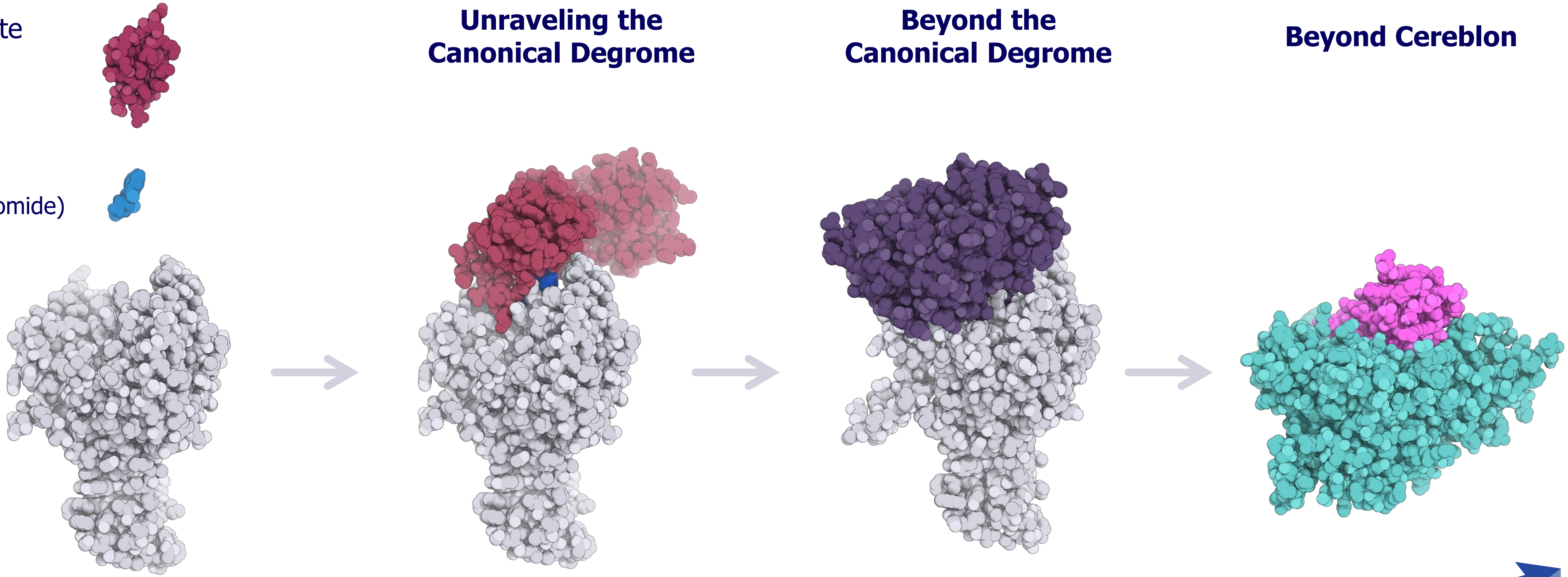
Teaching CRBN New Tricks

Georg Petzold | Keystone Symposia Proximity-Induced Therapeutics | January 23rd, 2024

Monte Rosa Therapeutics

Our Rational Approach to Unleash the Full Potential of MGDs



Neosubstrate
(e.g. IKZF1)

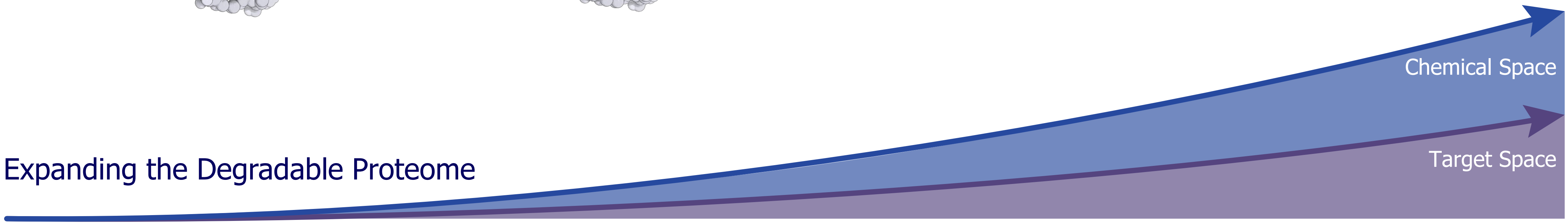
MGD
(e.g. Lenalidomide)

Cereblon

Unraveling the Canonical Degrome

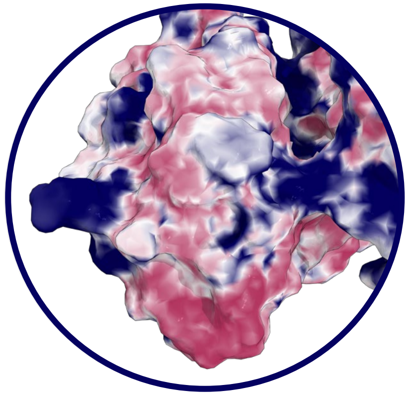
Beyond the Canonical Degrome

Beyond Cereblon



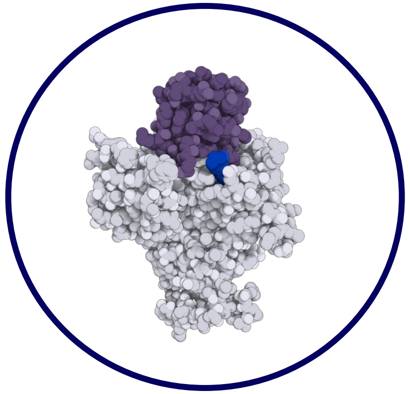
Expanding the Degradable Proteome

The QuEEN™ Engine – Unique Capabilities for MGD Discovery and Design



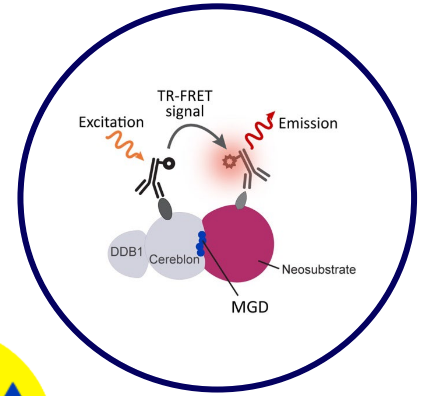
AI/ML

In silico degron & ternary complex discovery using proprietary AI-powered algorithms



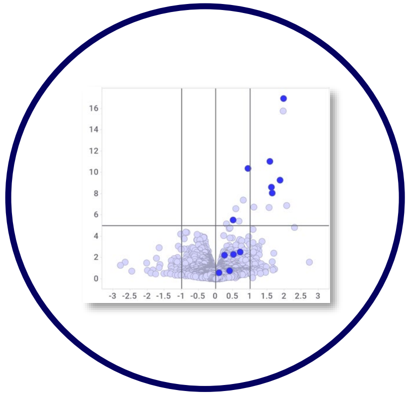
Structure-based Design

Proprietary database of >100 cryo-EM and crystal structures to enable rapid structure-based MGD design



Proximity Screening

Specialized suite of biochemical and cellular assays to assess proximity and degradation in high throughput



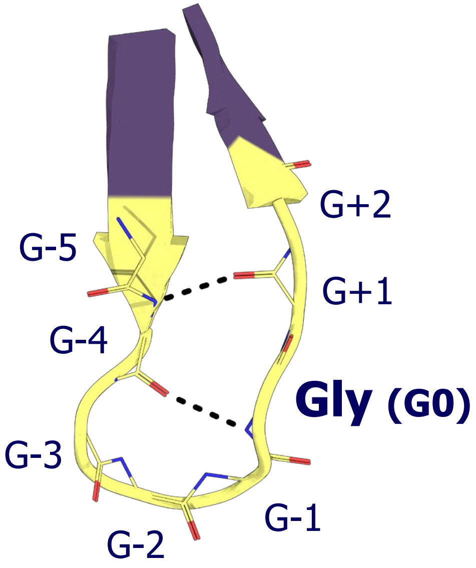
Proteomics

Integrated proteomics engine and database to identify novel targets and to explore cellular complex formation and degradation

Neosubstrates Engage CRBN Through a Shared Structural Motif

The G-loop degron: a β-hairpin α-turn with a conserved glycine

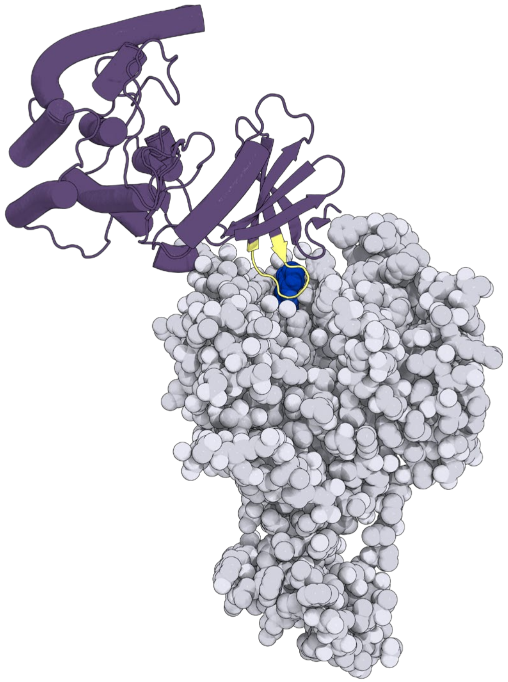
G-loop motif



CK1a	INITNGEE
GSPT1	VDKKSGEK
ZNF692	QCEICGFT
SALL4	VCSVCGHR
IKZF2	HCNQCGAS

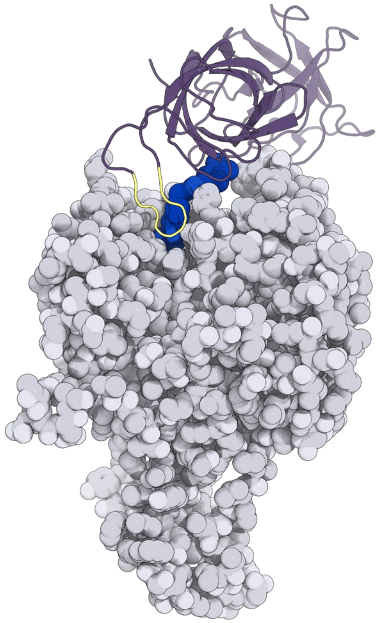
Structurally characterized neosubstrates in the public domain

