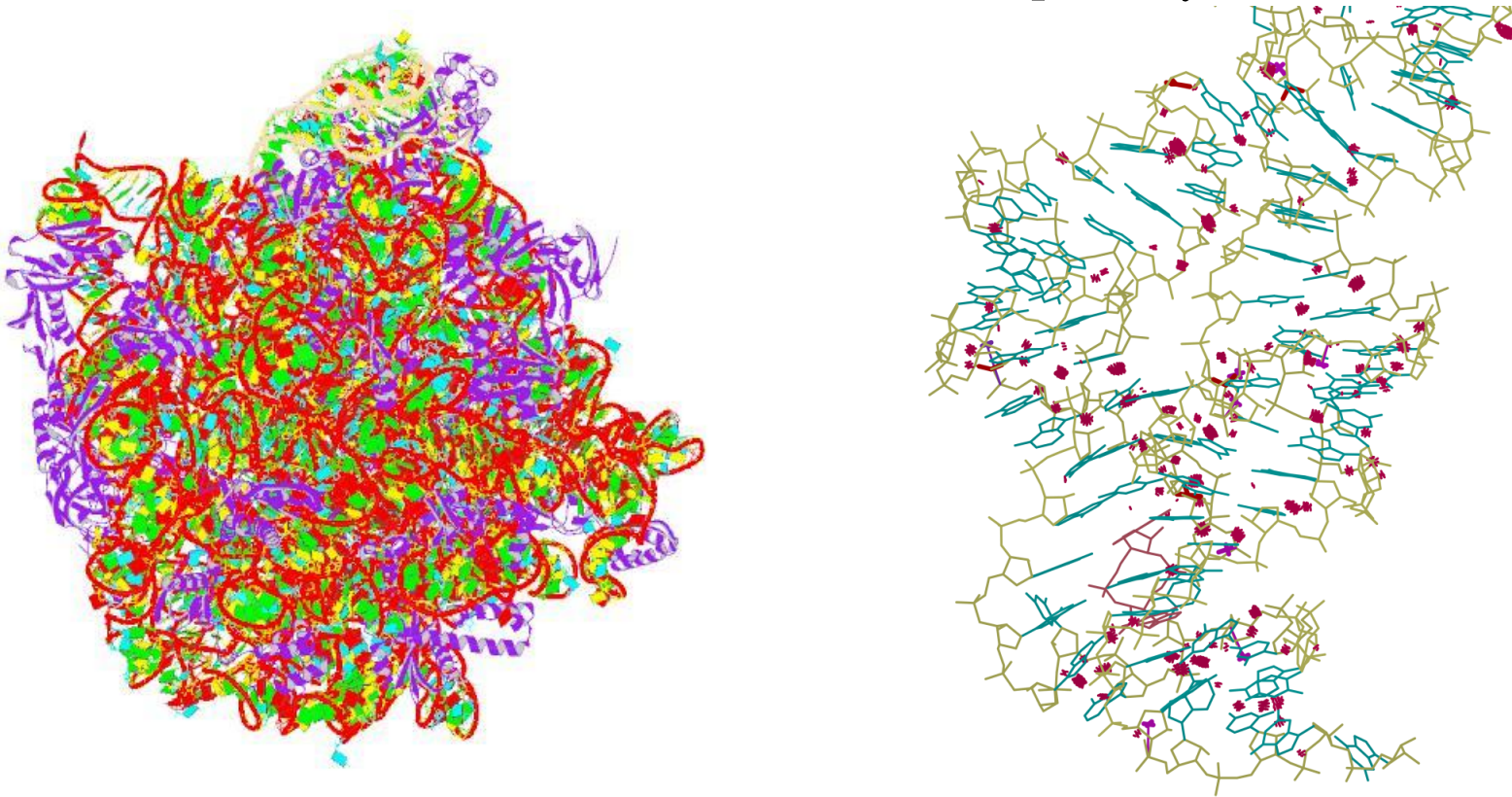


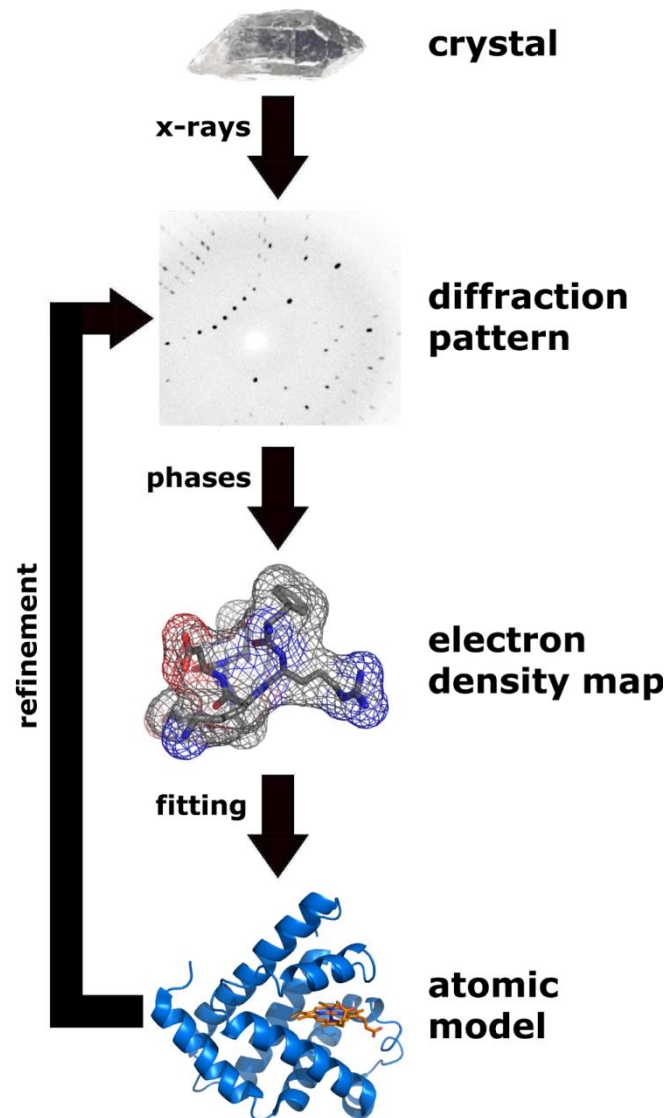
Improving RNA Crystallographic Models Using Rosetta

Fang-Chieh Chou (Das Lab)
For RosettaCon 2011

- The number RNA crystal structures increased explosively after the first Ribosome crystal structure is solved in 2000 (118 → 1484).
- However multiple errors exist in the current RNA crystal structures, which can be revealed in Molprobity.

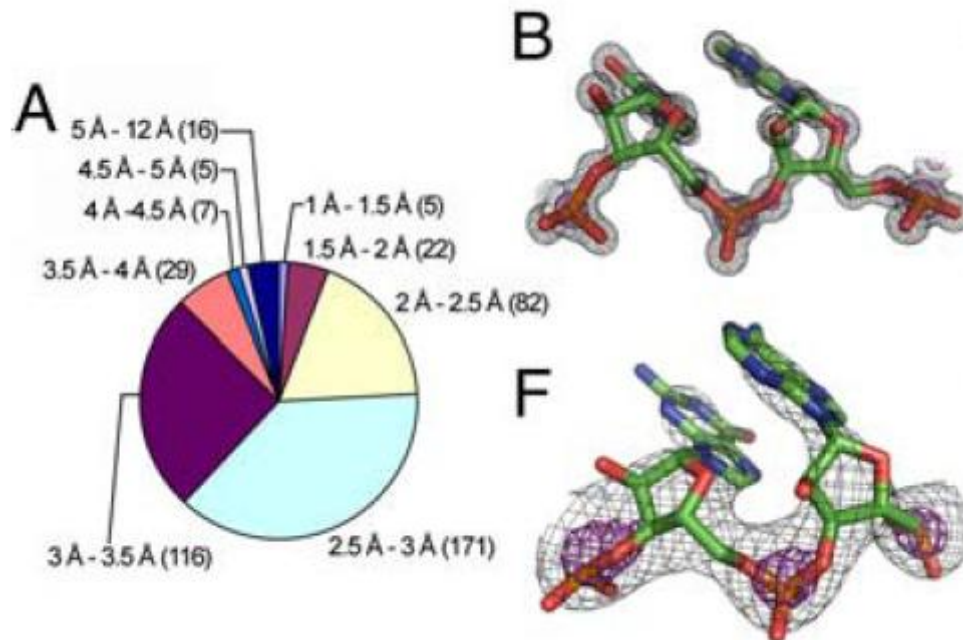


Determination of X-ray Structure



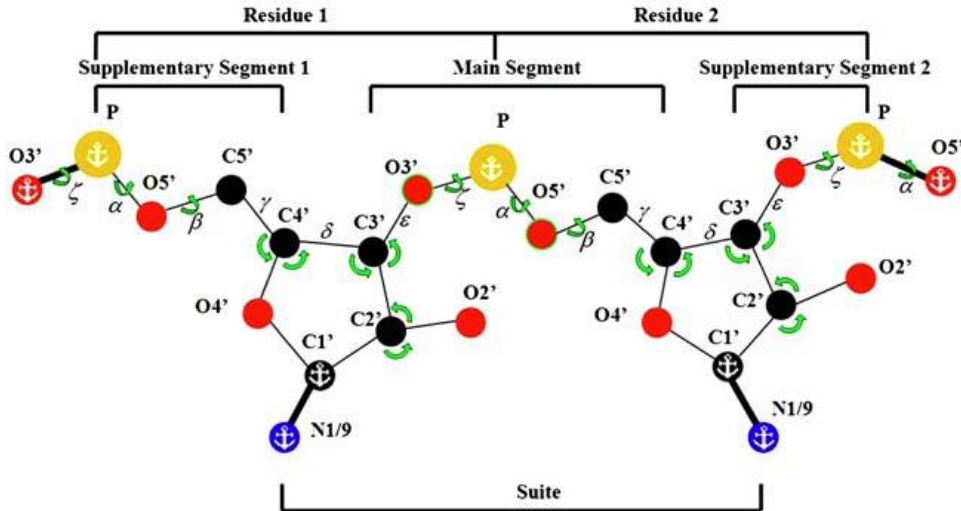
Limits of Crystallography

- Position of atoms become ambiguous when the resolution is low (especially for RNA sugar ring).
- The majority of the current RNA structure has a resolution below 2.5 Å, and they may contain multiple errors.

