HEXABODY-CD27 ENHANCES T-CELL ACTIVATION, PROLIFERATION, CYTOKINE SECRETION AND CYTOTOXIC ACTIVITY INDEPENDENTLY OF FC GAMMA RECEPTOR-MEDIATED CROSSLINKING


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Background Clustering of CD27 on the plasma membrane of T cells induces T-cell activation, proliferation, and differentiation. Therefore, this costimulatory receptor represents a target for cancer immunotherapy. Multiple monoclonal antibodies (mAbs) targeting CD27 are being explored in the clinic, which require Fc gamma receptor (FcγR)-mediated crosslinking to induce CD27 agonism. HexaBody-CD27 (GEN1053/BNT313) is a novel anti-CD27 mAb with an IgG Fc domain engineered to induce CD27 agonist activity independently of FcγR-bearing cells, which may be scarce in tumors. The Fc domain was further modified to silence Fc-mediated antibody effector functions, with the aim to prevent T-cell depletion. Here we present preclinical characterization of the mechanism of action of HexaBody-CD27.

Methods Target binding characteristics and functional activity of HexaBody-CD27 were analyzed in vitro using flow cytometry, cell-based reporter assays and primary human lymphocyte assays. The capacity of HexaBody-CD27 to induce tumor-infiltrating lymphocyte (TIL) proliferation was assessed ex vivo using non-small cell lung cancer (NSCLC) tissue resected from patients. HexaBody-CD27 activity in vivo was investigated in human CD27 knock-in mice that were immunized with ovalbumin and treated with HexaBody-CD27 by characterizing peripheral blood and splenic T cells using flow cytometry.

Results HexaBody-CD27 exhibited dose-dependent CD27 agonist activity in reporter assays, independent of crosslinking via FcγR-expressing cells. In contrast, agonist activity of benchmark anti-CD27 antibody analogs was dependent on FcγR-mediated crosslinking. HexaBody-CD27 did not functionally engage with FcγRs, and membrane-bound HexaBody-CD27 was unable to bind C1q, confirming functional silencing of the IgG Fc domain. In vitro, HexaBody-CD27 enhanced activation, proliferation, and proinflammatory cytokine secretion of TCR-stimulated human CD4+ and CD8+ T cells as well as CD8+ T-cell mediated cytotoxic activity towards cognate antigen-expressing tumor cells. In TIL assays with human NSCLC tumor tissue, HexaBody-CD27 promoted expansion of CD8+ T cells. In human CD27 knock-in mice, HexaBody-CD27 enhanced expansion and IFN-γ secretion of antigen-specific CD8+ T cells. No decrease in percentages of circulating or splenic T cells was detected in vivo after treatment with HexaBody-CD27, whereas treatment with a benchmark anti-CD27 mAb analog resulted in a marked reduction of T cells.

Conclusions HexaBody-CD27 has a functionally inert Fc domain and exhibits FcγR-crosslinking-independent CD27 agonist activity, a unique mechanism of action that distinguishes HexaBody-CD27 from benchmark mAbs targeting CD27. In preclinical studies in vitro and in vivo, HexaBody-CD27 increased T-cell activation, proliferation, cytokine secretion, and cytotoxic activity. A first-in-human clinical trial has been initiated to evaluate HexaBody-CD27 in patients with advanced solid tumors (NCT05435339).