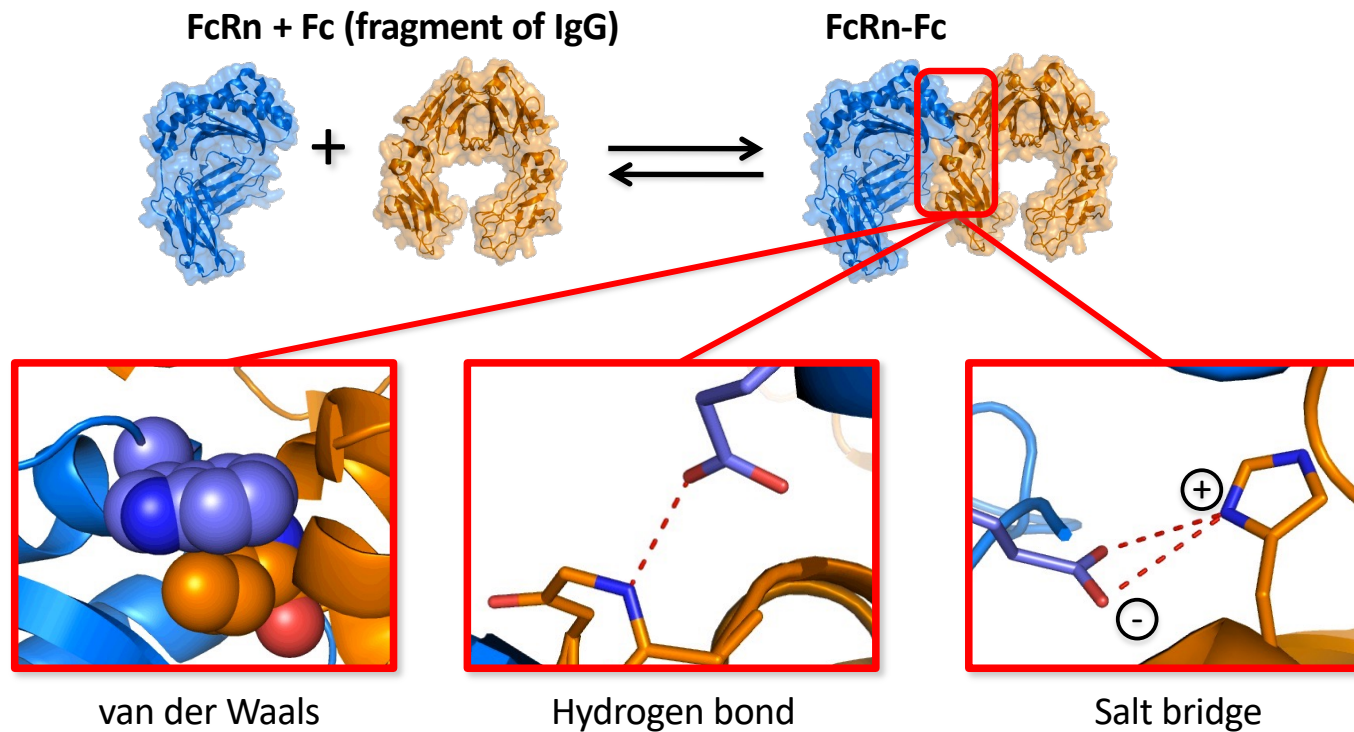


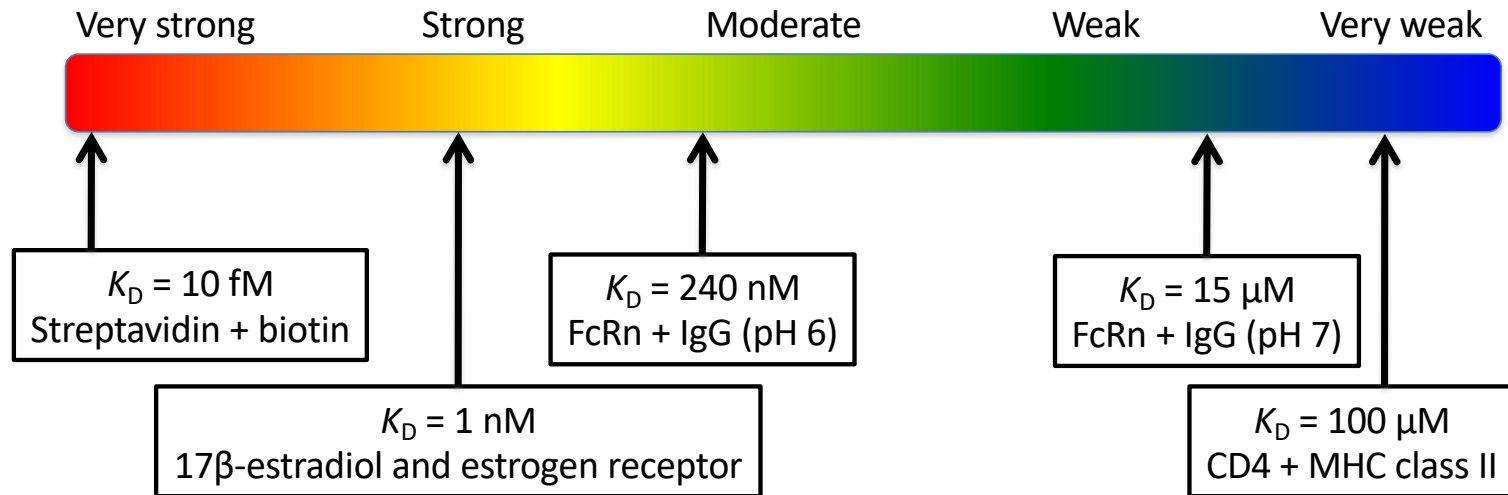
# Non-covalent protein-protein interactions

Many protein interactions involve two species that form a temporary complex.



$$\Delta G = \Delta G_{vdW} + \Delta G_{hbond} + \Delta G_{sb} + \Delta G_{solvP} + \dots = -RT \ln(K_{eq})$$

## Examples of some $K_D$ values



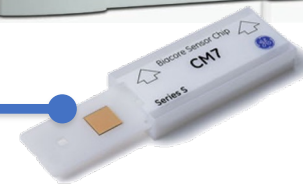
In adult mammals, the half-life of IgG in the blood is very long (about 3 weeks). This neonatal Fc receptor (FcRn) is expressed on epithelial cells and salvages endocytosed IgG from degradation.

# Biacore T200

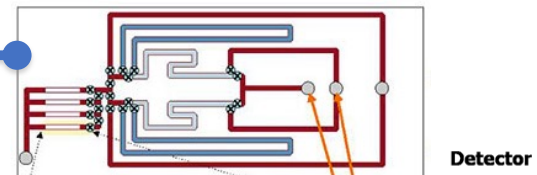
**Reagent compartment** samples and injection needle



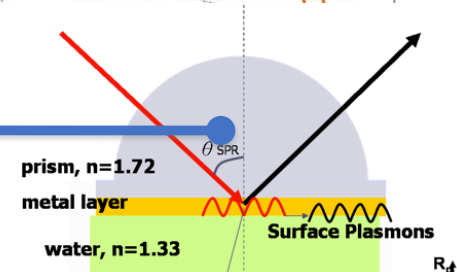
**Sensor chip** four “reaction vessels” in series



**IFC** integrated microfluidics cartridge

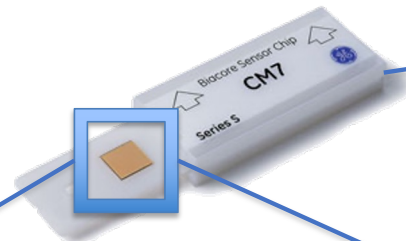


**Optics** measures change in refractive index near the sensor chip surface

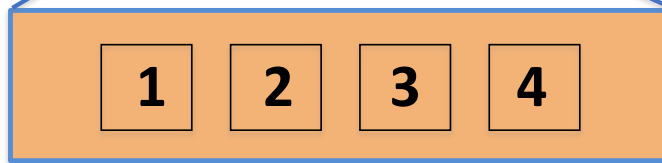


# Biacore T200

Sensor chip



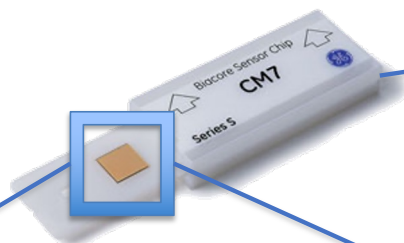
Top view



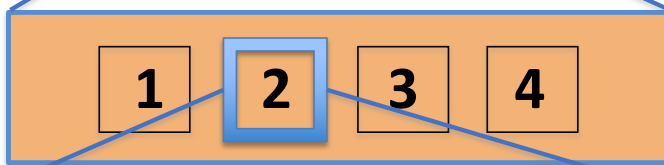
Gold-dextran surface with four **flow cells**  
(reaction vessels)

# Biacore T200

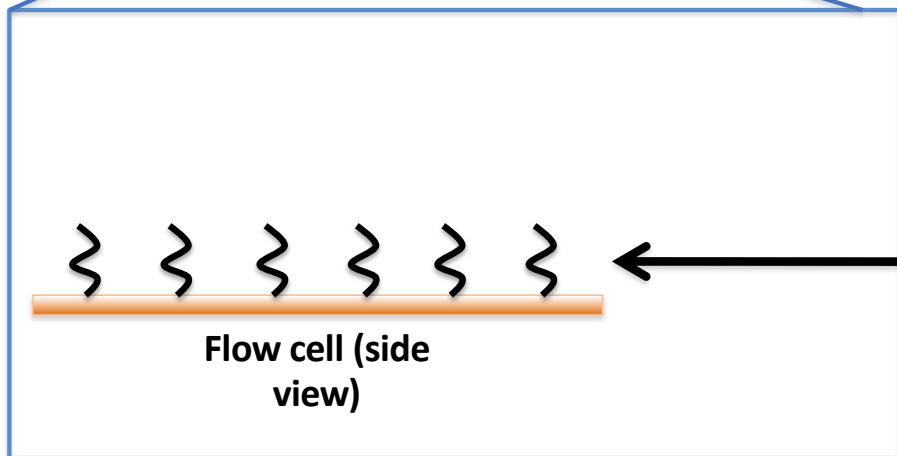
Sensor chip



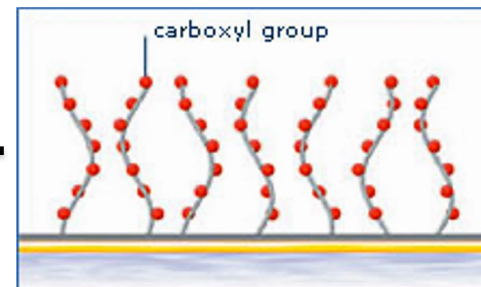
Top view



Gold-dextran surface with four **flow cells** (reaction vessels)

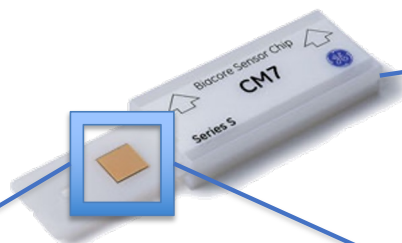


100 nm carboxydextran

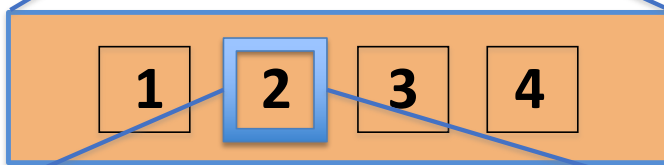


# Biacore T200

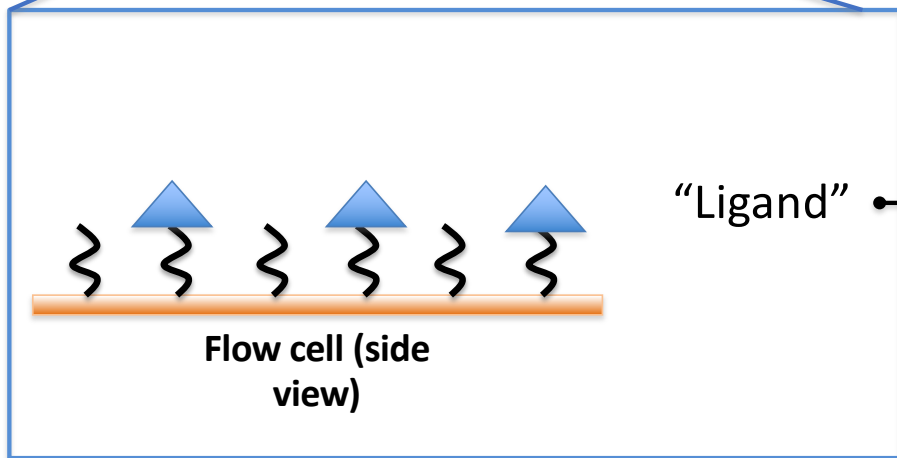
Sensor chip



Top view



Gold-dextran surface with four **flow cells** (reaction vessels)

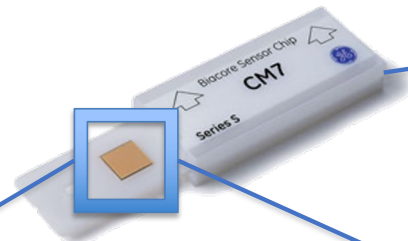


"Ligand"

