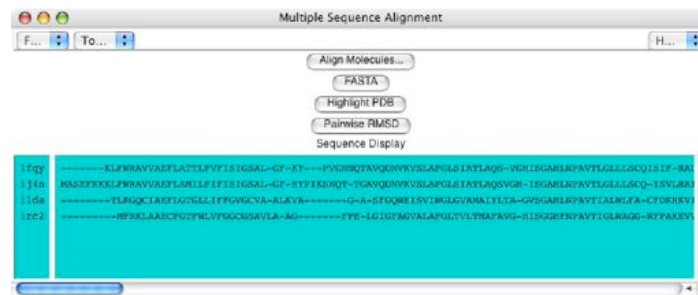
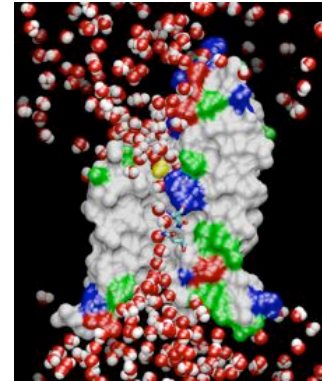
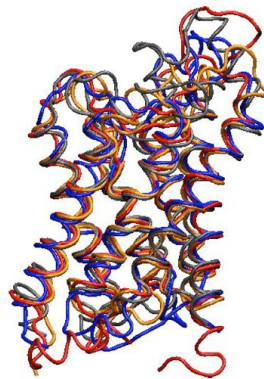


# Sequence and Structure Alignment - Illustrated for the Water Channel Aquaporin



```
Multiple Sequence Alignment
F... To... H...
Align Molecules...
FASTA
Highlight PDB
Pairwise RMSD
Sequence Display

119g -----KLFWRVAVSTLATTLPVYISIGSAL-GY-ET---FVGRNINVDYVYVSLAPVLEIETLADG-PKIDGARIYAVTIGLLGKQIISIF-RAI
119b MADEFKLLWRVAVSTLARIIFETISIGSAL-GY-ETVETADQV-YSAYQVYVYVSLAPVLEIETLADQVGR-IGARLIRYAVTIGLLGKQIISIF-RAI
119a -----ELRQCIASPTIGLLIFPQVCA-ALAVK-----GQ-A-STQVMEISYIMGLVYRNALVLTG-SPGARLIRYAVTIGLRFKFA-CTQKSEV
1002 -----DRKLAACVDFPMLYVQCSAVLA-AG-----FPE-LELUTMVALAPVETVLTAYAVG-RIIDGRIRYAVTIGLIRAGG-KYFAKSEV
```

