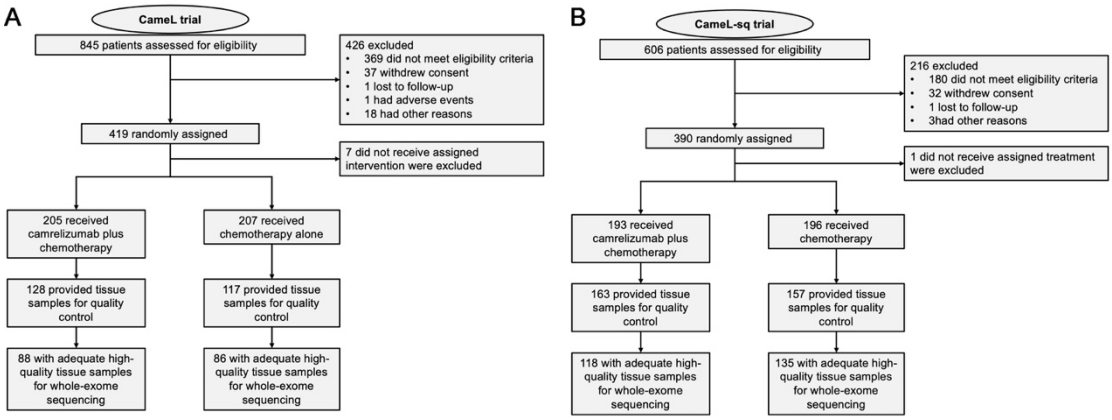
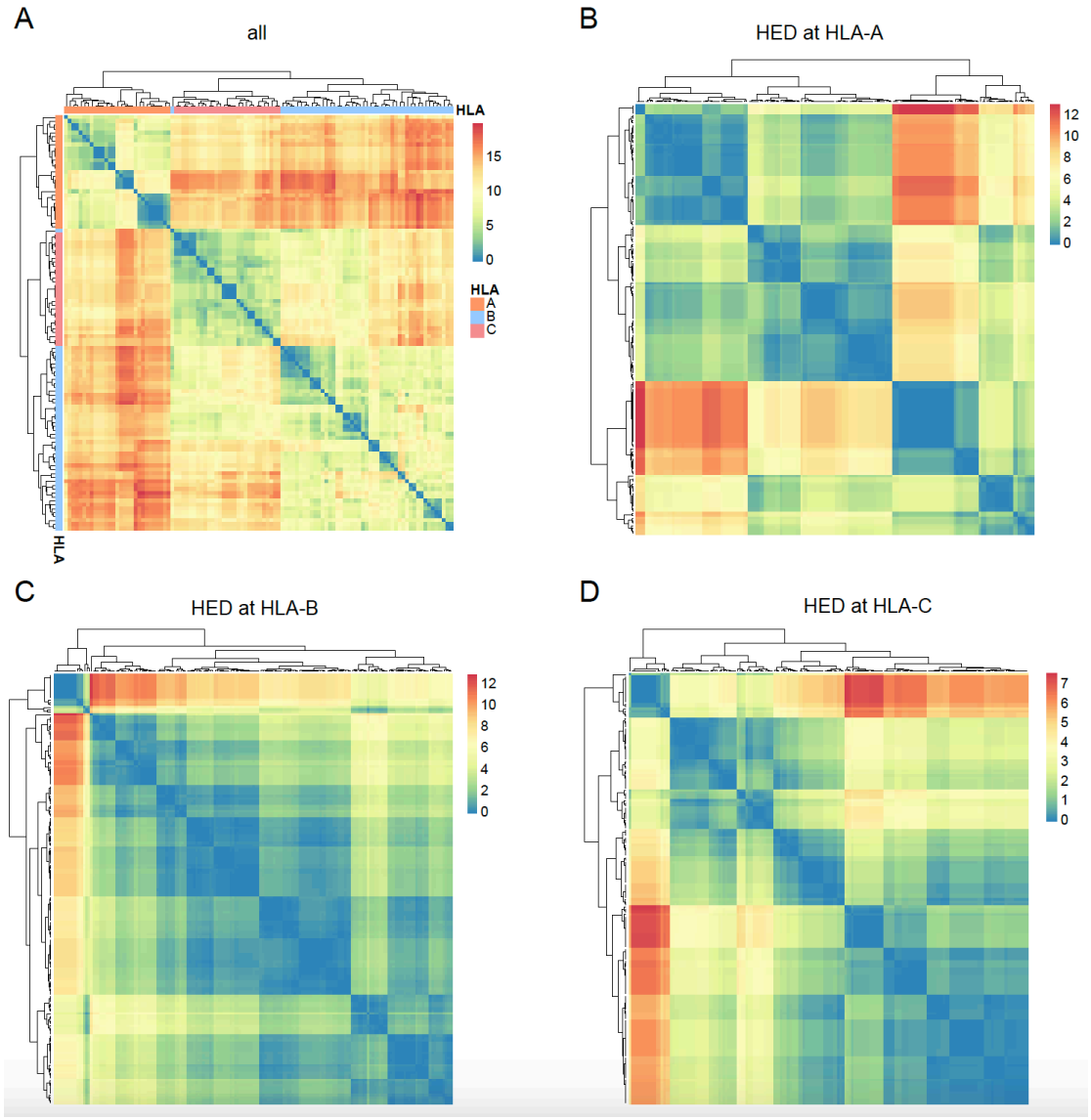
Supplementary Materials

Supplementary Figure S1. Flowchart of patients' selection



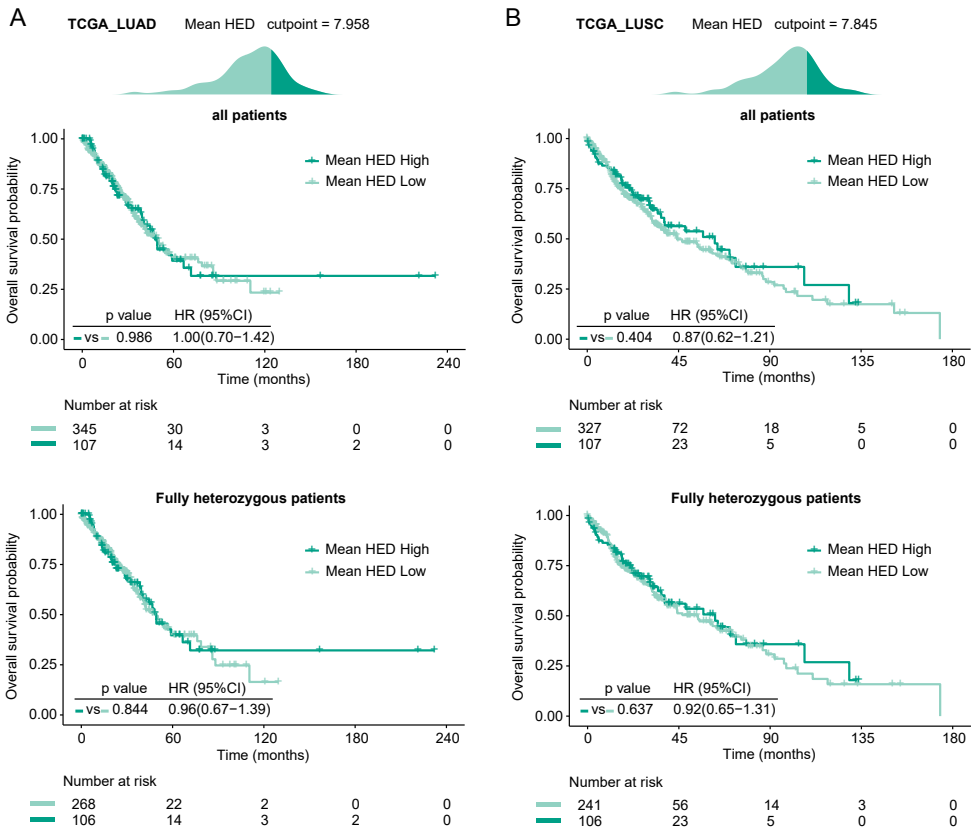
A, Flowchart of patients' selection in the CameL trial. **B,** Flowchart of patients' selection in the CameL-sq trial. mIF: multiplex immunofluorescence.

