Non-canonical Peptide and Macrocycle design with Rosetta

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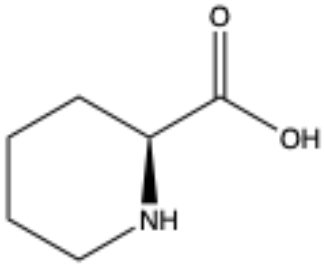
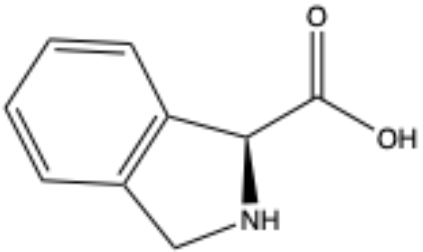
Rosetta Workshop

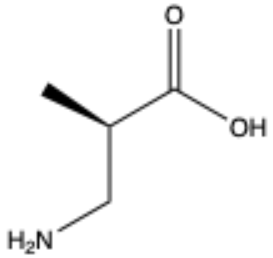
12/6/2023

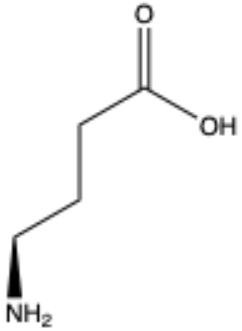
The magical world of non-canonical amino acids (NCAAs)



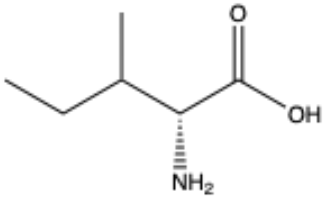


Sidechain conjugation

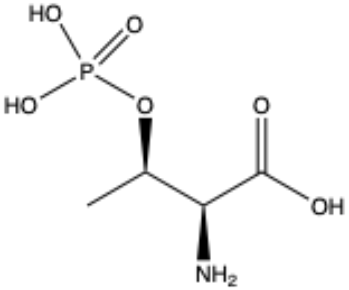




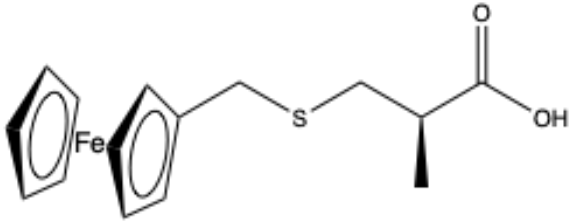
Non-canonical backbones



D-AAs



PTMs



…or just, whatever

Rosetta can do

Rosetta can’t do

Anatomy of a Rosetta amino acid

```
ATOM  N   Nbb  NH1  -0.6046255 -0.350
ATOM  CA  CAbb CT2  -0.0257287  0.100
ATOM  C   CObb C     0.6884871  0.550
ATOM  O   OCbb O    -0.6884871 -0.550
ATOM  H   HNbb H     0.3987955  0.250
ATOM 1HA  Hapo HB    0.1157793  0.000
ATOM 2HA  Hapo HB    0.1157793  0.000

ATOM_ALIAS 1HA   HA2
ATOM_ALIAS 2HA   HA3

LOWER_CONNECT N
UPPER_CONNECT C
BOND  N    CA
BOND  N    H
BOND  CA   C
BOND  CA  1HA
BOND  CA  2HA
BOND_TYPE  C    O  2
```

Atom/bond block: Atom names, atom types, partial charges, atom connectivity

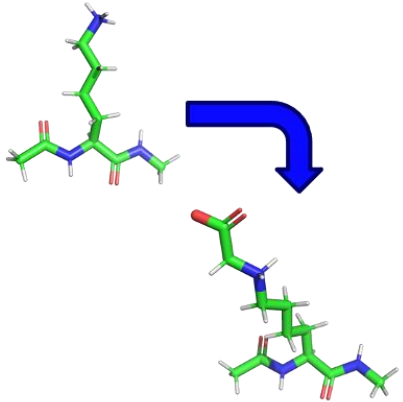
```
NAME GLY
IO_STRING GLY G
TYPE POLYMER #residue type
AA GLY
ROTAMER_AA GLY
PROPERTIES PROTEIN CANONICAL_AA ALPHA_AA METALBINDING ACHIRAL_BACKBONE
METAL_BINDING_ATOMS O
NBR_ATOM CA
# APL CA to O distance -- not yet measured; default to max CB to O dist
NBR_RADIUS 3.4473
FIRST_SIDECHAIN_ATOM NONE
RAMA_PREPRO_FILENAME all.ramaProb prepro.ramaProb
```

