

# Effect of a long-acting IL-7 (NT-I7), on CD4 T cells during chronic LCMV

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## ABSTRACT

Failure of the immune system to respond effectively during chronic disease is largely attributed to T cell exhaustion. Our lab has shown the heterogeneity of exhausted CD8 T cells, with PD1+TCF1+ stem-like CD8 T cells being the treatment responsive subset. Long-acting IL-7, NT-I7, selectively expands stem-like CD8 T cells. Much less is known about CD4 T cells in chronic viral infections. Because of the role of CD4 T cells in CD8 T cell responses, we sought to characterize the CD4 T cell subsets, their response during chronic viral infection and the effect of NT-I7. Our data show that administration of NT-I7 during chronic LCMV:

- Increases the number of antigen-specific CD4 T cells in lymphoid and non-lymphoid tissues.
  - Increases the number of antigen-specific PD1+TCF1+ CD4 T cells.
  - Increases Th1 antigen-specific CD4 T cells,
- suggesting NT-I7 as an effective way to increase CD4 T cells. Future studies are ongoing describing the effect of NT-I7 on progenitor cells, mechanisms of CD4 T cell differentiation, their effect on B cell and antibody responses and combination therapy of NT-I7 with PD-1 blockade.

## INTRODUCTION

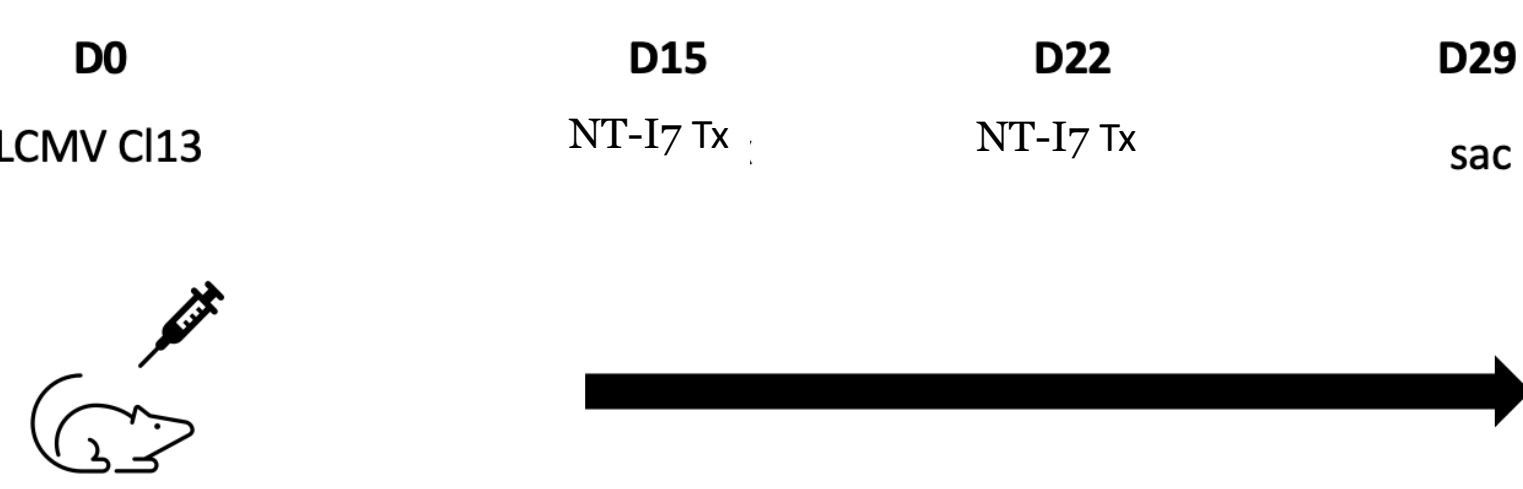
Despite their key role during antiviral responses, effector CD4 T cell activity is often reduced by antigen persistence, causing T cell exhaustion and inability to clear the infection.<sup>1</sup> In contrast to the Th1 effector responses during an acute infection, CD4 responses skew towards Tfh during chronic antigen stimulation.<sup>2</sup> IL-7, a signal of T cell survival and growth, can cause CD4 T cell expansion during chronic LCMV.<sup>3,4</sup> We aim to characterize CD4 T cell differentiation mechanisms, subsets and their phenotype and function during chronic LCMV upon NT-I7 treatment to inform a potentially more efficient T cell immunotherapy.

