PROTACS: MAKE "UNDRUGGABLE" TARGETS DRUGGABLE REVIEW AND SUPPORT FROM MEDICILON

Zhuo Mao, Xingquan Ma, and Xuedong Dai

Abstract

Proteolysis targeting chimeras (PROTACs) offer a fast and reversible chemical knock-down approach to control protein function. The impact of PROTAC platform has changed the landscape of drug discovery and development[1-3].

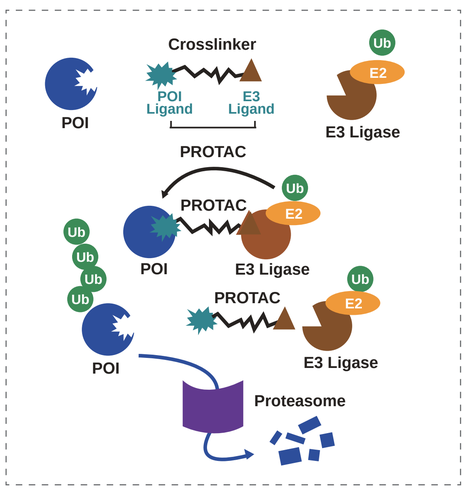
Medicilon's PROTAC drug discovery technology platform covers the currently popular target protein ligands. We have established a linker system with an extensive collection of bifunctional linkers. Together with our expanding E3 ubiquitin ligase binder library, we can efficiently synthesize a substantial amount of highly active PROTAC bispecific small molecules, which would have the potential to significantly facilitate ithe drug discovery and development process. In addition, Medicilon has established as well as improved the PROTAC biological screening and testing platform throughout the pre-clinical stages.

Medicilon's strong technical expertise and flexible service models allow individualized and customized projects ranging from sole chemical synthesis to in vitro and/or in vivo service, and to more comprehensive integrated package support. Our laboratories are US FDA and China NMPA accredited, and we will soon receive the European OECD GLP accreditation as well. We have successfully filled over 150 IND submissions worldwide. Medicilon is confident in providing efficient, cost-effective, and professional services to support our clients in reaching their drug development milestones.

Background

Proteolysis targeting chimeras (PROTACs), also known as bivalent chemical protein degraders, are heterobifunctional molecules that degrade specific endogenous proteins through the E3 ubiquitin ligase pathway. A PROTAC molecule structurally connects the protein of interest (POI)-binding ligand and the E3 ubiquitin ligase (E3) ligand through an appropriate linker.

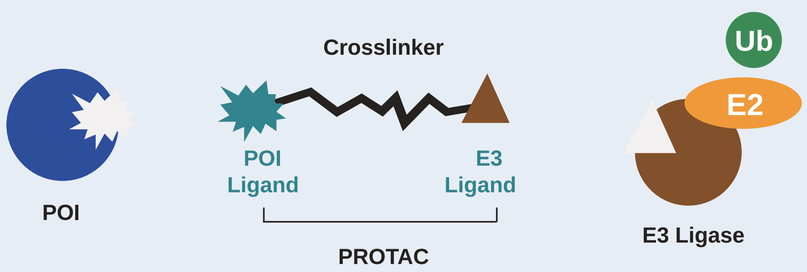
PROTAC technology is an effective tool to degrade endogenous target proteins through the ubiquitin-proteasome system (UPS)[1-3].



Medicilon PROTAC Technology Platform

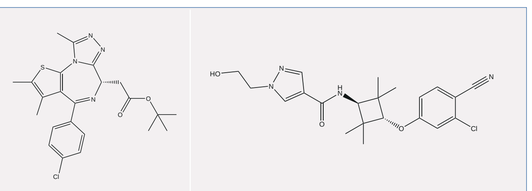
- >6 years PROTAC experience
- >300 Advanced linkers
- >1200 Chemists in total
- >200 PROTAC dedicated chemists
- >20 Ongoing projects
- >Fee for Service / FTE

