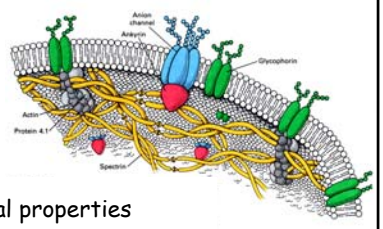
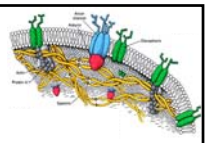
Biological Membranes



- Structure
- Function
- Composition
- Physicochemical properties
- Self-assembly
- Molecular models

Lipid Membranes



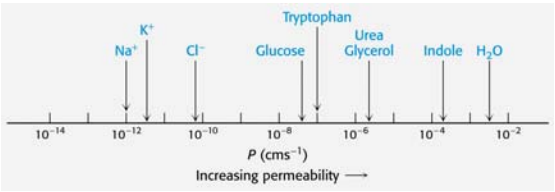
- Receptors, detecting the signals from outside:
  - Light
  - Odorant
  - Taste
  - Chemicals
    - Hormones
    - Neurotransmitters
    - Drugs
- Channels, gates and pumps
- Electric/chemical potential
  - Neurophysiology
  - Energy
- Energy transduction:
  - Photosynthesis
  - Oxidative phosphorylation

A highly selective permeability barrier

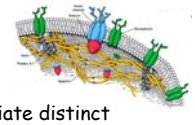
Internal membranes for organelles

Bilayer Permeability

- Low permeability to charged and polar substances
- Water is an exception: small size, lack of charge, and its high concentration
- Shedding solvation shells for ions is very unlikely



Common Features of Biological Membranes

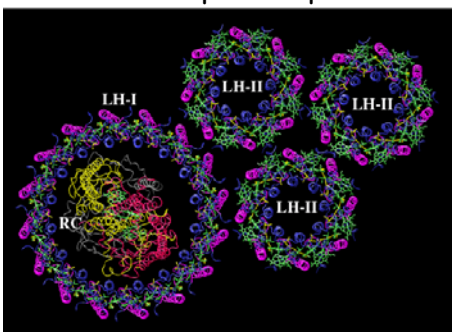


- Sheet-like structure
- TWO-molecule thick (60-100Å)
- Lipids, Proteins, and carbohydrates
- Lipids form the barrier. Proteins mediate distinct functions.
- Non-covalent assemblies (self-assembly, protein-lipid interaction)
- Asymmetric (always)
- Fluid structures: 2-dimensional solution of oriented lipids and proteins
- Electrically polarized (inside negative ~-60mV)
- Spontaneously forming in water
- Protein/lipid ratio = 1/4 – 4/1
- Carbohydrate moieties are always outside the cell

Protein/Lipid ratio

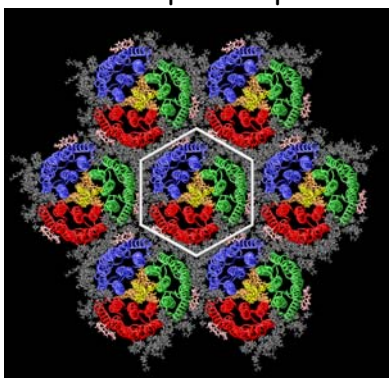
- Pure lipid: insulation (neuronal cells)
- Other membranes: on average 50%
- Energy transduction membranes (75%)
  Internal membranes of mitocondria and chloroplast
  Purple membrane of halobacteria
- Different functions = different protein composition

Protein / Lipid Composition



Light harvesting complex of purple bacteria

Protein / Lipid Composition

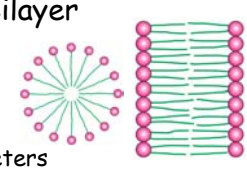


The purple membrane of halobacteria

General features of Lipids

- Small molecules
- Amphipathic (amphiphilic) Hydrophobic/hydrophilic moieties
- Spontaneously form vesicles, micelles, and **bilayers** in aqueous solution

