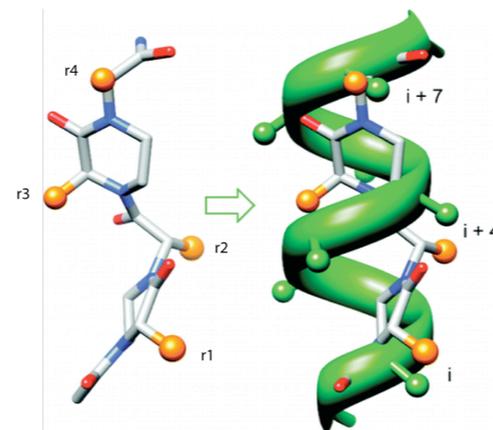# Rational design of protein inhibitors using Oligooxopiperazines (OOPs)

RosettaCon 2012
Kevin Drew
Tuesday, July 31, 2012

Tosovska, P., 2010

Outline

- ## Motivation and Background

  - helical mimetics
  - MDM2 - P53 protein interface
  - OOPs

- ## Inhibitor Design

  - Design Protocol
  - Designs
  - Binding Mode
  - Scaffold Library

Outline

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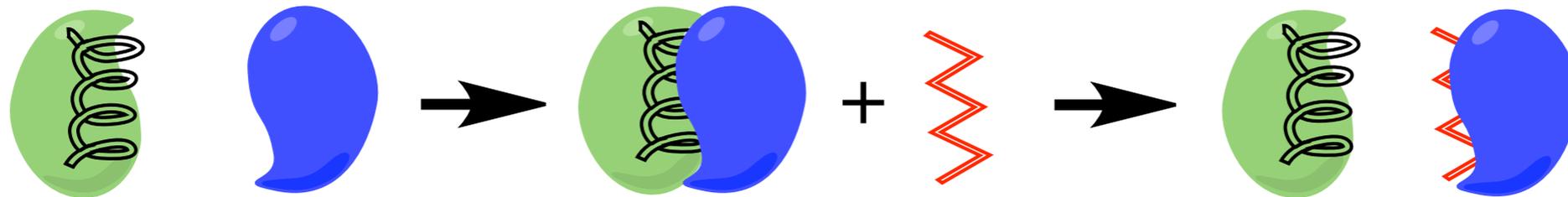 - MDM2 - P53 protein interface
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Inhibitor Design - Goal

Rationally design a molecule to disrupt a specific protein interaction

- screening is expensive and laborious

- small molecules do not span full interaction interface

- use natural interface as starting point
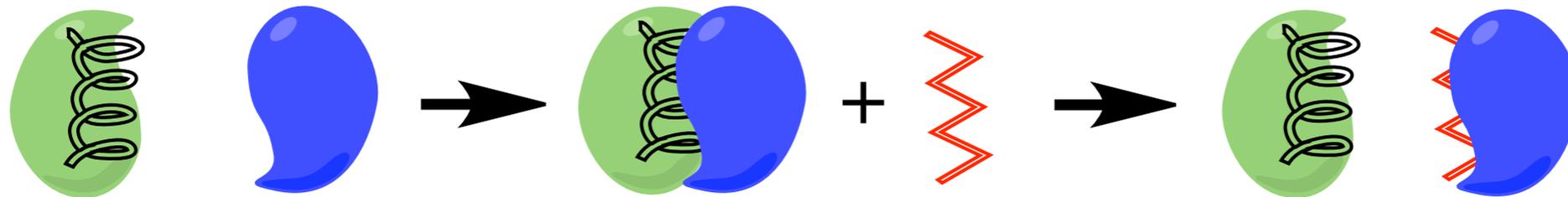
Inhibitor Design - Goal

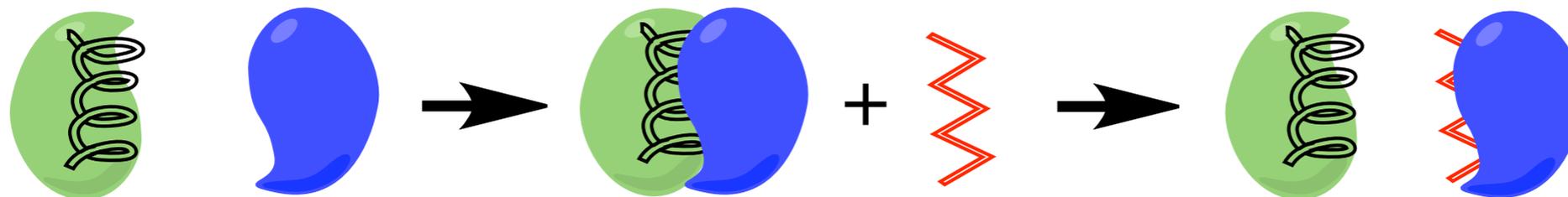Rationally design a molecule to disrupt a specific protein interaction

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  Wadman Nature Biotechnology (2012)

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- **- use natural interface as starting point**

  • mimic hotspot residues on a stable proteolytic resistant scaffold
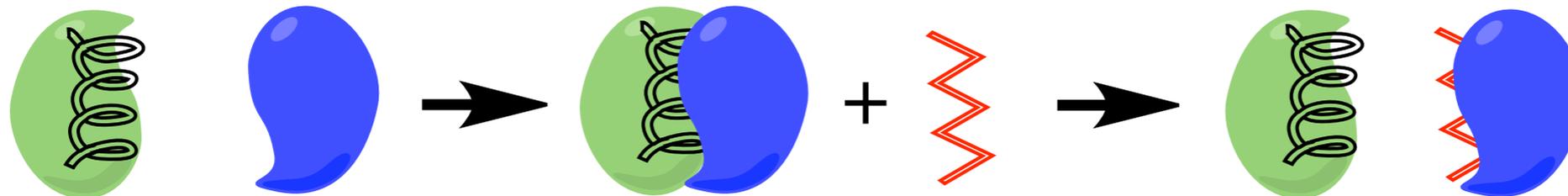
Inhibitor Design - Background
Helical memetic successes

Hydrogen bond surrogate -

inhibits Hypoxia Inducible Factor 1 / coactivator interaction
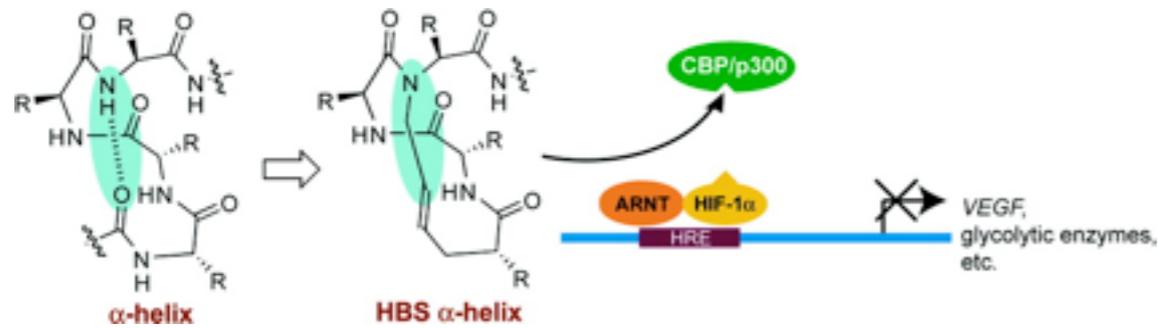
Henchey et al JACS 2010

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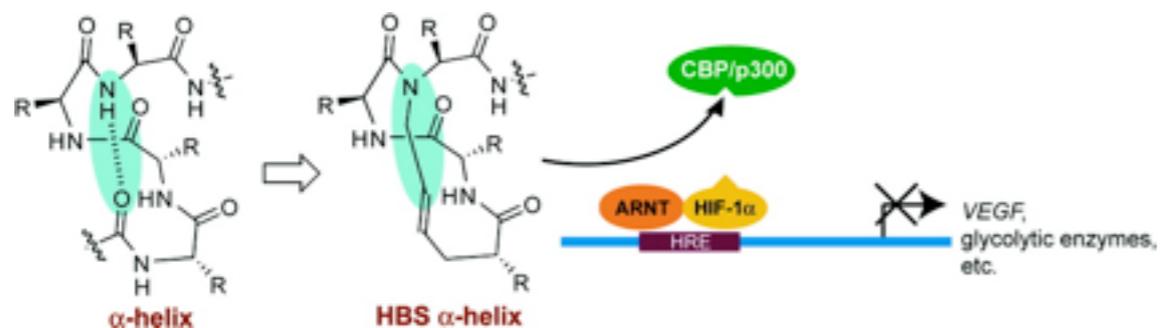

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Alpha-beta peptides -
binds bcl2 family (anti-apoptotic protein)
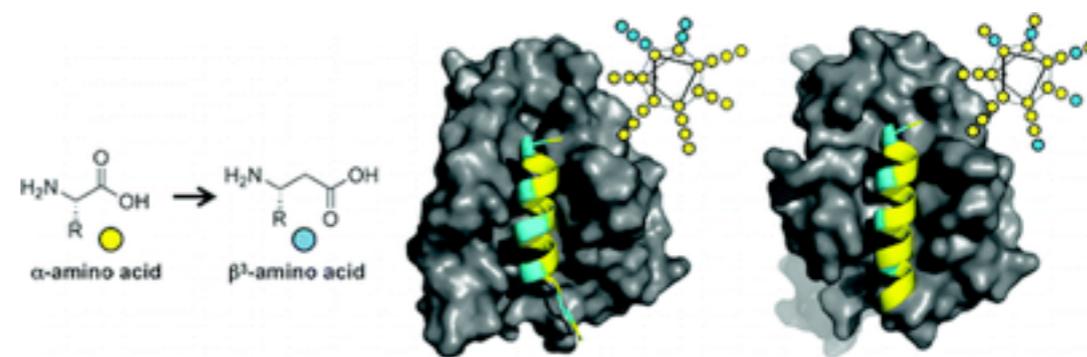
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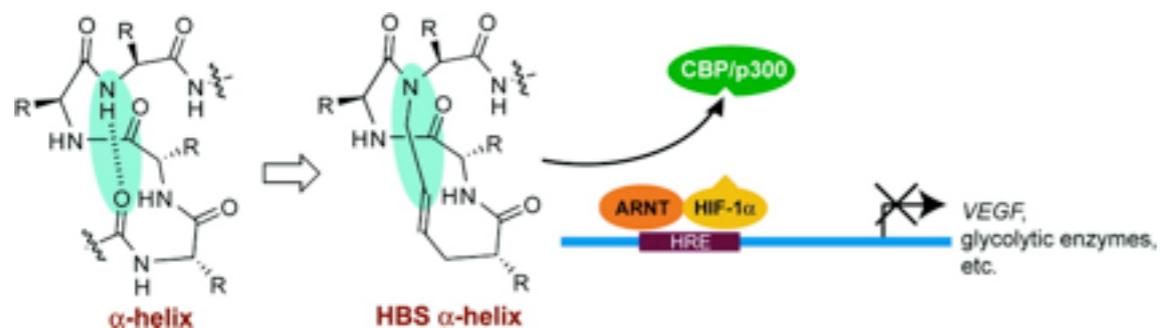
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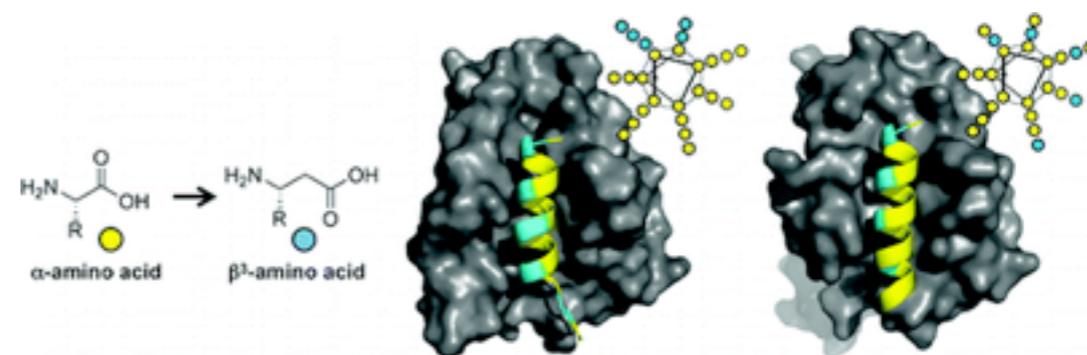
Boersma et al JACS 2011

Terphenyl -
disruption of gp41 oligomerization
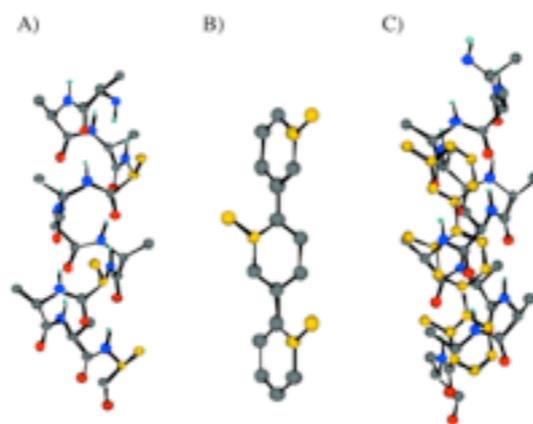
Ernst et al ACIE 2002

Inhibitor Design - Model System

P53 - MDM2 Protein Interaction
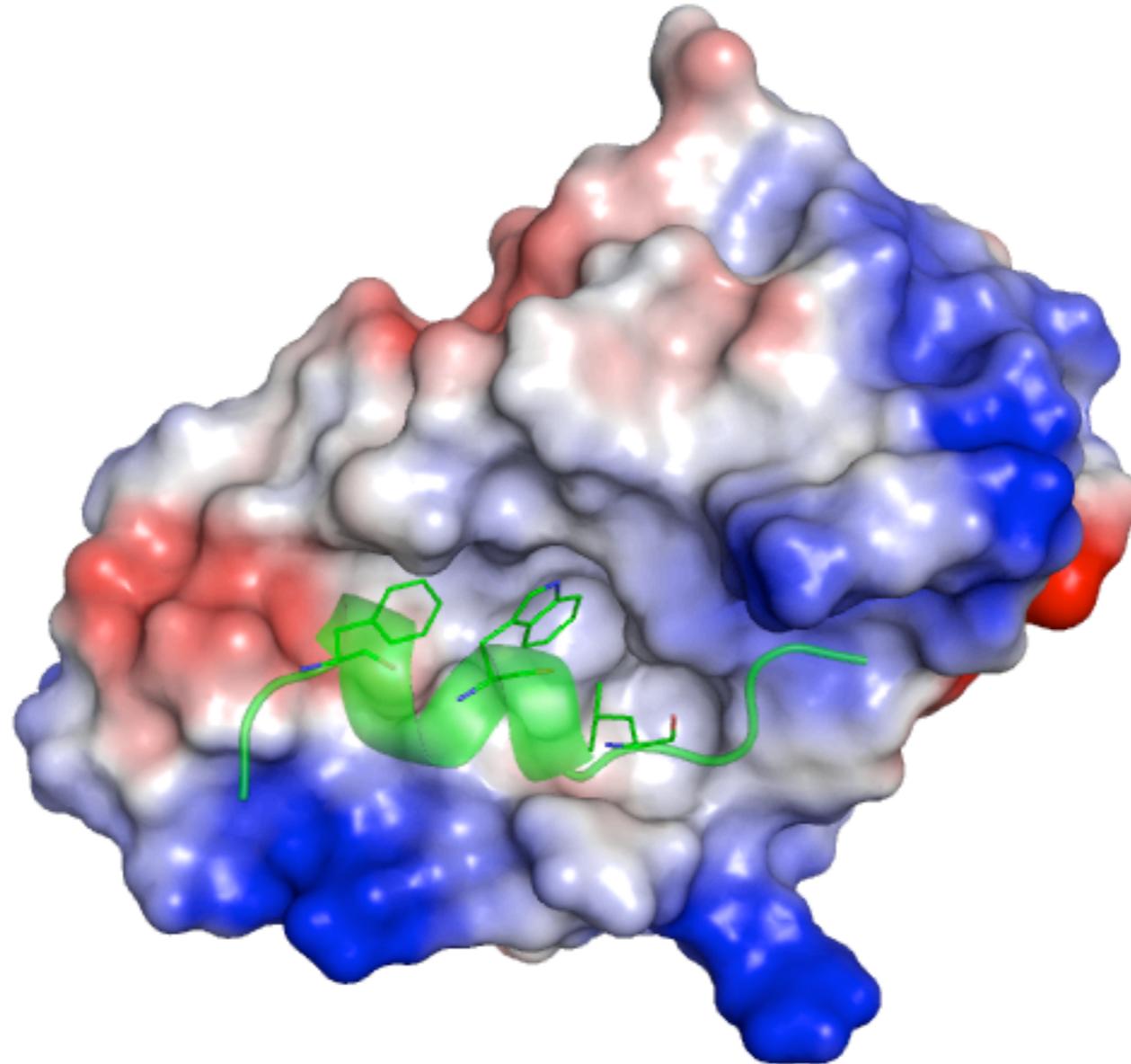
P53 transactivating domain (green) bound to MDM2 (electrostatic)
pdbid: 1YCR. (Kussie et al. Science 1996)

Side chains important for binding shown in lines.