PROTAC Synthesis Services



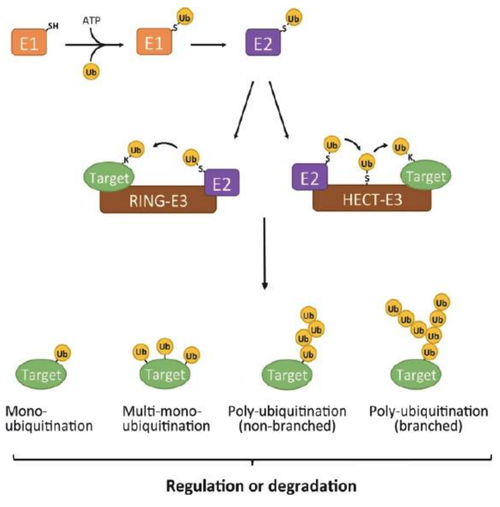
POI Ligands:

AR/ER/BTK/BRD4/ALK/CDK, etc. as well as proprietary target protein ligands.

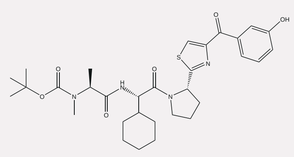


PROTAC Linkers: over 300 in total (for now) PEG chains/Alkyl chains/Hydrophobic chains

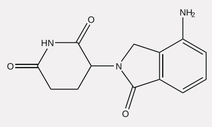
E3 Ubiquitin ligase binders:



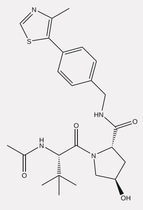
cIAP



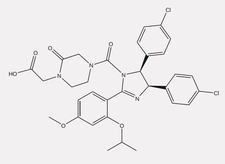
Cereblon



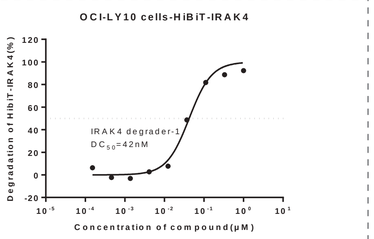
von Hippel-Lindau (VHL)



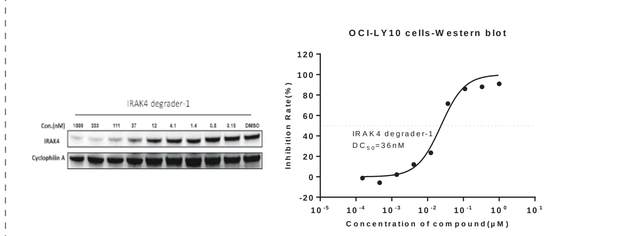
MDM2



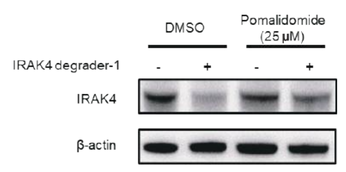
PROTAC *in Vitro* Evaluation

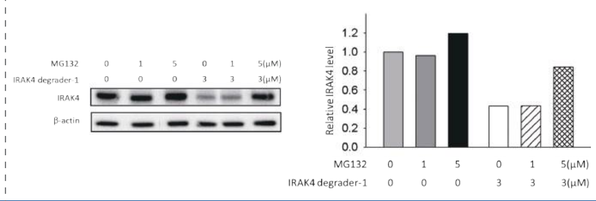


Percentage degradation of HiBiT-IRAK4 in HiBiT-IRAK4 overexpression OCI-LY10 cells.



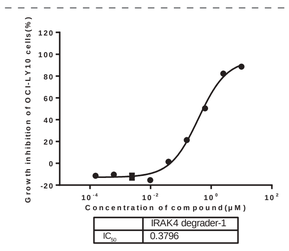
Western blot of anti-IRAK4 on OCI-LY10 cells treated with IRAK4 degrader-1.

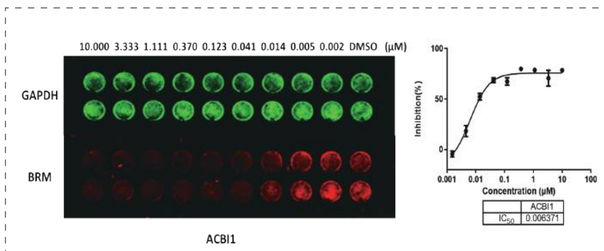




Pomalidomide(CRBN binder) competition experiment show that IRAK4 degrader-1 degrades IRAK4 through CRBN.