Property assignment: Rotamers, Ramachandrans, AA name, molecular properties, rotamers, etc.

Internal Coordinates block: Bond lengths, bond angles, dihedral angles

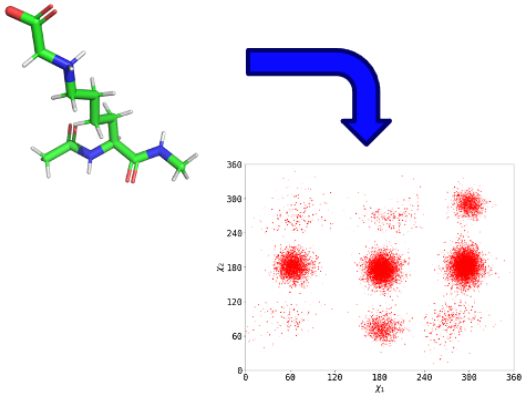
```
ICOOR_INTERNAL    N      0.000000    0.000000    0.000000   N     CA    C
ICOOR_INTERNAL    CA     0.000000  180.000000    1.458001   N     CA    C
ICOOR_INTERNAL    C      0.000000   68.799995    1.523259   CA    N     C
ICOOR_INTERNAL  UPPER  149.999969   63.800018    1.328685   C     CA    N
ICOOR_INTERNAL    O   -179.999985   59.200005    1.231015   C     CA  UPPER
ICOOR_INTERNAL   1HA   121.400000   70.500000    1.090168   CA    N     C
ICOOR_INTERNAL   2HA   117.200000   70.500000    1.089353   CA    N    1HA
ICOOR_INTERNAL  LOWER -150.000015   58.300003    1.328685   N     CA    C
ICOOR_INTERNAL    H    180.000000   60.850040    1.010000   N     CA  LOWER
```

Three methods of NCAA rotamer generation



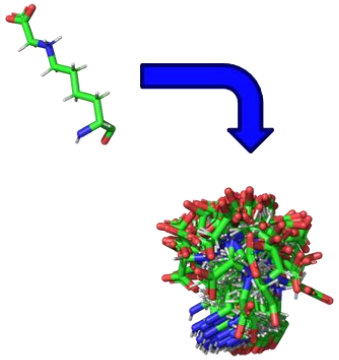
+Quick and easy
+Ensures “dunbrack-like” behavior
-Requires the NCAA resemble a CAA

“Parent” Rotamers



+Able to parameterize many NCAA using CHARMM energy
-Longest runtime
-Max chi count of 4
-Best with prior knowledge of chi distributions