Inhibitor Design - Model System

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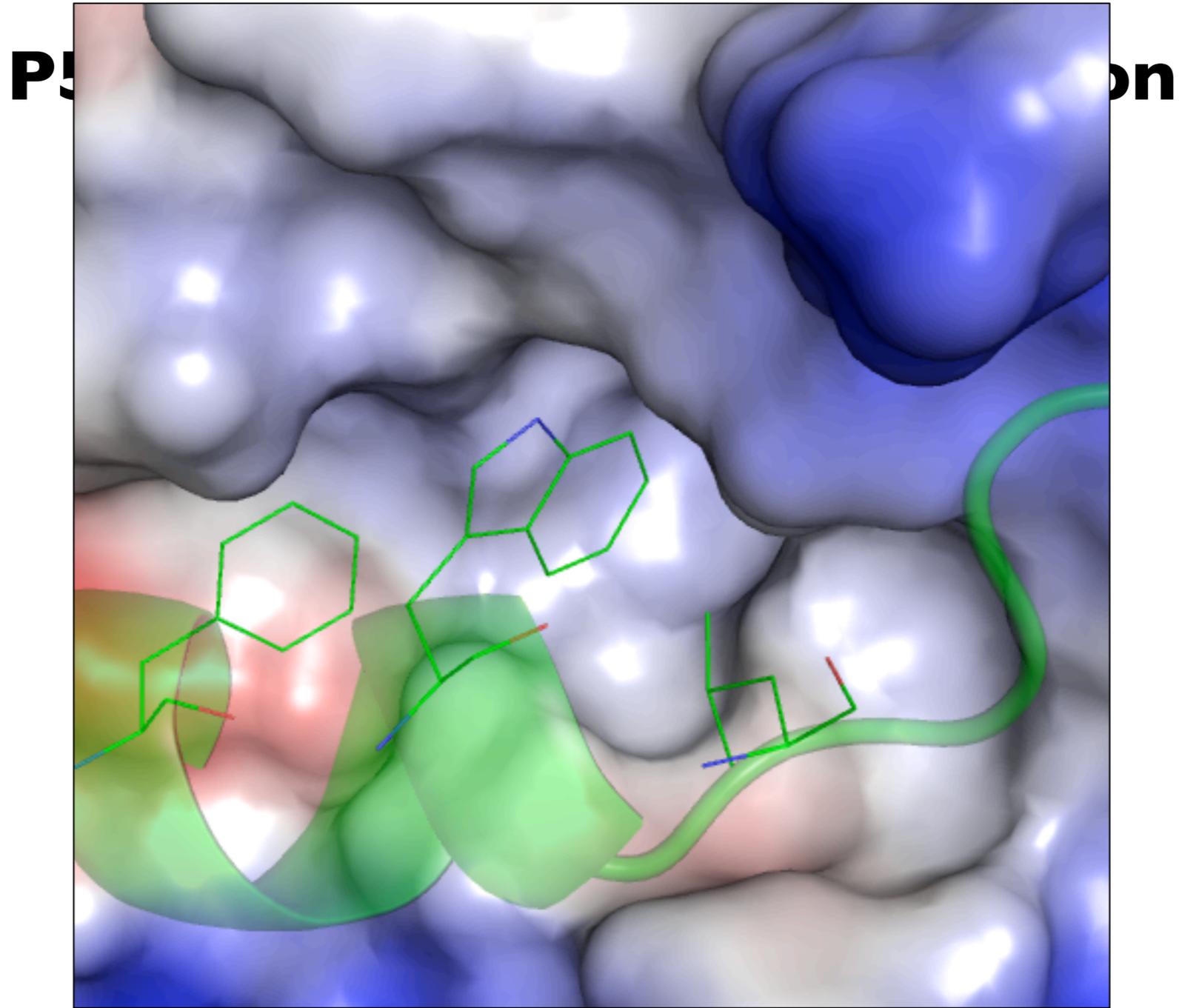
Inhibitor Design - Model System

P53 transactivating domain (green) bound to MDM2 (electrostatic)
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Side chains important for binding shown in lines.

Nutlins - small molecules known to disrupt interaction, ~140nM IC_{50}
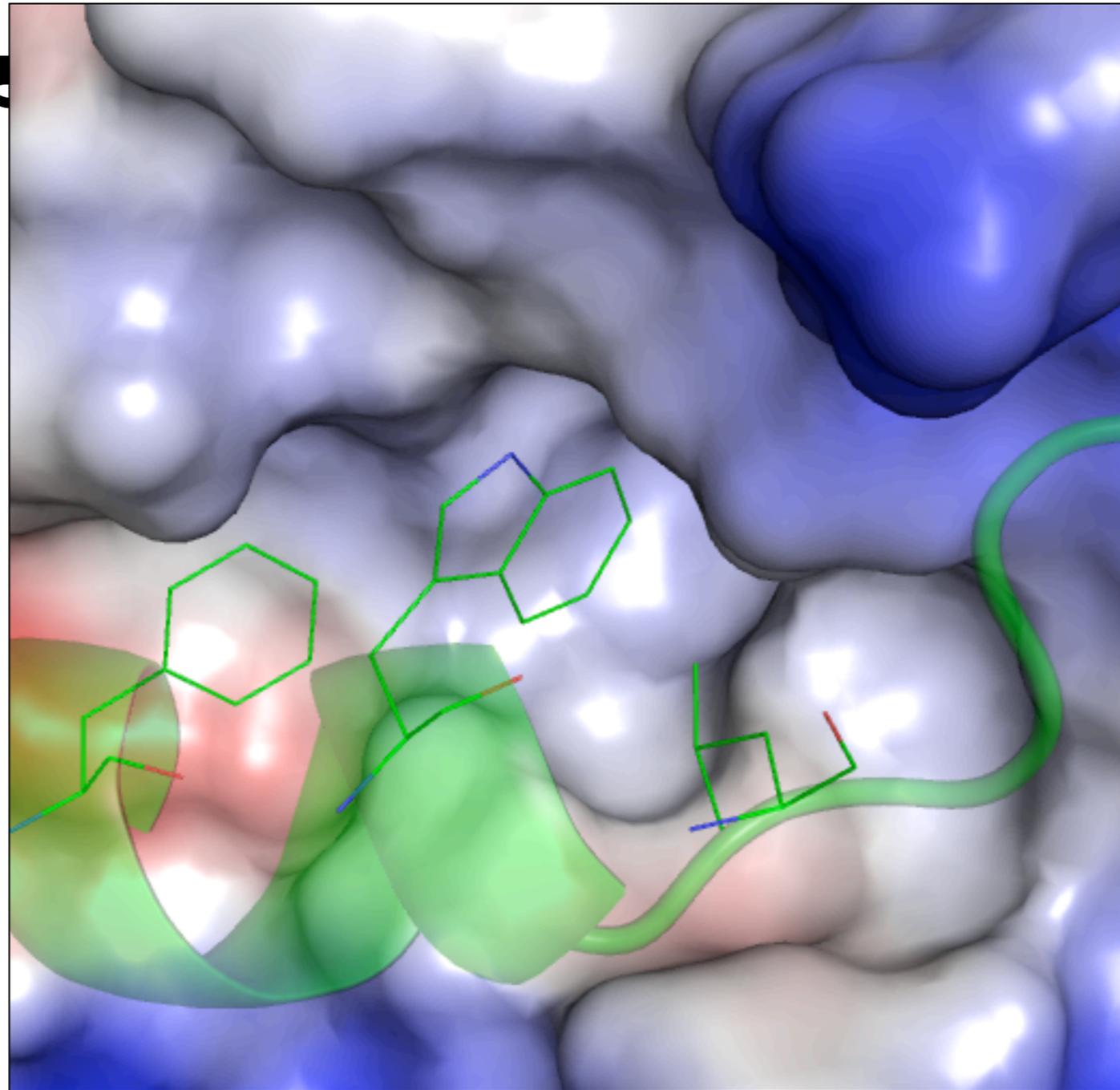
Inhibitor Design - Scaffold

Oligooxopiperazines (OOPs)
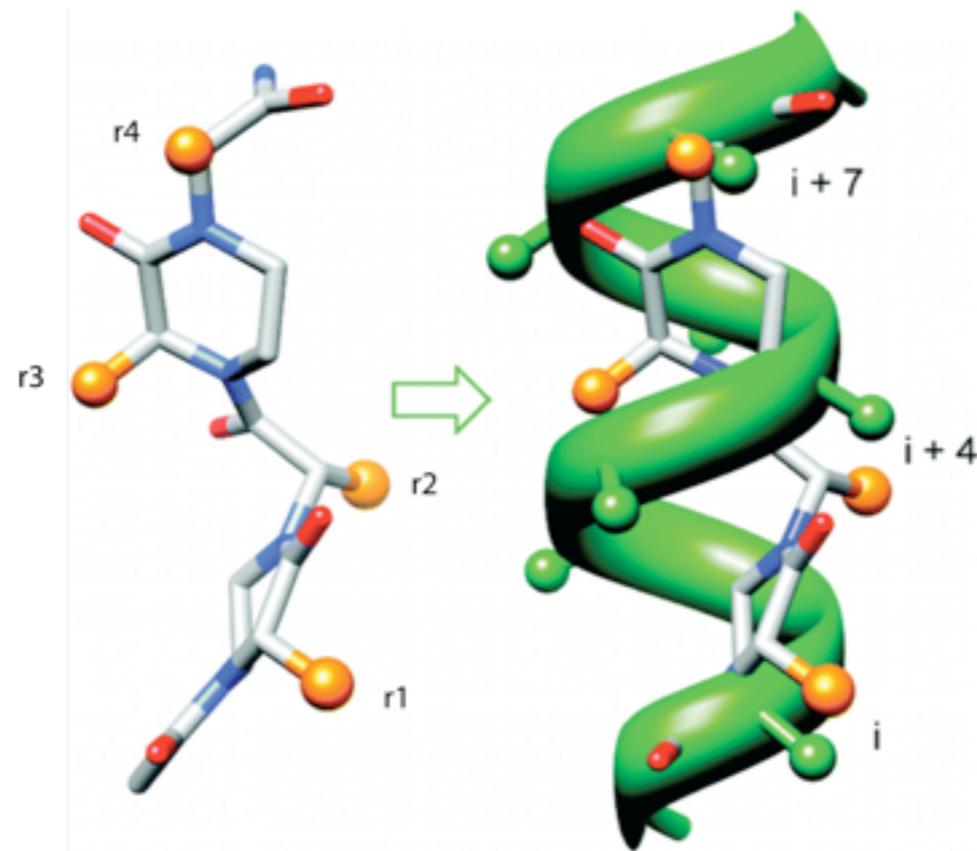
Tosovska, P. 2010

Mimics i, i+4 and i+7 residues of helix

Peptide backbone with C-C bond

Easy to synthesize (solid phase)
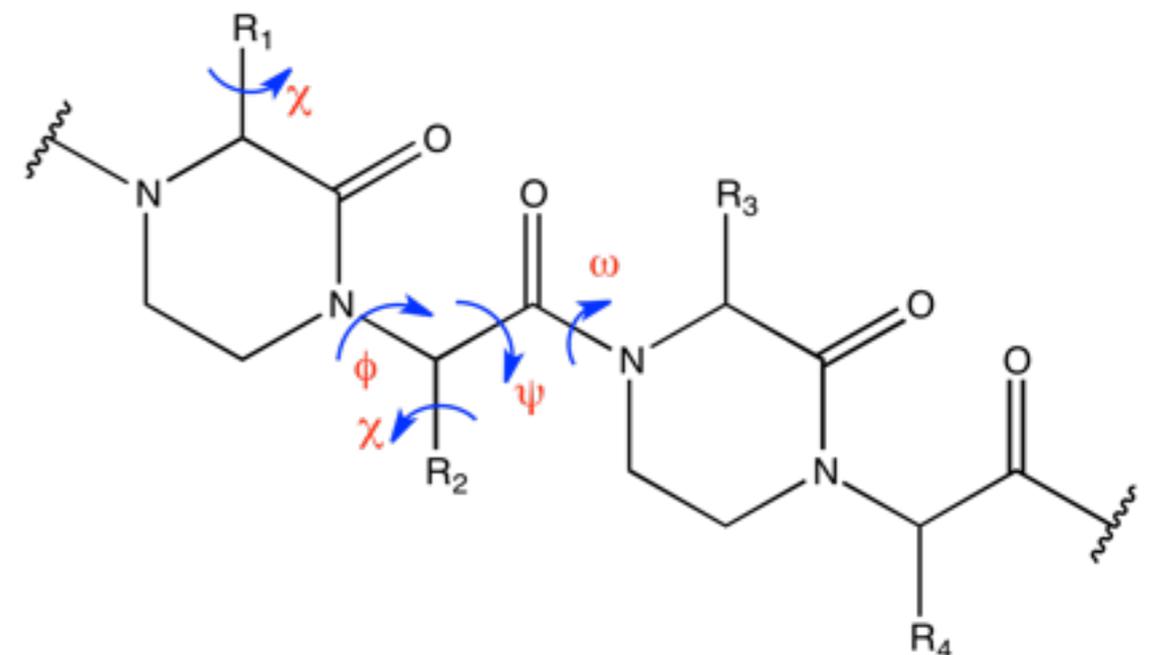
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OOP quantum characterization

How does Rosetta energy function compare?

OOP quantum characterization

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Quantum Calculations

Approach - Gaussian QM Software

Rotate each bond 360° (increment 15°)

Hartree-Fock optimization

B3LYP 6-31G(d) energy calculation

MP2 6-31G(d) energy calculation

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Ramachandran Map

Ψ

ϕ

JS Richardson 1981

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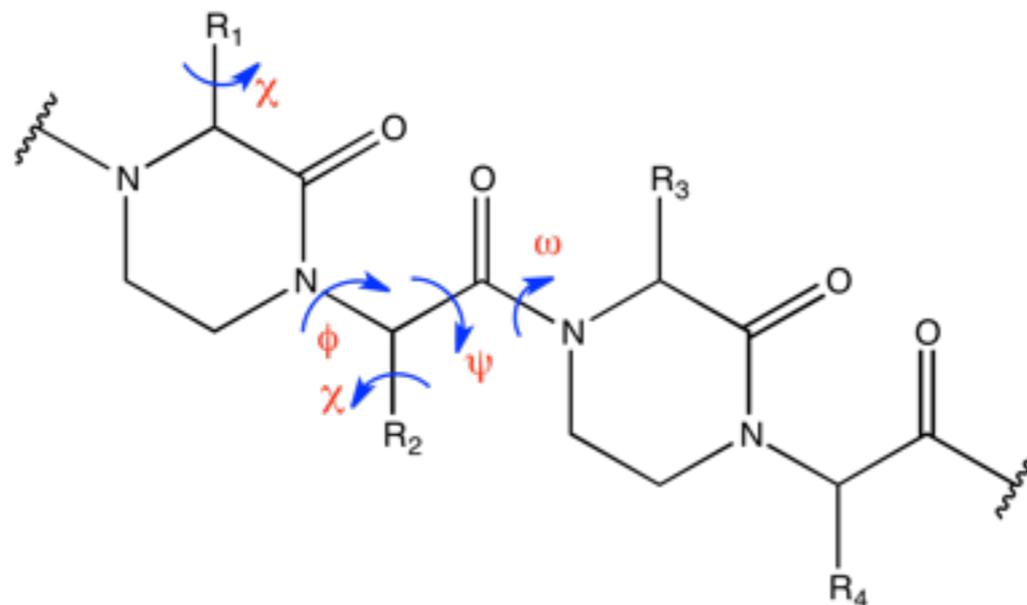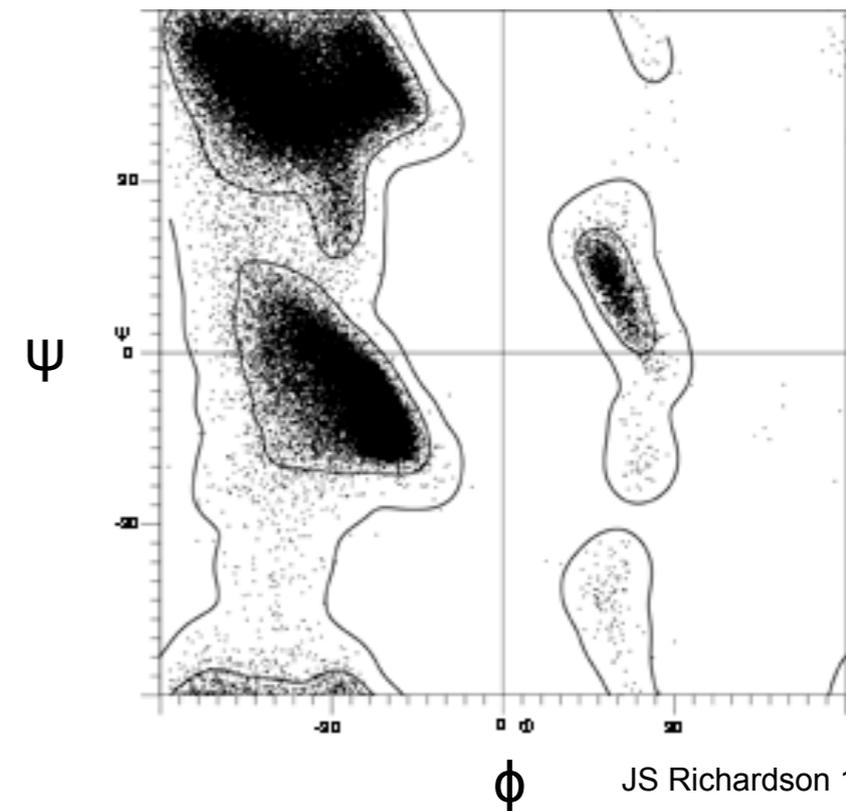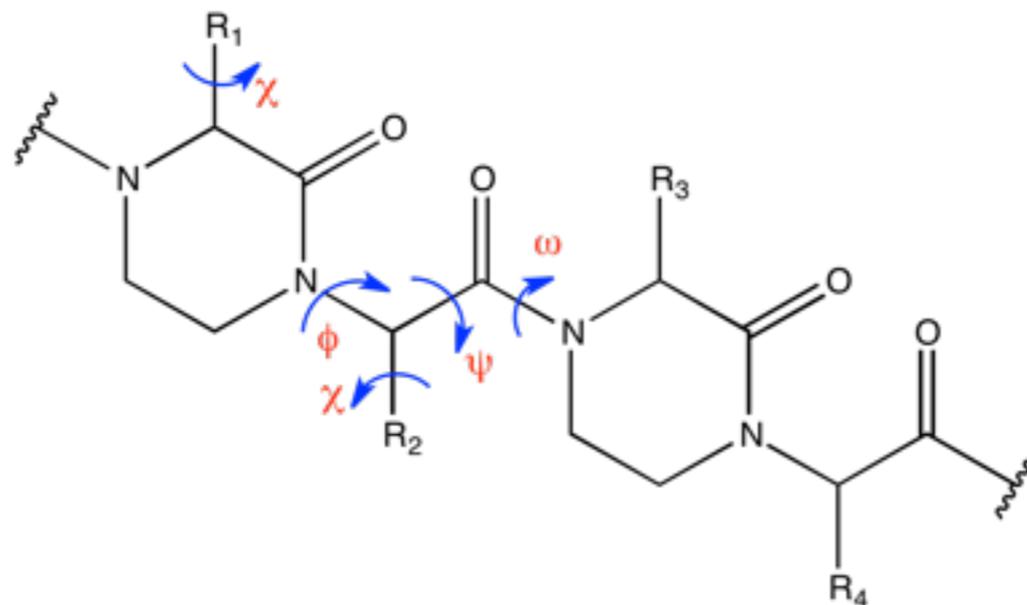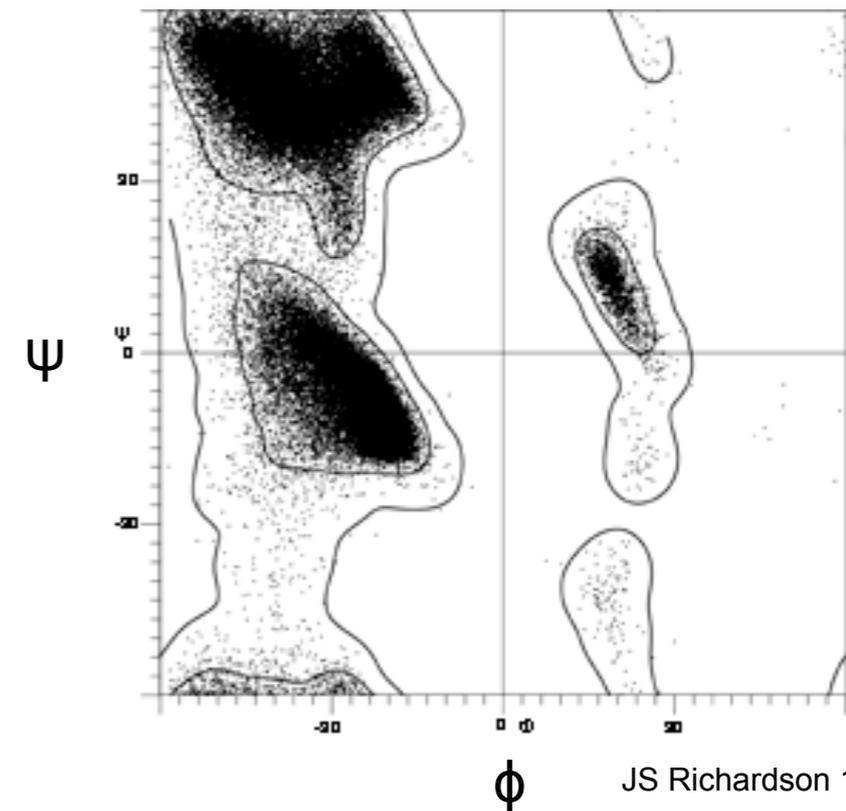
OOP characterization

