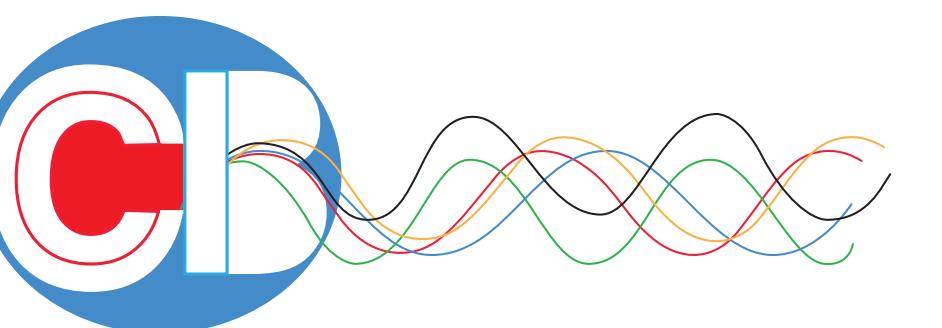
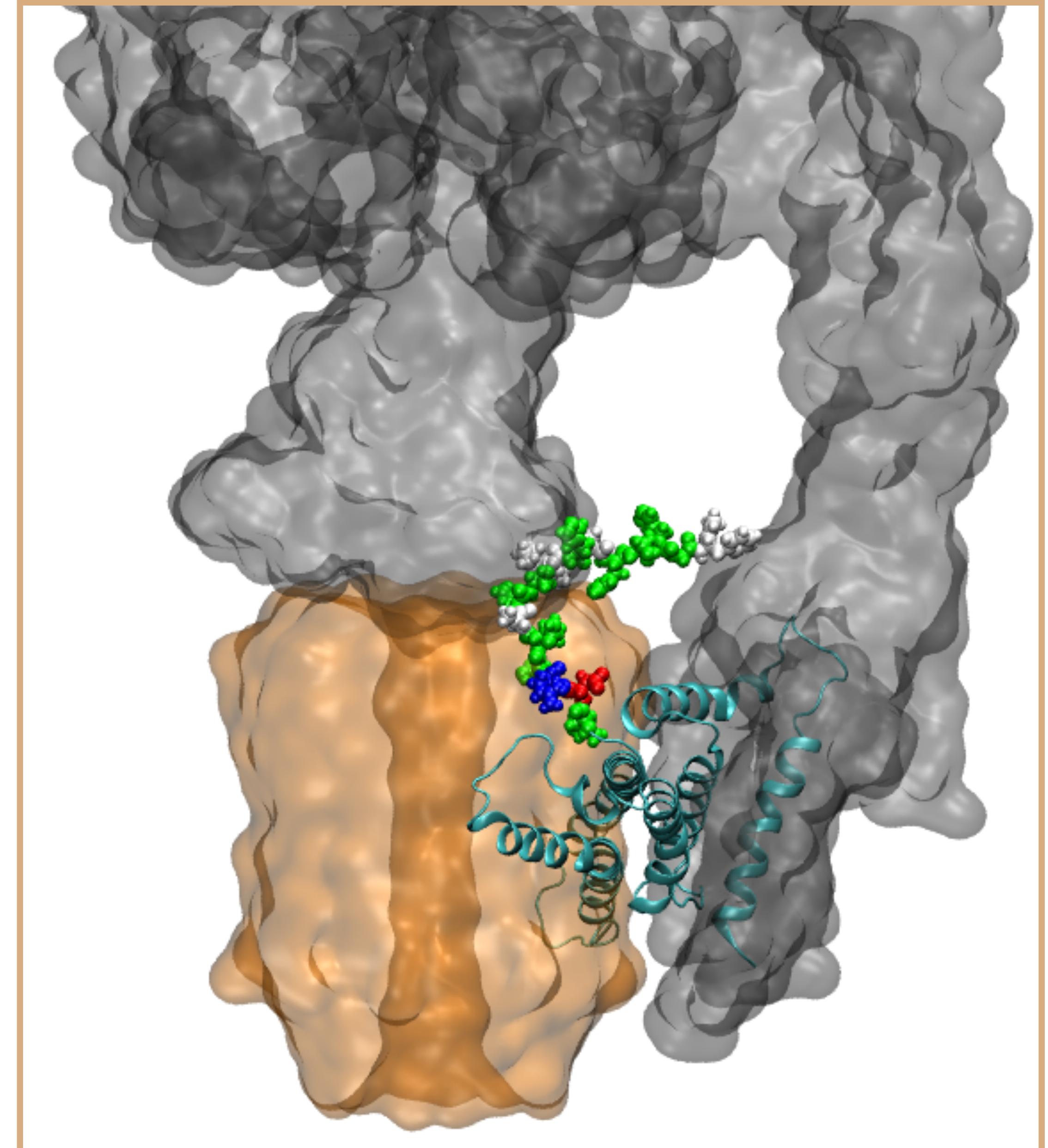


Molecular Mechanism of Reversed Temperature Dependence of ATP Synthesis in Glacier Ice Worms

Noureen Abdelrahman
April, 14th 2023



Advisor: Dr. Grace Brannigan



Outline

- Background & Motivation
- Approach
- Results
 - ATP6 Conservation Scores
 - ATP6 Flexibility
 - Rotational Diffusion of the c-ring
 - ATP6 Extension Orientation At Different Temperatures
- Summary

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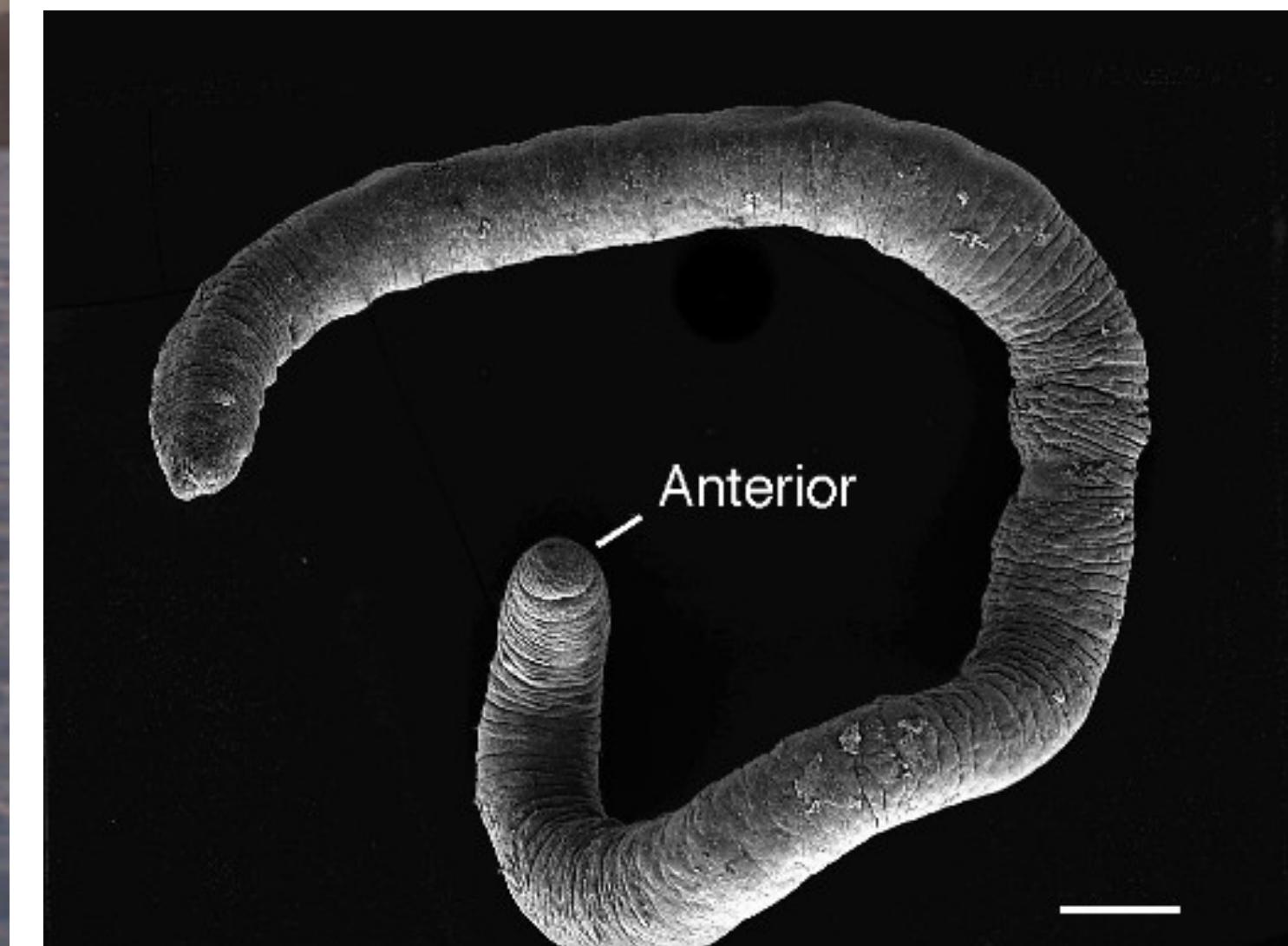
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Glacier ice worms complete their entire life cycle below 0 °C



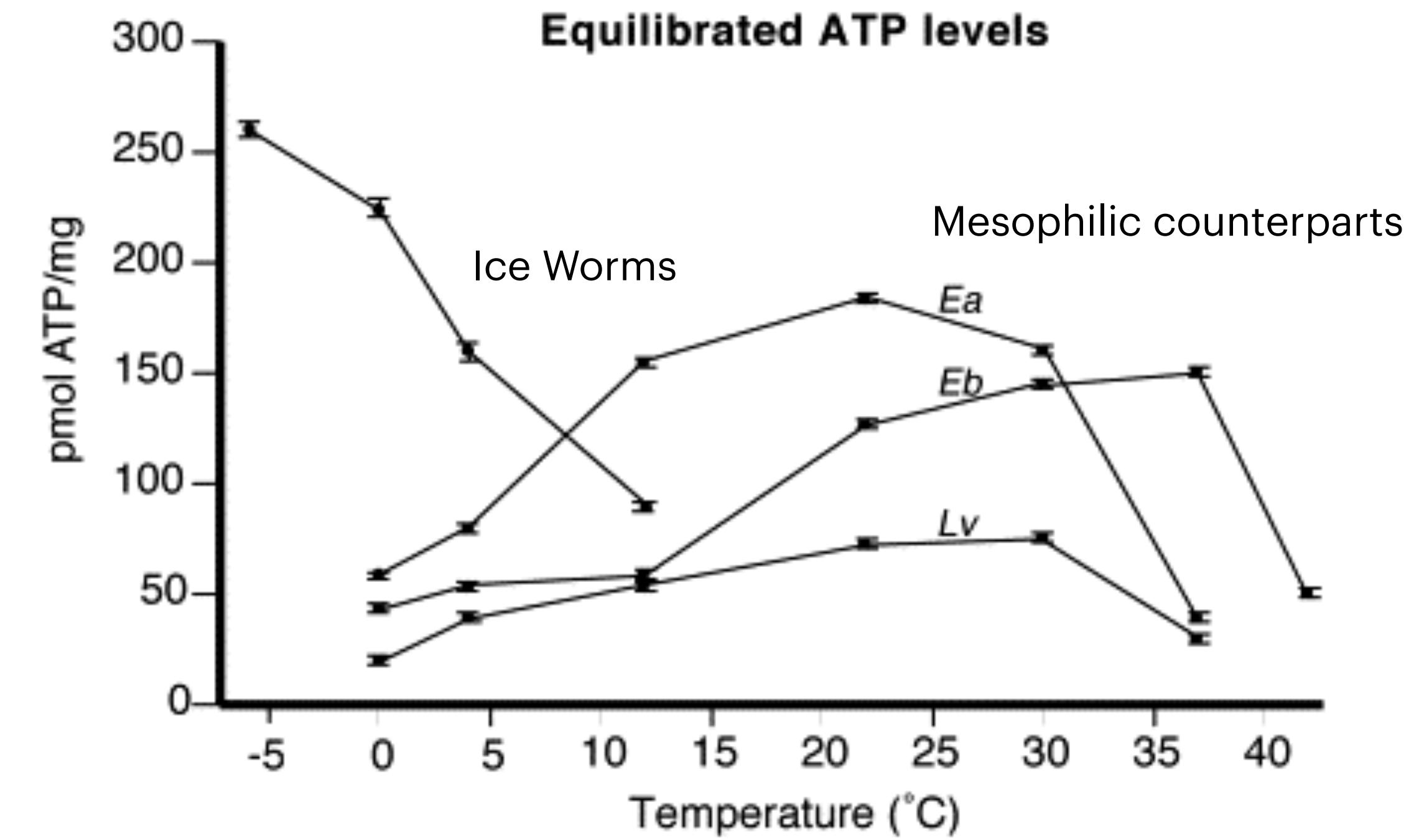
Pictures from: Dr. Daniel Shain and coworkers

