Supplementary Methods

Eligibility criteria

Eligible patients were > 18 years of age, had voluntarily signed a written informed consent form, pathologically confirmed advanced and/or metastatic solid tumor with recurrent/progressing and/or refractory disease, HLA-A*02:01 expression, MAGEA1-positive tumor as assessed by qPCR from a fresh biopsy, ECOG performance status of 0 to 1, adequate organ and marrow function (absolute neutrophil count ≥ 1.0 $x10^9$ /L without granulocyte colony-stimulating factor support, platelets ≥ 75,000 /µL, hemoglobin ≥ 8 g/dL), measurable disease according to RECIST 1.1, adequate hepatic function (bilirubin level ≤ 2.5x ULN, unless the patient has known Gilbert's syndrome, and alanine aminotransferase/aspartate aminotransferase ≤ 3x ULN or ≤ 5x ULN for patients with liver tumor lesions) and pulmonary function, life expectancy > 3 months, serum creatinine within 1.5x normal range for age or creatinine clearance with a recommended estimated glomerular filtration rate ≥ 50 mL/min/1.73m^2, acceptable coagulation status (international normalized ratio of prothrombin time of blood coagulation ≤ 2.0x ULN and partial thromboplastin time ≤ 2.0x ULN), received available standard-of-care treatments, recovered from any side effects of prior therapy to ≤ grade 1 and confirmed and adequate production timelines and capacities for the patient’s IMA202 product. HCC patients had a Child-Pugh score of < 6. Male patients had to agree to use effective contraception or be abstinent on trial for six months after IMA202 infusion. Female patients of childbearing potential had to use adequate contraception prior to trial entry until twelve months after IMA202 infusion.

Patients were excluded if they fulfilled one of the following criteria: history of other malignancies (except for adequately treated basal or SCC or carcinoma *in situ*) within the last three years, pregnancy or breastfeeding, prior allogenic stem-cell transplantation or solid-organ transplantation, being a liver transplant candidate, clinically detectable hepatic encephalopathy or hepatic encephalopathy requiring medication, any condition contraindicating leukapheresis, human immunodeficiency virus infection, active hepatitis B virus (HBV) and/or hepatitis C virus (HCV) infection, ongoing active anti-HCV treatment or detectable HBV or HCV viral load (except resolved, or chronic stable HBV infection), receipt of systemic corticosteroids (≥ 10 mg/day prednisone or equivalent) within two weeks prior to leukapheresis, concurrent severe and/or uncontrolled medical disease (*e.g.*, uncontrolled diabetes, severe infection requiring active treatment, severe malnutrition, chronic severe liver or renal disease), history of, or current immunodeficiency disease or prior treatment compromising immune function (inclusion at the discretion of the investigator), history of hypersensitivity (to CY, FLU, IL-2 or any rescue medication), solid tumor indications with low likelihood of MAGEA1 expression (colon adenocarcinoma, glioblastoma multiforme, kidney chromophobe, lower-grade glioma, mesothelioma, pancreatic adenocarcinoma, pheochromocytoma, paraganglioma, prostate adenocarcinoma, rectum adenocarcinoma, or thyroid carcinoma), active brain metastases (patients with a history of brain metastases are eligible if imaging studies performed ≥ 4 weeks following treatment indicate stable disease, the patient is asymptomatic, and steroid therapy was discontinued for ≥ 2 weeks). Furthermore, patients with a history of serious autoimmune diseases are excluded, such as active serious inflammatory bowel disease (including Crohn's disease and ulcerative colitis), and autoimmune disorders such as rheumatoid arthritis, multiple sclerosis,

