



Optimizing CD45 Targeting Immunotherapy

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Background

Clinical Significance of CD45 Antibodies

- Despite advances in AML therapy, outcomes remain poor
- CD45 = broadly expressed hematopoietic target
- CD45-targeted therapies are being explored:
 - Antibody drug conjugates (ADCs)
 - Radioimmunotherapy (RIT)
 - Chimeric antigen receptor (CAR-T) cells
- Novel CD45 antibodies may provide improved therapeutic targeting

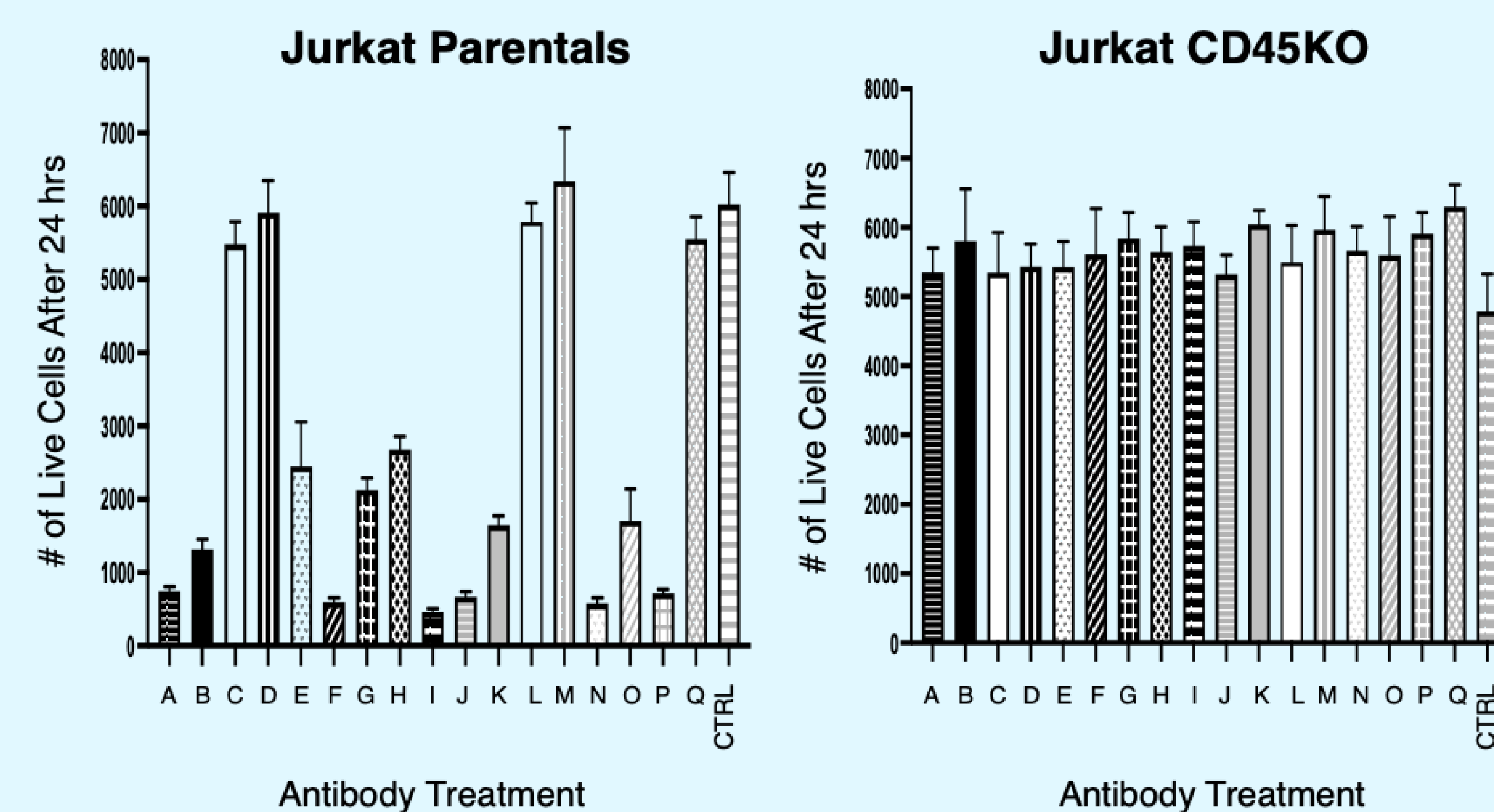


Figure 1. CD45 G4 Ab killing assays in Jurkat parental/CD45 KO pair (n=3)