## RESULTS

Figure 1

### Experimental design

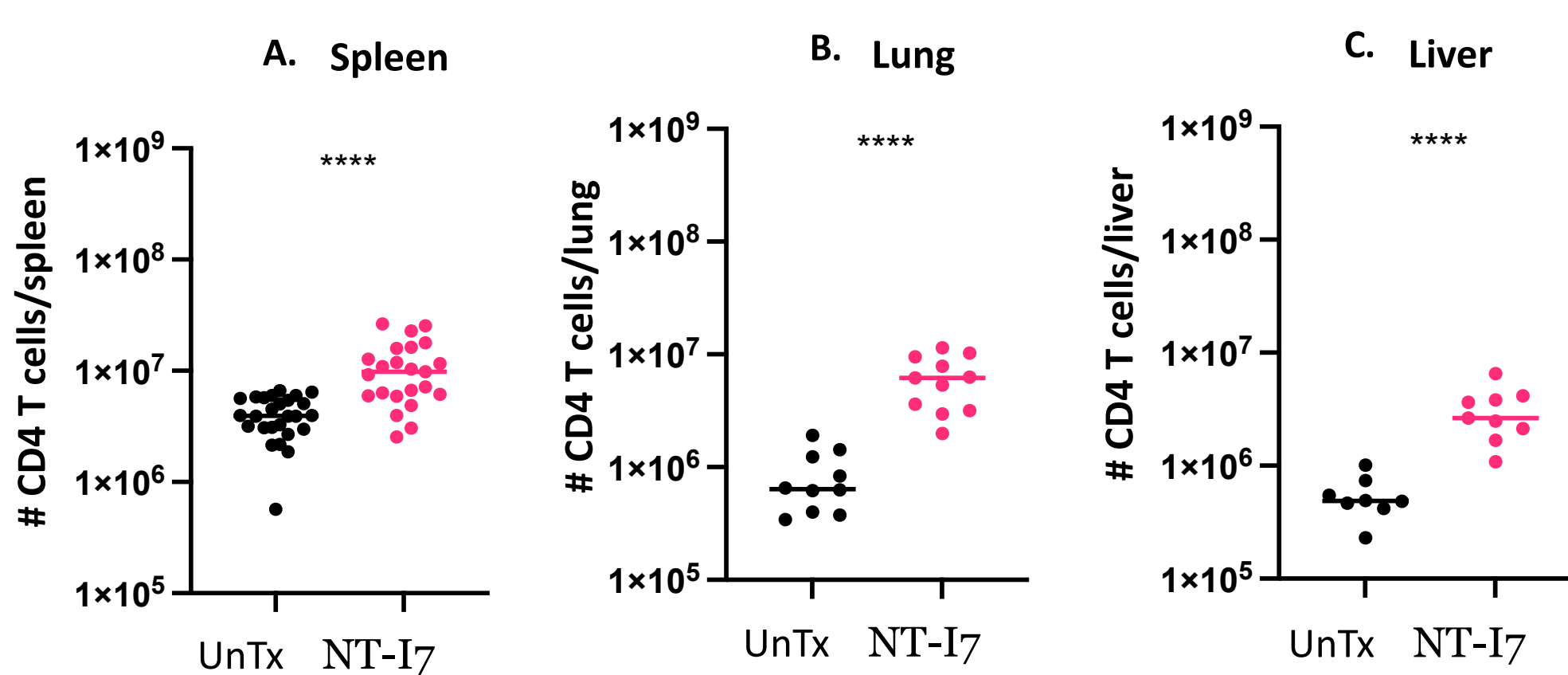
1. LCMV Clone 13 infected mice, were treated subcutaneously with 2 doses of NT-I7 (1 week apart) at 200µg /mouse, starting 15 days p.i.
2. 1 week after the second dose, tissues were collected for CD4 exhausted T cell phenotyping (immunostaining, flow cytometry analysis).



NT-I7: long-acting IL-7, product of NeolImmuneTech, Inc.

