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Supporting Information

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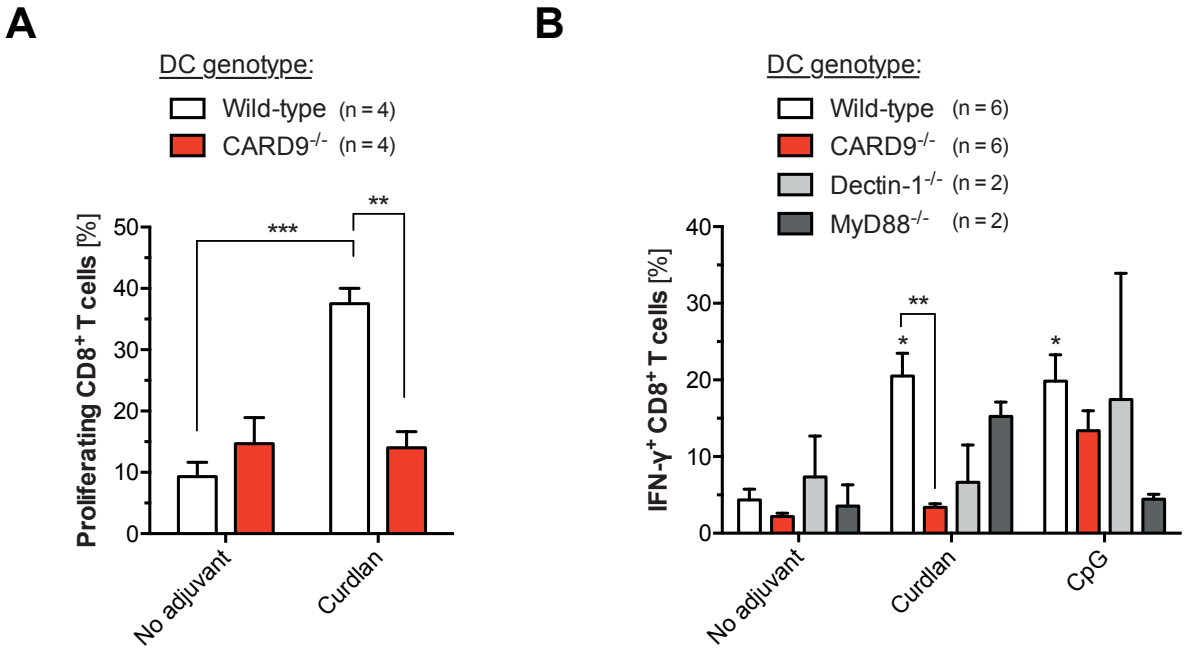
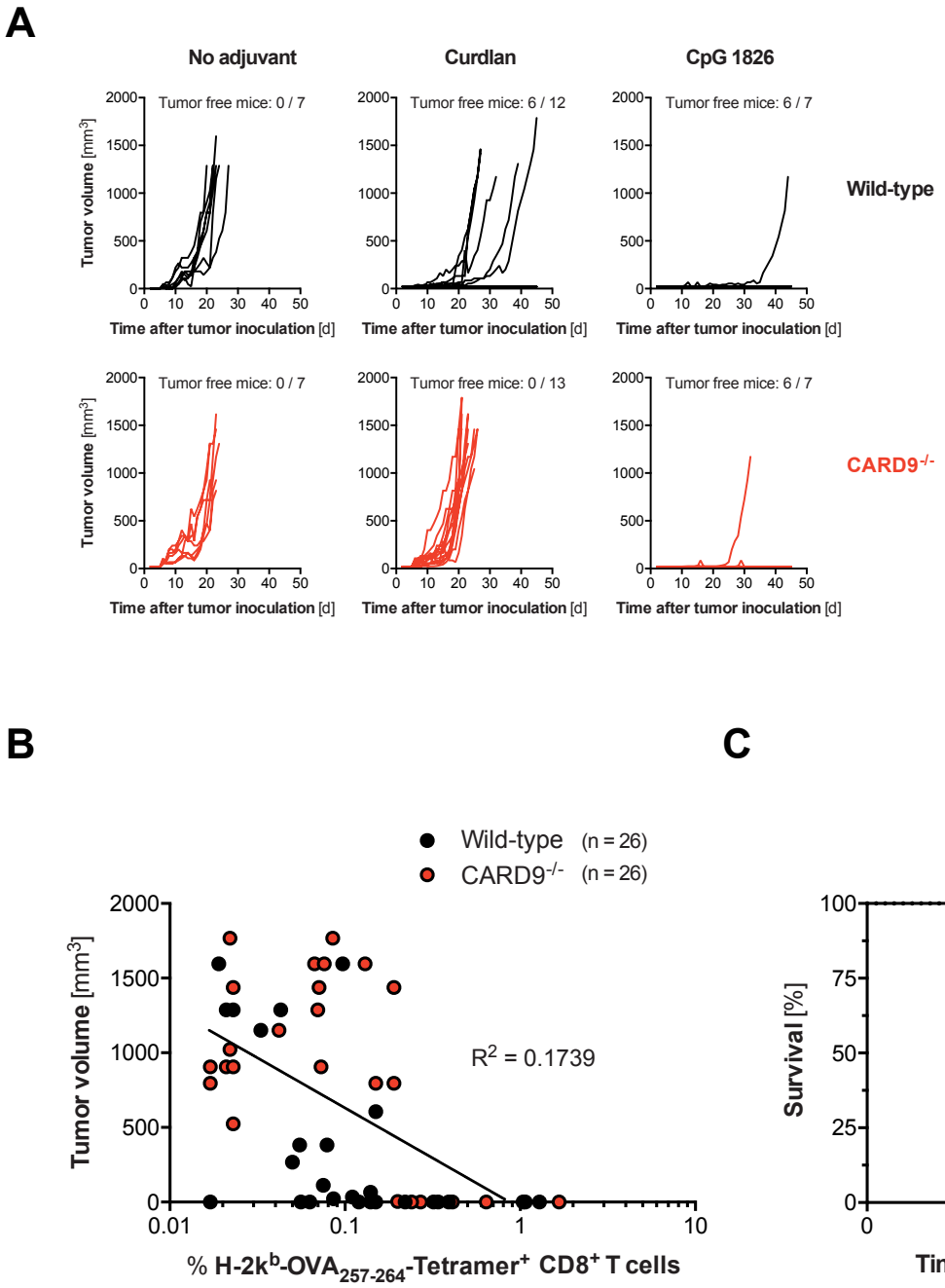
Card9 controls Dectin-1-induced T-cell cytotoxicity and tumor growth in mice



