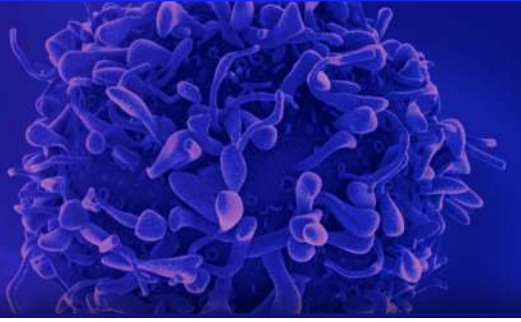
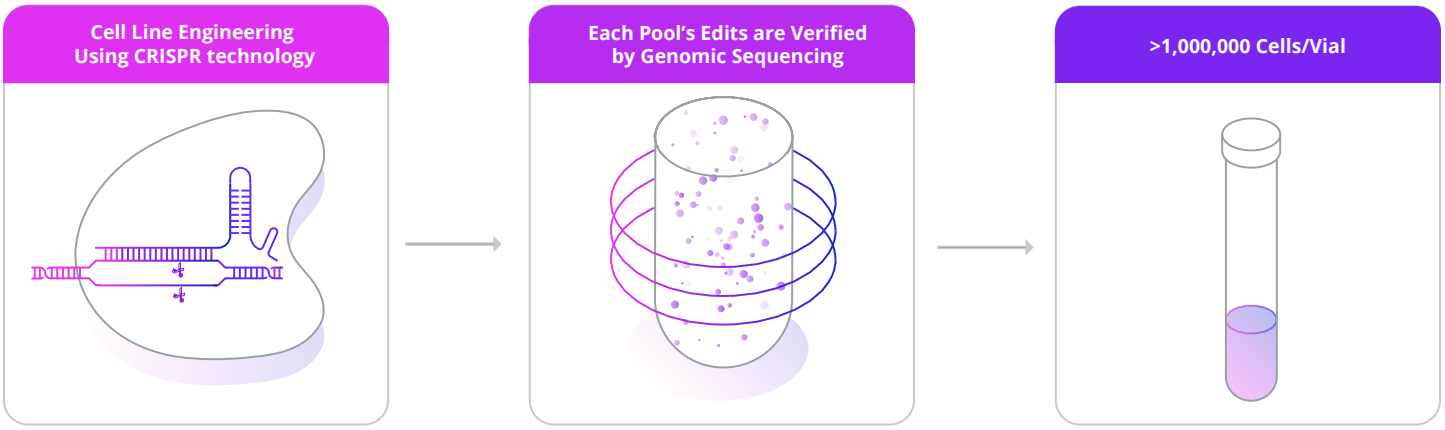
# Primary Cell KO Pools:

## Advance your immunology research with edited-to-order primary cells



High editing efficiency with reliable post-editing functionality. Order Primary Cells supplied by EditCo or work with our team to edit your primary cell type of interest.




OVERVIEW

### Skip to the main experiments with edited T cells

- ✓ **Confident Results:** 80% editing efficiency guaranteed smart multi-guide design.
- ✓ **Faster Results:** A 7-day editing protocol delivers results in 2 weeks or faster.
- ✓ **Flexibility:** Choose from EditCo-supplied cells or onboard your T cells.
- ✓ **Functionality:** High cell viability and editing levels without affecting functionality.

### T cells: Key Product Deliverables

- ✓ Regular updates on your order's progress
- ✓ 2 vials of edited cell pools with >1,000,000 cells/vial (pools consist of a heterogeneous population of edited and unedited cells)
- ✓ Control-transfected cell pools (2 vials)
- ✓ Sequence of synthetic gRNA used
- ✓ Primer sequences used for PCR and Sanger Sequencing
- ✓ ICE Sanger sequencing analysis reports for each edited clone after expansion
- ✓ Comprehensive QC report that includes the following information: mycoplasma test (positive/negative) and passage number

Using other primary cell lines?
**Reach out to us about custom projects!**

**Scan to learn more**



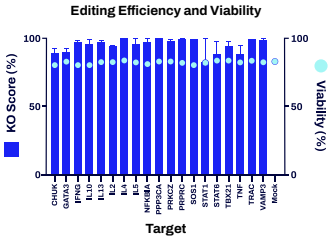
EditCo

**Efficient genome editing:** EditCo guarantees no less than 80% editing efficiency and >70% cell viability at time of freeze for both edited and CD8+ T cells.

## CD4+ T Cells

| Donor ID | Donor 1 | Donor 2 | Donor 3 |
|---|---|---|---|
| Race | White | White | White |
| Smoker | Non-Smoker | Non-Smoker | Non-Smoker |
| Age | 59 | 24 | 52 |
| Gender | F | M | F |

**Table 1.** Characteristics of edited T cell donors. Donors are screened for all major pathogens such as HIV, CMV and others.

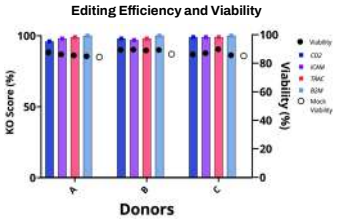



**CD4+ T Cells Viability and Editing efficiency.** Values represent the average of 3 donors. Error bars are S.E.M.

## CD8+ T Cells

| Donor ID | Donor A | Donor B | Donor C |
|---|---|---|---|
| Race | White | AA | White |
| Smoker | Non-Smoker | Non-Smoker | Non-Smoker |
| Age | 29 | 29 | 23 |
| Gender | F | F | M |

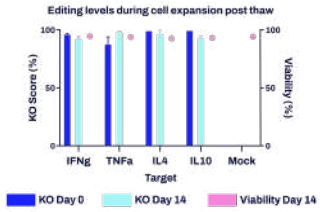
**Table 2.** Characteristics of CD8+ T cell donors. Donors are screened for all major pathogens such as HIV, CMV and others.




