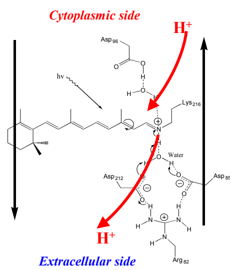
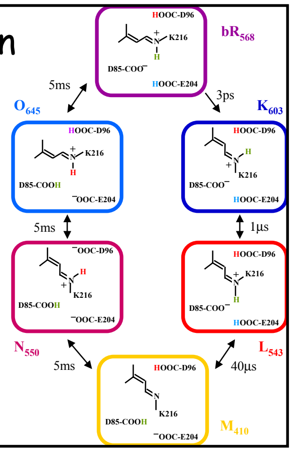
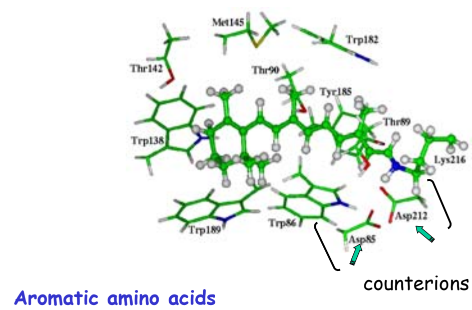
Bacteriorhodopsin



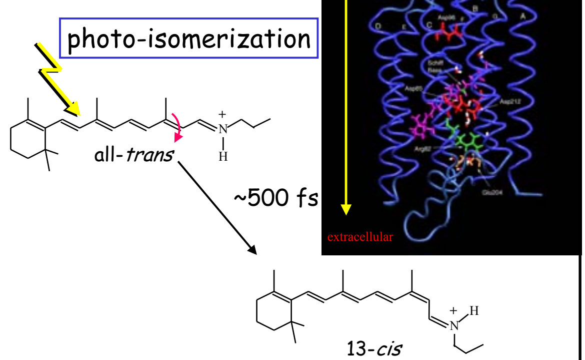


Photoabsorption -> isomerization
-> proton transfer

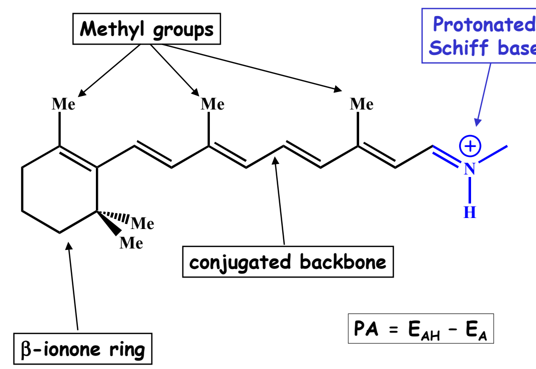
Chromophore-binding Pocket



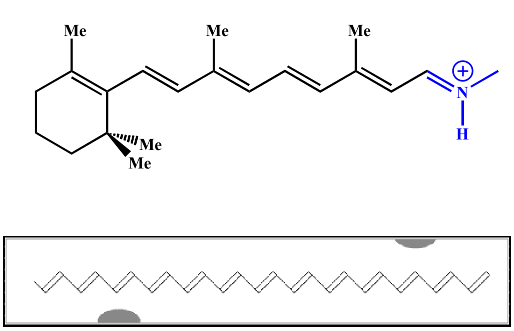
Proton accessibility!



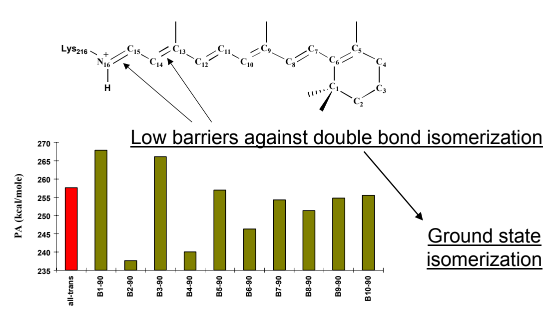
All structural details in the retinal chromophore are functionally important



