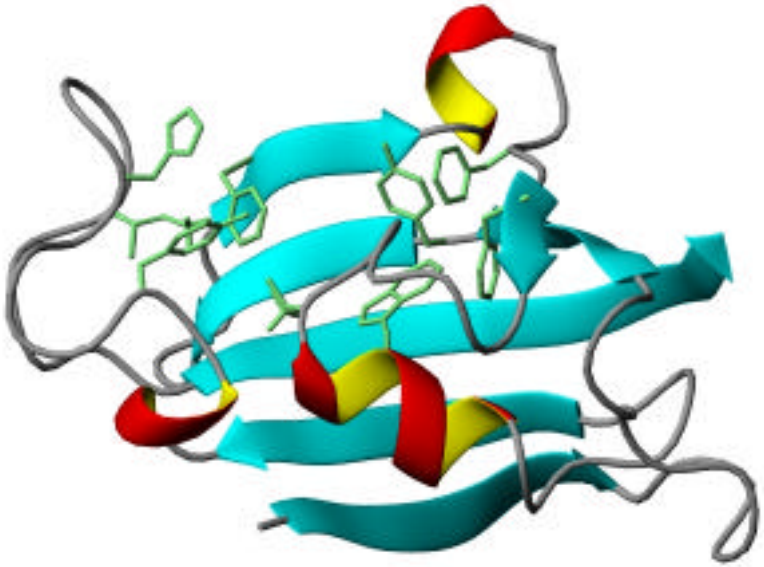
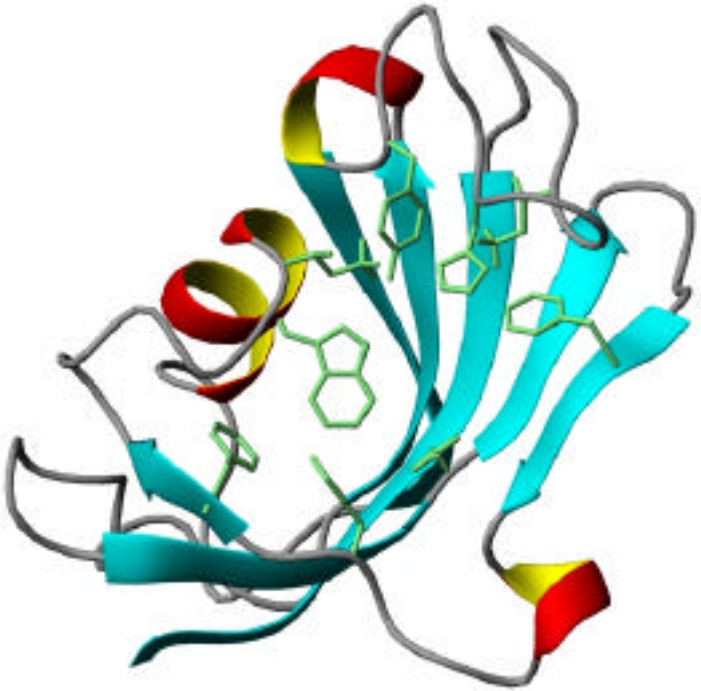
FKBP12 : FK506 Binding Protein