Micelle / Bilayer



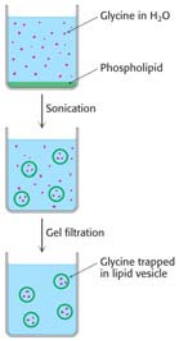
- Fatty acids (one tail)
- Phospholipids (two tails)
- Micelle max 20 nm
- Bilayer up to millimeters
- Self-assembly process
- Hydrophobic interaction is the driving force (also in protein folding and in DNA stacking)

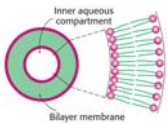
- Extensive; tendency to close on themselves; self-sealing (a hole is unfavorable)

Vesicles

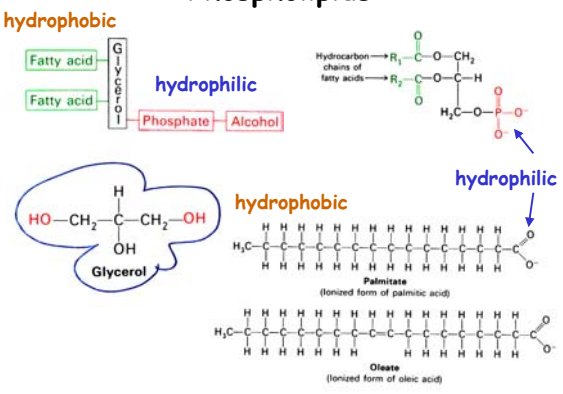
- Phospholipids
- Sonicating (~50nm)
- Evaporation (1 micron)

- Experimental tool for studying membrane proteins
- Clinical use (drug delivery)





Phospholipids



Structure of fatty acids

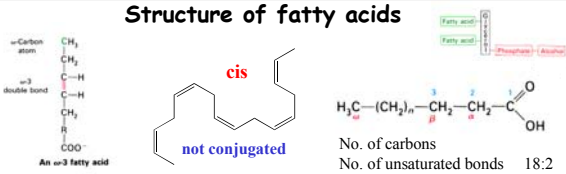
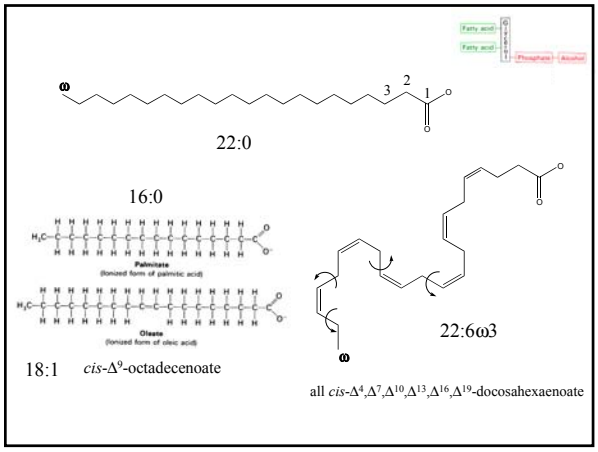
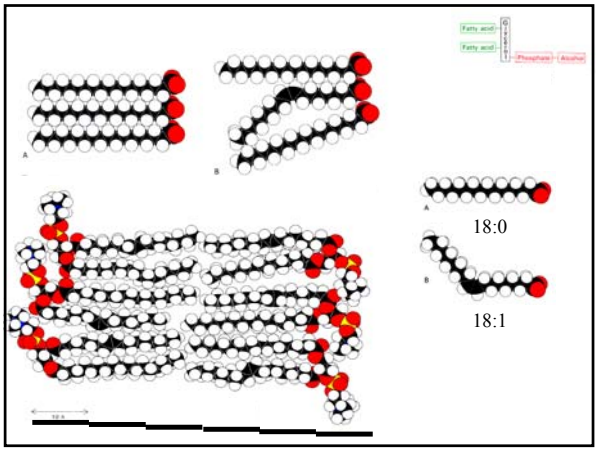