Quantum vs Rosetta: Phi/Psi energy comparison

OOP characterization
Quantum vs Rosetta: Phi/Psi energy comparison

Quantum
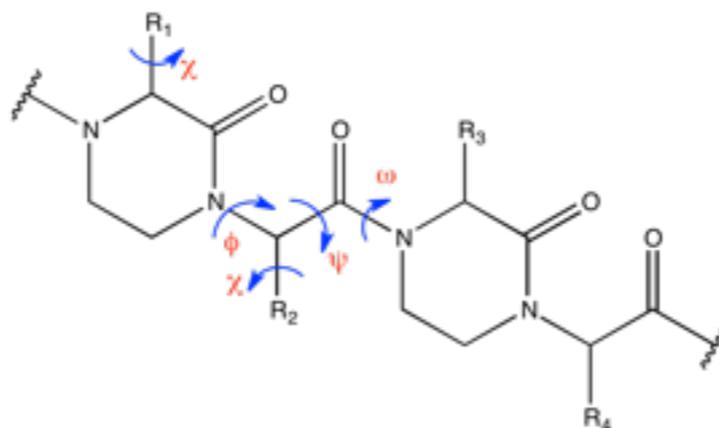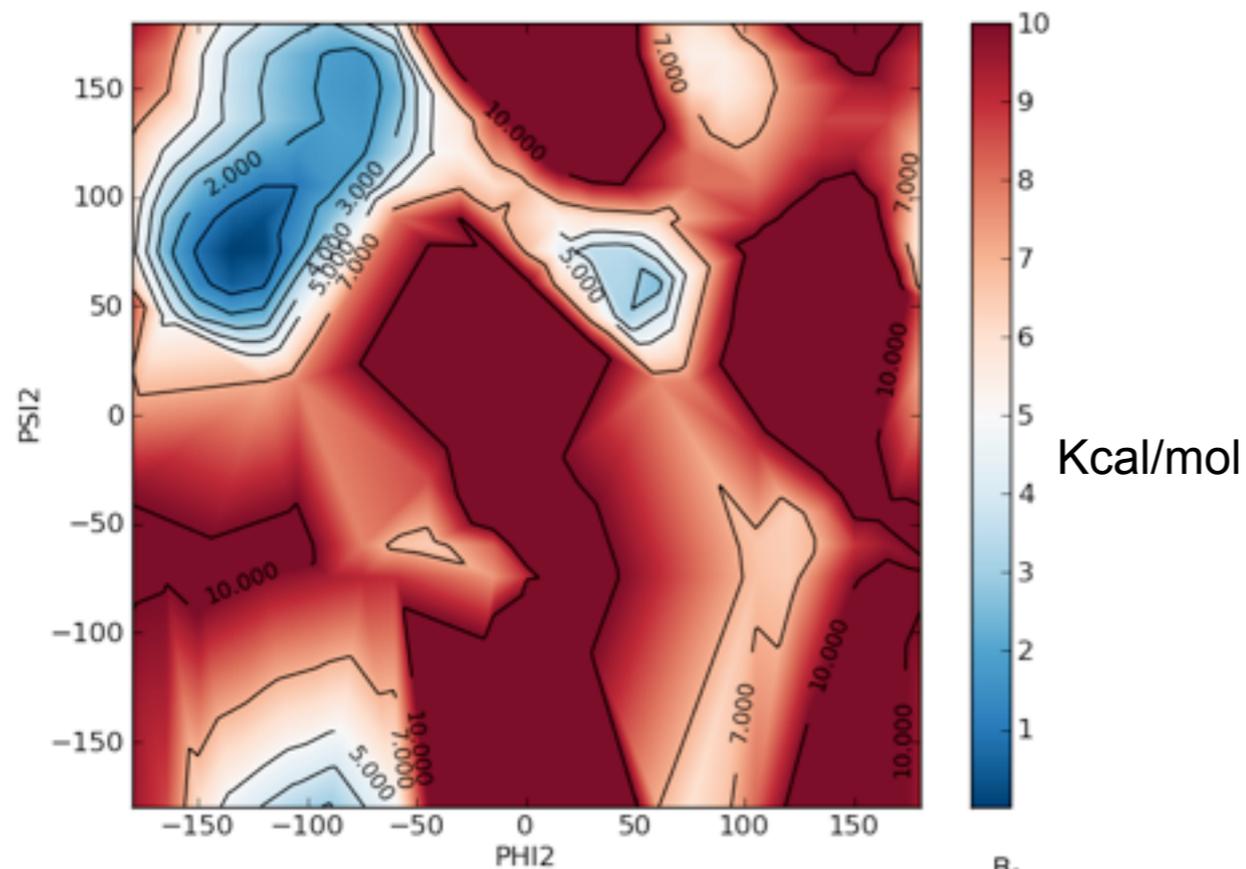
B3LYP SOLV 6-31G(d)

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Quantum
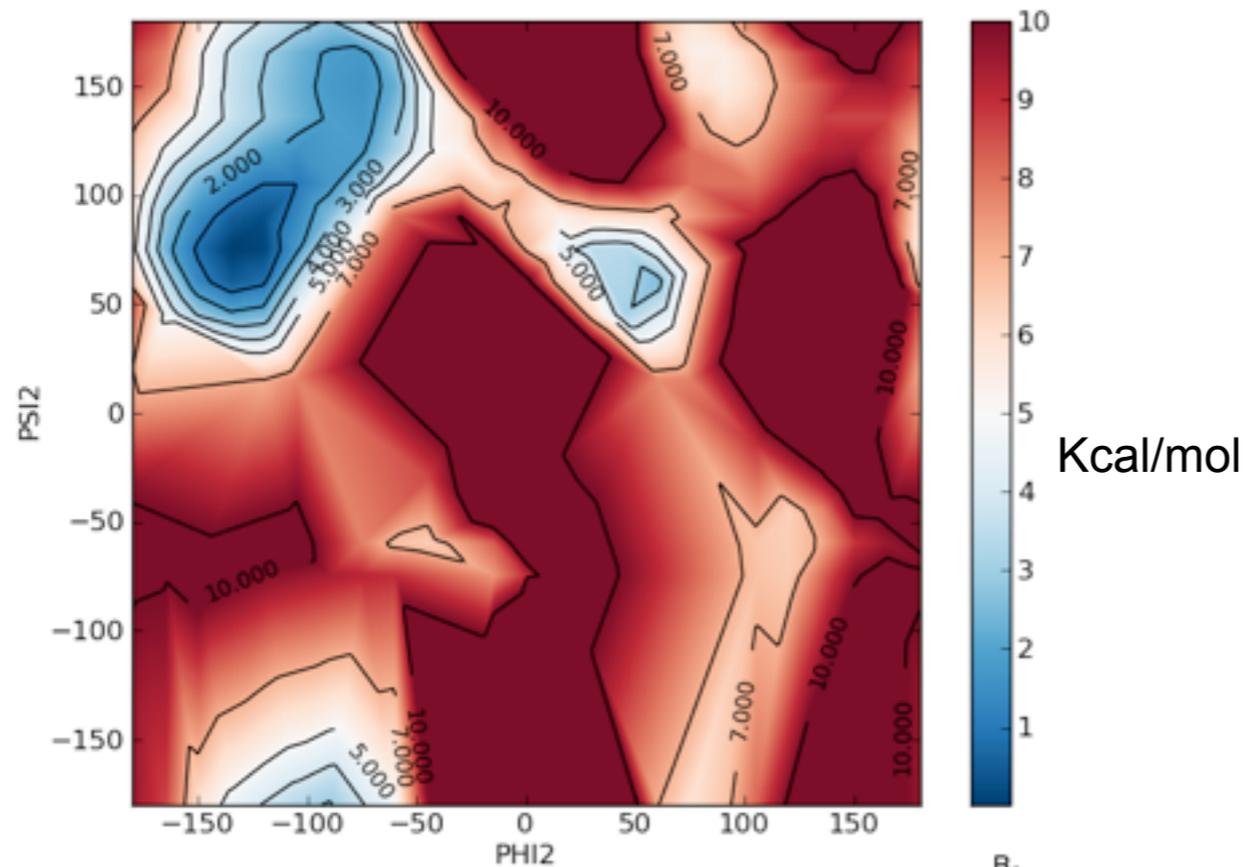
B3LYP SOLV 6-31G(d)

Rosetta

MM: Lennard-Jones potential, Lazaridius Karplus solvation, Hbond, reference energy

OOP characterization

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Design Movie

1st pos	2nd pos	4th pos
dimethyl-PHE	hydroxy-phenylglycine	dehydro-LEU
3methyl-PHE	phenyglycine	fluoro-LEU
4methyl-PHE		
naphthyl-alanine		

10

Design Movie

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10
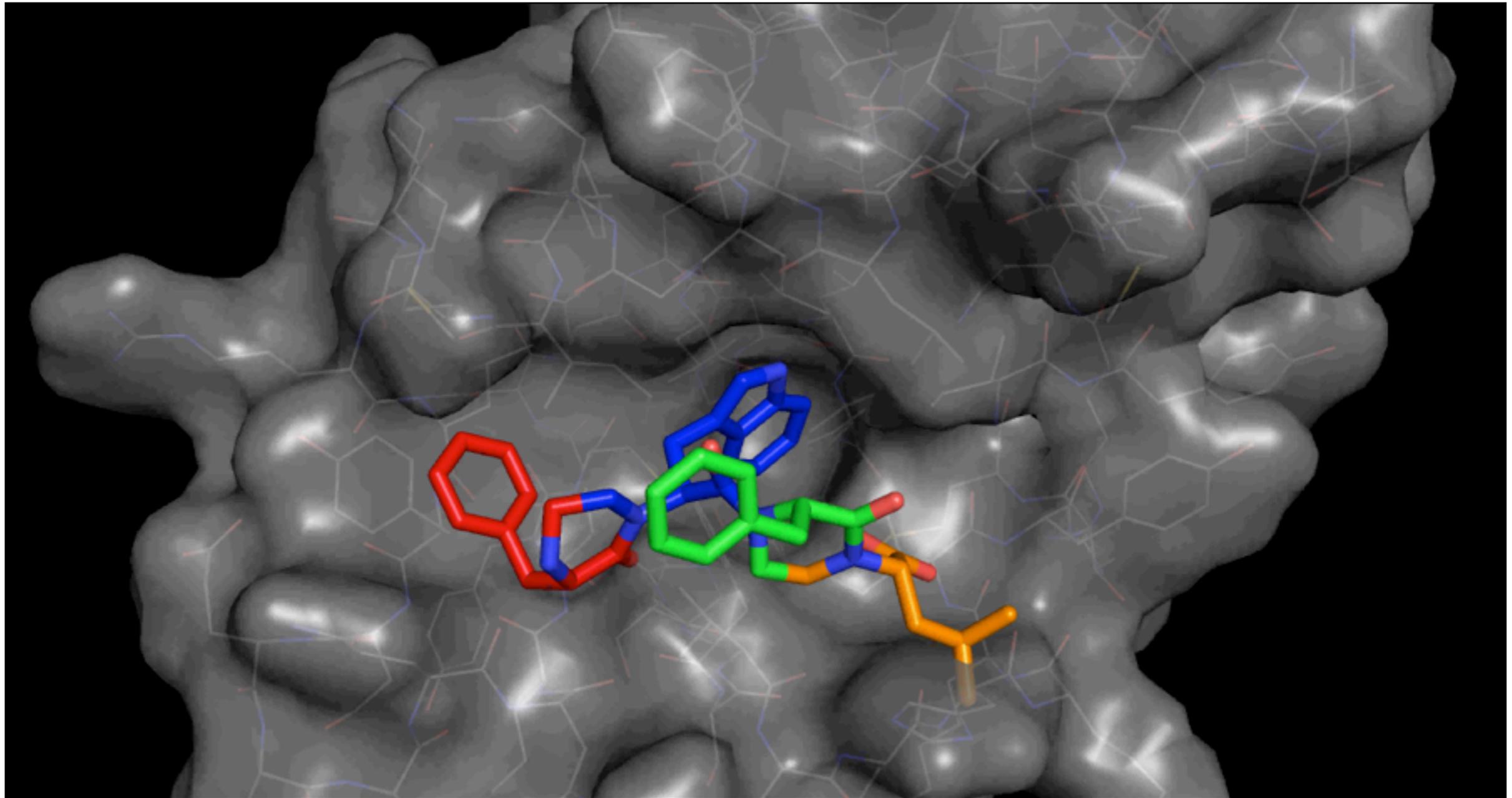
OOP Design

OOP Design

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FWFL
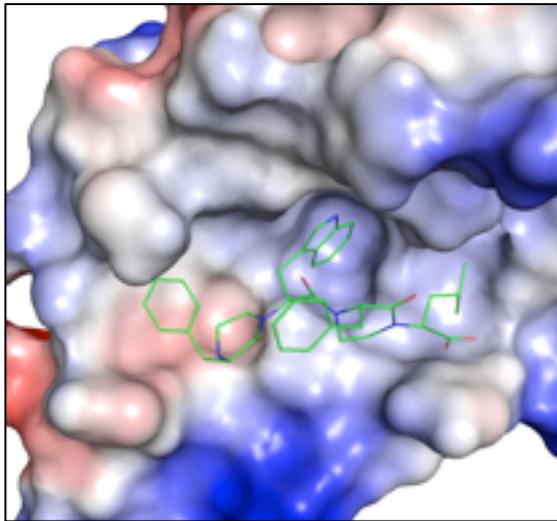
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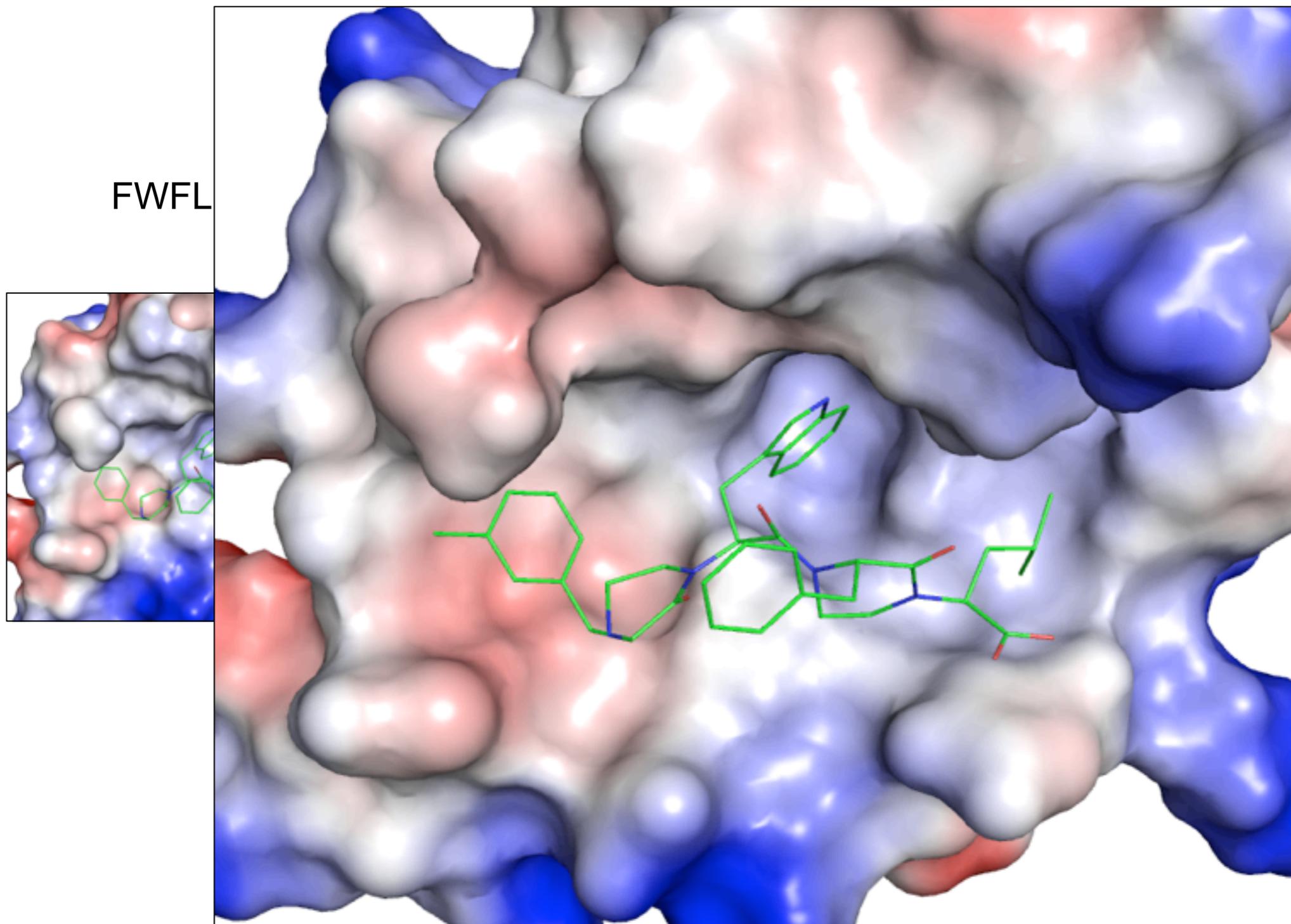
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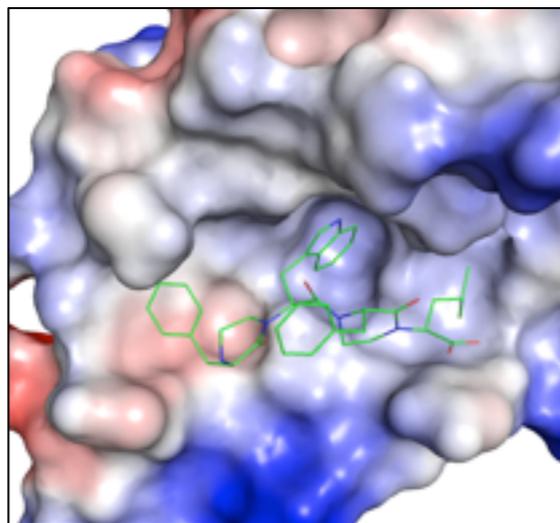
OOP Design

