Pharmacodynamic biomarkers for anti-TIGIT treatment and prevalence of TIGIT expression in multiple solid tumor types


INTRODUCTION

TIGIT (T cell immunoreceptor with Ig and ITIM domains) is a co-inhibitory receptor on T cell and feature killer (NK) cell activity in the healthy immune system.

In tumors, TIGIT is highly expressed on a subset of desmoplastic T and NK cells and on highly expressive regulatory T cells (Treg).

Loss of TIGIT signaling enhances NK activity, CD4+ T cell priming and CD8+ T cell effector functions, suggesting a role in anti-tumor immunity.

We have generated an anti-TIGIT antibody, DMP-3184,32 to stimulate anti-tumor immunity. As a routine surrogate for preclinical studies, we generated 3184R, an anti-TIGIT blocking antibody that showed dose dependent, potent single agent efficacy in multiple syngeneic mouse models. Pharmacodynamic (PD) biomarkers in blood and in tumor were also identified, using flow cytometry, immunohistochemistry and gene expression analysis.

Our preclinical biomarker data can be utilized to demonstrate target engagement for our clinical stage anti-TIGIT antibody, DMP-3184.

MATERIALS AND METHODS

3184R is a murine antibody mouse IgG2a monoclonal antibody that binds to TIGIT (produced by Oncomed Pharmaceuticals).

Female BALB/c and C57Bl/6 (NCR-/-) mice, 6-8 weeks old, were obtained from Charles River Laboratories. The antibodies were produced by Oncomed Pharmaceuticals and purified by Protein A column chromatography. The antibodies were formulated in saline or PBS and administered via intraperitoneal injection. All animals were housed in a pathogen-free facility and treated according to the National Institutes of Health guidelines.

For flow cytometry analysis, mice were sacrificed by CO2 asphyxiation. Single cell suspensions were prepared from tumors and spleens. Cells were stained with surface and intracellular markers and analyzed by flow cytometry using a FACSCanto II and FACSCalibur electronic systems (BD Immunocytometry Systems). Data were acquired and analyzed using FlowJo v10.2 software (TreeStar). For intracellular cytokine staining, cells were fixed and permeabilized using BD Cytofix/Cytoperm (BD Biosciences) for intracellular cytokines and MHC class II (BD Pharmingen) for CD83. Data were acquired and analyzed using FlowJo v10.2 software (Tree Star).

All data were analyzed by Oncomed Pharmaceuticals and the data are presented as mean ± SEM. Differences between two groups were analyzed using Student’s t test. Data were analyzed using GraphPad Prism v6 software. The experiment was performed in duplicate.

RESULTS

Anti-TIGIT treatment shows antitumor efficacy in multiple syngeneic models

We generated a human anti-TIGIT antibody, which has been shown to have significant antitumor efficacy in preclinical studies. We administered this antibody to mice with established tumor burdens and found that it significantly reduced tumor growth. Furthermore, we observed increased infiltration of CD8+ T cells and NK cells into the tumors, indicating a potential mechanism of action.

Prevalence of TIGIT-expressing immune cells in human tissues

We performed a flow cytometry analysis of primary tumor samples from patients with various tumor types. We found that TIGIT expression was highest in melanoma and colorectal cancer tissues. In contrast, TIGIT expression was low in lung cancer tissues.

CONCLUSIONS

Using a surrogate anti-TIGIT blocking antibody, we show potent single agent dose-dependent antitumor efficacy on large established tumors from different syngeneic models.

Anti-TIGIT treatment promoted dose-dependent infiltration and activation of CD8+ and CD4+ T cells in the tumor microenvironment.

Anti-TIGIT treatment increased NK cell activation in tumors and increased NK cell cytotoxicity in the spleen, which correlated with dose and efficacy.

Concurrent with the mechanism of action, anti-TIGIT treatment promoted plegia expression of immune genes associated with activation of T and NK cells, TH1 response and cytotoxic activity. Results from blood and tumors were consistent, suggesting that blood may be a good surrogate tissue for measuring PD markers.

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anti-TIGIT treatment or saline control. Two tumor types in the Cancer Genome Atlas by RNA-Seq showed a global increase in the expression level of TIGIT and T cell markers (supplementary graphics showing correlation of TIGIT and CD3B in human prostate cancer). The data were analyzed using GraphPad Prism v6 software. The experiment was performed in triplicate.

Table in panel A shows the ranking of indications with high, moderate, low levels of TIGIT expression across all tumor types. The expression level of TIGIT is higher in melanoma and colorectal cancer than in other tumor types. The expression level of TIGIT is lower in lung cancer than in other tumor types.

The overall incidence of TIGIT receptor on the plasma membrane is less in tumor cells than in normal tissues. The data show that TIGIT expression is decreased in tumor cells from patients with advanced stages of cancer. The data were analyzed using GraphPad Prism v6 software. The experiment was performed in triplicate.

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