Poster No. 1281

SGN-B7H4V shows immunomodulatory activity through induction of immunogenic cell death

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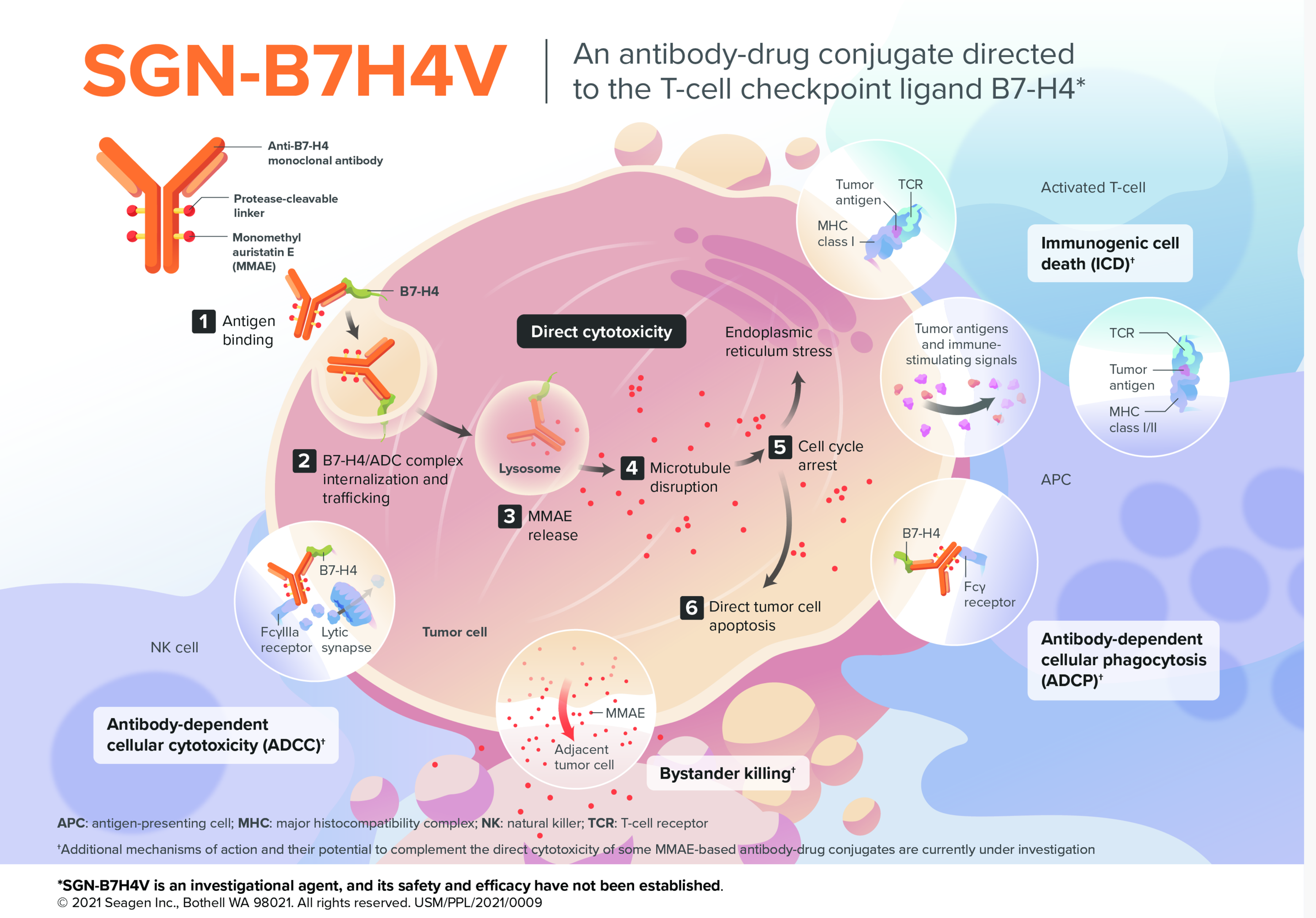
Background

