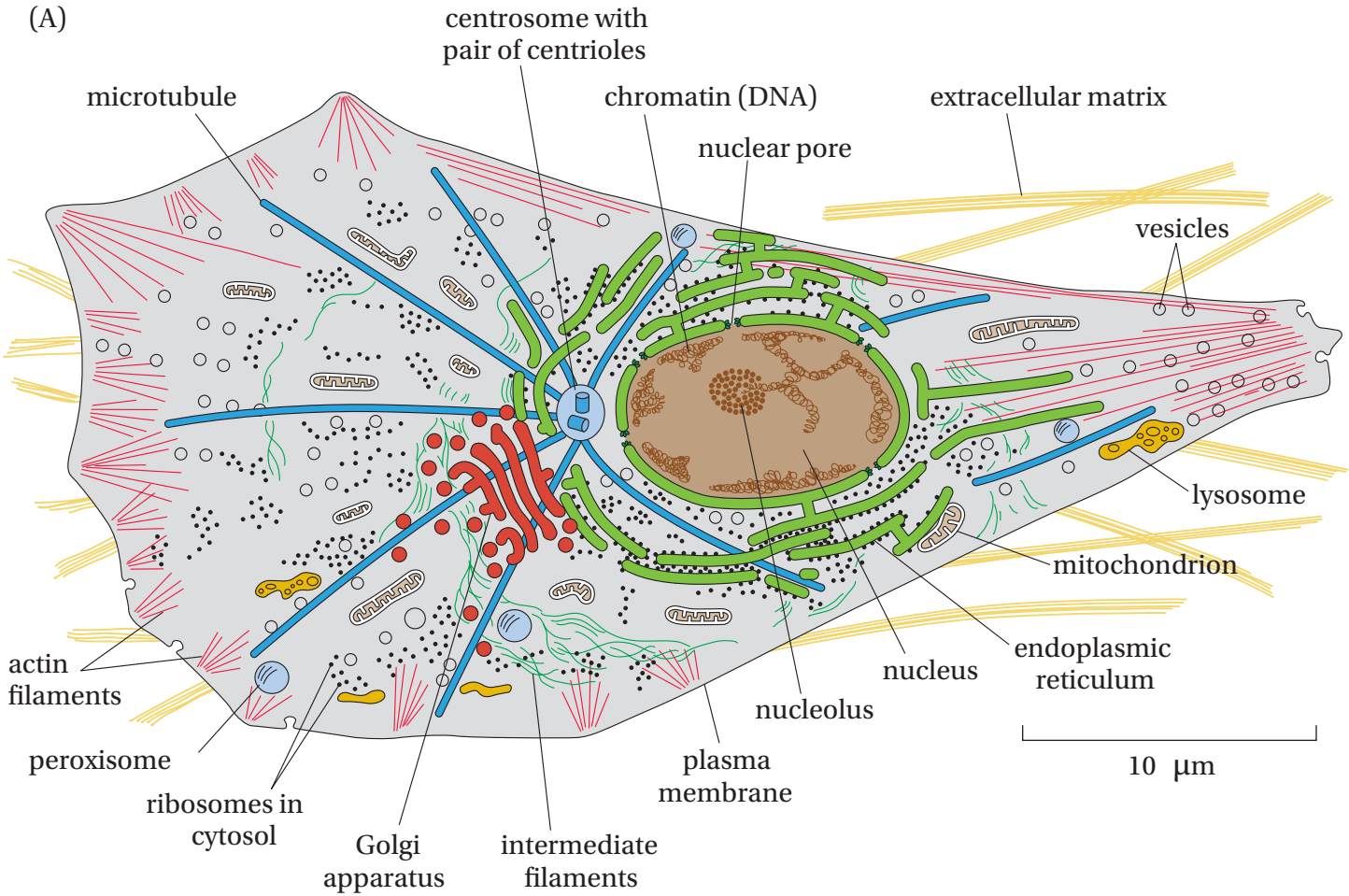
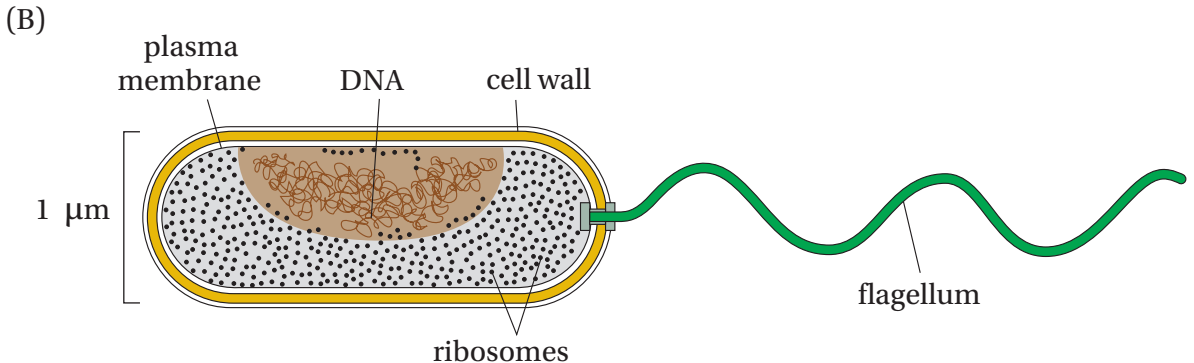
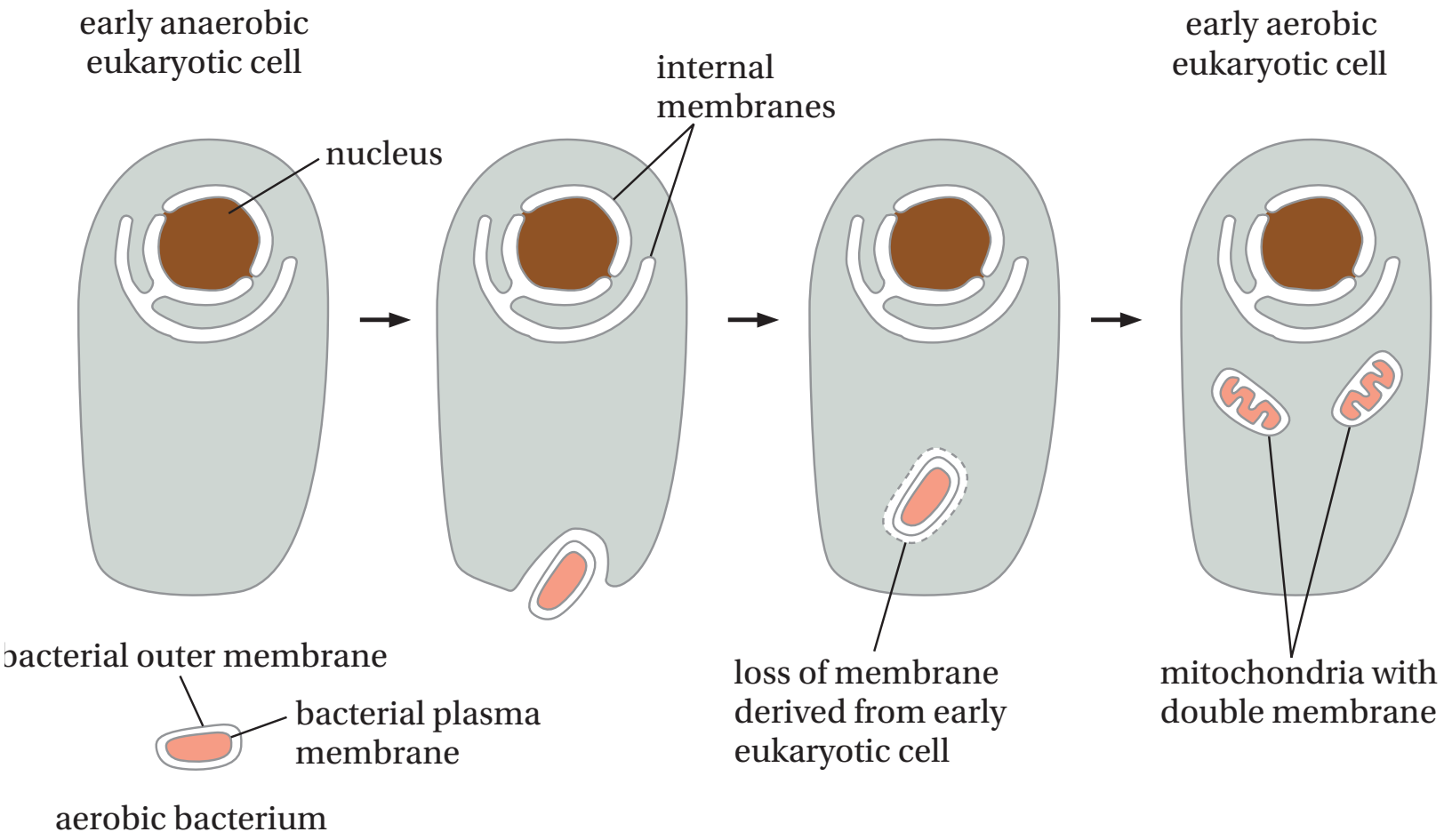
Membrane Proteins

Anatomies of two types of biological cells



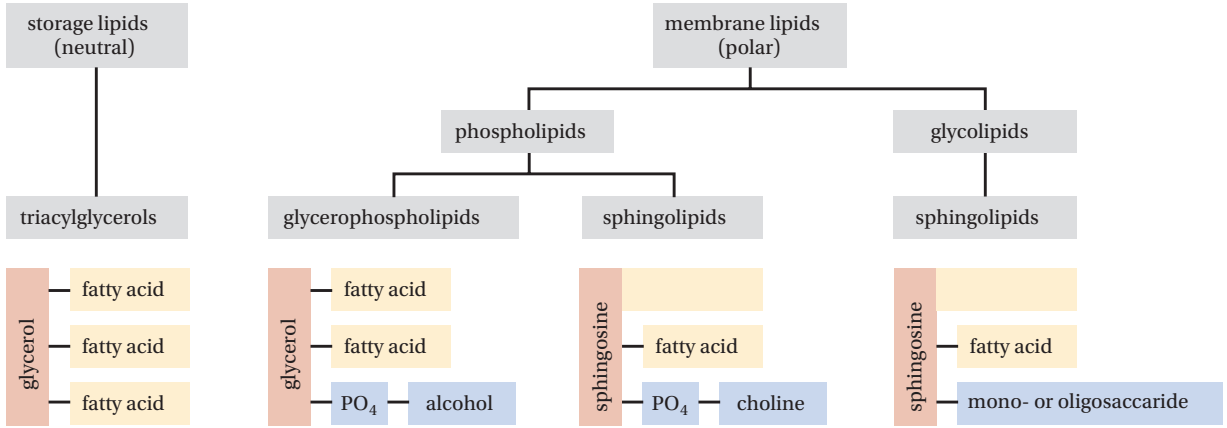


Evolution of mitochondria

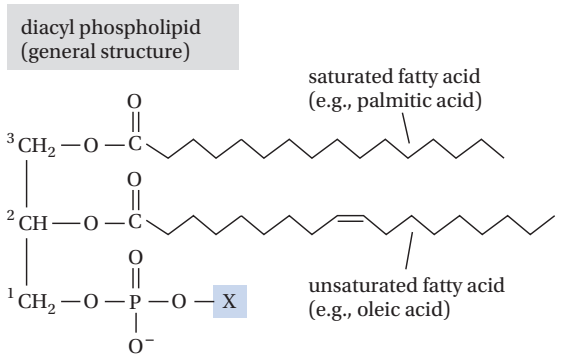


Membrane Lipids Form Bilayers in Water

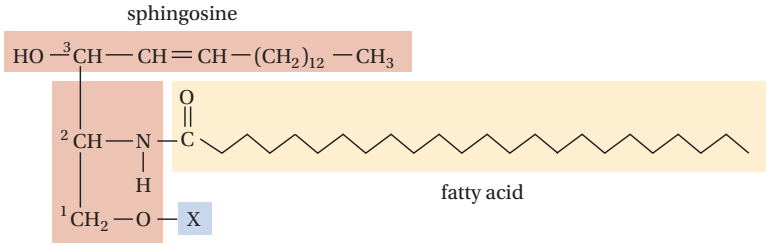
(A)



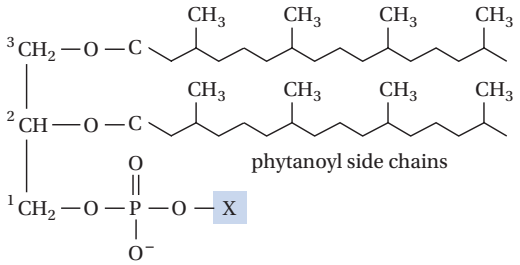
(B)



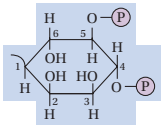
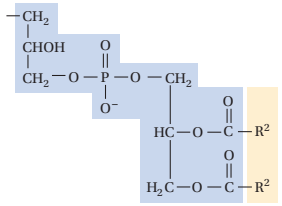
(C)



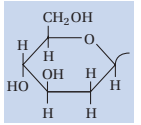
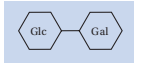
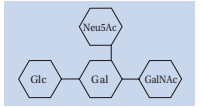
(D)



(E)

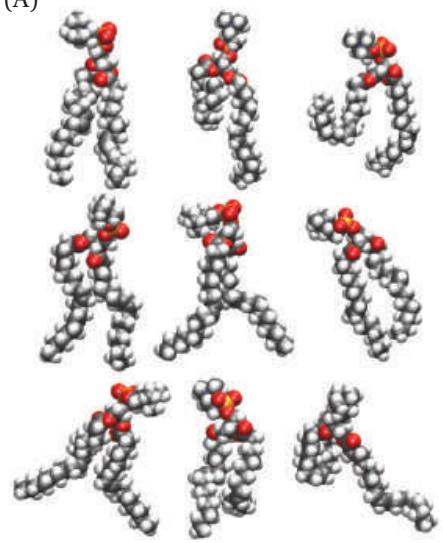
name of glycerophospholipid	name of X	formula of X	net charge (at pH 7)
phosphatidic acid	–	—H	–1
phosphatidylethanolamine	ethanolamine	$-CH_2-CH_2-\overset{+}{N}H_3$	0
phosphatidylcholine	choline	$-CH_2-CH_2-\overset{+}{N}(CH_3)_3$	0
phosphatidylserine	serine	$-CH_2-CH(COO^-)-\overset{+}{N}H_3$	–1
phosphatidylglycerol	glycerol	$-CH_2-CH(OH)-CH_2-OH$	–1
phosphatidylinositol 4,5-bisphosphate	4,5-bisphosphate		–5
cardiolipin	phosphatidylglycerol		–2

(F)

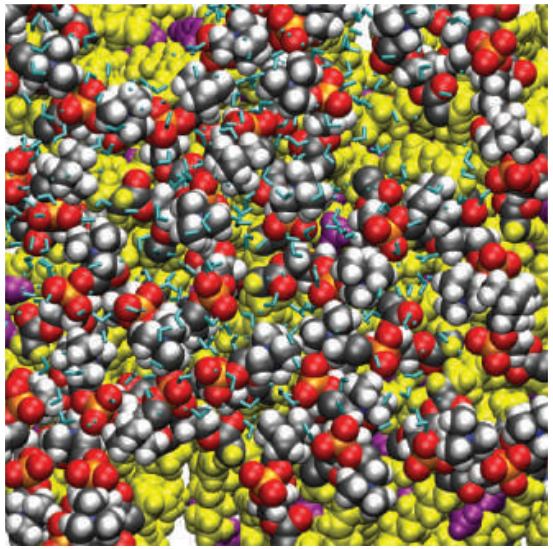
name of sphingolipid	name of X	formula of X
ceramide	–	—H
sphingomyelin	phosphocholine	$-O-P(=O)(O^-)-O-CH_2-CH_2-\overset{+}{N}(CH_3)_3$
neutral glycolipids glucosylcerebroside	glucose	
lactosylceramide	di-, tri-, or tetrasaccharide	
ganglioside GM2	complex oligosaccharide	

Molecular dynamics

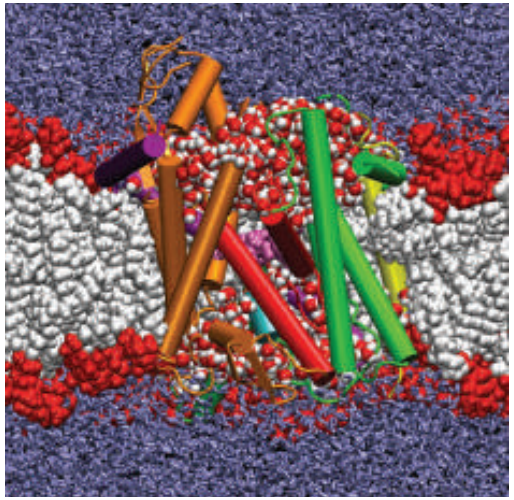
(A)



(B)



(A)



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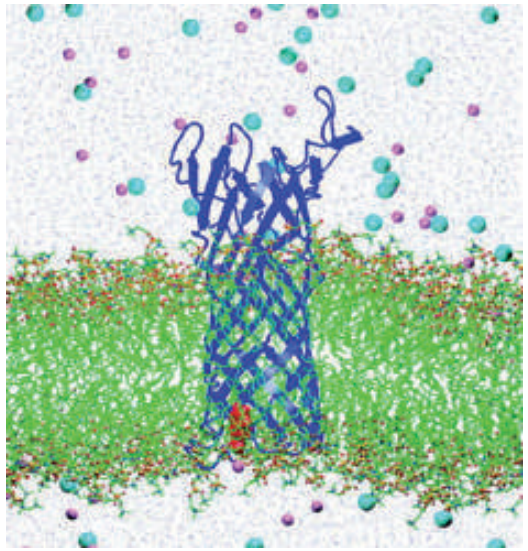
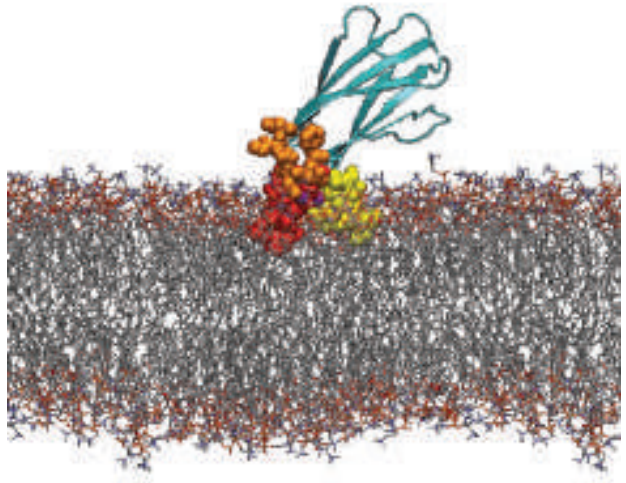


(C)

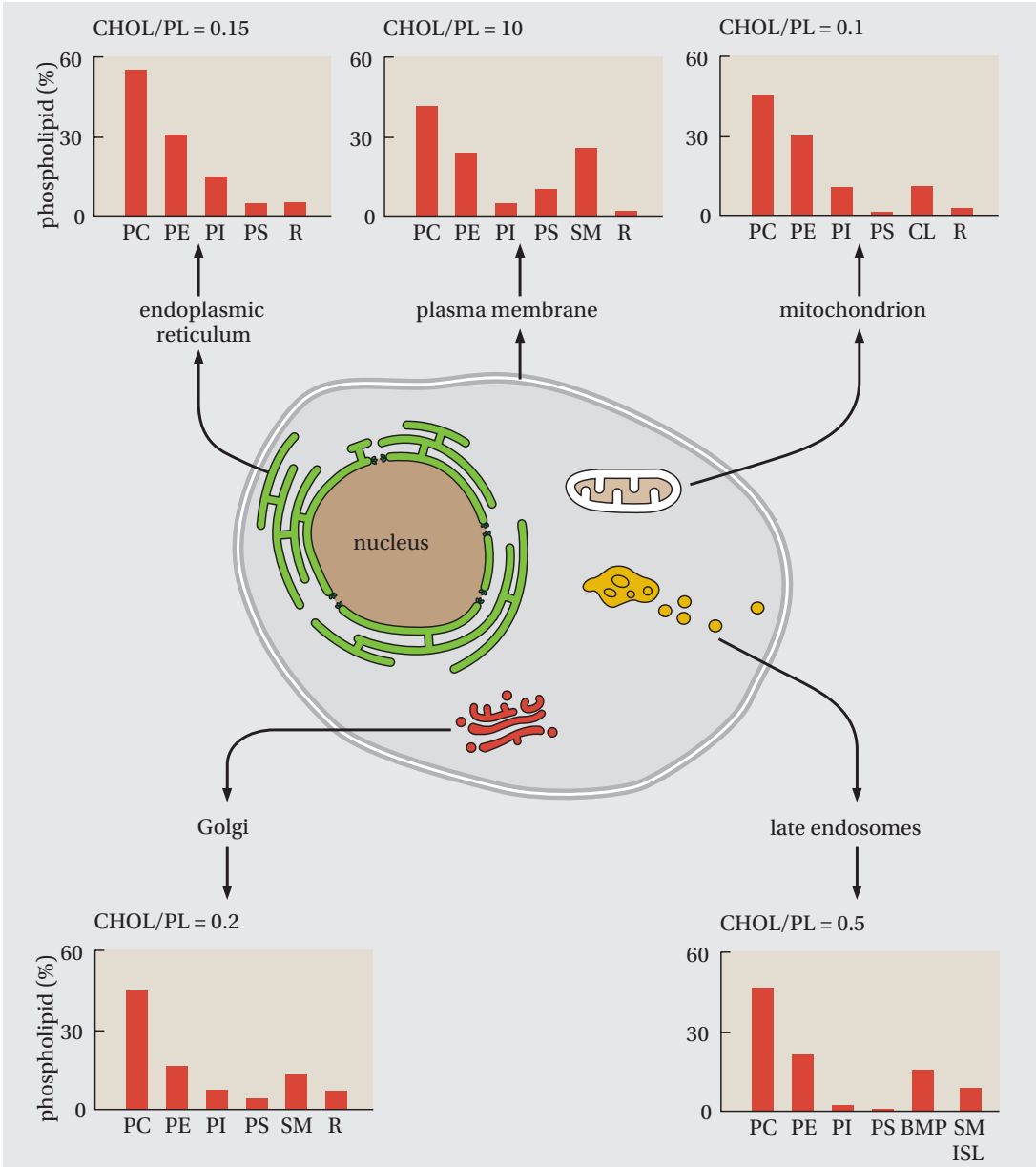


Composition of the membranes

Composition of Membranes (% by weight)			
Membrane	**Protein**	**Lipid**	**Carbohydrate**
Myelin	20	75	5
Red blood cell ghosts	49	43	8
Plasma membranes			
Liver cells	58	42	(5–10)[a]
Ehrlich ascites	67	33	(5–10)[a]
Amoeba *Proteus*	25	32	15
Mitochondrion			
Outer membrane	52	48	(2–4)[a]
Inner membrane	76	24	(1–2)[a]
Bacteria (Gram-positive)	75	25	(10)[a]

[a]Carbohydrate is due to glycosylated proteins and lipids.

Based on data provided in Guidott G [1972] *Arch Intern Med* 129: 194–301.



