Applying (Mostly QM) for Rational Drug Design

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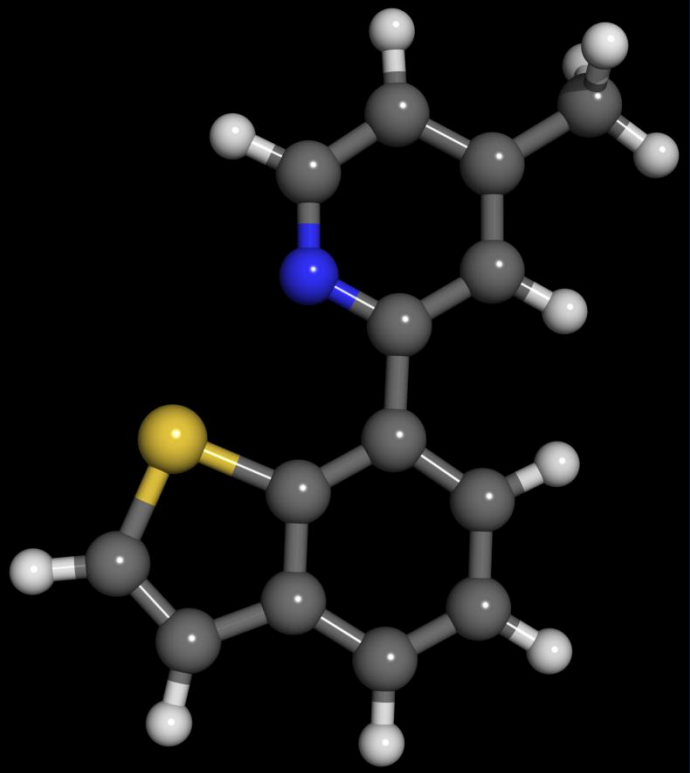
Part 1:

lp → σ* Driven

Conformational Effects in

GKRP Inhibition (T2D)

7-Substituted Benzothiophene: $lp_{(N)} \rightarrow \sigma^*_{(S\text{-}C)}$ and Effect on Conformation



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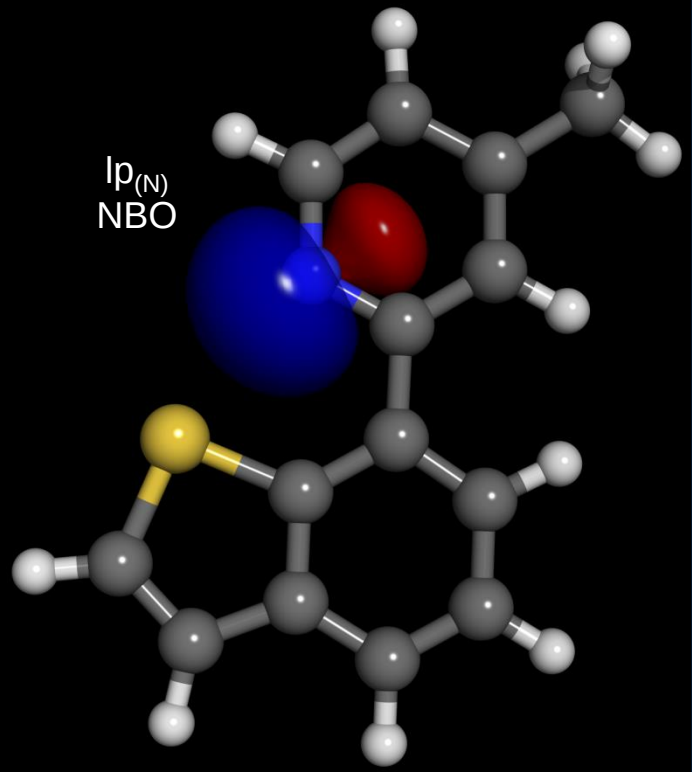

(M06-2X / 6-31+G(d,p))
NBO 6.0

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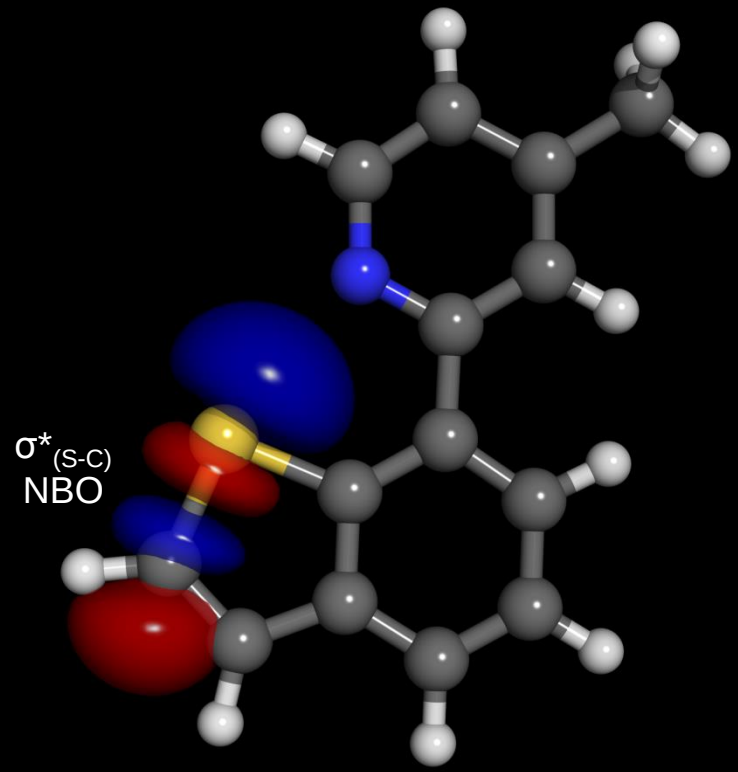

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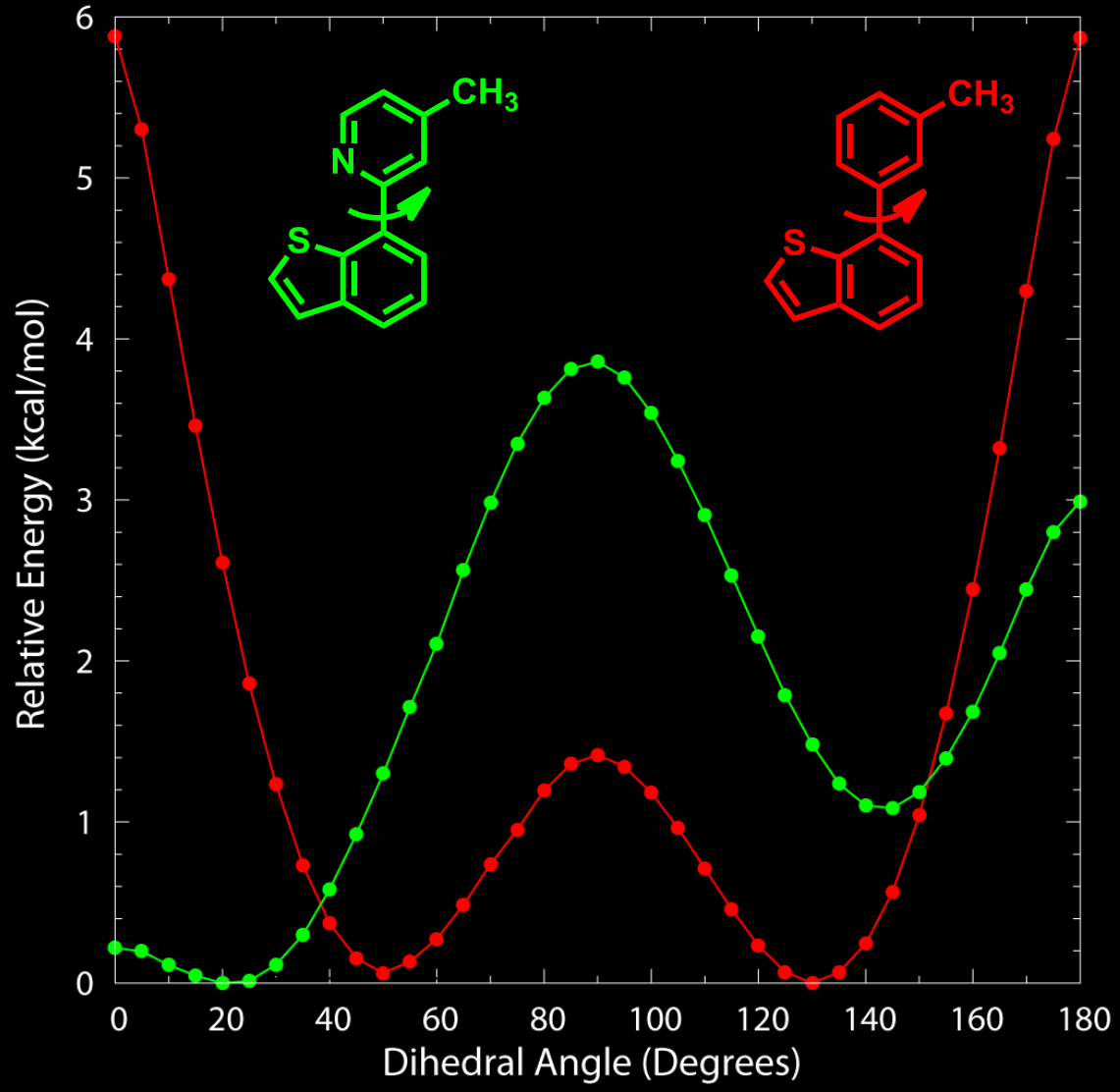
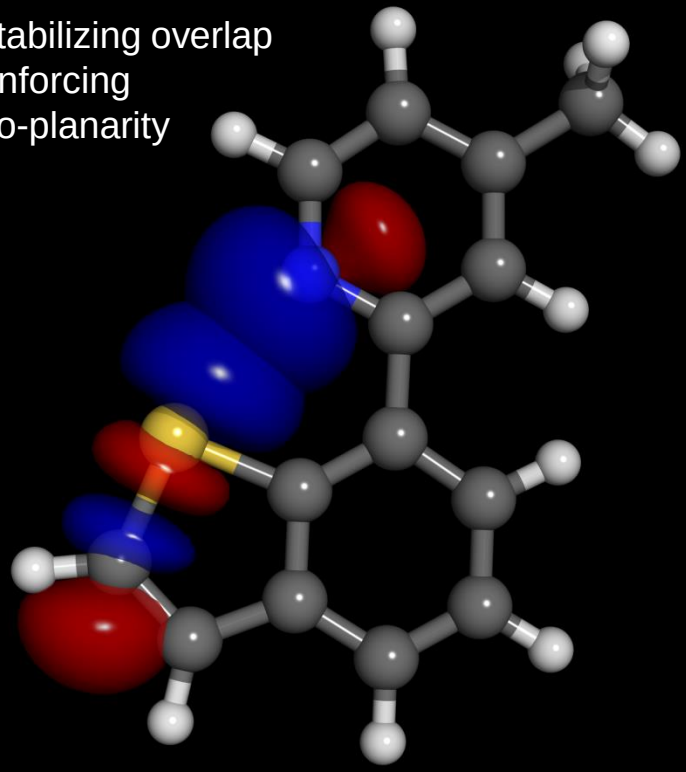


stabilizing overlap enforcing co-planarity



(M06-2X / 6-31+G(d,p))
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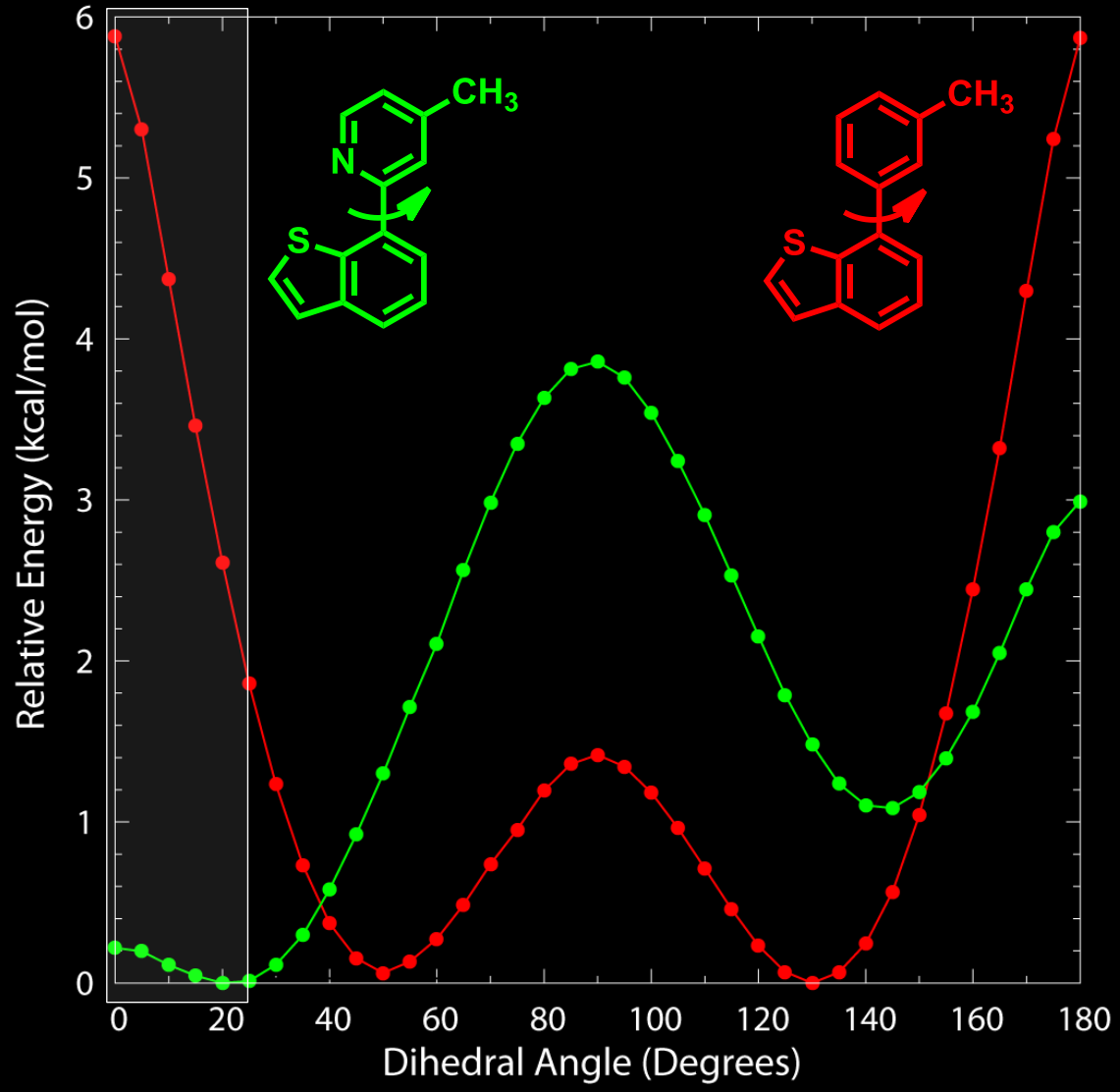
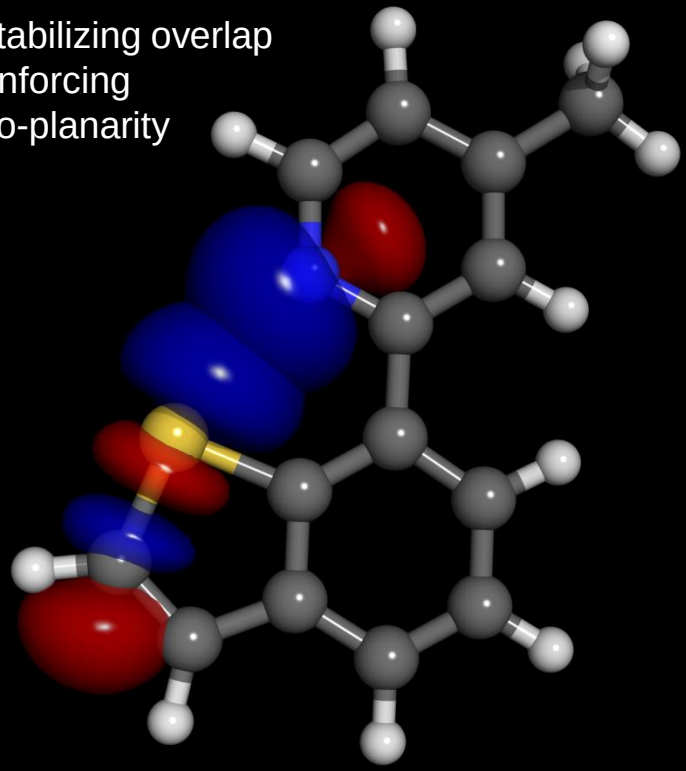


