

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 (<i>n</i>) 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 <i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i>
☐	☒ 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. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted <i>Give P values as exact values whenever suitable.</i>
☒	☐ 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 <i>d</i> , Pearson's <i>r</i>), 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	Single cell RNA sequencing data was collected using the 10x Chromium™ Single Cell 3' Solution (V3 chemistry) and DNBelab C Series Single-Cell Library Prep Set (MGI, 940-000507-00). Single-cell libraries constructed by Chromium single-cell kit were subjected to 150-bp paired-end sequencing on the Illumina NavoSeq6000 platform. The libraries constructed by DNBelab C Series Single-Cell Library Prep Set were then sequenced on a DNBSEQ platform at the China National GeneBank in Shenzhen, China. Sequencing was performed with a 47-bp read length for read 1 and a 100-bp read length for read 2. Flow cytometry was performed using Aurora Spectrum flow cytometry (Cytek Biosciences) with SpectroFlo (Cytek, V3.1.0).
Data analysis	For scRNA-seq, After obtaining the paired-end raw reads, we used CellRanger (10x Genomics, v3.1.0) to preprocess the single-cell RNA-seq data. The cell barcodes and unique molecular identifiers (UMIs) of the library were extracted from read1. Then, the reads were split according to their cell (barcode) IDs, and the UMI sequences from read2 were simultaneously recorded for each cell. Quality control of these raw readings was subsequently performed to eliminate adapter contamination, duplicates, and low-quality bases. After filtering barcodes and low-quality readings that were not related to cells, we aligned the clean reads to the human genome, and we retained the uniquely mapped reads for UMI counts. Next, we estimated the molecular counts and generated a UMI count matrix for each cell by counting the UMIs for each sample. Finally, we generated a gene-barcode matrix that showed the barcoded cells and gene expression counts. Based on the number of total reads, the number of detected gene features, and the percentage of mitochondrial genes, we performed quality control filtering through Seurat (v4.9.9.9045) to discard low-quality cells. Briefly, cells with the gene number less than 400 or the proportion of mitochondrial genes greater than 15% were removed. To remove doublets, cells with greater than 8000 genes were first removed. Then we used DoubletFinder to further detect the doublets. Then single cells that passed the QC criteria were used for downstream analyses. Seurat objects from all samples were integrated using canonical correlation analysis (CCA) with 2000 features to identify integration anchors and remove batch effects. The same method was also used to remove batch effects between in-house data and public datasets. For heatmap and violin plots, the SCT-transformed data from the Seurat pipeline were used. Using the Seurat 'FindMarkers' function, we found differentially expressed genes

(DEGs) between the two cell types. We also used the R package clusterProfiler (v4.4.4) with the default parameters to identify gene sets that exhibited significant and consistent differences between two given biological states. Raw count matrices of ADSC subsets were used to estimate their developmental trajectories using the Monocle2 package (version 2.32.0) in R. We investigated cell–cell interactions and receptor–ligand pairs using CellChat (version 1.6.1). Mitochondrial reads for each ADSC and endothelial cell were subsequently extracted from BAM files using the pysam package (v0.21.0). For data visualization, t-SNE was performed using the Rtsne package (v0.16), followed by hierarchical clustering via the hclust function and the construction of an evolutionary tree using the ggtree package (v3.8.2).

Flow cytometry analysis was performed by SpectroFlo (Cytek, V3.1.0) and FlowJo (V10.8.1). Immunofluorescence quantification was analysed by ImagePro Plus (Ver. 6.0, Media Cybernetics).

For immunofluorescence staining of human samples, the stained tissue was scanned with a confocal microscope (TCS-SP8 gSTED 3X, Leica) and viewed with Leica Application Suite X (Ver. 3.7.4, Leica). The positively stained area of the carotid PVAT cross-sections was quantified with ImagePro Plus (Ver. 6.0, Media Cybernetics).

For histological analysis, the relative abundances of type I collagen (yellow-red birefringence) and type III collagen (green birefringence) were distinguished and quantified based on their birefringence patterns under polarized light using Image-Pro Plus software (v. 6.0). For morphological analysis of adipocytes, adjacent sections were stained with hematoxylin and eosin (H&E) constant dye kit (G1076, Servicebio, China). The size of adipocytes was measured in randomly selected fields from each sample using ImageJ software (v. 1.45g).

