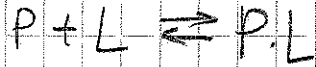
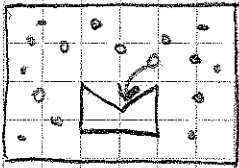


①

Haemoglobin and MyoglobinOutline

1. Binding reaction, Scatchard plot
2. Binding to n independent sites
3. Hill equation, Hill coefficient
4. MWC model

1. Simple binding reaction


 E_b - binding energy

$$Y = \frac{[PL]}{[PL] + [P]}$$

↑ site occupancy

— free F_{free}

$$F_{\text{free}} = 0 - TS$$

— bound F_{bound}

$$F_{\text{bound}} = -E_b \quad (E_b > 0)$$

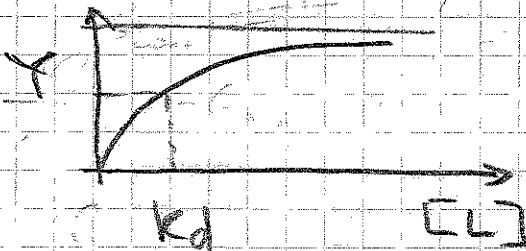
Estimate entropy in the free state: $S^f = \log \omega$

$$= \log \omega - \# \text{ of states per molecule} = \frac{V}{N_L} = \frac{1}{[L]}, \text{ where}$$

$$[L] = \frac{N_L}{V} \text{ - concentration}$$

$$\Rightarrow S = -\log [L]$$

$$Y = \frac{e^{-F_{\text{bound}}/T}}{e^{-F_{\text{bound}}/T} + e^{-F_{\text{free}}/T}} = \frac{e^{E_b/T}}{e^{E_b/T} + e^S} = \frac{[L]}{K_d + [L]}$$

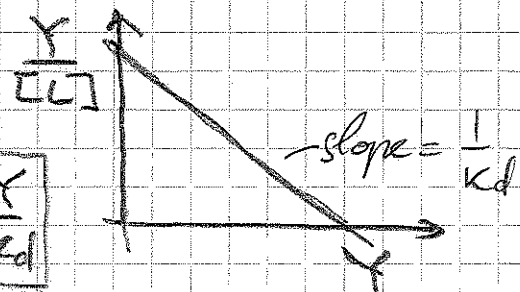


where $K_d = e^{-E_b/T}$

* Scatchard Plot

$$Y(K_d + [L]) = [L]$$

$$\frac{Y}{[L]} K_d + Y = 1 \Rightarrow \boxed{\frac{Y}{[L]} = \frac{1}{K_d} - \frac{Y}{K_d}}$$



②

2. Independent sites

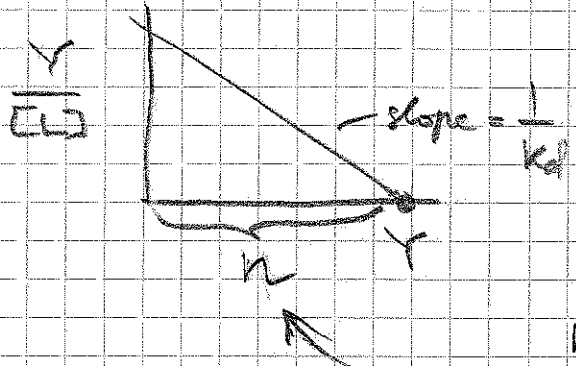


occupancy of a site

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

occupancy of the protein

$$Y_P = \frac{n[L]}{K_d + [L]}$$



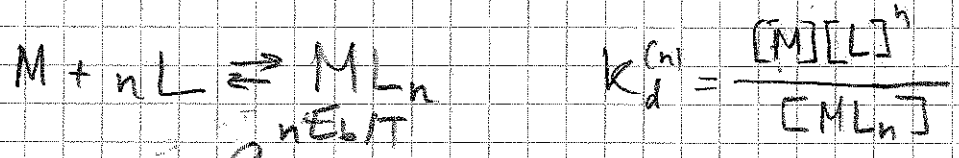
Scatchard plot

$$\frac{Y_P}{n[L]} = \frac{1}{K_d} - \frac{Y}{nK_d} \Rightarrow \frac{Y_P}{[L]} = \frac{n}{K_d} - \frac{Y}{K_d}$$

