# Immunomodulatory effects of soluble CD5 on experimental tumor models

## SUPPLEMENTARY MATERIALS
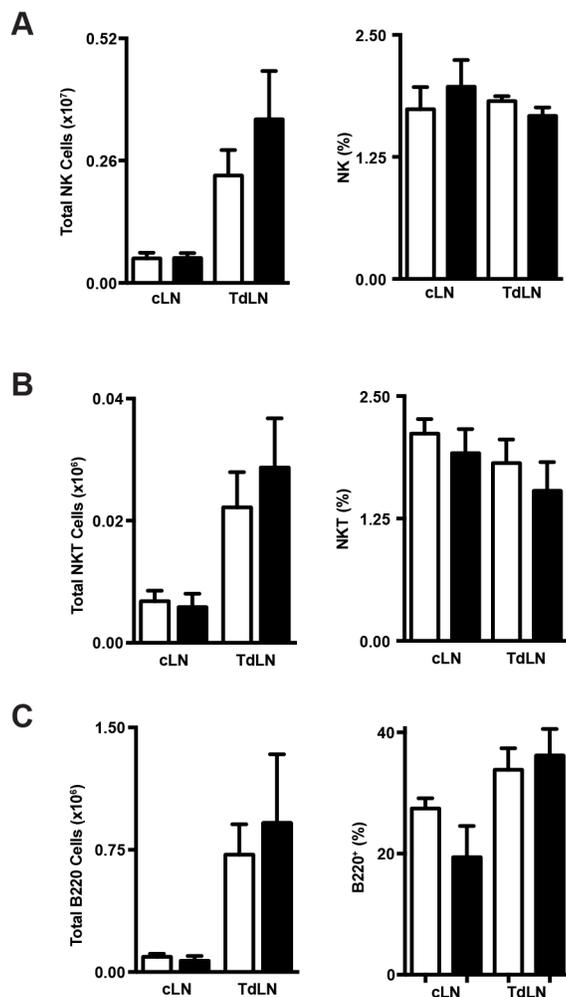


**Supplementary Figure 1: Absence of significant LN changes in NK, NKT and B cell populations of tumor-challenged shCD5EµTg mice.** Total cell numbers **(left)** and percentage **(right)** of NK **(A)**, NKT **(B)** and B **(C)** cells from TdLN and cLN from the same B16-F0-challenged shCD5EµTg mice as in Figure 2. Values are represented as mean ± SEM.
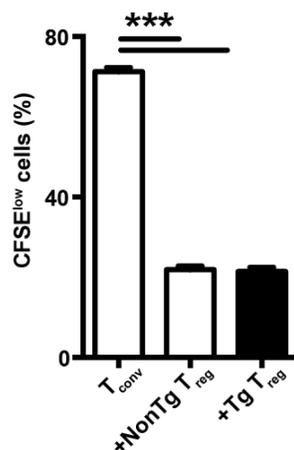
**Supplementary Figure 2: Absence of significant differences in the suppressive activity of $T_{reg}$ cells from shCD5EµTg and NonTg mice.** CFSE-stained $T_{conv}$ cells ($1 \times 10^5$ cells) from LN of NonTg mice were cultured in anti-CD3 mAb–coated plates plus soluble anti-CD28 mAb (1 µg/ml) in the absence or presence of $T_{reg}$ cells from NonTg and shCD5EµTg mice LN at 2:1 ratio ($5 \times 10^4$ cells). Shown is the percentage of CFSE$^{low}$ lymphocytes at day 3 of culture from one representative experiment out of three performed. Values are represented as mean ± SEM of duplicates from cells pooled from 3 mice of each phenotype. ***, p <0.0001 (unpaired *t* test).
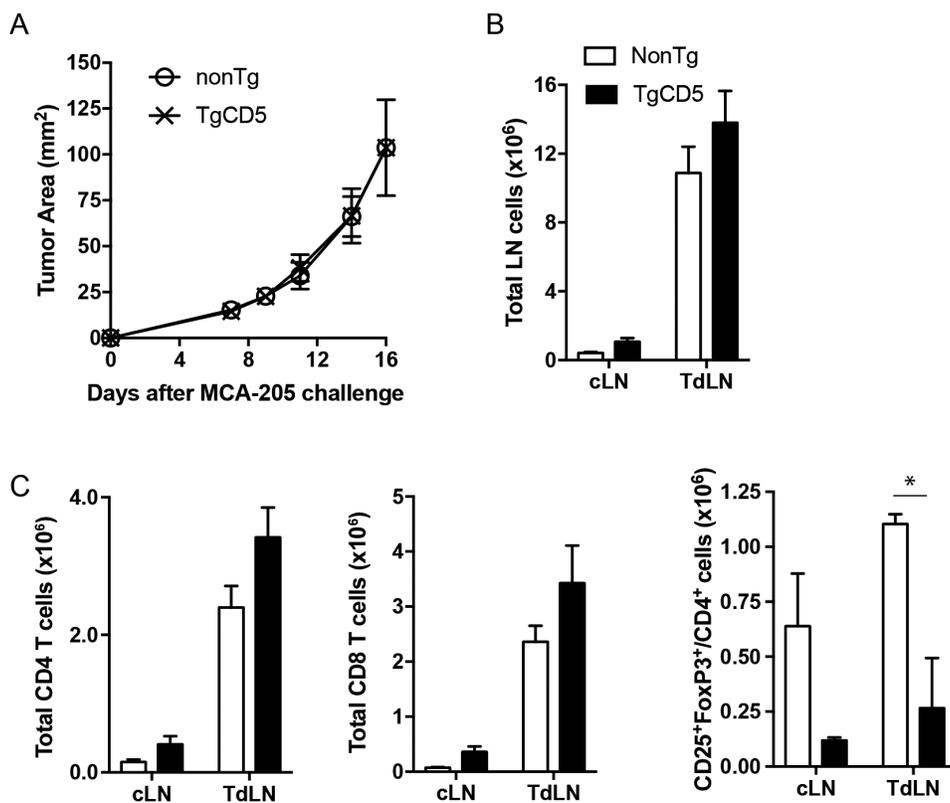


**Supplementary Figure 3: Analysis of lymphocyte changes in cLN and TdLN from MCA-205-challenged shCD5EµTg mice.** **(A)** shCD5EµTg (n = 4) and NonTg (n = 4) mice were injected *s.c.* with MCA-205 cells ($5 \times 10^4$) and tumor area measured every other day. **(B-C)** Total cell numbers, and total CD4$^+$, CD8$^+$ and $T_{reg}$ (CD25$^+$FoxP3$^+$CD4$^+$) T cells from cLN and TdLN of the same mice as in (A) were counted from single cell suspensions and analyzed by flow cytometry at day 16. Values are represented as mean ± SEM. *p< 0.05 (unpaired *t* test).