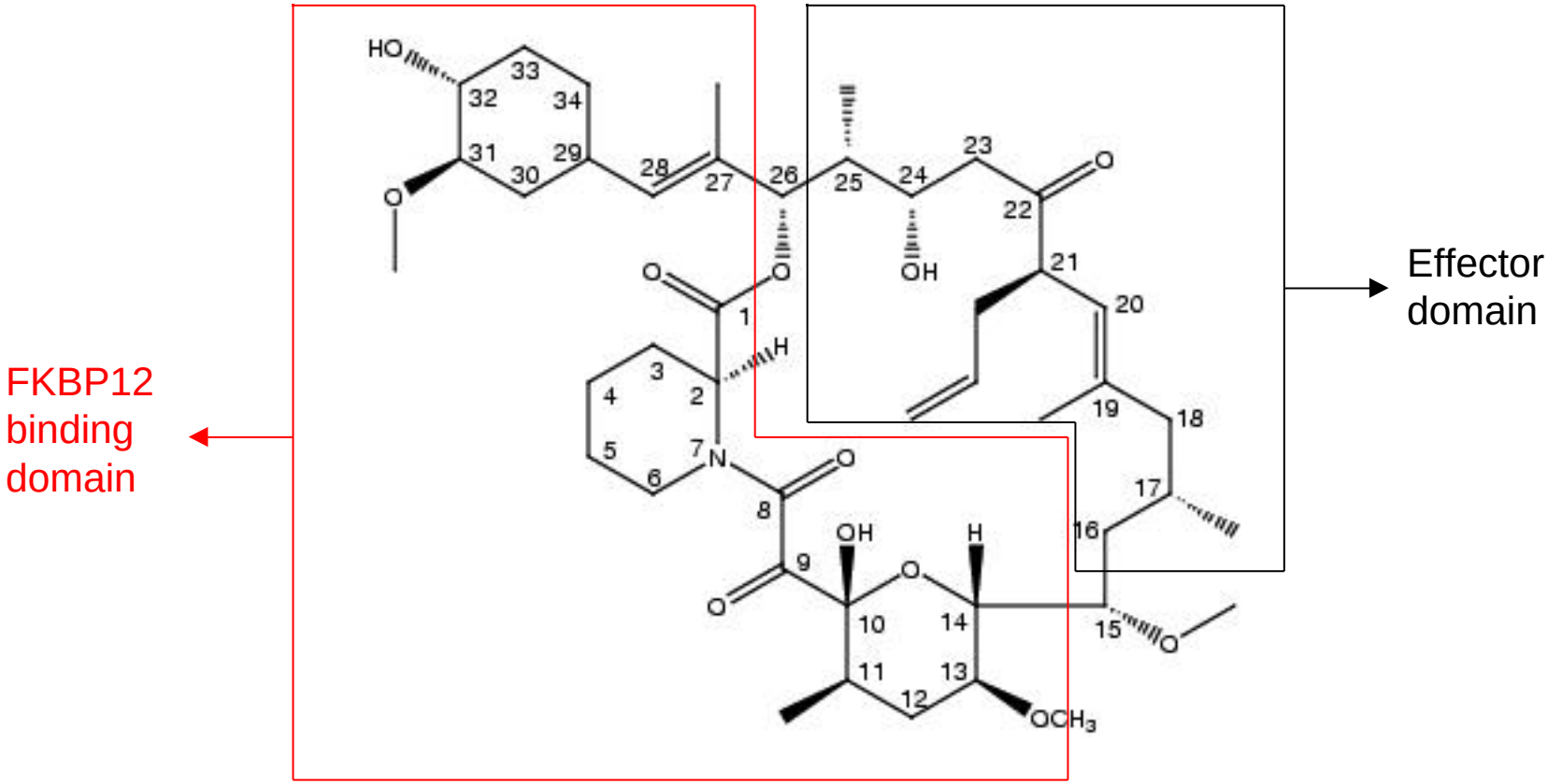


•Famille des immunophilines : récepteur des principales molécules Immunosupressantes (cyclosporine, rapamycine, FK506)
•Possède aussi une activité neurotrophe
•Protéine α/β, poche hydrophobe

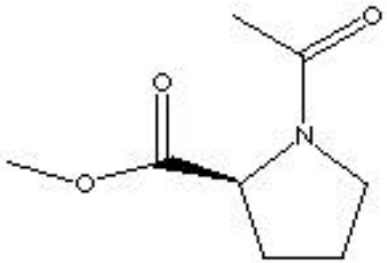
Babine, R.E., Bender, S.L. (1997) ***Chem. Rev.***, 97: 1359-1472 {1437}
Gold, B.G., (1997) ***Molecular Neurobiology,*** 15: 285-305

FK506 and FKBP12

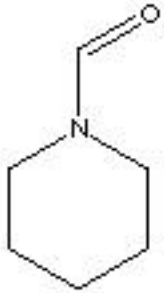
•K_d(FKBP12-FK506) = 0.4nM



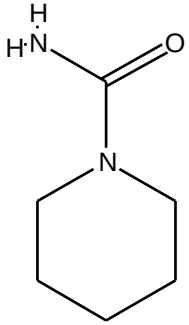
Groups



(2S)1-acetylproline methyl ester



1-formyl piperidine

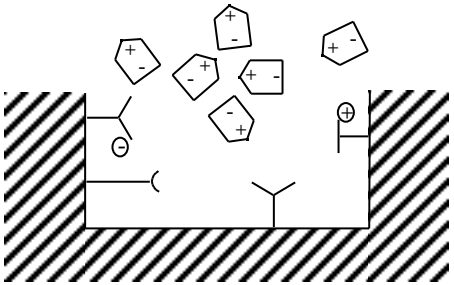


1-piperidine carboxamide

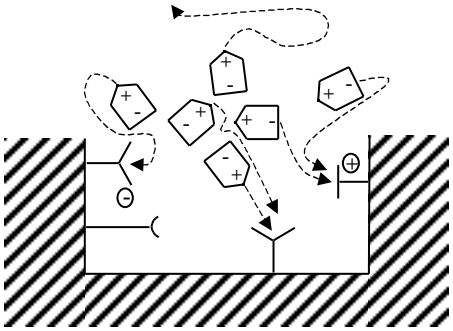
- Comparison with NMR experiments done by Dr. C. Sich and Dr. S. Improta at Sanofi~Synthélabo (Strasbourg)
  - Groups have common features with FK506
  - Commercially available
  - Expected to be weak but specific FKBP12 ligands
- Groups have been parametrized and inserted into MCSS fragments library

MCSS

500-5000 copies of the group are randomly placed in the pocket

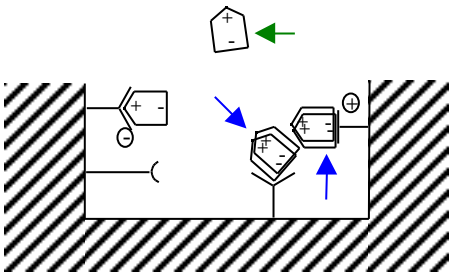


500 steps of steepest descent
500 steps of conjugate gradient

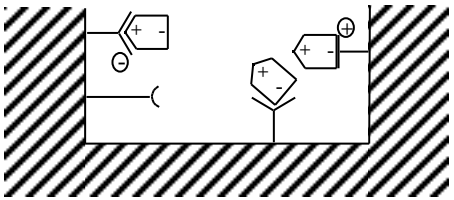


Duplicate elimination based on RMS
Non-interacting copies elimination based on $E_{interaction}$

