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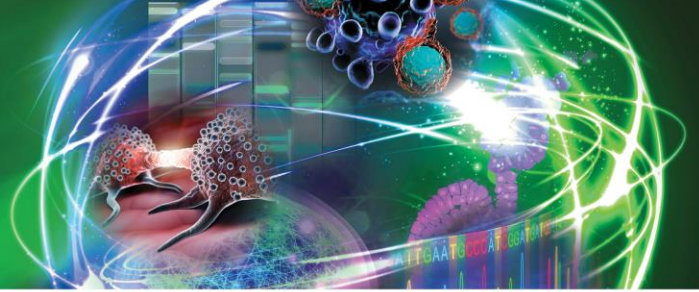
ANNUAL MEETING 2021
VIRTUAL #AACR21
APRIL 10-15, 2021 AND MAY 17-21, 2021 ★ AACR.ORG ★ AACR.ORG/AACR2021



Integrative surfaceome profiling identifies immunotherapeutic targets in osteosarcoma and preclinical testing of BT1769, an MT1-MMP-targeted *Bicycle*® toxin conjugate, in osteosarcoma by the Pediatric Preclinical Testing Consortium (PPTC)

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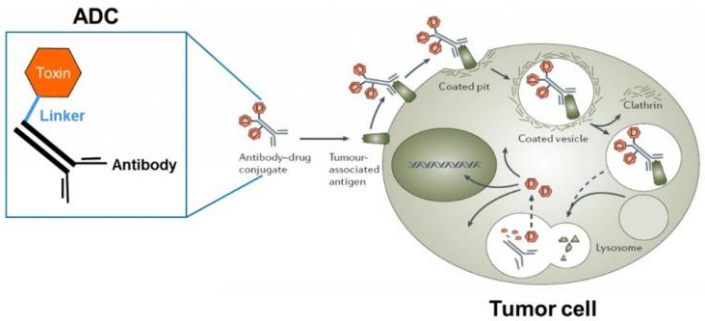
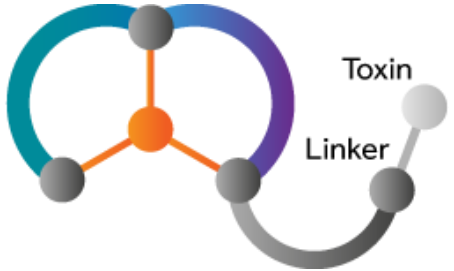
Disclosure Information

I have no financial relationships to disclose.

Introduction

- Current developments in the immunotherapeutic strategies show potential in the treatment of solid tumors.
- In osteosarcoma, surface proteins such as GD2 and HER2 have been identified as cell surface antigens of interest, but therapeutic mAb such as dinutuximab and trastuzumab were inadequate to generate robust clinical responses.
- Immunotherapies such as immunoconjugates or CAR-T cell therapy have not been sufficiently studied in OS.
- *Bicycle* Toxin Conjugates have low molecular weight (~4 kDa) compared to antibody drug conjugates (~150 kDa) - rapid tumor penetration and short half-life.