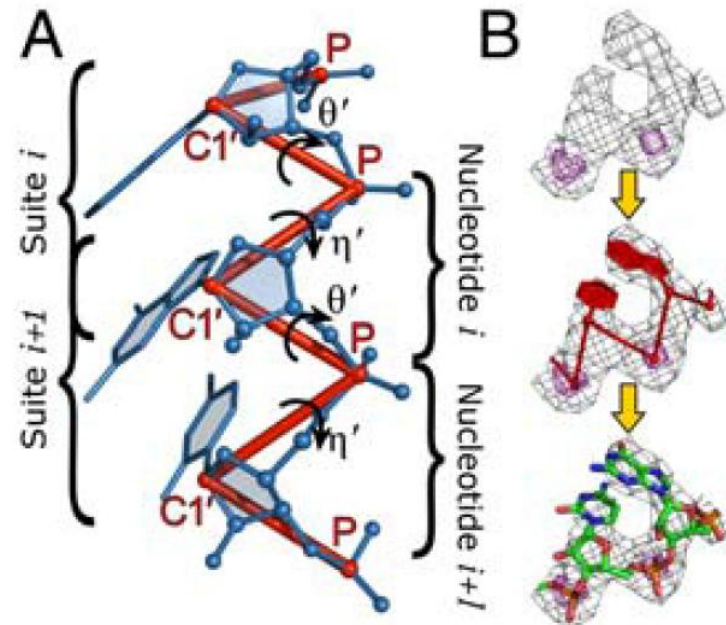


Current Status of RNA Model Improvement Tools

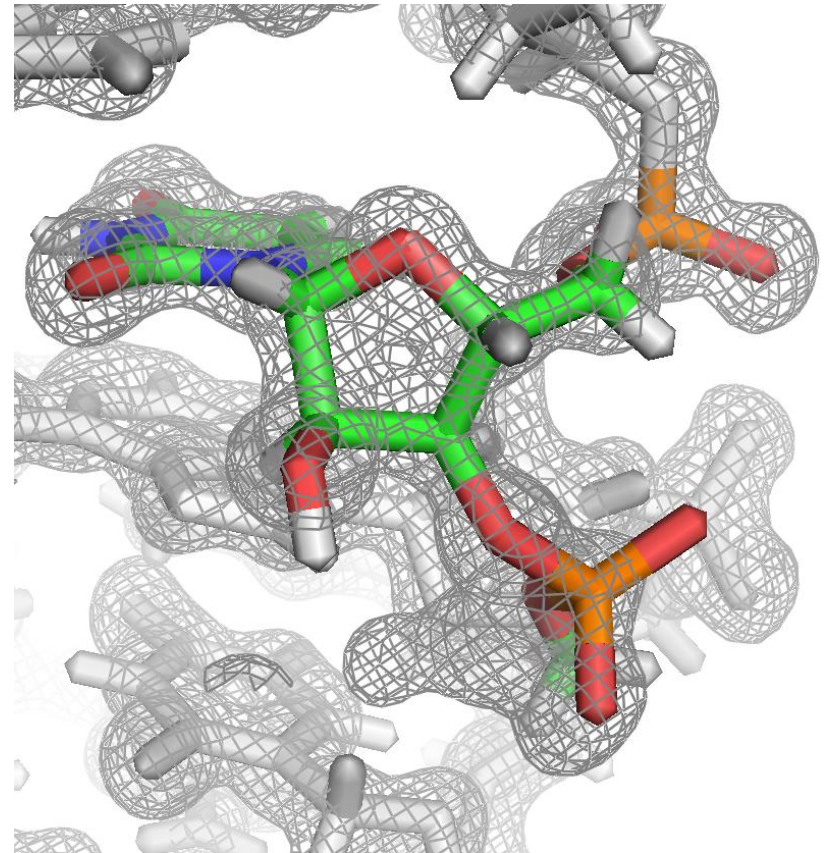
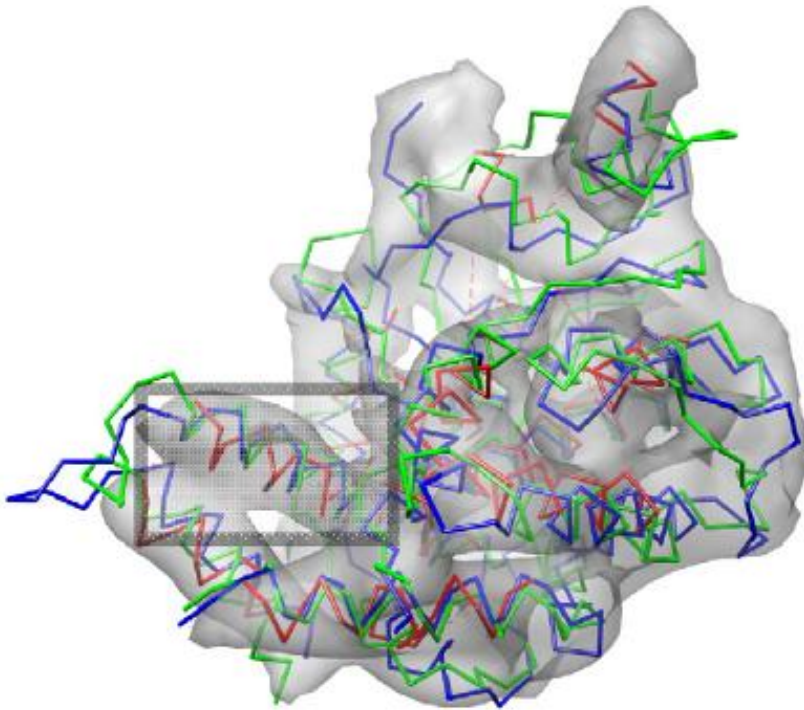
RNABC



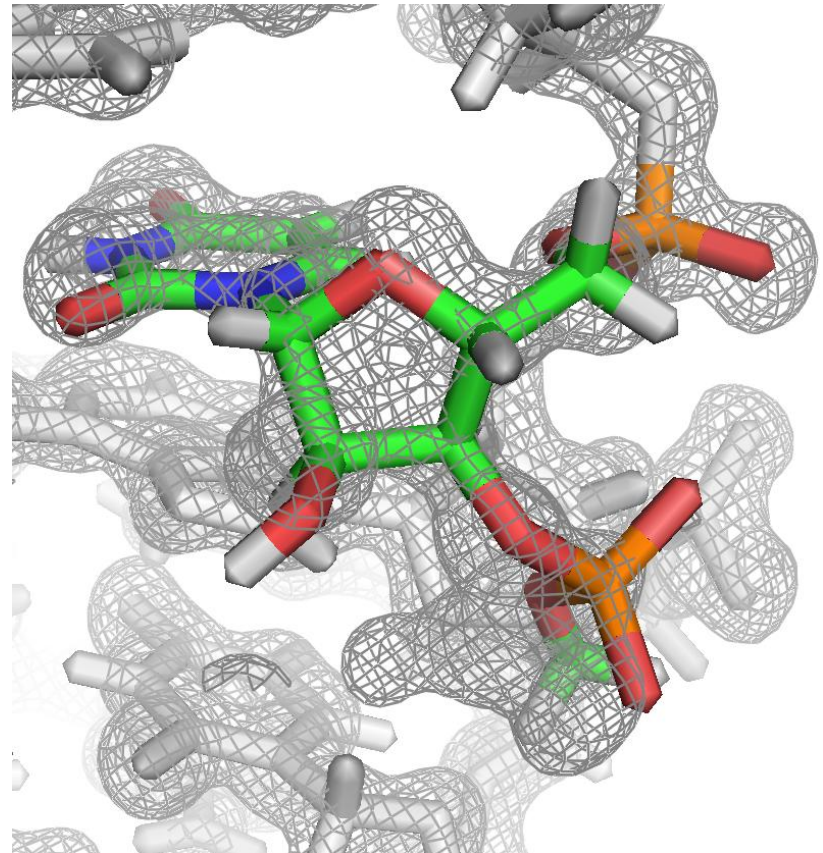
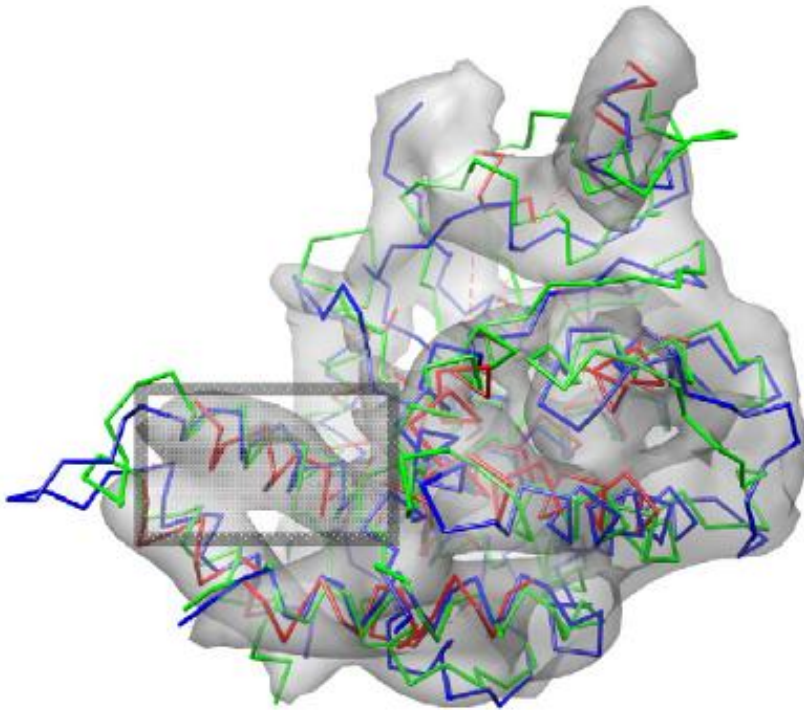
RCrane



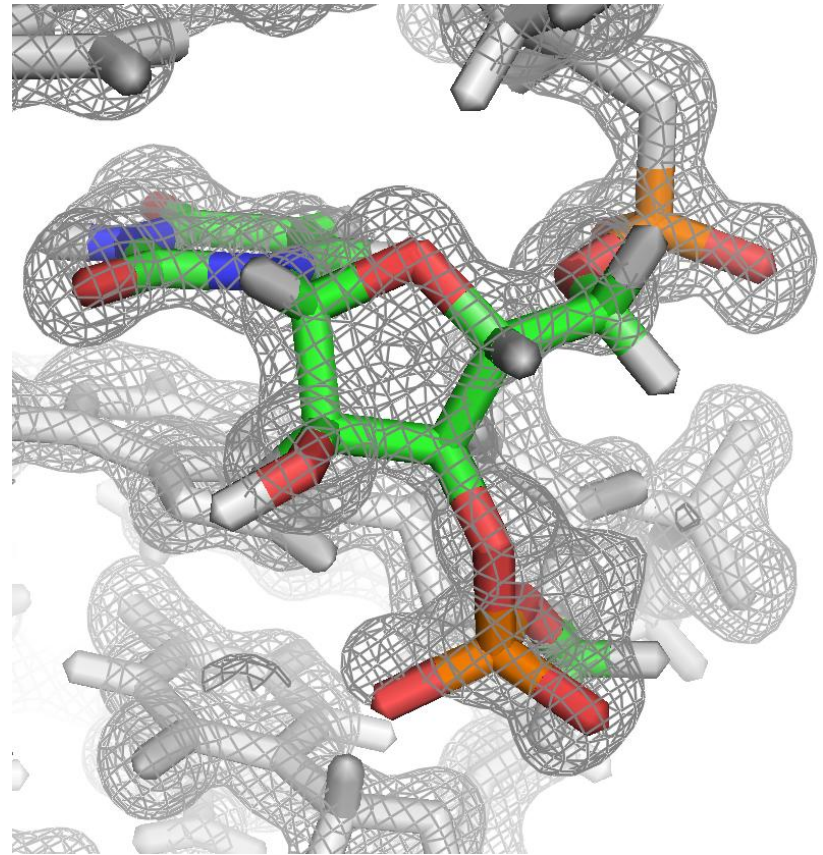
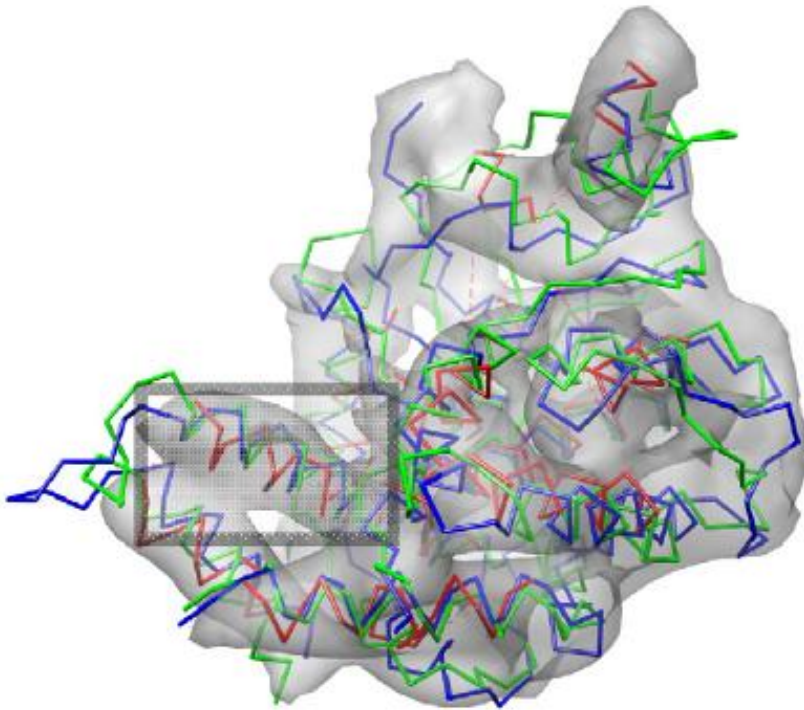
Tools in Rosetta: Electron Density Scoring and Stepwise Assembly (SWA)



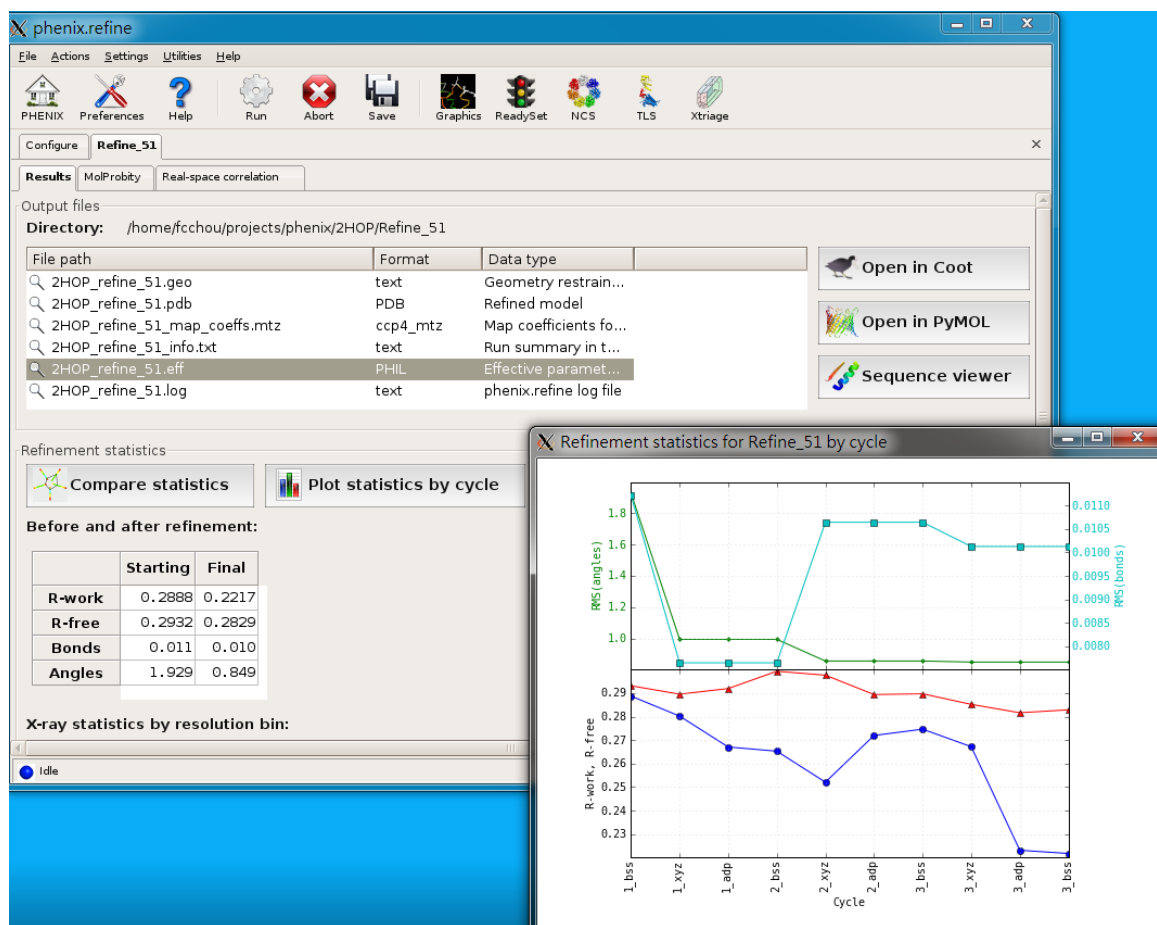
Tools in Rosetta: Electron Density Scoring and Stepwise Assembly (SWA)