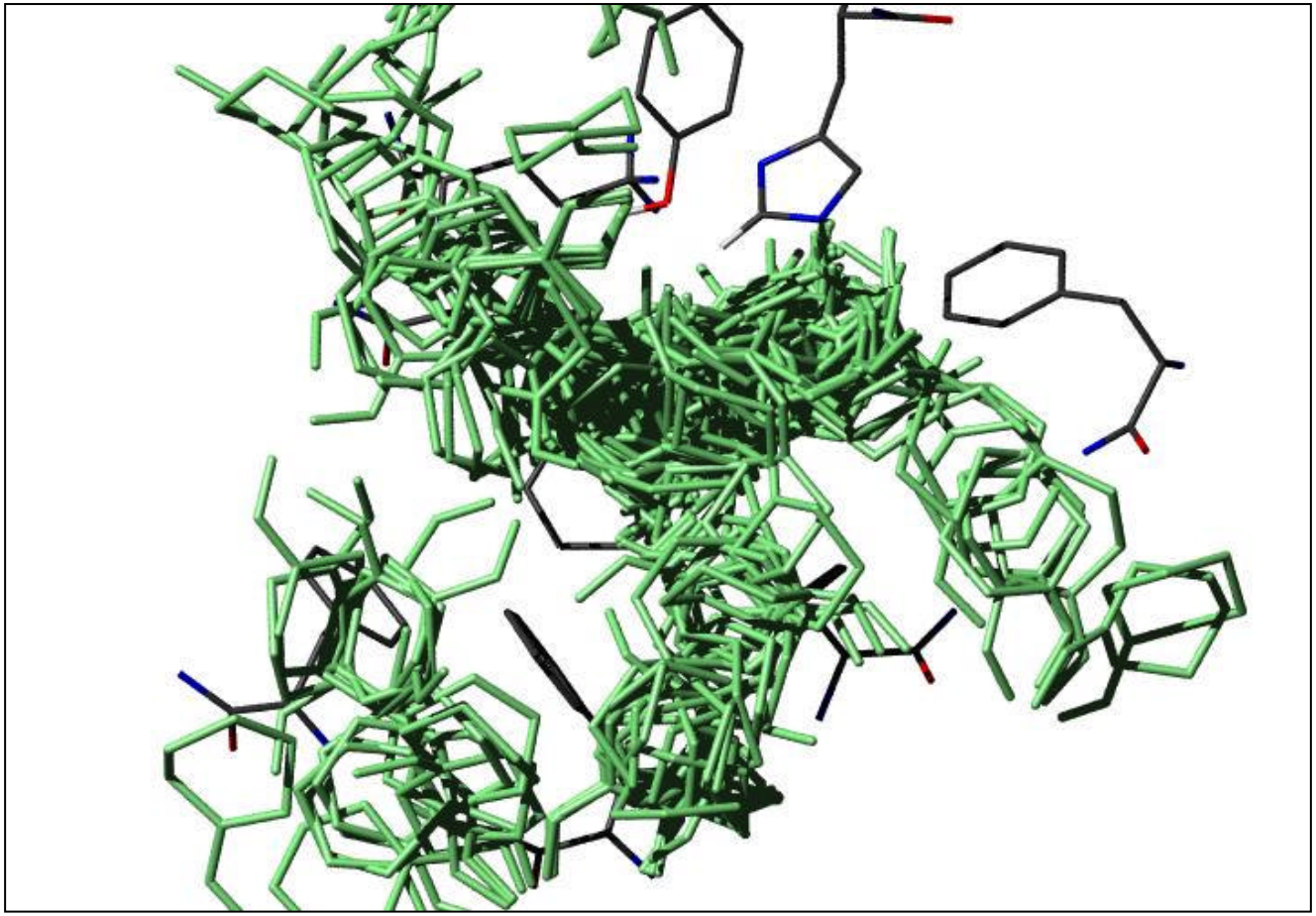
Iterations - New copies are placed an the pocket



Functionality map of binding for the group



Carte fonctionnelle MCSS



MCSS

✓Détermination ***exhaustive*** des positions et orientations

✓Repose sur CHARMM : << ***customisable*** >>

oPas de prise en compte du solvant

oBeaucoup de minima : analyse visuelle ardue

oBeaucoup d’opérations manuelles

Taking the solvent into account

•The solvent is treated as a continuum : Linear Poisson-Boltzmann

$$\Delta G_{binding} = \Delta E^{fragm} + \Delta E_{VdW}^{Interm} + \Delta G_{elec}^{Interm} + \Delta G_{ele\,desolv}^{protein} + \Delta G_{ele\,desolv}^{fragm} + \Delta G_{non\,polar}^{complex}$$

