DIFFERENT NEOANTIGEN EXPRESSION PATTERNS IMPACT THE STRENGTH OF ANTI-TUMOR IMMUNE RESPONSES

Kim Nguyen*, Stefani Spranger, Christopher Copeland. Massachusetts Institute of Technology, Cambridge, USA

Background Many cancer immunotherapies depend on the ability of cytotoxic CD8+ T cells to recognize neoantigens on MHCI complexes to effectively eliminate tumor cells. However, patient response following immunotherapy is highly variable, with recent work suggesting that neoantigen expression patterns can impair patient response. Specifically, it was observed that the immune response is dampened when neoantigens are expressed only by a subset of tumor cells (heterogeneous expression). 1 To study why anti-tumor immunity is reduced in a heterogeneous setting we developed a transplant murine tumor model engineered to express neoantigens in a heterogeneous pattern or homogeneously.

Methods A curated list of neoantigens with varying predicted MHC-I binding affinities was used to establish an array of cell lines expressing at one to three neoantigens. The lines were inoculated subcutaneously in immunocompetent mice as mixtures (heterogenous) or as a single line (homogenous) to study the resulting immune response. Tumors were harvested at days 7, 10 and 14 and flow cytometry analysis was used to phenotype infiltrating immune populations, including antigen-specific CD8+ T cells. ELISpot assays were performed using splenocytes from the same timepoints to determine the frequency of antigen-specific T cells in the periphery.

Results Compared to neoantigens predicted to bind weakly to MHCI, neoantigens predicted to bind strongly elicited robust expansion of antigen-specific T cells in the periphery and tumors expressing these antigens alone exhibited greater numbers of tumor infiltrating T cells. Homogenous expression of two neoantigens was found to enhance anti-tumor immunity by increasing the frequency of tumor-reactive T cells. Further, homogenous expression of two neoantigens induced protective immunity against antigens, including those that failed to be controlled when expressed alone.

Conclusions Using our novel reductionist tumor model, our results suggest that a more robust response against weak antigens could be induced if a response against a strong, highly immunogenic neoantigen is mounted simultaneously. This observation has direct implications for the design of neoantigen vaccines either as mono- or combination immunotherapies, especially in the setting of a heterogeneous neoantigen expression pattern.

REFERENCES

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0508

509 POTENT AND SELECTIVE INHIBITION OF AXL RECEPTOR TYROSINE KINASE FOR THE TREATMENT OF CANCER

Susan Papocka*, Akshata Udyavar, Subhasree Sridhar, Dillon Miles, Yu Chen, Sean Cho, Corinne Foley, Rebecca Grange, Mammohan Leleti, Sharon Zhao, Lixia Jin, Stephen Young, Jay Powens, Matthew Walters. Arcus Biosciences, Hayward, CA, USA

Background AXL receptor tyrosine kinase (AXL) is a transmembrane protein that is over-expressed in a variety of cancer and immune cells. AXL signaling has been implicated in creating an immunosuppressive tumor microenvironment (TME) through both tumor-intrinsic and immunomodulatory mechanisms1,2,3,4,5 promoting resistance to various therapies.6,7,8,9

Methods Compound inhibition potency against the kinase activity of AXL and other kinases was determined by detecting phosphorylated substrate using homogeneous time-resolved fluorescence (HTRF). Binding affinity of inhibitor to intracellular AXL kinase was determined by monitoring displacement of a competitive fluorescent tracer using an AXL NanoBRET assay. Recombinant Gas6, cancer cell lines, whole blood or isolated cells from healthy donors were used to determine the reduction in AXL-mediated signaling in-in vitro. PK/PD and anti-tumor effects of selected AXL inhibitors were evaluated in murine models.

Results AXL is highly expressed on a subset of immune cells, including DC’s, NK cells and M2 macrophages as well as fibroblasts, which contribute to a blunted anti-tumor response. Consistent with these observations, AXL is strongly associated with increased infiltration of macrophages, exhausted NK and T-cells, as well as significantly increased CD73 expression in multiple cancer types in TCGA. Additionally, AXL expression is strongly and significantly correlated with epithelial-mesenchymal transition (EMT), which further generates an immunosuppressive TME and promotes resistance to immune, targeted and chemotherapies. High expression of AXL is also strongly associated with poor survival in NSCLC, pancreatic, breast, head & neck, stomach, colorectal, ovarian & prostate adenocarcinomas, especially in the metastatic setting. AXL inhibitors that exhibit high potency in both biochemical (IC <5nM) and cell-based (IC <25nM) assays in addition to good selectivity against closely related kinases MER and TYRO3 (>90x and >25x fold selectivity, respectively) as well as other kinases involved in downstream signaling such as PI3K have been developed. Initial studies in animal models indicate a favorable pharmacokinetic profile and anti-tumor efficacy.

Conclusions AXL is a promising therapeutic target involving both immunomodulatory and tumor-intrinsic mechanisms. AXL inhibition reduces the immunosuppressive TME, enables activation of an anti-tumor immune response and renders tumors more susceptible to previously resistant therapies. Highly potent and selective AXL inhibitors have been designed, displaying biological profiles superior to those of less-selective molecules currently advancing through clinical development.

REFERENCES