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Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Research policies, see Authors & Referees and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed	
☐	☒	The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒	A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒	The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐	A description of all covariates tested
☒	☐	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒	A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒	For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐	Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	Electronic Case Report Forms (eCRF) (version: 3.0) for clinical data. Nucleofector 2b device (Lonza, Germany) for preparation of edited T cells, Veriti thermocycler (Applied Biosystems, USA) for T7E1 cleavage assay, FACSAria II flow cytometry (BD Biosciences, CA), Ventana Ultra instrument (USA) and optical microscope (Olympus, Leica S3, Japan) for immunohistochemistry staining, IMMAGE 800 (Beckman Coulter, USA) for IL-6 data, IMMULITE 1000 (Siemens, UK) for IL-8 and TNF-a data,, Illumina (San Diego, CA), HiSeq X Ten instrument (Illumina, USA) and MiSeq Reporter (Illumina, USA) for tracking of edited cells; NovaSeq 6000 platform (Illumina) for WGS data, Fastq join (version 1.3.1) program for NGS data; sgRNAcas9 software (version 2.0.6) for predicting off-targets by NGS; Cas-OFFinder (version 2.4) for predicting potential off-target sites by WGS.
Data analysis	MedDRA (version 21.0) for AE analysis; Paired Student's t test for IFN-γ production analysis; The exact Wilcoxon test for TCR diversity analysis; ImageJ software (version 1.47) for immunohistochemistry staining density; samtools (version1.1), sambamba (version 0.6.8), strelka2 (version 125 2.8.3), Fusionmap (version 10.0.1.29) and ANNOVAR (version 2019Oct24) for WGS data analysis; SPSS 22.0 for OS and PFS data analysis; FlowJo software (version 7.6.1) for flow cytometry analysis; Details of the all data analysis are included in the Methods.

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors/reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

The data that support the findings of this study will be provided by corresponding authors upon reasonable request.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	This is a standard 3+3 designed for phase I dose-escalation trial. And added a cohort Pre-A aims to assure the safety of trial. So size of enrolled 10 patients were designed in four cohorts (cohort Pre-A, A, B and C). During the trial, cohort Pre-A added one patient for consideration of safety because of the first patient (Pre-A-01) suffering from arrhythmia. Cohort A added one patient (A-04) due to patient (A-03) early withdrawal from trial. Finally, the actual sample size was 12 patients for evaluation of safety and feasibility in this trial.
Data exclusions	No data was excluded from the analysis.
Replication	We included data on all available patients/samples in all figures.
Randomization	There was no randomization in this phase I single arm study.
Blinding	There was no blinding as the paper reports a phase I single arm study.

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology
☒	☐ Animals and other organisms
☐	☒ Human research participants
☐	☒ Clinical data

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☐	☒ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used

1. Antibodies used for flow cytometry
Antigen, Antibody Clone, Manufacturer, Catalog number, Working dilution, Lot number
CD3-BV510, HIT3a, BD Biosciences, 564713, 1:50, 9164820
CD4-BV421, RPA-T4, BD Biosciences, 562424, 1:50, 8187880
CD8-PE, HIT8a, BD Biosciences, 555635, 1:50, 8299501
PD-1-APC, MIH4, BD Biosciences, 558694, 1:50, 7075610
IFN-γ-PE, B27, BD Biosciences, 554701, 1:50, 8354864
CD8-APC-Cy7, BD Biosciences, 557834, 1:50, 8184768
2. Antibodies used for immunohistochemstry
Antigen, Antibody Clone, Manufacturer, Catalog number, working dilution, lot number.
PD-L1, Ventana SP142, Roche, 740-4859, neat, E29397
secondary antibody: DAB, Roche, 760-500, neat, F13568

