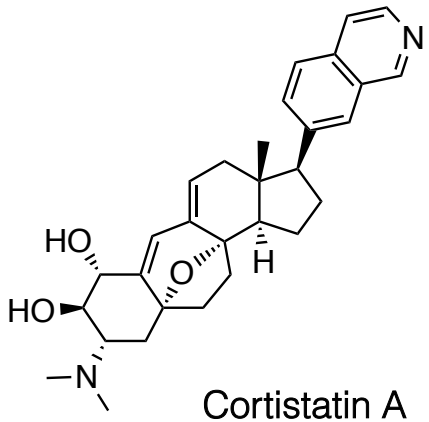
Current Literature

Development of Highly Potent and Selective Steroidal Inhibitors and Degraders of CDK8

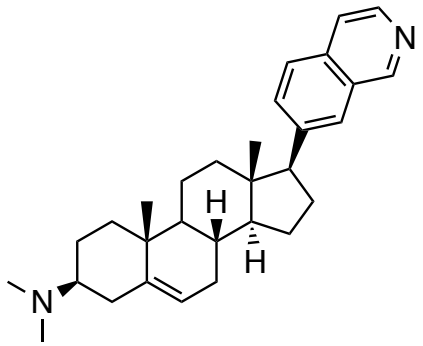
ACS Med. Chem. Lett. 2018, ASAP



Cortistatin A
16-30 steps; 0.012-2%
CDK8 Enzyme IC_{50}: 15 nM

Rational Drug Development

simplification

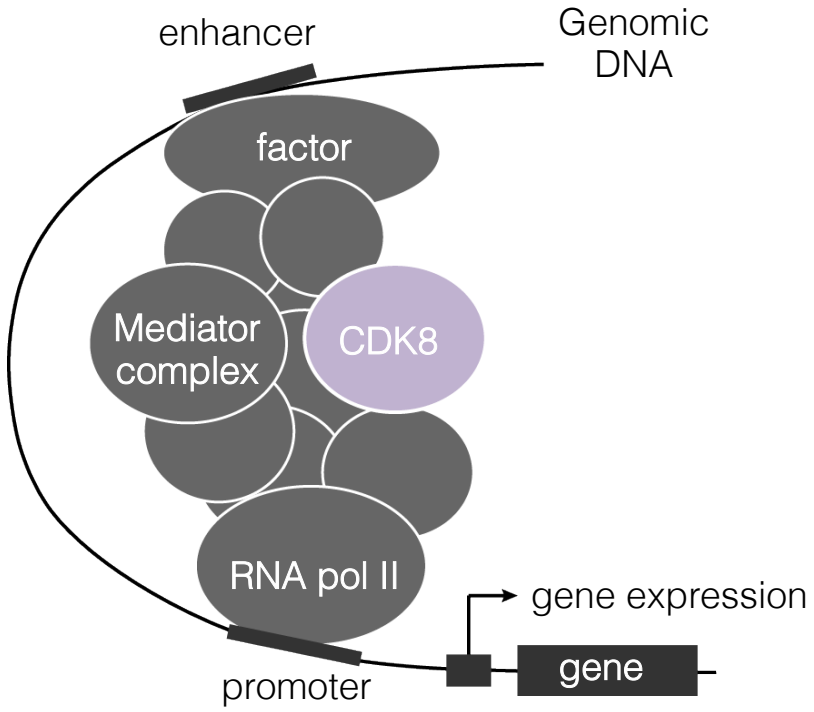


JH-VIII-49
8 steps; 33%
CDK8 Enzyme IC_{50}: 16 nM

Evan Carder
Wipf Group Current Literature
May 12, 2018

Cyclin-dependent kinase 8 (CDK8)

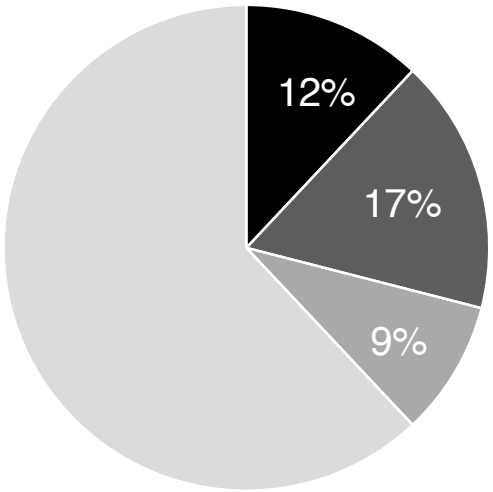
- Serine and threonine protein kinase
- Component of the mediator complex – multiprotein assembly compromising up to 30 subunits that regulates cell cycle and gene transcription
- Demonstrates both positive and negative regulation of transcription
- Elevated expression is associated with enhanced mortality colorectal, breast, and ovarian cancer
- Knock down of CDK8 in cancer cells reduced cell proliferation, induced cell cycle arrest, and promoted apoptosis
- Inhibiting CDK8 may be a promising approach for cancer therapy.

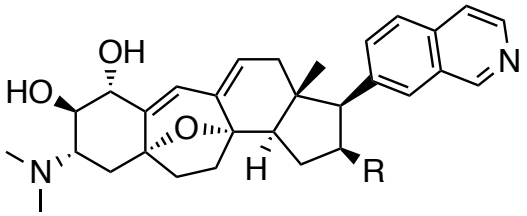


Genome Biol. **2014**, 15, 122.
Nature **2008**,455, 547.

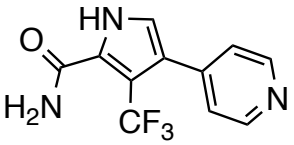
Target modulation - where to begin?

Small-molecule anticancer drugs
(1940-2014; n = 136)

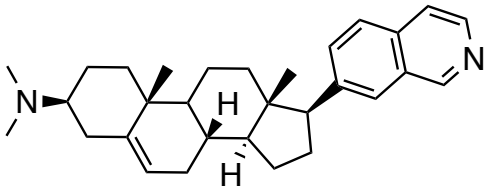




Cortistatin A



16

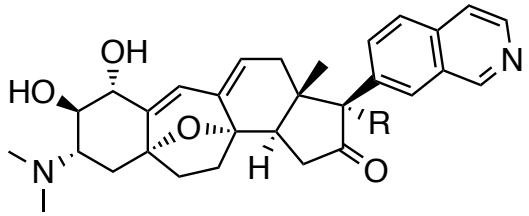


JH-VII-49
(this work)

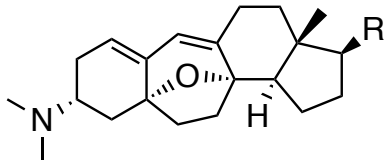
N	Unaltered natural product – semi or total synthetic
S	Synthetic – found by random screening
S*	Synthetic – natural product pharmacophore

J. Nat. Prod. **2016**, 79, 629; *Bioorganic & Medicinal Chem. Lett.* **2017**, 27, 4488; *ACS Med. Chem.* **2018**, ASAP