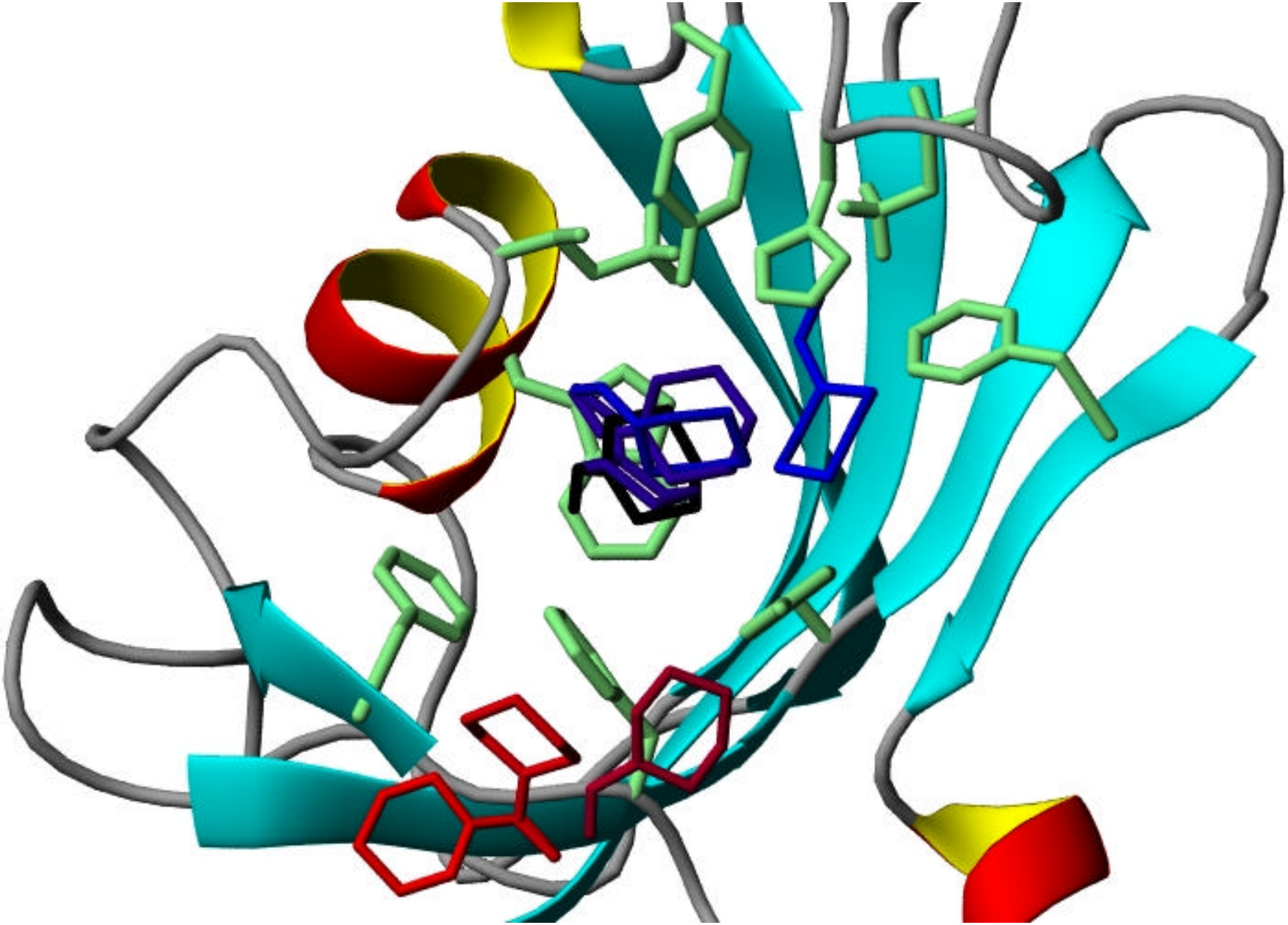
CHARMM ❶ UHBD(LPB) ❷ CHARMM ❸

❶ Internal energy of a minimum, group/protein Van der Waals interaction energy

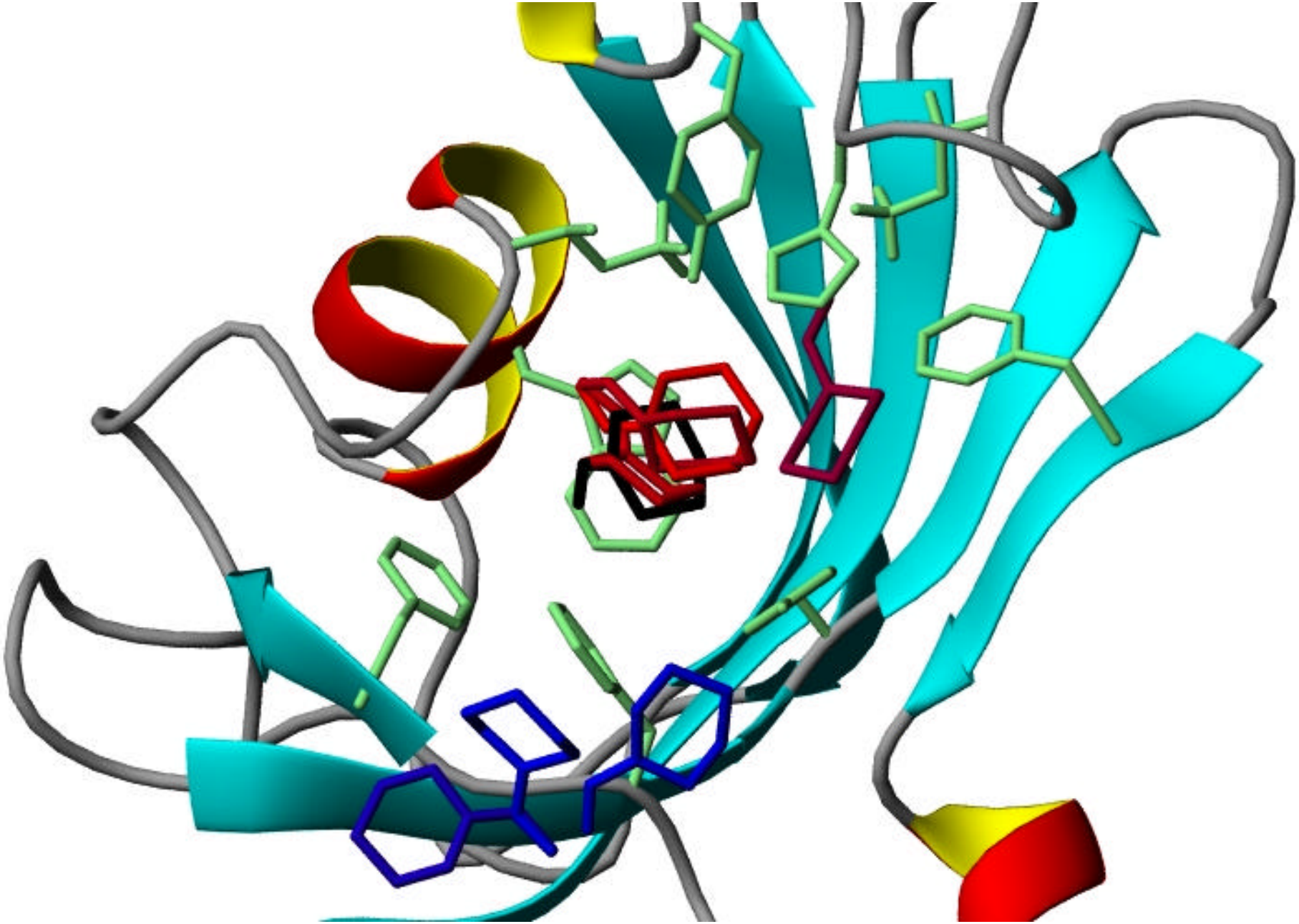
❷ Numerical resolution of finite differences LPB

❸ Non-polar interaction energy is proportional to the loss in solvent accessible surface (SAS)