- SGN-B7H4V is a novel, investigational antibody drug conjugate (ADC) composed of a B7-H4-directed antibody (B7H41001 mAb) conjugated via a protease cleavable linker to the clinically validated vedotin payload [2-4].
- Previously, we have demonstrated that expression of the immune checkpoint ligand B7-H4 is elevated on a variety of solid tumors including breast, ovarian, and endometrial tumors [1,5,8].
- SGN-B7H4V is designed to bind and internalize the immune checkpoint ligand B7-H4/ADC complex from the surface of malignant cells and release the cytotoxic payload MMAE.
- SGN-B7H4V can induce tumor cell death through several mechanisms, including MMAE-mediated direct and bystander cytotoxicity as well as antibody-dependent cellular cytotoxicity (ADCC) and phagocytosis (ADCP) [8].
- Vedotin ADCs have been described to elicit antitumor immune responses in part through MMAE-mediated induction of immunogenic cell death (ICD). These immunomodulatory effects potentially position vedotin ADCs to uniquely combine with checkpoint inhibitors, supported by recent clinical activity observed when vedotin ADCs are paired with anti-PD1 agents [6,7].
- Here, we characterize SGN-B7H4V-mediated ICD and subsequent immunomodulatory activity. We also evaluate the contribution of SGN-B7H4V-induced immune activation to antitumor activity in combination with an anti-PD1 agent.

Proposed Mechanism of Action



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Disclosures: All authors are employees of and/or hold stock in Seagen, Inc.

SGN-B7H4V Induces Hallmarks of ICD in Vitro

SGN-B7H4V induces ATP release and calreticulin exposure

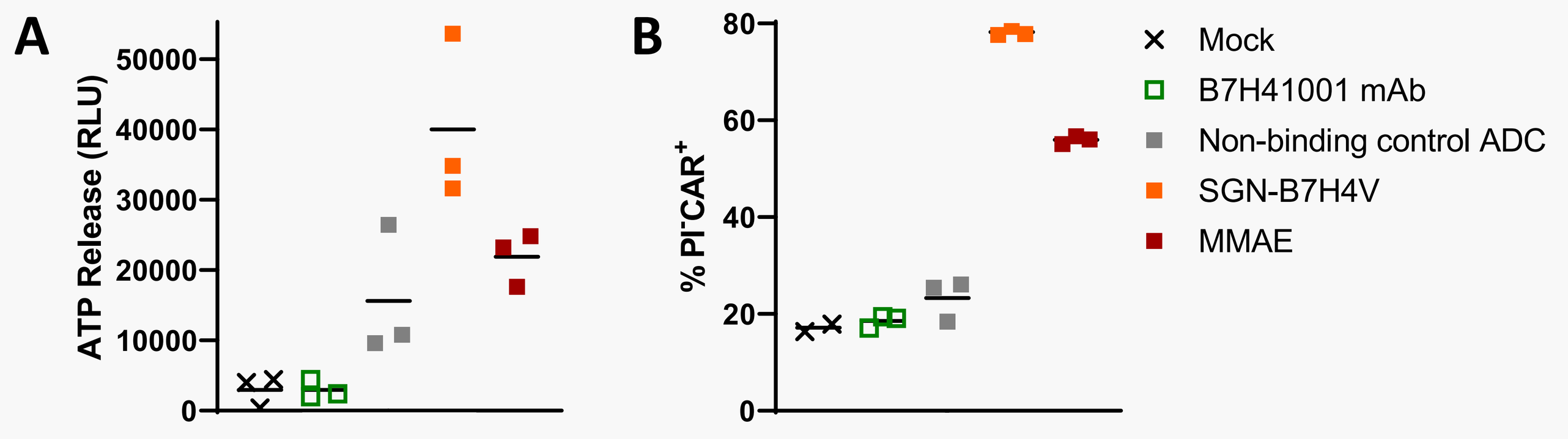

Figure 1. SGN-B7H4V induces secretion of ATP as well as surface exposure of calreticulin. SGN-B7H4V and MMAE drove ATP release **(A)** as well as cell surface exposure of calreticulin **(B)** by SKBR3 cells 48 hours following treatment with 1 µg/mL SGN-B7H4V or non-binding control ADC or 100 nM MMAE free drug.

