



# Targeting antigens to CD180 induces a robust humoral response by activating immature B cells and promoting survival and proliferation of Ag-specific B cells

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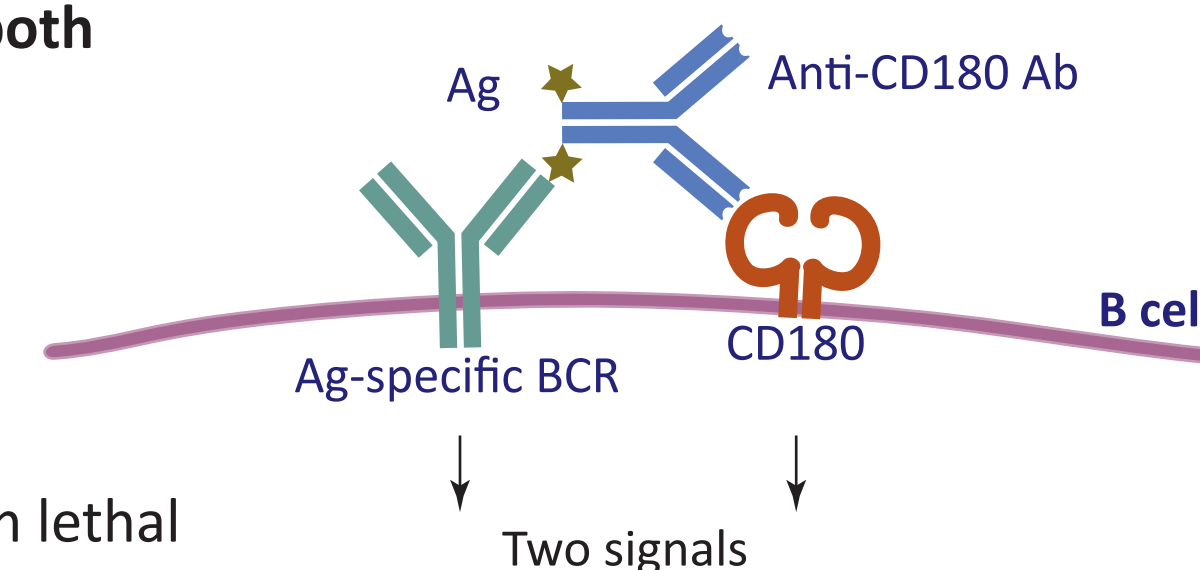
DEPARTMENT OF IMMUNOLOGY

## INTRODUCTION

**Antigen (Ag) targeting to CD180 delivers Ag directly to B cells and dendritic cells, activating both CD180 and B cell receptor (BCR) signaling.**

- Induces rapid Ag-specific high affinity IgG antibody (Ab) responses
- Ag must be directly conjugated to  $\alpha$ CD180 Ab for Ag-specific responses
- Generates Ag-specific long-lived plasma cells and robust B and T cell memory
- BAFFR<sup>-/-</sup> mice (lack mature B cells), but not  $\mu$ MT mice (lack all B cells) can be protected from lethal West Nile virus (WNV) infection by prior vaccination with a WNVprotein- $\alpha$ CD180 conjugate

Chaplin et al. JEM, 2013. Giordano et al. PLoSPathogens, 2017.