CK1α



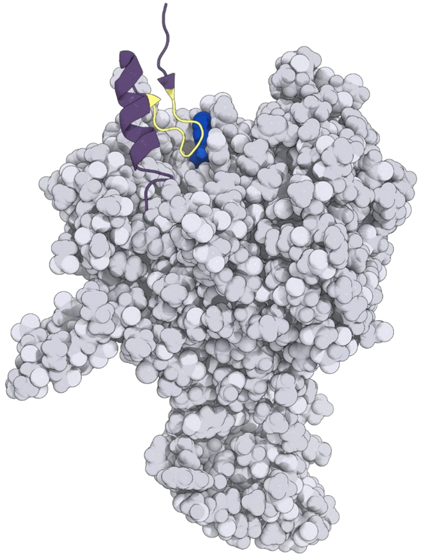
Petzold et al. 2016

GSPT1



Matyskiela et al. 2016

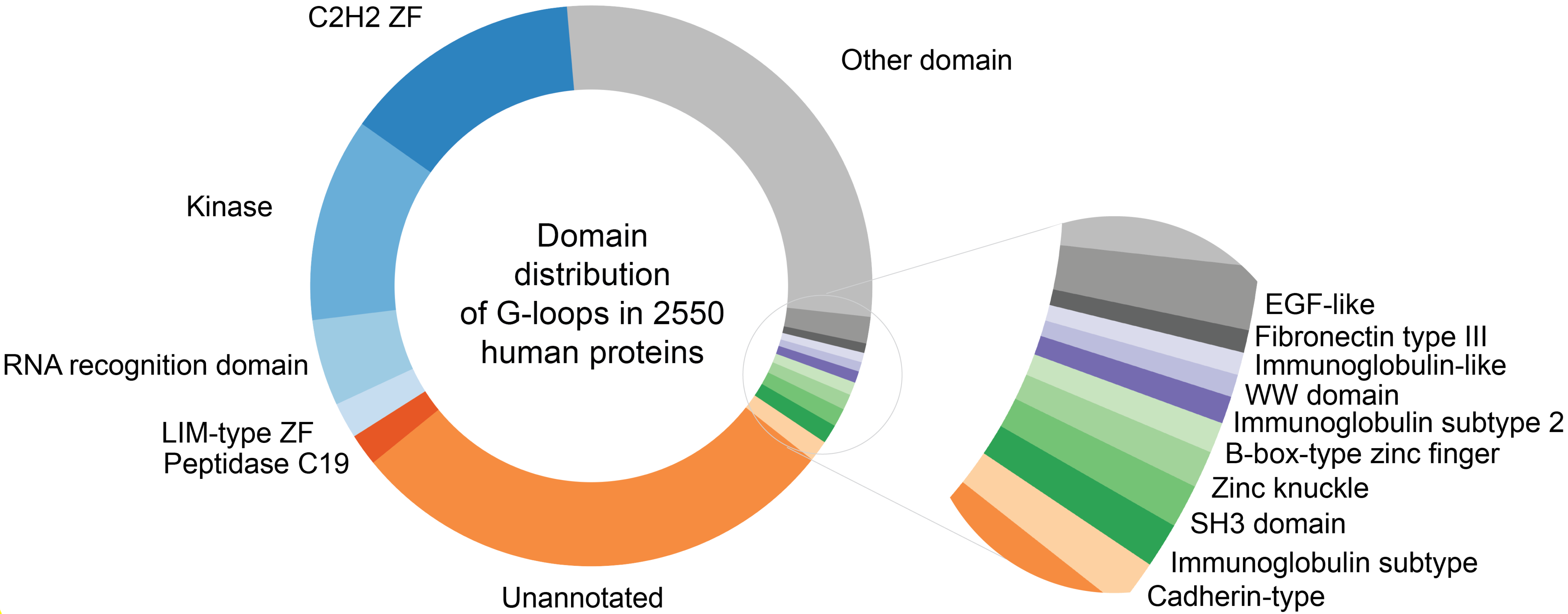
C2H2 ZFs



Sievers/Petzold et al. 2018

Mining the Human Proteome for Canonical G-loop Degrons

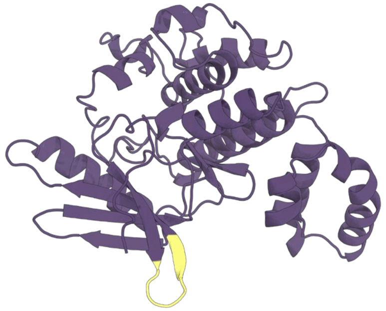
Over 2,500 human proteins contain a G-loop-like motif, many in undruggable domains



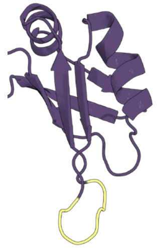
* Based on AlphaFold2 structures

The G-loop Degron Recruits Different Domain Types to CRBN

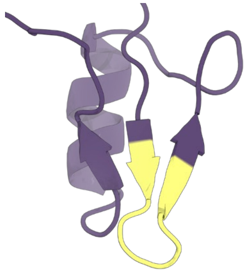
Mined G-loops validate in proximity readouts (NanoBRET, TurboID, ...)



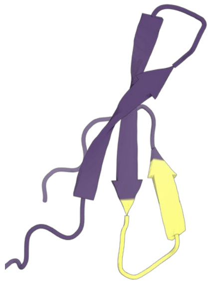
Kinases (beyond CK1α)



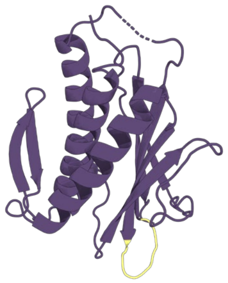
RNA-recognition domain



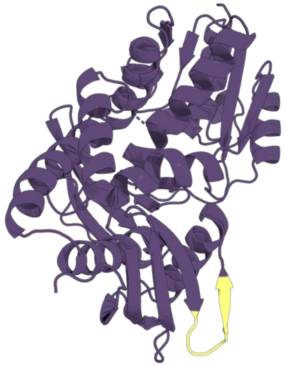
BRK domain



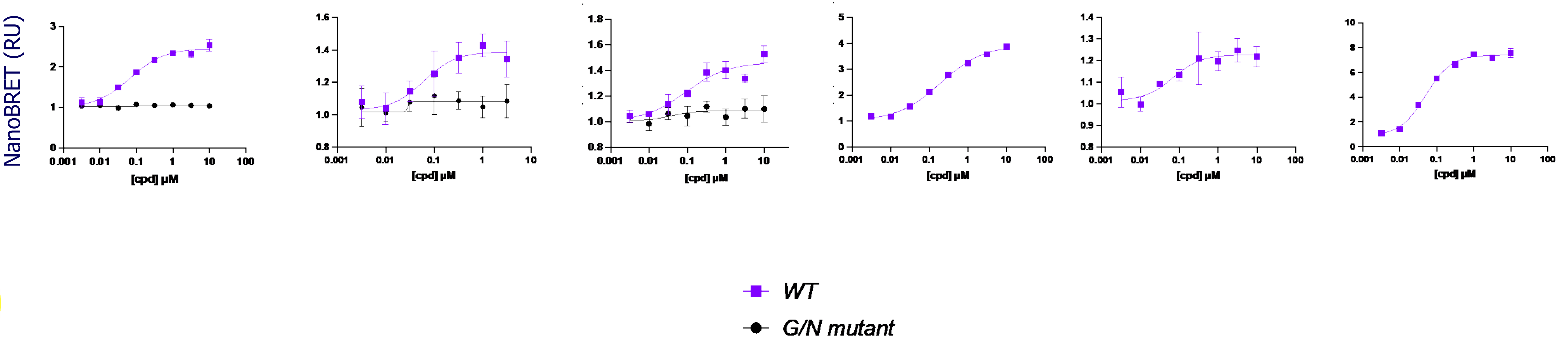
WW domain



Horma domain



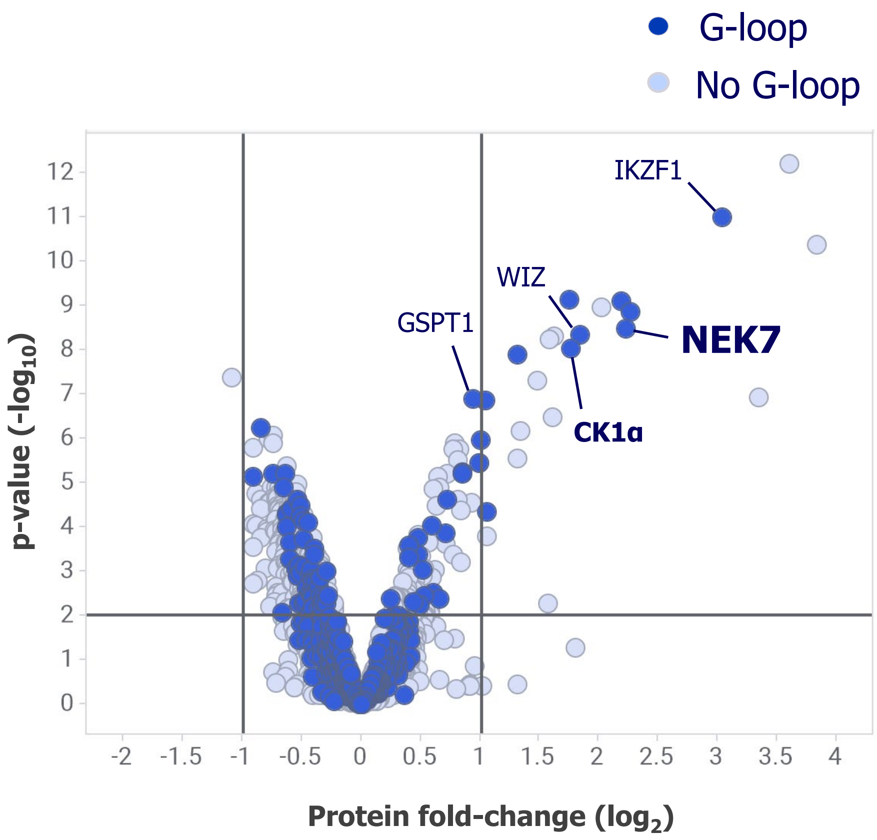
HUP domain



Engineering Selective MGDs for G-loop Targets – Exemplified by NEK7

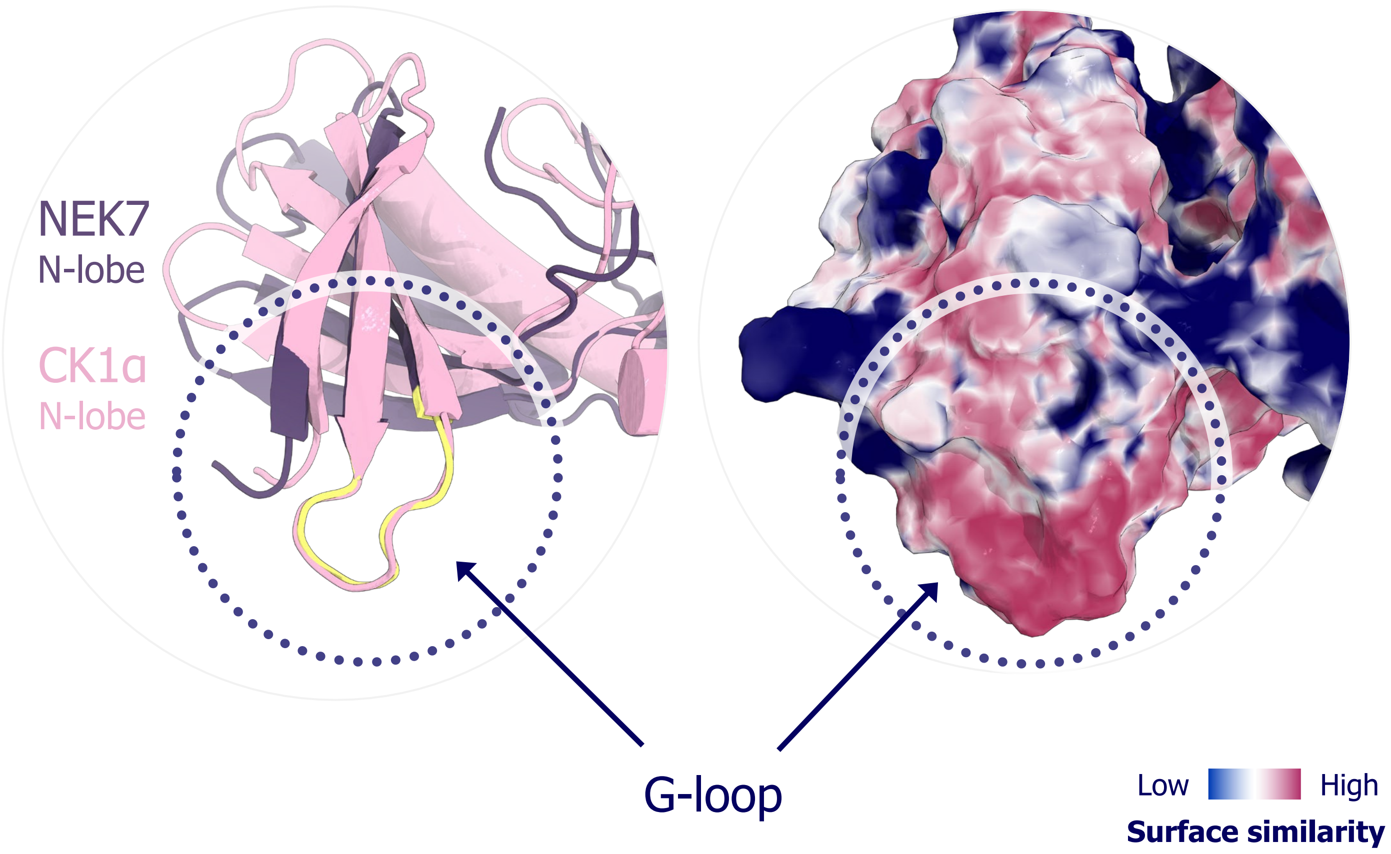
NEK7 and CK1α show high degree of G-loop surface similarity