For immunofluorescence staining of animal studies. Image analysis and quantification were conducted by two independent, blinded researchers using the computer-based image analysis systems Cell Profiler (v4.2.8, Broad Institute) and Image Pro Plus (version 6.0, Media Cybernetics).

For tube formation assay, the resulting networks were imaged with an inverted microscope, and the total segment length was quantified using the ImageJ (v. 1.45g) Angiogenesis Analyzer plugin.

For transwell assay, the number of cells was determined using Image Pro Plus (version 6.0, Media Cybernetics).

Western blot band intensities were measured using ImageJ software (v. 1.45g).

GraphPad Prism software (ver. 9.5.0, GraphPad, USA) or R software (<https://www.r-project.org/>) were used for statistical calculation and figure presentation.

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](#)

The single-cell sequencing data could be obtained from the GSA-human database (<https://ngdc.cnbc.ac.cn/gsa-human/>) with accession number HRA001023 and CNGB Sequence Archive (CNSA) of the China National GeneBank DataBase (CNGBdb) (<https://db.cngb.org/cnsa>) with the accession number: CNP0004894. A GitHub repository containing the complete source code and analytical pipelines for our scRNA-seq analysis, includes detailed documentation of all state-of-the-art computational methods and packages used in this study. Access is available at: <https://github.com/SongMei233/PVAT>. The processed data and analysis codes are available upon reasonable request from the corresponding author.

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

Gender classification was based on patient self-reporting. The study design did not incorporate gender-based stratification as participant selection was determined by surgical eligibility and specific inclusion criteria. Our findings did not reveal a clear influence of gender on the distinction between symptomatic and asymptomatic presentations of carotid artery stenosis.

Reporting on race, ethnicity, or other socially relevant groupings

This research did not provide any details regarding the social or ethnic background of the human participants.

Population characteristics

Patients characteristics and covariates are reported in Supplementary Table 2.

Recruitment

Patients with asymptomatic carotid artery stenosis with a stenosis degree of 70%~99% or symptomatic carotid artery stenosis with a degree of 50%~99% (North American Symptomatic Carotid Endarterectomy, NASCET criteria) who underwent standard carotid endarterectomy (CEA) or reversed carotid endarterectomy (eversionCEA, eCEA) and patients diagnosed with benign carotid body tumours who underwent carotid body tumour resection with revascularization (healthy donors, control group) in the Department of Vascular Surgery, Peking Union Medical College Hospital, were included in the cohort (Cohort of Optimized Assessment Strategy of Carotid Artery Stenosis, COAS-CAS, NCT05629000). Eligible subjects gave informed written consent before enrolling in this study. Patients who smoked one cigarette or more per day for one year or more were considered smokers.

Ethics oversight

The study was approved by the Ethics Review Committee of Peking Union Medical College Hospital (No. JS-2966 and No.

I-22PJ019).

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](https://www.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	For this investigation, a formal sample size calculation was not conducted. Instead, the sample size was ascertained based on the availability and dimensions of the tissue specimens.
Data exclusions	Six samples from immunofluorescence verification were excluded due to the predominance of perivascular lymphoid node tissue within the specimens.
Replication	In the single-cell sequencing, a total of 10 samples were observed, with 7 samples in the carotid artery stenosis group and 3 samples in the carotid body tumor control group. Flow cytometry included a total of 21 cases, with 14 duplicates in the carotid artery stenosis group and seven duplicates in the carotid body tumor control group. Pathological staining encompassed 142 samples, with 125 duplicates in the carotid artery stenosis group and 17 duplicates in the carotid body tumor control group. Among these, 98 cases underwent both Sirius red staining and H&E staining. For each sample subjected to immunofluorescence staining, the staining procedure was performed in duplicate. Three samples from immunofluorescence staining were shared with flow cytometry, and two samples were shared with single-cell sequencing.
Randomization	Samples for single-cell sequencing, flow cytometry and pathological staining were collected using a prospective random sampling method at the time of patient enrollment. Five to nine random photographs of pathological staining in each sample were taken for statistical analysis.
Blinding	Researchers processing the carotid samples were blinded. Flow cytometry analyses and pathological quantification analyses were performed blindly.