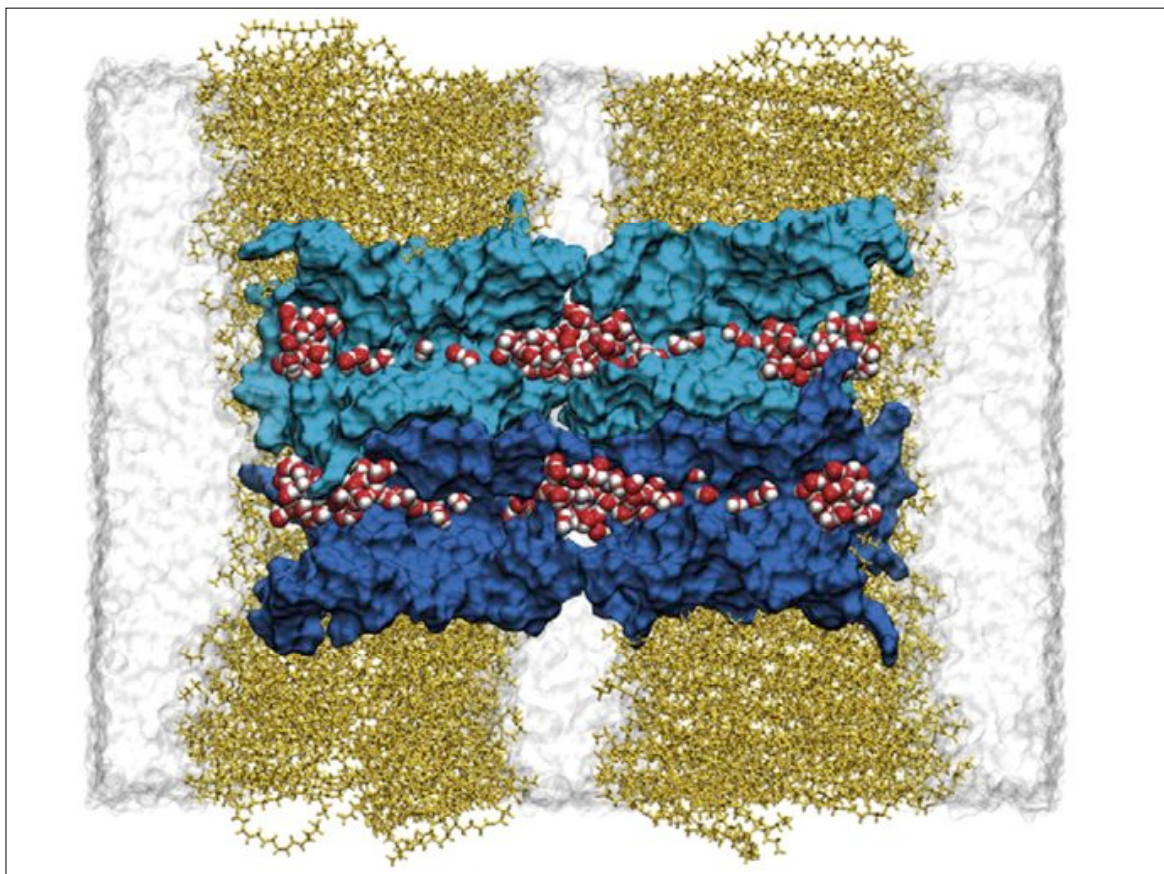
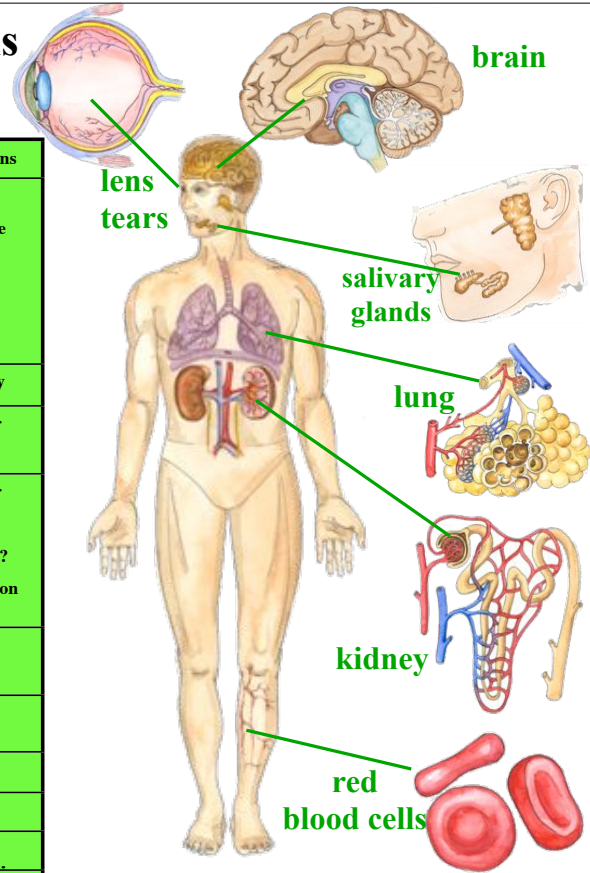
## Physical Bioinformatics - A Case Study

Sequence and structure information are the bedrock on which an understanding of cellular functions and the underlying physical mechanisms can be built. This lecture illustrates how the two sources of information are combined to investigate by means of the program VMD function and mechanism of the aquaporin family of membrane channels that transport water and certain small solutes across cell walls. Introducing first the key architectural features of a single aquaporin, structures and sequences of four aquaporins are aligned and common features recognized. The shared and distinct features are examined closely and used as guideposts leading quickly to key questions regarding the mechanism underlying aquaporin's efficient conduction and selection.



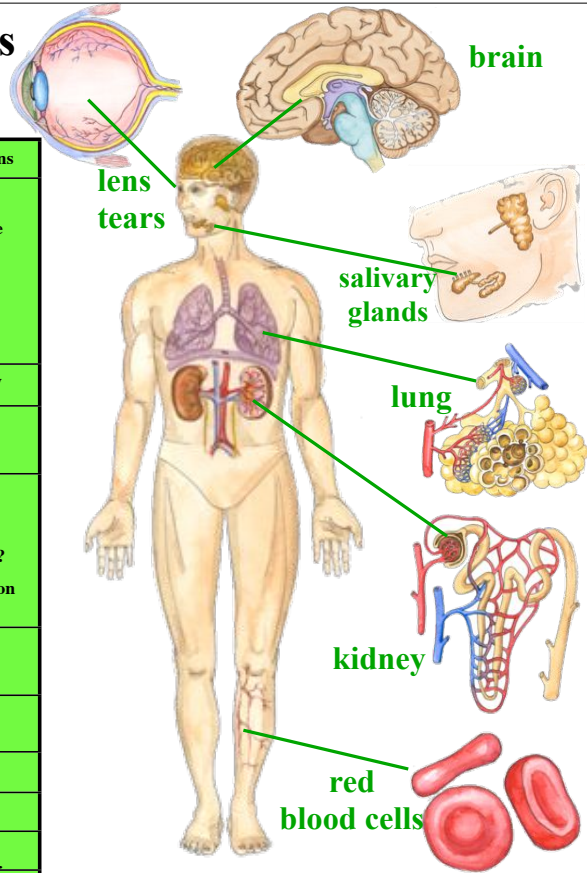
# Water and Glycerol Channels in the Human Body

