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Reporting Summary

Nature Portfolio 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 Portfolio policies, see our Editorial Policies 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	FACS Diva (version 8.0.0.1); LAS X (version 3.3.0); CytExpert (Version 2.4); ZEN Lite (version 2.3)
Data analysis	ImageJ Version 2.0.0-rc-43/1.51q; FlowJo 10.2; GraphPad Prism v9;

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 and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio 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 description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

Source data are provided with this paper. Further data supporting the findings of this study are available from the corresponding author 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	All human experiments were performed from at least three different donors. For mouse studies at least three differents animals were used. For the in vivo studies all mice were gender (male) and age-matched and were purchased from Charles River to minimize difference between experimental group. All animals were kept in safe and no stress environment. Sample size was determined according to similar experiments carried out priviously by authors of the work (Lanna et al 2014, 2017).
Data exclusions	No data were excluded in both human and mouse studies.
Replication	Every experiment was reproduced three or more time (up to 9 times). Where indicated some experiments were performed twice. Statistical analysis were performed throughout the study to ensure significance of results. All replications of experimets were successful.
Randomization	The experimets were not randomized. Groups were labelled and defined (e.g. Tel- vs Tel+) and randomization was not necessary.
Blinding	Investigators were not blinded to the group allocations,. There were defined groups (e.g T cells without vesicles vs T cell with vesicles) and blinding was not necessary (for instance Thapa, N et al. 2020 and Chen, M., Choi, S., Wen, T. et al. all in Nature Cell Biology 2022), except for the microscopic analysis of IF-FISH staining and in vivo manipulation executed by technical staff.

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 and archaeology
☐	☒ Animals and other organisms
☐	☒ Human research participants
☒	☐ Clinical data
☒	☐ Dual use research of concern

Methods

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

Antibodies

Antibodies used

α-Rad51 ab63801, Abcam
α-TRF2 ab13579, Abcam
α-POT1 PA-66996, ThermoFisher
α-POT1 NB500-176 (Novusbio)
α-Sestrin 1 ab134091, Abcam
α-TERT sc-7212 , Santa Cruz
α-CD3 BIO-RAD MCA463G
Goat α-rabbit IgG AlexaFluor 405 A-31556, Life Technologies
Goat α-mouse IgG1 AlexaFluor 647 A-21240, Life Technologies
α-p-H2AX S139 Abcam, ab26350
α-BRCA1 GeneTex, GTX70113
α-Cytochrome C BD pharmigen, 556432
α-MHC II Abcam, ab55152
α-TZAP Abnova, H00003104-B01P
α-CD63 GeneTex, GTX28219
α-BRCA2 Abcam, ab123491
α-TSG101 GeneTex, GTX70255
α-IgG control Cell Signaling, 3900S
Goat α-Rabbit IgG-HRP conjugated 12-348, Sigma-Aldrich

Goat α-Mouse IgG-HRP conjugated 12-349, Sigma-Aldrich
α-CD4 Alexa 700 clone OKT4, Biolegend, Cat. 317426
α-CD27 PerCP5.5 clone M-T271, Biolegend, Cat. 356408
α-CD28 APC-Vio770 clone REA612, Miltenyi Biotec, Cat. 130-116-506
α-CD45RA BV421 clone HI100, Biolegend, Cat. 304130
α-CD45RO ECD clone UCHL1, Beckman Coulter, Cat. B49192
α-PD1 APC clone NAT105, Biolegend, Cat. 367406
α-FasL PE-Cy7 clone NOK-1, Biolegend, Cat. 306418
α-CTLA4-594 clone L3D10, Biolegend, Cat. 349922
α-TIM-3 VioBright FITC clone REA635, Miltenyi Biotec, Cat. 130-124--442
α-Strepavidin-collodial 10nm gold conjugate S9059, Sigma Aldrich
α-DNA Abcam, ab27156
α-BrdU abcam, ab6326
Goat α-rabbit IgG AlexaFluor 594 Thermofisher Cat # A32740
α-CD4 APC-Vio 770 Miltenyi, 130-118-955
α-CD44 Miltenyi, Cat. 130-119-121
α-CD62L PE-Vio 770 Miltenyi Cat. 130-112-649
α-CD45.1 PerCP-Vio 700 Miltenyi Cat. 130-121-221
α-CD45.2 PE Milteny Cat. 130-124-080
α-CD45.2-biotin Miltenyi Cat.130-124-089
α-CD95-Fitc Miltenyi Cat. 130-122-950
Goat α-rabbit IgG HRP-conjugated Cell signalling, 7074P2
α-H3 ab18521, Abcam
α-Caspase 3 9661T, Cell Signaling)
α-CD3 86022706 Sigma Aldrich
α-CD28 37407 MAB342 R&D systems
α-mouse IgG HRP-conjugated 31430 Invitrogen
α-rabbit IgG hHRP-conjugated 31460 Invitrogen