**CD180 (RP105) is a pattern recognition receptor related to TLR-4.**

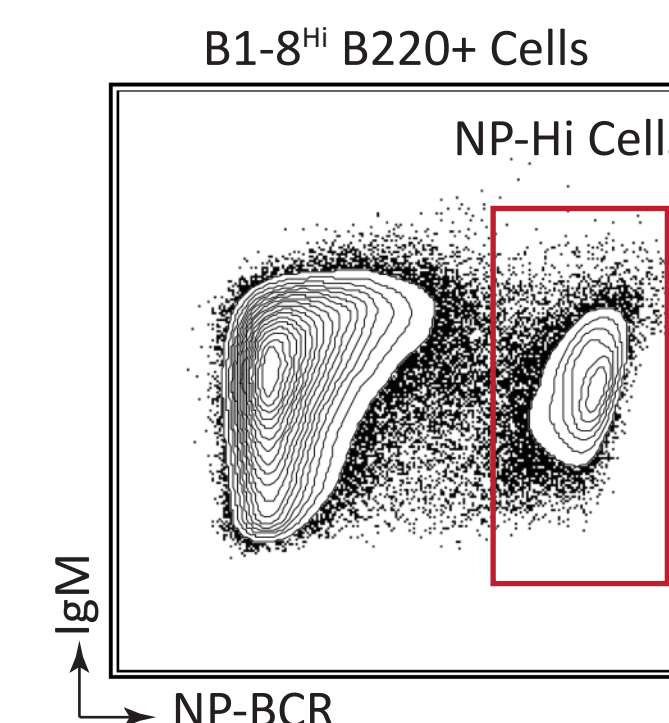
- Does not rely on the adaptor MyD88 for signal transduction
- Signaling resembles BCR signaling, relying on CD19, Lyn and PI3K
- Much of the signaling cascade in B cells remains ill-defined

### Hypothesis

- BCR plus CD180 signaling induces a balanced proliferative and differentiation program in immature and follicular (FO) B cells necessary for the robust formation of Ag-specific long-lived plasma cells