systemic progressive sclerosis (scleroderma), systemic lupus erythematosus, or autoimmune vasculitis (e.g., Wegener's granulomatosis). Patients may be included if their disease is well controlled without the use of immunosuppressive agents. Other exclusion criteria were unfavorable cardiac conditions, such as uncontrolled hypertension (blood pressure > 140/100) despite optimal therapy, uncontrolled angina, ventricular arrhythmias, congestive heart failure (New York Heart Association Class II or above), baseline left ventricular ejection fraction ≤ 50%, prior or current cardiomyopathy, atrial fibrillation with heart rate > 100 beats per minute, unstable ischemic heart disease (myocardial infarction within six months prior to leukapheresis or angina requiring use of nitrates more than once weekly). Patients with active viral infection (e.g., coronavirus disease 2019 [COVID-19], influenza, severe acute respiratory syndrome [SARS], middle east respiratory syndrome) were excluded during initial screening, but screening may have started/continued after recovery. For patients with confirmed or suspected COVID-19 infection, screening or lymphodepletion may have started or continued at the earliest two weeks after full recovery or a negative SARS-CoV-2 test. For patients recovered from other viral infections, lymphodepletion may have started after full recovery. No patients which received any anti-tumor therapy, such as chemotherapy, surgery, palliative radiotherapy, tyrosine kinase inhibitors (e.g., erlotinib, gefitinib) and investigational therapies seven days prior to leukapheresis were included. The same was true for patients receiving i) chemotherapy, palliative radiotherapy, or investigational therapies within one week, ii) major surgery within two weeks, iii) extensive radiotherapy to the lung or liver within four months, iv) live vaccines within six weeks, or v) inactivated vaccines within two weeks prior to lymphodepletion. The existence of any other condition that would, in the investigator’s judgment, contraindicate the

patient's participation in the clinical trial because of safety concerns or compliance with clinical trial procedures, and concurrent participation in an interventional part of another clinical trial were also part of the exclusion criteria.

Supplementary Figure legends

Supplementary Figure 1: Distribution of MAGEA1 expression in samples of selected tumor entities from TCGA and resulting MAGEA1 prevalences and comparison with IMA202 patient target expression data and MAGEA1 expression in cell lines used for TCR characterization

MAGEA1 prevalence according to TCGA cohorts is indicated by fill color of histograms. Samples are considered MAGEA1-positive if MAGEA1 expression in the sample is above a mass spectrometry-guided threshold (red line). Target expression data in cell lines and IMA202 patients with observed tumor shrinkage (filled dots) or no observed tumor shrinkage (white dots) are indicated.

Abbreviations: FPKM, fragments per kilo base per million mapped reads; TCGA, The Cancer Genome Atlas.

Supplementary Figure 2: The ability to bind to pHLA multimer and to elicit cytokine response upon antigen exposure is a CD8 co-receptor dependent function for TCR-positive IMA202 cells

A) Detection of TCR expression by tetramer binding on $CD8^+$ and $CD8^-$ cells among total $CD3^+$ population shown in four different healthy donors. B, C) Cytokine response of $CD8^+$ and $CD8^-$ cells present in IMA202 products measured in response to various target-positive tumor cell lines by intracellular cytokine staining. B) Flow cytometry plots from three donors demonstrating TNF-α production by $CD8^+$ MAGEA1-positive TCR-positive IMA202 T cells and lack of cytokine secretion by $CD8^-$ IMA202 T cells six hours post co-culturing with U138

HLA-A*02-positive cell line engineered to overexpress MAGEA1. C) Comparison of IFN-γ production by CD8$^+$ and CD4$^+$ IMA202 T cells from four different products after 16 hours co-culturing with U2OS and UACC-257 (cell lines endogenously expressing MAGEA1), U138 HLA-A*02-positive MAGEA1-positive cell line and PMA-Ionomycin as a positive control for stimulation.

Abbreviation: NT, non-transduced.

Supplementary Figure 3: Presentation overview of MAGEA1 pHLA similar peptides in normal healthy tissues

Columns indicate HLA-A*02-positive normal healthy tissues analyzed by LC-MS/MS-based immunopeptidomics and each row indicates one of the similar peptides of MAGEA1 pHLA. Black tiles indicate that the peptide was identified via LC-MS/MS in at least one sample derived from the respective healthy normal tissue. White tiles indicate that the similar peptide was never detected in any sample derived from the respective tissue.

Supplementary Figure 4: MAGEA1 RNA expression for each treated patient

The graph depicts MAGEA1 RNA expression relative to the threshold for treated patients which either had progressive disease (PD; left plot) or stable disease (SD; right plot) as best overall response. Red dotted line indicates the defined threshold for which a reasonable HLA-presentation of the target peptide is likely.