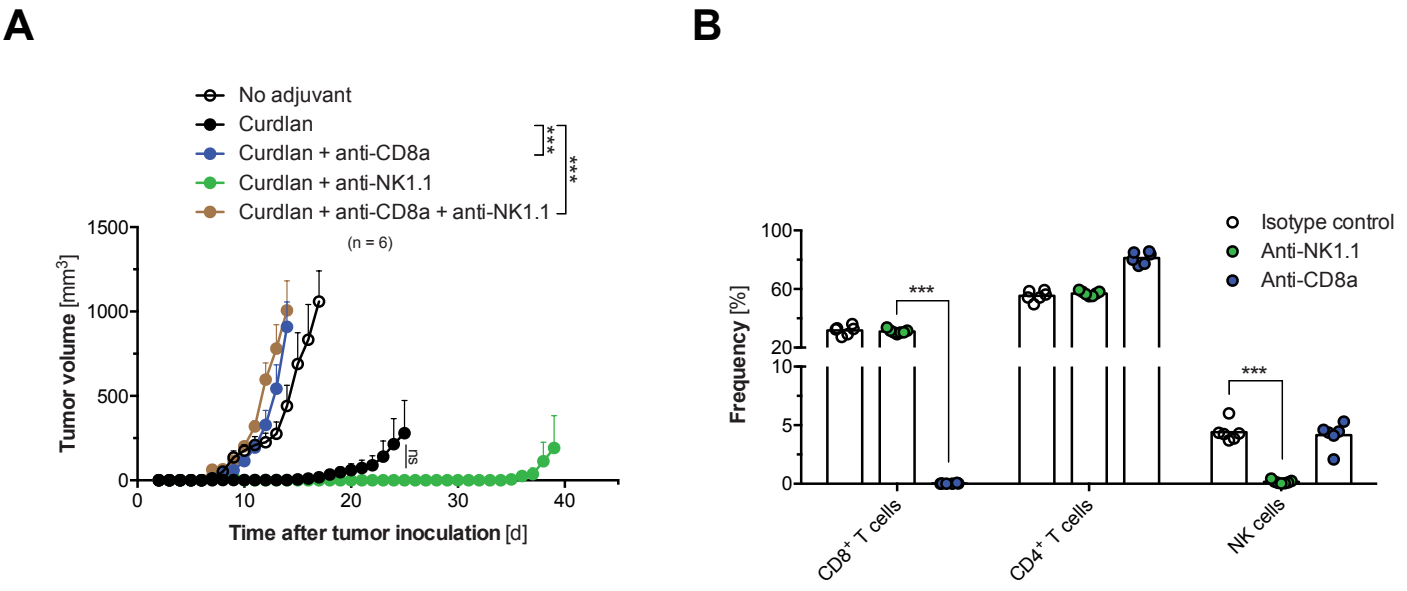
Supporting Information Figure 1: BM-DCs from wild-type, Card9- (Card9$^{-/-}$), Dectin-1- (Dectin-1$^{-/-}$) and MyD88-deficient (MyD88$^{-/-}$) donor mice were stimulated with either Curdlan or CpG and were then co-cultured with magnetically purified, CFSE-labeled CD8^{+} OT-I T cells in the presence of OVA protein, as described for Figure 1B. **(A)** CD8^{+} T cell proliferation as per CFSE dye dilution and **(B)** intracellular IFN-γ expression were analyzed by flow cytometry. All data give mean value ± S.E.M. and were pooled from six independent experiments. Statistical significance was calculated using one-way ANOVA with Bonferroni post-test (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$). An asterisk without brackets indicates comparison to the wild-type 'No adjuvant' condition.

