Controlled Local Expression of IL-12 as Gene Therapy Concomitant with Systemic Chemotherapy Improves Survival in Glioma (IMMU-33)

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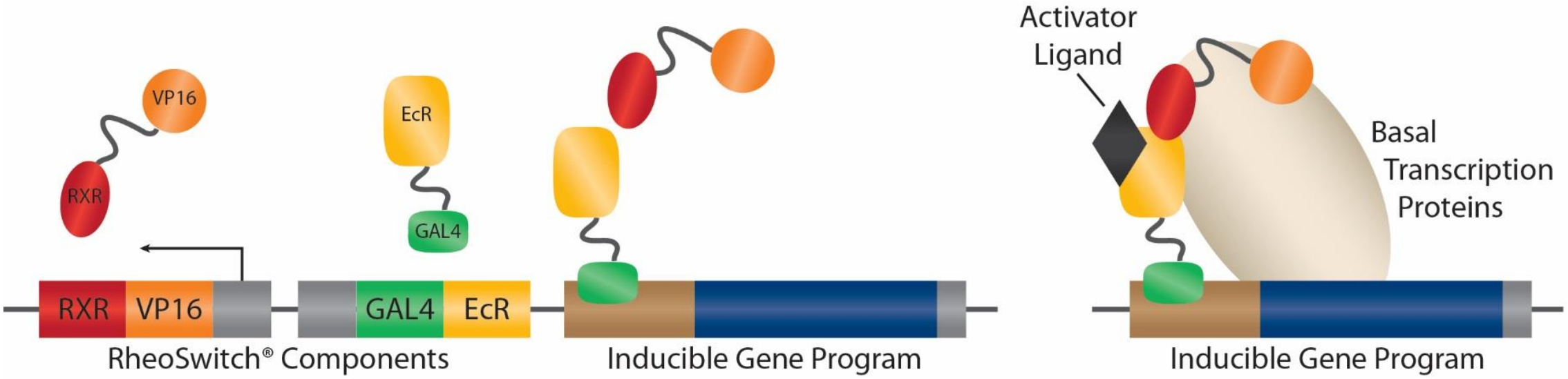
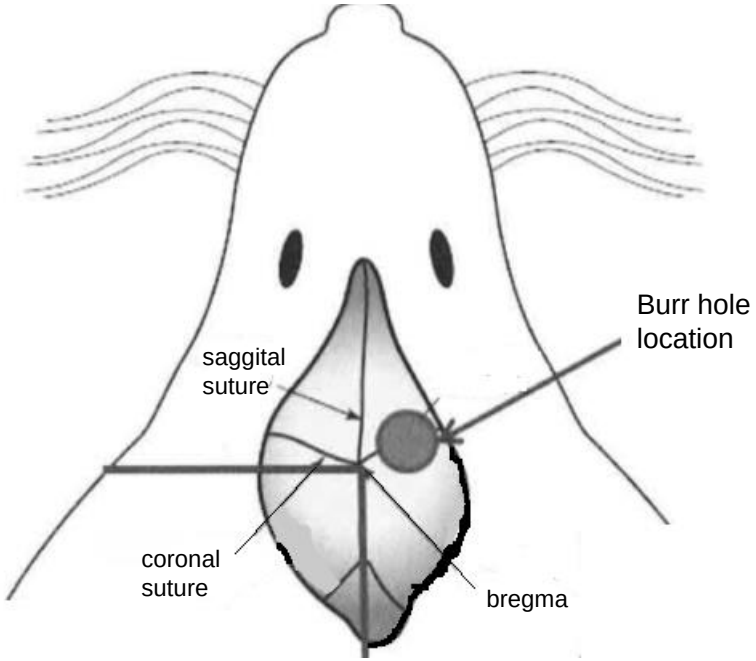
Test Articles Studied

Ad-RTS-IL-12 + Veledimex: controlled local expression of IL-12 via a RheoSwitch Therapeutic System® (RTS®) is a 3-component transcriptional regulator