Promiscuous MGDs induce proximity of multiple G-loop targets to CRBN



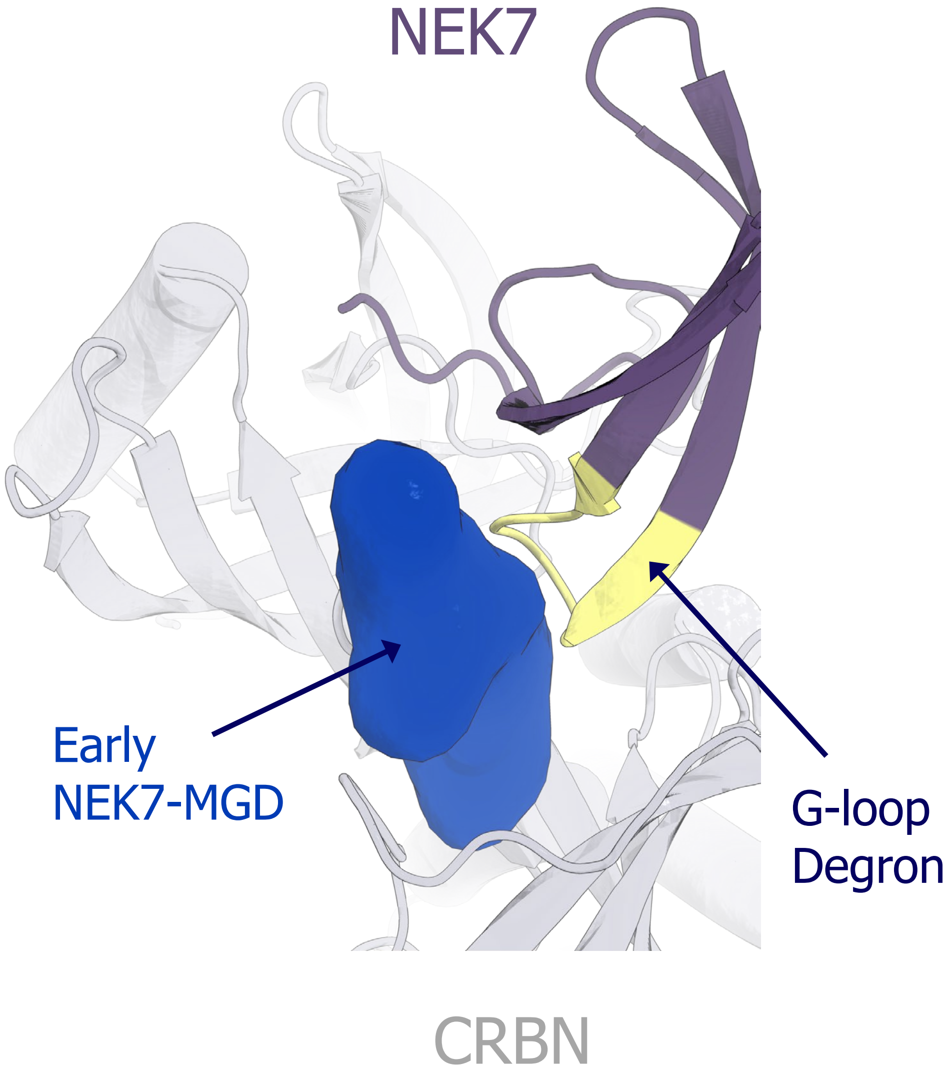
TurboID-CRBN (U937)

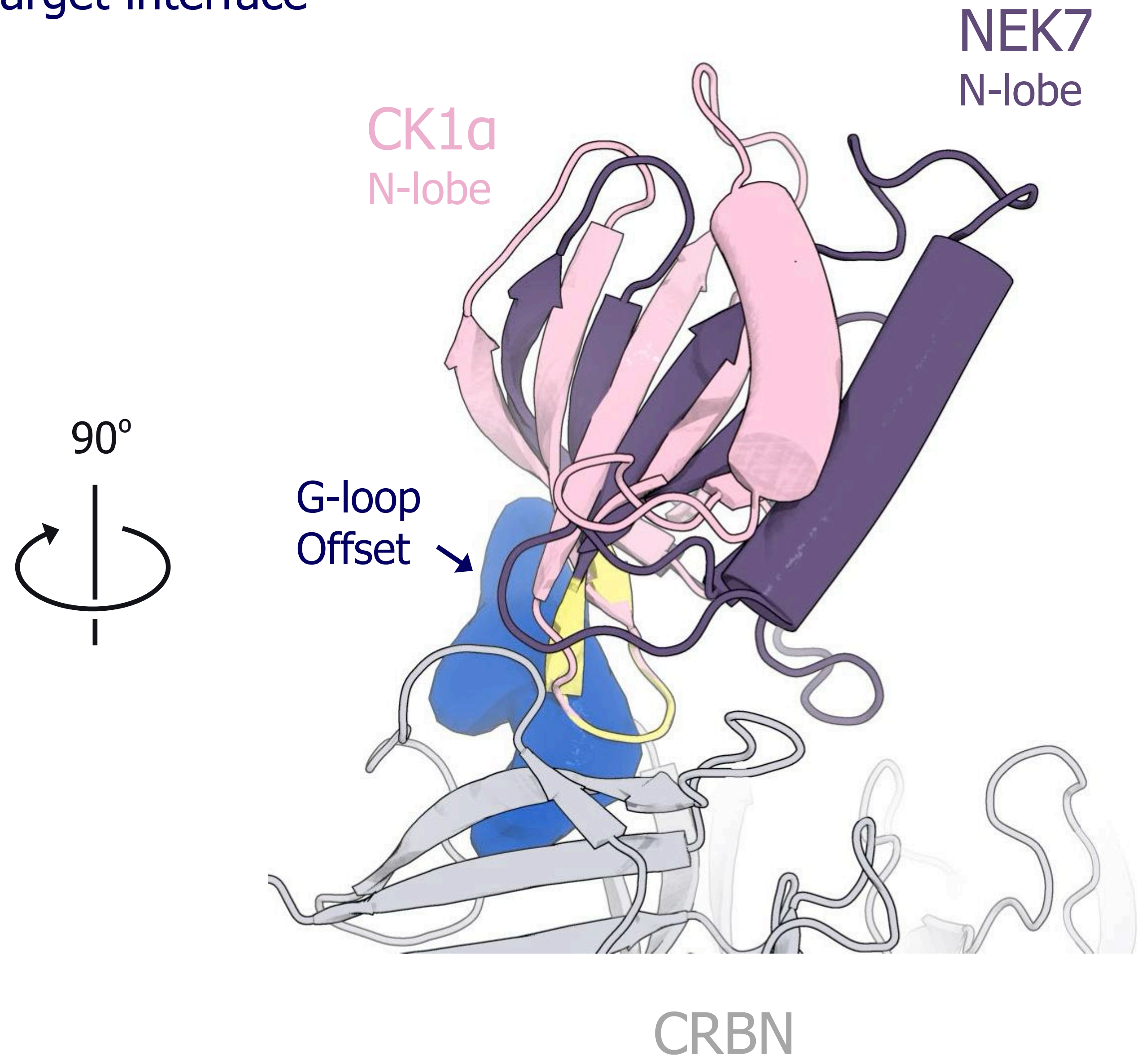
NEK7 and CK1α share high G-loop surface similarity



Early Structural Insights into NEK7 Binding Enabled Rational MGD Design

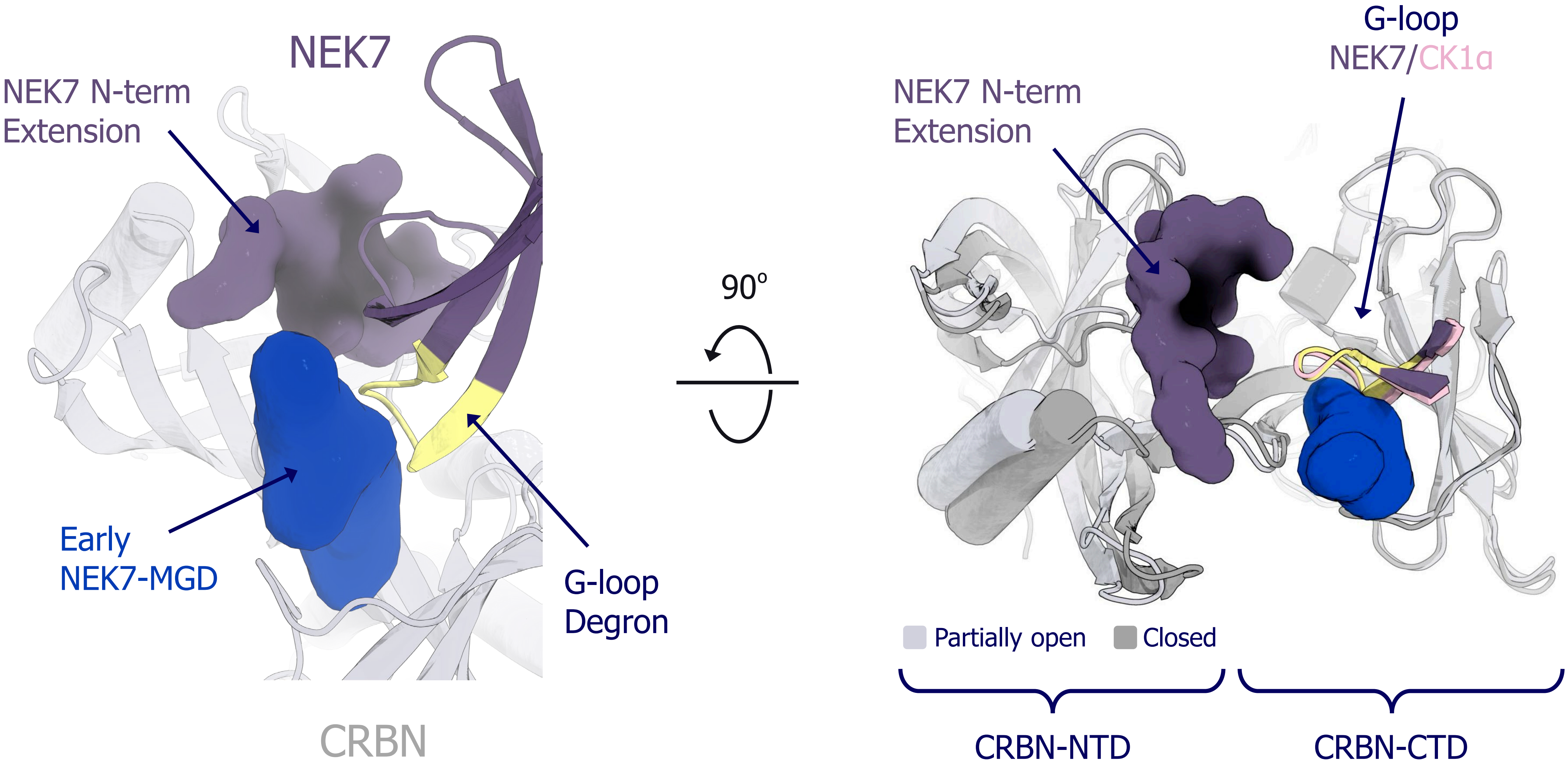
NEK7 G-loop offset creates unique MGD:target interface





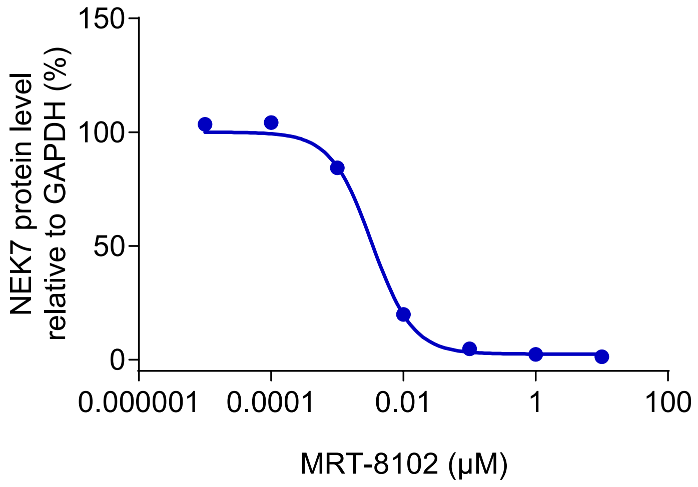
NEK7 Engages CRBN in a Partially Open Conformation