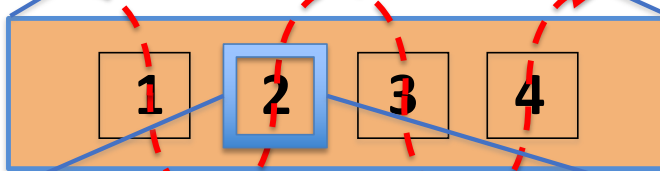


# Biacore T200

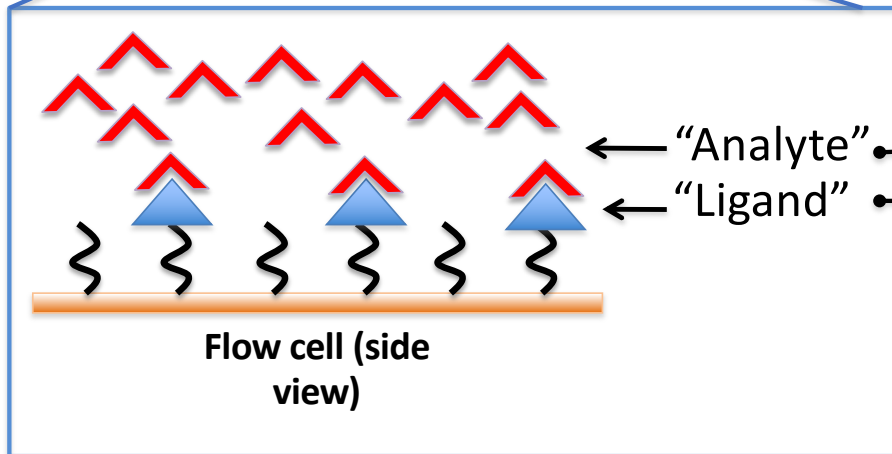
Sensor chip



Top view



Gold-dextran surface with four **flow cells** (reaction vessels)

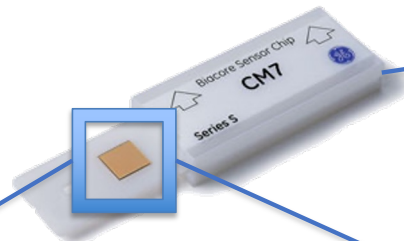


← "Analyte"

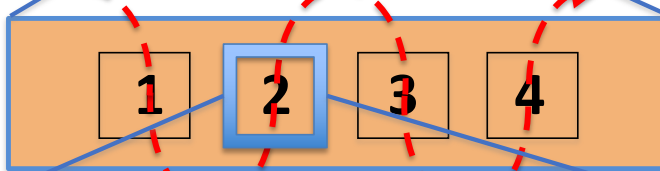
← "Ligand"

# Biacore T200

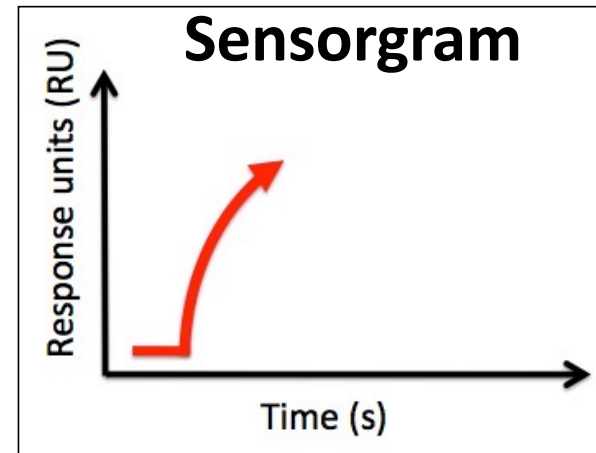
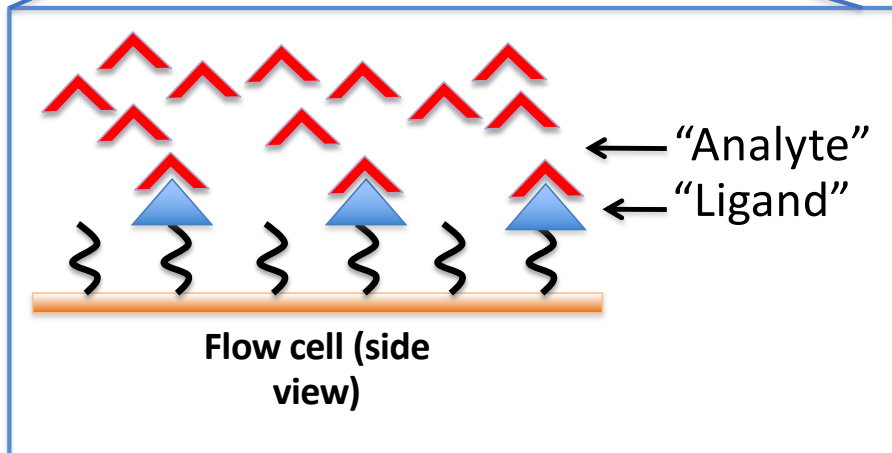
Sensor chip



Top view

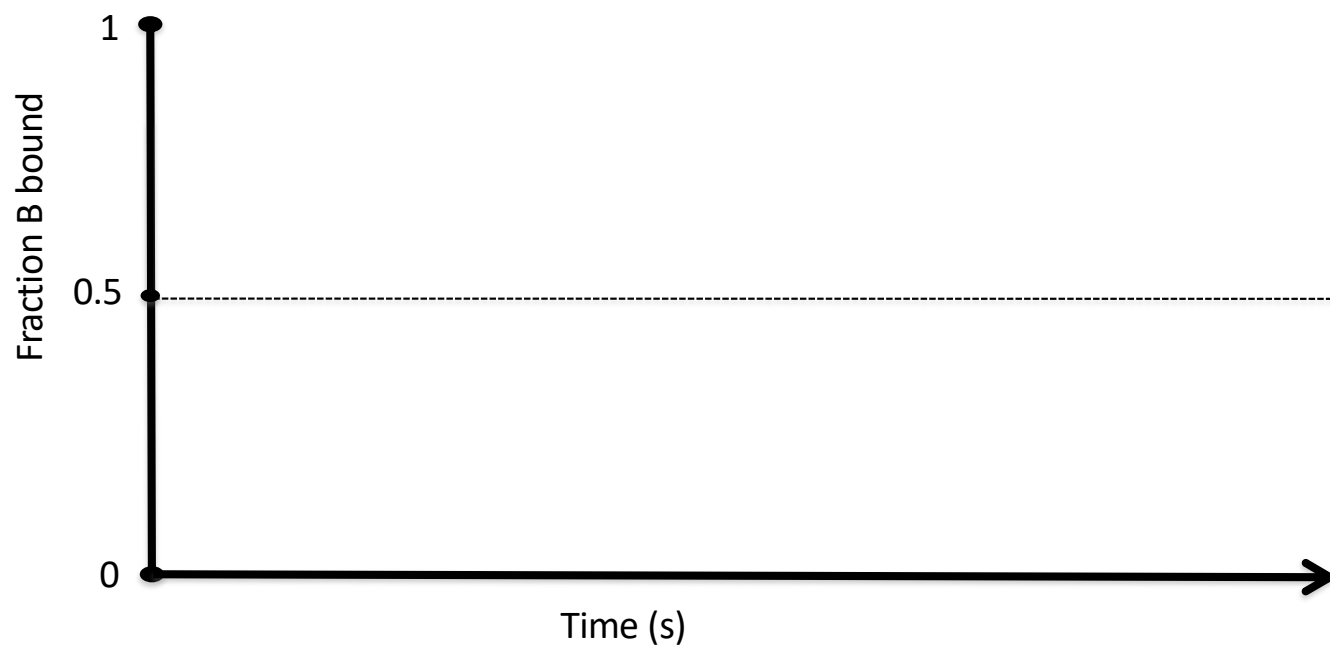
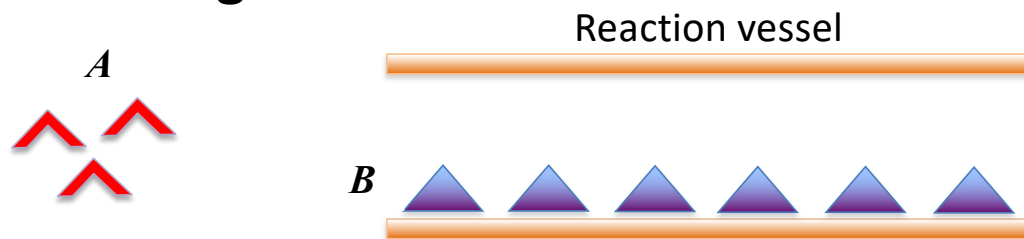


Gold-dextran surface with four **flow cells** (reaction vessels)

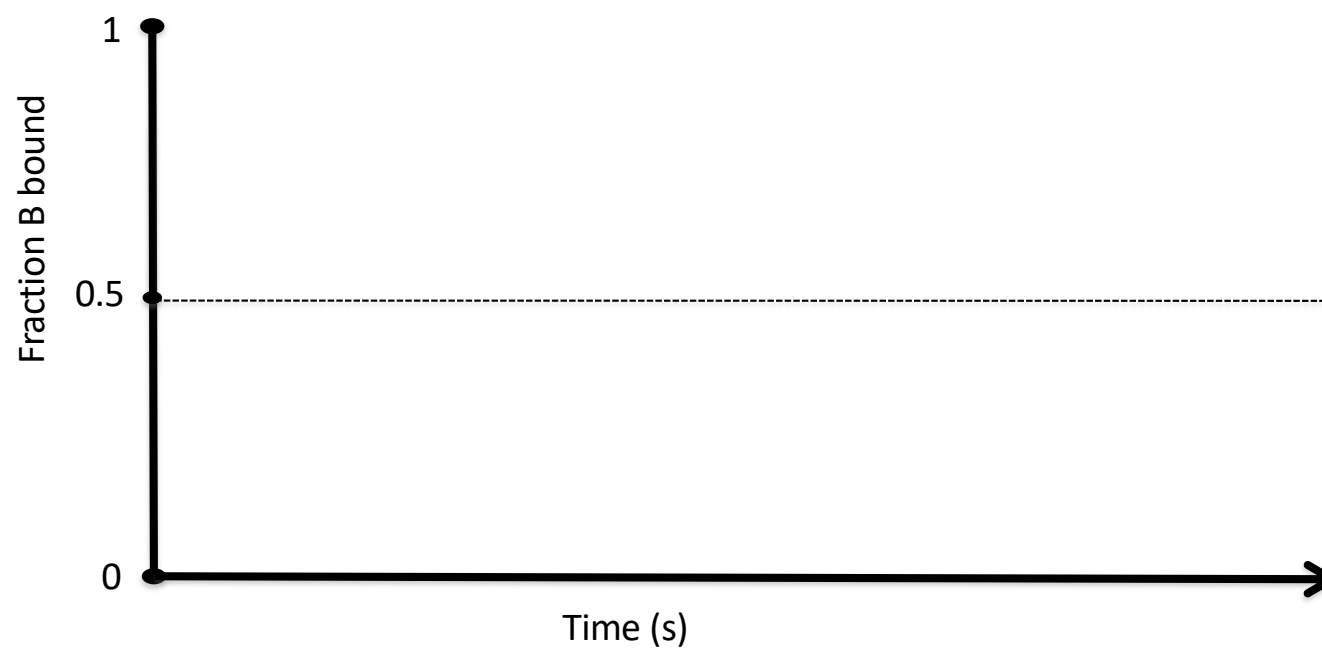
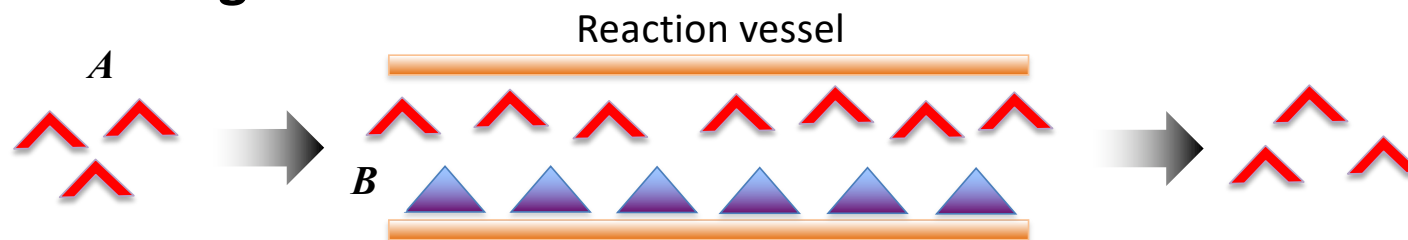