Aquaporin-0	Eye: lens fiber cells	Fluid balance of the lens
Aquaporin-1	Red blood cells Kidney: proximal tubules Eye: ciliary epithelium Brain: choroid plexus Lung: alveolar epithelial cells	Osmotic protection Concentration of urine Aqueous humor Production of CSF Alveolar hydration
Aquaporin-2	Kidney: collecting ducts	ADH hormone activity
Aquaporin-3	Kidney: collecting ducts Trachea: epithelial cells	Reabsorption of water Secretion of water
Aquaporin-4	Kidney: collecting ducts Brain: ependymal cells Brain: hypothalamus Lung: bronchial epithelium	Reabsorption of water CSF fluid balance Osmosensing function? Bronchial fluid secretion
Aquaporin-5	Salivary glands Lacrimal glands	Production of saliva Production of tears
Aquaporin-6	Kidney	Very low water permeability!
Aquaporin-7	Testis and sperm	
Aquaporin-8	Testis, pancreas, liver	
Aquaporin-9	Testis	
Additional members are suspected to exist.		



# Water and Glycerol Channels in the Human Body

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Aquaporin-8	Testis, pancreas, liver	
Aquaporin-9	Leukocytes	
Additional members are suspected to exist.		



## Functionally Important Features of Aquaporins

- Water, gas, and glycerol transport
- Exclusion of ions and protons
- Tetrameric arrangement in membrane

Aquaporins of known structure:

**GlpF** – E. coli glycerol channel (aquaglyceroporin)

– Fu, et al., Science (2000)

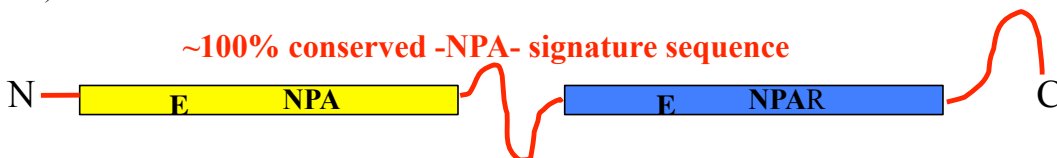
**AQP1** – Mammalian aquaporin-1 (pure water channel) -Sui et al, Nature (2001)

AQP1 - Bovine - Murata et al, Nature (2000)

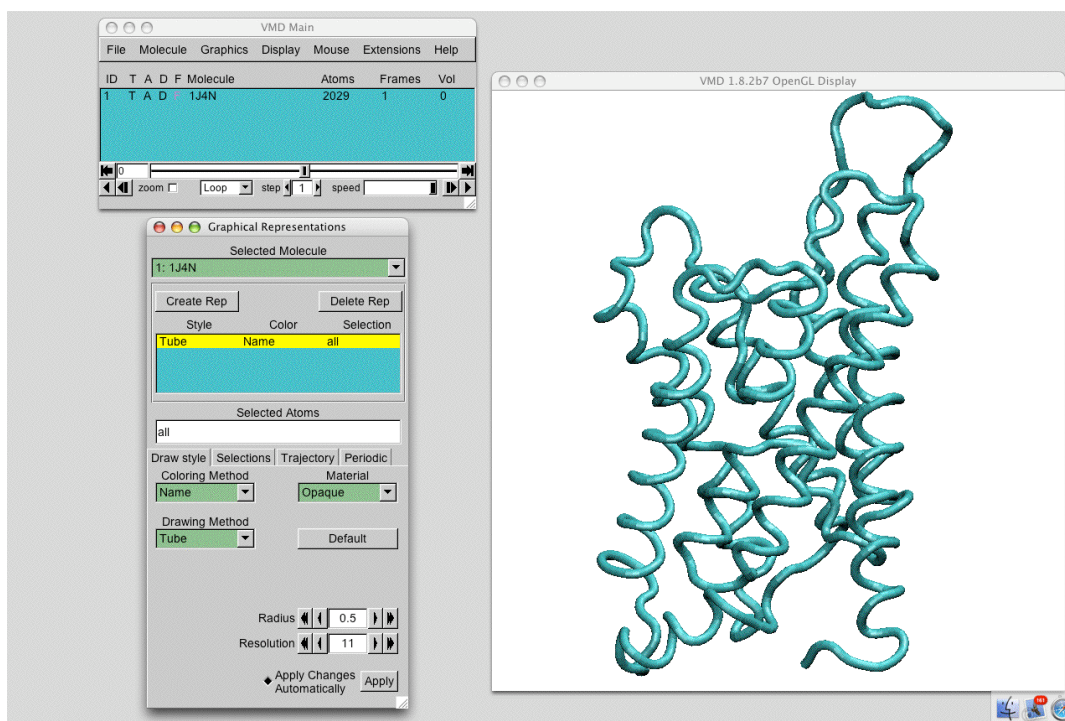
AQPZ - E. coli water channel - Savage et al, PLOS Biol (2003)