Curvature model of membrane vesicles

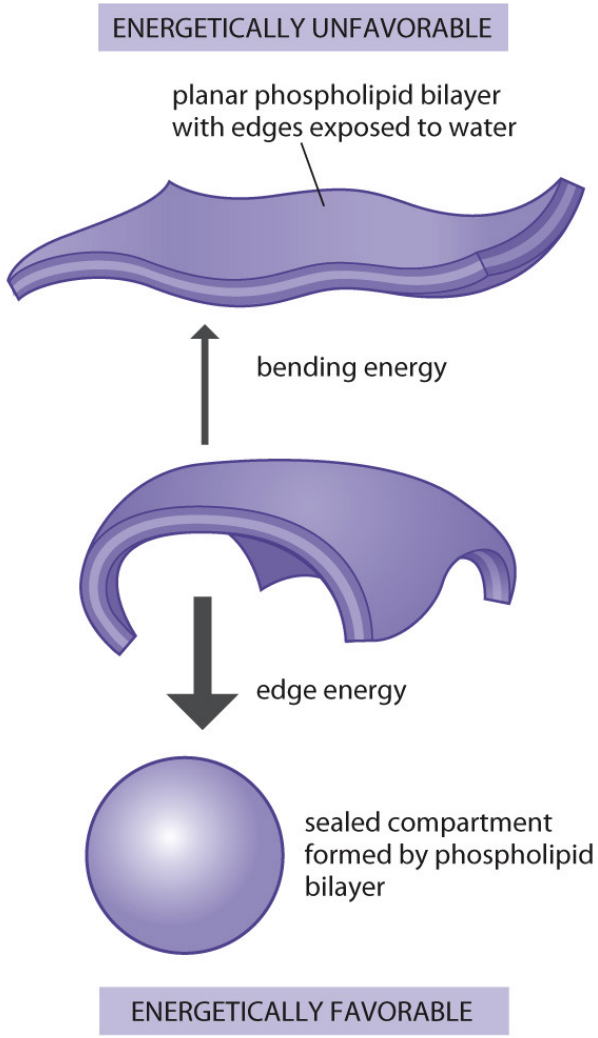
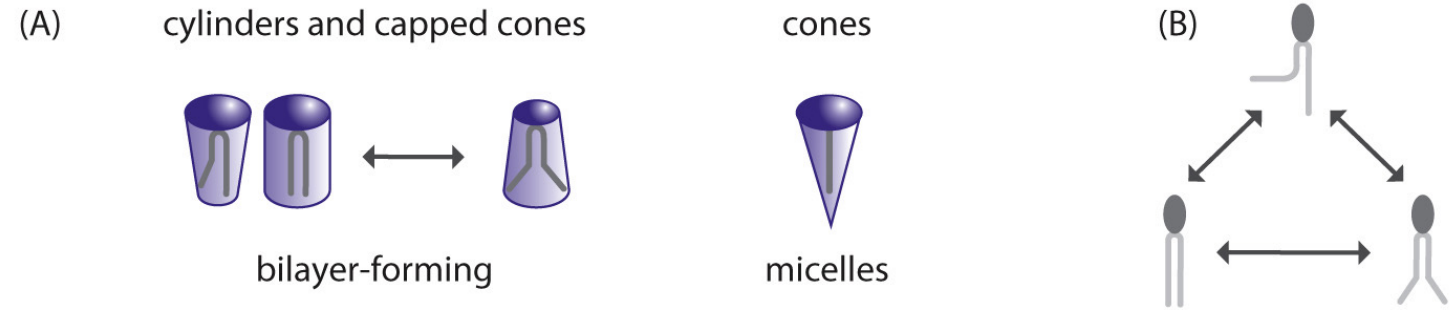


Figure 2.17 Cell Membranes (© Garland Science 2016)



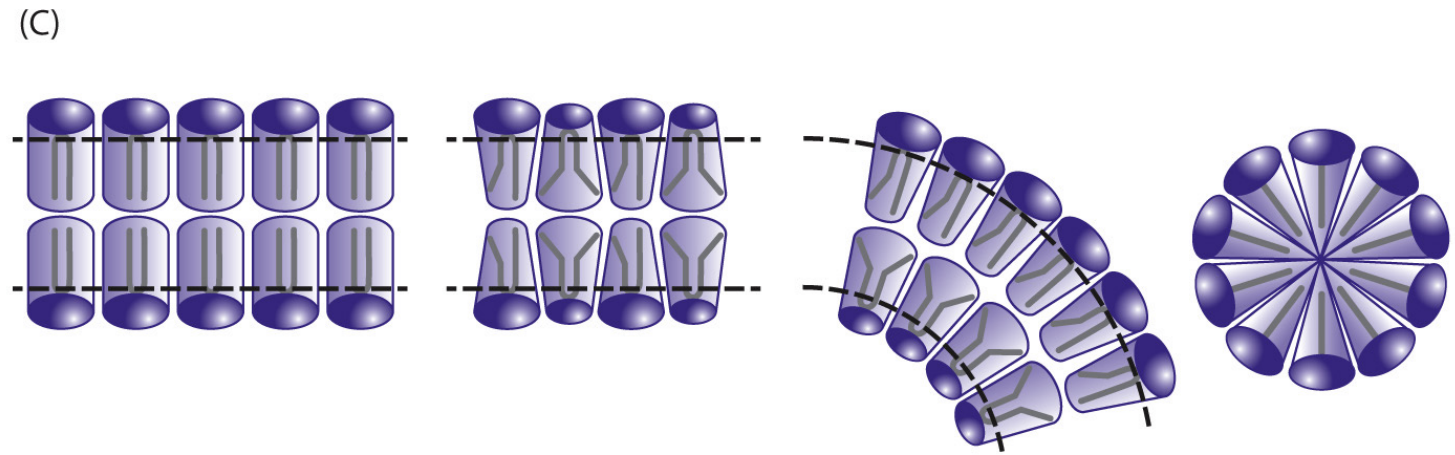
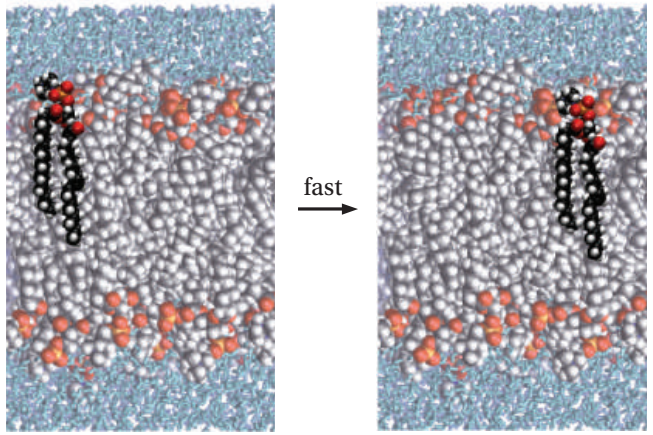


Figure 2.20 Cell Membranes (© Garland Science 2016)

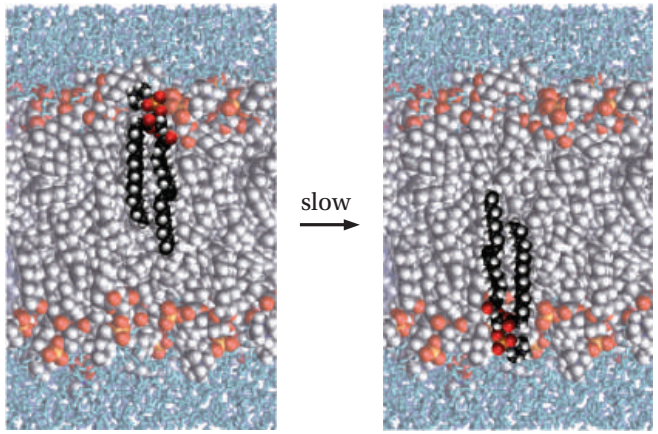
	P	volume	conformation	curvature	lipid
	>1	l	reverse cone	negative	PE
	=1	l	cylinder	zero	PC
	<1	l	cone	positive	lyso PC

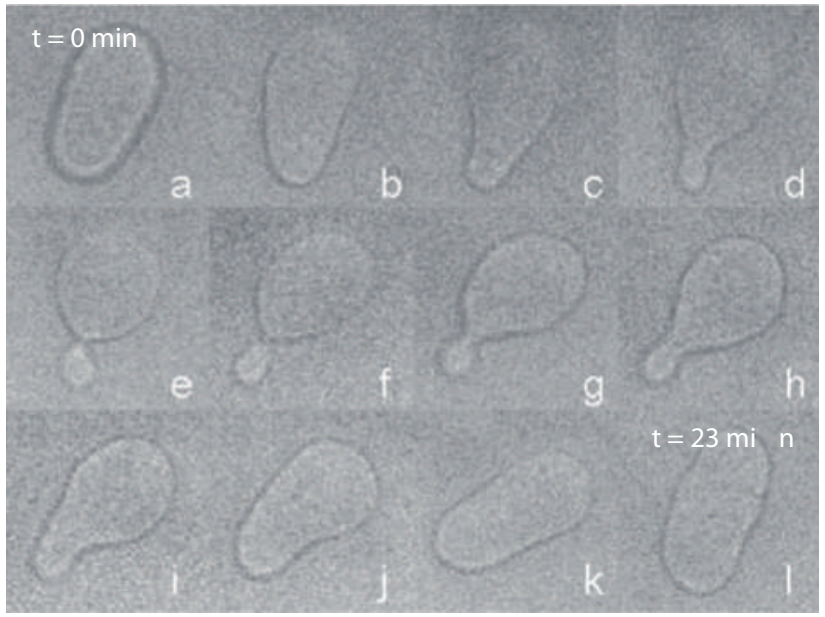
Lipid diffusion

lateral diffusion



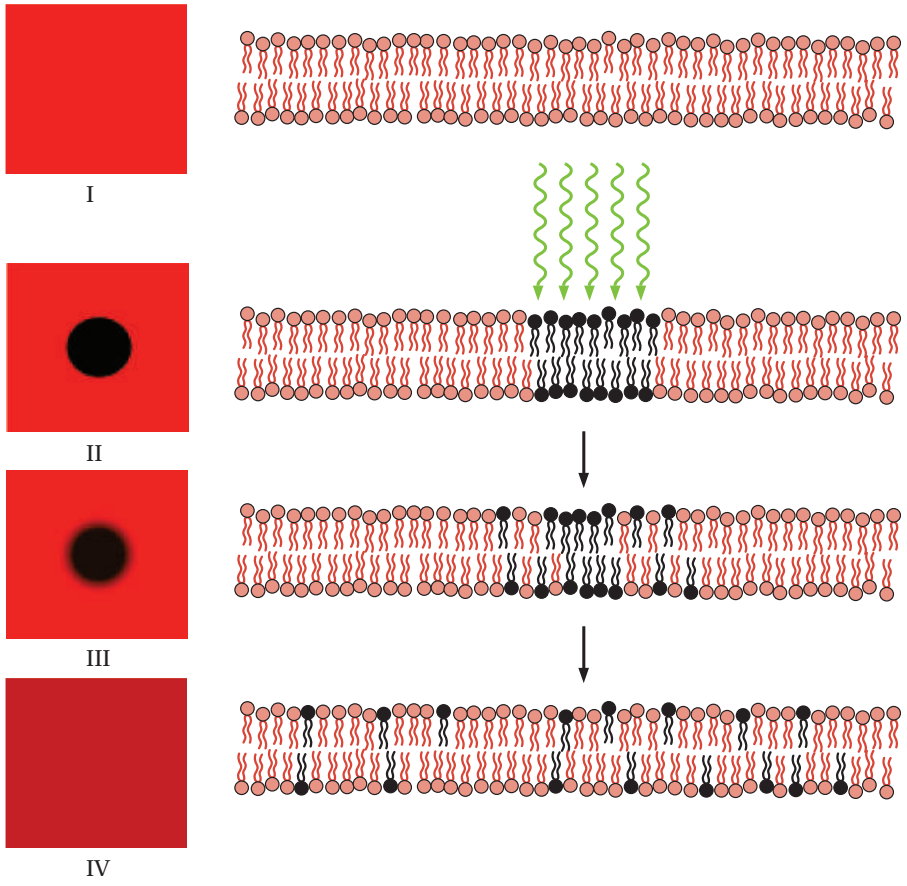
transverse diffusion (flip-flop)

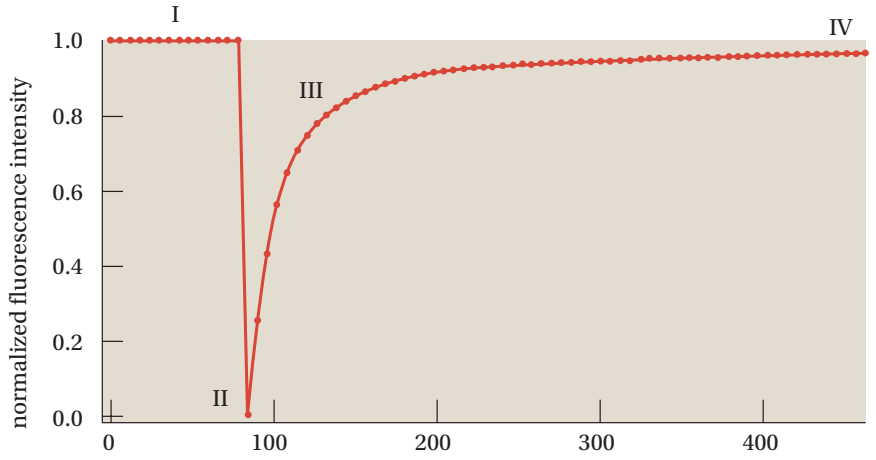




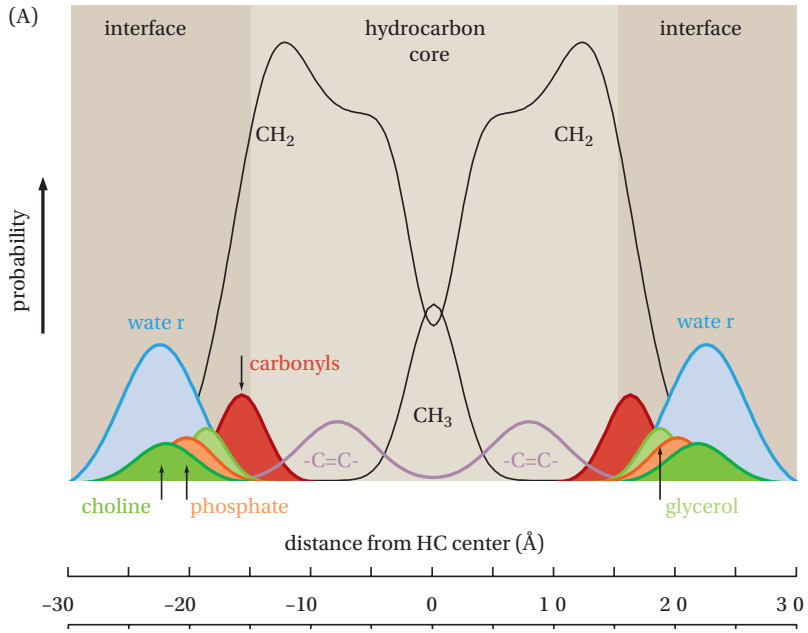
top view

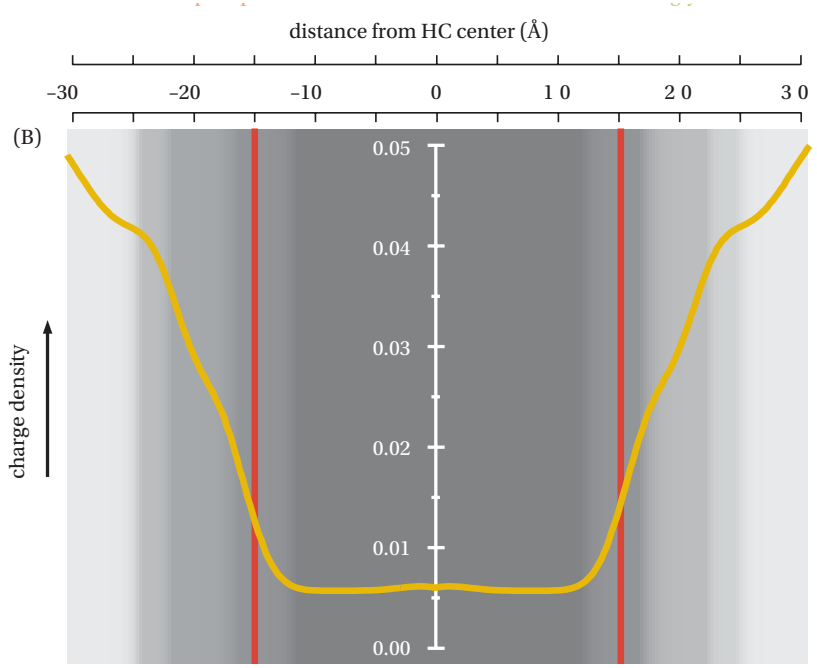
side view

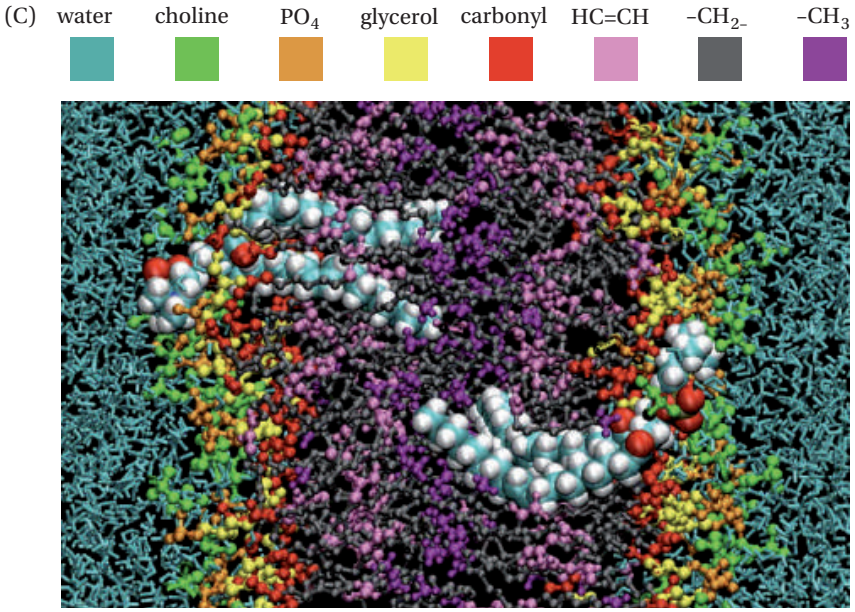




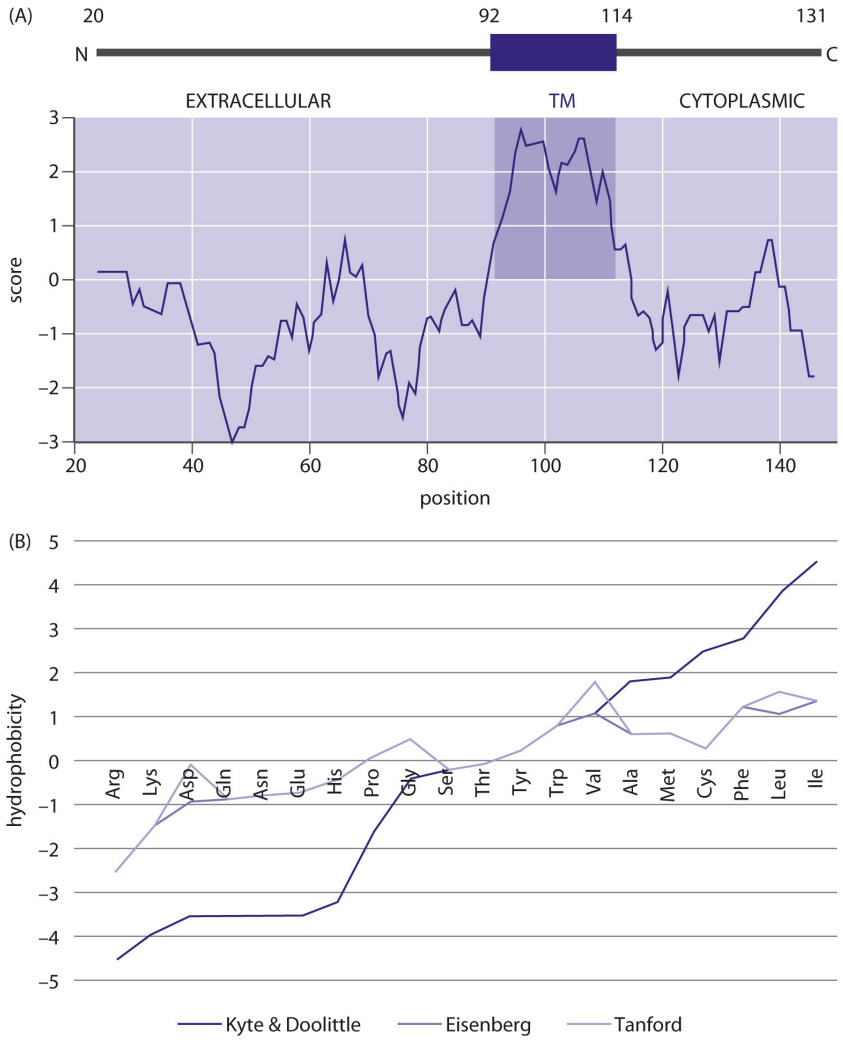
Structure of a DOPC bilayer







Predicting transmembrane helices



(C)

hydrophobicity amino acid values

Kyte and Doolittle	Eisenberg	Tanford
Ala: 1.800	Ala: 0.620	Ala: 0.620
Arg: -4.500	Arg: -2.530	Arg: -2.530
Asn: -3.500	Asn: -0.780	Asn: -0.780
Asp: -3.500	Asp: -0.900	Asp: -0.090
Cys: 2.500	Cys: 0.290	Cys: 0.290
Gln: -3.500	Gln: -0.850	Gln: -0.850
Glu: -3.500	Glu: -0.740	Glu: -0.740
Gly: -0.400	Gly: 0.480	Gly: 0.480
His: -3.200	His: -0.400	His: -0.400
Ile: 4.500	Ile: 1.380	Ile: 1.380
Leu: 3.800	Leu: 1.060	Leu: 1.530
Lys: -3.900	Lys: -1.500	Lys: -1.500
Met: 1.900	Met: 0.640	Met: 0.640
Phe: 2.800	Phe: 1.190	Phe: 1.190
Pro: -1.600	Pro: 0.120	Pro: 0.120
Ser: -0.800	Ser: -0.180	Ser: -0.180
Thr: -0.700	Thr: -0.050	Thr: -0.050
Trp: -0.900	Trp: 0.810	Trp: 0.810
Tyr: -1.300	Tyr: 0.260	Tyr: 0.260
Val: 4.200	Val: 1.080	Val: 1.800

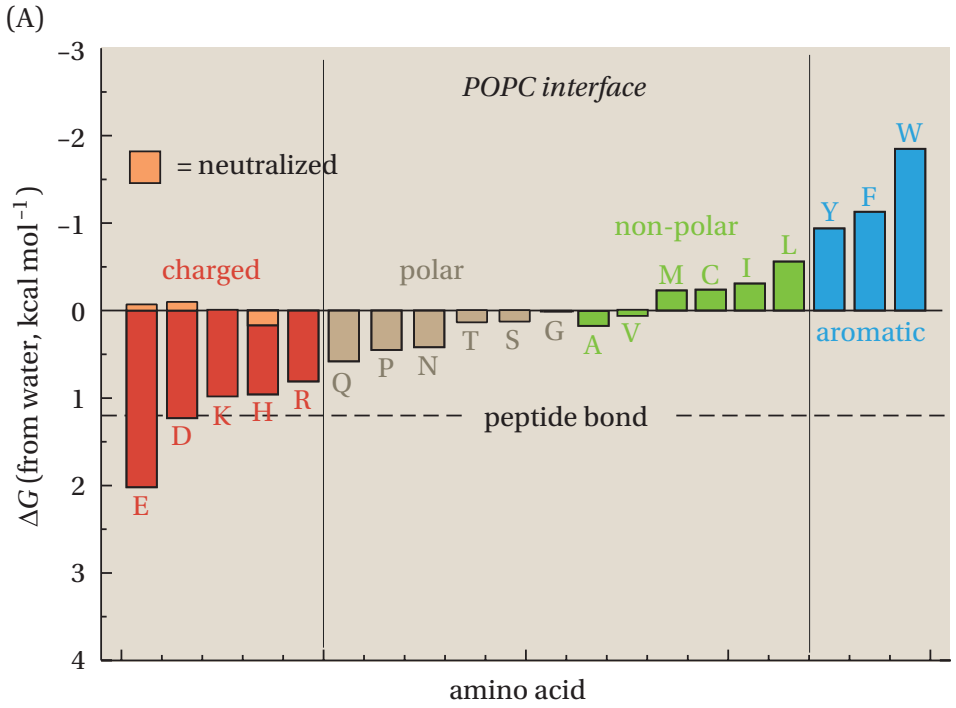
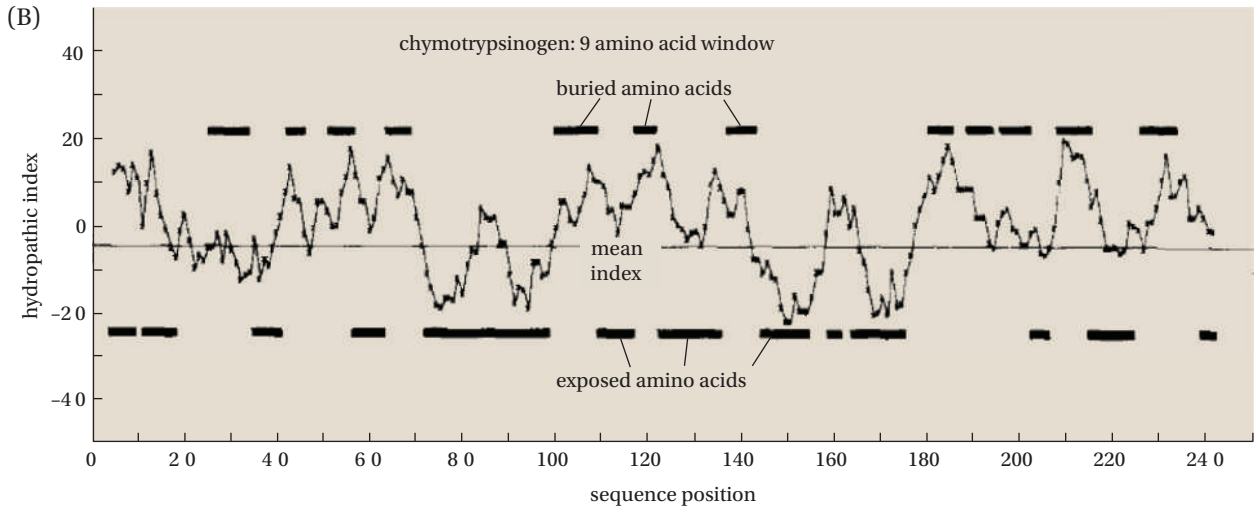
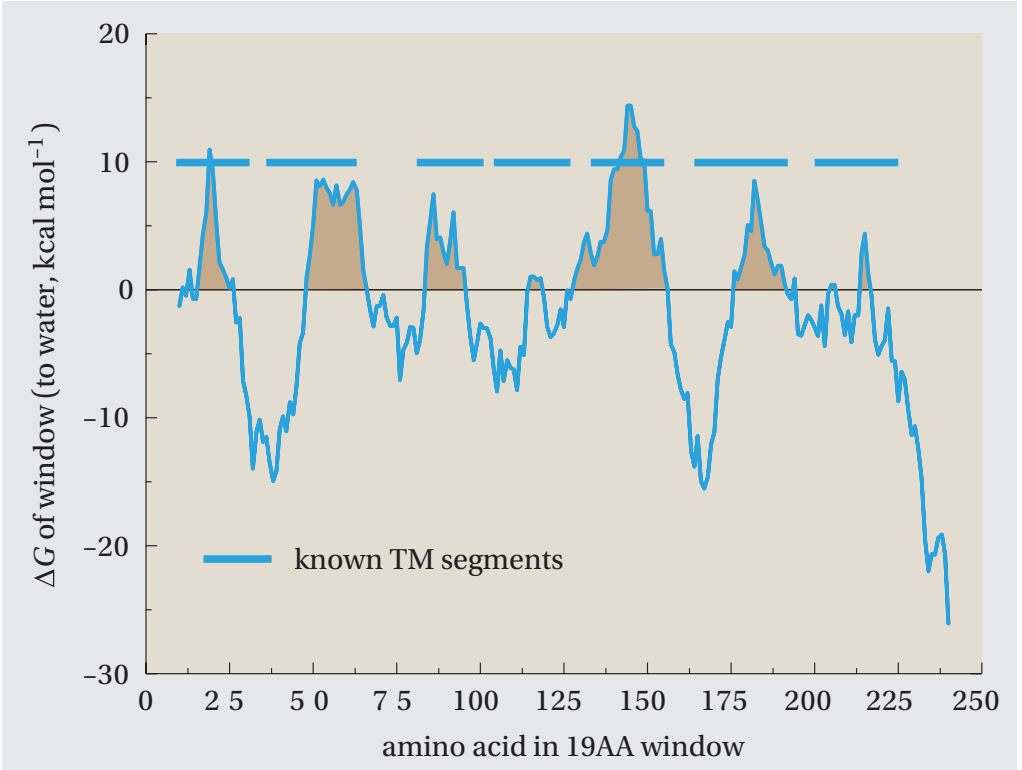


Figure 3.15 Cell Membranes (© Garland Science 2016)