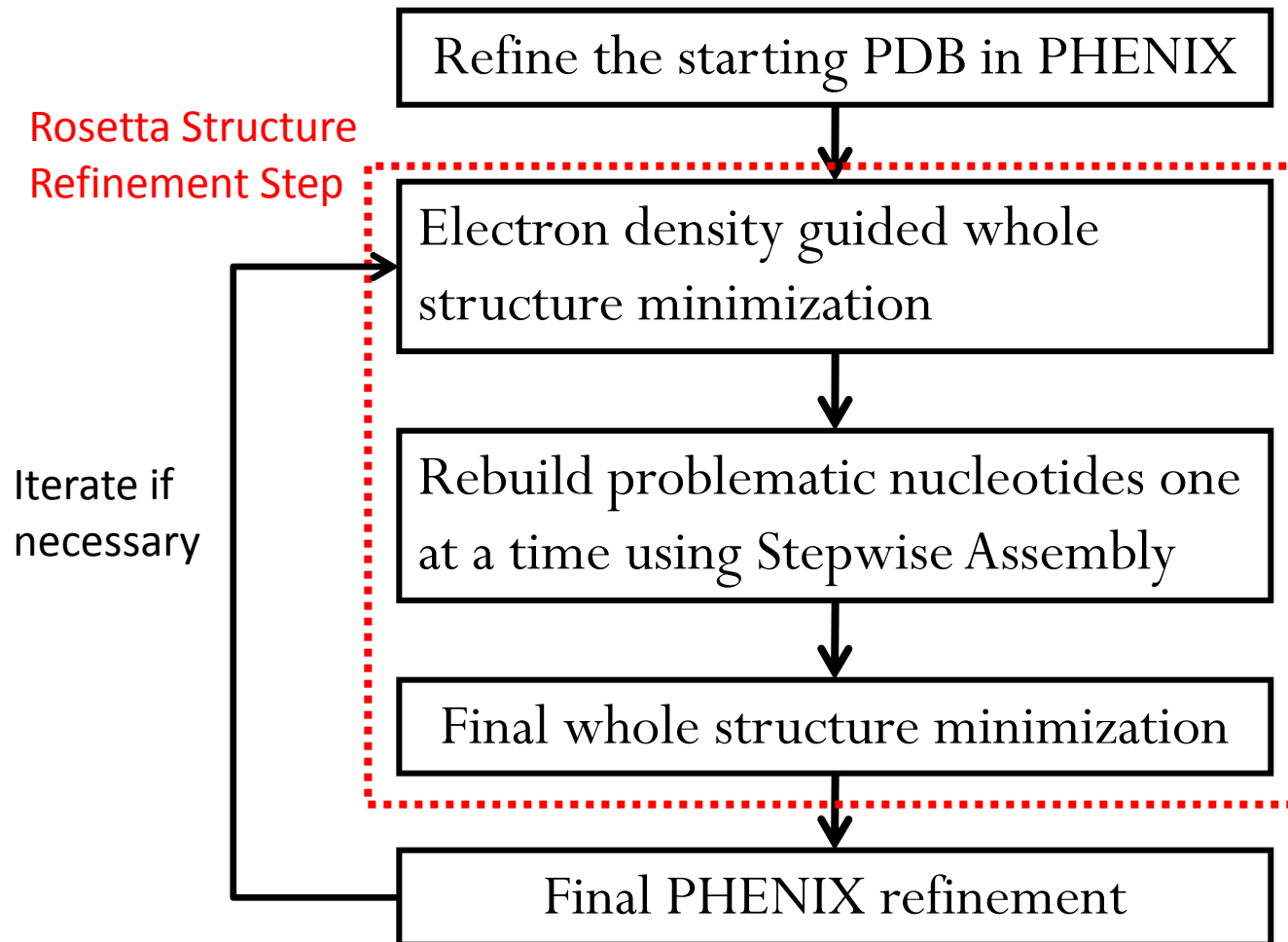
Tools in Rosetta: Electron Density Scoring and Stepwise Assembly (SWA)



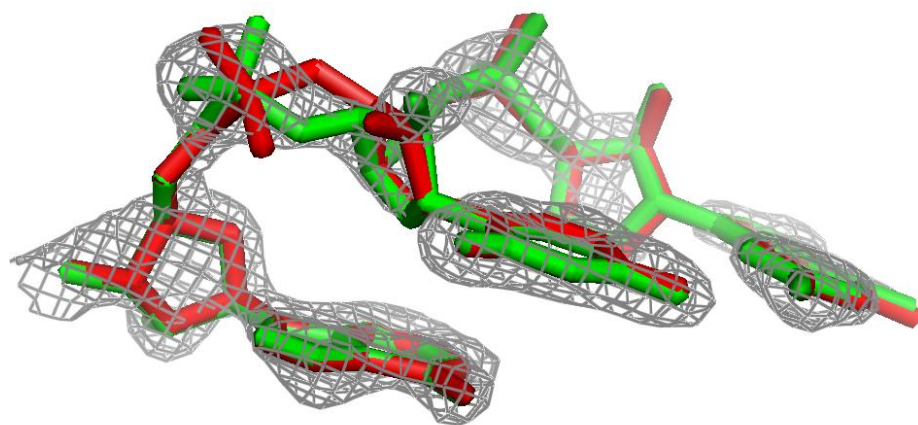
PHENIX: Structure Refinement Tool



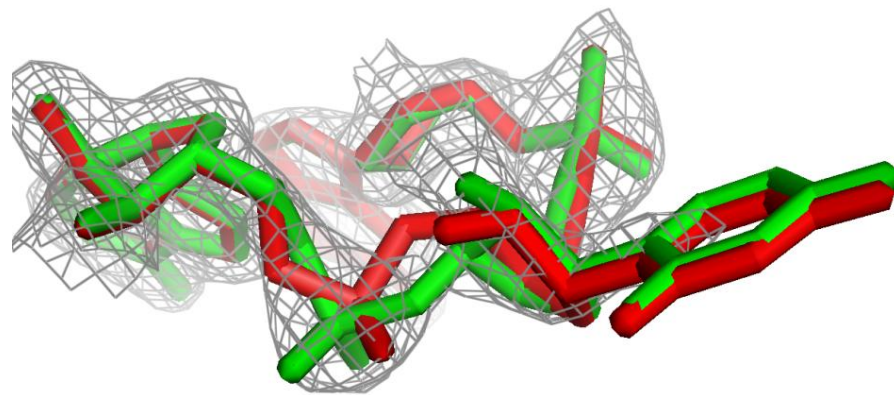
Overall Strategy



Improvement of Backbone



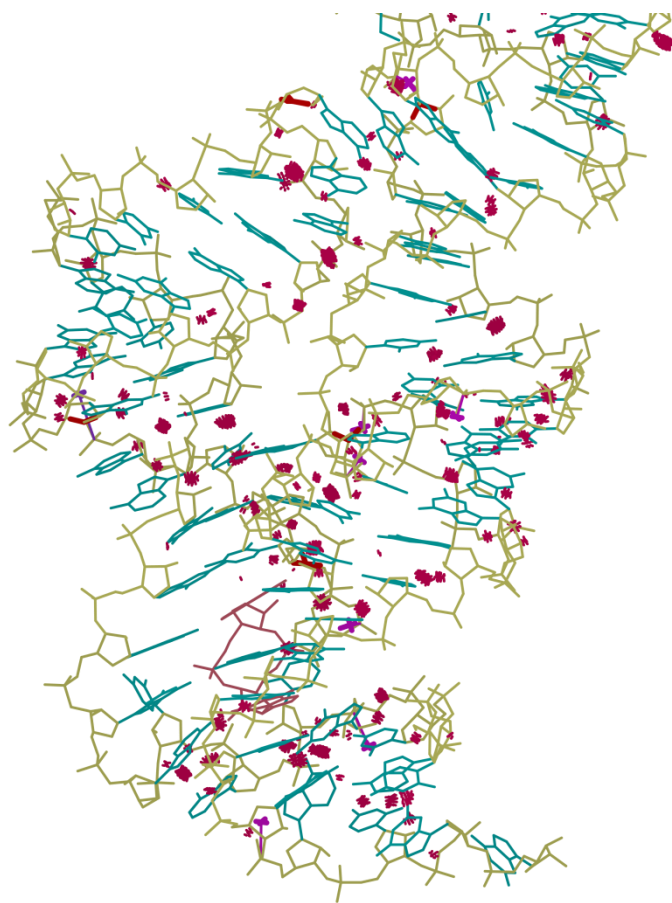
1U8D (1.95 Å)



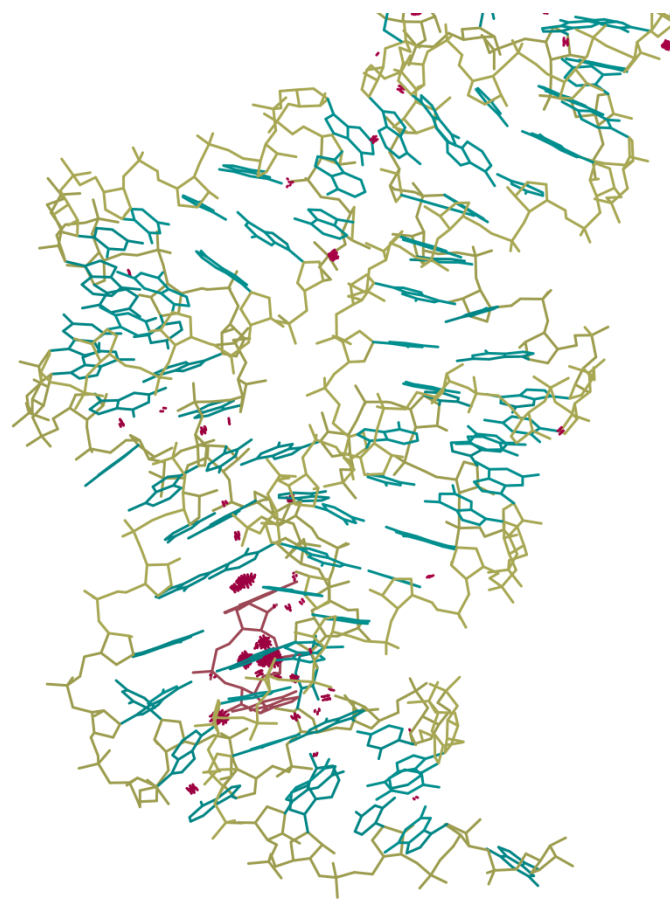
2GDI (2.05 Å)