**CD8+ T Cells Viability and Editing efficiency.** Viability and editing efficiencies across all KO edits were measured in CD8+ T cell pools.

**Cell fitness is not affected by the editing process.** EditCo T cells maintain high cell viability and editing levels after thawing and expansion.

A



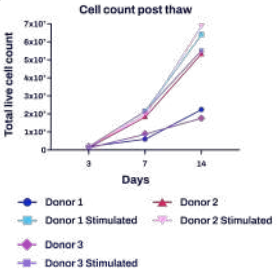

**CD4+ T cell pool stability after thaw.**

**A.** Pools of 4 different edits from 3 separate donors showed >90% viability with no decrease in editing efficiency after 14 days in culture.

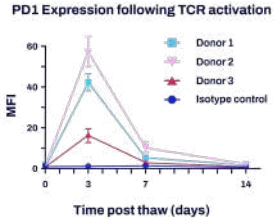
**B.** Total live cell count over a 14 day period from 3 separate donors expanded with CD3/CD2/CD28 stimulation (stimulated) or IL2 alone.

**C.** PD1 expression in TCR activator-treated cultures peaked on day 3 in all tested donors (edited and mock samples). Isotype control staining over the same period is shown for reference.

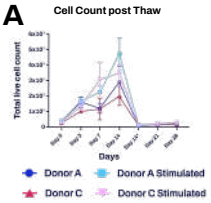
B



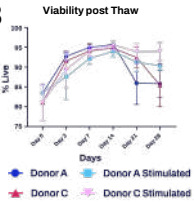

C



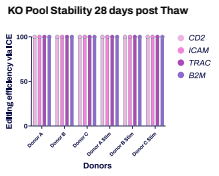

A



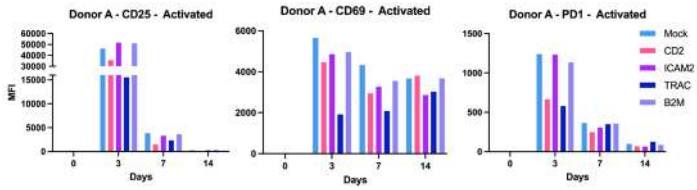

B




C

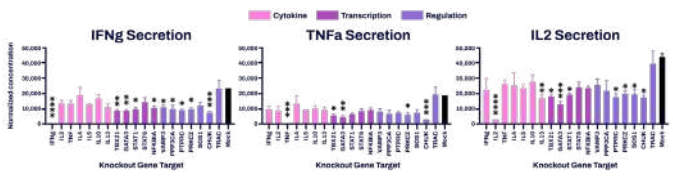



**CD8+ T cell pool stability after thaw.** **A.** Cell numbers and **B.** Viability were measured at the indicated time points. (*) Cell number was reduced to $10^6$/well on Day 14 to avoid overcrowding. **C.** Editing stability was measured at Day 28 in all cultures by PCR/Sanger sequencing. Charts show average values and Standard Error.

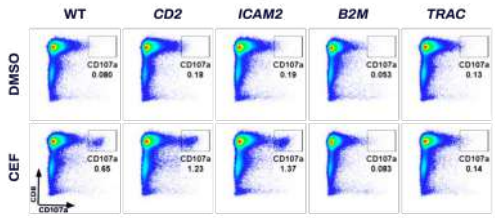



**Transient expression of activation and exhaustion markers.** Median Fluorescence Intensity (MFI) of activation markers (CD25 and CD69) or exhaustion markers (PD-1) in cultures of edited CD8+ T cells after thaw. Charts show average MFI and Standard Error.

**Functional cells following editing.** Edited CD4+ T cells produced levels of IFNg, IL2 and TNFa after mitogenic stimulation correlating with known target gene function. CD8+ T cells demonstrated antigen specific CD107a mobilization following CEF peptide stimulation.




**Cytokine secretion in edited cell pools following CD4+ T cell expansion.** Edited cells from 3 donors listed in Table 1 were thawed and then stimulated with PMA/ionomycin for 6 hours. Supernatants were harvested and measured for concentrations of IFNG (left panel), IL2 (center panel) and TNFA (right panel) as well as IL10 and IL5 (not shown) by FACS-ELISA (Miltenyi). Values were quantile normalized to reduce batch effects. Following ANOVA, Tukey's multiple comparison test was performed between Mock and each gene target to determine statistical significance. (*) p<0.05; (**) p<0.01; (***) p<0.005; (****) p< 0.0001.




**CD8+ T cell antigen-specific cytotoxic activity.** Edited CD8+ T cells were thawed and stimulated for 10 days with HLA-I matched, activated B cells loaded with CMV, EBV and IAV peptide mix (CEF). Antigen re-stimulated cells were incubated for 4 hours with CEF peptides in the presence of FITC labeled anti-CD107a antibody. Levels of CD107a expression in gated live, CD3+ cells were compared to unedited control (Mock). As expected, antigen specific responses were completely abrogated in TRAC and B2M edited CD8+ cells, compared to Mock, CD2 or ICAM-2 edited cells. These results are consistent with the canonical roles of B2M and TRAC.



EditCo