IASOC XII

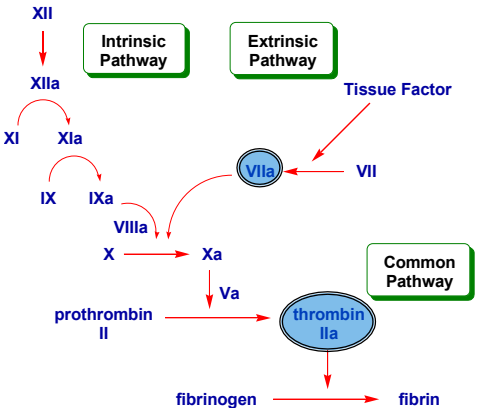
"Structure-based Organic Synthesis of Drug Prototypes"

Stephen Hanessian

University of Montreal, Canada
University of California, Irvine, USA

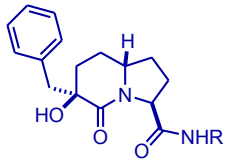
Thrombin and factor VIIa

- Members of the family of serine proteases
- Thrombin holds central position in the final steps of the blood coagulation cascade
- Factor VIIa holds a major role in the extrinsic pathway
- They both regulate hemostasis
- Lead to the conversion of Fibrinogen to Fibrin
- Stimulate platelets aggregation
- Current treatments → side effects

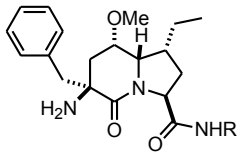


Reviews: *Clin. Appl. Thrombosis/Hemostasis* ***2001****, 7*, 195-204
Am. Heart J. **2001**, *142*, S3-8
Exp. Opin. Invest. Drugs **2001**, *10*, 845-864
Current Medicinal Chemistry **1998**, *5*, 289-304

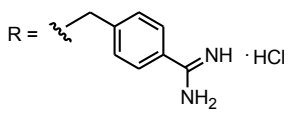
Biological Results

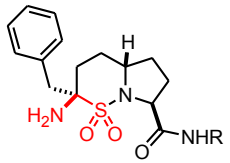


thrombin: 18 nM

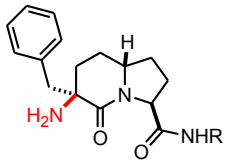


thrombin: 210 nM

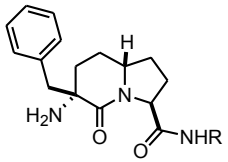




thrombin: 494 nM



thrombin: 4.7 nM



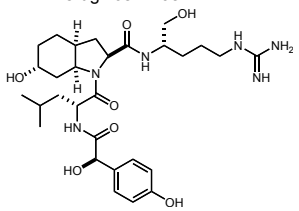
thrombin: 2 550 nM

S. Hanessian, E. Balaux, D. Musil, L. L. Olsson, I. Nilsson, I.*Bioorg. Med. Chem. Lett.* **2000**, *10,* 243;
S. Hanessian, E. Therrien, K. Granberg, I. Nilsson, *Bioorg. Med. Chem. Lett.* **2002** *12*, 2907;
S. Hanessian, H. Sailes E. Therrien, *Tetrahedron* **2003**, *59*, 7047;
S. Hanessian, A. Munro, H. Sailes E. Therrien, *J. Org. Chem.* **2003**, *68*, 7219.

Bioactive Compounds Produced by Cyanobacteria

- Isolated from the blue-green algae *Microcystis aeruginosa*
- Thrombin and trypsin inhibitors

