Localized Regulated Expression of IL-12 as a Gene Therapy Concomitant with Blockade of PD-1 for Treatment of Glioma

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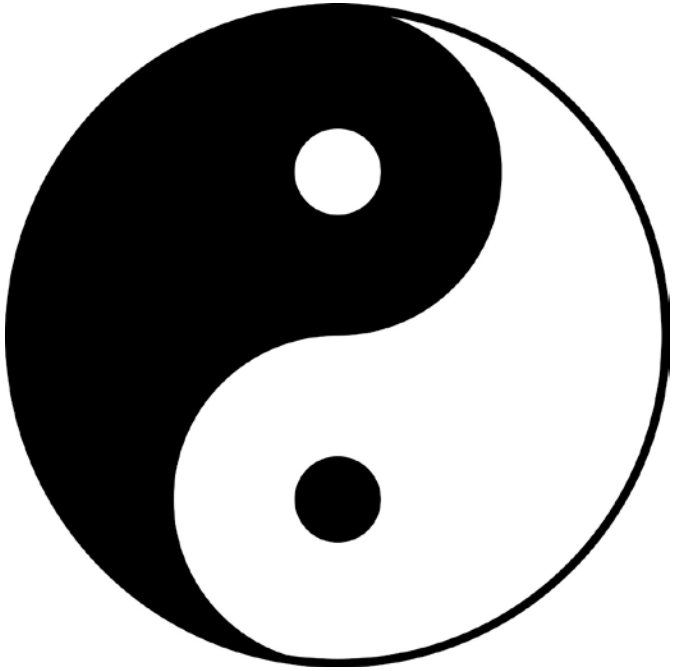
Immuno-oncology

Immune Activation



Immune Suppression

CTLA4: Brakes

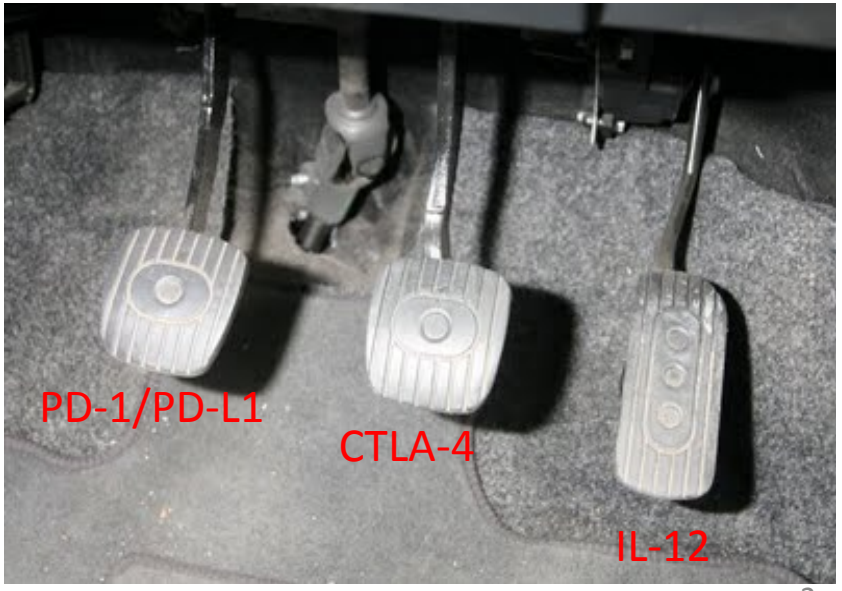
- Proportionally dampens cytotoxic T cell activation