Location: American Pacific Northwest



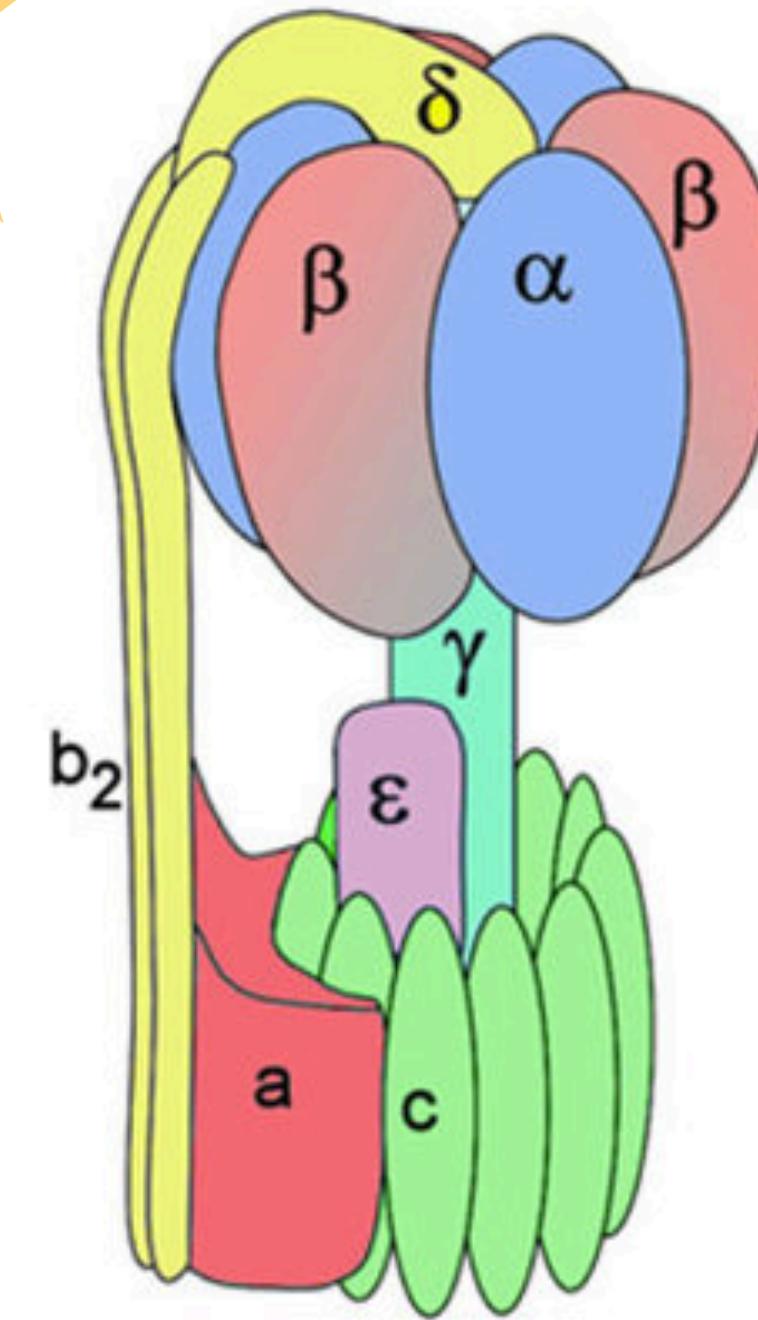
Ice Worms have elevated ATP levels at declining temperatures

- Survive exclusively in glacier ice in 0 °C temperature
- Compensatory Mechanism: Enhanced ATP production with respect to ATP consumption —> Elevated ATP levels

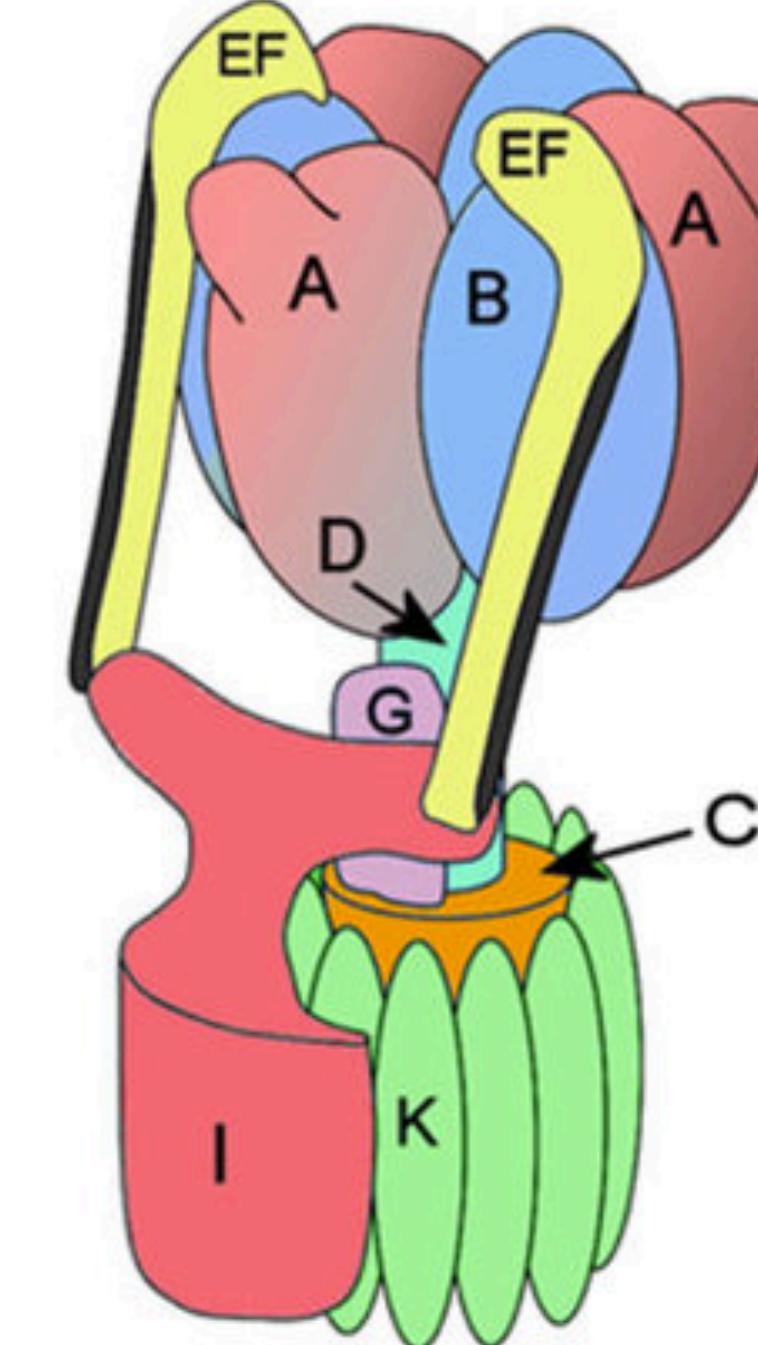


Michael Napolitano, Robert Nagele,
Daniel Shain
2004

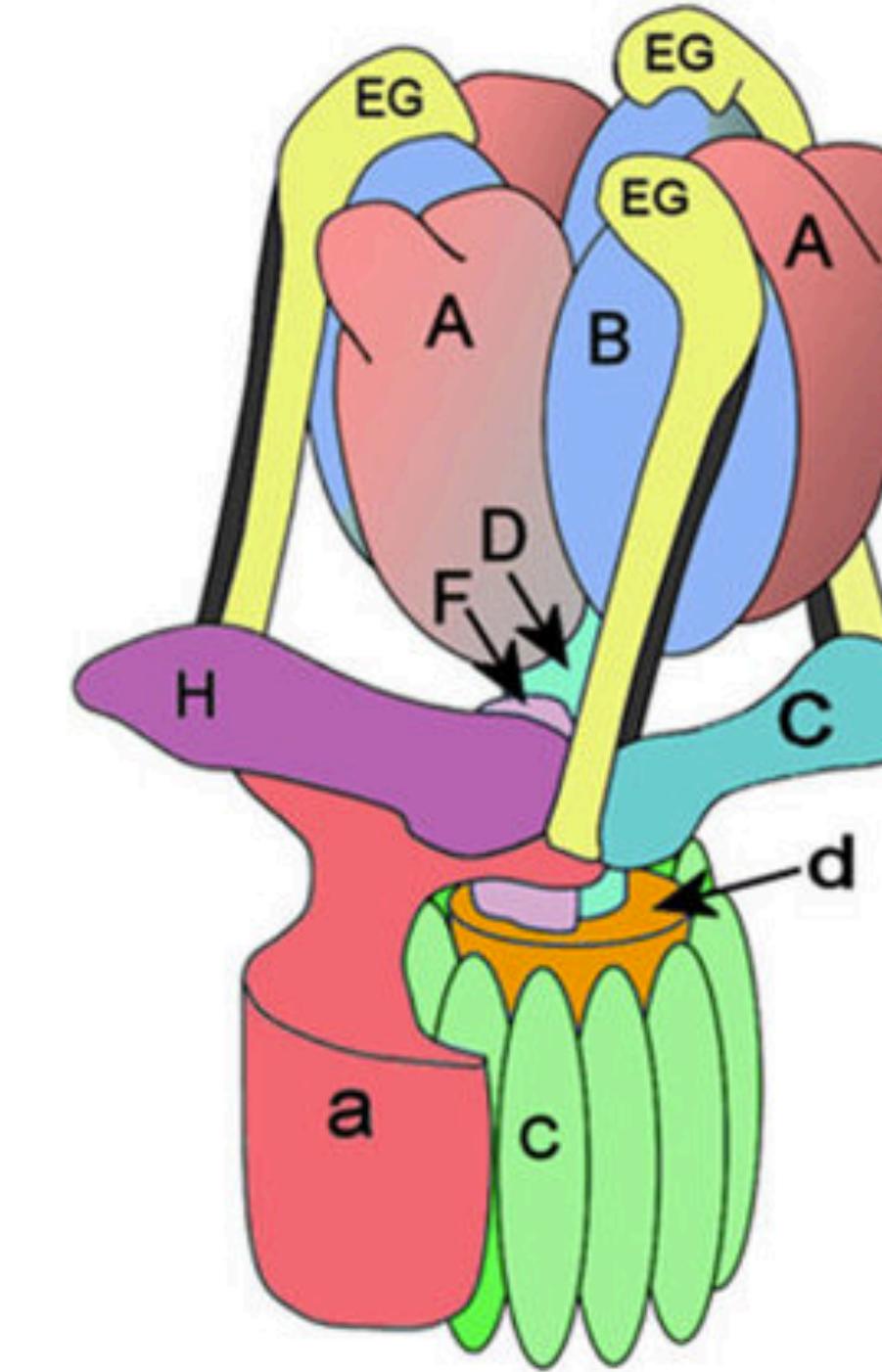
ATP levels are controlled by the rotary motor ATPases family