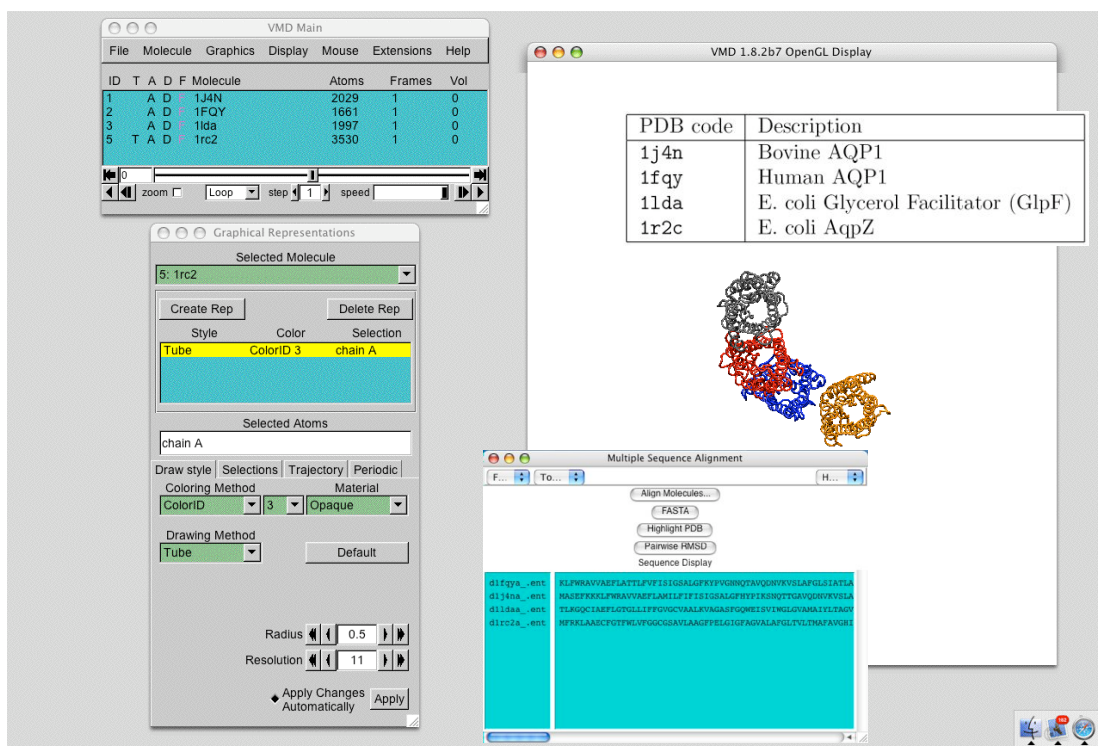
**~100% conserved -NPA- signature sequence**



# Load Aquaporin 1J4N into VMD



# Load Aquaporins 1j4n, 1fqy, 1lda, 1rc2 into VMD



# Aligning Structures and Sequences

The screenshot displays the VMD (Visual Molecular Dynamics) interface. The main window shows a 3D ribbon representation of a protein structure, colored by chain (chain A is blue, chain B is red, chain C is yellow, chain D is green). The 'VMD Main' window contains a table of loaded molecules:

ID	T	A	D	F	Molecule	Atoms	Frames	Vol
1	A	D	F		1J4N	2029	1	0
2	A	D	F		1FQY	1661	1	0
3	A	D	F		1lda	1997	1	0
5	T	A	D	F	1rc2	3530	1	0

The 'Graphical Representations' window shows the selected molecule '5: 1rc2' with a 'Tube' style and 'ColorID 3' for 'chain A'. The 'Multiple Sequence Alignment' window shows a sequence alignment of the four proteins, with the 'Sequence Display' option selected.