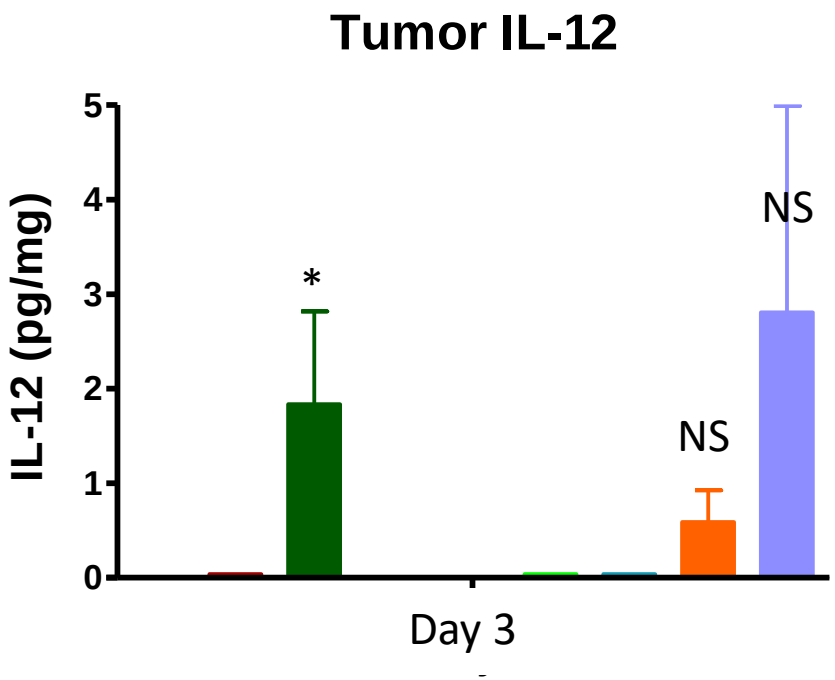
Lomustine: is a lipid soluble alkylating nitrosourea compound which crosses the BBB and used in the treatment of glioma. Lomustine causes interstrand and intrastrand cross-linking of DNA resulting in cell death.