F-ATPase



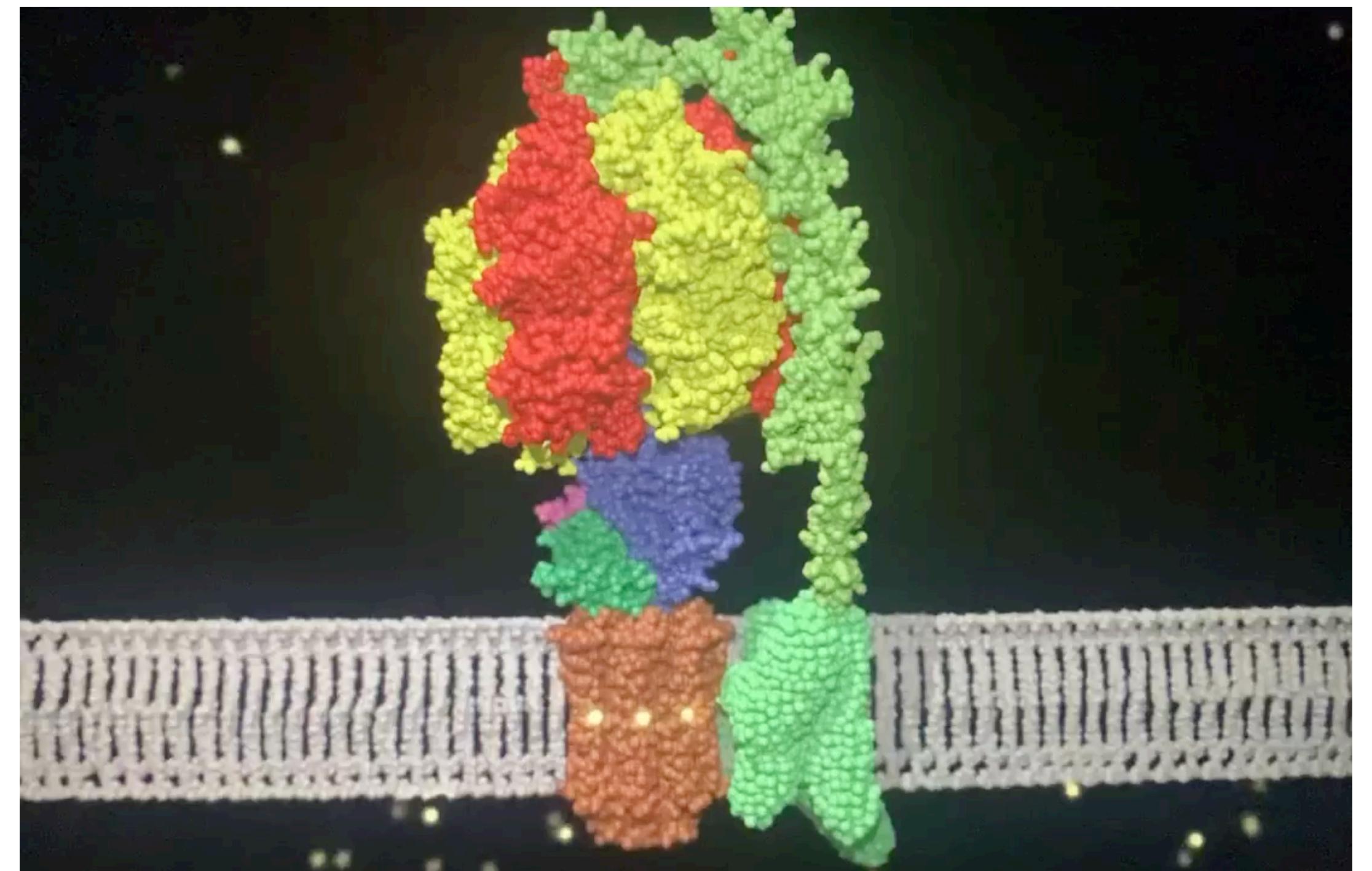
A-ATPase



V-ATPase

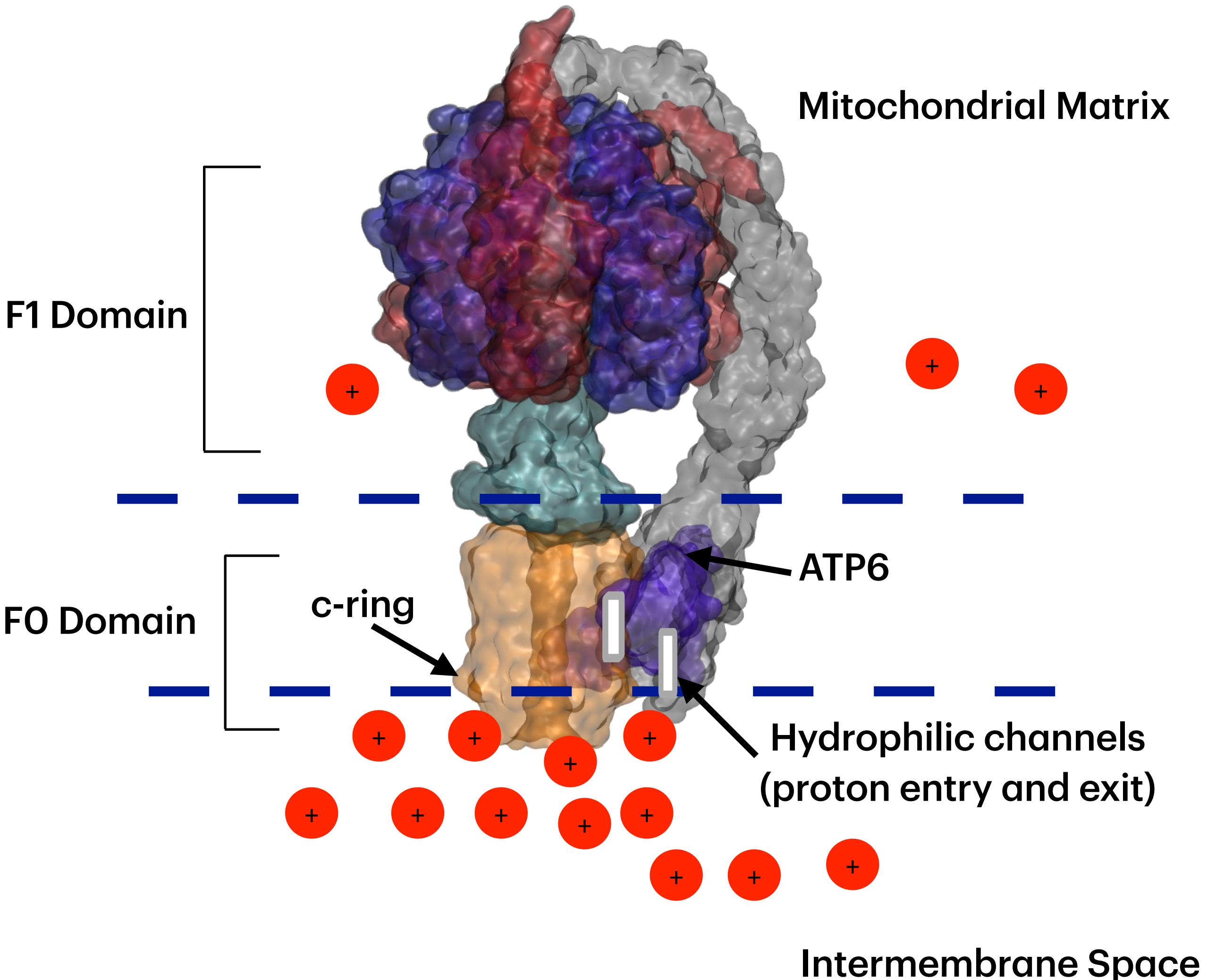
System of Interest: F0F1 ATP Synthase Complex

- Synthesizes and Hydrolyzes ATP
- Highly conserved among all species
- 22 subunits
- ATP synthesis is dependent upon a proton gradient

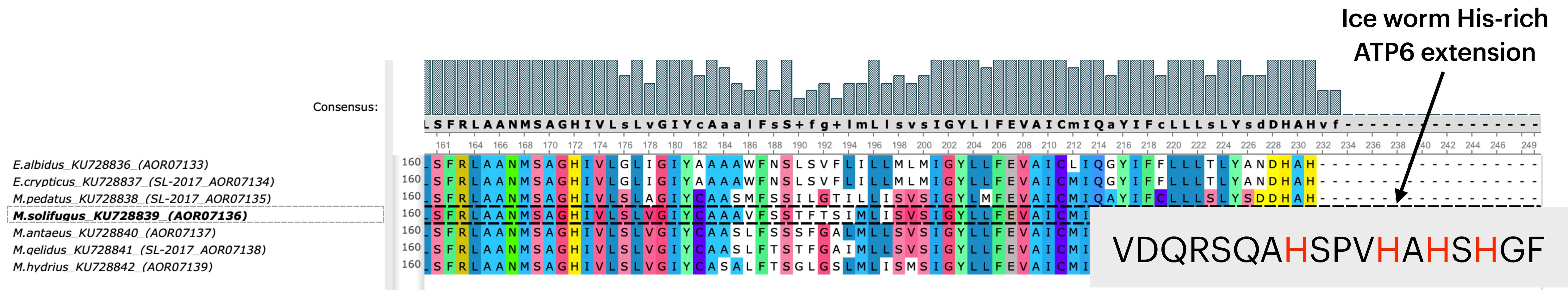


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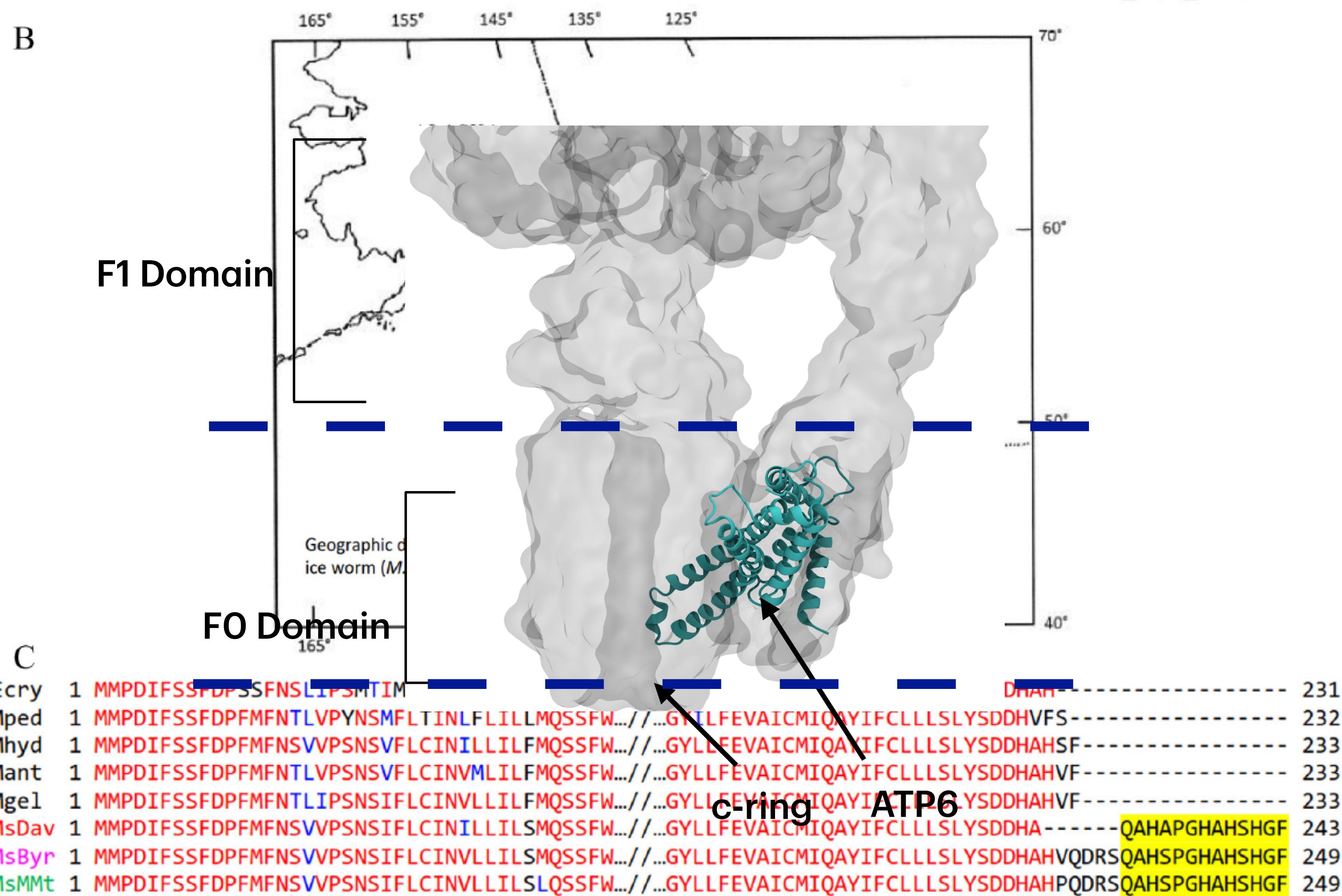


Ice worms have an extended His-rich ATP6 domain at the C-terminus



The ATP6 insertion is likely to have a major impact on functional activity

- Davidson and Byron Glacier ice worms diverged from each other 5-10 million years ago
- ATP6 is highly sensitive to modification: cause for most mt-diseases
- ATP6 extension is Histidine-rich

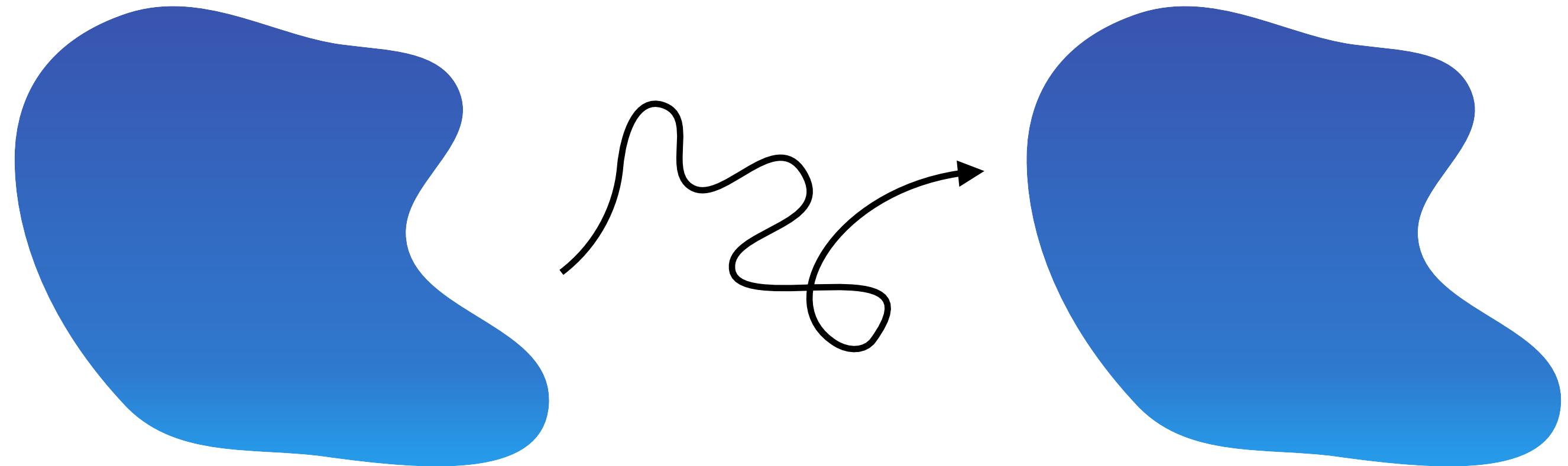


Research Question:

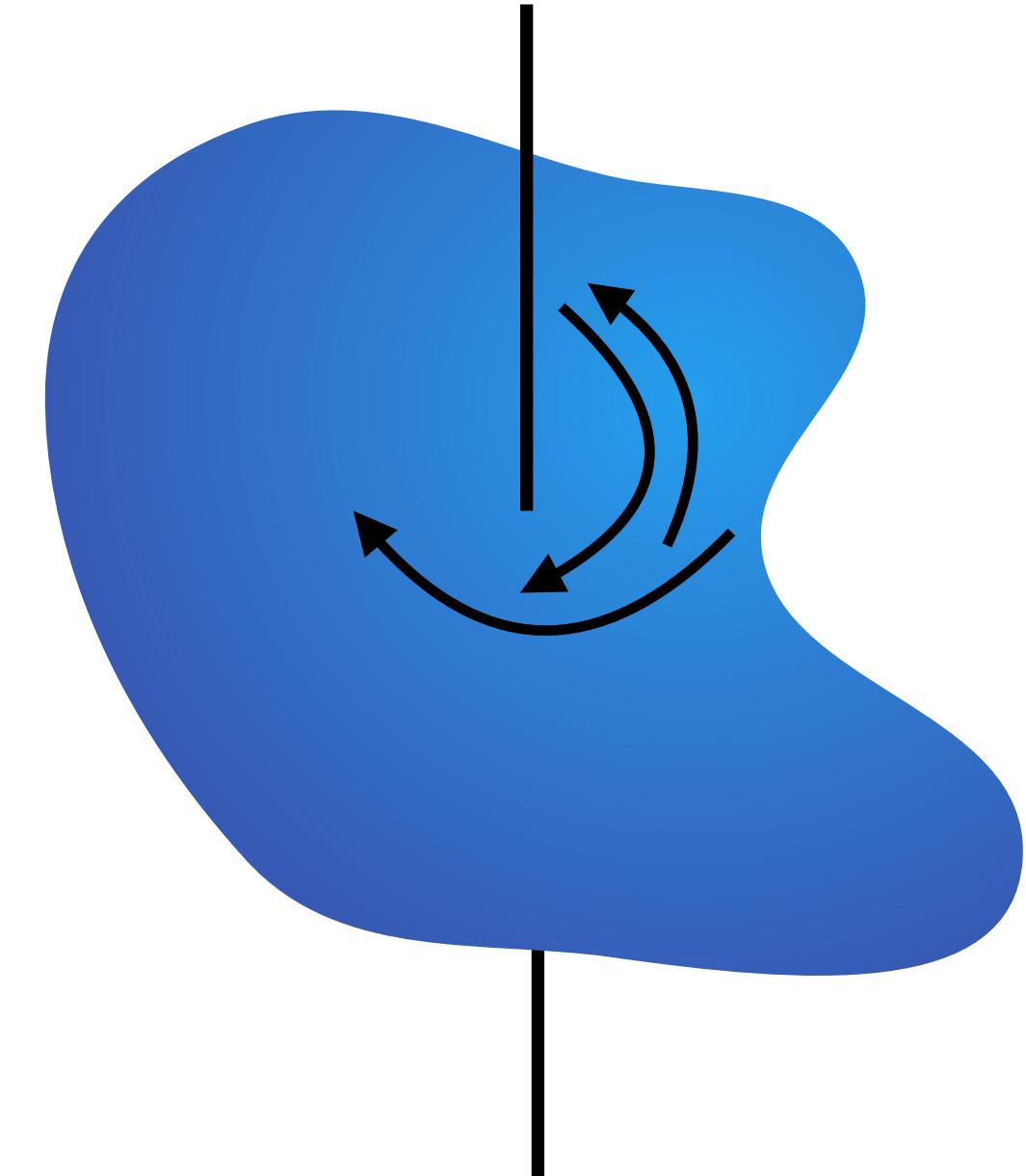
How does sequence affect the Temperature sensitivity of the c-ring?