Figure 2

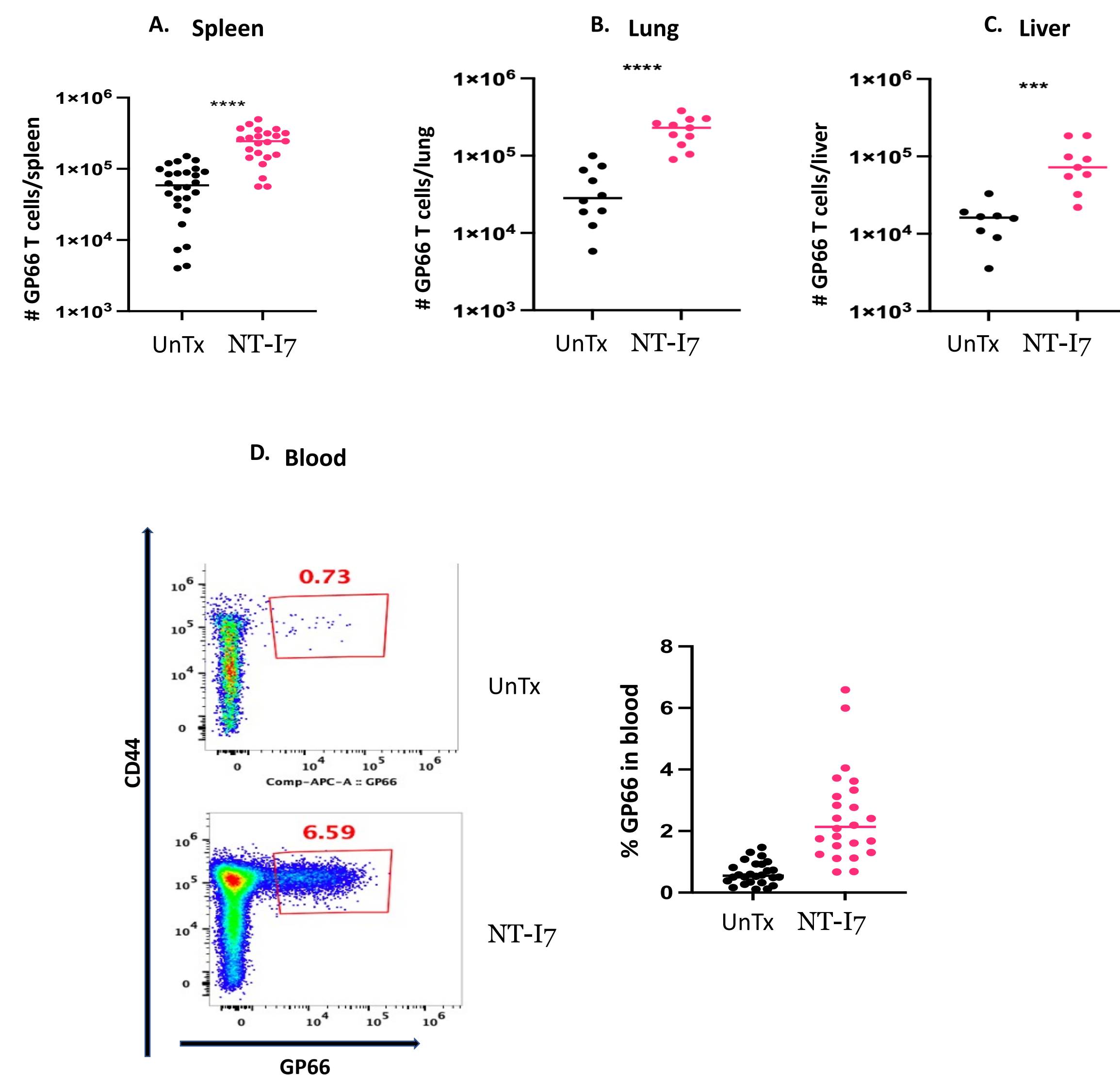
### NT-I7 therapy induces expansion of CD4 T cells in lymphoid and non-lymphoid tissues.



