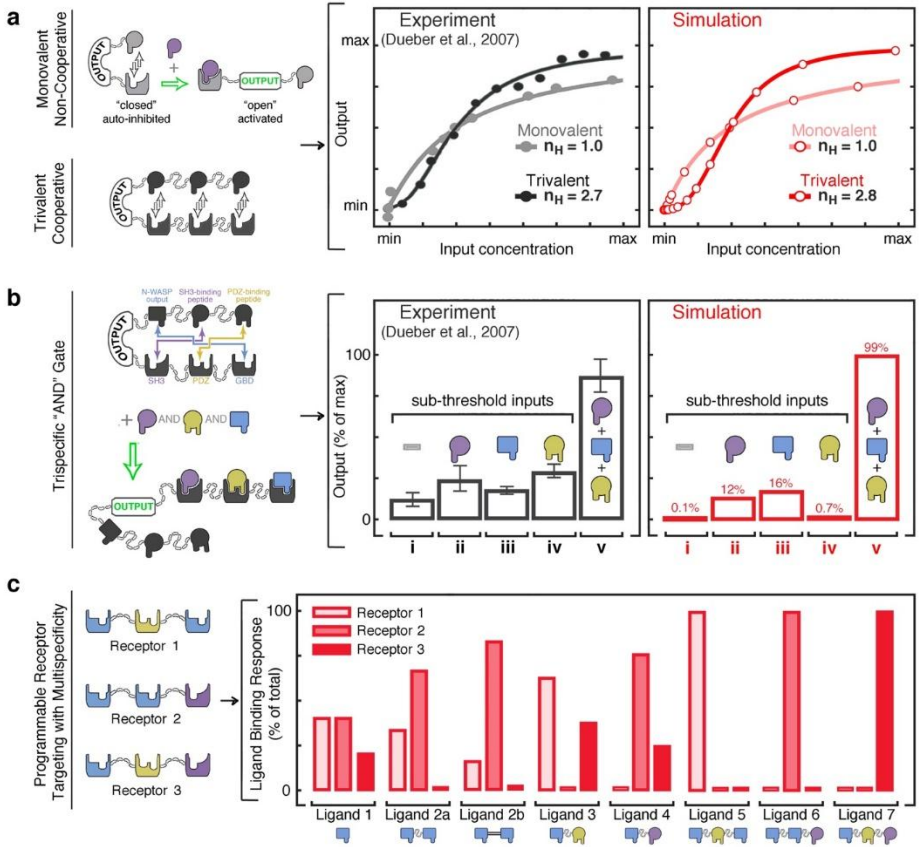
- AbLec (under development)