Validation

Immunoblots: Rad51 (ab63801, Abcam): https://www.abcam.com/rad51-antibody-ab63801.html
TRF2 (ab13579, Abcam): https://www.abcam.com/trf2-antibody-4a794-ab13579.html
POT1 (PA-66996, ThermoFisher): https://www.thermofisher.com/antibody/product/POT1-Antibody-Polyclonal/PA5-66996
POT1 (NB500-176 (Novusbio): https://www.novusbio.com/products/pot1-antibody_nb500-176
Sestrin 1 (ab134091, Abcam):https://www.abcam.com/sesn1-antibody-epr19302-ab134091.html
TERT (sc-7212 , Santa Cruz): https://www.citeab.com/antibodies/831165-sc-7212-tert-antibody-h-231
CD3(BIO-RAD MCA463G): https://www.bio-rad-antibodies.com/monoclonal/human-cd3-antibody-ucht1-mca463.html?f=purified&JSESSIONID_STERLING=D8BF321DC023536B7AE2367D6C6D8CB9.ecommerce1&evCntryLang=IT-itthirdPartyCookieEnabled
Goat anti-rabbit IgG AlexaFluor 405 (A-31556, Life Technologies): https://www.thermofisher.com/antibody/product/Goat-anti-Rabbit-IgG-H-L-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-31556
Goat anti-mouse IgG1 AlexaFluor 647 (A-21240, Life Technologies): https://www.thermofisher.com/antibody/product/Goat-anti-Mouse-IgG1-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-21240
p-H2AX S139 (Abcam, ab26350): https://www.abcam.com/gamma-h2ax-phospho-s139-antibody-9f3-ab26350.html
BRCA1 (GeneTex, GTX70113): https://www.genetex.com/Product/Detail/BRCA1-antibody-8F7/GTX70113
Cytochrome C (BD pharmigen, 556432): https://www.genetex.com/Product/Detail/BRCA1-antibody-8F7/GTX70113
MHC II (Abcam, ab55152): https://www.abcam.com/mhc-class-ii-antibody-6c6-ab55152.html
TZAP (Abnova, H00003104-B01P): http://www.abnova.com/products/products_detail.asp?catalog_id=H00003104-B01P
CD63 (GeneTex, GTX28219): https://www.genetex.com/Product/Detail/CD63-antibody-MEM-259/GTX28219
BRCA2 (Abcam ab123491): https://www.abcam.com/brca2-antibody-ab123491.html
TSG101 (GeneTex, GTX70255): https://www.genetex.com/Product/Detail/TSG101-antibody-4A10/GTX70255
IgG control(Cell Signaling, 3900S): https://www.cellsignal.com/products/primary-antibodies/rabbit-da1e-mab-igg-xp-isotype-control/3900
Goat Anti-Rabbit IgG-HRP conjugated (12-348, Sigma-Aldrich): https://www.sigmaaldrich.com/IT/it/product/mm/12348
Goat Anti-Mouse IgG-HRP conjugated (12-349, Sigma-Aldrich): https://www.sigmaaldrich.com/IT/it/product/mm/12349
CD4 Alexa 700 (clone OKT4, Biolegend, Cat. 317426): https://www.biolegend.com/fr-ch/products/alexa-fluor-700-anti-human-cd4-antibody-3661
CD27 PerCP5.5 (clone M-T271, Biolegend, Cat. 356408): https://www.biolegend.com/de-at/products/percp-cyanine5-5-anti-human-cd27-antibody-8416
CD28 APC-Vio770 (clone REA612, Miltenyi Biotec, Cat. 130-116-506): https://www.miltenyibiotec.com/IT-en/products/cd28-antibody-anti-human-reafinity-rea612.html#apc-vio-770:30-tests-in-60-ul
CD45RA BV421 (clone HI100, Biolegend, Cat. 304130): https://www.biolegend.com/de-de/products/brilliant-violet-421-anti-human-cd45ra-antibody-7200
CD45RO ECD (clone UCHL1, Beckman Coulter, Cat. B49192): https://www.beckman.it/reagents/coulter-flow-cytometry/antibodies-and-kits/single-color-antibodies/cd45ro/b49192
PD1 APC (clone NAT105, Biolegend, Cat. 367406): https://www.biolegend.com/fr-lu/products/apc-anti-human-cd279-pd-1-antibody-12184?GroupID=GROUP28
FasL PE-Cy7 (clone NOK-1, Biolegend, Cat. 306418): https://www.biolegend.com/en-us/search-results/pecyanine7-anti-human-cd178-fas-l-antibody-18204?GroupID=BLG10280
CTLA4-594 (clone L3D10, Biolegend, Cat. 349922): https://www.biolegend.com/en-us/products/pe-dazzle-594-anti-human-cd152-ctla-4-antibody-12216
TIM-3 VioBright FITC (clone REA635, Miltenyi Biotec, Cat. 130-124--442): https://www.miltenyibiotec.com/IT-en/products/cd366-tim-3-antibody-anti-human-reafinity-rea635.html?countryRedirected=1#vio-bright-fitc:30-tests-in-60-ul
Strepavidin-collodial 10nm gold conjugate (S9059, Sigma Aldrich): https://www.sigmaaldrich.com/IT/it/product/sigma/s9059
DNA (Abcam, ab27156): https://www.abcam.com/ds-DNA-antibody-35I9-DNA-BSA-and-Azide-free-ab27156.html?gclsrc=aw.ds%7Caw.ds&gclid=EAIaIQobChMIjZfL-5_2-AIVpxOtBh1Bpwo4EAAYASAAEglymvD_BwE