Number of CD4 T cells in chronically infected mice with LCMV clone 13 (day 29 after infection) with or without NT-I7 treatment. A. Spleen; Mice from 6 experiments (total n=26). B. Lung; Mice from 2 experiments (total n=10), C. Liver; Mice from 2 experiments (total n=9). Unpaired t-test, \*\*\*\* P ≤ 0.0001

Figure 3

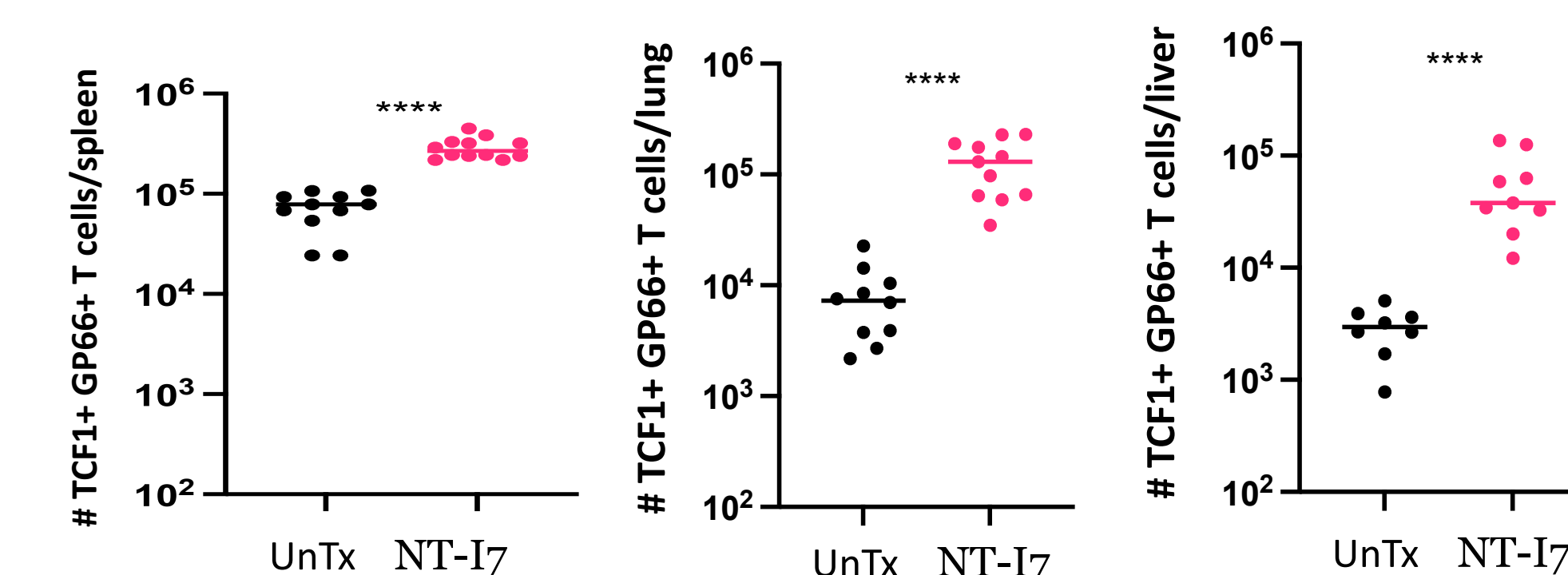
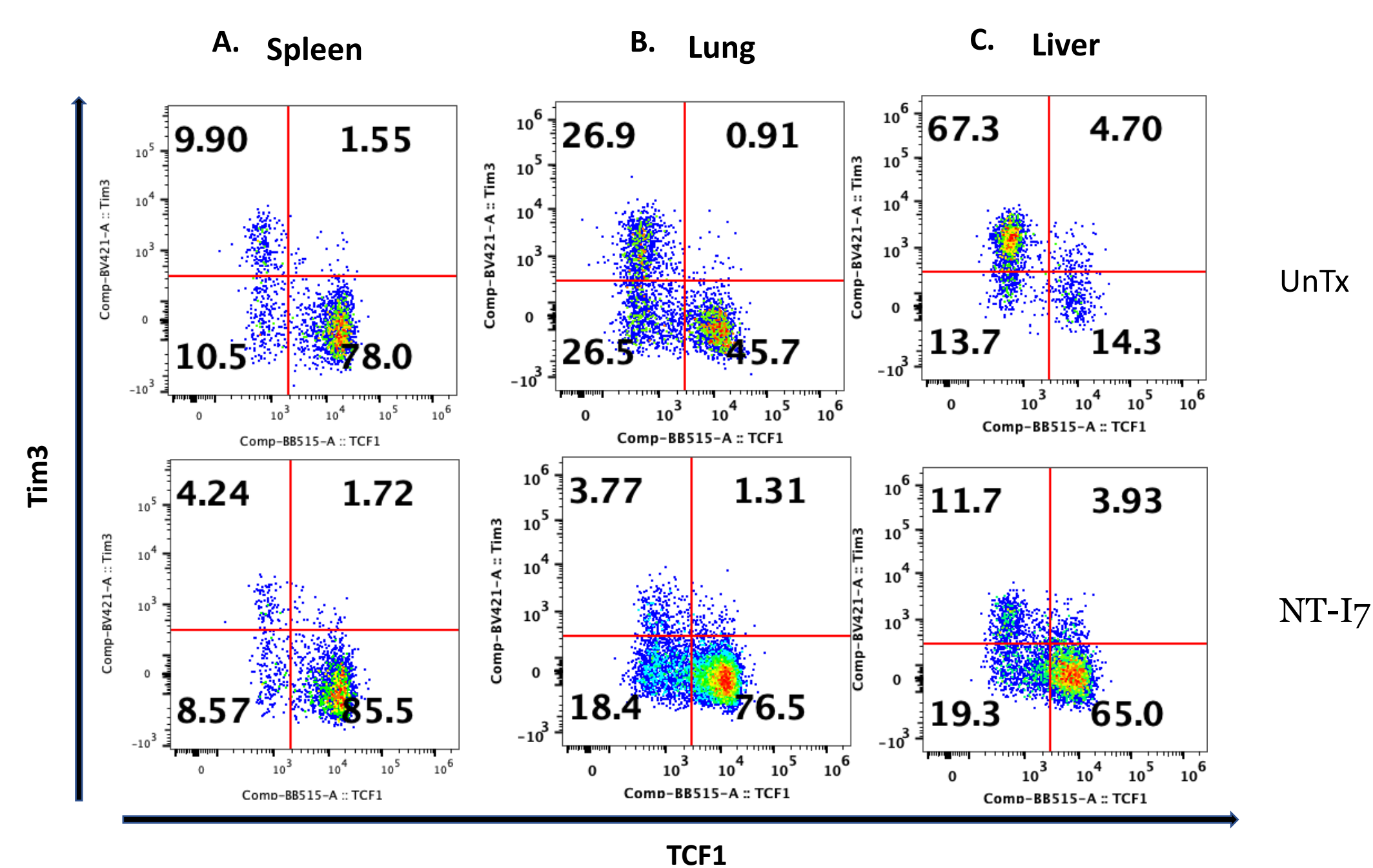
### NT-I7 induces expansion of LCMV-specific CD4 T cells.