Cortistatin natural products

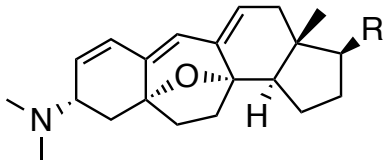


cortistatin C: R = H
cortistatin D: R = OH

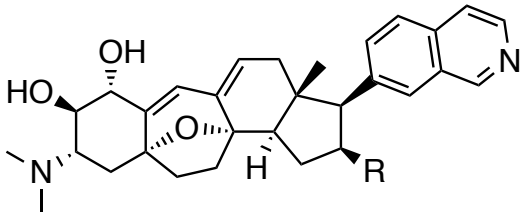


cortistatin E: R = S_1
cortistatin G: R = S_2

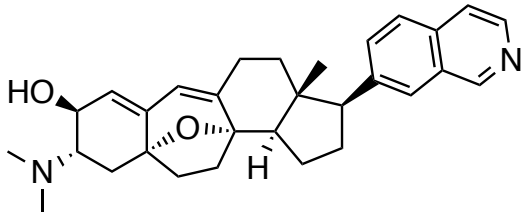
cortistatin H: R = S_3
cortistatin K: R = S_4



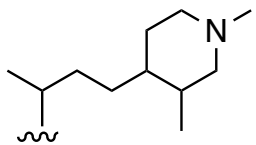
cortistatin F: R = S_1
cortistatin J: R = S_4



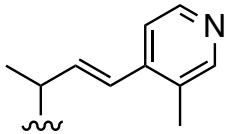
cortistatin A: R = H
cortistatin B: R = OH



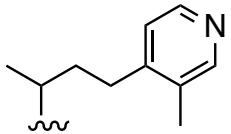
cortistatin L



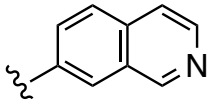
S_1



S_2



S_3



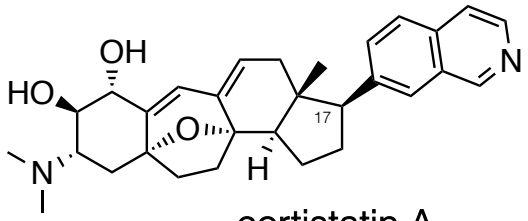
S_4

J. Am. Chem. Soc. **2006**, 128, 3148; *Tetrahedron* **2007**, 63, 4074; *Tetrahedron Lett.* **2007**, 48, 4485.

Cortistatin A

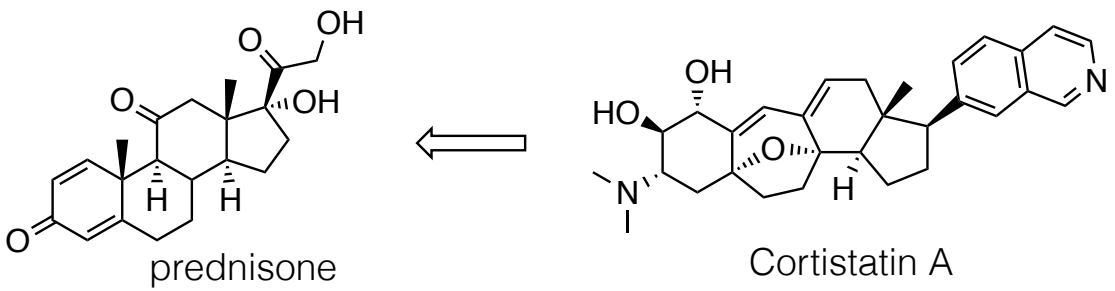
- Family of 11 steroidal alkaloid
- 9-(10,19)-*abeo*-androstane type
- High affinity binder to CDK8 (Kd = 17 nM)
- Active against human umbilical vein endothelial cells (HUVEC)
- Attractive lead compounds for drug discovery as an anti-angiogenic cancer chemotherapy
- Semi, formal, and total synthesis:

  Baran *et. al. J. Am. Chem. Soc.* **2008**, 130, 7241.
  Shair *et. al. J. Am. Chem. Soc.* **2008**, 130, 16864.
  Nicolaou *et. al. Angew. Chem. Int. Ed.* **2008**, 47, 7310.
  Hirama *et. al. Tet. Lett.* **2009**, 50, 3277–3279.
  Myers *et. al. Nat. Chem.* **2010**, 2, 886.

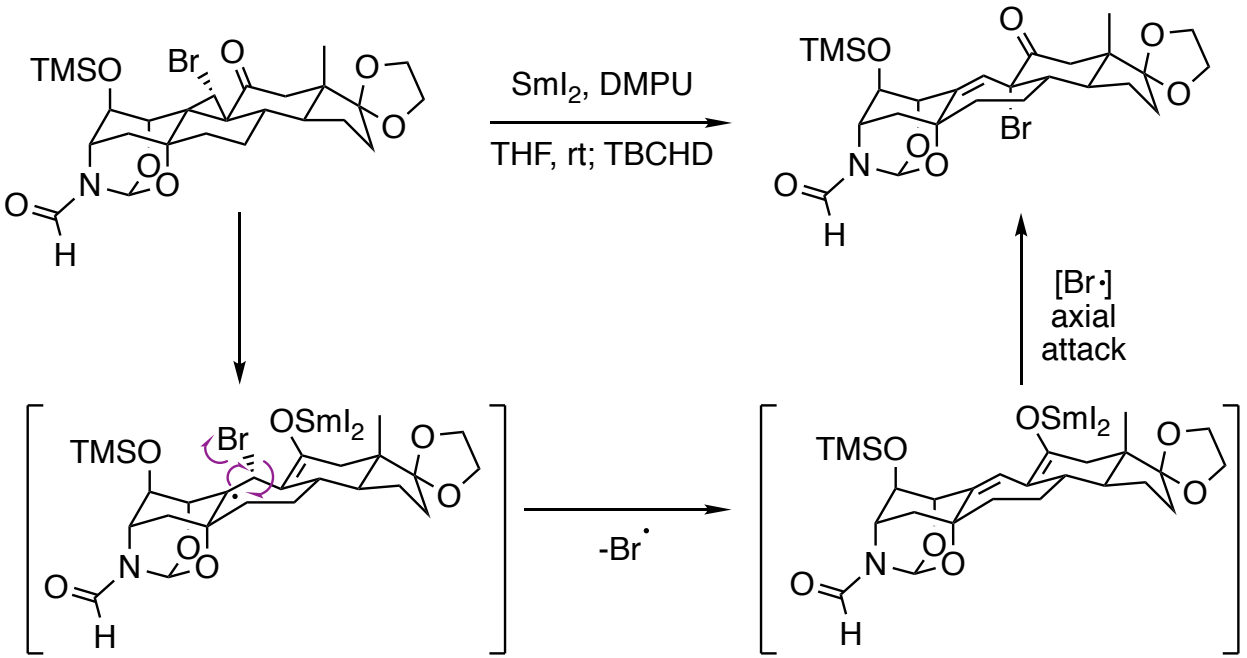


cortistatin A

Baran *et. al.* semi-synthesis

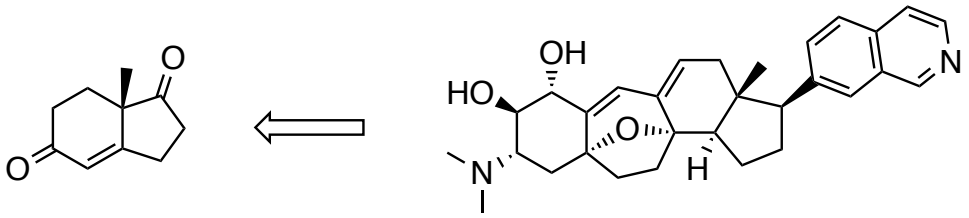


Key step:



J. Am. Chem. Soc. **2008**, 130, 7241.

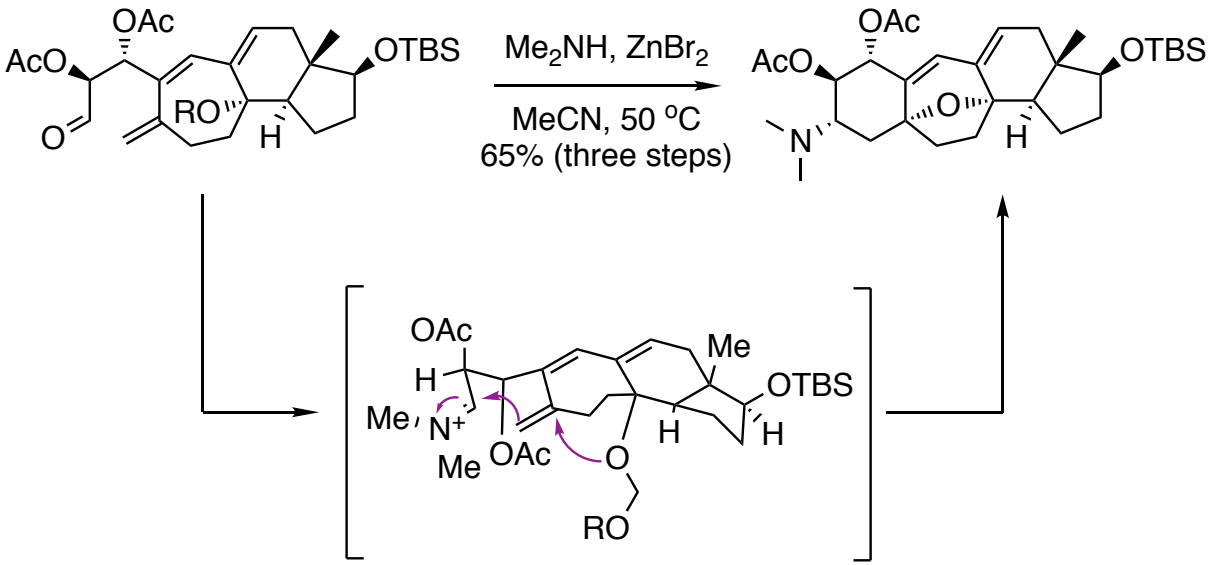
Shair *et. al.* synthesis



Hajos-Parrish ketone

Cortistatin A

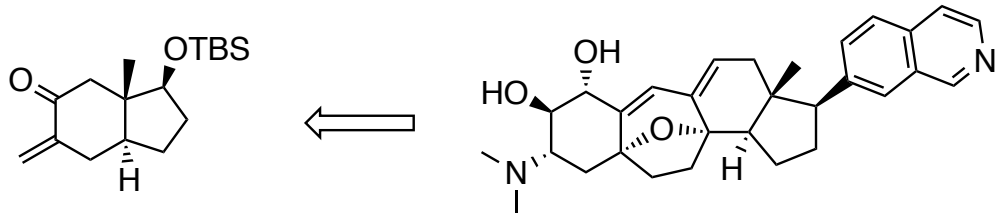
Key step:



Aza-Prins/transannular cyclization

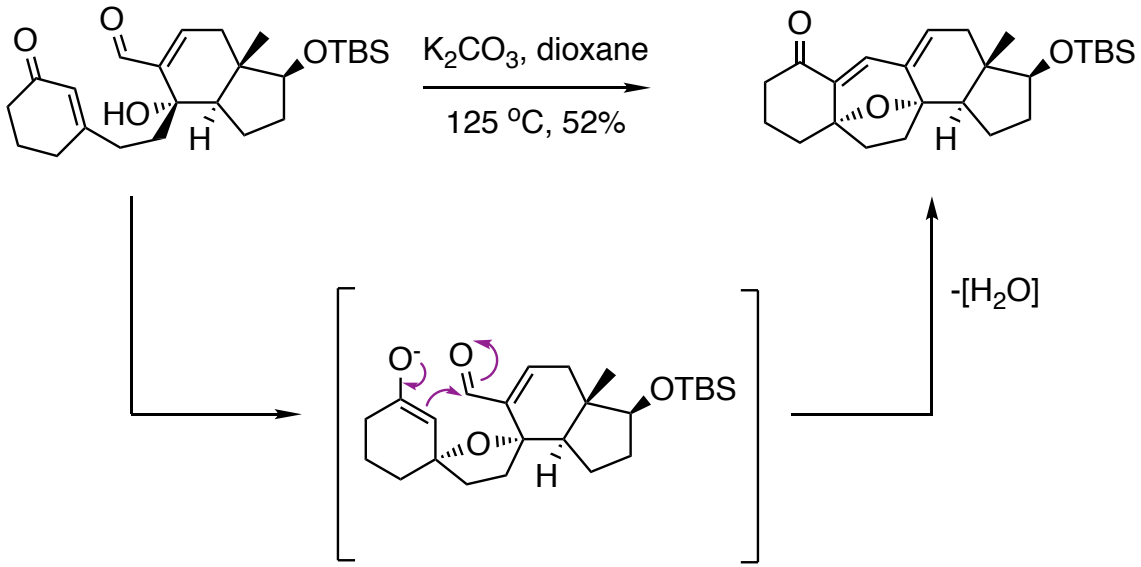
J. Am. Chem. Soc. **2008**, 130, 16864.