stabilizing overlap enforcing co-planarity



(M06-2X / 6-31+G(d,p))
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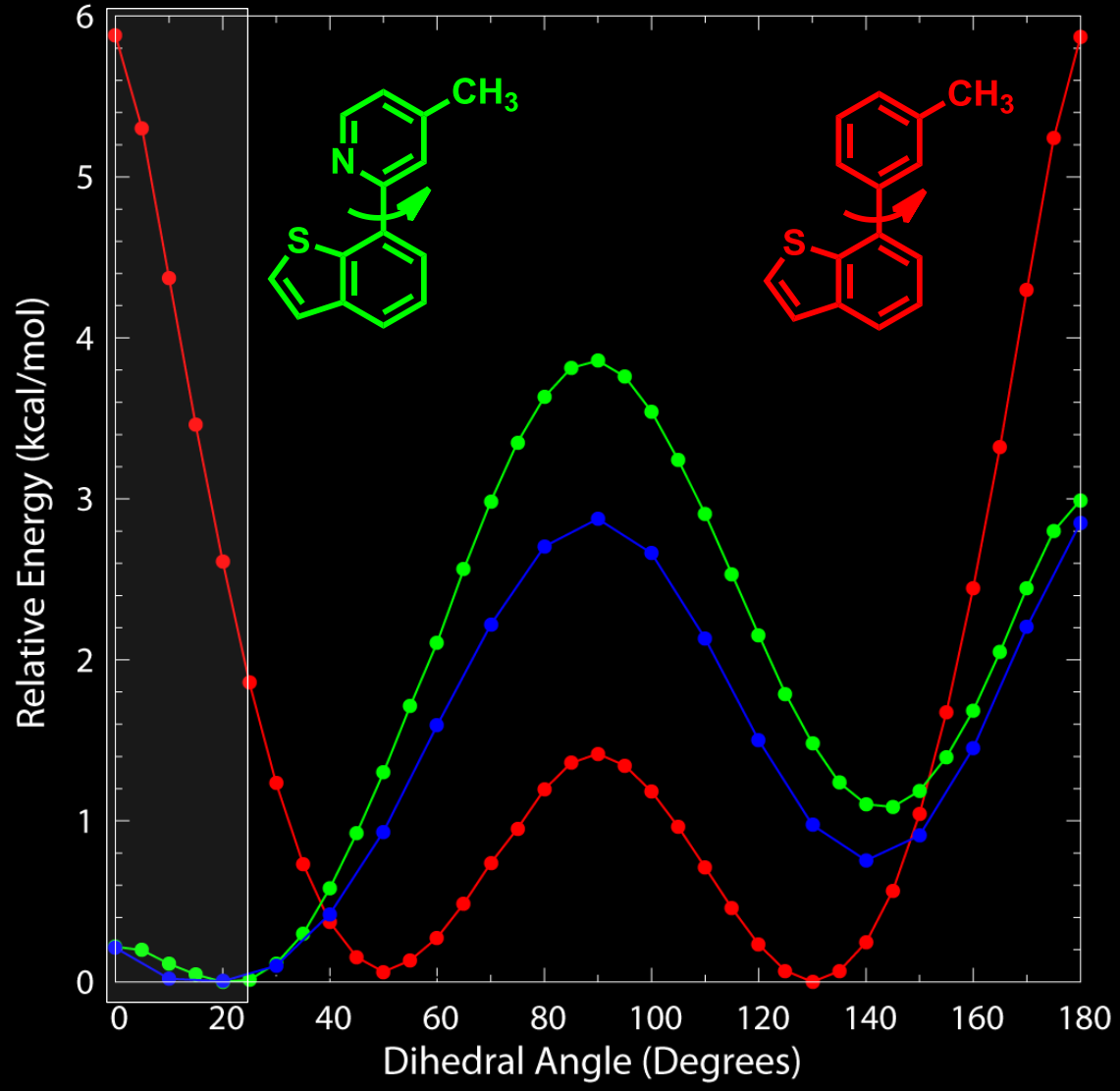
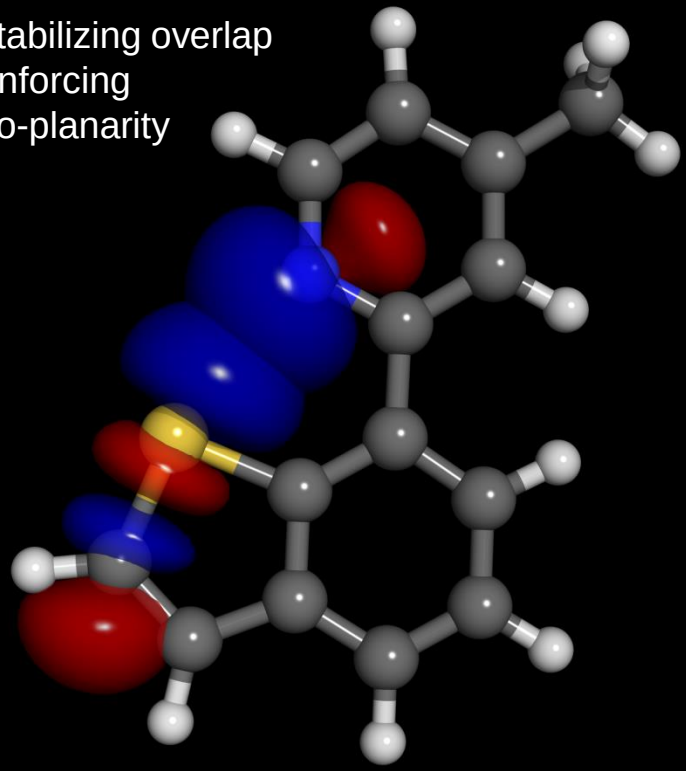
7-Substituted Benzothiophene: $lp_{(N)} \rightarrow \sigma^*_{(S\text{-}C)}$ and Effect on Conformation



stabilizing overlap enforcing co-planarity



(CCSD(T) / aug-cc-PVDZ // M06-2X / 6-31+G(d,p))

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Aside: Conformational Characteristics of Sulfur Heterocycles

Journal of Medicinal Chemistry

Perspective

pubs.acs.org/jmc

A Survey of the Role of Noncovalent Sulfur Interactions in Drug Design

Brett R. Beno,[†,∥] Kap-Sun Yeung,[‡,∥] Michael D. Bartberger,[§] Lewis D. Pennington,[§] and Nicholas A. Meanwell*[,‡]

J. Med. Chem. **2015**, *58*, 4383.

IN THE PIPELINE

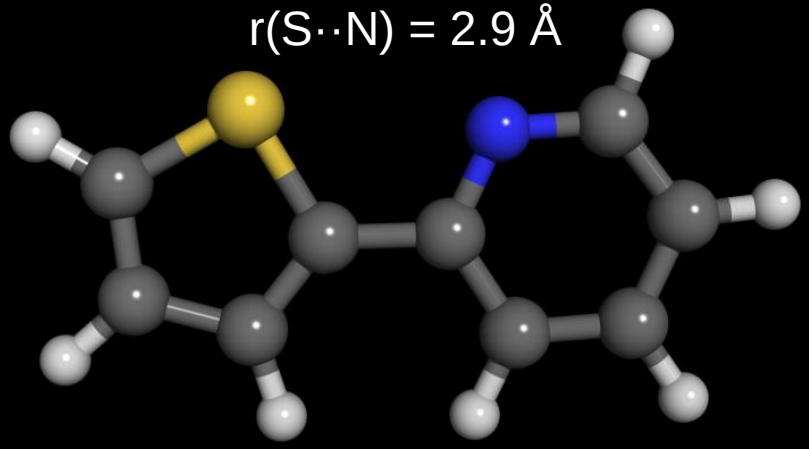
Derek Lowe's commentary on drug discovery and the pharma industry. An editorially independent blog from the publishers of *Science Translational Medicine*.

By Derek Lowe

Our Friend the Sulfur Atom

By Derek Lowe | March 4, 2015

r(S··N) = 2.9 Å



In the blogosphere:
http://blogs.sciencemag.org/pipeline/archives/2015/03/04/our_friend_the_sulfur_atom

	vdW radius (Å)	∑ X + S (Å)
S	1.80	
O	1.52	O + S: 3.32
N	1.55	N + S: 3.35

X-ray of 2-(2'-thienyl)pyridine archetype:

Acta Crystallogr. C: Cryst. Struct. Commun. **1993**, *C49*, 1031.

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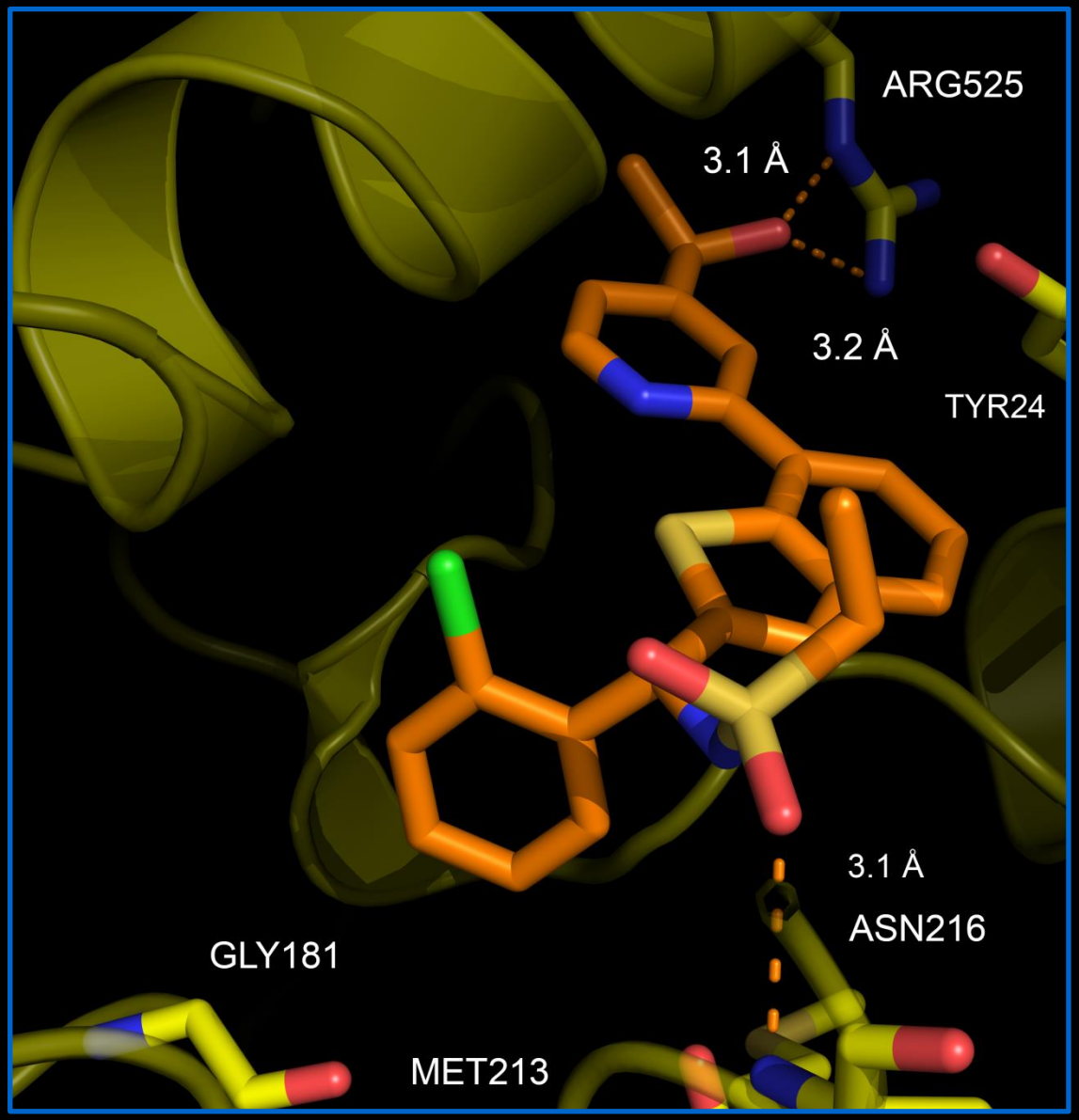
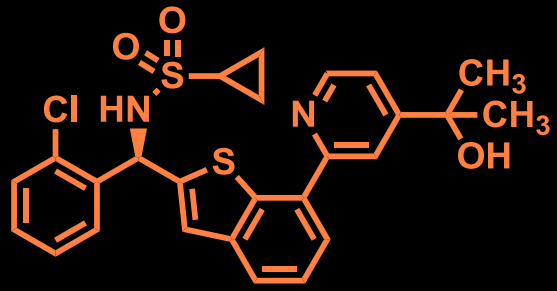
Structure of AMG-0265 in GKRP Confirms ARG525 Contact





AMG-0265
GK–GKRP IC_{50} = 0.11 μM
RLM CL_{int} = 34
cLogP = 4.8

co-crystal structure of AMG-0265 bound to GKRP (front view)

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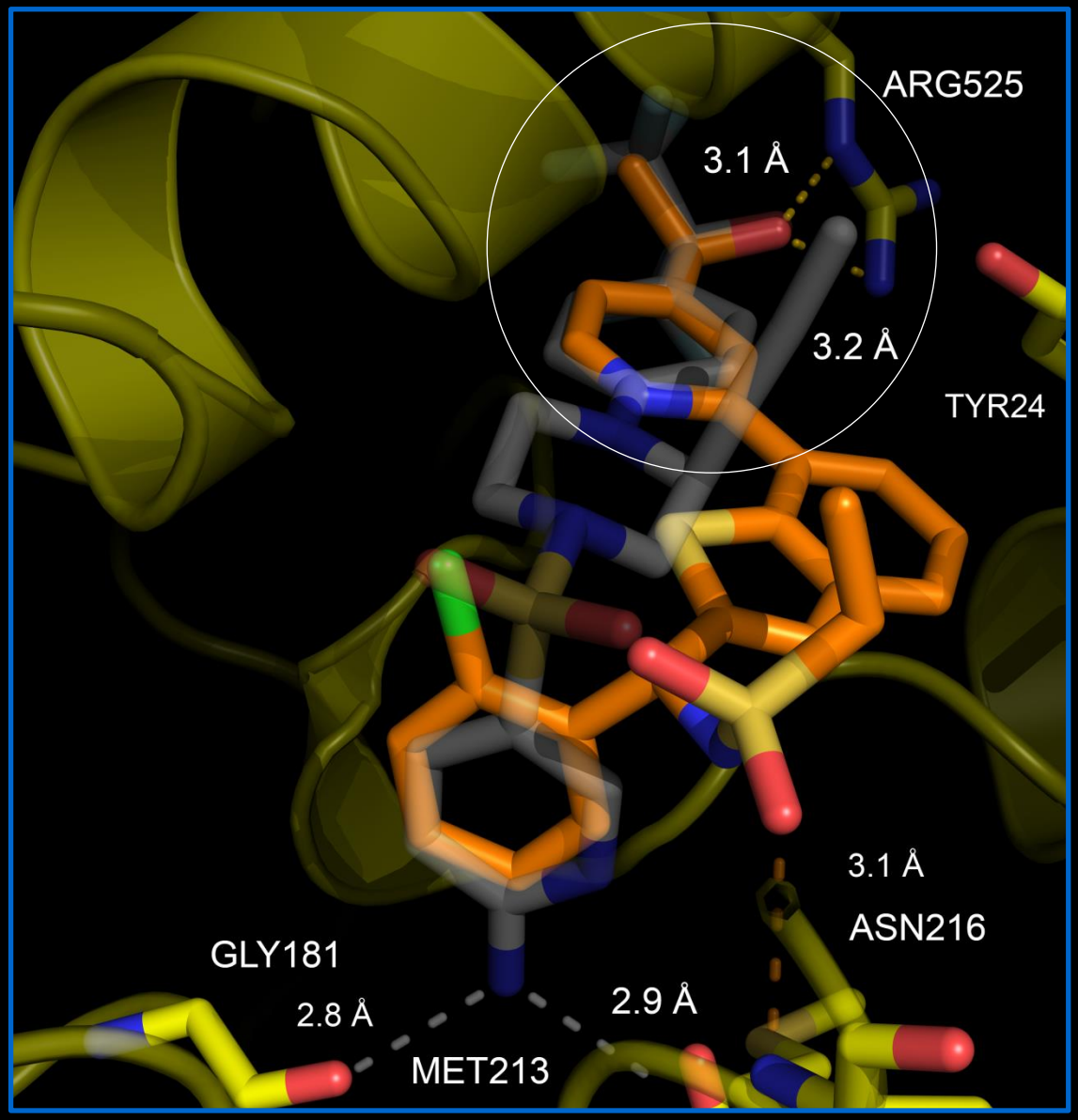
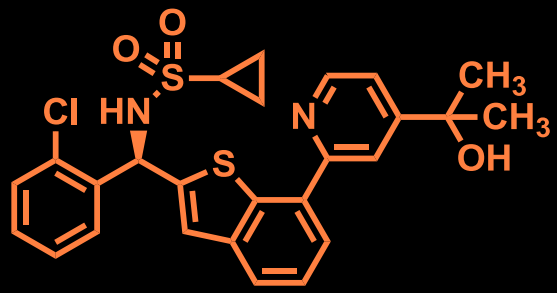
Structure of AMG-0265 in GKRP Confirms ARG525 Contact





AMG-0265
GK–GKRP IC_{50} = 0.11 μM
RLM CL_{int} = 34
cLogP = 4.8

near-perfect overlap of ARG525-engaging ring and carbinol moiety

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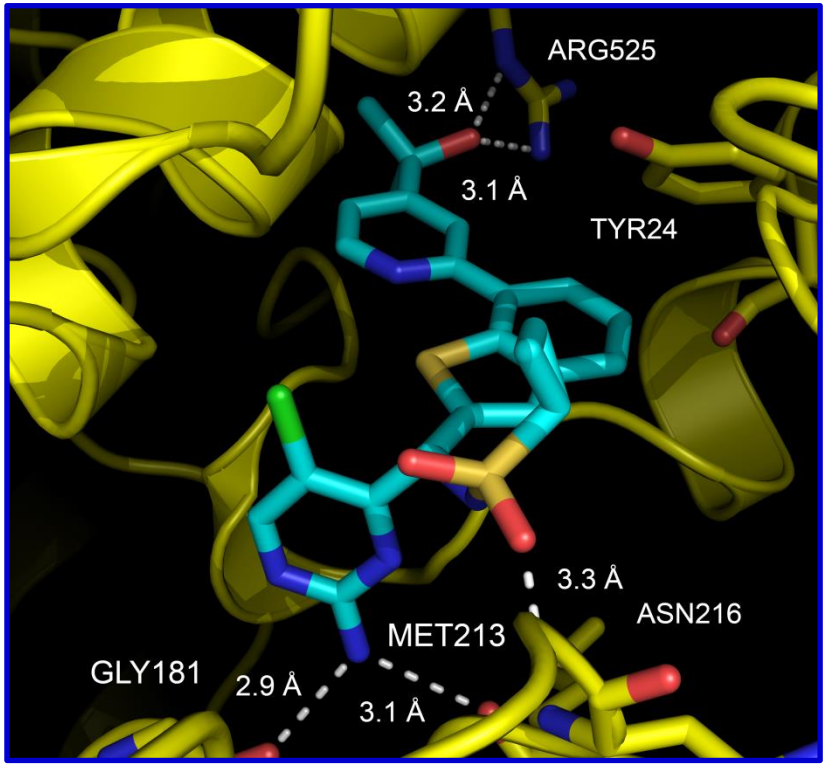
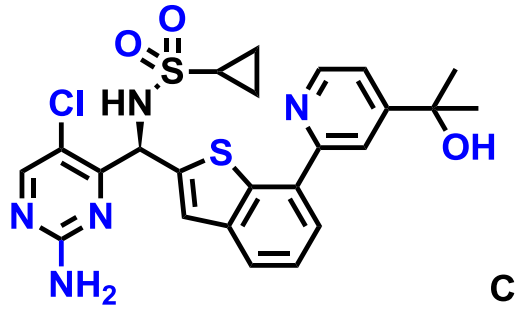
AMG-7549: PK, PD, and Efficacy





Rat PK in vivo

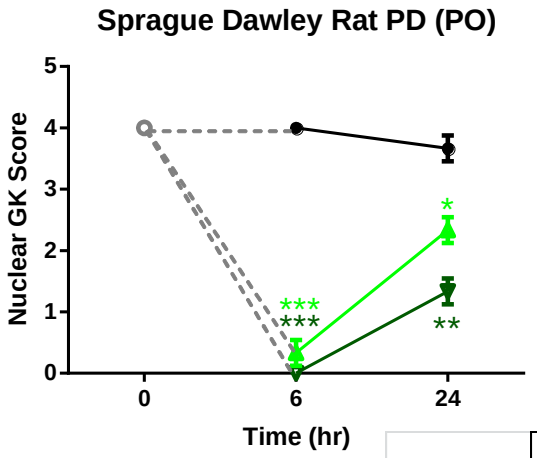
CL = 0.3 L/h/kg

V_{dss} = 0.3 L/kg

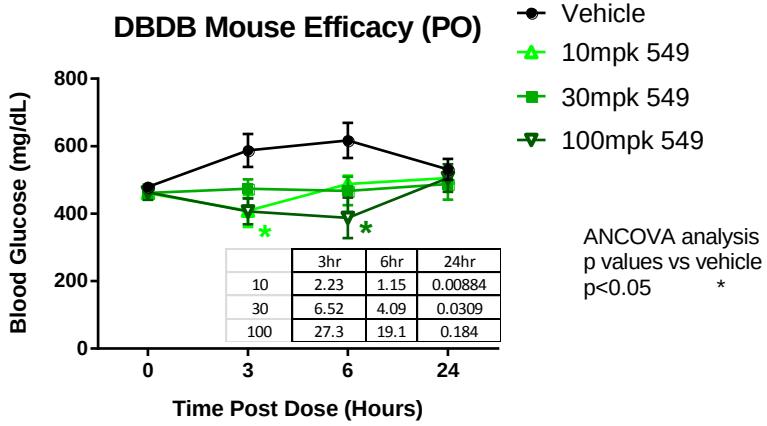
%*F* = 25

plasma fu = 0.01

CMax = 1.8 µM (4.7 hr)