BrdU (abcam, ab6326): https://www.abcam.com/brdu-antibody-bu175-icr1-proliferation-marker-ab6326.html
Goat Anti-Rat IgG Antibody, HRP conjugate (Sigma aldrich; AP136P): https://www.sigmaaldrich.com/IT/it/product/mm/ap136p?gclid=EAIaIQobChMIh7Hz7aD2-AIV8wytBh314wriEAAYASAAEgJCGvD_BwE
CD4 APC-Vio 770 (Miltenyi, 130-118-955): https://www.miltenyibiotec.com/IT-en/products/cd4-antibody-anti-mouse-reafinity-rea604.html#apc-vio-770:150-ug-in-1-ml
CD44 (Miltenyi, Cat. 130-119-121): https://www.miltenyibiotec.com/IT-en/products/cd44-antibody-anti-mouse-reafinity-rea664.html#gref
CD62L PE-Vio 770 (Miltenyi Cat. 130-112-649): https://www.miltenyibiotec.com/IT-en/products/cd62l-antibody-anti-mouse-reafinity-rea828.html#pe-vio-770:150-ug-in-1-ml
CD45.1 PerCP-Vio 700 (Miltenyi Cat. 130-121-221): https://www.miltenyibiotec.com/IT-en/products/cd45-1-antibody-anti-mouse-reafinity-rea1179.html#percp-vio-700:30-ug-in-200-ul
CD45.2 PE (Milteny Cat. 130-124-080): https://www.miltenyibiotec.com/IT-en/products/cd45-2-antibody-anti-mouse-reafinity-rea1223.html#pe:30-ug-in-200-ul
CD45.2-biotin (Miltenyi Cat.130-124-089): https://www.miltenyibiotec.com/IT-en/products/cd45-2-antibody-anti-mouse-reafinity-rea1223.html#biotin:30-ug-in-200-ul
CD95-Fitc (Miltenyi Cat. 130-122-950): https://www.miltenyibiotec.com/IT-en/products/cd95-fas-antibody-anti-mouse-reafinity-rea453.html#gref

