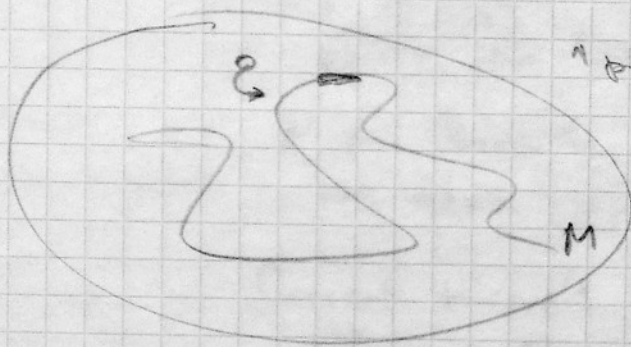


③ Kinetics of search

How long does it take for a protein to find its site?



1 protein, 1 specific site
M alternative sites.

Search can be quantified
by either k_{on} - association
rate

$$k_{on} [M^{-1}s^{-1}]$$

or t_s - search time

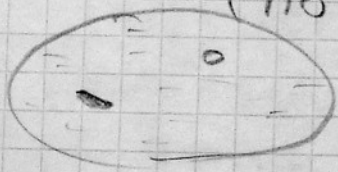
$$t_s [sec]$$

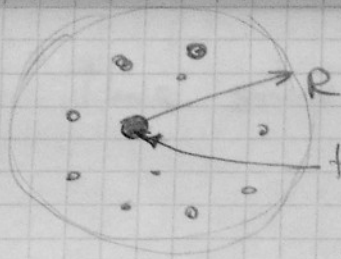
$$t_s = \frac{1}{k_{on} [P]}$$

⊗ Smoluchowski

theory of bi-molecular reactions.

(no long DNA, just a single protein
and its site on DNA)





Diffusion to a sphere

Diffusion equation:

$$\frac{\partial c(\mathbf{r}, t)}{\partial t} = D \nabla^2 c(\mathbf{r}, t)$$

target (immobile) of size r_0 .

$c(r)$ - concentration of proteins searching for their targets

$$k_{on} = I$$

← current of proteins through the surface of adsorbing sphere of radius r_0 .

First we calculate the density of proteins at $c(r_0)$ in steady state.

Diffusion equation: $\nabla^2 c(r) = 0$

Boundary conditions $c(R) = c(\infty)$

$$c(r_0) = 0 \quad (\text{adsorbing})$$

Only radial part

of the Laplacian is relevant (due to spherical sym)

$$\frac{1}{r^2} \frac{\partial}{\partial r} \left(r^2 \frac{\partial}{\partial r} c(r) \right) = 0 \quad \rightarrow c(r) = \frac{A}{r} + B$$

From Boundary conditions

we obtain $c(r) = \left(1 - \frac{r_0}{r}\right) c(\infty)$ for $R \gg r_0$.

Current through the element of the sphere \vec{J}

$$\vec{J} = -D \vec{\nabla} c(r) = +D \frac{r_0}{r^2} c(\infty)$$

$$\text{Total current } I = J(r_0) 4\pi r_0^2 = 4\pi D \frac{r_0}{r_0^2} \cdot r_0^2 c(\infty)$$

$$k_{on} = 4\pi D r_0 \times a$$

↳ fraction of the molecule's surface that is reactive.

Calculate k_{on}

for a protein binding site on DNA (in water).

$r_0 = 0.34 \text{ nm} = 1 \text{ bp}$ (since displacement by 1 bp leads to a different sequence)

$$D = 3 \cdot 10^{-7} \frac{\text{cm}^2}{\text{s}}$$

$$a \approx 0.2$$

× Avagadro's number to get $\text{M}^{-1} \text{s}^{-1}$ units

This gives

$$k_{on} \approx 10^8 \text{ M}^{-1} \text{ s}^{-1}$$

Diffusion limit for rate of this
bi molecular reaction.

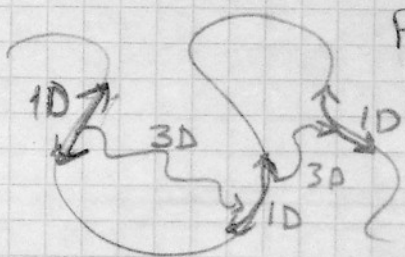
Experiment

$$k_{on} = 10^{10} \text{ M}^{-1} \text{ s}^{-1} \quad [\text{Riggs et al 1970}]$$

How can the rate be above the diffusion limit?

* Theory of facilitated diffusion

Instead of calculating k_{on} , we turn to t_s - mean search time.



Protein is capable of binding DNA non-specifically and sliding along it (1D)

Each round of search consists of 3D + 1D

$$t_s = K (\tau_{3D} + \tau_{1D})$$

\uparrow mean search time \uparrow # of rounds \uparrow mean duration of 3D phase of each round \uparrow mean duration of the 1D phase of each round.

M - length of the genome, n - # of sites visited in each 1D sliding.

$$\left\{ \begin{array}{l} \text{Prob. to hit a site} \\ \text{on a given 1D slide} \end{array} \right\} = \frac{n}{M} \Rightarrow K = \frac{M}{n}$$

$$t_s = \frac{M}{n} (\tau_{1D} + \tau_{3D})$$

$$n = \sqrt{4D_{1D} \tau_{1D}}$$

\uparrow 1D diffusion.

Find τ_{1D} that minimizes search time.

$$t_s = \frac{M}{\sqrt{4D_{1D} \tau_{1D}}} (\tau_{1D} + \tau_{3D})$$

$$\frac{\partial t}{\partial \tau_{1D}} = 0 \Rightarrow \tau_{1D}^{opt} = \tau_{3D}$$

$$\Rightarrow t_s^{opt} = 2 \frac{M}{n} \tau_{3D}$$

* 3D search alone: $n=1, \tau_{1D}=0$
 $t_s^{3D} = M \tau_{3D}$

* 1D search alone $\tau_{3D}=0, n=M \Rightarrow \tau_{1D} = \frac{M^2}{D_{1D}}$
 $t_s^{1D} = \frac{M}{M} \cdot \frac{M^2}{D_{1D}} = \frac{M^2}{D_{1D}}$

|| t_s^{opt} is $\frac{n}{2}$ times smaller than t_s^{3D}
and $\frac{M}{2n}$ times smaller than t_s^{1D}

$n \approx 100$ bp \Rightarrow Combined 3D/1D search
is about 100 faster
than diffusion limit.