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

For full spectrum flow cytometry:

1. Anti-human CD142-BV421 (dilution 2 µl/100 µl, BD, 744003, HTF-1)
2. Anti-human Glypican 3-PE (dilution 2 µl/100 µl, Abcam, ab275697, 024)
3. Anti-human CD55-BV786 (dilution 2 µl/100 µl, BD, 742681, IA10)
4. Anti-human CD45-APC-Fire810 (dilution 2 µl/100 µl, Biolegend, 304076, HI30)
5. Anti-human CD3-APC-Fire750 (dilution 2 µl/100 µl, Biolegend, 300470, UCHT1)
6. Anti-human CD4-R718 (dilution 2 µl/100 µl, BD, 566930, SK3)
7. Anti-human CD279-BV605 (dilution 2 µl/100 µl, BD, 745139, MIH4)
8. Anti-human CD8-FITC (dilution 2 µl/100 µl, Biolegend, 344704, SK1)
9. Anti-human CD74-BV711 (dilution 2 µl/100 µl, BD, 743735, LN2)
10. Anti-human CD19-PerCP-Cy5.5 (dilution 2 µl/100 µl, Biolegend, 363016, SJ25C1)
11. Anti-human CD56-BV480 (dilution 2 µl/100 µl, BD, 746586, MY31)
12. Anti-human CD16-Pacific Blue (dilution 2 µl/100 µl, Biolegend, 302032, 3G8)

13. Anti-human CD14-BV650 (dilution 2 µl/100 µl, Biolegend, 301836, M5E2)
14. Anti-human CD163-PE-Cy7 (dilution 2 µl/100 µl, Biolegend, 326514, RM3/1)
15. Anti-human MRP-14 (S100A9) APC (dilution 2 µl/100 µl, Biolegend, 350708, MRP, 1H9)

For cell sorting:

1. Anti-mouse/rat CD29-APC (dilution 2 µl/100 µl, 102215, Biolegend, HMβ1-1)
2. Anti-mouse CD45-PerCP/Cyanine5.5 (dilution 2 µl/100 µl, 103132, Biolegend, 30-F11)
3. Anti-mouse CD55 (DAF)-PE (dilution 2 µl/100 µl, 131803, Biolegend, RIKO-3)
4. Anti-mouse Ly-6A/E (Sca-1)-BV421 (dilution 2 µl/100 µl, 108128, Biolegend, D7)
5. Anti-human CD34-APC (dilution 2 µl/100 µl, 343607, Biolegend, 4H11)
6. Anti-human CD45-BV510 (dilution 2 µl/100 µl, 304035, Biolegend, HI30)
7. Anti-human CD142-BV421 (dilution 2 µl/100 µl, 744003, BD, HTF-1)
8. Anti-human CD55-BV786 (dilution 2 µl/100 µl, 742681, BD, IA10)
9. Anti-human GPC3-PE (dilution 2 µl/100 µl, ab275697, Abcam, 024)

For immunofluorescence:

1. Anti-human CD142 (dilution 1:200, ab228968, Abcam, EPR22548-232)
2. Anti-human CD55 (dilution 1:200, A22324, Abclonal, ARC52244)
3. Isolectin B4 (dilution 5 µg/ml, GS-IB4, Flexa Fluor 488, I21411, Invitrogen)
4. Anti-human CD31 (dilution 1:200, GB153151, Servicebio, SB166).
5. Anti-human/mouse PDGFRA (dilution 1:200, ab203491, Abcam, EPR22059-270)
6. Anti-human/mousePDGFRB (dilution 1:200, A19531, Abclonal, ARC0009)
7. Anti-human GPC3 (dilution 1:200, A11686, Abclonal, ARC0675)
8. FlexAble CoraLite Plus 555 (dilution 1:500, KFA002, Proteintech)
9. FlexAble CoraLite Plus 647 (dilution 1:500, KFA003, Proteintech)
10. Alexa Fluor 488 Goat anti-Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody (dilution 1:500, A11008, Invitrogen)
11. Anti-mouse CD31 (dilution 1:200, GB15063, Servicebio, SB149)
12. Goat Anti-Mouse IgG H&L (Alexa Fluor® 555) preadsorbed (dilution 1:500, ab150118, Abcam)
13. Anti-human CD31 (dilution 1:200, GB12064, Servicebio, 3A9C6)
14. Anti-human/mouse GFP (dilution 1:200, ab252881, Abcam, 3H9)
15. Goat Anti-Rat IgG H&L (Alexa Fluor® 647) preadsorbed (ab150167, Abcam)
16. Goat Anti-Rat IgG H&L (Alexa Fluor® 488) preadsorbed (ab150165, Abcam)