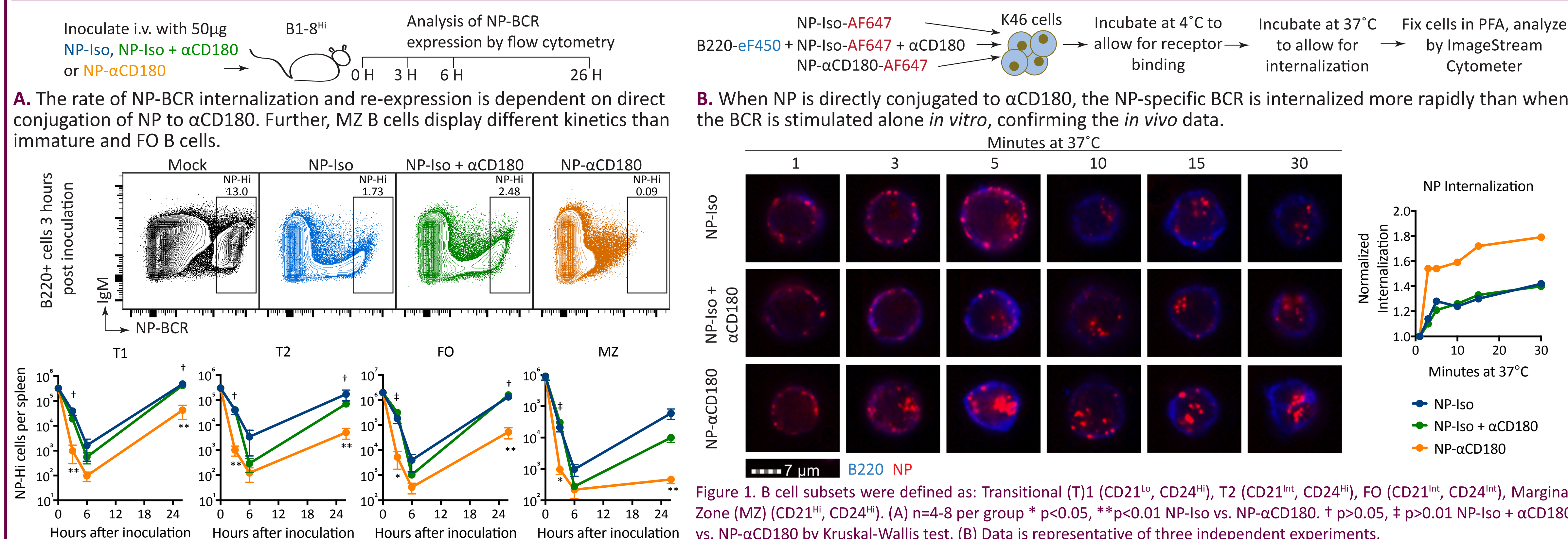
### Experimental Models

- B1-8<sup>hi</sup> mice: Engineered to express a BCR that binds to NP-hapten on 12-15% of splenic B cells.
- K46 $\mu$ M17 B cell line: Expresses both CD180 and NP-specific BCR.