Different Types of Movement in the Membrane:

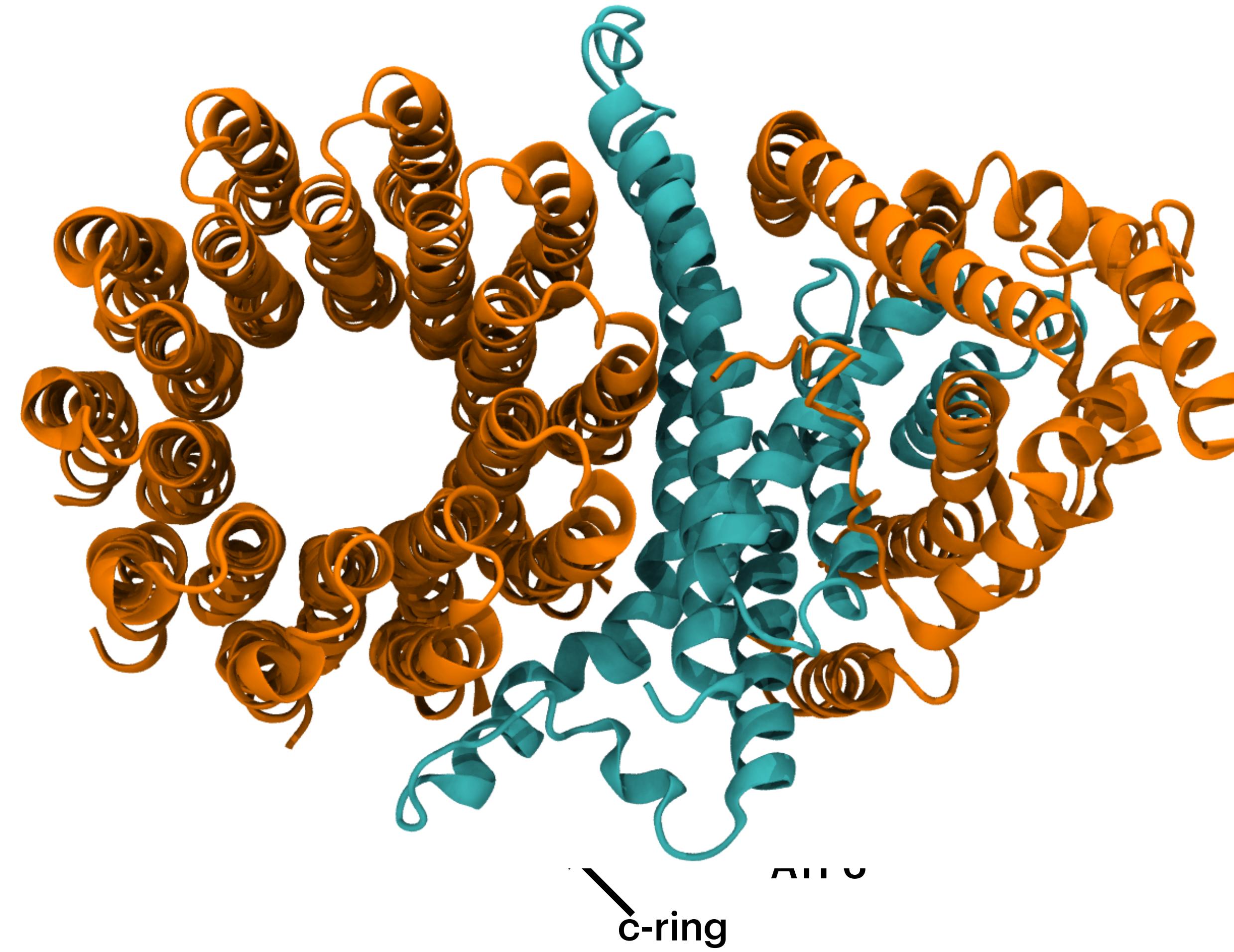
Translational Diffusion



Rotational Diffusion



The c-ring of the F0 domain rotates around its central axis



Research Question:

How much does sequence affect the Temperature sensitivity of the c-ring?

- How much does Temperature impact the flexibility and dynamics of ATP6 in ice worms?
- How much does Temperature affect the rotational diffusion of the c-ring?
- How much does the ice worm ATP6 extension affect the temperature sensitivity of the c-ring?

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Molecular Dynamics as a Computational Microscope

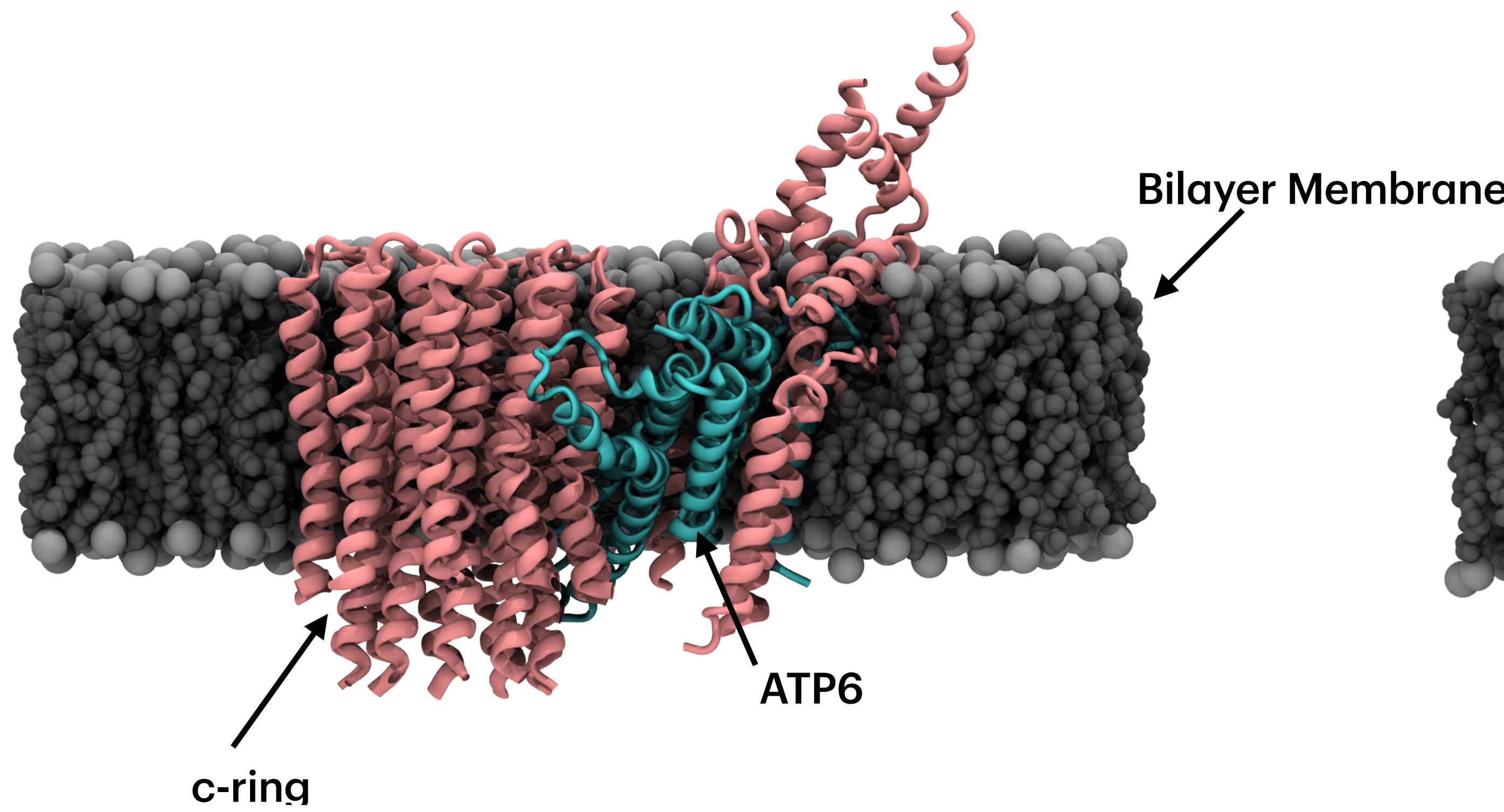
- MD simulations are used to visualize the dynamic motion of molecules in our cells
- In atomistic simulations, the motions of atoms in the molecules are observed in a collection of “movie frames” over time
- To get accurate predictions for the motions of atoms, very small time steps are required

Approach: Atomistic Simulations

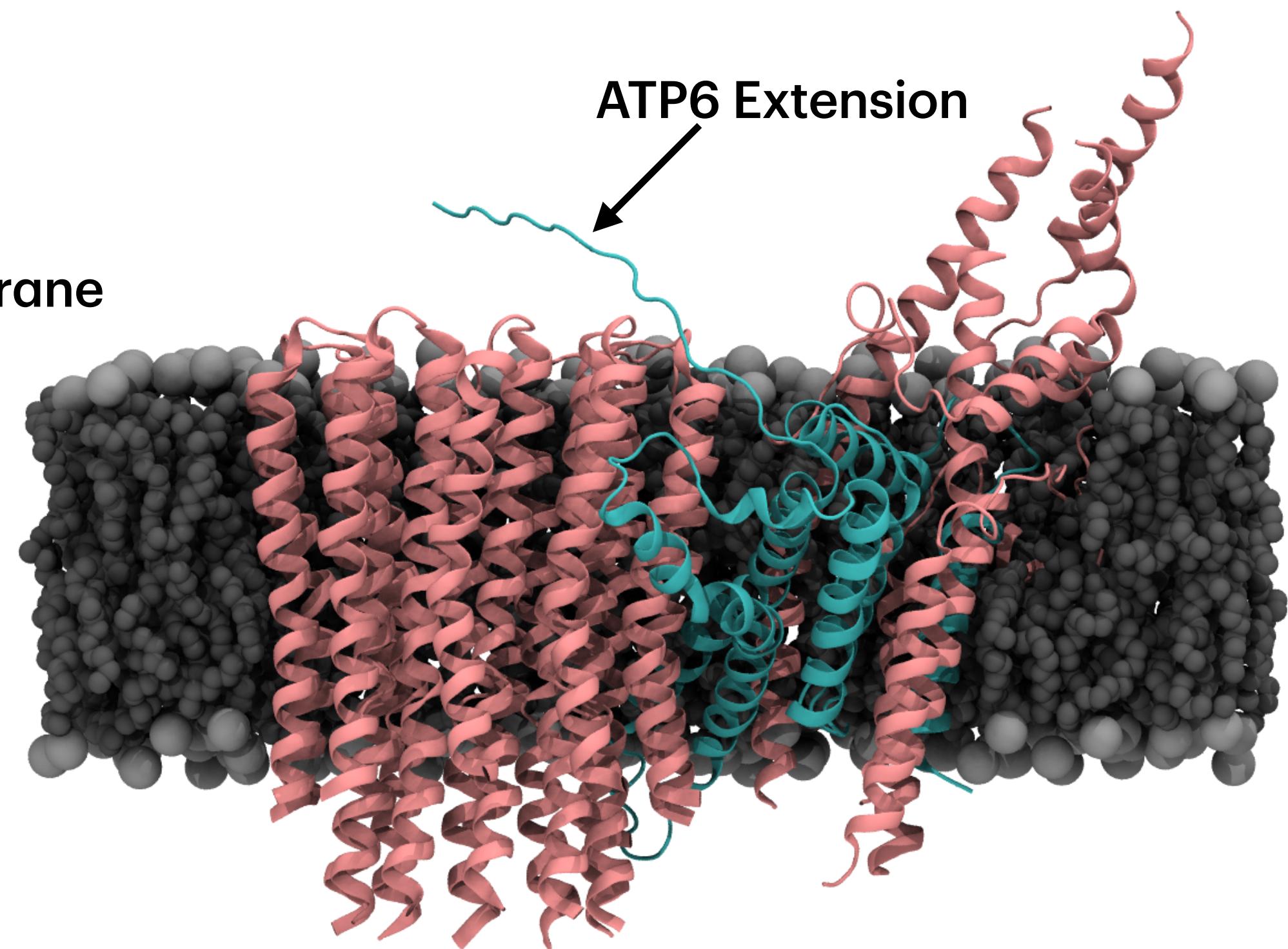
Sequence	277 K	310 K
Yeast		
Ice Worm		
Ice Worm Without ATP6 Extension		