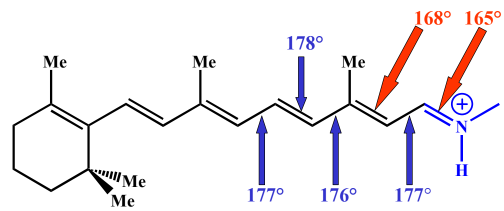
Delocalization of positive charge



Isomerization barriers in retinal

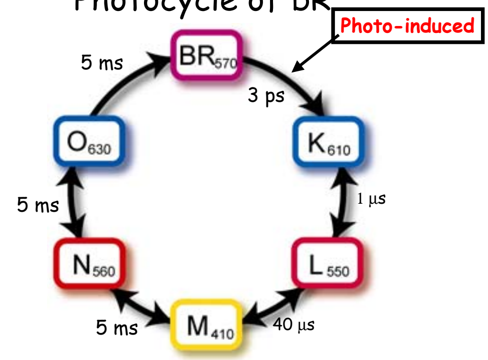


A twisted chromophore in bR?



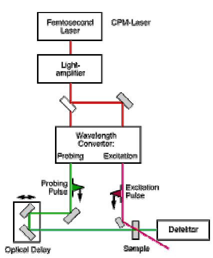
- A twisted chromophore is also experimentally reported.
- X-ray structures of bR report the twisted form of chromophore
- The twist is found around the terminal double bonds
- It may influence pK_a of the chromophore

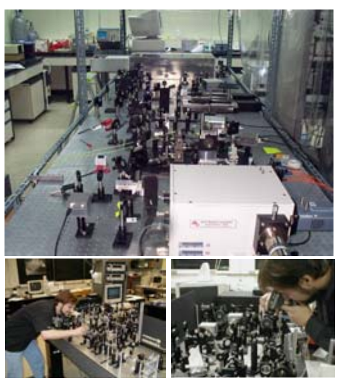
Photocycle of bR



All intermediates are trapped in low temperature and have been characterized by vibrational and absorption spectroscopy.

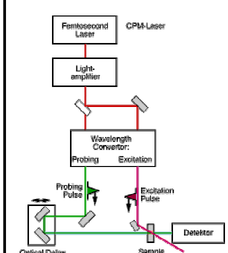
Ultrafast spectroscopy

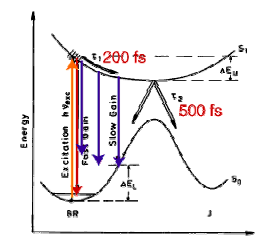




1 fs : 3 x 10^{-4} mm
1 ps : 3 x 10^{-1} mm

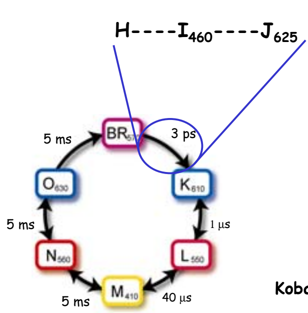
Ultrafast spectroscopy of bR

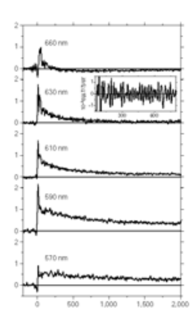




Ultrafast spectroscopy of bR

Femtosecond time resolution



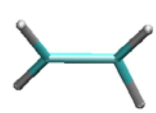


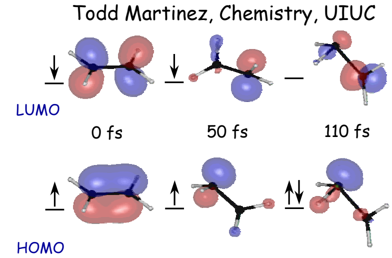
Kobayashi et al., Nature, Nov 2001

Calculation of the Excited state Dynamics of Photoactive Molecules by *ab initio* Techniques

Ab initio (First-Principles) dynamics of ethylene in vacuum

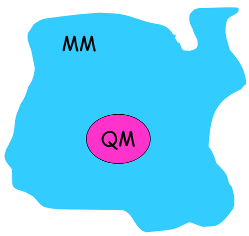
Todd Martinez, Chemistry, UIUC

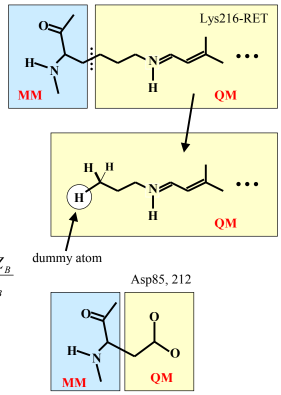




But what happens in the protein?