Red: PDB, Green: Rosetta/PHENIX

Improvement of Clashes



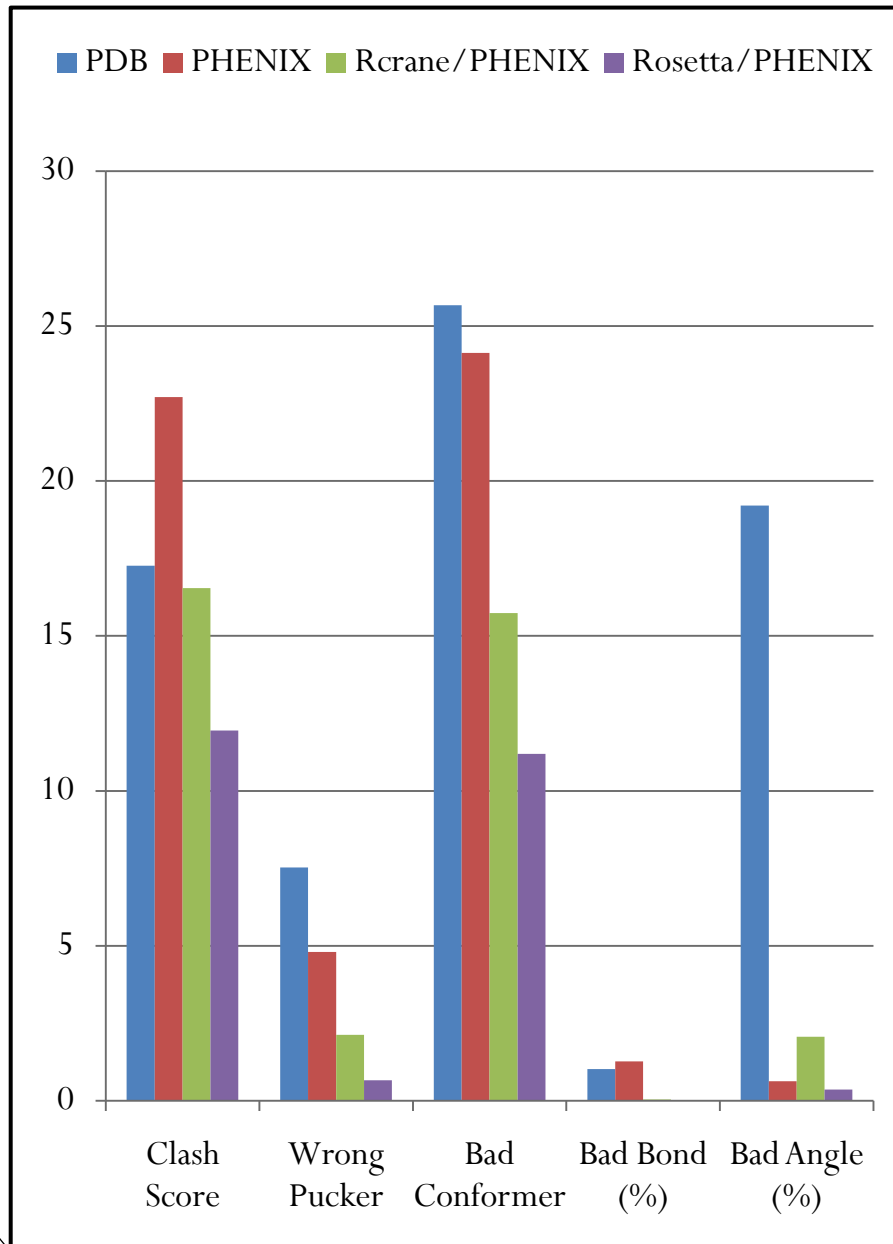
PDB



Rosetta/PHENIX

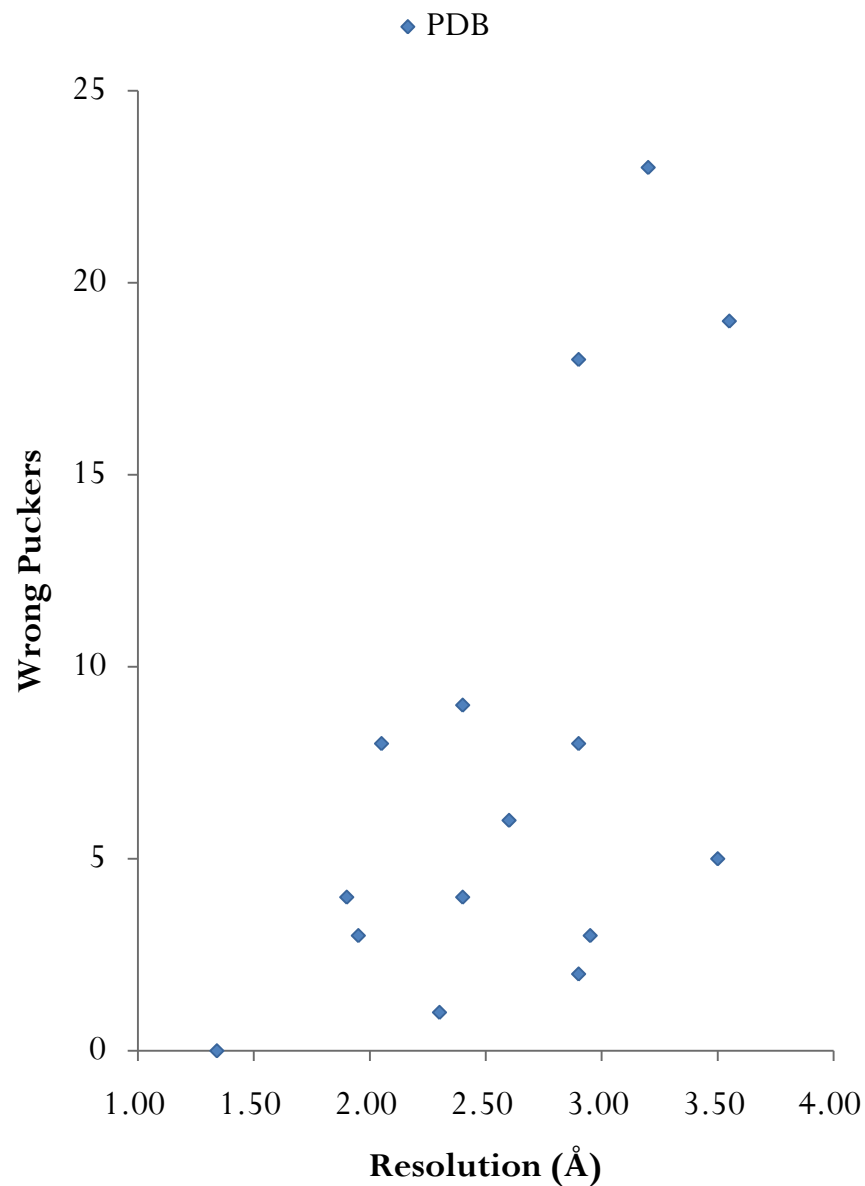
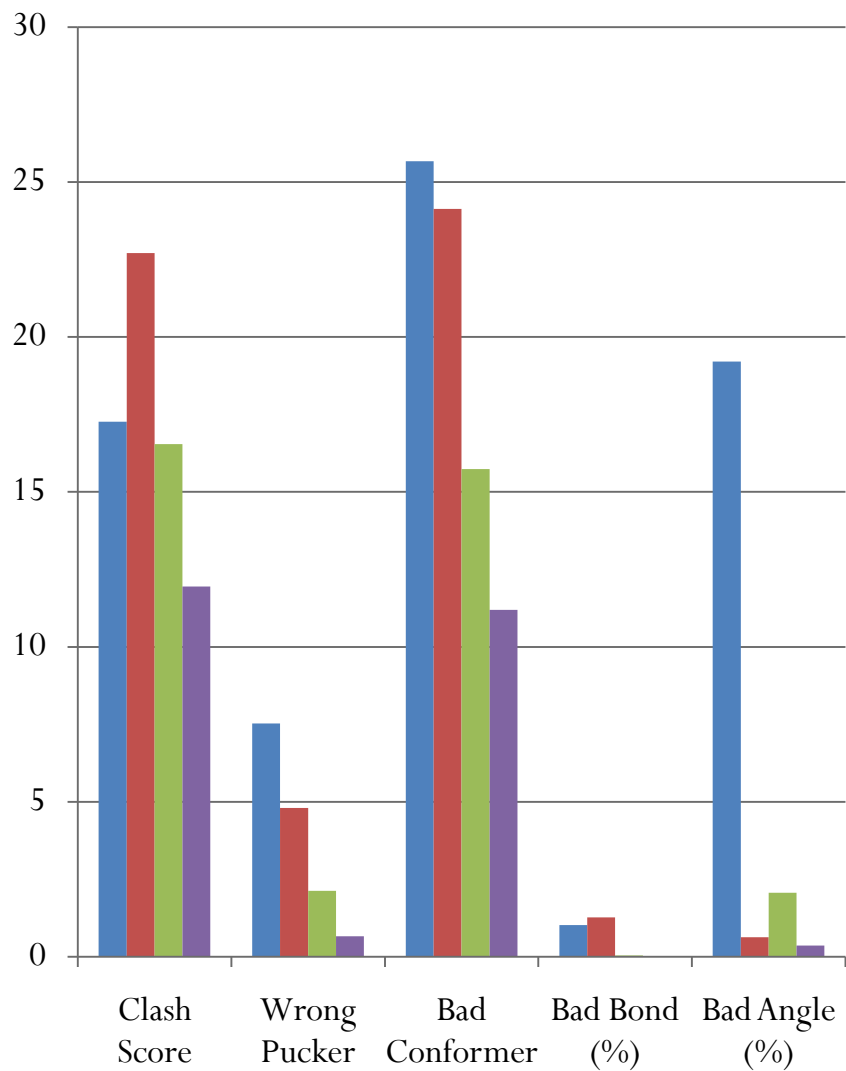
From 3IWN(3.2 Å)