Nicolaou *et. al.* synthesis



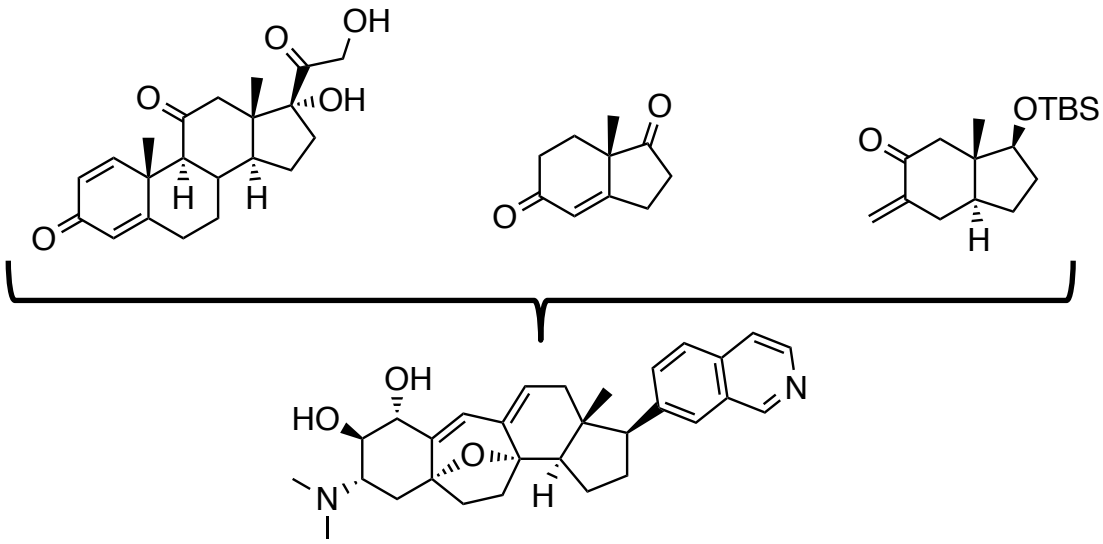
Cortistatin A

Key step:



1,4-addition / Adol / dehydration

Angew. Chem. **2008**, 120, 7420.



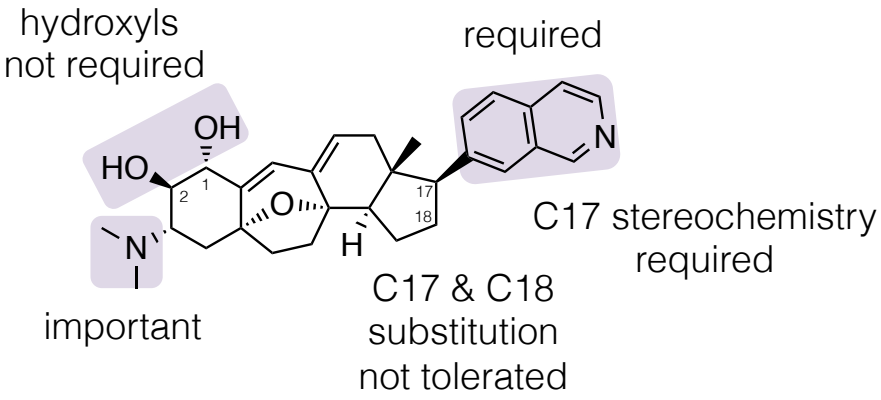
cortistatin A
16-30 steps; over yields: 0.012-2%

Current Work

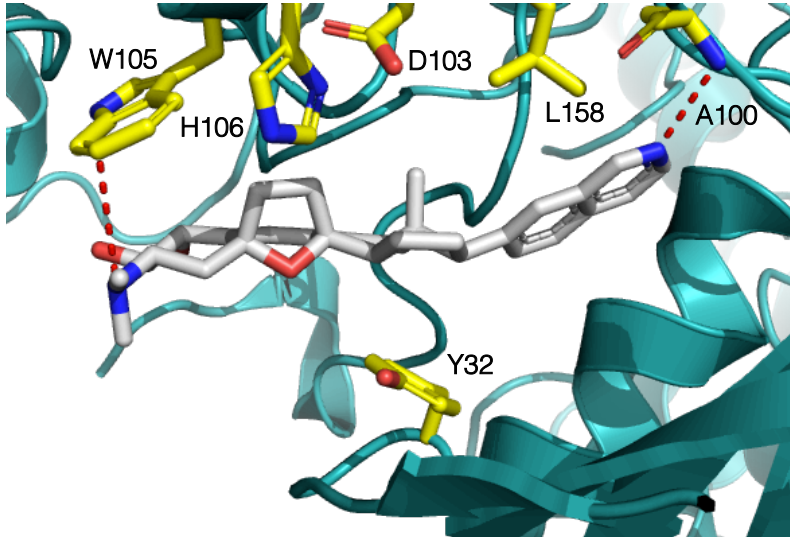
Initiate a campaign to design simple, more easily prepared CDK8 inhibitors based on a steroid scaffold that would be more convenient for large-scale synthesis.

Structural Analysis

Structure-activity relationship



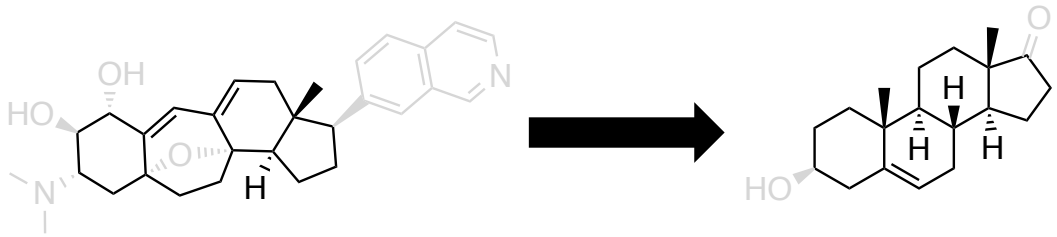
Cortistatin A-CDK8 co-crystal structure



PDB: 4CRL

Baran *et. al. Angew. Chem. Int. Ed.* **2009**, 48, 4328; Corey *et. al. J. Am. Chem. Soc.* **2009**, 131, 9014; Nicolaou *et. al. J. Am. Chem. Soc.* **2009**, 131, 10587.

Alternative steroid scaffold

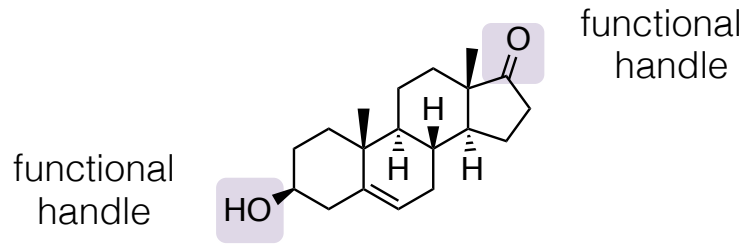


Cortistatin A core

DHEA core

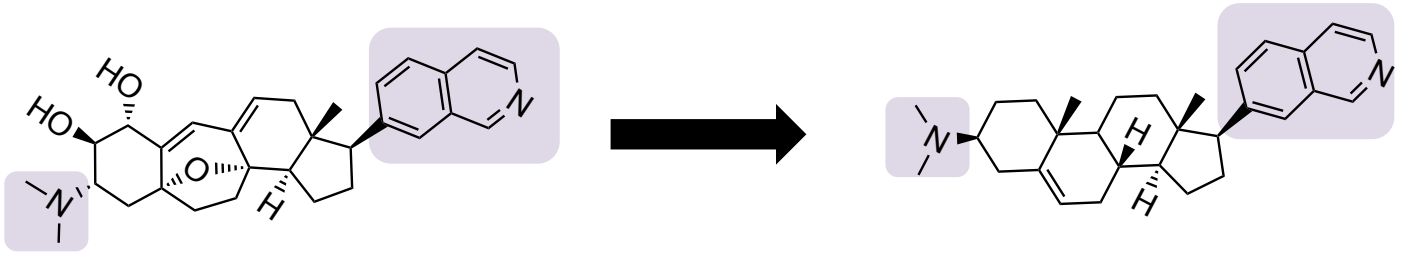
Dehydroepiandosterone (DHEA)

