BGB-B2033, a novel 4-1BB/GPC3 bispecific antibody, exhibits potent in vitro and in vivo antitumor activity in preclinical models

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Abstract

The tumor necrosis factor receptor superfamily member 4-1BB (CD137) is a key costimulatory receptor of T cells and a promising therapeutic target in cancer. Glypican-3 (GPC3) is a well-established tumor-associated antigen (TAA) overexpressed in a variety of solid tumors, including hepatocellular carcinoma, germ cell tumors, squamous non-small cell lung cancer, alpha-fetoprotein producing gastric cancer, pancreatic carcinoma, and to a lesser extent in other tumors. GPC3 is not expressed in adult normal tissues (except endometrium and placenta), thus is recognized as an ideal TAA.

BGB-B2033 is a novel IgG-based bispecific antibody targeting 4-1BB and GPC3 and is under clinical development for the treatment of advanced or metastatic solid tumors in humans. BGB-B2033 binds to its target proteins with high specificity and affinity. Potent and GPC3-dependent functional activities were demonstrated using the human peripheral blood mononuclear cell (PBMC)-based immune cell activation and cytotoxicity assays. In humanized 4-1BB knock-in mice bearing human GPC3-expressing tumors, BGB-B2033 exhibited potent, dose-associated single-agent efficacy as well as synergistic antitumor activity in combination with anti-PD-1 antibody.

The phase 1 study of BGB-B2033, alone or in combination with Tislelizumab is ongoing (NCT06427941). Here, we describe the characterization of BGB-B2033 regarding its mechanism of action and preclinical activities.

Molecule Design

BGB-B2033 is a novel IgG-based bispecific antibody (BsAb) targeting GPC3 and 4-1BB. It includes a bivalent F(ab')2 fragment that binds to GPC3, a fusion of 4-1BB-binding heavy chain variable (VH) domain fragments, and a silenced Fc that prevents binding to FcγRs (**Figure 1**). BGB-B2033 can only activate 4-1BB receptors when GPC3 is present, thus resulting in the immune cell stimulation in the tumor microenvironment while greatly reducing risk of systemic toxicity (**Figure 2**).

Figure 1. BsAb design

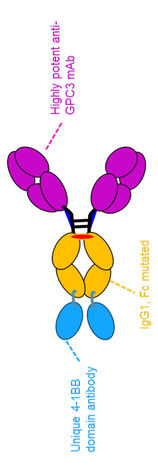
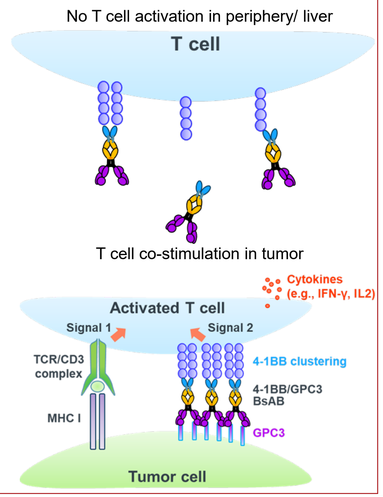
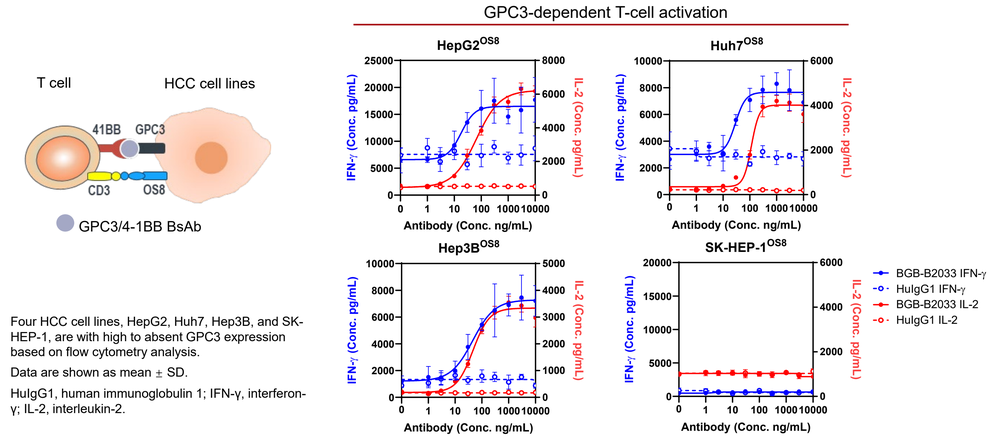