15 test cases are remodeled



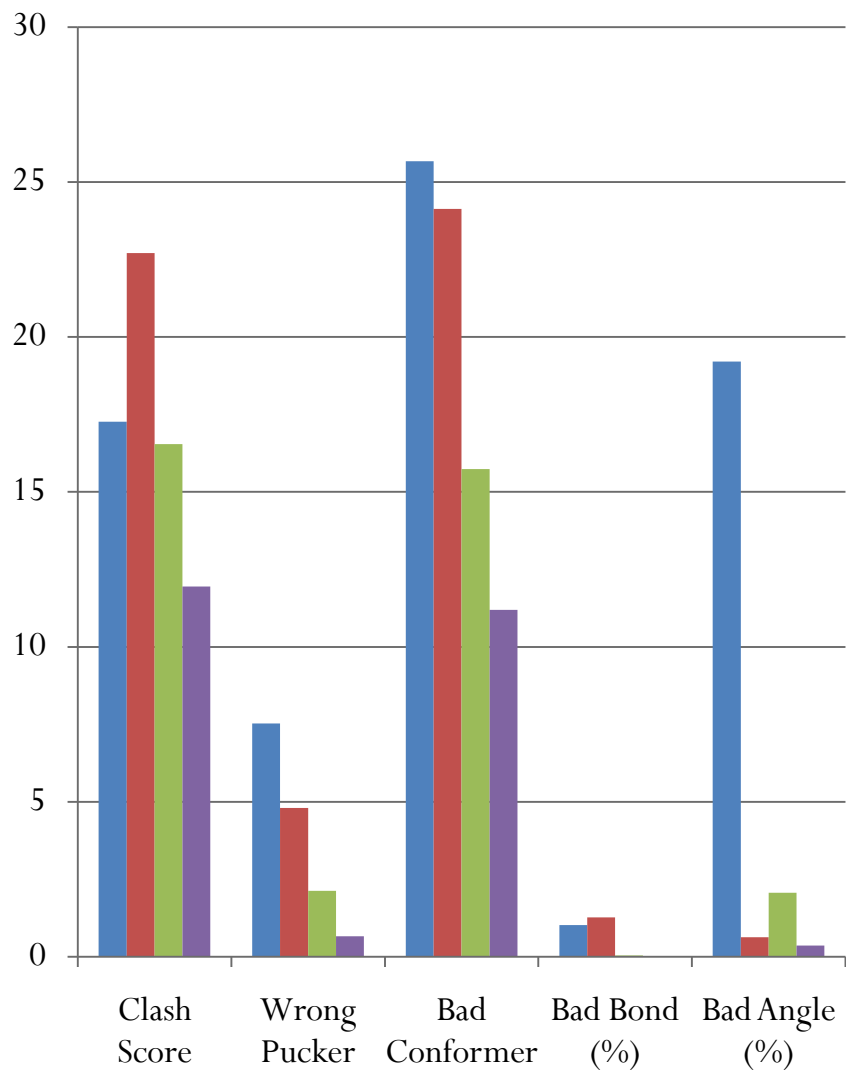
15 test cases are remodeled

■ PDB ■ PHENIX ■ Rcrane/PHENIX ■ Rosetta/PHENIX

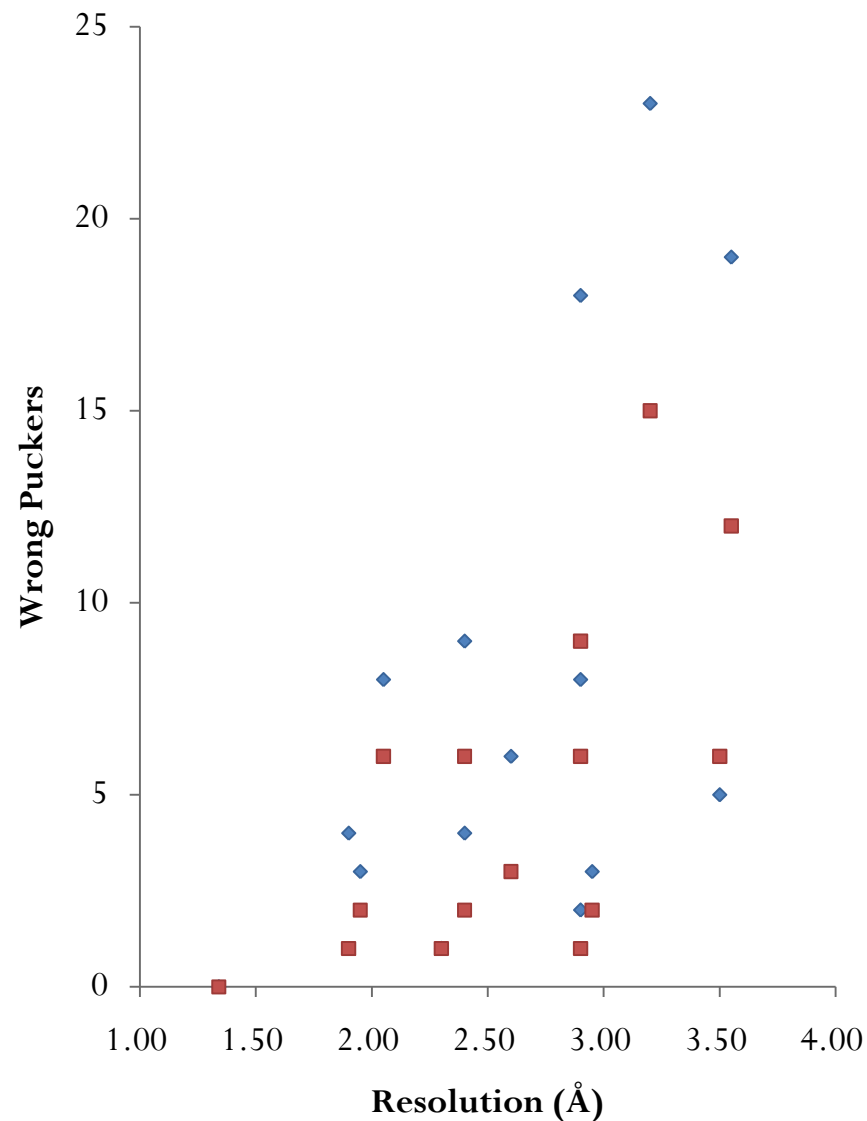


15 test cases are remodeled

■ PDB ■ PHENIX ■ Rcrane/PHENIX ■ Rosetta/PHENIX

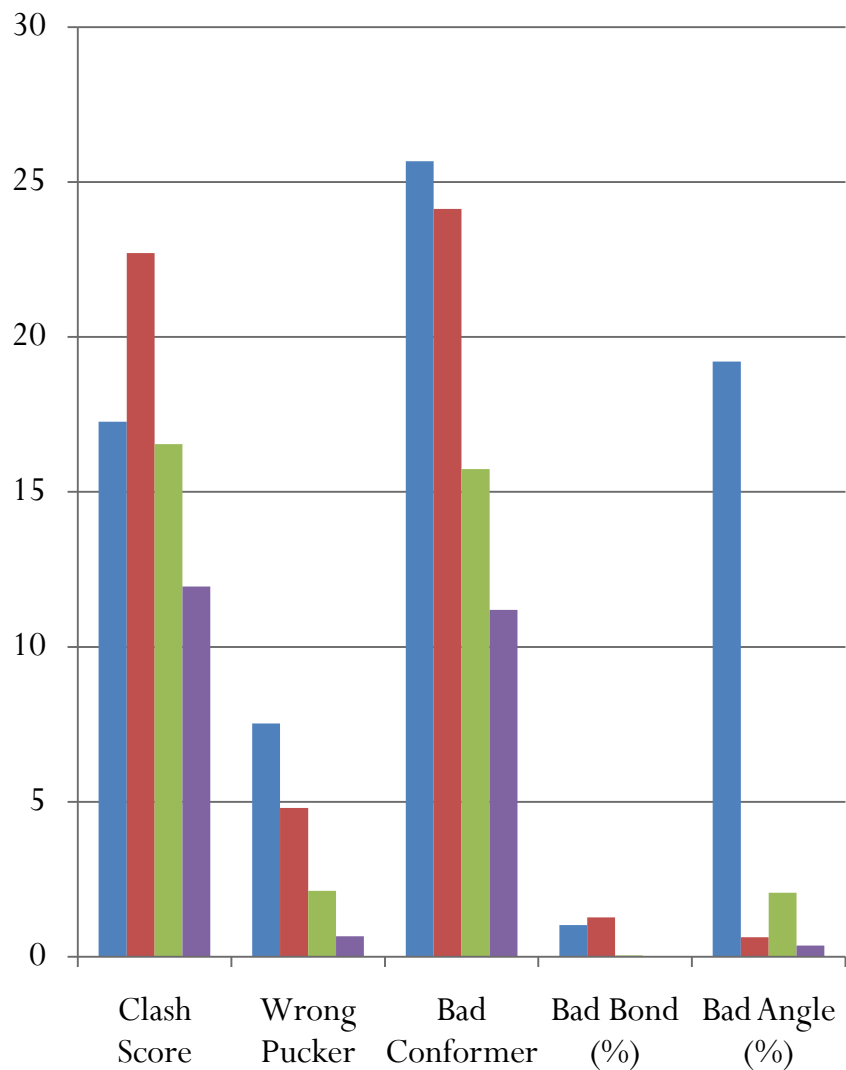


◆ PDB ■ PHENIX

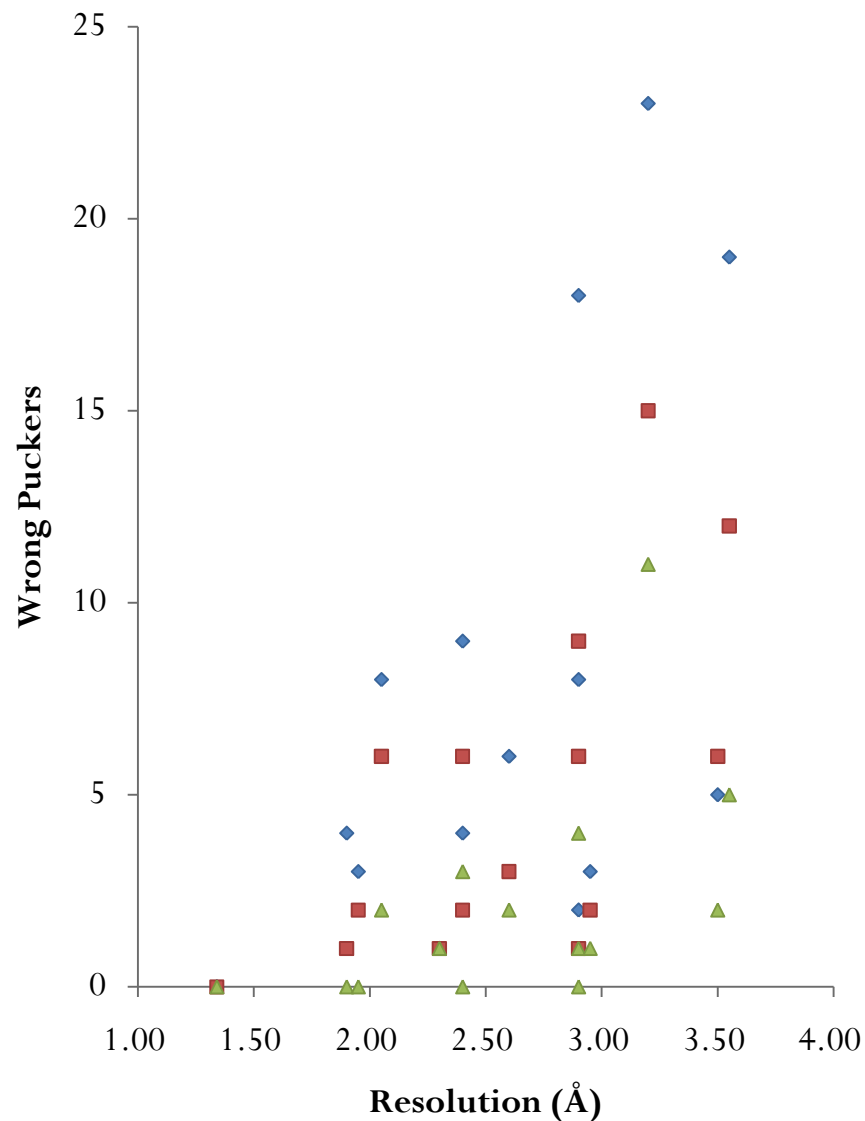


15 test cases are remodeled