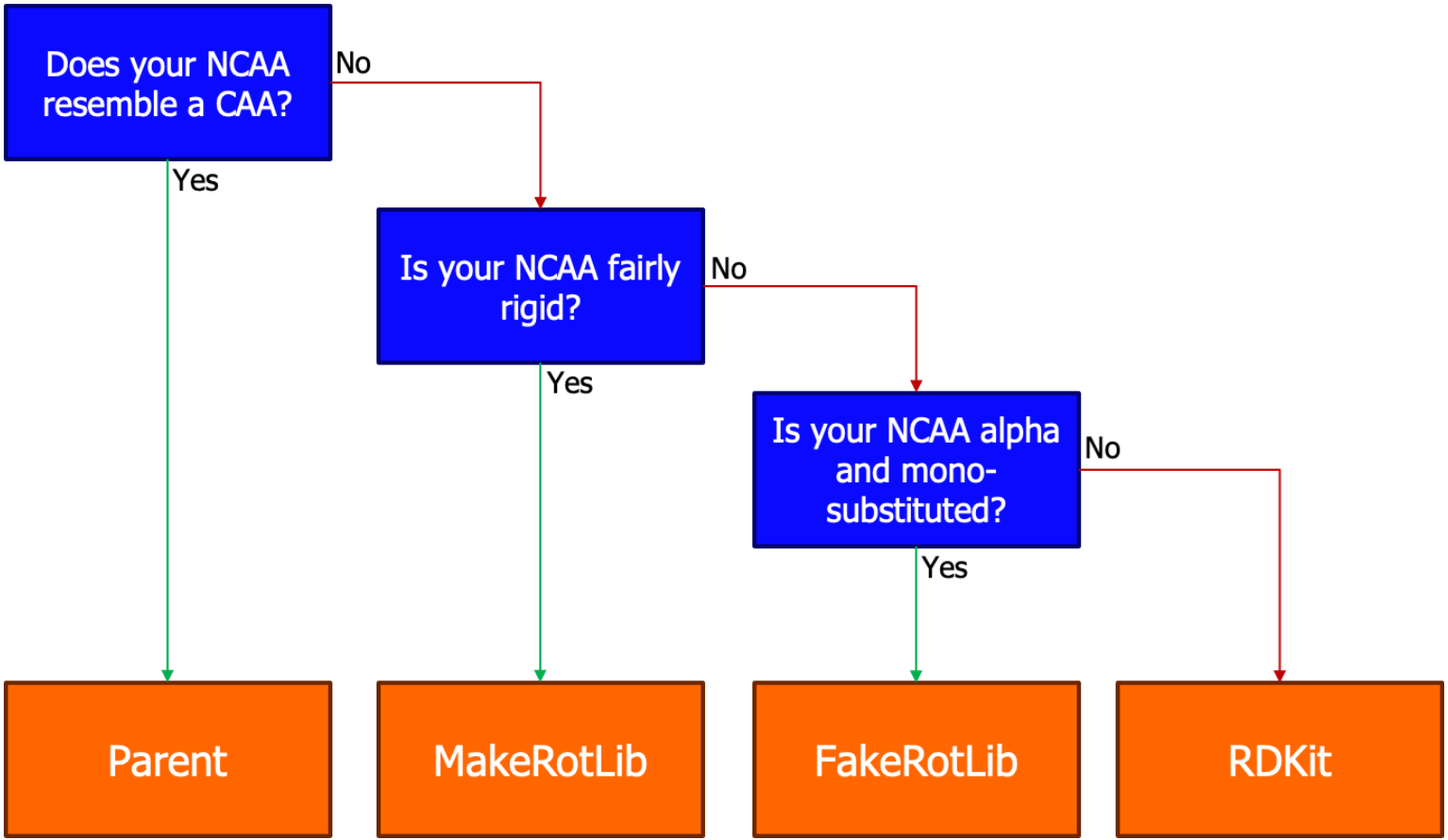
MakeRotLib



+Quick because of ligand methods
+Handles all NCAAs
-Lacks “dunbrack-like” behavior
-Worst performing method

Small Molecule approach

NCAA parameterization flowchart



RESEARCH ARTICLE | BIOCHEMISTRY |

Computationally designed peptide macrocycle inhibitors of New Delhi metallo-β-lactamase 1

Vikram Khipple Mulligan, Sean Workman, Tianjun Sun, +13, and David Baker Authors Info & Affiliations

