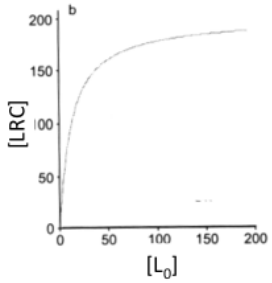
Module 03: Cellular Dynamics and High Throughput Biological Data
Assignment
Total Point Value = 30

Due by midnight on Day 7 of Module 3
This should be submitted to Blackboard as a pdf.

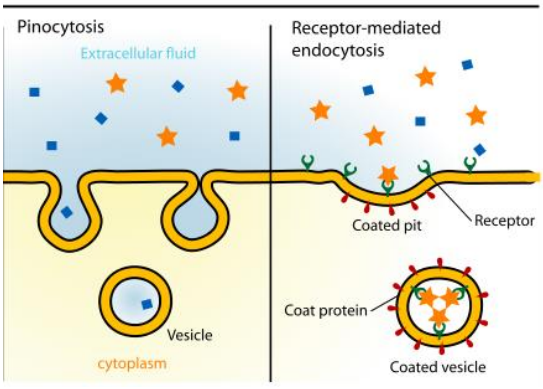
1. From *Tissue Engineering*, Saltzman
Exercise 4.2

a) The amount of cell-associated HRP is linear because it does not depend on ligand-binding kinetics --- there is no receptor to bind. When HRP is associated with the cell it is going through fluid-phase endocytosis. Therefore cell-associated HRP is simply proportionally to its abundance in the extracellular space. The amount of cell-associated EGF is hyperbolic because it is dependent on the **receptor concentration**. As we saw in the text (Figure 4.12 below) and lecture 2, at equilibrium the relationship between ligand, L_0 (EGF in this case) and the complex, LRC (which is the amount of cell-associate ligand) is hyperbolic. This is because at equilibrium with, assuming constant receptor concentration ($R_0 = R_{Total}$), the amount of receptor limits the amount of complex formed. Therefore increases in ligand/EGF concentration past the saturation of EGF receptor will not further increase the amount of LRC/cell-associated EGF.



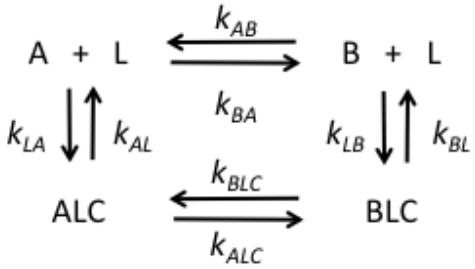
$$[LRC]|_{equilibrim} = \frac{R_T[L_0]}{[L_0] + K_d}$$

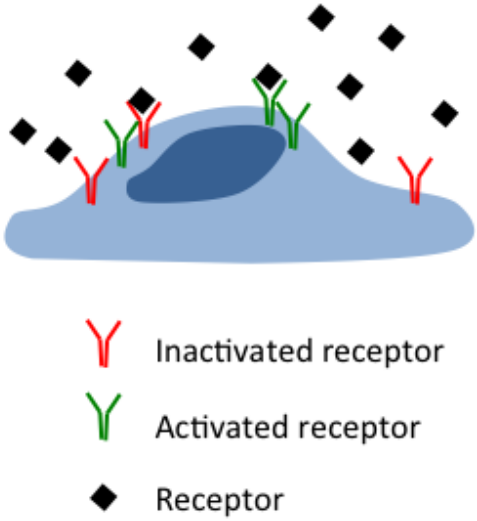
b) The rate of uptake with respect to ligand concentration is equal to the slope of the lines given in the graph 4.27. Looking at the graph you can see that the rate of uptake is faster for EGF than HRP (steeper slope) at low concentrations. This is because EGF associates with a specific cell receptor (the EGF receptor). At low concentration **the receptor is not limiting** and as EGF gets to the cell surface it can easily bind a receptor (becoming cell-associated). At low concentration of HRP the cell has no active mechanism to select HRP out of the bulk medium; instead the association of HRP is completely dependent on its concentration, which determines its likelihood of running into the membrane and being non-specifically associated with the cell during fluid-phase endocytosis. (Image source: http://www.newworldencyclopedia.org)



2. In order to model the protein dynamics of a ligand binding to a receptor you need to both write appropriate equations and know appropriate parameter values. In this problem you will investigate a 2 state receptor-ligand network. In this network the receptor is either active (called A) or inactive (called B). As we discussed earlier this semester – regulation occurs on many levels in the body and changing the activity state of a receptor is one level of regulation that allows for quick changes to the cell behavior. Instead of turning on a gene, transcribing , translating , folding and translocating – the cell can keep all of the receptors made in an inactive state and simply activate them when needed.

a. Please write ODEs to describe this system (following the Laws of Mass action). There should be 5 equations, one for each species present in this system (for example d[A]/dt)





$$\frac{dA}{dt} = k_{AL} * [ALC] - k_{LA} * [A][L] + k_{AB} * [B][L] - k_{BA}[A][L]$$
$$\frac{dB}{dt} = k_{BL} * [BLC] - k_{LB} * [B][L] + k_{BA} * [A][L] - k_{AB}[B][L]$$
$$\frac{d[ALC]}{dt} = k_{LA} * [A][L] - k_{AL}[ALC] + k_{BLC}[BLC] - k_{ALC}[ALC]$$
$$\frac{d[BLC]}{dt} = k_{LB}[B][L] - k_{BL}[BLC] + k_{ALC}[ALC] - k_{BLC}[BLC]$$
$$\frac{d[L]}{dt} = k_{AL}[ALC] - k_{LA}\{A][L] + k_{BL}[BLC] - k_{LB}[B][L]$$

(1.2 points for each equation, 0,3 points for each term)

b. Please describe *methods* you could use to measure the necessary parameters including rate constants and species concentration.

The quick and dirty answer here is (notes on these methods found in lecture 1):
Species concentrations can be measured using FCS or FCCS
Rate constants can be measured using FRAP, FRET or FCS.

Assignment Rubric

Question	Component	Total Point Value
1	a	7
	b	7
2	a	6
	b	10

Total Point Value = 30