- Important endogenous steroid hormone
- Precursor in the synthesis of steroidal drugs; e.g. abiraterone acetate (Zytiga) – treatment of CRPC
- Commercially available – 100 g/$150.00 (ArkPharm)

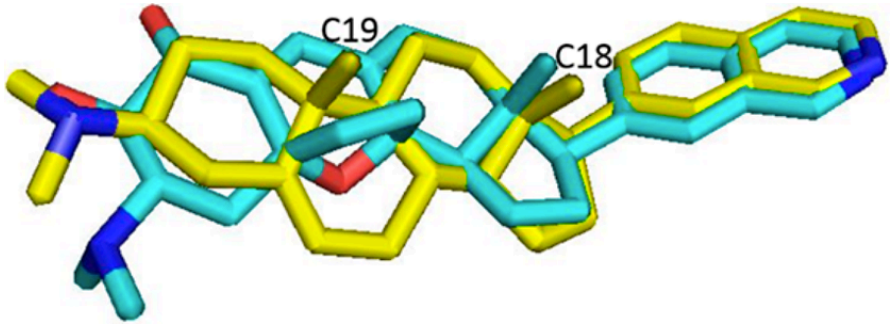


DHEA

Rational drug design



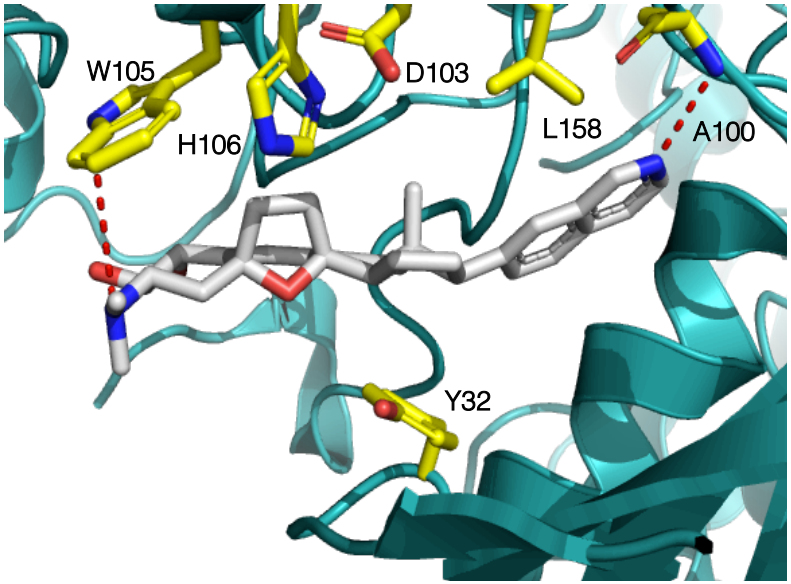
Design of DHEA derivative



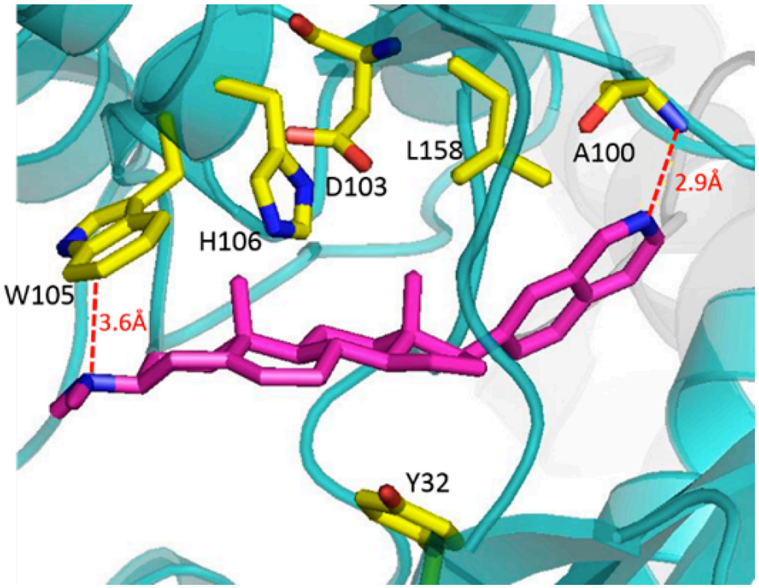
3D structure overlay of cortistatin A and DHEA derivative