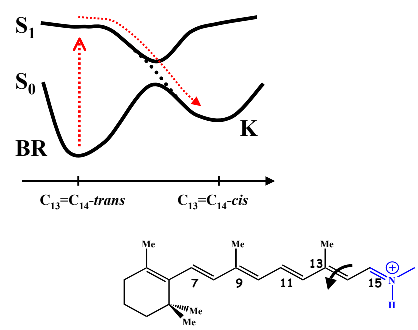
QM/MM calculations



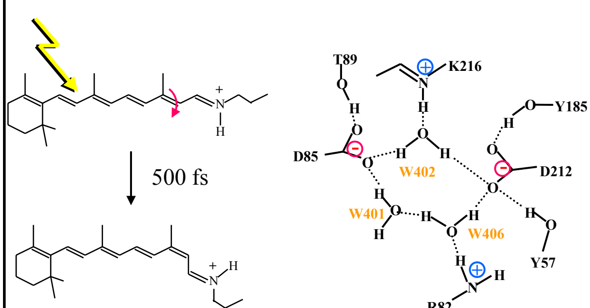


$$\hat{H} = \sum_i \frac{1}{2} p_i^2 + \sum_i \sum_A \frac{Z_A}{r_{iA}} + \sum_{i>j} \frac{1}{r_{ij}} + \sum_{A>B} \frac{Z_A Z_B}{r_{AB}} + \sum_i \sum_p \frac{q_p}{r_{ip}} + \sum_A \sum_p \frac{Z_A q_p}{r_{Ap}} + V^{MM}_{QM-MM} + V^{MM}_{MM}$$

Coupling of electronic excitation and conformational change in bR



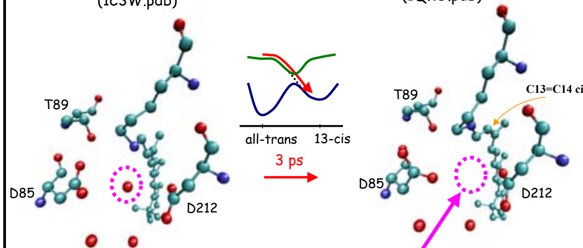
Hydrogen bond network in the retinal binding pocket



Water movement after the photoisomerization

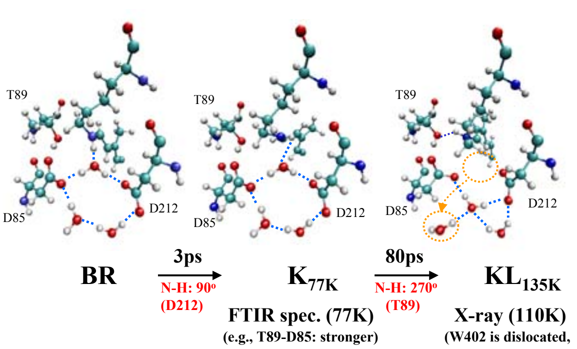
Structure of the ground state (1C3W.pdb)

Structure of the first intermediate (1QKO.pdb)



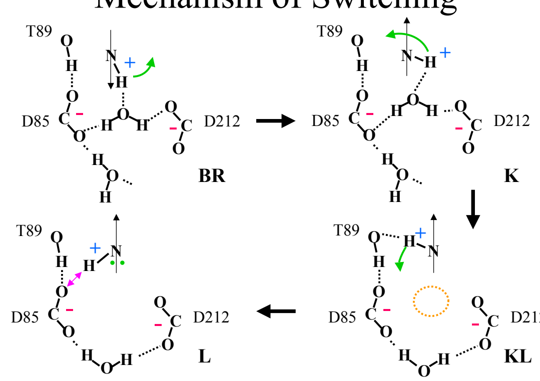
One water molecule is dislocated, but where is it?