Supplementary Figure S2. Hierarchical clustering of HED.



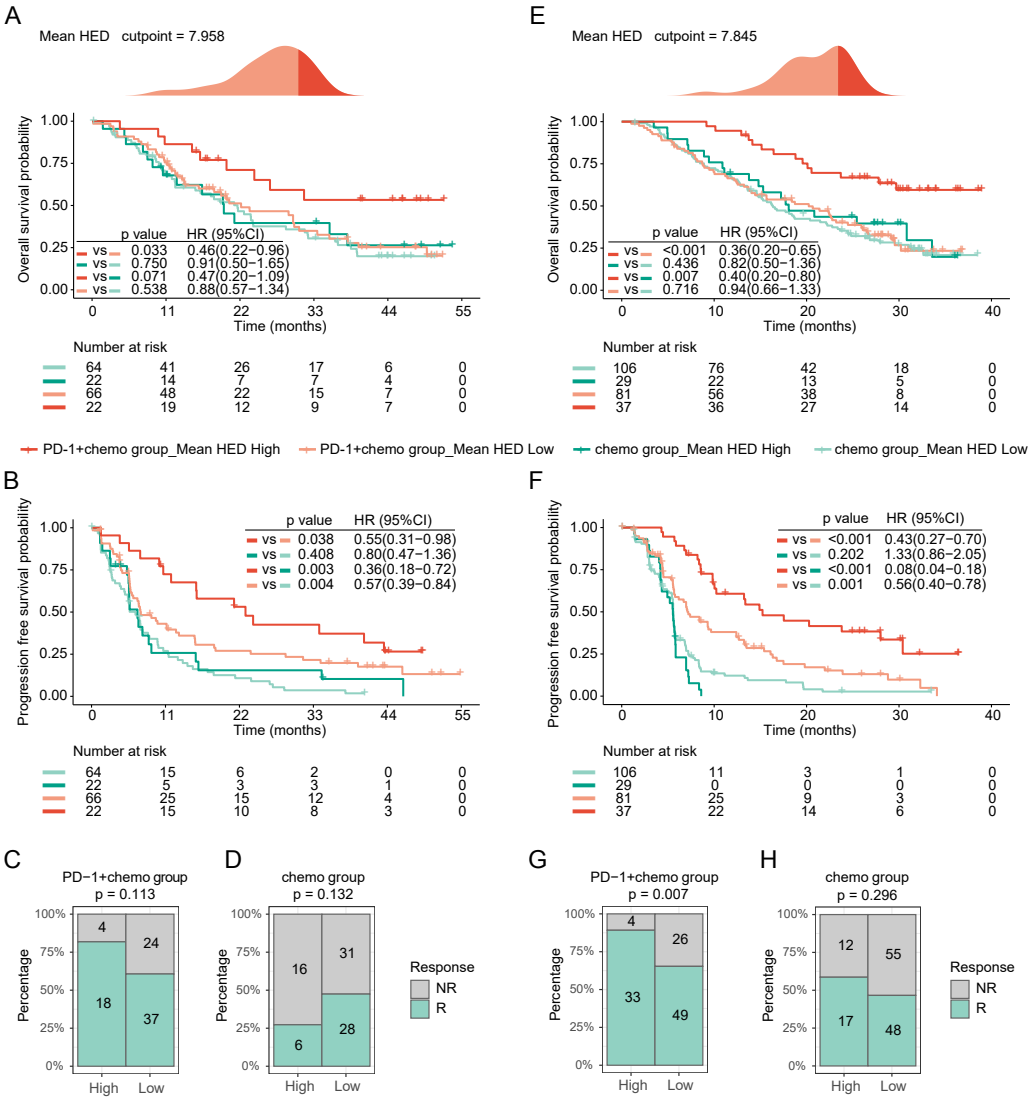
A, Hierarchical clustering of HED at individual HLA-I loci in CameL trial. **B,** Hierarchical clustering of HED at HLA-A using all HLA-A alleles from all patient of CameL trial. **C,** Hierarchical clustering of HED at HLA-B using all HLA-B alleles from all patient of CameL trial. **D,** Hierarchical clustering of HED at HLA-C using all HLA-C alleles from all patient of CameL trial. The heatmap shows z score-normalized HED across all alleles in all patient of CameL trial. The color gradient of blue to red indicates low HED between allele pairs to high HED between allele pairs, respectively.

Supplementary Figure S3. Association of HED with prognosis in TCGA lung cancer patients.



A, HED (upper left) and full heterozygosity at HLA-I (bottom left) is not associated with prognosis in TCGA lung adenocarcinoma. **B,** HED (upper right) and full heterozygosity at HLA-I (bottom right) is not associated with prognosis in TCGA lung squamous cell carcinoma.

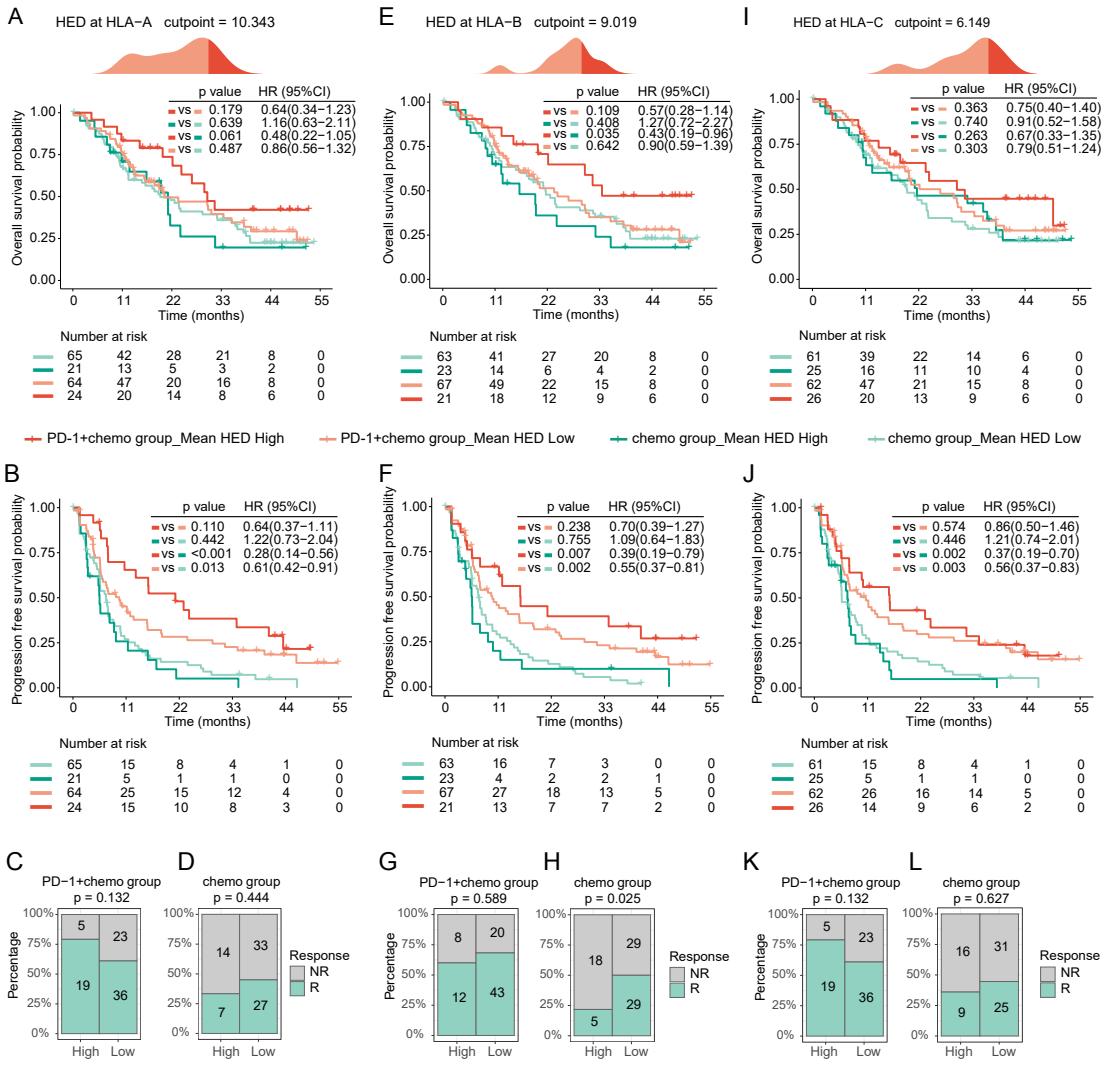
Supplementary Figure S4. HED^{high} predicts outcomes of PD-1 blockade plus chemotherapy in all patients.