PD-1/PD-L1: Clutch

- Regulates cytotoxic T cell activity to decrease tissue damage

Local IL-12: Gas/GPS

- Activates cytotoxic T cell
- Decreases T regs, T cell homing



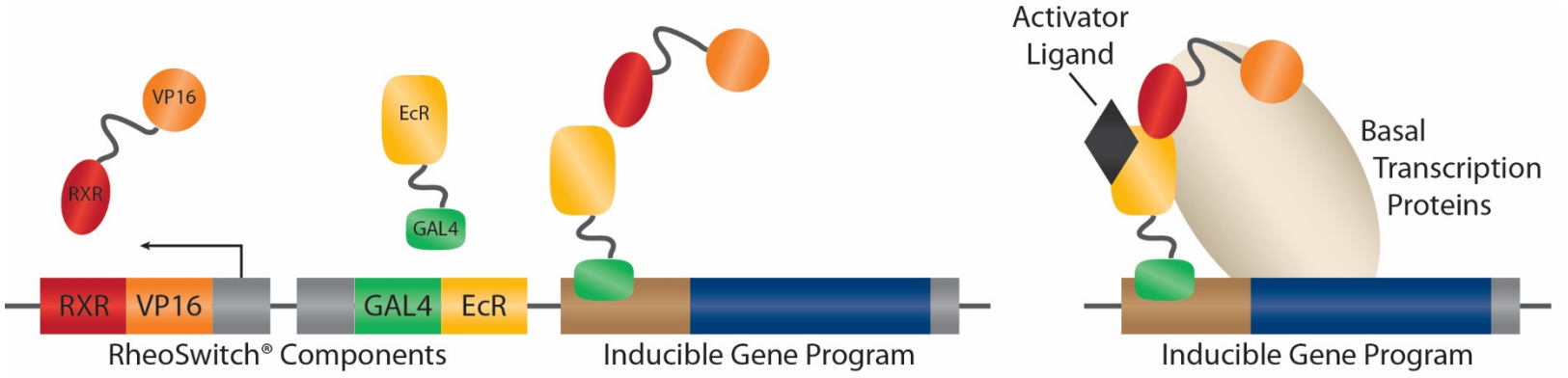
Rationale for tumor IL-12 production in combination with anti-PD-1 therapy

- **Interleukin-12 (IL-12)**
  - Master regulator of cell-mediated immunity to pathogens and malignant cells
  - Produced by innate immune cells in response to pathogens
- **PD-1**
  - Negative regulator of lymphocyte activation
  - PD-1 is expressed on activated CD4 & CD8 T cells after MHC-TCR engraftment modulates & T cell activation.
  - Inhibition of PD-1 reverses the immunosuppression

- Tumor immunostimulation via IL-12 coupled with the reduction in innate tumor immunosuppression by anti-PD-1 should result in enhanced efficacy over monotherapy.