FWFL

[3-methyl-PHE] WFL
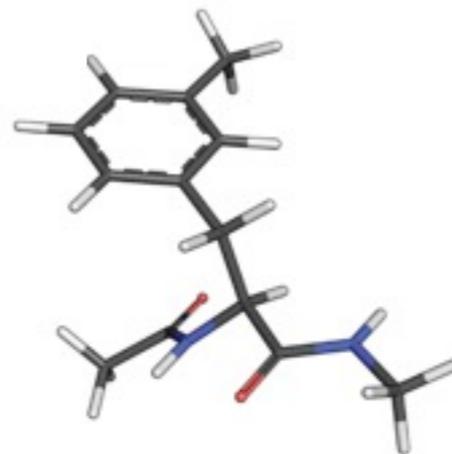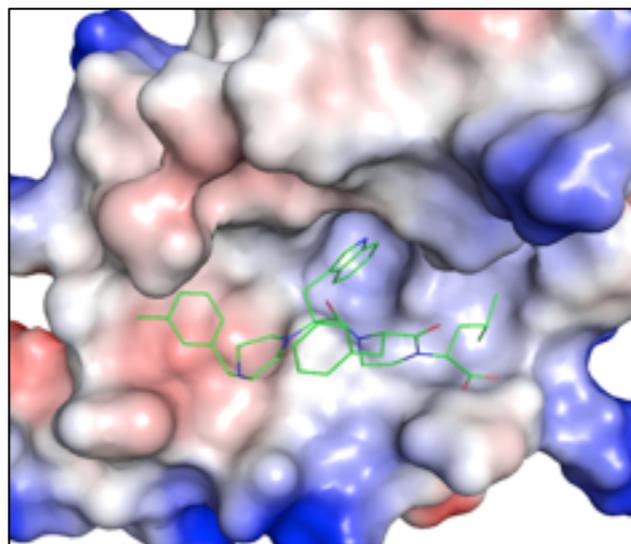
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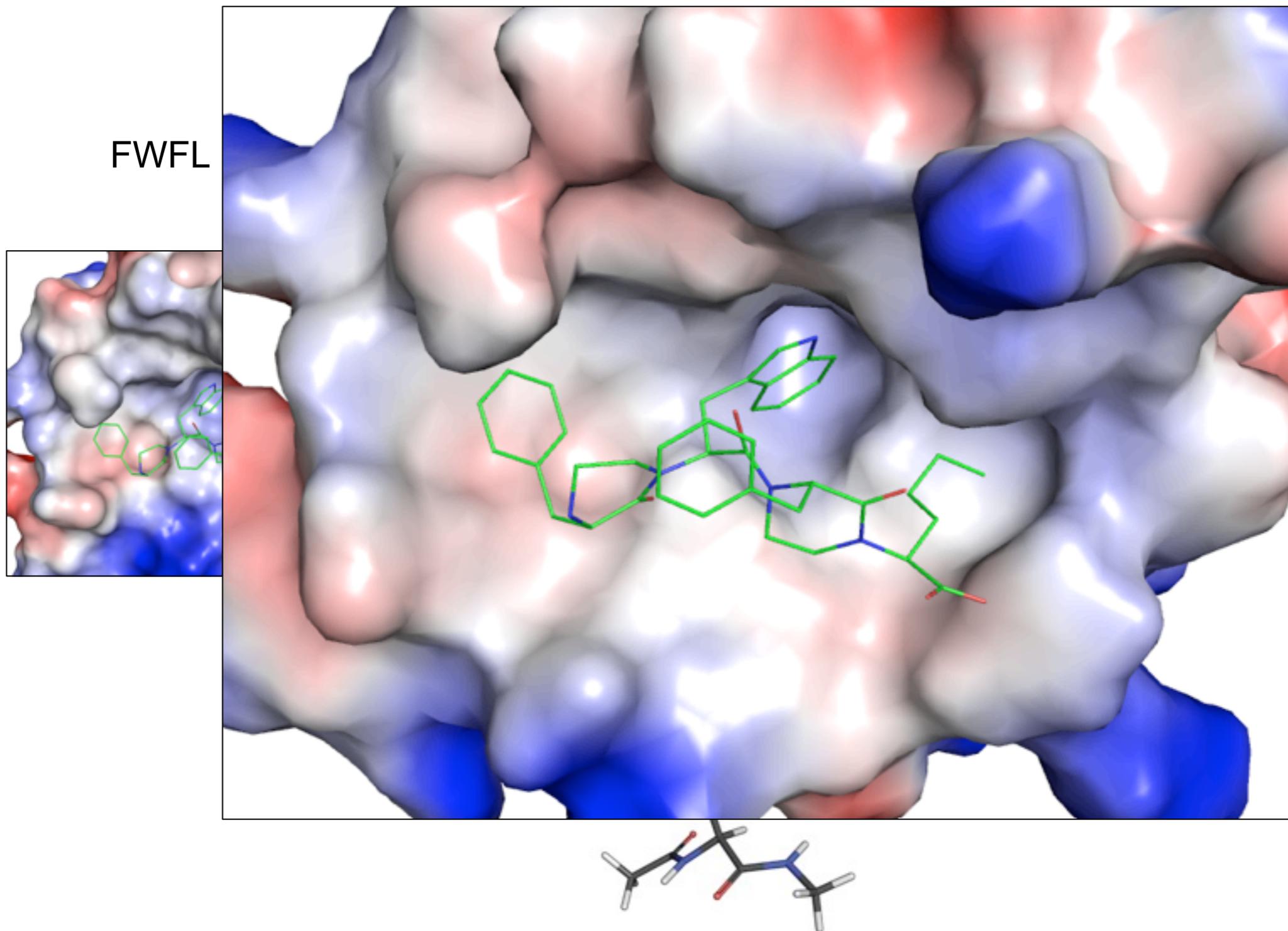
FWFL
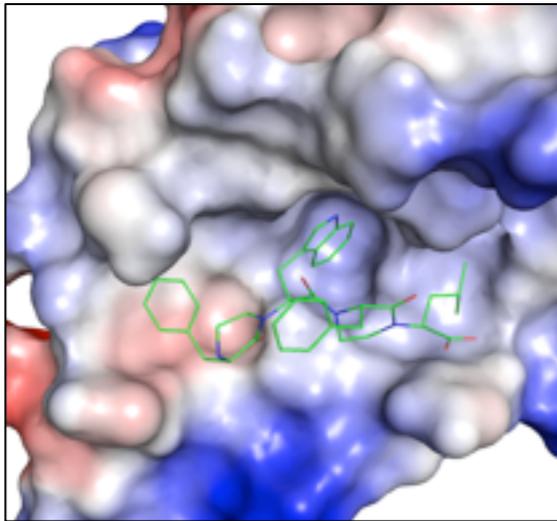
OOP Design

FWFL
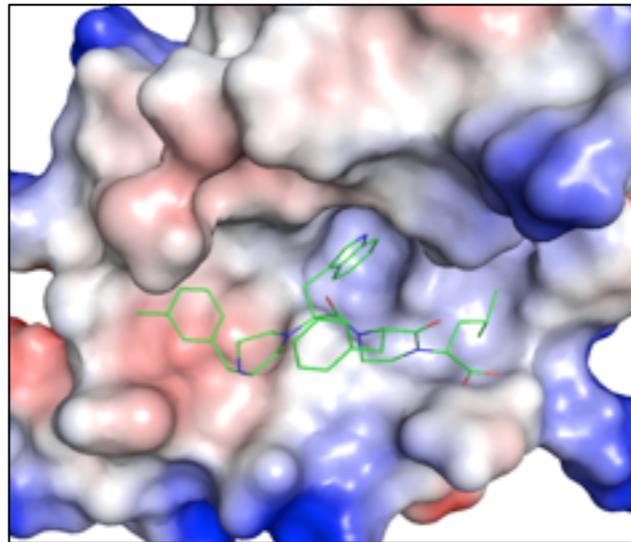
[3-methyl-PHE] WFL
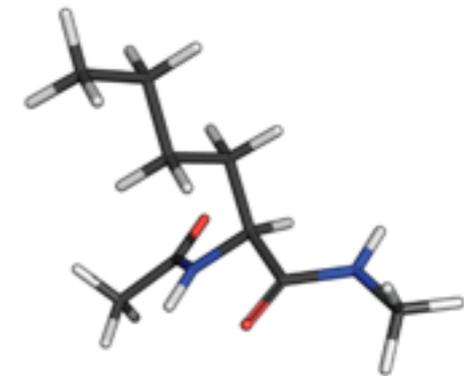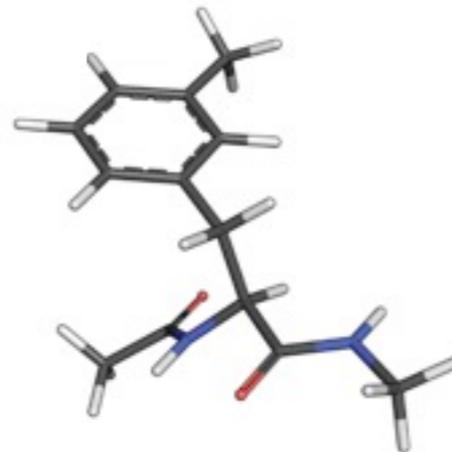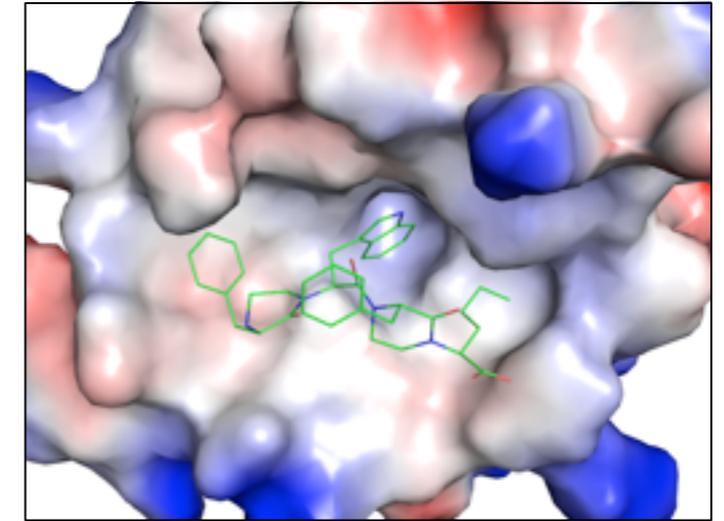
FWF Norleucine

OOP Design
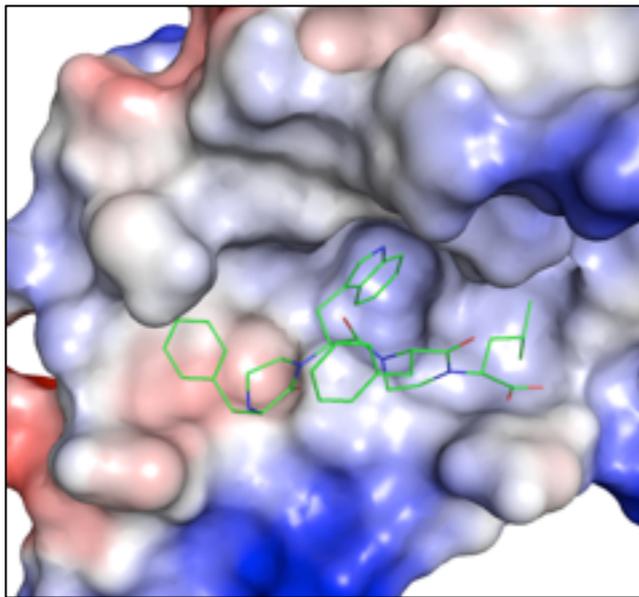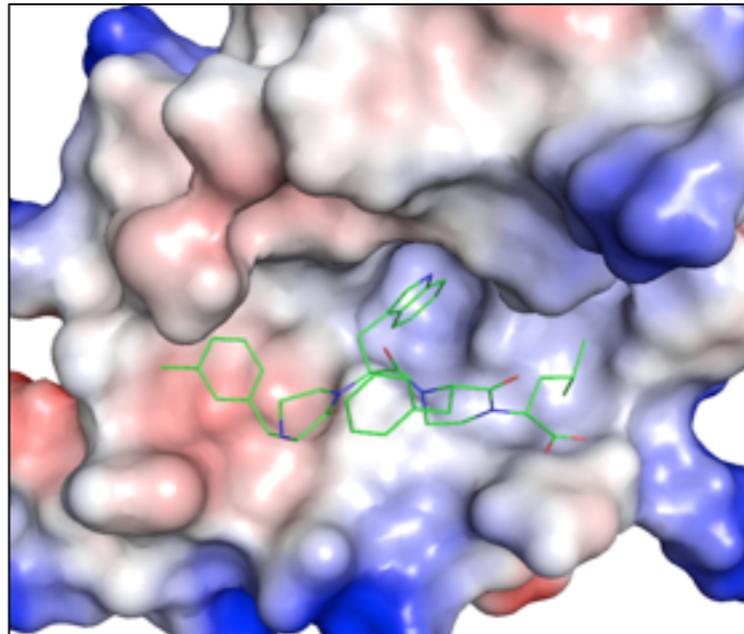
FWFL   [3-methyl-PHE] WFL   FWF Norleucine
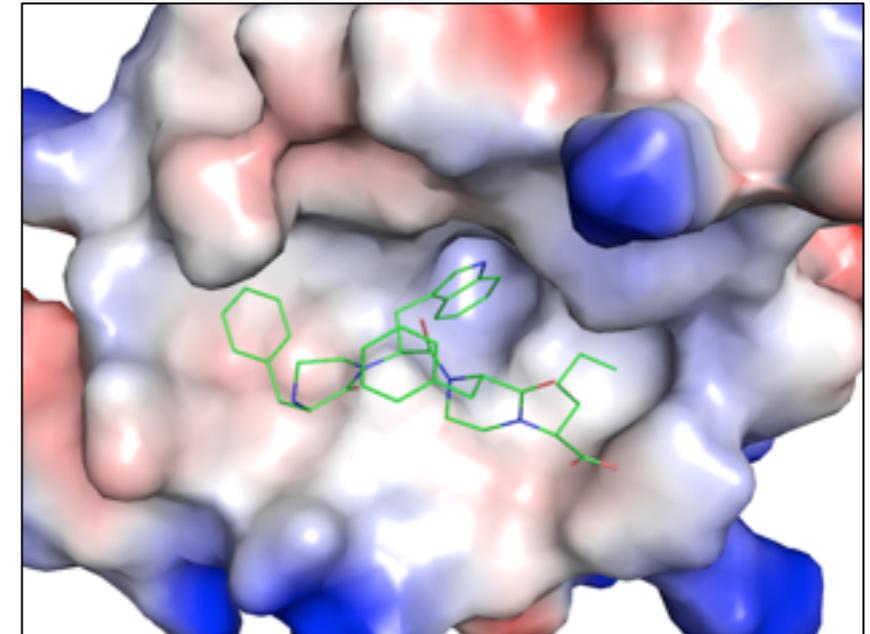
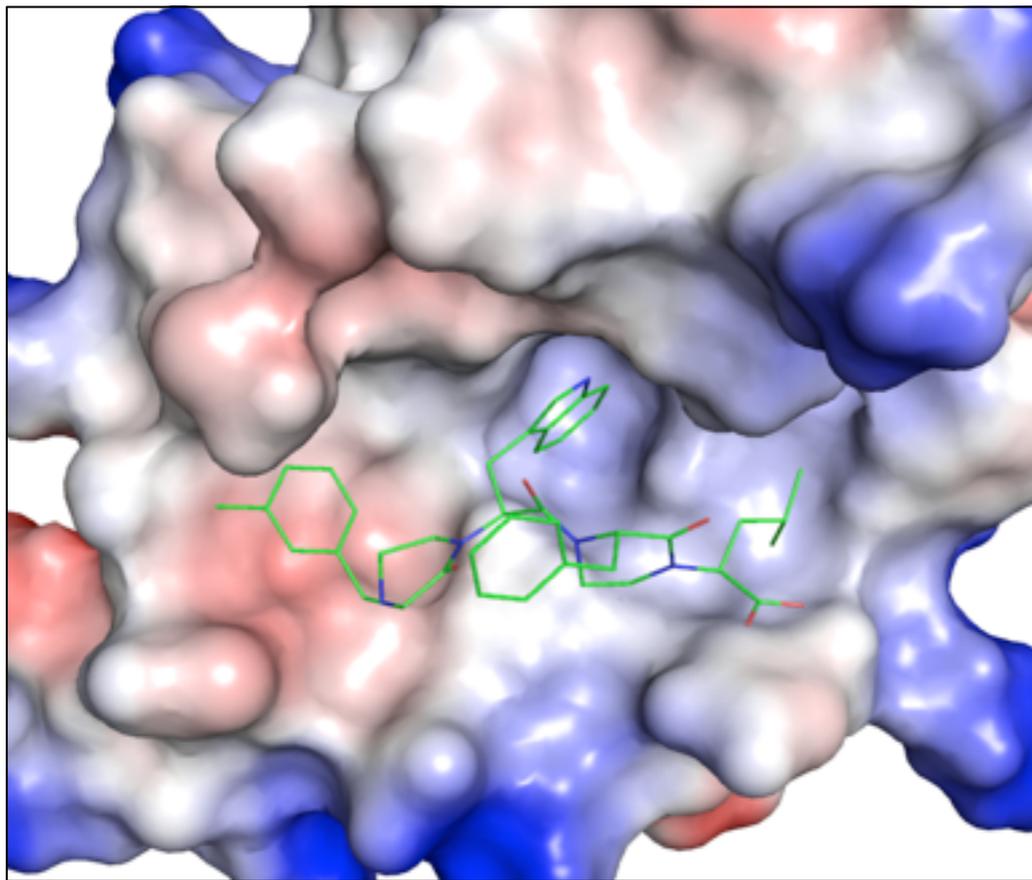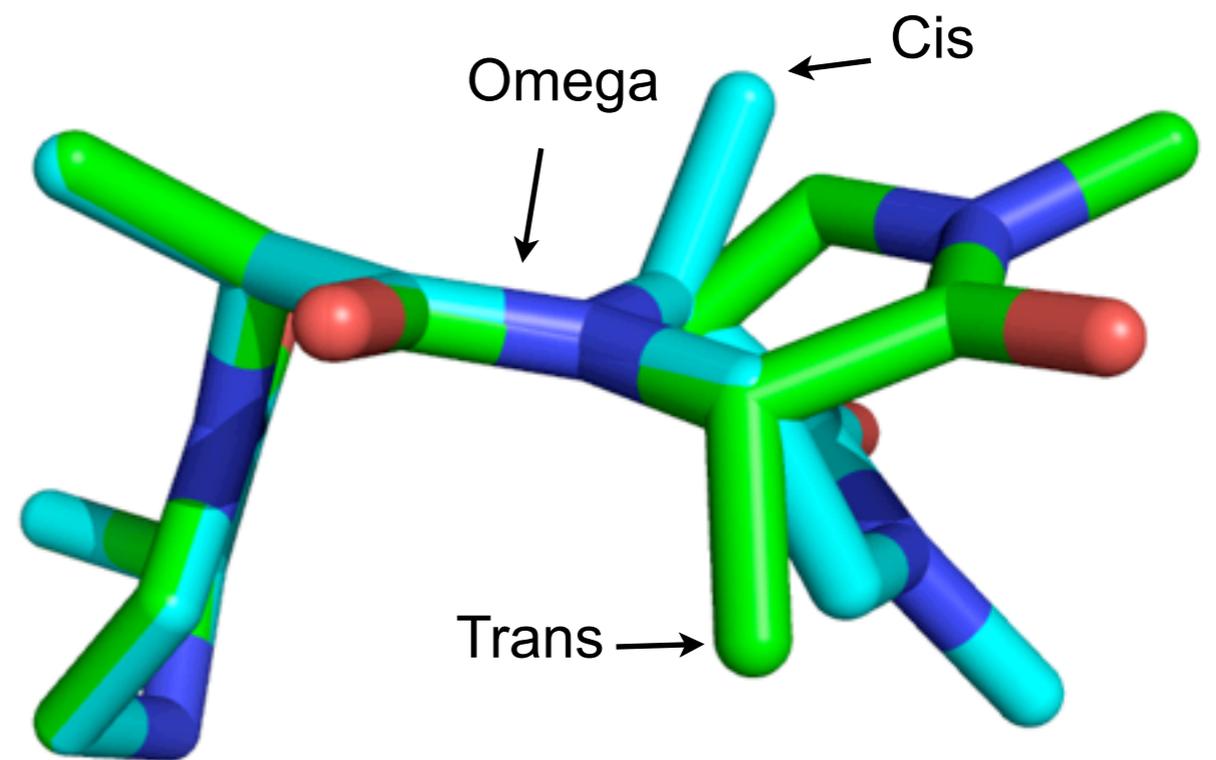| Metric	FWFL	[3-methyl-PHE]WFL	FWF Norleucine
K_d	6.9 uM Bullock (Arora Lab)	pending	pending

