## Physical properties of membranes

(Müller & Rudin “black lipid bilayer” model membrane)

<table>
<thead>
<tr>
<th>Physical property</th>
<th>Cell membrane</th>
<th>Lipid bilayer</th>
</tr>
</thead>
<tbody>
<tr>
<td>Capacitance</td>
<td>~ 1 µF/cm²</td>
<td>~ 1 µF/cm²</td>
</tr>
<tr>
<td>Resistance</td>
<td>~ $10^3$ Ω cm²</td>
<td>$10^6$–$10^9$ Ω cm²</td>
</tr>
<tr>
<td>H₂O permeability</td>
<td>90–400 µm/sec</td>
<td>17–104 µm/sec</td>
</tr>
<tr>
<td>Surface tension</td>
<td>0.3–1 dyne/cm</td>
<td>0.5–2 dyne/cm</td>
</tr>
</tbody>
</table>

**Diagram:**

- $I_i(x)$: Current at position $x$.
- $R_i$: Resistance.
- $C_m$: Capacitance.
- $R_m$: Resistance.
- $V_m$: Voltage.
- $I_i(x+dx)$: Current at position $x+dx$.
- $j_i(x)$: Current density.
Membrane Electrostatics

Complex Electrostatic Environment

- Many atoms
- large density of charges and dipoles
- 10’s mV potential across 6 nm!
- large variation in dielectric constant

DMPC
Transmembrane Potential

The potential difference between two bulk phases.

Charge separation across water ($\varepsilon = 80$) will tend to create a relatively small electric field.

Charge separation across organic phases ($\varepsilon = 1.89$) will create large electric fields.
Two parallel planes of opposite charges

The electrical potential difference between the plates is

$$\Delta \psi = - \int_{0}^{x} E \, dx = - \int_{0}^{x} \frac{q}{\varepsilon \varepsilon_0} \, dx = -q \frac{x}{\varepsilon \varepsilon_0}$$

$$E = \frac{q}{\varepsilon \varepsilon_0}$$

$$C = \frac{q}{\Delta \psi} = A \varepsilon \varepsilon_0 \frac{x}{x}$$

The capacitance of biological membrane is about 1 $\mu$F/cm$^2$. 
Membrane potential is the force that drives ions across the membrane.

The membrane potential partly determines the energy stored in ion concentration gradients.

The electric field associated with the membrane potential acts on dipolar groups in membrane proteins and may regulate the activities of these proteins.

Voltage-dependent channels open and close in response to changes in the membrane potential.
Membranes have a planar symmetry

A charge smoothly distributed on a plane will create an electric field perpendicular to the plane.

\[ E = \frac{-q_s}{2 \varepsilon_o} \]

\[ \psi(x) \approx \psi_0 e^{-x/\lambda} \]

Surface potential controls protein adsorption and can regulate membrane transport.
**Origin of surface charge**

- **Ionisation of surface groups** (ionisation of carboxyl and amino groups to give \( \text{COO}^- \) and \( \text{NH}_3^+ \) ions)

- **Ion adsorption**
  - Cations more hydrated than anions, have greater tendency to reside in bulk.
  - Anions have greater tendency to be specifically adsorbed.

- Any ion whose adsorption at surface is influenced by forces other than simple electrostatic potential can be regarded as specifically adsorbed.

![Specific Adsorption Diagram](image)
The structure of several common mammalian phospholipids that can be found in the plasma membrane along with their charge ranges under physiological conditions.
The charge separation depends on:
- a dynamic tension with diffusion pushing the counterions away from the surface
- electrical attraction pulling them toward the surface
- a dynamic tension with diffusion pushing the counterions away from the surface

Generally membranes have a negative charge (10 - 20% anionic lipids, charge from gangliosides and proteins)

Membrane surface electrostatic potential

A plane of fixed charges with a surface charge density \( \sigma_s \).

Space charge density \( \rho_v(x) \).
STERN MODEL (1924)

- **Surface potential**
  - $\psi_0$

- **Stern potential**
  - $\psi_\delta$

- **Stern layer**

- **Stern layer thickness** $\delta$

- $\psi(x)$
Strong specific adsorption in the Stern Layer

Counter-ions

Co-ions
**Electrical double layer**

- **Helmholtz model (1879)**
- **Guoy-Chapman model (1914)**
The Gouy-Chapman theory

Assumptions:

1. The surface charge is smeared
2. Ions are point charges
3. No specific ion adsorption
4. Dielectric constant is constant

The ion distributions as a function of distance from the membrane surface.

