

Supplementary Fig. S1 Flowchart of identification, sorting and cloning of HLA-A2-restricted cancer-specific CTLs. Mononuclear cells from cancer patient was initially isolated and stimulated with cancer peptides *in vitro* for 14 days at 37°C. After 14days, mononuclear cells were stained with HLA-A2/tumor peptide tetramer and CD103 and sorted for CD103+ and CD103- cancer-specific CTLs by FACS. Sorted cells were then clonally expanded *in vitro* for 14 days at 37°C. After 14 days, CD103 and tetramer specificity is validated using flow cytometry. Clonality of CTLs were determined by RNA transcript extraction, cDNA generation and subsequent TCR sequencing.



Supplementary Fig. S2 Phenotype of paired CD103⁺ and CD103⁻ cancer-specific T cells. A. Representative of FACS dot plots of the differentiation stage of CD103⁺ and CD103⁻ cancer-specific T cells (SSX-2-specific). B. The frequency of CD103⁺ and CD103⁻ cancer-specific T cells expressing the standard Trm marker of CD69. C. T cell receptor sequences of CD103⁺ or CD103⁻ T cells of SSX-2-specific (*top*) and NY-ESO-1-specific (*bottom*) T cells. D. T cell receptor densities (by OKT3 staining MFI) of either CD103⁺ or CD103⁻ T cells of SSX-2-specific T cells (*left*) or NY-ESO-1-specific T cells (*right*) following OKT3 binding staining of 0.5µg.



Supplementary Fig. S3. T cell-mediated response of cancer-specific T cells without external antigen stimulation. Flow cytometry plots of IFN γ and TNF α responses by CD103+ of CD103-SSX-2-specific or NY-ESO-1-specific T cells following co-culture with HCT116 cancer cell line without external tumor antigen peptide stimulation.



Supplementary Fig. S4. T cell cytotoxic events. Brightfield and Annexin V images of antigen stimulated HCT116 cell death following co-exposure with either CD103⁺ (*left*) or CD103⁻ (*right*) SSX-specific T cells at 0,6 and 12 hours, with 0µM-stimulated HCT116 co-culture as controls. Annexin V excited at 632nm excitation and illustrated in blue.



Supplementary Fig. S5 Anti-tumor T cells cytotoxicity E-Cadherin- THP-1. Percentage of THP-1 cell death across a 12-hour time period following co-culture with no T cells as controls. (N, number of repeats=3). Data represented as median±s.e.m.



Supplementary Fig. S6 CD103+ or CD103- cancer-specific T cells migration towards E-Cadherin- THP-1. The number of T cells (either CD103⁺ or CD103⁻T cells) that migrated across a transwell membrane over a time period of 2 hours. SSX-2-specific T cell clones shown on the *left* and NY-ESO-1-specific T clones shown on the *right*. (N, number of repeats=3).



Supplementary Fig. S7 *Ex vivo* phenotypic analysis of CD103⁺ TILs. Representative flow cytometry plot of CD103⁺ CD39^{high}, CD103⁺ CD39^{low}, CD103⁺ CD39⁻ or total CD8⁺ T cells expressing either PD-1 or Tim3, derived from the paired tumor and paratumor tissue of one lung cancer patient.

Supplementary Table 1 Clinical parameters of cancer patients (for ex vivo analysis)

Clinical parameters of cancer patients						
Age	40-59	2				
	60-69	1				
	≥70	2				
	average (range)	65.6 (59-80)				
	Total	5				
Sex	Male	3				
	Female	2				
Tumor type	Lung	5				
Tumor origin	metastatic	0				
	non-metastatic	5				
Clinical tumor TNM stage	T1N0M0	4				
	T2N1M0	1				
Identification of cancer	Adenocarcinoma	3				
	Squamous cell carcinoma	2				
	-					

Clone	TRAV	CDR3(aa)	TRAJ	TRBV	CDR3(aa)	TRBJ
SSX2_CD103-Pos	8-6	CAVSDQNRDDKIIFGKG	30	6-1	CASSQGLTYGPSSYEQYFGPG	2-7
SSX2_CD103-Neg	8-6	CAVSDQNRDDKIIFGKG	30	6-1	CASSQGLTYGPSSYEQYFGPG	2-7
Ny-ESO_CD103-Pos	12-2	CAVDNNARLMFGDG	31	12-4	CASSQGAYGYTFGSG	1-2
Ny-ESO_CD103-Neg	12-2	CAVDNNARLMFGDG	31	12-4	CASSQGAYGYTFGSG	1-2