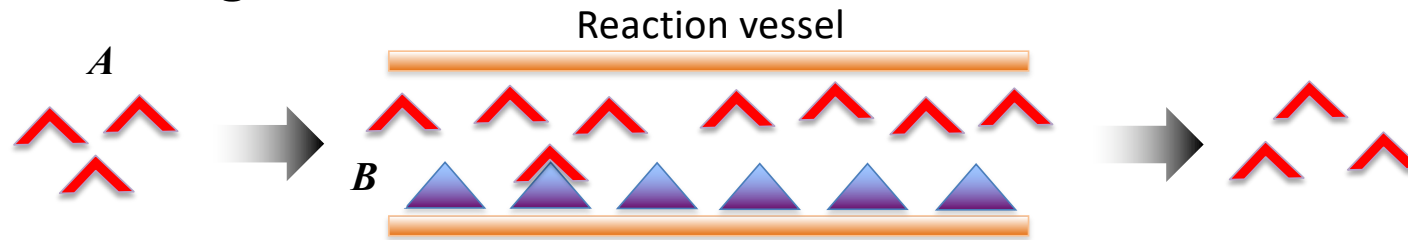
# Watching reactions in real-time



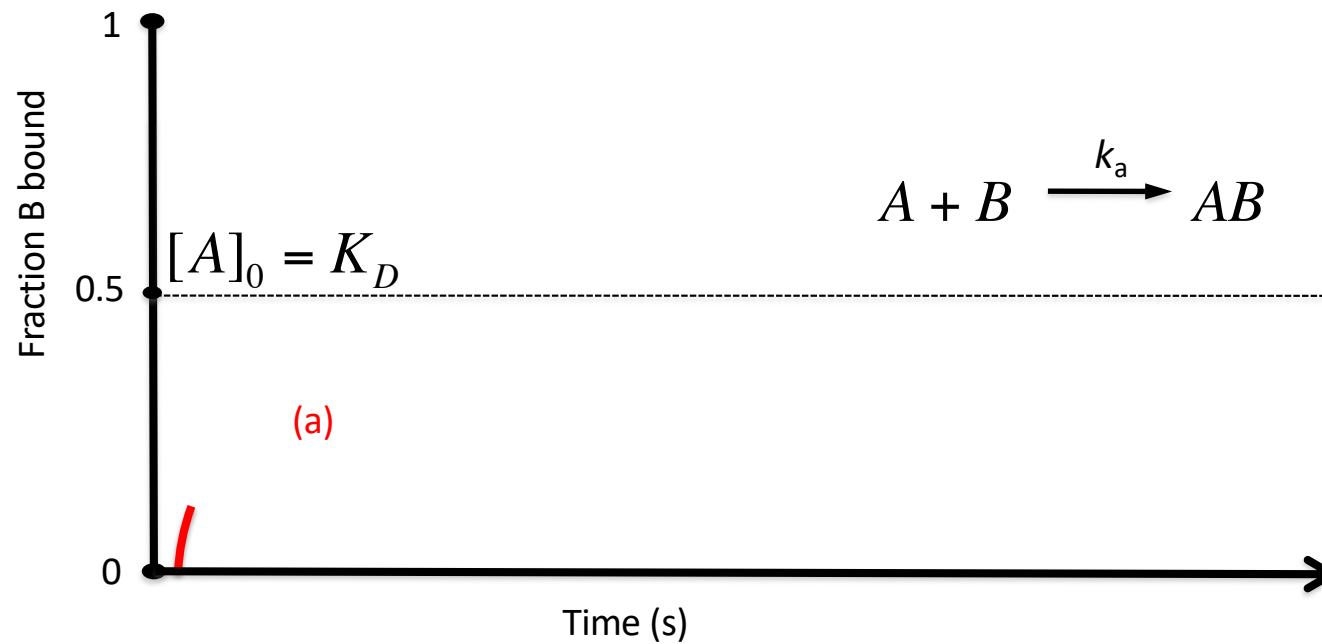
## Watching reactions in real-time



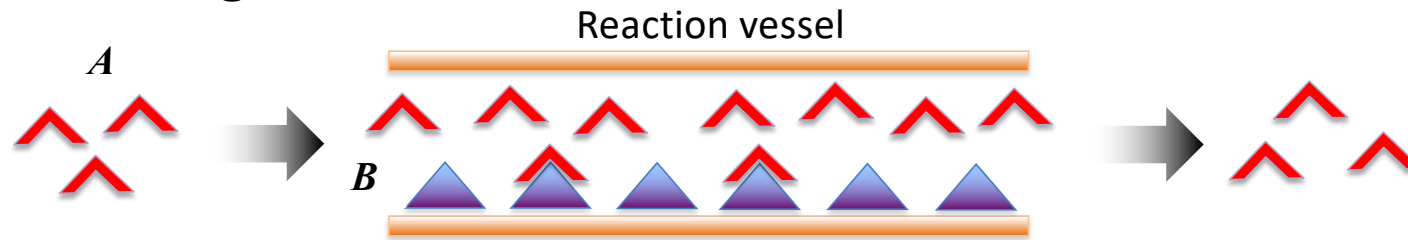
## Watching reactions in real-time



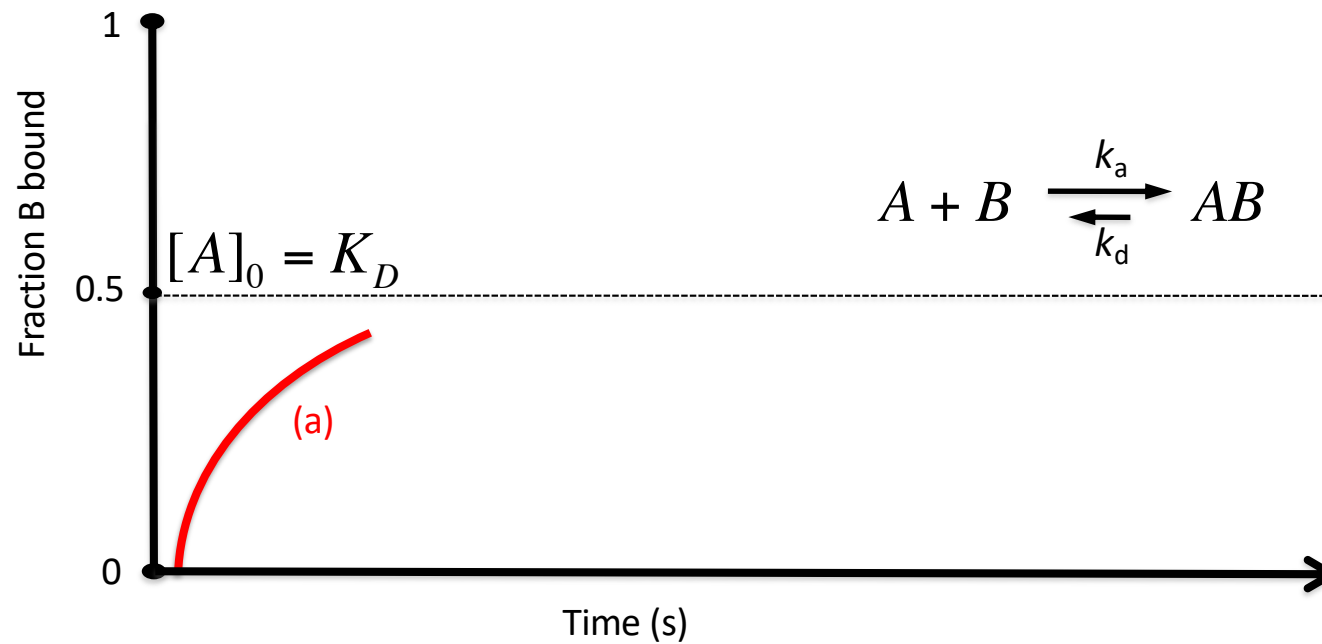
(a) Association phase:  $A$  is injected and allowed to bind to  $B$ .



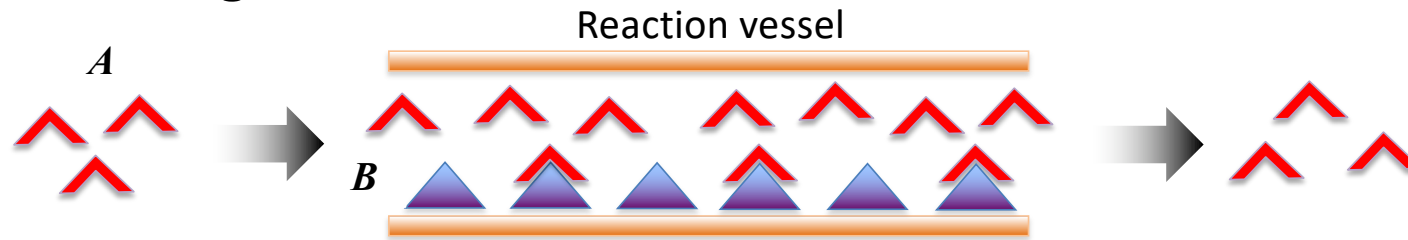
## Watching reactions in real-time



(a) Association phase:  $A$  is injected and allowed to bind to  $B$ .

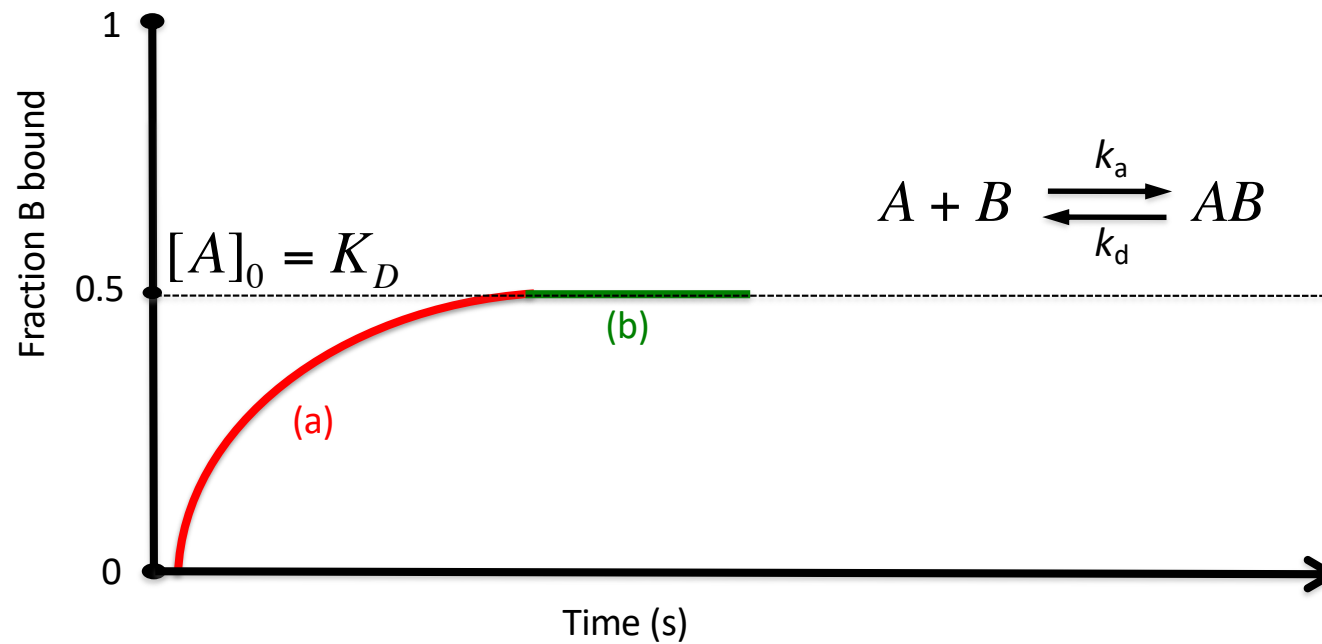


## Watching reactions in real-time

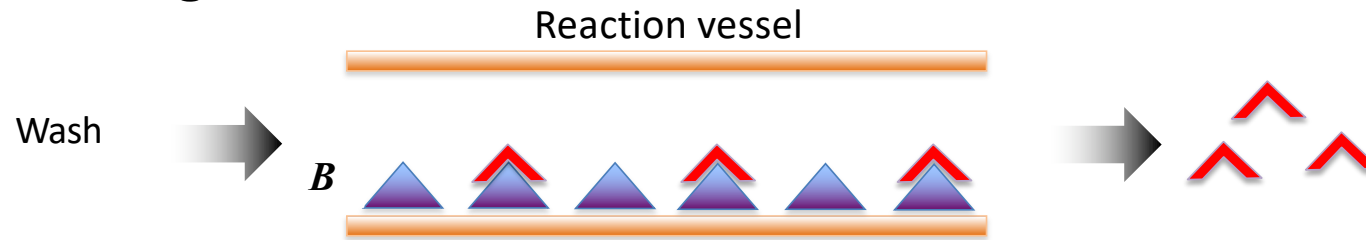


(a) Association phase:  $A$  is injected and allowed to bind to  $B$ .