Atomistic molecular dynamics model of F0 domain

Yeast F0 domain in a POPC Membrane



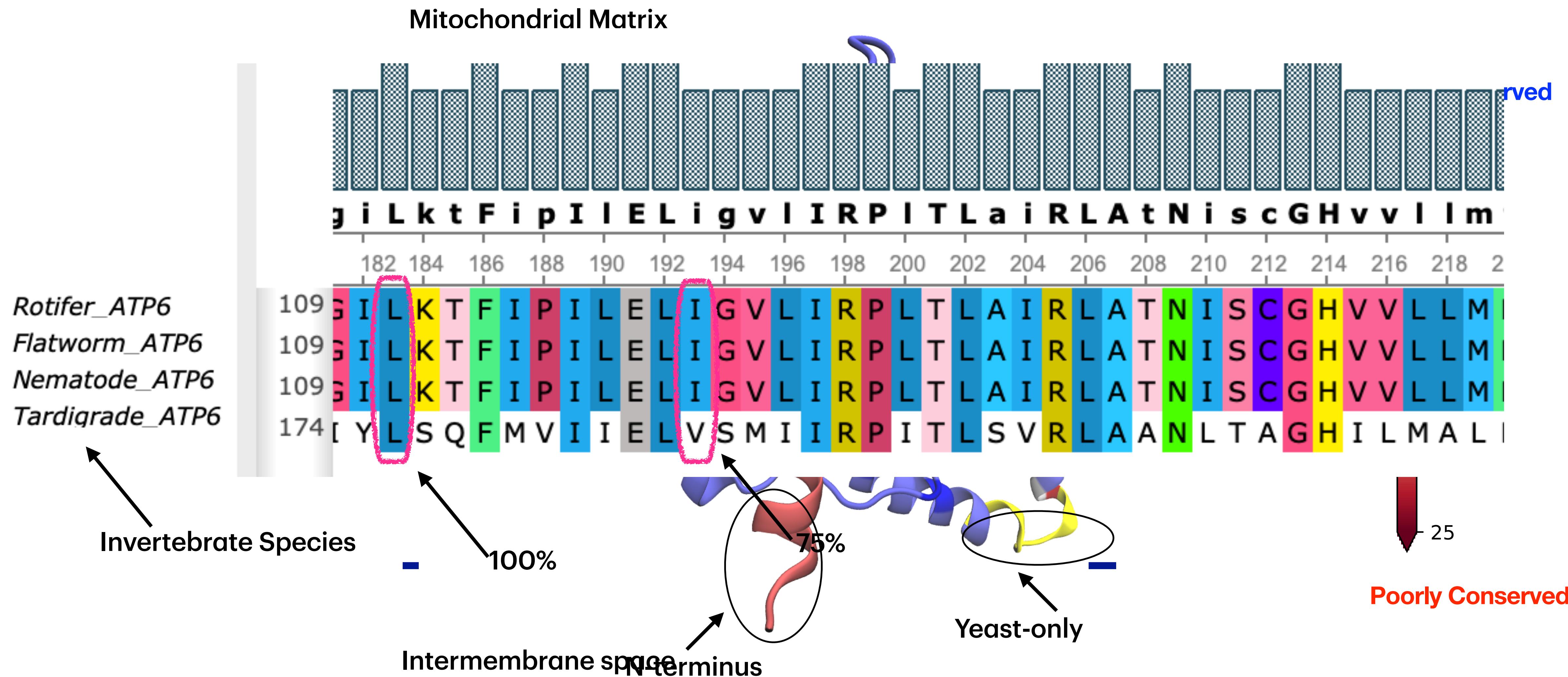
Ice Worm F0 domain in a POPC Membrane



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Poorly conserved regions of ATP6 lie at the terminus



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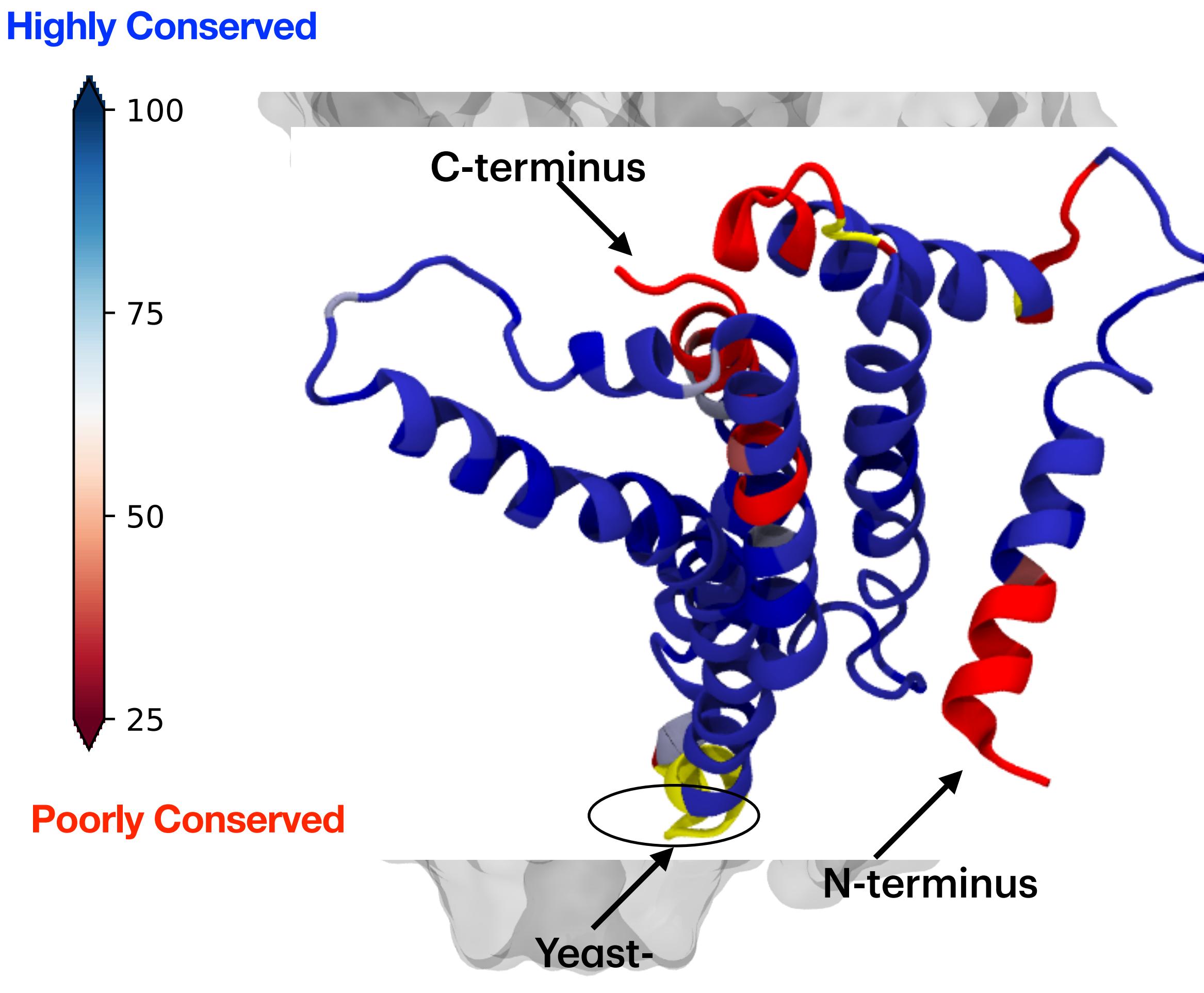
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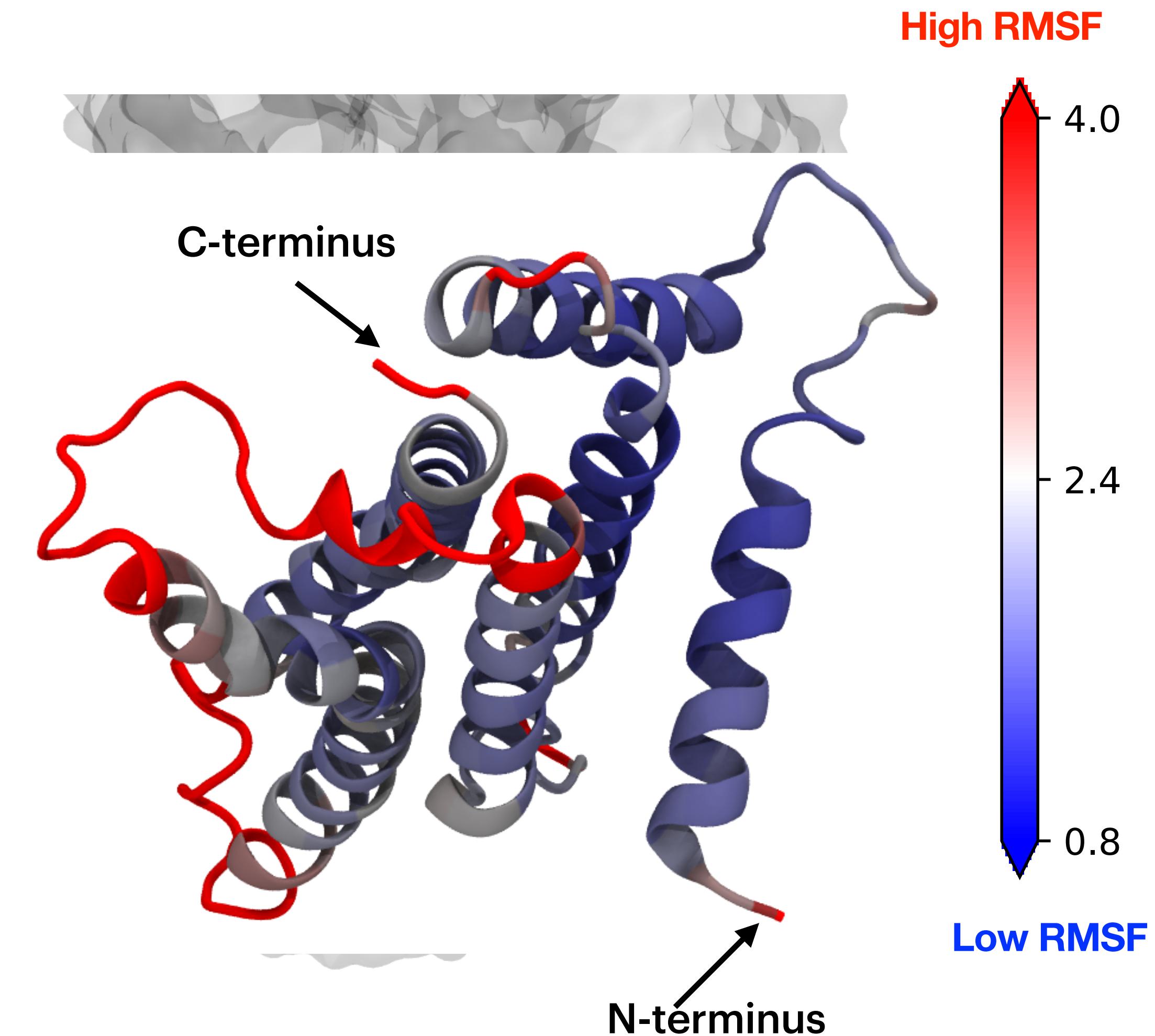
Poorly conserved residues in ATP6 have higher RMSF than the highly conserved residues

Yeast at 310 K

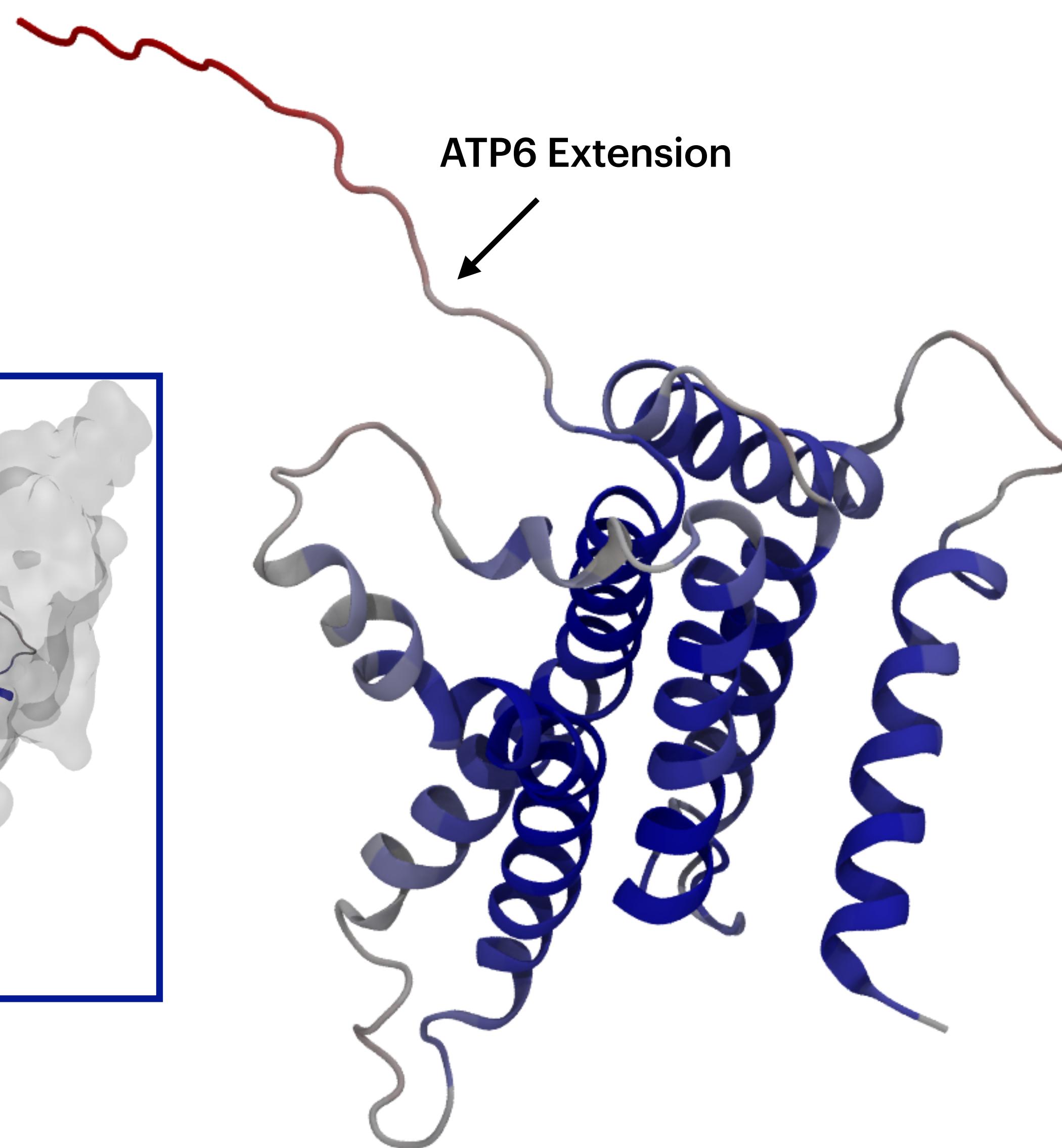
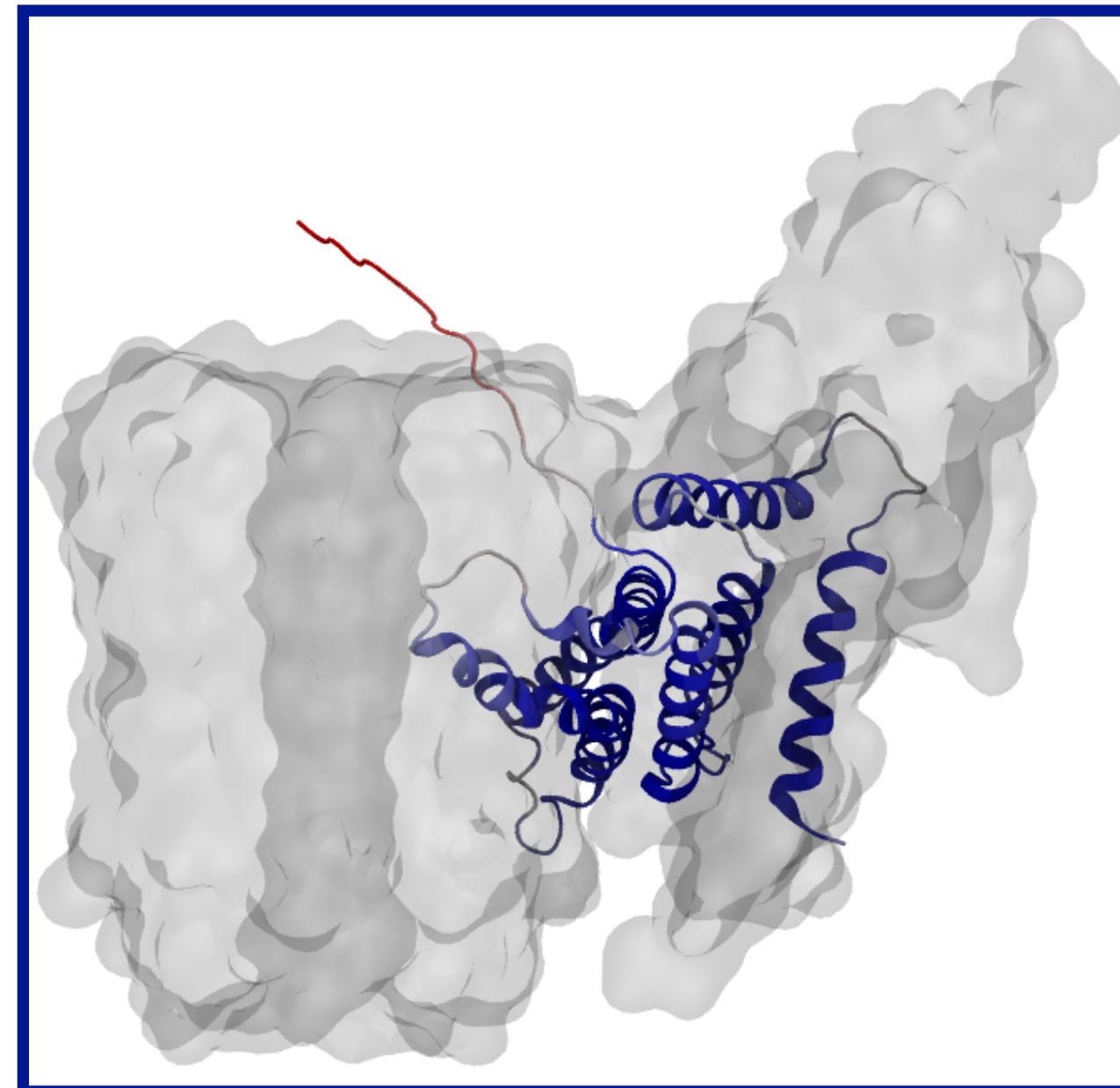
ATP6 Colored by Conservation