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	*State the source of each cell line used.*
Authentication	*Describe the authentication procedures for each cell line used OR declare that none of the cell lines used were authenticated.*
Mycoplasma contamination	*Confirm that all cell lines tested negative for mycoplasma contamination OR describe the results of the testing for mycoplasma contamination OR declare that the cell lines were not tested for mycoplasma contamination.*
Commonly misidentified lines (See ICLAC register)	*Name any commonly misidentified cell lines used in the study and provide a rationale for their use.*

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals	All animals were age and gender-matched (six-week old male mice) and were simultaneously purchased from Charles River. We used C57BL6J mice (B6.WT mice) and C57BL6-Tg (TcraTcrb)425Cbn/Crl (B6.OTII mice) throughout the study
Wild animals	No wild animals were used in this study.
Field-collected samples	No field-collected samples were used.
Ethics oversight	All mice experiments were carried out under approval of Italian Minister of Health (454/2020) and UK Home office PPL number P69E3D849

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

Human research participants

Policy information about studies involving human research participants

Population characteristics	We collected heparinized peripheral blood samples from 140 healthy donors (aged 20-65, male 55% and female 45%).
Recruitment	Recruitment took place by email or poster advertising at the University of Oxford and University College London, respectively or the Fondazione Santa Lucia.
Ethics oversight	This research complies with all relevant ethical regulations. All human specimens were obtained with the approval of Fondazione Santa Lucia Ethical Committee or the Ethical Committee of Royal Free and University College Medical School or the University of Oxford, with voluntary informed consent in accordance with the Declaration of Helsinki. No compensation was offered to any participant.

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

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	Primary human CD27/CD28 CD4+ T cell subsets were purified from peripheral blood by immunomagnetic separation using commercially available kits from Miltenyi, as described below. Briefly, peripheral blood mononuclear cells (PBMCs) were isolated by Ficoll followed by negative selection for CD4 (c.n.130-096-533). The untouched population was then purified by CD27 microbeads first (c.n.130-051-601). Non-senescent early-stage CD27+ CD28+ CD4+ T cells (Terl) were used throughout the study. For isolation of mouse T cells from splenocytes, CD4 MicroBeads (L3T4; c.n. 130-117-043) were used. For APC purification, human CD3 MicroBeads (c.n.130-050-101) and mouse CD3e Microbead kit (c.n.130-094-973) were used to deplete T cells. To form conjugates, APCs were pre-loaded with antigen mix (HBV, Influenza and CMV lysates) or OVA antigens at 37°C for 4h followed by addition of autologous T cells in a 3:1 ratio, as described in 'Methods'. Telomere vesicles were extracted from APCs live-labelled with Cy3 TelC telomere probes and the membrane lipid dye PKH67 (Sigma Aldrich). After labelling, APCs were activated with ionomycin as above described, and the culture media were collected 18h later. After sequential centrifugation (300g, 2000g and 10000g), the APC supernatants were analysed by fluorescence activated vesicle sorting using the MoFlo Astrios-EQ flow cytometer (Beckman Coulter).
Instrument	LSR-2 BD flow-cytometer (BD) and CytoFLEX Flow Cytometer (Beckman Coulter) for flow cytometry. MoFlo Astrios-EQ flow cytometer (Beckman Coulter) was used for sorting of Telomere vesicles.
Software	All events were acquired using the BD FACS Diva software and CytExpert software. Data were analyzed with FlowJo software v.10.2.
Cell population abundance	Sample purity (>90%) was confirmed by flow-cytometry.
Gating strategy	Lentiviral vector-transduced T cells were identified and gated on the GFP+ compartment as indicated. For adoptive transfers, donor T cells were identified by either Cell Trace Violet (CTV; in vivo telomere transfer studies) or GFP+ transduction (long-term memory studies). For in vivo telomere transfer studies, CTV+ donor OT-II cells were analyzed for uptake of Cy3-TelC-labelled. For telomere vesicles sorting, gating was done using the Ultra Rainbow Fluorescent Particles 3.0 µm (Spherotech, URFP-30-2). The triggering threshold was applied to the side scatter on the 488-SSC channel and the relative applied voltage was determined by using size reference beads (Apogee flow systems, 1493) to exclude vesicle aggregates >300 nm. The Instrument setup to identify microvesicles was obtained by balancing triggering threshold and SSC voltage in order to reduce and maintain over time the background noise at 150-300 events/sec.

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