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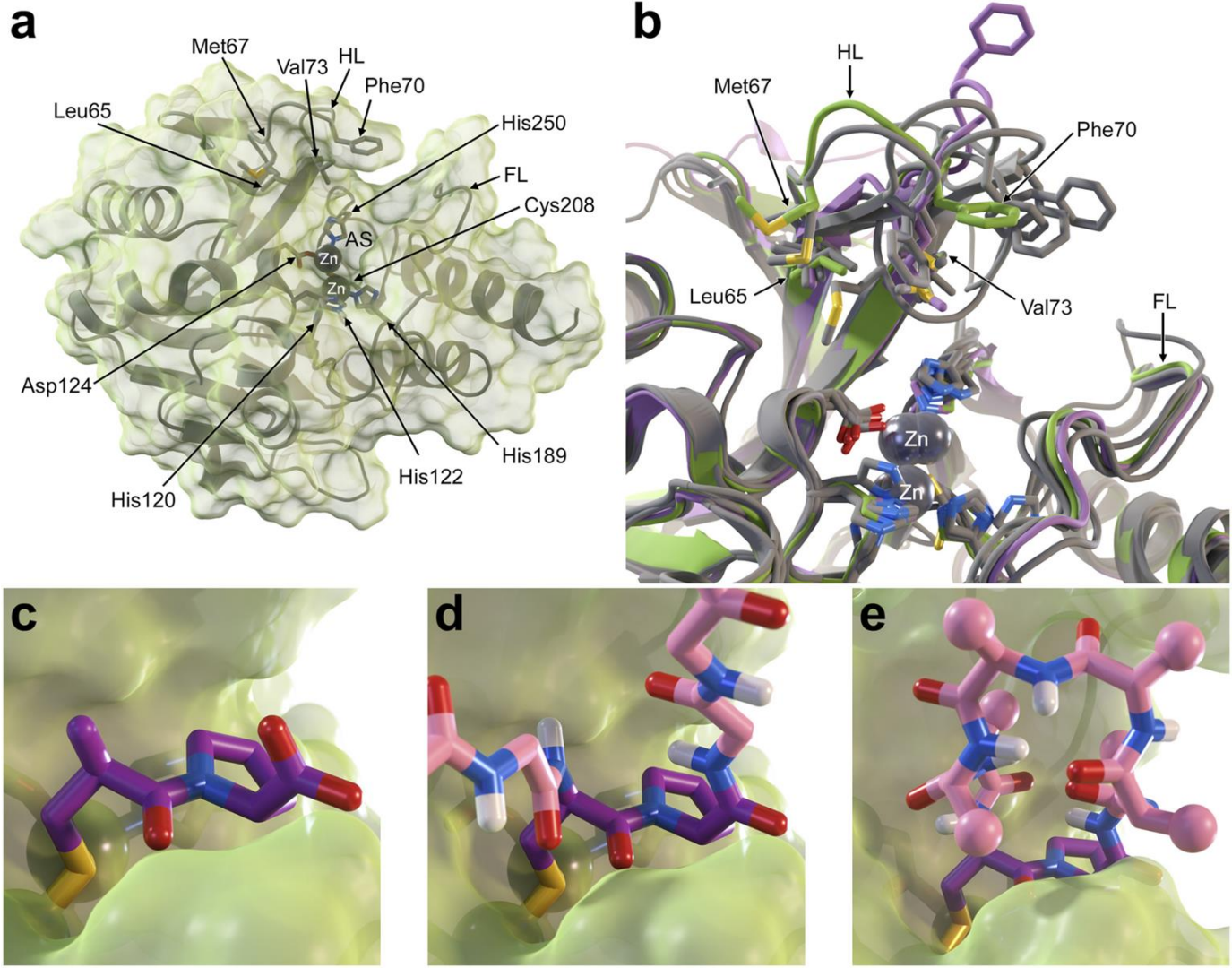