SANS SOLVATATION

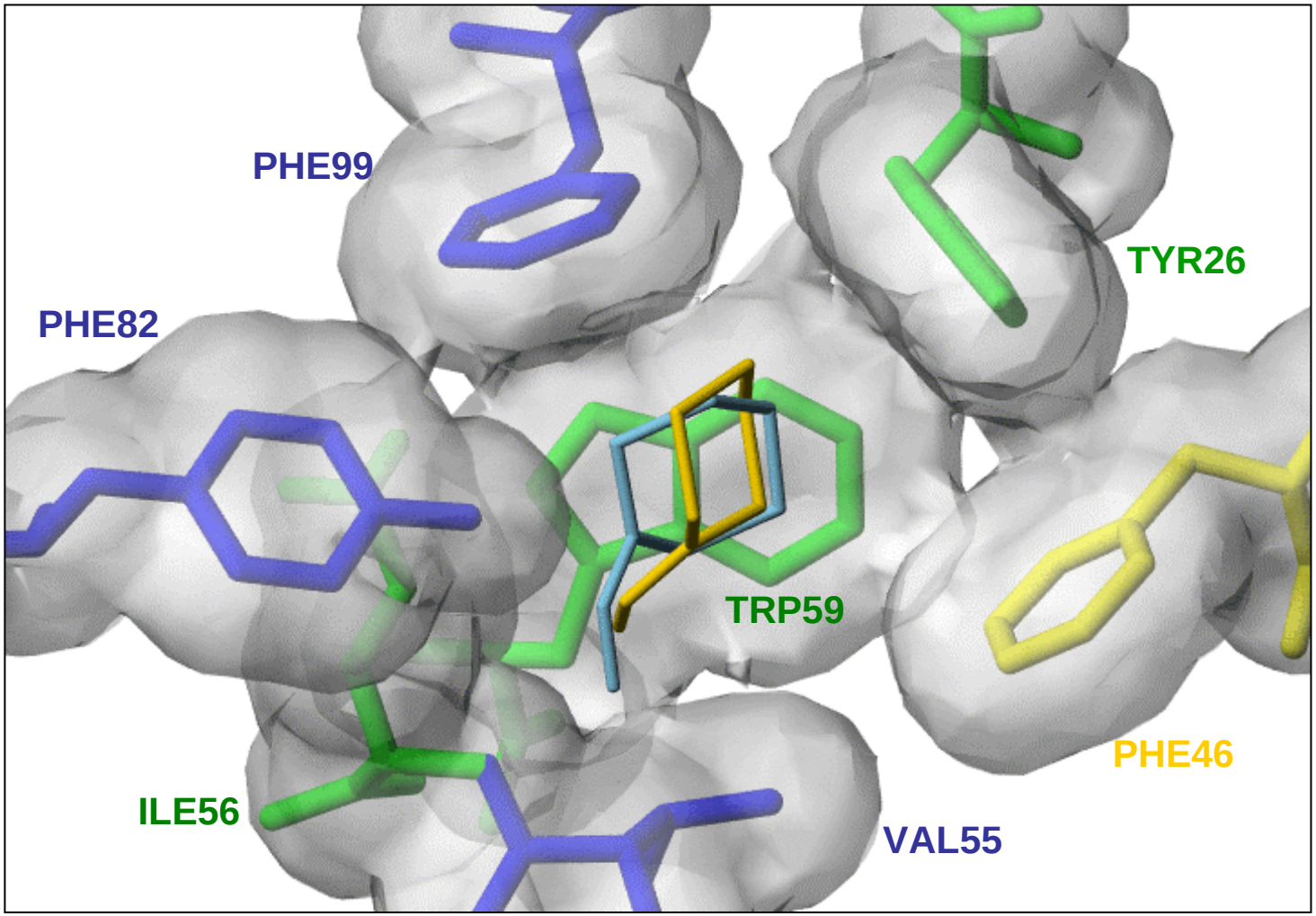


AVEC SOLVATATION



CLUSVDW

- Clustering based on protein/group Van der Waals contacts
- Each fragment is identified by a Van der Waals“fingerprint”
- Complete linkage algorithm

