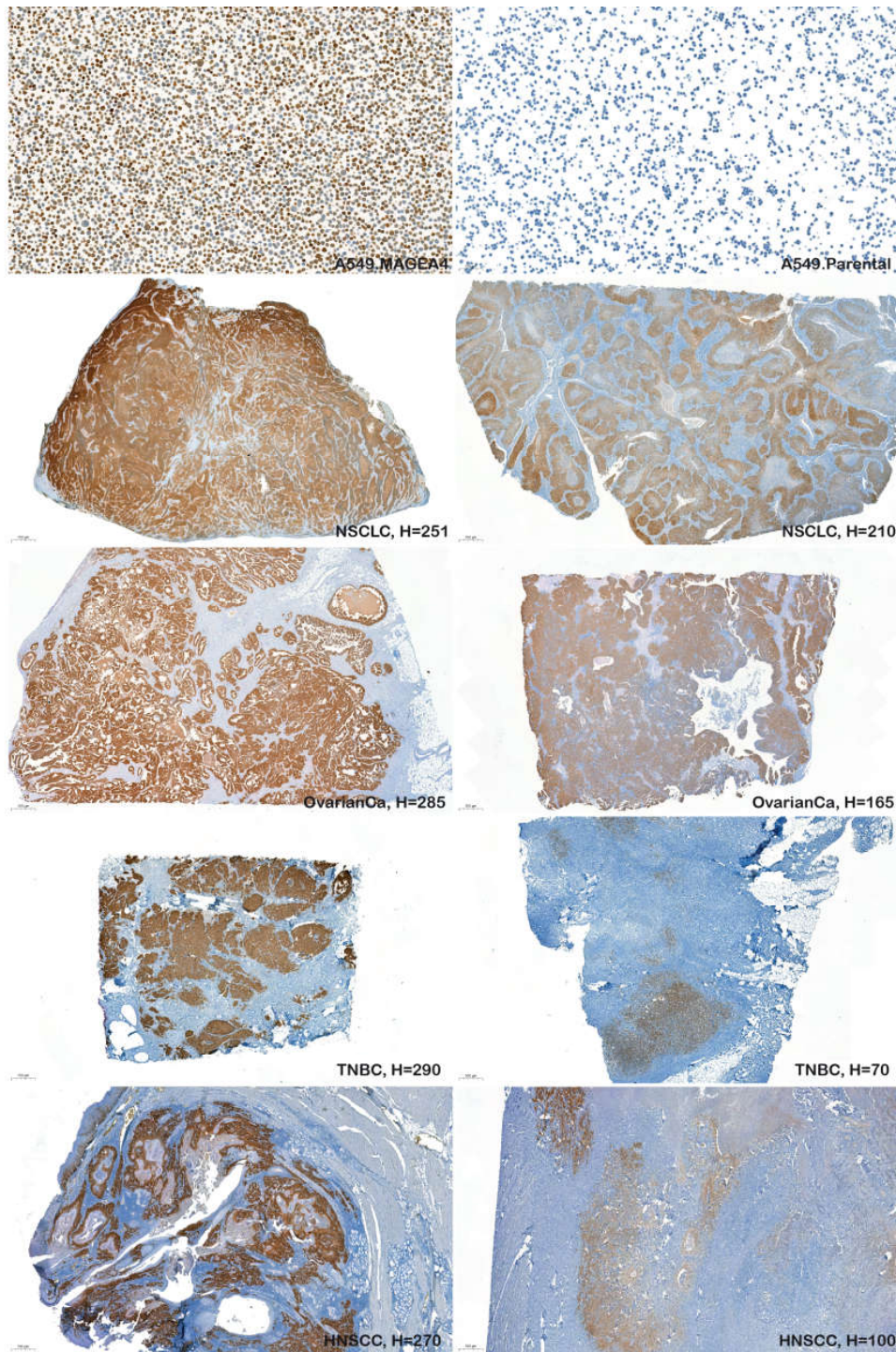## Supplementary Figure 1

Figure S1: MAGE-A4 is expressed in different solid tumors
Representative photomicrographs of primary patient-derived cancer tissue sections. A549 cells transduced with MAGE-A4 or not (parental) serve as technical controls. Brown = MAGE-A4 signal, blue = hematoxylin counterstain. H-score (H) is stated for each respective photograph in