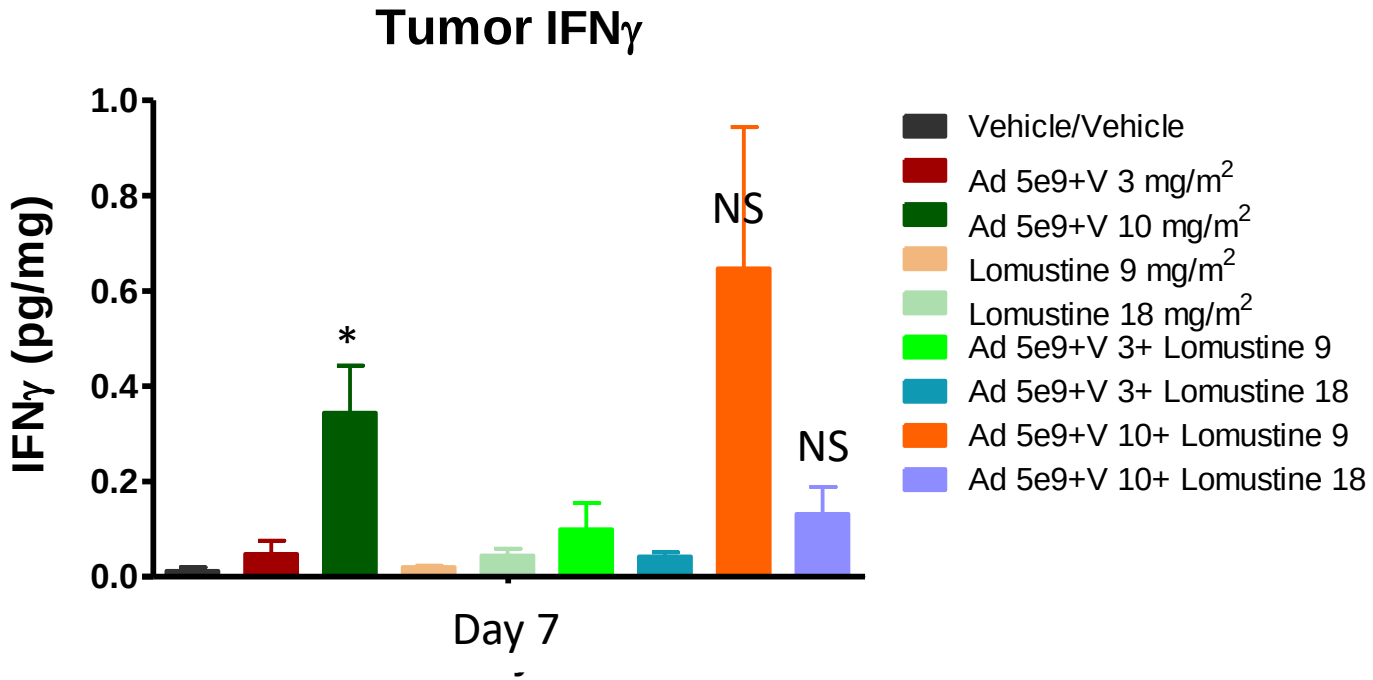
GL-261 Orthotopic Glioma Model



- Five Days prior to therapy 1 x 10^5 GL-261 glioma cells volume 3 µl were administered into the brain of C57BL/6 mice.
- On Day 1 a single dose of Ad-RTS-mIL-12 at 5x10^9vp 5 µl followed by the activator ligand, veledimex p.o. QDx14 CCNU i.p. QDx5.
- The time to disease progression and death was studied.

Expression of Tumor IL-12 & IFNγ with Ad-RTS-mIL-12 + Veledimex Alone or in Combination with CCNU