SGN-B7H4V Demonstrates Immunomodulatory Activity in a TNBC Xenograft Tumor in Vivo

SGN-B7H4V recruits macrophages to a xenograft tumor

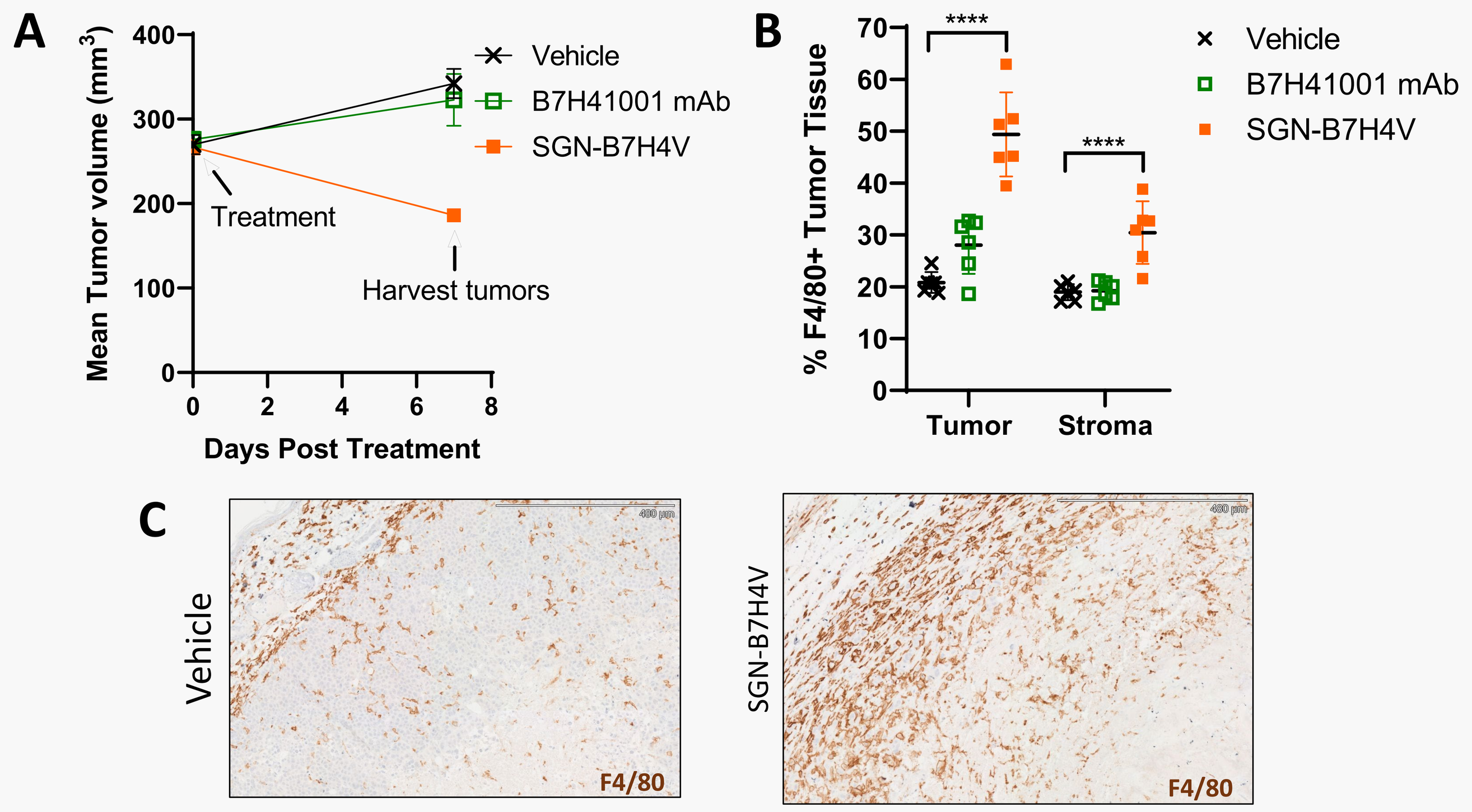

Figure 2. SGN-B7H4V recruits mouse macrophages to a triple-negative breast cancer (TNBC) xenograft tumor. MDA-MB-468 tumor-bearing NSG mice were treated with a single 3 mg/kg dose of SGN-B7H4V, unconjugated B7H41001 mAb, or vehicle control. Tumors were harvested 7 days post-treatment and processed for either RNAseq or immunohistochemistry (IHC) **(A)**. IHC staining revealed an increase in F4/80+ macrophages at the tumor site following treatment with SGN-B7H4V **(B, C)**.