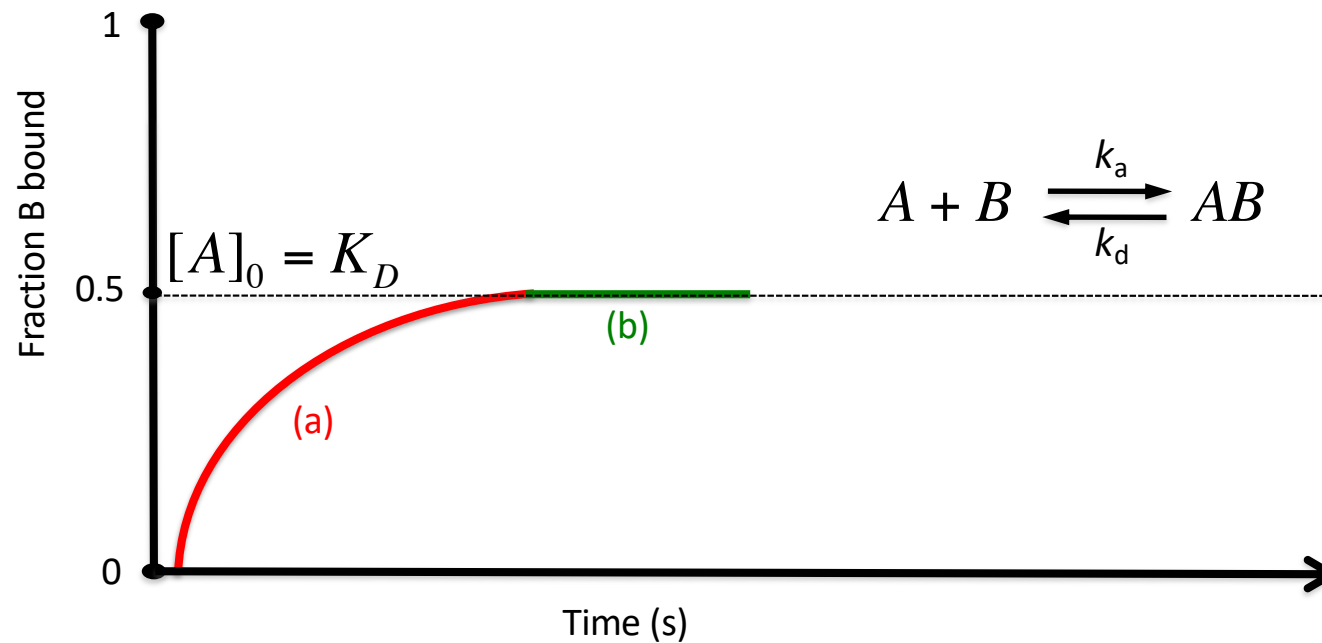
(b) Equilibrium phase:  $A$  is still injected, but  $dAB/dt = 0$ .



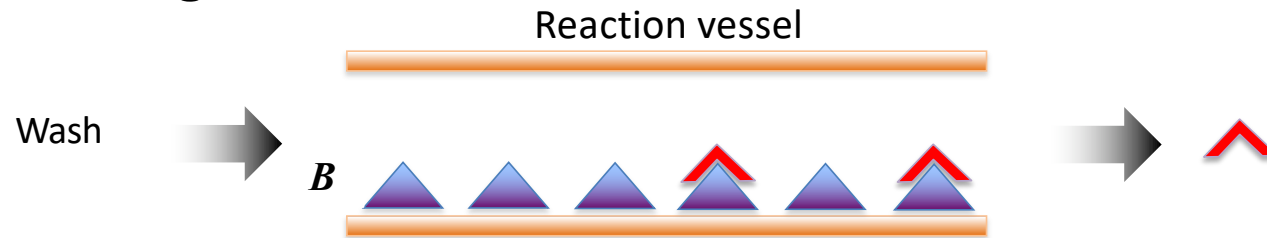
## Watching reactions in real-time



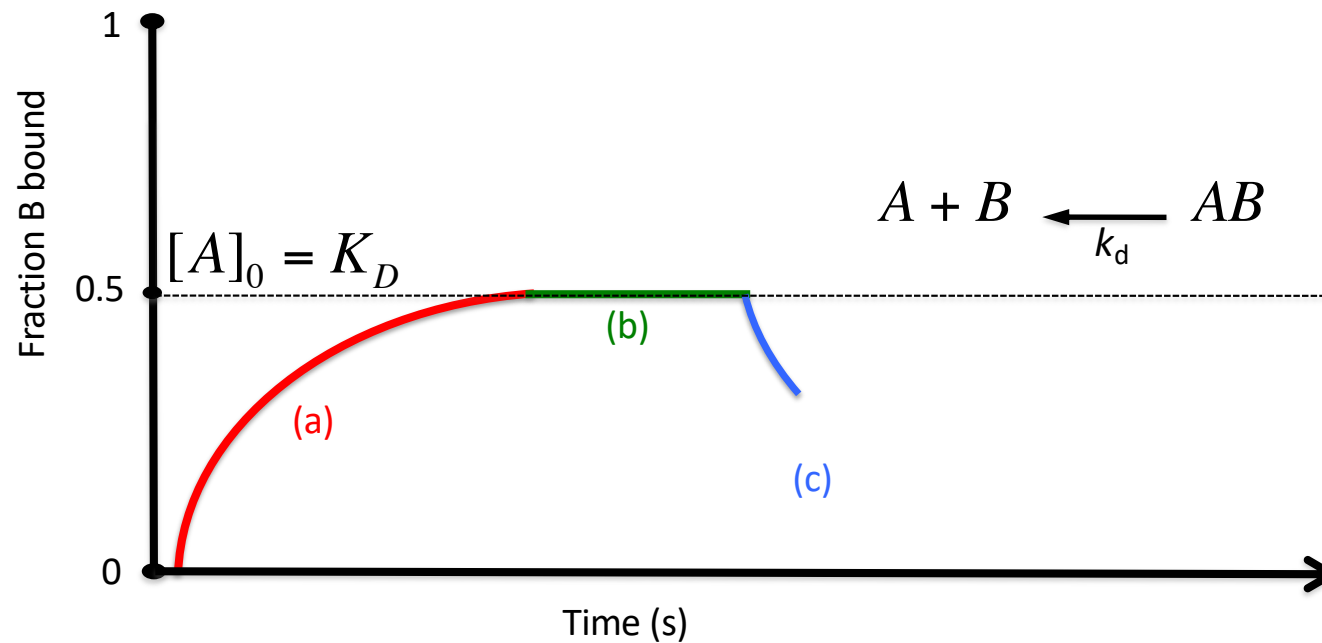
- (a) Association phase:  $A$  is injected and allowed to bind to  $B$ .
- (b) Equilibrium phase:  $A$  is still injected, but  $dAB/dt = 0$ .
- (c) Dissociation phase:  $A$  is replaced with buffer, leaving only  $AB$  to dissociate.



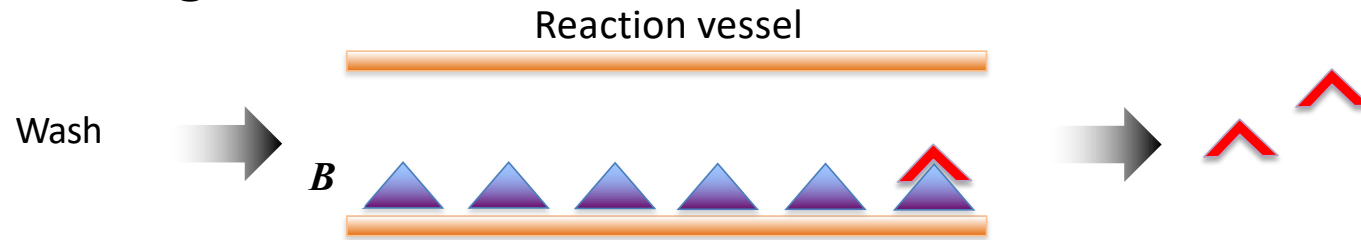
## Watching reactions in real-time



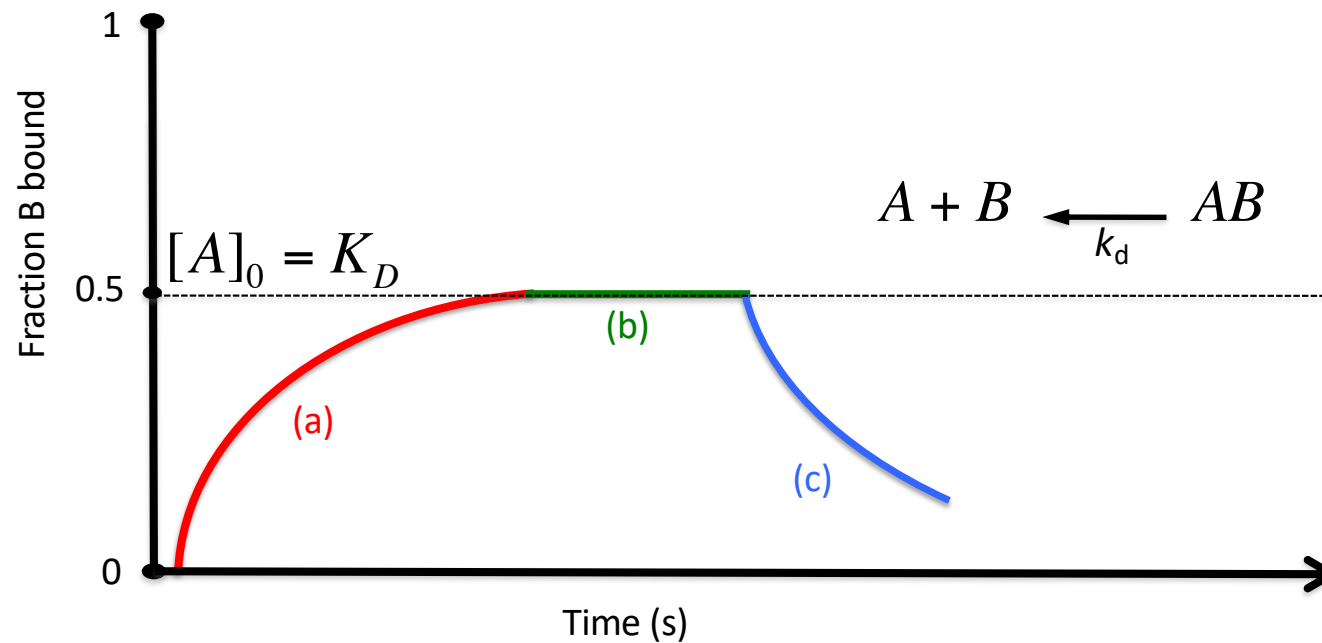
- (a) Association phase:  $A$  is injected and allowed to bind to  $B$ .
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## Watching reactions in real-time

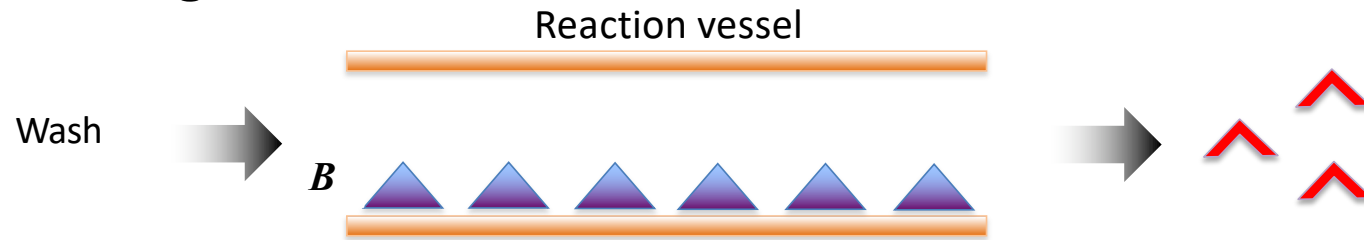


- (a) Association phase:  $A$  is injected and allowed to bind to  $B$ .
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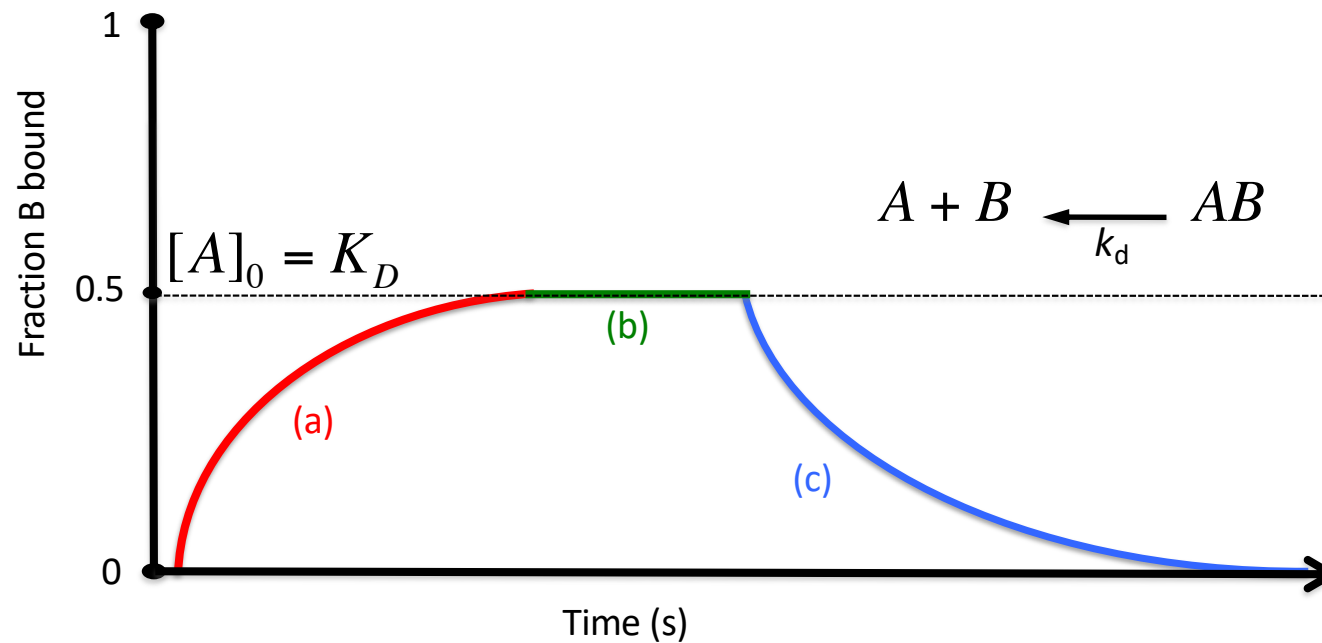