A, Association of HED^{high} with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). Density plots show the distribution and cut point (top quartile) for mean HED used in Kaplan-Meier analysis. **B,** Association of HED^{high} with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). **V,** Comparison of response rates between HED^{high} and HED^{low} groups in PD-1 blockade plus chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). **D,** Comparison of response rates between HED^{high} and HED^{low} groups in chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). **E,** Association of HED^{high} with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL-sq trial of all patients (two-sided log-rank test). Density plots show the distribution and cut point (top quartile) for

mean HED used in Kaplan-Meier analysis. **F,** Association of HED^{high} with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL-sq trial of all patients (two-sided log-rank test). **G,** Comparison of response rates between HED^{high} and HED^{low} groups in PD-1 blockade plus chemotherapy arm in CameL-sq trial of all patients (two-sided Fisher's exact test). **H,** Comparison of response rates between HED^{high} and HED^{low} groups in chemotherapy arm in CameL-sq trial of all patients (two-sided Fisher's exact test). HR, hazard ratio; CI, confidence interval; PD-1+chemo, PD-1 blockade plus chemotherapy; Chemo, chemotherapy; TPS, tumor proportional score; TMB, tumor mutational burden. R, response, included patients with complete and partial response per RECIST v1.1; NR, not response included patients with stable disease and disease progression per RECIST v1.1.

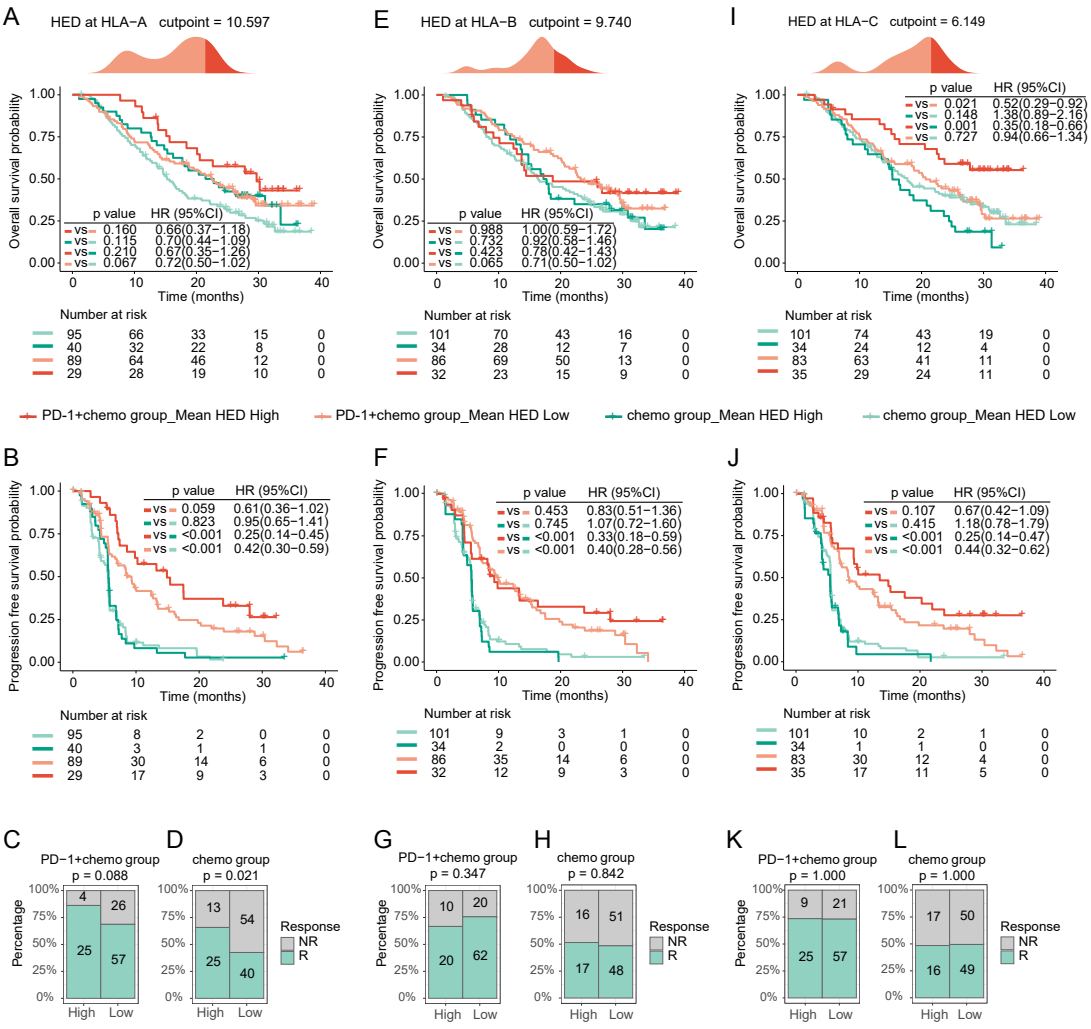
Supplementary Figure S5. Association of HED at each HLA class I locus with treatment response and outcomes in CameL trial.



A, Association of HLA-A HED^{high} with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). Density plots show the distribution and cut point (top quartile) for HLA-A mean HED used in Kaplan-Meier analysis. **B,** Association of HLA-A HED^{high} with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). **C,** Comparison of response rates between HLA-A HED^{high} and HED^{low} groups in PD-1 blockade plus chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). **D,** Comparison of response rates between HLA-A HED^{high} and HED^{low} groups in chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). **E,** Association of HLA-B HED^{high} with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided

log-rank test). Density plots show the distribution and cut point (top quartile) for HLA-B mean HED used in Kaplan-Meier analysis. **F,** Association of HLA-B HED^{high} with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). **G,** Comparison of response rates between HLA-B HED^{high} and HED^{low} groups in PD-1 blockade plus chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). **H,** Comparison of response rates between HLA-B HED^{high} and HED^{low} groups in chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). **I,** Association of HLA-C HED^{high} with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). Density plots show the distribution and cut point (top quartile) for HLA-C mean HED used in Kaplan-Meier analysis. **J,** Association of HLA-C HED^{high} with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). **K,** Comparison of response rates between HLA-C HED^{high} and HED^{low} groups in PD-1 blockade plus chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). **L,** Comparison of response rates between HLA-C HED^{high} and HED^{low} groups in chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). HR, hazard ratio; CI, confidence interval; PD-1+chemo, PD-1 blockade plus chemotherapy; Chemo, chemotherapy; TPS, tumor proportional score; TMB, tumor mutational burden. R, response, included patients with complete and partial response per RECIST v1.1; NR, not response included patients with stable disease and disease progression per RECIST v1.1.