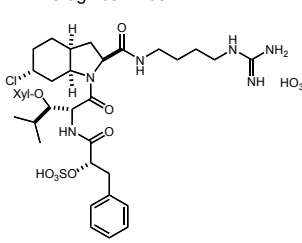
Aeruginosin 298-A



Trypsin IC_{50} 1.0 μM
Thrombin IC_{50} 0.5 μM

Microcystis aeruginosa

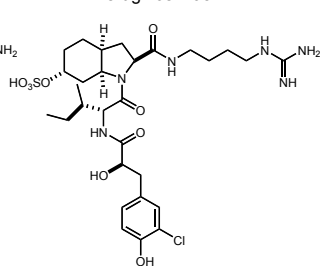
Aeruginosin 205-A



Trypsin IC_{50} 0.07 μM
Thrombin IC_{50} 0.1 μM

Oscillatoria agardhii

Aeruginosin 98A

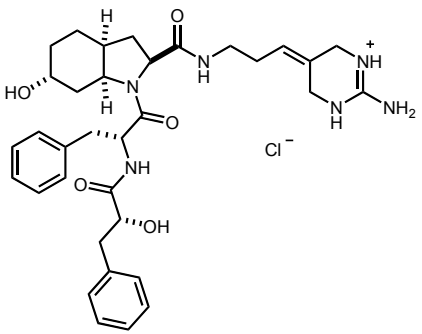


Trypsin IC_{50} 0.6 μM
Thrombin IC_{50} 7.0 μM

Microcystis aeruginosa

Reviews: H. Leusch, *et al Curr. Med. Chem.* **2002**, *9*, 179.
M. Murakami, *et al Tetrahedron* **1999**, *55*, 10971
M. Namikoshi, K. L. Rinehart, *J. Indust. Microbiol.* **1996**, *17*, 373.

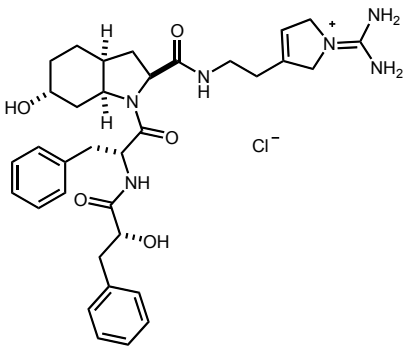
New Aeruginosins from *Oscillatoria agardhii*



Presumed Oscillarin

Boehringer Mannheim

WO 96/11941



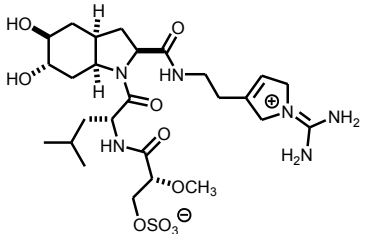
Oscillarin (D-Pla-D-Phe-L-Choi-Adc)

Boehringer Mannheim

WO 97/21725; U.S. 2002/0026034

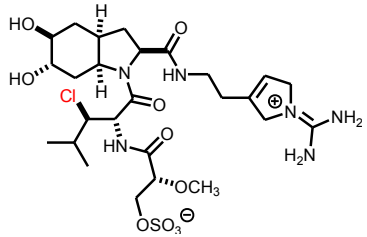
S. Hanessian et al, *J. Am. Chem. Soc.* **2004**, *126*, 6064.

Oceans Apart



Dysinosin A

Lizard Island, Australia



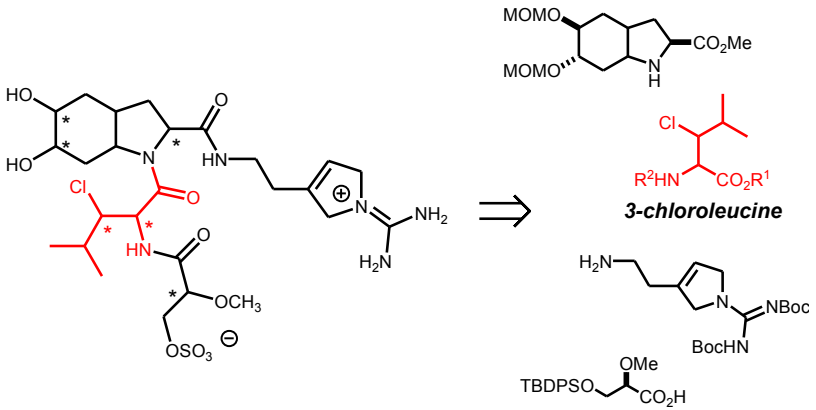
"Chlorodysinosin A"

Scripps Institute of Oceanography
La Jolla, CA

Pharmacia PCT WO03/051831.