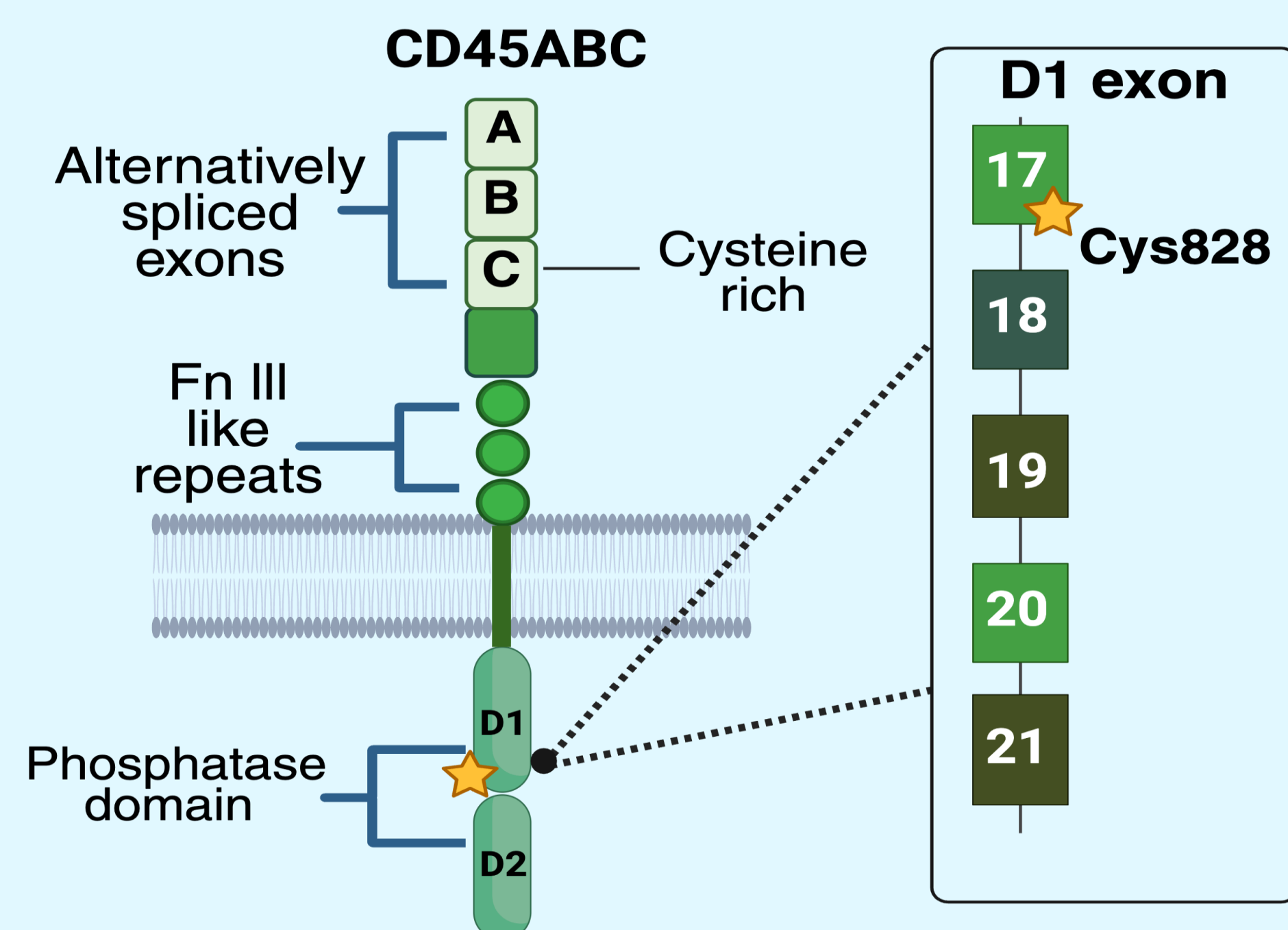


Figure 2. CD45ABC structure: alternatively spliced exons, Fn III like repeats, and phosphatase intracellular domains (ICD)

CD45 Antibody Killing

- CD45 antibodies show differential cytotoxicity
- Interested in manipulating CD45 domains to alter antibody mediated cytotoxicity

Research Questions

- Can novel CD45 mAbs be generated and evaluated for therapeutic potential?
- Do CD45 signaling properties influence the efficacy of CD45 mAbs?