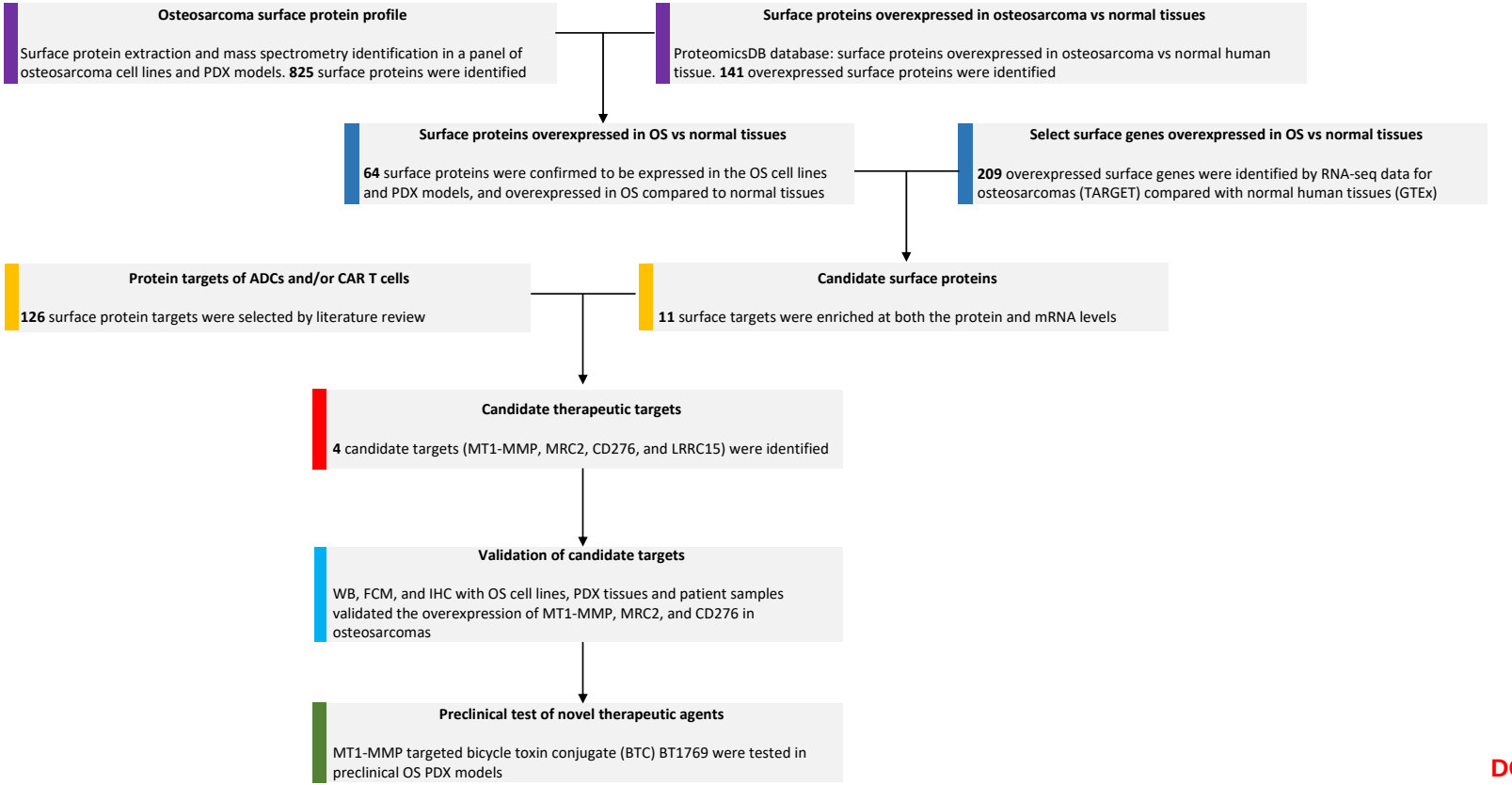




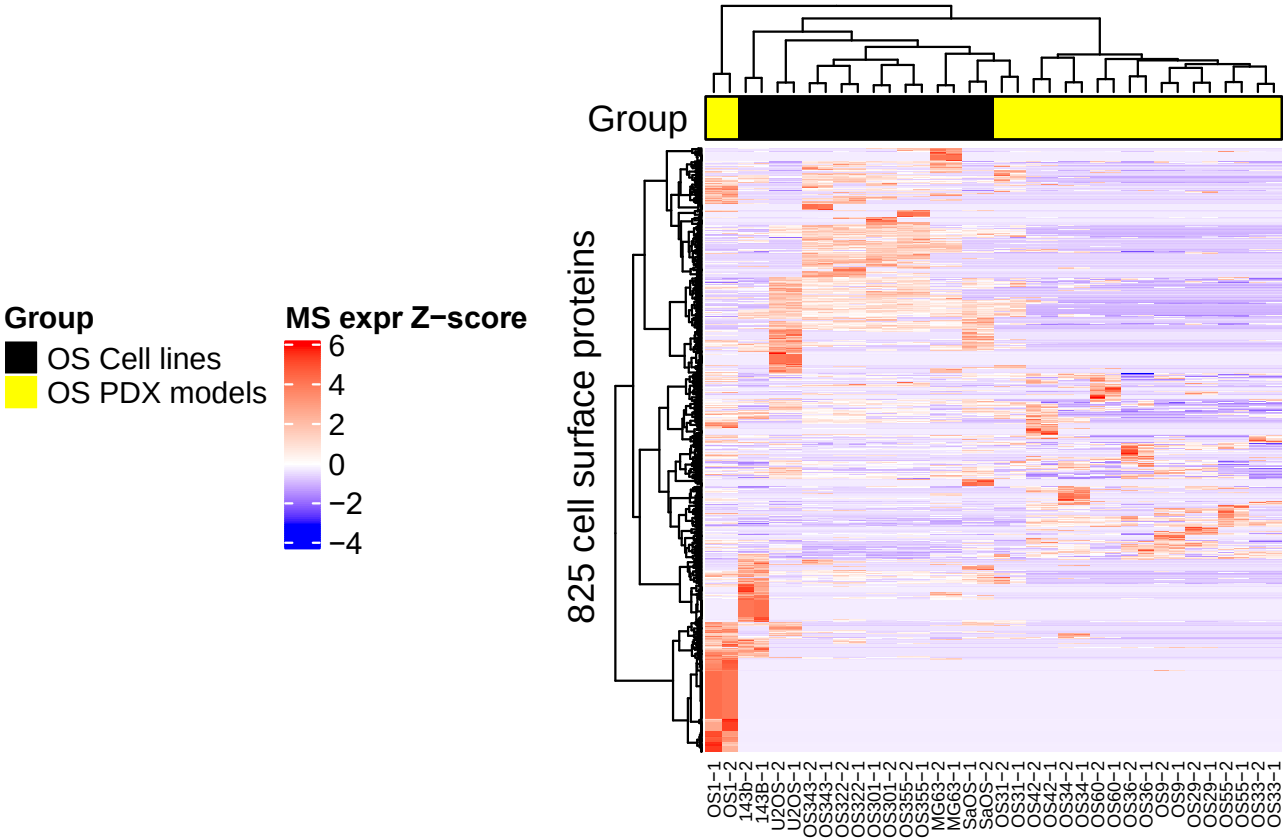
Identifying New Targets in Osteosarcoma

- Hypothesis: surface proteins such as those related to bone differentiation or other consistent osteosarcoma features may be targeted therapeutically.

Identification of candidate OS cell surface antigens: approach



825 surface proteins are identified in OS cell lines/PDX models by mass spectrometry



Integrative proteomic and transcriptomic analysis identifies overexpressed cell-surface proteins in OS

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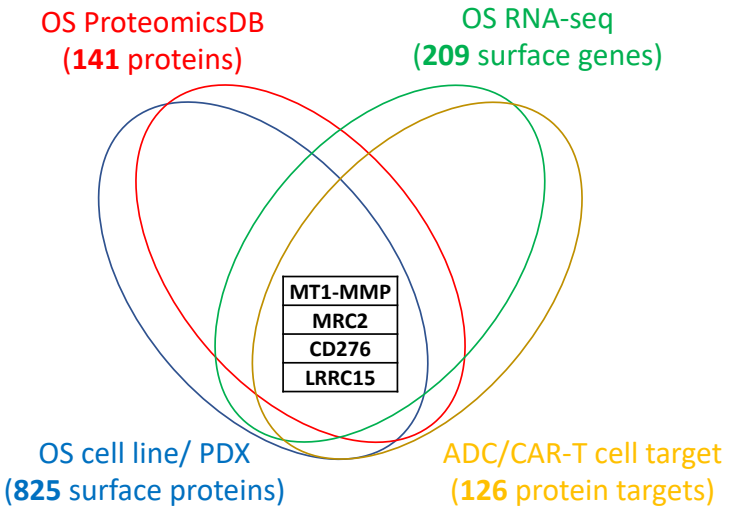
- Mass spectrometry: **825** surface proteins that were highly expressed in OS cell lines/PDX were identified by quantitative proteomics mass spectrometry.
- RNAseq: **209** OS specific plasma membrane-associated genes were identified.
- MS public database: **141** proteins overexpressed in OS were identified.
- The overlapped targets among these 3 datasets were filtered by current **126** ADC/CAR-T targets
- **4** surface proteins (MT1-MMP, MRC2, CD276, and LRRC15) were found to be enriched in OS by all RNA-seq and mass spectrometry datasets. There are immunoconjugates and/or *Bicycle* toxin conjugates targeting these 4 surface proteins.



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The expression of CD276, MRC2 and MT1-MMP in OS vs Normal tissues