Inducible Gene Regulation: RheoSwitch Therapeutic System®

RheoSwitch Therapeutic System® (RTS®) is a 3-component transcriptional regulator

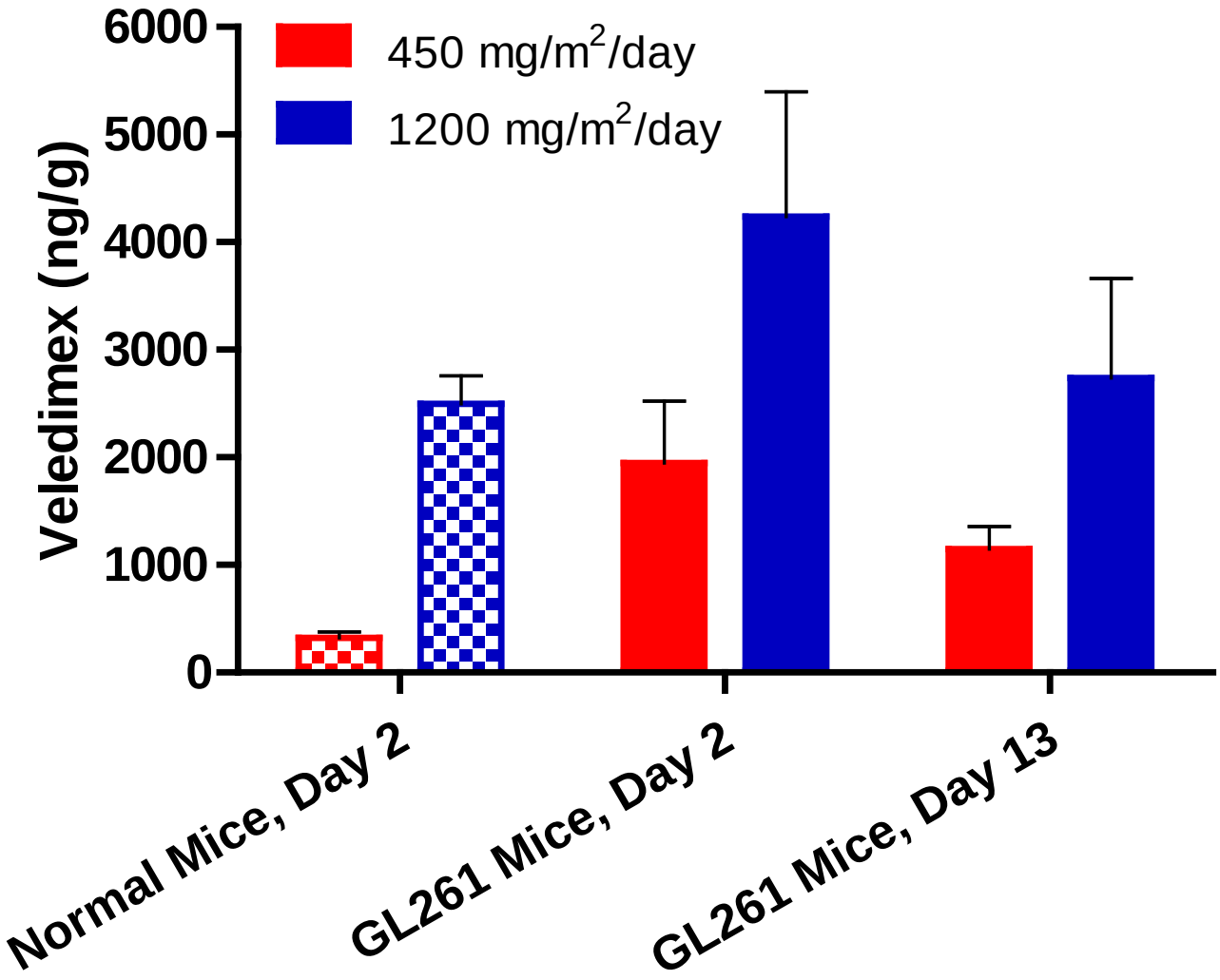


1. **The Switch Components**: The RTS® gene program includes 2 receptor protein fusions: VP16-RXR (co-activation partner, CAP) and Gal4-EcR (ligand-inducible transcription factor, LTF). In the absence of ligand, the LTF binds to the inducible promoter and does not form a stable interaction with the CAP.

2. **The Inducible Promoter**: A customizable promoter to which basal transcription proteins are recruited and the target gene is transcribed.

3. **The Activator Ligand (veledimex):** An ecdysone analog, diacylhydrazine-based small molecule functions as an activator. In the presence of the ligand, a conformational change in the LTF leads to a stable, high affinity interaction with the CAP.

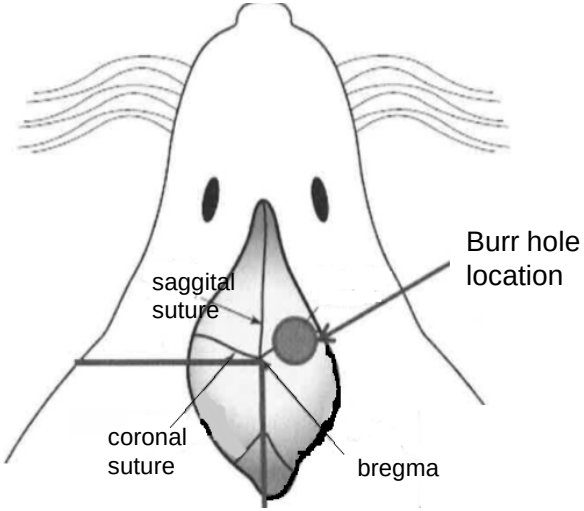
Veledimex Crosses the Blood Brain Barrier in GL261 Orthotopic Glioma and Normal Mice