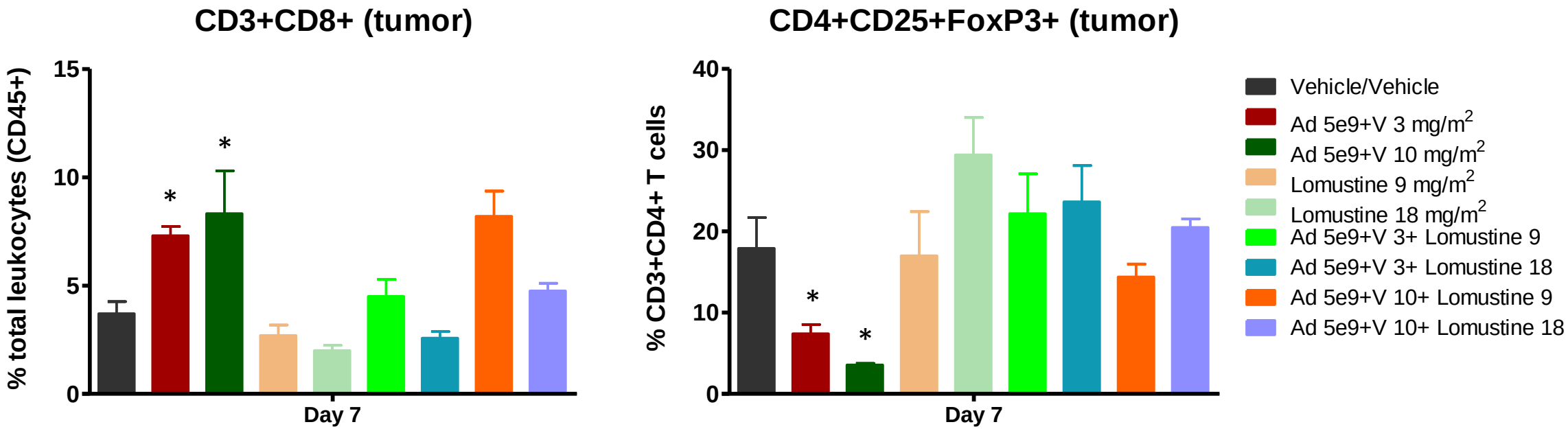




Serum levels of IL-12 and IFNγ ~ 30 times lower than tumor

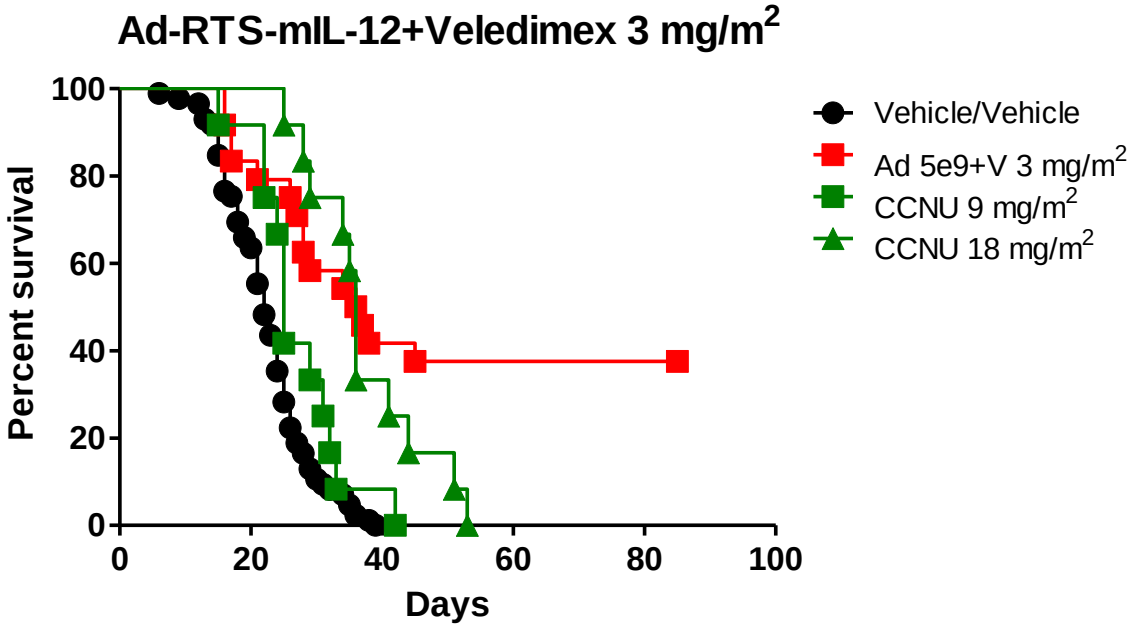
- * P<0.05 vehicle vs treatment
- NS= No significant differences between Ad-RTS-mIL-12 + veledimex vs. combination

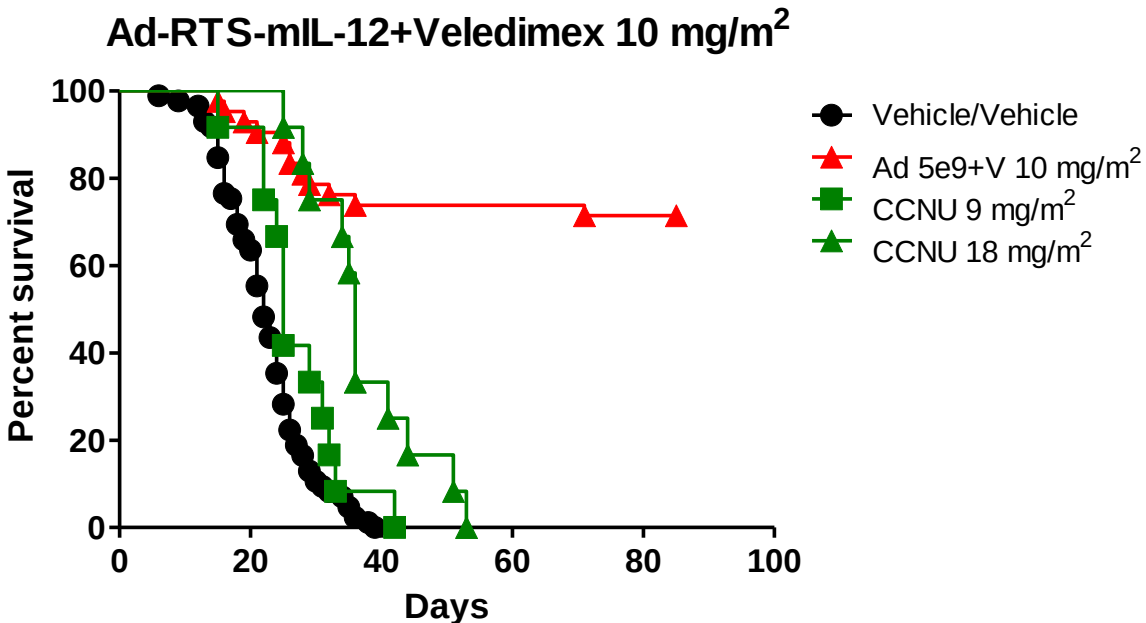
Ad-RTS-mIL-12 + veledimex increases tumor cytotoxic T cells while decreasing Tregs via FACS