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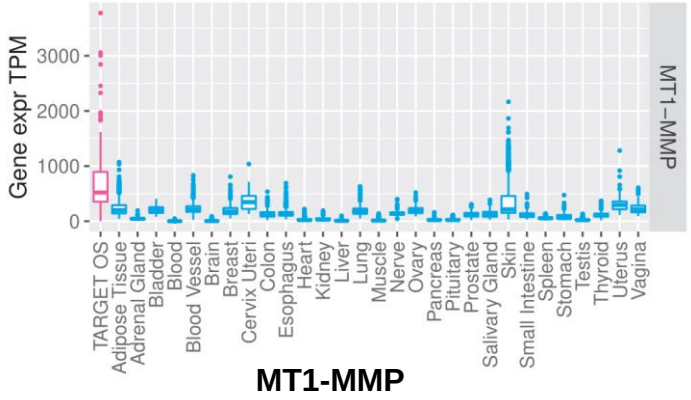
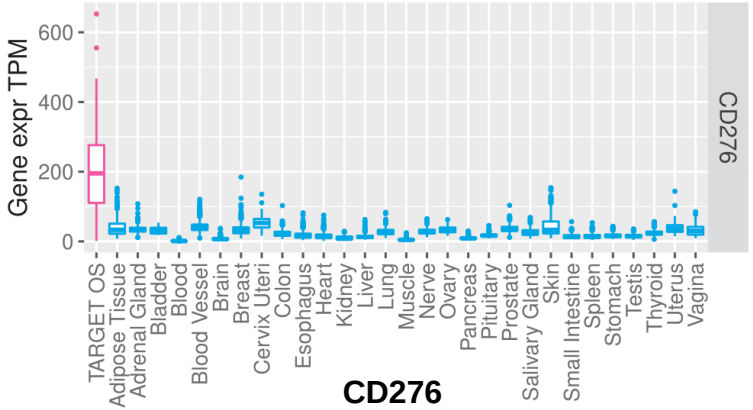
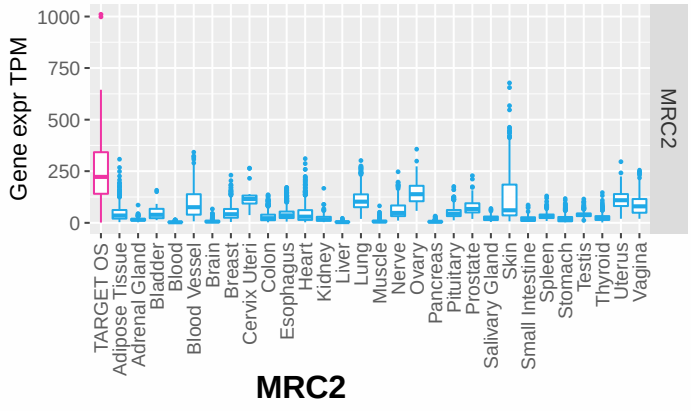


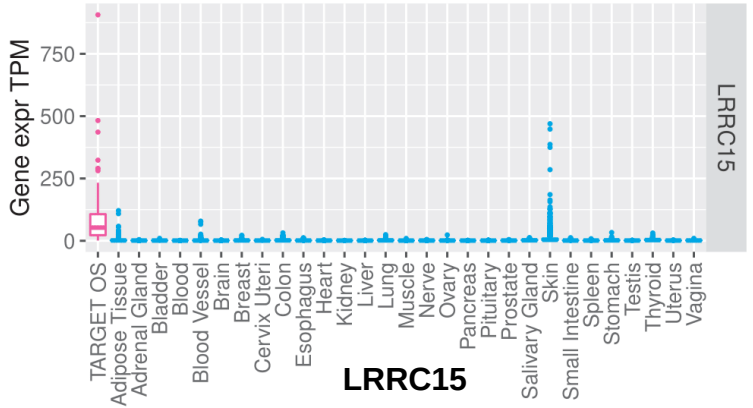
MT1-MMP



CD276



MRC2



LRRC15

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Validation of the expression of MT1-MMP, MRC2 and CD276 in OS cell lines and PDXs

