A Comprehensive Platform for Characterization and Evaluation of Degrader-Antibody Conjugates

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Introduction

Degrader-antibody conjugates (DACs) represent a promising modality for cancer therapy, integrating a tumor-targeting antibody, a protein degrader payload, and a chemical linker into a single molecule. This design combines antibody-mediated tumor specificity with the degrader's ability to eliminate disease-associated proteins. To accelerate DAC development, ICE Bioscience has established a comprehensive evaluation platform enabling end-to-end characterization and assessment of DACs, from in vitro profiling to in vivo pharmacology.

Methods

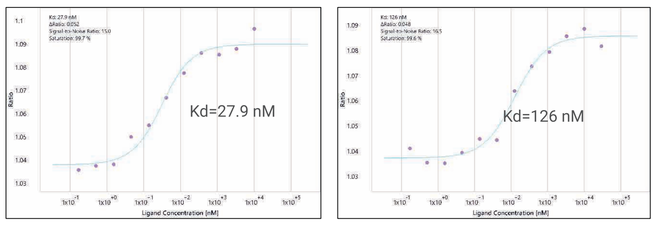
- **Degrader Payload Screening**
  Binary and ternary complex formation: Spectral shift, HTRF
  Target degradation: HiBiT, flow cytometry
- **Degrader-Antibody Conjugate Evaluation**
  In vitro degradation and cytotoxic effect on tumor cells: WB, CellTiter-Glo
  Pharmacokinetics and Drug-Antibody Ratio Analysis: ELISA, LC-HRMS
  In vivo Antitumor Efficacy: CDX model

Degrader: Binary and ternary complex formation

A

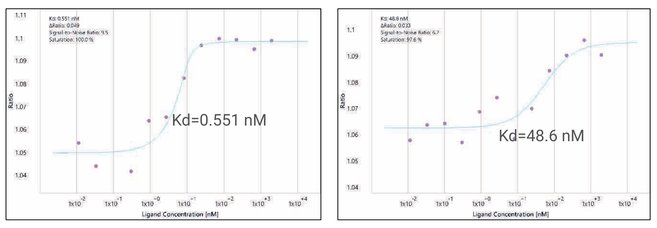
CRBN-degrader binary complex formation

CC-885 | CC-90009



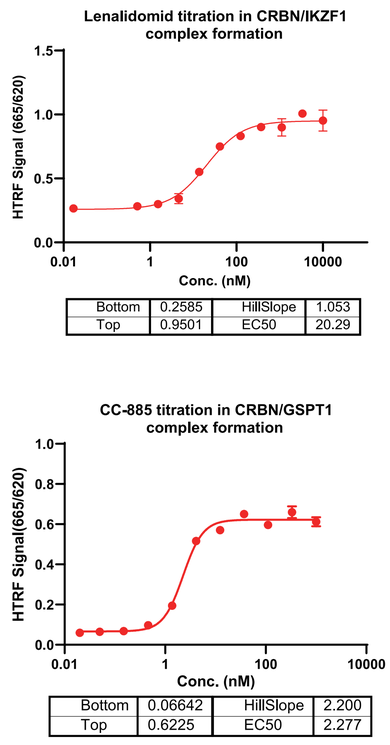
CRBN-degrader-GSPT1 ternary complex formation

CC-885 | CC-90009



Degrader	Assay type	Kd (nM)	Cooperativity factor, α
CC-885	Binary	27.9	50.6
	Ternary	0.551	
CC-90009	Binary	126	2.59
	Ternary	48.6	