* P< 0.05 one-way analysis of variance; Dunnets test

Survival: Ad-RTS-mIL-12 + Veledimex or CCNU Alone & In Combination



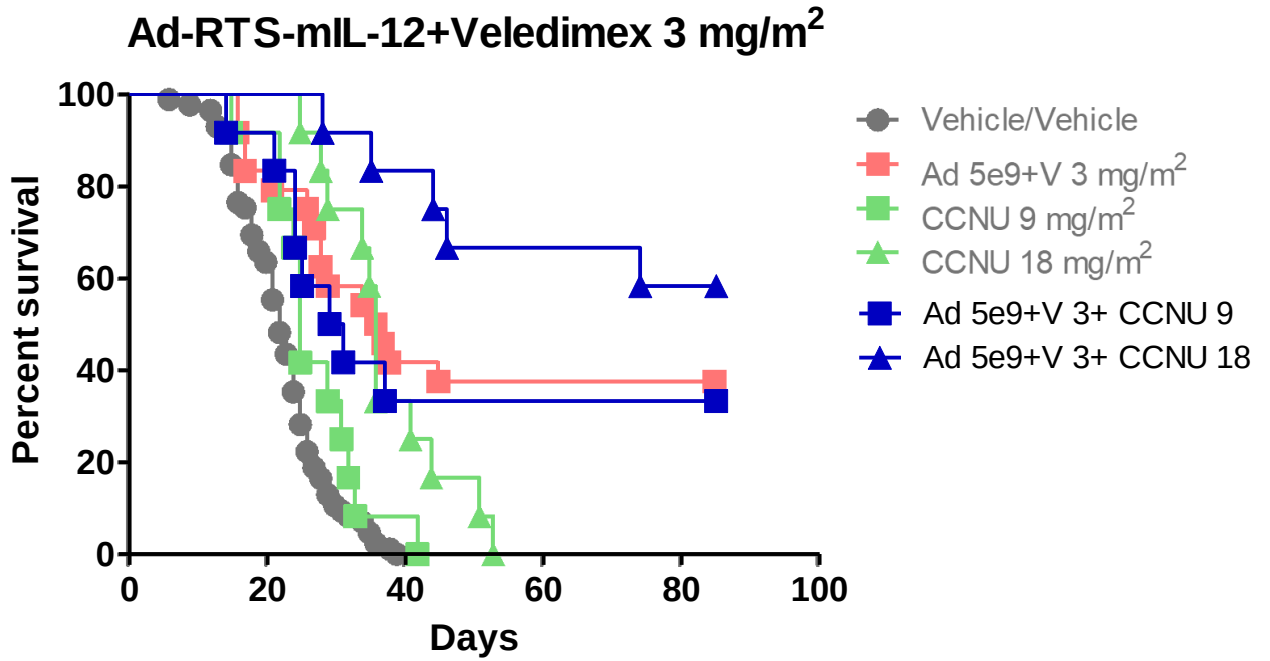


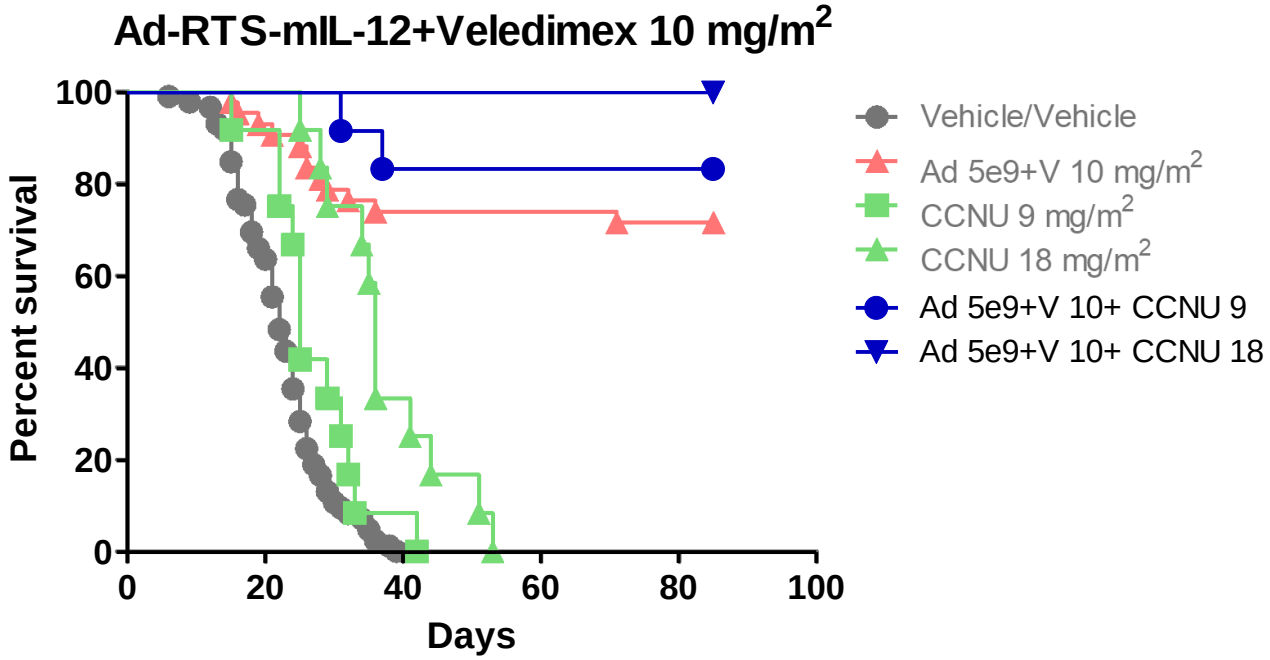
Treatment (mg/m^2)	Median Survival (Days)	Increase in Life Span (ILS) (%)[a]	Percent Survival at Day 85
Vehicle/Vehicle	22		0
Ad 5e9+V 3 mg/m^2	37	66	38
Ad 5e9+V 10 mg/m^2	>85[b]	>286[b]	71
Lomustine 9 mg/m^2	25	14	0
Lomustine 18 mg/m^2	36	64	0

[a] ILS=%T/C -100%;T/C = quotient of median survival of treated vs. control

[b] Animals survived to the end of the study (Day 85), TTE >85 days& ILS >286%

Survival: Ad-RTS-mIL-12+Veledimex & CCNU Alone & In Combination





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Lomustine 18 mg/m^2	36	64	0
Ad 5e9+V 3+ Lomustine 9	30	36	33
Ad 5e9+V 3+ Lomustine 18	>85[b]	>286[b]	58
Ad 5e9+V 10+ Lomustine 9	>85[b]	>286[b]	83
Ad 5e9+V 10+ Lomustine 18	>85[b]	>286[b]	100

[a] ILS=%T/C -100%;T/C = quotient of median survival of treated vs. control

[b] Animals survived to the end of the study (Day 85), TTE >85 days& ILS >286%

Summary