A larger spectrum of CRBN conformations are accessible for TPD



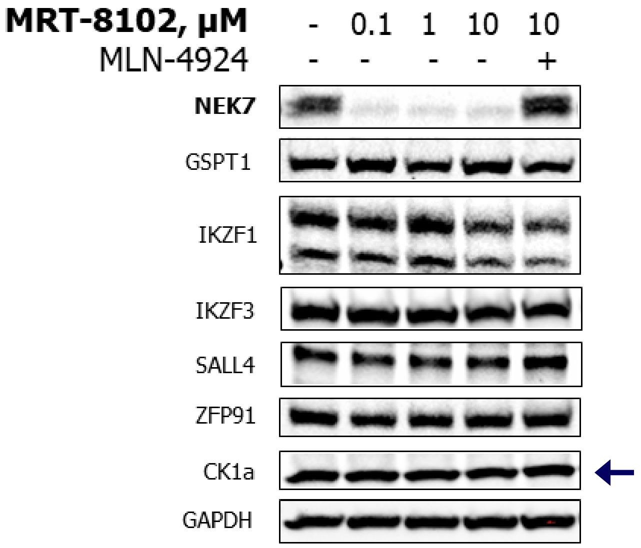
MRT-8102 is a Potent and Selective NEK7-directed MGD

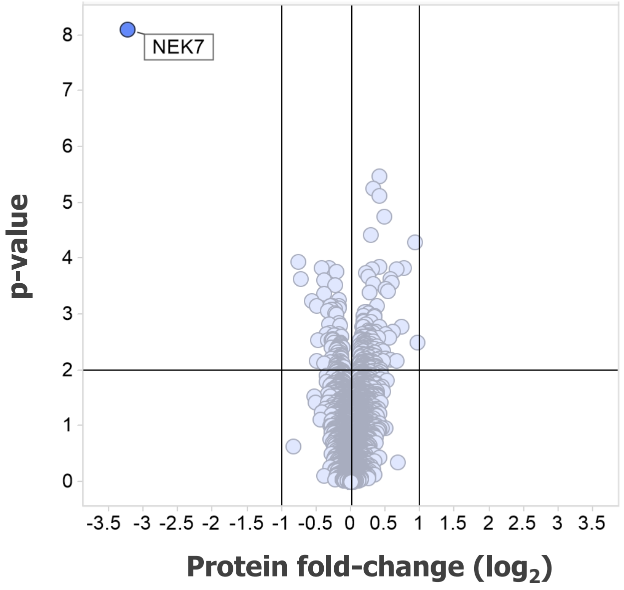
MRT-8102 is a potent NEK7-directed MGD



in vitro **data**	
CRBN binding, IC_{50}	200 nM
Degradation, DC_{50} /D_{max} (hPBMC; 24hr)	10 nM / 89 %

MRT-8102 induces selective NEK7 degradation and has favorable ADME/DMPK properties

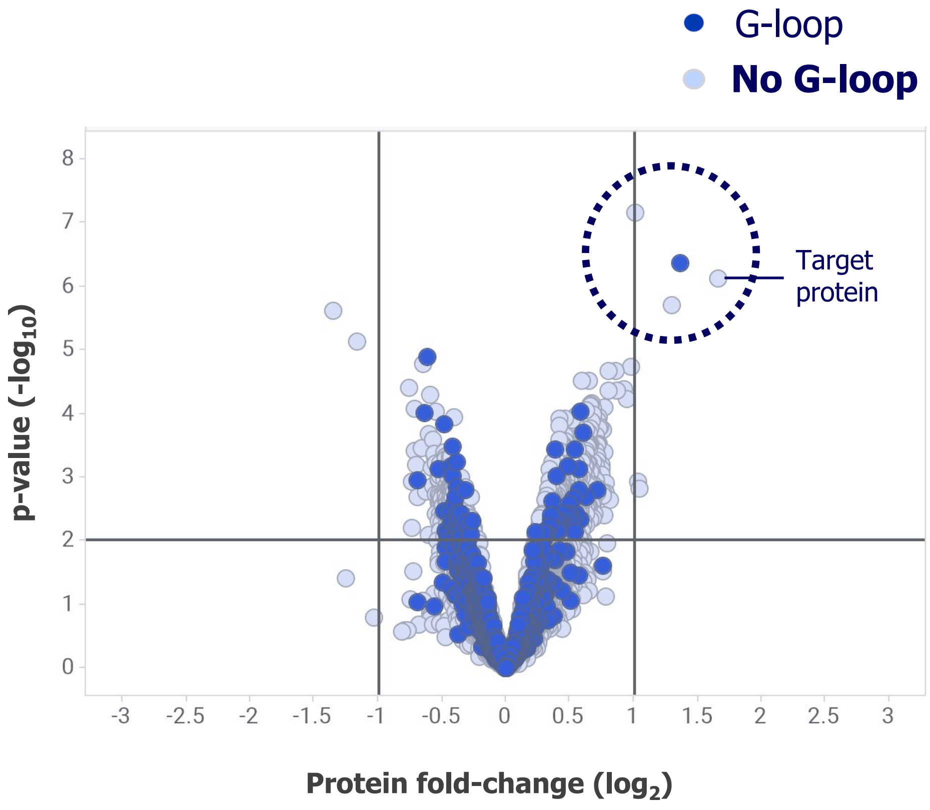




ADMET profile	
CEREP panel	No inhibition
hERG inhibition patch clamp	EC_{50} > 30 μM
Oral bioavailability	Yes

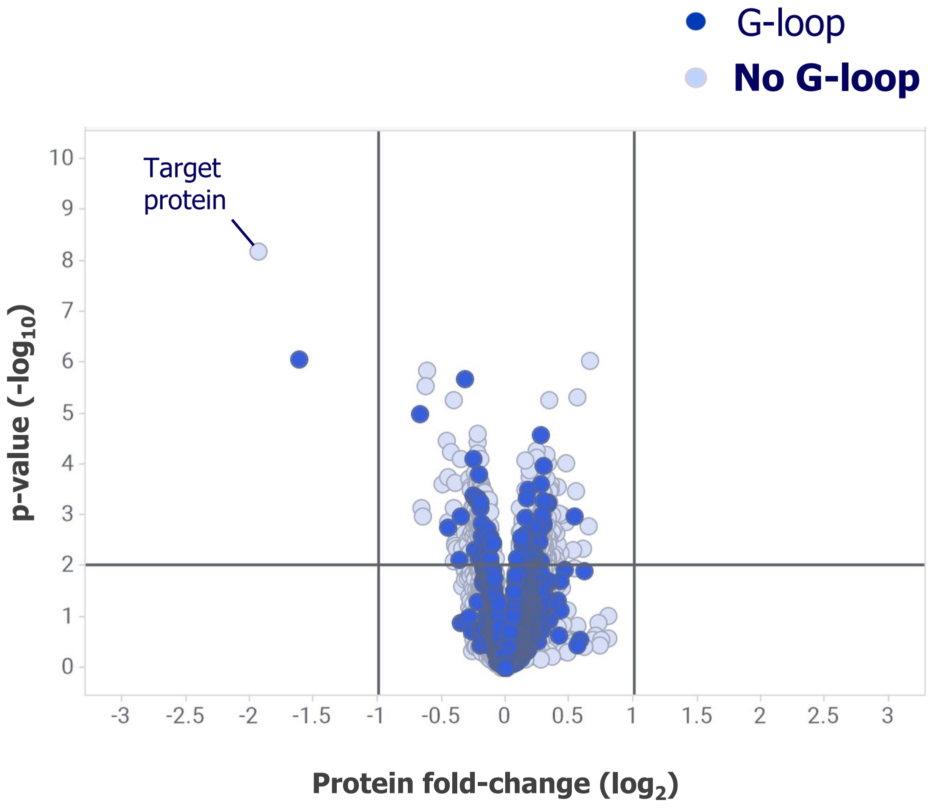
Non-G-loop Targets can be Recruited to CRBN and Degraded

Enrichment of non-G-loop proteins in CRBN proximity-ligation



TurboID-CRBN (CAL51)

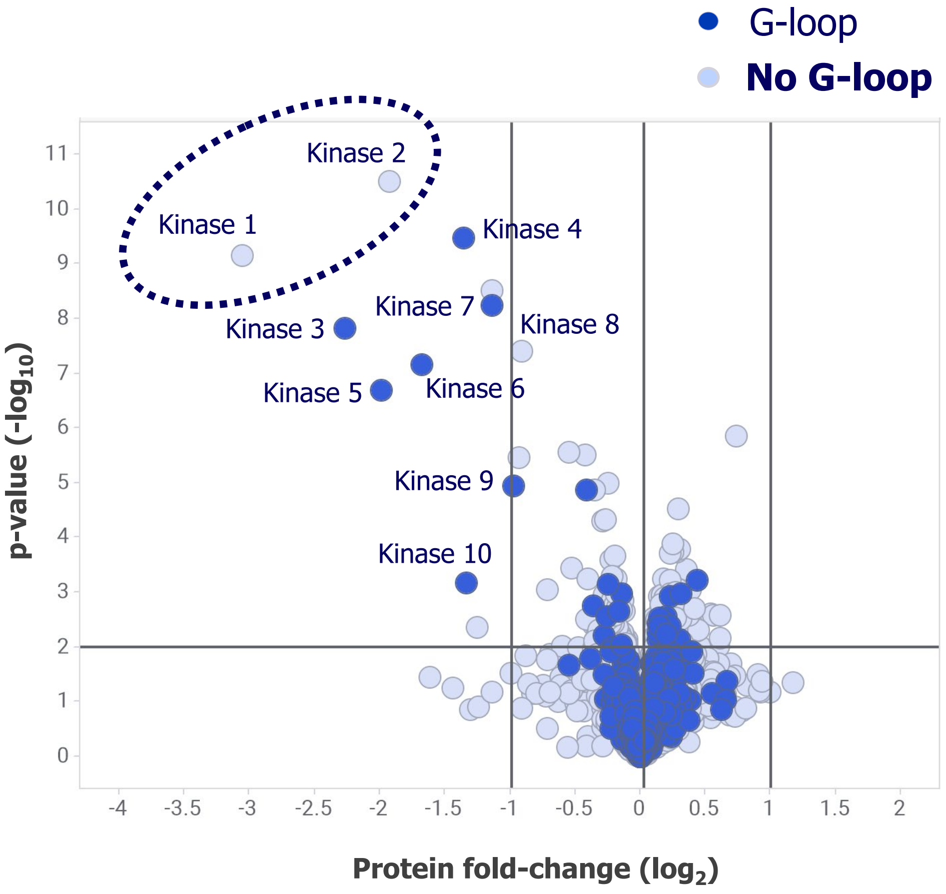
Degradation of a non-G-loop target in TMT-proteomics



TMT-Proteomics (Jurkat)

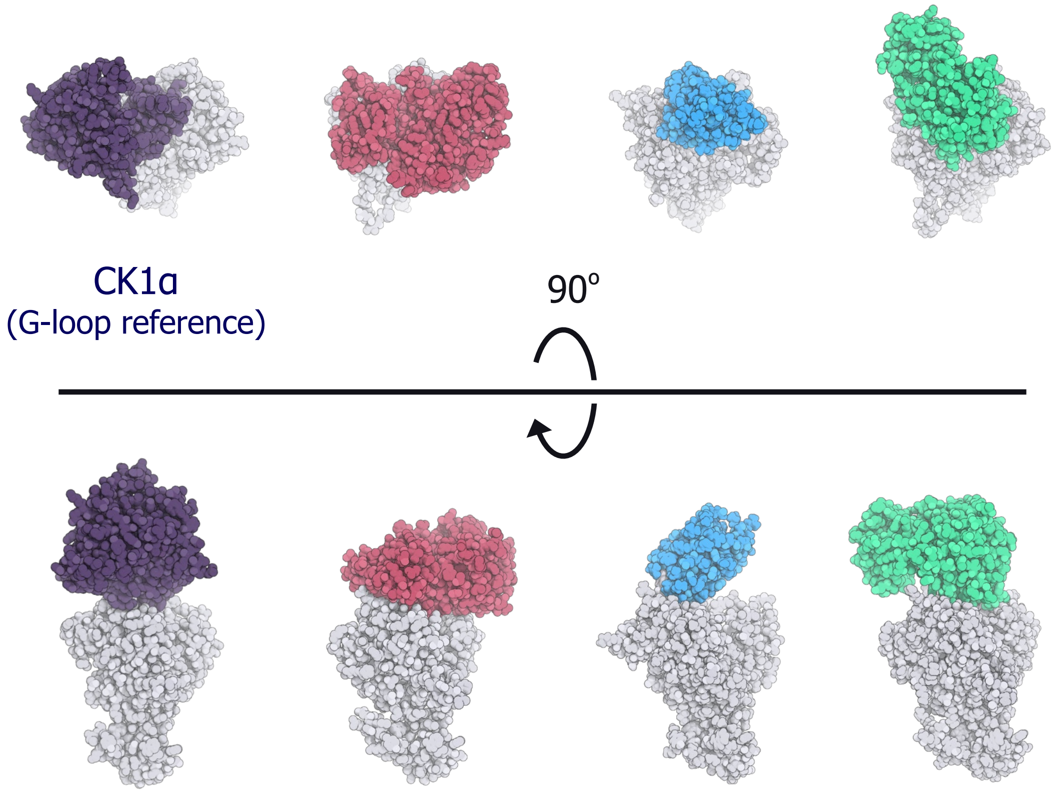
Non-G-loop Targets can be Recruited to CRBN and Degraded