For Western blot:

1. PDGFRA (1:1000, ab203491, Abcam, EPR22059-270)
2. PDGFRB (1:1000, A19531, Abclonal, ARC0009)
3. ACTA2 (1:1000, A17910, Abclonal, ARC0298)
4. TAGLN/SM22 (1:1000, 60213, Proteintech, 2A10C2)
5. β-actin (1:1000, 66009-1-Ig, Proteintech, 2D4H5)
6. HRP-conjugated secondary antibodies goat anti-rabbit (1:10000, ZB-2301, ZSGB-BIO)
7. HRP-conjugated secondary antibodies goat anti-rabbitgoat anti-mouse (1:10000, ZB-2305, ZSGB-BIO)

Validation

For full spectrum flow cytometry:

1. CD142-BV421 (BD, 744003, HTF-1): <https://www.bdbiosciences.com/zh-cn/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/bv421-mouse-anti-human-cd142.744003>.
2. Glypican 3-PE (Abcam, ab275697, 024): <https://www.abcam.cn/products/primary-antibodies/pe-glypican-3-antibody-024-ab275697.html>.
3. CD55-BV786 (BD, 742681, IA10): <https://www.bdbiosciences.com/zh-cn/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/bv786-mouse-anti-human-cd55.742681>.
4. CD45-APC-Fire810 (Biolegend, 304076, HI30): <https://www.biolegend.com/en-us/products/apc-fire-810-anti-human-cd45-antibody-19538>.
5. CD3-APC-Fire750 (Biolegend, 300470, UCHT1): <https://www.biolegend.com/en-us/products/apc-fire-750-anti-human-cd3-antibody-13562>.
6. CD4-R718 (BD, 566930, SK3): <https://www.bdbiosciences.com/zh-cn/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/r718-mouse-anti-human-cd4.566930>.
7. CD279-BV605 (BD, 745139, MIH4): <https://www.bdbiosciences.com/zh-cn/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/bv605-mouse-anti-human-cd279-pd-1.745139>.
8. CD8-FITC (Biolegend, 344704, SK1): <https://www.biolegend.com/en-us/products/fitc-anti-human-cd8-antibody-6149>
9. CD74-BV711 (BD, 743735, LN2): <https://www.bdbiosciences.com/zh-cn/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/bv711-mouse-anti-human-cd74.743735>.
10. CD19-PerCP-Cy5.5 (Biolegend, 363016, SJ25C1): <https://www.biolegend.com/en-us/products/percp-cyanine5-5-anti-human-cd19-antibody-10305>.
11. CD56-BV480 (BD, 746586, MY31): <https://www.bdbiosciences.com/zh-cn/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/bv480-mouse-anti-human-cd56.746586>.
12. CD16-Pacific Blue (Biolegend, 302032, 3G8): <https://www.biolegend.com/en-us/products/pacific-blue-anti-human-cd16-antibody-2854>.
13. CD14-BV650 (Biolegend, 301836, M5E2): <https://www.biolegend.com/en-us/products/brilliant-violet-650-anti-human-cd14-antibody-7819>.
14. CD163-PE-Cy7 (Biolegend, 326514, RM3/1): <https://www.biolegend.com/en-us/products/pe-cyanine7-anti-human-cd163-antibody-10756>.
15. MRP-14 (S100A9) APC (Biolegend, 350708, MRP, 1H9): <https://www.biolegend.com/en-us/products/apc-anti-human-mrp-14-s100a9-antibody-10079>.