- **Cytokines:**
  - Ad-RTS-mIL-12 + veledimex increased tumor cytokines in a dose-related manner
  - CCNU (lomustine) alone does not affect tumor cytokine levels at the doses studied
  - Ad-RTS-mIL-12 + veledimex + CCNU does not enhance tumor cytokines when compared to Ad-RTS-mIL-12 + veledimex alone
- **Tumor FACS**:
  - Ad-RTS-mIL-12 + veledimex increased tumor cytotoxic T cells with concomitant decrease in tumor Tregs
  - CCNU alone had no effect on tumor cytotoxic T cells or tumor Tregs
  - Ad-RTS-mIL-12 + veledimex + CCNU does not further increase cytotoxic T cells when compared to Ad-RTS-mIL-12 + veledimex alone
- **Survival:**
  - Ad-RTS-mIL-12 + veledimex demonstrated dose-related increase in survival vs. vehicle
  - CCNU 18 mg/m^2 demonstrated minimal increase in survival vs. vehicle
  - Ad-RTS-mIL-12 + veledimex + CCNU resulted in an increase in survival over Ad-RTS-mIL-12 + veledimex monotherapy. 100% survival with Ad + V 10 mg/m^2 + CCNU 18 mg/m^2
- **Controlled local immunostimulation with IL-12 combined with CCNU, warrants further nonclinical investigation**