Binding mode comparison

Cortistatin A-CDK8 co-crystal structure

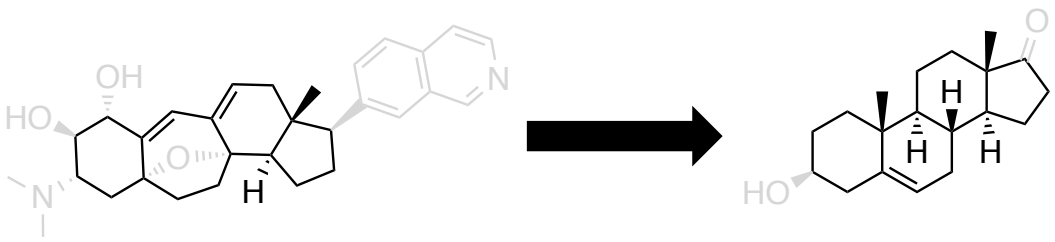


DHEA derivative-CDK8 model



Hypothesis

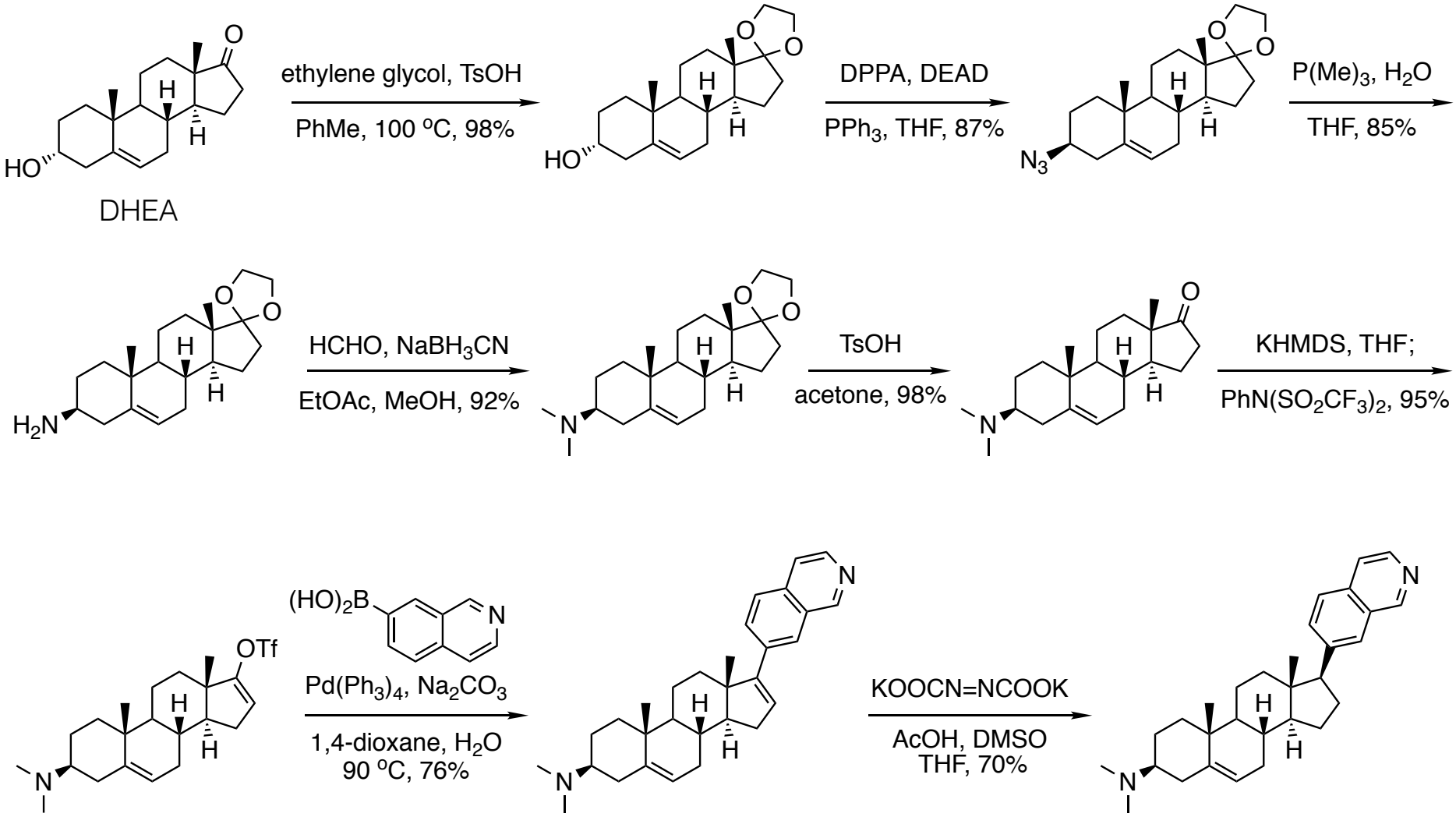
The replacement cortistatin A's core with dehydroepiandrosterone's (DHEA) carbocyclic framework will lead to a potent, selective, and scalable CDK8 inhibitor.

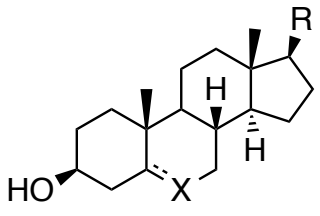


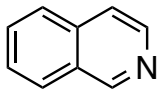
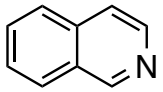
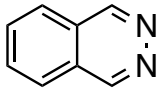
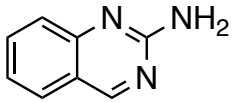
Cortistatin A core

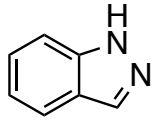
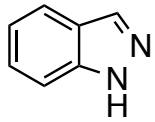
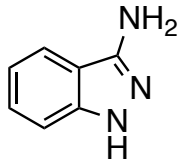
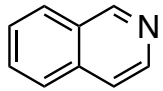
DHEA core

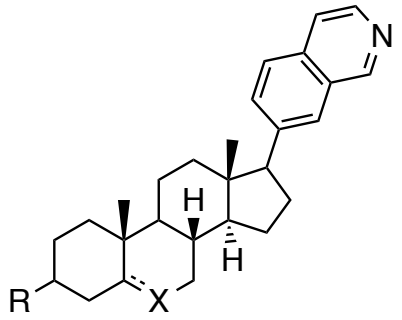
Representative synthesis



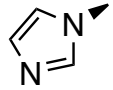
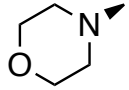
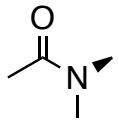


X	R	CDK8 IC_{50} (nM)
=		48
—		208
=		10,000
=		108

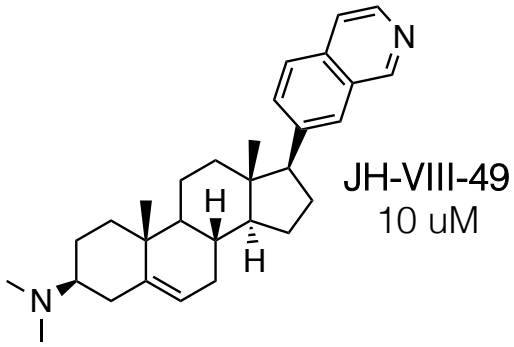
X	R	CDK8 IC_{50} (nM)
=		10,000
=		10,000
=		682
=		6000



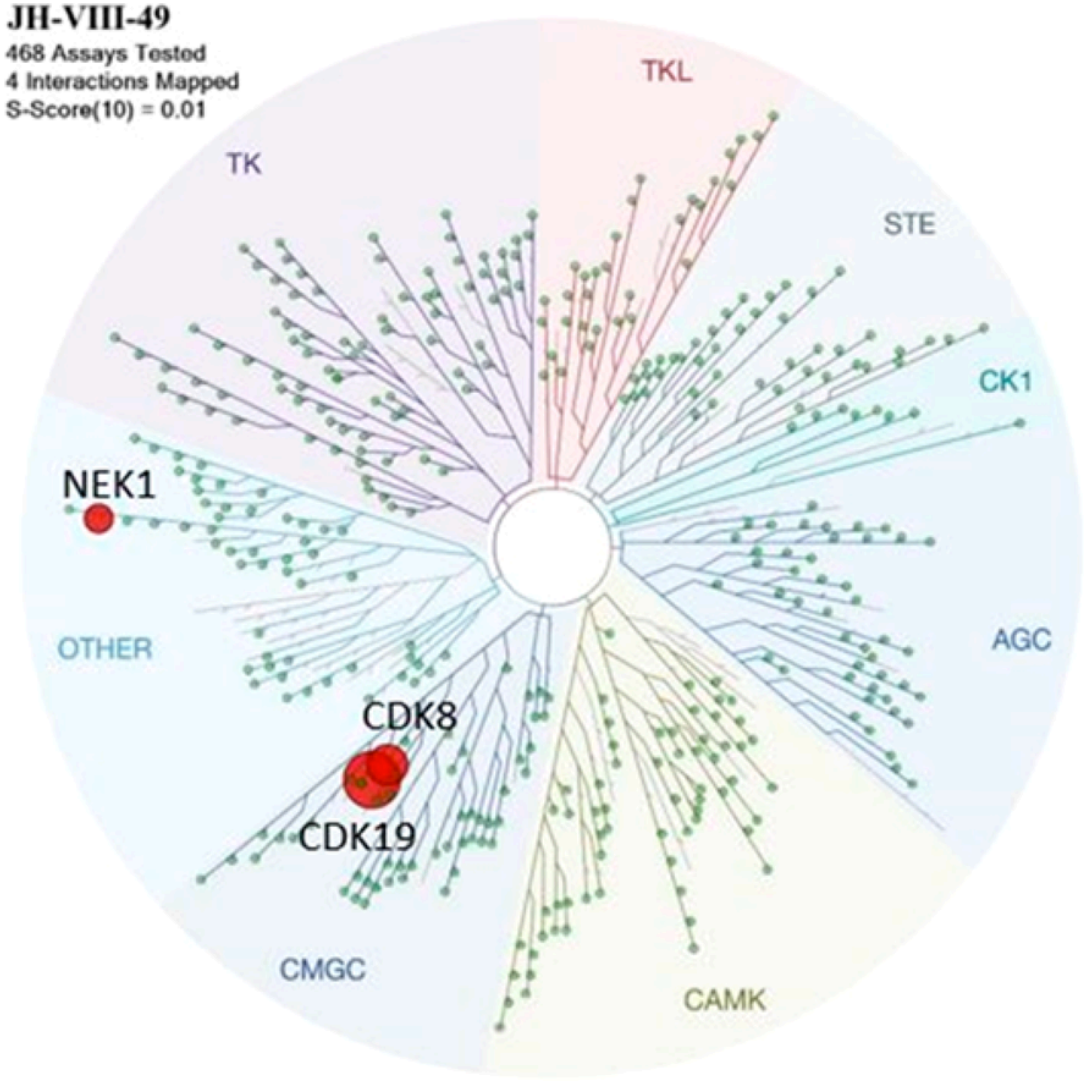
X	R	CDK8 IC_{50} (nM)
—		24
═		16
—		64
—		1320