B

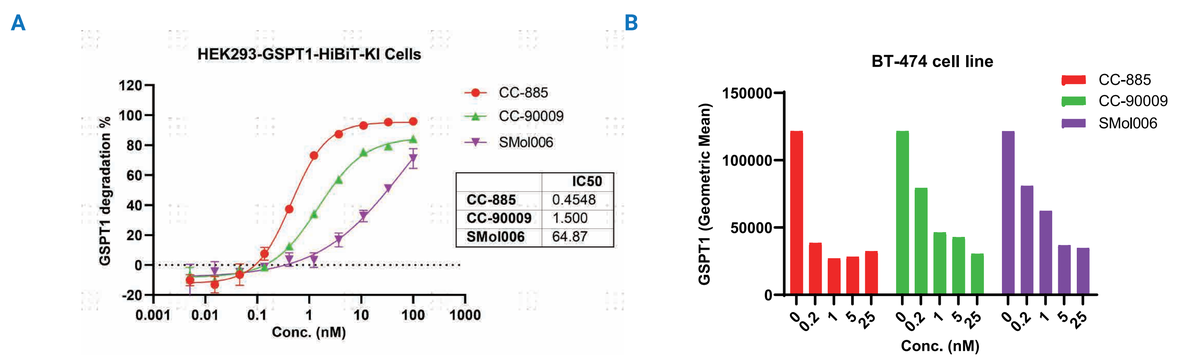


Bottom	0.2585	HillSlope	1.053
Top	0.9501	EC50	20.29

Bottom	0.06642	HillSlope	2.200
Top	0.6225	EC50	2.277

Figure 1. A, Binary and ternary complex formation was detected by the spectral shift assay. Two GSPT1 degraders exhibited cooperativity in ternary complex formation, as indicated by a cooperativity factor α>1. B, An ICE-established HTRF assay was used to detect the formation of CRBN-degrader-IKZF1 and CRBN-degrader-GSPT1 ternary complex.

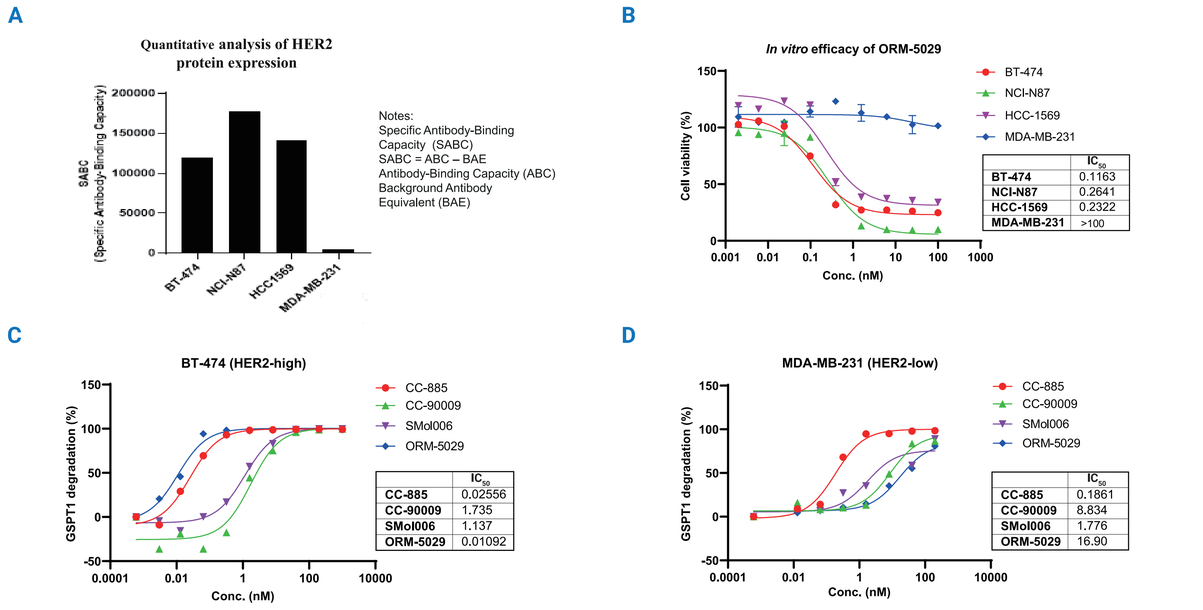
Degrader: Target degradation



	IC50
CC-885	0.4548
CC-90009	1.500
SMol006	64.87

Figure 2. GSPT1 degradation was assessed in a HEK293 GSPT1-HiBiT knock-in reporter cell line using a luminescence-based plate reader assay (A), and in a breast cancer cell line BT-474 by flow cytometry (B). SMol006, the GSPT1-degrader payload of the DAC ORM-5029, induced weaker GSPT1 degradation than the degraders CC-885 and CC-90009.