Promiscuous kinase degrader



TMT-Proteomics (Jurkat)

Kinases engage CRBN through canonical and non-canonical binding modes

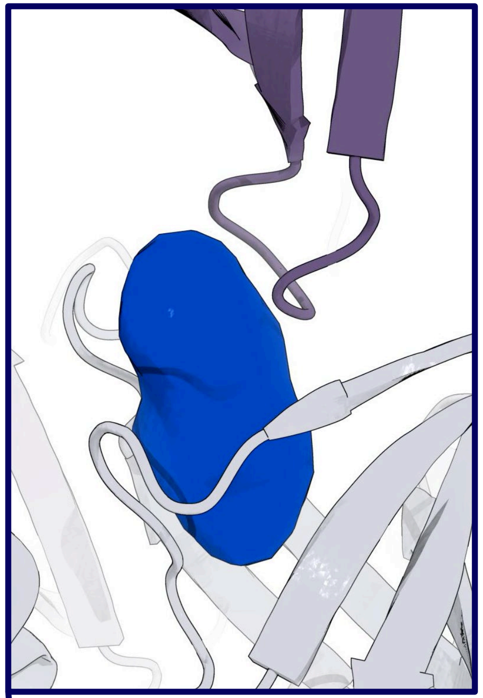


CRBN

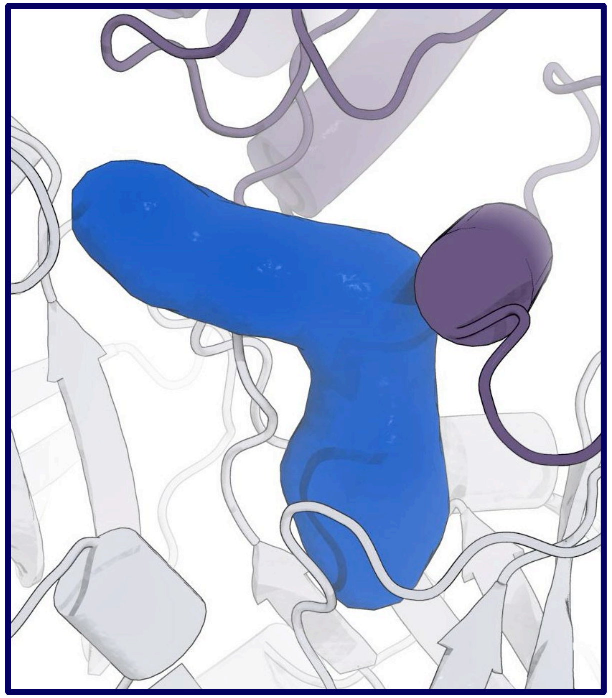
Novel Binding Modes Employ Otherwise Undruggable Surface Patches

Surface complementarity with CRBN:MGD drives target engagement

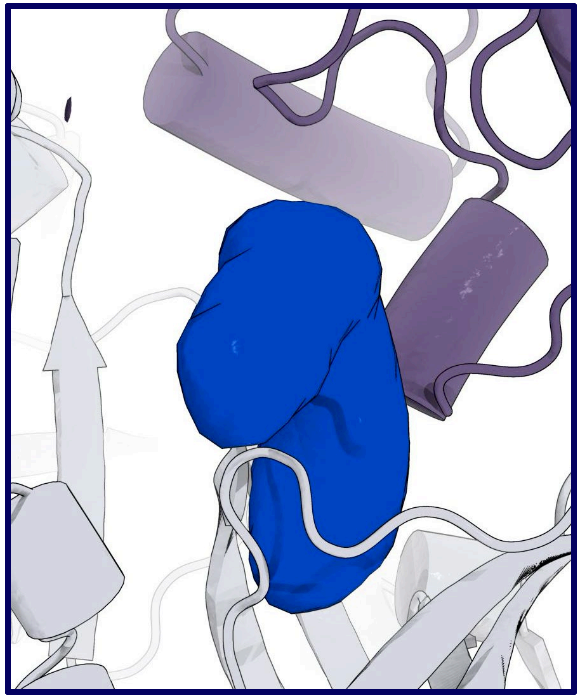
Canonical
G-loop degron



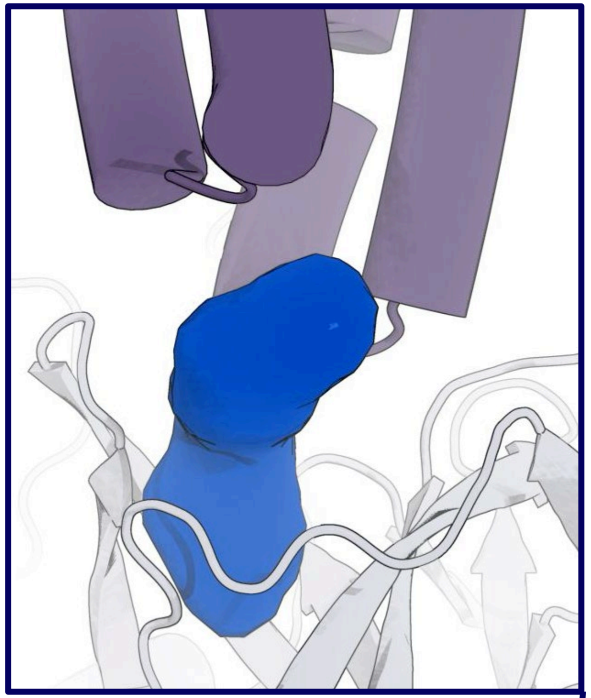
Novel binding mode
Type 1



Novel binding mode
Type 2



Novel binding mode
Type 3

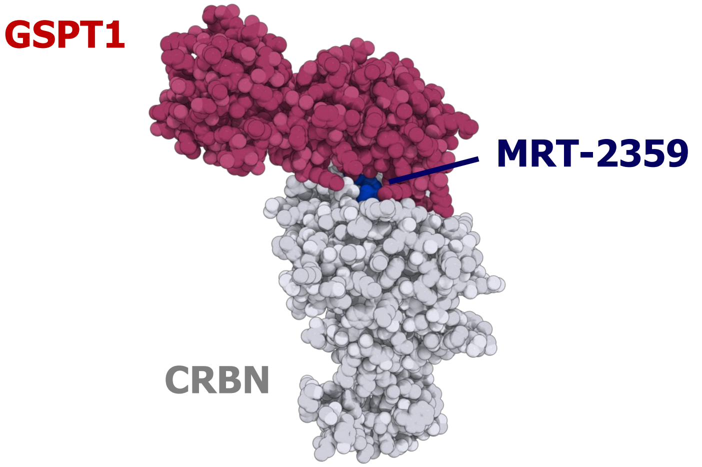


CRBN:MGD

QuEEN™ AI Finds Degrons Using Surfaces

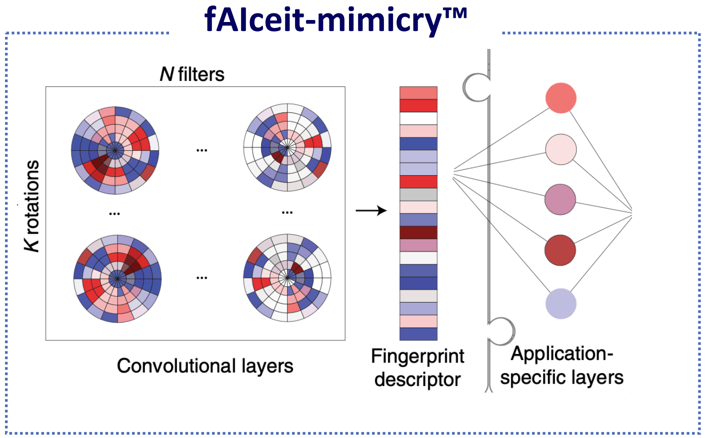
Leverage knowledge of ternary complex structures