Figure 2. Mechanism of action



Enhanced IFN-γ and IL-2 Secretion from PBMCs in a GPC3-Dependent Manner

Figure 3. PBMCswere co-cultured with GPC3-expressing HCC cells in the presence of serial diluted BGB-B2033. BGB-B2033 enhanced IFN-γ and IL-2 secretion in GPC3-dependent manner.



Four HCC cell lines, HepG2, Huh7, Hep3B, and SK-HEP-1, are with high to absent GPC3 expression based on flow cytometry analysis.

Data are shown as mean ± SD.

HuIgG1, human immunoglobulin 1; IFN-γ, interferon-γ; IL-2, interleukin-2.

Dose-Dependent Antitumor Monotherapy Activity in Human 4-1BB Knock-in Mice

Figure 6. Hepa1-6/hGPC3 cells were implanted into human 4-1BB knock-in mice. BGB-B2033 significantly inhibited tumor growth in the Hepa1-6/hGPC3 model; tumor-free rates at study end were 30% (3/10) and 90% (9/10) for dose level 1 and 2 treatment groups.

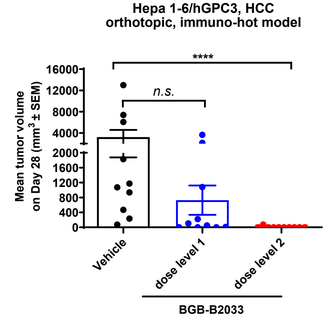
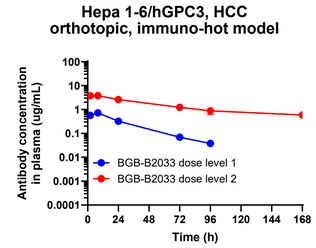


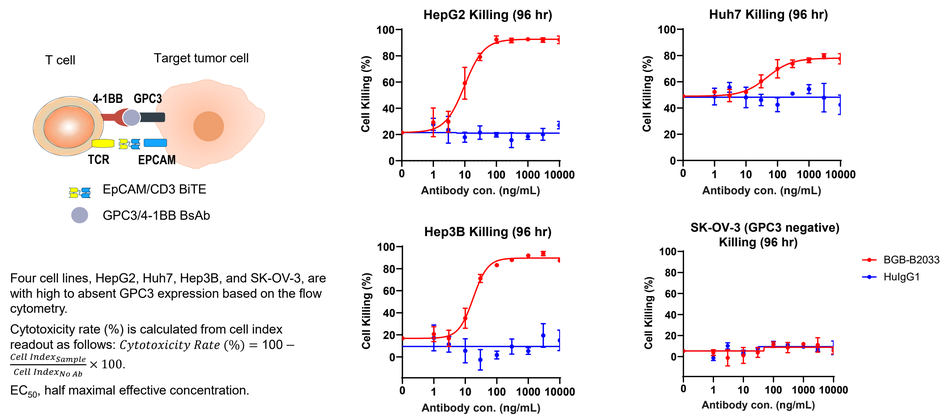
Figure 7. Hepa1-6/hGPC3 tumor bearing human 4-1BB knock-in mice were treated with BGB-B2033. Plasma concentration was quantified by ELISA.

Hepa1-6/hGPC3 model: human GPC3 expressed in mouse hepatoma cell line Hepa1-6.