3. Cooperative binding to n sites

Hill model of hemoglobin (1910)

n sites, either all occupied or all vacant



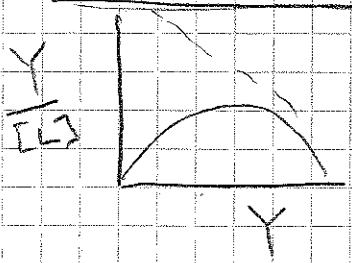
$$Y = \frac{e^{-nE_b/T}}{e^{-nE_b/T} + e^{-n \log(L)}} =$$

entropy loss upon binding of n ligands

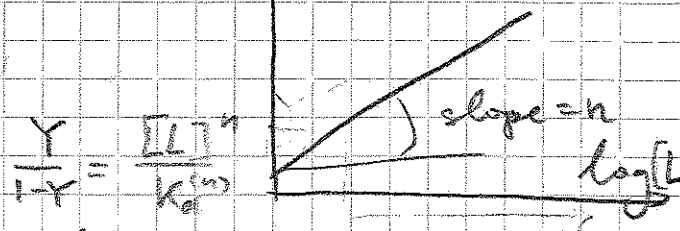
$$Y = \frac{[L]^n}{K_d^{(n)} + [L]^n}$$

Hill equation

Scatchard is non-linear



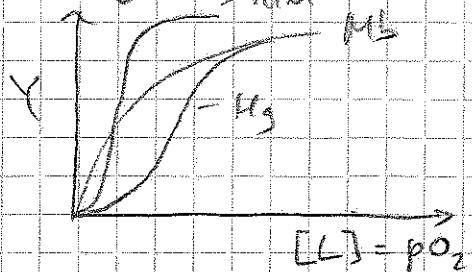
Hill plot $\log \frac{Y}{1-Y}$



$$\frac{Y}{1-Y} = \frac{[L]^n}{K_d^{(n)}}$$

$$\log \frac{Y}{1-Y} = n \log [L] - \log K_d^{(n)}$$

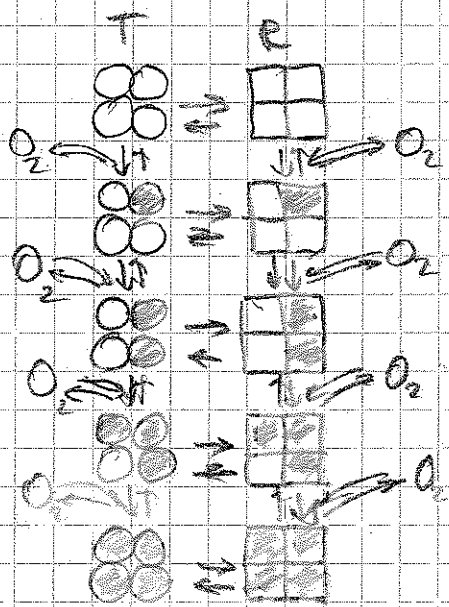
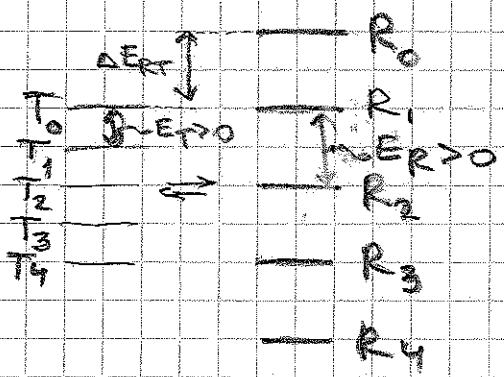
③ Hemoglobin Hill (1910)



=> need a better model

* Monod - Wyman - Changeux model (1965)
(MWC)

- 1) Hemoglobin consists of $n=4$ domains
- 2) All domains can be either in R or in T states.
- 3) O_2 binds to the R state with higher affinity
- 4) In the unbound form, T state has a lower energy