ATP6 Colored by RMSF



At 277 K, ATP6 Extension is very dynamic compared to the rest of the subunit

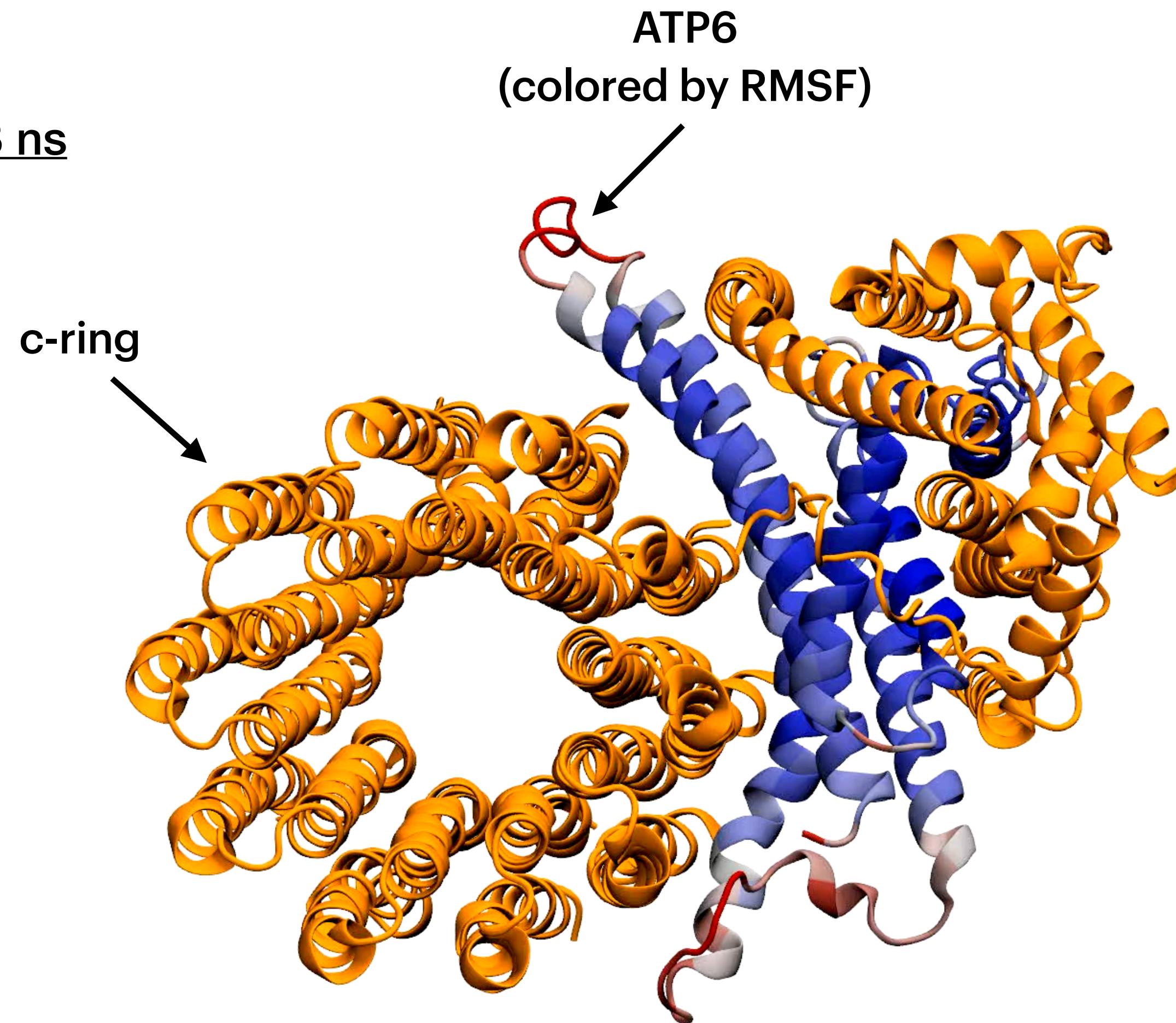


ATP6 Extension RMSF: 9 - 19 Å

Rest of ATP6 RMSF: 0.8 - 5 Å

Let's watch it LIVE

Yeast F0 domain at 310 K, 238 ns

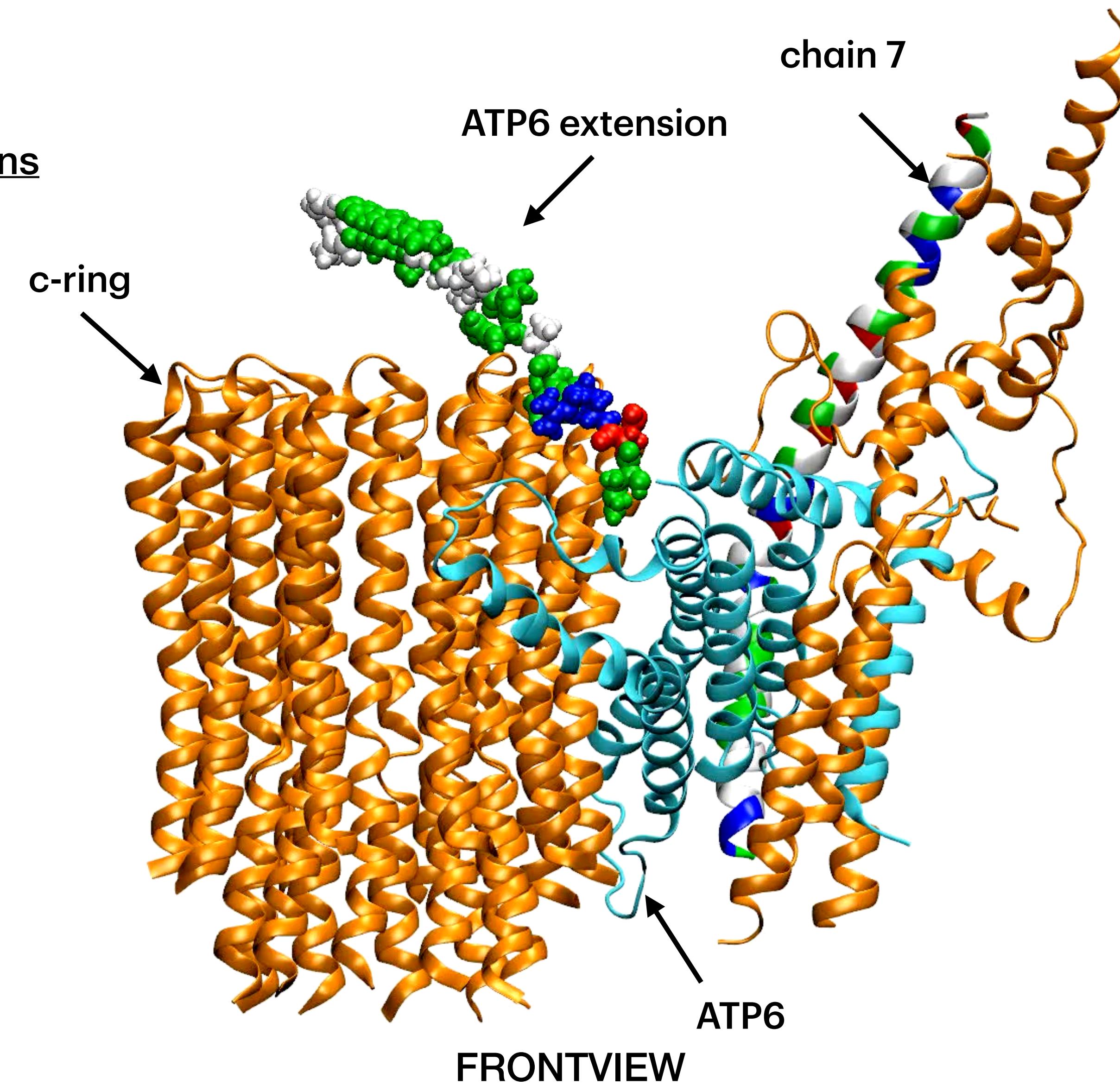


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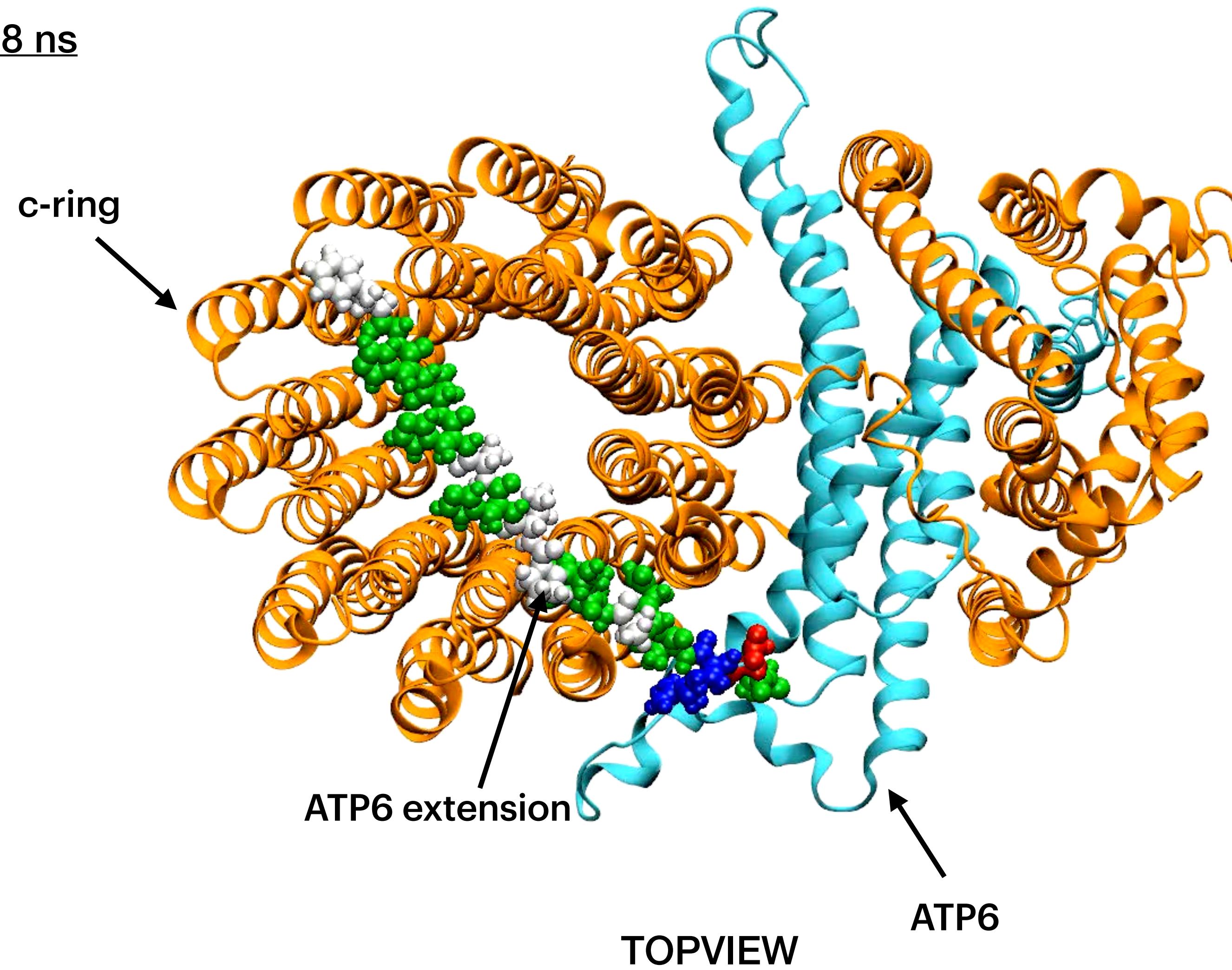
Hydrophobic interactions between ATP6 extension and structural chain 7 at 277 K

