Lecture 15

Going behind steady state (s.s) approximation:

1-Solving diffusion equation with no approximation for a point source in 1D.

2- A Lesson from part (1): Diffusion time scales...

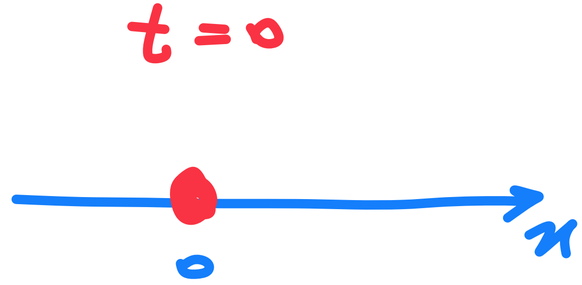
3-A Lesson from part (2): Pseudo (quasi) s.s approximation

1-Solving diffusion equation with no approximation for a point in 1D.

Example 1: Assume we have a point-like source of a species located at the center, similar to a small droplet of ink. As time passes, we know it will spread, meaning it is not at steady state (S.S.). The question is: can we calculate the concentration of this ink, CC, as a function of space and time?

a) $\frac{\partial C}{\partial t} = D \nabla^2 C$ no S.S

b) coordinate? 1D, only x



c) $\frac{\partial C}{\partial t} = D \frac{\partial^2 C}{\partial x^2}$ → trick → this is a PDE, but we can solve it with change of variables: $\eta \equiv \frac{x}{\sqrt{4Dt}}$ → dimensionless variable

now we can assume: $C(x,t) \equiv \frac{M}{\sqrt{4\pi Dt}} \times f(\eta)$

$\downarrow$

$C \text{ in 1D} \propto \frac{M}{L} \propto \frac{M}{\sqrt{Dt}} \propto \frac{M}{\sqrt{4\pi Dt}}$

$$\rightarrow \frac{\partial C}{\partial t} = \frac{M}{\sqrt{4\pi Dt}} \left(-\frac{f(\eta)}{2t} - \frac{\eta}{2t}\frac{\partial f}{\partial \eta}\right)$$

$$\frac{\partial^2 C}{\partial x^2} = \frac{M}{\sqrt{4\pi Dt}} \frac{\partial^2 f}{\partial \eta^2} \times \frac{1}{4Dt}$$

putting back to diffusion equation: $\rightarrow$

$$\Rightarrow \frac{M}{\sqrt{4\pi Dt}} \left(-\frac{f(\eta)}{2t} - \frac{\eta}{2t}\frac{\partial f}{\partial \eta}\right) = D \frac{M}{\sqrt{4\pi Dt}} \frac{\partial^2 f}{\partial \eta^2} \frac{1}{4Dt}$$

$$\Rightarrow -f(\eta) - \eta\frac{\partial f}{\partial \eta} = 2\frac{\partial^2 f}{\partial \eta^2} \Rightarrow \frac{d}{d\eta}\left(\frac{\partial f}{\partial \eta} + 2\eta f\right) = 0$$

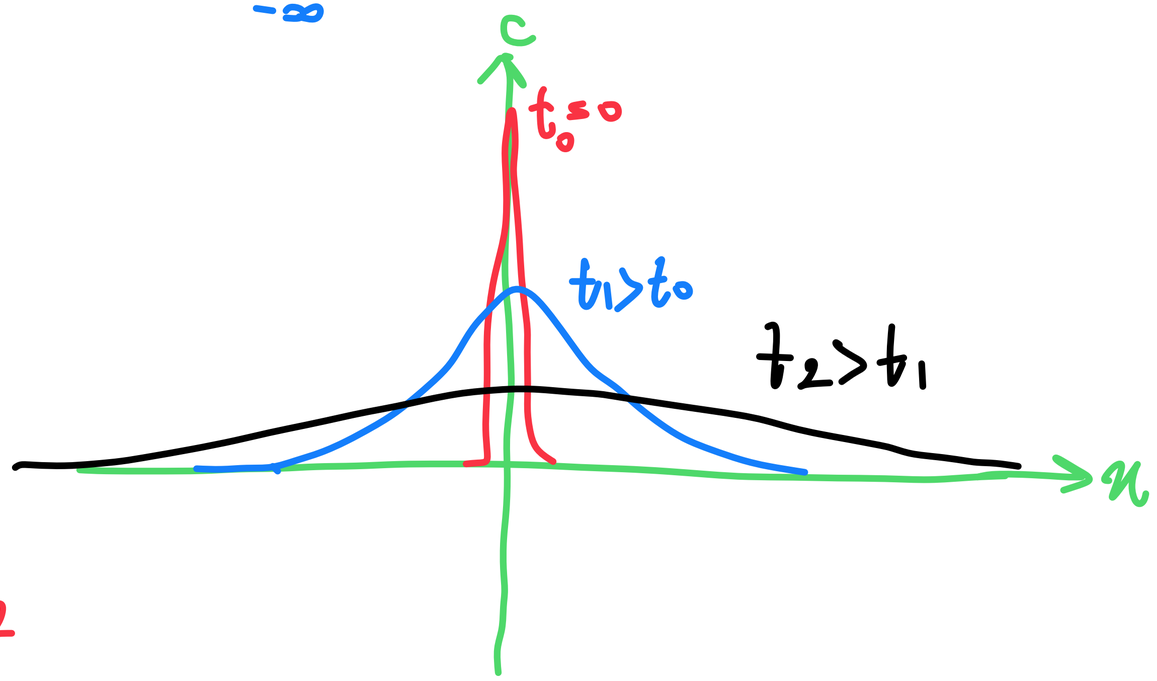
one solution is $\left(\cdots\right) = 0$ $\rightarrow$