Parameters of the model:

$$L = \frac{[T_0]}{[R_0]} = e^{-\Delta E_{RT}} \gg 1$$

$$\alpha = \frac{\alpha_R}{\alpha_T} = \frac{[L]}{K_d^R} = [L] e^{E_R/RT}$$

$$c = \frac{K_R}{K_T} = \frac{\alpha_T}{\alpha_R} = e^{(E_T - E_R)/RT} \ll 1$$

$$Z = \sum_{i=0}^n [w(R_i) + w(T_i)] \text{ , where } w(R_i) = \alpha^i C_n^i$$

$$w(T_i) = L \cdot (\alpha c)^i C_n^i$$

$$= \sum_{i=0}^n C_n^i \alpha^i + L \sum_{i=0}^n C_n^i (\alpha c)^i =$$

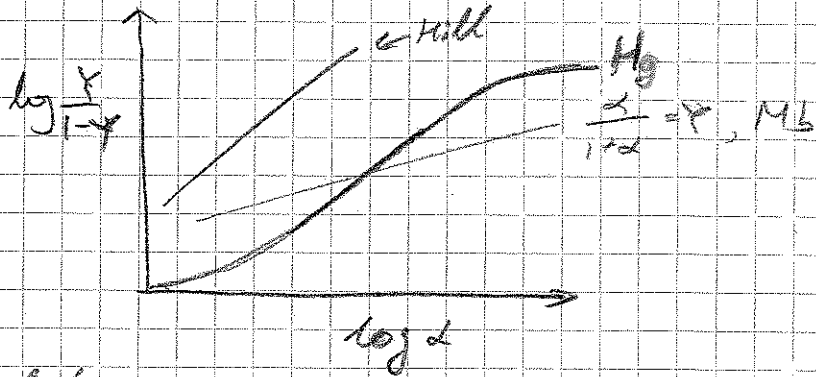
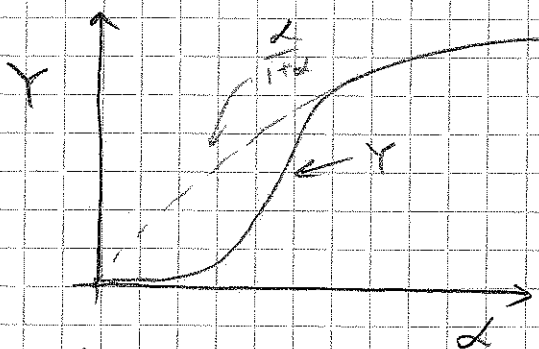
$$= (1 + \alpha)^n + L(1 + \alpha c)^n$$

④

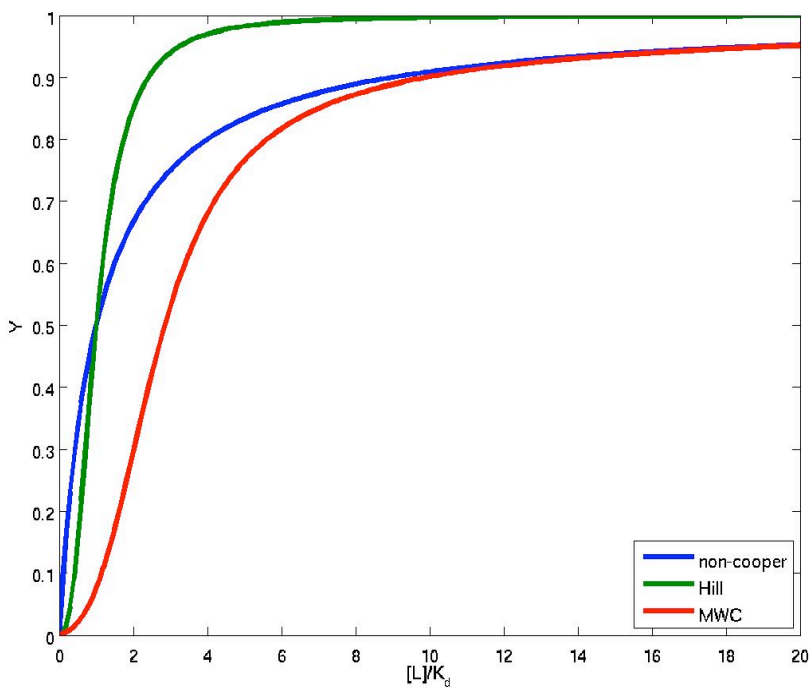
$$Y = \frac{\sum_{i=0}^n i \cdot C_n^i [w(R_i) + w(O_i)]}{\sum_{i=0}^n C_n^i [w(R_i) + w(O_i)]}$$