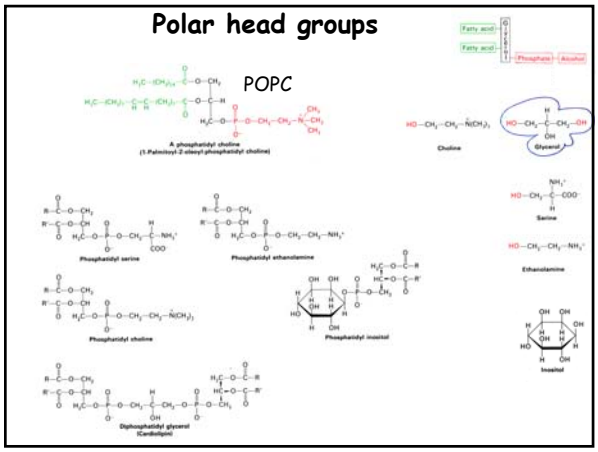
TABLE 12.1 Some naturally occurring fatty acids in animals

Number of carbons	Number of double bonds	Common name	Systematic name	Formula
12	0	Laurate	n-Dodecanoate	$CH_3(CH_2)_{10}COO^-$
14	0	Myristate	n-Tetradecanoate	$CH_3(CH_2)_{12}COO^-$
16	0	Palmitate	n-Hexadecanoate	$CH_3(CH_2)_{14}COO^-$
18	0	Stearate	n-Octadecanoate	$CH_3(CH_2)_{16}COO^-$
20	0	Arachidate	n-Eicosanoate	$CH_3(CH_2)_{18}COO^-$
22	0	Behenate	n-Docosanoate	$CH_3(CH_2)_{20}COO^-$
24	0	Lignocerate	n-Tetracosanoate	$CH_3(CH_2)_{22}COO^-$
16	1	Palmitoleate	cis-Δ^9-Hexadecenoate	$CH_3(CH_2)_5CH{=}CH(CH_2)_7COO^-$
18	1	Oleate	cis-Δ^9-Octadecenoate	$CH_3(CH_2)_7CH{=}CH(CH_2)_7COO^-$
18	2	Linoleate	cis,cis-Δ^9,Δ^{12}-Octadecadienoate	$CH_3(CH_2)_4(CH{=}CHCH_2)_2(CH_2)_6COO^-$
18	3	Linolenate	all-cis-$\Delta^9,\Delta^{12},\Delta^{15}$-Octadecatrienoate	$CH_3CH_2(CH{=}CHCH_2)_3(CH_2)_6COO^-$
20	4	Arachidonate	all-cis-$\Delta^5,\Delta^8,\Delta^{11},\Delta^{14}$-Eicosatetraenoate	$CH_3(CH_2)_4(CH{=}CHCH_2)_4(CH_2)_2COO^-$





Polar head groups

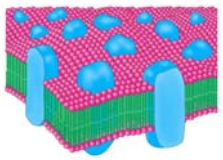


Diffusion in Membrane

Einstein relation for diffusion:

$$6Dt = \left\langle \left| \mathbf{r}_i(t) - \mathbf{r}_i(0) \right|^2 \right\rangle$$

$$\left| \mathbf{r}_i(t) - \mathbf{r}_i(0) \right| = S$$

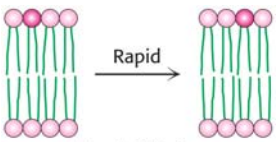


$6Dt = \langle S^2 \rangle$ in a 3 dimensional space

$4Dt = \langle S^2 \rangle$ in 2 dimensions (in-plane diffusion in a membrane)

$2Dt = \langle S^2 \rangle$ in 1 dimension

Lipid Diffusion in Membrane



Lateral diffusion

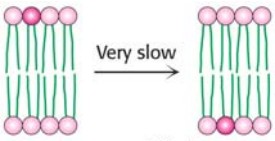
D = 1 μm².s⁻¹

50 Å in ~ 2.5 x 10⁻⁵ s

~9 orders of magnitude difference

D_{lip} = 10⁻⁸ cm².s⁻¹

D_{wat} = 2.5 x 10⁻⁵ cm².s⁻¹

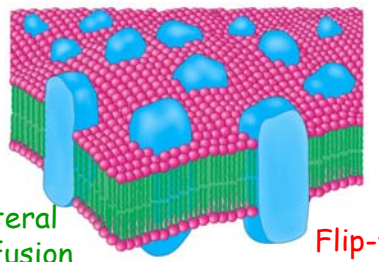


Tranverse diffusion (flip-flop)

Once in several hours!