Hydropathy plot analysis of protein sequences





Topology and orientation of helical integral membrane proteins

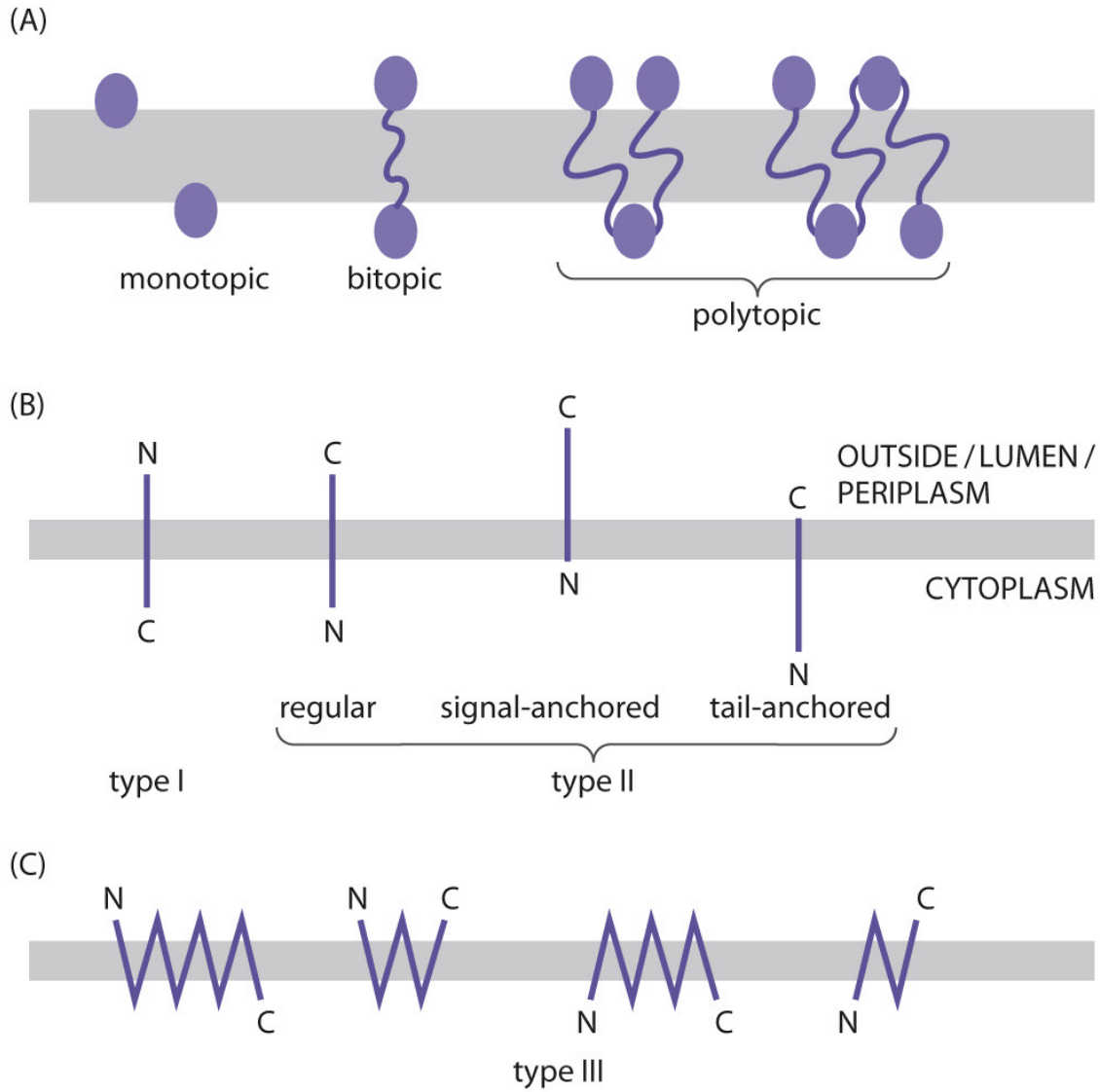
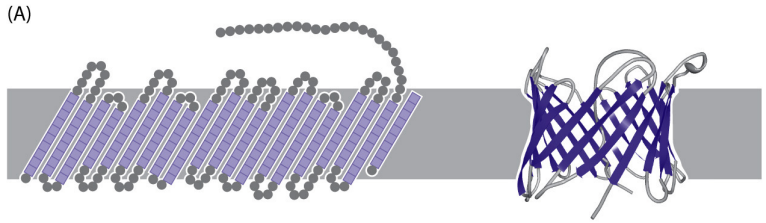


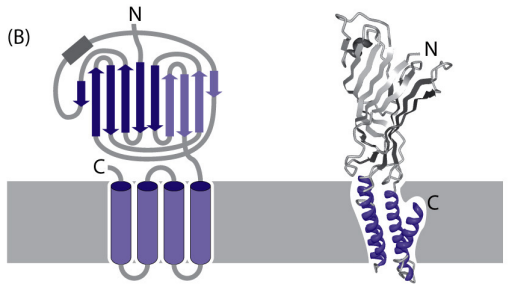
Figure 3.4 Cell Membranes (© Garland Science 2016)

Topology diagrams and X-ray structures for various transmembrane proteins

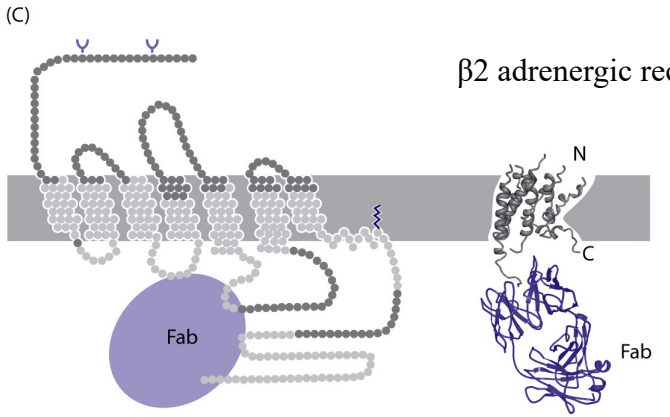


voltage-dependent anion channel (VDAC

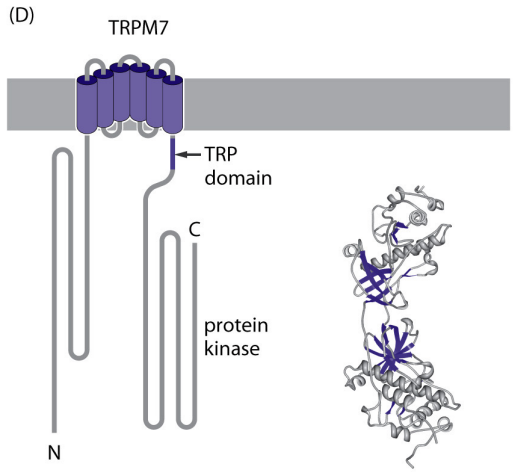
ELIC, pentameric ligand-gated ion channel

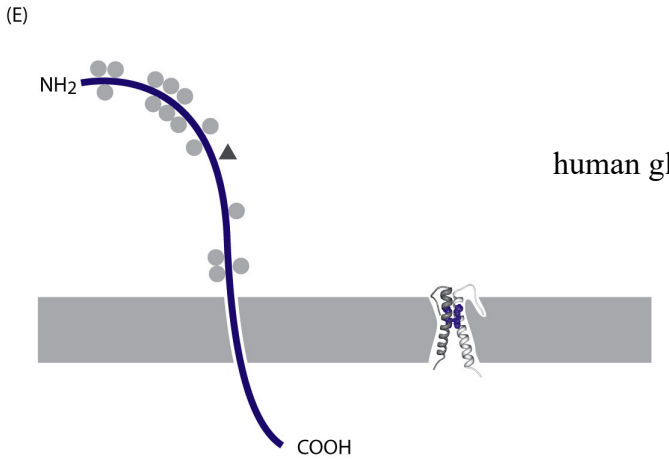


β2 adrenergic receptor protein



cold receptor





human glycophorin A

Figure 3.11 Cell Membranes (© Garland Science 2016)

Characteristics of transmembrane α helices

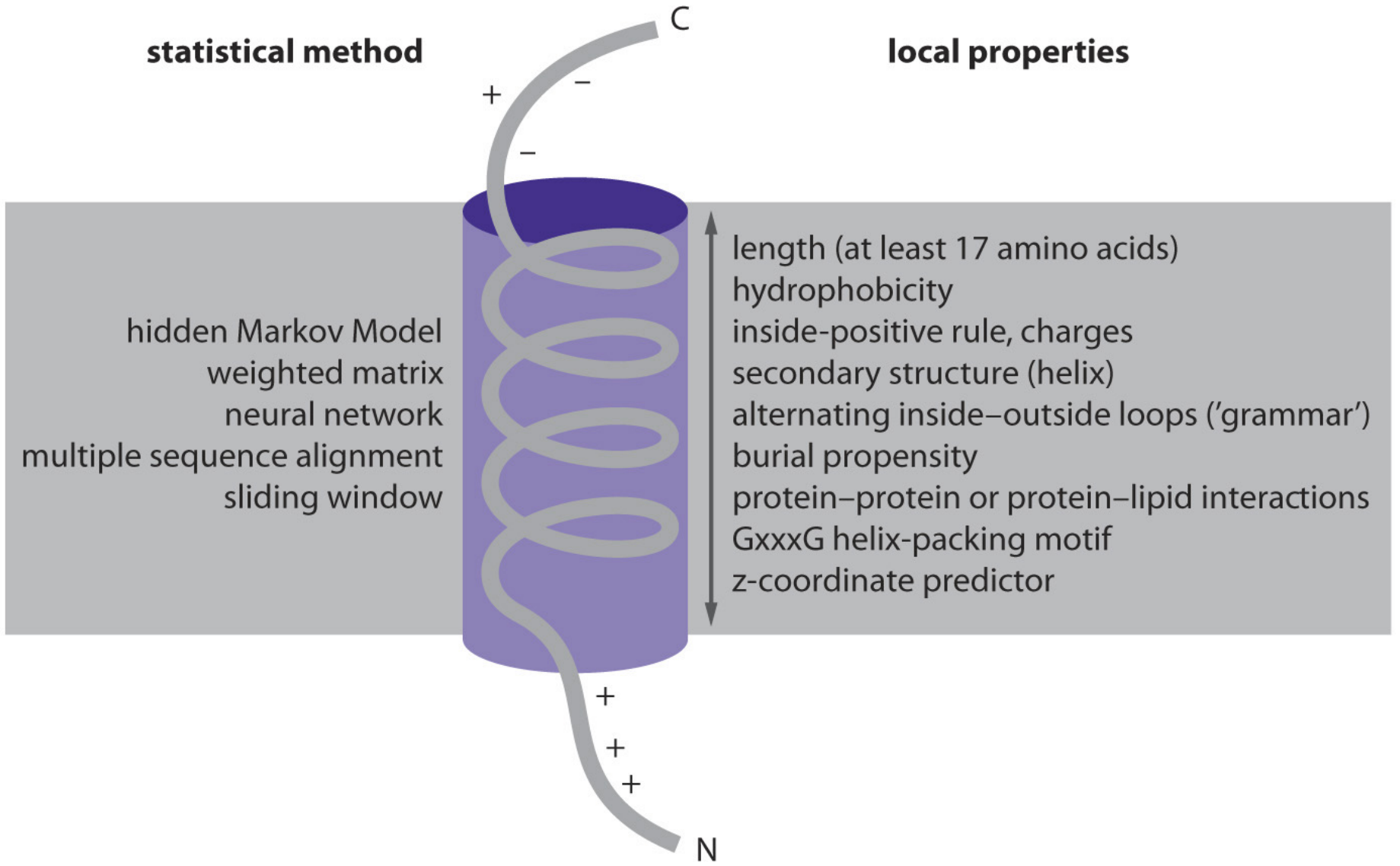


Figure 3.14a Cell Membranes (© Garland Science 2016)

Characteristics of transmembrane α helices

phospholemman (2JO1)

glycophorin (1AFO)

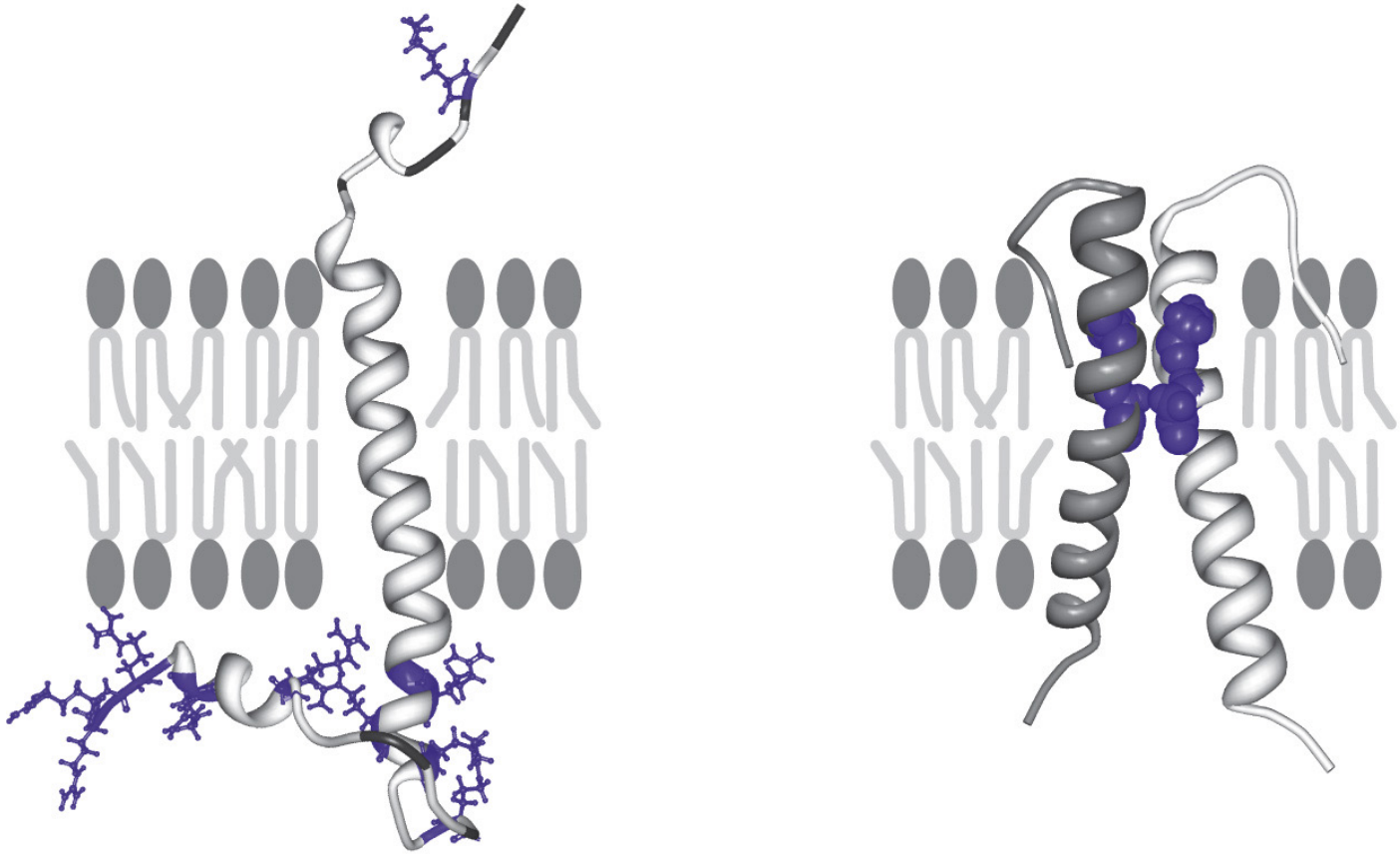
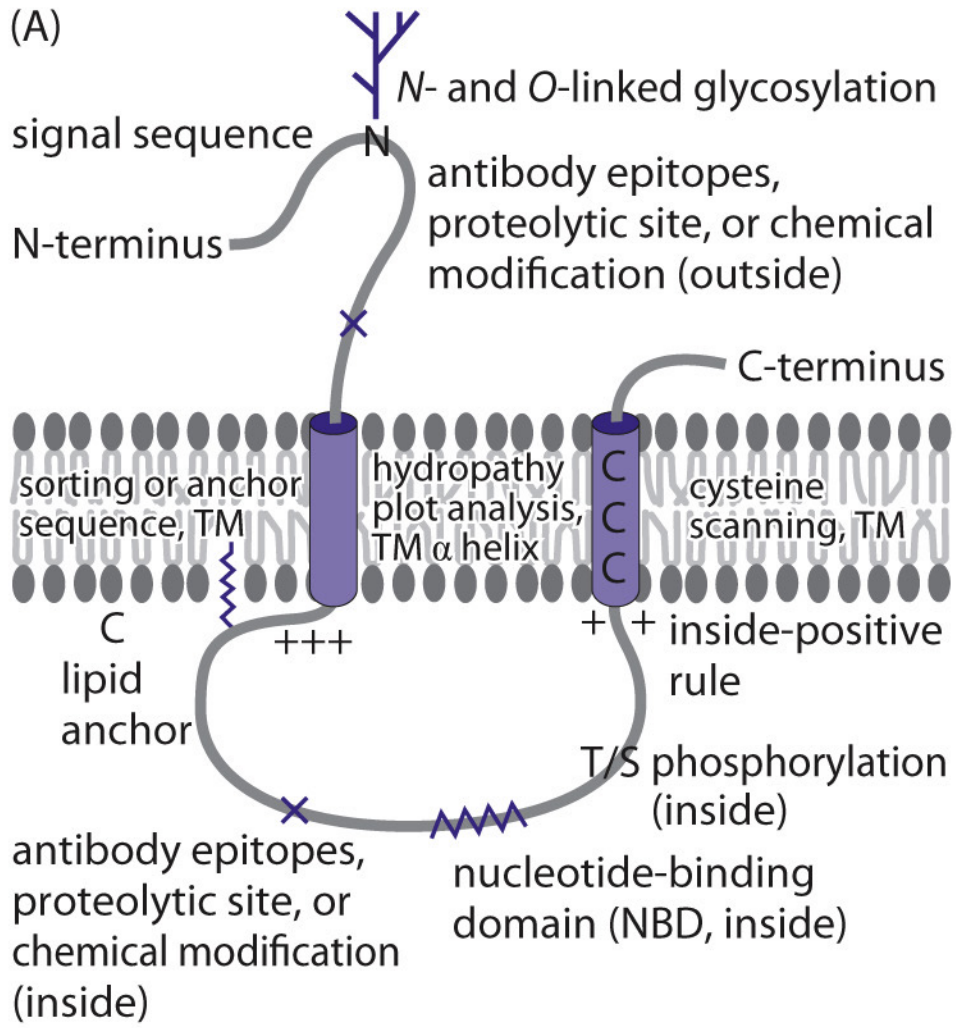


Figure 3.14b Cell Membranes (© Garland Science 2016)

Topology diagrams to probe intra- and extracellular orientation



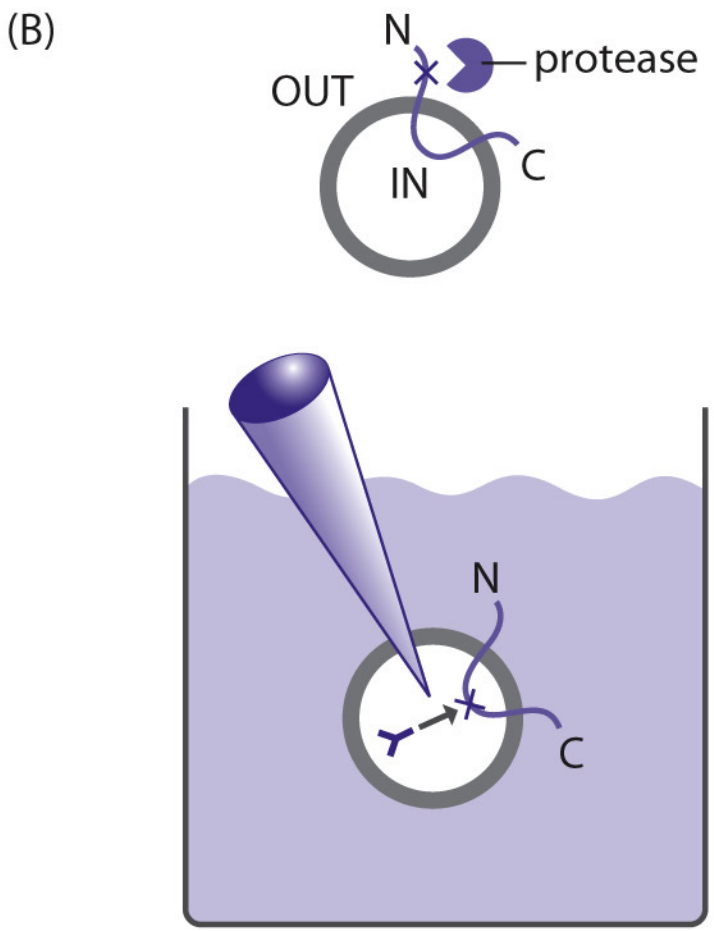


Figure 3.13 Cell Membranes (© Garland Science 2016)

Transmembrane segment variations

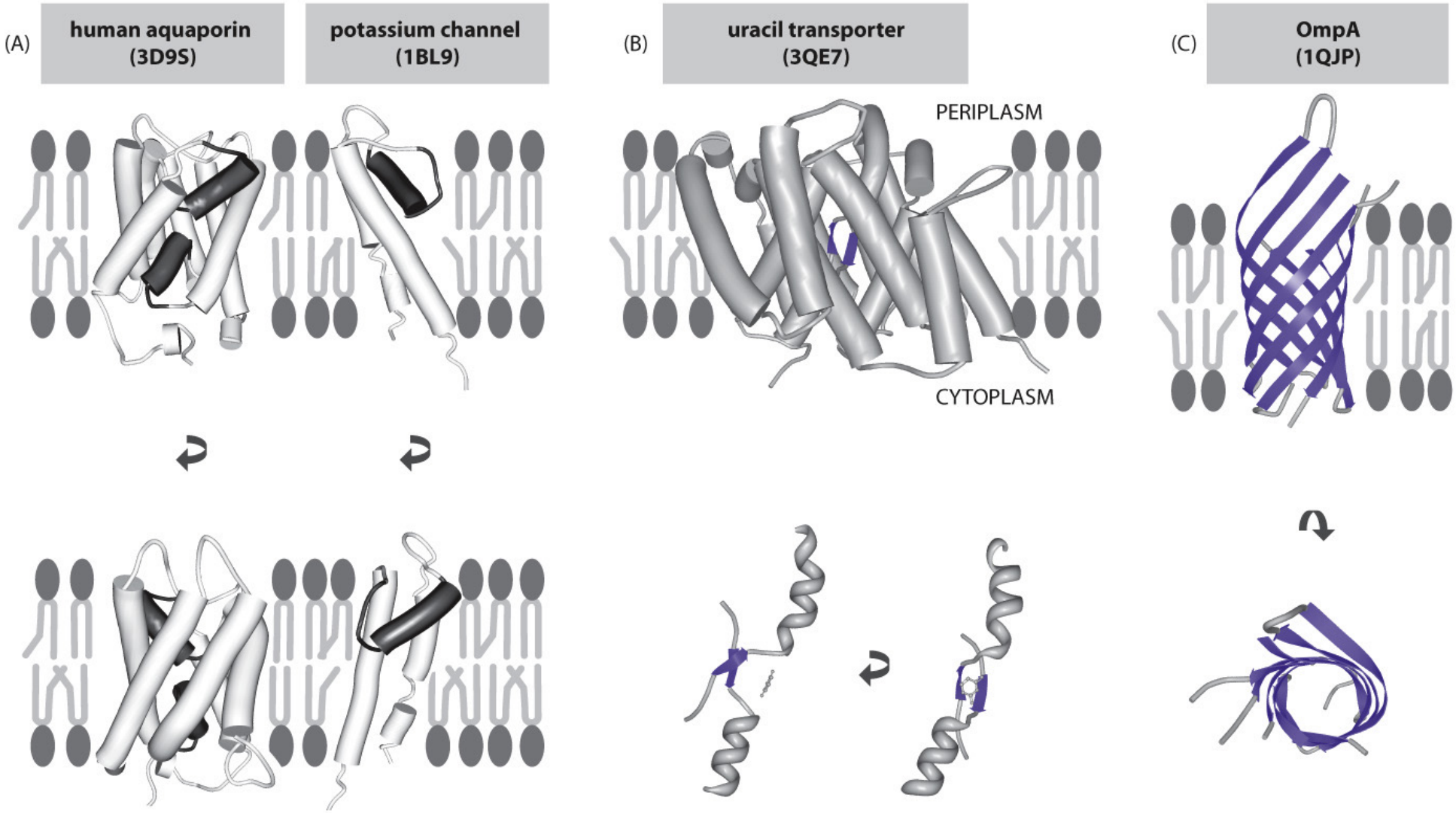


Figure 3.18 Cell Membranes (© Garland Science 2016)