Proprietary ternary complexes
X-ray & cryoEM



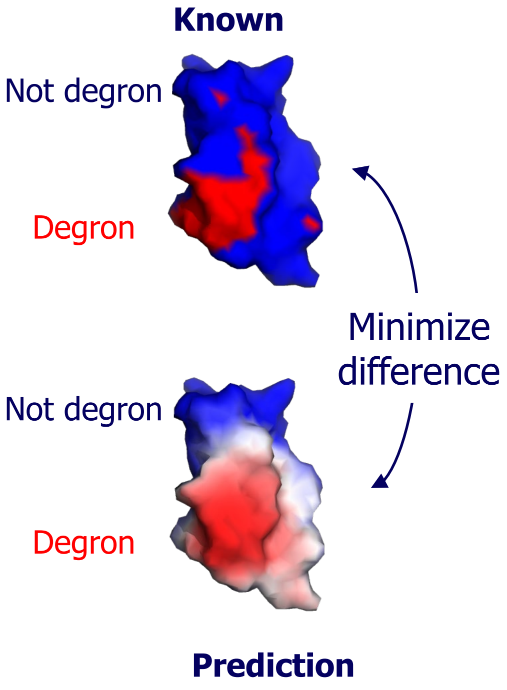
Algorithms tailored to degron discovery

Ultra-fast fingerprint search for surfaces
that mimic known degron surfaces



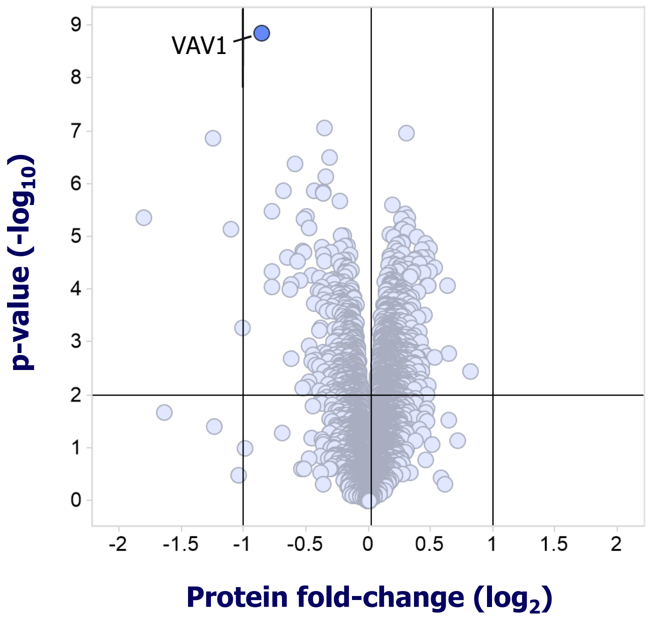
Expand analysis across the proteome

Identify surface patches that
match known degron interfaces



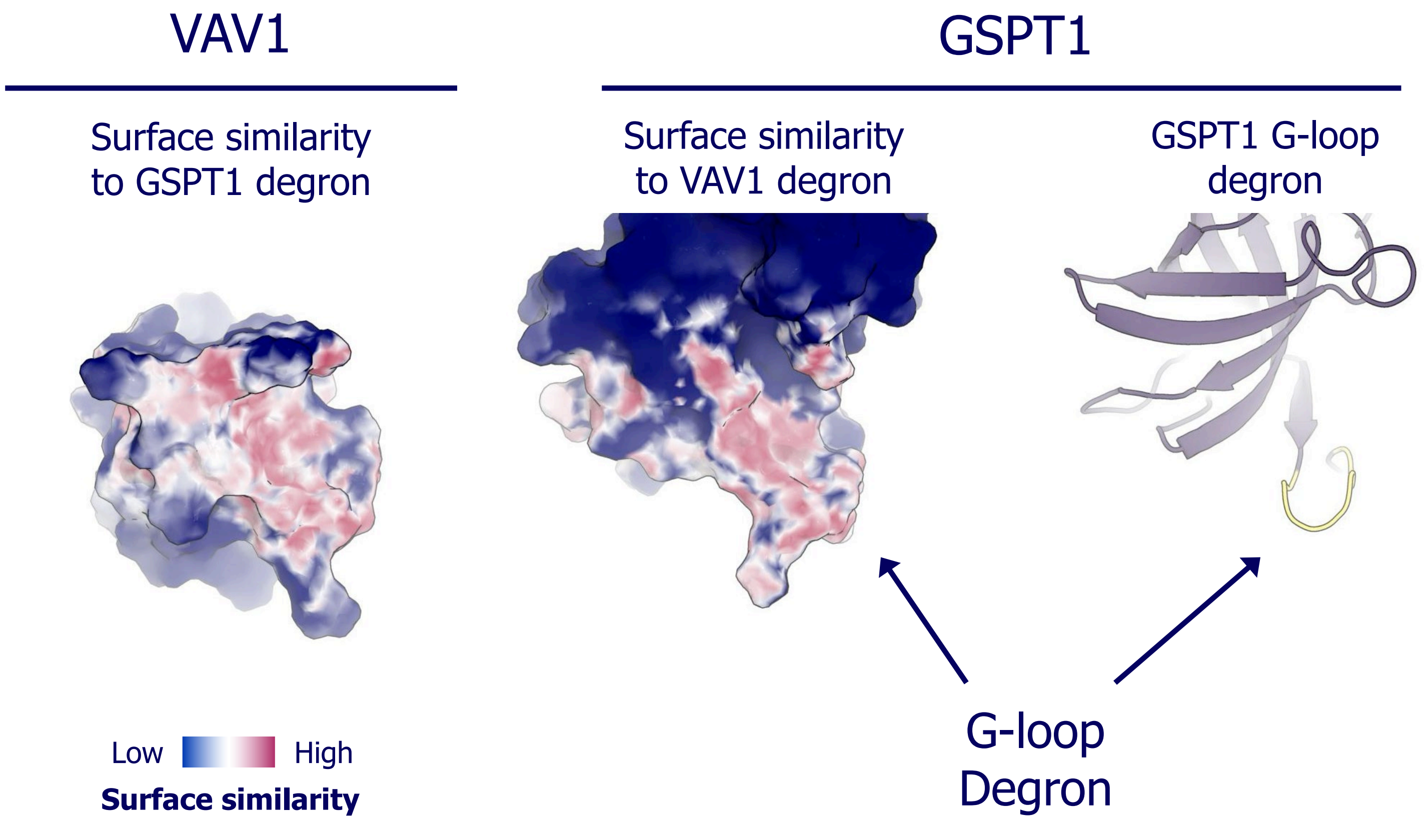
A Patch on VAV1 Shows Surface-similarity to the GSPT1 G-loop Degron

VAV1 identified from cell-based whole proteome screening



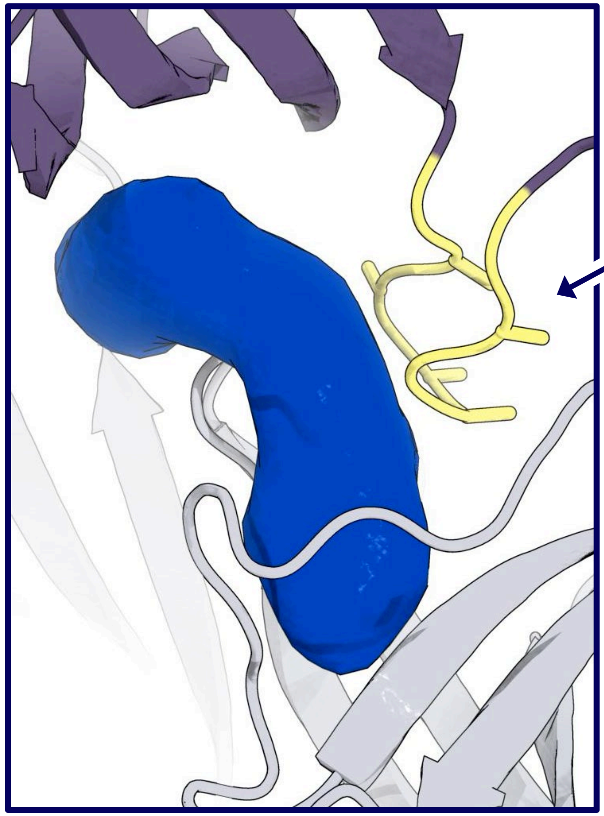
TMT-Proteomics (MOLM13)

VAV1 shows surface similarity to the G-loop degron of GSPT1



Novel Binding Mode for VAV1 Despite Surface-similarities to GSPT1

GSPT1
Canonical G-loop
degron

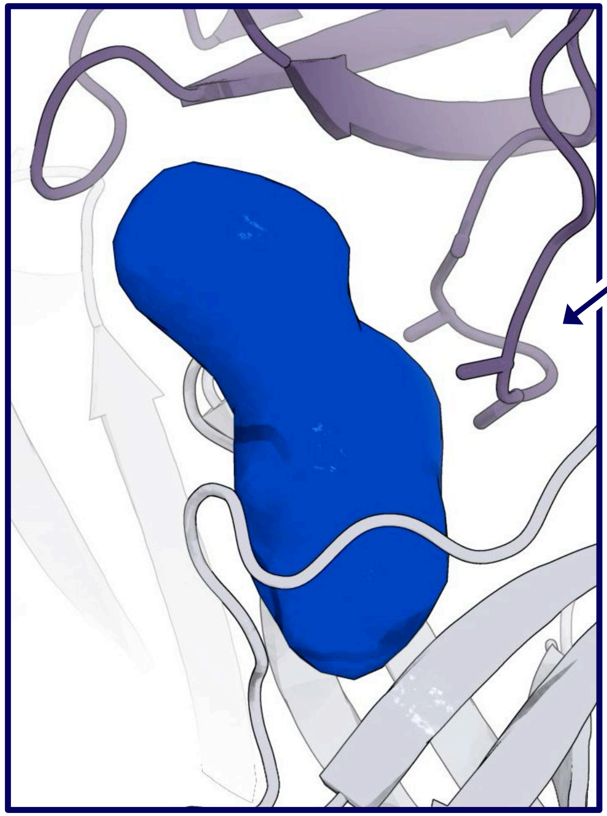


CRBN:MGD

MGD:GSPT1
interactions

Backbone-driven

VAV1
Novel binding mode
Type 4



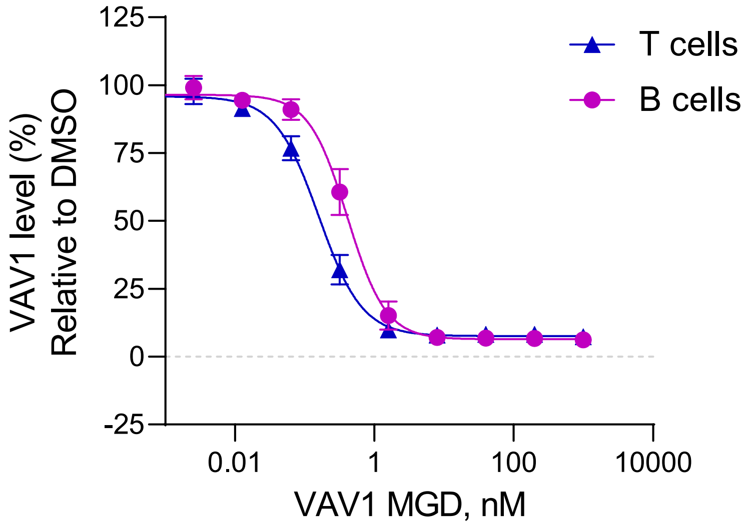
CRBN:MGD

MGD:VAV1
interactions

Side chain-driven

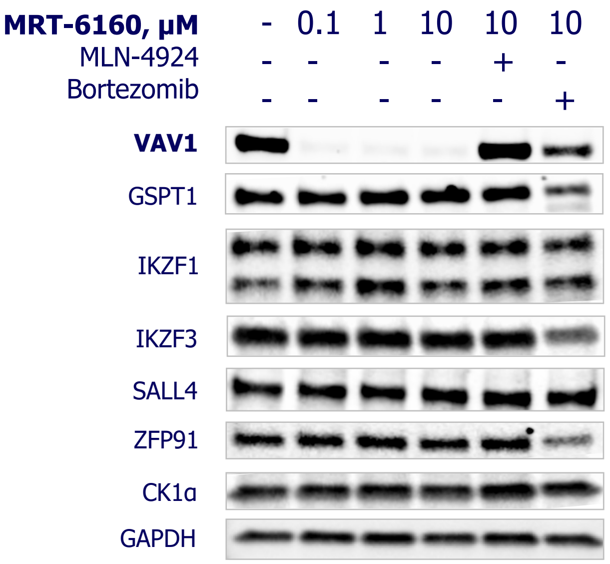
MRT-6160 is a Potent and Selective VAV1-directed MGD

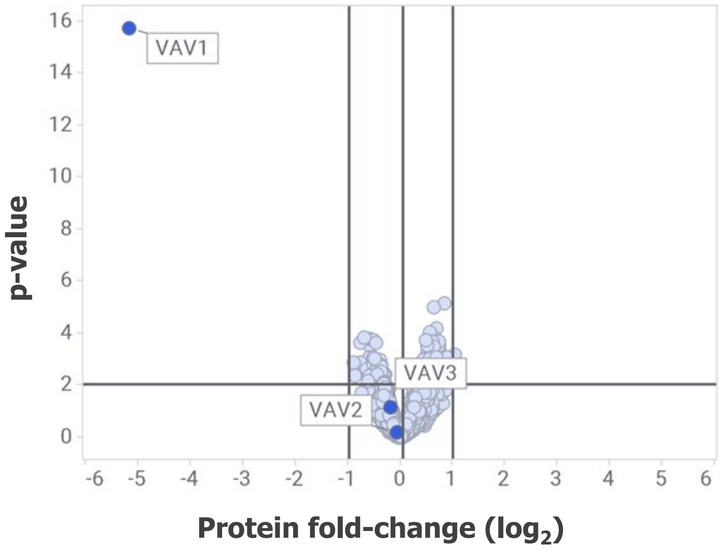
MRT-6160 is a potent VAV1-directed MGD



in vitro **data**	
CRBN binding, IC_{50}	670 nM
Ternary complex, EC_{50}	11 nM
Degradation, DC_{50} / D_{max} (Jurkat)	7 nM / 97 %

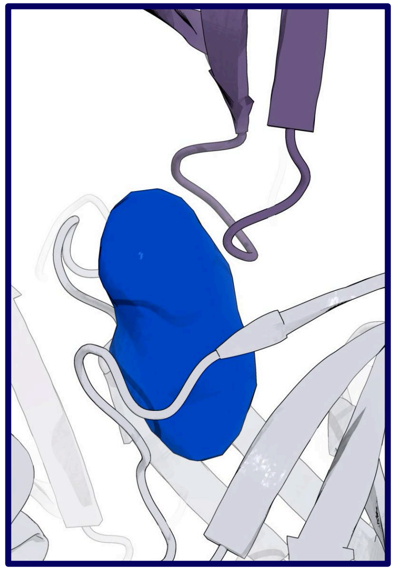
MRT-6160 induces selective VAV1 degradation and has a favorable ADME/DMPK profile