Veledimex levels at 24 hr posttreatment

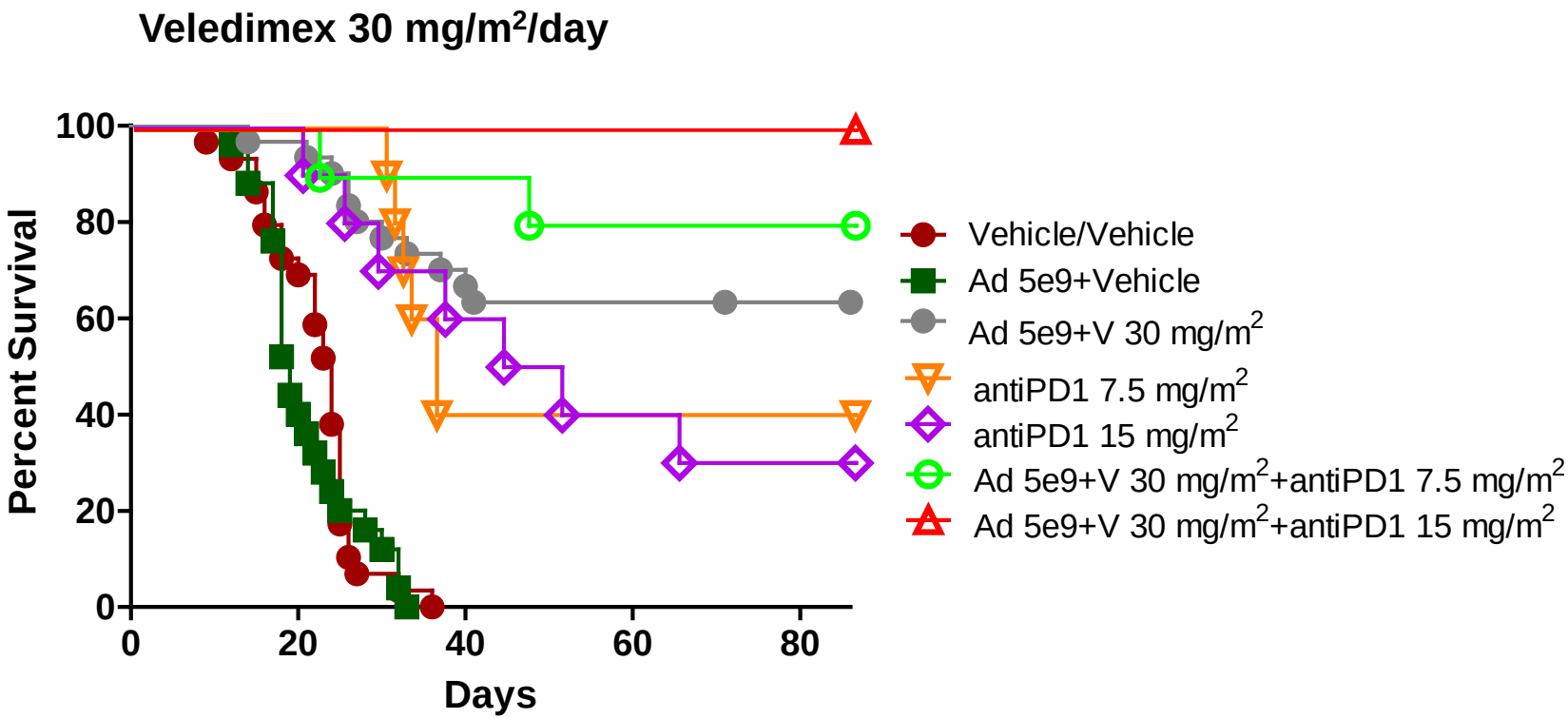
Ad-RTS-mIL12 + Veledimex in Combination with anti PD-1 in GL-261 Orthotopic Model