X	R	CDK8 IC_{50} (nM)
—		24
—		120
—		34

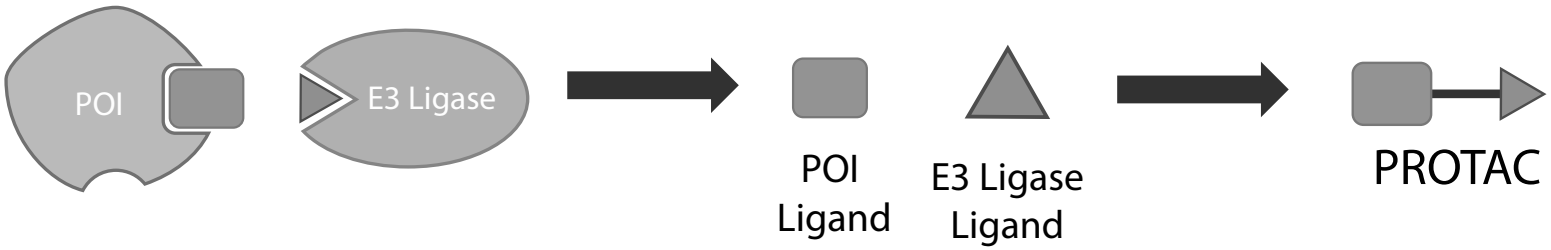
Kinase selectivity



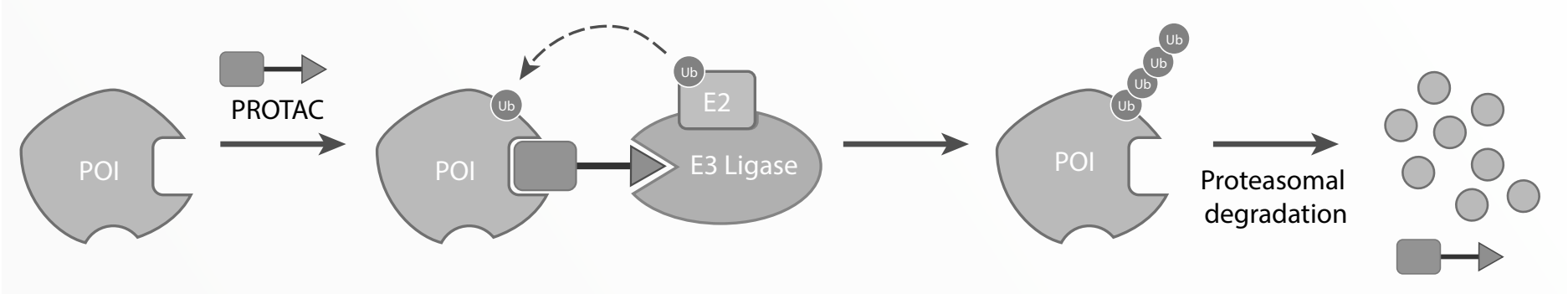
Kinase	% Control at 10 uM
CDK11	0.4
CDK8	2.9
PIKFVE	8.5
NEK1	8.7
RIOK2	11



Proteolysis Targeting Chimeras - PROTACs

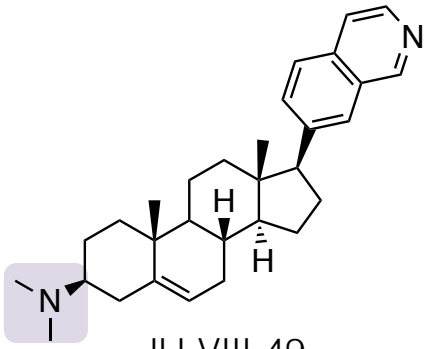


Small-molecule-mediated protein degradation:

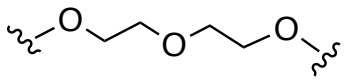


Natl. Acad, Sci. USA **2001**, 98, 8554.

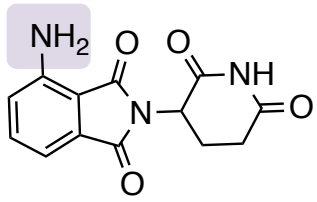
Design of PROTAC



JH-VIII-49
CDK8 Ligand

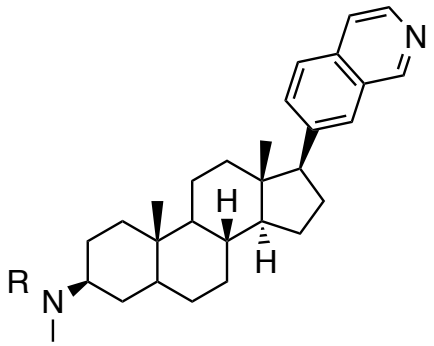


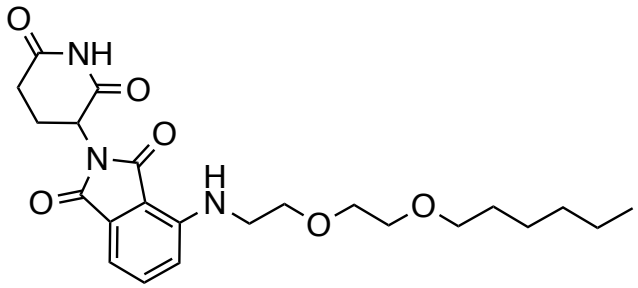
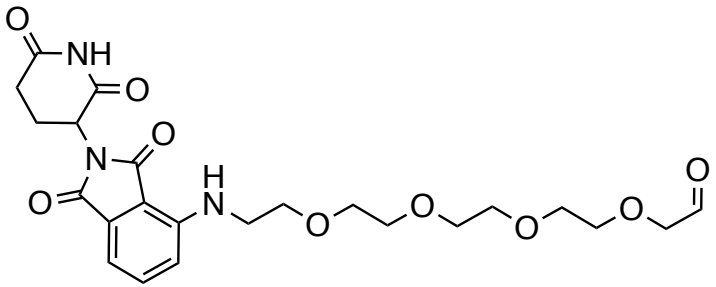
ethylene glycol linker