Lipid–protein interactions

human glycophorin A

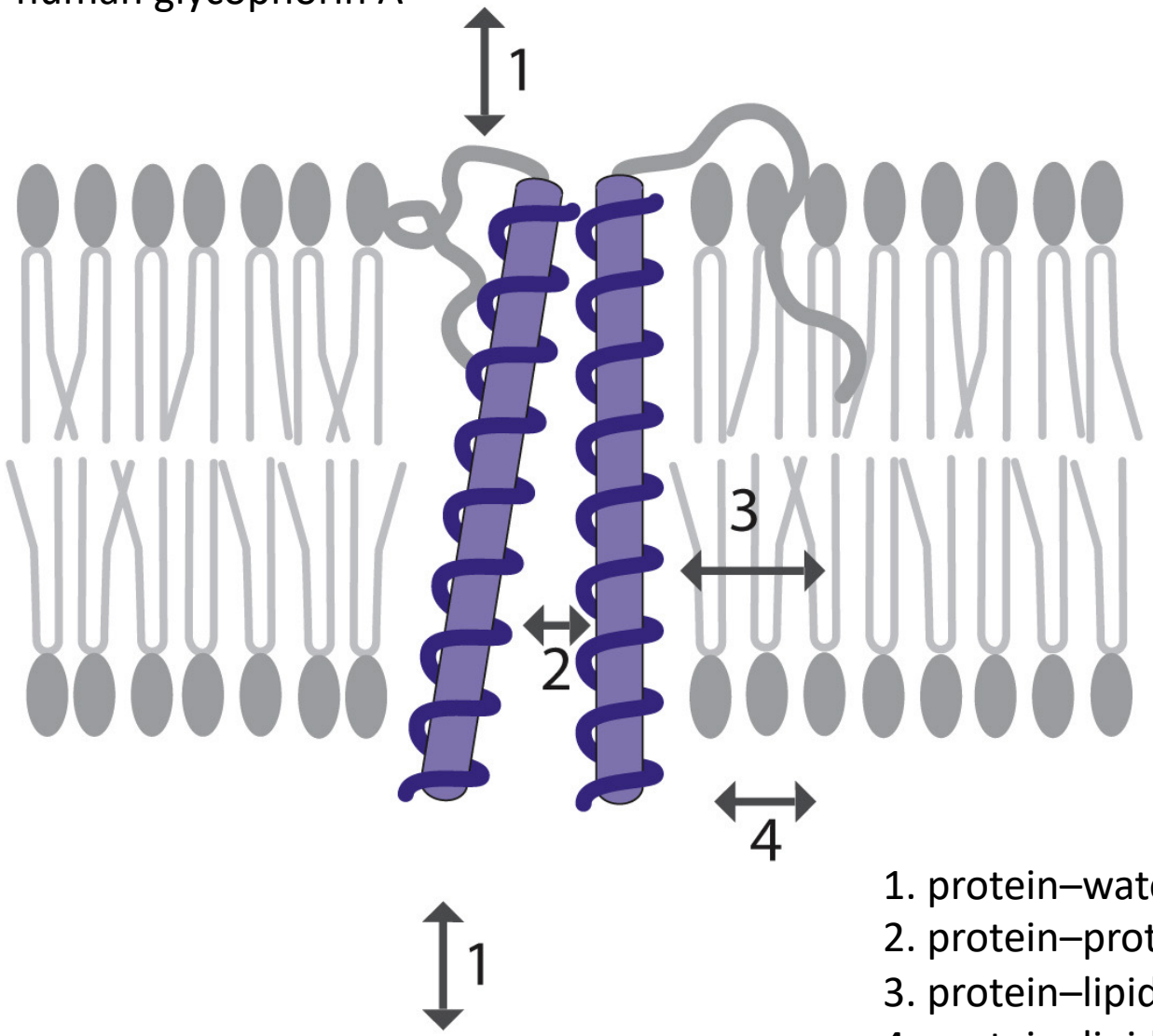
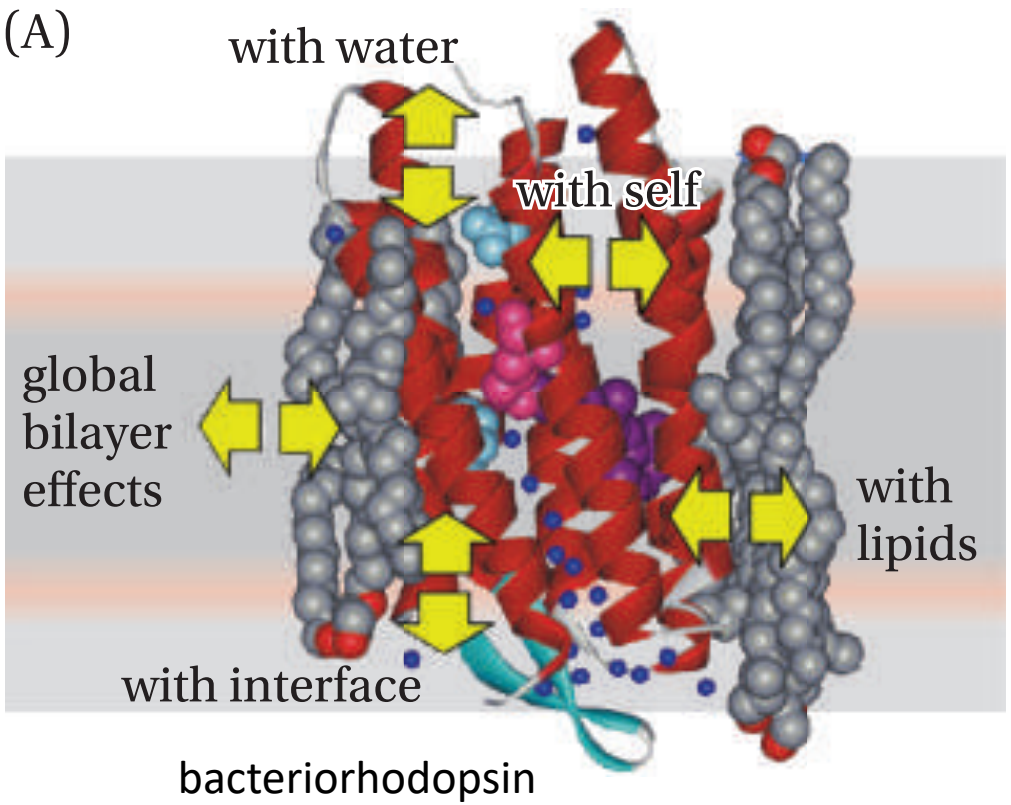


Figure 3.23a Cell Membranes (© Garland Science 2016)

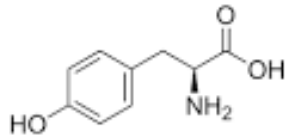


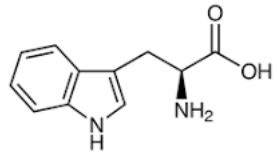
bacteriorhodopsin

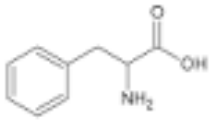
1. protein–water interactions
2. protein–protein interactions
3. protein–lipid tail (hydrophobic core) interactions
4. protein–lipid head group (bilayer interface) interactions

Aromatic side chains as membrane anchors

tyrosine, phenylalanine, tryptophan







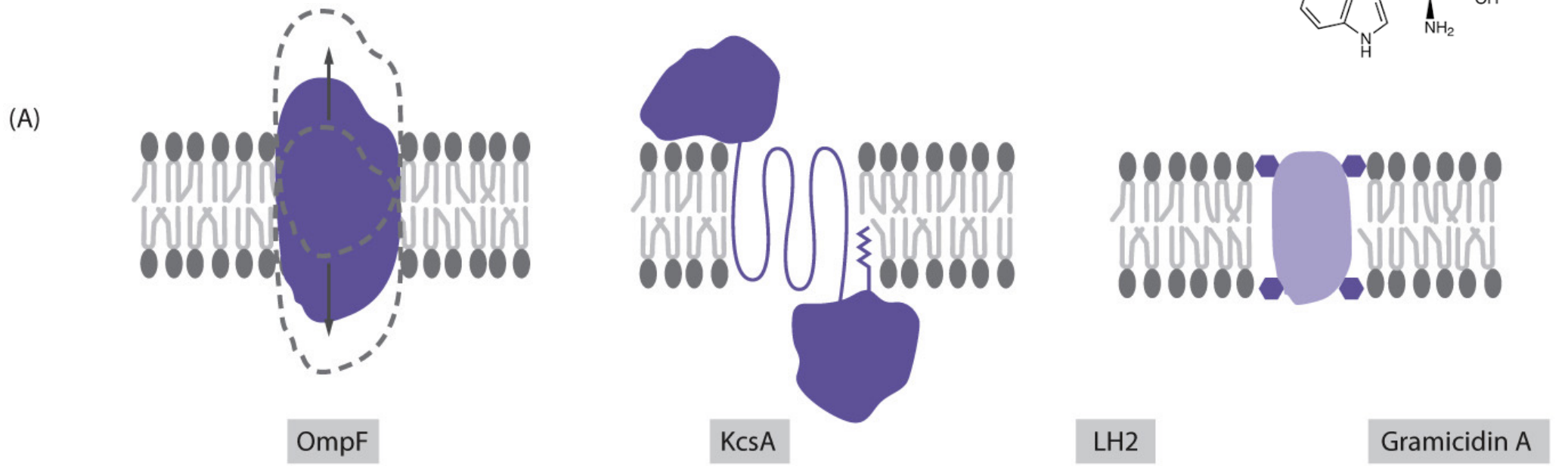
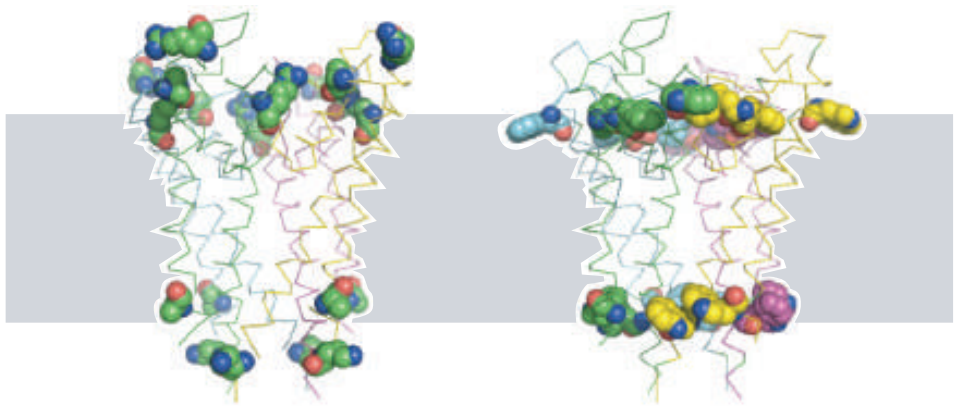


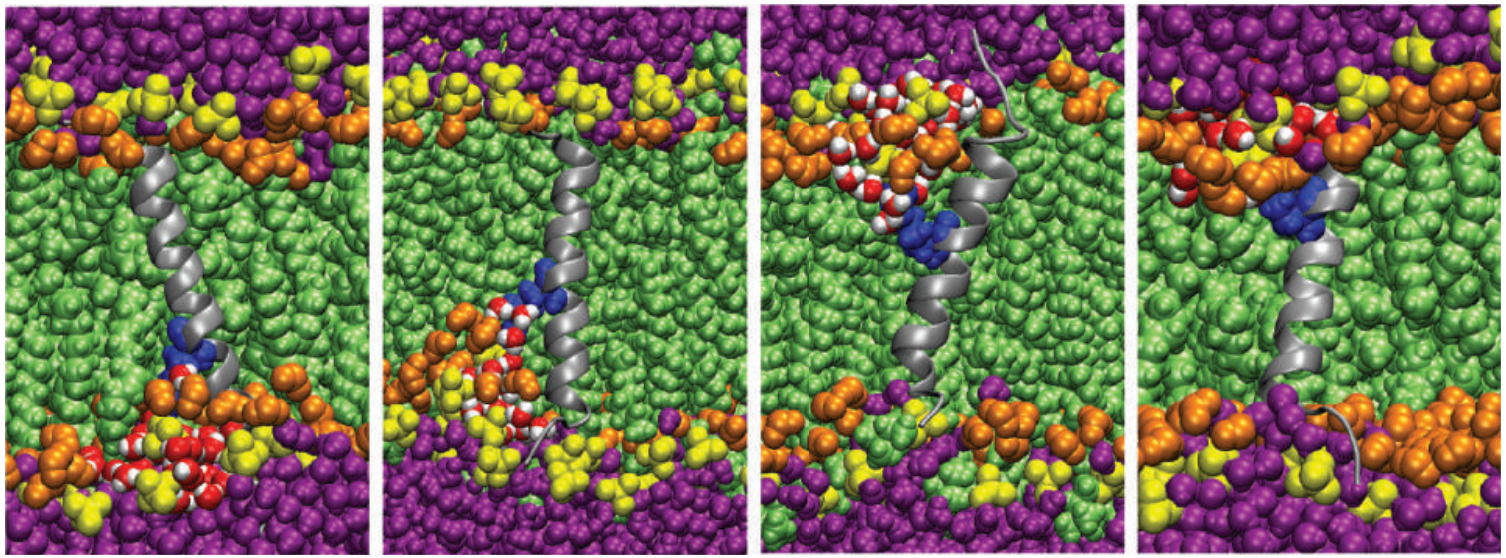
Figure 3.21 Cell Membranes (© Garland Science 2016)

(A) KcsA potassium channel

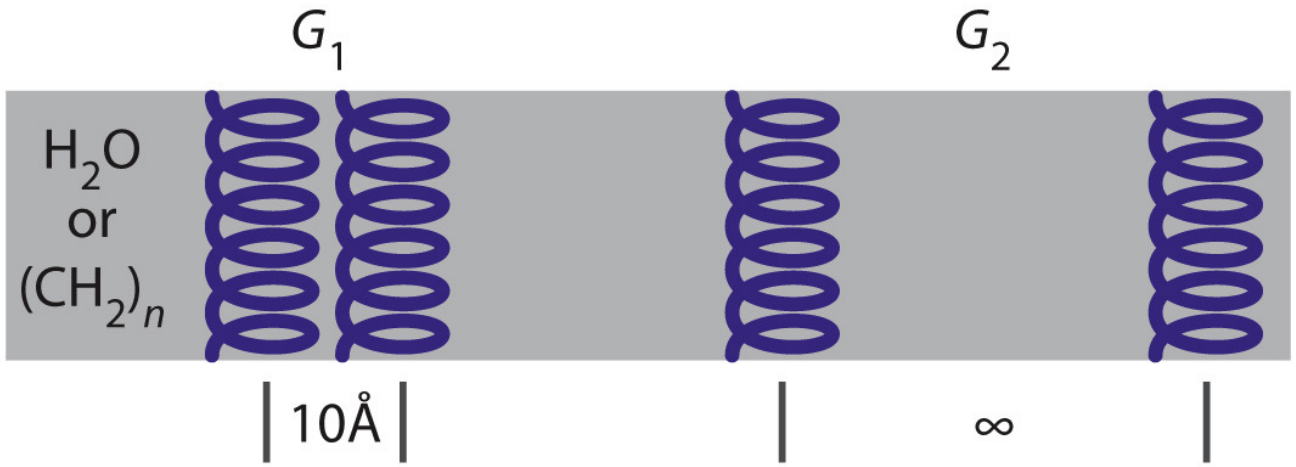


G Gly, P Pro, L Leu, R Arg

(C)



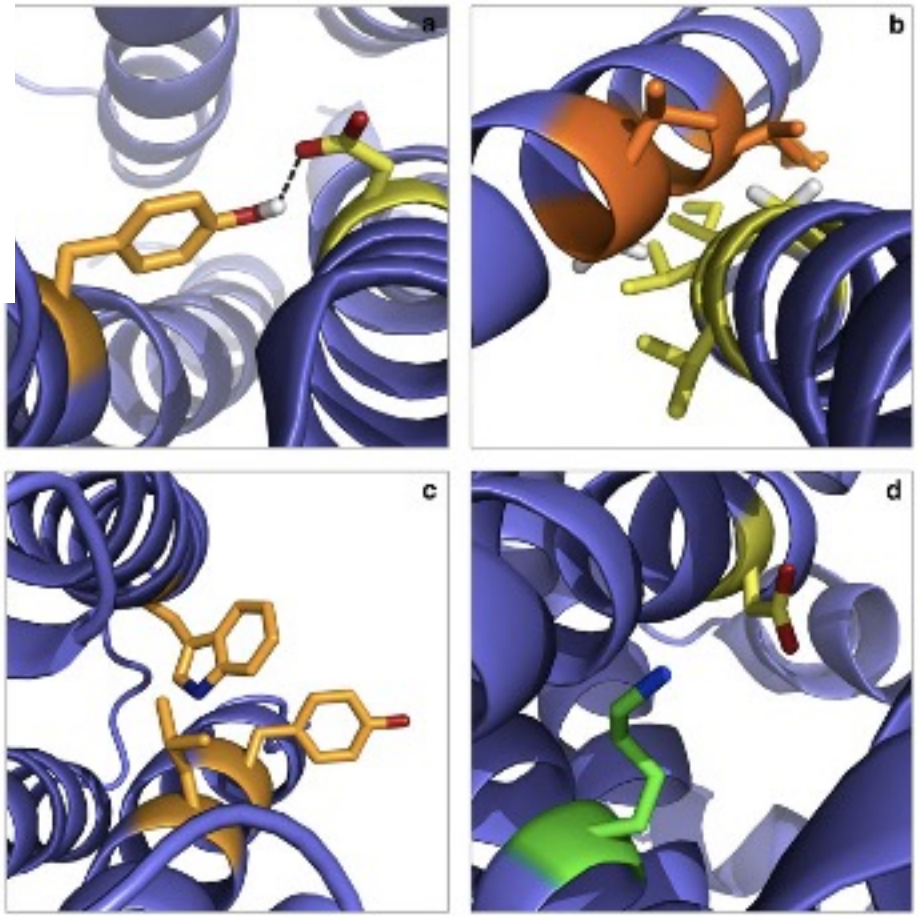
Interactions of helices in lipid bilayers



	$\Delta G = G_2 - G_1$ (kcal/mol) in H_2O	in $(CH_2)_n$
hydrophobic	20	0
AB ion pair	2	20 – 30
AB strong H bond	0	10 – 20
AB H bond	–1	5
conformation entropy	–5	–5

Figure 3.23b Cell Membranes (© Garland Science 2016)

Interhelical hydrogen bonds. In bacteriorhodopsin from Halobacterium salinarum Tyr185 (orange) of helix 6 and Asp212 (yellow) of helix 7 form an interhelical hydrogen bond (dashed line).

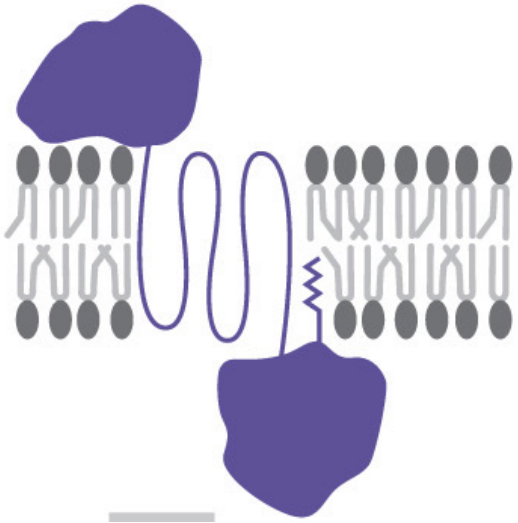


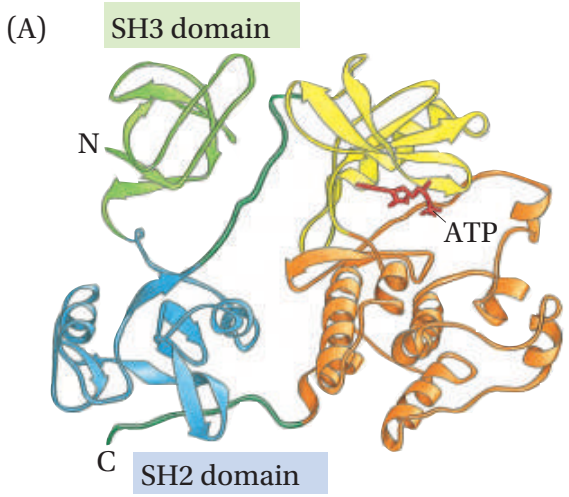
van der Waals contacts. Small residues (orange and yellow) increase the homodimer interface and allow for extensive van der Waals contacts in human glycophorin A Hydrogens are shown for Gly residues only

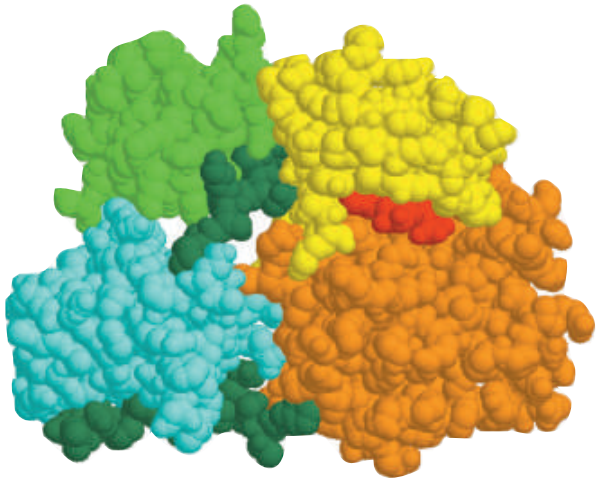
Aromatic–aromatic interactions. In subunit I of aberrant ba3-cytochrome c oxidase from Thermus thermophilus Trp110 of helix 4 interacts with Tyr23 and Leu27 of helix 1, although it is partly exposed to the lipid bilayer.

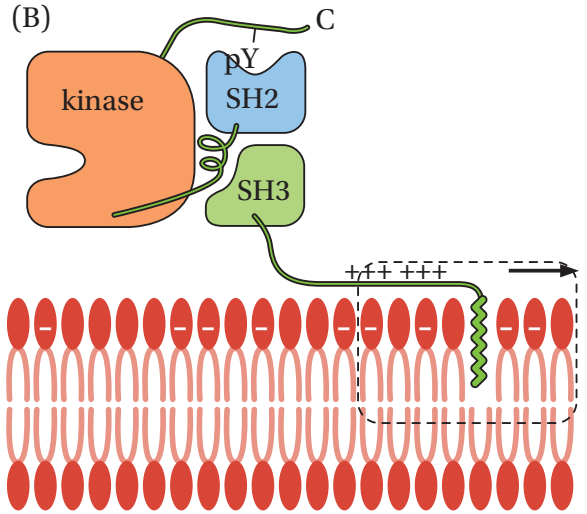
Salt bridges. Lys358 and Asp237 are crucial for membrane insertion of lac permease from E. coli and have been suggested to form a salt bridge within the protein's transmembrane region

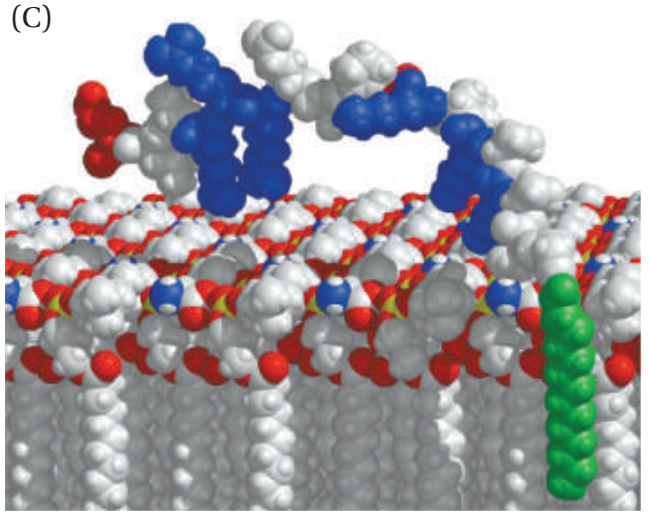
Bind to Membranes with the Help of Covalently Linked Lipids









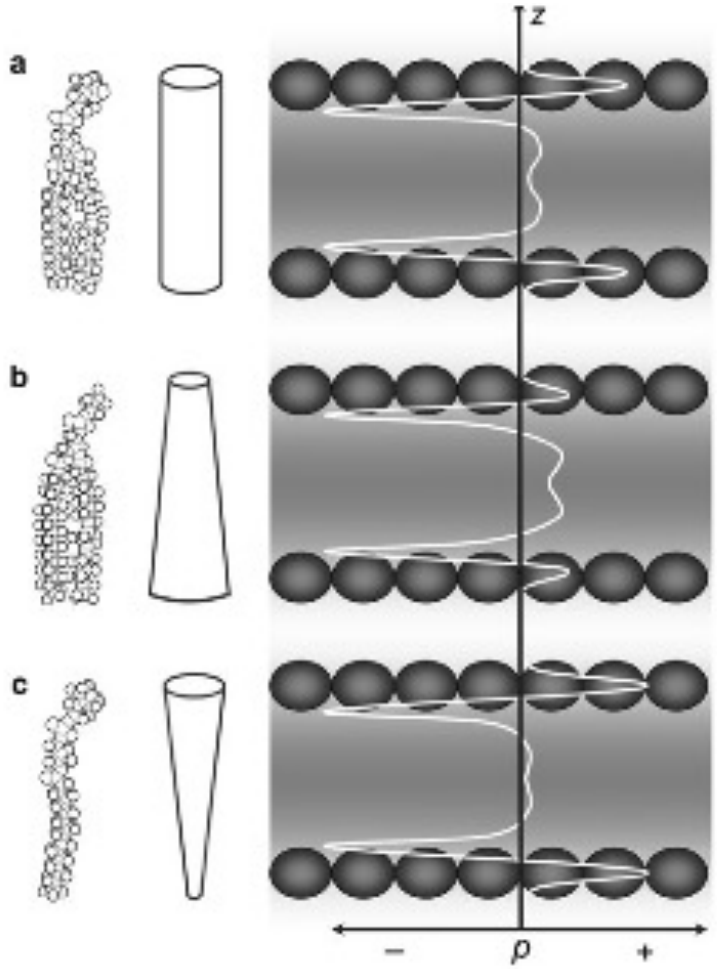


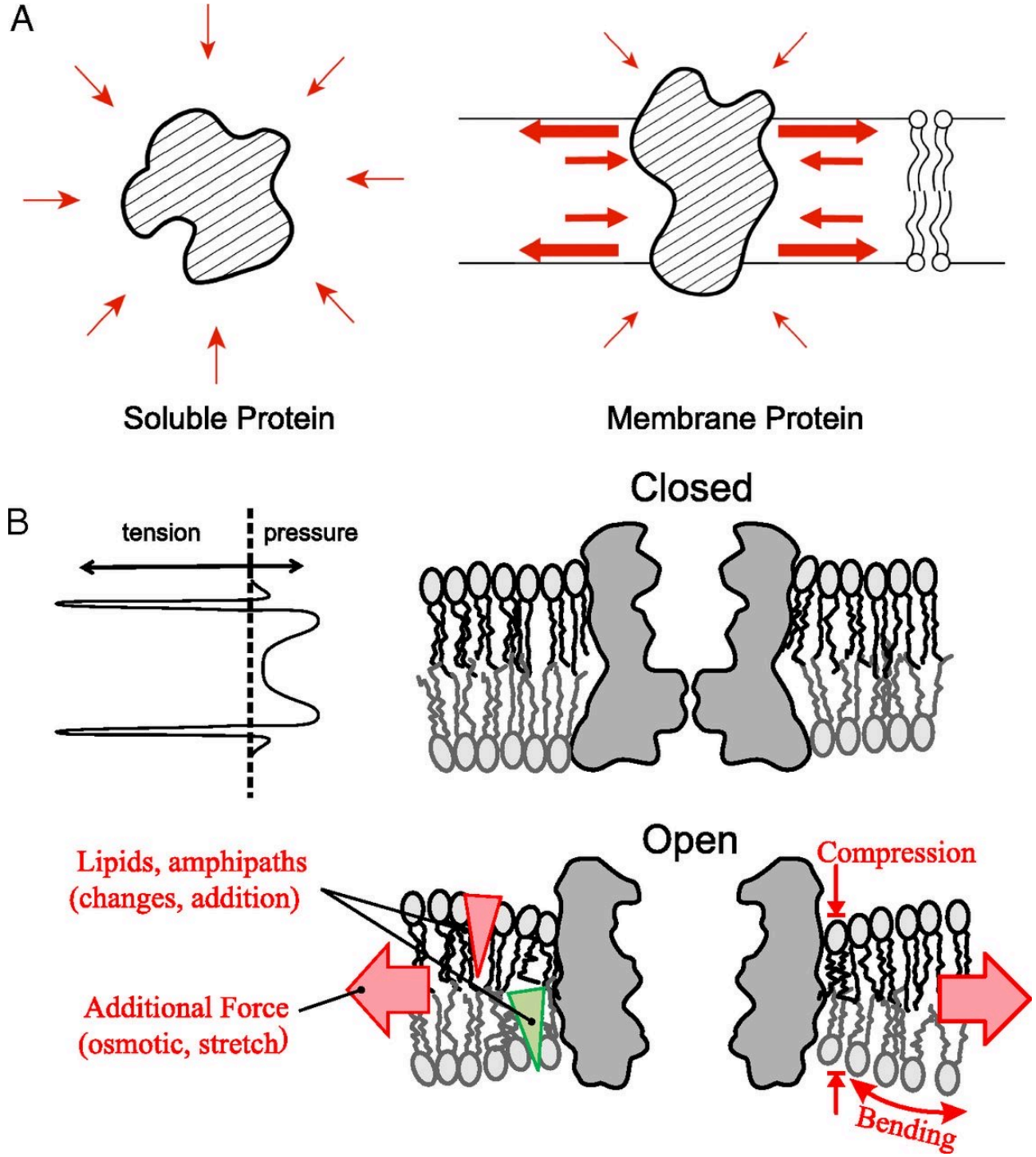
(D)