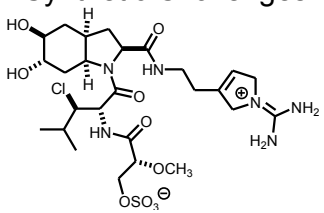
S. Hanessian et al, *J. Am. Chem. Soc.* **2002**, *124*, 13342;
S. Hanessian et al., *J. Am. Chem. Soc.* **2006**, *128*, 10491.

Disconnection



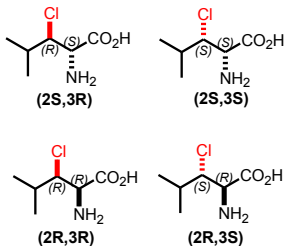
3-chloroleucine

Synthetic Challenges



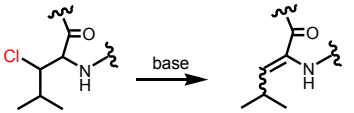
"chlorodysinosin A"

Which isomer of β-chloroleucine?



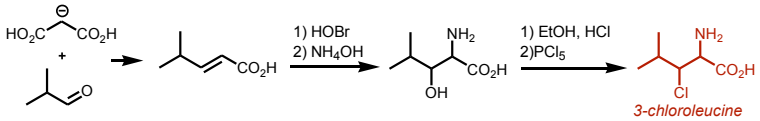
(2S,3R) **(2S,3S)** **(2R,3R)** **(2R,3S)**

Elimination!



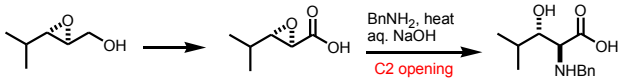
3-Chloroleucine and 3-Hydroxyleucine

One known synthesis of racemic amino acid (Shive et *al.*):

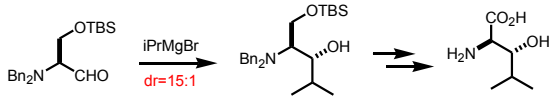


Shive, W. et al. *J. Org. Chem.* **1961**, *26*, 1167

3-Hydroxyleucine synthesis:

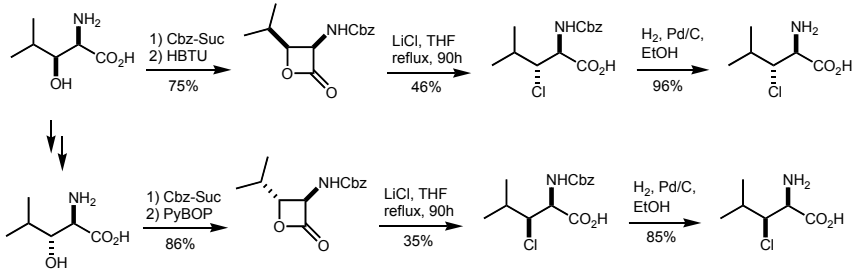


Caldwell, C. G. and Bondy, S. S. *Synthesis* **1990**, 35.



J. Zhu, et al. *J. Org. Chem.* **1998**, *63*, 1709.

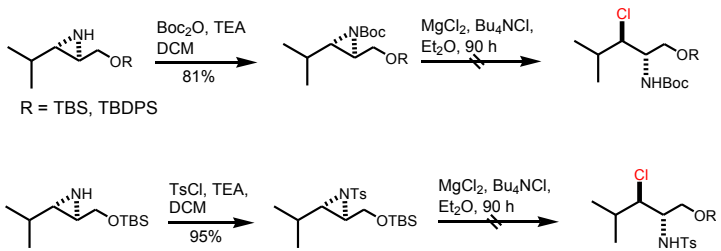
A Synthesis of 3-Chloroleucine



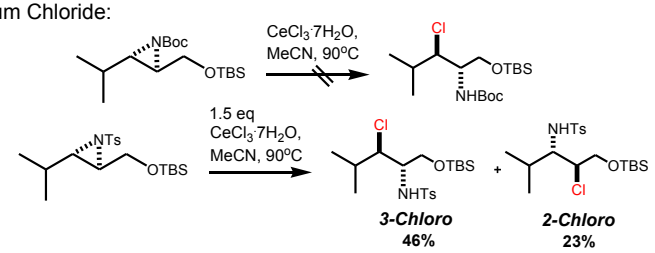
J. Bonjoch, et al. *Tetrahedron Lett.* **2006**, *47*, 3701.