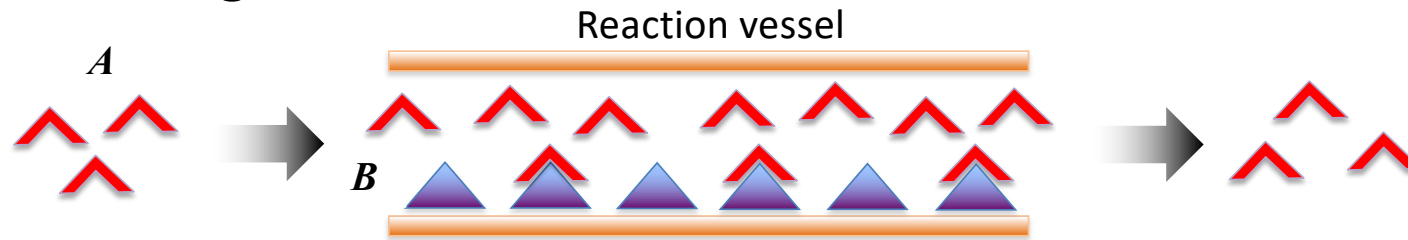
## Watching reactions in real-time



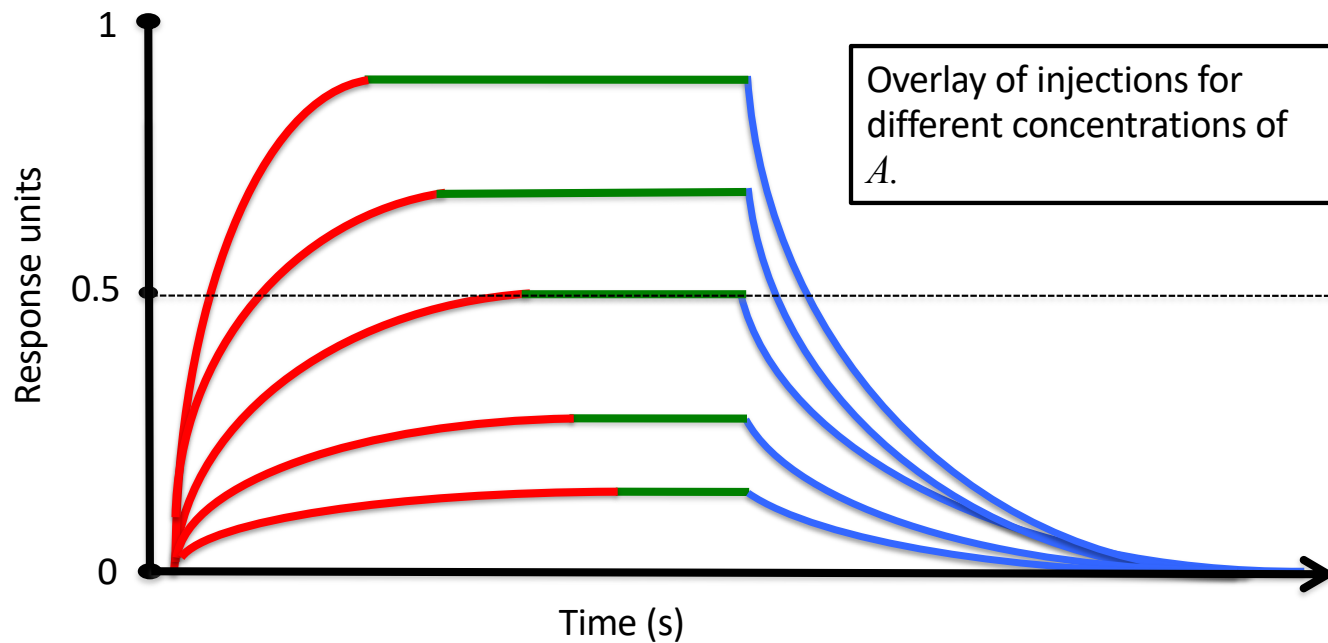
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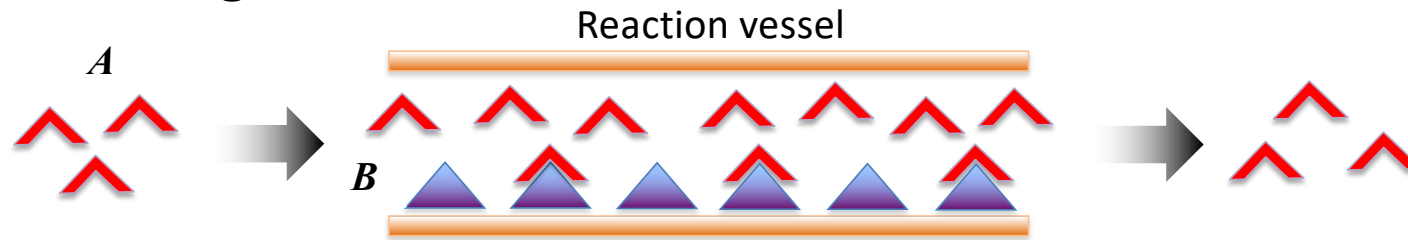
## Watching reactions in real-time



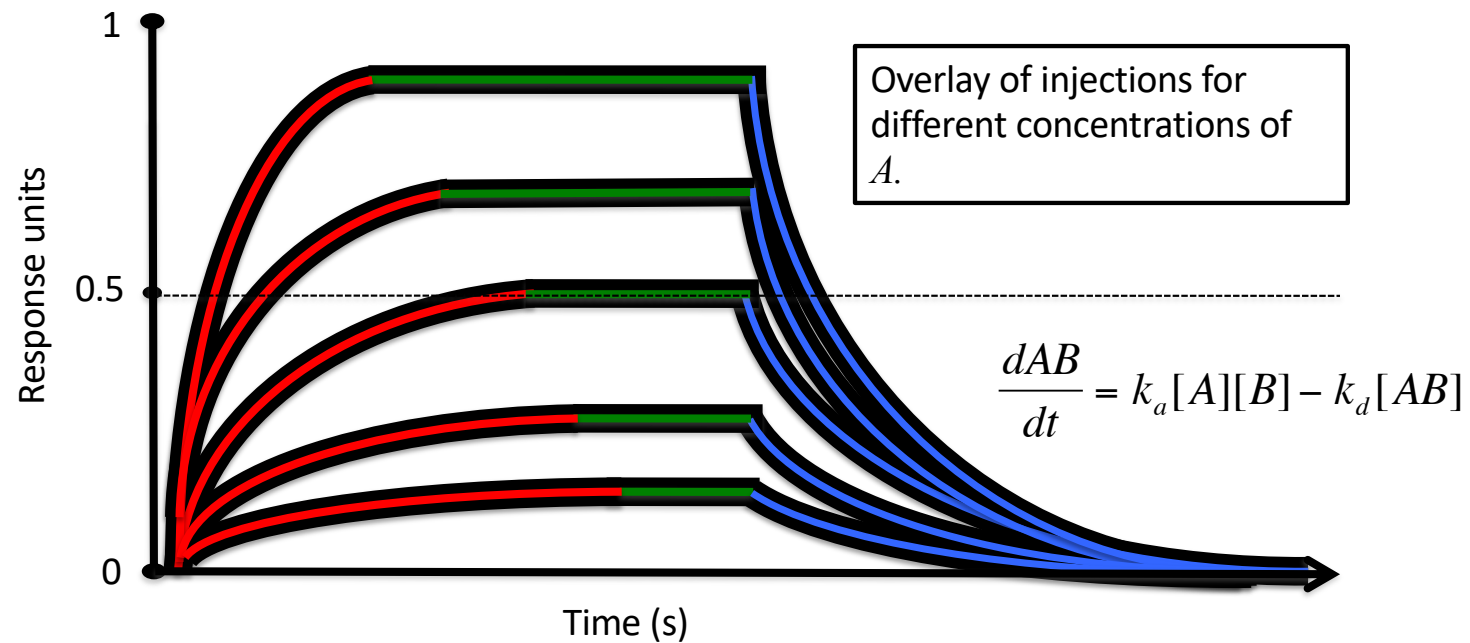
- (a) Association phase: *A* is injected and allowed to bind to *B*.
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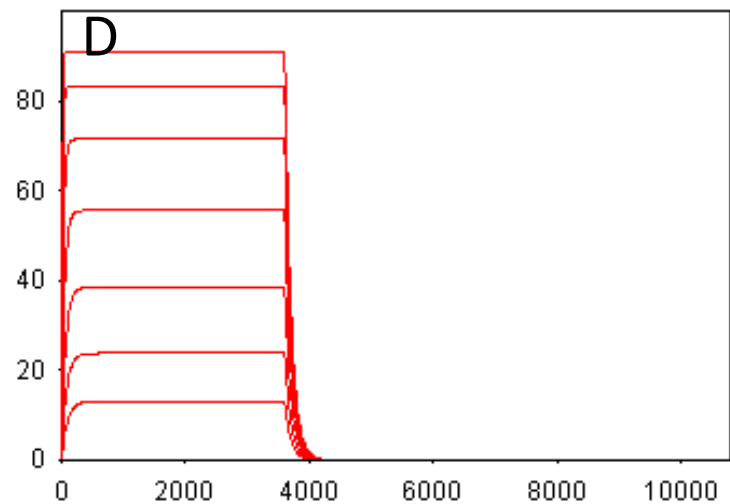
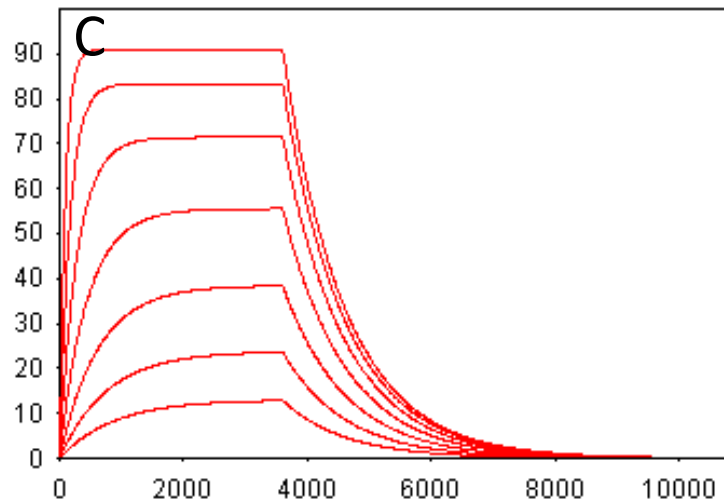
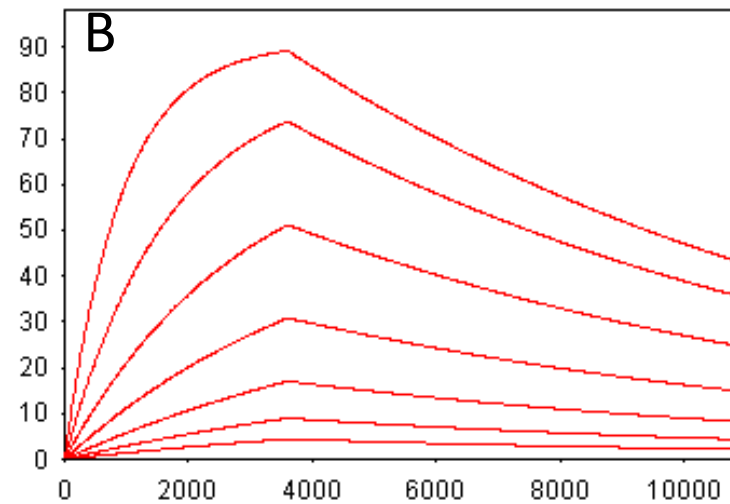
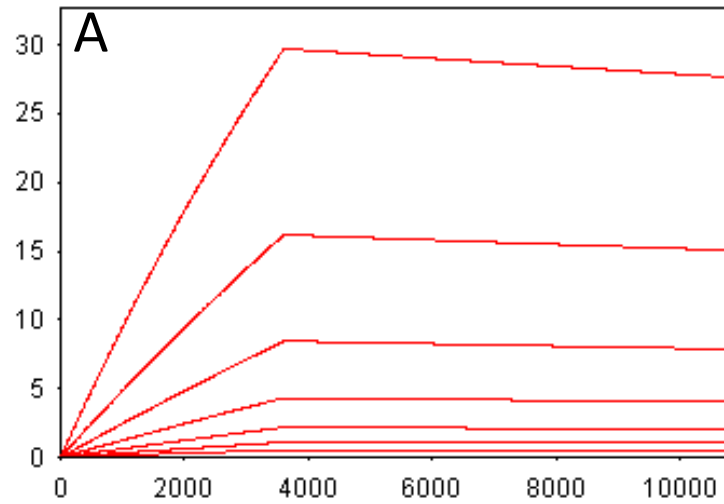
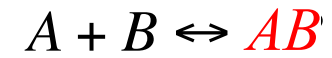
## Watching reactions in real-time