$$\sum_{i=0}^n i \cdot C_n^i \alpha^i = \sum_{i=0}^n i \cdot C_n^i \alpha^i = \alpha \sum_{i=0}^n i \cdot C_n^i \alpha^{i-1} = \alpha \frac{d}{d\alpha} \sum_{i=0}^n C_n^i \alpha^i = \alpha \frac{d}{d\alpha} (1+\alpha)^n = \alpha n (1+\alpha)^{n-1}$$

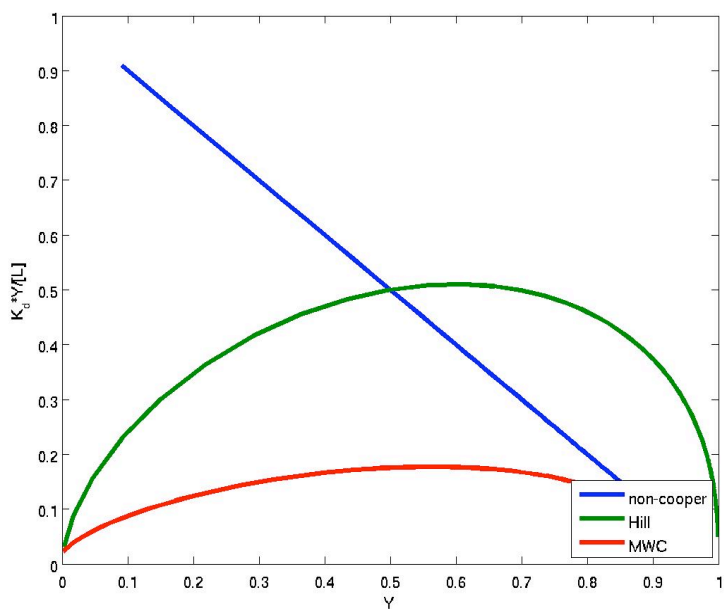
$$Y = \frac{K(\alpha(1+\alpha)^{n-1} + L\alpha(1+\alpha C)^{n-1})}{K(1+\alpha)^n + L(1+\alpha C)^n} = \alpha \frac{(1+\alpha)^{n-1} + LC(1+\alpha C)^{n-1}}{(1+\alpha)^n + L(1+\alpha C)^n}$$



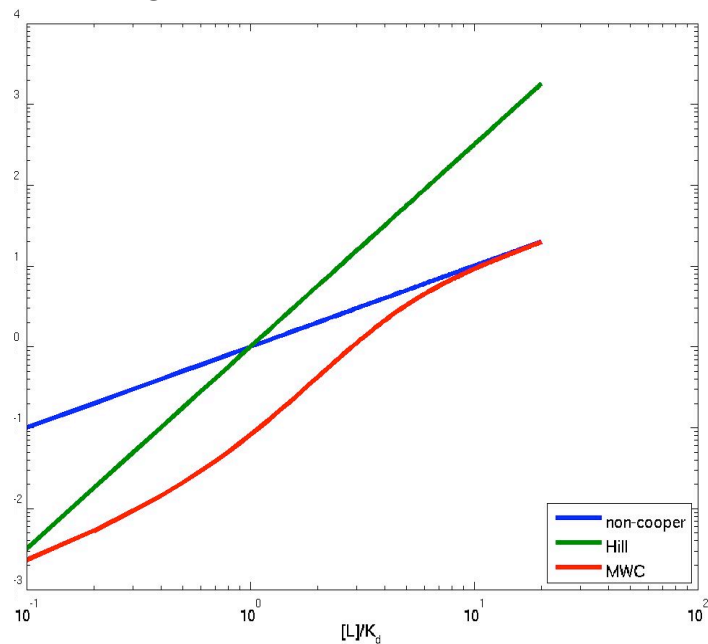
- effect of parameters C and L
- Bohr effect
- Heterotropic regulation



SCATCHARD PLOT



HILL PLOT



SENSITIVITY OF MWC TO PARAMETERS