	6hr	24hr
10	2.880	0.198
100	23.300	0.212



	3hr	6hr	24hr
10	2.23	1.15	0.00884
30	6.52	4.09	0.0309
100	27.3	19.1	0.184

J. Med. Chem. **2015**, *58*, 9663. *(ACS Editor's Choice)*

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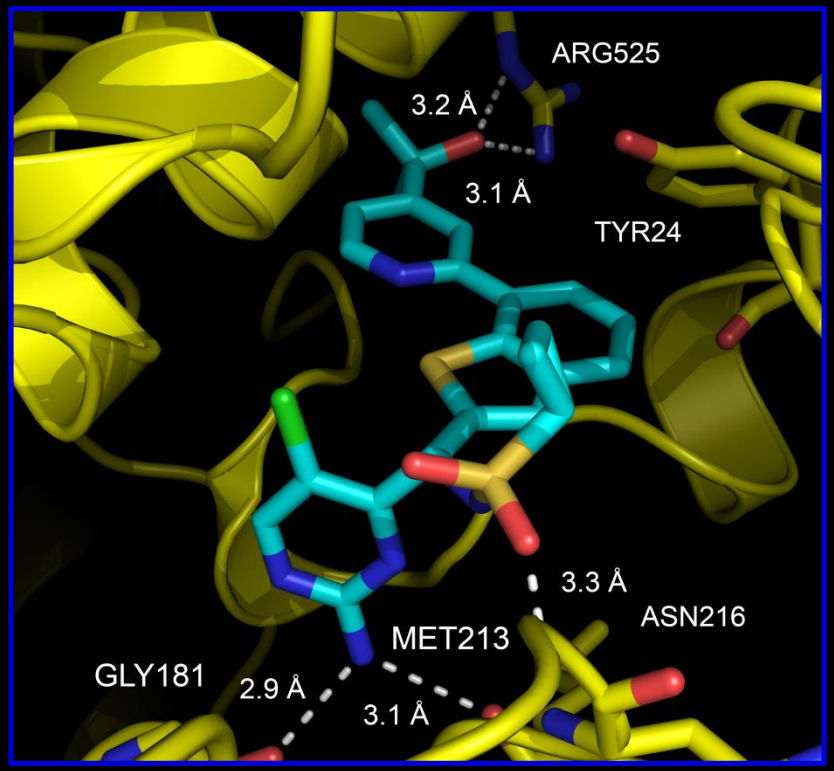
AMG-7549: PK, PD, and Efficacy



J. Med. Chem. **2015**, *58*, 9663. *(ACS Editor's Choice)*

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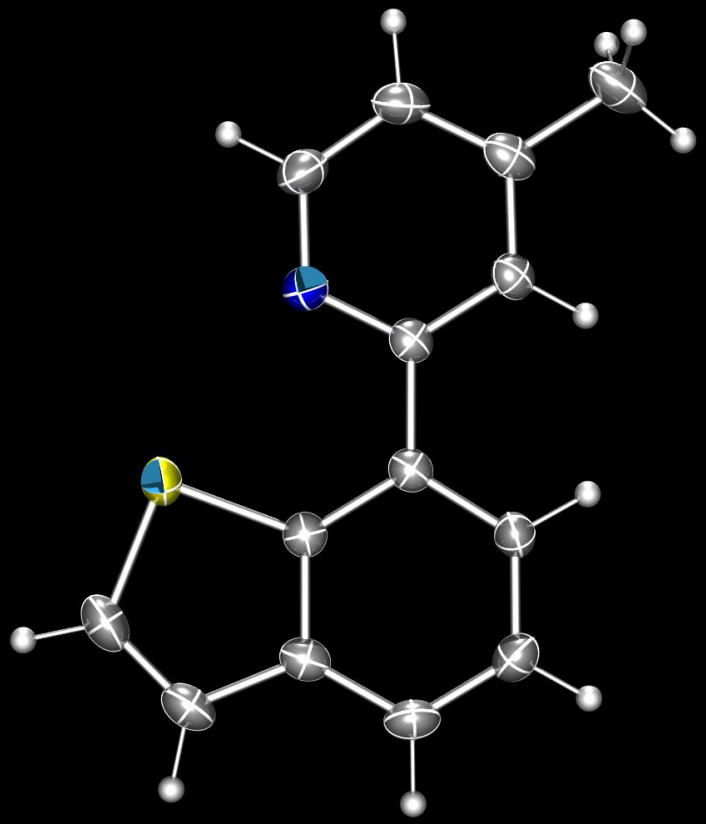
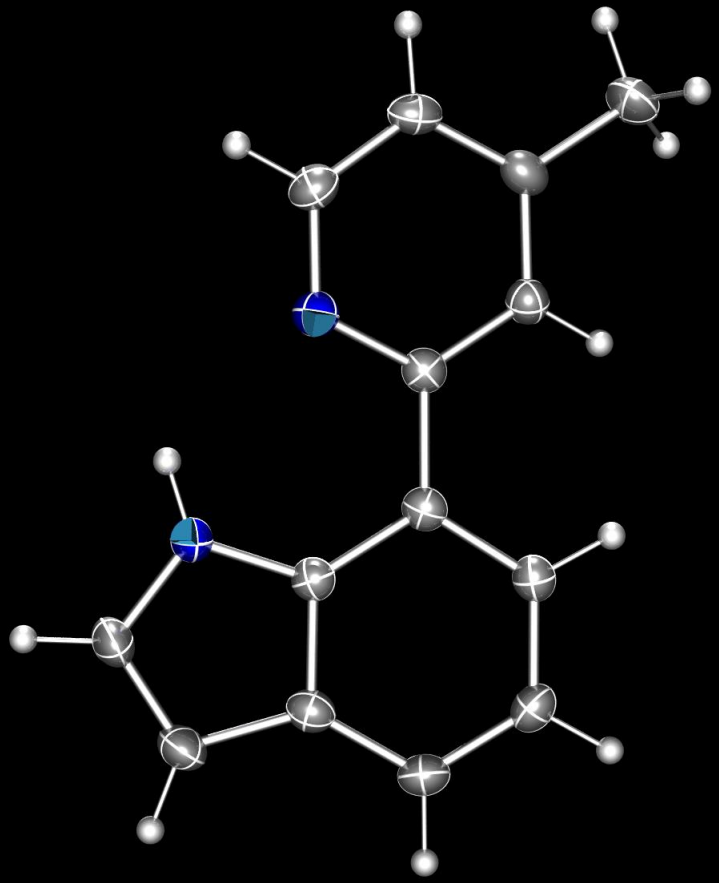
Single Crystal Structures Also Corroborate Prediction



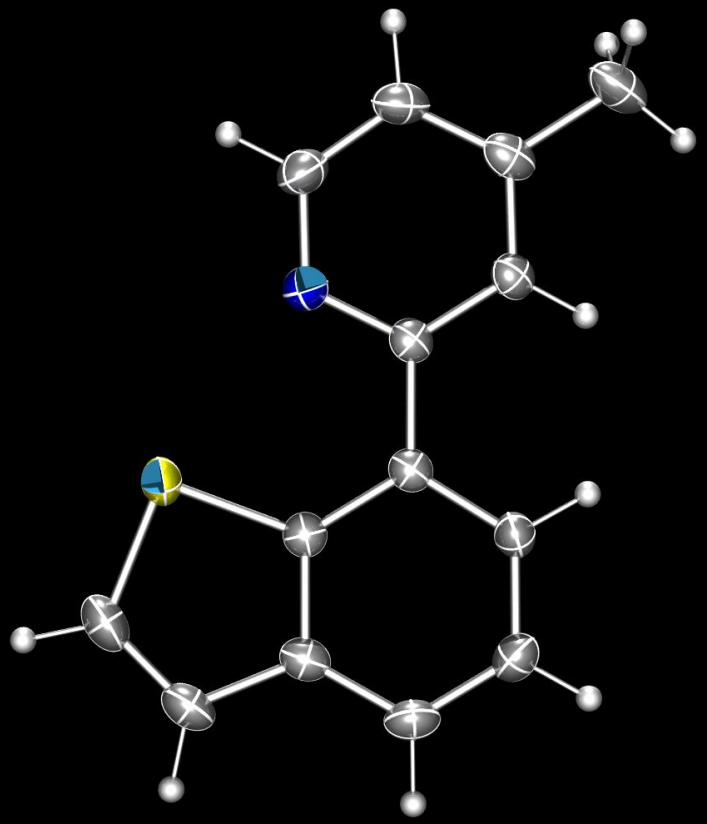
benzothiophene



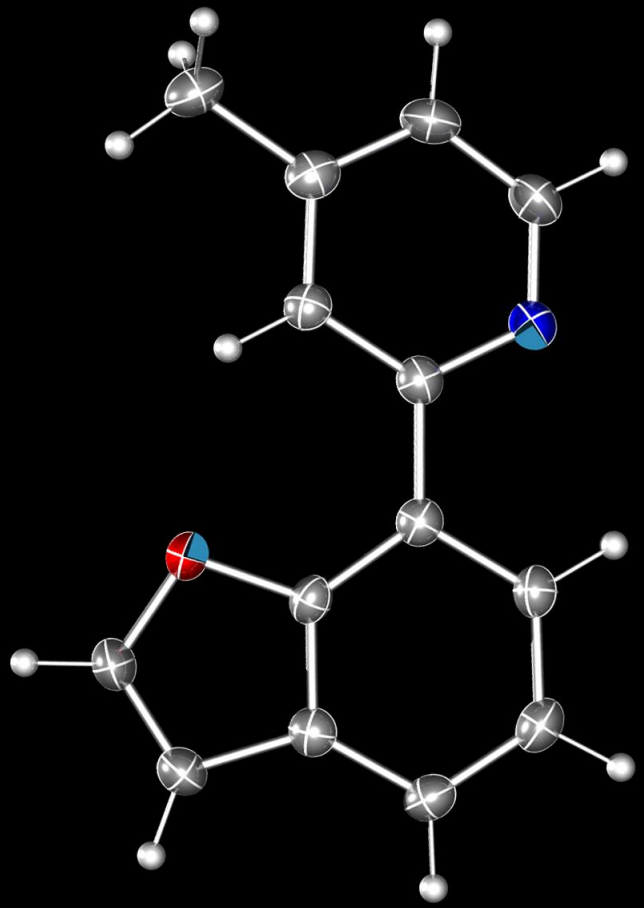
indole

S ≈ NH

Single Crystal Structures Also Corroborate Prediction



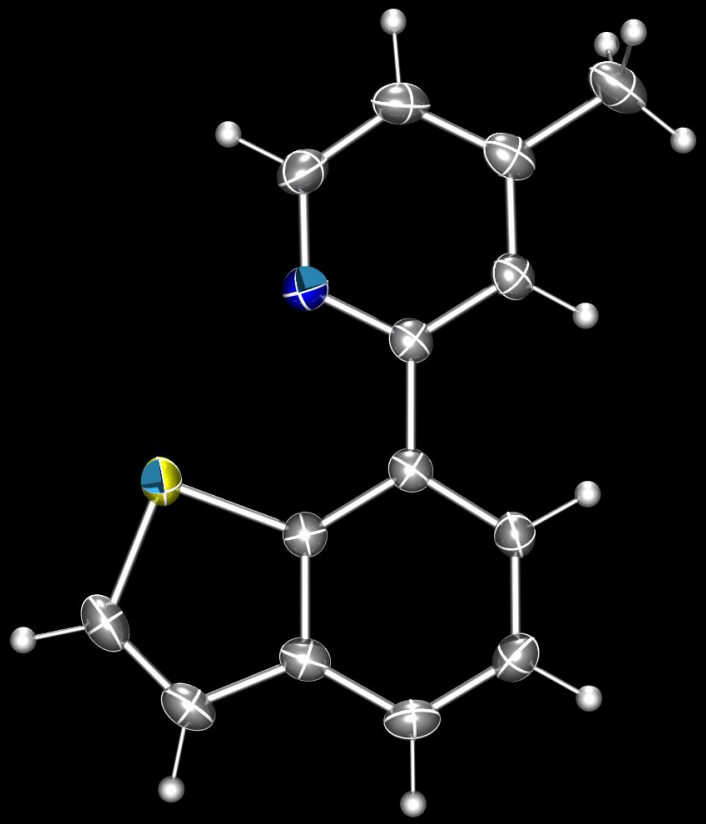
benzothiophene



benzofuran

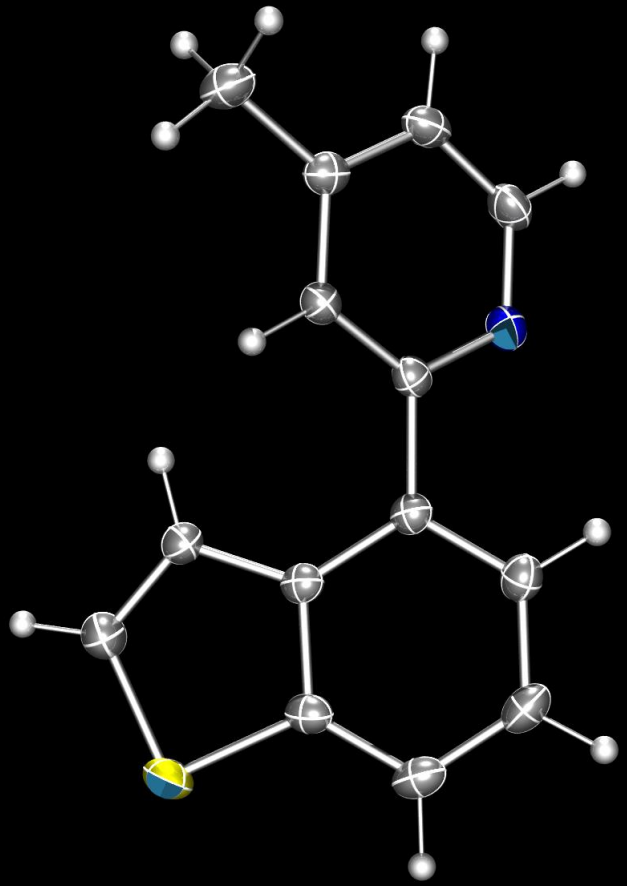
S ≠ O

Single Crystal Structures Also Corroborate Prediction



benzothiophene

S ≠ C



benzothiophene
regioisomer

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Part 2:

Spectra (VCD)

and

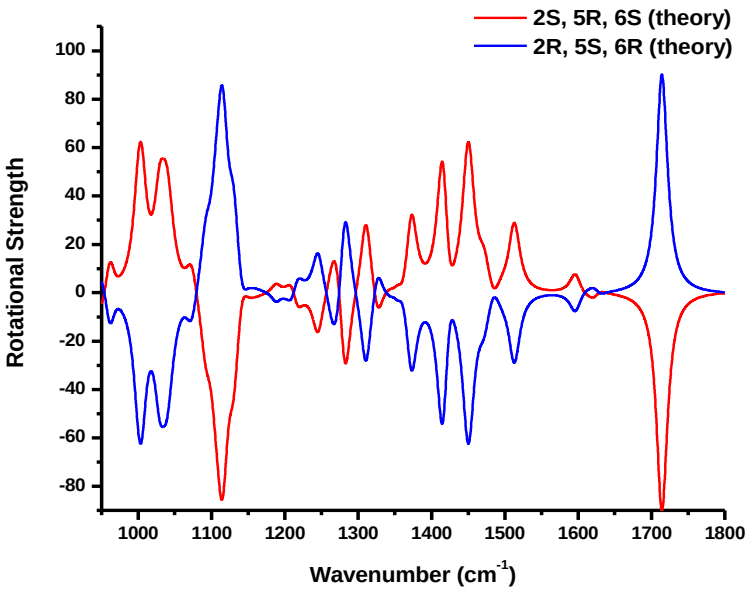
Conformational Effects, II :

Sterically-Driven Interactions in

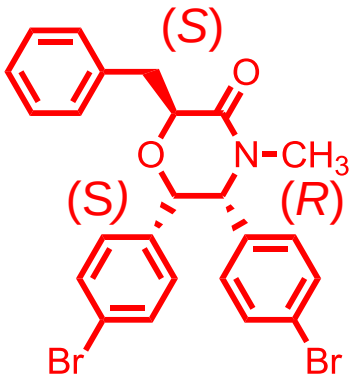
MDM2-p53 Disruption

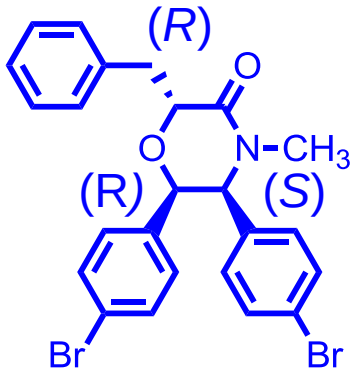
(Oncology)

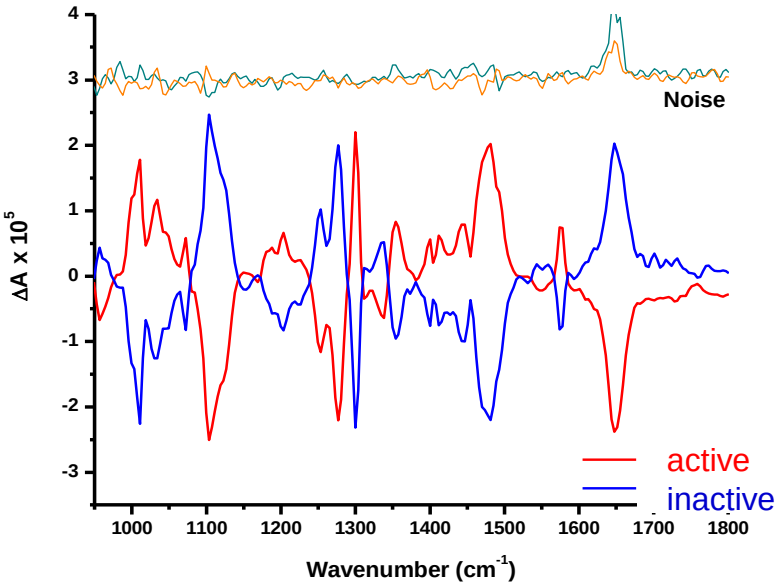
Determination of Absolute Configuration *in Solution*: VCD



Theoretical Vibrational Circular Dichroism Spectra (B3LYP/6-31G*)