DAC: Degradation and cytotoxic effect on tumor cells

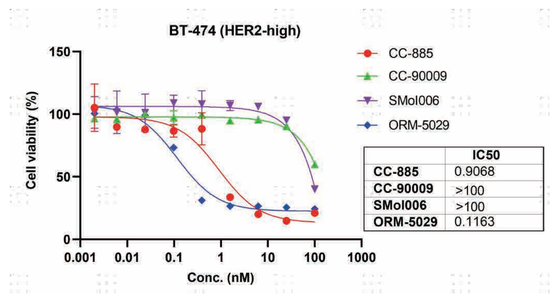


	IC_{50}
BT-474	0.1163
NCI-N87	0.2641
HCC-1569	0.2322
MDA-MB-231	>100

	IC_{50}
CC-885	0.02556
CC-90009	1.735
SMol006	1.137
ORM-5029	0.01092

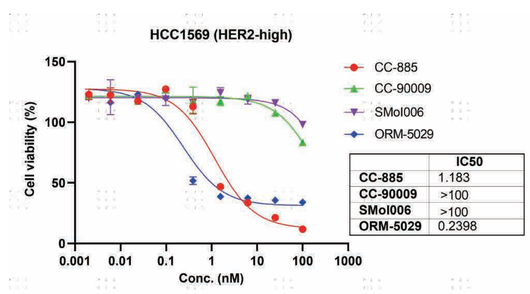
	IC_{50}
CC-885	0.1861
CC-90009	8.834
SMol006	1.776
ORM-5029	16.90

E



	IC50
CC-885	0.9068
CC-90009	>100
SMol006	>100
ORM-5029	0.1163

F

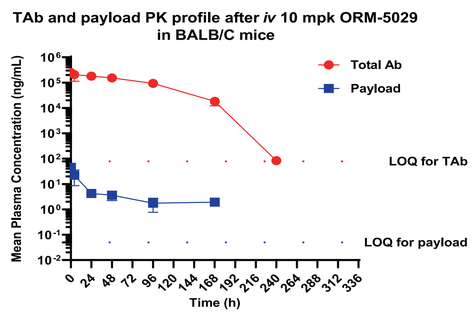


	IC50
CC-885	1.183
CC-90009	>100
SMol006	>100
ORM-5029	0.2398

Figure 3. A, Quantitative analysis of HER2 expression level in different cells via flow cytometry using QIFIKIT. B, In vitro potency of ORM-5029 in cell lines with differential HER2 expression levels assessed by the CellTiter-Glo viability assay. Comparison of GSPT1 degradation (C, D) and cytotoxicity (E, F) between GSPT1 degraders and ORM-5029 in cell lines with different HER2 expression levels. Both the cytotoxicity and target degradation of ORM-5029 were positively correlated with cell-surface HER2 expression levels. ORM-5029 demonstrated more potent cytotoxicity than the unconjugated GSPT1 degraders.

DAC: Pharmacokinetics and drug-antibody ratio analysis

A



B

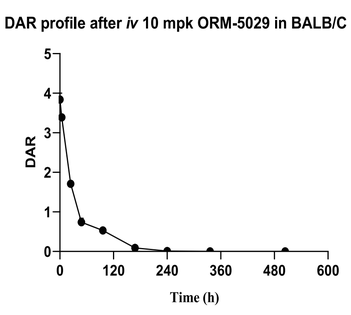
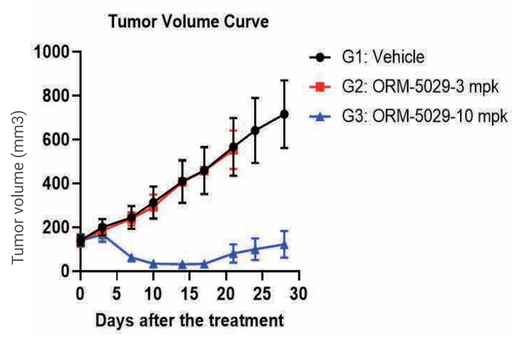


Figure 4. A, Plasma concentration of total antibody and payload analyzed via ELISA/LC-MS/MS, following a single dose of 10 mg/kg ORM-5029 in mice (n = 3 per group). B, Drug-to-antibody ratio (DAR) was detected by IC-LC-HRMS. The t½ of ORM-5029 was approximately 34 h, and payload clearance was 332,926 mL/day/kg. DAR values correlated well with the PK profile of ORM-5029.