decreasing order (left to right). NSCLC = non-small cell lung carcinoma, Ovarian = ovarian carcinoma, HNSCC = head and neck squamous cell carcinoma, TNBC = triple-negative breast carcinoma.

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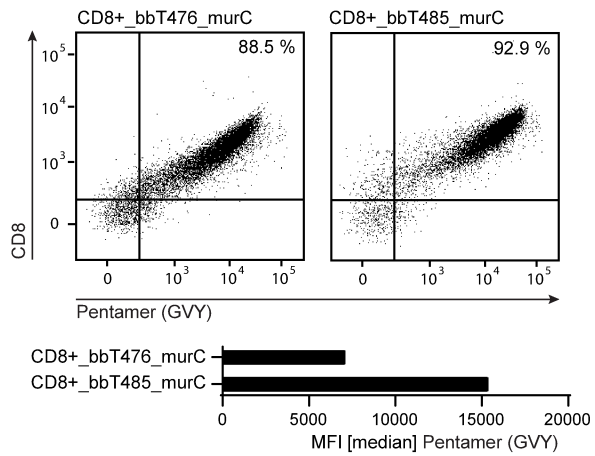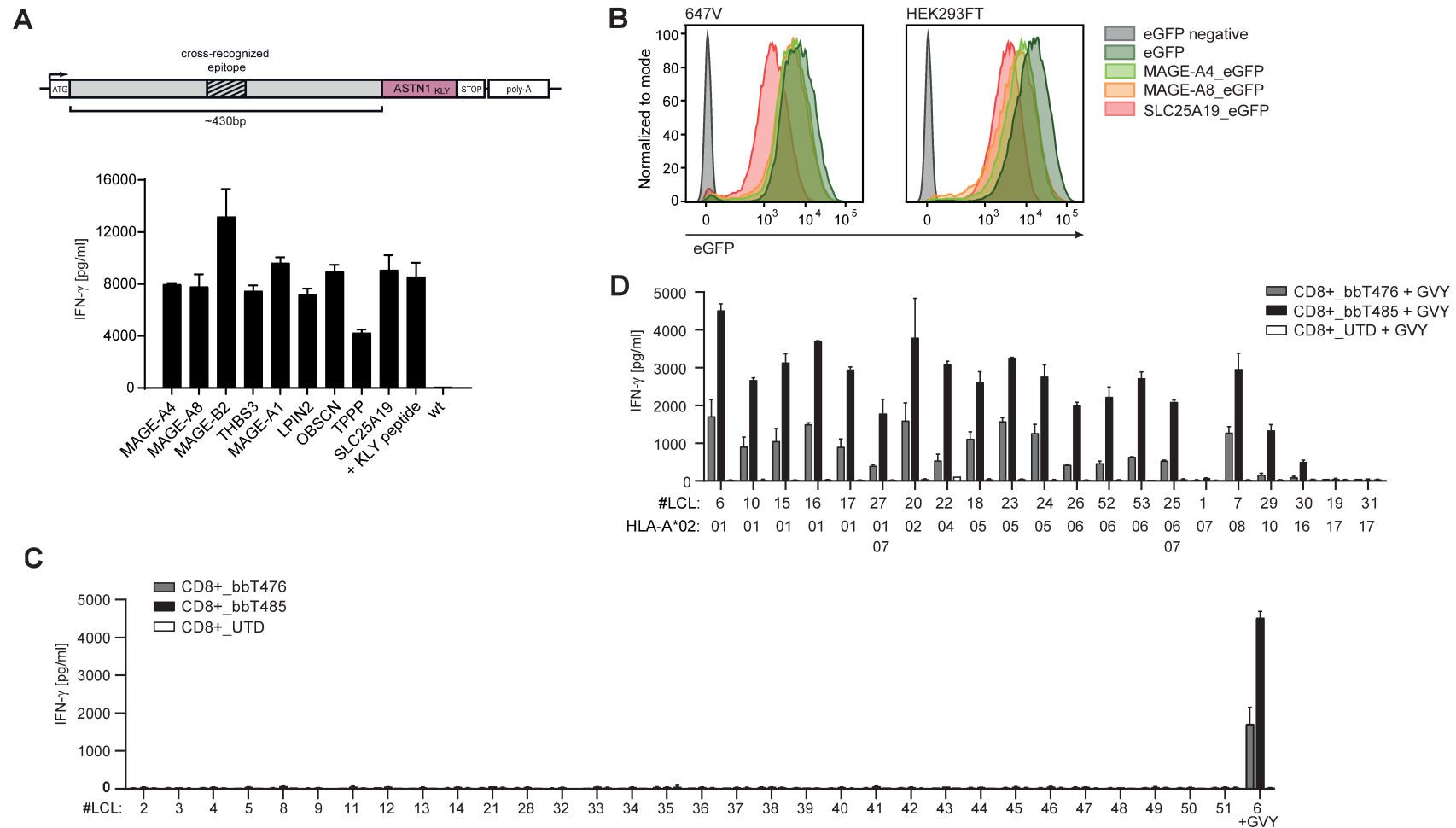
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Supplementary Figure 2