Iceworm F0 domain, 229 ns



ATP6 extension inserts itself in the c-ring lumen at 310 K

Iceworm F0 domain, 238 ns



Outline

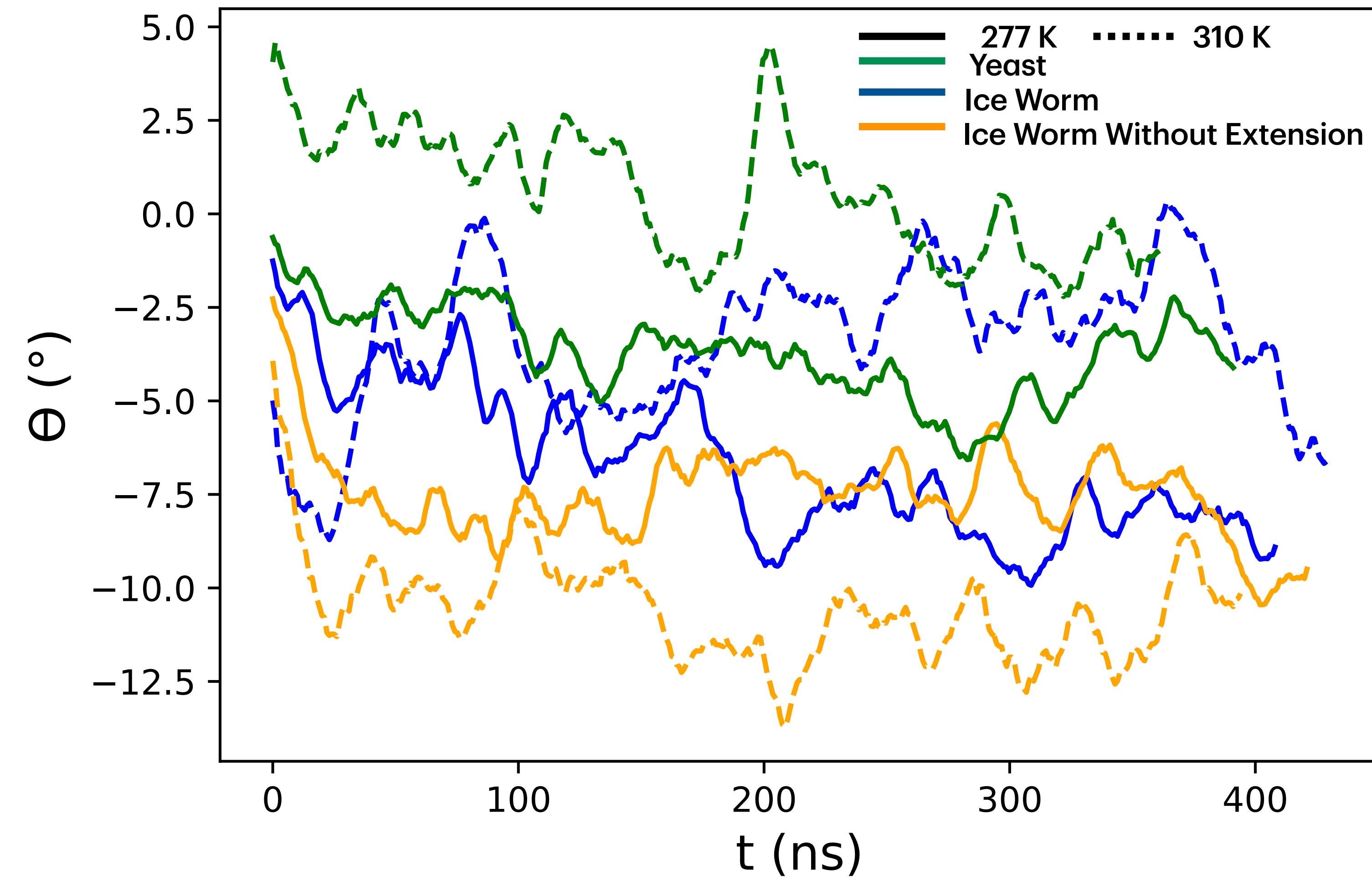
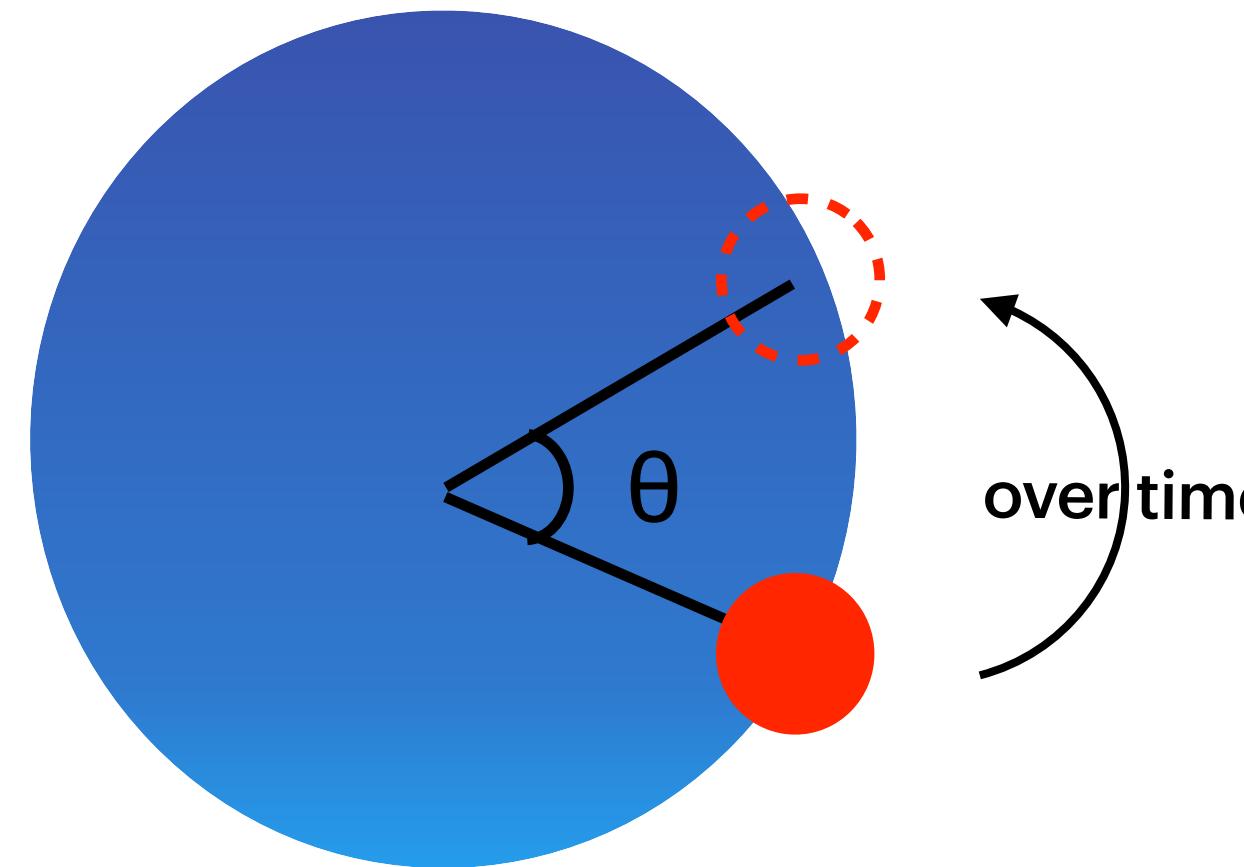
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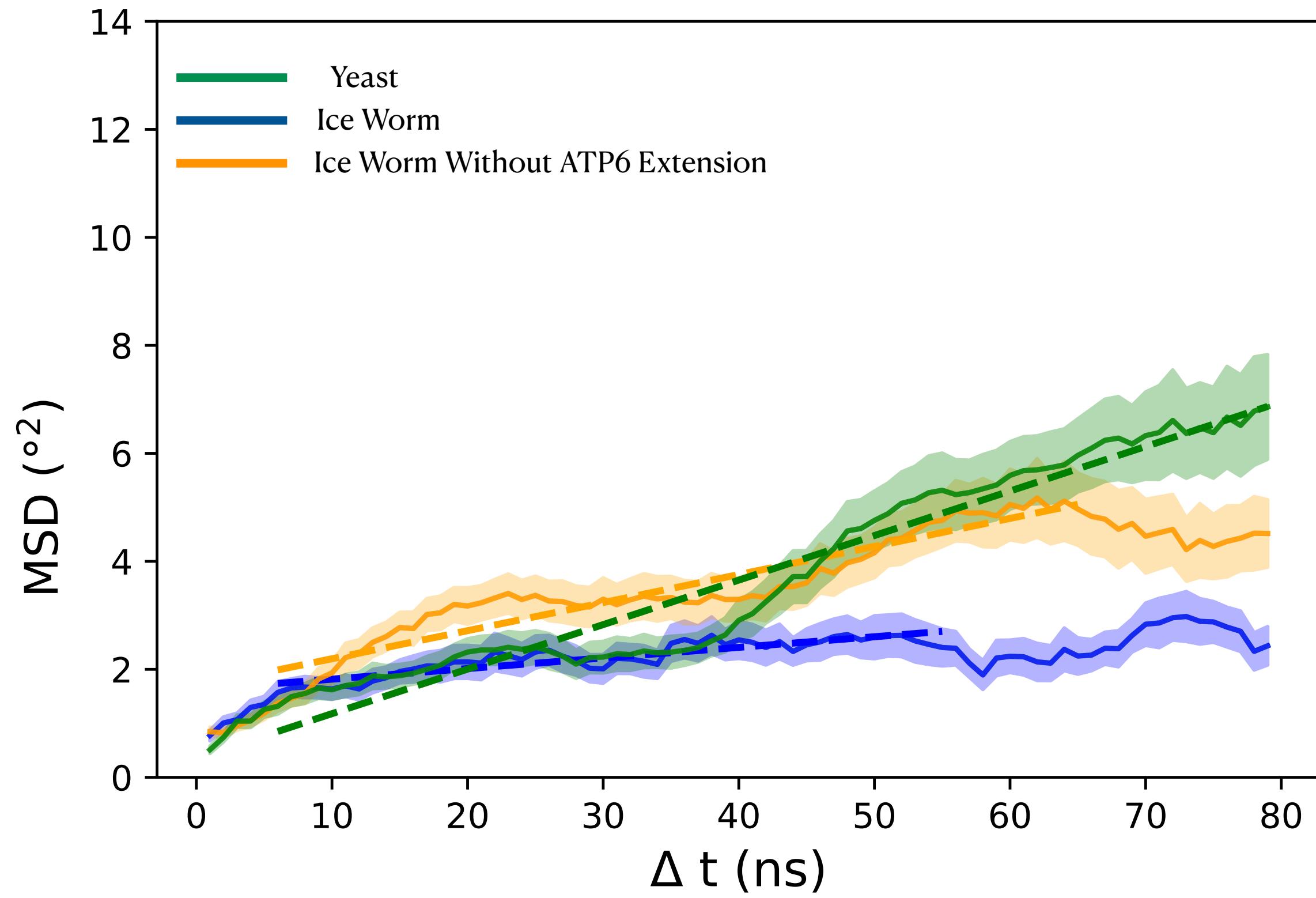
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Rotational Angles at 277 K and 310 K for each protein sequence

