

The N-terminus of α -Subunit Isoforms is Involved in
Signaling between V-ATPase and Cytohesin-2*^S

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SUPPLEMENTAL DATA

SUPPLEMENTAL LEGENDS

Table S1. Synthesis of V-ATPase α 1-, α 2-, α 3- and α 4-subunit isoforms derived peptides coupled with PEG or TAT sequence to render them soluble and cell permeable. Peptides were further coupled at the N-terminus with either: i) biotin (in blue), ii) fluorescein isothiocyanate (FITC)(in green), iii) 5-carboxy-fluorescein (Fluor)(in green) or 5-carboxy-teramethyl rhodamine (CTMR)(in red). After testing all peptides for their solubility and cell permeability in our experiments following peptides: i) Biotin- α 2N₁₋₁₇-TAT; ii) FITC- α 2N₁₋₁₇-TAT; iii) FITC- α 1N₁₋₁₇-TAT; iv) FITC- α 3N₁₋₁₇-TAT; v) FITC- α 4N₁₋₁₇-TAT as well as FITC-TAT as control were used.

Table S2. Structural statistics of the peptide α 2N₁₋₁₇.

Movie S1. Arf1/Sec7/ α 2N₁₋₁₇ complex.mov

Movie S2. Arf6/Sec7/ α 2N₁₋₁₇ complex.mov

Table S1

Peptide Synthesis #1: Peptides labeling #1:	<p>PEG - a2N₁₋₁₇ Biotin - PEG - a2N₁₋₁₇ Fluor - PEG - a2N₁₋₁₇</p>	<p>PEG - MGSLFRSESMCLAQLFL Biotin - PEG - MGSLFRSESMCLAQLFL Fluor - PEG - MGSLFRSESMCLAQLFL</p>
Peptide Synthesis #2: Peptides labeling #2:	<p>a2N₁₋₁₇ - PEG Biotin - a2N₁₋₁₇ - PEG Fluor - a2N₁₋₁₇ - PEG</p>	<p>MGSLFRSESMCLAQLFL - PEG Biotin - MGSLFRSESMCLAQLFL - PEG Fluor - MGSLFRSESMCLAQLFL - PEG</p>
Peptide Synthesis #3: Peptides labeling #3:	<p>TAT - a2N₁₋₁₇ Biotin - TAT - a2N₁₋₁₇ Fluor - TAT - a2N₁₋₁₇</p>	<p>YGRKKRRQRRRMGSLFRSESMCLAQLFL Biotin - YGRKKRRQRRRMGSLFRSESMCLAQLFL Fluor - YGRKKRRQRRRMGSLFRSESMCLAQLFL</p>
Peptide Synthesis #4: Peptides labeling #4:	<p>a2N₁₋₁₇ - TAT Biotin - a2N₁₋₁₇ - TAT Fluor - a2N₁₋₁₇ - TAT FITC - a2N₁₋₁₇ - TAT CTMR - a2N₁₋₁₇ - TAT</p>	<p>MGSLFRSESMCLAQLFLYGRKKRRQRRR Biotin - MGSLFRSESMCLAQLFLYGRKKRRQRRR Fluor - MGSLFRSESMCLAQLFLYGRKKRRQRRR FITC - MGSLFRSESMCLAQLFLYGRKKRRQRRR CTMR - MGSLFRSESMCLAQLFLYGRKKRRQRRR</p>
Peptide Synthesis #5: Peptides labeling #5:	<p>TAT FITC - TAT CTMR - TAT</p>	<p>YGRKKRRQRRR FITC - YGRKKRRQRRR CTMR - YGRKKRRQRRR</p>
Peptide Synthesis #6: Peptides labeling #6:	<p>a1N₁₋₁₇ - TAT a3N₁₋₁₇ - TAT a4N₁₋₁₇ - TAT</p>	<p>MGELFRSEEMTLAQLFLYGRKKRRQRRR MGSMFRSEEVQVLLLYGRKKRRQRRR MASVFRSEEMCLSQVFLYGRKKRRQRRR</p>
	<p>FITC - a1N₁₋₁₇ - TAT FITC - a3N₁₋₁₇ - TAT FITC - a4N₁₋₁₇ - TAT</p>	<p>FITC - MGELFRSEEMTLAQLFLYGRKKRRQRRR FITC - MGSMFRSEEVQVLLLYGRKKRRQRRR FITC - MASVFRSEEMCLSQVFLYGRKKRRQRRR</p>

Table S2**Structural statistics of the peptide a2N₁₋₁₇**

Distance restraints

Total	233
Intraresidue ($i - j = 0$)	77
Sequential ($ i-j = 1$)	81
Medium-range ($2 \leq i-j \leq 4$)	75
Long-range ($ i-j \geq 5$)	0

Average number of violations

Distance violations $> 0.5 \text{ \AA}$	0
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Ramachandran plot² (%)

Residues in most favoured regions	78.2
Residues in additionally allowed regions	21.8
Residues in generously allowed regions	0
Residues in disallowed regions	0

Average RMSD to Mean (\AA)

Residues 11-19, Backbone (C^α , C' , and N)	0.311
rmsd heavy atoms	1.355