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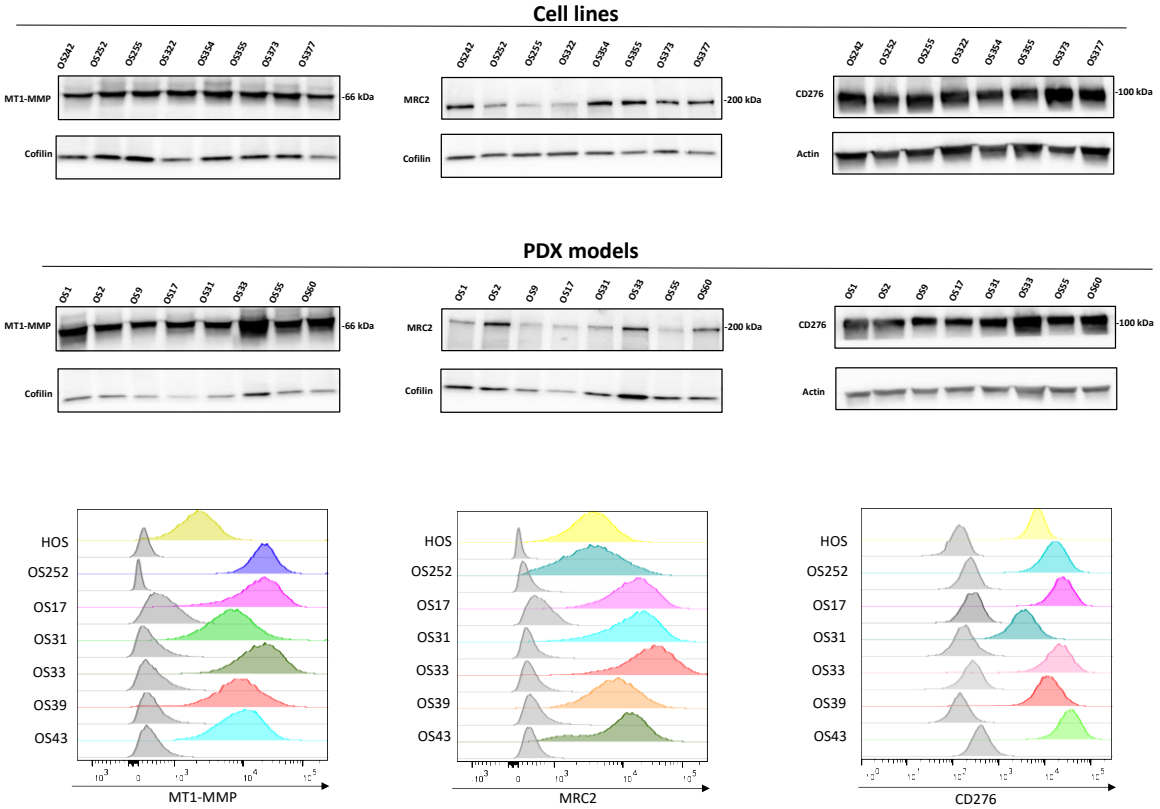

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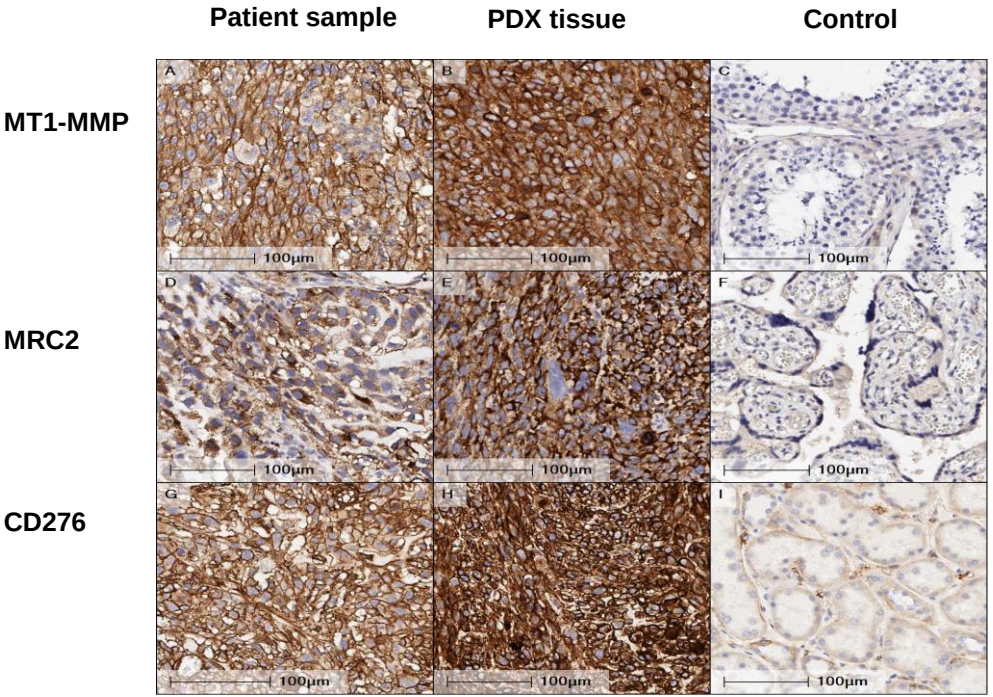