Mean field approximation

Each ion is moving under the influence of an electric potential created by the average charge density of the others $<\rho_q>$. 
**Salt ions shield charged objects in solution**

\[ \mu = \mu^0 + k_B T \ln(c) \]

\[ \mu' = \mu^0 + k_B T \ln(c) + ze\Psi(x) \]

**Equilibrium:**

\[ \begin{align*}
\mu'_1 &= \mu'_2 \\
\mu^0 &= \text{const} \\
k_B T \ln[c(x_1)] + ze\Psi(x_1) &= k_B T \ln[c(x_2)] + ze\Psi(x_2)
\end{align*} \]
Salt ions shield charged objects in solution

Equilibrium:

\[ \begin{align*}
   \mu'_1 &= \mu'_2 \\
   \mu^0 &= \text{const} \\
   k_B T \ln[c(x_1)] + ze\Psi(x_1) &= k_B T \ln[c(x_2)] + ze\Psi(x_2)
\end{align*} \]

Nernst Equation:

\[ \ln \frac{c(x_1)}{c(x_2)} = \frac{-ze[\Psi(x_2) - \Psi(x_1)]}{k_B T}, \quad \text{or} \]

\[ c(x_2) = c(x_1) \exp \left[ \frac{-ze[\Psi(x_2) - \Psi(x_1)]}{k_B T} \right] \]
Salt ions shield charged objects in solution

\[
n_+(x) = n_\infty \exp\left(\frac{-ze\Psi(x)}{k_BT}\right)
\]

\[
n_-(x) = n_\infty \exp\left(\frac{+ze\Psi(x)}{k_BT}\right)
\]

\[
\rho(x) = ze[n_+(x) - n_-(x)]
\]
Salt ions shield charged objects in solution

At a distance $x$ from the surface:

\[
\begin{align*}
n_-(x) &= n_\infty \exp\left(\frac{+ze\Psi(x)}{k_B T}\right) \\
n_+(x) &= n_\infty \exp\left(-\frac{ze\Psi(x)}{k_B T}\right)
\end{align*}
\]

\[\rho(x) = ze[n_+(x) - n_-(x)]\]

Poisson Equation:

\[\nabla^2 \Psi = -\frac{\rho}{\varepsilon \varepsilon_0}\]

Poisson-Boltzmann Equation:

\[
\nabla^2 \Psi = -\frac{\rho}{\varepsilon \varepsilon_0} = \frac{zen_\infty}{\varepsilon \varepsilon_0} \left[\exp\left(\frac{ze\Psi}{k_B T}\right) - \exp\left(-\frac{ze\Psi}{k_B T}\right)\right]
\]

Linearized Poisson-Boltzmann Equation (Debye-Hückel equation):

\[\nabla^2 \Psi = \frac{2zen_\infty}{\varepsilon \varepsilon_0} \left(\frac{ze\Psi}{k_B T}\right) = \kappa^2 \Psi\]

\[1/\kappa \text{ is the Debye length}\]
**Poisson equation**
(describes variation of potential in a spatial charge distribution)

$$\frac{d^2 \psi}{dx^2} = - \frac{1}{\varepsilon \varepsilon_0} \rho_e \sum_{i} z_i e n_i(x)$$

**Boltzmann equation**
(describes variation of ion concentration in an electric field)

$$\frac{n_i(x)}{n_i(\infty)} = \exp\left(\frac{-z_i e \psi(x)}{kT}\right)$$

**Poisson-Boltzmann equation**

$$\frac{d^2 \psi}{dx^2} = - \frac{1}{\varepsilon \varepsilon_0} \sum z_i e n_i(\infty) \exp\left(\frac{-z_i e \psi}{kT}\right)$$
For a single, symmetric (z-z) electrolyte…

\[
\frac{d^2\psi}{dx^2} = -\frac{ze n(\infty)}{\varepsilon\varepsilon_0} \left[ \exp\left(\frac{ze\psi}{kT}\right) - \exp\left(\frac{-ze\psi}{kT}\right) \right],
\]

for low potentials \(|z\psi_0| \leq 25 \text{ mV}, \sinh y \approx y, \text{ and}\)

\[
\frac{d^2\psi}{dx^2} = \frac{2z^2e^2n(\infty)}{\varepsilon\varepsilon_0 kT} \psi = \kappa^2 \psi
\]