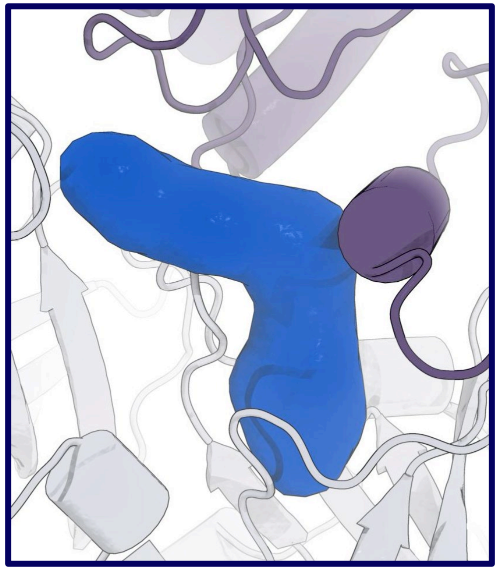


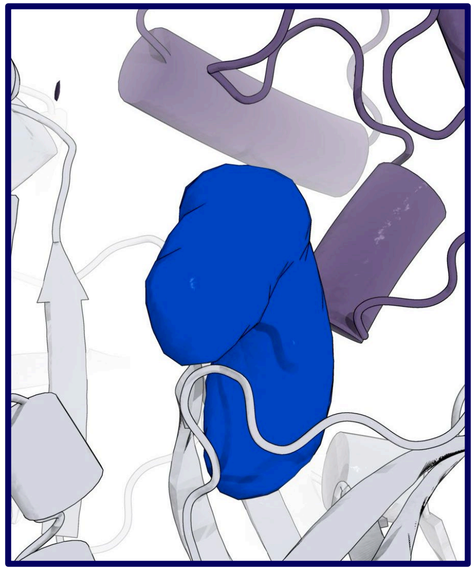


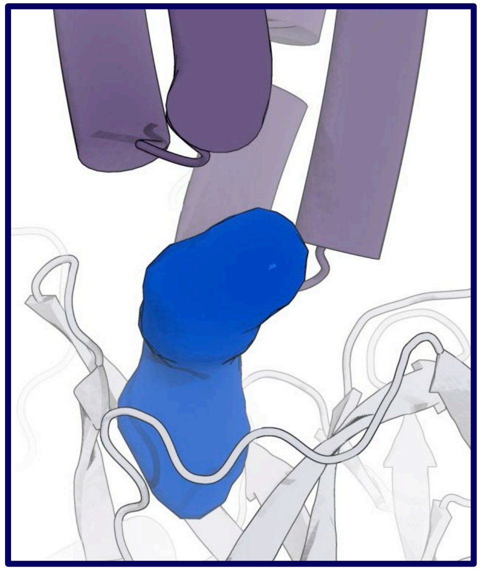
ADMET profile	
CYP DDIs	IC_{50} > 30 µM
hERG inhibition patch clamp	EC_{50} > 30 µM
Oral bioavailability all species	> 50%

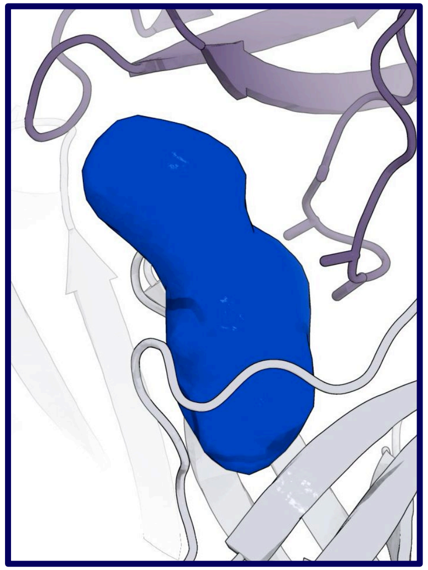
Novel Binding Modes are Highly Diverse in Structure, Sequence and MGD


Canonical G-loop


Novel Mode Type 1


Novel Mode Type 2


Novel Mode Type 3

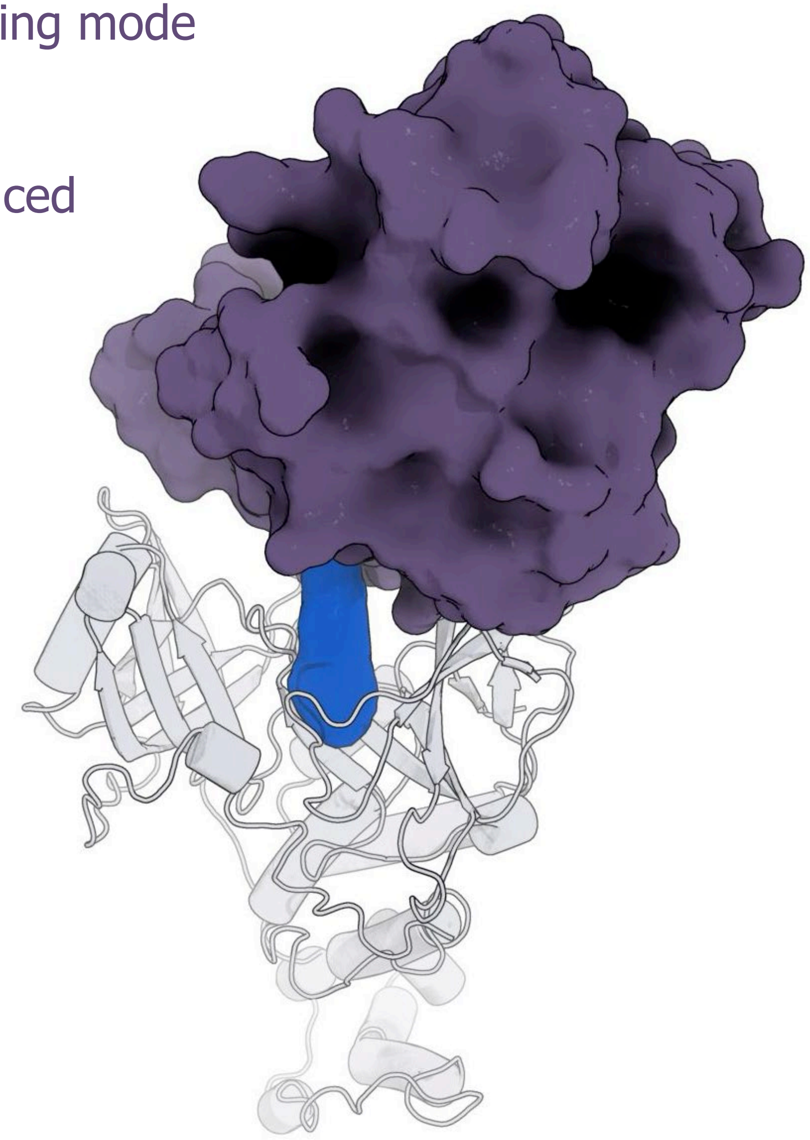

Novel Mode Type 4

- Novel binding modes broaden target space
- Potential benefits for selective MGD design

Example of an MGD Engaging a Cryptic Pocket at the Target Interface

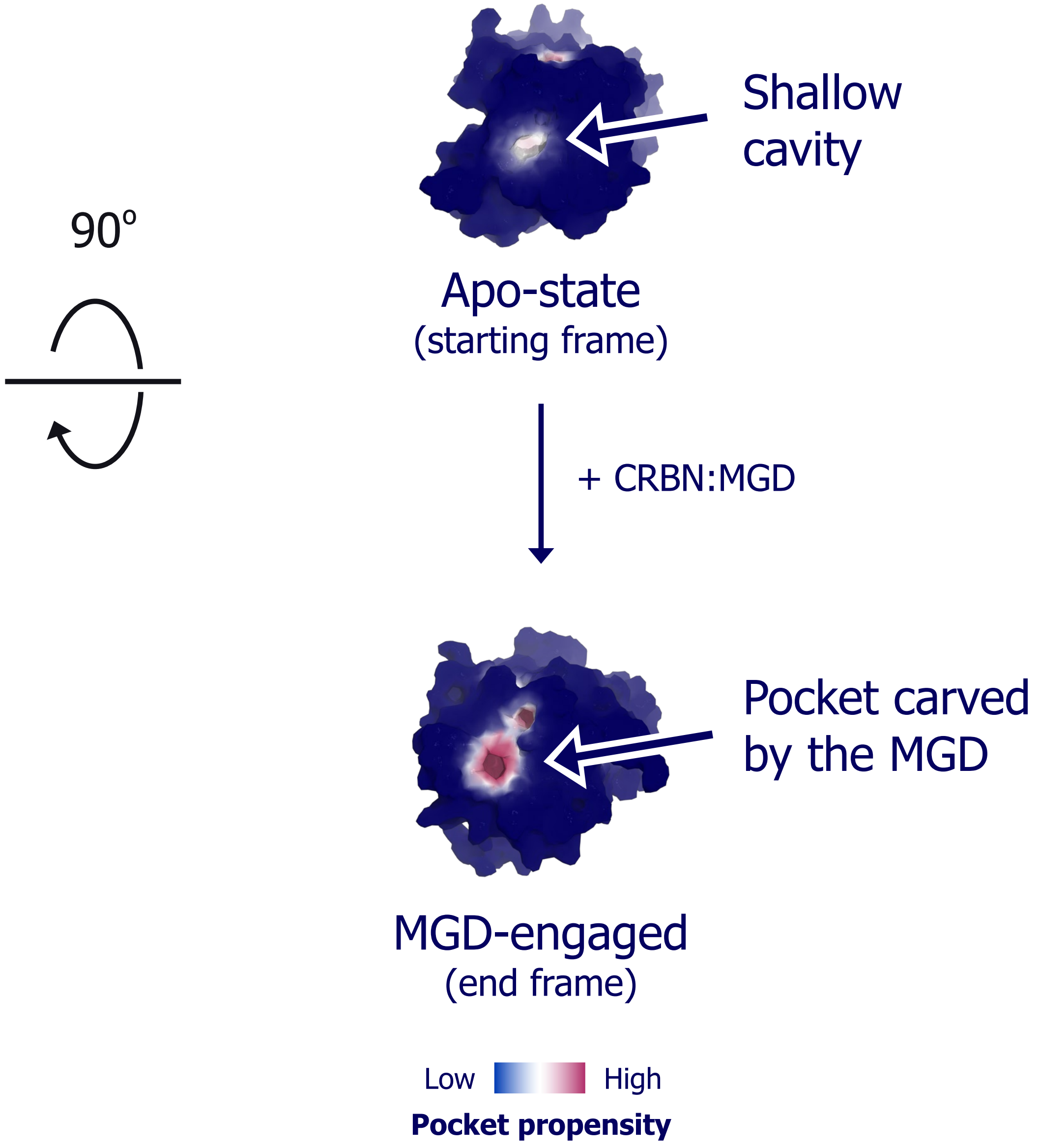
Novel binding mode
Type 5

(MGD-induced
pocket)

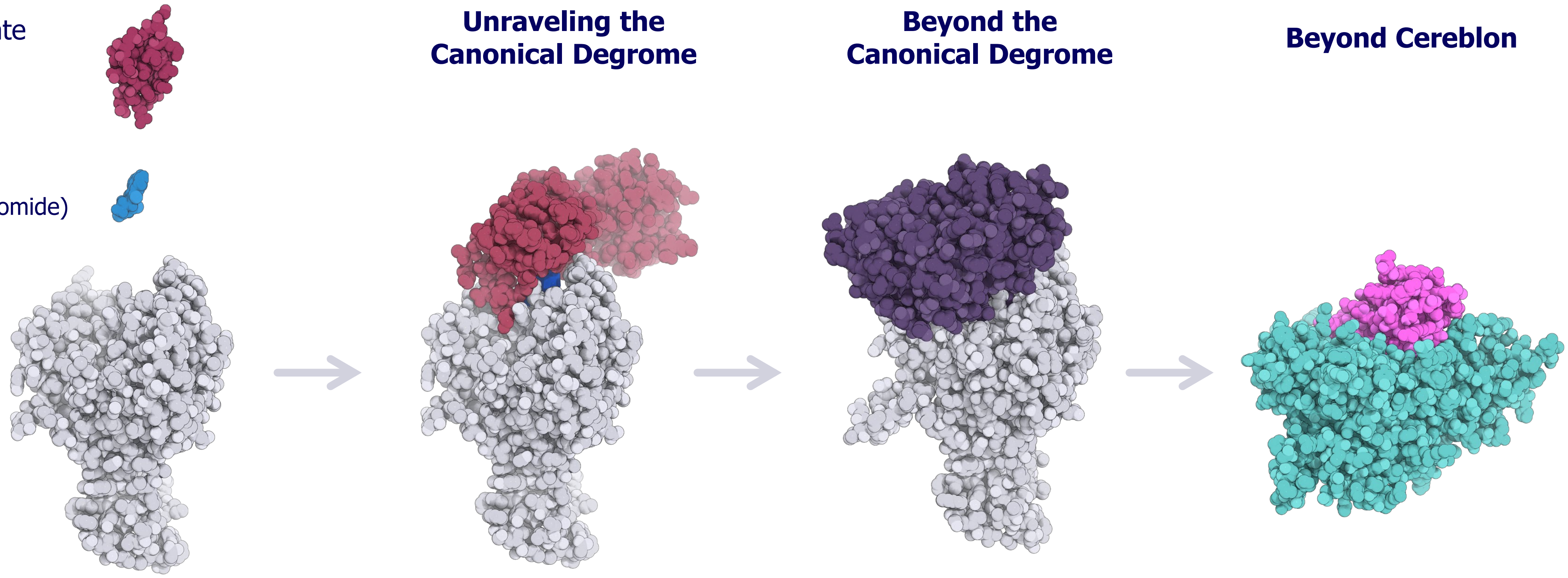


MGD

CRBN



Our Rational Approach to Unleash the Full Potential of MGDs



Neosubstrate
(e.g. IKZF1)