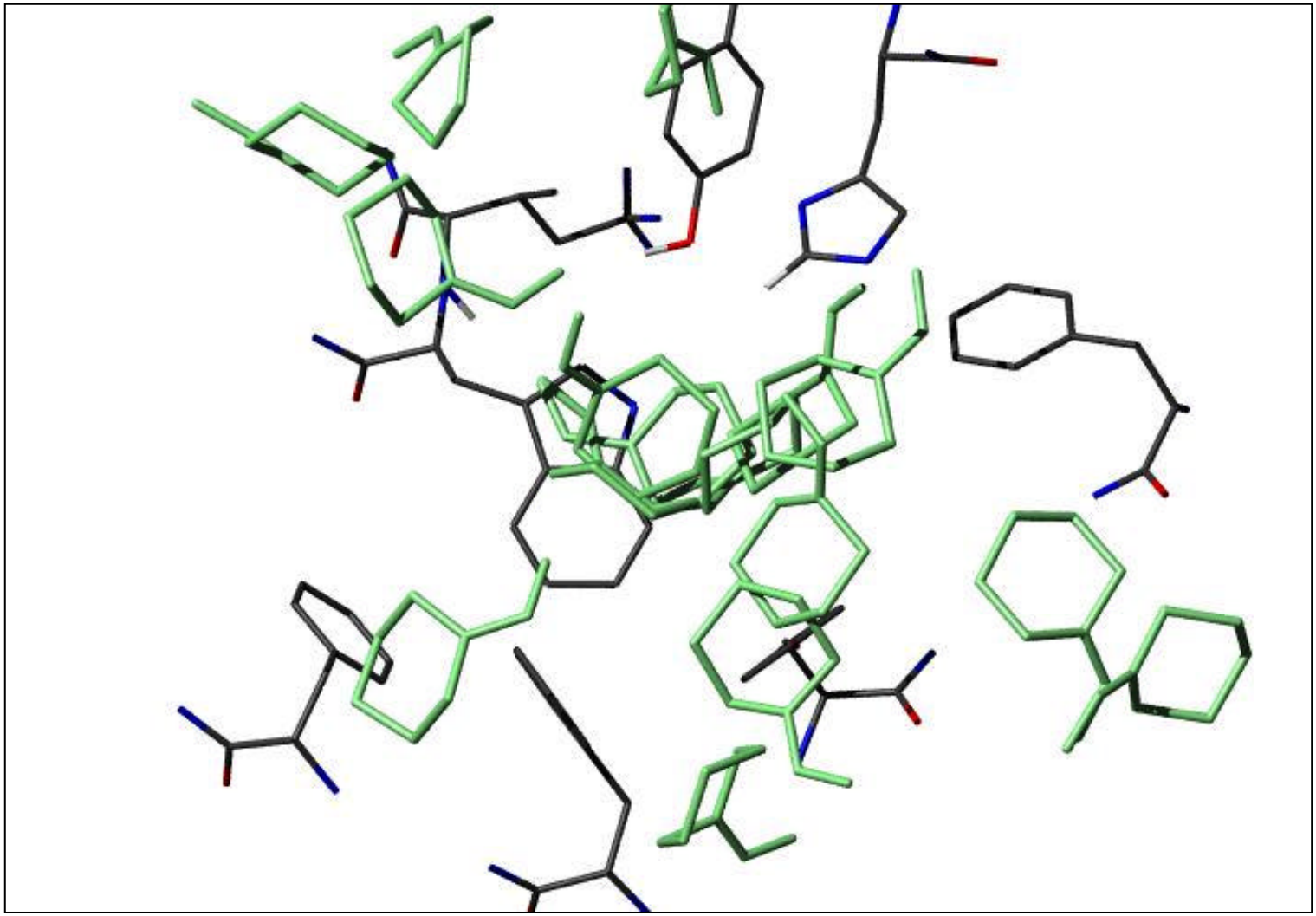


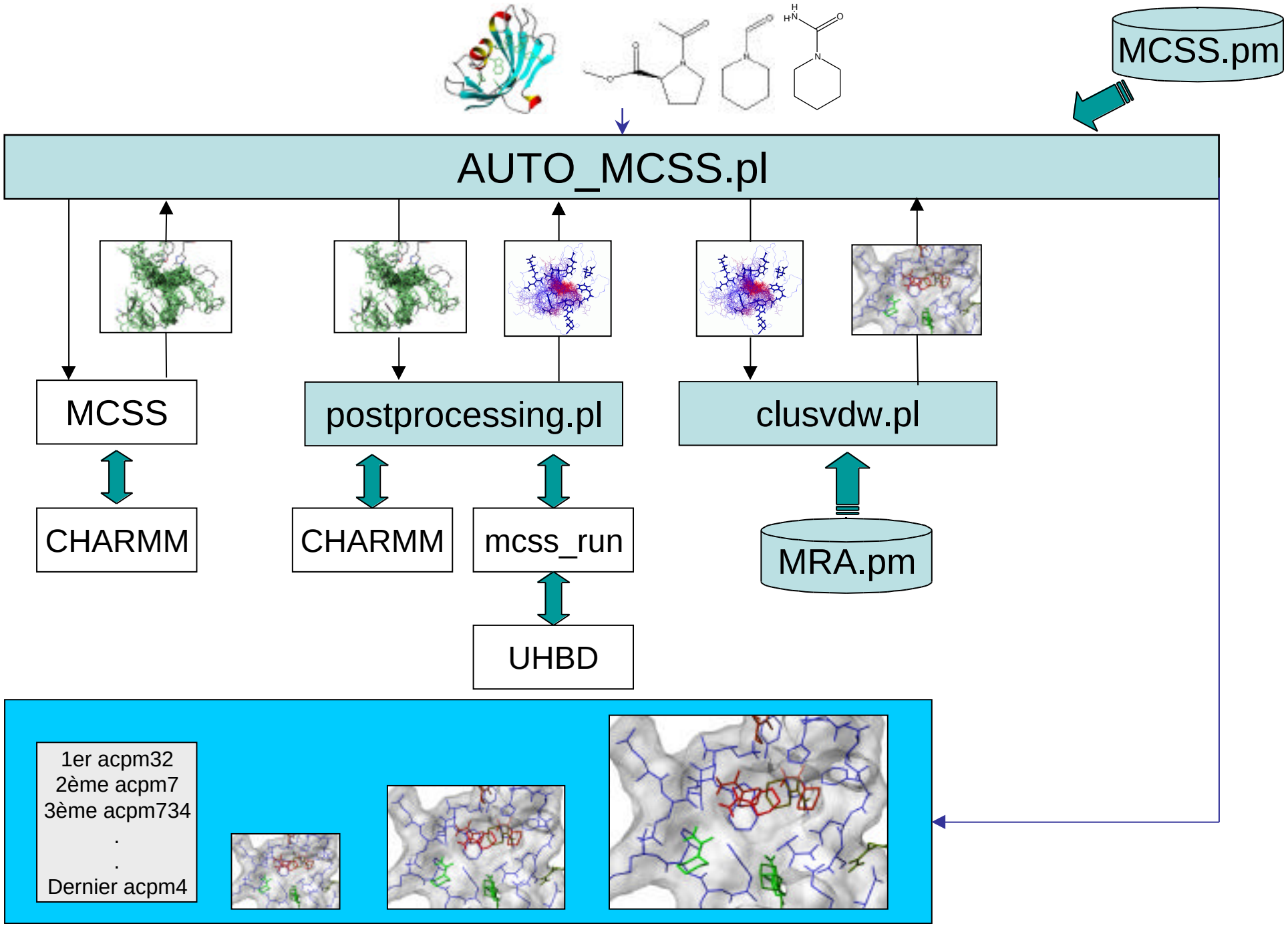
Minimum 6
TYR26 PHE46
ILE56 TRP59

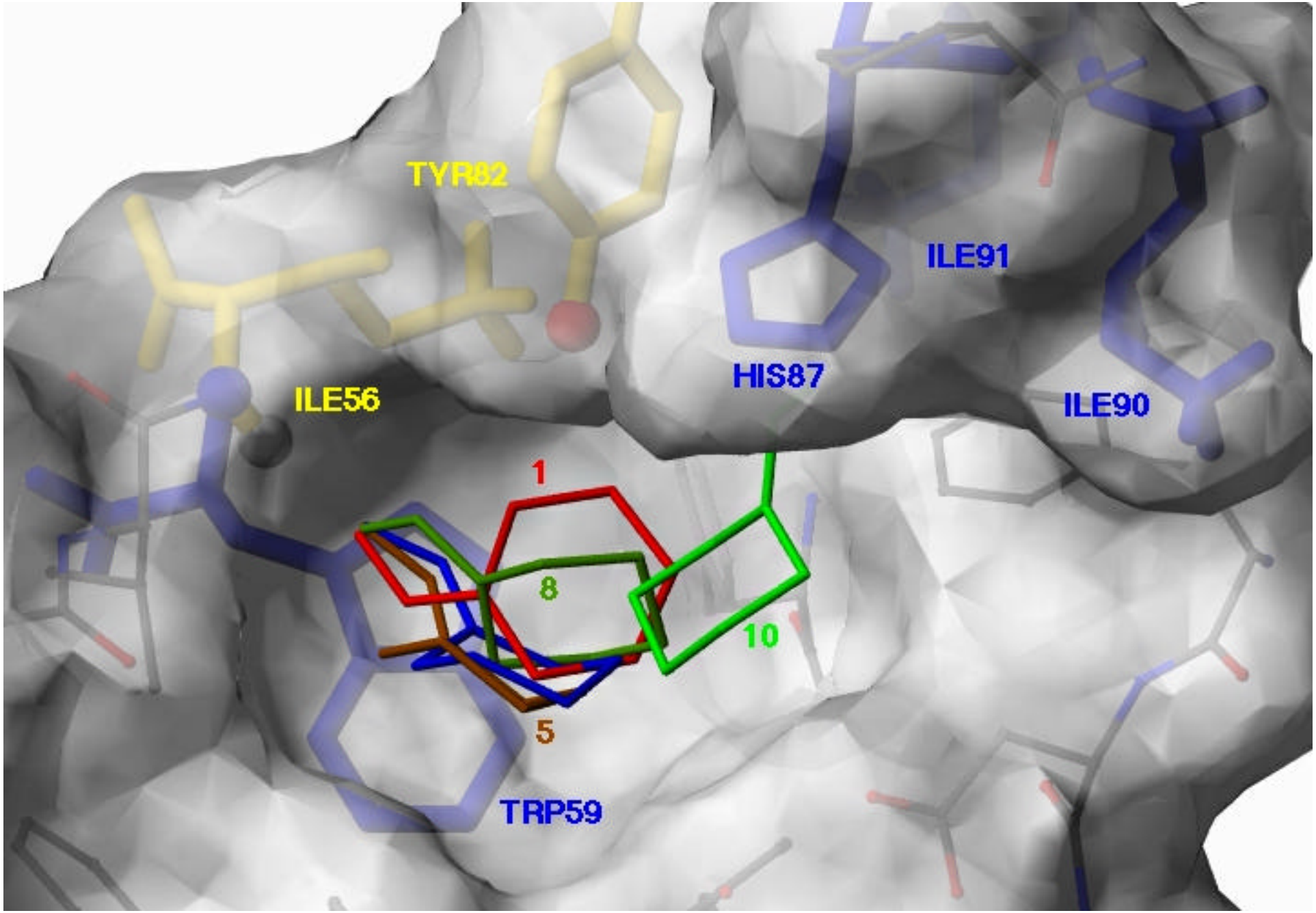
Minimum 51
TYR26 VAL55
ILE56 TRP59
TYR82 PHE99

Clusters

- Only one minimum is used to represent a given cluster
- Binding mode diversity information is visible




MCSS.pm
AUTO_MCSS.pl
MCSS
postprocessing.pl
clusvdw.pl
CHARMM
CHARMM
mcss_run
MRA.pm
UHBD
1er acpm32
2ème acpm7
3ème acpm734
.
.
Dernier acpm4


TYR82
ILE91
HIS87
ILE56
ILE90
1
8
10
5
TRP59

- Comparaison MCSS / RMN menée indépendemment
- Le placement de petit groupe est efficace grâce à la prise en compte du solvant (postprocessing)
- Le clustering permet de visualiser facilement les principaux modes de liaison du groupe chimique
- La procédure est complètement automatisée tout en permettant une souplesse de choix de paramètres pour l'utilisateur
- Auto_mcss.pl peut piloter plusieurs machines à un ou plusieurs processeurs