SGN-B7H4V induces upregulation of cytokine and type I interferon response genes by human tumor cells

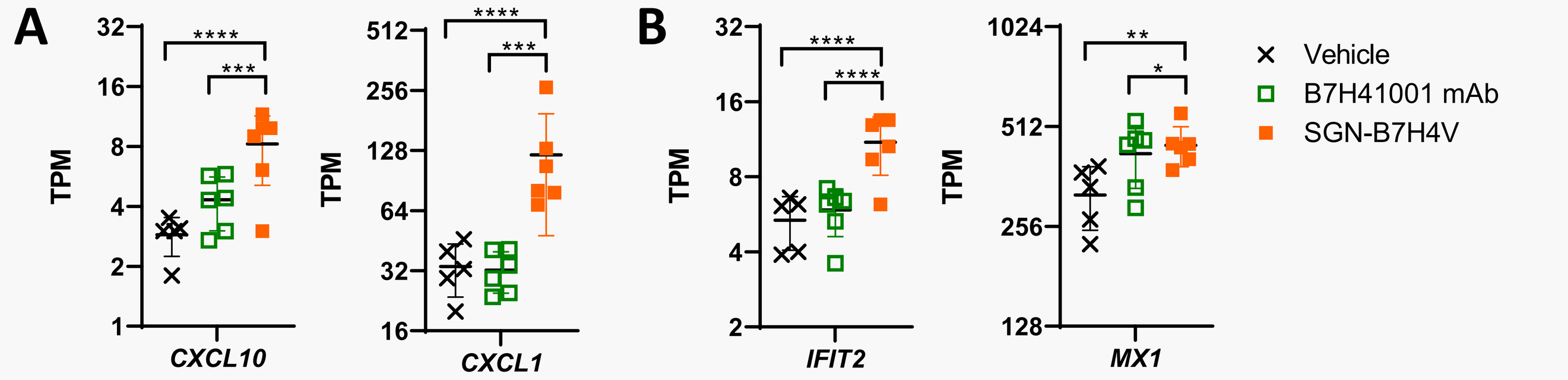

Figure 3. SGN-B7H4V induces upregulation of cytokines and type I interferon response genes by human tumors cells. RNAseq analysis of MDA-MB-468 tumors treated as in **Figure 2** revealed an increase in human transcripts encoding cytokines **(A)** and type I IFN response genes **(B)** in tumor cells following treatment with SGN-B7H4V compared to the vehicle control.

SGN-B7H4V Drives Antitumor Activity in an Immunocompetent Murine Tumor Model

SGN-B7H4V elicits tumor regression in a syngeneic murine B7-H4-expressing Renca tumor model