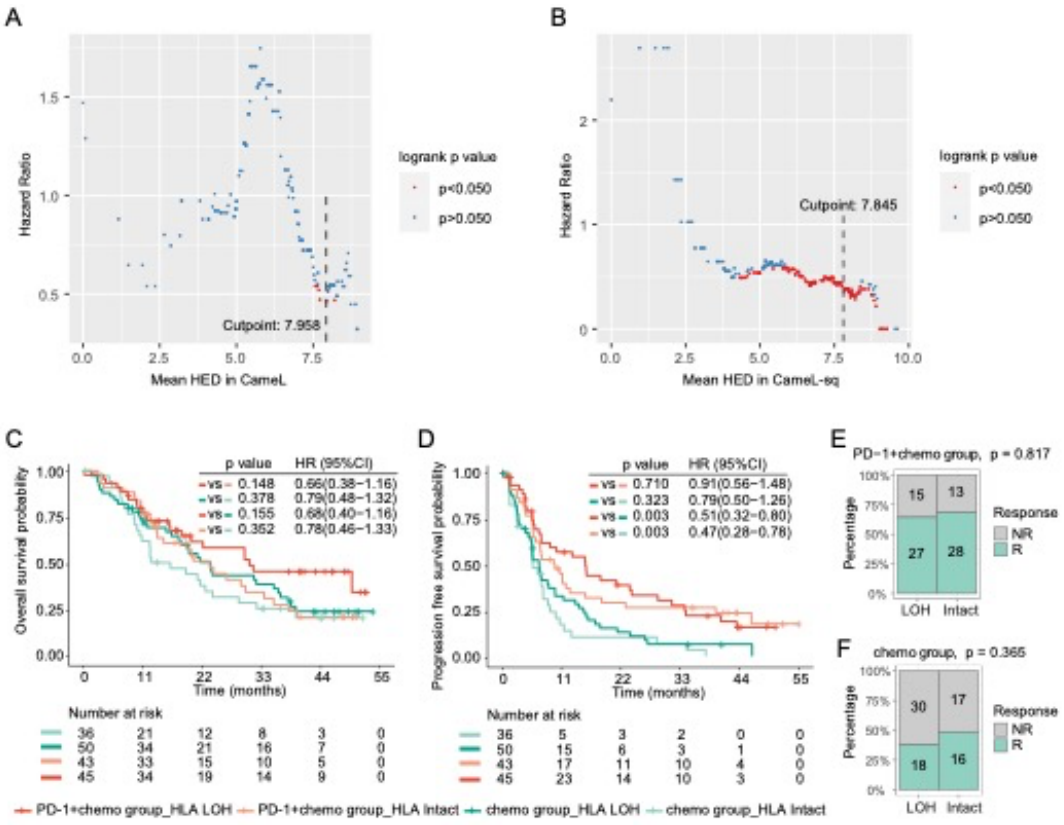
Supplementary Figure S6. Association of HED at each HLA class I locus with treatment response and outcomes in CameL-sq trial.



A, Association of HLA-A HED^{high} with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL-sq trial of all patients (two-sided log-rank test). Density plots show the distribution and cut point (top quartile) for HLA-A mean HED used in Kaplan-Meier analysis. **B,** Association of HLA-A HED^{high} with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL-sq trial of all patients (two-sided log-rank test). **C,** Comparison of response rates between HLA-A HED^{high} and HED^{low} groups in PD-1 blockade plus chemotherapy arm in CameL-sq trial of all patients (two-sided Fisher's exact test). **D,** Comparison of response rates between HLA-A HED^{high} and HED^{low} groups in chemotherapy arm in CameL-sq trial of all patients (two-sided Fisher's exact test). **E,** Association of HLA-B HED^{high} with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL-sq trial of all patients (two-sided log-rank test). Density plots show the distribution and cut point (top

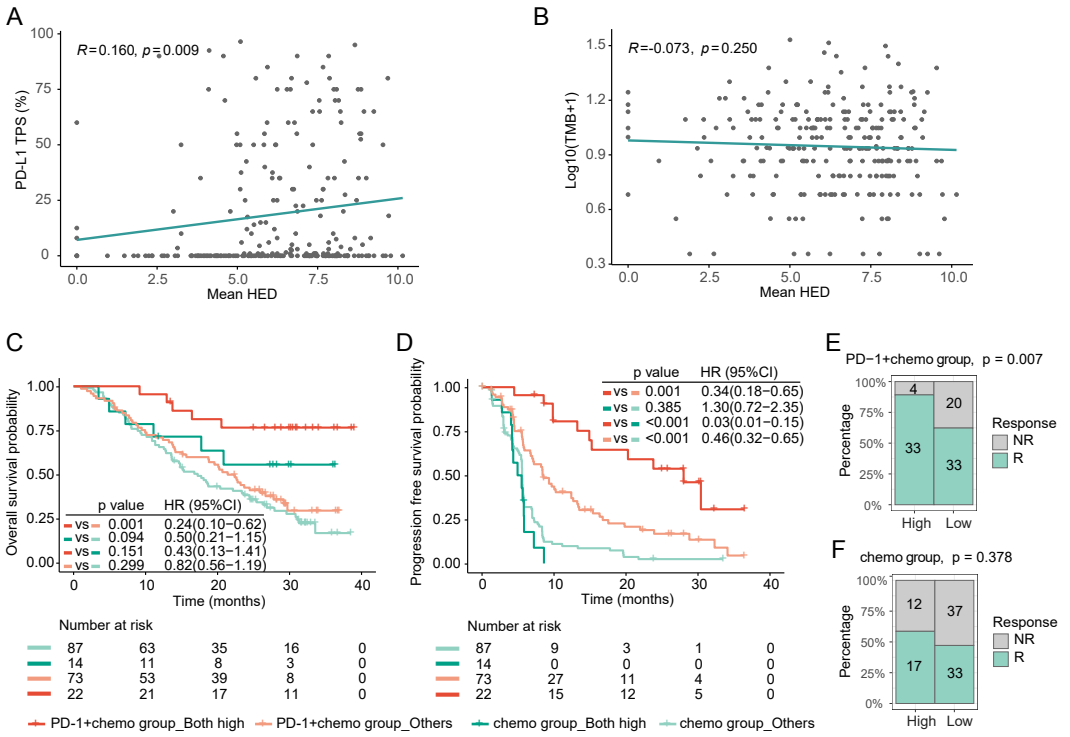
quartile) for HLA-B mean HED used in Kaplan-Meier analysis. F**f,** Association of HLA-B HED^{high} with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL-sq trial of all patients (two-sided log-rank test). **G,** Comparison of response rates between HLA-B HED^{high} and HED^{low} groups in PD-1 blockade plus chemotherapy arm in CameL-sq trial of all patients (two-sided Fisher's exact test). **H,** Comparison of response rates between HLA-B HED^{high} and HED^{low} groups in chemotherapy arm in CameL-sq trial of all patients (two-sided Fisher's exact test). I**i,** Association of HLA-C HED^{high} with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL-sq trial of all patients (two-sided log-rank test). Density plots show the distribution and cut point (top quartile) for HLA-C mean HED used in Kaplan-Meier analysis. **J,** Association of HLA-C HED^{high} with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL-sq trial of all patients (two-sided log-rank test). **K,** Comparison of response rates between HLA-C HED^{high} and HED^{low} groups in PD-1 blockade plus chemotherapy arm in CameL-sq trial of all patients (two-sided Fisher's exact test). **L,** Comparison of response rates between HLA-C HED^{high} and HED^{low} groups in chemotherapy arm in CameL-sq trial of all patients (two-sided Fisher's exact test). HR, hazard ratio; CI, confidence interval; PD-1+chemo, PD-1 blockade plus chemotherapy; Chemo, chemotherapy; TPS, tumor proportional score; TMB, tumor mutational burden. R, response, included patients with complete and partial response per RECIST v1.1; NR, not response included patients with stable disease and disease progression per RECIST v1.1.