MGD
(e.g. Lenalidomide)

Cereblon

Unraveling the Canonical Degrome

>2,500 G-loop proteins

↓

>120 G-loops validated

Beyond the Canonical Degrome

fAIceit-mimicry™ and screening

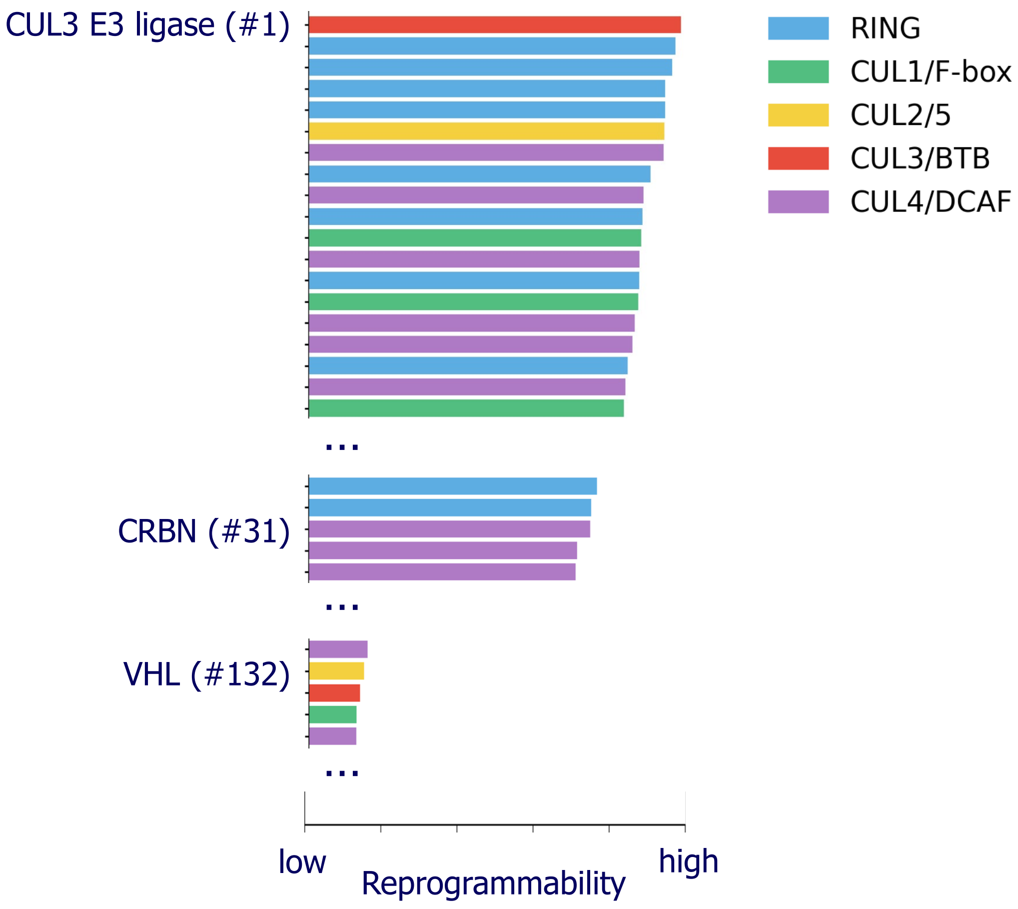
↓

>10 novel binding modes identified

Beyond Cereblon

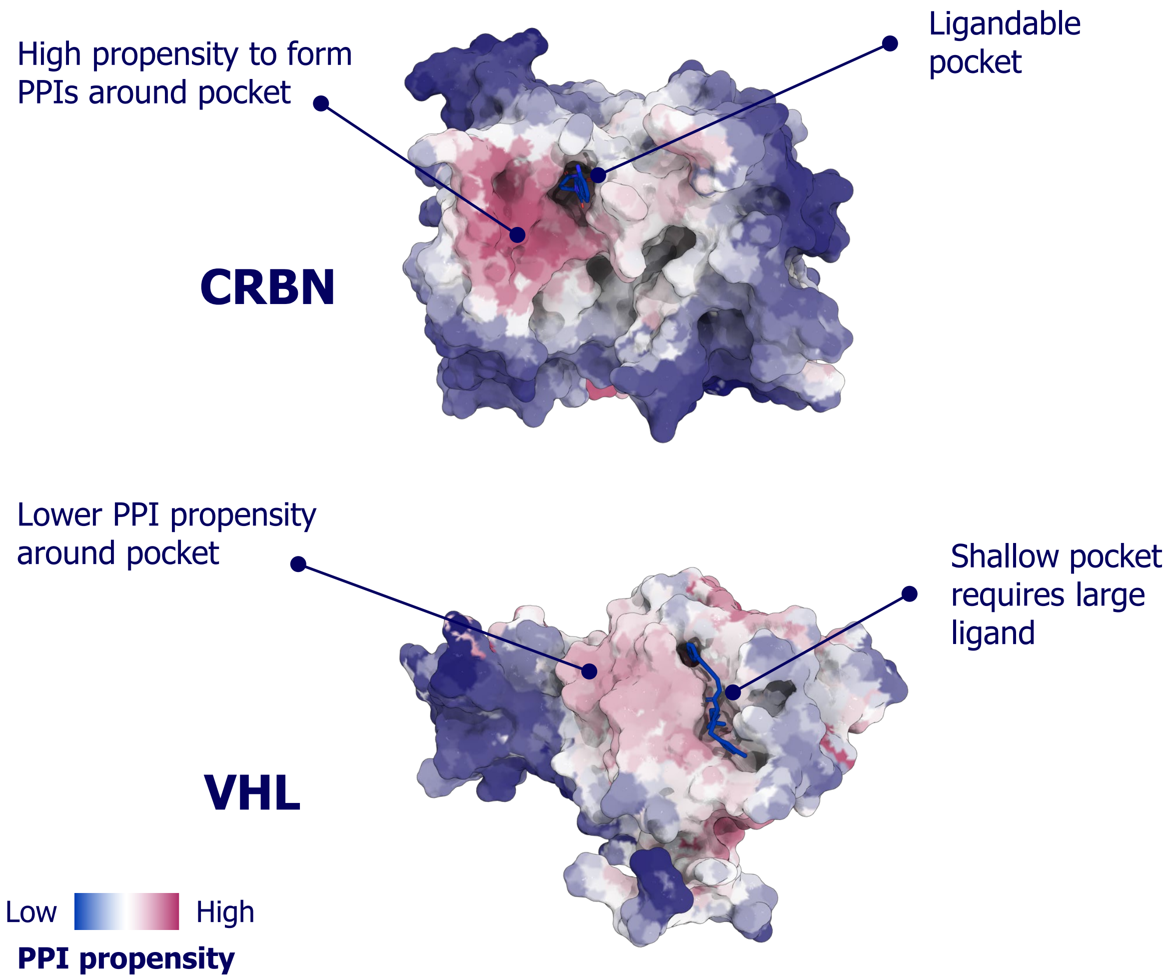
Prioritizing E3 Ligases Based on Reprogrammability Potential

fAIceit AI-powered surface characterization



CRL & RING families shown

CRBN is one of a handful of high-scoring E3 ligases



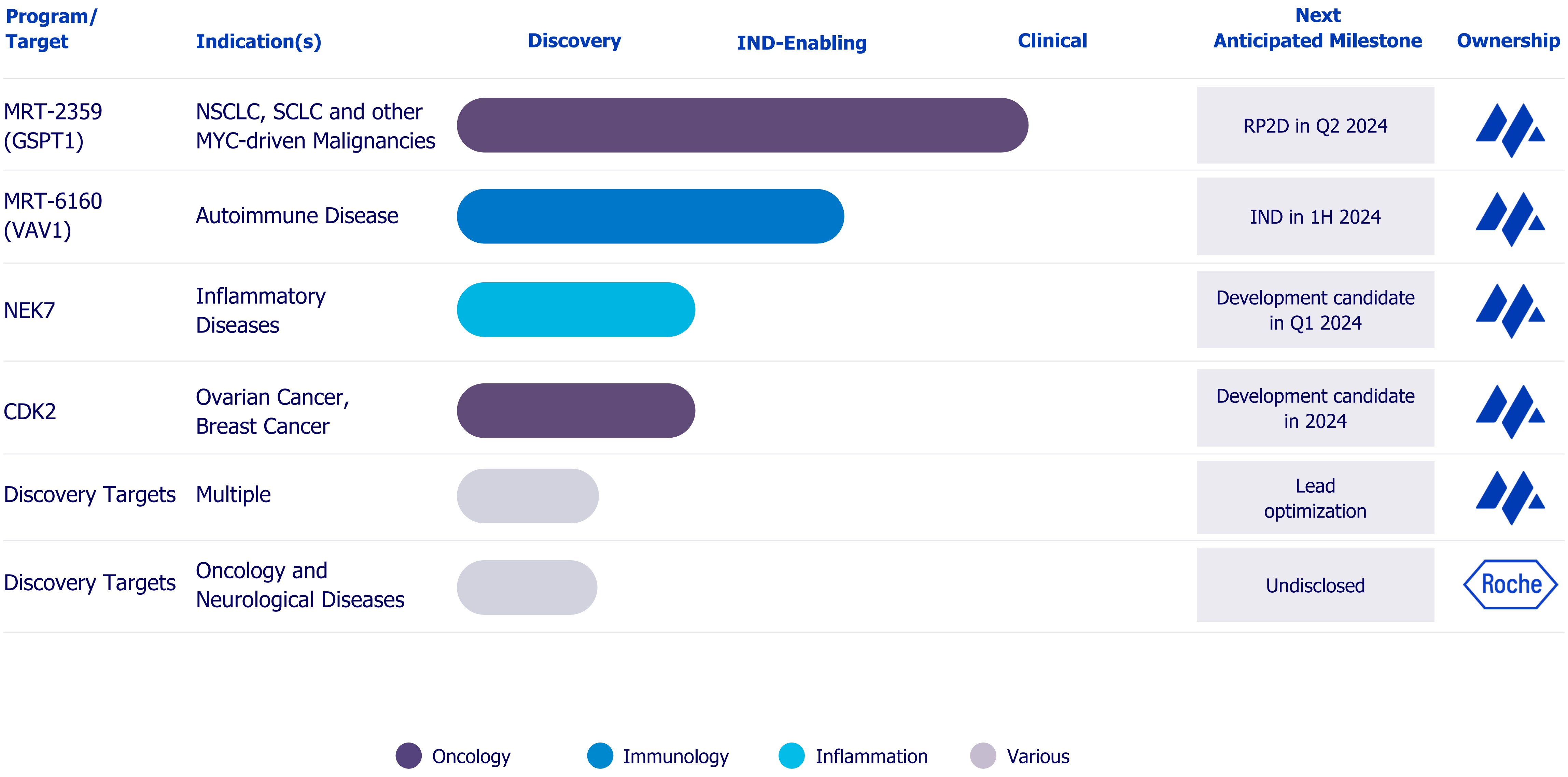
High propensity to form PPIs around pocket

Ligandable pocket

Lower PPI propensity around pocket

Shallow pocket requires large ligand

Monte Rosa Pipeline



Program/ Target	**Indication(s)**	**Discovery**	**IND-Enabling**	**Clinical**	**Next Anticipated Milestone**	**Ownership**
MRT-2359 (GSPT1)	NSCLC, SCLC and other MYC-driven Malignancies	●	●	●	RP2D in Q2 2024	
MRT-6160 (VAV1)	Autoimmune Disease	●	●		IND in 1H 2024	
NEK7	Inflammatory Diseases	●			Development candidate in Q1 2024	
CDK2	Ovarian Cancer, Breast Cancer	●			Development candidate in 2024	
Discovery Targets	Multiple	●			Lead optimization	
Discovery Targets	Oncology and Neurological Diseases	●			Undisclosed	Roche

● Oncology ● Immunology ● Inflammation ● Various

Acknowledgments – It's a HUGE Team Effort!