



**Supplementary Figure 2** High-throughput personalized screening strategy to detect T cell reactivity to cancer somatic mutations in peripheral blood of melanoma patients. **(i)** Peripheral blood mononuclear cells (PBMC) were obtained from a pre-treatment leukapheresis product. Tumor was resected to obtain DNA and RNA. **(iia)** PBMC were labeled (anti-CD3/ CD8/ PD-1), sorted based on PD-1 expression, and expanded *in vitro*. In parallel, the adherent fraction of the leukapheresis was cultured to generate autologous immature dendritic cells (DCs). **(iib)** Tumors underwent whole-exome sequencing (WES) and RNA sequencing to identify non-synonymous somatic mutations. Tandem minigene (TMG) constructs encoding up to 16 minigenes (mutant 25-mers) were used as templates to generate *in vitro* transcribed (IVT) RNA. **(iii)** IVT TMG RNA was used to transfect immature autologous dendritic cells (DCs) employed as targets in a coculture with the *in vitro* expanded peripheral blood subsets. At 20 h, T cell reactivity was analyzed by IFN- $\gamma$  enzyme-linked immunosorbent spot assay (ELISPOT), and by determining the percentage of the activation marker 4-1BB.