Figure S2: bbT485 TCR-Ts expressing an allo-derived murinized TCR exhibits superior epitope binding characteristics compared to bbT476 expressing the auto-derived murinized TCR
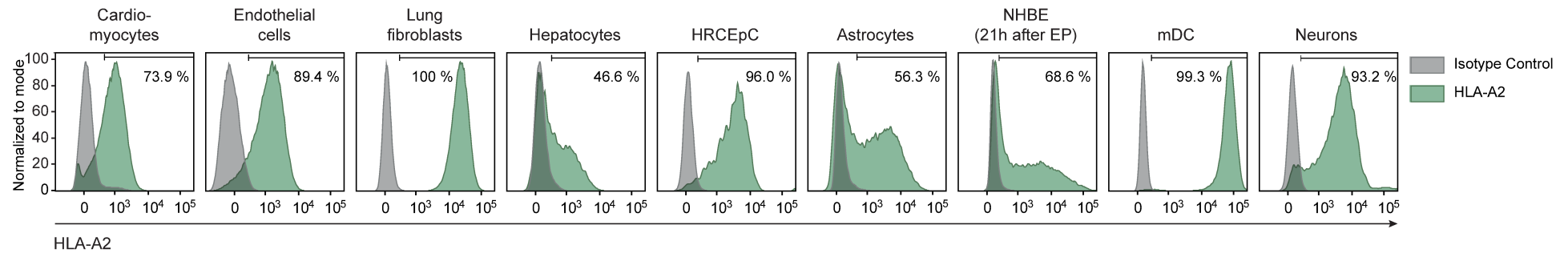Dot plots indicating the percentage of HLA-A2-GVYDGREHTV pentamer-positive CD8+ T cells and bar graphs showing median fluorescence intensity within respective gates, murC indicates use of murinized constant regions for TCR expression. Data shown are representative of 3 different donors for each tested TCR.