OOP Binding Mode
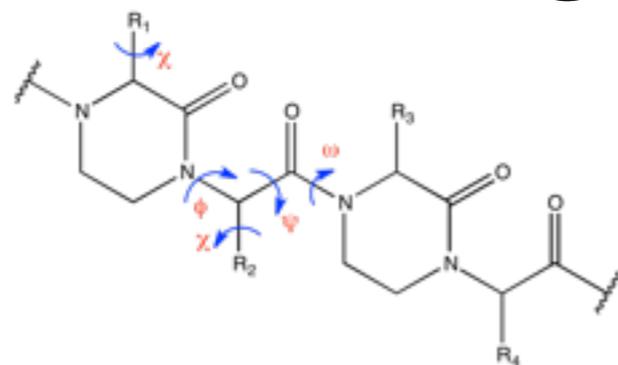
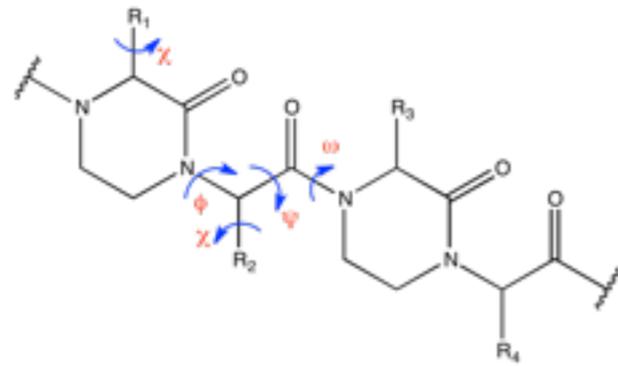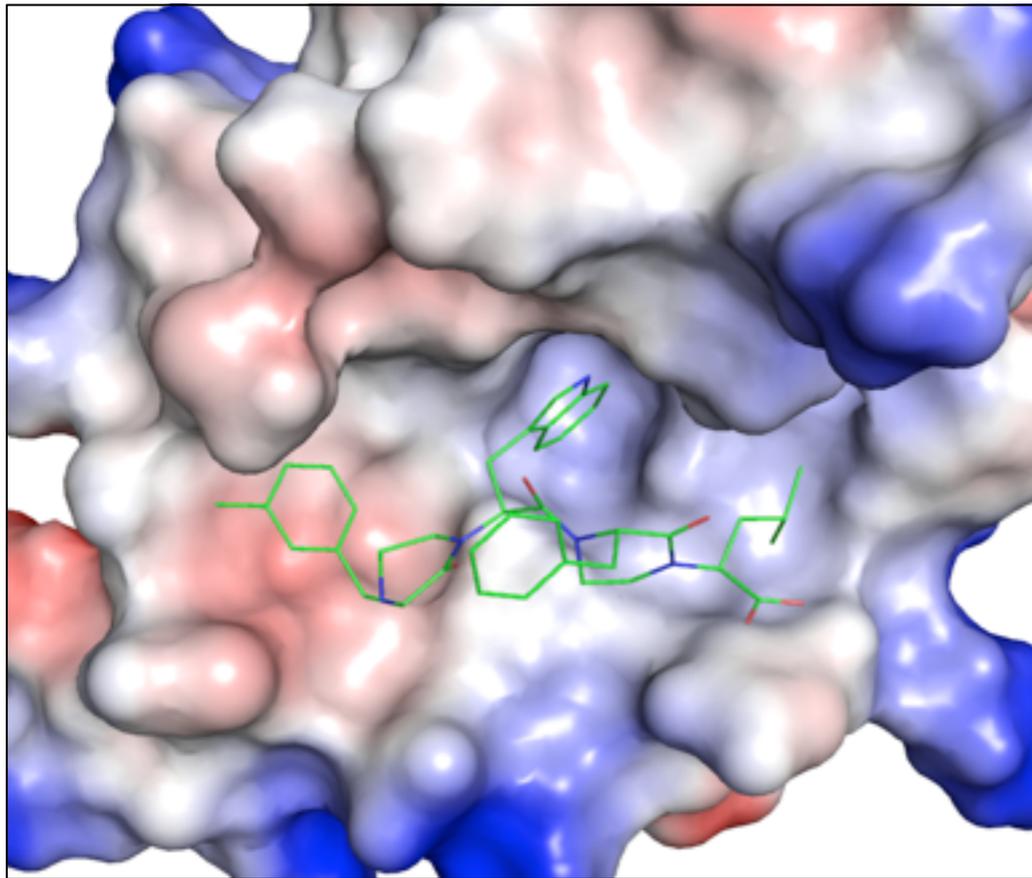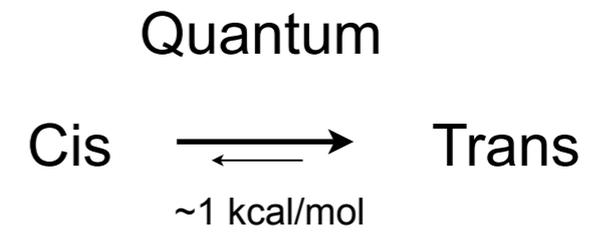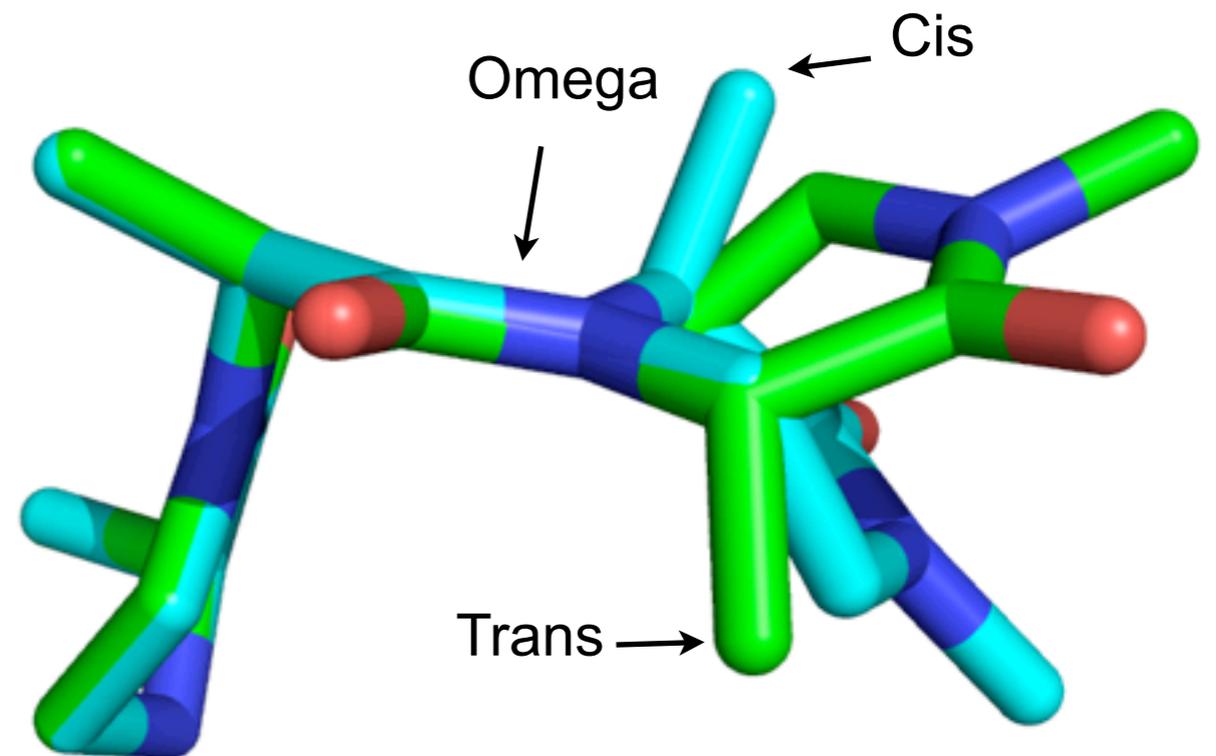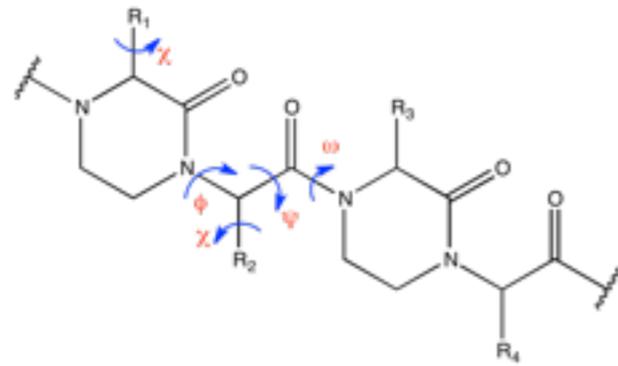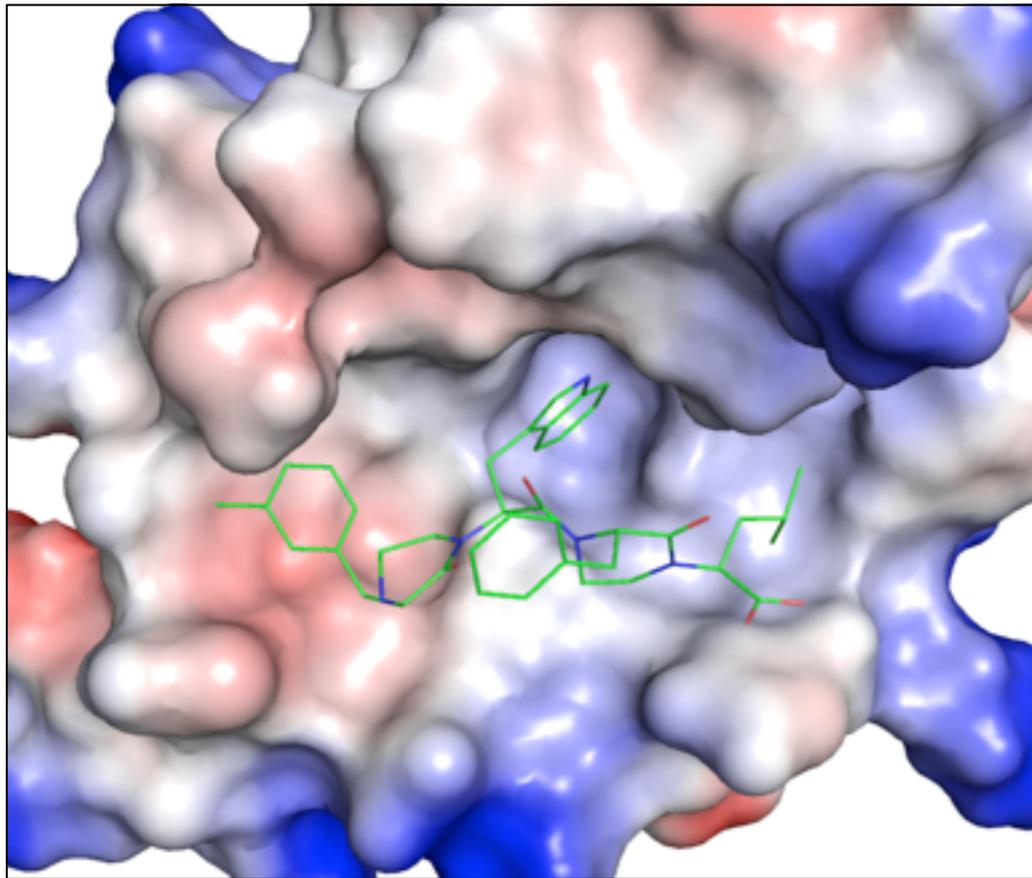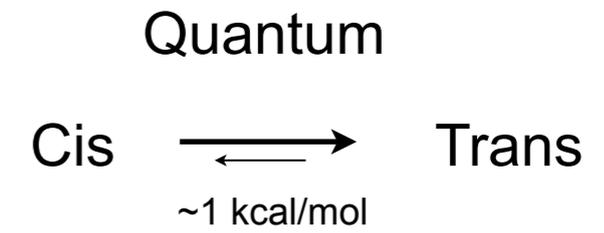

