

**TRAV4-4/DV10\*01(1)**

**TRAV4-4/DV10\*01(2)**

**A**

**B**

**TRBV12-2\*01**

**UT**



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**C**

**Fig. S1.** (**A**) mTCR Expression of human PBL co-transduced with oligoclonal TCR alpha and beta chains. Two oligoclonal alpha chain and one oligoclonal beta chains were identified from murine KRAS G12D-reactive splenocytes (1M) by 5’RACE (table S4) and individually cloned into the retroviral vector, pMSGV1. Allogeneic PBL were stimulated with anti-CD3 (50ng/ml) for 2 days and co-transduced twice with retroviruses encoding oligoclonal TCR alpha and beta chains at 0.5 x 106 cells per well in a 24-well plate. Three days after transduction, T cells transduced with candidate pairs of TCR alpha and beta chains were labeled with anti-CD3, anti-CD8 and anti-mouse TCR beta antibodies, and analyzed on FACS Canto II. Data was gated on live CD3+ population. Untransduced T cells were used as controls. (**B**) Reactivity of PBL co-transduced with oligoclonal TCR alpha and beta chains. Anti-CD3 stimulated human PBL co-transduced with 2 paired alpha and beta chains were co-cultured with COS7/A11 transduced with minigenes encoding 23 aa of KRAS WT, KRAS G12D, and KRAS G12V, or KRAS genes encoding 165 aa of the protein, including WT (D1+D2), G12D (D1+D2), and G12V (D1+D2). Functional analysis was done by assessing IFN- production from the co-culture supernatant after overnight incubation. (**C**) Correlation between KRAS G12D expression and reactivity of KRAS G12D-reactive TCR. KRAS G12D expression was presented as % of ACTB, and reactivity of KRAS G12D-reactive TCR was presented as IFN- production of T cells transduced with TRAV4-4\*01/BV12-2\*01 against a panel of pancreatic tumor lines with or without G12D mutation (R2=0.978, P<0.001).