Experimental Vibrational Circular Dichroism Spectra in $CDCl_3$

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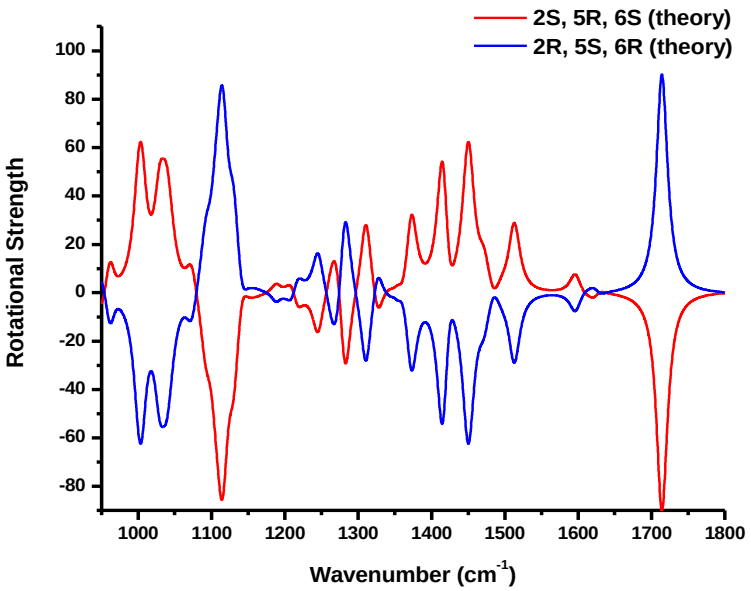
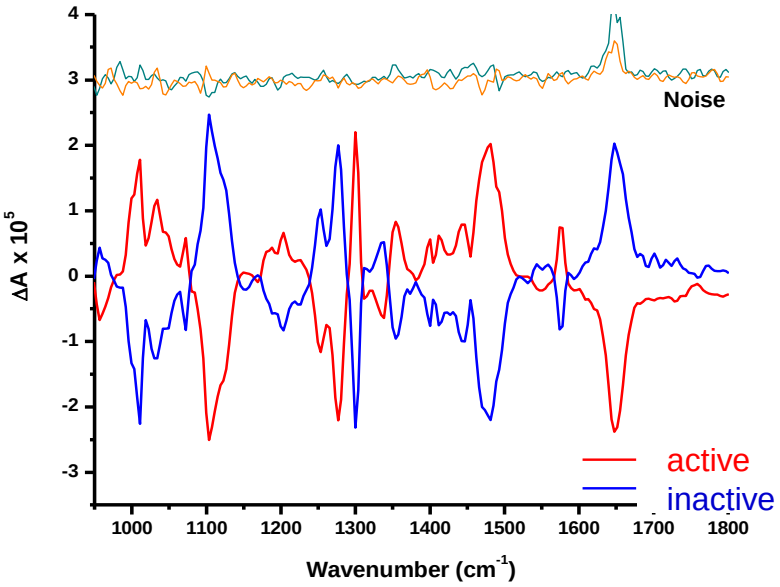
Determination of Absolute Configuration *in Solution*: VCD





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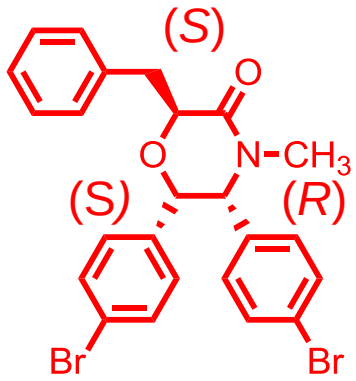

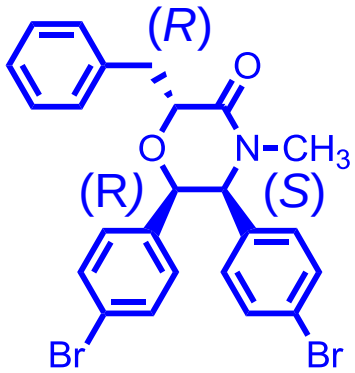

α_D (calc) = +200°
α_D (exp) = +166.2°

active is (2*S*,5*R*,6*S*)

α_D (calc) = -200°
α_D (exp) = -154.7°

inactive is (2*R*,5*S*,6*R*)

Experimental Vibrational Circular Dichroism Spectra in $CDCl_3$

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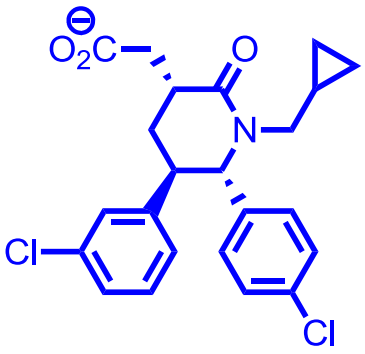
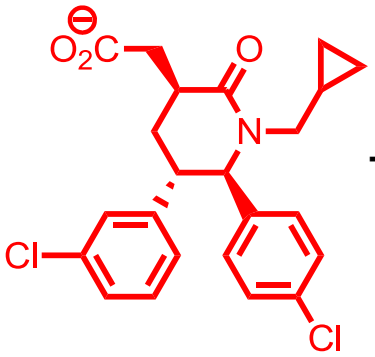
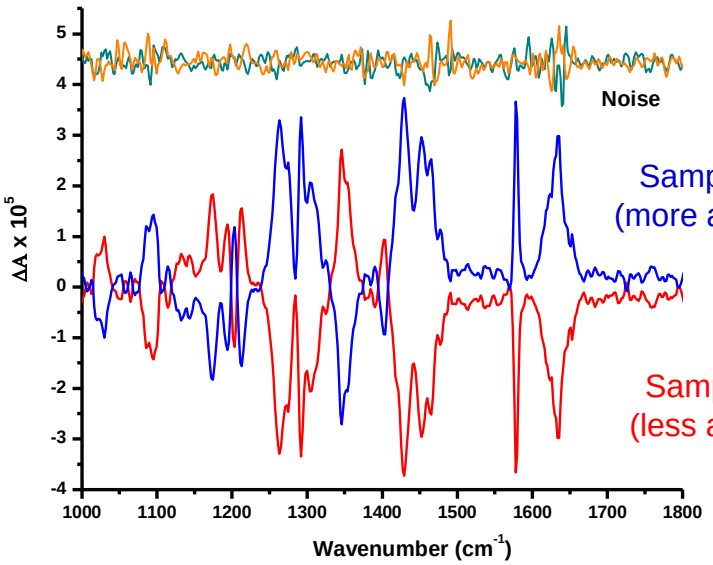
Absolute Configuration Determined via VCD

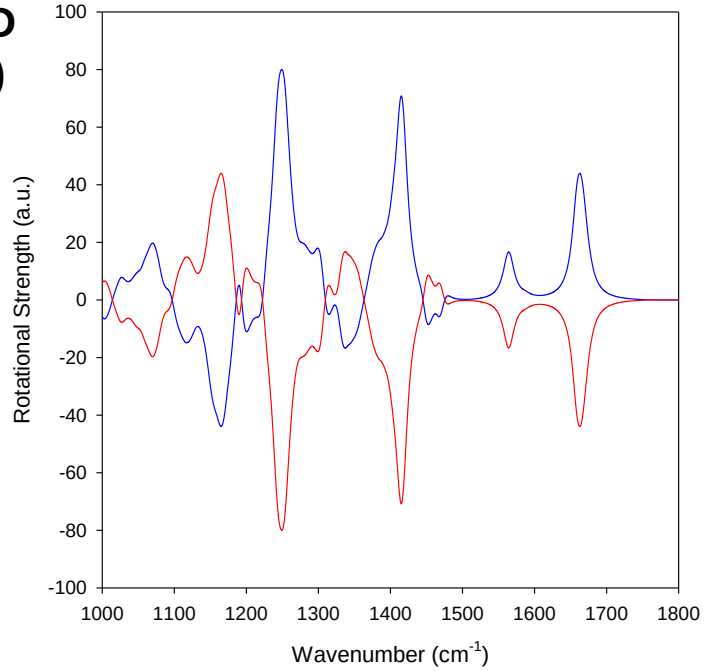


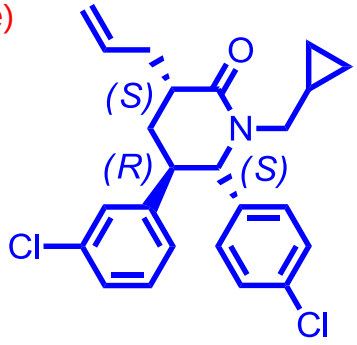


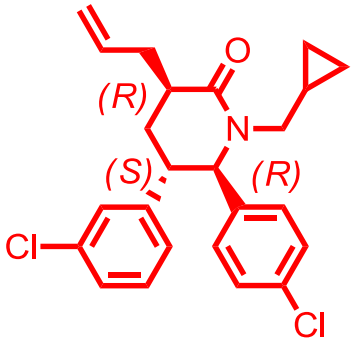


Experimental VCD in $CDCl_3$

Theoretical VCD (B3LYP/6-31G*)







VCD-based assignment corroborated using QM-vs-experimental optical rotations

(Boltzmann-weighted average of thermally-accessible conformational ensemble)

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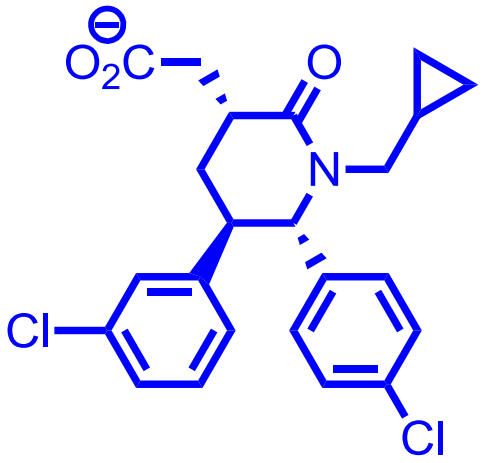
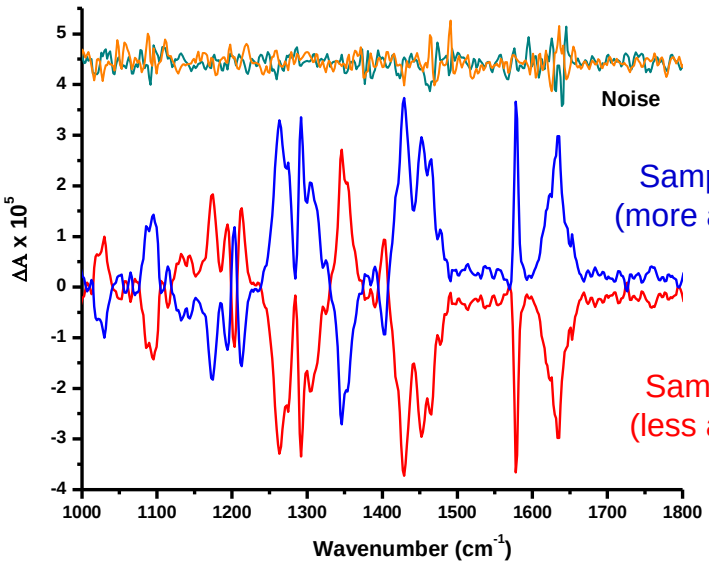
Absolute Configuration Determined via VCD

active form

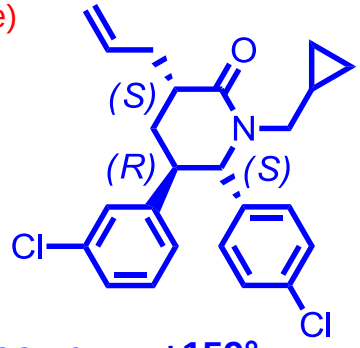




exp. α_D ~ +148°

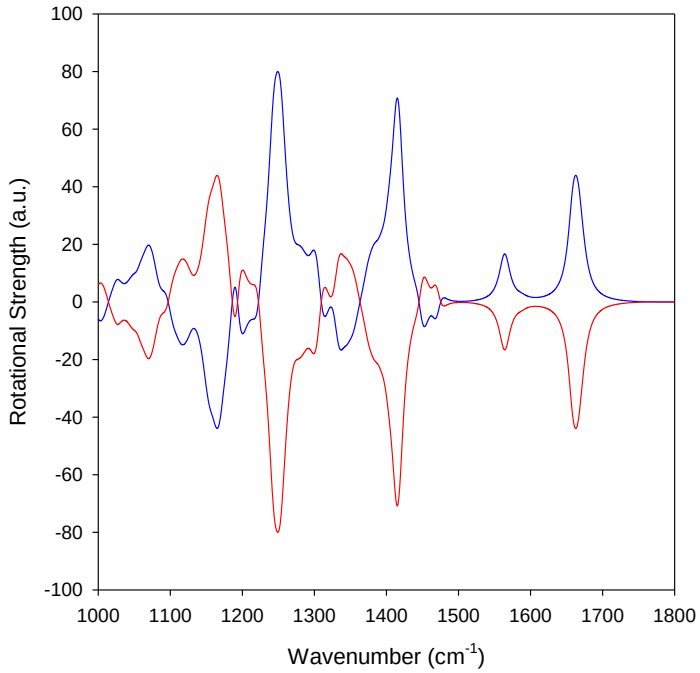
Experimental VCD in $CDCl_3$

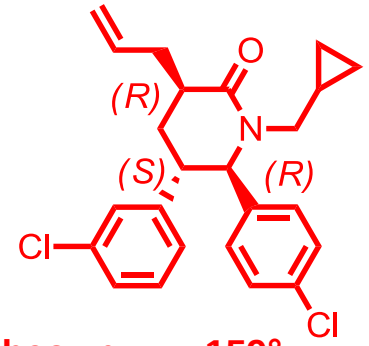
(exp. α_D ~ -85°)



theo. α_D = +159°

Theoretical VCD (B3LYP/6-31G*)





theo. α_D = -159°

VCD-based assignment corroborated using QM-vs-experimental optical rotations

(Boltzmann-weighted average of thermally-accessible conformational ensemble)

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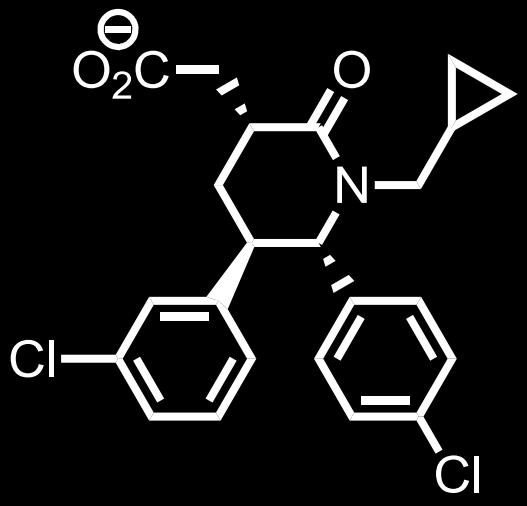
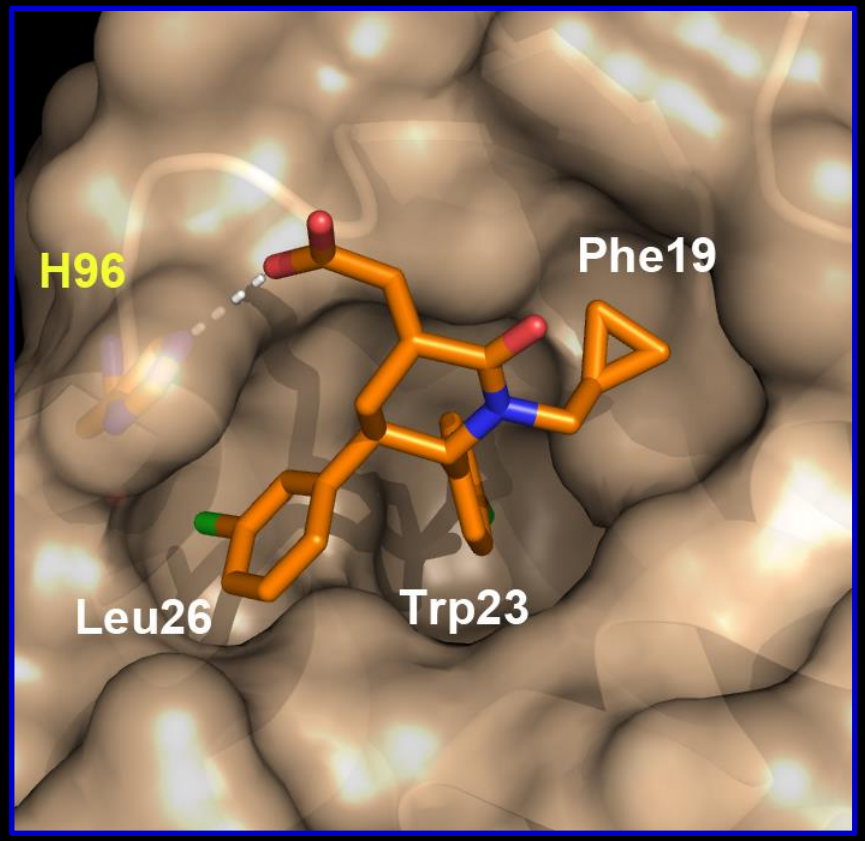
Leads to Predicted Binding Mode (Docking / MM-GBSA)





predicted binding mode (docking followed by MMGB minimization in MDM2 protein)

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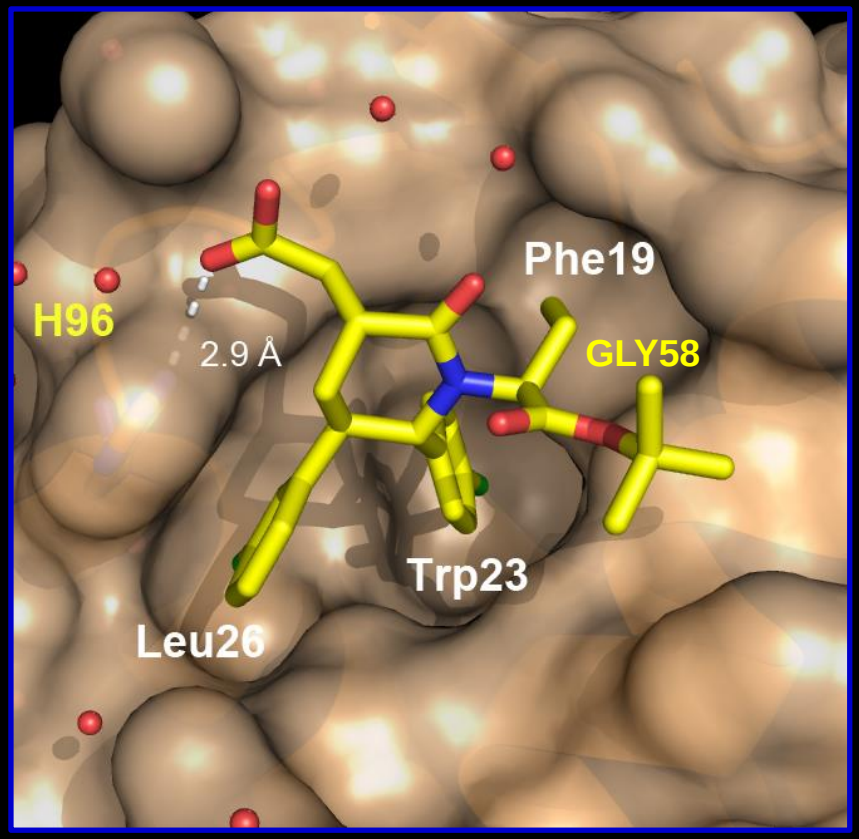
… Subsequently Verified Crystallographically



X-ray co-crystal structure in MDM2 protein
(superimposable with model)