Supplementary Figure S7. Effect of mean HED on HR from PFS across all possible cut points and association of HLA-I LOH with survival in all patients.



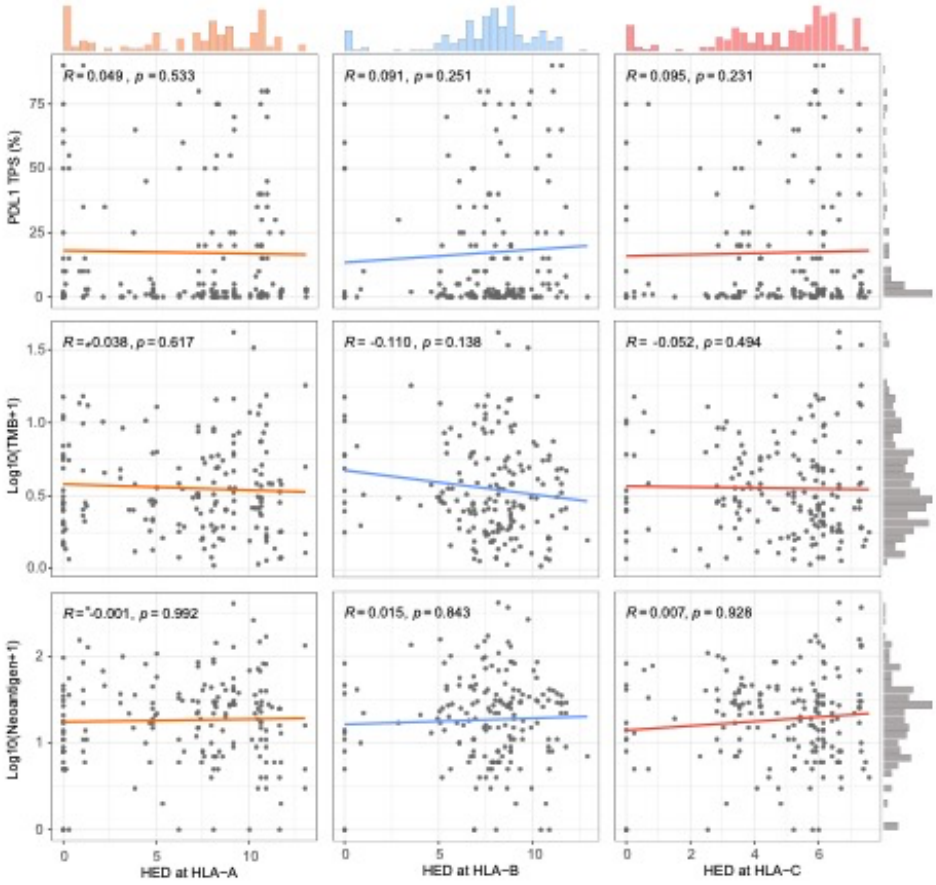
A, Effect of mean HED on HR from PFS across all possible cut points in CameL trial. **B,** Effect of mean HED on HR from PFS across all possible cut points in CameL-sq trial. **C,** Association of HLA-I LOH with OS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). **D,** Association of HLA-I LOH with PFS after PD-1 blockade plus chemotherapy or chemotherapy in CameL trial of all patients (two-sided log-rank test). **E,** Comparison of response rates between patients with and without HLA-I LOH in PD-1 blockade plus chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). **D,** Comparison of response rates between patients with and without HLA-I LOH in chemotherapy arm in CameL trial of all patients (two-sided Fisher's exact test). HR, hazard ratio; CI, confidence interval; HLA LOH, HLA class I loss of heterozygosity; PD-1+chemo, PD-1 blockade plus chemotherapy; Chemo, chemotherapy; R, response, included patients with complete and partial response per RECIST v1.1; NR, not response included patients with stable disease and disease progression per RECIST v1.1.

Supplementary Figure S8. Combination of mean HED and PD-L1 expression showed better predictive performance in CameL-sq trial.



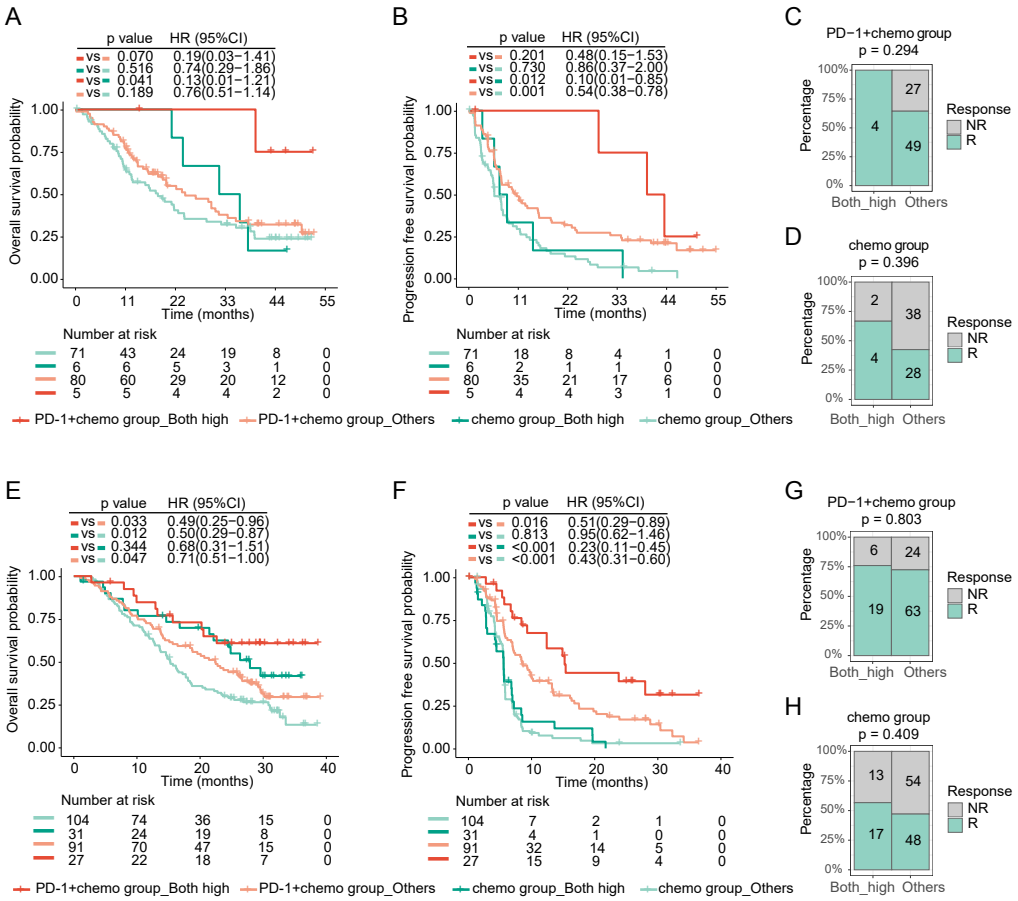
A, Correlation of mean HED with PD-L1 TPS (*P* = 0.157; two-sided Spearman's correlation). **B,** Correlation of mean HED with TMB (*P* = 0.229; two-sided Kendall's rank correlation). **C,** OS comparisons between patients with both high and others after PD-1 blockade plus chemotherapy or chemotherapy among fully heterozygous patients (log-rank test). When combining mean HED and PD-L1 expression level, patients with both hed^{high} and positive PD-L1 expression were defined as the 'Both high' group, while the other patients belonged to the 'Others' group. **D,** PFS comparisons between patients with both high and others after PD-1 blockade plus chemotherapy or chemotherapy among fully heterozygous patients (log-rank test). **E,** Response rates comparison between patients with both high and others after PD-1 blockade plus chemotherapy among fully heterozygous patients (two-sided Fisher's exact test test). **F,** Response rates comparison between patients with both high and others after chemotherapy among fully heterozygous patients (two-sided Fisher's exact test test). HR, hazard ratio; CI, confidence interval; TPS, tumor proportional score; TMB, tumor mutational burden; PD-1+chemo, PD-1 blockade plus chemotherapy; Chemo, chemotherapy; R, response, included patients with complete and partial response per RECIST v1.1; NR, not response included patients with stable disease and disease progression per RECIST v1.1.