■ PDB ■ PHENIX ■ Rcrane/PHENIX ■ Rosetta/PHENIX

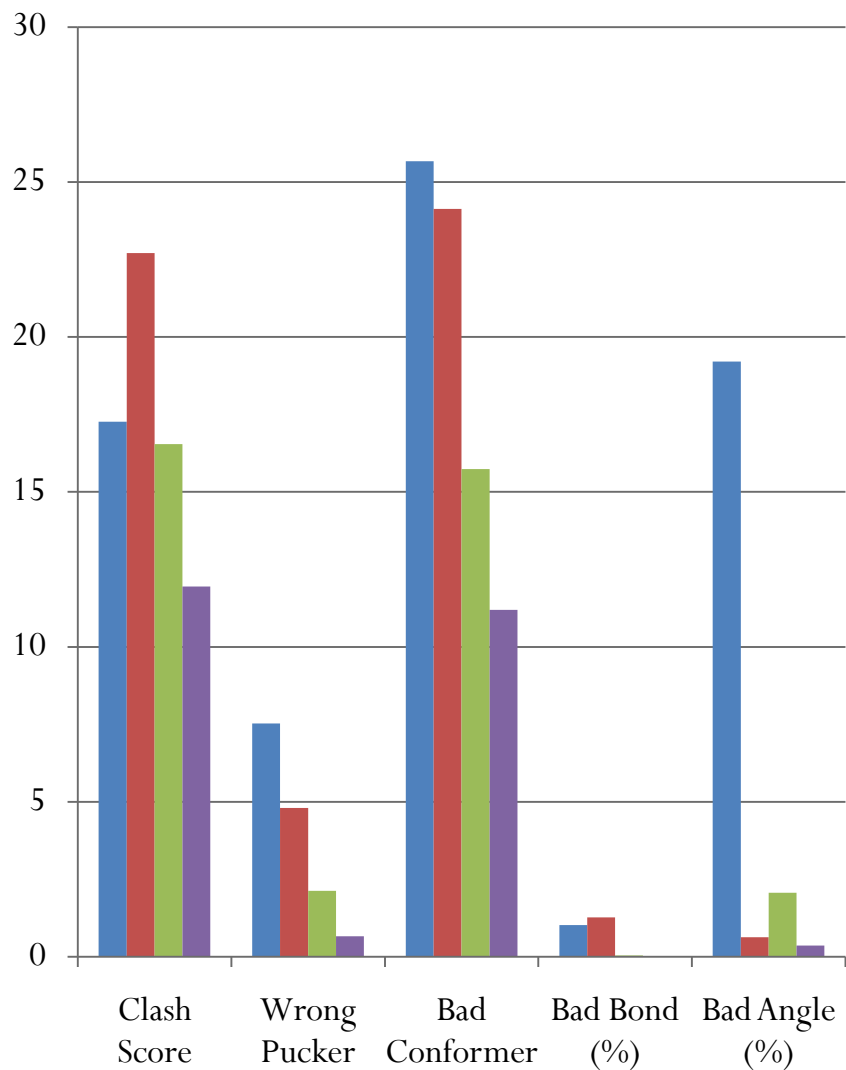


◆ PDB ■ PHENIX ▲ Rcrane/PHENIX

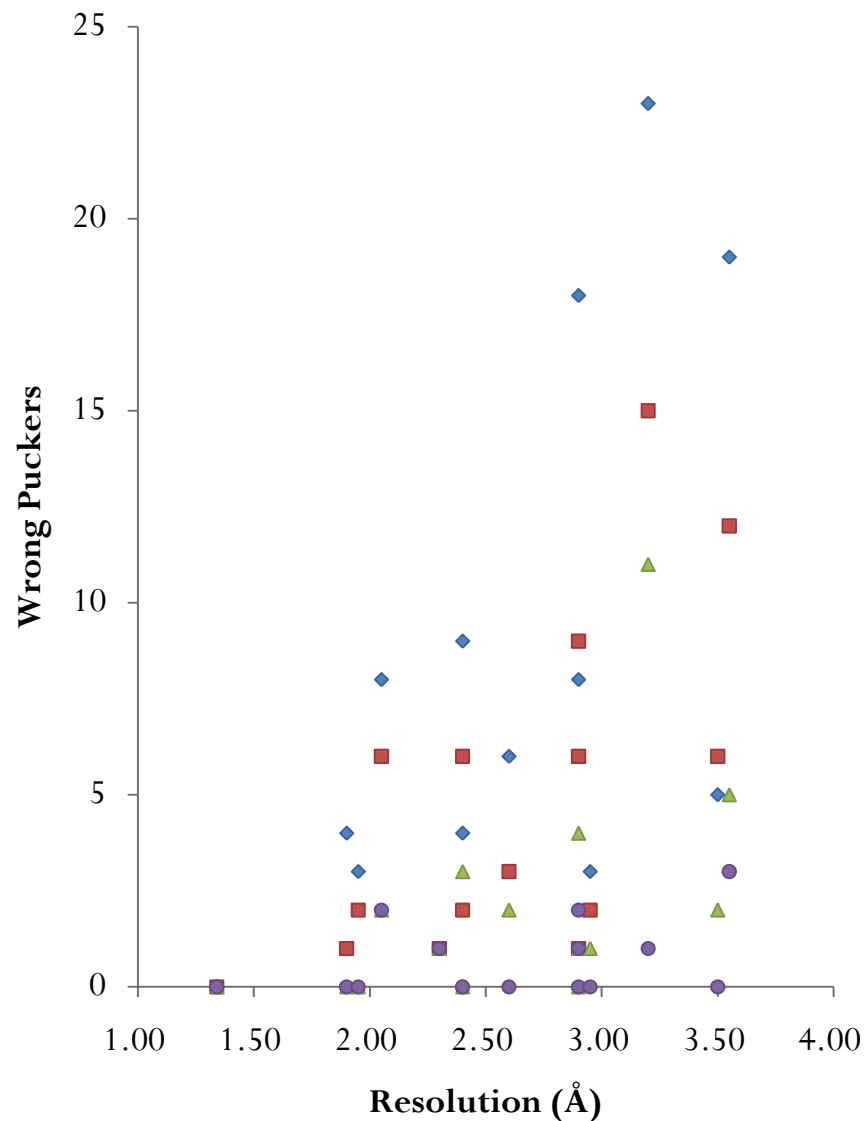


15 test cases are remodeled

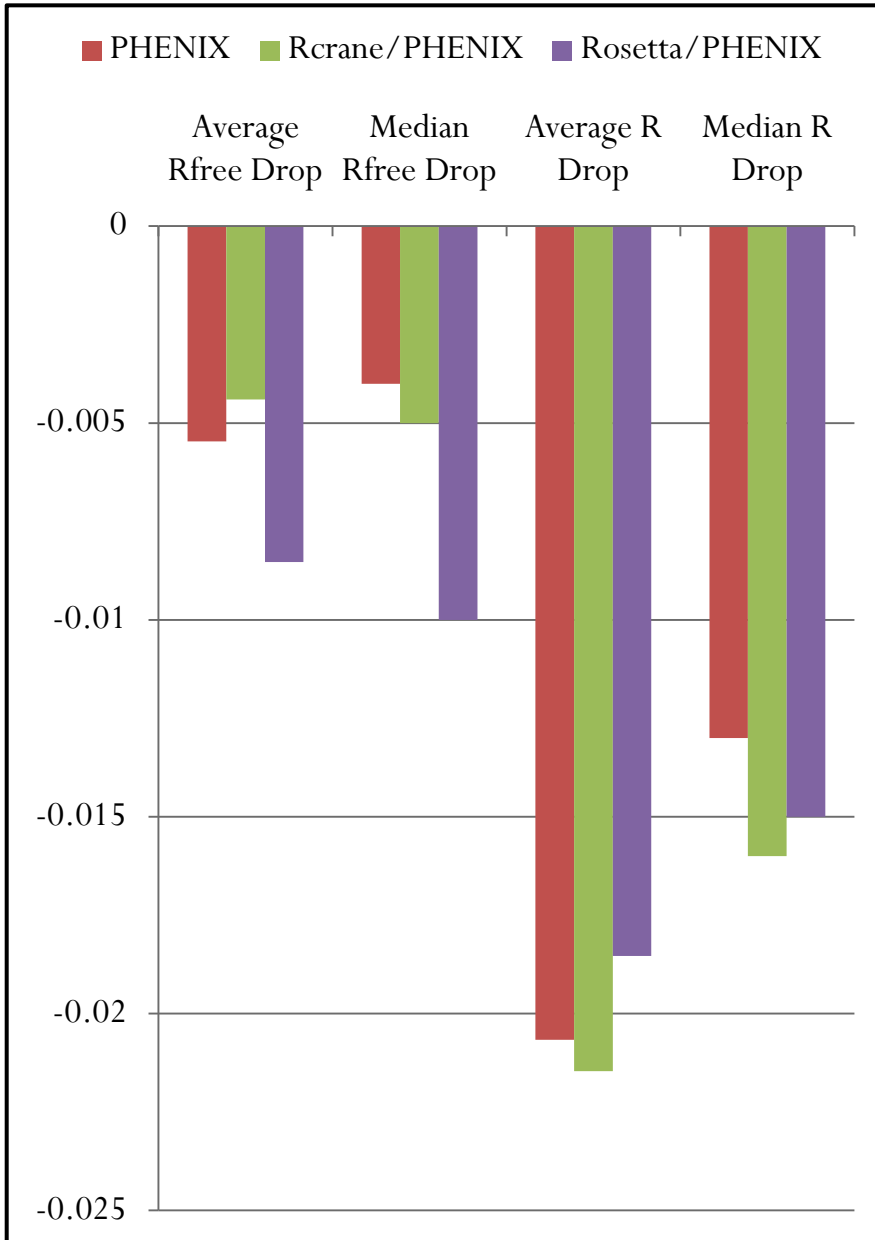
■ PDB ■ PHENIX ■ Rcrane/PHENIX ■ Rosetta/PHENIX



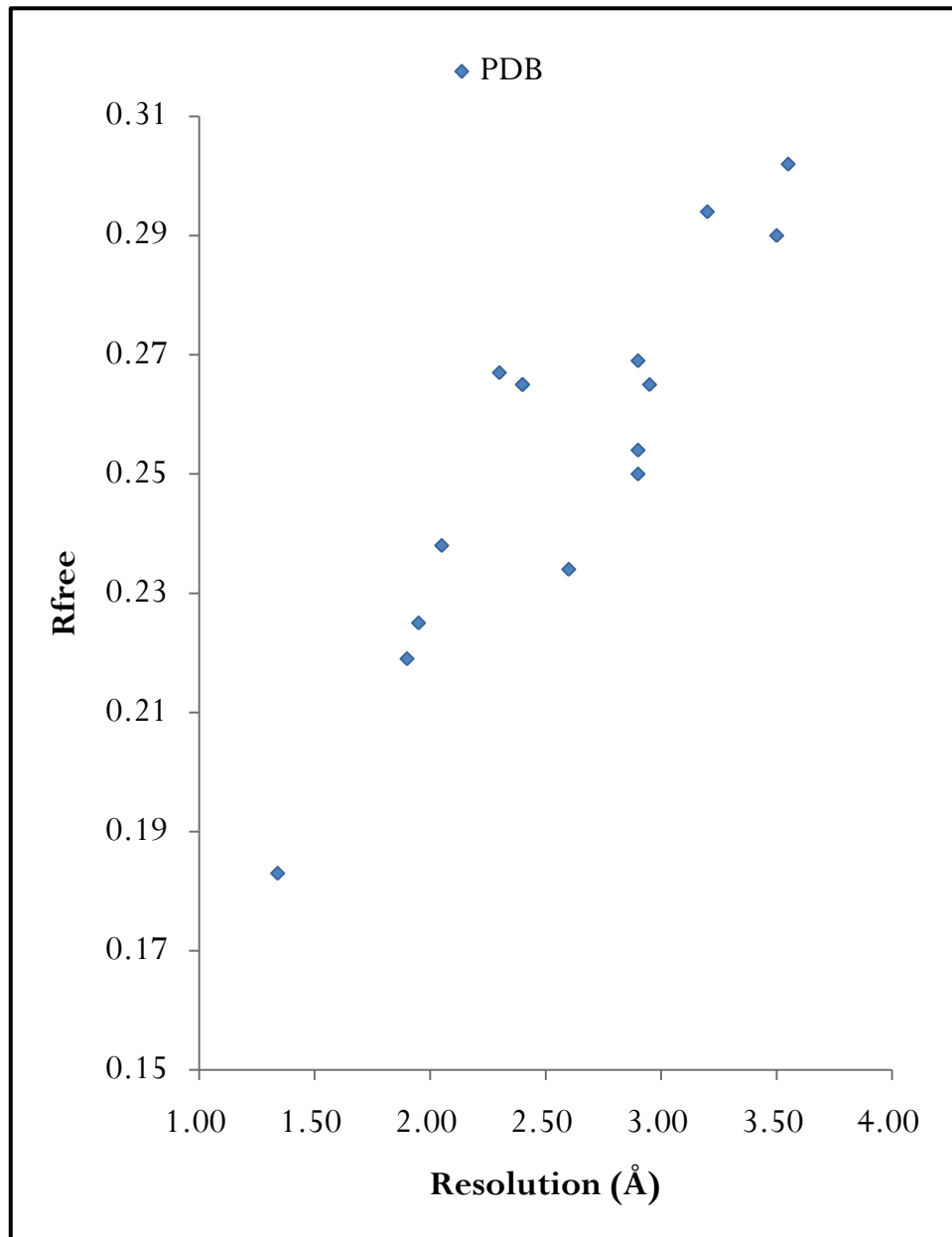
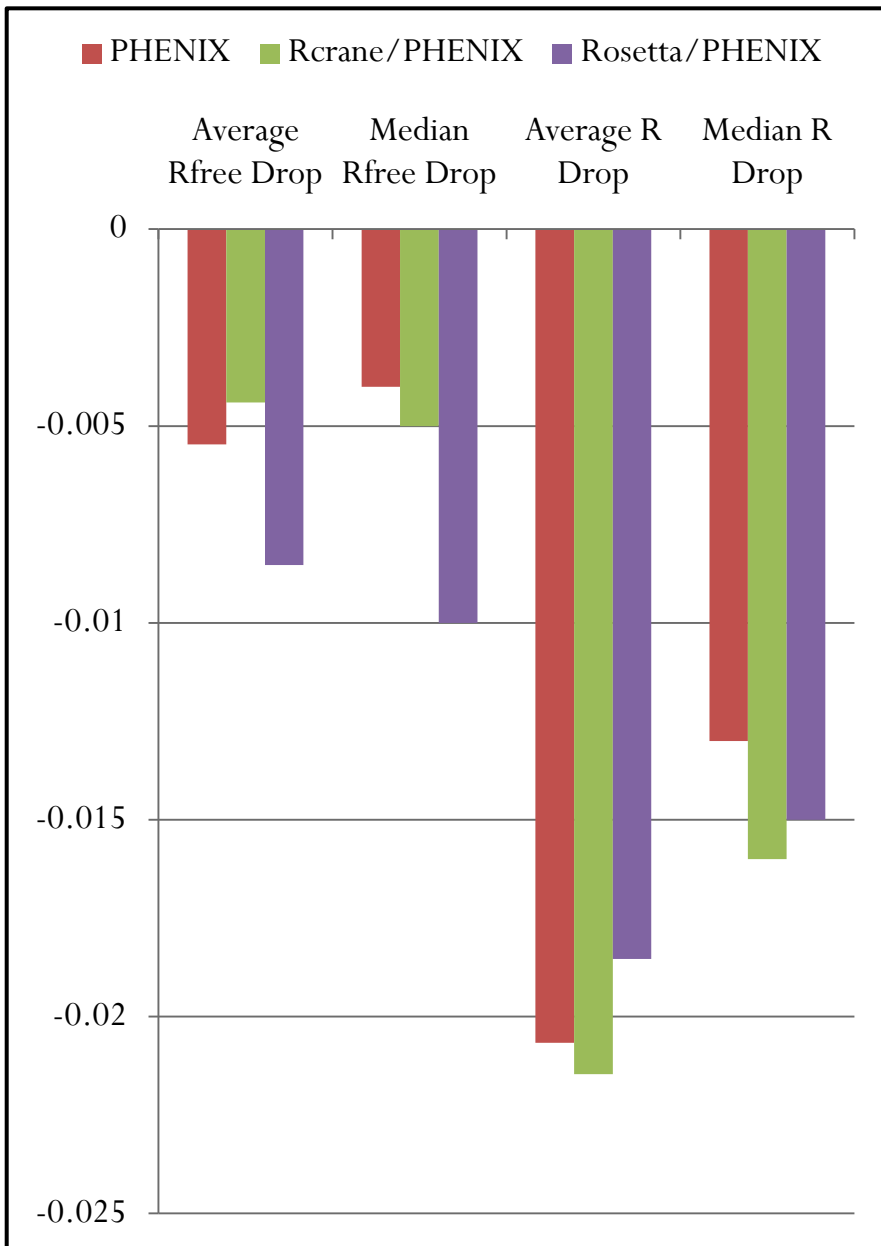
◆ PDB ■ PHENIX ▲ Rcrane/PHENIX ● Rosetta/PHENIX