DAC: In vivo antitumor efficacy

A



B

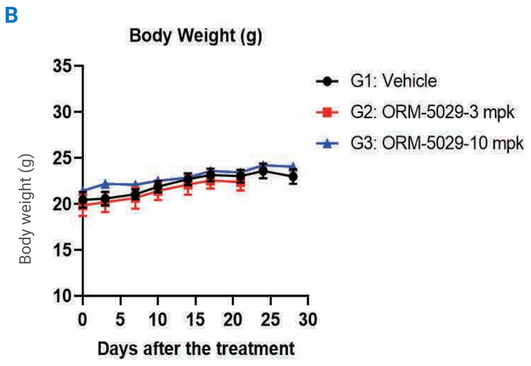


Figure 5. A-B, The in vivo antitumor efficacy of ORM-5029 was evaluated in a CDX model bearing HCC1569 cells, a HER2-high breast cancer cell line (n = 3 per group). ORM-5029 (10 mg/kg) demonstrated marked tumor growth inhibition compared with vehicle control, with no appreciable body weight loss.

Degrader and DAC: proteomics analysis

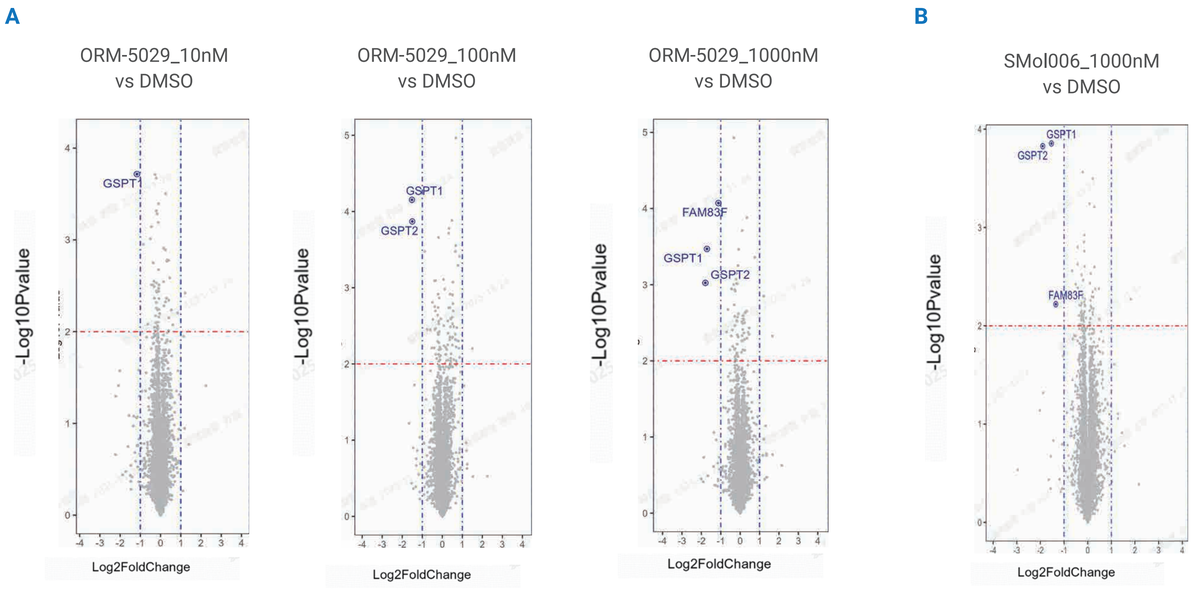


Figure 6. Proteomic analysis was performed in HCC1569 cells following 8-hour treatment with ORM-5029 (A) or SMol006 (B), with three biological replicates. Both compounds significantly depleted GSPT1, confirming efficient target engagement. Notably, at 1000 nM, both the ORM-5029 and SMol006 induced degradation of the off-targets GSPT2 and FAM83F. In addition, ORM-5029 showed a dose-dependent effect on additional candidate off-target proteins.

Summary

Drawing on 15 years of early drug discovery expertise—from target validation to preclinical candidate selection, ICE Bioscience provides end-to-end screening and evaluation platforms for degraders, antibodies, and DACs, empowering partners to accelerate DAC therapeutic R&D.

References

- Tsuchikama, K. et al. Exploring the next generation of antibody-drug conjugates. Nat Rev Clin Oncol. 2024, 21(3):203-223.
- Saini, S. et al. Assessment in Breast Cancer Patients from Phase I Clinical Trial Of ORM-5029, a Potent GSPT1 Degrader. AACR 2023. Abstract #2118.

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