F. Sirockin

C. Sich

S. Improta

V. Saudek

N. Froloff

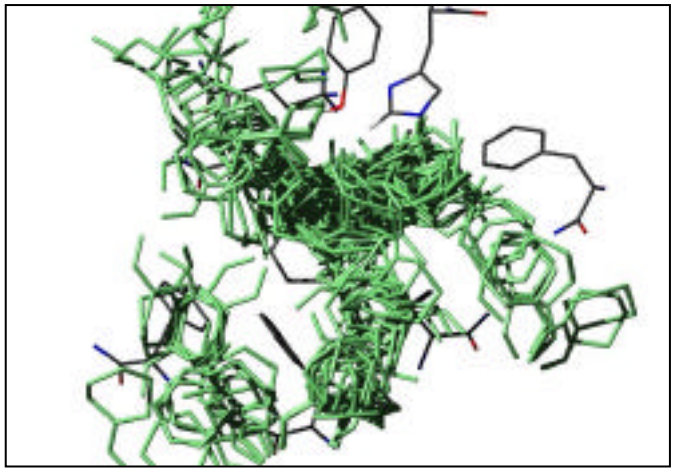
M. Karplus

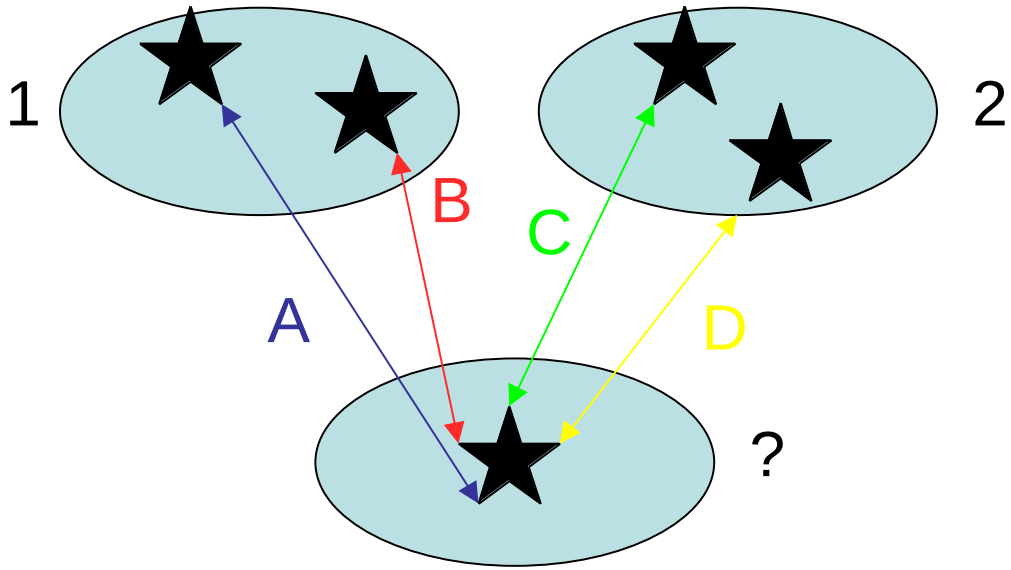
M. Schaefer

V. Lafont

M. Schechner

Clustering





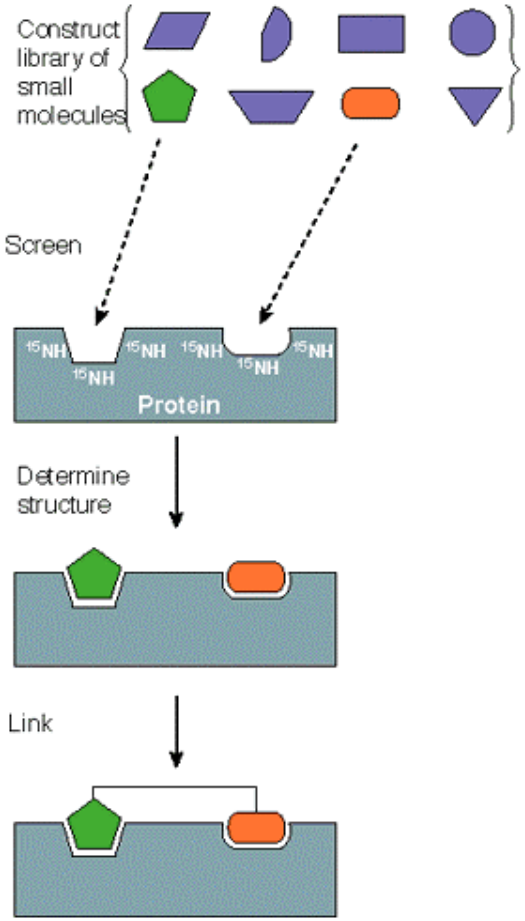
Clustering algorithm:

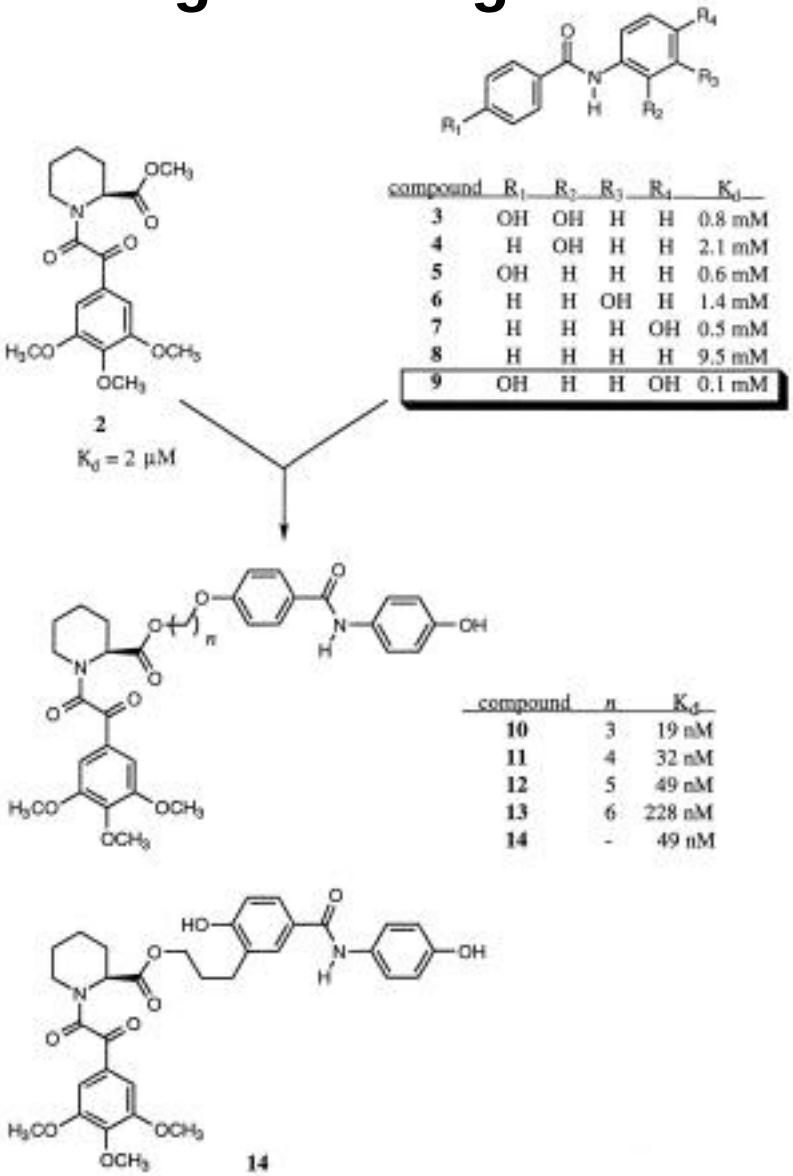
•Proximity index (similarity or dissimilarity)
eg: euclidian distance is a dissimilarity index

•Grouping method
•Single linkage -> goes in 2
•Complete linkage -> goes in 1

A=2
B=3
C=5
D=1

Stepwise approach for ligand design





compound	R_1	R_2	R_3	R_4	K_d
3	OH	OH	H	H	0.8 mM
4	H	OH	H	H	2.1 mM
5	OH	H	H	H	0.6 mM
6	H	H	OH	H	1.4 mM
7	H	H	H	OH	0.5 mM
8	H	H	H	H	9.5 mM
9	OH	H	H	OH	0.1 mM

compound	n	K_d
10	3	19 nM
11	4	32 nM
12	5	49 nM
13	6	228 nM
14	-	49 nM

Shuker, S.B., Hajduk, P.J., Meadows, R.P., Fesik, S.W., (1996) ***Science*** 274: 1531-1534