Aziridine Opening

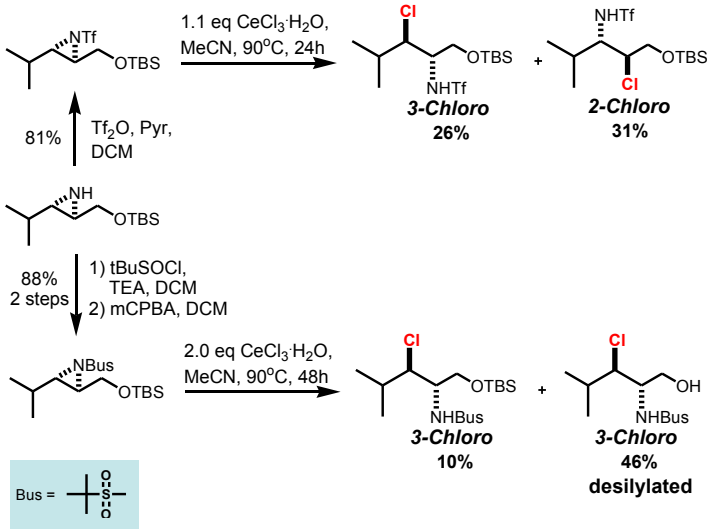
Attempted Cleavage with Magnesium Chloride:



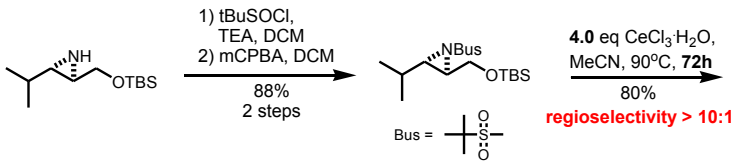
With Cerium Chloride:

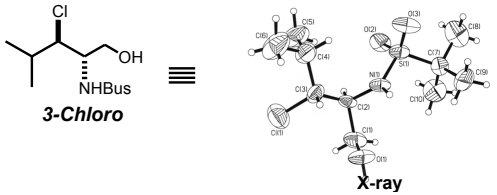


Opening of *N*-Sulfonyl Aziridines

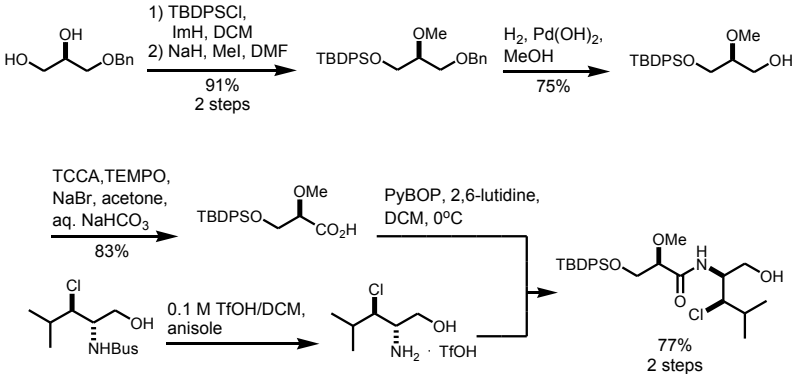


Regioselective Opening of *N*-Bus Aziridine

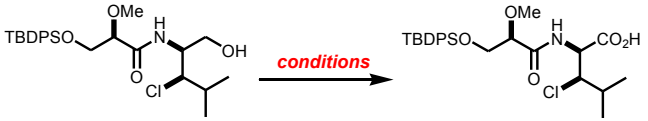




Assembly of the *N*-Terminal Fragment



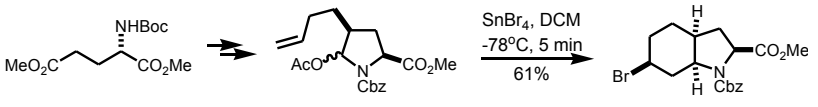
Oxidations

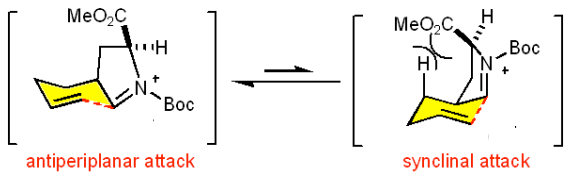


$NaClO_2$, NaClO, TEMPO, $MeCN/H_2O$	*no rxn*
$(COCl_2)$, DMSO, TEA, DCM	*decomposition*
$RuCl_3$, $NaIO_4$, $CCl_4/MeCN/H_2O$	42%
TCCA, TEMPO, NaBr, Me_2CO/H_2O	40% + (37% TBDPSO, OMe, CO_2H)
H_5IO_6, CrO_3, MeCN	75-83%

Reider, P. et al. *Tetrahedron Lett.* **1998**, *39*, 85323.

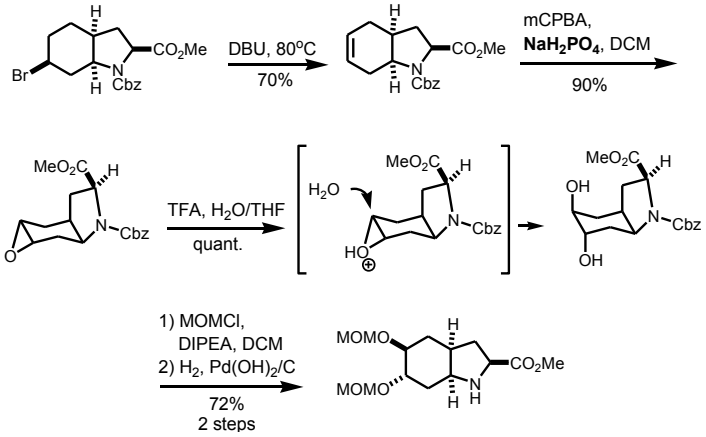
Octahydroindole Synthesis (Azonia-Prins)



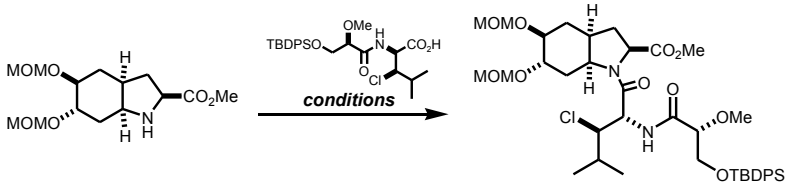


S. Hanessian, M. Tremblay, J. F. W. Petersen, *J. Am. Chem. Soc.* **2004**, 6064.

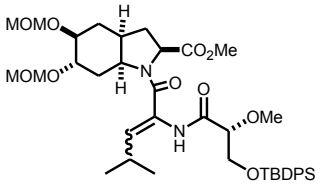
Octahydroindole Synthesis



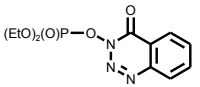
Coupling of the 3-Chloro Leucine Amide



HBTU/HOBT, DIEA, DMF	no product
EDC/HOBt, TEA, DCM	9%
PyBOP, TEA, DCM	15%
PyBOP, 2,6-lutidine, DCM	18-30%
EDC/HOBt, 2,6-lutidine, DCM	17%
BOPCl, 2,6-lutidine, DCM	15%
PyAOP, 2,6-lutidine, DCM	22%
DEPBT, 2,6-lutidine, DCM	57%