**b.c.’s**
1. \(x = 0; \psi = \psi_0\)
2. \(x \to \infty; \psi = 0\)

**Potential profile**
\[
\psi(x) = \psi_0 e^{-\kappa x}
\]

“Debye-Hückel approximation”
**Ion concentration profile**

\[ C_i(x) = C \exp \left[ -\frac{z_i e}{kT} \psi_0 \exp(-\kappa x) \right] \]

For water, 25°C:

\[ \kappa^{-1} \approx \frac{0.30}{z \sqrt{C(M)}} \text{ nm} \]

0 < \kappa^{-1} < 1000 nm

**Double layer thickness**

"Double layer thickness"
1/κ is a measure of the thickness of the diffuse double layer.

**Debye length for**
- 0.1 M NaCl, \( L_D = 0.96 \) nm,
- 0.01 M NaCl \( L_D = 3.04 \) nm
- 0.01 M MgCl\(_2\) \( L_D = 1.75 \) nm

**The decay of potential from a surface**

Univalent electrolyte at 10 mM concentration, \( \varepsilon = 80 \) and \( T = 20^\circ\text{C} \).

The surface charge density = 0.0158 C/m\(^2\).

Univalent electrolyte, \( \varepsilon = 80 \) and \( T = 20^\circ\text{C} \).
For the aqueous solution ($\varepsilon = 80$) the Debye length for ultrapure water is 190 nm and for 1 mM KCl 9.7 nm.
Surface charge has a number of effects

- A negative surface charge attracts cations to the membrane surface
  - increasing the conductance of the membrane to cations
  - enhancing the binding of cations to the membrane surface.

- The surface potential has a greater affect on the distributions of divalent ions than on distributions of monovalent ions (high Ca$^{2+}$ concentrations at the membrane surface).

- The surface potential changes the pH near the surface of the membrane.

- The surface potential may act to repel or attract other surfaces.
How much electrostatic energy is stored in double-layer?

A fully dissociating bilayer membrane can have one unit of charge per lipid headgroup

\[
\left| \frac{\sigma_q}{e} \right| = 0.7 \text{ nm}^{-2}
\]

A spherical vesicle of radius 10 µm then carries stored free energy

\[
\Delta G \approx 4\pi (10 \mu m)^2 \times \frac{0.7}{nm^2} k_B T \approx 10^9 k_B T
\]
An entropic force between charged surfaces in solution

- Each surface, together with its counterion cloud, is an electrically neutral object!

- The repulsion of between like-charged surfaces can only arise as an entropic effect.

- The diffused counterion clouds get squeezed; they then resist with an osmotic pressure.

\[
\frac{f}{\text{area}} = c_0 k_B T
\]

Opositely charged surfaces attract by counterion release

- As the surfaces approach they shed counterion pairs while preserving the system’s neutrality.

- The released ions gain entropy lowering the free energy and driving the surfaces together.
Origin of a dipole potential $\Psi_d$

$-P - N^+$ dipole
$P = O$ bonds of phosphate groups

Primery determinant $sn_2$-carbonyls

Little effect of $sn_1$-carbonyls

Dipoles of hydration water which are ordered and oriented at the membrane surface.

Orientational polarization of water

\[ \langle \cos(\theta) \rangle \]

Displacement from bilayer center (Å)
Sketch of typical contributions to the electrostatic potential of a bilayer membrane made of zwitterionic lipids.
The dipole potential is a major factor in determining the ionic permeability of the lipid bilayer.

Dipole potential modifies the electric field inside the membrane, producing a virtual positive charge in the apolar bilayer center.

Lipid membranes exhibit a substantial (up to six orders of magnitude) difference in the penetration rates between positively and negatively charge hydrophobic ions.
The dipole potential \[ \Delta \psi = \frac{D}{\varepsilon \varepsilon_0} \] D is the surface dipole density in C/m.