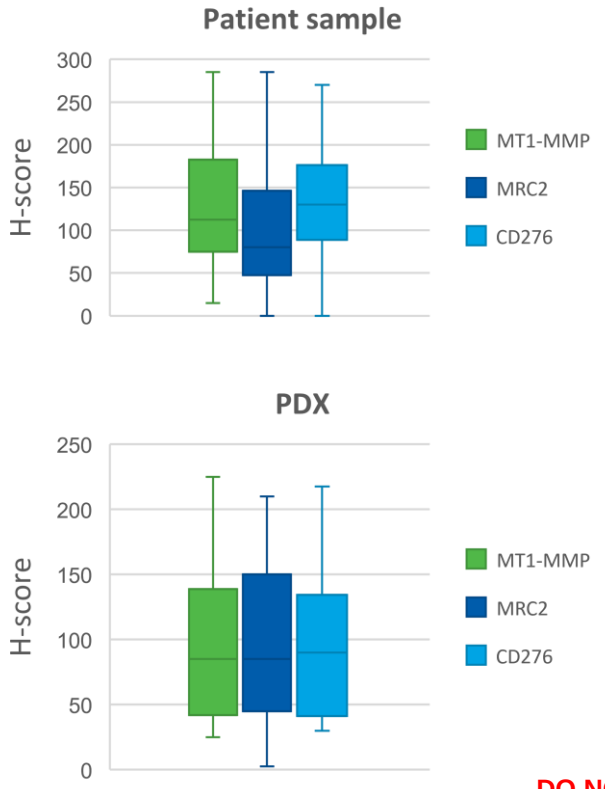
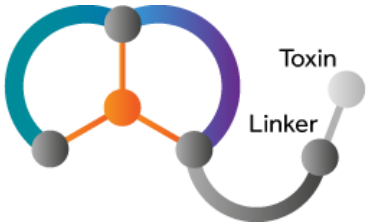
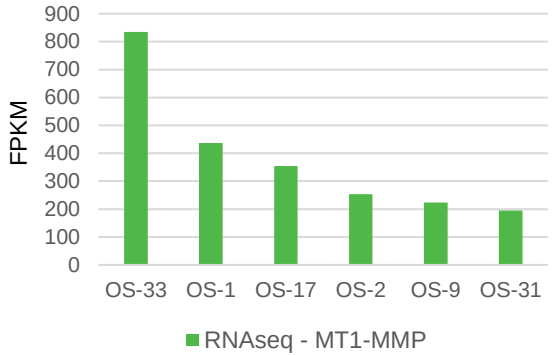
Validation of the expression of the candidate surface proteins - IHC with patient and PDX TMA