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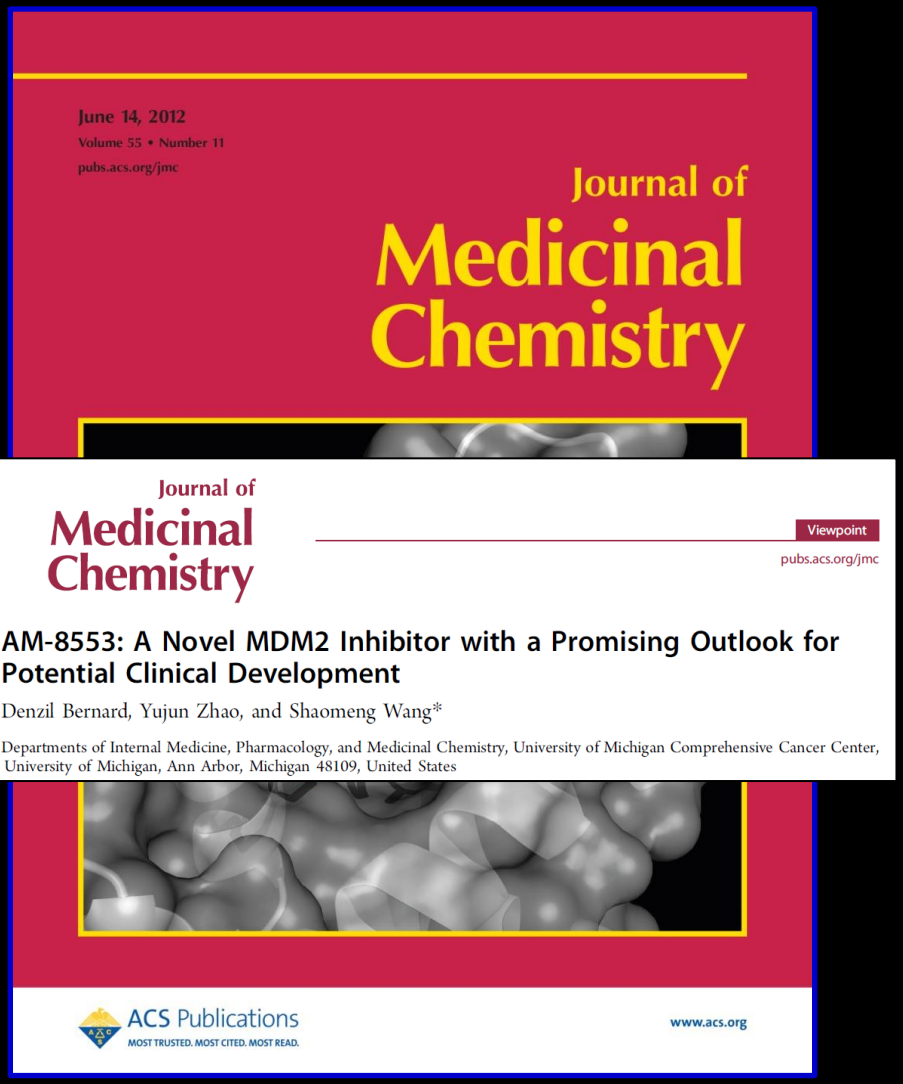
… Subsequently Verified Crystallographically



Journal of
Medicinal
Chemistry

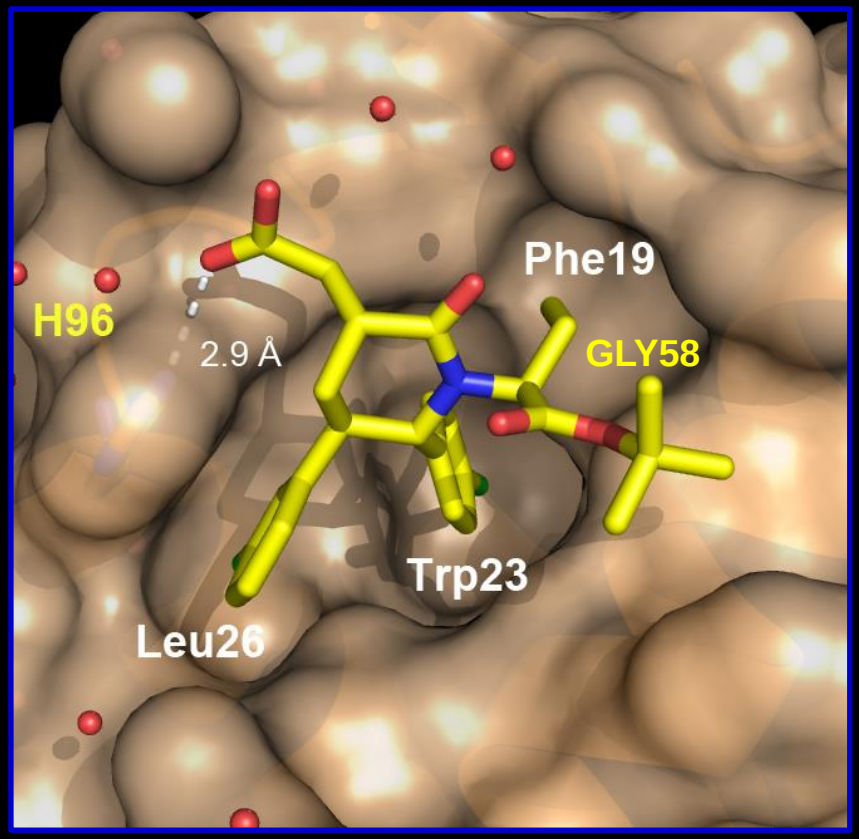
Viewpoint

pubs.acs.org/jmc

AM-8553: A Novel MDM2 Inhibitor with a Promising Outlook for Potential Clinical Development

Denzil Bernard, Yujun Zhao, and Shaomeng Wang*

Departments of Internal Medicine, Pharmacology, and Medicinal Chemistry, University of Michigan Comprehensive Cancer Center, University of Michigan, Ann Arbor, Michigan 48109, United States



X-ray co-crystal structure in MDM2 protein (superimposable with model)

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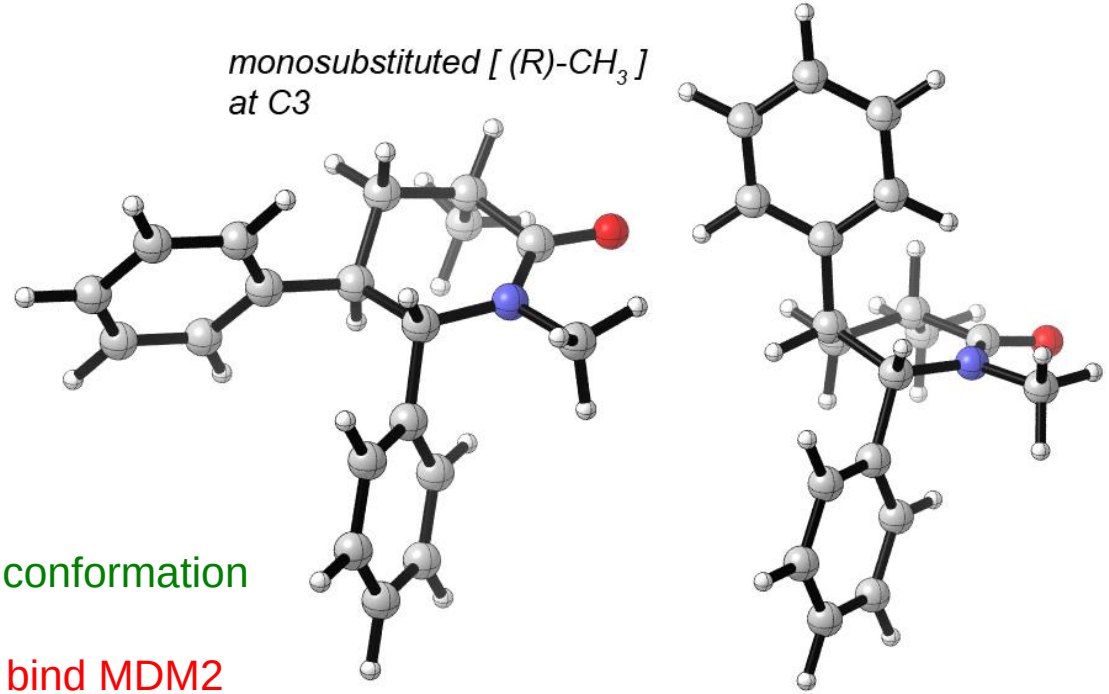
Crystallographic Binding Conformation is not the Free State Global Minimum



"gauche-like*"* orientation: binding conformation

"anti-like*"* orientation: can not bind MDM2

	gauche	*anti*
ΔE (B3LYP/6-31G*)	+0.5	+0.0

prediction: only ~30 % of compound exists in binding conformation (ca. 30 : 70)

proposal: introduce a group tolerated by protein target which stabilizes gauche-like orientation (via destabilization of anti) , enriching population of binding conformer

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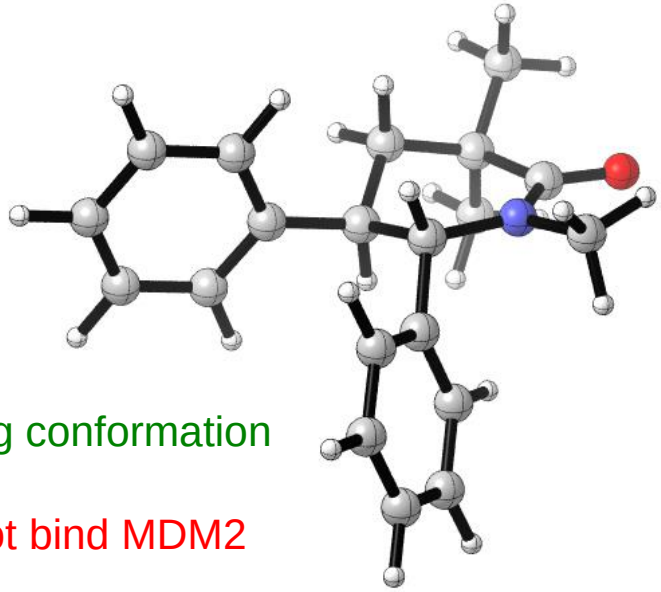
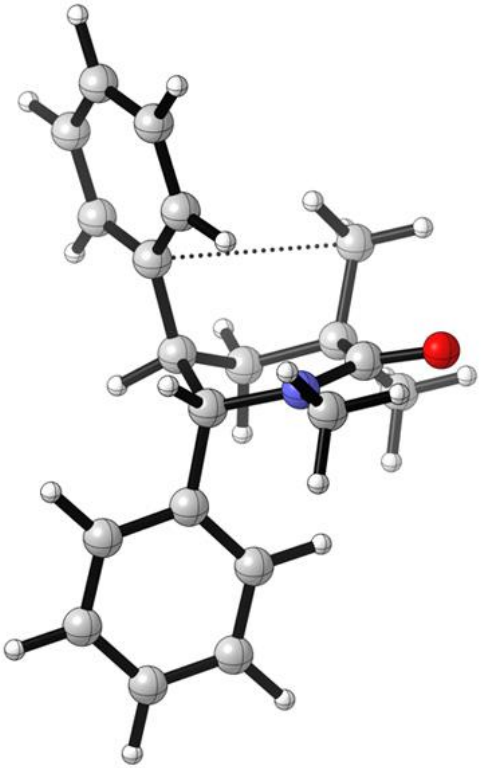
Prediction: C3 Substitution Stabilizes Binding Conformation

disubstituted at C3





"gauche-like" orientation: binding conformation

"anti-like" orientation: can not bind MDM2

	gauche	*anti*
ΔE (B3LYP/6-31G*)	+0.0	+2.4

(ca. 99 : 1)

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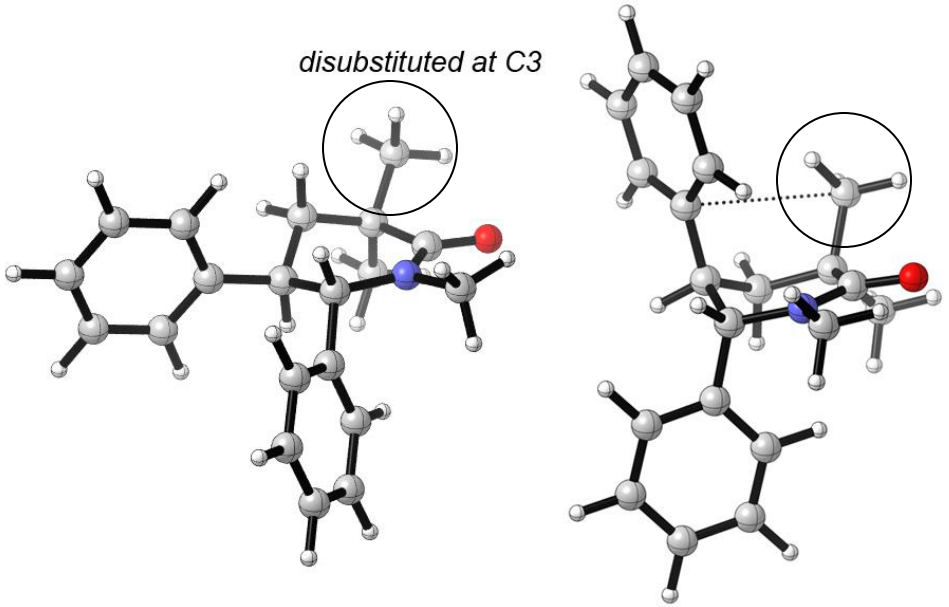
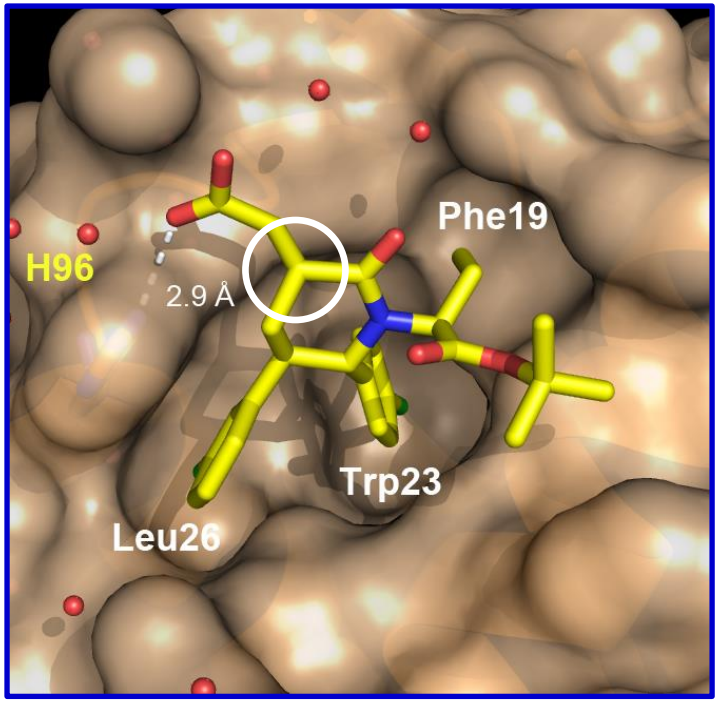
Prediction: C3 Substitution Stabilizes Binding Conformation

"gauche-like" orientation: binding conformation

"anti-like" orientation: can not bind MDM2





	gauche	anti
ΔE (B3LYP/6-31G*)	+0.0	+2.4

(ca. 99 : 1)

prediction: conformation-stabilizing C3 substituent would project out toward solvent and not interfere with MDM2 binding

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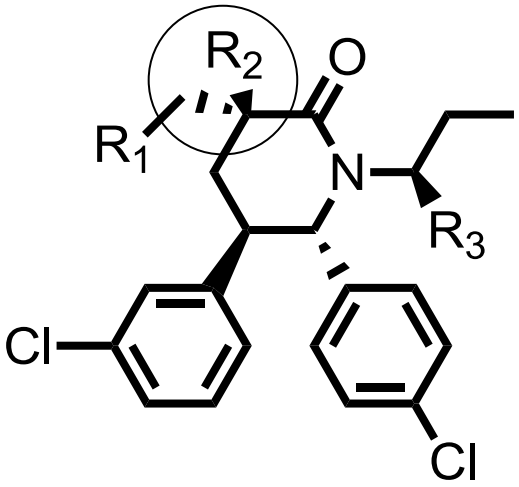
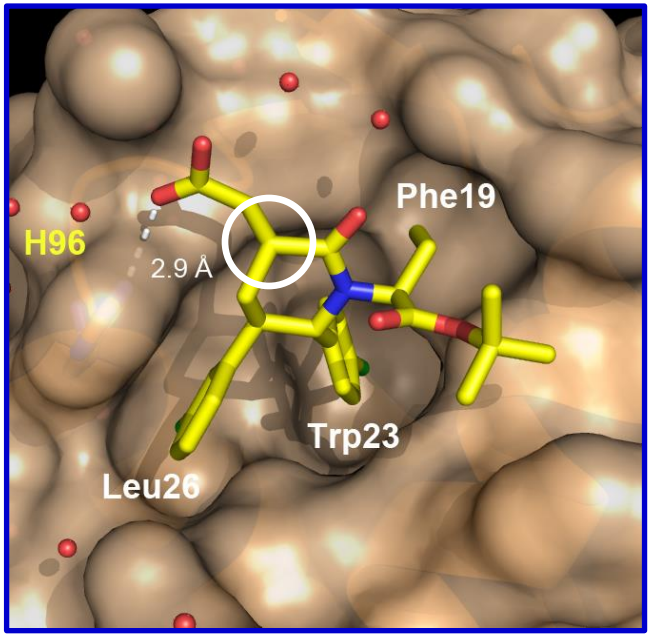
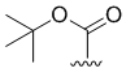
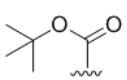
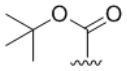
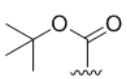
C3 Substitution Results in Increased Activity (2-3x)





Compd	R¹	R²	R³	Biochemical Potency		Cellular Potency (SJSA-1)	
				HTRF IC50 (nM)[a]	HTRF (15% HS[b]) IC50 (nM)[a]	p21 (10% HS[b]) IC50 (μM)[a]	EdU (10% HS[b]) IC50 (μM)[a]
23				4.2 ± 0.9	43.3 ± 5.9	4.3 ± 0.7	0.48 ± 0.09
24				1.8 ± 0.7	20.3 ± 3.5	3.1 ± 0.9	0.31 ± 0.09
25				2.2 ± 0.7	19.9 ± 7.5	1.9 ± 0.5	0.19 ± 0.06
26				0.90 ± 0.21	9.0 ± 2.7	1.6 ± 0.7	0.17 ± 0.03

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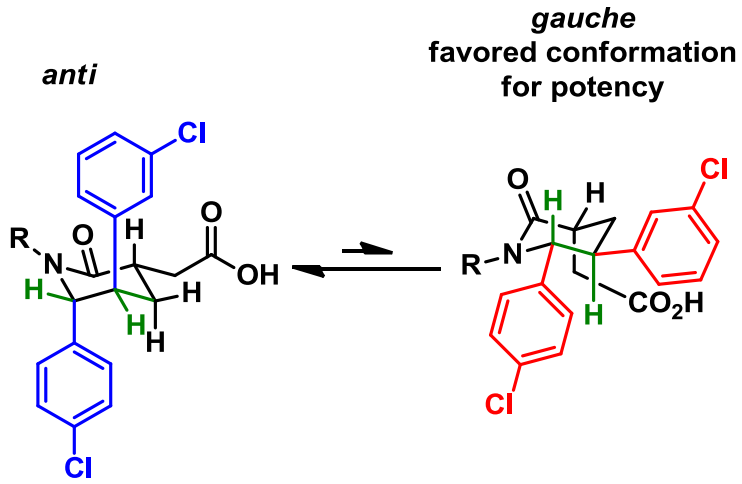
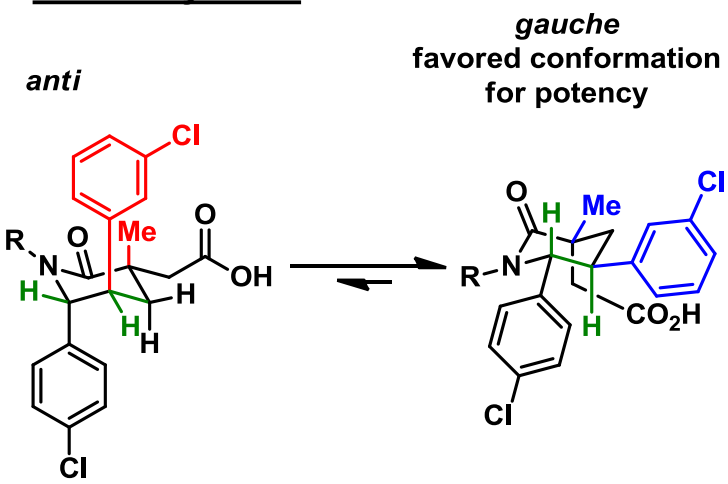
VT ^{1}H NMR Study Validates Distribution in Free State