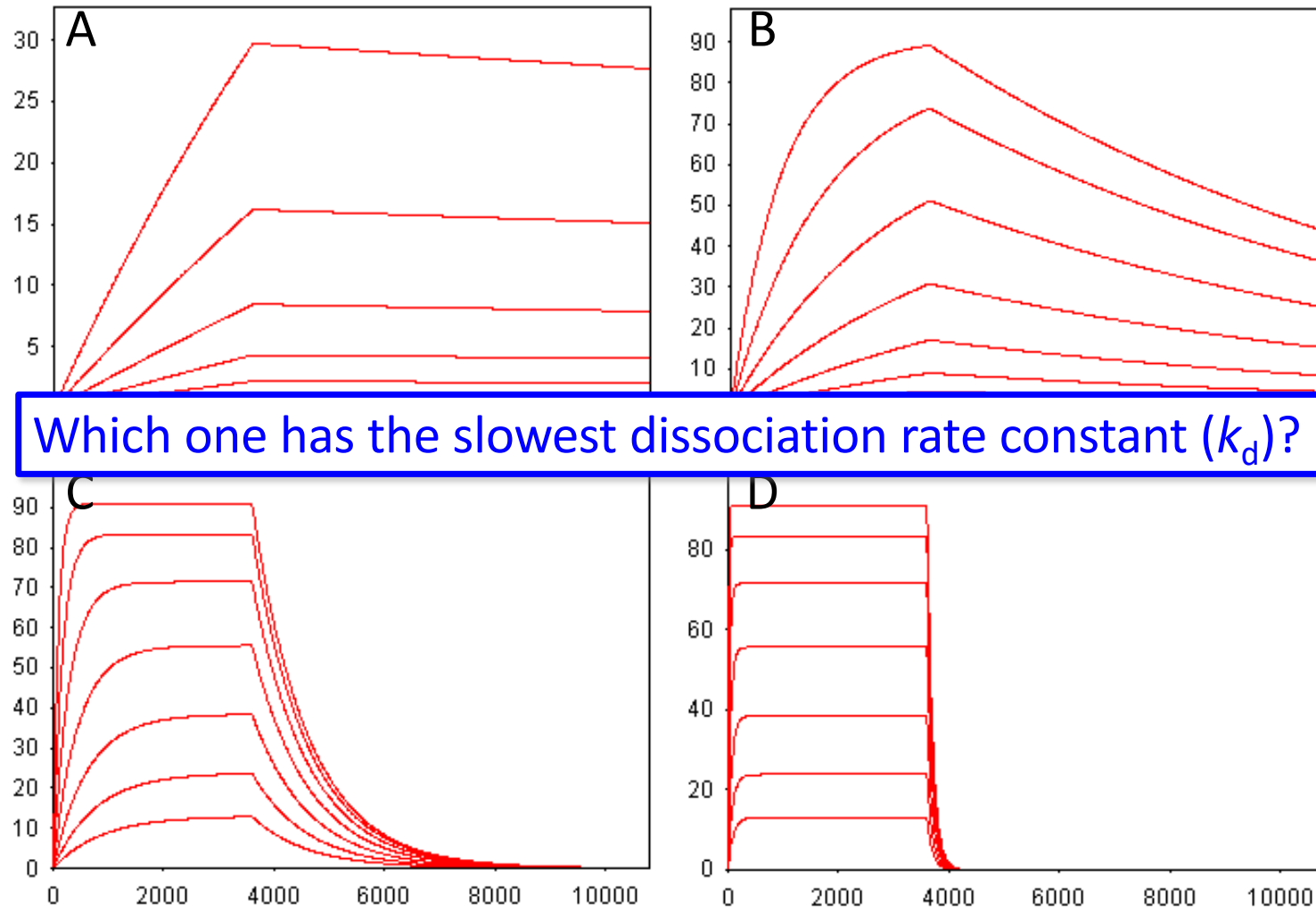
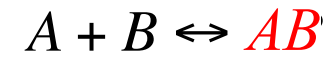
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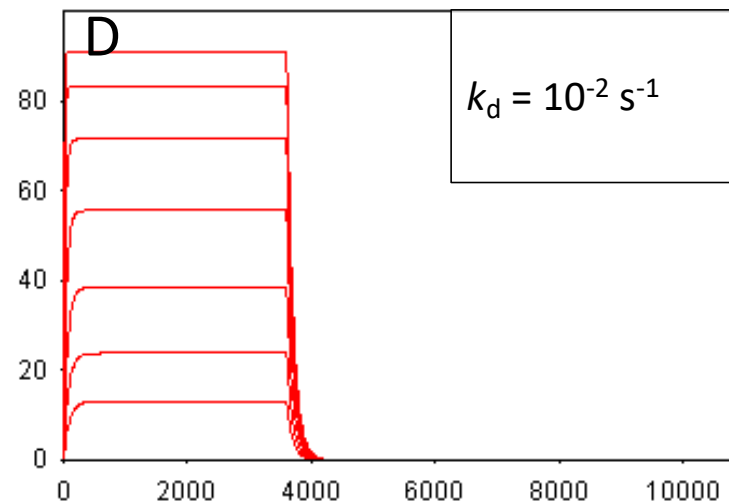
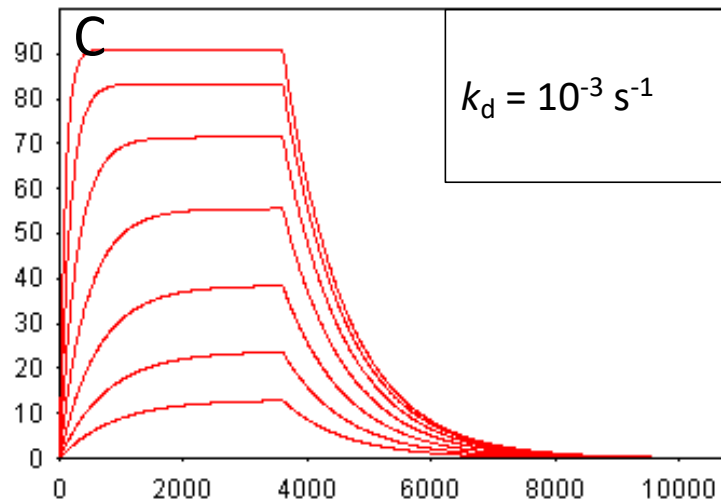
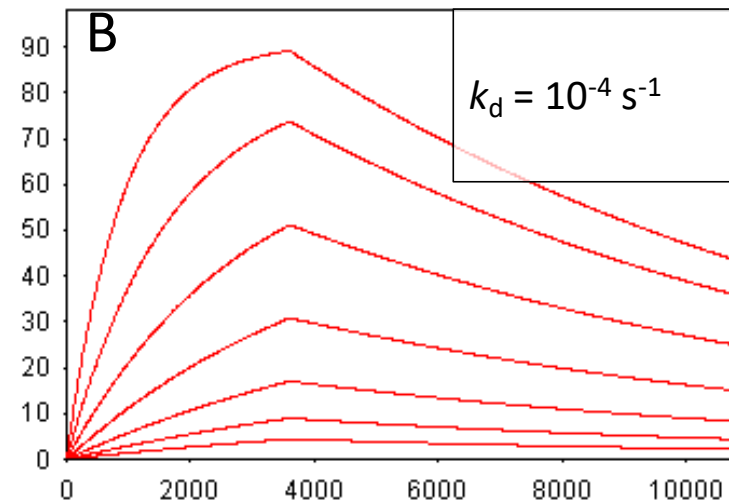
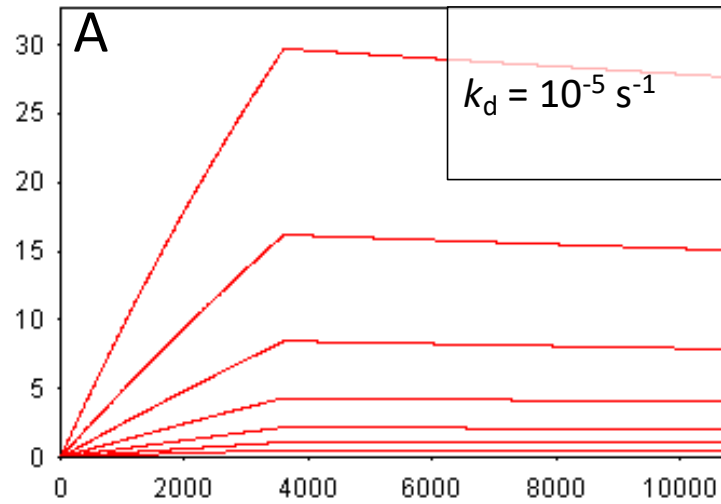
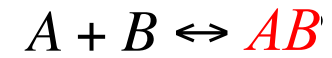
# Comparison of simulations for 1-to-1 binding