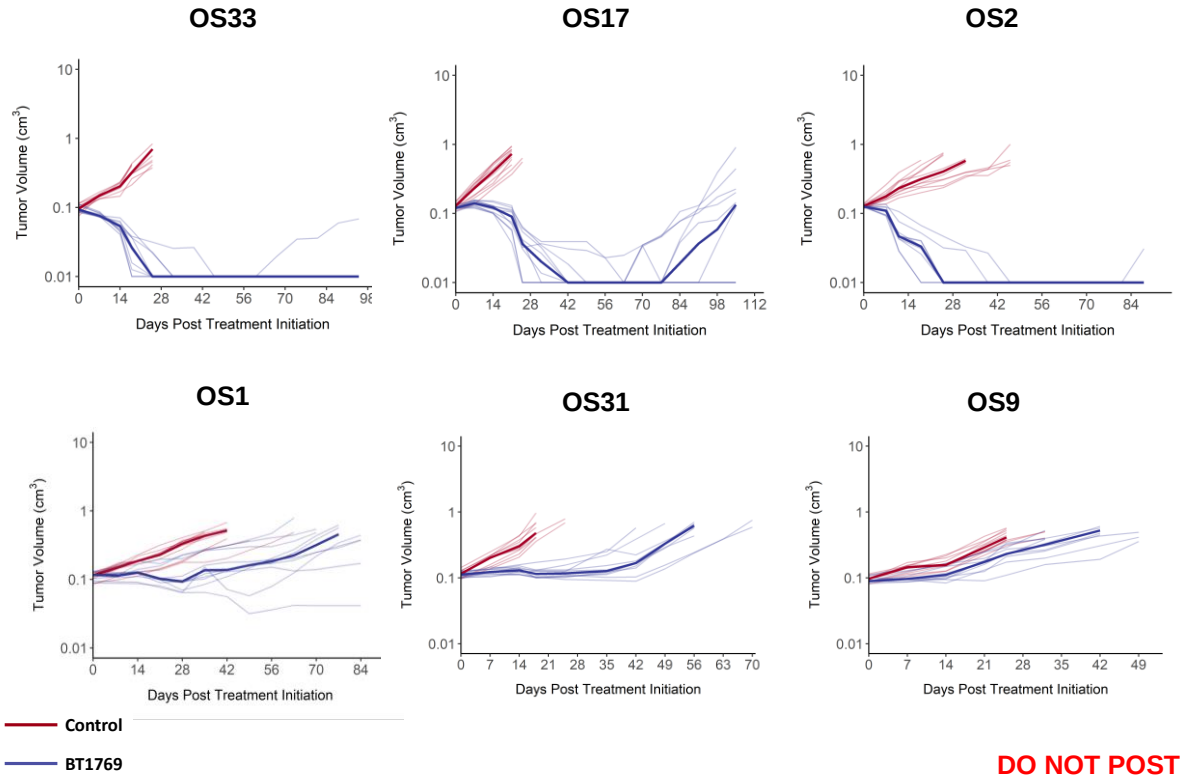




Preclinical test of MT1-MMP targeted BT1769







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Post-treatment recurrent tumor showed a lower MT1-MMP expression level

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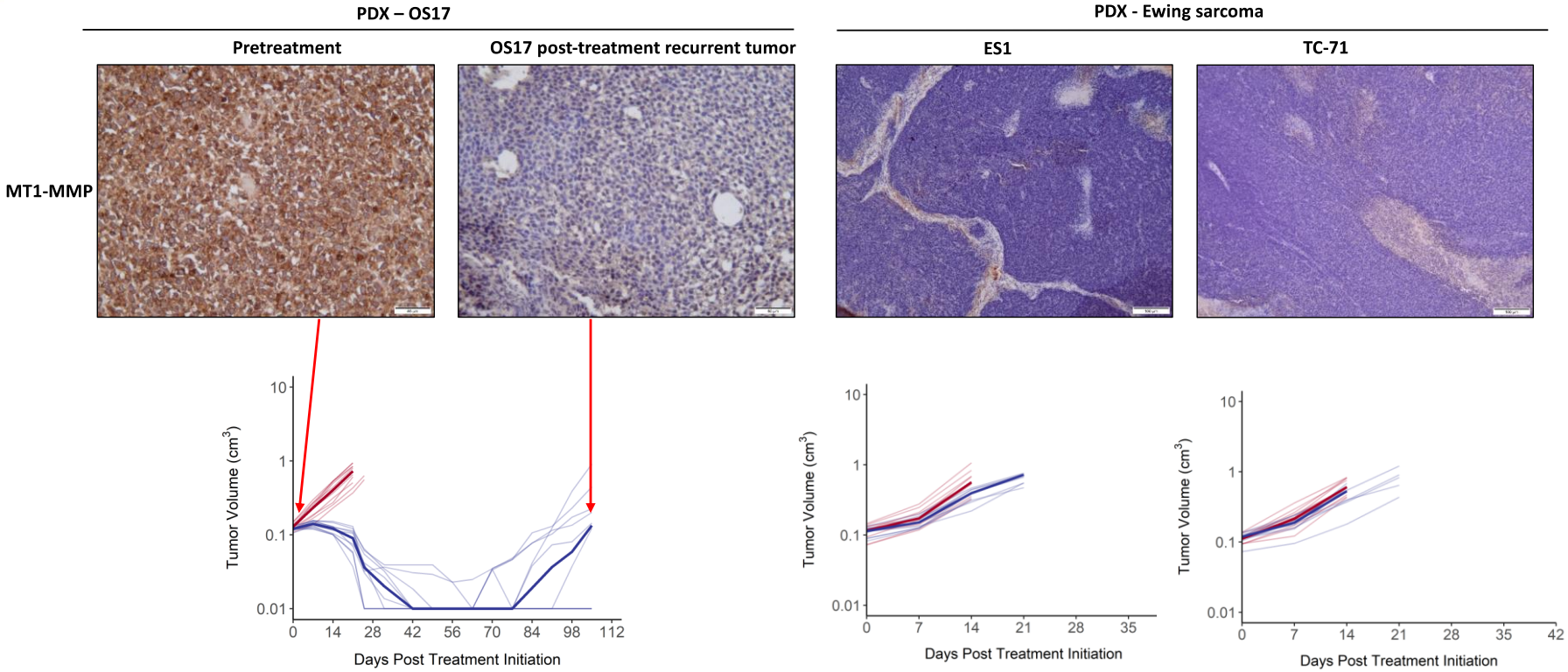