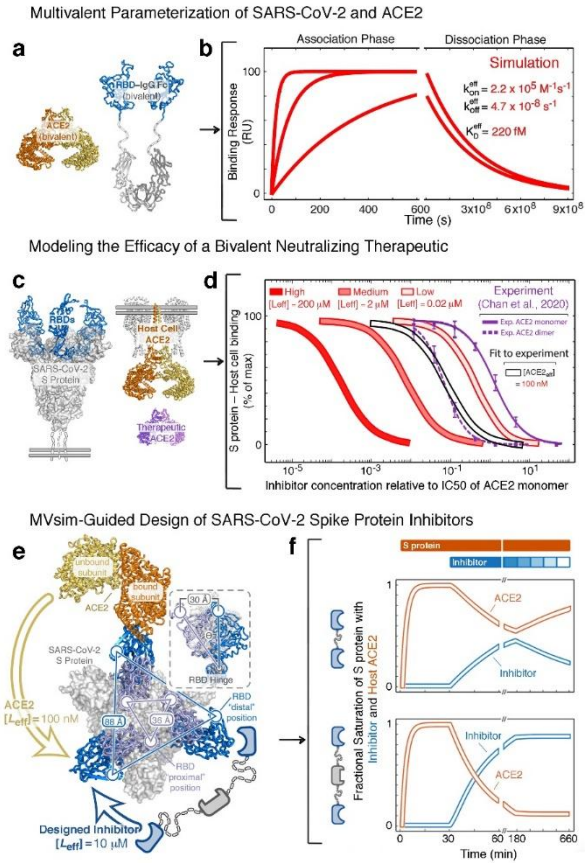


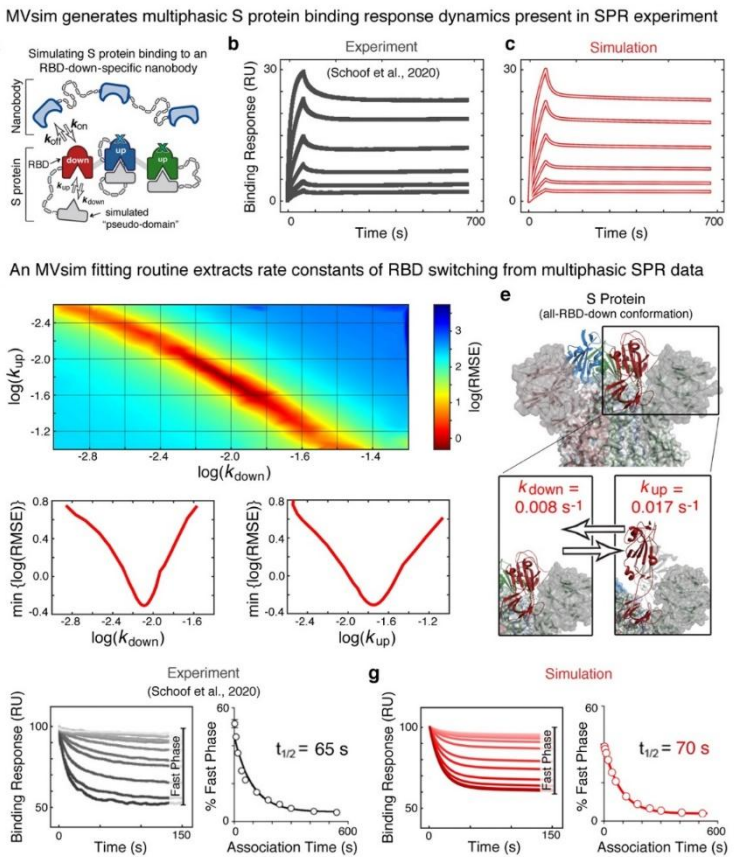
APPLICATION EXAMPLE – MOLECULAR SWITCHES

- Molecular switch
- Ultrasensitive reaction
- Multispecificity
- Importance of genetic switches



SARS-COV-2 S PROTEIN RBD AND ACE2 INTERACTION





THANK YOU FOR YOUR ATTENTION!

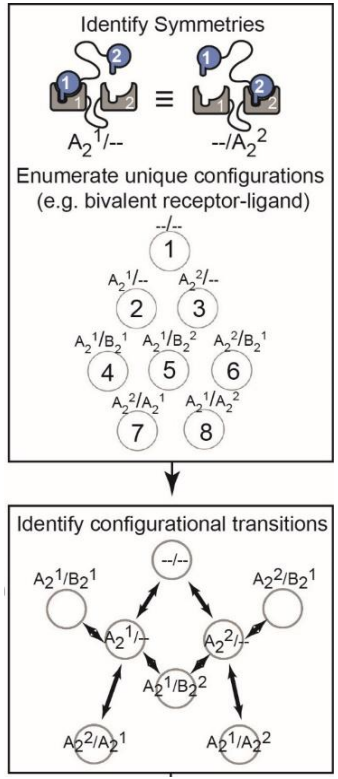
ODES – LETS MAKE SOMETHING SIMPLE COMPLICATED

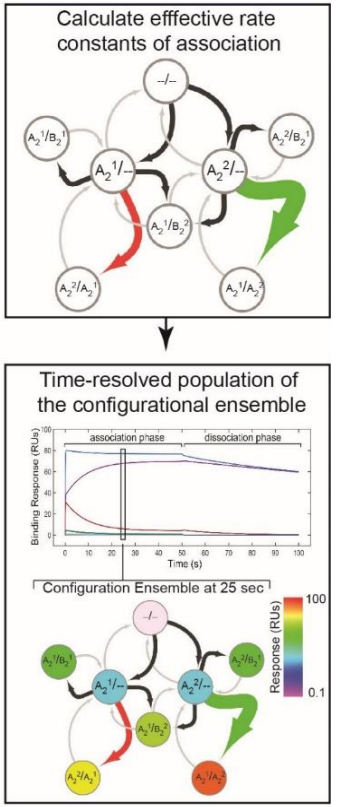
Distance s				

$$\int \sqrt{(-g\,x + c1)^2 + c2}\; dx = \frac{(g\,x - c1)\sqrt{(c1 - g\,x)^2 + c2} - c2\log\left(\sqrt{(c1 - g\,x)^2 + c2} + c1 - g\,x\right)}{2\,g} + \text{constant}$$

FRAMEWORK

1. Identifying unique configurations
   - Reduction by symmetry or similarity
2. Identifying transitions
3. Describing the transitions
   - Approximating the speed of transitions
4. Calculating time-dependent concentrations





Experiment

Simulation