GL-261 Orthotopic Glioma Model



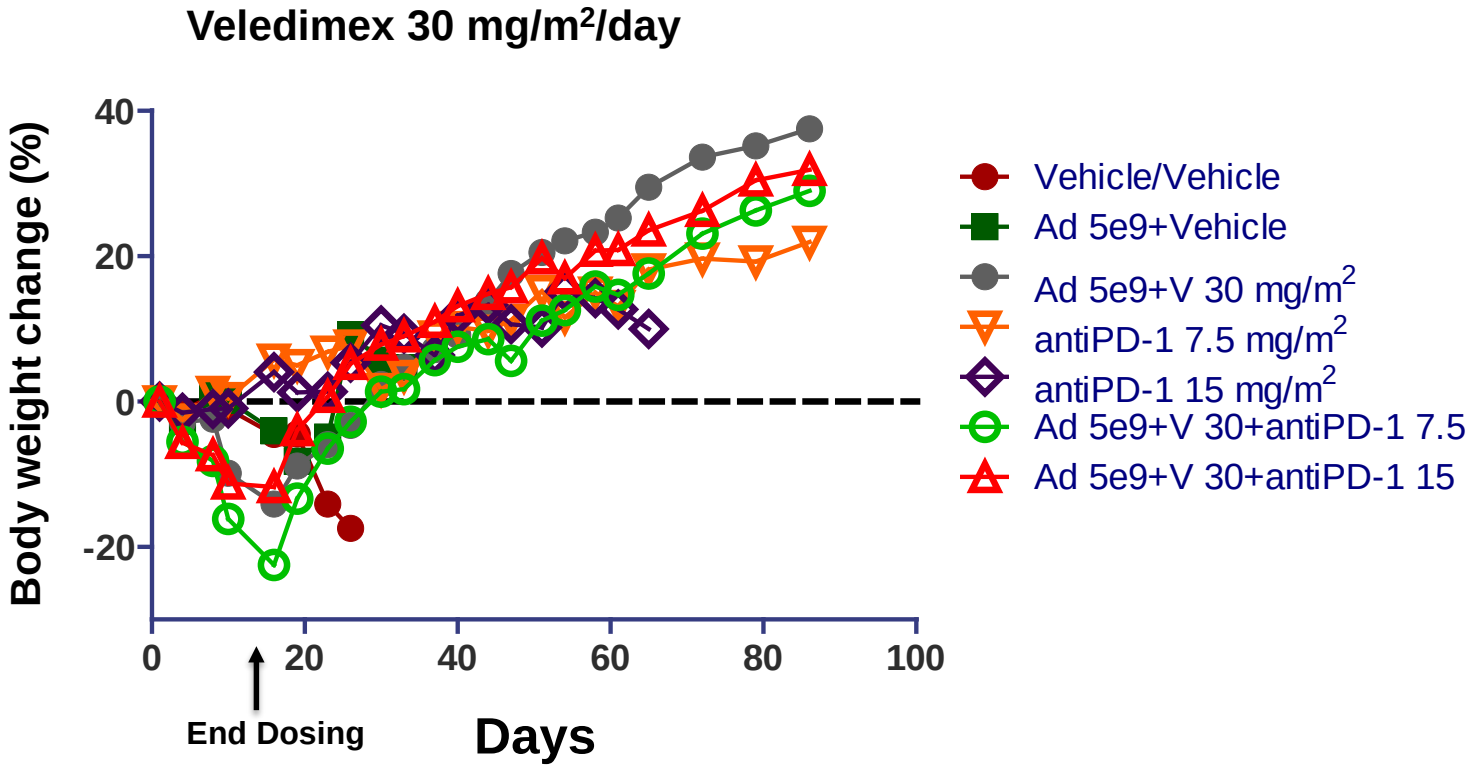
iCPI	Antibody	Route	Schedule	Dose (mg/m2)	
antiPD-1	RMP 1-14	i.p.	Q4Dx5	7.5	15

