Bacteriorhodopsin

Photoabsorption → isomerization
→ proton transfer

Chromophore-binding Pocket

Aromatic amino acids

Proton accessibility!

All structural details in the retinal chromophore are functionally important

Delocalization of positive charge

Isomerization barriers in retinal

Ground state isomerization
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.

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\[
\begin{align*}
\text{Me} & \quad 178^\circ \\
\text{Me} & \quad 177^\circ \\
\text{Me} & \quad 176^\circ \\
\text{Me} & \quad 177^\circ \\
\text{N} & \quad 165^\circ \\
\text{Me} & \quad 168^\circ \\
\end{align*}
\]

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Photocycle of bR

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

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Ultrafast spectroscopy

1 fs: $3 \times 10^{-4}$ mm
1 ps: $3 \times 10^{-1}$ mm

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Ultrafast spectroscopy of bR

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Ultrafast spectroscopy of bR

Femtosecond time resolution

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Calculation of the Excited state Dynamics of Photoactive Molecules by \textit{ab initio} Techniques

\textit{Ab initio} (First-Principles) dynamics of ethylene in vacuum

But what happens in the protein?
QM/MM calculations

Coupling of electronic excitation and conformational change in bR

Hydrogen bond network in the retinal binding pocket

Water movement after the photoisomerization

Early intermediates of bR’s photocycle

Mechanism of Switching

\[
H = \sum_{r} \frac{1}{r} \rho_{r} + \frac{1}{2} \sum_{r} \frac{Z_{r}^{2}}{r_{a}^{2}} + \sum_{r} \frac{Z_{r} \Delta_{r}}{r_{a}^{2}}
\]

\[
+ \sum_{r} \frac{Z_{r} \Delta_{r}}{r_{a}^{2}} + \frac{1}{2} \sum_{r} \frac{Z_{r} \Delta_{r}}{r_{a}^{2}}
\]

\[
+ \sum_{r} \frac{Z_{r} \Delta_{r}}{r_{a}^{2}} + \frac{1}{2} \sum_{r} \frac{Z_{r} \Delta_{r}}{r_{a}^{2}}
\]

\[
+ \sum_{r} \frac{Z_{r} \Delta_{r}}{r_{a}^{2}} + \frac{1}{2} \sum_{r} \frac{Z_{r} \Delta_{r}}{r_{a}^{2}}
\]
Role of water in proton transfer

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

The Purple membrane of *Halobacterium salinarum*

The Bacteriorhodopsin

© 2000. Peter Galajda
Institute of Biophysics
Biological Research Center
Szeged, Hungary

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

Cytoplasmic
Extracellular

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.