Supplementary Figure S9. Association of HED at each HLA class I locus with PD-L1 expression level, TMB and neoantigen burden.



There was no any correlation between mean HED at each HLA-I locus and PD-L1 expression level, TMB or neoantigen burden.

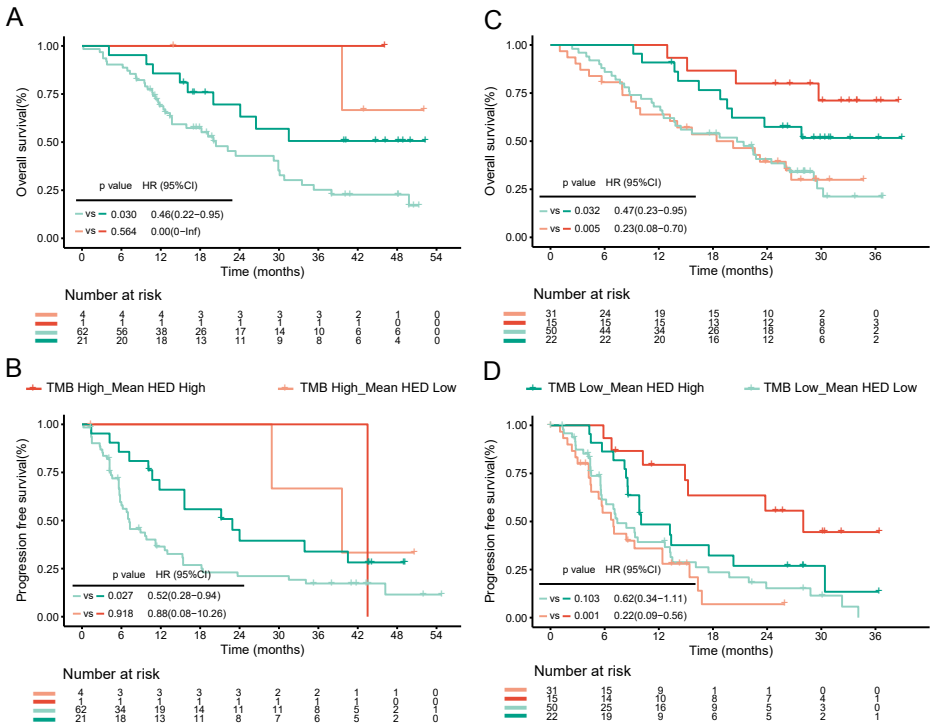
Supplementary Figure S10. Predictive value of joint utility of mean HED and TMB.



A, OS comparisons between patients with both high and others after PD-1 blockade plus chemotherapy or chemotherapy among fully heterozygous patients of CameL trial (log-rank test). When combining mean HED and TMB level, patients with both hed^{high} and high TMB level (≥10 Muts/Mb) were defined as the 'Both high' group, while the other patients belonged to the 'Others' group. **B,** PFS comparisons between patients with both high and others after PD-1 blockade plus chemotherapy or chemotherapy among fully heterozygous patients of CameL trial (log-rank test). **C,** Response rates comparison between patients with both high and others after PD-1 blockade plus chemotherapy among fully heterozygous patients of CameL trial (two-sided Fisher's exact test test). **D,** Response rates comparison between patients with both high and others after chemotherapy among fully heterozygous patients of CameL trial (two-sided Fisher's exact test test). **E,** OS comparisons between patients with both high and others after PD-1 blockade plus chemotherapy or chemotherapy among fully heterozygous patients of CameL-sq trial (log-rank test). **F,** PFS comparisons between patients with both high and others after PD-1 blockade plus chemotherapy or chemotherapy among fully heterozygous patients of

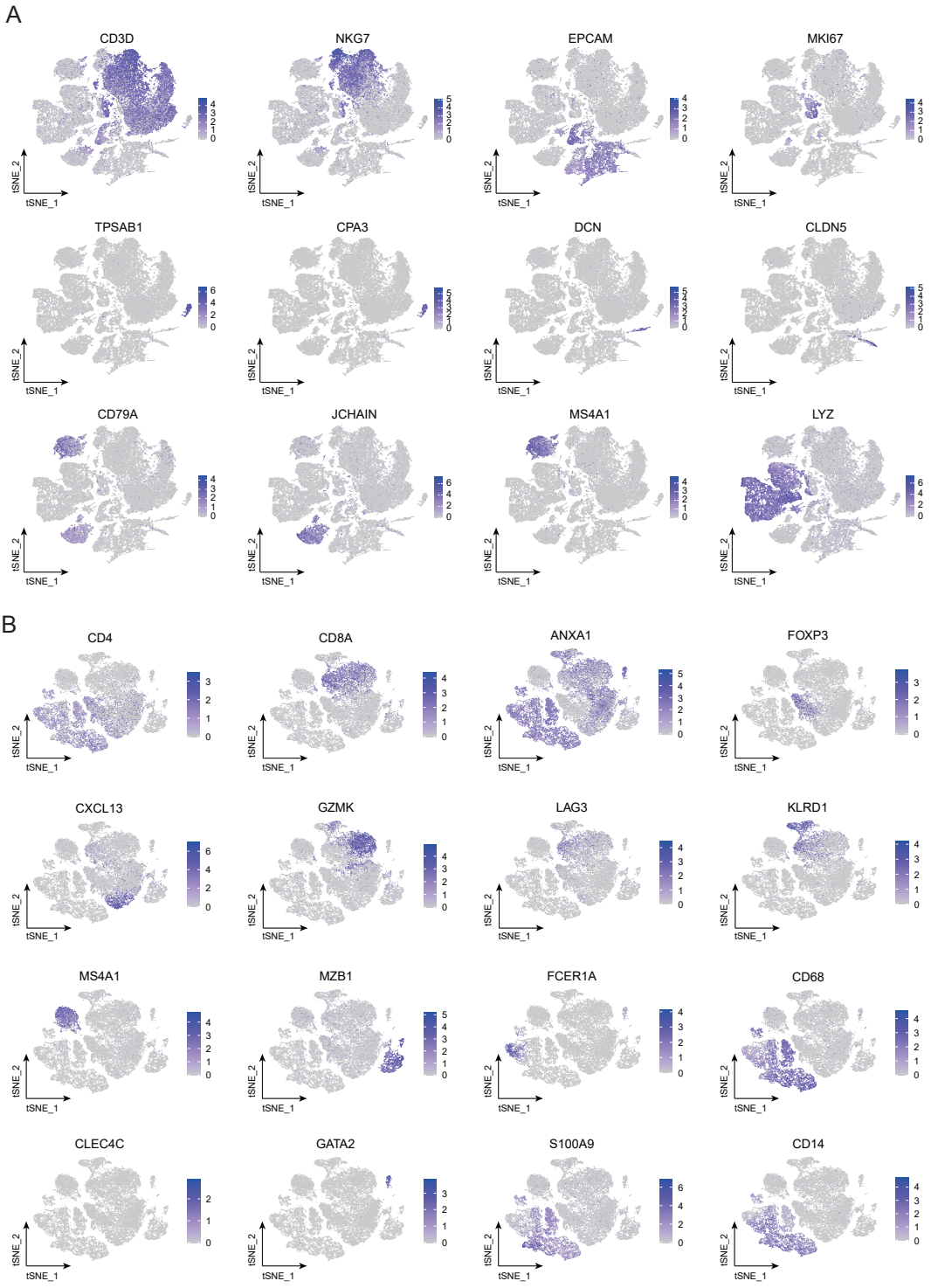
CameL-sq trial (log-rank test). **G,** Response rates comparison between patients with both high and others after PD-1 blockade plus chemotherapy among fully heterozygous patients of CameL-sq trial (two-sided Fisher's exact test test). **H,** Response rates comparison between patients with both high and others after chemotherapy among fully heterozygous patients of CameL-sq trial (two-sided Fisher's exact test test). HR, hazard ratio; CI, confidence interval; TMB, tumor mutational burden; PD-1+chemo, PD-1 blockade plus chemotherapy; Chemo, chemotherapy; R, response, included patients with complete and partial response per RECIST v1.1; NR, not response included patients with stable disease and disease progression per RECIST v1.1.