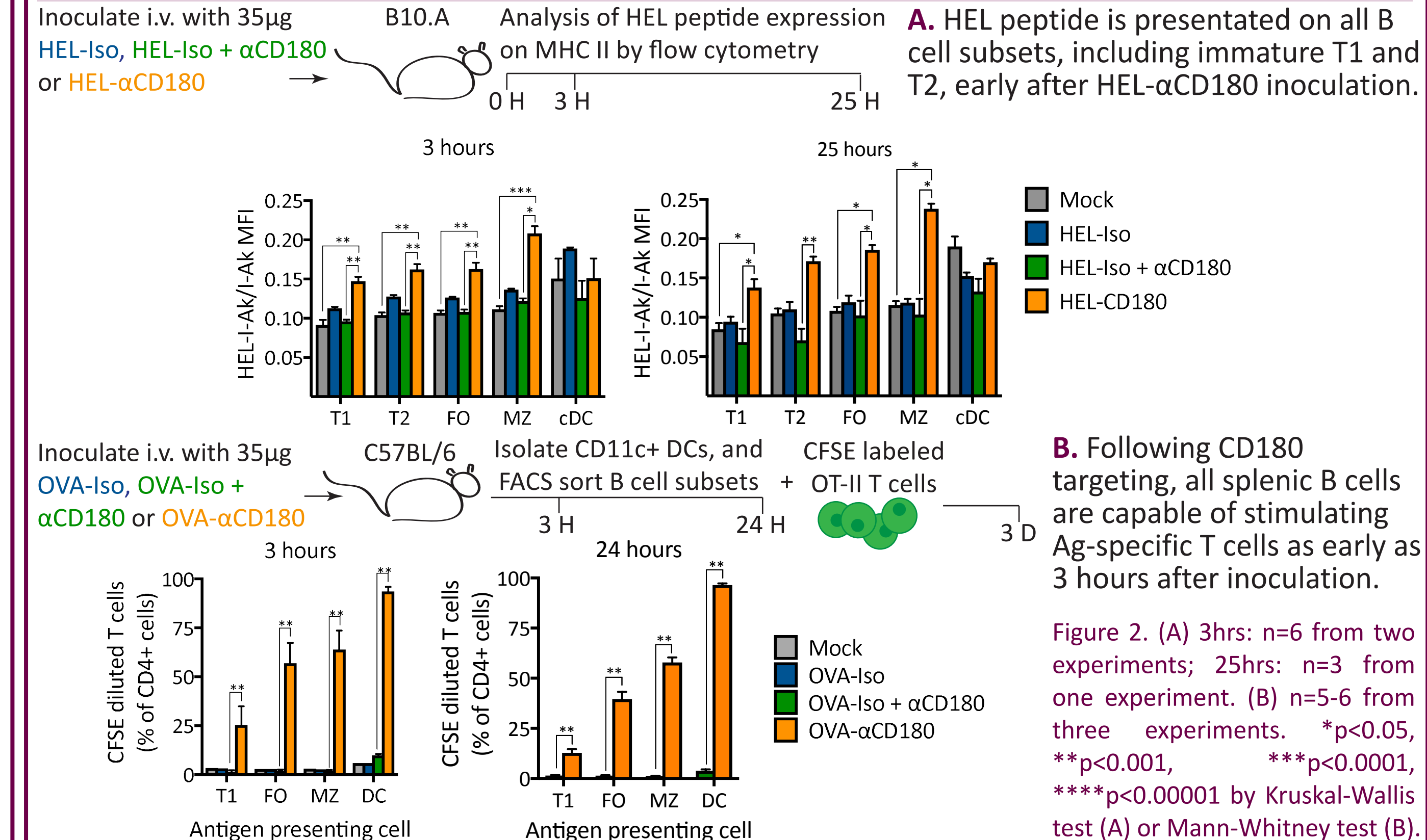


## RESULTS

**Figure 1. Ag-specific BCR internalization is more rapid upon contact with Ag- $\alpha$ CD180 than Ag-Iso +  $\alpha$ CD180**



**Figure 2. B cells present Ag and stimulate T cells following Ag targeting to CD180**



**Figure 3. Ag-specific B cell survival and proliferation depends on the direct conjugation between the Ag and  $\alpha$ CD180 in the inoculum and is T cell dependent**