Src(2–16)	myristate–GS SKSKPKDPSQRRR
MARCKS(151–175)	KKKKKRFSFKKSFKLSGFSFKKNKK
HIV-1 Gag (2–31)	myristate–GA RASVLSGGELDRWEKIRLRPGGKKKYKL
K-Ras 4B (174–185)	GKKKKKKSKTSC–farnesyl

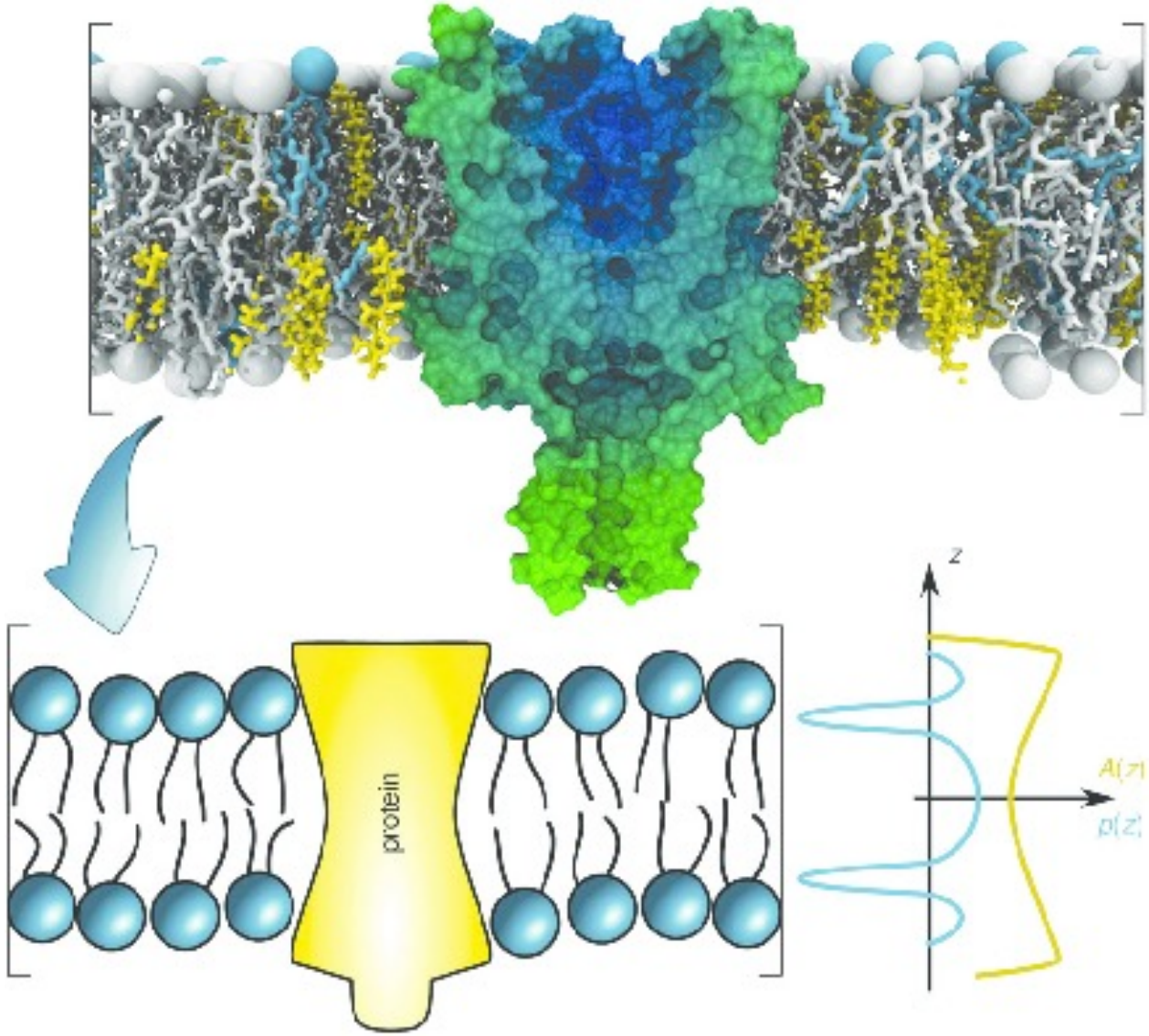
K Lys, R Arg, G Gly, S Ser, D Asp, E Glu, N Asn, V Val

Lateral Pressure

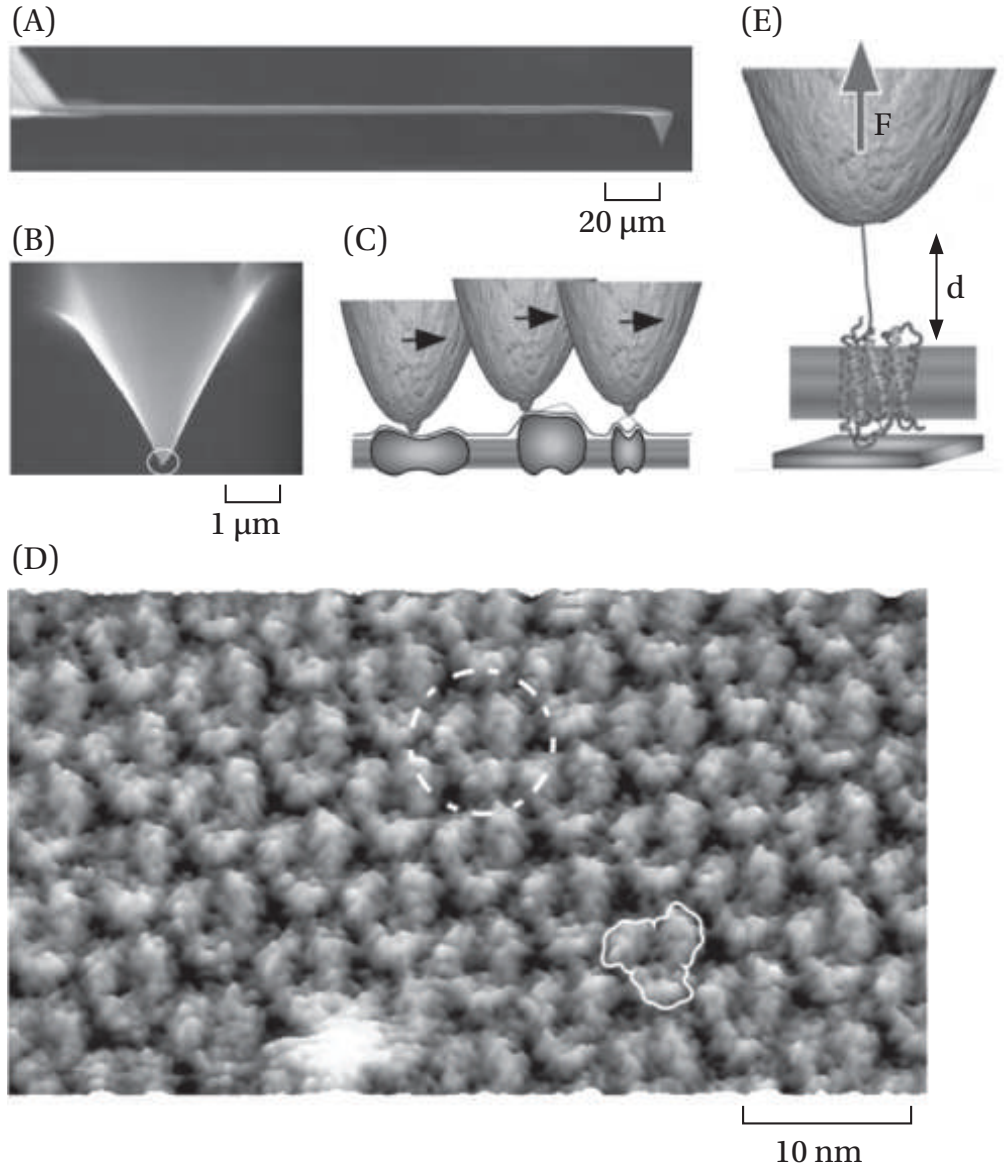




Lateral Pressure



Forced unfolding of membrane-bound BR



Protein–lipid complexes

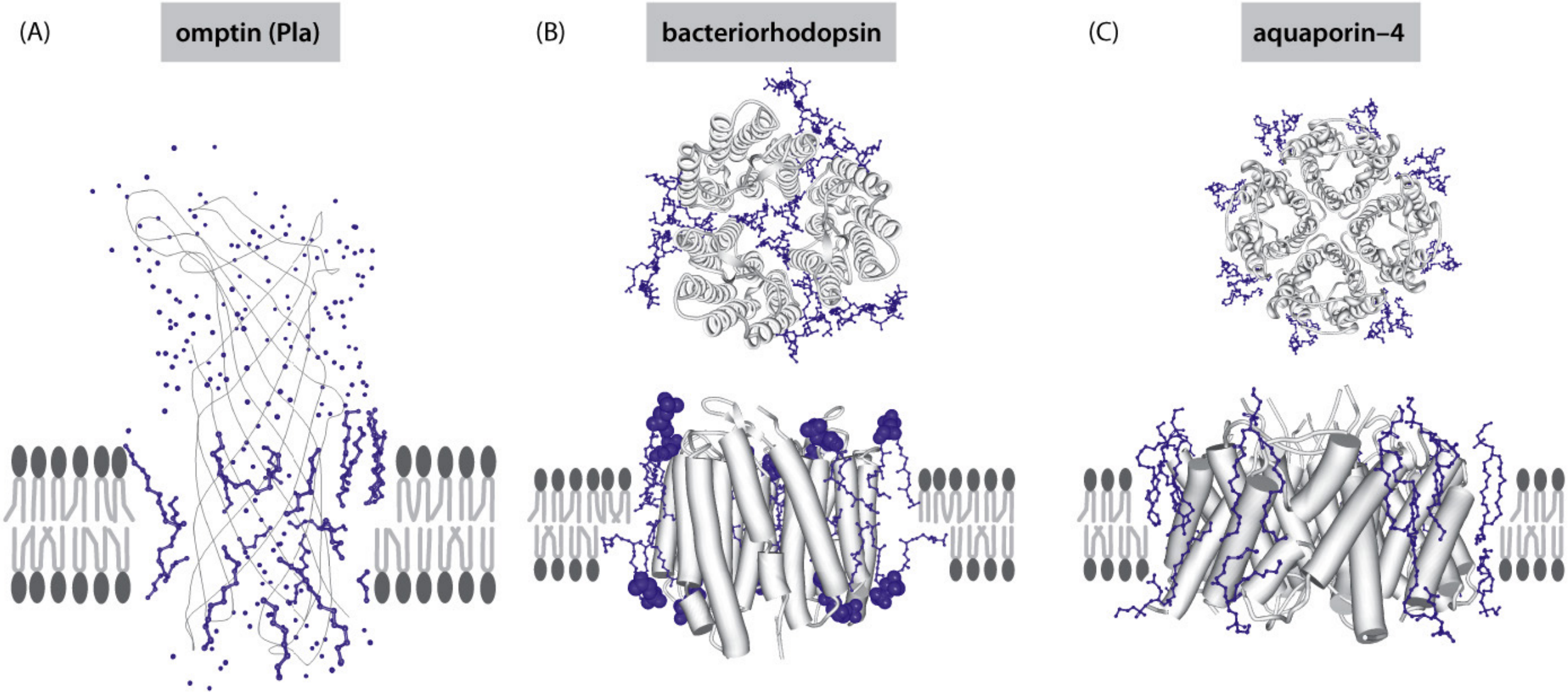


Figure 3.25 Cell Membranes (© Garland Science 2016)

Specific lipid effects

Lipids can act as co-factor that facilitate the folding or stabilise the structure of membrane proteins

Diacylglycerol kinase from E. coli, which requires 1,2-dioleoyl-sn- glycero-phosphoglycerol (DOPG) for proper folding.

Cardiolipin, a four-chain lipid binds to the large mitochondrial membrane protein bovine cytochrome c oxidase and is essential for its function. Cardiolipin is explicitly required for association of cytochrome c oxidase subunits IVa and IVb.

Several membrane-protein crystal structures show tightly bound lipid molecules and provide valuable insights into how these specifically interact with membrane proteins

The function of KvAP channel, a voltage- dependent K+-channel, depends on certain lipid species. KvAP senses voltage with the aid of Arg-containing structures located at the membrane interface and pointing into the membrane interior

The functional state of KvAP requires POPE or 1-palmitoyl-2-oleoyl-sn-glycero-phosphoglycerol (POPG) and that phosphate groups play a crucial role, as their enzymatic removal disrupts function

Oligomerization and clustering of membrane proteins

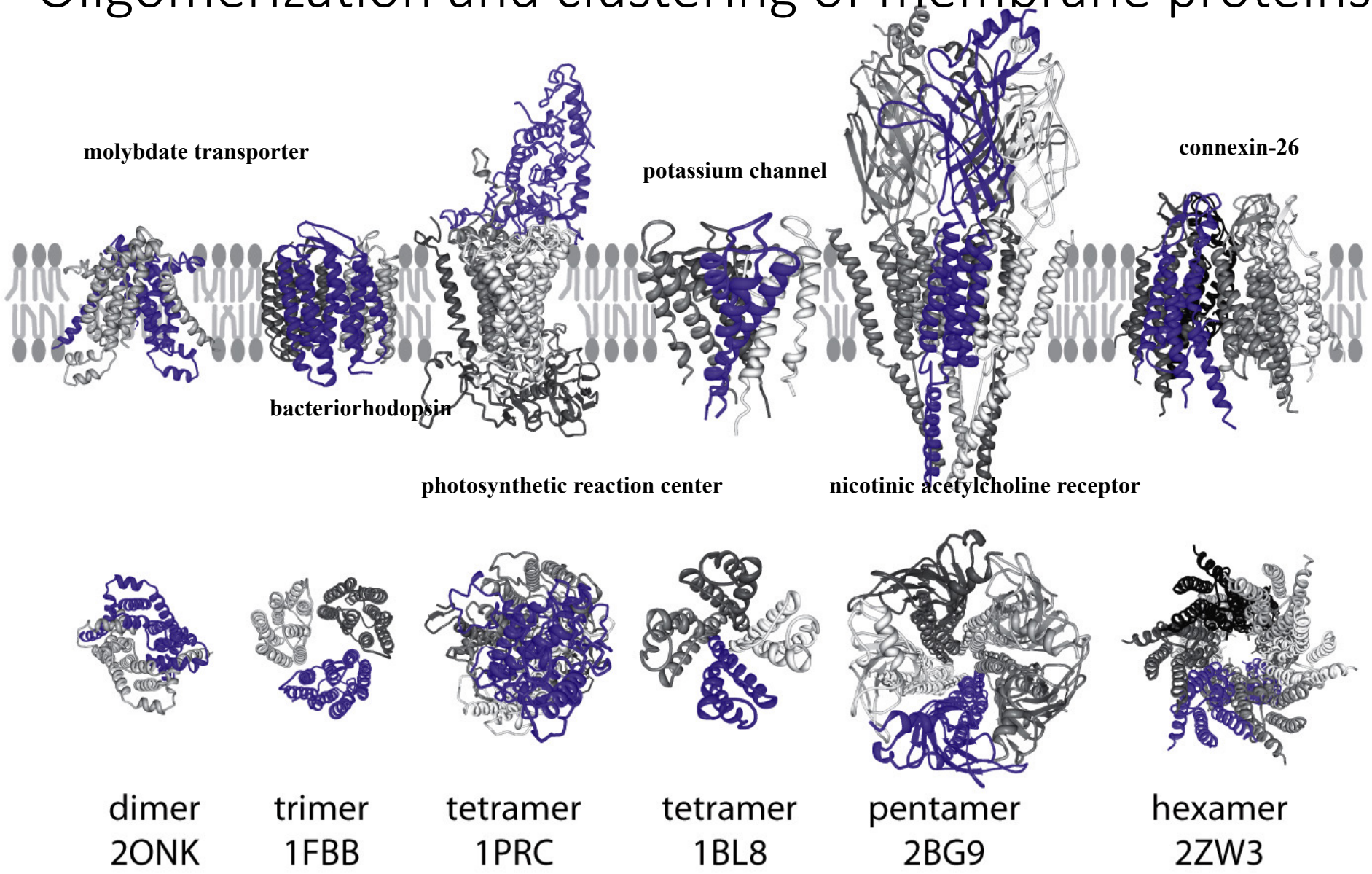


Figure 3.22c Cell Membranes (© Garland Science 2016)

Oligomerization and clustering of membrane proteins

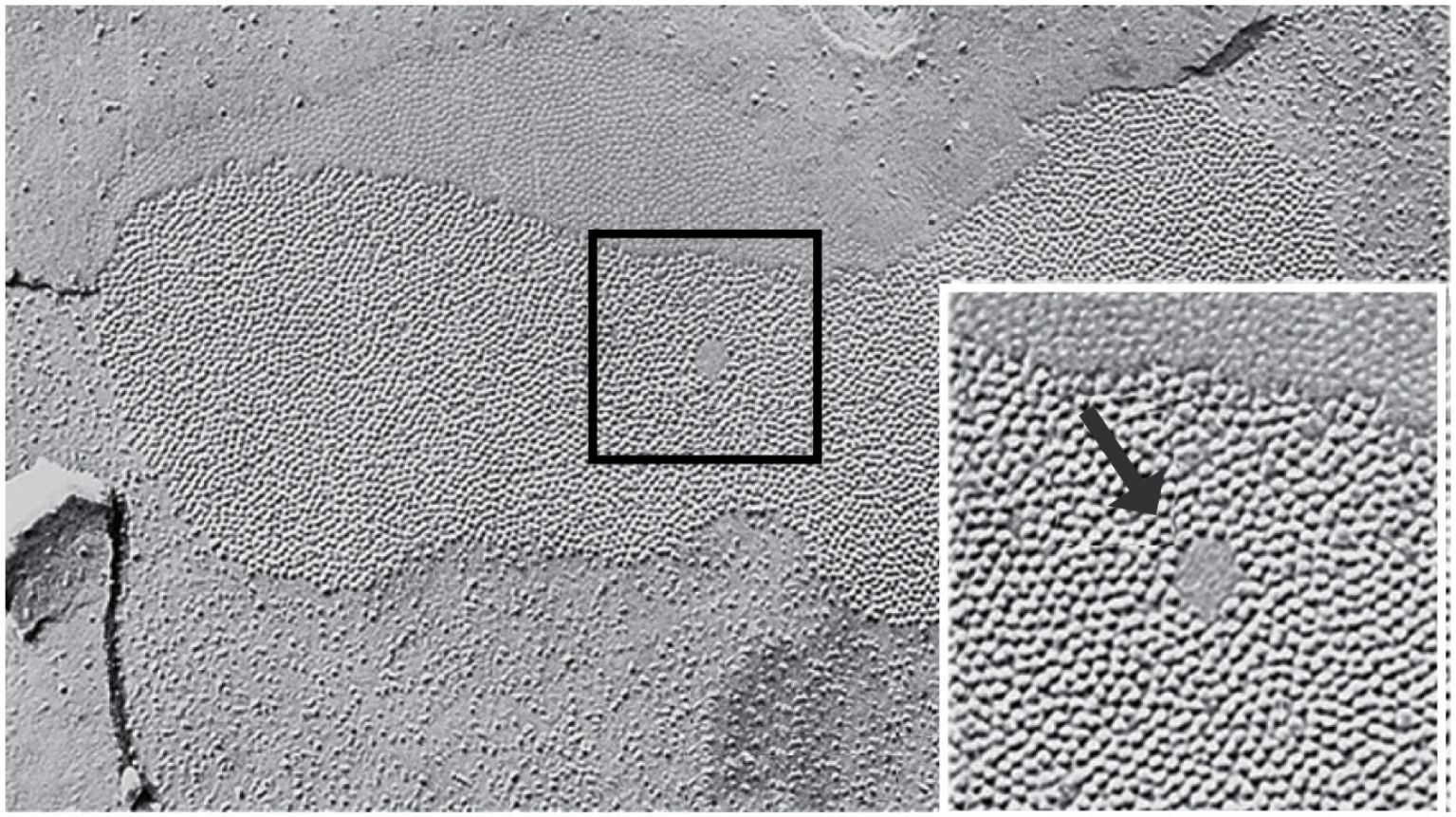
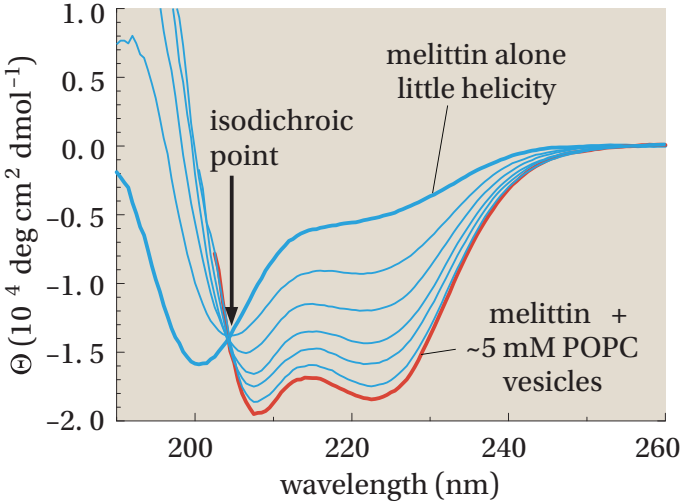


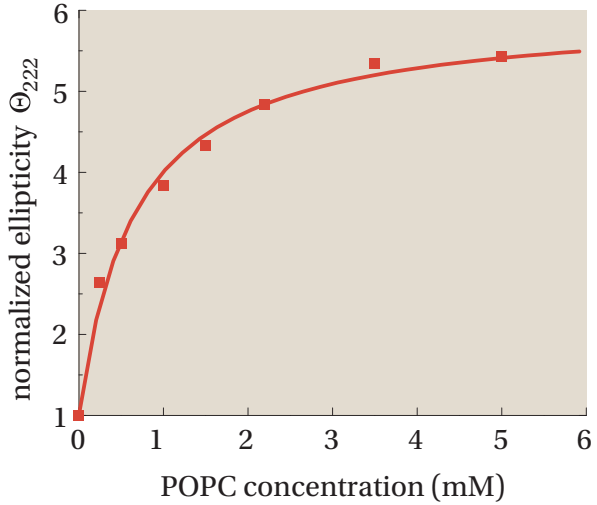
Figure 3.22b Cell Membranes (© Garland Science 2016)

Membrane active peptides

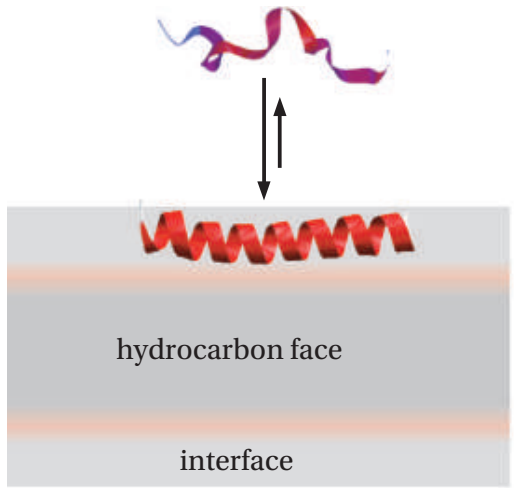
(A)



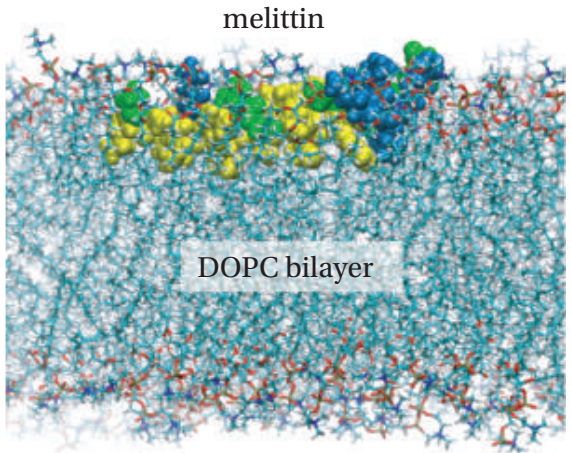
(C)



(B)



(D)



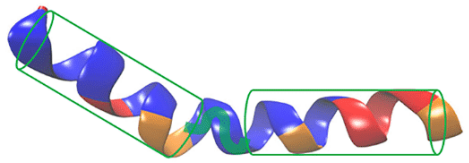
melittin

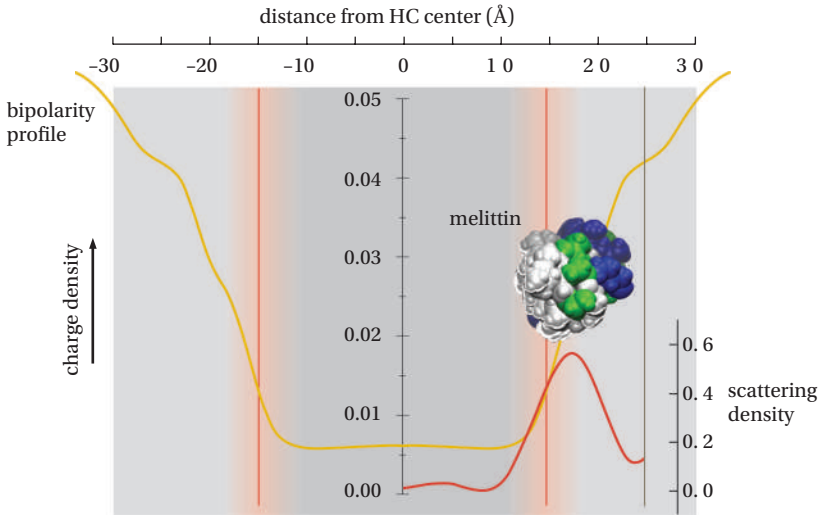
a

Peptide	Amino Acid Sequence *												
	1	2	3	4	5	6	7	8	9	10	11	12	13
Melittin	G	I	G	A	V	L	K	V	L	T	T	G	L
	14	15	16	17	18	19	20	21	22	23	24	25	26
	P	A	L	I	S	W	I	K	R	K	R	Q	Q