Supporting Information Figure 1



Supporting Information Figure 2: Wild-type and Card9-deficient (Card9$^{-/-}$) mice were vaccinated with OVA in combination with either Curdlan or CpG 1826. B16.OVA melanomas were subsequently induced as described for Figure 3A. **(A)** Data show the tumor growth of individual mice. **(B)** The Pearson correlation coefficient of tumor volume and the frequency of circulating tumor-specific CD8$^+$ T cells was calculated at day 20 post tumor induction. Each circle represents an individual mouse. **(C)** All animals independent of their genotype were grouped into low or high frequency of circulating tumor-specific CD8$^+$ T cells and group survival was analyzed using the Log-rank test. All data are pooled from two independent experiments.

Supporting Information Figure 2



Supporting Information Figure 3: Wild-type mice were vaccinated with OVA in combination with Curdlan. B16.OVA melanomas were subsequently induced as described for Figure 3A. Some mice were additionally injected with anti-CD8a- and/or anti-NK1.1-depleting antibodies, beginning two days prior to Curdlan-based vaccination as described for Figure 3D. **(A)** Data show mean tumor growth +/- S.E.M. of n = 6 individual mice per group. The mean tumor growth curve was discontinued when the first animal per group succumbed to tumor progression. Statistical significance was calculated using one-way ANOVA with Bonferroni post-test (***, $p < 0.001$; ns, not significant) based on the tumor volume at day 13, if not stated otherwise. **(B)** Frequency of indicated cell populations in peripheral blood of animals treated with depleting antibodies described in panel (A) as determined by flow cytometry. The analysis was performed on the day of tumor induction. Bars show mean cell population frequencies. Each circle represents an individual mouse. Statistical significance was calculated using one-way ANOVA with Bonferroni post-test (***, $p < 0.001$). Data is representative of two identical experiments.

Supporting Information Figure 3