Target Validation, Antibody Discovery and Preclinical Data Supporting ADAM9 as an Antibody-Drug Conjugate Therapeutic Target for Solid Tumors

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Abstract

Introduction: A target-validated approach based on intact cell immunizations with fetal progenitor cells or tumor stem cells followed by an immunotoxin technology (ITX) screen for cancer-specific candidates, led to the identification of anti-ADAM9 reactivity in a pan-cancer proteomics screen for ADAM9 overexpression.

Background: Overexpression of ADAM9 is observed in multiple carcinomas, including NSCLC, colon, gastric, pancreatic, and kidney cancers. ADAM9 is involved in the protein ectodomain shedding of membrane-bound molecules (disintegrins). Disintegrins are a family of metalloproteases with a Cystein-rich domain and a Metalloprotease domain.

Methods: Intact cell immunizations of mice with viable human fetal progenitor cells or tumor initiating/cancer stem-like cells were used to generate mAbs. Immunoprecipitation/mass spectrometry analysis of the panel of ADAM9-positive cell lines was performed to further evaluate the mAbs as ADC epitope-specificity. mAbs were also screened to identify those that strongly react with representative tumors to determine the therapeutic potential of anti-ADAM9 antibody-drug conjugates (ADCs) toward ADAM9-expressing solid cancers.

Results: Anti-ADAM9 mAbs exhibited strong reactivity toward the tumor epithelium of solid cancers, including pancreatic, kidney, prostate, bladder, breast, colon, lung, and ovarian cancer, but limited reactivity toward normal tissues. Anti-ADAM9 mAbs were efficiently internalized and processed by tumor cell lines, including lines with only modest ADAM9 expression. Anti-ADAM9 ADCs exhibited specific, dose-dependent cytotoxicity toward ADAM9-positive cancer cell lines in vitro, with IC50 values in the nanomolar range. Humanization and affinity maturation of the lead mAb yielded a development candidate that retains potent antitumor activity toward ADAM9-positive tumor cell lines and equivalent, high affinity binding to both human and cynomolgus monkey ADAM9.

Conclusion: ADAM9 is a cell surface antigen that is overexpressed on a wide range of solid tumors. Anti-ADAM9 mAbs that were strongly reactive with representative tumors exhibited high affinity for the antigen and were efficiently internalized and processed by ADAM9-bearing tumor cells. Anti-ADAM9 ADCs demonstrated dose-dependent cytotoxicity in vitro toward a panel of ADAM9-positive tumor cell lines. Our findings demonstrate that an ADC targeting ADAM9 may serve as a potential therapeutic for ADAM9-expressing solid tumors.

Antibody/Target Discovery Platform

ADAM9 is Highly Expressed on Range of Cancers

ADAM9 mAbs Retains Binding Affinity

ADAM9 mAbs are Rapidly Internalized and Processed

Background

- Intact cell immunizations of mice with viable human fetal progenitor cells or tumor stem cells were used to generate mAbs.
- An iPCR screen for cancer-specific mAbs identified a panel with high tumor-versus-normal differential and Immunoprofiling/mass spectrometry analysis of the panel identified mAbs that had specificity toward ADAM9.

Objectives

- Validate ADAM9 as a therapeutic target
- Evaluate the therapeutic potential of anti-ADAM9 antibody-drug conjugates

Antibody Target Discovery Platform

- Cancer Type
  - Pancreatic Cancer
  - Gastric Cancer
  - Breast Cancer
  - Prostate Cancer
- mAb [pM]
  - DU 145
  - NCI-H1703
  - MDA-MB-468
  - PA-1
  - SNU-16
  - SW48

Results

- ADAM9 is highly expressed on a range of cancers.
- Humanized ADAM9 mAbs retain binding affinity.
- ADAM9 mAbs are rapidly internalized and processed.

Conclusions

- ADAM9 is a cell surface antigen that is overexpressed on a wide range of solid tumors.
- Anti-ADAM9 mAbs are efficiently internalized and processed by ADAM9-expressing tumor cells.
- ADAM9 mAbs conjugated to a microtubule inhibitor (DM1) or a DNA alkylating agent (DN4) demonstrated dose-dependent cytotoxicity in vitro toward a panel of ADAM9-positive tumor cell lines.
- Humanized ADAM9-DM1 mAbs exhibited potent in vitro cytotoxicity toward ADAM9-expressing tumor cell lines.
- ADAM9-DM1 and ADAM9-DN4-based ADAM9 mAbs may serve as potential therapeutic targets for ADAM9-expressing solid tumors.

Summary of SPR Analysis

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<th>Cancer Type</th>
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<th>Binding of Cyno ADAM9</th>
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