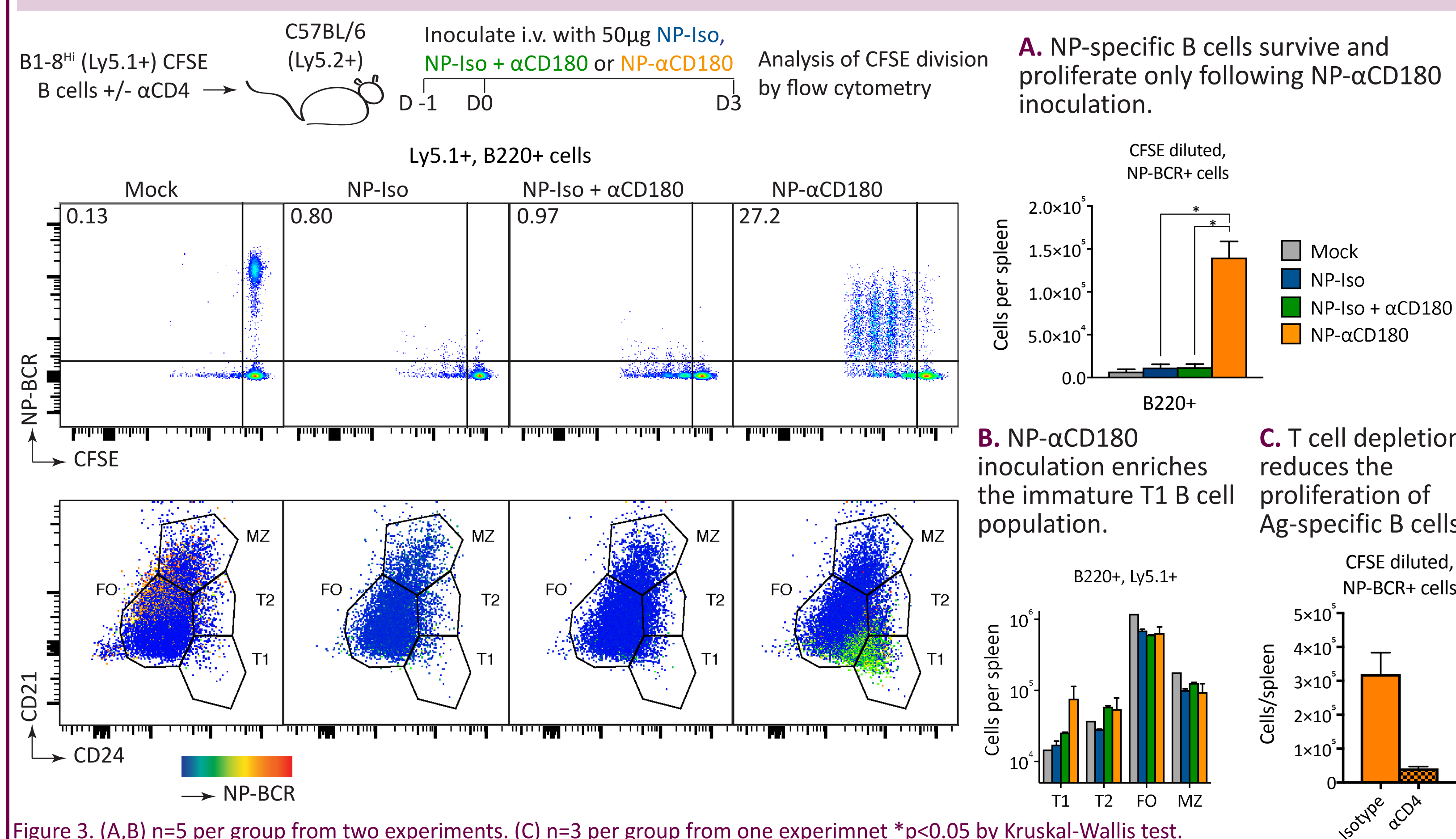


Figure 3. (A,B) n=5 per group from two experiments. (C) n=3 per group from one experiment \*p<0.05 by Kruskal-Wallis test.

**Figure 4. Ag presentation, Ag-specific B cell proliferation, T1 B cell expansion and the development of long-lived plasma cells occur following Ag- $\alpha$ CD180 inoculation, but not after Ag- $\alpha$ CD40 inoculation**