Supplementary Figure 5: Frequency of MAGEA1-specific T cells in the final product

A) The ratio of overall CD8$^+$ and CD4$^+$ T cells within CD3$^+$ cells in final products was assessed by flow cytometry (n=16). B) The frequency of MAGEA1-specific (tetramer-positive CD8$^+$ T cells) within CD3$^+$CD8$^+$ T cells in final products was assessed by flow cytometry (n=16).

Supplementary Figure 6: Product characteristics

A) Representative dot plot for memory T cell subsets on MAGEA1-specific CD8$^+$ T cells (tetramer-positive CD8$^+$ T cells) of a patient's frozen product. Memory T cell subsets were classified using the markers CD197 (CCR7) and CD45RA (naïve: CCR7$^+$CD45RA$^+$; central memory [CM]: CCR7$^+$CD45RA; effector memory [TEM]: CCR7$^-$CD45RA; terminally differentiated effector memory [TEMRA]: CCR7$^-$CD45RA$^+$). B) Frozen products were analyzed for expression of the markers CD62L, CD28 and, CD27 on MAGEA1-specific CD8$^+$ T cells (tetramer-positive CD8$^+$ T cells) . Representative dot plots are shown. Gating was performed according to fluorescence minus one (FMO) control.

Supplementary Figure 7: Longitudinal phenotyping of peripheral blood T cells

A) Frequency of memory T cell subsets of MAGEA1-specific T cells (tetramer-positive CD8$^+$ T cells) in the final product (FP) and post-infusion. B) Frequency of CD28, CD27, CD62L, PD-1, CD45RO, CD57, TIM-3 and LAG-3 of MAGEA1-specific T cells (tetramer-positive CD8$^+$ T cells) in FP and post-infusion (n=16). C) Frequency of memory T cell subsets of non-specific CD8$^+$ T cells (tetramer-negative CD8$^+$ T cells) in FP and post-infusion. D) Frequency of CD28, CD27, CD62L, PD-1, CD45RO, CD57, TIM-3 and LAG-3 of non-specific CD8$^+$ T cells (tetramer-negative CD8$^+$ T cells) in FP and post-infusion. n=16. E) Comparison of percentage change of TEM, CD62L expression, and PD-1 expression in MAGEA1-specific CD8$^+$ T cells and non-specific CD8$^+$ T cells at week 1 after infusion. Statistical significance was assessed by Mann-Whitney test. Boxes represent the interquartile range including the

median value (line within boxes). Whiskers depict minimum and maximum values and dots represent individual analyzed patient samples.

Abbreviations: CM, central memory T cells; FP, final product; TEM, effector memory T cells; TEMRA, effector memory T cells expressing CD45RA.

Supplementary Figure 8: Persistence of MAGEA1-specific IMA202 T cells post-infusion

A) Frequency of MAGEA1-specific $CD8^+$ T cells (tetramer-positive $CD8^+$ T cells) within $CD8^+$ T cells, and B) MAGEA1-specific $CD8^+$ T cells within all $CD3^+$ T cells, at baseline and post-infusion assessed by flow cytometry. The frequency of MAGEA1-specific T cells was only analyzed up to day 179 post-infusion due to limited sample availability.

Supplementary Figure 9: Correlation of peak IMA202 vector copies or infused cell dose with maximum change in tumor size

Peak IMA202 cell expansion and infused number of specific cells was correlated with % maximum change in sum of diameters of the target lesions. Standard test for Spearman correlation was performed.

Supplementary Figure 10: Correlation of circulating and tumor-infiltrating MAGEA1-specific T cells

(A) Presence of MAGEA1-specific T cells in biopsy at day 42 is positively correlating with MAGEA1-specific T cells in PBMCs at day 28 or day 56, respectively as assessed by qPCR specific for Psi sequence of the lentiviral construct. (B) Proportion of MAGEA1-specific T cells in PBMC is significantly higher compared to MAGEA1-specific T cells in the biopsy (assessed for two different timepoints of PBMC collection).