Ad-RTS-mIL-12 + Veledimex + Anti-PD-1: Overall Survival



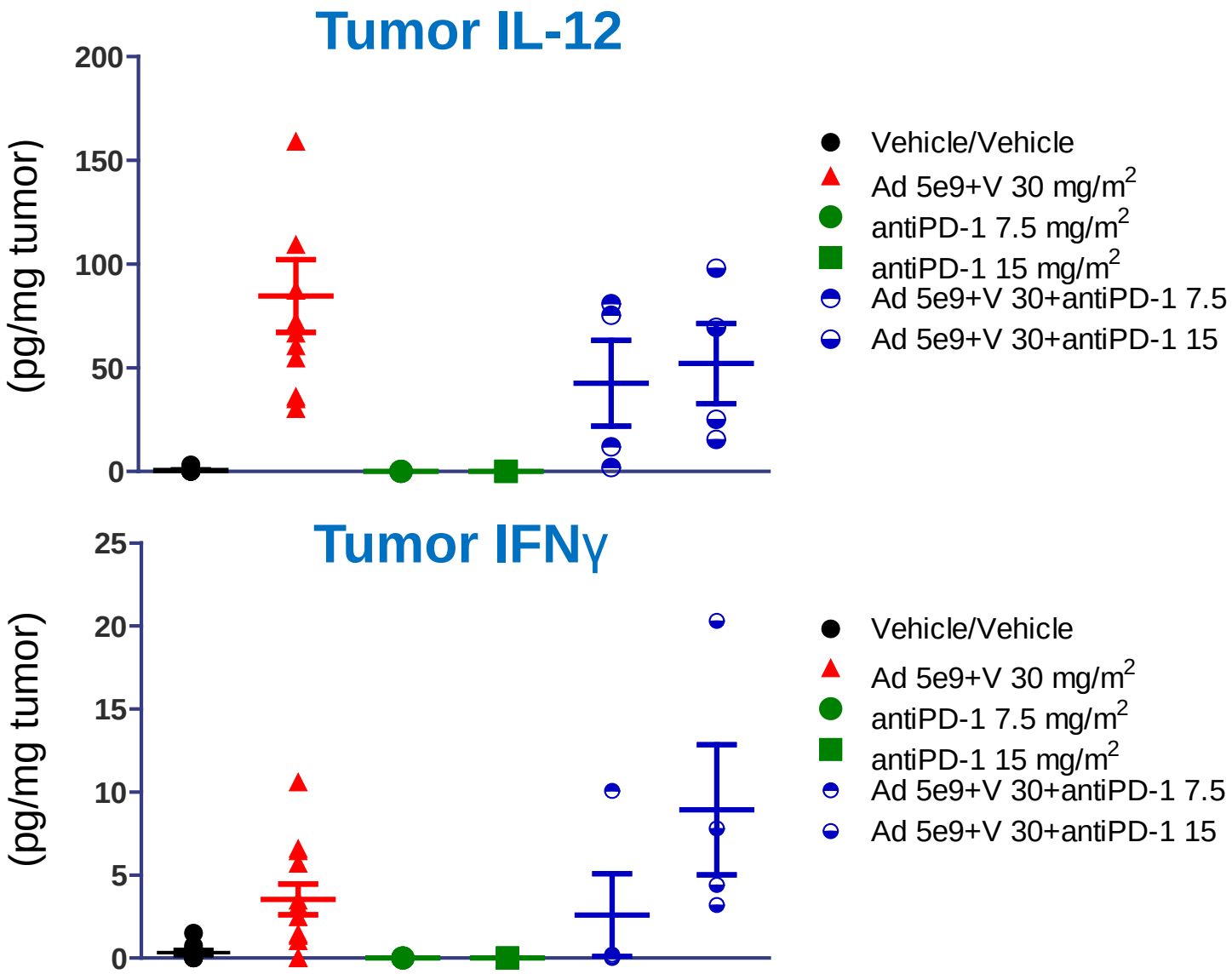
- Ad-RTS-mIL-12 + veledimex dose-related increase in survival
- Ad-RTS-mIL-12 + veledimex + anti PD1 increased survival over monotherapy
- Ad-RTS-mIL-12 + veledimex 30mg/m^2/day + anti PD1 15mg/m^2 results in 100% survival