C3-*des*-methylated:

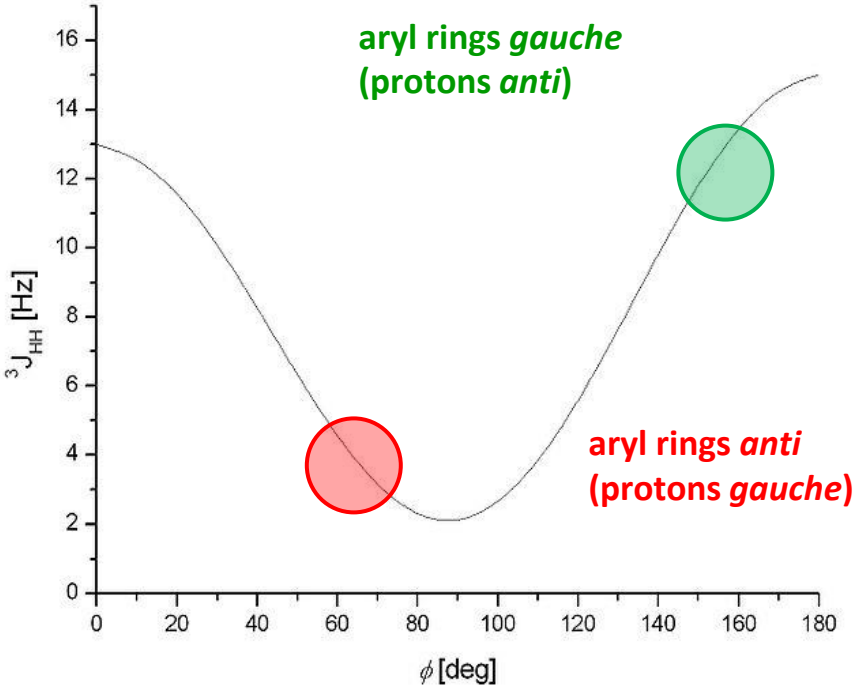


C3-methylated:



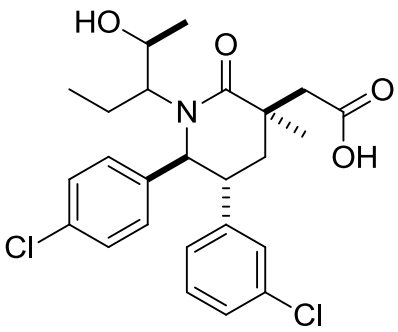
Summary:

		des-methylated	methylated
J_{ab} (Hz)	@ 298K	6.8 (mixture)	10.9 (rings *gauche*)
	@ 203K	0 (rings *anti*)	10.9 (rings *gauche*)



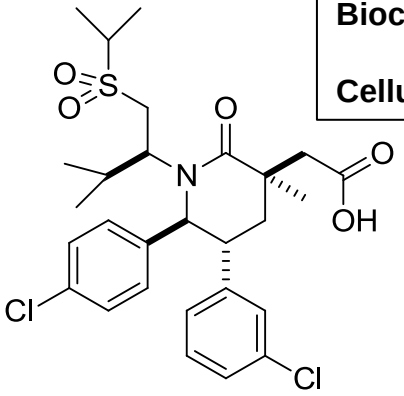
Karplus relationship for $^3J_{HH}$

Onward to the clinic… AMG232



AM-8553

J. Med. Chem. **2012**, *55*, 4936.



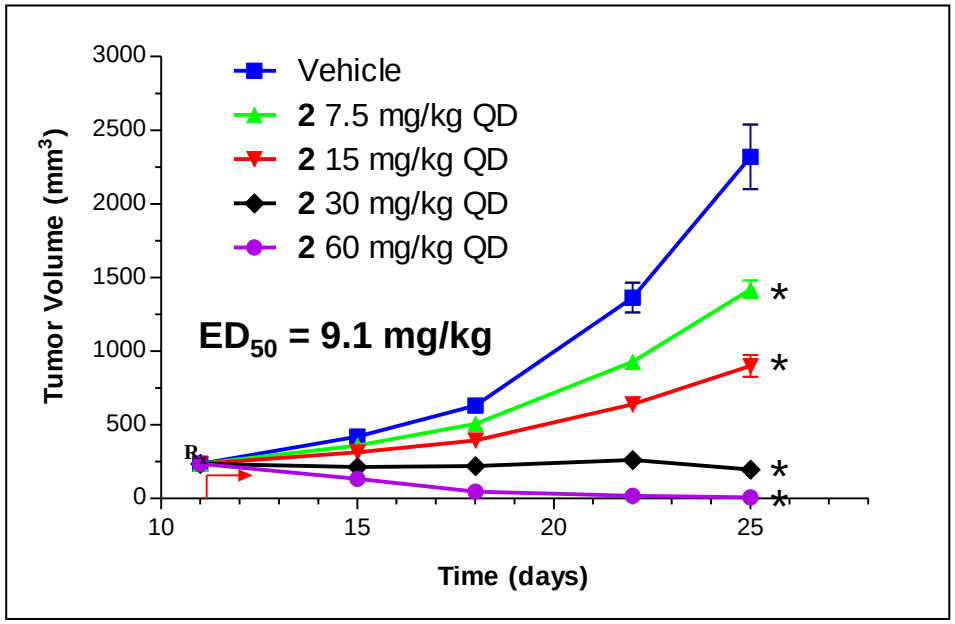
AMG232

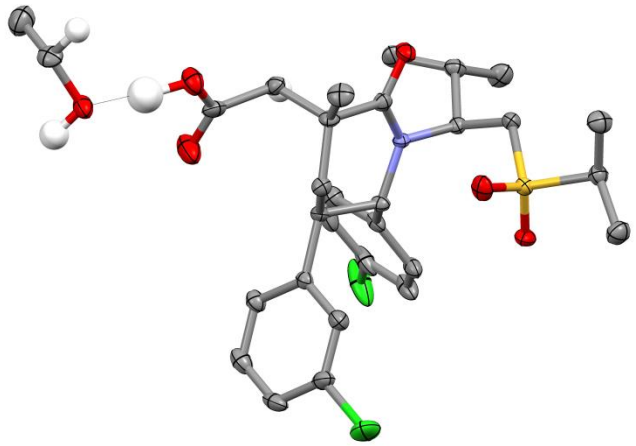
J. Med. Chem. **2014**, *57*, 1454.

Biochemical *in vitro* IC_{50} (HTRF) = 0.6 nM

Cellular activity (SJSA-1, +10%HS) = 9.1 nM

http://www.clinicaltrials.gov
(Ph 1b / 2a)





single-crystal X-ray structure of AMG232

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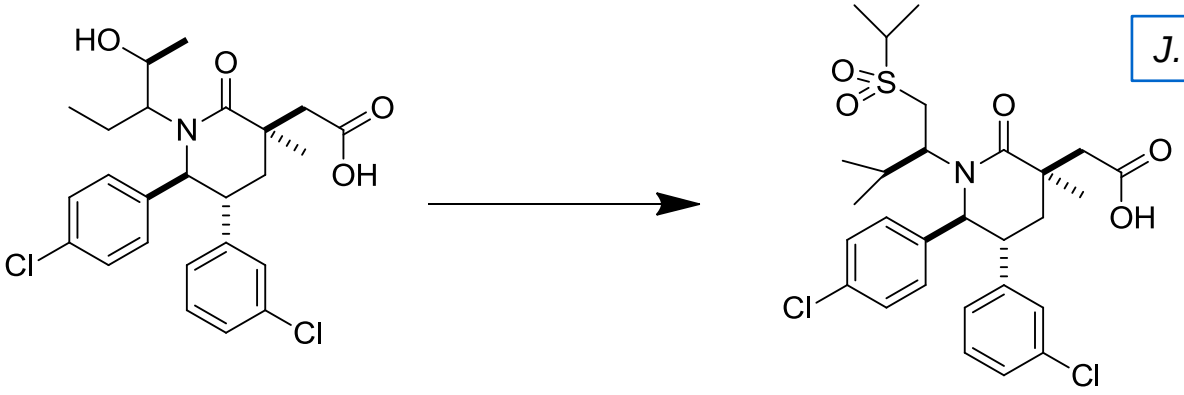
Onward to the clinic… AMG232



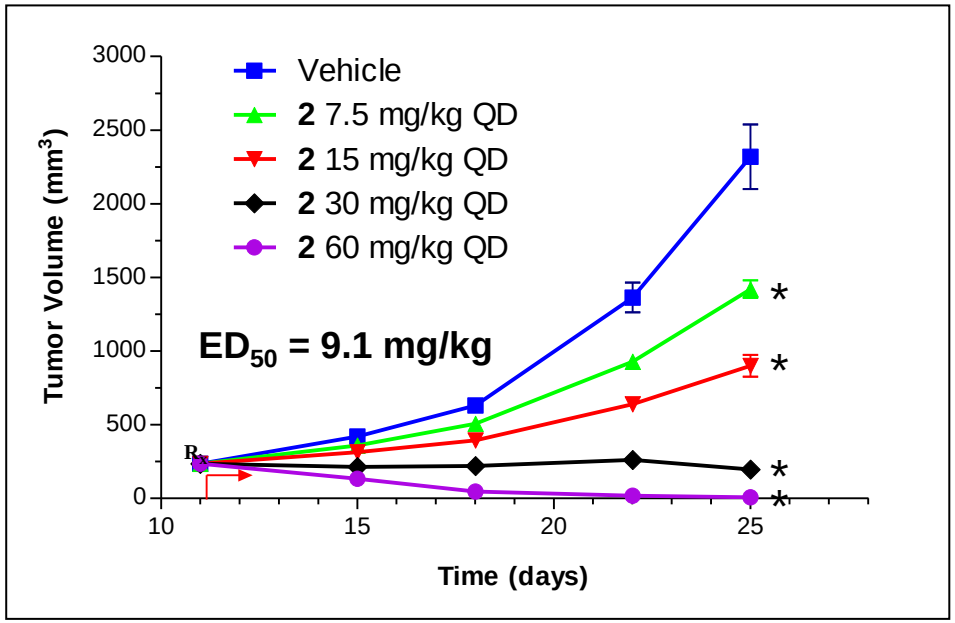
AM-8553

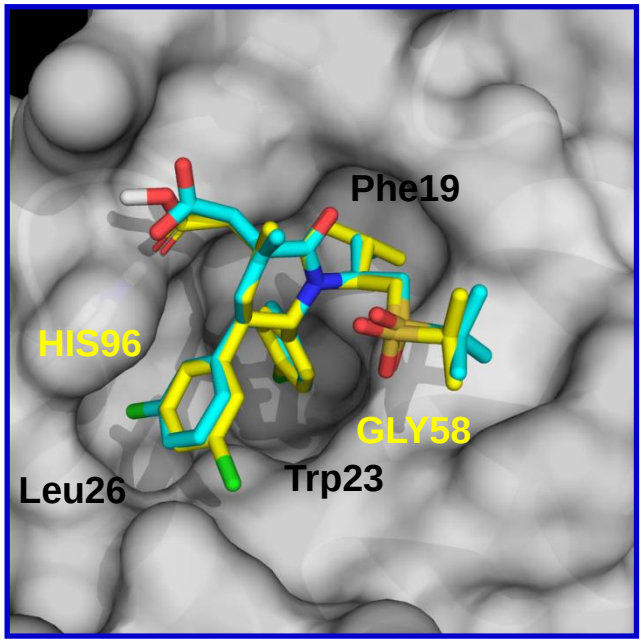
AMG232

J. Med. Chem. **2014**, *57*, 1454.

http://www.clinicaltrials.gov
(Ph 1b / 2a)

J. Med. Chem. **2012**, *55*, 4936.





single-crystal X-ray structure of AMG232 (yellow) overlaid with co-crystal structure of *tert*-butyl analog in MDM2 protein (cyan/grey)

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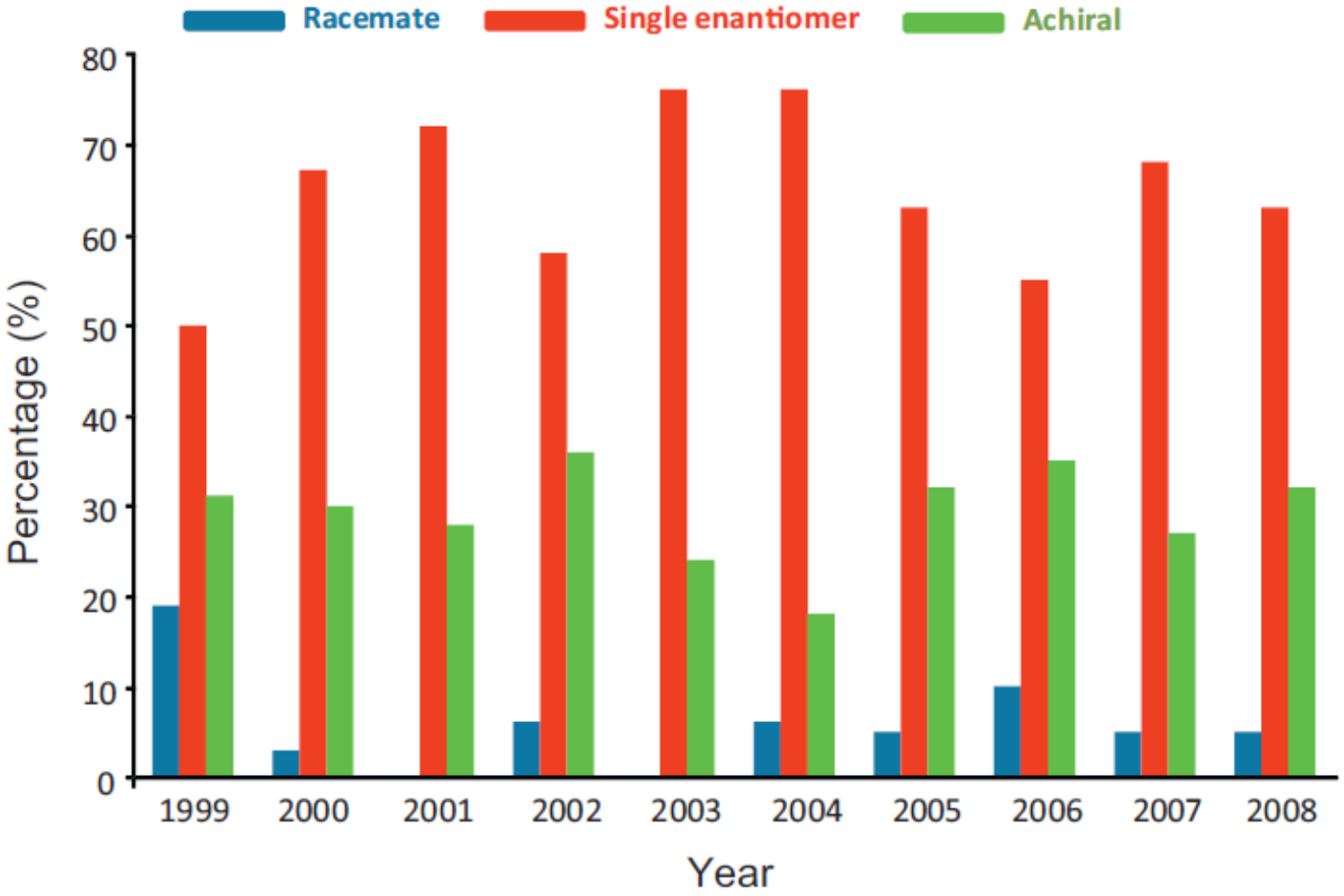
Part 3:

Ion Mobility and Density Functional Theory in Structural Elucidation of Diastereomers

Most Approved Drugs are Single Enantiomers of Chiral Molecules

Drugs approved by the FDA from 1999 to 2008



Wu, L. and Vogt, F.G. *J. Pharm and Biomed. Anal.*, **2012**, *69*, 133-147.

