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Model Building of PI3KC3-C1/RAB1A

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ABSTRACT

Details model building of the PI3KC3-C1/RAB!A complex using chimeraX1.5-1.6 and ISOLDE. Refinement was done using Phenix real space refinement.

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protocols.io

https://protocols.io/view/mod el-building-of-pi3kc3-c1rab1a-c2svyee6

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Pettersen, E. F. et al. UCSF ChimeraX: Structure visualization for researchers, educators, and developers. Protein Sci30, 70-82 (2021). https://doi.org:10.1002/pro.3 943

Croll, T. I. ISOLDE: a physically realistic environment for model building into low-resolution electron-density maps. Acta Crystallogr D Struct Biol 74, 519-530 (2018).

https://doi.org:10.1107/S205 9798318002425

Pavel V Afonine, et al. Realspace refinement in PHENIX for cryo-EM and crystallography, ,Acta Crystallogr D Struct Biol. 2018 Jun 1; 74(Pt 6): 531-544.

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Generate Starting model

1 Use Alphafold2 to generate a starting model containing the PI3KC3-C1 components (VPS34, VPS15, ATG14, BECN1) and RAB1A respectively.

Docking and model building/refinement

- 2 Dock each starting model into the experimental map
- 3 Use ISOLDE to relax the entire PI3KC3-C1 complex and RAB1A into the experimental data in ChimeraX, selectively using restraints and ignoring clashes until the model simulates correctly. Trim obvious disordered regions by deleting them in Chimera.
- **4** Build ligands into the map, use the ignore command to ignore clashes selectively until the clashes have been eliminated.

5	Iteratively refine the model by building sidechains into density features, and improving the Ramachandran angles with selective restraints.
6	Assess the model to map fit using the built in Q-score function and Ramachandran plot
7	When model fit to density and Ramachandran angles start to look good, export the model and refine using Phenix real space refinement.
8	Repeat steps 5-7 until you are satisfied with the final model and the metrics cease improvement.