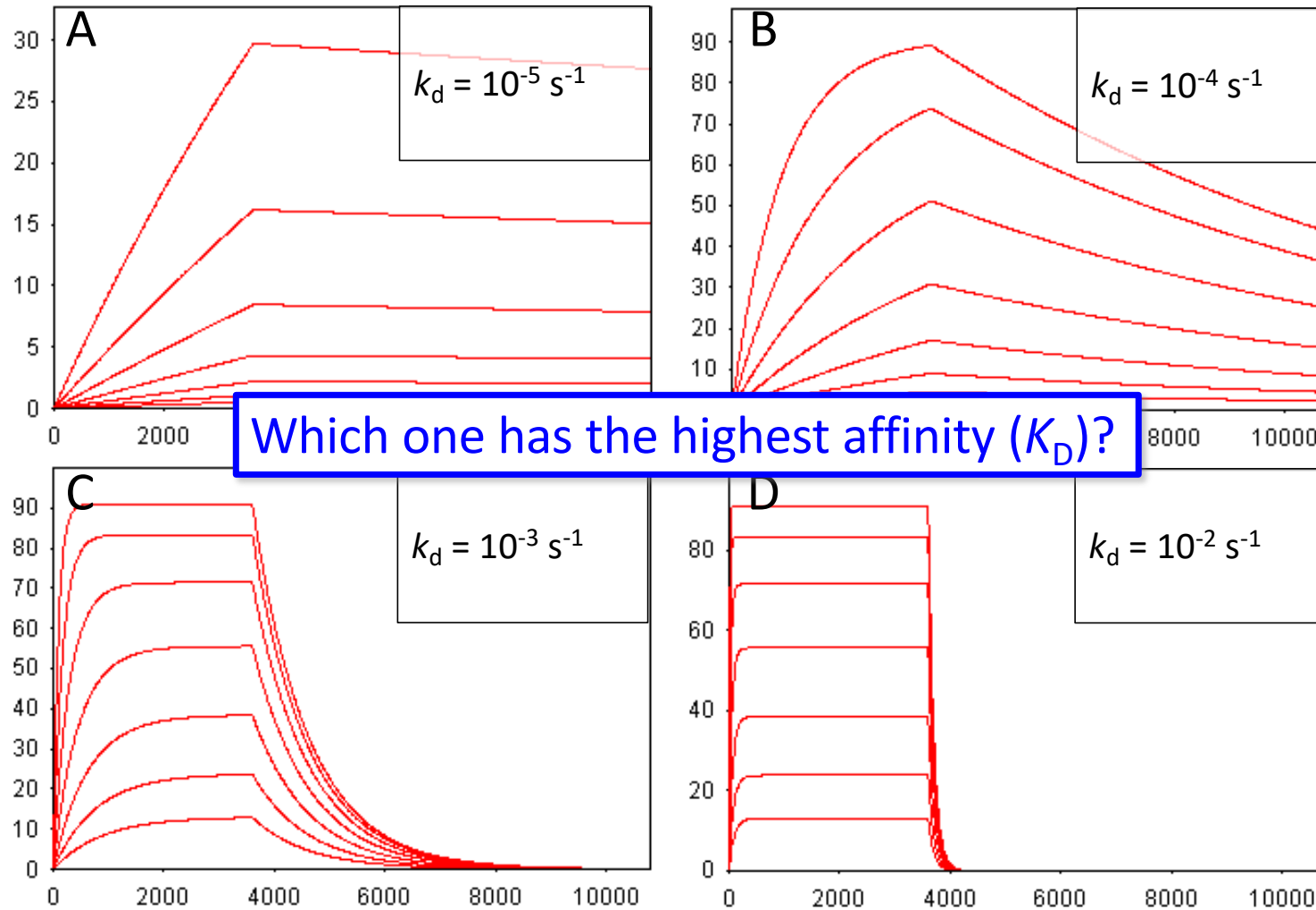
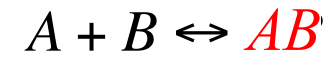
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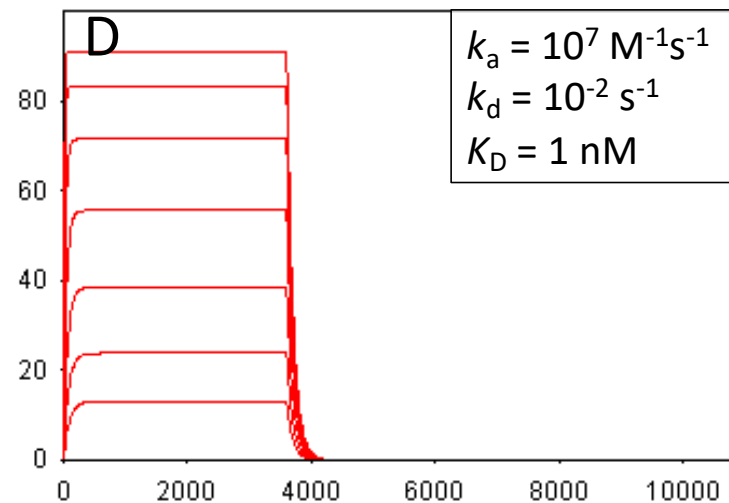
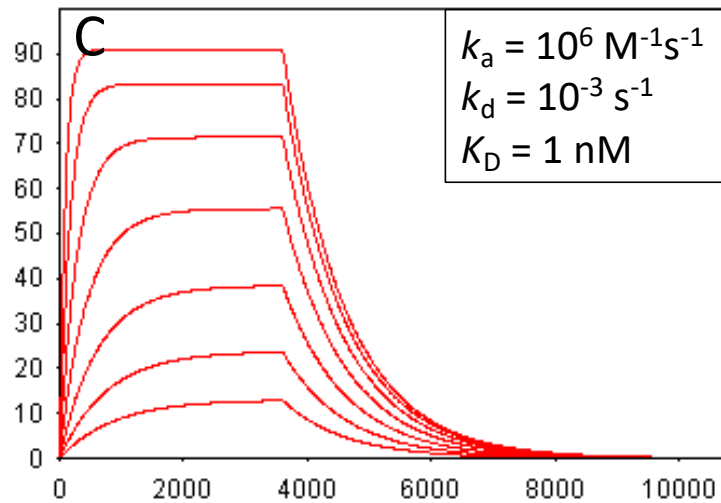
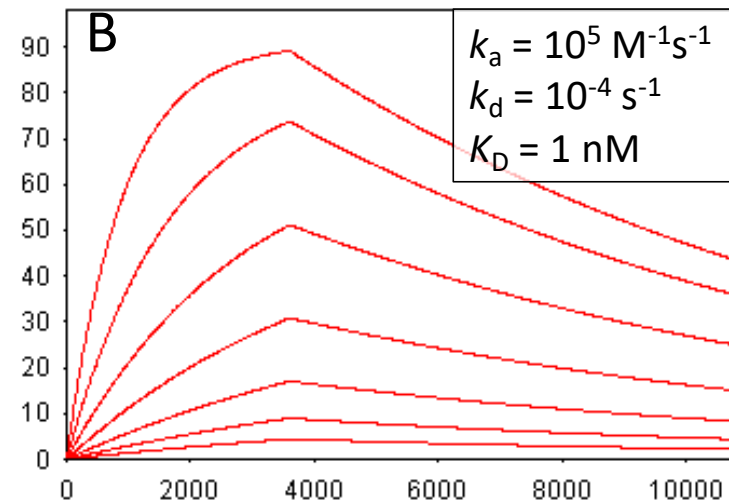
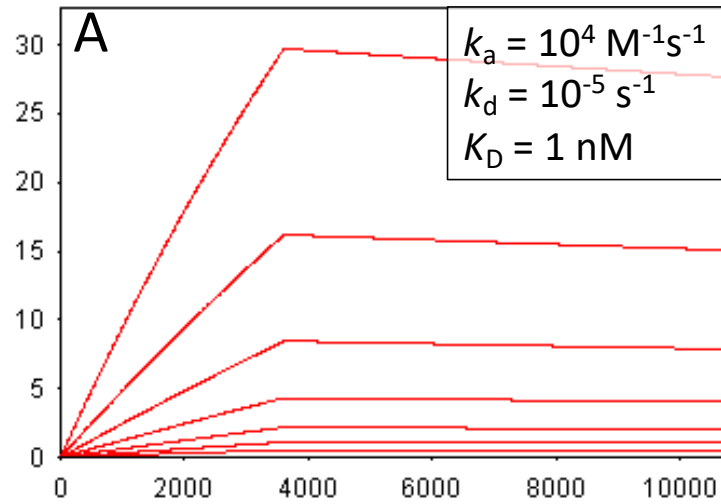
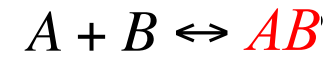
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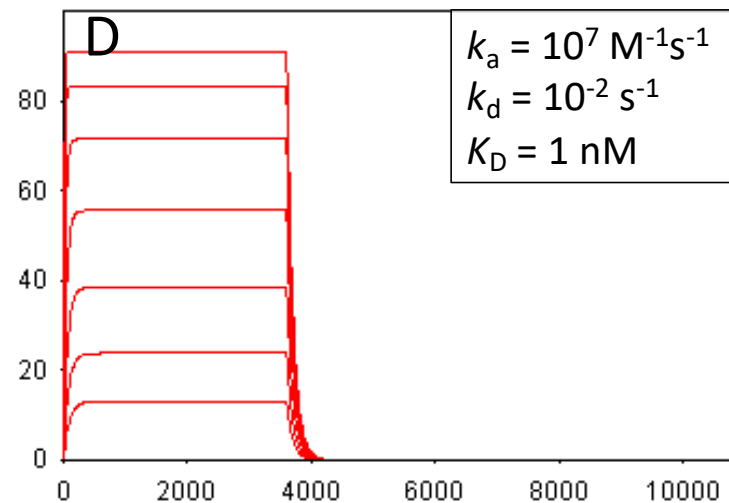
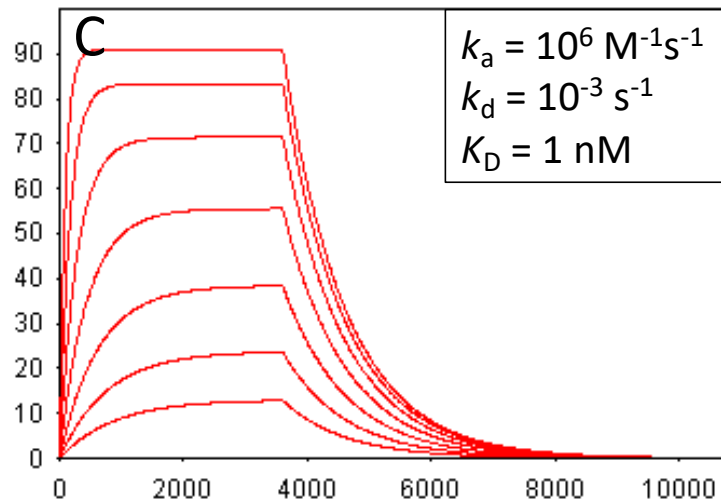
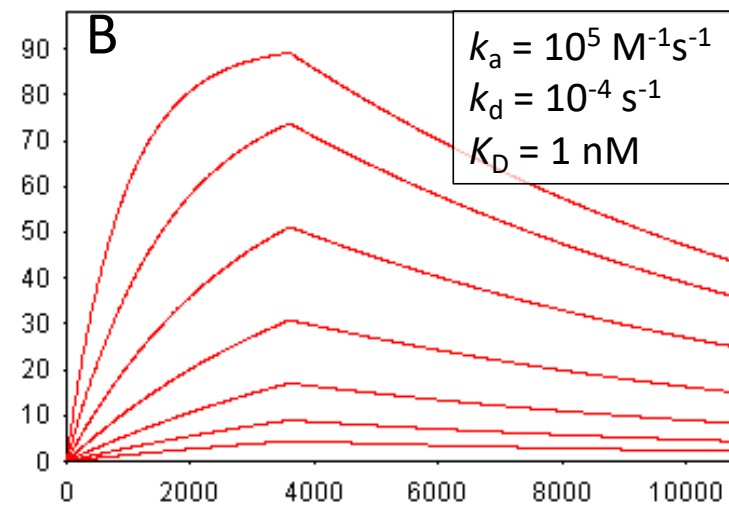
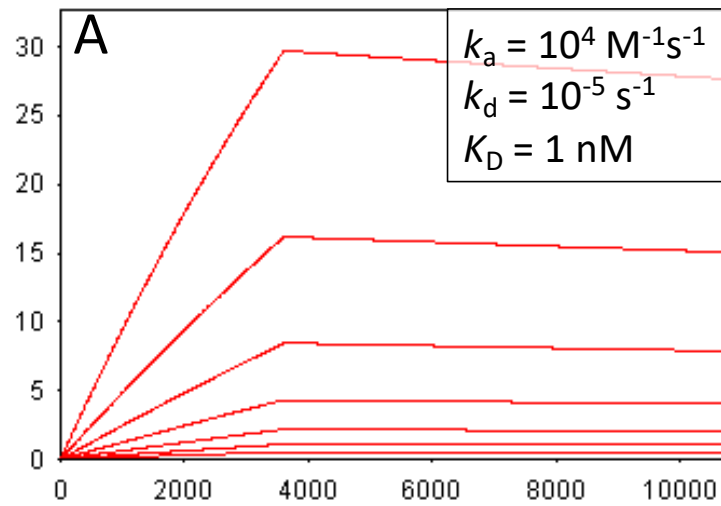
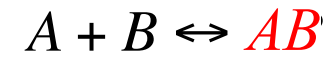


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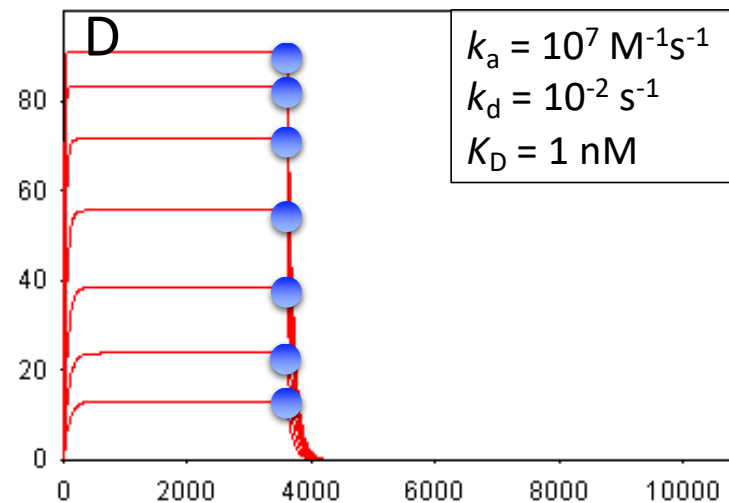
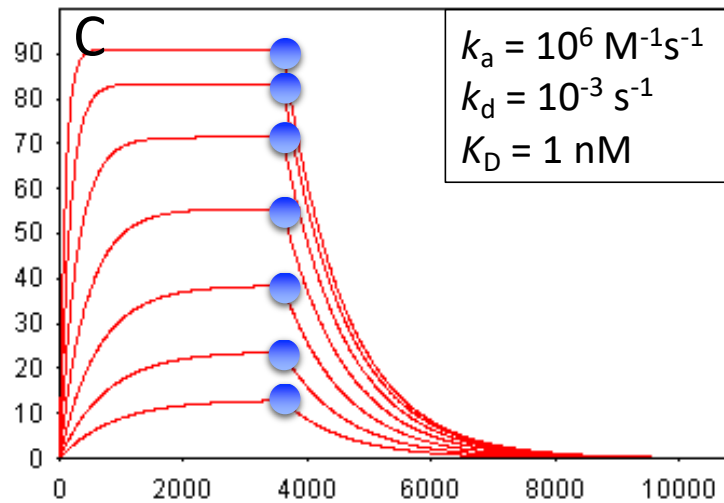
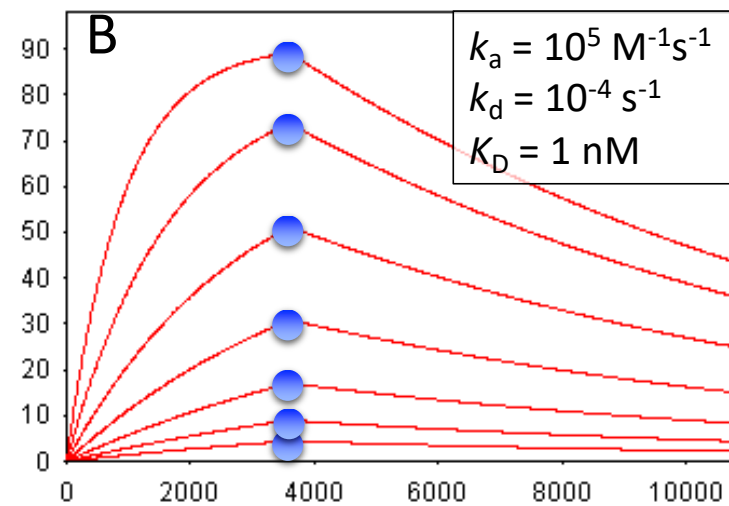
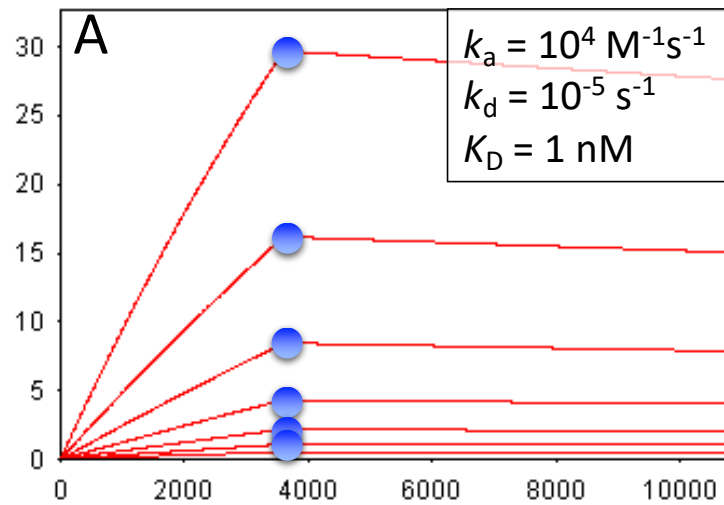