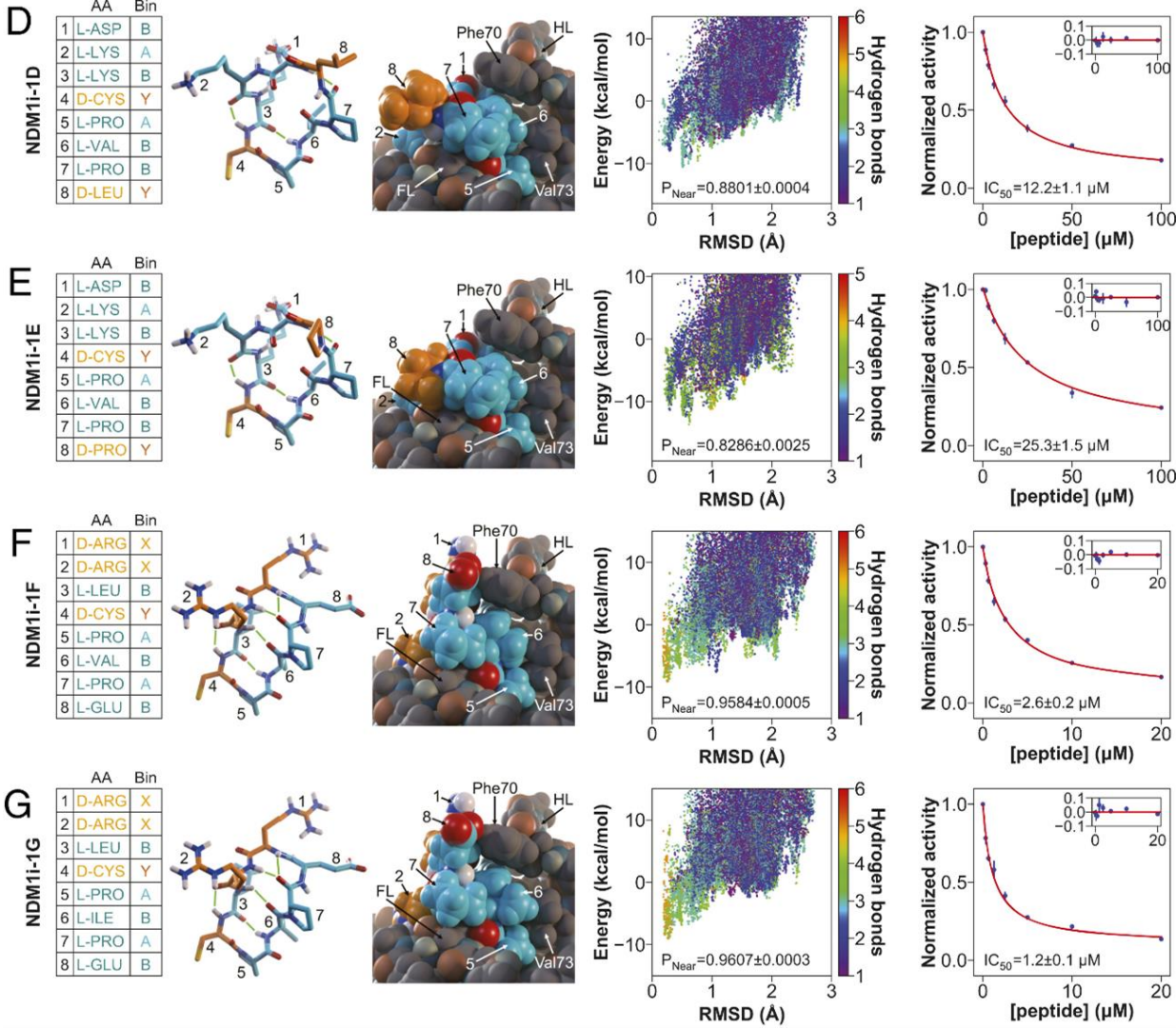
9,743 | 21

