Background Immune checkpoint inhibition (ICI) using Fc-containing monoclonal antibodies has emerged as a powerful therapeutic approach to augment anti-tumor immunity. While ICI has drastically improved patient outcomes in melanoma, there is still variability in patient response. 1, 2 We recently showed that FcγRIIB, the only inhibitory IgG-Fc receptor, is expressed on differentiated effector CD8 T cells in mice and humans. 3–4, raising the possibility that CD8 T cell responses may be directly modulated by checkpoint inhibitor binding to T cell-expressed FcγRIIB.

Methods Flow cytometric phenotyping was performed on PBMCs isolated from melanoma patients and healthy donors. For in vitro functional experiments, healthy human PBMCs were stimulated with CD3/28 Dynabeads and/or PMA/ionomycin. Effector function was assessed through intracellular cytokine staining. Anti-PD1 (clone J116) or anti-CTLA4 (clone 1C10) antibodies were used where mentioned and the anti-PD1 F(ab) was generated from Nivolumab. In vivo experiments were performed in mice with B16-hgp100, B16-OVA, or LLC-OVA tumors. Where mentioned, WT OT-I, Fcgr2b-/- OT-I, or WT pmel-17 CD8 T cells were adoptively transferred into these mice. For treatment, 250 μg of anti-PD1 (clone RMP1-14) and anti-FcγRIIB (clone 2.4G2) or isotype antibodies were administered for blockade experiments.

Results Here, we show that despite exhibiting strong proliferative and cytokine responses at baseline, human FcγRIIB+ CD8 T cells exhibited reduced responsiveness to both PD-1 and CTLA-4 checkpoint inhibition compared to FcγRIIB- CD8 T cells in vitro (p<0.05). Moreover, frequencies of FcγRIIB+ CD8 T cells were reduced following treatment of human melanoma patients with nivolumab in vivo (p<0.05). This reduced responsiveness was FcγRIIB-dependent, because conditional genetic deletion of FcγRIIB on tumor-specific CD8 T cells improved response to checkpoint blockade in a B16 mouse melanoma model (p<0.01). The limited responsiveness of FcγRIIB+ CD8 T cells was dependent on an intact Fc region of the checkpoint inhibitor, in that treatment with Fc-devoid anti-PD-1 F(ab) fragments resulted in a significant increase in proliferation of FcγRIIB+ CD8 T cells, without altering the response of FcγRIIB- CD8 T cells (p<0.05). Finally, blocking FcγRIIB in the context of PD-1 blockade significantly improved anti-tumor CD8 T cell responses in B16 melanoma and in Lewis lung cancer mouse models (p<0.05, p<0.001).

Conclusions These results illuminate an FcγRIIB-mediated, cell-autonomous mechanism of CD8 T-cell suppression which limits the efficacy of checkpoint inhibitors in vivo. The data presented here support the novel conclusion that CD8-expressed FcγRIIB is both a factor to consider in the development of therapeutic antibodies, and a new potential target for immunotherapeutic intervention.

REFERENCES