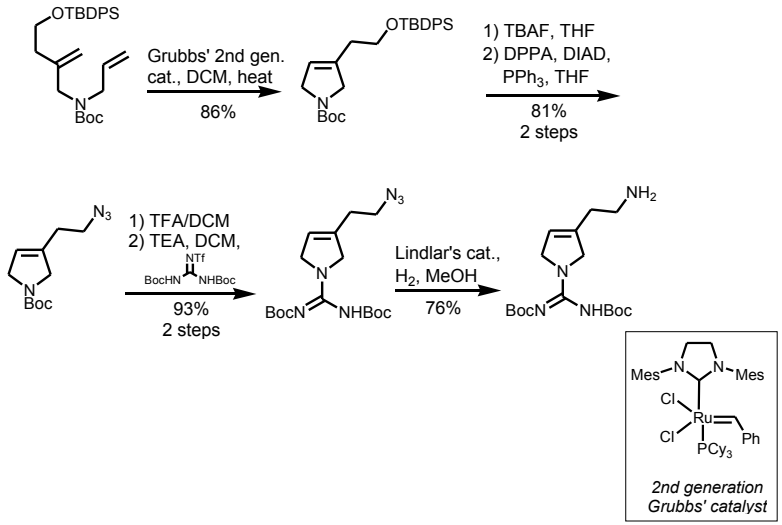


elimination product!

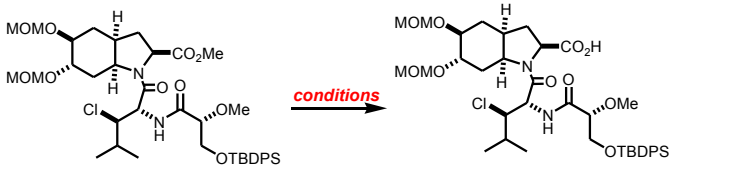


DEPBT

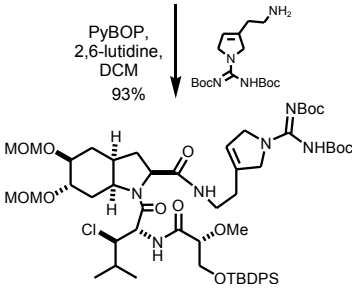
Synthesis of the Pyrroline Subunit



Assembly of Chlorodysinosin A

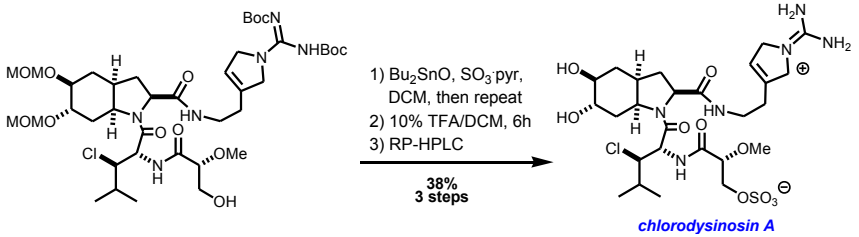


1M aq. NaOH/MeOH, 0°C	elimination
4 equiv. aq. LiOH/MeCN, 0°C	elimination
aq. NaOOH, MeOH, RT	no rxn
PLE, aq. Na_2HPO_4/acetone	no rxn
0.8M $MeTeAlMe_2$, PhMe, RT	MOM cleavage
20 eq. Me_3SnOH, DCE, 75°C, 48h	78-90%



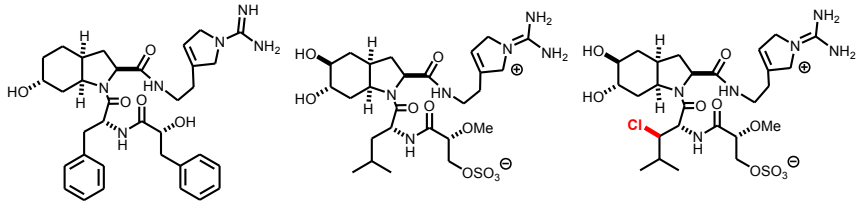
S. Hanessian, R. Margarita, A. Hall, S. Johnstone, M. Trembaly, L. Parlanti, *J. Am. Chem. Soc.* **2002**, 13342.