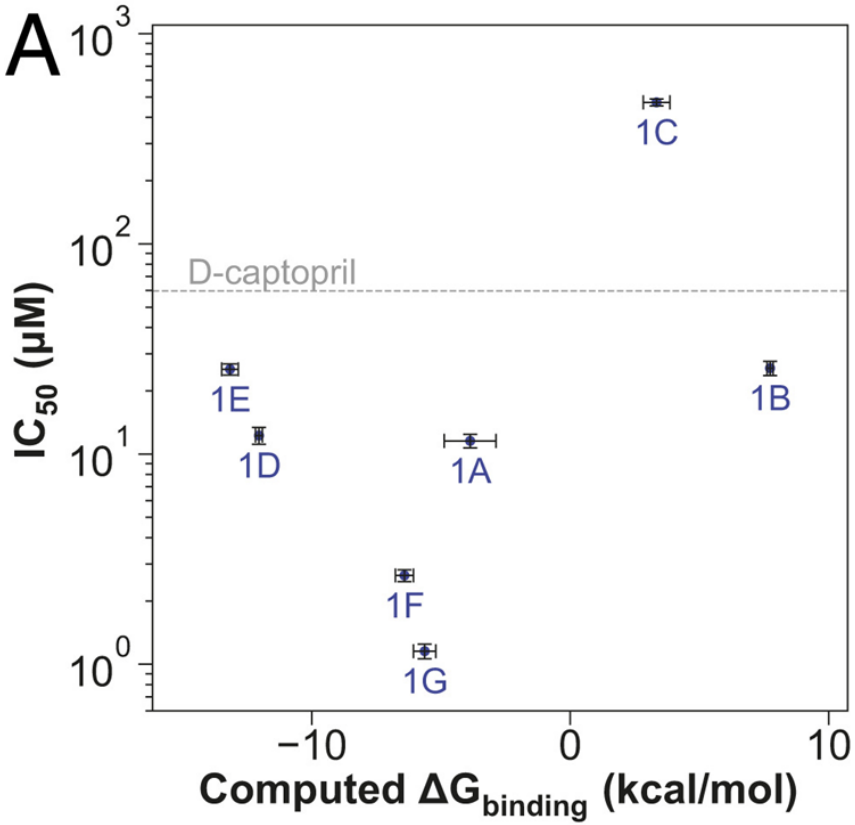
Peptide design starts from an L-Cys D-Pro stub