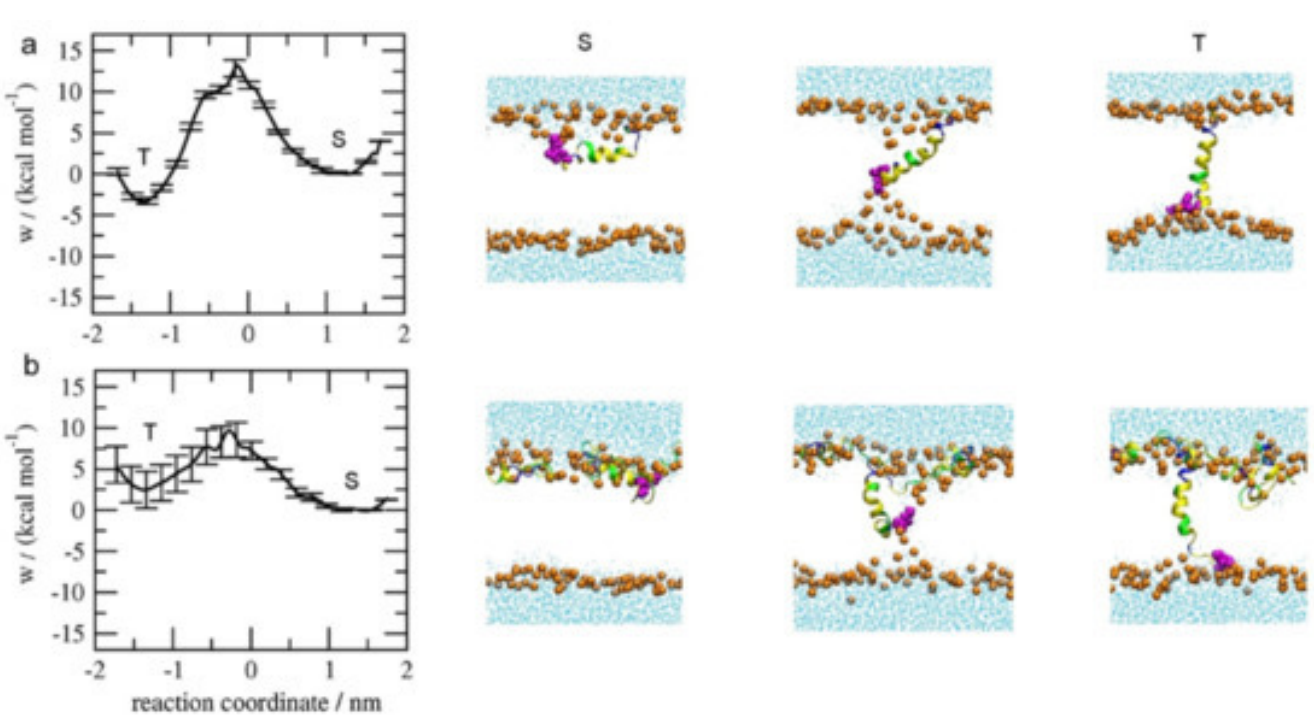
* Blue: hydrophobic; Orange: hydrophilic; Red: charged

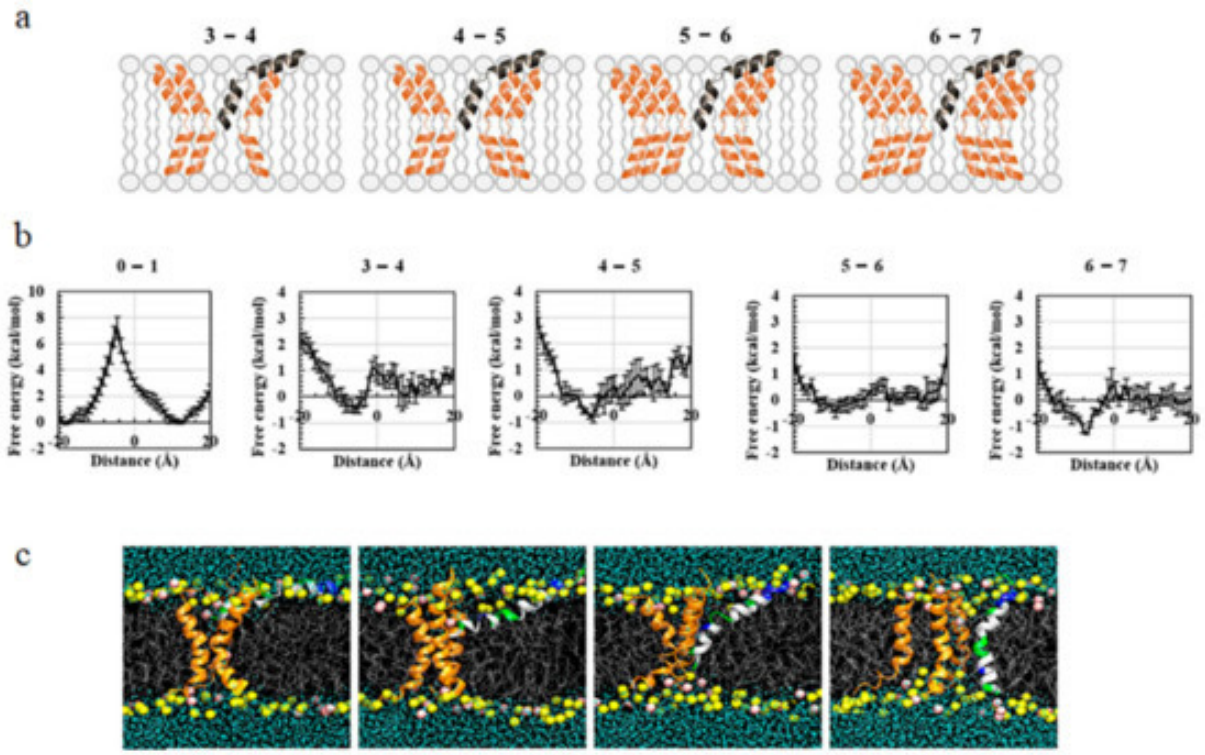
b





Melittin Insertion into Cell Membranes

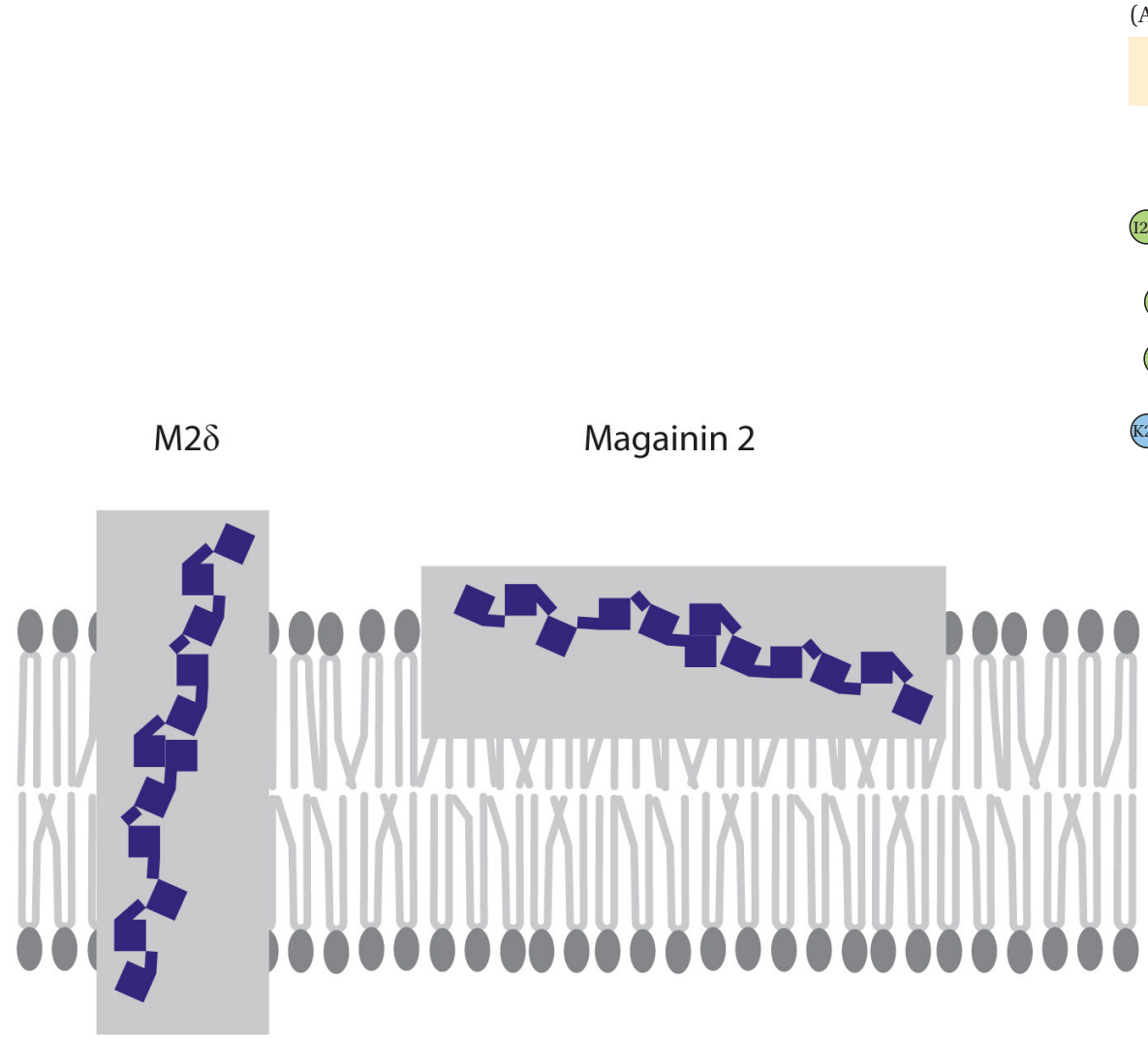


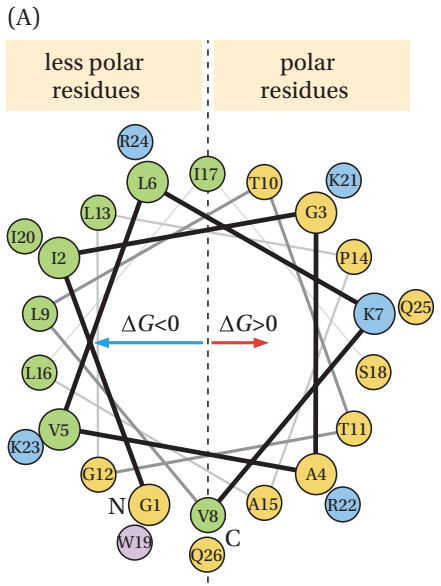


Molecules **2019**, *24*, 1775; doi:10.3390/molecules24091775

α-Helical ionophoric peptides

Gly-Ile-Gly-Lys-Phe-Leu-His-Ser-Ala-Lys-Lys-Phe-Gly-Lys Ala-Phe-Val-Gly-Glu-Ile-Met-Asn-Ser.





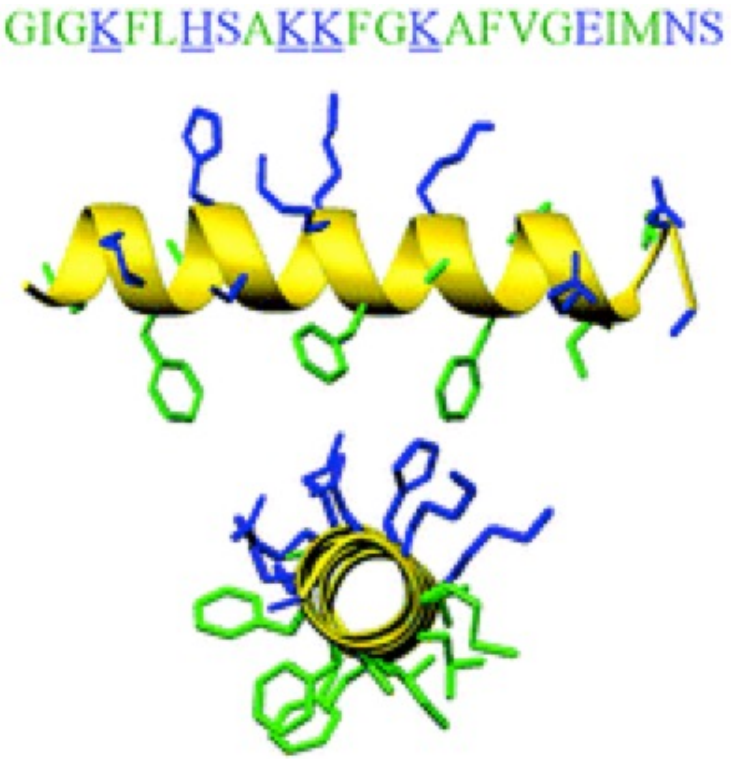


Figure 3.27a Cell Membranes (© Garland Science 2016)

α-Helical ionophoric peptides

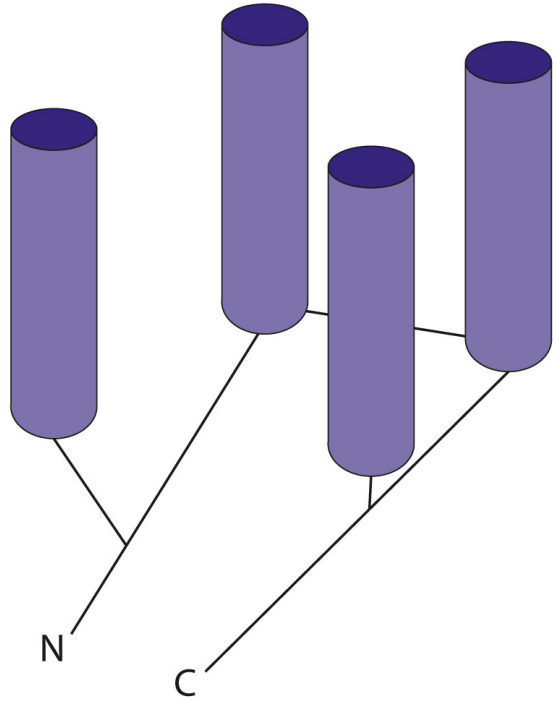
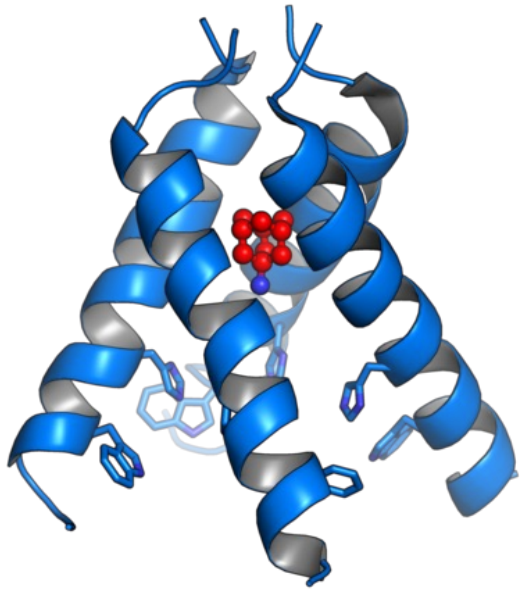


Figure 3.27c Cell Membranes (© Garland Science 2016)



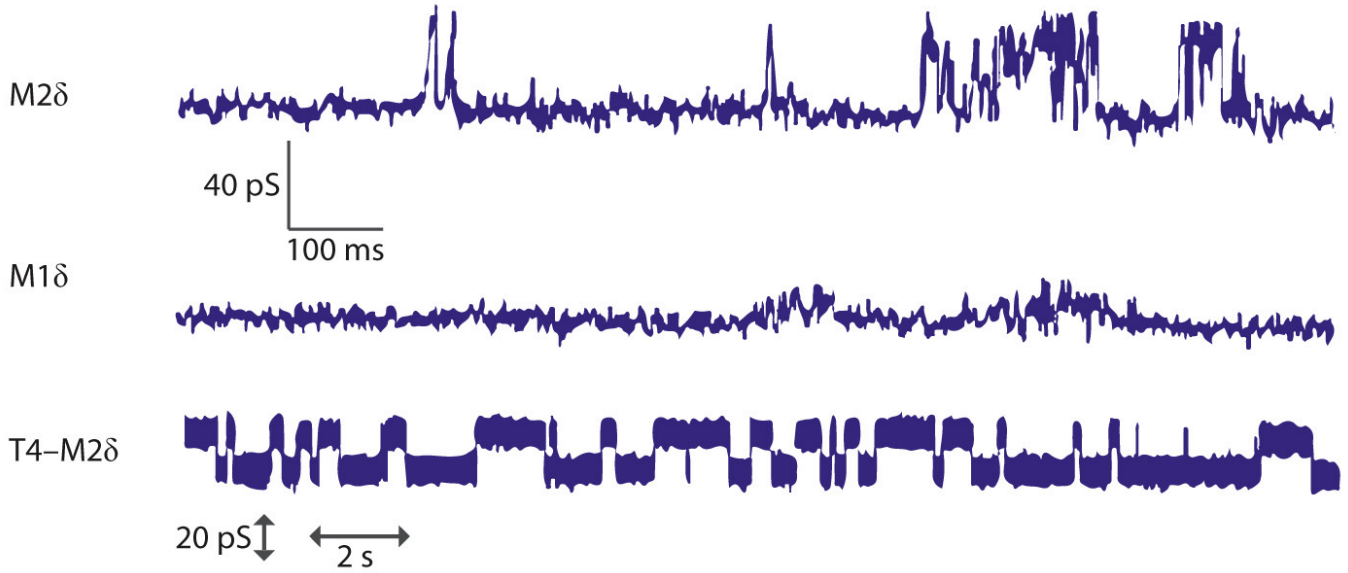
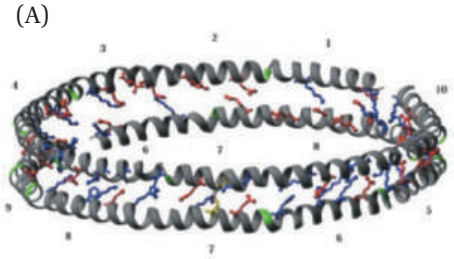
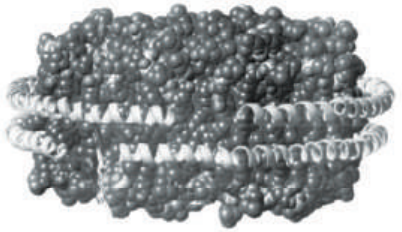


Figure 3.27d Cell Membranes (© Garland Science 2016)

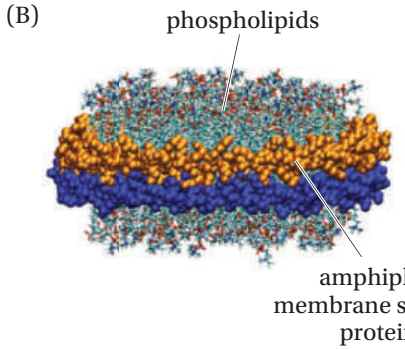
Amphiphilic helices

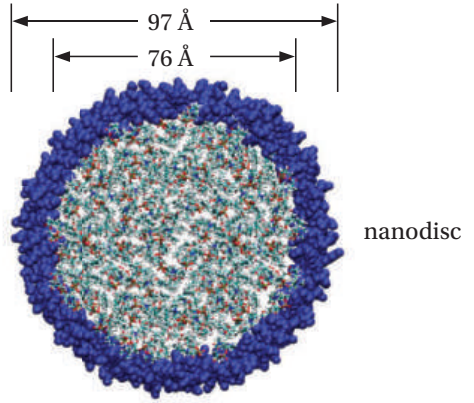


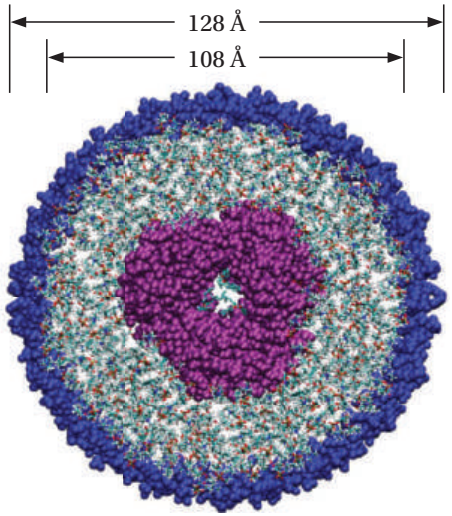
apolipoprotein A-I
tandem amphiphilic helices



lipoprotein A-I
tandem amphiphilic helices

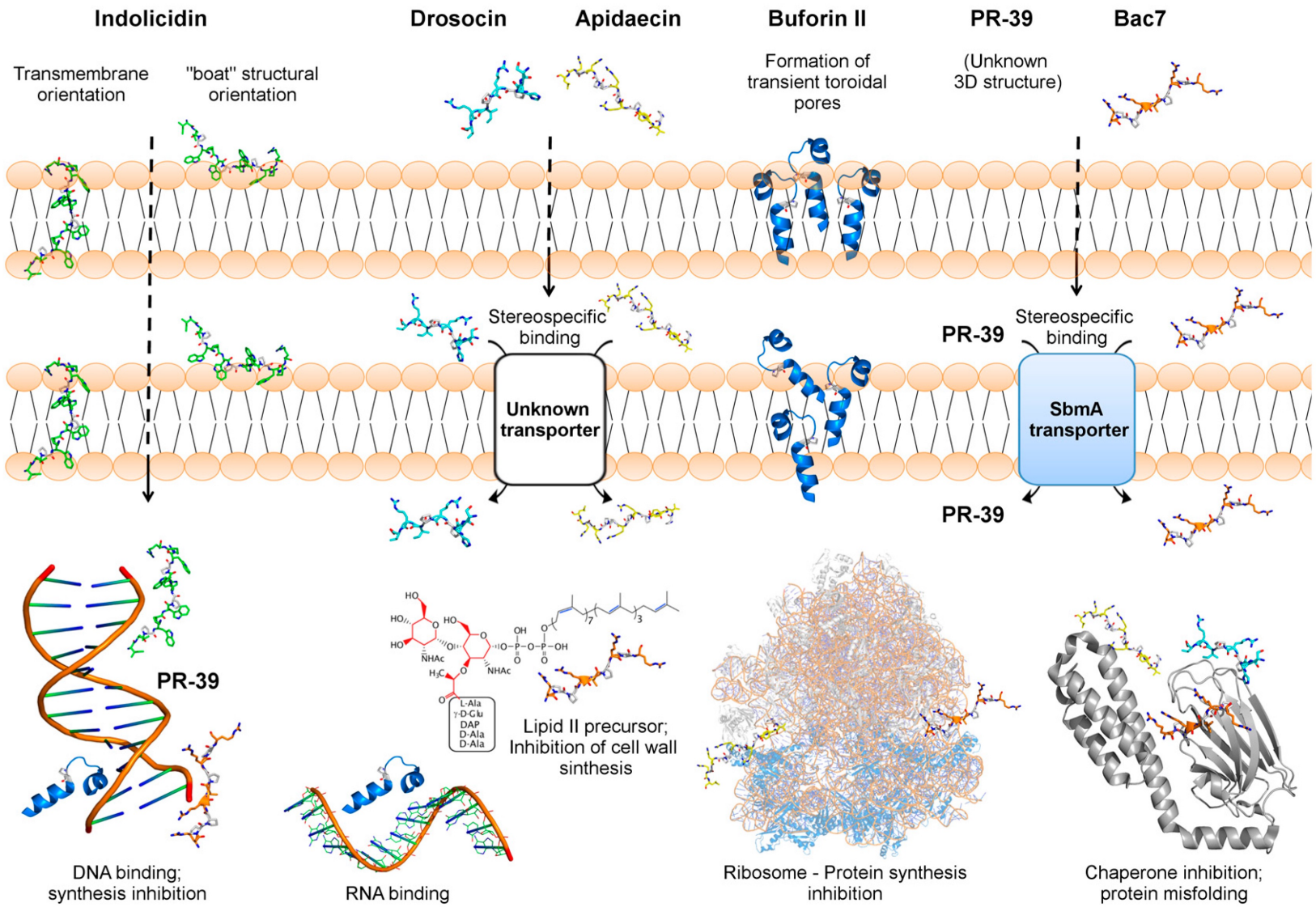






nanodisc
with bacteriorhodopsin
trimer

antimicrobial peptides (AMPs)



Cyclic peptides self-assemble into β-sheet type nanotubes

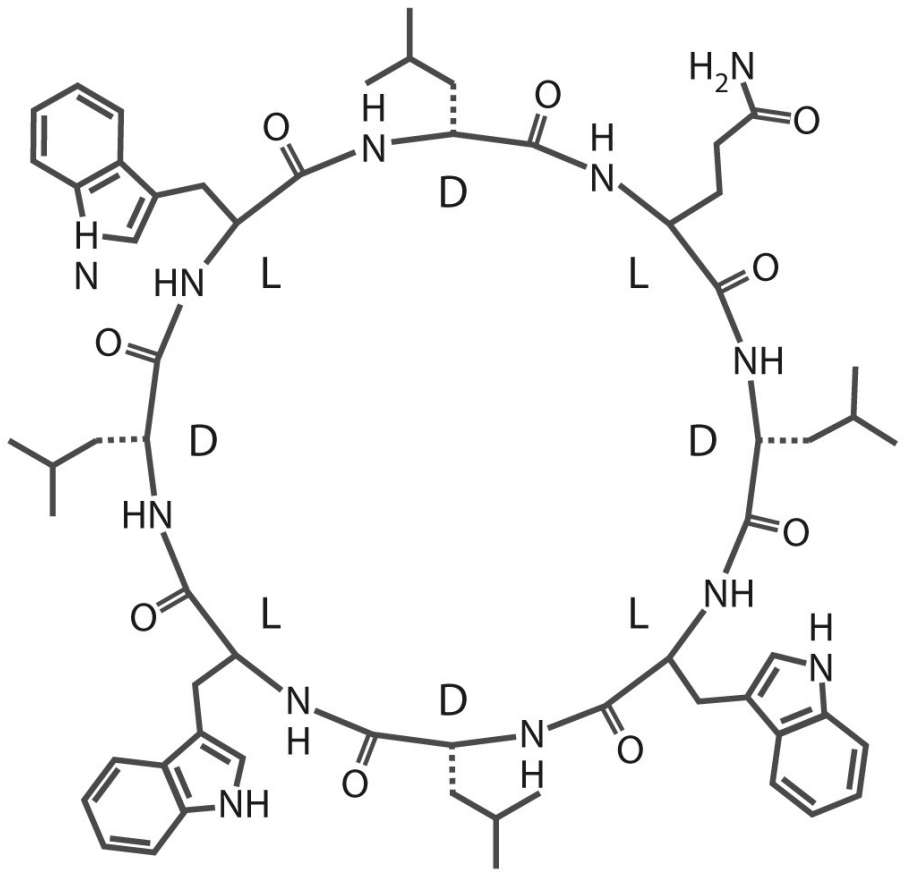
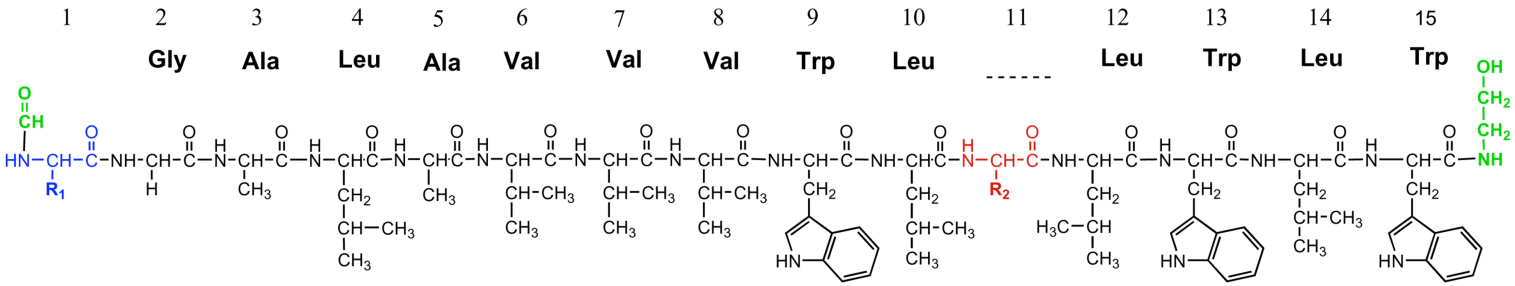