Completion of the Synthesis



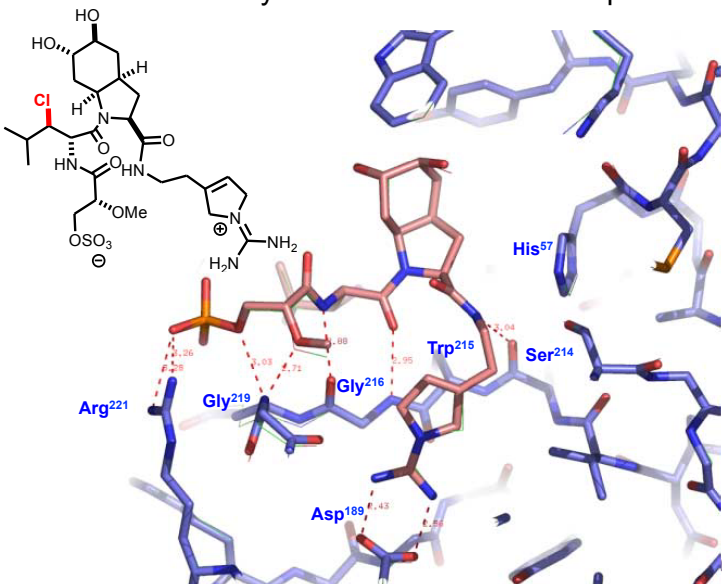
S. Hanessian, J. R. Del Valle, Y. Xue, N. Blomberg, *J. Am. Chem. Soc.* **2006**, *128*, 10491

The "Magic" Chlorine Atom?

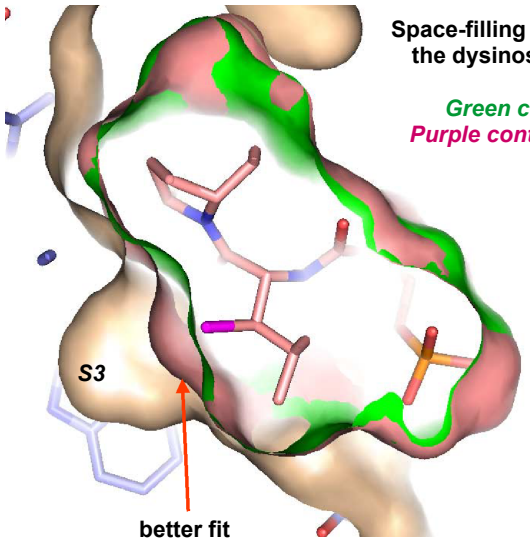


IC_{50}	*oscillarin*	*dysinosin A*	*"chlorodysinosin A"*
Thrombin	28 nM	46 nM	5.7 nM
Factor VIIa	3.9 μM	326 nM	39 nM
Factor Xa	-	5 μM	1.5 μM
Factor XIa	-	18 nM	4 nM

Chlorodysinosin A-Thrombin Complex



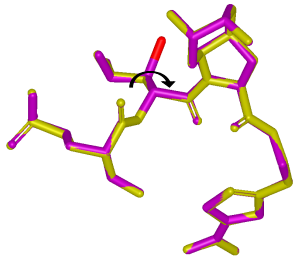
Topochemical Effect



Space-filling structural comparison of the dysinosins bound to thrombin

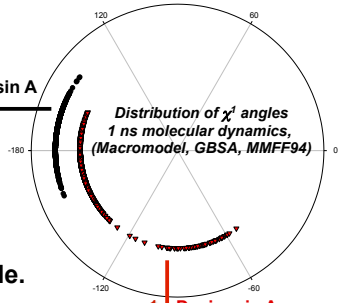
Green contour = Dysinosin A
Purple contour = ChlorodysinosinA

Conformational Effects

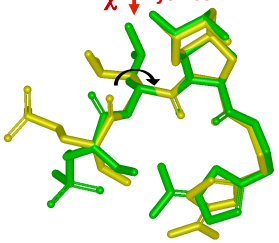


Chlorodysinosin A

χ^1



χ^1 Dysinosin A



- **3-Cl substituent restricts the χ^1 angle.**
- **3-Cl increases lipophilicity and hydrophobic interaction with the enzyme pocket.**
- **3-Cl fills the S3 site with release of water from the enzyme pocket (an entropic gain).**