Omega Trans or Cis?
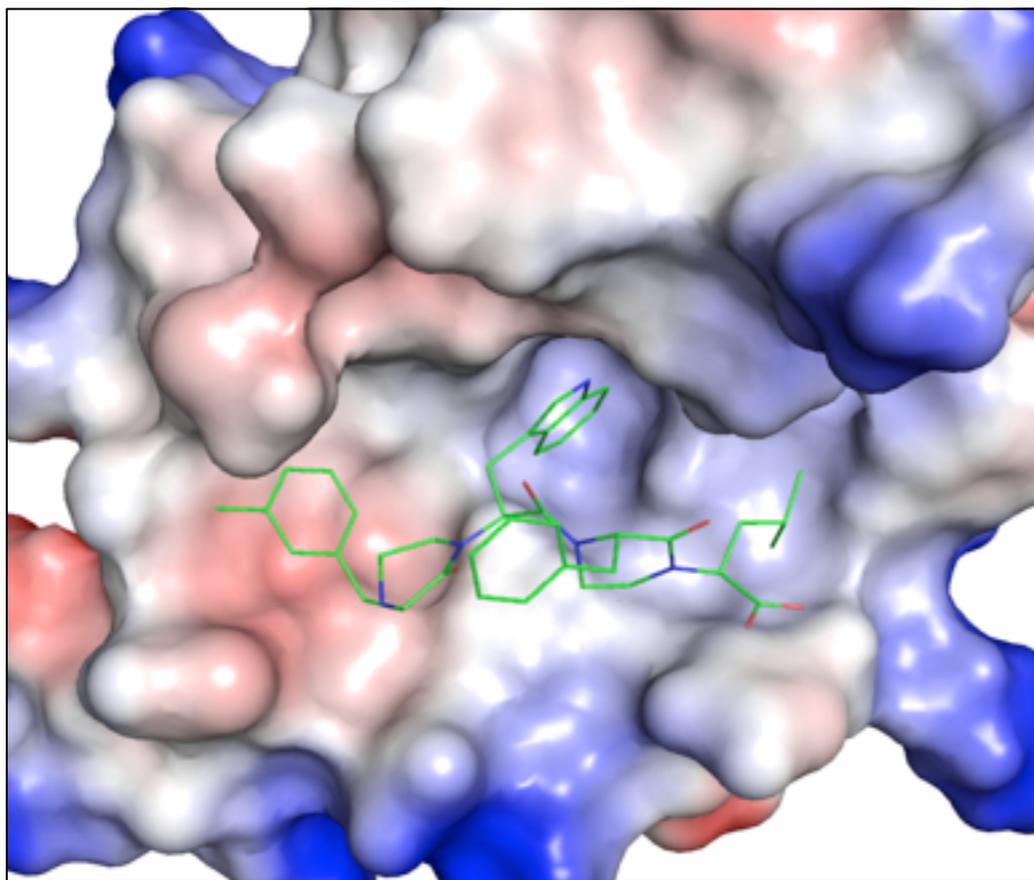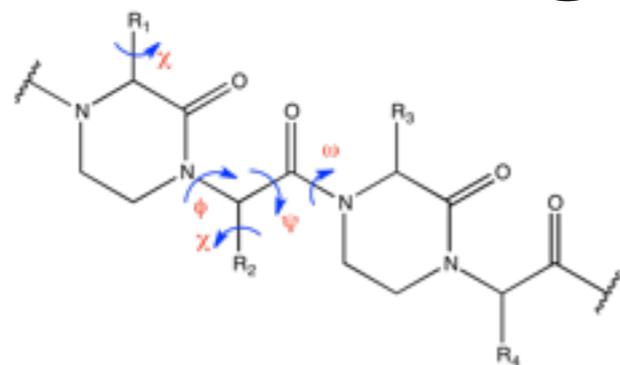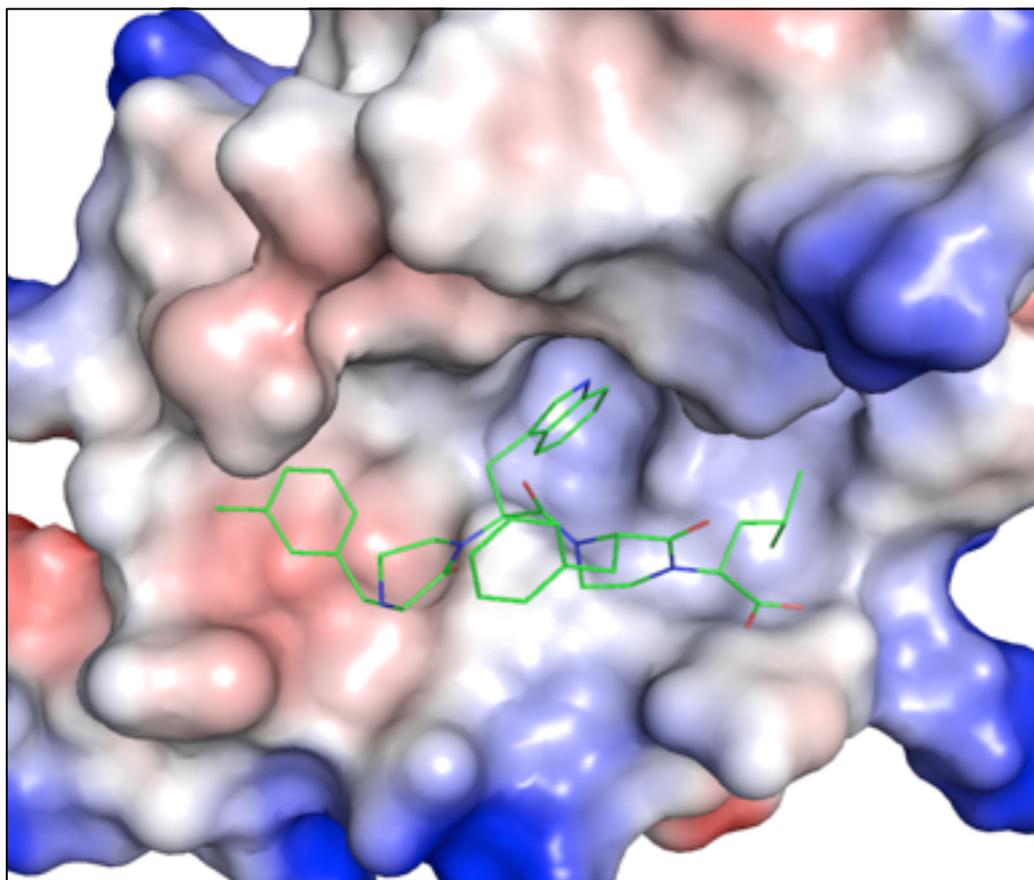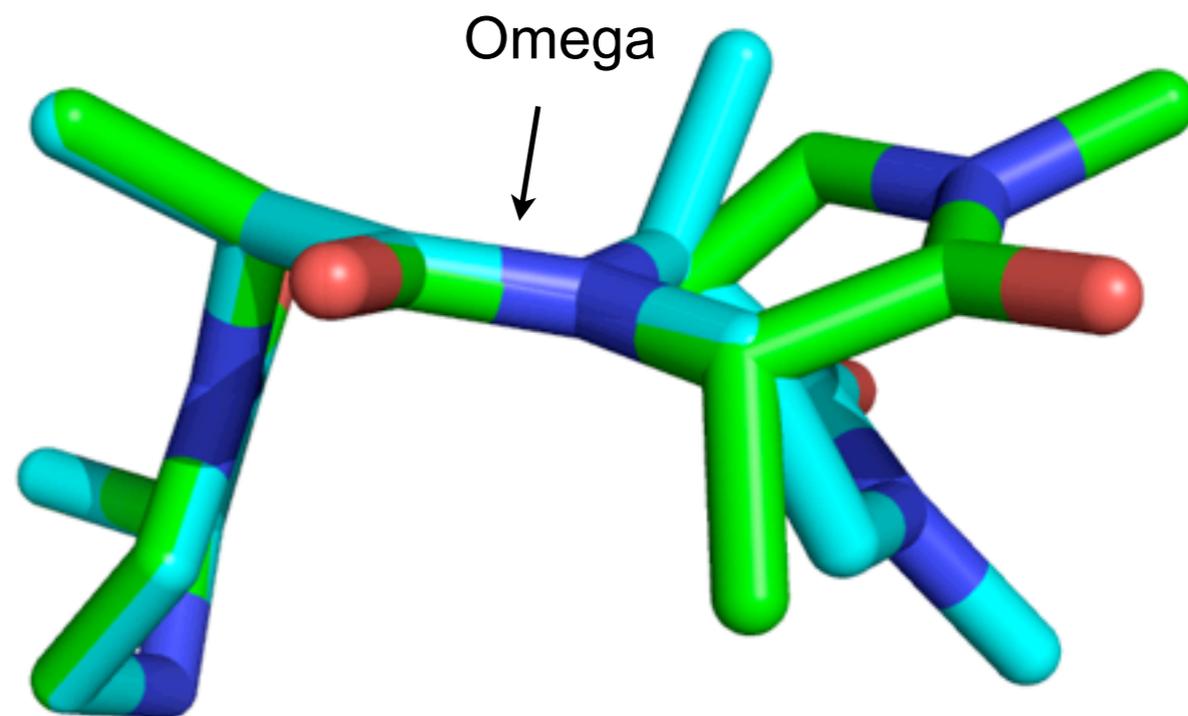
Trans
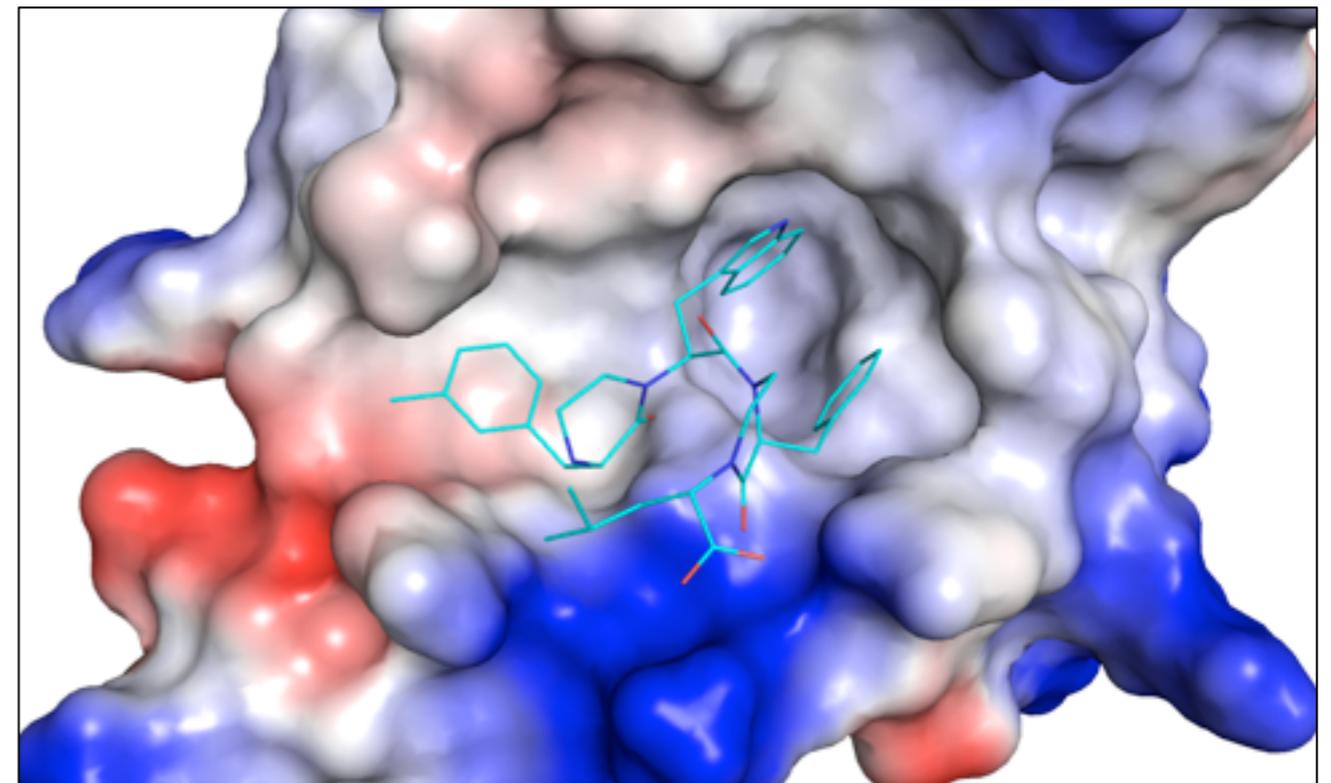
OOP Binding Mode

Omega Trans or Cis?

Trans

Omega

OOP Binding Mode

Omega Trans or Cis?

Trans

OOP Binding Mode

Omega Trans or Cis?

Quantum

Cis $\underset{\sim 1 \text{ kcal/mol}}{\rightleftharpoons}$ Trans

Trans

Omega

Cis

Trans

OOP Binding Mode

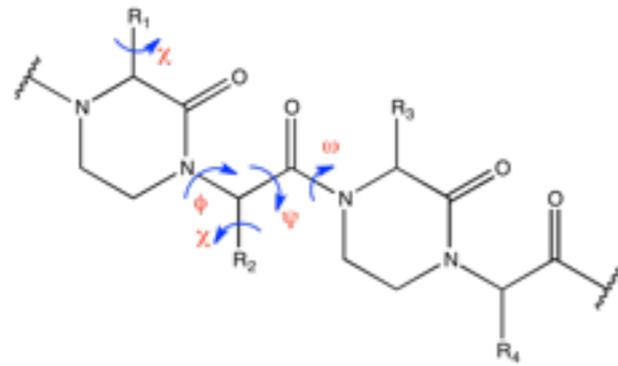Omega Trans or Cis?

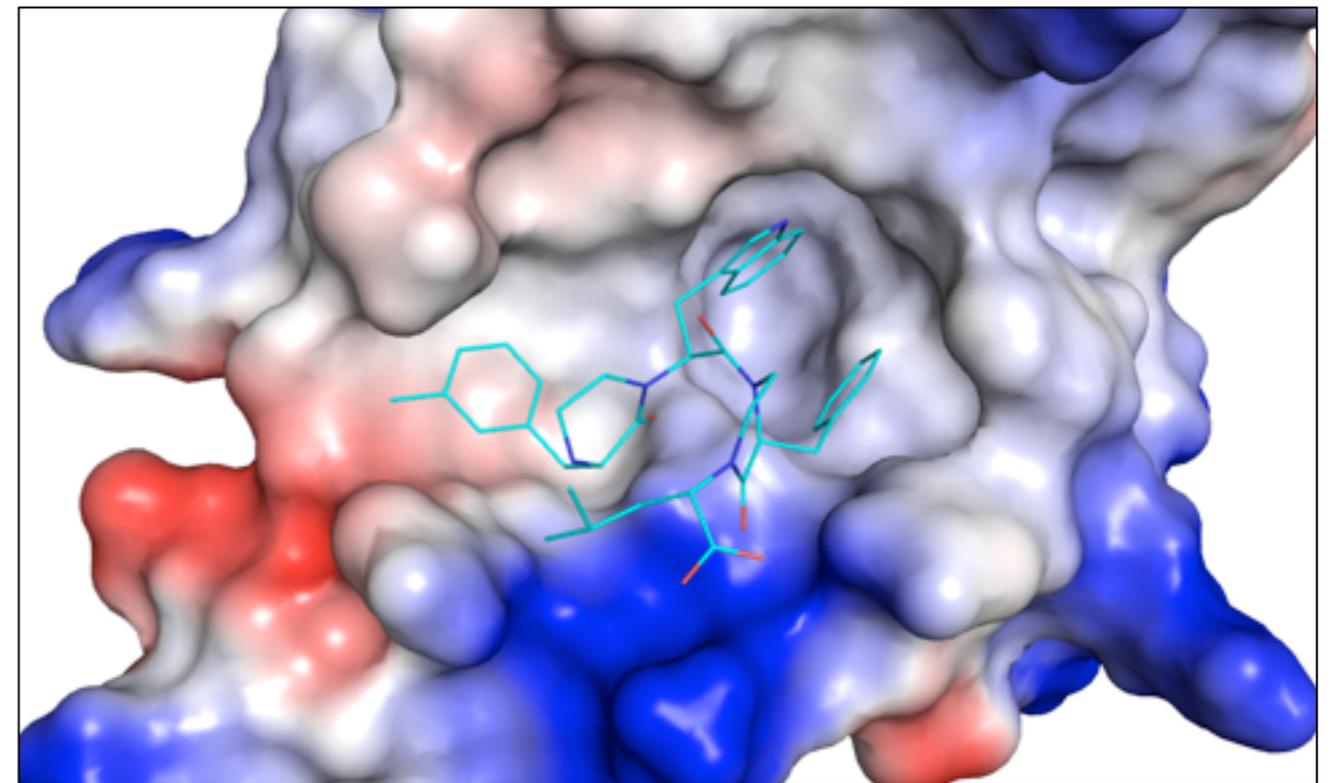Quantum

Cis ⇌ Trans

~1 kcal/mol
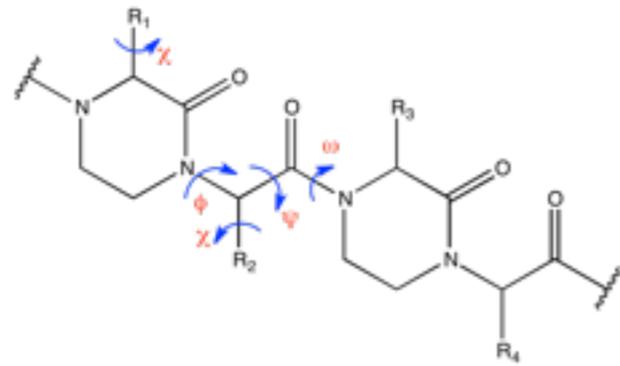
Trans

Cis

OOP Binding Mode

Omega Trans or Cis?