A dipole moment of phospholipid = 1.5 Debyes - 5 x 10^{-30} C/m
Surface area per lipid = 60 Å²
A surface dipole density = 8.34 x 10^{-12} C/m
A dipole potential = 1000/\varepsilon mV

The dipole potential is not a significant component of the membrane potential because the dipoles on opposite surfaces of the membrane are oriented in opposite directions and tend to cancel each other.
Schematics for measuring surface and dipole potentials in lipid monolayers

\[ \Psi_m = \Psi_w \]
Dipole potential functions:

- Effects the membrane permeability for lipophilic ions and drugs.
- Modulates the binding of peptides and biologically active molecules to cell plasma membranes.
- Directs the insertion and filling of peptides in the membranes.
- Modulates membrane enzymes activities.
Compounds modifying the bilayer dipole potential

**Phloretin** – strongly reduces the $\Psi_d$ value

**Sterols including cholesterol** - increases $\Psi_d$ value

**6-ketocholestanol** – strongly increases $\Psi_d$ value

**The removal of sn-carbonyls, which is realized in ether phospholipids decreases $\Psi_d$**
Fluorescence probes for membrane dipole potential monitoring

F4N1  BPPZ  di-8-ANEPPS
Electrochromism (Stark effect)

The shifts of absorption and emission bands caused by the interaction of an electric field with the ground-state and excited state dipole moments of the chromophore.

The spectral shift, $h\Delta \nu_{\text{obs}}$, is proportional to the electric field $E$, and the change of dipole moment associated with the spectroscopic transition $\Delta \mu$,

$$h\Delta \nu_{\text{obs}} = -\left(\frac{1}{\varepsilon_{\text{ef}}}\right)\Delta \mu \cdot |E| \cos \theta$$

$\theta$ – the angle between $\Delta \mu$ and $E$ vectors, and $\varepsilon_{\text{ef}}$ is a microscopic analog of dielectric constant, which accounts for dielectric screening.

Sensitivity depends on:

- change of the probe dipole moment on electronic excitation
- low-polar environment
- probe should be oriented parallel ($\cos \theta = 1$) or antiparallel ($\cos \theta = 1$) to the electric field.

Exited-state charge transfer in styryl dyes
Summary
Examples of potential profiles

- Negative surface charge on both sides.
- A large surface charge on one side.
- Surface charge on one side, dipole potential and transmembrane potential.
Biology and surface electrostatics
The electrostatic profile across the plasma membrane and at interfaces.
Long-range van der Waals attraction

Potential energy of interaction

\[ \Phi = \frac{Aa}{12s_0} \]

Hamaker constant, \( A \), proportional to intermolecular attraction constant

Attractive force of interaction

\[ F_{att} = \frac{d\Phi}{ds_0} = \frac{Aa}{12s_0^2} \]
Electrostatic repulsion retards aggregation

van der Waals attraction

electrostatic repulsion
Electrostatic repulsion depends on ....

1. The Stern potential, $\psi_\delta$
2. The thickness of the diffuse double layer, $\kappa^1$
Forces between particles (surfaces)

**Attractive forces**
- van der Waals forces

**Repulsive forces**
- electrostatic forces (aqueous or high $\varepsilon$ media)

**DLVO Theory**

When $\left(\frac{\Phi}{kT}\right) \geq 10$, $\rightarrow$ “stability”
Controlling DLVO interactions

\[
\frac{\Phi}{kT} = \frac{0.064 \pi a N_{Av} C}{\kappa^2} \tanh^2 \left( \frac{ze \psi_\delta}{4kT} \right) e^{-\kappa s} - \frac{A_{12} a}{12kT_s}
\]

\[
\kappa = \sqrt{\frac{2e^2 z^2 n_\infty}{\varepsilon \varepsilon_0 kT}} = \sqrt{\frac{2F^2 z^2 C}{\varepsilon \varepsilon_0 RT}} \approx 0.329 z\sqrt{C} \text{ Å}^{-1}
\]

**water, 25°C**
Controlling electrolyte valence and concentration

Jar testing of $\text{As}_2\text{S}_3$ sol with 1-1 electrolyte.

Schulze-Hardy Rule:

$$\text{CCC} \propto \frac{1}{z^6}$$

For $z = 2$, divide all $C$’s by 64;
for $z = 3$, divide all $C$’s by 729
Controlling effective Stern potential, $\psi_\delta$

Examples of surface potential control:

For oxides

$$\psi_0 = 59.2 \left( pH_{pzc} - pH \right) \text{ mV}$$

Specific ion adsorption

$$\psi_\delta = \psi_{\delta 0} + \frac{z_i e \Gamma_i}{\varepsilon \varepsilon_0 k}$$

$|\psi_\delta| < 25 \text{ mV generally inadequate for stability.}$