$$f = A_0 e^{-\eta^2}$$

$$\Rightarrow C(x,t) = \frac{A_0}{\sqrt{4\pi D t}} e^{-\frac{x^2}{4Dt}}$$

we know that for total mass: $M = \int_{-\infty}^{+\infty} C\, dx \Rightarrow A_0 = M$

$$\Rightarrow C(x,t) = \frac{M}{\sqrt{4\pi D t}} e^{-\frac{x^2}{4Dt}}$$



note that we have $e^{-\text{Some value}\left(\frac{x}{\sqrt{Dt}}\right)^2}$

Going behind steady state approximation:

2) About time scale of diffusion:

Looking at diffusion equation, what will determine the time-scale of diffusion?

- $\frac{\partial C}{\partial t} = D \nabla^2 C \longrightarrow$ looking at dimentions $D \propto \frac{[L]^2}{[S]}$

or from our 1D example:

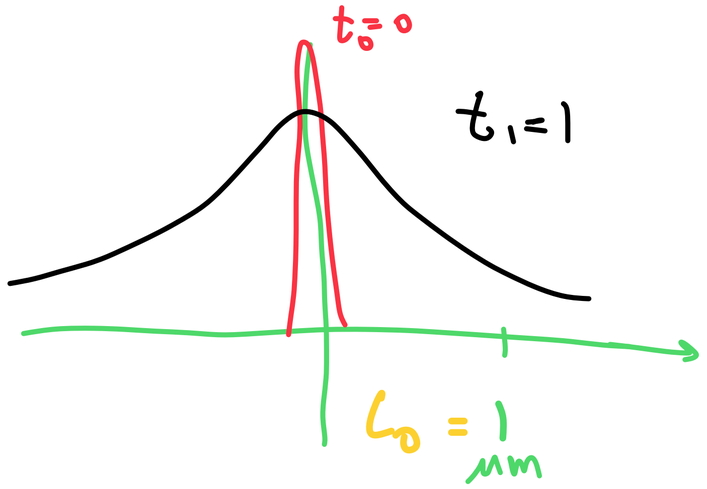
- $C(x,t) \propto e^{-(\text{something dimensionless}) \times \left(\frac{x^2}{Dt}\right)} \rightarrow \frac{[L]^2}{[D][S]} = 1 \rightarrow D \propto \frac{[L]^2}{[S]}$

Now we focus on our 1D example, how does the value of 'D' effect the dynamics of diffusion?

$$c(x,t) = \frac{M}{\sqrt{4\pi Dt}} e^{-\frac{1}{2}\left(\frac{x}{\sqrt{Dt}}\right)^2}$$

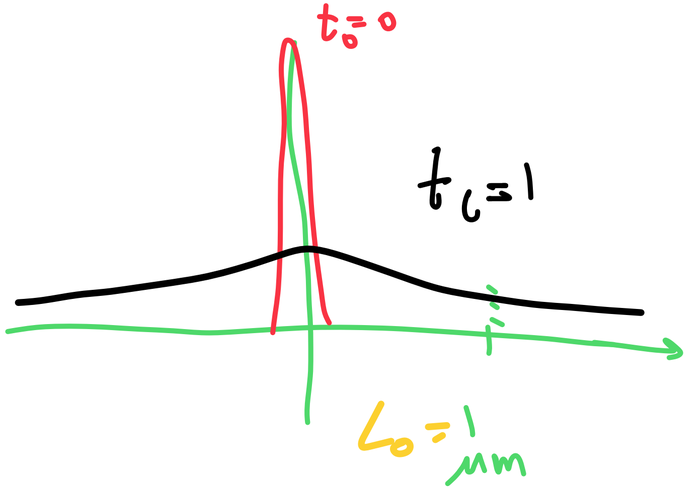
scenario 1:

$D_1 = 1 \frac{\mu m^2}{s}$



scenario 2:

$D_2 = 10 \frac{\mu m^2}{s}$



Spreads faster

Conclusion: The time-scale for particles to reach position L0, is at the order of:

$$\tau \approx \frac{(L_0)^2}{D}$$

We call this diffusion time scale, the shorter it is, the diffusion reaches S.S faster.

Let's look at some biologically relevant numbers [https://book.bionumbers.org/]

$$\gamma \propto \frac{L_0^2}{D}$$

molecule	measured context	diffusion coefficient ($\mu m^2/s$)
H_2O	water	2000
H_2O	nucleus of chicken erythrocyte	200
H^+ (from H_3O^+ to H_2O)	water	7000
O_2	water	2000
CO_2	water	2000
tRNA (≈20 kDa)	water	100
protein (≈30 kDa GFP)	water	100
protein (≈30 kDa GFP)	eukaryotic cell (CHO) cytoplasm	30
protein (≈30 kDa GFP)	rat liver mitochondria	30
protein (NLS-EGFP)	cytoplasm of *D. melanogaster* embryo	20
protein (≈30 kDa)	*E. coli* cytoplasm	7-8
protein (≈40 kDa)	*E. coli* cytoplasm	2-4
protein (≈70-250 kDa)	*E. coli* cytoplasm	0.4-2
protein (≈140 kDa Tar-YFP)	*E. coli* membrane	0.2
protein (≈70 kDa LacY-YFP)	*E. coli* membrane	0.03
fluorescent dye (carboxy-fluorescein)	*A. thaliana* cell wall	30
fluorescent dye (carboxy-fluorescein)	*A. thaliana* mature root epidermis	3
transcription factor (LacI)	movement along DNA (1D, *in vitro*)	0.04 (4×10^5 bp^2s^{-1})
morphogen (bicoid-GFP)	cytoplasm of *D. melanogaster* embryo	7
morphogen (wingless)	wing imaginal disk of *D. melanogaster*	0.05
mRNA	HeLa nucleus	0.03-0.10
mRNA	various localizations and sizes	0.005-1
ribosome	*E. coli*	0.04

mammal cell
$L_0 = 10\,mm$

$$\gamma \propto \frac{100\,mm^2}{2000\,\frac{mm^2}{s}} = 50\,ms$$

E coli
$L_0 = 1\,mm$

$$\gamma \propto \frac{1\,mm^2}{2000\,\frac{mm^2}{s}} = 0.5\,ms$$

$L_0 = 1\,\mu m$

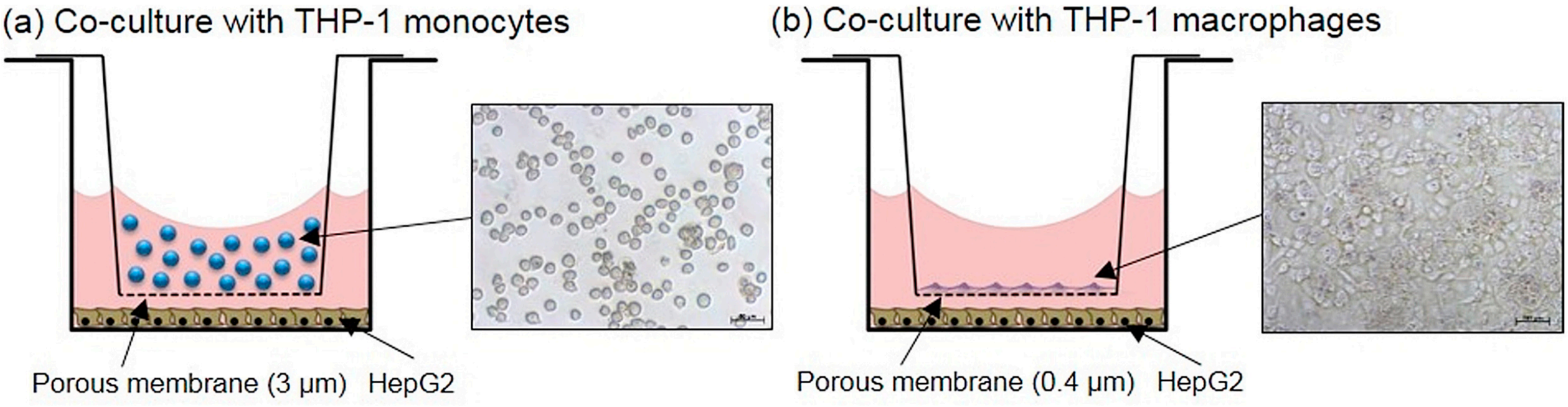
$$\gamma \propto \frac{1\,\mu m^2}{5\,\frac{\mu m^2}{s}} = 40\,ms$$