MG132 inhibition experiment show that IRAK4 degrader-1 degrades IRAK4 through proteasome pathway.





Growth inhibition effect of IRAK4 degrader-1 on OCI-LY10 cells.

In cell Western of BRM show that ACBI1(BRM degrader) degrades BRM in dose response manner.

Validated PROTAC Targets and Assays

Validated PROTACs	Targets	Validated assay
MP-A001	IRAK4	IRAK4 kinase assay; OCI-LY10 cytotoxicity assay; Western blot-based degradation assay; HiBiT-IRAK4 degradation assay (OCI-LY10 cells); HTRF-based IRAK4 degradation assay
ARV-110	AR	Western-based assay degradation assay; LNCAP cytotoxicity assay; AR dependent MDA-KB-2 cell reporter assay
Fulvestrant	ER	Western-based assay degradation assay; MCF-7 cytotoxicity assay; ER dependent MDA-KB-2 cell reporter assay
Lenalidomide	IKZF1	Western-based assay degradation assay; In-cell Western-based degradation assay
Lenalidomide	IKZF3	Western-based assay degradation assay; In-cell Western-based degradation assay
PROTAC-3	EGFR	EGFR kinase assay(EGFR-WT and mutants); Western-based degradation assay; NCI-H1975,HCC827 cytotoxicity assay; EGFR-HiBiT assay(HEK293 cells)
BSJ-04-132	CDK4	CDK4 kinase assay; Western-based degradation assay; HiBiT-CDK4 degaradation assay; Cytotoxicity assay
BSJ-04-132	CDK6	CDK6 kinase assay; Western-based degradation assay; HiBiT-CDK6 degaradation assay; Cytotoxicity assay
ACBI-2	SMARCA2	BRD domain binding assay; Western-based degradation assay; HiBiT-SMARCA2 degradation assay; MV-4-11 cytotoxicity assay
ACBI-2	SMARCA4	BRD domain binding assay; Western-based degradation assay; HiBiT-SMARCA4 degradation assay
PROTAC KRAS G12C degrader-1	KRAS G12C	KRAS G12C/SOS1 binding assay; Western-based degradation assay

PROTAC *in Vivo* Evaluation

Pharmacology Models

- 140 Xenograft models
- 25 Orthotopic Xenograft models
- 30 Syngeneic models
- 5 Humanized mice models
- 5 Transgenic models
- 40 Central Nervous System models
- 20 Cardiovascular and Metabolic Disease models
- 10 Inflammation and Immune System models
- 10 Inflammation and Immune System models
- 5 Other disease models

Summary

Medicilon has accumulated rich PROTAC experience working on a wide range of popular target proteins with high affinity small molecules and small molecule fragment compound libraries, a wide range of E3 ubiquitin ligase binders, and an extensive collection of bifunctional linkers. We leverage our broad chemistry capabilities and capacity with fully integrated biological and pre-clinical validation of the candidate PROTAC molecules. In addition, Medicilon supports a combination of chemical and biological drugs. We offer a variety of FTE models, both for chemistry as well as biology / preclinical studies. This also includes global project management and electronic lab notebook sharing (eLNB).

Medicilon offers integrated preclinical R&D services, including chemistry, biology, pharmacodynamics evaluation, pharmacokinetics evaluation, and toxicology evaluation.

References

[1]Si-Min Qi, et al. PROTAC: An Effective Targeted Protein Degradation Strategy for Cancer Therapy. Front Pharmacol. 2021 May 7;12:692574.

[2] Galen Andrew Collins, et al. The Logic of the 26S Proteasome. Cell. 2017 May 18;169(5):792-806.

[3]Jared A M Bard,et al. Structure and Function of the 26S Proteasome. Annu Rev Biochem. 2018 Jun 20;87:697-724.