# Comparing Structures by Similarity - Q Value

This screenshot shows the same VMD interface as above, but with the 'Multiple Sequence Alignment' window's 'Highlight Style' menu open. The 'Q per residue' option is selected, which highlights the alignment based on sequence identity. The 'Molecule Coloring' menu is also open, showing 'Q per residue' as the selected option.

The 'Multiple Sequence Alignment' window shows the following sequence alignment:

```

1f4y -----KLFWRVAVATLTTLPVYTSISDAL-GT-ET---PVGSRITVGGVYVYVSLAPLSEITFLAGS-VGRISGARLWVAVTGLLGGQIISIF-RAI
1j4n -----KASFPKXKLFWRVAVATLTTLPVYTSISDAL-GT-RYFTRKDT-TPGAVGVYVYVSLAPLSEITFLAGS-VGRISGARLWVAVTGLLGGQIISIF-RAI
1lda -----YLRGQCIRKPLGATGLLTPFGVGVVA-ALEVA-----G-A-SFVWKEIVVWGLQVARIITDQ-VVSGARLWVAVTGLLGGQIISIF-RAI
1rc2 -----WFRKLAARCPVTPVWVYVGGVGVVA-AQ-----PPE-LGILVAVAPGLVYVYVAFVAG-VIISGGHWRVAVTGLLGGQIISIF-RAI
  
```

# Comparing Structures by Similarity - Q Value

The screenshot displays the VMD (Visual Molecular Dynamics) interface. The main window shows a protein structure rendered as a multi-colored tube, with colors representing sequence identity. The 'VMD Main' window contains a table of loaded molecules:

ID	T	A	D	F	Molecule	Atoms	Frames	Vol
1	A	D	F		1J4N	2029	1	0
2	A	D	F		1FQY	1661	1	0
3	A	D	F		1lda	1997	1	0
5	T	A	D	F	1rc2	3530	1	0

The 'Graphical Representations' window shows the selected molecule '5: 1rc2' with a drawing method of 'Tube' and a coloring method of 'ColorID 3'. The 'Multiple Sequence Alignment' window shows a sequence alignment with a 'Molecule Coloring' menu set to 'Q per residue'.

# Exhibiting Sequence Identity - Side View

This screenshot shows the same VMD interface as above, but with the protein structure rotated to a side view. The 'Multiple Sequence Alignment' window now highlights specific residues in yellow, indicating high sequence identity between the structures. The 'Molecule Coloring' menu is still set to 'Q per residue'.

# Exhibiting Sequence Identity - Top View

The screenshot displays the VMD (Visual Molecular Dynamics) interface. The main window shows a top view of a protein structure composed of multiple chains, each represented by a different color (red, green, blue, yellow). The structure is highly complex and folded. The 'VMD Main' window shows a table of loaded molecules:

ID	T	A	D	F	Molecule	Atoms	Frames	Vol
1	A	D	F		1J4N	2029	1	0
2	A	D	F		1FQY	1661	1	0
3	A	D	F		1lda	1997	1	0
5	T	A	D	F	1rc2	3530	1	0

The 'Graphical Representations' window shows the selected molecule '5: 1rc2' with a drawing method of 'Tube' and a color of 'ColorID 3'. The 'Multiple Sequence Alignment' window shows a sequence alignment with conserved residues highlighted in yellow. The alignment is as follows:

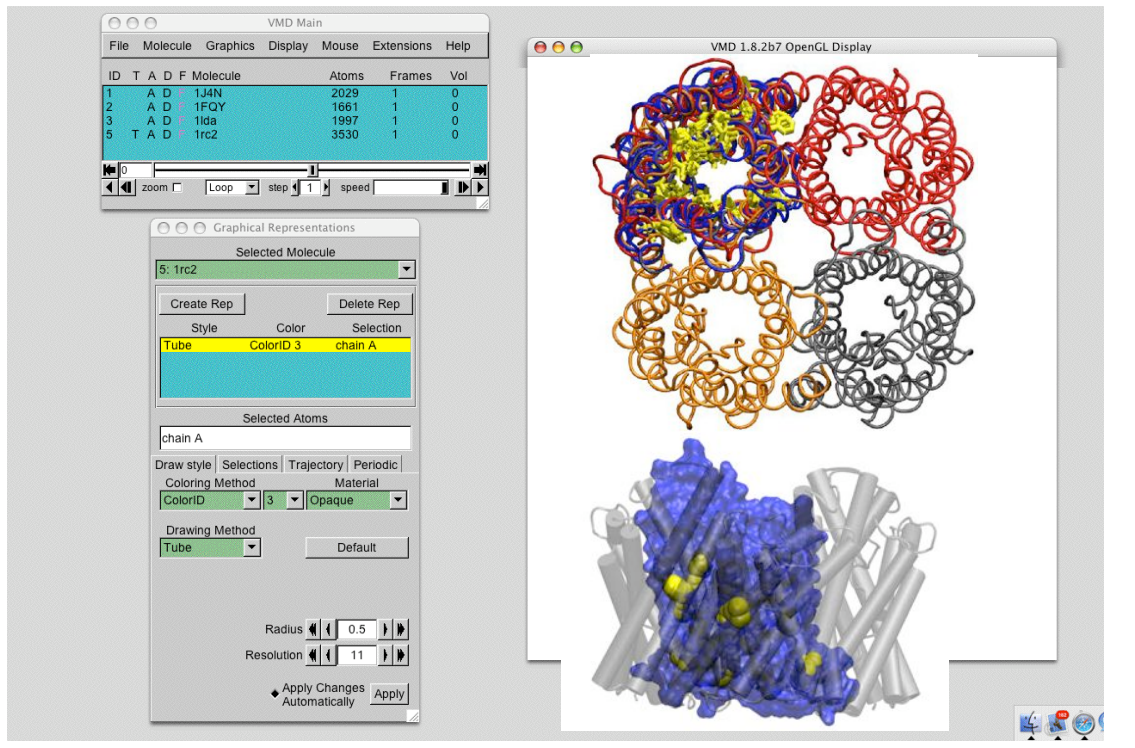
```
1fgy -----SLPPRYAVYAEFLACTLSPYETLGGAL-GP-EEY-FYVRRGQAVGQKVVYSLAQLIATLAGS-VRRERGRLRPAVTLIILSCQIISIF-R  
1j4l NKSEFKKLLPWRVAVAEFLAKILFLIETLGGAL-GP-HYFIREKGT-QWAVGQKVVYSLAQLIATLAGSVGR-SGRSLRPAVTLIILSCQIISIF-R  
1l0a -----TLRQGLAEFLZTGLLITPVVQVA-RLQVA-----GQ-SFGQRIQVWGLAAMAIVLTA-GQGLRPAVTLIILRFA-CFQRRS  
1rc2 -----RPVRLAARPTTFLVWGGQAVLA-NG-----FPL-LGTFAGVALAQLIUDWAFVQ-IISGHHKPAVTLIILMGG-RFPARR
```

# Showing Conserved Residues - Monomer

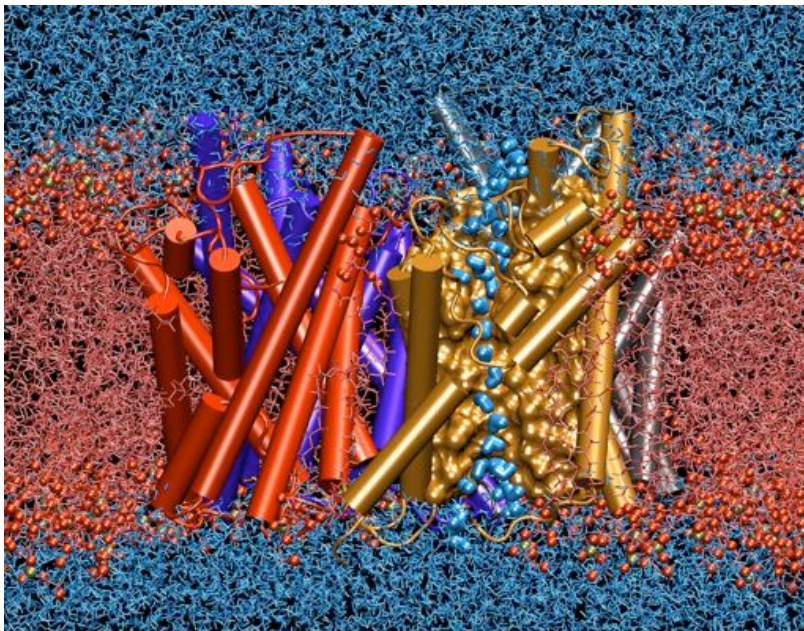
The screenshot displays the VMD interface showing a monomer view of a protein structure. The structure is a single chain, colored red, with conserved residues highlighted in yellow. The 'VMD Main' window shows the same table of loaded molecules as in the previous screenshot. The 'Graphical Representations' window shows the selected molecule '5: 1rc2' with a drawing method of 'Tube' and a color of 'ColorID 3'. The 'Multiple Sequence Alignment' window shows the same sequence alignment with conserved residues highlighted in yellow.