Supplementary Figure S11. Predictive value of HED^{high} in patients treated with PD-1 blockade plus chemotherapy according to TMB level.



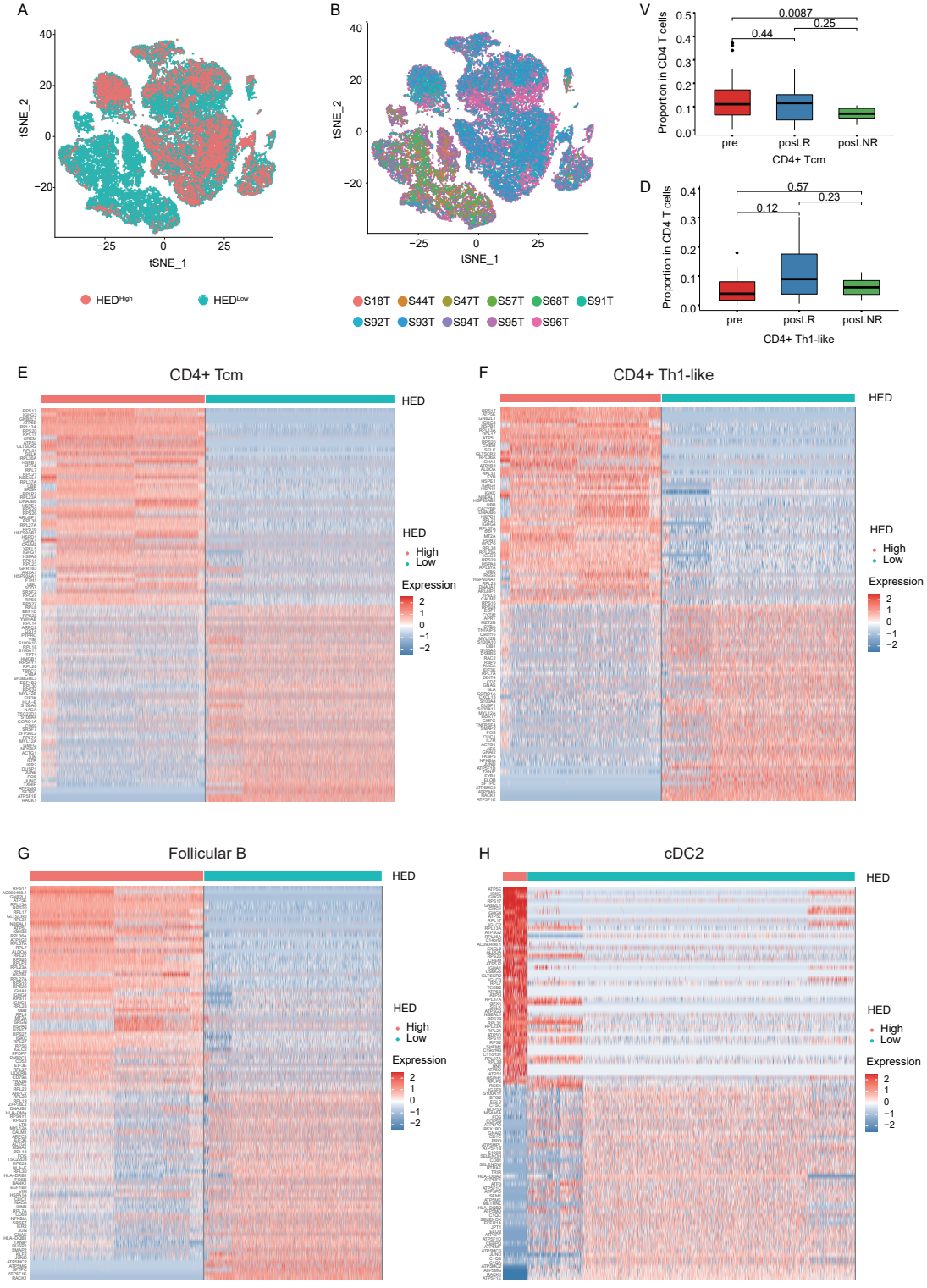
A, OS comparisons between patients with distinct TMB level and HED after PD-1 blockade plus chemotherapy in CameL study. **B,** PFS comparisons between patients with distinct TMB level and HED after PD-1 blockade plus chemotherapy in CameL study. **C,** OS comparisons between patients with distinct TMB level and HED after PD-1 blockade plus chemotherapy in CameL-sq study. **D,** PFS comparisons between patients with distinct TMB level and HED after PD-1 blockade plus chemotherapy in CameL-sq study.

Supplementary Figure S12. Identification of major cell types in patients with untreated NSCLC.



A, t-SNE plots show the expression levels of cell-type marker genes in total cells from 11 patients. **B,** t-SNE plots show the expression levels of cell-type marker genes in immune cells from 11 patients. tSNE, t-distributed Stochastic Neighborhood Embedding.

Supplementary Figure S13. Characterization of immune cell types in patients with untreated NSCLC.



A-B, t-SNE plots show 40,400 immune cells from 11 NSCLC patients. Points are color-coded by HED (**A**) and included patients (**B**). **C-D,** Boxplot shows the proportion of CD4+ Tcm (**C**) and

CD4+ Th1-like (**D**) in CD4 T cells at pre- and post-treatment (post.R represents responsive samples; post.NR represents non-responsive samples). *P*-values are calculated using two-sided student t-test. **E-H,** Heatmaps show the expression of top 100 differentially expressed gene with |log2fold Change| > 0.5 and adjusted *P* value < 0.01, selected from CD4+ Tcm (**E**), CD4+ Th1-like (**F**), Follicular B (**G**) and cdc2 (**H**) between hed^{high} and hed^{low} samples, respectively. tSNE, t-distributed Stochastic Neighborhood Embedding.

Supplementary Table S1: Baseline characteristics of included patients from the biomarker evaluable population and intention-to-treat cohort in CameL study.

	Biomarker evaluable trial		Intention-to-treat population		
Items	**Camrelizumb plus chemotherapy**	**Chemotherapy alone**	**Camrelizumb plus chemotherapy**	**Chemotherapy alone**	***P* value**
	(n=88)	**(n=86)**	**(n=205)**	**(n=207)**	
Age					
≥65 years	12 (14%)	28 (33%)	45 (22%)	53 (26%)	0.835
<65 years	76 (86%)	58 (67%)	160 (78%)	154 (74%)	
Sex					
Male	61 (69%)	68 (79%)	146 (71%)	149 (72%)	0.531
Female	27 (31%)	18 (21%)	59 (29%)	58 (28%)	
Smoking history					
≥400 cigarette-years	62 (70%)	57 (66%)	127 (62%)	130 (63%)	0.166
<400 cigarette-years or never	26 (30%)	29 (34%)	78 (38%)	77 (37%)	
ECOG performance status					
0	24 (27%)	17 (20%)	48 (23%)	36 (17%)	0.391
1	64 (73%)	69 (80%)	157 (77%)	171 (83%)	
Disease stage					
IIIB/IIIC	15 (17%)	13 (15%)	30 (15%)	41 (20%)	0.736
IV	73 (83%)	73 (85%)	175 (85%)	166 (80%)	
Histological type					
Adenocarcinoma	86 (98%)	84 (98%)	202 (99%)	204 (99%)	0.711
Non-adenocarcinoma	2 (2%)	2 (2%)	3 (1%)	3 (1%)	
Brain metastases at baseline					
Yes	2 (2%)	3 (3%)	10 (5%)	5 (2%)	0.640
No	86 (98%)	83 (97%)	195 (95%)	202 (98%)	
PD-L1 tumor proportion score					
<1%	19 (22%)	21 (24%)	49 (24%)	69 (34%)	0.115
≥1%	65 (74%)	56 (65%)	138 (67%)	117 (57%)	
Not evaluable	3 (3%)	9 (10%)	18 (9%)	21 (10%)	

ECOG, Eastern Cooperative Oncology Group.