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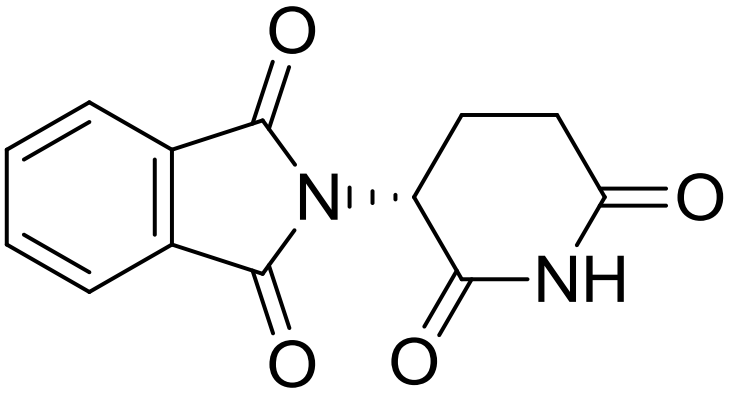
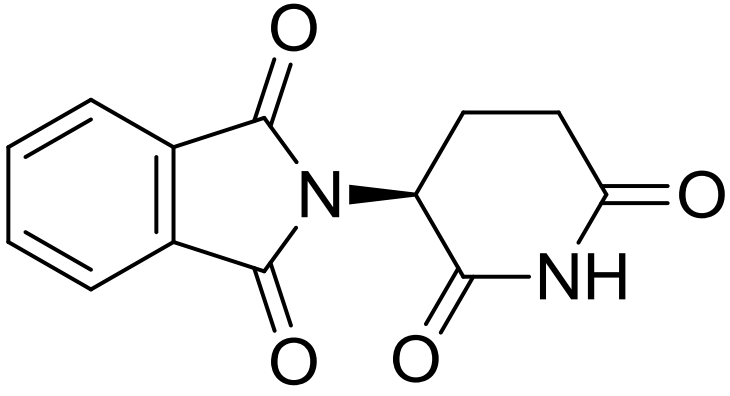
Characterization / Differentiation of Stereoisomers is Important



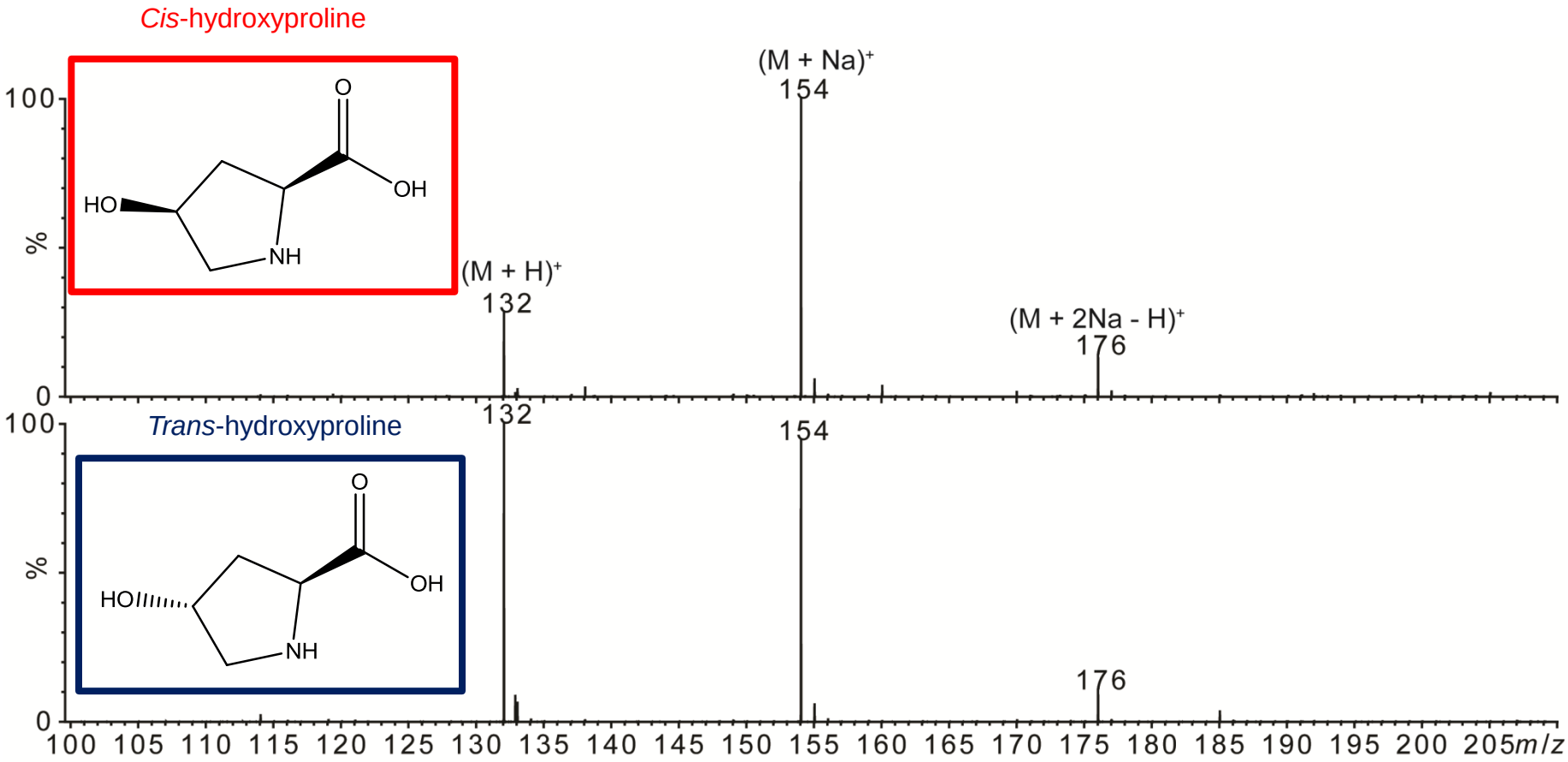
(R)-Thalidomide
Sedative



(S)-Thalidomide
Teratogenic

MS: Fast, Highly Sensitive Analytical Tool for Structural Determination…

… however, in many cases, can not differentiate cis/trans Isomers by MS (or MS^n) alone

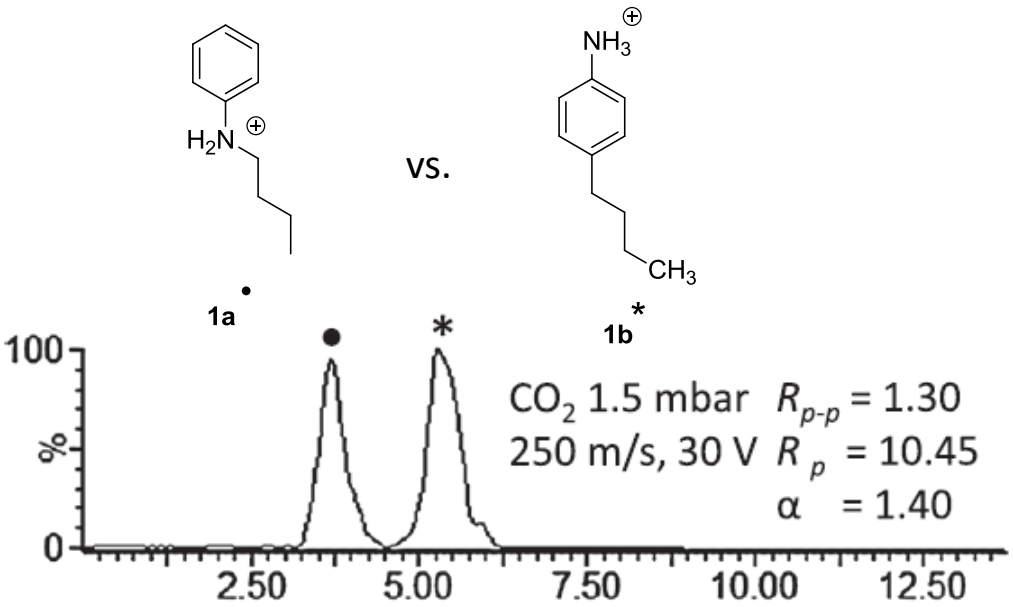


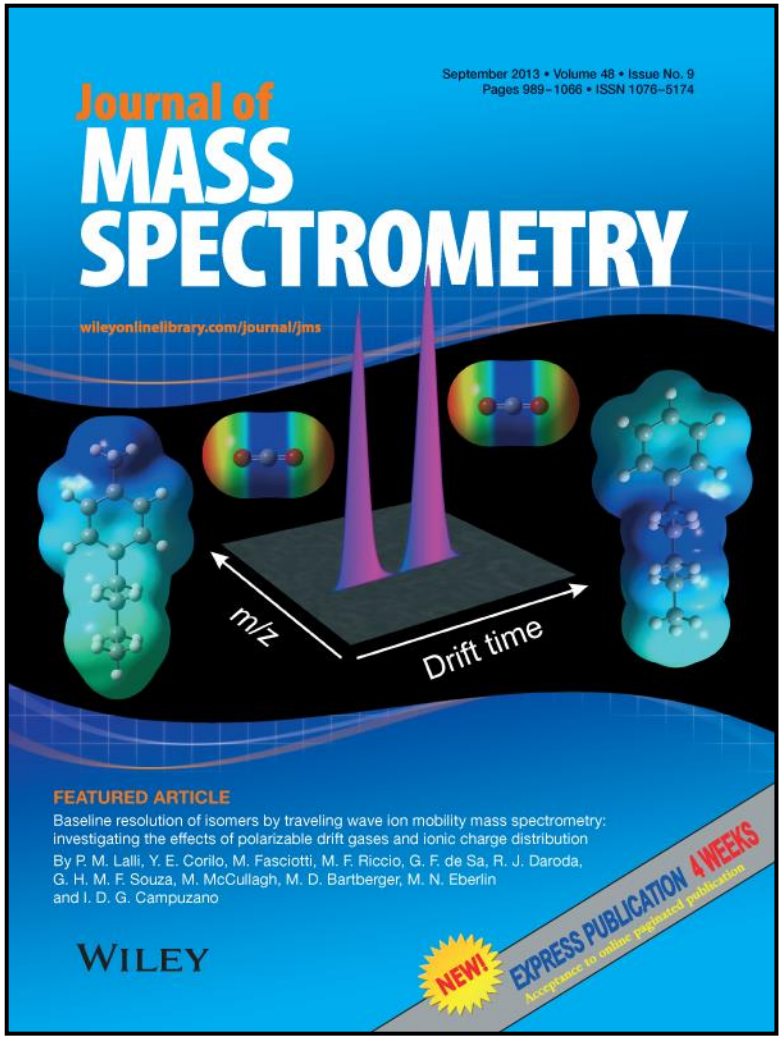
Differing Charge Distribution Allows Ion Separation via Mobility (Incl. Species w/Identical MW and/or Similar Fragmentation Pattern)

(wileyonlinelibrary.com) DOI 10.1002/jms.3245

Baseline resolution of isomers by traveling wave ion mobility mass spectrometry: investigating the effects of polarizable drift gases and ionic charge distribution

Priscila M. Lalli,[a] Yuri E. Corilo,[a] Maíra Fasciotti,[a] Maria Francesca Riccio,[a] Gilberto F. de Sa,[b,c] Romeu J. Daroda,[c] Gustavo H. M. F. Souza,[d] Michael McCullagh,[e] Michael D. Bartberger,[f] Marcos N. Eberlin[a]* and Iain D. G. Campuzano[f]*





Ion Mobility: Ashcroft, A.E., et al. *Int. J. Mass Spectrom.*, **2012**, *298*, 17-23.

Competitive Dissociation Kinetics: Cooks, R.G., et al. *JASMS*, **2003**, *14*, 152-160.

J. Mass Spectrom., **2013**, *48*, 989.

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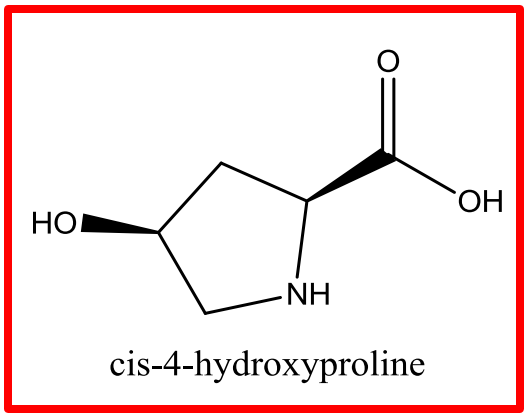
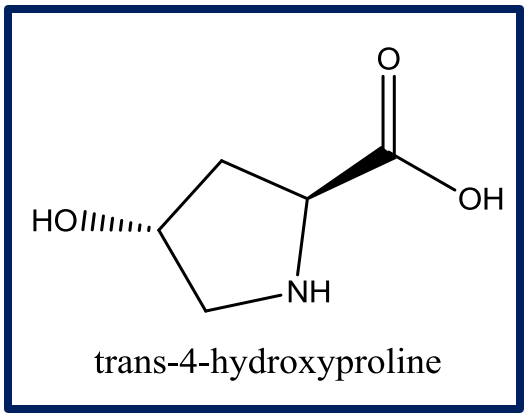
Metal Cationization Allowed for Differentiation of Stereoisomers Of Hydroxyproline, e.g. $(M + Na)^+$

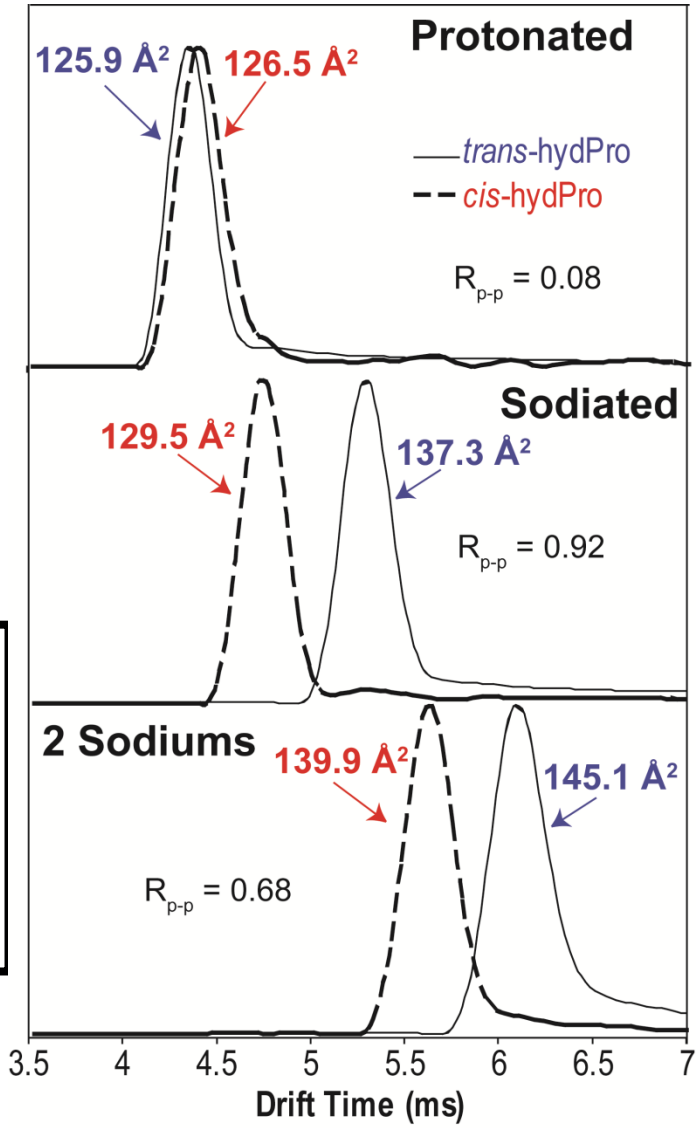


cis-4-hydroxyproline



trans-4-hydroxyproline

- $(M + Na)^+$ results in substantially improved Isomeric resolution compared to $(M + H)^+$
- Additional Na^+ adduction did not enhance differentiation



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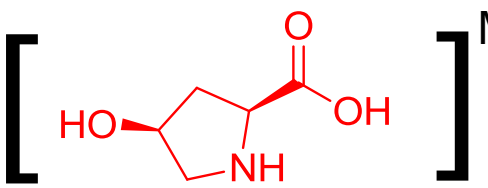
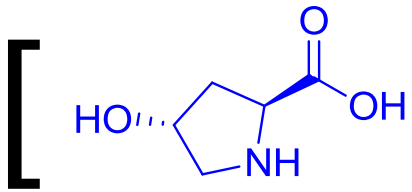
Effect of Metal Identity on Separation



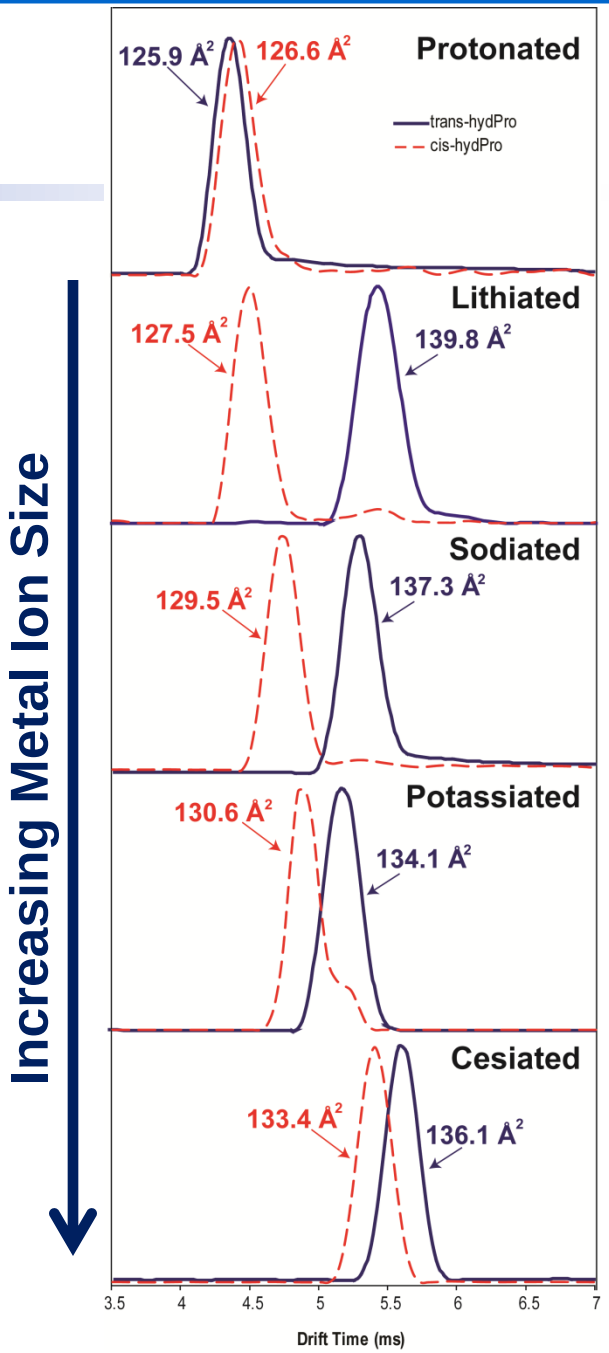
cis-4-hydroxyproline



trans-4-hydroxyproline

As metal ion size increases
(or charge density decreases),

- *cis* form steadily increases in ccs (increase ~6 Å^2)
- *trans* form decreases in ccs, but largely stays at a similar ccs



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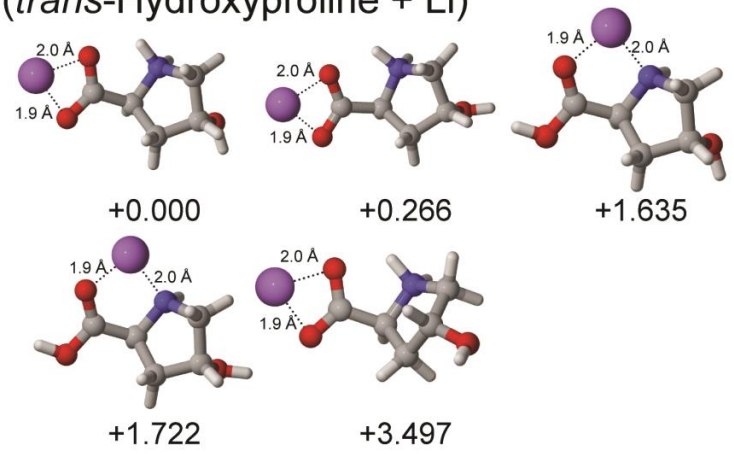
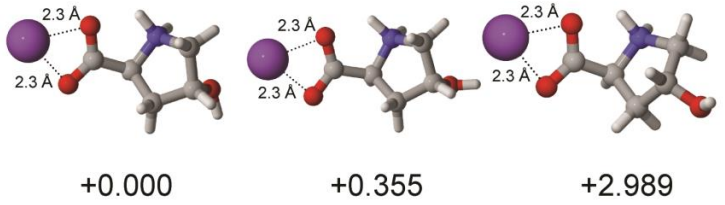
Trans-Hydroxyproline: Computational Investigation (B3LYP/6-31++G(d,p))