Early intermediates of bR's photocycle

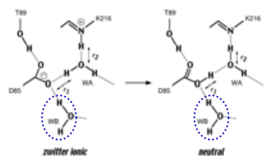


ΔE_{EX} (kcal/mol) -4.0 (-1.8) -2.9 (-1.8)

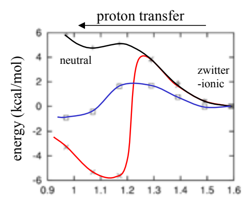
Mechanism of Switching



Role of water in proton transfer



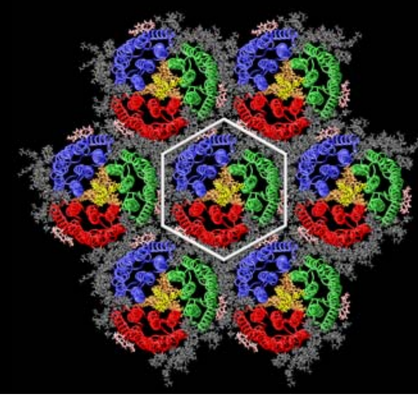
QM/MM calculation



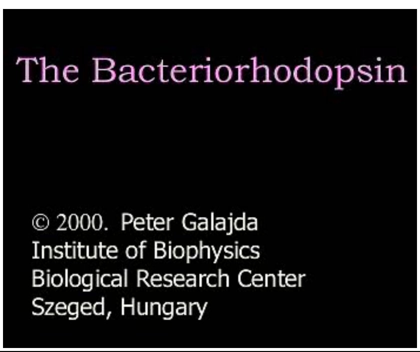
Rearrangement of the hydrogen-bond network can induce the proton transfer.

isomerization

THE PURPLE MEMBRANE



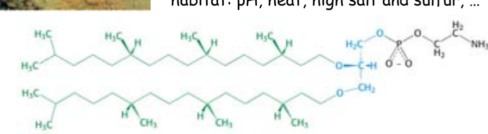
The Purple membrane of *Halobacterium salinarum*



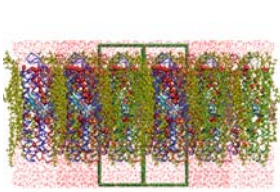
Archaeal Membranes

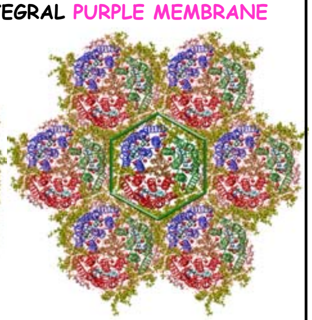
- Branched (less vulnerable to oxidation)
- Etheric bridge, not esteric (less sensitive to hydrolysis)
- Inverted glycerol stereochemistry

Higher resistance to harsh conditions of their habitat: pH, heat, high salt and sulfur, …



MODELING OF THE INTEGRAL PURPLE MEMBRANE

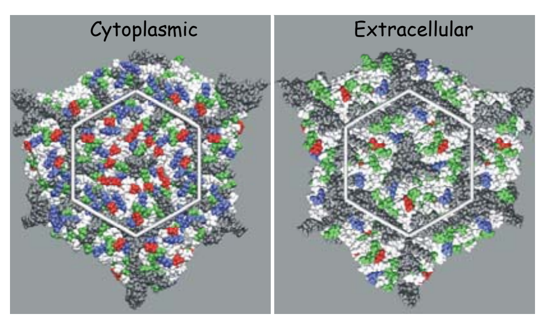




Bacteriorhodopsin trimer
Internal **water** molecules
Squalene molecules
Bulk *water*

Retinal chromophores
Intra-trimer lipids
Inter-trimer lipids

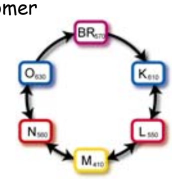
Charge distribution at different faces of the purple membrane



Basic : **Acidic** : **Polar** : **Lipids**

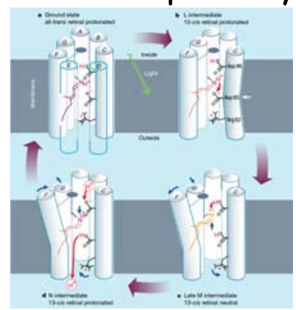
Kinetics of the photocycle is dependent on the lipid composition of the membrane

We have 10 molecules of lipid per bR monomer



PGP and squalene are necessary for the recovery of normal kinetics of the photocycle after detergent treatment of PM.

Helix dislocation at late stages of the photocycle



Possible involvement of lipid-protein interaction in the photocycle