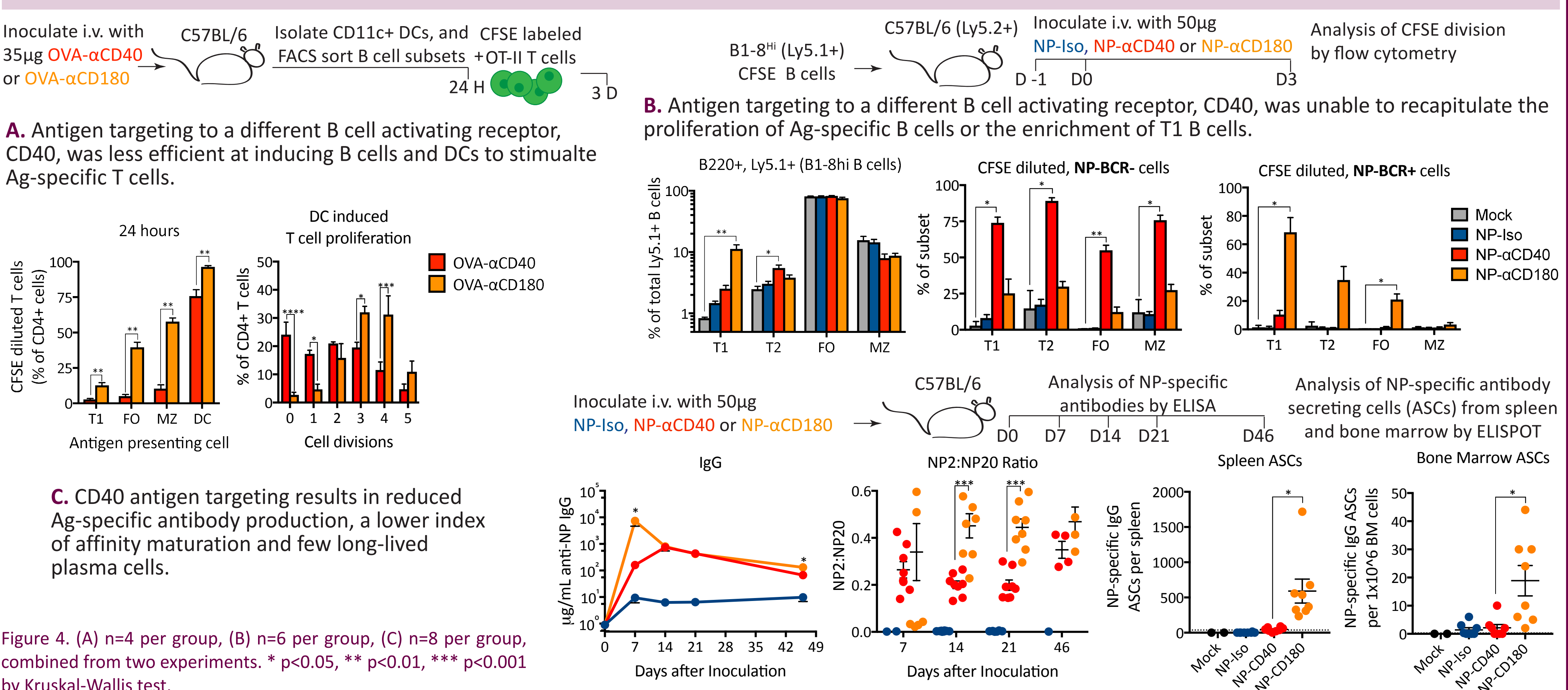
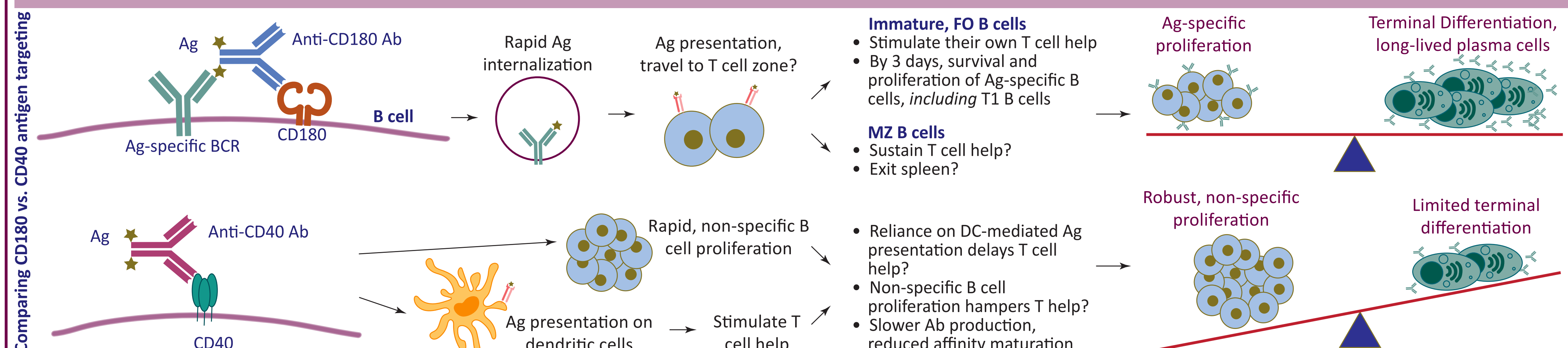


Figure 4. (A) n=4 per group, (B) n=6 per group, (C) n=8 per group, combined from two experiments. \* p<0.05, \*\* p<0.01, \*\*\* p<0.001 by Kruskal-Wallis test.

## MODELING CD180 ANTIGEN TARGETING



## FUTURE DIRECTIONS

- How does Ag targeting to CD180 affect the differentiation of plasma cells? How are different B cell subsets differentially programmed?
  - RNA seq to study the transcriptome of B cell subsets following Ag- $\alpha$ CD180 targeting.
- Can CD180 targeting induce Ag cross-presentation on B cells? What additional signals may be required?
- How and where do B cells and T cells interact in the spleen following CD180 targeting?
  - Studying changes in chemokine receptors on B and T cells.
  - Confocal microscopy following Ag- $\alpha$ CD180 targeting to observe migration of Ag-specific B and T cells.

## ACKNOWLEDGEMENTS

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