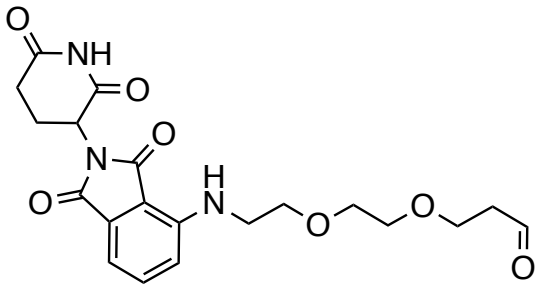
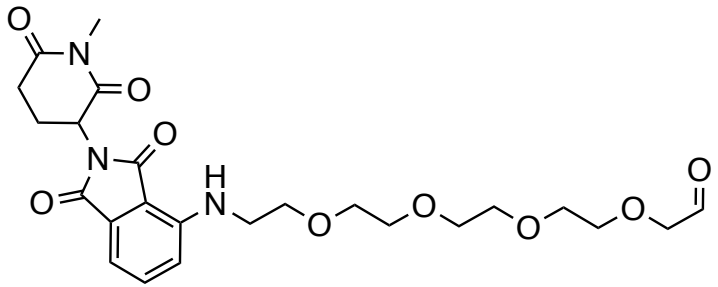


Pomalidomide
Cereblon (CRBN) ligand

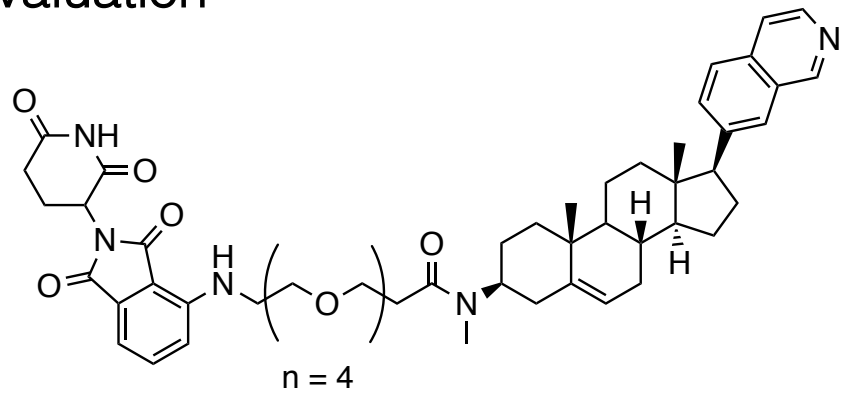
PROTAC



R	CDK8 IC_{50} (nM)
	N/A
	159

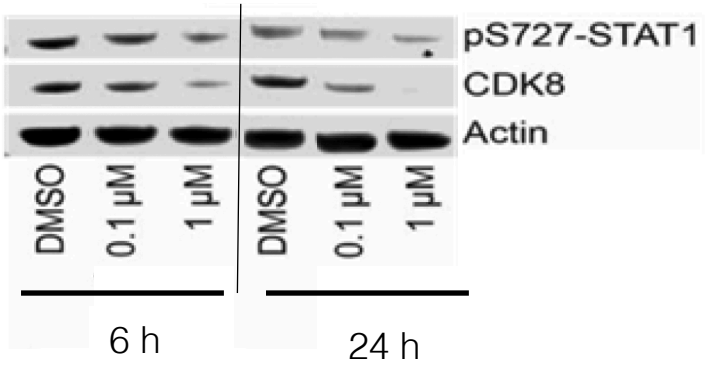
R	CDK8 IC_{50} (nM)
	192
	443

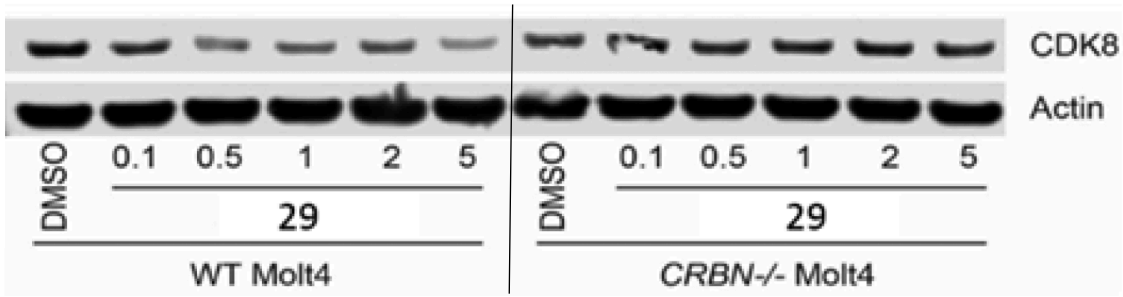
PROTAC biological evaluation



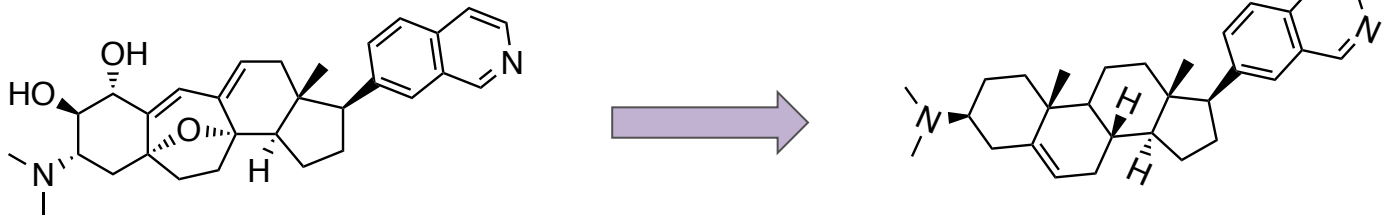
29

Jurkat cells





Summary



cortistatin A
16-30 steps; 0.012-2%
CDK8 Enzyme IC_{50}: 15 nM

JH-VIII-49
8 steps; 33%
CDK8 Enzyme IC_{50}: 16 nM

- Rationally designed and developed a potent and selective CDK8 kinase inhibitor
- 8 steps, 33% overall yield, and amenable for large-scale preparation
- Further developed a heterobifunctional CDK8 protein degrader

Continued work:

- Demonstrate synthesis on large scale
- Expand biological evaluation in both *in-vitro* and *in-vivo* models