Number of GP66+ CD4 T cells in chronically infected mice with LCMV clone 13 (day 29 after infection) with or without NT-I7 treatment. A. Spleen; Mice from 6 experiments (total n=26), B. Lung; Mice from 2 experiments (total n=10), C. Liver; Mice from 2 experiments (total n=9). D. Frequency of GP66+ CD4 T cells in blood; Mice from 6 experiments, (total n=26). Unpaired t-test, \*\*\* P ≤ 0.001 \*\*\*\* P ≤ 0.0001.

Figure 4

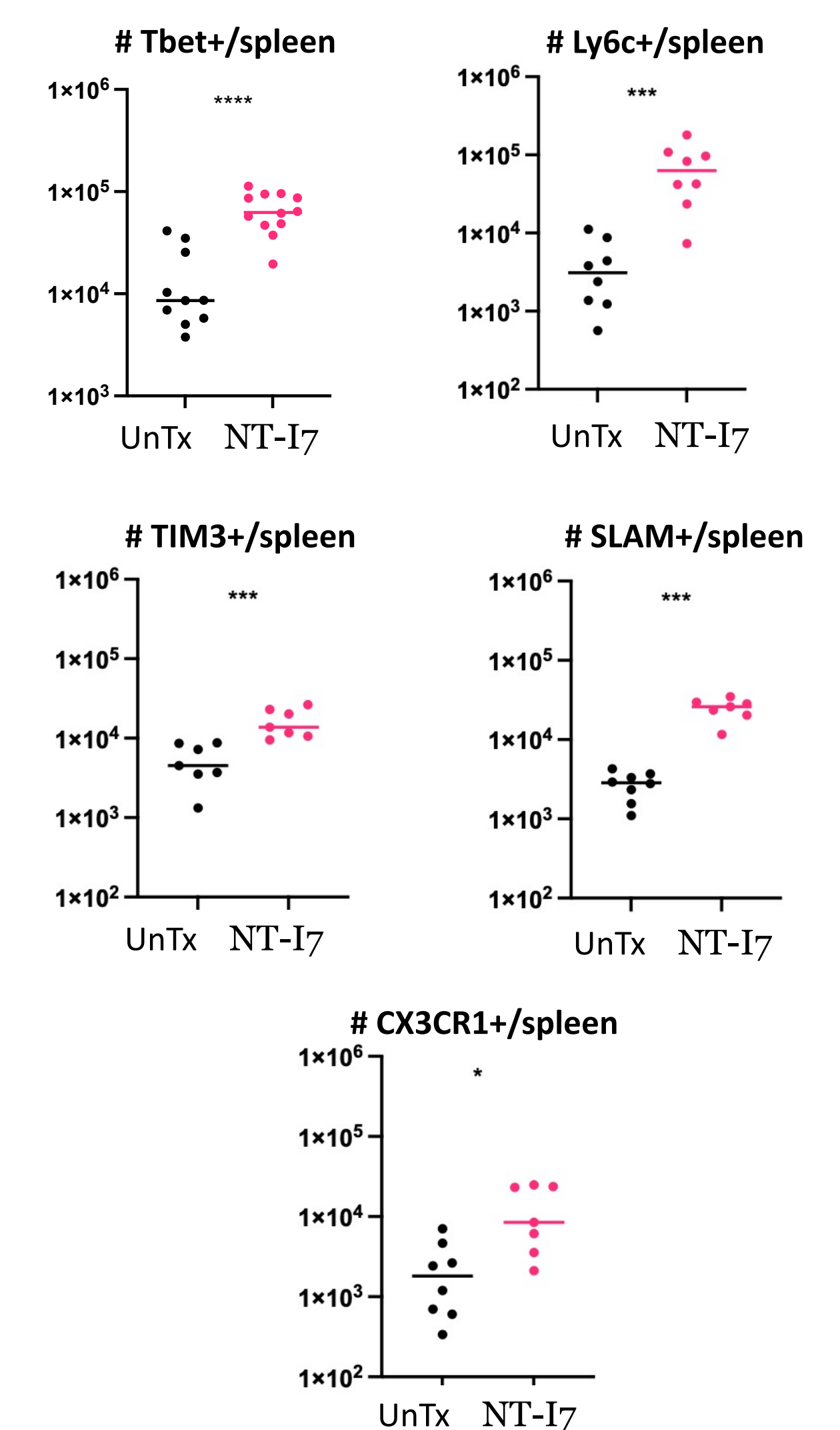
### NT-I7 induces expansion of LCMV-specific TCF1+ CD4 T cells.



Frequency (top) and number (bottom) of TCF1+ GP66+ CD4 T cells in chronically infected mice with LCMV clone 13 (day 29 after infection) with or without NT-I7 treatment. A. Spleen; Mice from 3 experiments (total n=11), B. Lung; Mice from 2 experiments (total n=10), C. Liver; Mice from 2 experiments (total n=9). Unpaired t-test, \*\*\*\* P ≤ 0.0001.

Figure 5

### Increased number of cells expressing Th1 markers upon NT-I7 treatment.



Number of TCF1+ GP66+ CD4 T cells in spleen of chronically infected mice with LCMV clone 13 (day 29 after infection) with or without NT-I7 treatment. Mice from 2-5 experiments (total n=7-20). Unpaired t-test, \*P ≤ 0.1 \*\*\*\* P ≤ 0.0001

## CONCLUSIONS

- NT-I7 increases the number of antigen-specific CD4 T cells in lymphoid and non-lymphoid tissues during chronic LCMV.
- NT-I7 increases the number of antigen-specific PD1+TCF1+ CD4 T cells.
- NT-I7 increases Th1 antigen-specific CD4 T cells.

## REFERENCES

1. Crawford, A., et al., Immunity, 2014
2. Fahey, L.M., et al, J Exp Med, 2011
3. Nanjappa, S.M., et al., Blood, 2011
4. Pellegrini, M., et al., Cell, 2011

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