Supplementary Figure 11: Serum cytokine levels in treated patients

Kinetics of cytokines IFN-γ (A), IL-6 (B), TNF-α (C), IL-10 (D), IL-8 (E), IL-18 (F) and GRO-alpha (G) as measured in patient serum taken on day of IMA202 infusion (day 0) and thereafter (day 1 to 28). Data is represented as geometric mean of $n \geq 12$ patients with standard error of mean. Cytokine values below the lower limit of quantification are considered as 1 for calculating the geometric mean. Y-axis was set in log-scale.

Abbreviations: IL, interleukin; GRO, growth-regulated protein.

Supplementary Tables

Supplementary Table 1: Overview of administered fludarabine, cyclophosphamide and IL-2 doses

Administered FLU doses ranged from 75 to 160 mg/m^2 BSA, CY doses from 1000 to 2000 mg/m^2 BSA and IL-2 doses from 8 x10^6 IU to 28 x10^6 IU. FLU and CY were applied from day -6 to day -3 before IMA202 infusion. IL-2 was administered subcutaneously approximately six hours after IMA202 infusion every twelve hours for 14 days. IL-2 was interrupted in nine patients.

Abbreviations: CY, cyclophosphamide; FLU, fludarabine; IL, interleukin; IU, international units.

Patient ID	Total FLU dose [mg/m^2]	Total CY dose [mg/m^2]	Total IL-2 dose [1x10^6 IU]
1	160	2000	28
2	160	2000	28
3	160	2000	28
4	75[1]	1200[1]	28
5	120[1]	2000	28
6	160	2000	25
7	160	2000	22
8	120[1]	1000[1]	26
9	120[2]	2000	14
10	160	2000	21
11	120[2]	1500[2]	15
12	120[2]	1500[2]	22
13	160	2000	28
14	80[3]	1000[3]	22
15	160	2000	8
16	80[3]	1000[3]	28

Dose reductions in the lymphodepletion regimen were mainly due to heavy pre-treatment[1], intercurrent adverse events[2] or indication (HCC)[3].

Supplementary Table 2: List of multimers and antibodies

Reagent	Distributor	Clone	Catalog Number	Lot
Alexa Fluor 488 CD8a	BioLegend	RPT-T8	301021	B201763 / B230874 / B284960
PerCP-Cy5.5 CD3	BioLegend	HIT3a	300328	B208442 / B338476
Streptavidin PE (Multi-mer)	Life Technology	-	S866	1865801 / 1973501
PE-Dazzle 594 CD27	BioLegend	M-T271	356422	B220331
Streptavidin PE-Cy7 (Multi-mer)	BioLegend	-	405206	B260327 / B287247 / B300682 / B335089
APC CD57	BioLegend	HNK.1	359610	B210684 / B340445
APC-Fire 750 CD45RA	BioLegend	HI100	304152	B218733
BV421 CCR7	BioLegend	G043H7	353208	B229001
BV510 FVS	BD	-	564406	7138874 / 8197539 / 9176704 / 0345532
BV510 CD19	BioLegend	H1B1-9	302242	B221987 / B239285 / B339100
BV605 CD45RO	BioLegend	UCHL1	304238	B246176 / B250227
BV785 CD62L	BioLegend	DREG-56	304830	B235783 / B277488
BV650 CD28	BioLegend	37.51	302946	B229718 / B329064
Alexa Fluor 488 TIM-3	R&D Systems	344823	FAB2365G	ABVG0415021 / ABVG0621091
APC LAG-3	eBioscience	3DS2223H	17-2239-42	4307377 / 2305268
Alexa Fluor 700 CD3	BioLegend	HIT3a	300324	B218705 / B279655
APC-Fire 750 CD45RA	BioLegend	HI100	304152	B218733

Reagent	Distributor	Clone	Catalog Number	Lot
BV421 CD8a	BioLegend	RPA-T8	301036	B242747 / B314490
BV650 PD-1	BioLegend	EH12	329950	B239981

Supplementary Table 3: Summary of included HLA alleles during the characterization of the IMA202 TCR

Summary of genotypes covered during IMA202 TCR characterization. None of the MAGEA1-negative tumor cell lines tested induced activation of IMA202-positive T cells, indicating not only absence of off-target reactivity in the context of HLA-A*02:01, but also the absence of allogenic off-target cross-recognition of peptides presented in the context of HLA isotypes different from HLA-A*02:01.