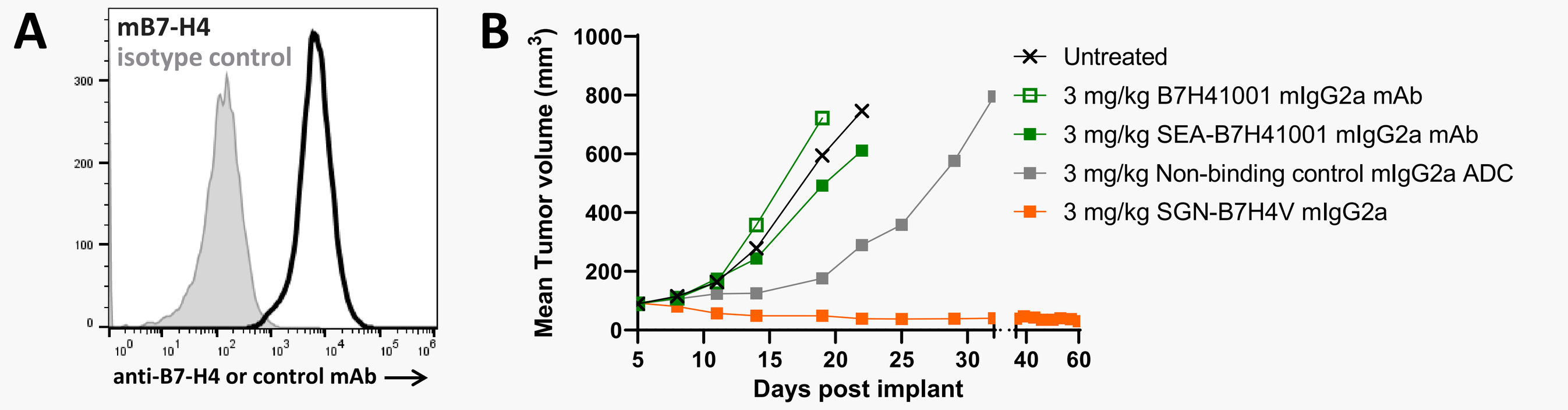

Figure 4. SGN-B7H4V drives robust antitumor responses in an immunocompetent tumor model. Renca tumor cells were engineered to express murine B7-H4 **(A)**. Treatment of B7-H4-Renca tumor-bearing mice with SGN-B7H4V mIgG2a* elicited durable tumor regression in 5/5 mice, while the non-binding control ADC, unconjugated mAb B7H41001, or the afucosylated mAb SEA-B7H41001 elicited minimal activity **(B)**.
ADCs and mAbs with a mIgG2a Fc backbone were used in all syngeneic models to avoid anti-drug antibody responses that can occur upon repeat treatment of hIgG1 antibodies in immunocompetent mice.

SGN-B7H4V recruits multiple immune cell types to murine B7-H4-expressing Renca tumors