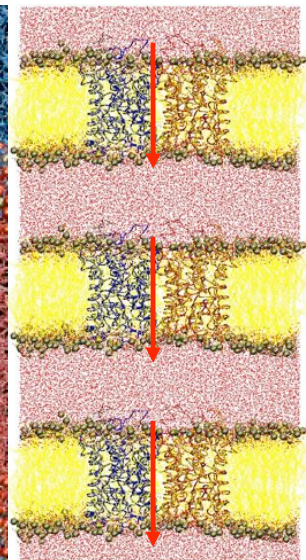
# Showing Conserved Residues - Tetramer



# Water Transport in Aquaporins



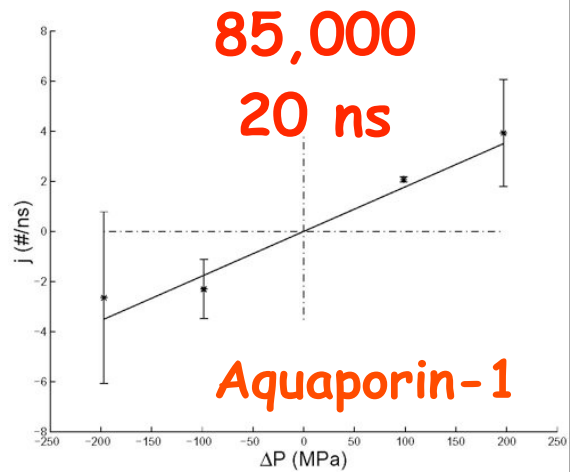
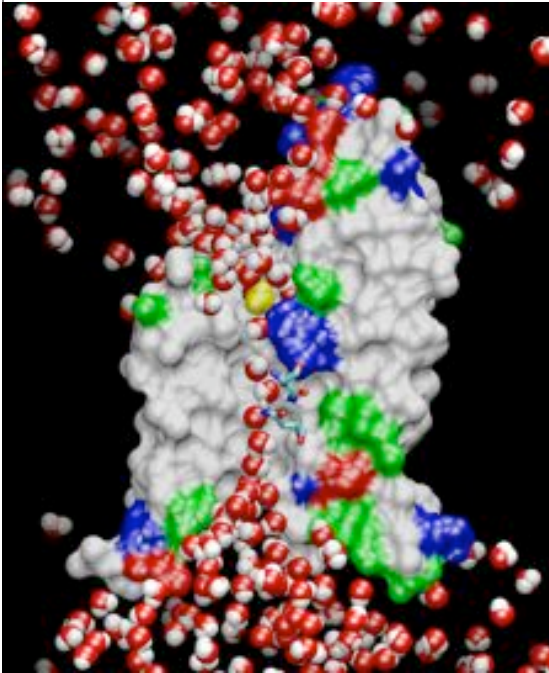
100,000 atoms



**Simulation:**

*Apply constant force  
on bulk water  
molecules*

# Osmotic permeability of water channels

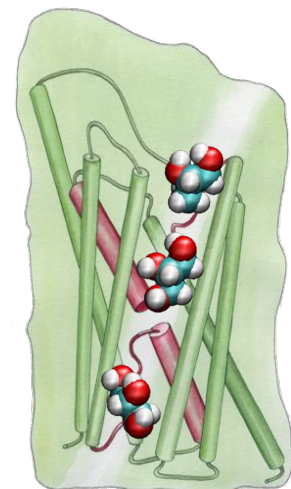
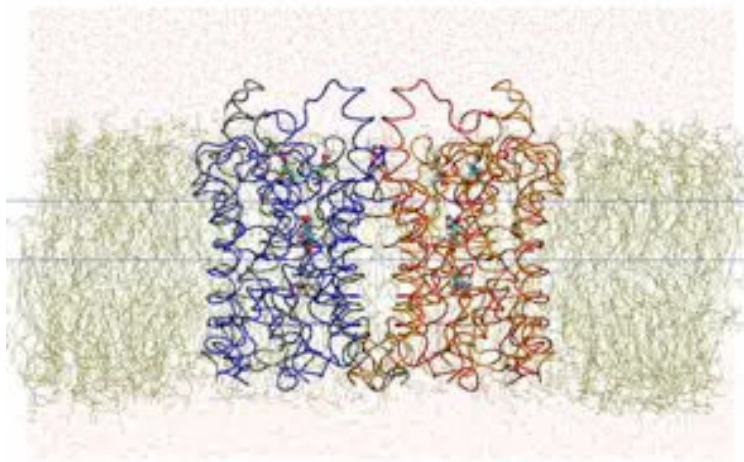


**Aquaporin-1**

$p_f: 7.0 \pm 0.9 \times 10^{-14} \text{ cm}^3/\text{s}$   
**Exp:  $5.4 - 11.7 \times 10^{-14} \text{ cm}^3/\text{s}$**