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Supplementary Figure 3

Figure S3: Engineered target cells for safety testing express mismatched sequences and HLA-allo cross reactivity

A, An example of used midigene-constructs including the minigene encoding the control epitope ASTN1 (KLYGLDWAEL) is shown above. Bar graphs displaying IFN-γ concentrations in supernatants 16 h after co-culture of CD8+ T cells expressing a transgenic control-TCR with A2+K562 cells transfected with ivt-RNA encoding mismatched peptide-sequences or GVYDGREHTV (MAGE-A4) in fusion with a respective control-epitope sequence at the 3' end. A2+K562 cells loaded with the KLYGLDWAEL (KLY) peptide serve as control. Parental protein-names indicate transfected midi-genes, wt = non-transfected A2+K562 cells. **B,** Histogram overlays displaying fluorescence intensity of eGFP determined by flow cytometry. Shown are data of 647V cells and HEK293FT cells transduced with eGFP, MAGE-A4_eGFP, MAGE-A8_eGFP or SLC25A19_eGFP. Untransduced T2 cells served as eGFP-negative control. **C,** LCLs expressing different HLA-A, -B and -C allotypes were tested for HLA allo-cross recognition in co-culture assays. HLA-typing of each LCL is given in supplementary table 1. HLA-A*02:01-positive LCL number 6 loaded with the GVY peptide served as positive controls. **D,** IFN-γ ELISA after co-culture with LCLs expressing different HLA-A*02 sub-alleles as indicated on x-axes loaded with the GVY peptide.

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Supplementary Figure 4

Figure S4: Normal tissue cells applied for safety testing express HLA-A*02:01-encoded surface protein
Histogram overlays displaying HLA-A2 expression determined by flow cytometry on indicated primary or iPSC-derived cells. Normal human bronchial epithelial cells (NHBE) were transfected with HLA-A*02:01-encoding ivt-RNA 21 h prior to assay. HRCEpC = human renal cortical epithelial cells; mDC = monocyte-derived dendritic cells, prepared as described in Material and Methods.

Supplementary Figure 5
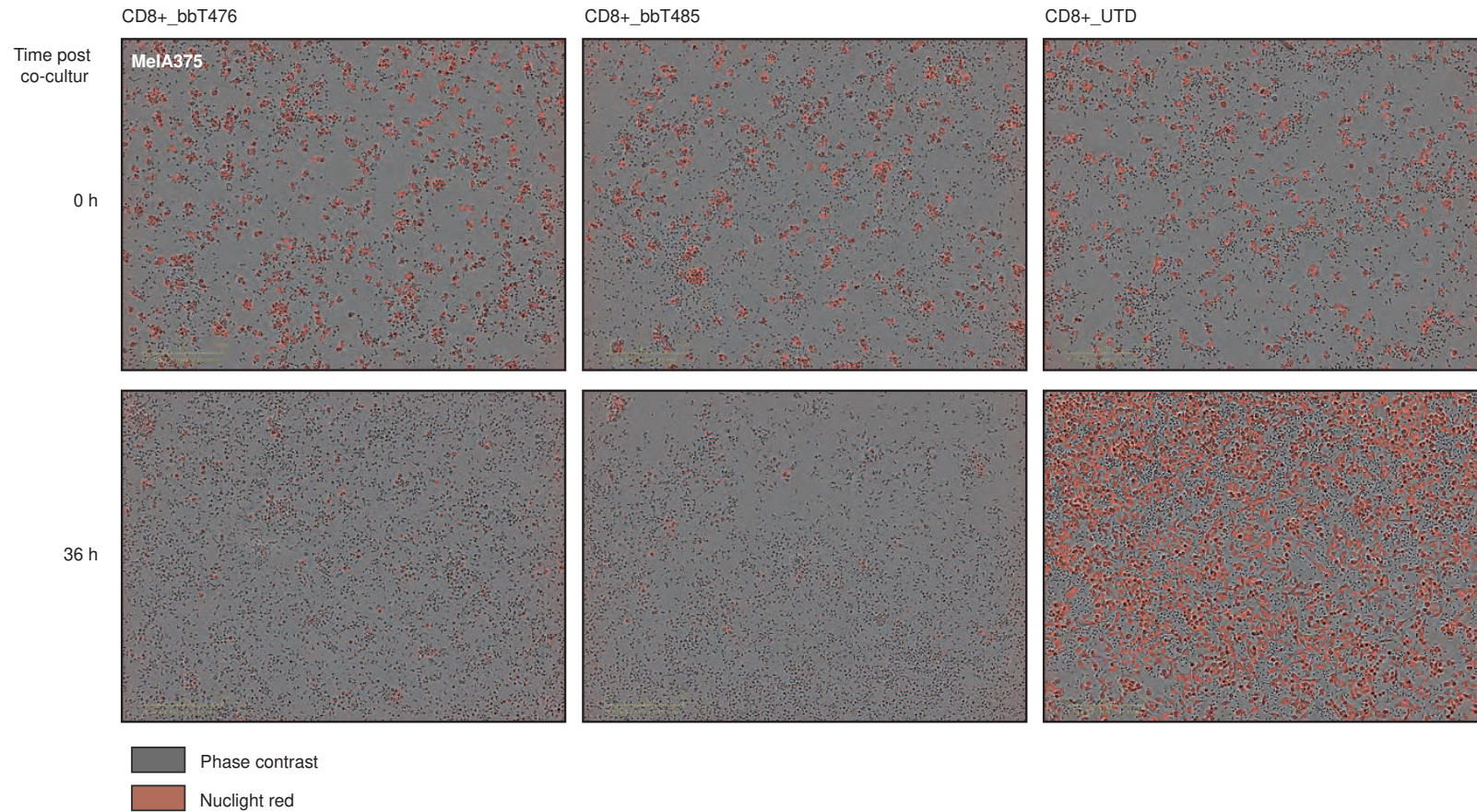
Figure S5: bbT476 and bbT485 TCR-Ts both lyse MelA375 cells

