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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	Flow cytometry data were collected with BD LSRFortessaTM X-20 using BD FACs Diva Software (BD Biosciences) or with Cytek® Aurora using SpectroFlo® software (Cytek Biosciences). Flow cytometry data files were analyzed using FlowJO software version 10.8.0 or 10.10.0 (Tree Star Inc., Ashland, OR, USA; BD). Light microscopy imaging data were acquired using Zeiss ZEN (blue edition) 3.1 software. Images from experiments involving culture in Matrigel were analyzed by Image J software (NIH) to quantify vascular cord length and branch number. Laser Doppler imaging data were collected and analyzed using moorLDI2 Research Software version v6.1 ((moorLD12-IR, Moor Instruments). Confocal microscopy data were acquired using LAS X software (Leica Microsystems), and immunofluorescence labeling images from some experiments analyzed using ImageJ software (NIH) to quantify cells expressing markers of interest. scRNA-seq raw data was analyzed using Cell Ranger v7.0.0 (10X Genomics). The processed data analyzed in R v4.1.3 using Seurat v4.1.1 R package and Monocle 3 R package. R packages Seurat, ggtree and ggplot2 were used for data visualization.
Data analysis	Statistical analyses were performed using Prism 9.0.0 or 10.0.2 (GraphPad).

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

A complete Data Availability statement has been included in the manuscript and can be seen below.

Data Availability

The single-cell RNA-seq data generated in this study has been deposited in the National Centre for Biotechnology Information Gene Expression Omnibus (83) under accession code GSE232625 (https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?&acc=GSE232625). The raw and/or processed flow cytometry, colony count, image analysis and qRT-PCR data are provided in the Supplementary Information and/or Source Data file. The raw imaging data generated in this study will be made available upon request. Source data are provided with this paper.

83. Edgar, R., Domrachev, M. & Lash, A.E. Gene Expression Omnibus: NCBI gene expression and hybridization array data repository. Nucleic Acids Res 30, 207-210 (2002).

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	Most experiments did not involve formal power calculation of sample sizes, but were performed using a minimal sample size of 3 different donor mice on at least two independent days. For adoptive transfer of aortic EndoMac progenitor cells into ischaemic hindlimb muscle, we determined that n=6 mice per group would allow us to detect a 40% increase in perfusion recovery with progenitors (0.40+/-0.10) compared to cell free control (0.24+/-0.10), with power of 80% and alpha 0.05. Both male and female mice were included in the study design for all experiments however data were not analysed on the basis of sex due to small sample sizes.
Data exclusions	When analyzing data using Prism (GraphPad) we applied the ROUT method to identify and exclude any outliers, with Q set to 1%. Rarely were any outlier data identified and excluded.
Replication	Experiments were performed using at least three different donor mice and on two or more independent experimental days. All data presented in the paper were reproducible.
Randomization	In the case of cell transplant and angiotensin II infusion experiments, mice were randomly allocated to a treatment group.
Blinding	Analysis was performed blinded where appropriate, including for the analysis of laser doppler scores, capillary density and arteriolar density in the hindlimb transfer experiments.

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
[ ]	[x] Antibodies
[x]	[ ] Eukaryotic cell lines
[x]	[ ] Palaeontology and archaeology
[ ]	[x] Animals and other organisms
[x]	[ ] Human research participants
[x]	[ ] Clinical data
[x]	[ ] Dual use research of concern

Methods

n/a	Involved in the study
[x]	[ ] ChIP-seq
[ ]	[x] Flow cytometry
[x]	[ ] MRI-based neuroimaging

Antibodies

Antibodies used	The list of all the antibodies used in this study with the sources and dilutions has been provided in Supplementary Data 12 in the Supplementary Data file.
Validation	All antibodies used for flow cytometry and immunofluorescence labeling were first validated and optimised in our own hands to determine the best working concentrations. Each experiment also used appropriate IgG isotype control antibodies and in the case of flow cytometry and FACS sorting, fluorescence-minus-one (FMO) controls also. Primary antibodies for immunofluorescence labelling were also validated by staining positive control tissues.

Animals and other organisms

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

Laboratory animals	The mouse strains used for in vitro and/or in vivo experiments, breeding strategies of lineage tracing mouse models and gender of mice used for experiments have been fully detailed in the Methods under 'Mouse Models' and in Supplementary Data 12. Ages of mice used for experiments has been specified in the figure legends and, where necessary, in the Results.
Wild animals	No wild animals were used in this study.
Field-collected samples	No field collected samples were used for this study.
Ethics oversight	The use of animals for this study was approved by the South Australian Health and Medical Research Institute Animal Ethics Committee (ID SAM117, SAM155, SAM308, SAM432.19 and SAM-23-008).

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

Flow Cytometry

Plots

Confirm that:

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

[x] 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).

[x] All plots are contour plots with outliers or pseudocolor plots.

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

Methodology

Sample preparation	Single cell suspensions from tissue digests or cultured-derived CFU-M were resuspended in aliquots of ≤ 10 x 10^6 cells/mL in PBS/5% FBS/0.2% sodium azide. 1 µL of fixable viability stain 700 (BD Horizon) was added to each sample and incubated for 15 min on ice. After washing, samples were blocked for 5 min on ice with purified rat anti-mouse CD16/CD32 (BD Pharmingen, San Diego CA, USA). Cells were then incubated for 30 min with primary antibodies and subsequently with secondary antibodies when required (Supplementary Data 12). Samples were then washed and fixed in formalin/PBS for analysis on a flow cytometer. For cell cycle analysis, BrdU Flow Kit (BD Biosciences) was used as per manufacturer's instructions.
Instrument	Flow cytometry was performed with BD LSRFortessaTM X-20 and BD FACs Diva Software (BD Biosciences) or Cytek® Aurora and SpectroFlo® software (Cytek Biosciences). A FACSAria Fusion cell sorter (BD Bioscience) was used for cell sorting.
Software	Data files were analyzed using FlowJO software version 10.8.0 or 10.10.0 (Tree Star Inc., Ashland, OR, USA; BD).
Cell population abundance	Purity checks of FACS sorted populations were performed by analysing the collected sample through the flow cytometer, as shown in Supplementary Figures 2C and 3F.

Gating strategy	Gating was performed based on FSC-A (to exclude cell debris), FSC-H vs FSC-A (for single cells), and viability stain selected cells (to exclude dead cells), and used Fluorescence-minus-one (FMO) controls to determine the positive percentage expression of different surface markers.

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