F. Zhu, E. Tajkhorshid, K. Schulten, *Biophys. J.* 86: 50-57 (2004)  
F. Zhu, E. Tajkhorshid, K. Schulten, *Phys. Rev. Lett.* 93: 224501 (2004)

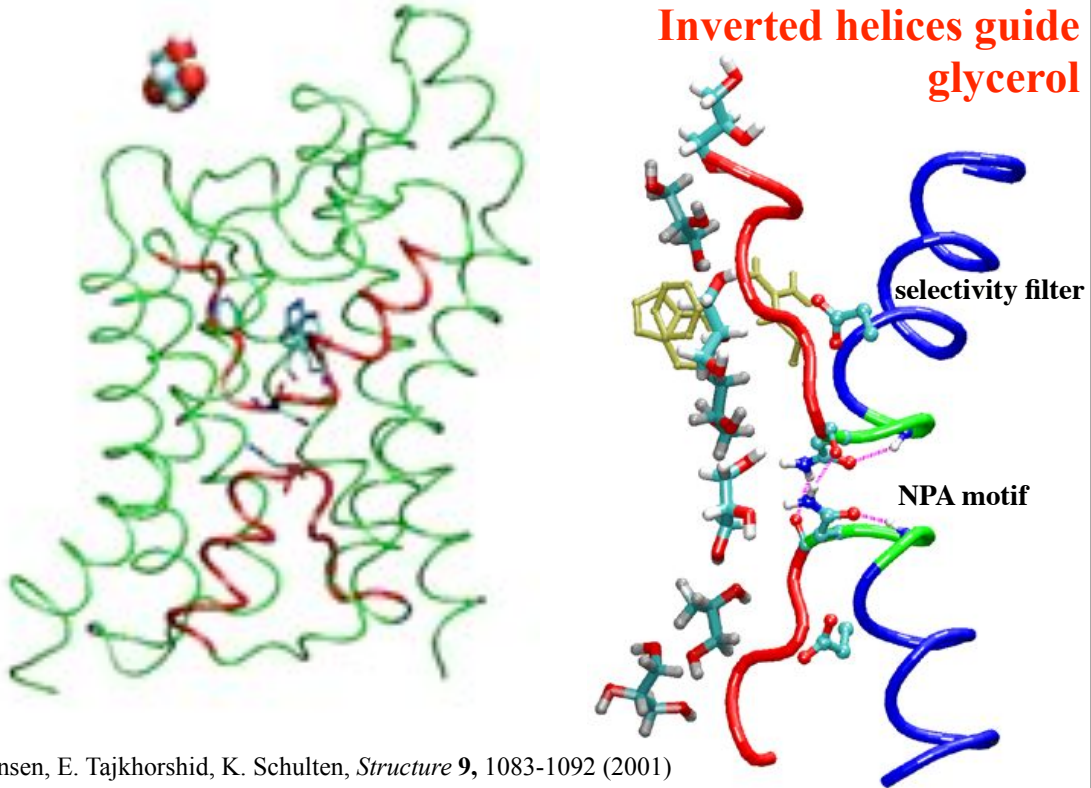
# Dynamics of Protein, Lipid, Water System



M. Jensen, E. Tajkhorshid, K. Schulten, *Structure* 9, 1083-1092 (2001)

# Glycerol Conduction

Inverted helices guide glycerol

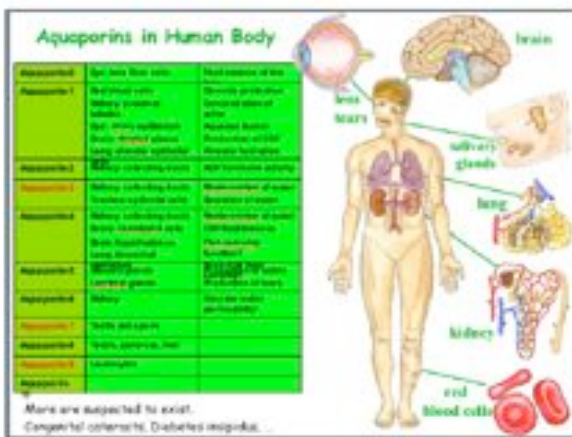


M. Jensen, E. Tajkhorshid, K. Schulten, *Structure* 9, 1083-1092 (2001)

University of Illinois at Urbana-Champaign  
 NIH Resource for Macromolecular Modeling and Bioinformatics  
 Beckman Institute

## Aquaporins

Case study, see at  
<http://www.ks.uiuc.edu/Training/CaseStudies/>



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 John Eargle

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 Elizabeth Villa  
 Emad Tajkhorshid  
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 Zan Luthey-Schulten