Hypothesis: CD45 domain and structure influence the cellular response to CD45-targeting mAbs, and understanding these properties can inform their therapeutic application

Methods

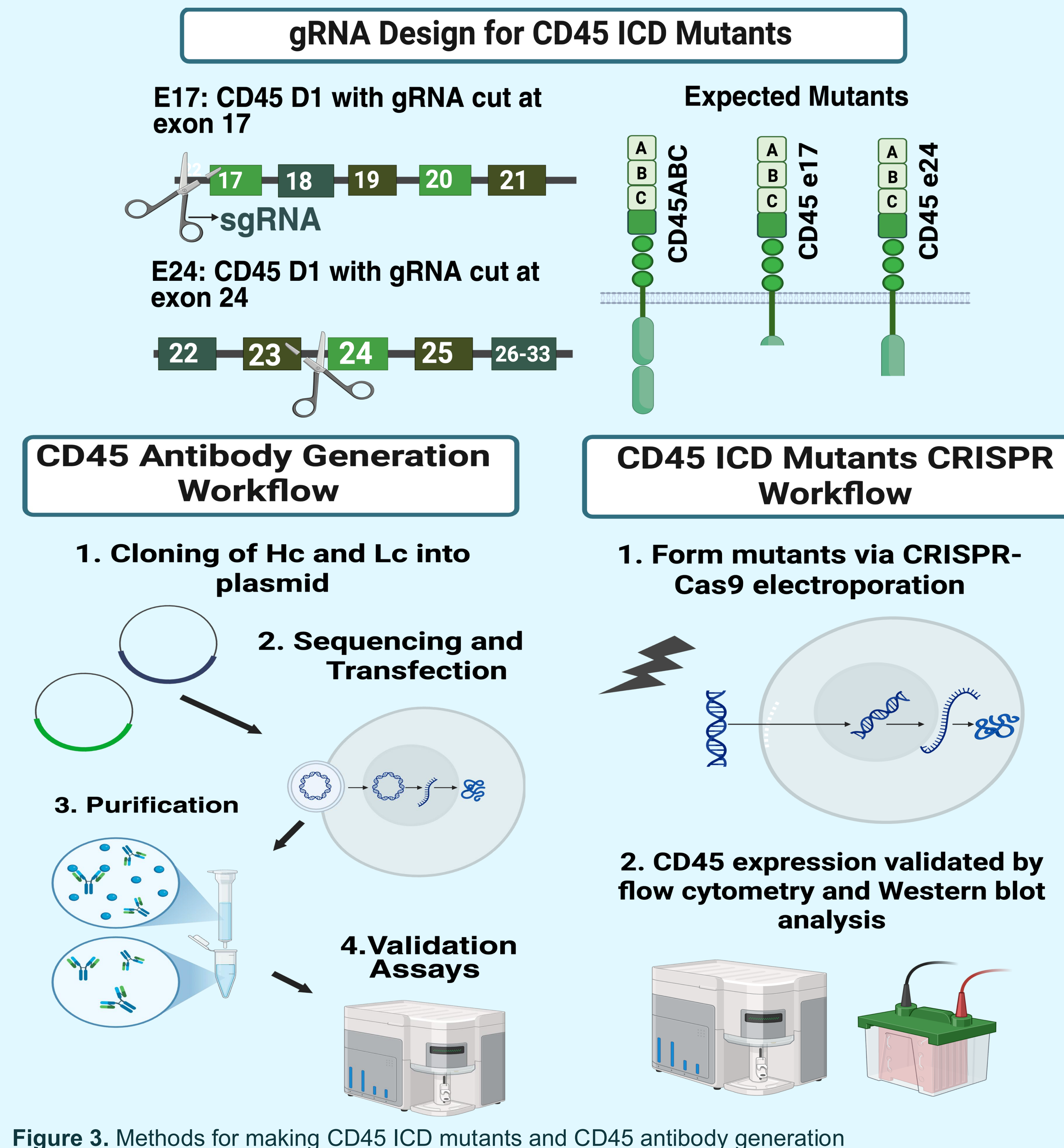


Figure 3. Methods for making CD45 ICD mutants and CD45 antibody generation

One mAb Successfully Detects CD45

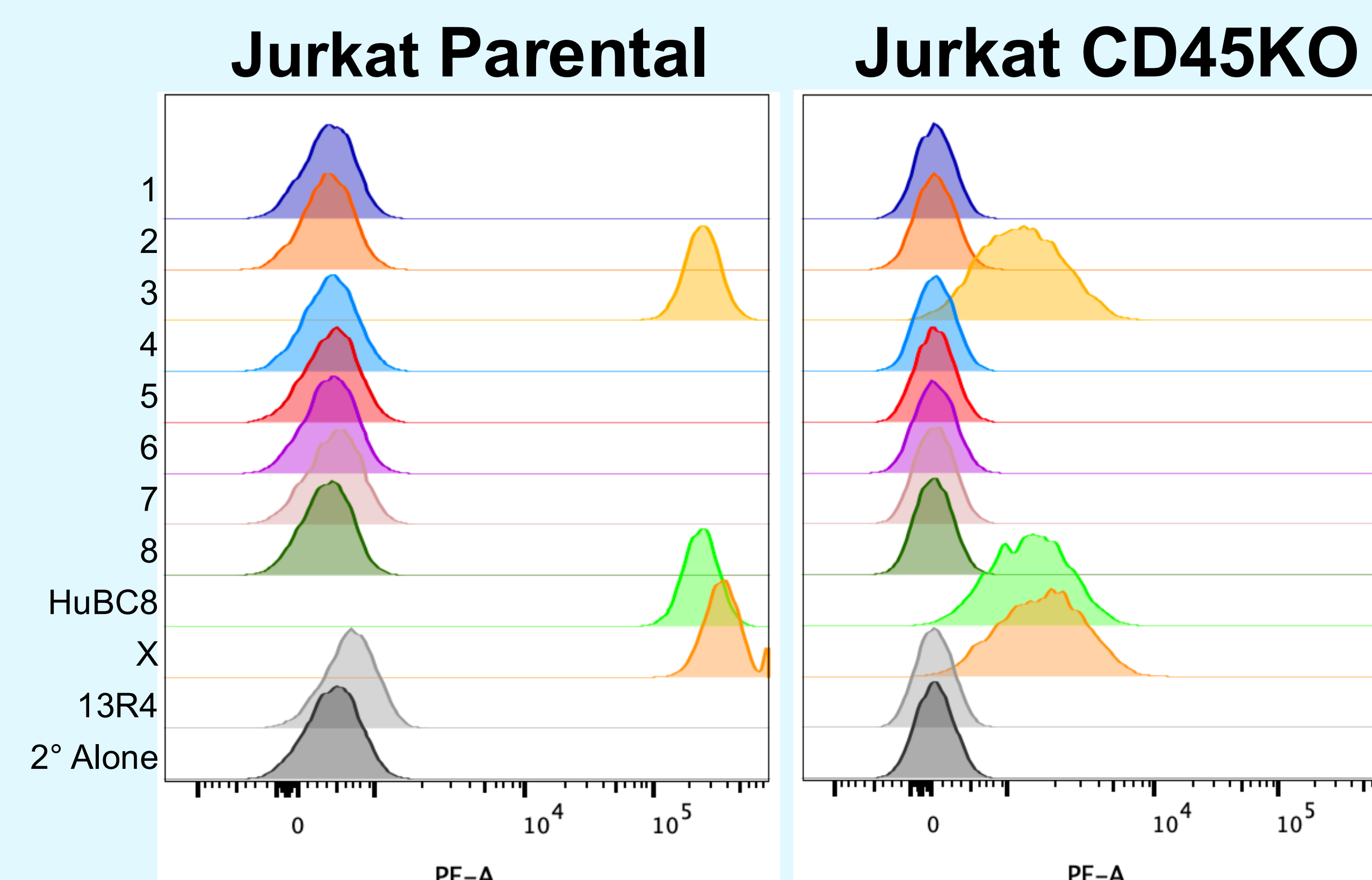


Figure 4. Flow cytometry staining for newly purified CD45 G4 abs