Supplementary Table S2: Baseline characteristics of included patients from the biomarker evaluable population and intention-to-treat cohort in CameL-sq study.

	Biomarker evaluable trial		Intention-to-treat population		
	Camrelizumb plus chemotherapy	Placebo plus chemotherapy	Camrelizumb plus chemotherapy	Placebo plus chemotherapy	*P* value
	(n=118)	(n=135)	(n=193)	(n=196)	
Age					
≥65 years	55 (47%)	52 (44%)	84 (44%)	71 (36%)	0.538
<65 years	63 (53%)	83 (56%)	109 (56%)	125 (64%)	
Sex					
Male	113 (96%)	127 (94%)	179 (93%)	180 (92%)	0.202
Female	5 (4%)	8 (6%)	14 (7%)	16 (8%)	
Smoking history					
≥400 cigarette-years	104 (88%)	111 (82%)	162 (84%)	157 (80%)	0.325
<400 cigarette-years or never	14 (12%)	24 (8%)	31 (16%)	39 (20%)	
ECOG performance status					
0	21 (18%)	30 (22%)	38 (20%)	43 (22%)	0.839
1	97 (82%)	105 (78%)	155 (80%)	153 (78%)	
Disease stage					
IIIB/IIIC	34 (29%)	40 (30%)	54 (28%)	55 (28%)	0.736
IV	84 (71%)	95 (70%)	139 (72%)	141 (72%)	
Brain metastases at enrollment*					
Yes	1 (1%)	3 (2%)	4 (2%)	3 (2%)	0.918
No	117 (99%)	132 (98%)	189 (98%)	193 (98%)	
PD-L1 tumor proportion score					
<1%	55 (47%)	68 (50%)	91 (47%)	97 (49%)	0.943
≥1%	63 (53%)	67 (50%)	95 (49%)	93 (47%)	
Not evaluable	0 (0%)	0 (0%)	7 (4%)	6 (3%)	

Data are n (%), unless otherwise indicated. * No patients with both liver and lung metastases were enrolled. ECOG, Eastern Cooperative Oncology Group.

Supplementary Table S3. Baseline parameters of included 11 patients.

Sample ID	Histology	Sex	Age	Smoking history	Stage	HED HLA-A	HED HLA-B	HED HLA-C	Mean HED
S18T	LUAD	Female	60	No	T1bN0M0	0.31	7.12	6.89	4.77
S44T	LUAD	Female	73	No	T1aN2M0	1.07	10.87	5.23	5.72
S47T	LUSC	Male	67	Yes	T2aN0M0	8.10	10.23	2.73	7.02
S57T	LUSC	Male	65	No	T2bN1M0	7.46	10.17	3.59	7.07
S68T	LUSC	Male	66	No	T2aN0M0	7.04	7.51	4.71	6.42
S91T	LUAD	Female	64	No	T1bN0M0	10.94	7.06	6.18	8.06
S92T	LUAD	Male	62	No	T1bN0M0	11.57	9.11	5.88	8.86
S93T	LUSC	Male	69	Yes	T1bN0M0	10.66	10.75	2.90	8.10
S94T	LUAD	Male	64	Yes	T1cN0M0	8.24	8.64	6.01	7.63
S95T	LUAD	Female	77	No	T2aN0M0	8.10	3.88	6.43	6.14
S96T	LUAD	Male	56	No	T1cN2M0	0.00	6.99	0.69	2.56

LUAD, lung adenocarcinoma; LUSC, lung squamous cell carcinoma.

Supplementary Table S4. Table of Research Resource Identifier.

Tool and Resources	RRID	Link
Agilent 2100 BioAnalyzer	RRID:SCR_019715	https://www.agilent.com/cs/library/posters/Public/BioAnalyzer.PDF
OptiType	RRID:SCR_022279	https://github.com/FRED-2/OptiType
IMGT/HLA	RRID:SCR_002971	http://www.ebi.ac.uk/imgt/hla/
LOHHLA	RRID:SCR_023690	https://github.com/slagtermaarten/LOHHLA
Illumina HiSeq4000	RRID:SCR_016386	https://www.illumina.com/systems/sequencing-platforms/hiseq-3000-4000.html
bcl2fastq	RRID:SCR_015058	https://support.illumina.com/sequencing/sequencing_software/bcl2fastq-conversion-software.html
Trimmomatic	RRID:SCR_011848	http://www.usadellab.org/cms/index.php?page=trimmomatic
NCBI Build 37.5 (NCBI Assembly Archive Viewer)	RRID:SCR_012917	https://www.ncbi.nlm.nih.gov/assembly
Burrows-Wheeler Aligner (BWA)	RRID:SCR_010910	http://bio-bwa.sourceforge.net/
Picard	RRID:SCR_006525	http://broadinstitute.github.io/picard/
Genome Analysis Toolkit (GATK)	RRID:SCR_001876	https://software.broadinstitute.org/gatk/
MuTect	RRID:SCR_000559	http://www.broadinstitute.org/cancer/cga/mutect
ENCODE	RRID:SCR_006793	http://genome.ucsc.edu/ENCODE
ANNOVAR	RRID:SCR_012821	http://www.openbioinformatics.org/annovar/
Exome Aggregation Consortium (ExAC)	RRID:SCR_004068	http://exac.broadinstitute.org/
Genome Aggregation Database (gnomAD)	RRID:SCR_014964	http://gnomad.broadinstitute.org/
COSMIC	RRID:SCR_002260	http://cancer.sanger.ac.uk/cancergenome/projects/cosmic/
CNVkit	RRID:SCR_021917	https://github.com/etal/cnvkit
dbSNP	RRID:SCR_002338	http://www.ncbi.nlm.nih.gov/SNP/
HLA-HD	RRID:SCR_022285	https://www.genome.med.kyoto-u.ac.jp/HLA-HD/
NetMHCpan 4.0	RRID:SCR_018182	http://www.cbs.dtu.dk/services/NetMHCpan/
Immune Epitope Database and Analysis Resource (IEDB)	RRID:SCR_006604	http://www.immuneepitope.org/
HLA-ATHLATES	RRID:SCR_023689	https://github.com/cliu32/athlates
Seurat	RRID:SCR_016341	https://satijalab.org/seurat/get_started.html
R (version 4.1.3)	RRID:SCR_001905	http://www.r-project.org/
clusterProfiler	RRID:SCR_016884	http://bioconductor.org/packages/release/bioc/html/clusterProfiler.html

BD Biosciences (Ethylene diamine tetraacetic acid (EDTA)-coated tubes)	RRID:SCR_013311	http://www.bdbiosciences.com/us/home
QIAGEN (QIAamp DNA FFPE Tissue Kit、DNeasy Blood and Tissue Kit、GeneRead DNA FFPE Kit)	RRID:SCR_008539	http://www.qiagen.com
Thermo Fisher Scientific (Qubit dsDNA High Sensitivity Assay Kit)	RRID:SCR_008452	http://www.fishersci.com
Roche (KAPA Hyper Prep Kit、KAPA Library Quanitification Kit)	RRID:SCR_001326	http://www.roche.com/
TIANGEN (TGuide S32 Magnetic Blood Genomic DNA Kit)	RRID:SCR_023688	https://www.tiangen.com/