Ad-RTS-mIL-12 + Veledimex + Anti-PD-1: Body Weight Change

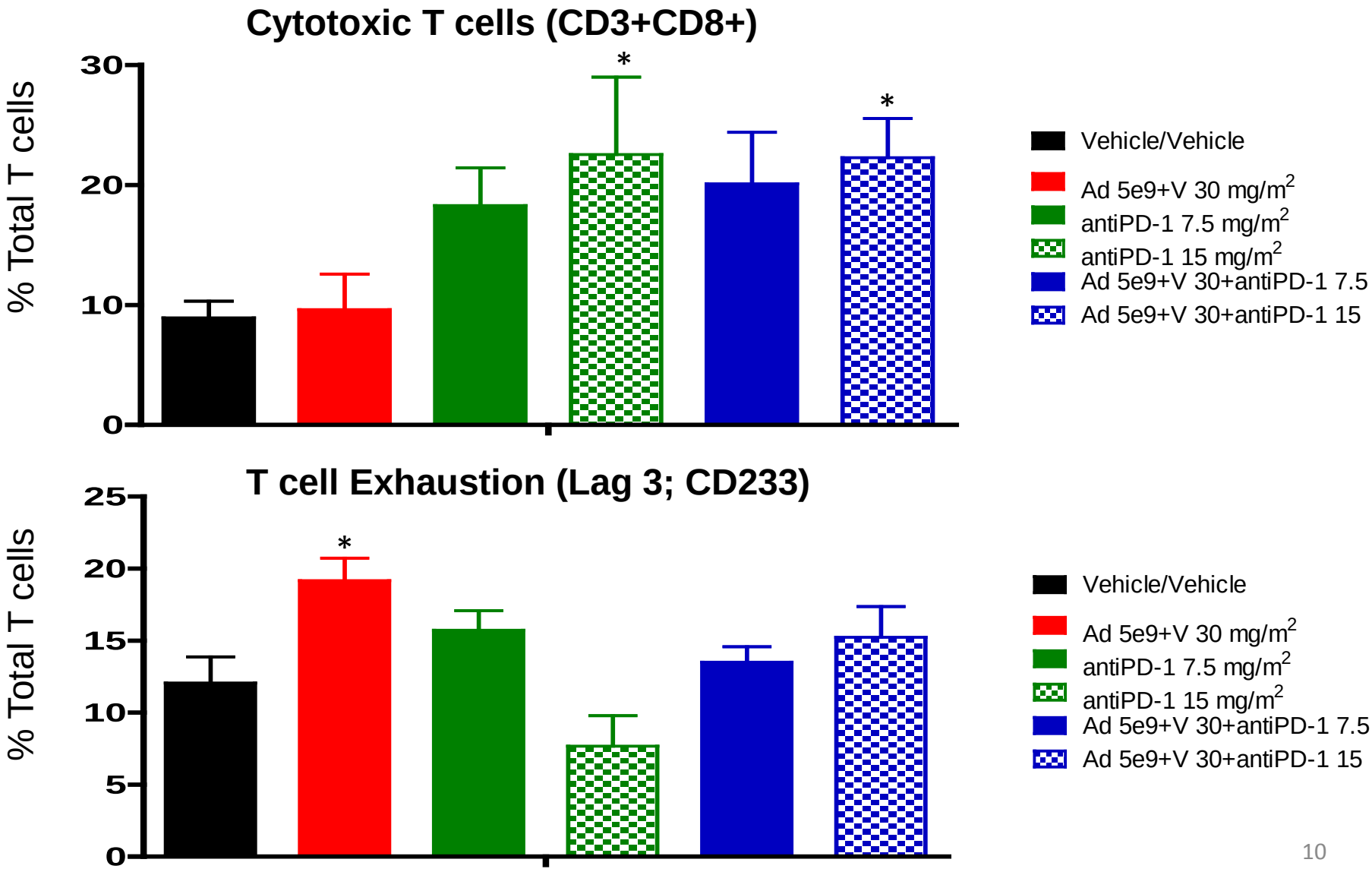


- Ad-RTS-mIL-12 + veledimex + anti-PD-1 therapy augmented the reduction in body weight over monotherapy.
- All groups recovered when veledimex was discontinued.