Figure 3.28a Cell Membranes (© Garland Science 2016)



Val Ile

R_1 = —CH(CH$_3$)(CH$_3$) —CH(CH$_3$)(CH$_2$CH$_3$)

Trp Phe Tyr

R_2 =

Gramicidin A Gramicidin B Gramicidin C

Cyclic peptides self-assemble into β-sheet type nanotubes

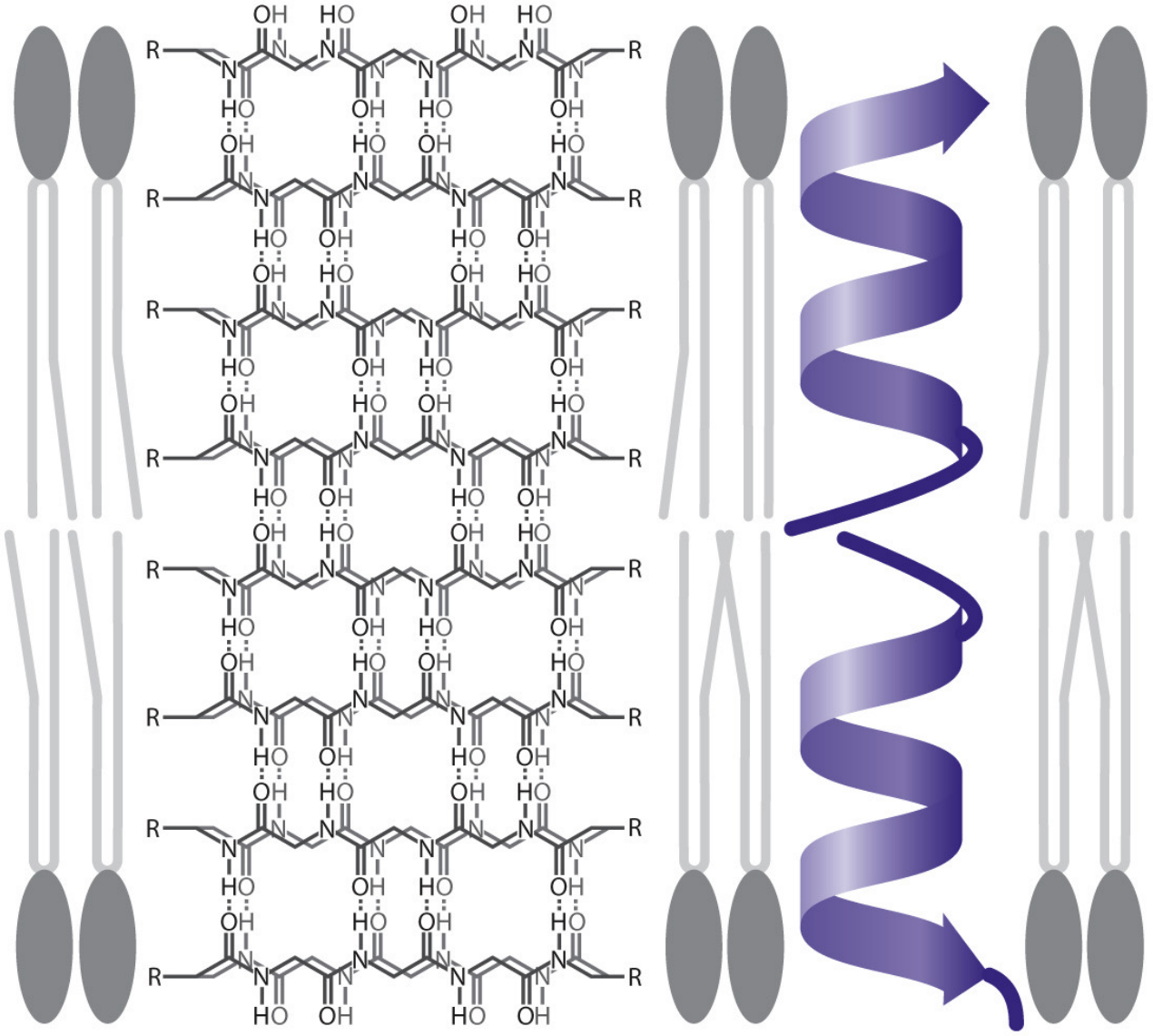
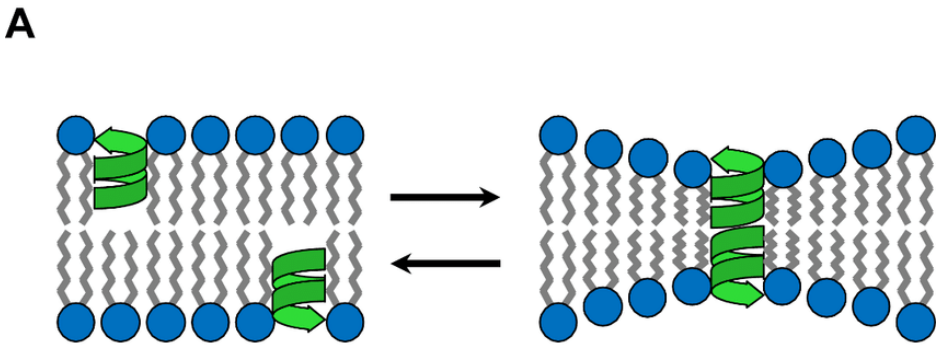
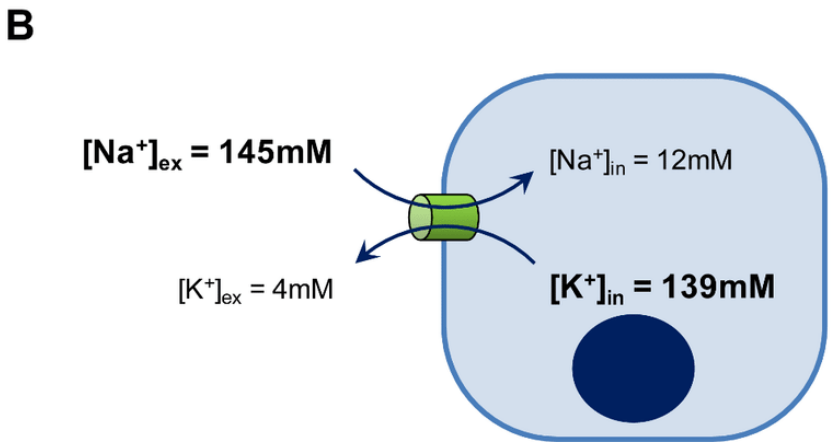
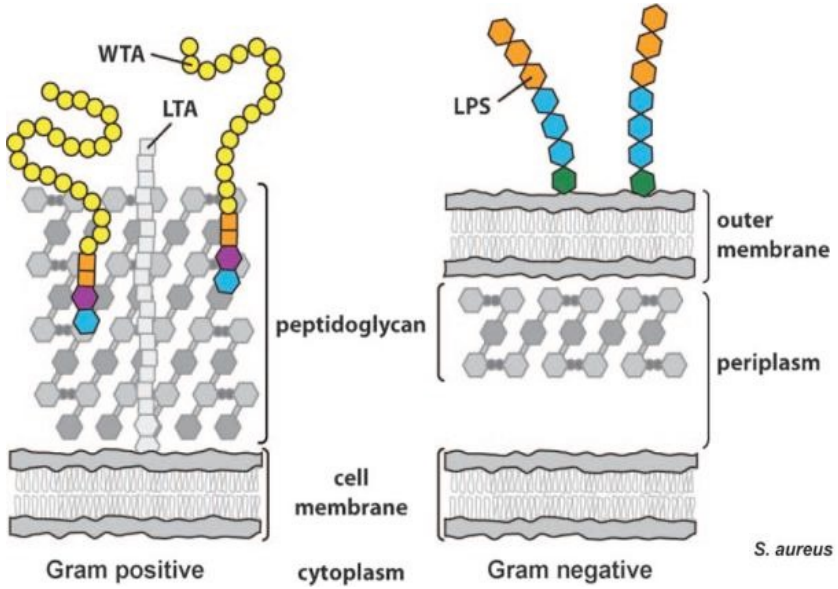


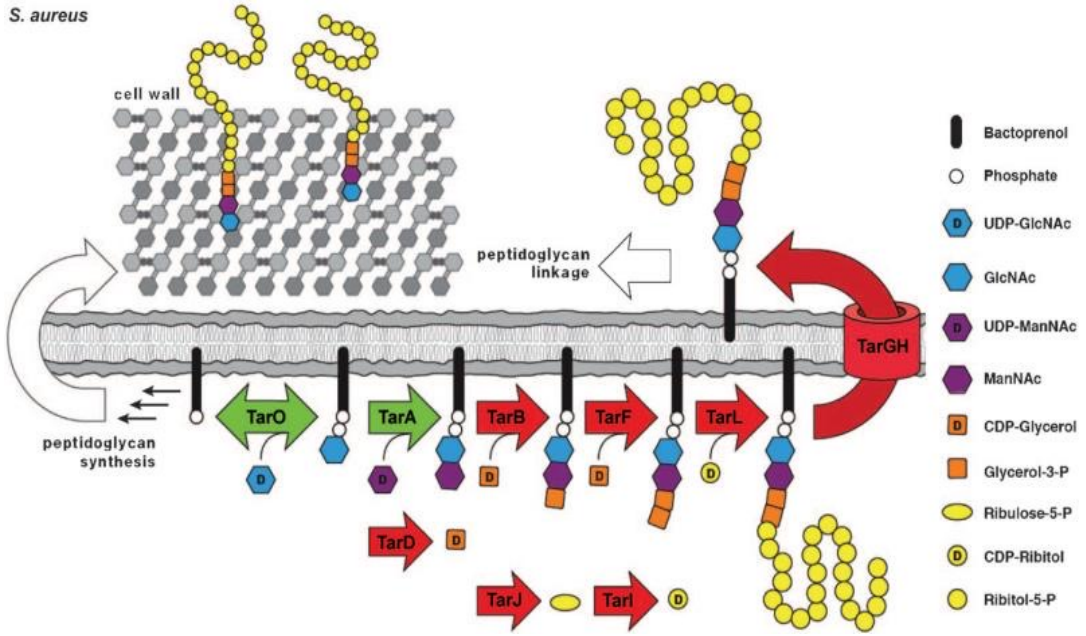
Figure 3.28b Cell Membranes (© Garland Science 2016)





Monotopic membrane proteins





teichoic acid biosynthesis protein F

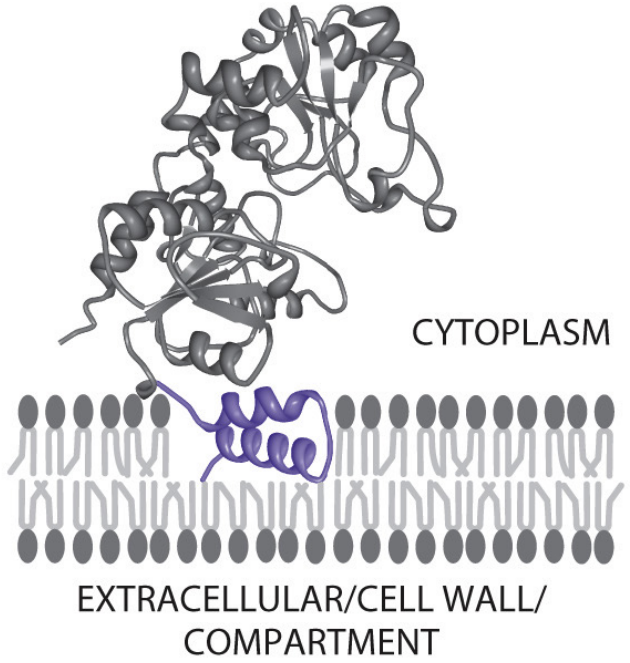
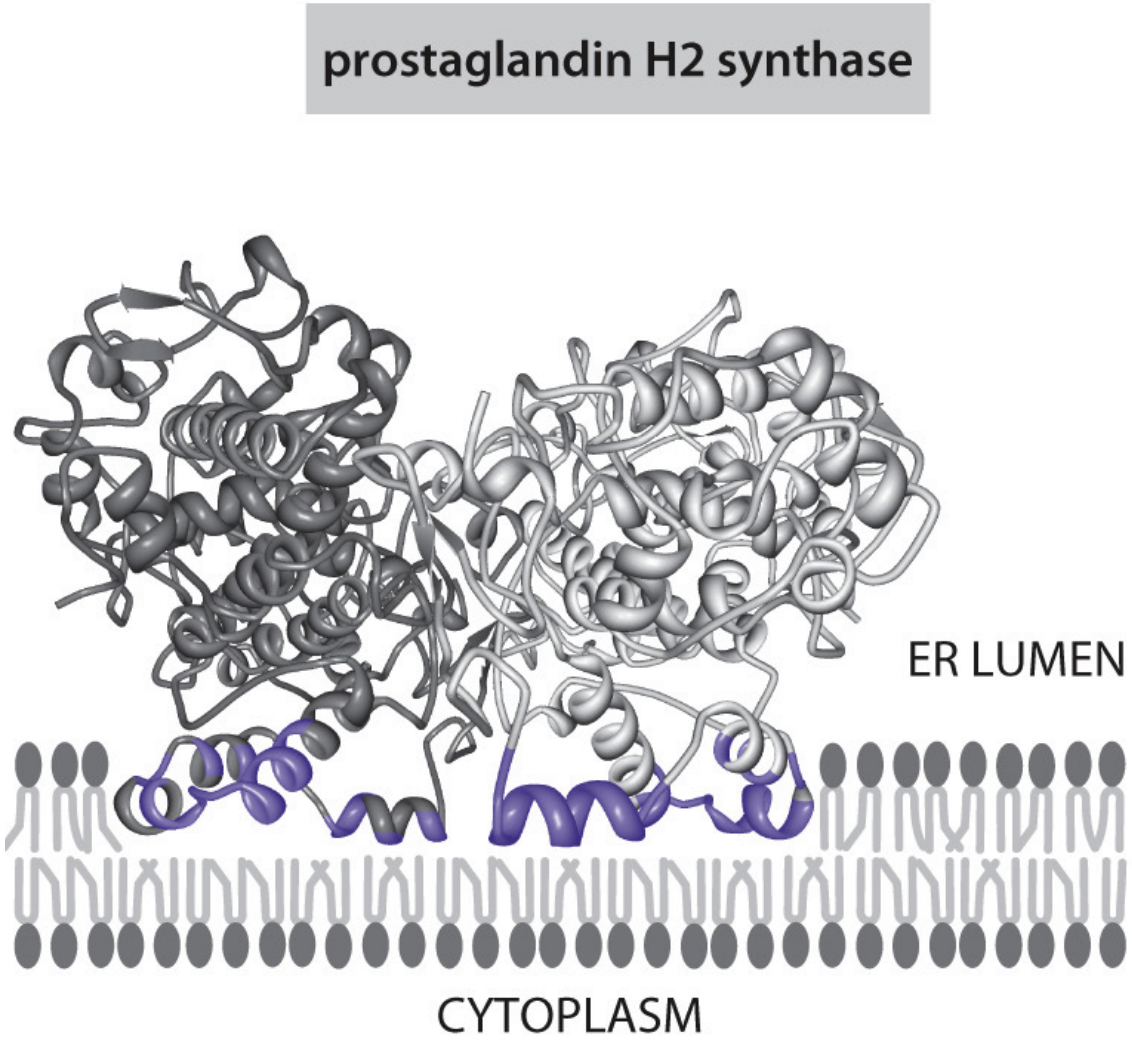


Figure 3.5 Cell Membranes (© Garland Science 2016)

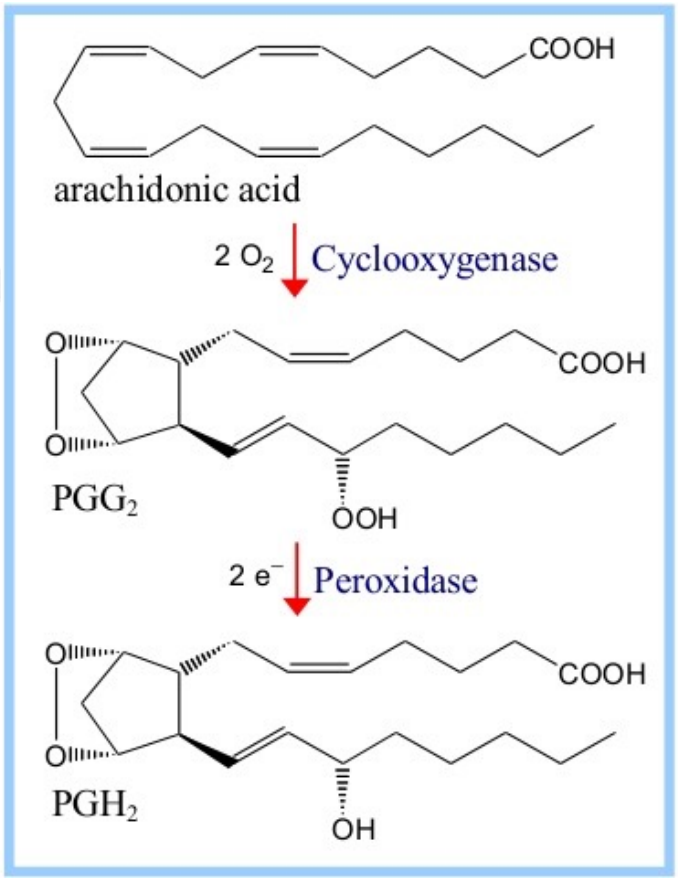
Monotopic membrane proteins



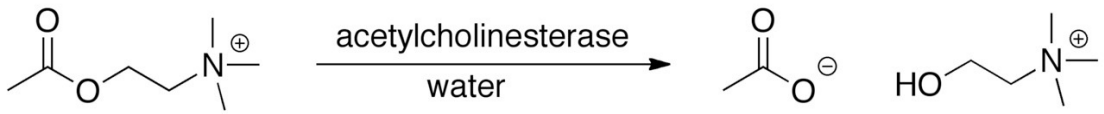
PGH_2 Synthase is a heme-containing **dioxygenase**, bound to ER membranes.

(A dioxygenase incorporates **O_2** into a substrate).

PGH_2 Synthase exhibits 2 activities: **cyclooxygenase** & **peroxidase**.



Lipid-anchored proteins



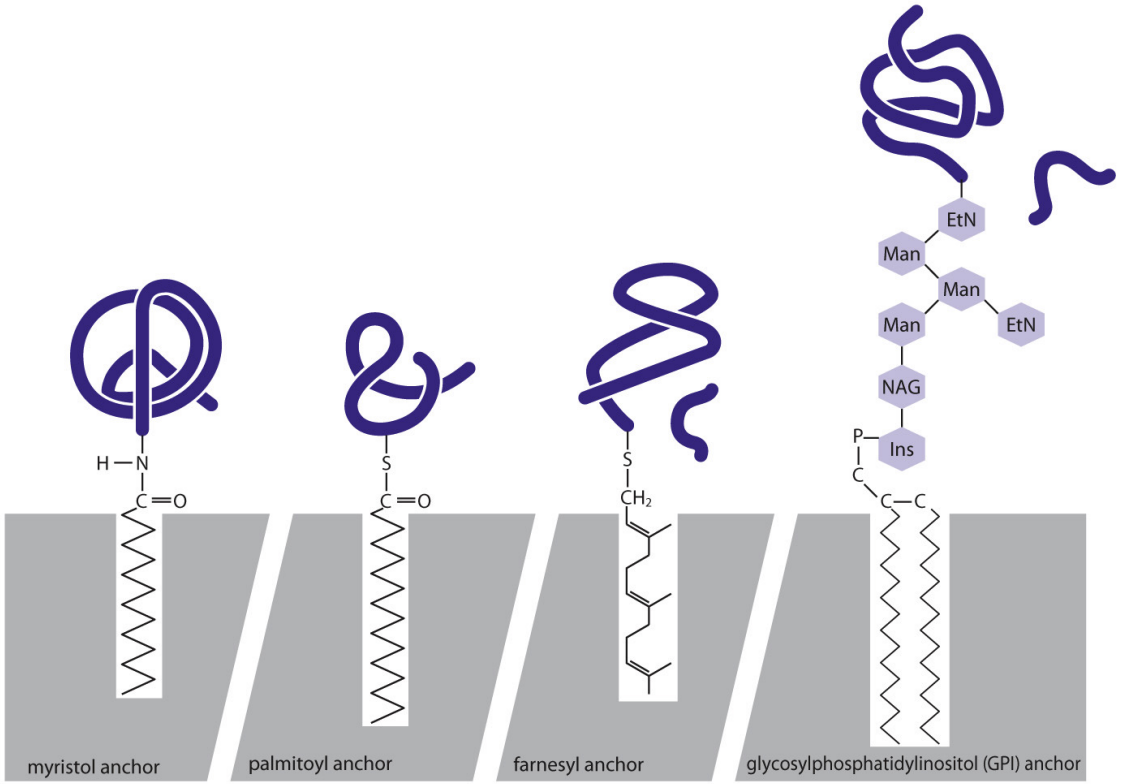


Figure 3.6a Cell Membranes (© Garland Science 2016)

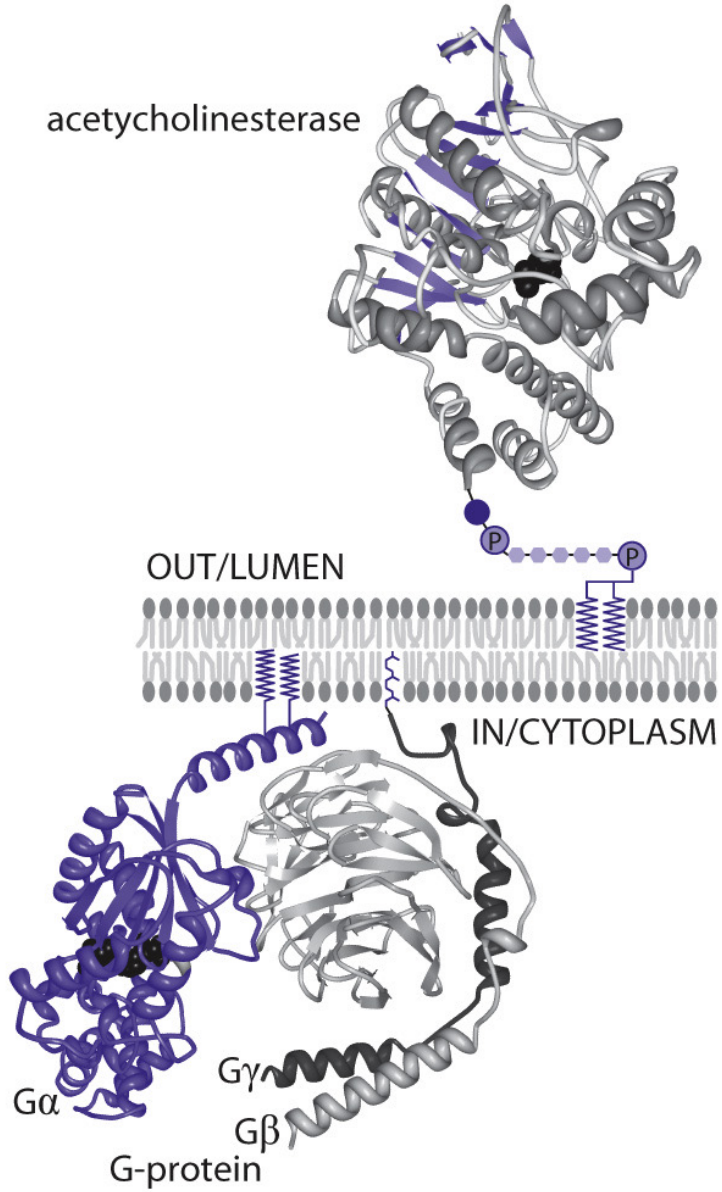


Figure 3.6b Cell Membranes (© Garland Science 2016)

Lipid anchor types and function

Table 3.2 Lipid anchor types and function

Common name	Lipid	Linker	Target membrane	Topology	Reversible	Function
Myristic acid	C14:0	N-term G (internal K)	Any	Cytosolic	No	Targeting
Palmitic acid	C16:0	Cys (K, S, T)	Any	Cytosolic	Yes	Sorting, trafficking
Farnesyl	C15	C-term (CaaX, CXC, XXCC)	Plasma	Cytosolic	No	Sorting, protein interaction
Geranylgeranyl	C20	C-term (CaaX, CXC, XXCC)	Plasma	Cytosolic	No	Sorting, protein interaction
GPI (SPI)	Glycerolipid (sphingolipid)	C-term	Plasma	Extracellular	Yes	Cell communication

GPI, glycosylphosphatidylinositol; SPI, sphingolipid inositol; C-term, C-terminal; N-term, N-terminal.