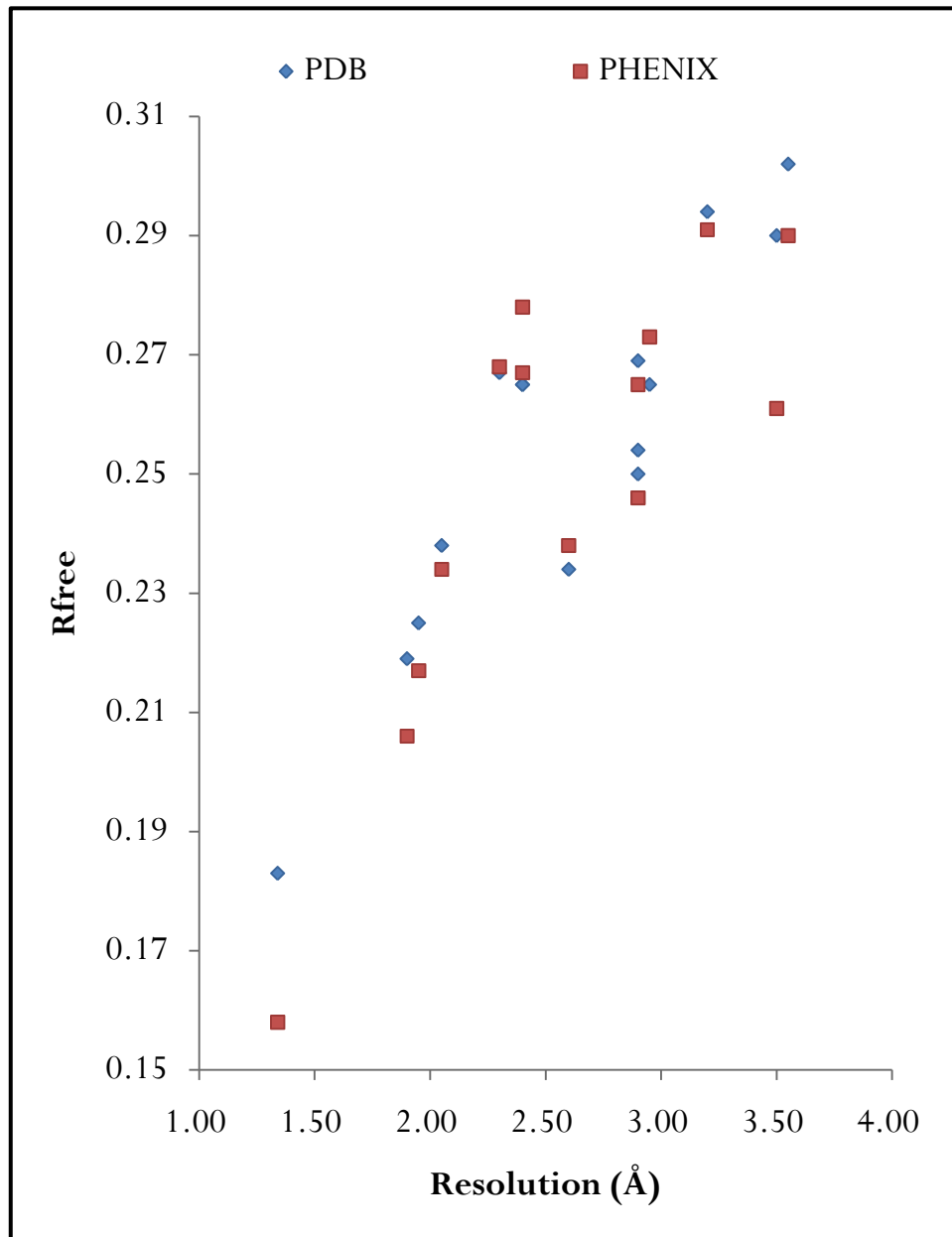
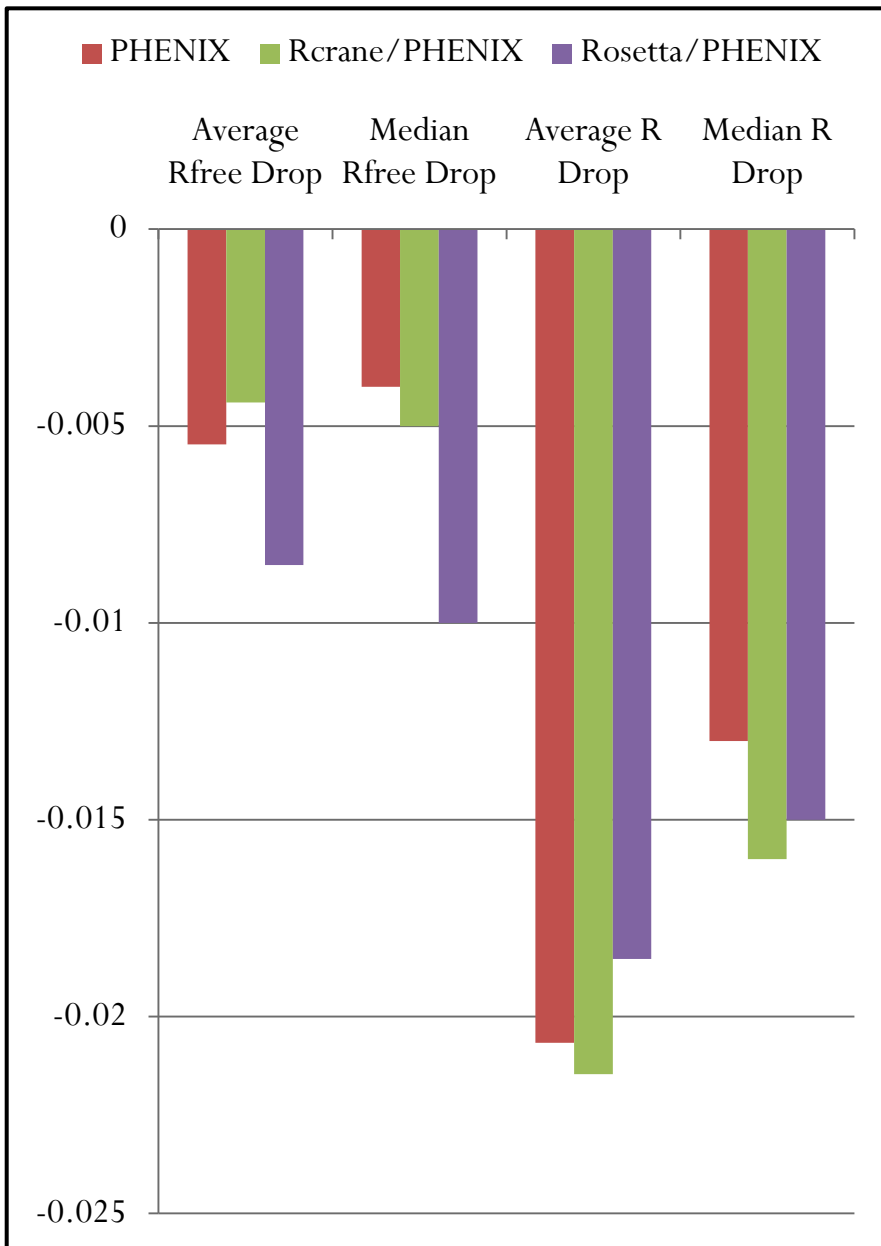
13 out 15 cases in Rosetta/PHENIX have equal or better R_{free} than in PHENIX



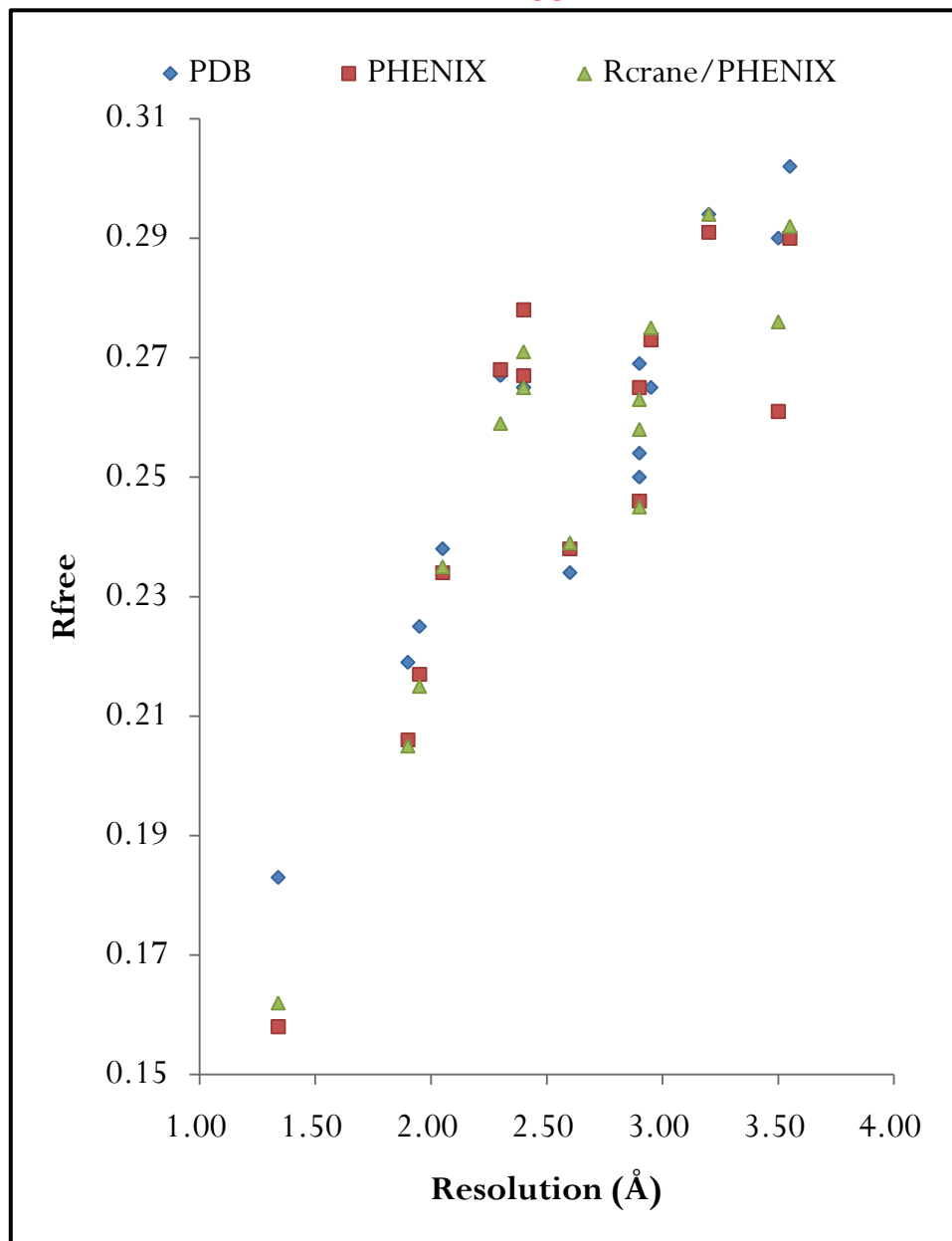
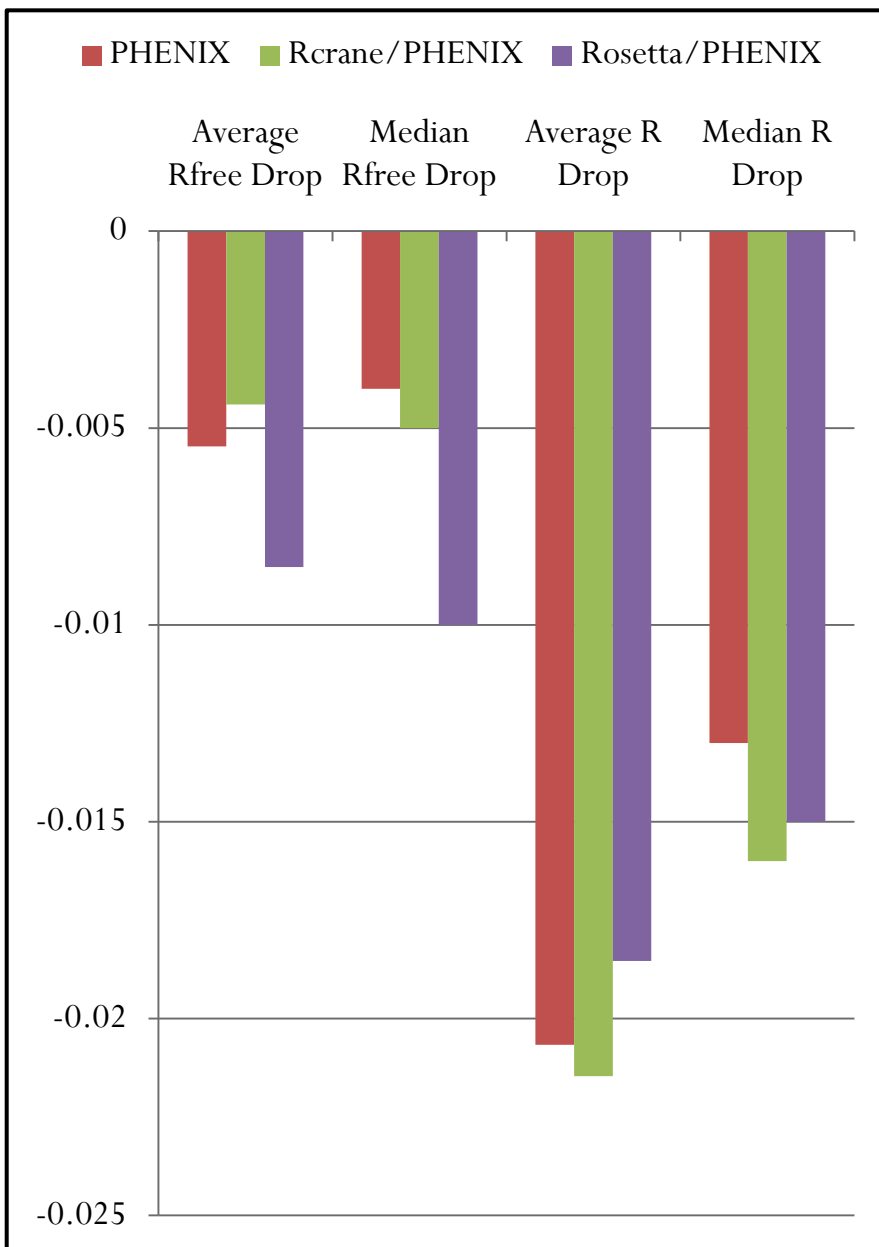
13 out 15 cases in Rosetta/PHENIX have equal or better R_{free} than in PHENIX