Included alleles during TCR characterization	HLA isotype					
	HLA-A	HLA-B	HLA-C	HLA-DP	HLA-DQ	HLA-DR
	01:01	07:02	02:02	DPA1*01:03	DQA1*01:02	DRB1*03:01
	02:01	08:01	03:03	DPA1*02:01	DQA1*01:03	DRB1*04:01
	02:05	15:01	03:04	DPB1*02:01	DQA1*01:05	DRB1*04:04
	02:06	18:10	04:01	DPB1*04:01	DQA1*02:01	DRB1*07:01
	02:11	27:05	05:01	DPB1*04:01	DQA1*03:01	DRB1*08:02
	03:01	35:01	06:02	DPB1*04:02	DQA1*03:19	DRB1*08:04
	23°	35:12	07:01	DPB1*04:02	DQA1*04:02	DRB1*10:01
	29:02	37:01	07:02	DPB1*11:01	DQA1*05:01	DRB1*11:01
	31:01	40:01	15:02		DQA1*05:05	DRB1*12:01
	68°	40:02	16:01		DQA1*05:09	DRB1*13:01
		40:06	17:01		DQB1*02:01	DRB1*15:01
		42:01			DQB1*02:02	DRB3*01:01
		44°			DQB1*03:01	DRB3*02:02
		44:02			DQB1*03:02	DRB4*01:01
		44:03			DQB1*06:01	DRB4*01:03
		50:01			DQB1*06:02	DRB5*01:01
		51:01			DQB1*06:03	

Summarized genotyping information for tumor cell lines (1) and primary cells that were included in the IMA202 TCR characterization. ° = no high resolution typing information available.

Supplementary Table 4: MAGEA1 and HLA-A*02 expression in cell lines used for TCR characterization

RNAseq-based expression data for the exon encoding the MAGEA1 target peptide (ENSE00001452168) and for the HLA-A gene (ENSG00000206503) are shown.

Cell line	MAGEA1 expression	HLA-A expression
UACC-257	25 FPKM	81 FPKM
U266B1	77 FPKM	154 FPKM
U2OS	46 FPKM	44 FPKM

Supplementary Table 5: $CD8^+/CD4^+$ T cell ratio and frequency of MAGEA1-specific $CD8^+$ T cells for each treated patients

The ratio of overall $CD8^+$ and $CD4^+$ T cells within $CD3^+$ cells in final products was assessed by flow cytometry. The frequency of MAGEA1-specific (tetramer-positive $CD8^+$ T cells) within $CD3^+CD8^+$ T cells in final products was assessed by flow cytometry.

Patient ID	$CD8^+/CD4^+$ T cell ratio	MAGEA1-specific $CD8^+$ T cells
1	N/A	22.5
2	0.85	40.9
3	0.33	18.0
4	1.33	11.7
5	0.60	44.2
6	0.34	43.6
7	0.27	30.5
8	N/A	41.5
9	0.82	37.9
10	0.51	42.2
11	0.36	46.0
12	0.38	42.5
13	1.49	44.2
14	1.73	70.7
15	0.92	72.8
16	1.31	46.7

Supplementary Table 6: Timepoint of peak expansion of infused MAGEA1-specific T cells for each treated patient

MAGEA1-specific T cells were assessed in DNA samples from patients' PBMC using qPCR. T_{max} refers to the timepoint (days) when the maximum number of T cells was detected in a given sample of a patient.

Patient ID	T_{max} [days]
1	3
2	1
3	3
4	1
5	3
6	3
7	7
8	1
9	7
10	1
11	1
12	1
13	3
14	3
15	7
16	7
Median (Min, Max)	**3 (1, 7)**

Supplementary References

1. Scholtalbers J, Boegel S, Bukur T, Byl M, Goerges S, Sorn P, et al. TCLP: an online cancer cell line catalogue integrating HLA type, predicted neo-epitopes, virus and gene expression. Genome Med. 2015;7:118.