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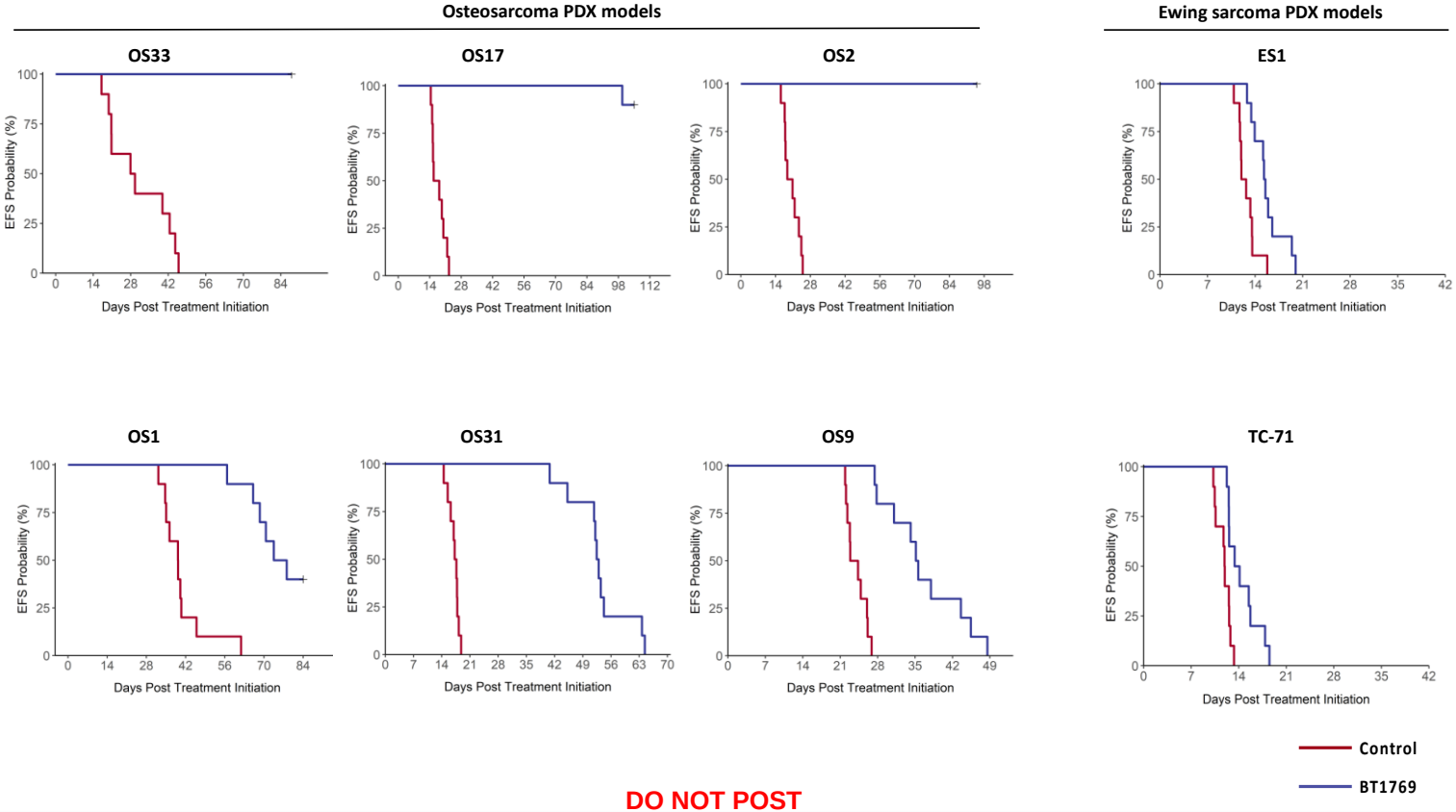
Preclinical test of MT1-MMP targeted BT1769



Summary

- Integrated proteomic and transcriptomic osteosarcoma surfaceome profiling identified high-confidence osteosarcoma cell-surface antigens (MT1-MMP, CD276, MRC2, and LRRC15) as therapeutic targets.
  - ADC targeting LRRC15 is active against osteosarcoma PDXs (Hingorani, et al. *Mol Cancer Ther* 2021)
  - ADC targeting CD276 is active against osteosarcoma PDXs (Kendsersky, et al. *Clinical Cancer Research*, 2021)

- MT1-MMP's high expression and BT1769's preclinical activity support the potential of MT1-MMP-targeted *Bicycles* in osteosarcoma.

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