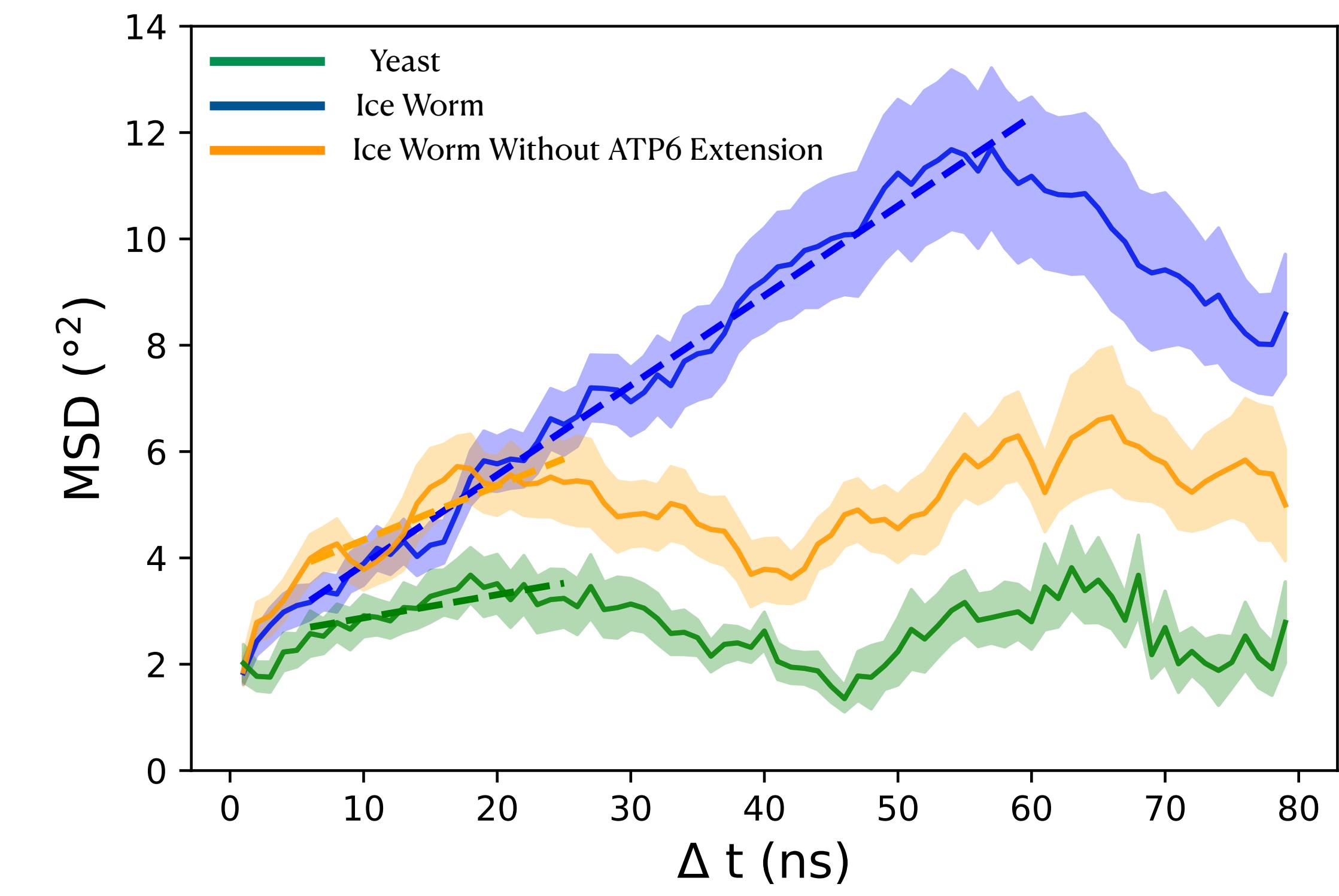


Rotational Diffusion is faster at 310 K for all sequences

a) 277 K



b) 310 K

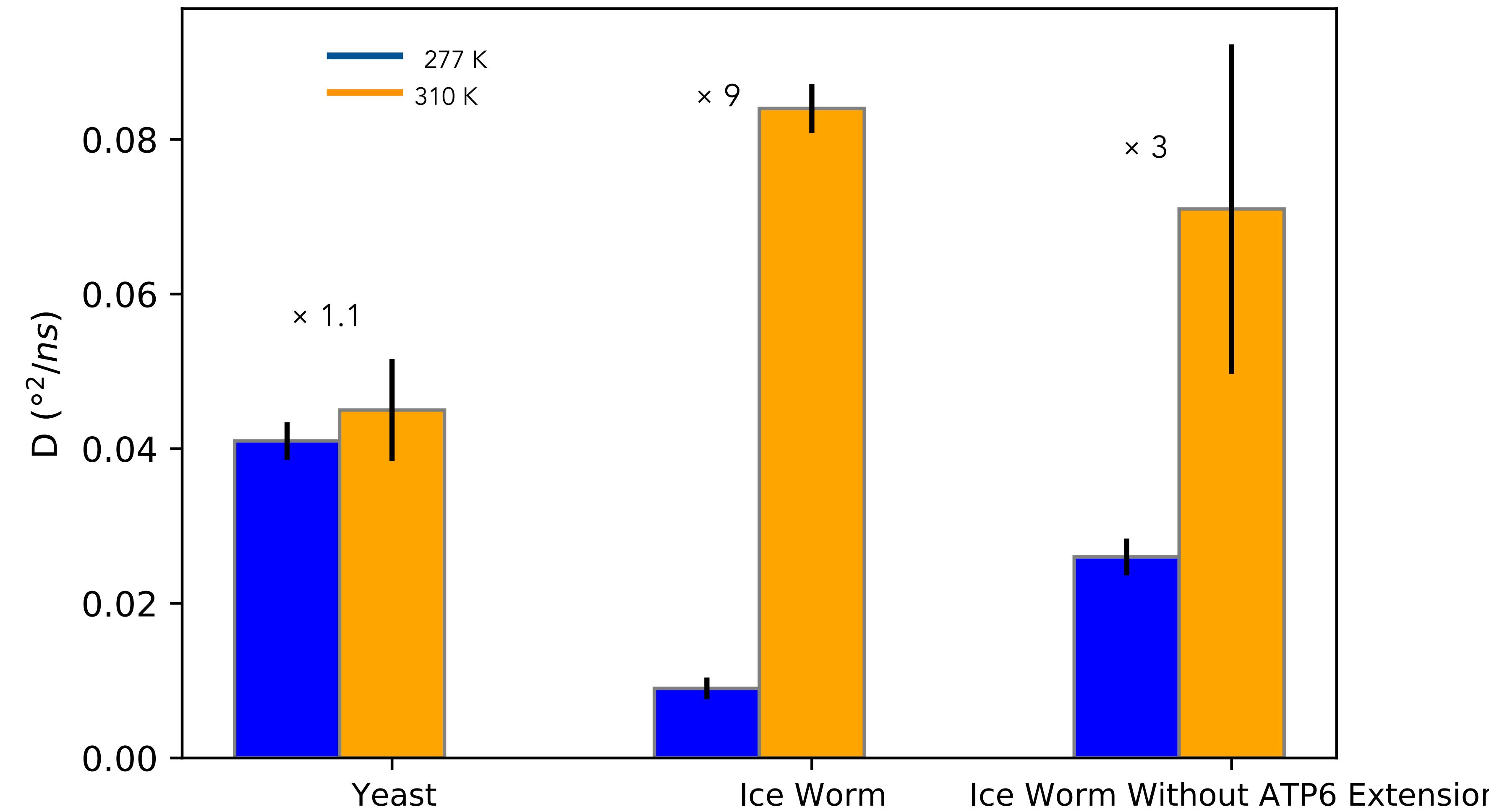


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ATP6 Extension exacerbates the difference in rotational diffusion at varying temperatures



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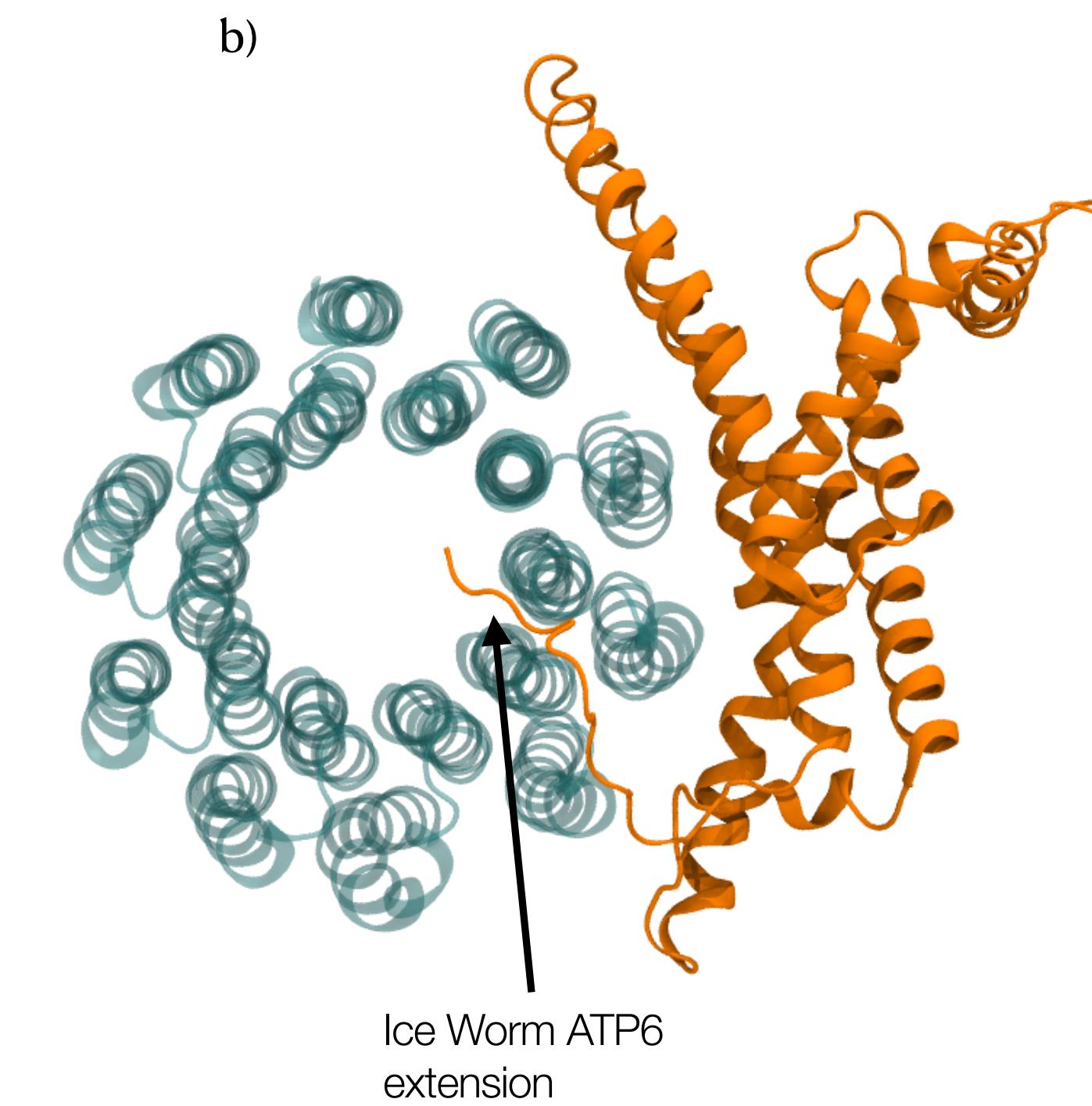
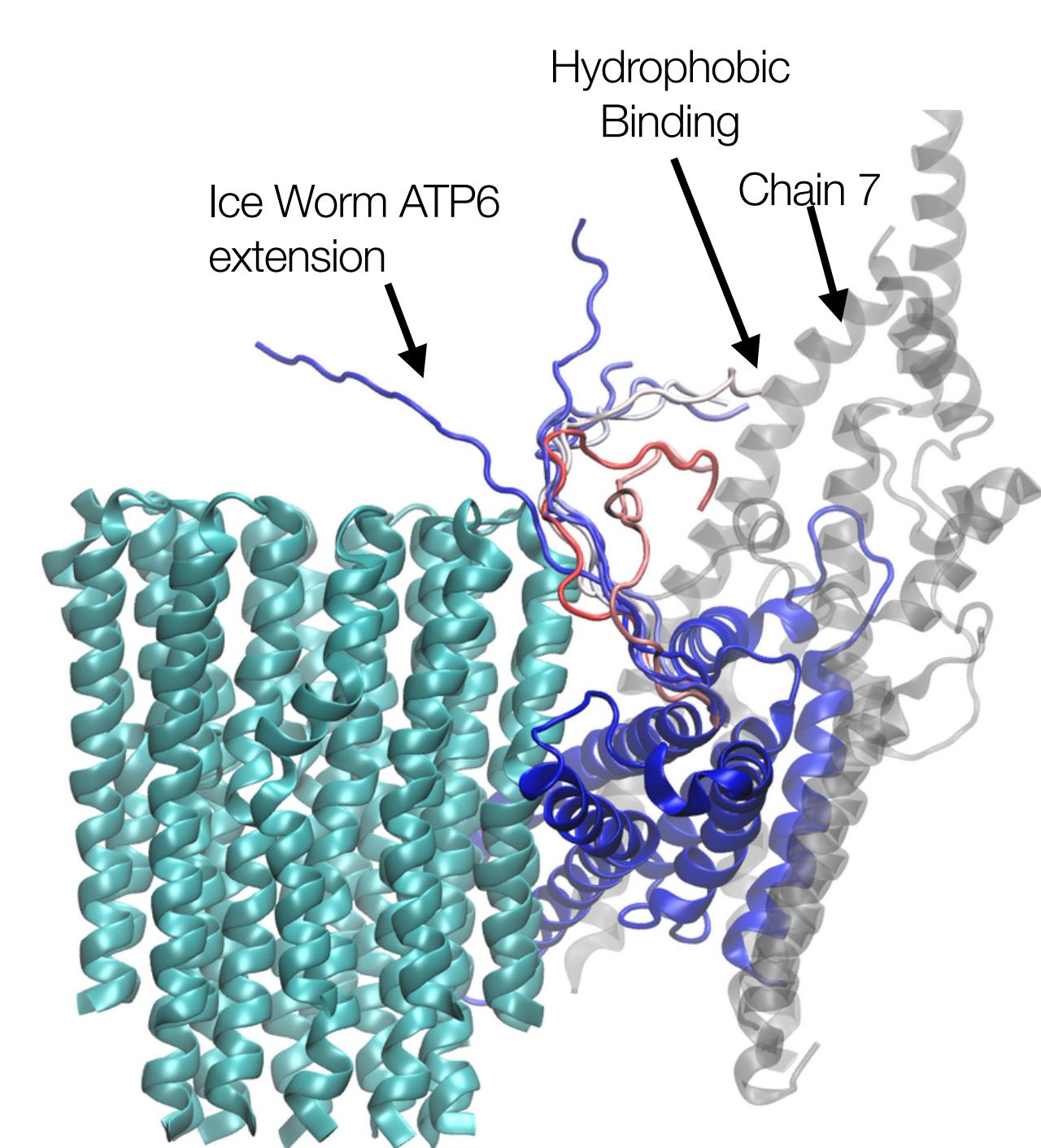
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Summary

- Calculated conservation scores and RMSF for ATP6: The termini are poorly conserved and the most dynamic in simulations
- Calculated Rotational Diffusion of the c-ring for the Yeast, Iceworm and Ice worm without the ATP6 extension F0 domains:
 - Rotational diffusion of the F0 c-ring is temperature-dependent. Faster diffusion at 310 K
 - Difference in rotational diffusion in the ice worm is significantly larger, compared to the other sequences: ATP6 extension has a role in increasing the temperature sensitivity of the protein

Summary

- At 277 K, the ATP6 extension binds to chain 7, which connects the F0 and F1 domains, a behavior that was not found in the ice worm system at 310 K



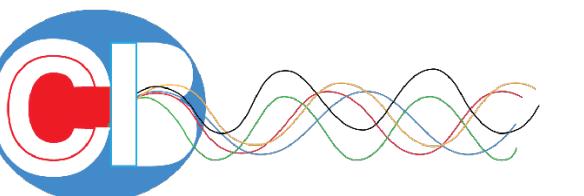
Thank You!

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Center (PSC)
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Questions?