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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	n/a
Data analysis	GraphPad Prism (v10.3.1) software R Studio (version 2022.12.0 Build 353) Python programming language (version 3. 9.7) Matplotlib (version 3.4.3) for data visualization, Pandas (version 1.3.4) for data management, NumPy (version 1.20.3) for numerical computations, and Jupyter Notebook (version 6.4.5); g:Profiler (version e108_eg55_p17_0254fbf) with g:SCS ;Ingenuity Pathway Analysis (IPA; Ingenuity® Inc, Redwood city, CA) and Metascape comparison analysis (https://metascape.org); RVAideMemoir' R package

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

RNA sequencing data is available (open access) on Array Express accession E-MTAB-13849.
All raw data provided with the manuscript (excel file).

Research involving human participants, their data, or biological material

Policy information about studies with human participants or human data. See also policy information about sex, gender (identity/presentation), and sexual orientation and race, ethnicity and racism.

Reporting on sex and gender	n/a
Reporting on race, ethnicity, or other socially relevant groupings	n/a
Population characteristics	n/a
Recruitment	n/a
Ethics oversight	n/a

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

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	Pilot studies were performed prior to study commencement using n=4/group to determine mean and SD.
Data exclusions	No data was excluded.
Replication	Multiple people across different collaborating laboratories performed the experiments and each time the data was replicated at least 3 x independently
Randomization	Animals were randomly assigned to experimental groups. Group allocation was performed without prior knowledge of treatment conditions to reduce allocation bias. Where applicable, littermates were distributed across groups to avoid litter effects.
Blinding	Blinding was not always possible as multiple researchers collected and processed the samples. Blinding was applied during outcome assessment -all data in the manuscript was analysed blinded to the phenotype). Experiments where blinding was possible: electrocardiography, flow cytometry, histology (fibrosis), cytokine analysis, NMR spectroscopy (1H NMR), RNA sequencing, biochemical analysis (plasma analysis).

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
☒	☐ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

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

Antibodies

Antibodies used

Dilution for all antibodies used 1:200.

Dilution used 1:200

Live/ Dead Dead Zombie Yellow™ 423104 BioLegend
Leukocytes CD45 FITC REA737 130-110-803 Miltenyi
T cells (cytotoxic) CD8 Per-CP-Vio-700 REA601 130-128-228 Miltenyi
Macrophages F4/80 APC REA126 130-131-632 Miltenyi
Neutrophils Ly6G APC-Vio 770 REA526 130-128-232 Miltenyi
Monocytes LyC VioBlue REA796 130-128-235 Miltenyi
B cells B220 Vio green REA755 130-110-852 Miltenyi
T cells (memory) CD62L BV785 CDF IgG2a, κ 564109 BD Biosciences
Dendritic cells CD11c PE REA754 130-110-837 Miltenyi
Monocytes/ macrophages CD11b PE-Vio® 615 REA592 130-113-811 Miltenyi
T cells (regulatory) CD4 PE-Vio-770 REA604 130-127-473 Miltenyi
T cells CD3 UV395 IgG2b, κ 569614 BD Biosciences

Validation

These are FACS antibodies and all have been validated by manufacturers as stated on their websites. Validation was performed by the company, see website for details. No antibody was used for a procedure that had not previously been validated.

Animals and other research organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in Research

Laboratory animals

Commercially available type 2 diabetic mice (db/db mouse, The Jackson Laboratory homozygous BKS.Cg-Dock7m +/+ Leprdb/J, male, Charles River, Italy) were purchased at 8 weeks with corresponding lean littermate controls (heterozygous Dock7m +/+ Leprdb, Charles River, Italy). Animals were kept under pathogen-free conditions, 12h light–dark cycle, controlled temperature (20–22°C), and fed chow and water ad libitum. This investigation conformed to UK Home Office Guidance on the Operation of the Animals (Scientific Procedures) Act, 1986.

Wild animals

n/a

Reporting on sex

Male mice were used.

Field-collected samples

n/a

Ethics oversight

All experiments were conducted using adult male mice. The exclusive use of male subjects was based on the need to minimize variability due to hormonal fluctuations associated with the estrous cycle in females, which could confound interpretation of the results in this specific study design. This approach is commonly used in initial mechanistic studies to establish baseline findings before expanding to both sexes in follow-up research. All experiments were approved by Queen Mary University of London AWERB ethics committee in accordance with the UK Home Office Guidance on the Operation of the Animals Scientific Procedures Act 1986.