Incucyte-derived snapshots of MelA375 cells expressing NucLight Red live-cell labeling reagent (mKate) in co-culture with CD8+ T cells expressing transgenic TCRs (bbT476 or bbT485) or not (UTD = untransduced) immediately after assay set-up and 36 h later. Depicted is mKate-dependent fluorescence (red) and phase contrast (grey). Data shown are representative of 3 different donors for each tested TCR.

Supplementary table 1: List of LCLs with HLA-type

# LCL	HLA-A		HLA-B		HLA-C	
1	01:01:01G	02:07:01G	08:01:01G	27:04:01G	07:01:01G	12:02:01G
2	01:01:01G		52:01:01G		12:02:01G	
3	01:01:01G		57:01:01G		06:02:01G	
4	01:01:01G		35:02:01G		04:01:01G	
5	01:01:01G		37:01:01G		06:02:01G	
6	01:01:01G	02:01:01G	07:02:01G	39:01:01G	07:02:01G	
7	01:01:01G	02:08	08:01:01G	50:01:01G	06:02:01G	07:01:01G
8	01:01:01G	29:02:01G	44:03:01G	44:05:01	02:02:02G	16:01:01G
9	01:01:01G	24:02:01G	39:06:02G	58:01:01G	07:01:01G	07:02:01G
10	02:01:01G		45:01:01G		16:01:01G	
11	02:01:01G		40:01:01G		03:04:01G	
12	02:01:01G		44:02:01G		05:01:01G	
13	02:01:01G		13:02:01G		06:02:01G	
14	02:01:01G		18:01:01G		07:01:01G	
15	02:01:01G		44:02:01G		05:01:01G	
16	02:01:01G		15:01:01G		03:04:01G	
17	02:01:01G		08:01:01G	51:08:01	07:01:01G	16:02:01G
18	02:05:01G	23:01:01G	41:01:01	49:01:01G	07:01:01G	17:01:01G
19	02:17:01G		15:01:01G		03:03:01G	
20	02:02:01G	03:01:01G	35:01:01G	53:01:01G	04:01:01G	
21	26:01:01G	68:01:01G	18:01:01G	35:02:01G	04:01:01G	07:01:01G
22	02:04:01G		51:01:01G		15:02:01G	
23	02:05:01G	32:01:01G	40:01:01G	49:01:01G	03:04:01G	07:01:01G
24	02:05:01G	03:01:01G	41:02:01G	49:01:01G	04:01:01G	07:01:01G
25	02:06:01G	02:07:01G	46:01:01G		01:02:01G	08:01:01G
26	02:06:01G	24:02:01G	08:01:01G	35:03:01G	07:01:01G	07:02:01G
27	02:01:01G	02:07:01G	46:01:01G		01:02:01G	
28	02:01:01G	03:01:01G	35:02:01G	38:01:01	04:01:01G	12:03:01G
29	02:10:01G	30:01:01G	13:02:01G	40:06:01G	06:02:01G	08:01:01G
30	02:16:01G	03:01:01G	51:01:01G		07:04:01G	15:02:01G
31	02:17:01G		15:01:01G		03:03:01G	
32	03:01:01G		47:01:01G		06:02:01G	
33	03:01:01G		35:01:01G	35:03:01G	04:01:01G	
34	03:01:01G		35:01:01G	56:01:01G	01:02:01G	04:01:01G
35	03:01:01G	32:01:01G	07:02:01G	40:02:01G	02:02:02G	07:02:01G
36	03:01:01G	11:01:01G	40:01:01G		03:04:01G	
37	23:01:01G		14:01:01		08:02:01G	
38	24:02:01G		15:17:01G	51:01:01G	07:01:01G	15:04:01G
39	24:02:01G		35:08:01		04:01:01G	
40	24:02:01G	26:02:01	40:06:01G	51:01:01G	08:01:01G	14:02:01G
41	24:02:01G	29:01:01G	07:05:01G	27:02:01G	02:02:02G	15:05:01G
42	24:02:01:02L		55:01:01G		01:02:01G	
43	25:01:01G		51:01:01G		01:02:01G	
44	24:02:01G		40:01:01G		03:04:01G	
45	29:02:01G		40:02:01G		02:02:02G	
46	30:01:01G	68:02:01G	42:01:01		17:01:01G	
47	30:02:01G		18:01:01G		05:01:01G	
48	31:01:02G		15:01:01G		01:02:01G	
49	33:01:01G		14:02:01G		08:02:01G	
50	33:03:01G		44:03:01G		14:03	
51	68:02:01G		53:01:01G		04:01:01G	
52	02:06	26:01	35:01	38:01	04:01	12:03
53	02:06:01	24:02:01:01	07:02:01	51:03	14:02:01	07:02:01

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Supplementary table 2: HLA-type of primary human and induced pluripotent stem cell (iPSC)-derived cells

Cell type	HLA-A		HLA-B		HLA-C	
iCell Cardiomyocytes2	02:01	02:01	40:01	82:01	03:02	03:04
iCell Hepatocytes 2.0	02:01	02:01	40:01	82:01	03:02	03:04
iCell Astrocytes	02:01	02:01	40:01	82:01	03:02	03:04
iCell GABANeurons	02:01	02:01	40:01	82:01	03:02	03:04
iCell Endothelilal Cells	02:01	02:01	40:01	82:01	03:02	03:04
Normal Human Lung Fibroblasts	02:01:01G	11:01:01G	44:02:01G	55:01:01G	03:03:01G	05:01:01G
Normal Human Bronchial Epithelial cells	03:01:01G	23:01:01G	07:02:01G	14:02:01G	07:02:01G	08:02:01G
Human Renal Cortical Epithelial cells	02:01:01G	03:01:01G	15:01:01G	51:01:01G	02:02:02G	15:02:01G
Monocyte derived DCs	02:01:01G	31:01:02G	40:01:01G	51:01:01G	01:02:01G	03:04:01G