(10⁴ s)

Fluid Mosaic Model of Membrane

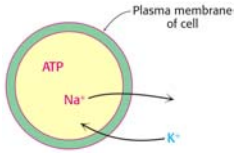


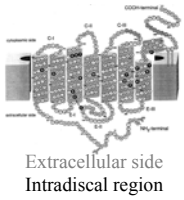
Lateral Diffusion Allowed

Flip-flap Forbidden

Ensuring the conservation of membrane asymmetric structure

Importance of Asymmetry





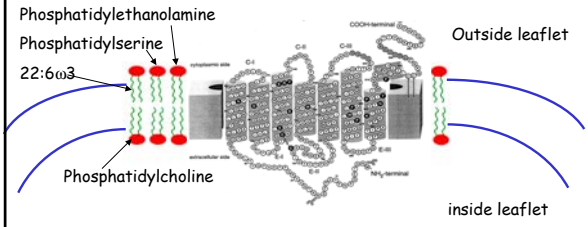
Apart from some passive transport mechanisms, all membrane proteins function in a directed way, and their correct insertion in the cell membrane is essential for their biological function.

Highly asymmetric and inhomogeneous lipid composition of membrane

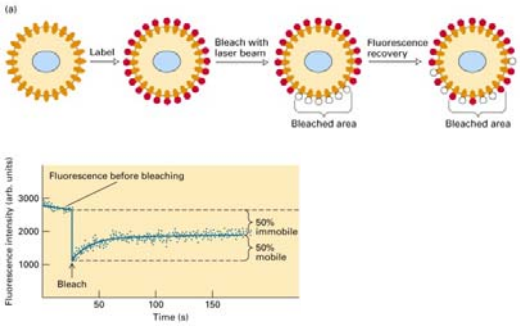
Polyunsaturated lipids → 22:6ω3: 47% in membrane

PC : PE : PS

45 : 42 : 14

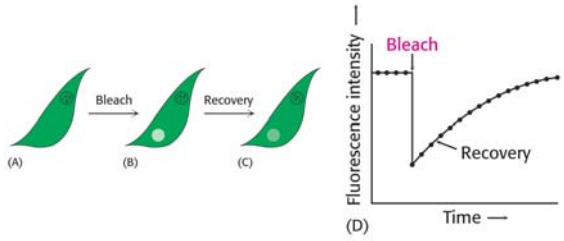


Fluorescence recovery after photobleaching (FRAP)



Lipid Diffusion in Membrane

- FRAP - fluorescent recovery after photobleaching (Albert's movie)



"FRAP experiment movie"
The Molecular Biology of the Cell

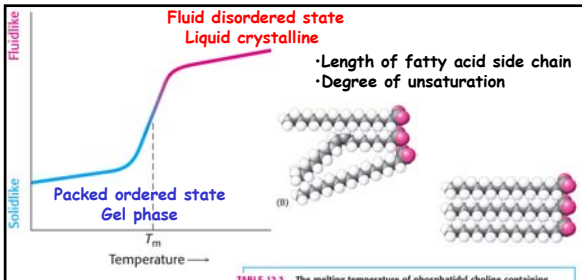


TABLE 12.3 The melting temperature of phosphatidyl choline containing different pairs of identical fatty acid chains

Number of carbons	Number of double bonds	Fatty acid: Common name	Fatty acid: Systematic name	T_m (°C)
22	0	Behenate	n-Docosanoate	75
18	0	Stearate	n-Octadecanoate	58
16	0	Palmitate	n-Hexadecanoate	41
14	0	Myristate	n-Tetradecanoate	24
18	1	Oleate	cis-Δ^9-Octadecenoate	−22

E. coli:

@ 42°C sat/unsat = 1.6

@ 27°C sat/unsat = 1.0

A Brief Introduction to Molecular Dynamics Simulations

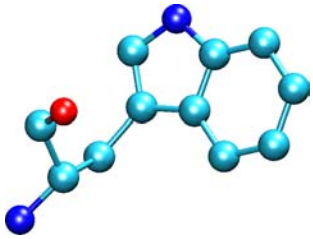
Macroscopic properties are often determined by atomic-level behavior.