Quantum

Cis $\longleftrightarrow$ Trans

~1 kcal/mol
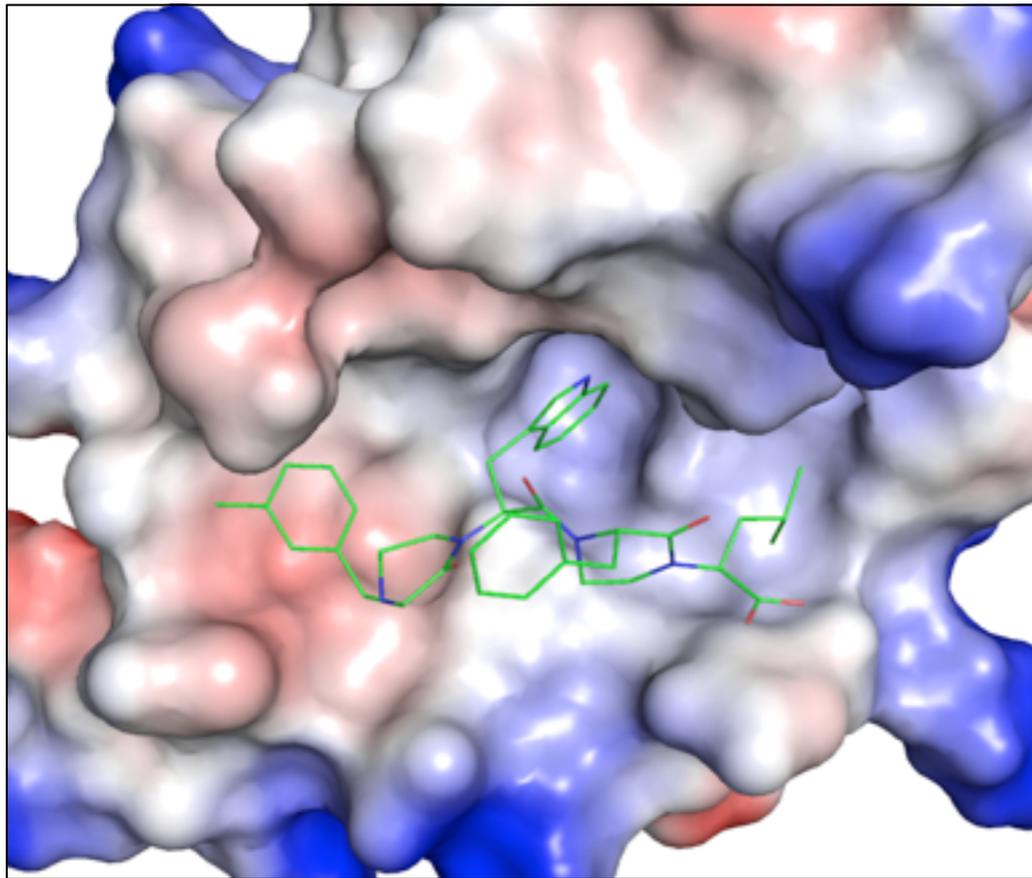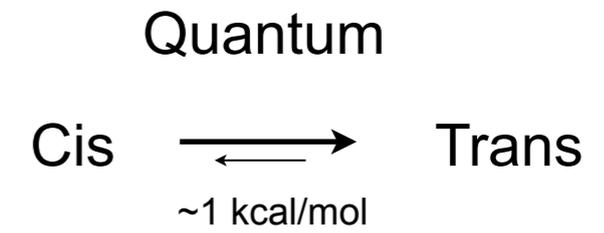
Trans
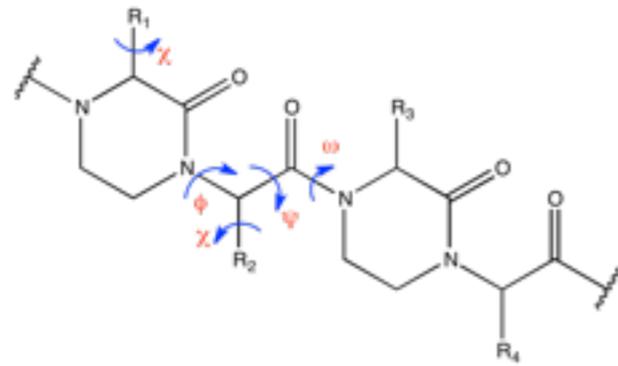
Energy Complex (REU) = -31.264

Cis

Energy Complex (REU) = -32.493

OOP Binding Mode

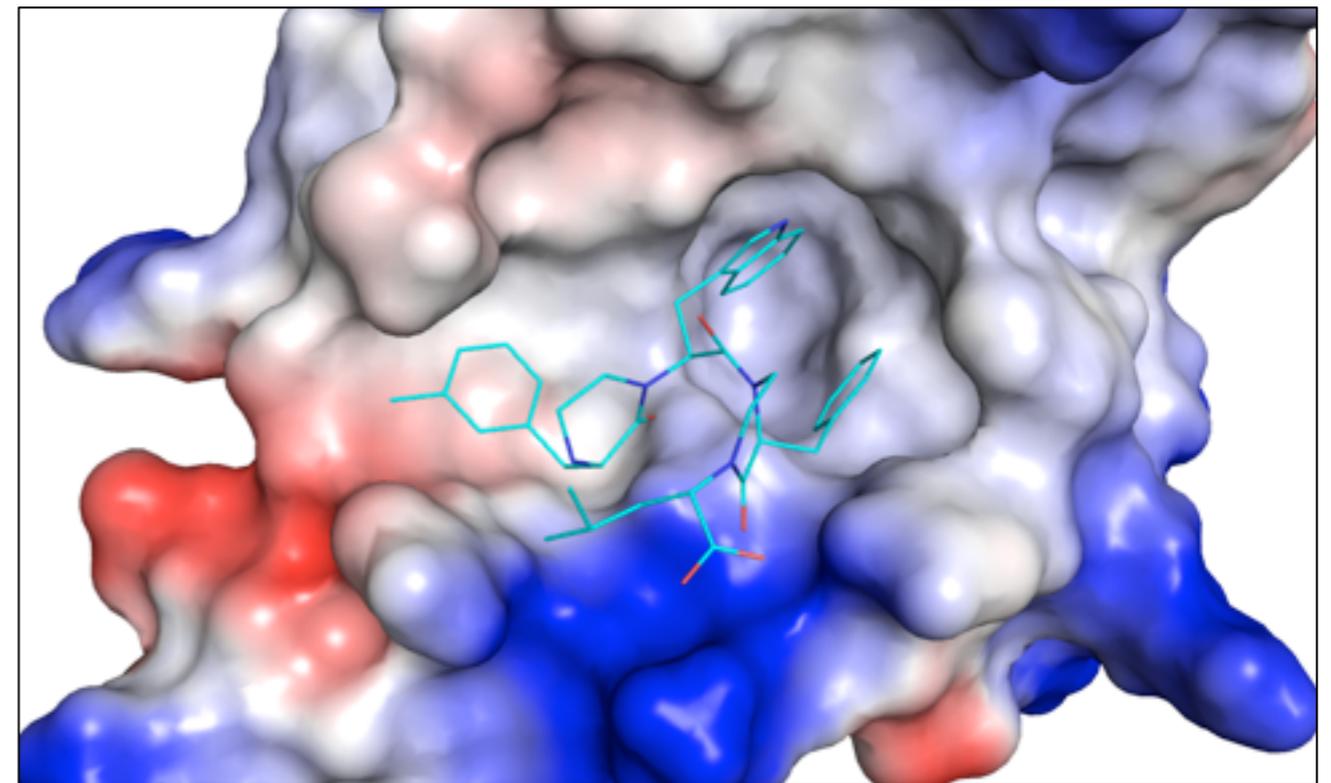Omega Trans or Cis?

Quantum

Cis $\rightleftharpoons$ Trans

~1 kcal/mol

Trans

Cis

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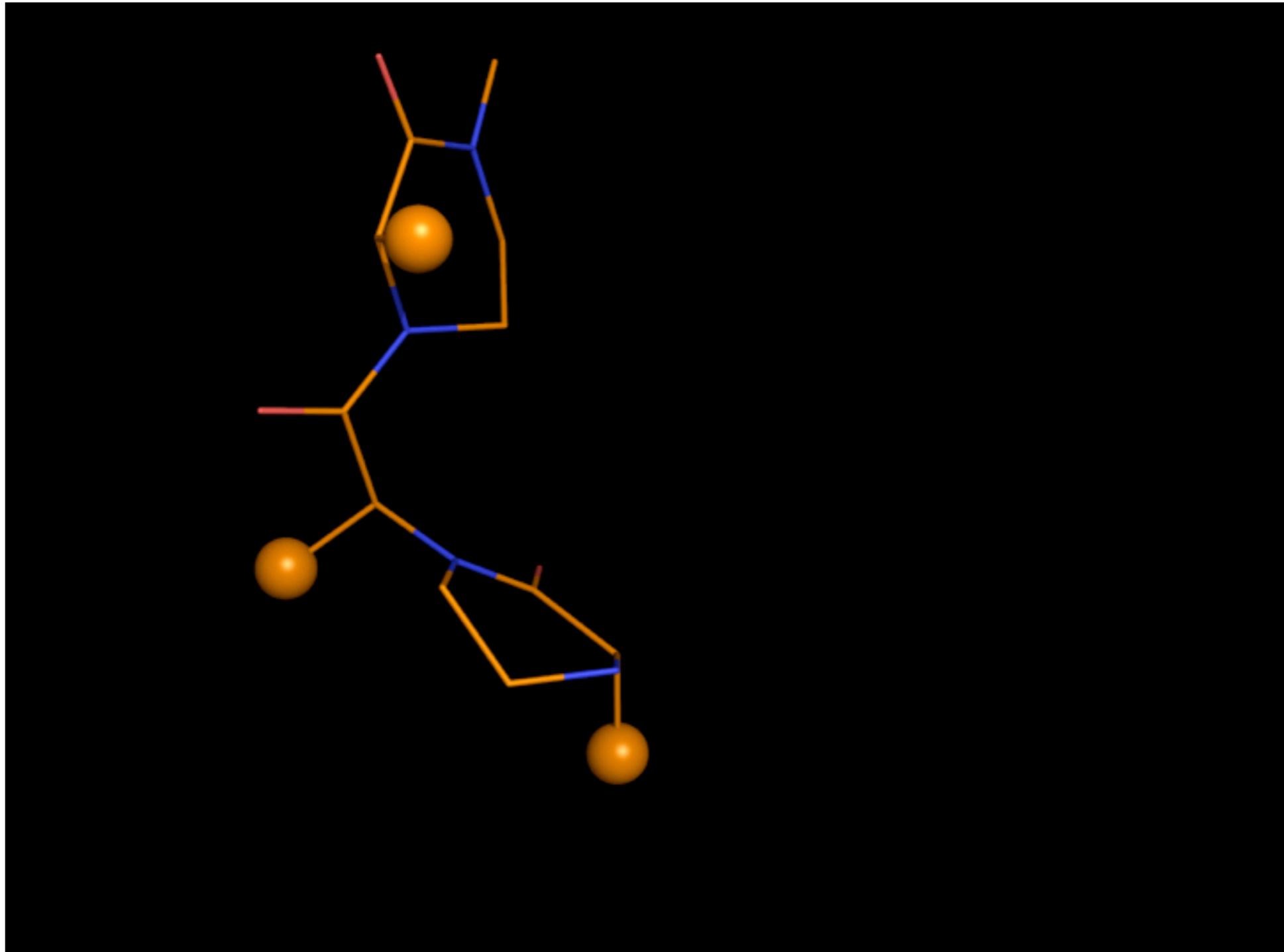
Energy Complex (REU) = -32.493

Cis orientation explains experiment:

FWFL K_d = **6.9uM**

FWKL K_d > **200uM**
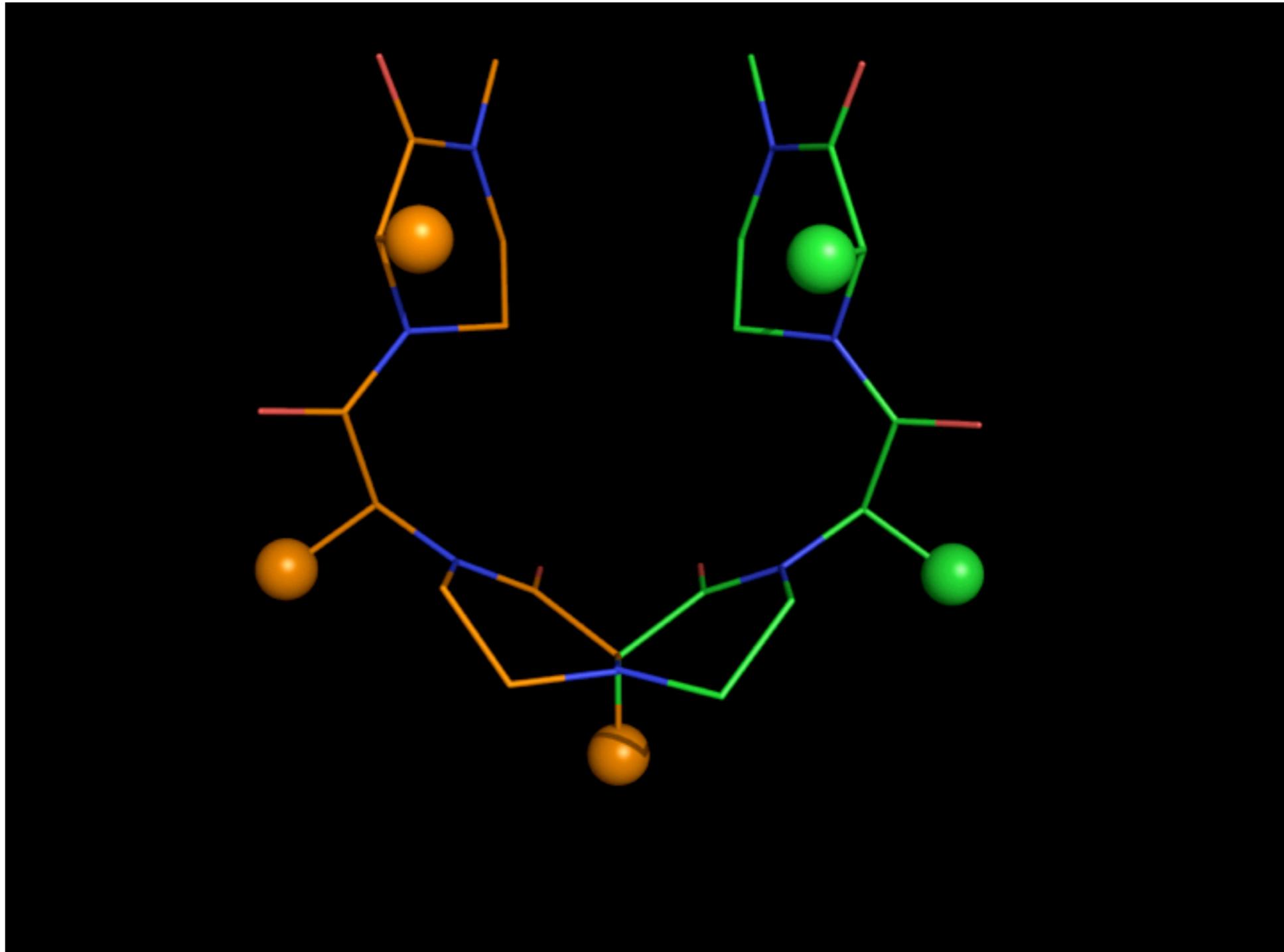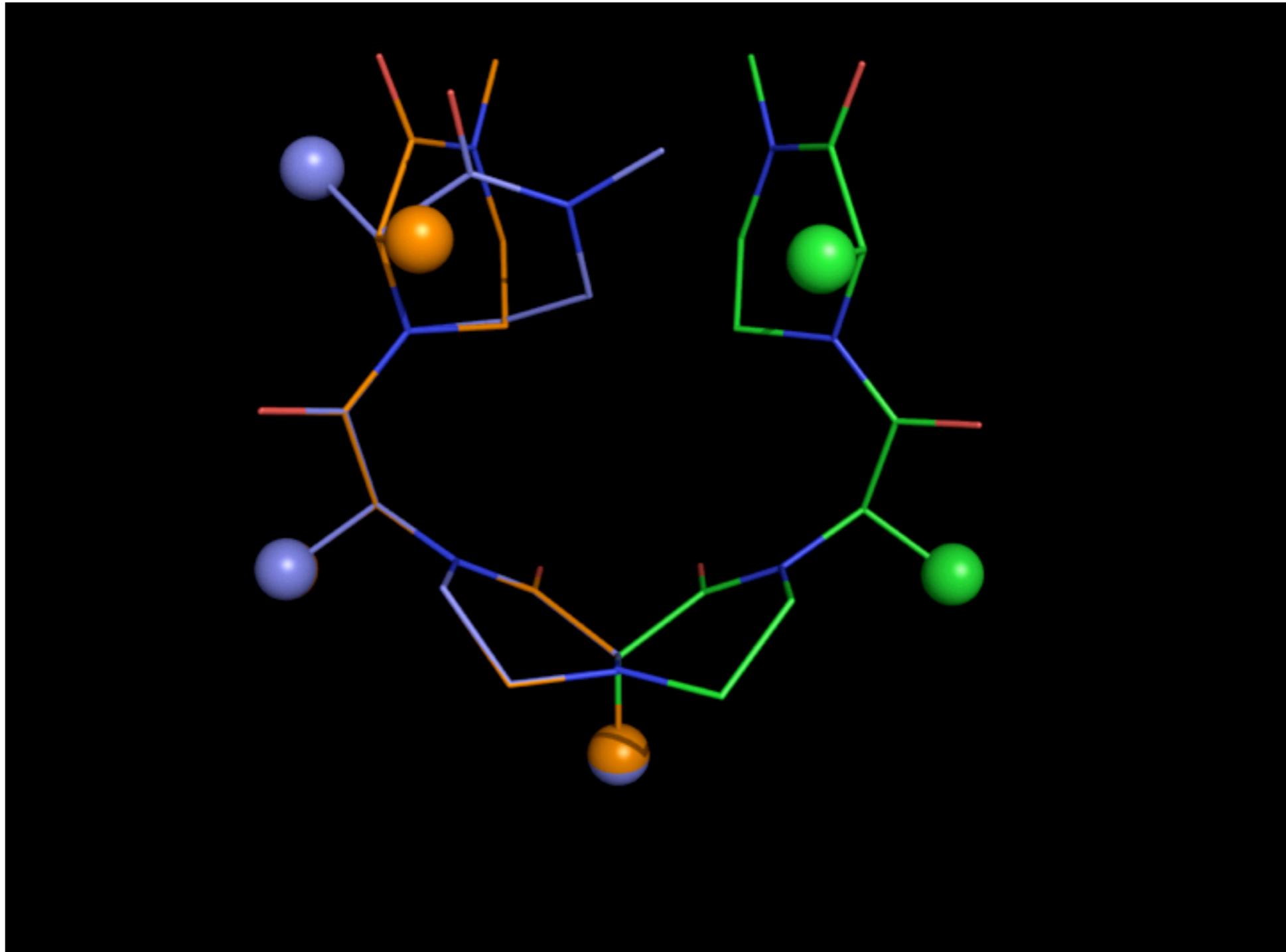
Increasing Versatility of OOPs