3-Pseudo (quasi) s.s approximation:

We learned that when analyzing diffusion in biological systems, under certain conditions (such as when the system is small (L0)and enough time has passed ($t>>\tau$)), we can use the steady-state (S.S.) approximation ($dc/dt=0$). Here, $c(x,t)$ represents the concentration profile within the compartment of interest (e.g., cell membrane, lung tissue, etc.).

However, when considering a larger system, where diffusive transport occurs only in a small part of it, and if the timescale of diffusion is much shorter than the timescale of concentration changes in the larger system, we can assume that diffusion is always at steady state while the overall system's concentration is changing. Let's consider a concrete example to see how this approach works. Note that this method is powerful when analyzing biological systems with different part and different timescales involved.

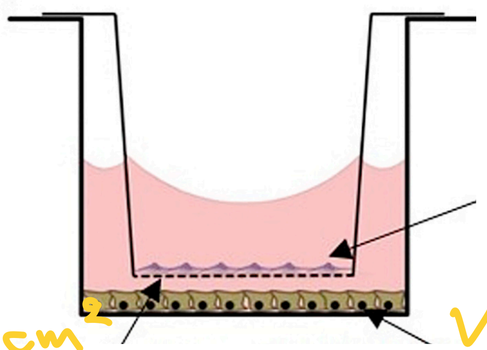
Background: Co-culture systems are widely used in bioengineering, tissue engineering, and biomedical research because they allow scientists to study the interactions between different cell types in a controlled environment. Unlike traditional monoculture systems, where a single cell type is grown in isolation, co-cultures more closely mimic the complexity of natural biological systems, such as tissues or organs, by allowing multiple cell types to communicate through soluble factors (like cytokines) or direct cell-cell contact.



Granitzny, Anne, et al. "Evaluation of a human in vitro hepatocyte-NPC co-culture model for the prediction of idiosyncratic drug-induced liver injury: a pilot study." *Toxicology reports* 4 (2017): 89-103.

Example 2: An engineer might face the challenge of designing a thin membrane for cytokine exchange. How will the thickness of the membrane, where materials diffuse, and its area and it's diffusivity impact the dynamics of cytokine changes in the whole system?

Let's consider a system with two chambers, 1 and 2, separated by a membrane (m) as shown below. Initially, cells are cultured in chamber 1 (with volume V1), where the cytokine concentration reaches C0. Then, chamber 2 (with volume V2) is added, separated by a porous membrane with thickness L0 and area A and diffusivity of D. The entire system is placed on a slow shaker to ensure that the media in each chamber is well-mixed. This setup allows us to explore how membrane properties like thickness and area affect cytokine exchange between the chambers.