Potent Cytotoxicity of PBMCs Against Cancer Cells

Figure 4. PBMCs were co-cultured with GPC3-expressing target tumor cell in presence of BGB-B2033. BGB-B2033 dose-dependently enhanced T-cell killing activity in GPC3-expressing cells, and no killing activity observed in GPC3-negative cells.



Four cell lines, HepG2, Huh7, Hep3B, and SK-OV-3, are with high to absent GPC3 expression based on the flow cytometry.

Cytotoxicity rate (%) is calculated from cell index readout as follows: $Cytotoxicity\ Rate\ (\%) = 100 - \frac{Cell\ Index_{Sample}}{Cell\ Index_{No\ Ab}} \times 100$.

EC_{50}, half maximal effective concentration.

Enhanced Antitumor Activity when Combined with Anti-PD1 Ab

H22/hGPC3 or LL2/hGPC3 cells were implanted into human 4-1BB knock-in mice. The mice were treated with BGB-B2033, anti-mouse PD-1 antibody Ch15mt, or a combination of both.

Figure 8. Tumor growth inhibition rate in the combination group was significantly higher in H22/hGPC3 HCC model

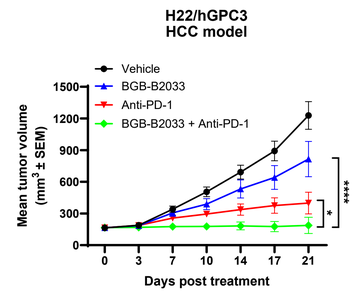
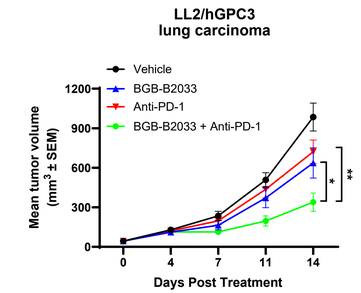
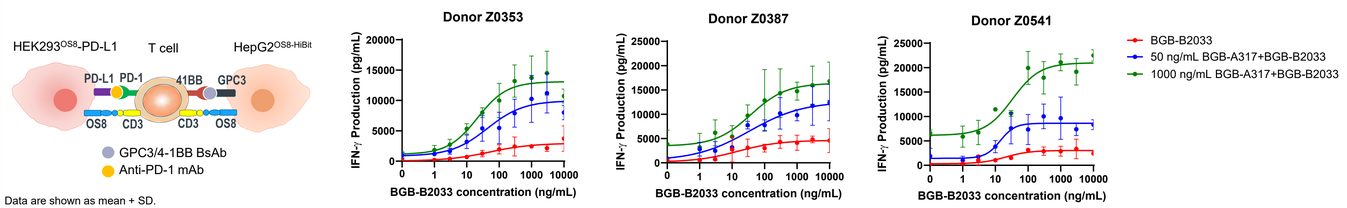


Figure 9. Tumor growth inhibition rate in the combination group was significantly higher in LL2/hGPC3 lung carcinoma model



Combination with Anti-PD1 Ab Promotes IFN-γ Secretion from PBMCs

Figure 5. PBMCs were co-cultured with GPC3-expressing HepG2 cells and engineered HEK293 cells (with PD-L1) in the presence of serial diluted BGB-B2033 and BGB-A317 (anti-PD1 Ab). Combination of BGB-A317 and BGB-B2033 enhanced the maximum IFN-γ production.



Data are shown as mean + SD.

Conclusion

BGB-B2033 is a selective GPC3-dependent 4-1BB targeting bispecific antibody and demonstrated strong antitumor effects as monotherapy or in combination with anti-PD1 antibody in pre-clinical setting with the following evidences:

- GPC3-dependent T-cell activation in *in vitro*
- Strong antitumor effects in animal studies
- Enhanced antitumor activity in combination with anti-PD1 antibody.