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

Plants

Seed stocks	N/A
Novel plant genotypes	N/A
Authentication	N/A

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	To generate leukocyte single cell suspensions for characterisation of immune cell populations, tissues were isolated from mice and digested. Mice were euthanised using an overdose of anaesthesia with 5 % isoflurane in 2 L/min O2. Cessation of pedal and corneal reflexes were checked and death was confirmed by cervical dislocation. Cardiac tissue suspensions were prepared by perfusing hearts with cold HBSS for 5 min prior to removing the atria and mincing the ventricular tissue that was digested in collagenase I (Worthington Laboratories, C1639, 450 units/mL), collagenase XI (Worthington Laboratories, C7657, 125 units/mL), DNase 1 (Worthington Laboratories, D4527, 60 units/mL) Hyaluronidase (Sigma Aldrich, H3506, 60 units/mL) and 20mM Hepes in PBS for 20 minutes at 37℃ with gentle agitation (Thermomixer, 750 rpm). Spleens were mechanically dissociated by mashing through a cell strainer using the plunger end of a sterile syringe. Samples were passed through a 70 µm cell strainer, rinsed with 10 mL cold 2% FBS/ PBS and centrifuged at 400g for 10 min at 4℃. Supernatant was removed and pellets resuspended in 5 mL red blood cell lysis buffer (BioLegend) and incubated on ice for 10 min with occasional agitation after which 10 mL cold 2% FBS/ PBS was added to neutralise the lysis. Samples were then centrifuged for 8 min at 4℃ and 400g, supernatant was removed and cells were resuspended and incubated with FC-block (BioLegend 101320, 1 µL per 1 □ 106/ mL cells). Cells were then washed again and resuspended in PBS ready for counting and antibody staining. Flow cytometry Cells isolated from hearts and spleen were resuspended (~107/ml) and incubated for 30 minutes at room temperature with fluorochrome-conjugated antibodies (Supplementary Table 1) in 100 □l of flow cytometry buffer made of PBS containing 0.1% sodium azide (SigmaAldrich) and 1% FBS. For intracellular marker staining, cells were fixed and permeabilized for 30 minutes at 4oC using fixation/permeabilization kit (eBioscience), washed in 1X permeabilization buffer (eBioscience) and stained with fluorochrome-conjugated antibodies in 1x permeabilization buffer for 30 minutes at 4□C. A final wash with 1x permeabilization buffer was performed, centrifuged and resuspended in 200µl of flow cytometry buffer. Alternatively, cells were fixed (Fix/Perfm kit, BioLegend 426803) and stored at 4℃. Cell viability was assessed using incubation with viability dyes (Supplementary Table 1). Samples were analysed on FACSAriaIII (BD Biosciences) running FACSDiVa v.8.0 software (BD Biosciences). CD3 beads (Miltenyi, UK) were routinely used to calibrate the cytometer. Single stain and fluorescence minus one control were acquired for compensation and precise gating (Supplementary Fig1-3 gating strategies). Compensation was automatically calculated, and samples analyzed using FlowJo software (version 10, FlowJo LLC, Oregon, USA).
Instrument	LSR Fortessa (BD Biosciences); FACSAriaIII (BD Biosciences)
Software	FlowJo V10 was used to analyse the data, along with Prism v9; FACSDiVa v.8.0 software (BD Biosciences)
Cell population abundance	We didnt use any cell sorting. We isolated cells from the hearts as described above.
Gating strategy	Gating strategy provided in supplementary fig 1 and 2: Treg (CD4CD25FoxP3): Lymphocytes, Single Cells, Live, CD4, Treg. T cell (CD4 and CD8)panel: CD45+, Viability, Single Cells, CD45, leukocytes, B cells, CD3 Te cells, CD4 and CD8 Tcells. Non-T cell panel: CD45+, Viable Cells, Leukocytes, B cells, T cells (CD4+, CD8+) or CD3-(Macrophages-Ly6Clow, Lyc6hi); CD11b- (Neutorphils and Ly6G-; Dentritic Cells and CD11c-).

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