Quantitative and/or qualitative information about macroscopic behavior of macromolecules can be obtained from simulation of a system at atomistic level.

Molecular dynamics simulations calculate the motion of the atoms in a molecular assembly using Newtonian dynamics to determine the net force and acceleration experienced by each atom. Each atom i at position r_i, is treated as a point with a mass m_i and a fixed charge q_i.

What is the Force Field?

In molecular dynamics a molecule is described as a series of charged points (atoms) linked by springs (bonds).

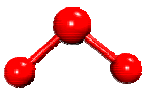


To describe the time evolution of bond lengths, bond angles and torsions, also the non-bonding van der Waals and elecrostatic interactions between atoms, one uses a force field.
The force field is a collection of equations and associated constants designed to reproduce molecular geometry and selected properties of tested structures.

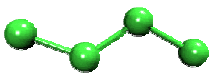
Energy Terms Described in the CHARMm Force Field

Bond

Angle



Dihedral



Improper

Energy Functions

$$U(\vec{R}) = \underbrace{\sum_{bonds} k_i^{bond}(r_i - r_0)^2}_{U_{bond}} + \underbrace{\sum_{angles} k_i^{angle}(\theta_i - \theta_0)^2}_{U_{angle}} + \underbrace{\sum_{dihedrals} k_i^{dihe}[1 + \cos(n_i\phi_i + \delta_i)]}_{U_{dihedral}} + \underbrace{\sum_i \sum_{j \neq i} 4\epsilon_{ij}\left[\left(\frac{\sigma_{ij}}{r_{ij}}\right)^{12} - \left(\frac{\sigma_{ij}}{r_{ij}}\right)^{6}\right] + \sum_i \sum_{j \neq i} \frac{q_i q_j}{\epsilon r_{ij}}}_{U_{nonbond}}$$

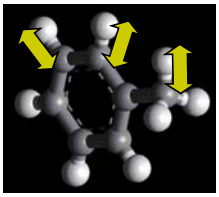
U_{bond} = oscillations about the equilibrium bond length
U_{angle} = oscillations of 3 atoms about an equilibrium bond angle
$U_{dihedral}$ = torsional rotation of 4 atoms about a central bond
$U_{nonbond}$ = non-bonded energy terms (electrostatics and Lenard-Jones)

Time Scale of Biological Events

Motion	Time Scale (sec)
Bond stretching	10^{-14} to 10^{-13}
Elastic vibrations	10^{-12} to 10^{-11}
Rotations of surface sidechains	10^{-11} to 10^{-10}
Hinge bending	10^{-11} to 10^{-7}
Rotation of buried side chains	10^{-4} to 1 sec
Allosteric transistions	10^{-5} to 1 sec
Local denaturations	10^{-5} to 10 sec

The 1 fs Time Step Limit

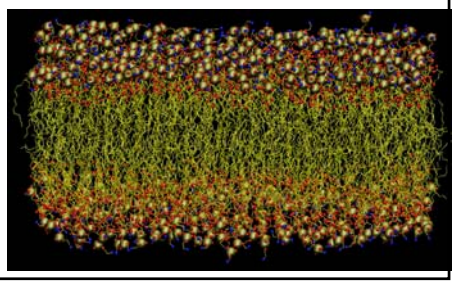
- Dynamics simulations are limited by the highest frequency vibration.
- Ideally the timestep should be 1/10 of the period of the highest frequency vibration.
- X-H bond stretching (~10^{-14} s) is the fastest mode.



Technical difficulties in Simulations of Biological Membranes

- Time scale
- Inhomogeneity of biological membranes

60 x 60 Å
Pure POPE
5 ns
~100,000
atoms



Coarse grain modeling of lipids

by: J. Siewert-Jan Marrink and
Prof. Dr. Alan E. Mark, University of Groningen, The Netherlands

150 particles

9 particles!