LLL

Increasing Versatility of OOPs

LLL

DDD

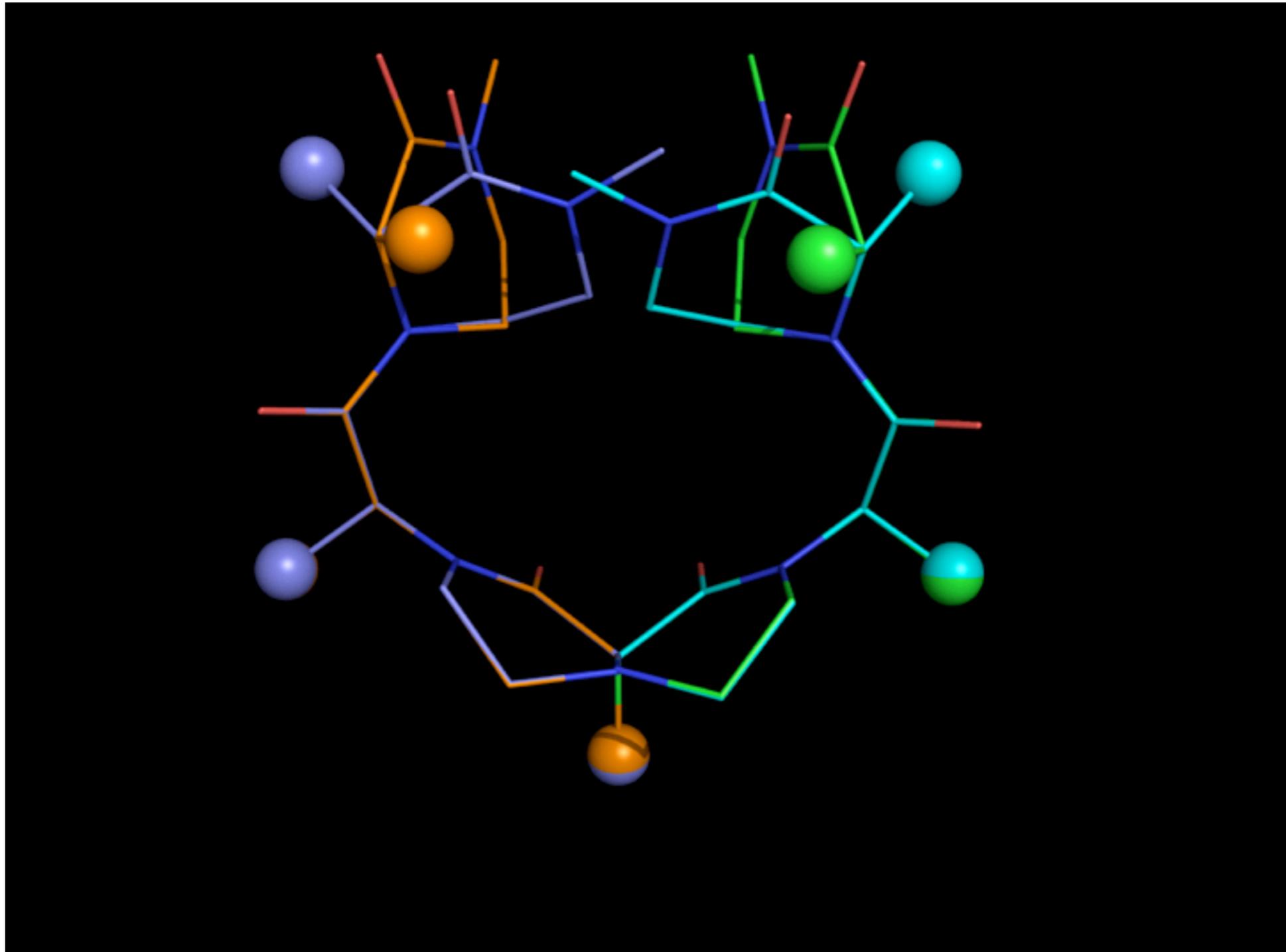# Increasing Versatility of OOPs

LLL

LLD

DDD

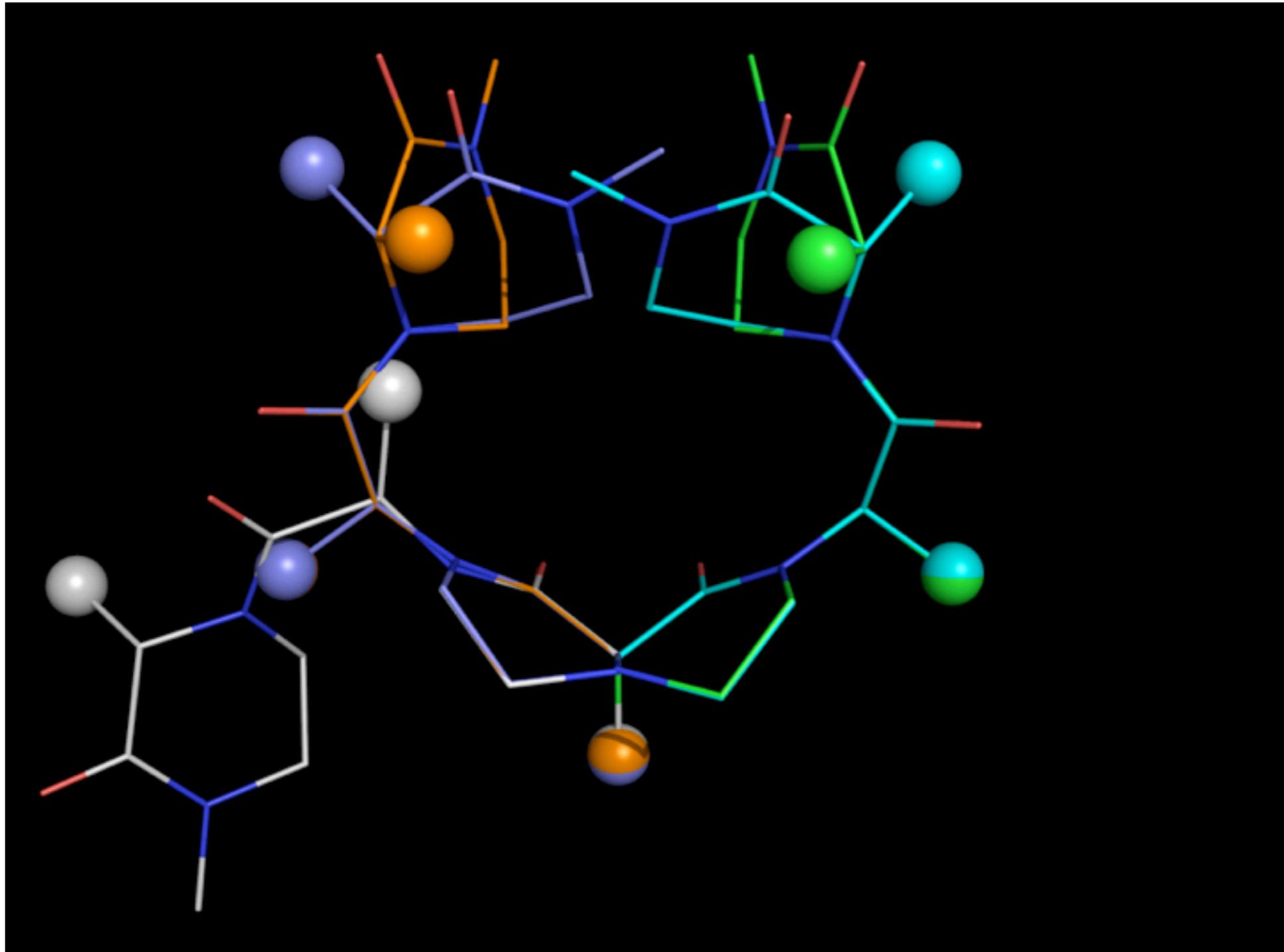# Increasing Versatility of OOPs

LLL

LLD

DDD

DDL

Increasing Versatility of OOPs
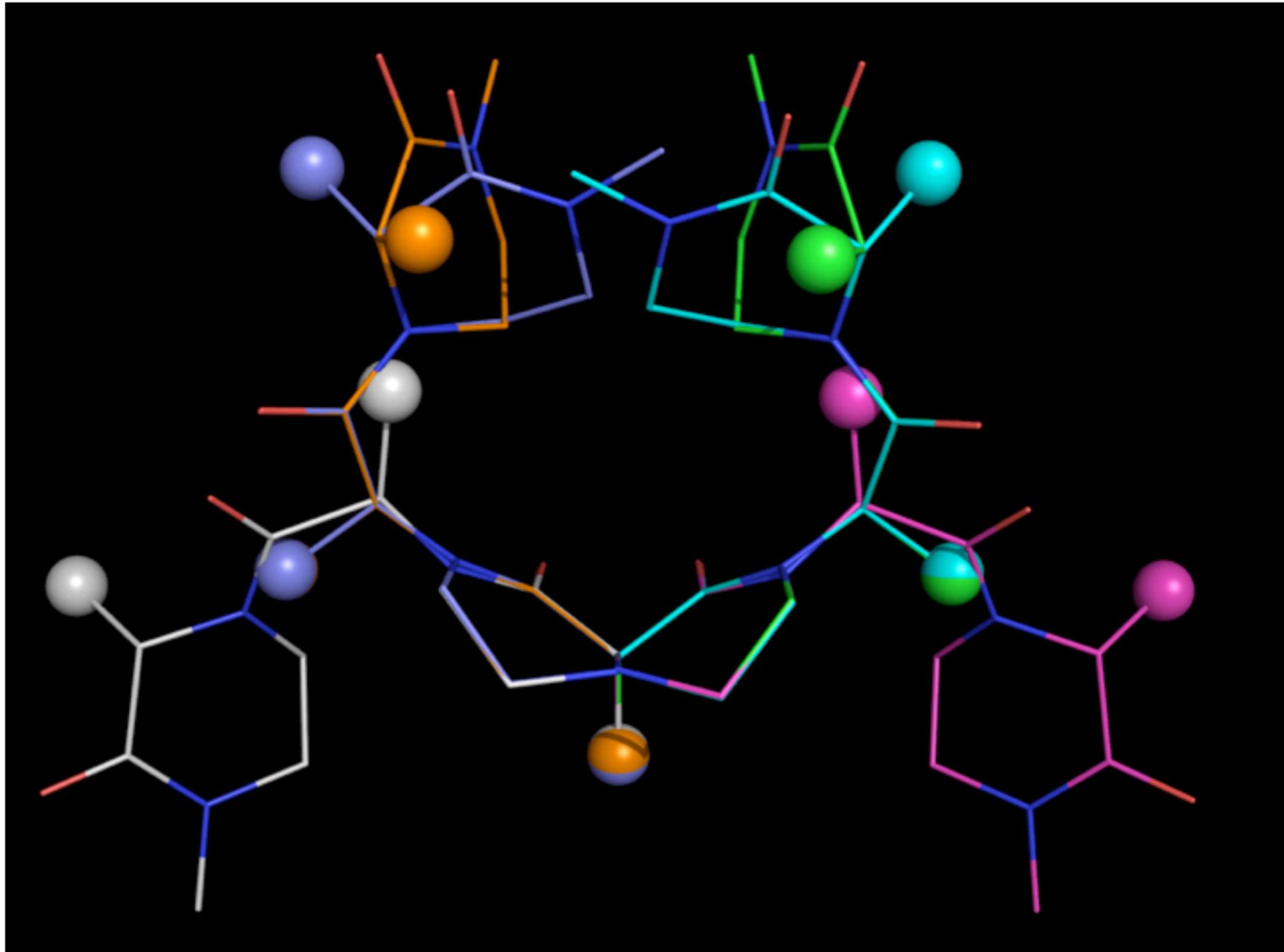

LLL

LLD

LDL

DDD

DDL

Increasing Versatility of OOPs
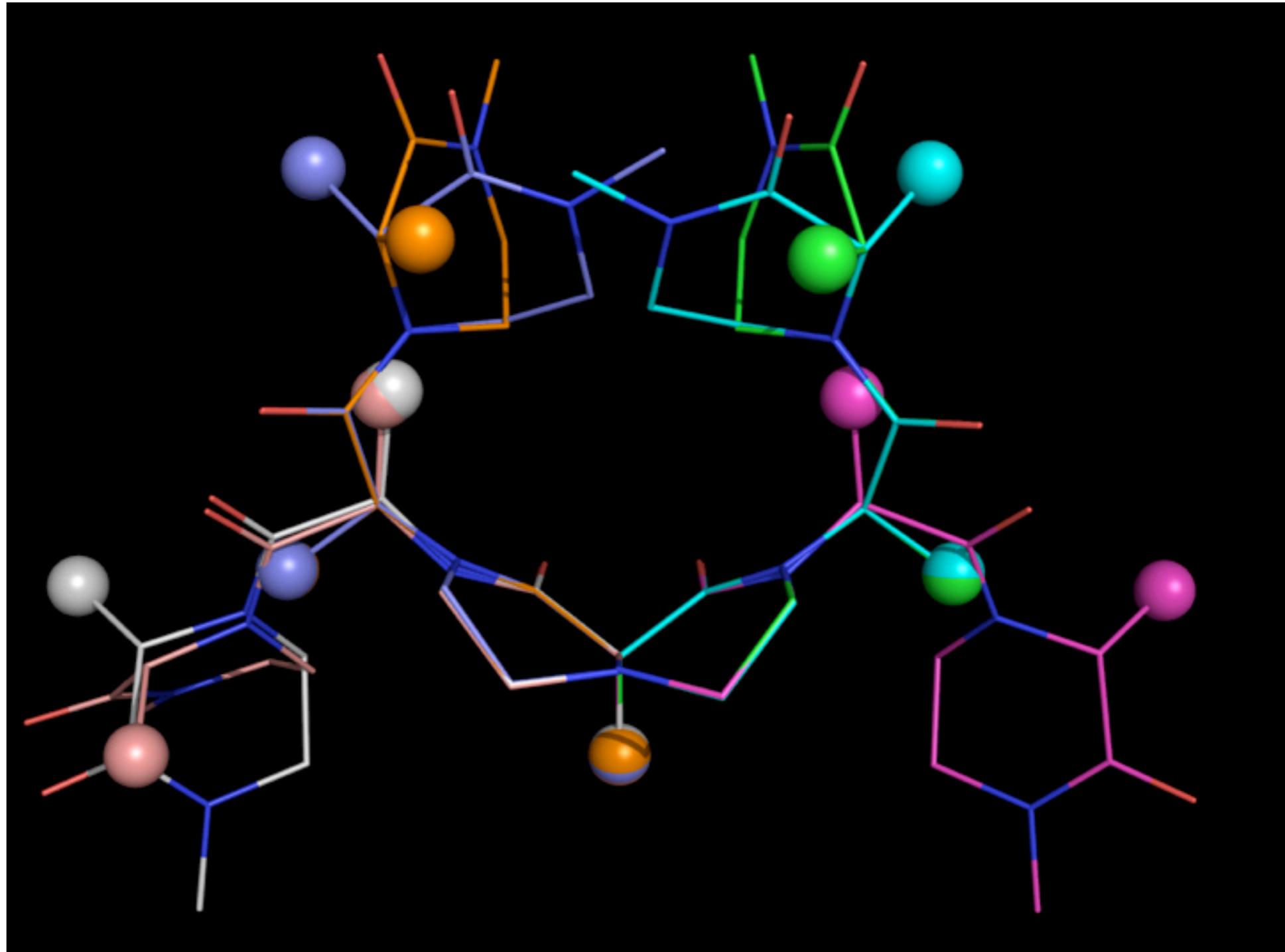
LLL

LLD

LDL

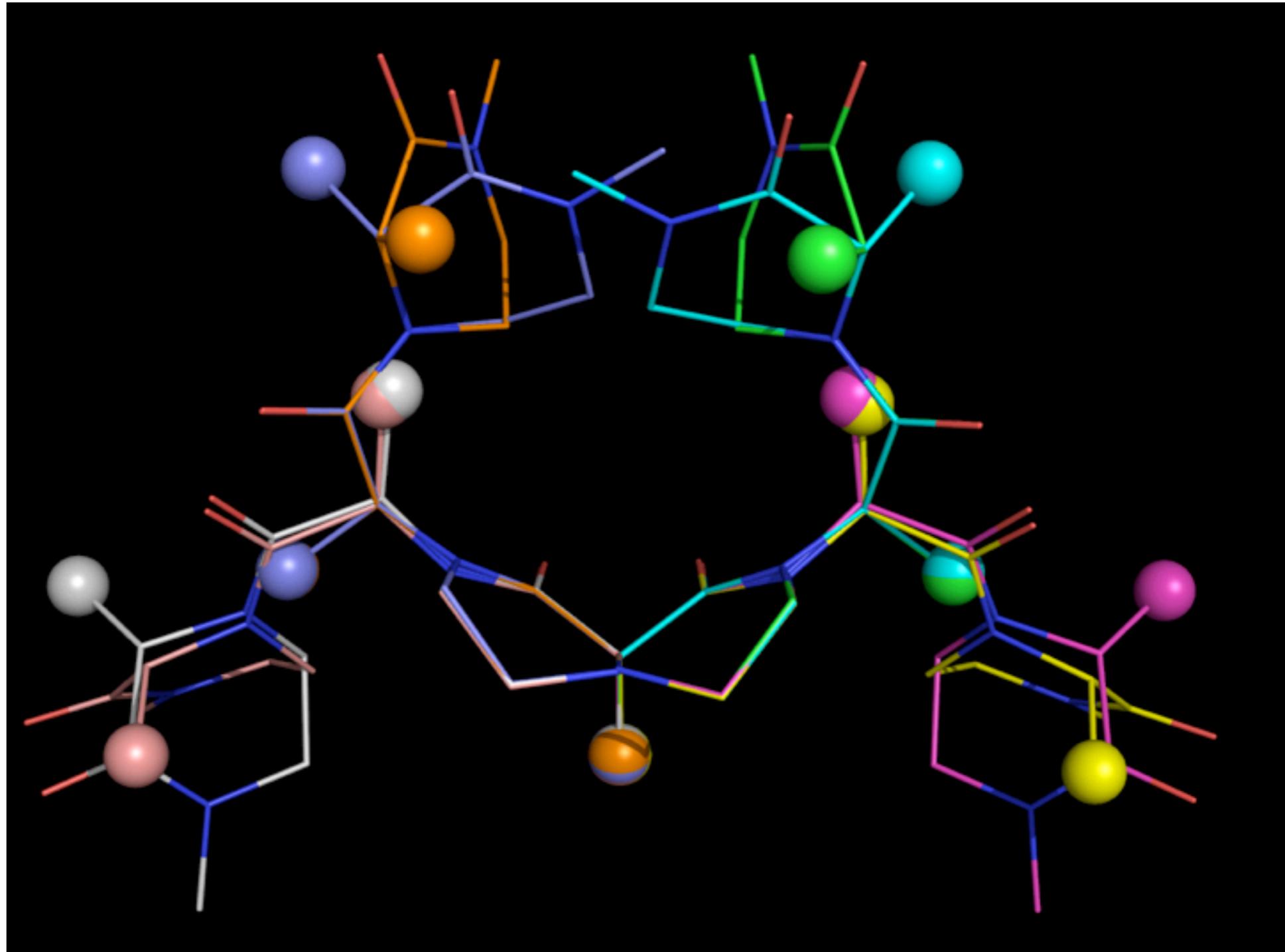DDD

DDL

DLD

Increasing Versatility of OOPs

LLL

LLD

LDL

LDD

DDD

DDL

DLD

Increasing Versatility of OOPs

LLL

LLD

LDL

LDD

DDD

DDL

DLD

DLL

Acknowledgements

Rosetta-Commons

Arora Lab

Brooke Bullock*

Bonneau Lab

P. Douglas Renfrew*
Glenn Butterfoss*
Alex Greenfield
Aviv Madar
Chris Poultney
Duncan Penfold Brown
Leif Halvorsen
Kieran Mace
Noah Youngs
Christoph Hafemeister
Evan Baugh
Tim Craven
Abba Leffler
Patrick Winters (former)

Funding

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Committee

Alex Morozov (Rutgers Physics)
Christine Vogel (NYU Biology)
Jane Carlton (NYU Parasitology)
Paramjit Arora (NYU Chemistry)
Richard Bonneau (NYU Biology)