Lateral diffusion is faster for lipid-anchored than for transmembrane proteins

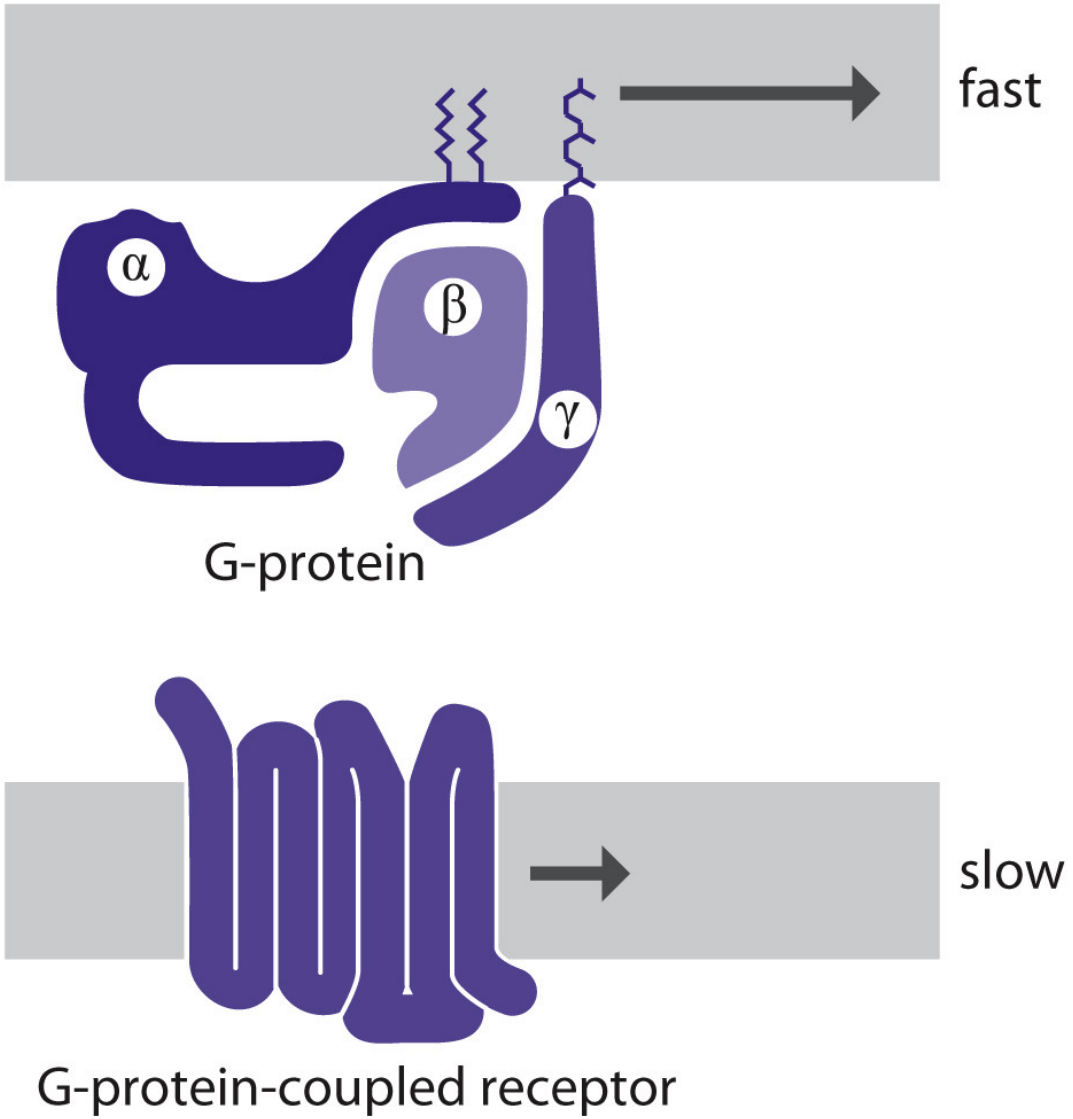


Figure 3.7 Cell Membranes (© Garland Science 2016)

Peripheral membrane proteins as subunits of protein complexes

ATP + H_2O + $maltose_{out}$ <—> ADP + phosphate + $maltose_{in}$

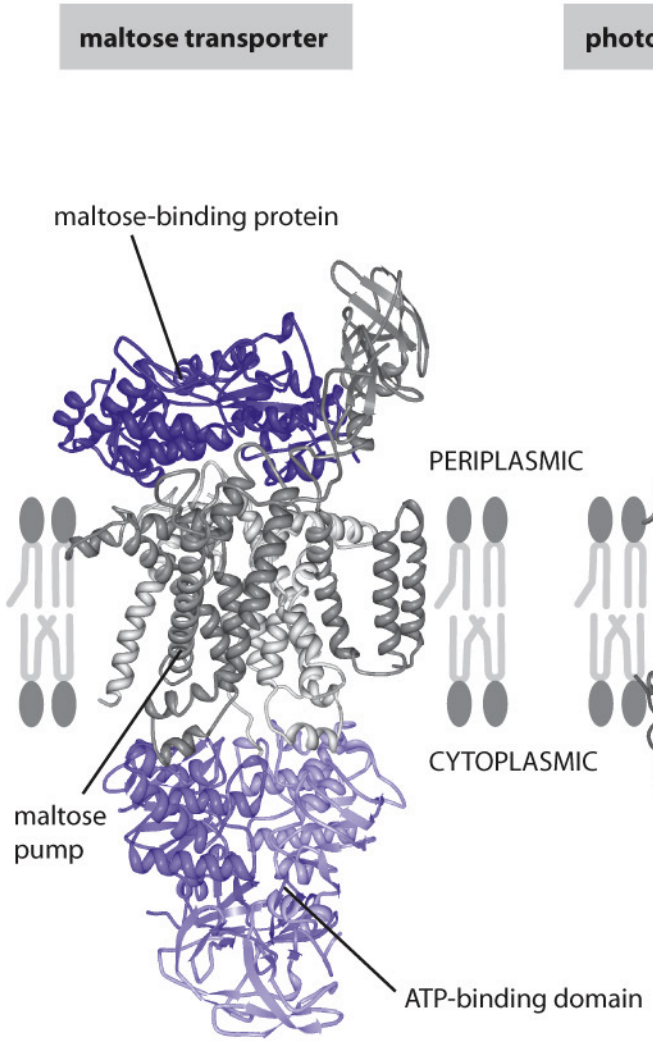
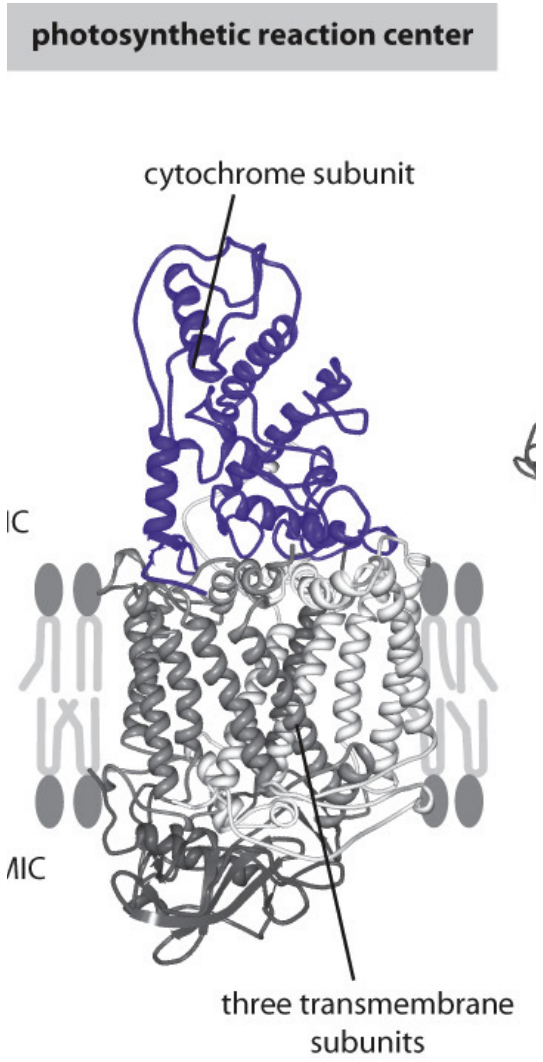


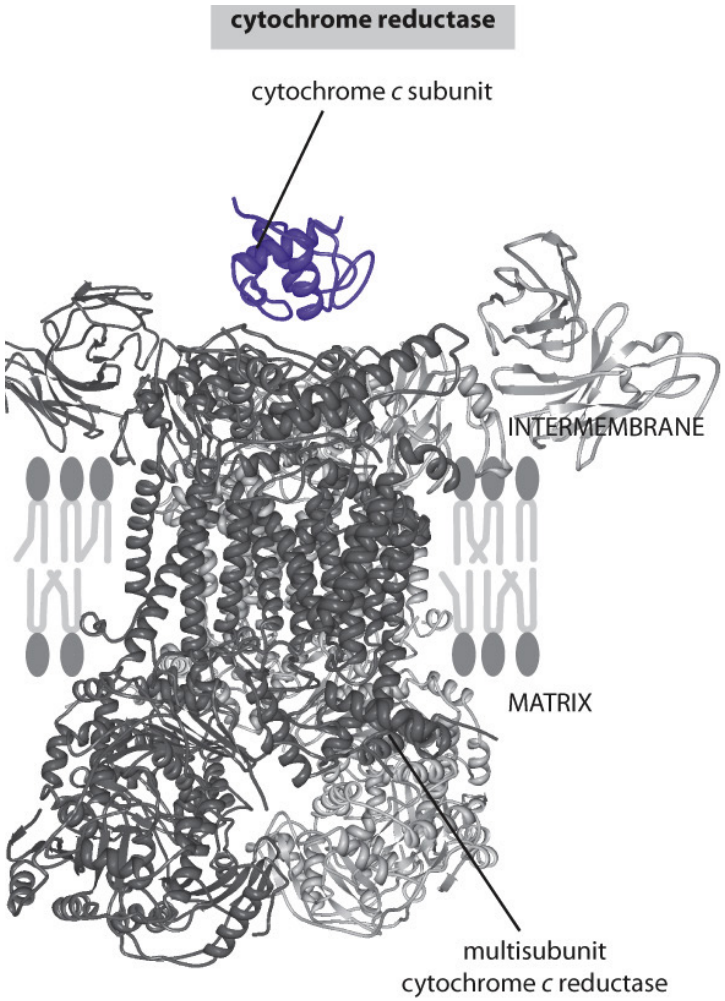
Figure 3.8 Cell Membranes (© Garland Science 2016)

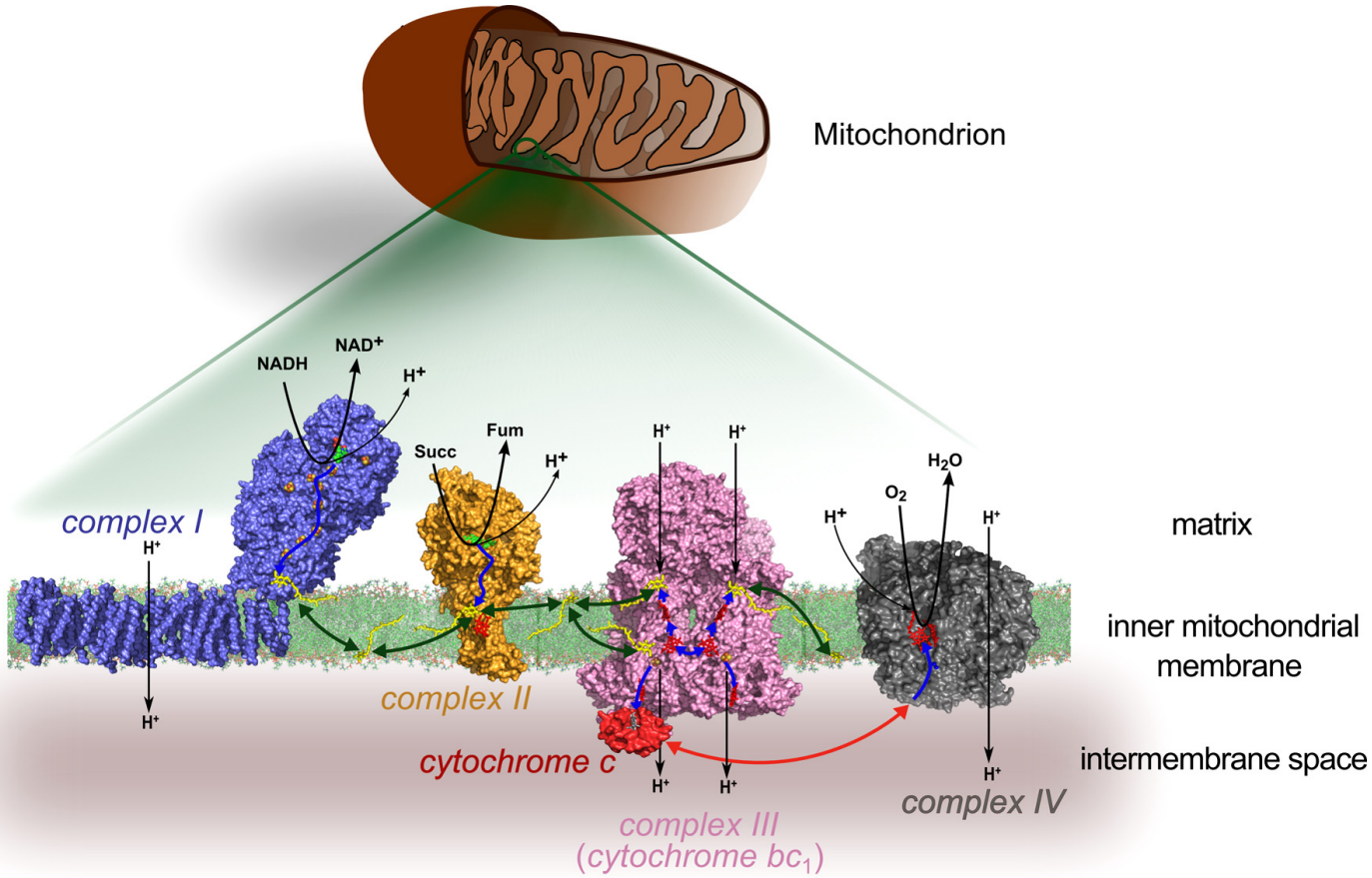
Peripheral membrane proteins as subunits of protein complexes

$$DPIA + h\nu \rightarrow DP^{*}IA \rightarrow DP^{+}I^{-}A \rightarrow DP^{+}IA^{-} \rightarrow D^{+}PIA^{-}$$



Peripheral membrane proteins as subunits of protein complexes





Peripheral lipid-binding membrane proteins

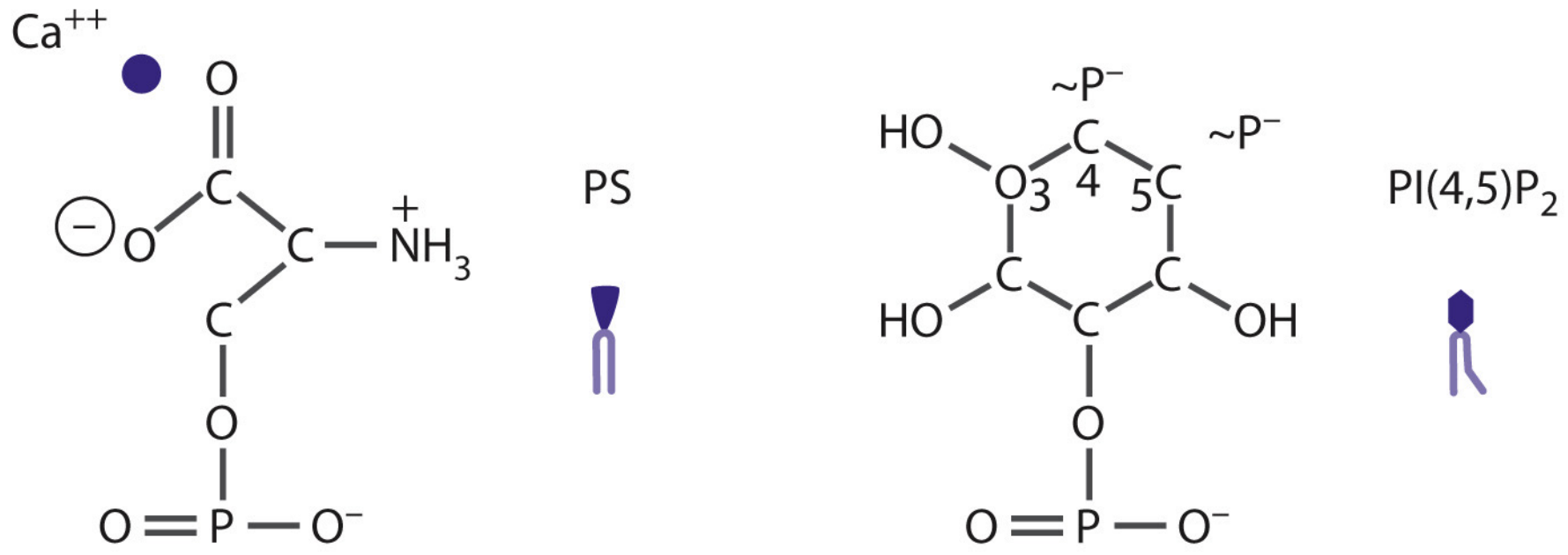


Figure 3.9b Cell Membranes (© Garland Science 2016)

Peripheral lipid-binding membrane proteins

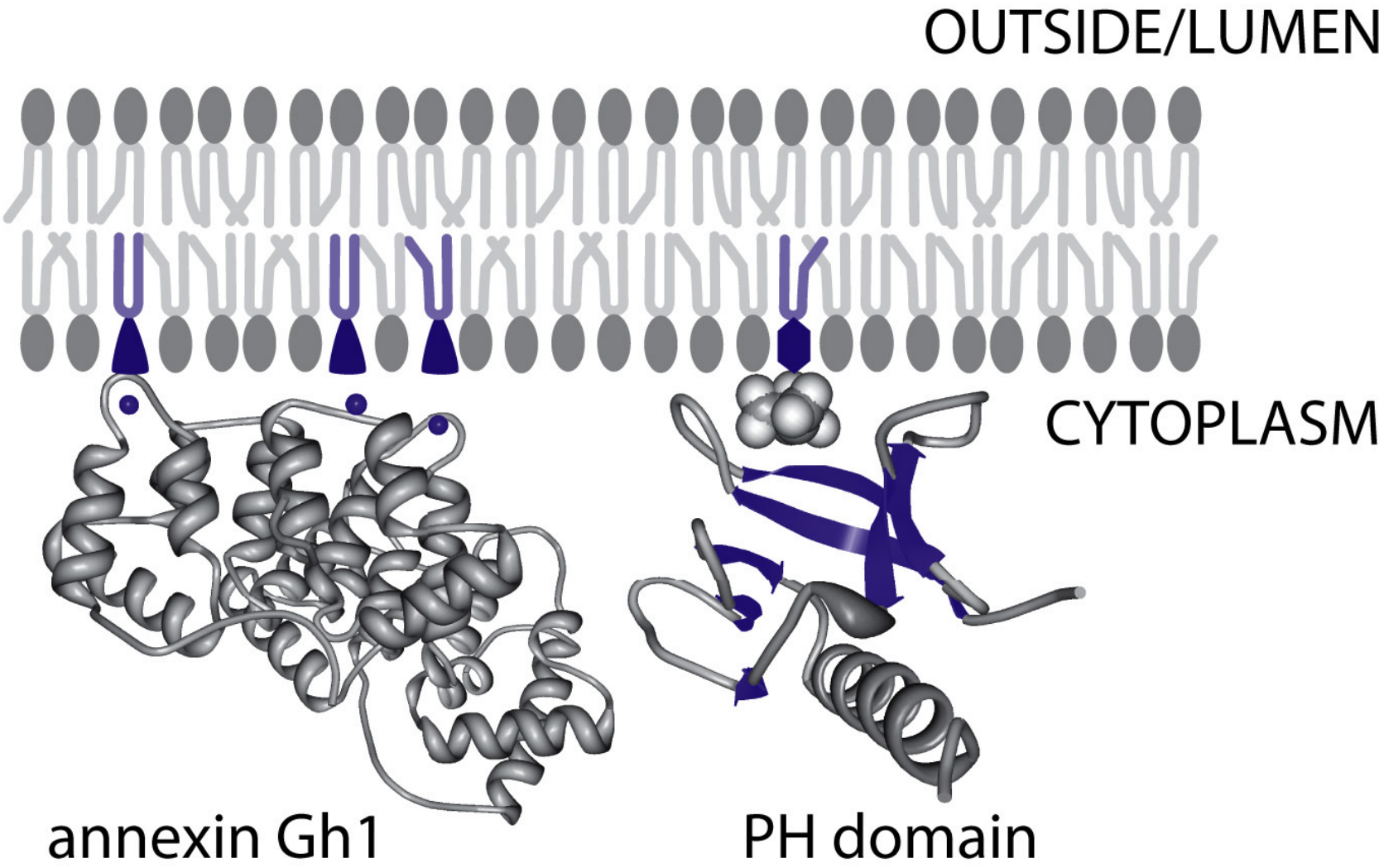
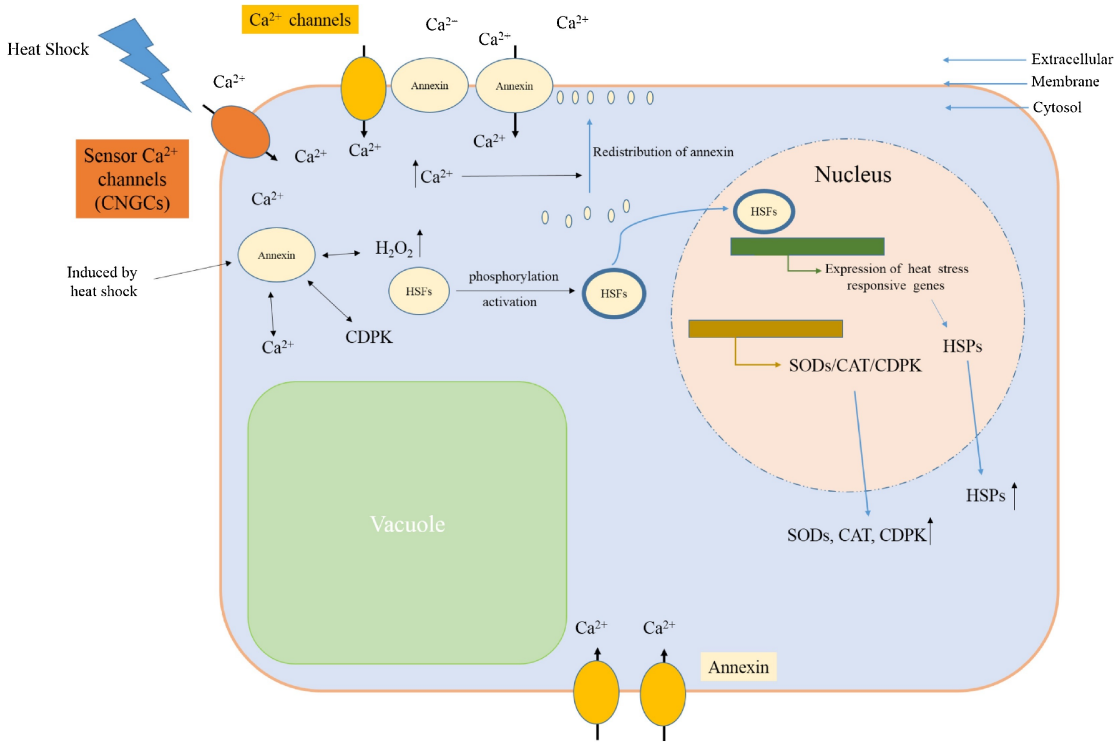
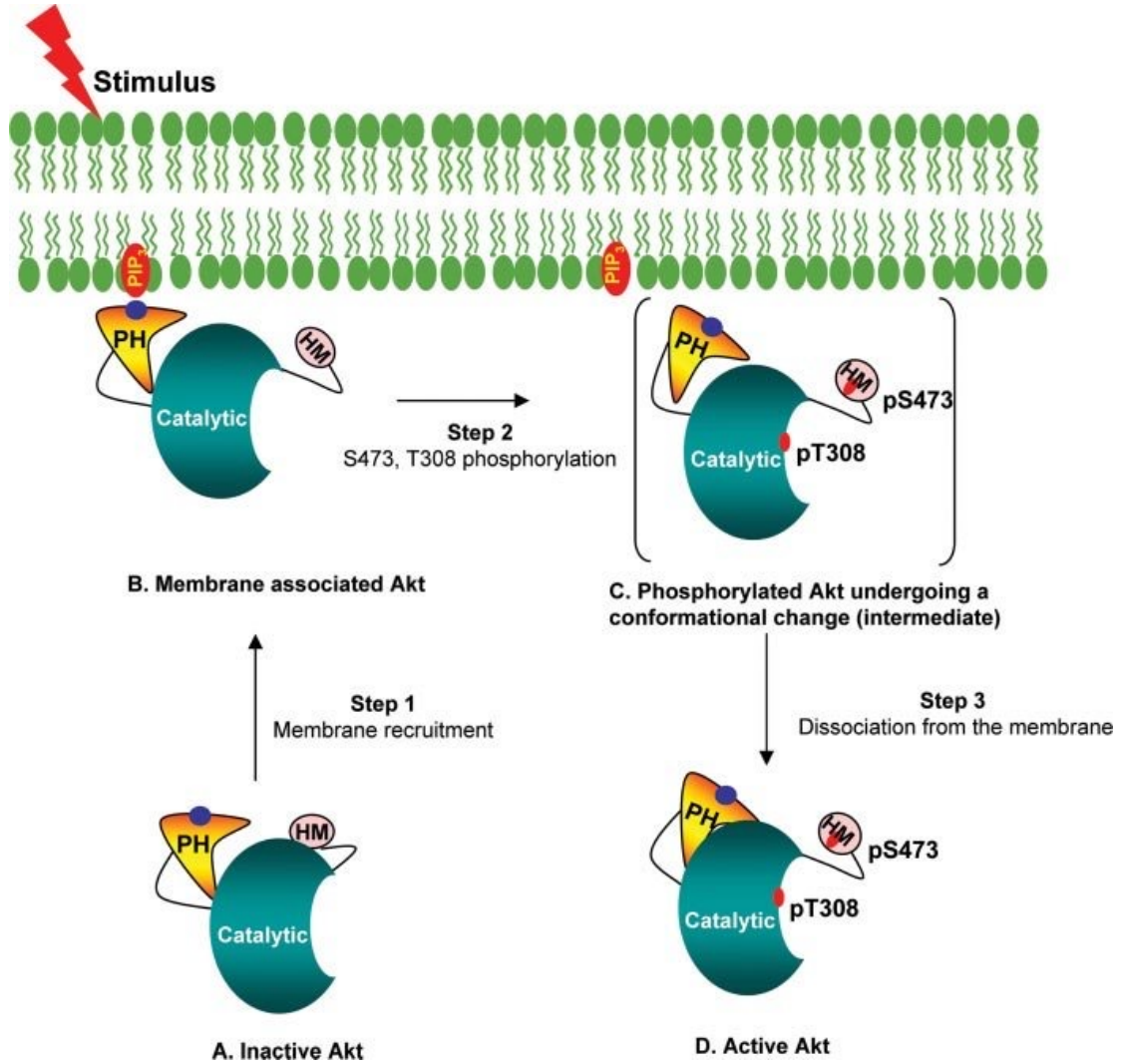


Figure 3.9a Cell Membranes (© Garland Science 2016)

pleckstrin homology

Action


Heat Shock
Ca2+ channels
Sensor Ca2+ channels (CNGCs)
Induced by heat shock
Annexin
Annexin
Annexin
Annexin
Extracellular
Membrane
Cytosol
Redistribution of annexin
Nucleus
HSFs
Expression of heat stress responsive genes
HSPs
SODs/CAT/CDPK
HSPs
SODs, CAT, CDPK
H2O2
HSFs
phosphorylation
activation
HSFs
CDPK
Vacuole
Annexin


Stimulus
PIP3
PIP3
PH
HM
Catalytic
B. Membrane associated Akt
Step 2
S473, T308 phosphorylation
pS473
pT308
C. Phosphorylated Akt undergoing a conformational change (intermediate)
Step 1
Membrane recruitment
Step 3
Dissociation from the membrane
A. Inactive Akt
D. Active Akt