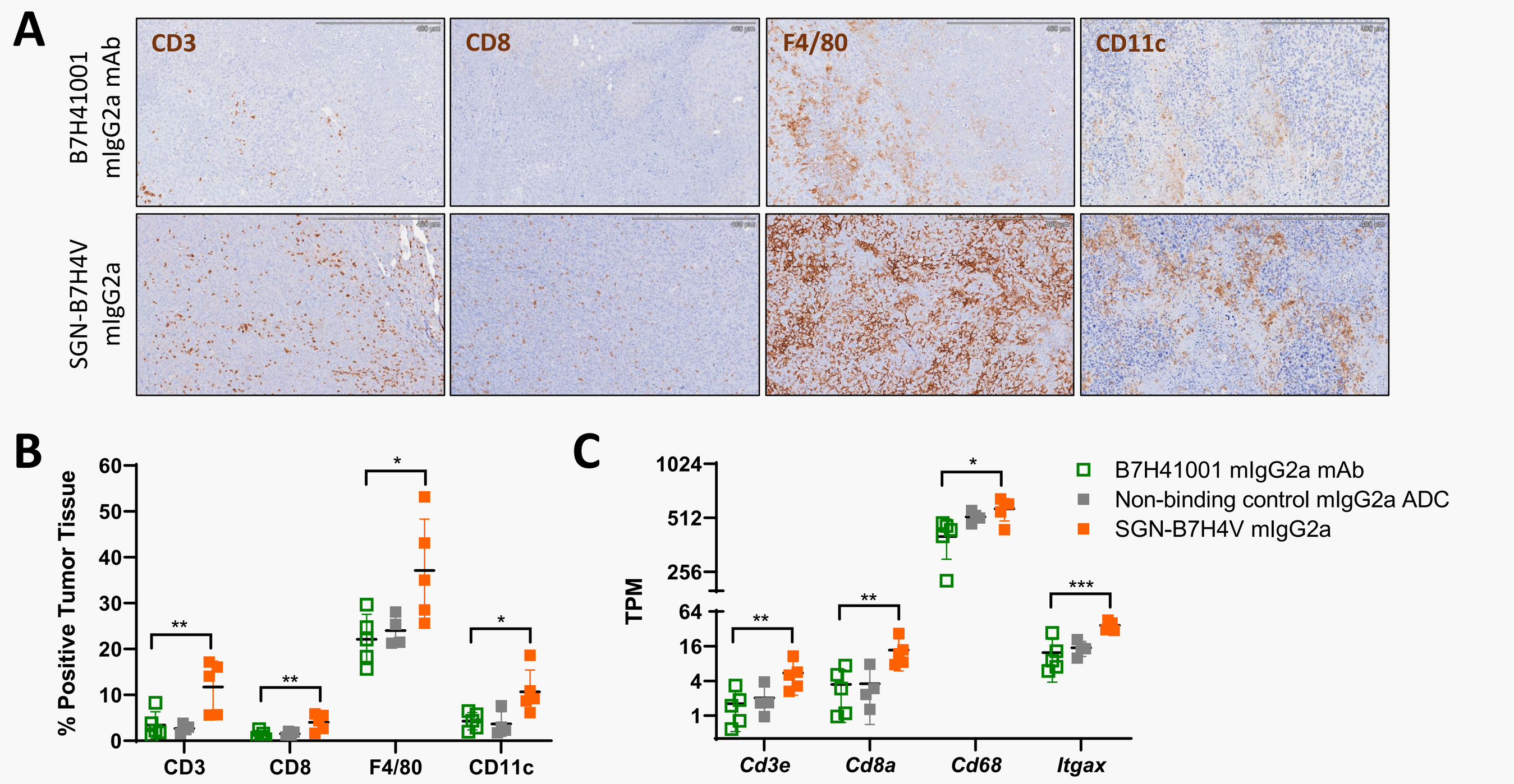

Figure 5. SGN-B7H4V elicits recruitment of T cells, macrophages, and dendritic cells to murine B7-H4-expressing Renca tumors. B7-H4-Renca tumor-bearing mice were treated with a single 3 mg/kg dose of naked B7H41001 mAb, SGN-B7H4V, or non-binding ADC. Tumors were harvested 7 days after treatment and processed for RNAseq or IHC. SGN-B7H4V treatment led to an increase in CD3/CD8+ T cells, F4/80+ macrophages, and CD11c+ antigen-presenting cells at the tumor site **(A, B)**. A significant increase in *Cd3e* (encodes CD3e), *Cd8a* (encodes CD8a), *Cd68* (encodes the macrophage marker CD68) and *Itgax* (encodes CD11c) transcripts was also observed following treatment with SGN-B7H4V **(C)**.

SGN-B7H4V Pairs Well With an Anti-PD-1 mAb

Combination of SGN-B7H4V with an anti-PD-1 mAb demonstrates enhanced antitumor activity

