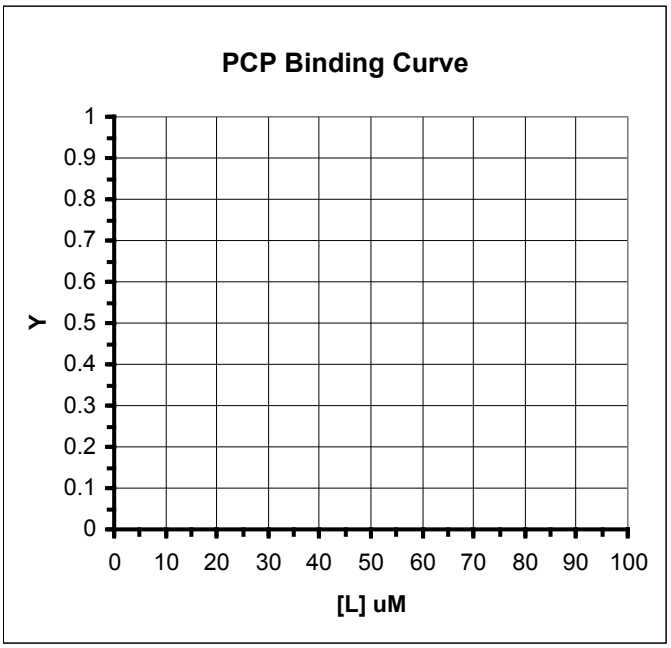
Ligand Binding Problem - Single Site Non-cooperative.

a) An equilibrium dialysis experiment is set-up to measure the binding affinity of PCP to the F_v fragment of an antibody. A total of 1 □M of F_v fragment was placed in the bag and radioactive PCP was placed outside the bag. At equilibrium, the concentration of PCP outside the bag was found to be 40 □M and the concentration of PCP inside the bag was 40.8□M. Calculate the fractional saturation, Y, at this particular ligand concentration.

$$Y = \frac{[ML]}{[M]+[ML]} = \frac{[ML]}{[M_T]}$$



b) The experiment was repeated with two additional ligand concentrations. Estimate the K_D from the binding curve.

$$Y = \frac{[L]}{K_D + [L]}$$

[L](□M)	Y	Y/L
0	0	-
3.4	0.25	0.075
40.0		
100.0	0.91	0.009

Determine K_D using a Scatchard Plot.

Scatchard Plot:

$$\frac{Y}{[L]} = \frac{-1}{K_D} Y + \frac{1}{K_D}$$

A plot of Y/[L] *versus* Y will give a straight line with a slope of -1/K_D.