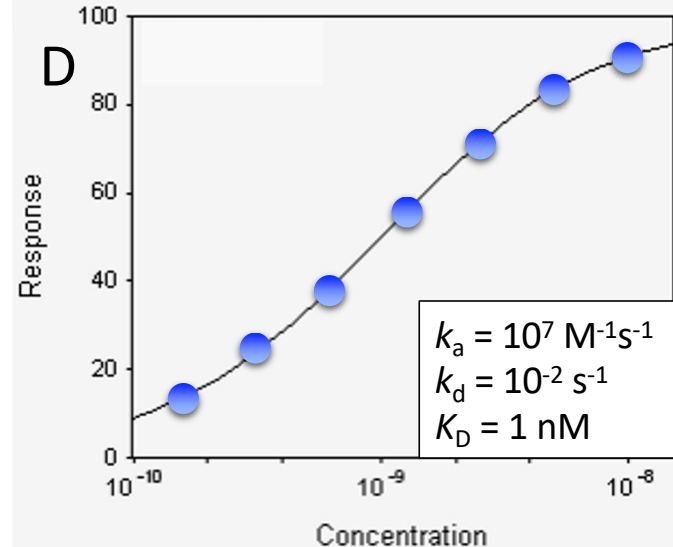
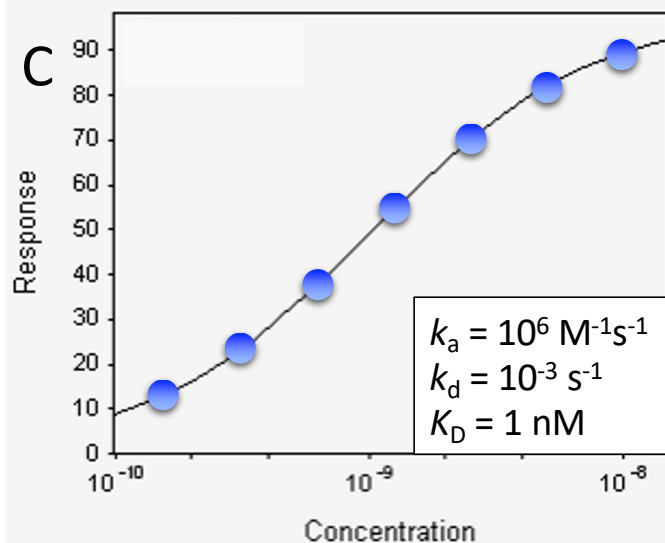
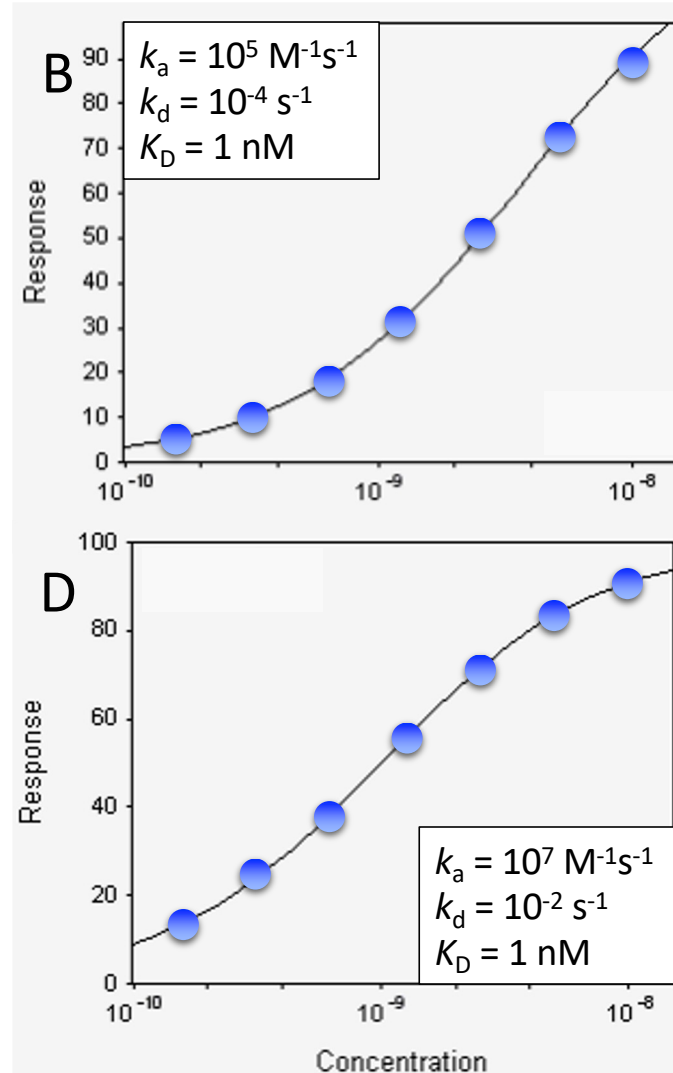
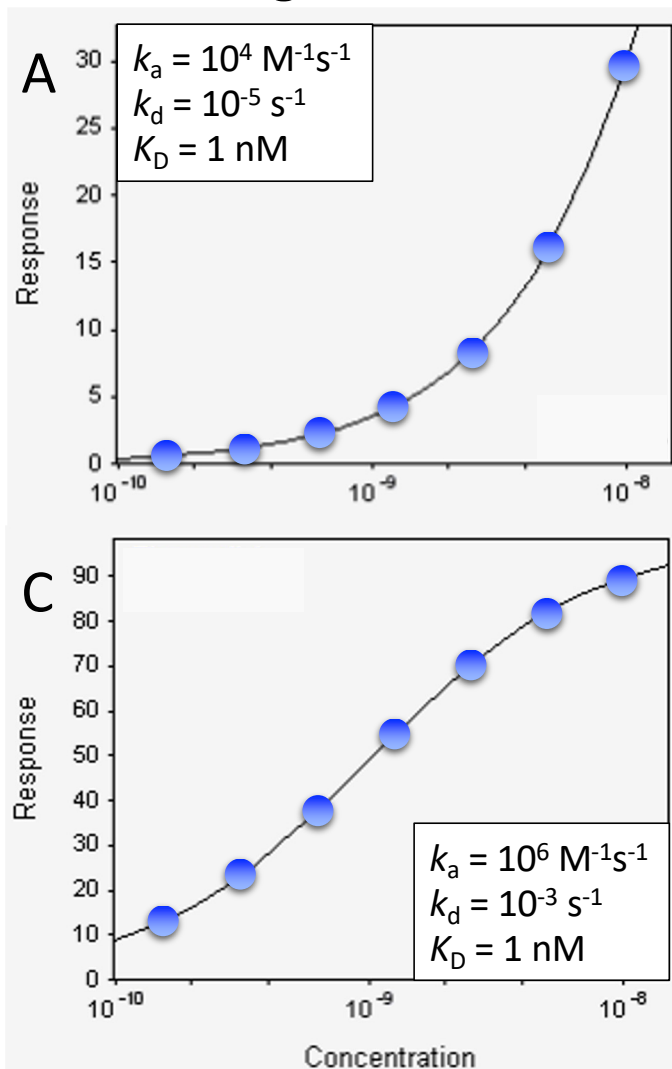
# Comparison of simulations for 1-to-1 binding



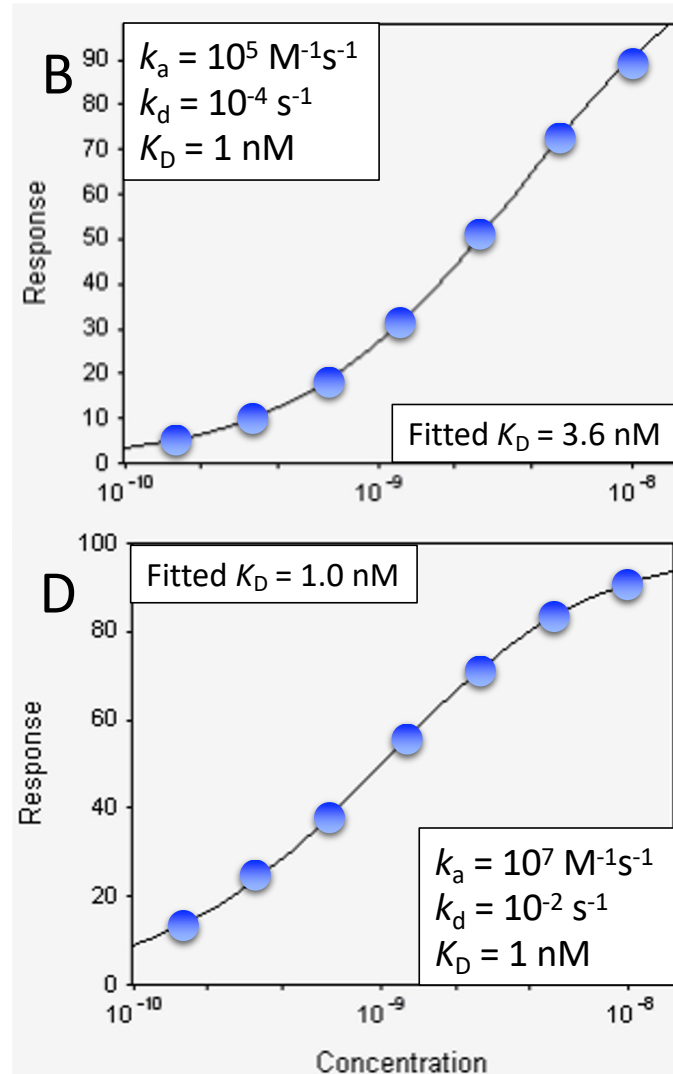
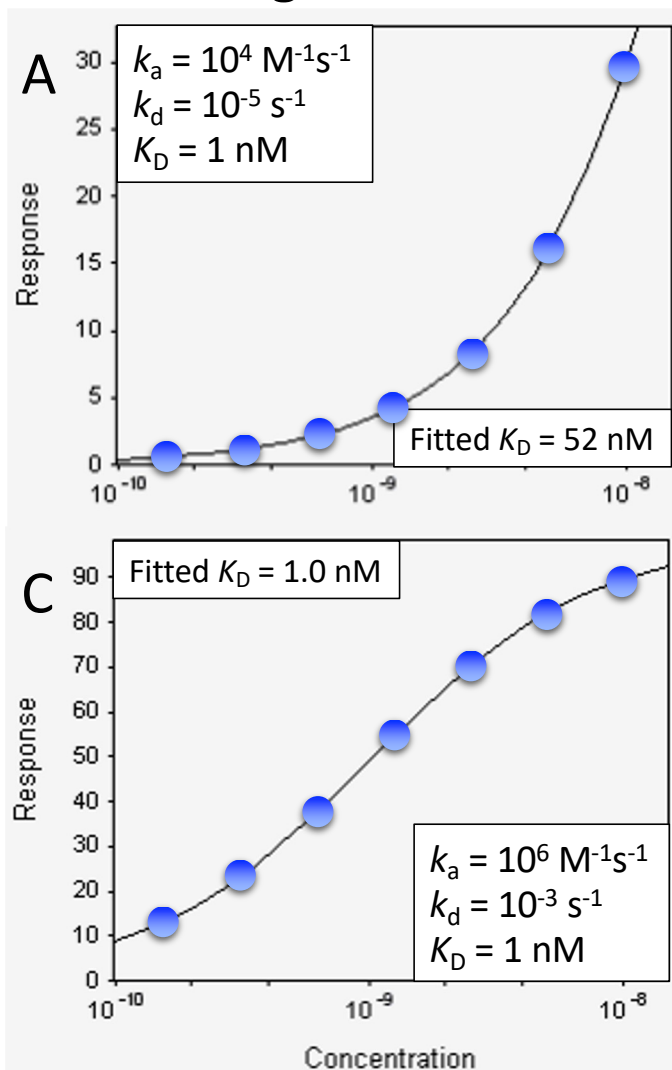
## What if this was an ELISA with a 1 hour incubation?



# Plot of binding isotherms at 1 hour



# Plot of binding isotherms at 1 hour



## What if this was an ELISA with a 1 hour incubation?