CD3, LN10, ZSGB-BIO, ZM0417, 1:100, 2005005
secondary antibody: DAB, Roche, 760-700, neat, F11441
CD8, C8/144B, DAKO, m7103, 1:100, 20042547
secondary antibody: DAB, Roche, 760-500, neat, F13568
CD68, PGM-1, DAKO, m0876, 1:100, 20036057
secondary antibody: DAB, Roche, 760-500, neat, F13568
3. Antibodies used for T cell expansion and activation
Antibody, Manufacturer, Catalog number, Working dilution, Lot number.
anti-CD3 antibody, OKT3, Novoprotein Scientific Inc, GMP-A018, 1:100, 0331305
anti-CD28 antibody, Novoprotein Scientific Inc, GMP-A063, 1:100, 0331306
IFN-γ Novoprotein Scientific Inc, GMP-CI57, 1:1000, 0331197
IL-2 Beijing Shuanglu Pharmaceutical Co., Ltd., Sinopharm Standard S19991010, 1:1000, 20160308

Validation

All antibodies used in this study are commercially available. Antibodies used in a specific species or application have been appropriately validated by manufacturers for that application and this information is provided on their website and product information datasheets. All antibodies described here have been further optimized for an appropriate concentration by testing several dilutions.

Human research participants

Policy information about studies involving human research participants

Population characteristics	Twelve patients received the planned PD-1 gene edited T-cell therapy are eligible for analysis, the population characteristics of them were provided in Table 1. The median age of the patients was 54.5 (range: 31-68). Four patients were female and 8 were male. Nine patients had 3 previous systemic treatment regimens. Three patients had more than 3 previous systemic treatment regimens. Two patients were previously treated with PD1 antibody.
Recruitment	Patients were recruited through recruitment advertisement which was approved by the Institutional Review Board of West China Hospital, Sichuan University. The recruitment advertisement was put up as a poster in bulletin board of GCP, West China Hospital. Patients were enrolled on a first-come-first-served basis. There was no potential self-selection bias in recruiting patients. Twenty-two patients were enrolled based upon the inclusion and exclusion criteria in this phase I clinical trial. All patients were explained and written and signed informed consent was obtained prior to enrollment.
Ethics oversight	The study protocol and subsequent amendments (Necessary additional data) were approved by the Institutional Review Board of West China Hospital, Sichuan University, Chengdu, China (2016 Review, No. 123).

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Clinical data

Policy information about clinical studies

All manuscripts should comply with the ICMJE guidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.

Clinical trial registration	MHC-001 ClinicalTrials.gov number, NCT02793856
Study protocol	The full trial protocol are available in the supplementary materials for this article.
Data collection	For study using Electronic Data Capture (EDC), Version 3.61.3 PROD XKL 20180710 (https://edc.edetek.cn:8443/MHC001/index.action), the designated investigator staff will enter the data required by the protocol into the Electronic Case Report Forms (eCRF). The principal investigator is responsible for assuring that the data entered into eCRF is complete, accurate, and that entry and updates are performed in a timely manner. All data collected at between Aug 26, 2016 and Jan 31, 2020.
Outcomes	The primary endpoints of this phase 1 dose escalation study was safety and feasibility. Secondary endpoint was efficacy. Exploration objectives included assessments of T-cell receptor (TCR) clones and in vivo tracking of edited T cells.

Flow Cytometry

Plots

Confirm that:

☒ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).

☒ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).

☒ All plots are contour plots with outliers or pseudocolor plots.

☒ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation	Human blood was collected in Red-top tubes (CDRICH®, BD Biosciences) and was processing in research lab. The PD-1 edited T cells were prepared by. MedGenCell, Co., Ltd (Chengdu, China).

Instrument	FACSAria II flow cytometry (BD Biosciences, CA)
Software	FlowJo software (version 7.6.1)
Cell population abundance	N/A
Gating strategy	FSC-A/FSC-H plots were used to determine singlet gates. FSC-A/SSC-A plots were used to determine cell population gates.Isotype controls were used to indicate the boundaries between positive and negative populations.

☒ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.