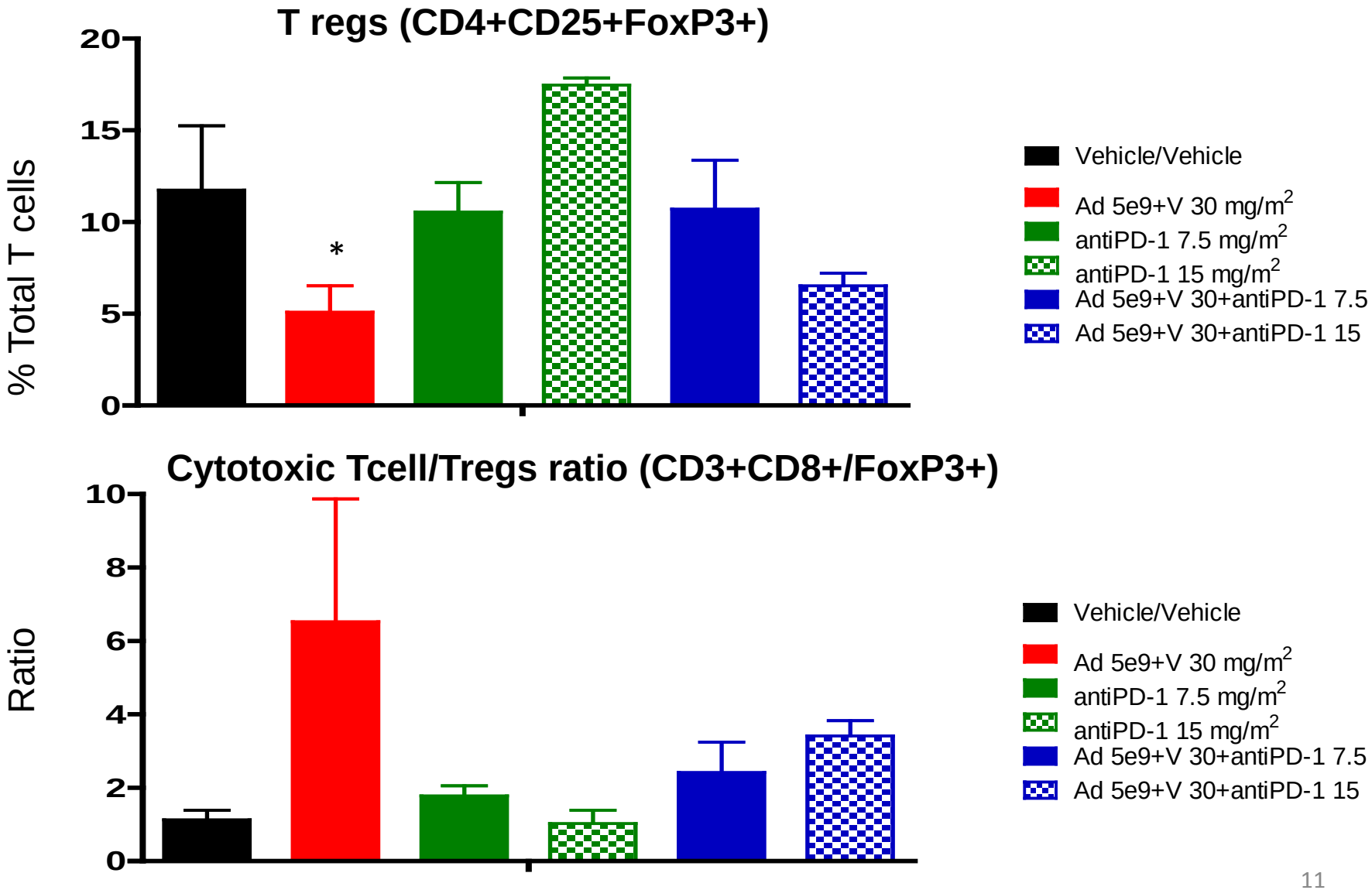
Ad-RTS-mIL-12 + Veledimex in Combination with Anti-PD-1 (RPM 1-14) on Tumor Cytokines



Ad-RTS-mIL-12 + Veledimex + Anti-PD-1 (RPM 1-14) Tumor FACS



Ad-RTS-mIL-12 + Veledimex + Anti-PD-1 (RPM 1-14) Tumor FACS



Conclusions

- Controlled tumor IL-12 production was able to stimulate the immune system in the presence of innate tumor immunosuppression
- The addition of anti-PD-1 therapy resulted in a reduction in tumor innate immunosuppression
- The combination of both therapies resulted in decreased immunosuppression coupled with local immunostimulation proved to be beneficial in the treatment of glioma with a profound increase in survival over controls & monotherapy
- **Clinical study design to assess combination therapy in recurrent GBM in progress**