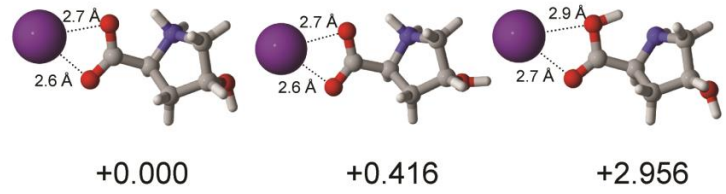
(*trans*-Hydroxyproline + Li)$^+$



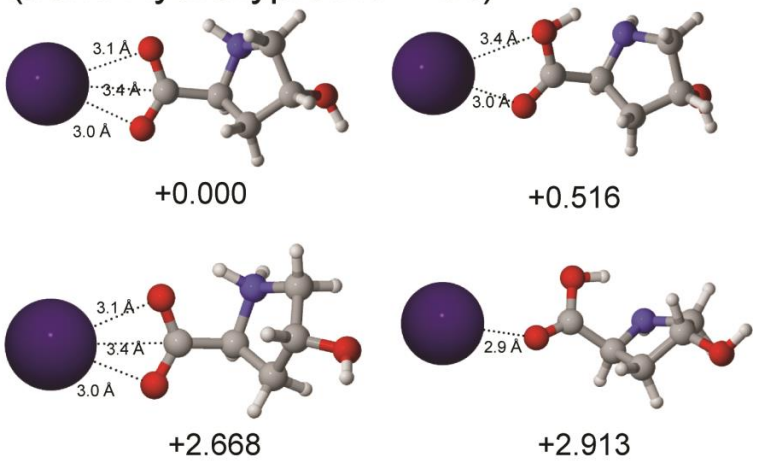
(*trans*-Hydroxyproline + Na)$^+$



(*trans*-Hydroxyproline + K)$^+$



(*trans*-Hydroxyproline + Cs)$^+$



B3LYP/SVPD basis used for Cs$^+$

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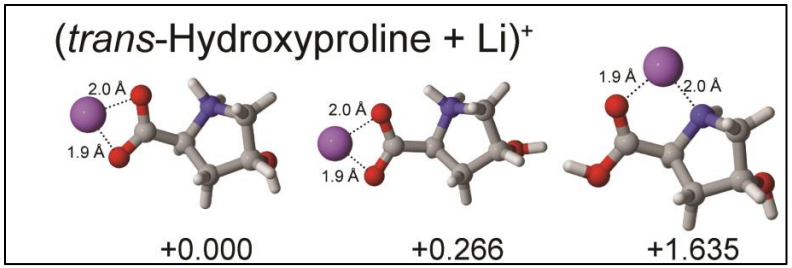
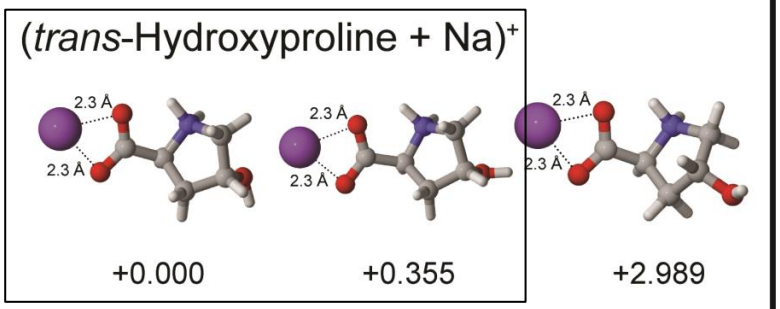
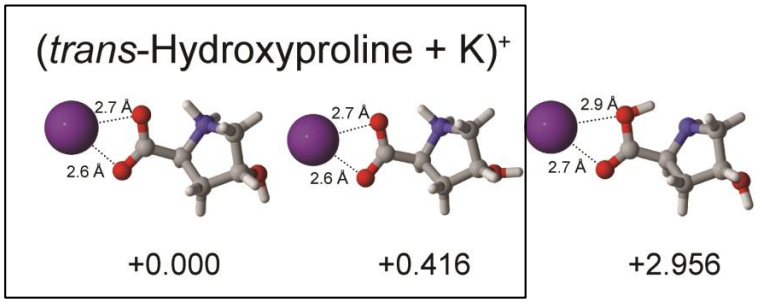
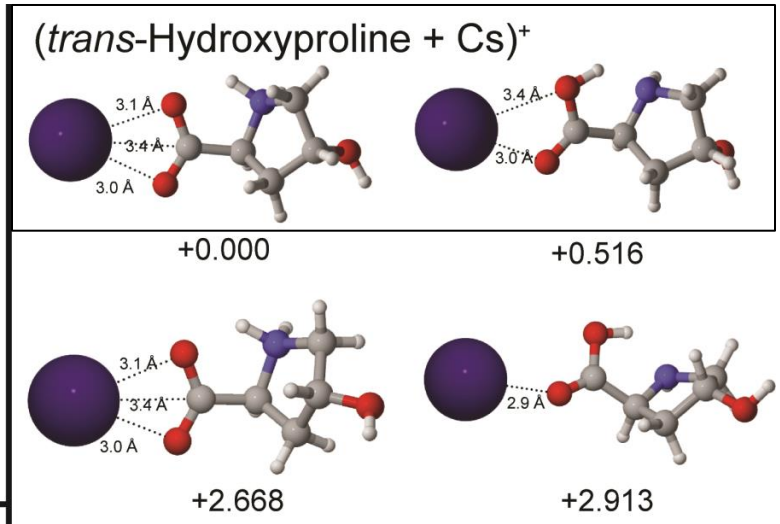
Trans-Hydroxyproline: Computational Investigation (B3LYP/6-31++G(d,p))









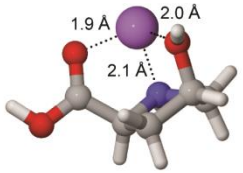
Salt-bridge structure is favored for *trans*-hydroxyproline, regardless of metal ion identity

B3LYP/SVPD basis used for Cs^+

AMGEN

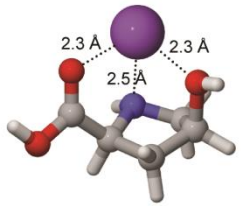
Cis-Hydroxyproline: Computational Investigation (B3LYP/6-31++G(d,p))

(*cis*-Proline + Li)$^+$



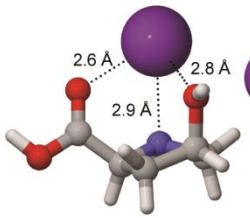
+0.000

(*cis*-Proline + Na)$^+$

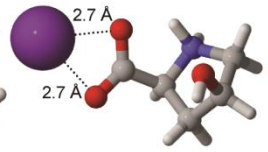


+0.000

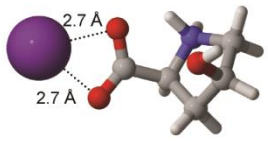
(*cis*-Proline + K)$^+$



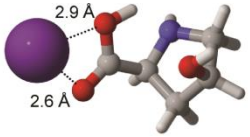
+0.000



+1.604

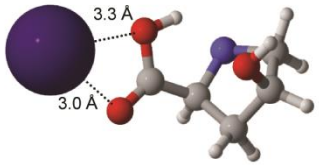


+2.063



+2.310

(*cis*-Proline + Cs)$^+$



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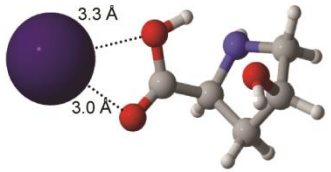
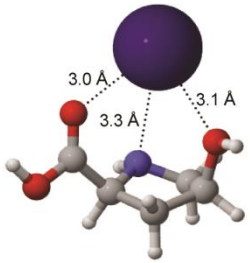
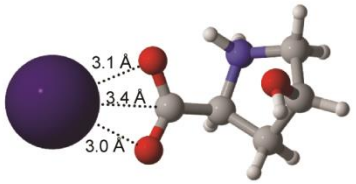


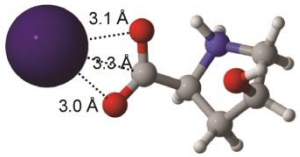
+0.543



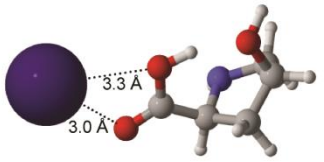
+1.010



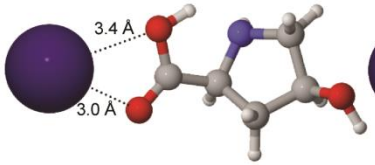
+1.397



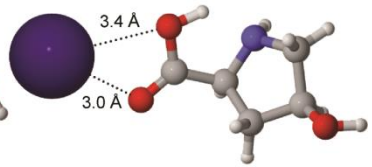
+1.840



+2.422

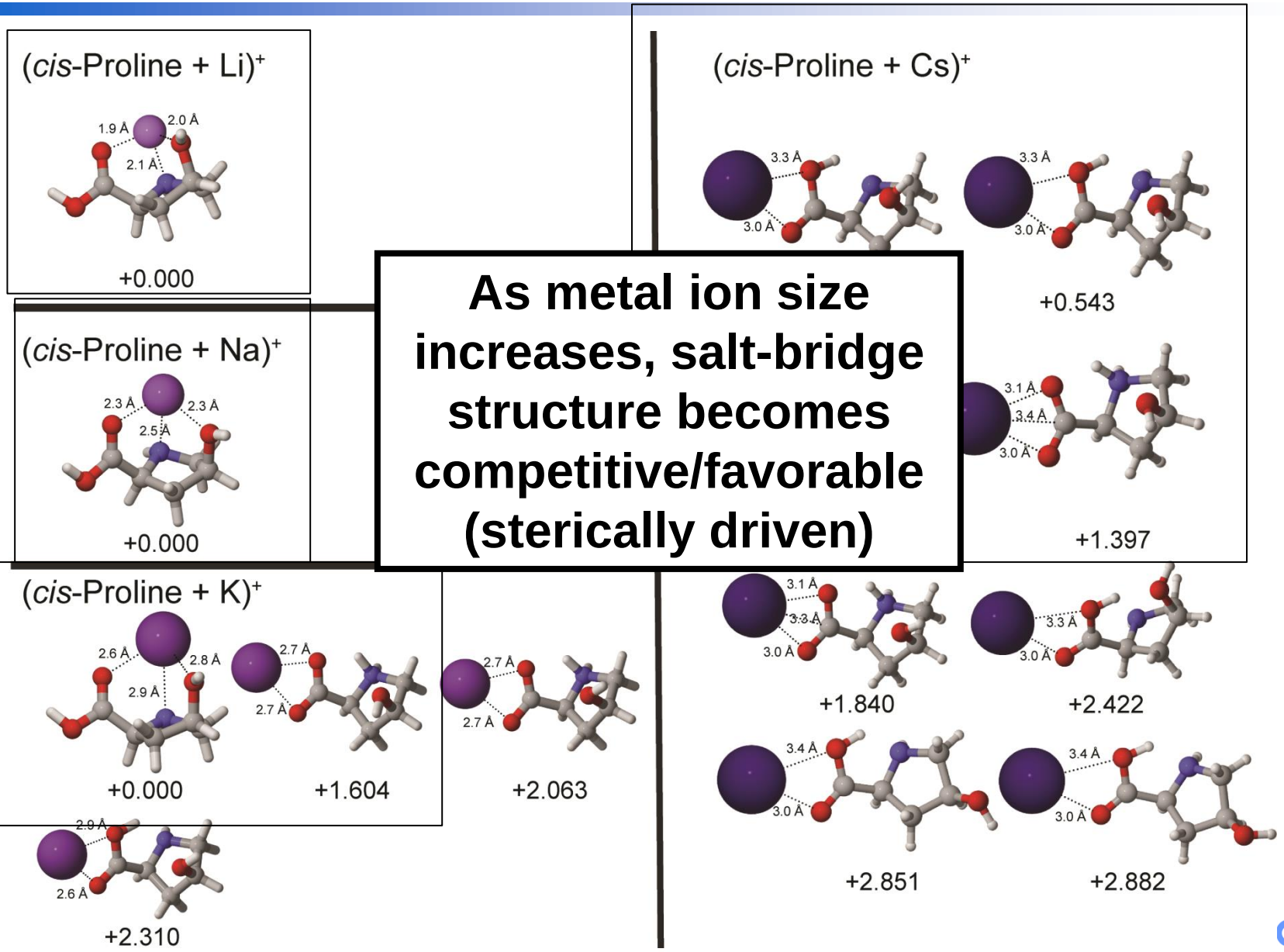


+2.851

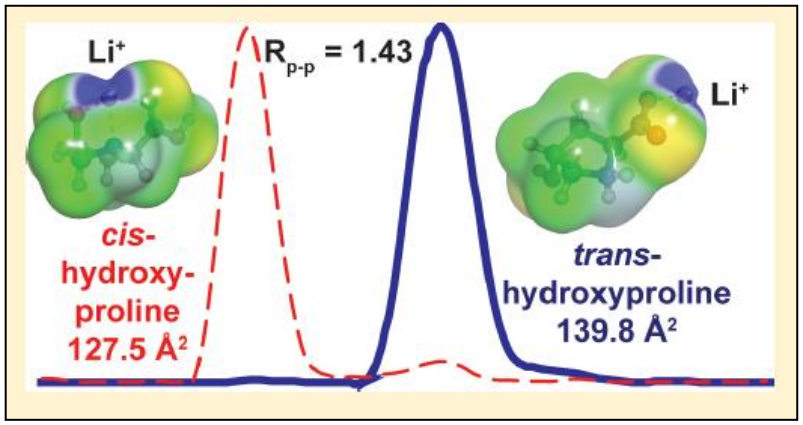


+2.882

Cis-Hydroxyproline: Computational Investigation (B3LYP/6-31++G(d,p))



Δ(CCS) (trans-cis) Values Track Well With Experiment



(B3LYP/6-31++G(d,p)

Δ(calc.)

Δ(exp.)

	cis-hydroxyproline		*trans*-hydroxyproline		
charge carrier	exp.	theor.	exp.	theor.	
H	126.5	120.7 ± 1.4	125.9	120.3 ± 1.5	-0.6
Li	127.5	120.0 ± 1.3	139.8	128.0 ± 1.4	+12.3
Na	129.5	122.8 ± 1.0	137.3	129.0 ± 1.6	+7.8
K	130.6	124.6 ± 1.1	134.1	129.6 ± 1.5	+3.5
Cs	133.4	129.4 ± 1.4	136.0	129.5 ± 1.5	+2.6

-0.4
+8.0
+6.2
+5.0
+0.1

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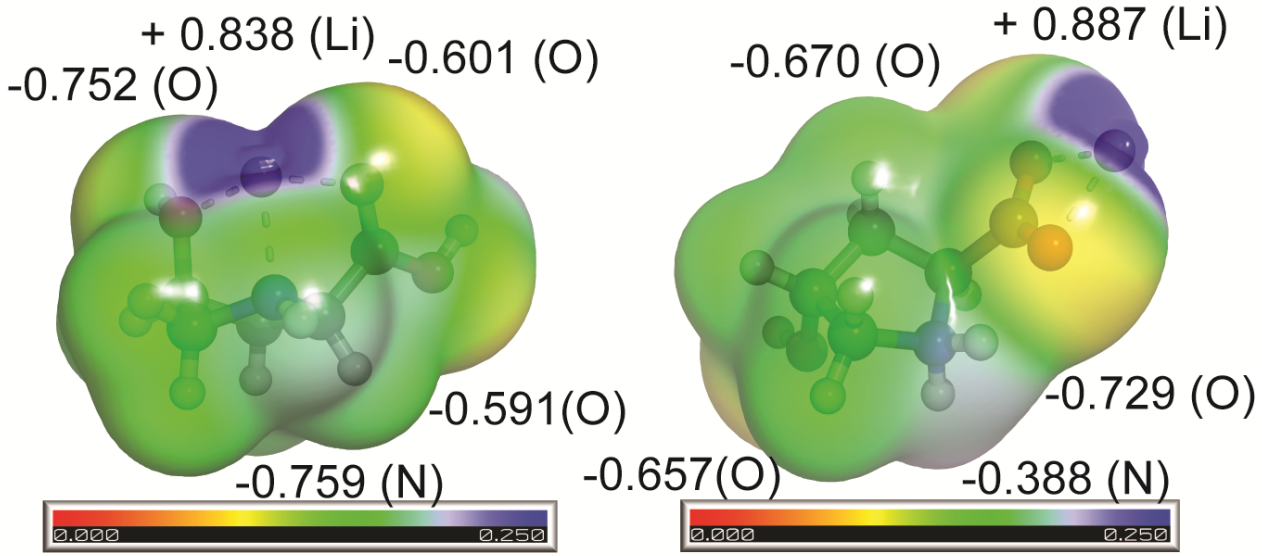
Effect on Electrostatic Potential / Charge Distribution, Dipole Moment



B3LYP/6-31++G(d,p)

δ = 2.08 Debye

δ = 3.45 Debye

- Ions with different charge densities will have differing ion-dipole interactions with drift gas
- *Trans*- form will should greater interaction with more polarizable drift gas and be less mobile

Analytical. Chem. **2015**, *87*, 3300.

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Lunch and Learn Workshop:

Small Molecule Geometry Optimizations and CCS Calculations

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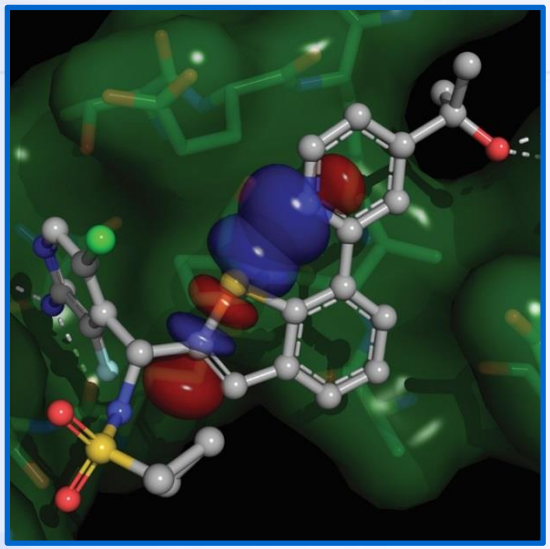
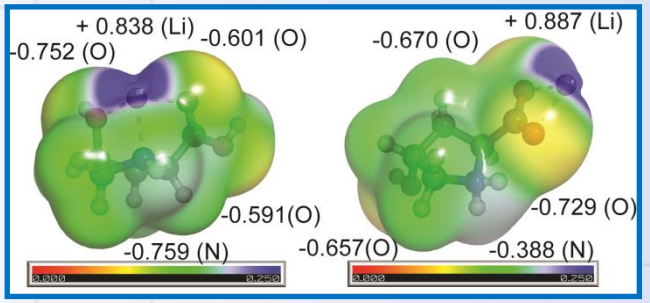
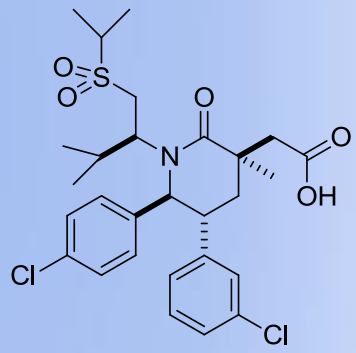


IUPUI

Carlos Larriba-Andaluz



Tawnya Flick
Iain Campuzano



Steve Olson
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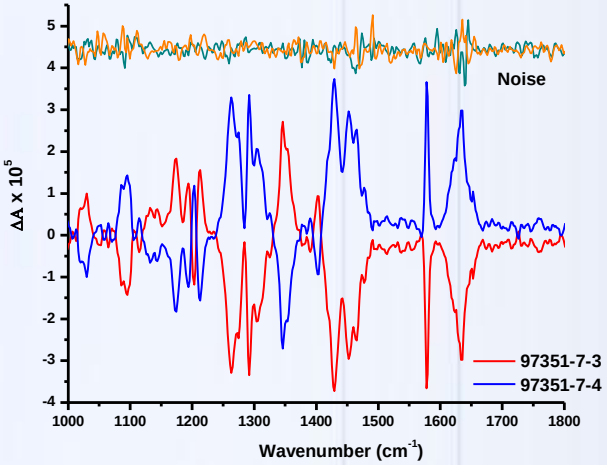
The MDM2 Team



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