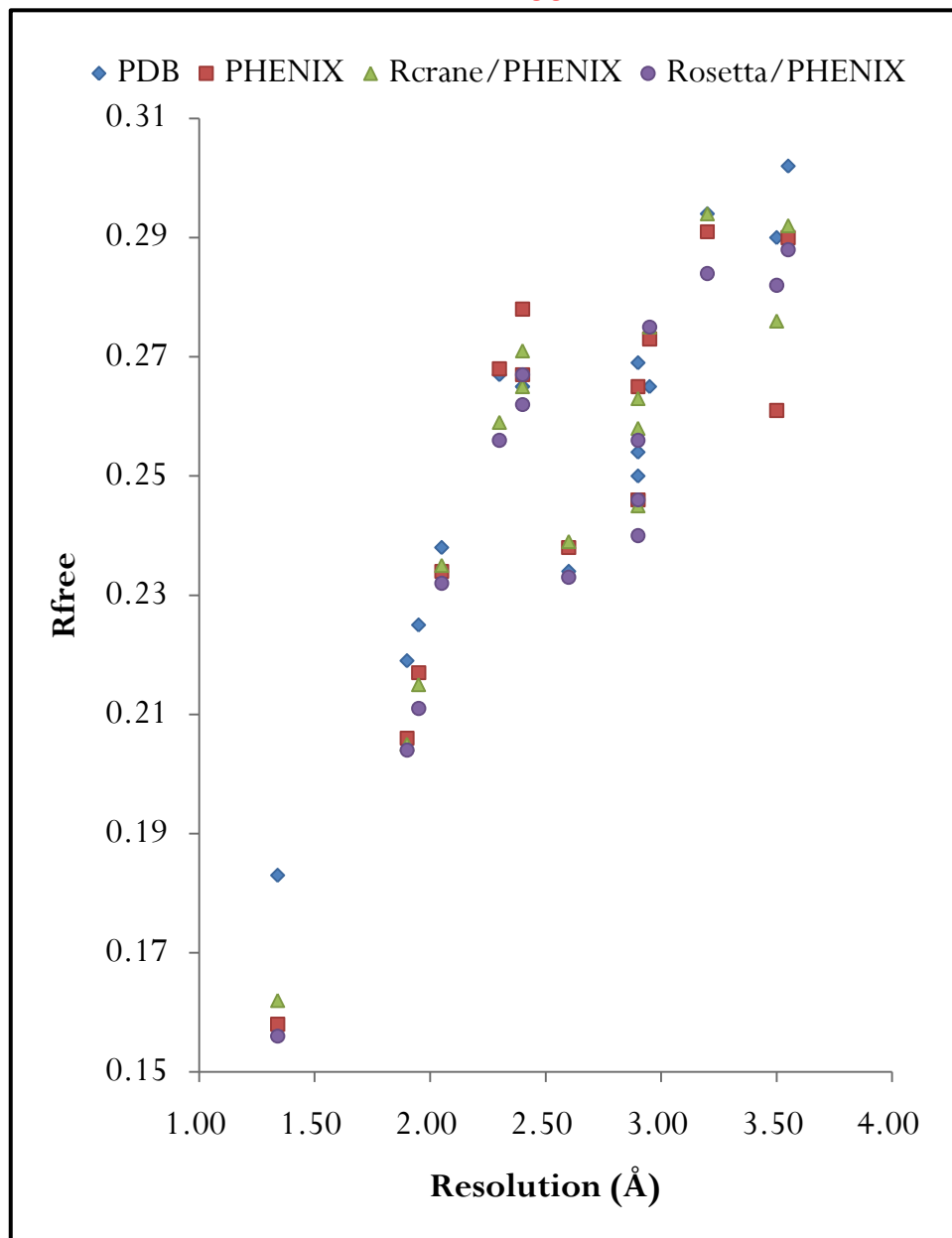
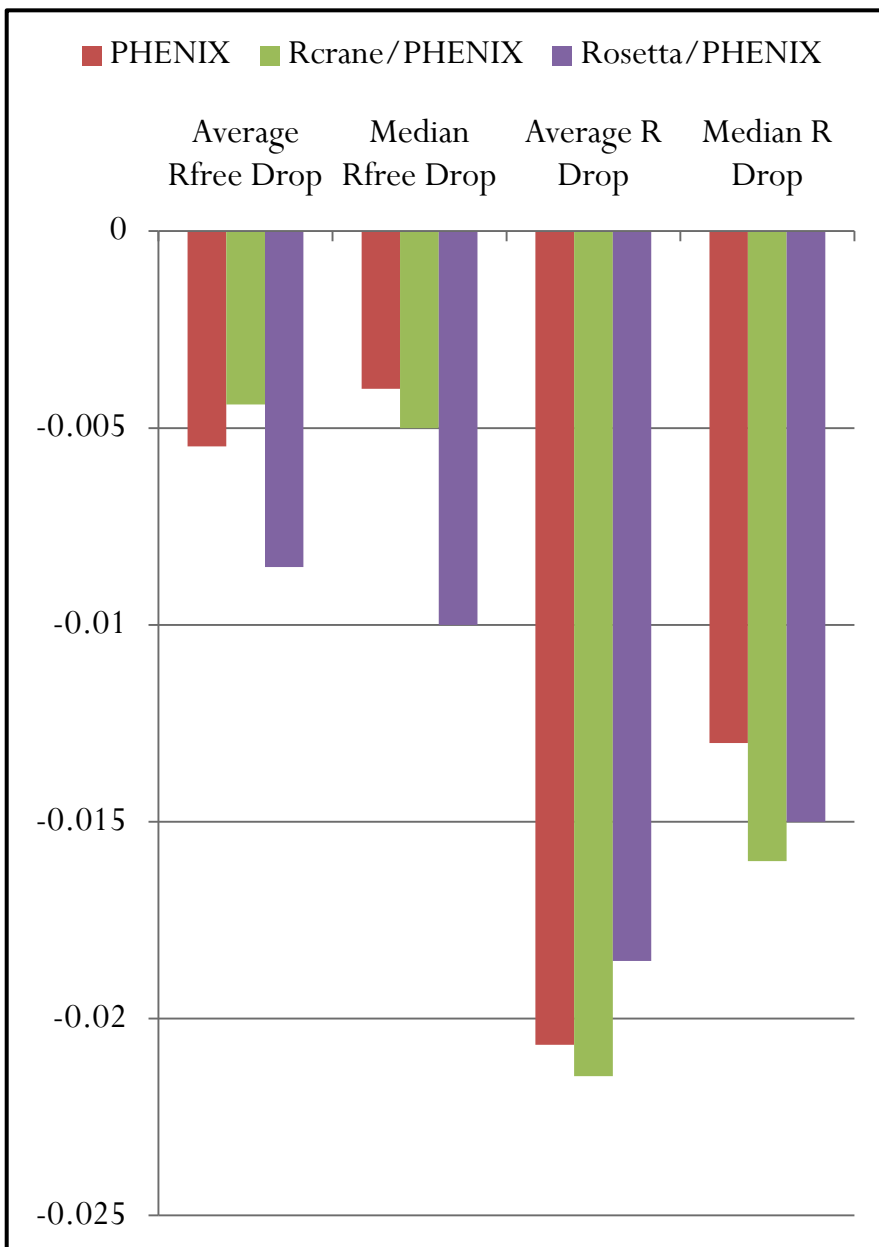
13 out 15 cases in Rosetta/PHENIX have equal or better R_{free} than in PHENIX



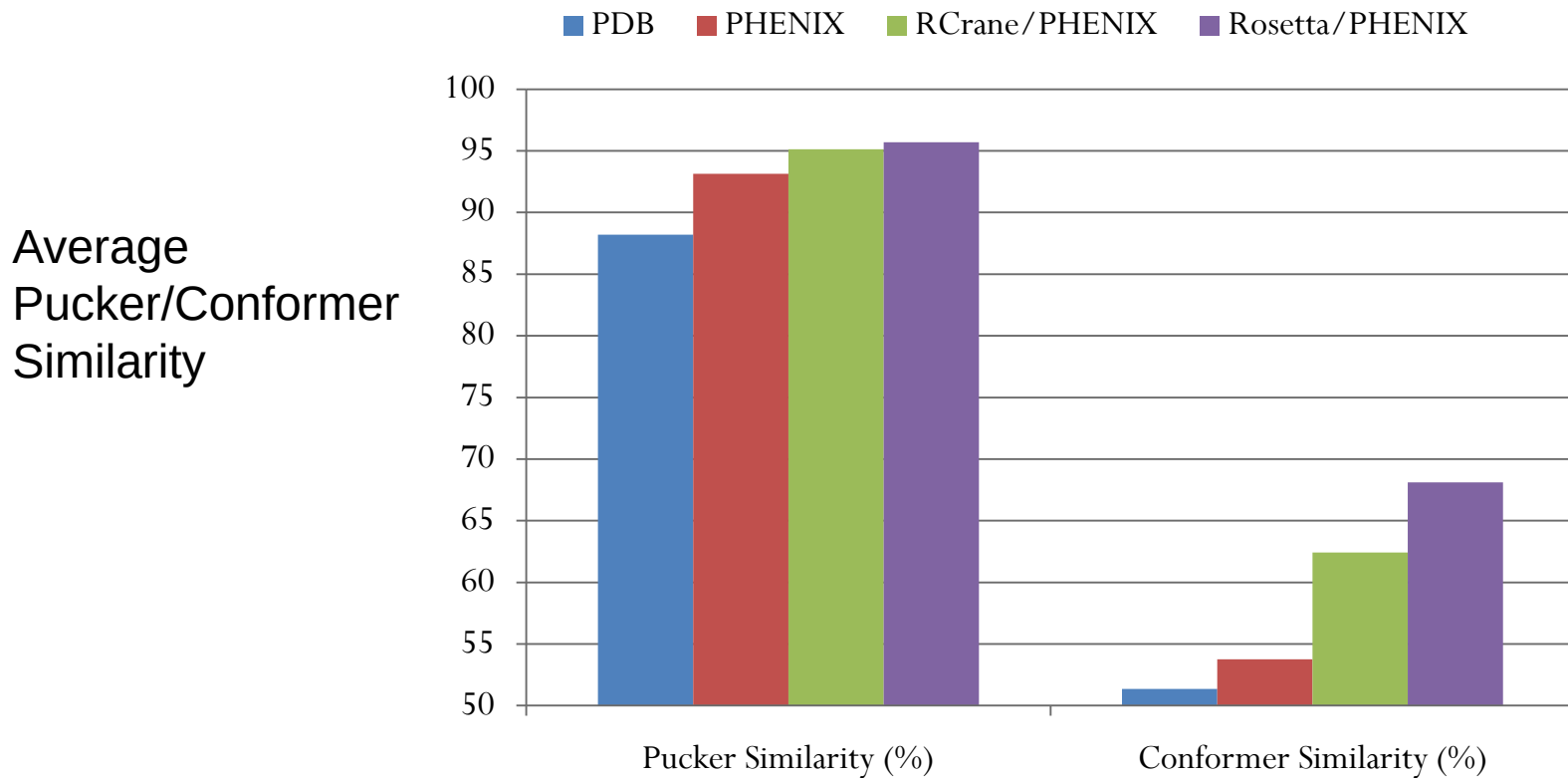
13 out of 15 cases in Rosetta/PHENIX have equal or better R_{free} than in PHENIX



13 out 15 cases in Rosetta/PHENIX have equal or better R_{free} than in PHENIX



We compared the similarity of sugar puckers and backbone conformers for the two chains of PDB with two copies in the structure and two PDBs of the same sequence by different groups. The two aptamer stems of glycine riboswitch (3P49) are also compared.



Summary

- Rosetta/PHENIX improves RNA crystallographic models.
 - Consistent reduction of clashes and bad geometry (Molprobity)
 - Lower or maintain R_{free} in 13 out of 15 cases
- Reasonable computational cost.
 - Less than one day to fix a ~ 170 residues RNA in a single core PC.
- We plan to add it into the next release of Rosetta and also set up a server to host this code after we publish it.

Acknowledgement

- Rhiju Das, my advisor.
- Parin Sripakdeevong, for providing the SWA code and helping me with all the Rosetta questions.
- All other members in the Das lab for their advices.
- Bio-X² cluster at Stanford for the computational resource.
- Rosetta community for making the code publicly available.

Thank you for your attention!