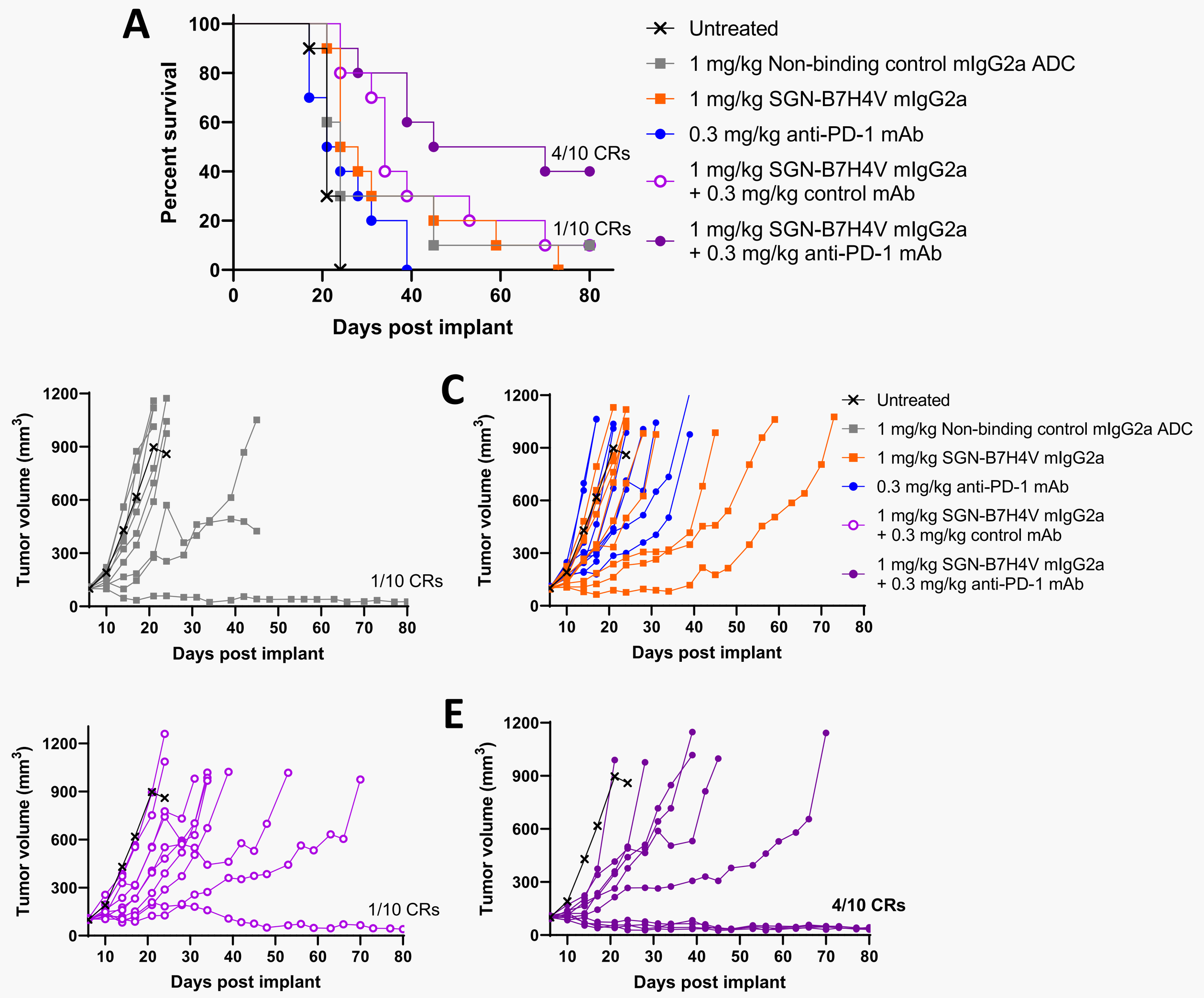

Figure 6. SGN-B7H4V in combination with an anti-PD-1 mAb elicits enhanced antitumor activity. Treatment of B7-H4-Renca tumor-bearing mice with a sub-therapeutic dose of SGN-B7H4V in combination with an anti-PD-1 mAb led to improved survival (**A**, with survival defined by tumor outgrowth to ~1000mm³) and enhanced antitumor activity **(E)** compared to the non-binding control ADC **(B)**, either treatment alone **(C)**, or SGN-B7H4V in combination with a rat isotype control mAb **(D)**.

SGN-B7H4V in combination with an anti-PD-1 mAb drives robust immune memory

Mice with complete responses (CR) being rechallenged	Tumor cells used for rechallenge	% Protection from tumor rechallenge
3 mg/kg SGN-B7H4V mIgG2a (Figure 4B)	Parental Renca	40% (2/5 mice)
1 mg/kg SGN-B7H4V mIgG2a + 0.3 mg/kg anti-PD-1 mAb (Figure 6)	Parental Renca	100% (4/4 mice)

Figure 7. SGN-B7H4V in combination with an anti-PD-1 mAb elicits robust immune memory. All four mice from **Figure 6** that achieved a CR after treatment with SGN-B7H4V in combination with an anti-PD-1 mAb were protected from rechallenge with parental Renca tumor cells compared to 40% of mice from **Figure 4** that achieved a CR after treatment with SGN-B7H4V alone.

Conclusions

- SGN-B7H4V induces hallmarks of immunogenic cell death in vitro, driven by the MMAE (vedotin) payload.
- Moreover, SGN-B7H4V led to immune changes in the tumor microenvironment in vivo, including recruitment of macrophages and T cells to tumors, suggesting the potential to drive both innate and adaptive antitumor immunity.
- Finally, SGN-B7H4V drove robust antitumor activity in an immunocompetent tumor model as a monotherapy and shows combination activity with an anti-PD1 agent.
- Altogether, these data support the evaluation of SGN-B7H4V as a monotherapy in the ongoing Phase 1 Study of SGN-B7H4V in Advanced Solid Tumors (NCT05194072) and potential future clinical combinations with immunotherapies.

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