$C_0 = 10 \frac{mg}{L}$

$V_2 = 2$ mL = 2 cm³

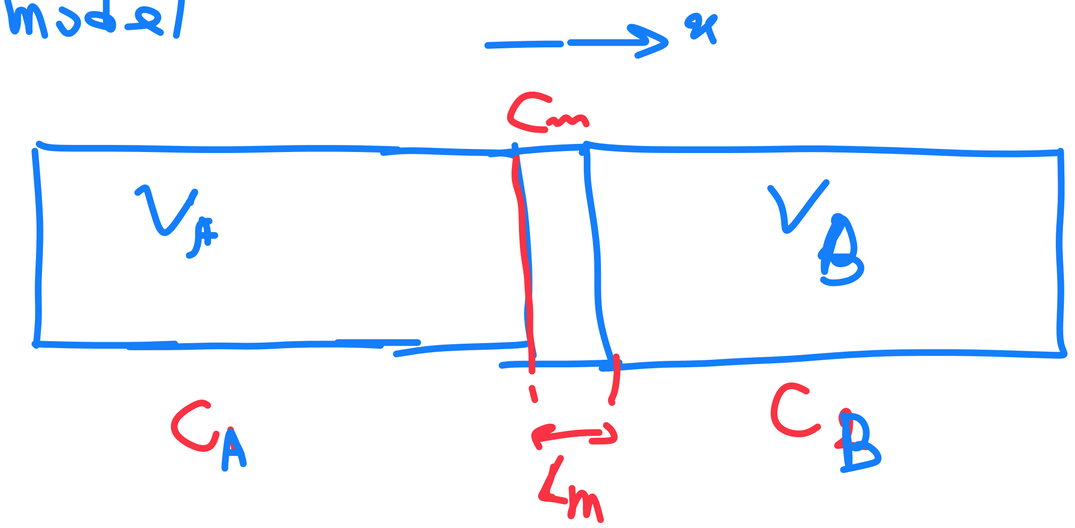
$V_1 = 2$ mL = 2 cm³

$A = 1\ cm^2$

$L_0 = 1\ \mu m$

$D = 10 \frac{mm^2}{s}$

our model



a) assuming Q.S.S is valid so diffusion in membrane is at S.S all the time while C_A and C_B are changing slowly

b) we know from before: $C_m = C_A - (C_A - C_B)\frac{x}{L_0}$

$$\Rightarrow J = \frac{D}{L_0}(C_A - C_B)$$

c) mass balance for chamber B: $\frac{dm}{dt} = r_i - r_o + r_g - r_c$

$\frac{dm}{dt} = C_B \times V_B$

$r_i = J(x = L_0) \times A_m$

$r_o = 0$, $r_g = 0$, $r_c = 0$

$$\Rightarrow V_B \frac{dC_B}{dt} = \frac{A_m \times D_m}{L_m} (C_A - C_B) \quad (1)$$

d) now let's write mass balance for the whole system:

$$\frac{dm}{dt} = \cancel{F_i} - \cancel{F_o} + \cancel{r_A} - \cancel{r_C}$$

$$\downarrow$$

$$\left(\frac{dC_A}{dt}\right)V_A + \cancel{\left(\frac{dC_m}{dt}\right)V_m} + \left(\frac{dC_B}{dt}\right)V_B = 0 \quad \text{(Q.S.S} \Rightarrow 0\text{)}$$

$$\frac{dC_A}{dt} = -\frac{V_B}{V_A}\frac{dC_B}{dt}$$

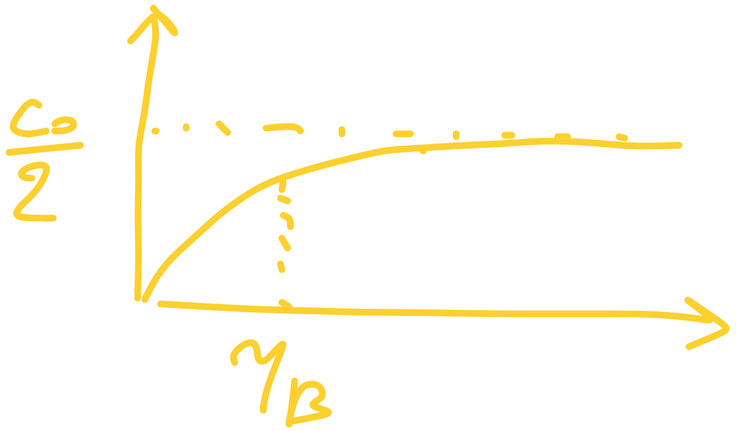
$$\Rightarrow V_A = V_B, \; C_B(t=0) = 0$$

$$C_B = C_0 - C_A$$

(1) & (2) $\Rightarrow$

$$V_B \frac{dC_B}{dt} = \left(\frac{A_m D_m}{L_m}\right)(C_0 - 2C_B(t))$$

$$\Rightarrow C_B(t) = \frac{C_0}{2}\left(1 - e^{-\left(\frac{A_m D_m}{L_m V_B}\right)t}\right)$$



γ_B is the time scale of this, let's take a look at it:

$$\gamma_B \propto \frac{L_m V_B}{A_m D_m} = \frac{1\,\mu m \times 2\,cm^3}{1\,cm^2 \times 10\,\frac{\mu m^2}{s}} \approx 1000\,s$$

what about diffusion in membrane? $\gamma_m \propto \frac{L_m^2}{D} = \frac{1}{10}\,s$

$\gamma_B \gg \gamma_m \longrightarrow$ membrane reaches S.S faster $\Rightarrow$ Q.SS ✓

also note that γ_B and similarly γ_A are function of

$$\gamma_{A \text{ or } B} \propto \frac{L_m}{D_m A_m} \times V_B$$

Questions?