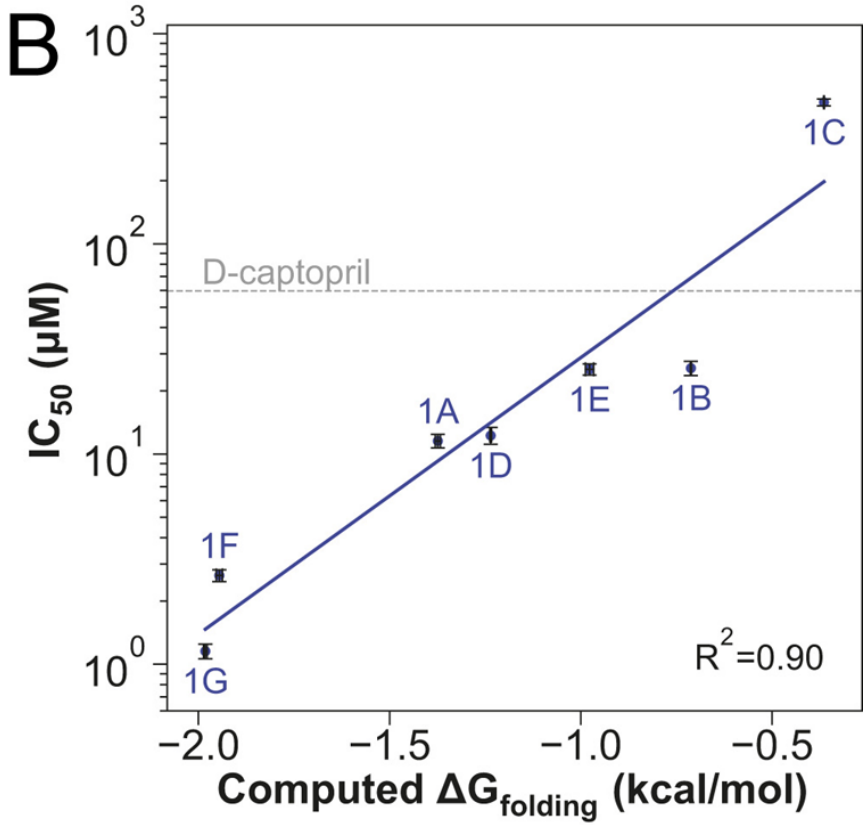


Macrocycle design produces peptides with varying activities

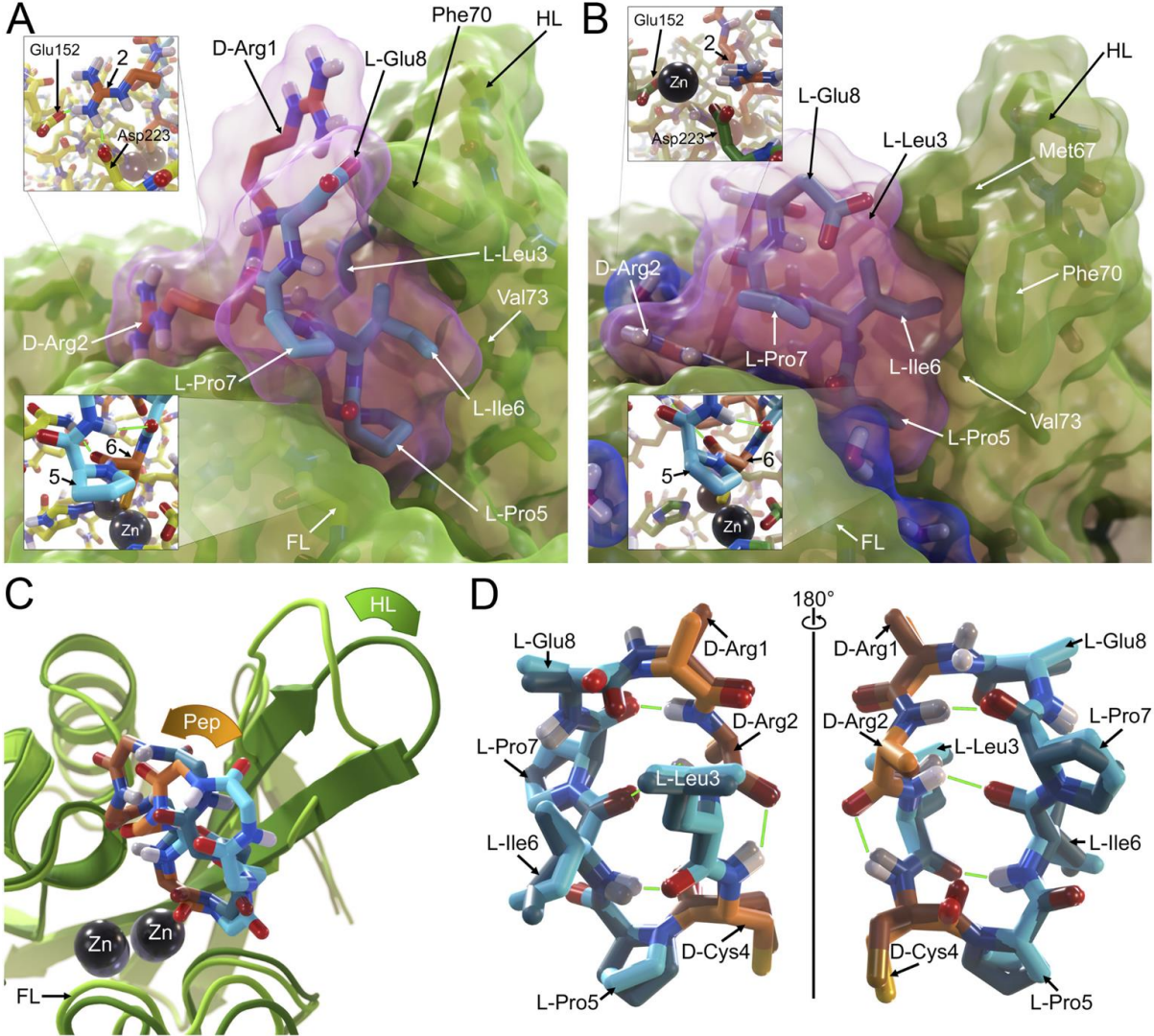


Peptide folding is more predictive of bioactivity





Designed peptide NDM1i-1G binds the enzyme pocket



Designed peptide NDM1i-3D contains NCAA