ICD Truncations Result in Loss of CD45 Expression

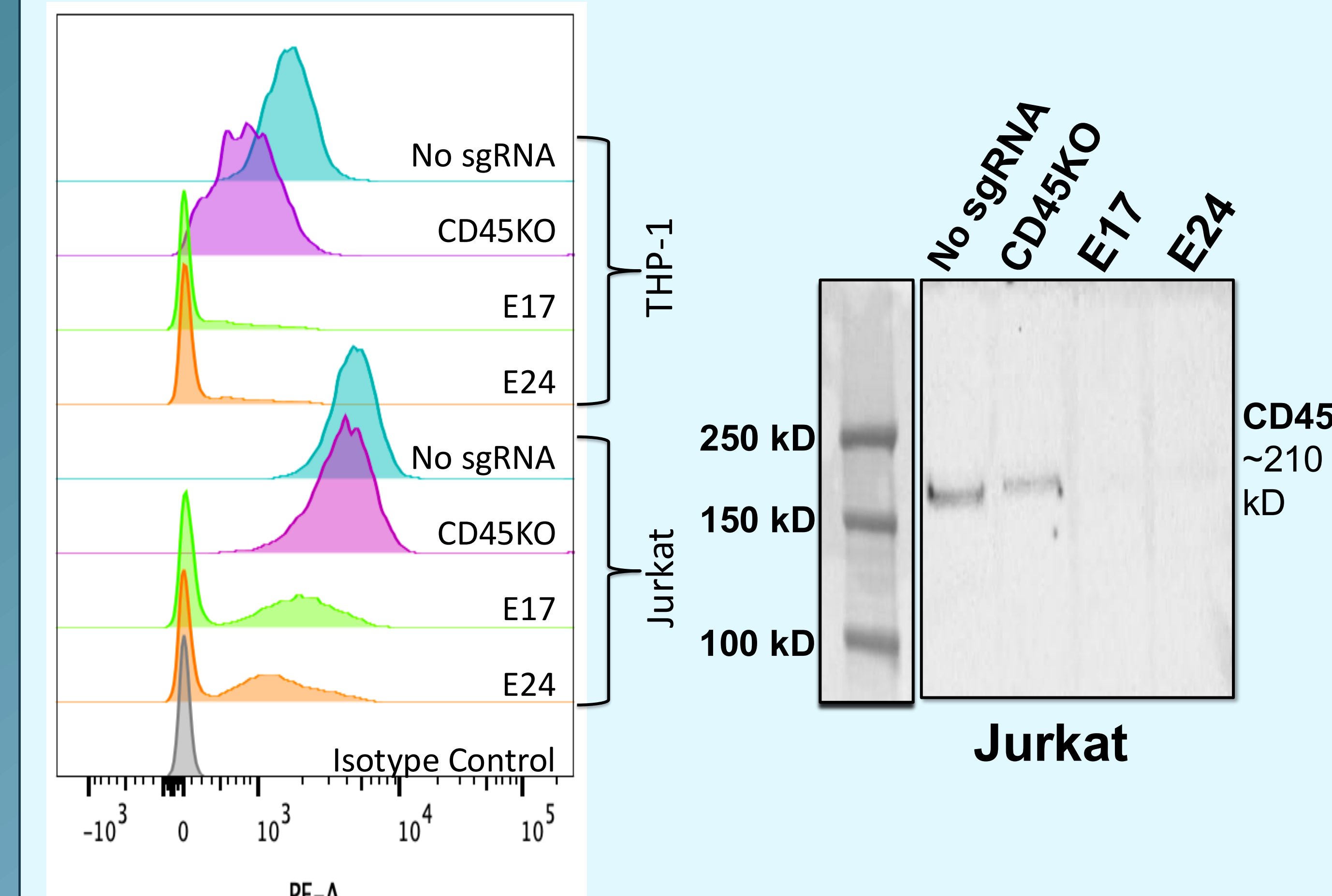


Figure 5. Flow cytometry results for ICD mutants

Figure 6. Western blot results for ICD mutants

Discussion

Loss of CD45 Expression

- Exon 17/24 targeting resulted in loss of CD45 protein
- NHEJ-mediated indels may have caused frameshifts and premature stop codons

CD45 Antibody Binding

- A single newly generated mAb demonstrated CD45 binding
- Differential binding may reflect epitope specificity, and accessibility
- Non-binding mabs may recognize altered or absent epitopes

Future Directions

CD45 Antibody Development

- Re-transfect CD45 antibodies to validate mAb binding
- Assess epitope specificity and accessibility
- Evaluate antibody 3 for therapeutic applications, including RIT

Acknowledgements

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