For cell sorting:

1. Anti-mouse/rat CD29-APC (dilution 2 µl/100 µl, 102215, Biolegend, HMβ1-1): <https://www.biolegend.com/en-us/products/apc-anti-mouse-rat-cd29-antibody-3090>.
2. Anti-mouse CD45-PerCP/Cyanine5.5 (dilution 2 µl/100 µl, 103132, Biolegend, 30-F11): <https://www.biolegend.com/en-us/products/percp-cyanine5-5-anti-mouse-cd45-antibody-4264>.
3. Anti-mouse CD55 (DAF)-PE (dilution 2 µl/100 µl, 131803, Biolegend, RIKO-3): <https://www.biolegend.com/en-us/products/pe-anti-mouse-cd55-daf-antibody-5514>.
4. Anti-mouse Ly-6A/E (Sca-1)-BV421 (dilution 2 µl/100 µl, 108128, Biolegend, D7): <https://www.biolegend.com/en-us/products/brilliant-violet-421-anti-mouse-ly-6a-e-sca-1-antibody-7198>.
5. Anti-human CD34-APC (dilution 2 µl/100 µl, 343607, Biolegend, 4H11): <https://www.biolegend.com/en-us/products/apc-anti-human-cd34-antibody-6204>.
6. Anti-human CD45-BV510 (dilution 2 µl/100 µl, 304035, Biolegend, HI30): <https://www.biolegend.com/en-us/products/brilliant-violet-510-anti-human-cd45-antibody-8006>.
7. Anti-human CD142-BV421 (dilution 2 µl/100 µl, 744003, BD, HTF-1): <https://www.bdbiosciences.com/zh-cn/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/bv421-mouse-anti-human-cd142.744003>.
8. Anti-human CD55-BV786 (dilution 2 µl/100 µl, 742681, BD, IA10): https://www.bdbiosciences.com/zh-cn/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/bv786-mouse-anti-human-cd55.742681?tab=product_details.
9. Anti-human GPC3-PE (dilution 2 µl/100 µl, ab275697, Abcam, 024): <https://www.abcam.cn/products/primary-antibodies/pe-glypican-3-antibody-024-ab275697>.

For immunofluorescence:

1. Anti-human CD142 (dilution 1:200, ab228968, Abcam, EPR22548-232): <https://www.abcam.cn/products/primary-antibodies/tissue-factor-antibody-epr22548-232-ab228968.html>.
2. Anti-human CD55 (dilution 1:200, A22324, Abclonal, ARC52244): <https://abclonal.com.cn/catalog/A22324>.
3. Isolectin B4 (dilution 5 µg/ml, GS-IB4, Flexa Fluor 488, I21411, Invitrogen): <https://www.thermofisher.cn/order/catalog/product/I21411?SID=srch-srp-I21411>.
4. CD31 (1:200, GB153151, Servicebio, SB166): <https://www.service-bio.com/goodsdetail?id=22007>.
5. PDGFRA (1:200, ab203491, Abcam, EPR22059-270): <https://www.abcam.cn/products/primary-antibodies/pdgfr-alpha-antibody-epr22059-270-ab203491>.
6. PDGFRB (1:200, A19531, Abclonal, ARC0009): <https://abclonal.com.cn/catalog/A19531>.
7. GPC3 (1:200, A11686, Abclonal, ARC0675): <https://abclonal.com.cn/Datasheet/Antibodies/A11686.pdf>.
8. FlexAble CoraLite Plus 555 (1:500, KFA002, Proteintech): <https://www.ptgcn.com/products/FlexAble-CoraLite-Plus-555-Antibody-Labeling-Kit-for-Rabbit-IgG-KFA002.htm>.
9. FlexAble CoraLite Plus 647 (1:500, KFA003, Proteintech): <https://www.ptgcn.com/products/FlexAble-CoraLite-Plus-647-Antibody-Labeling-Kit-for-Rabbit-IgG-KFA003.htm>.
10. Alexa Fluor 488 Goat anti-Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody (1:500, A11008, Invitrogen): <https://www.thermofisher.cn/cn/zh/antibody/product/Goat-anti-Rabbit-IgG-H-L-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-11008>.
11. Anti-mouse CD31 (dilution 1:200, GB15063, Servicebio, SB149): <https://www.service-bio.com/goodsdetail?id=21920>.
12. Goat Anti-Mouse IgG H&L (Alexa Fluor® 555) preadsorbed (dilution 1:500, ab150118, Abcam): <https://www.abcam.cn/products/secondary-antibodies/goat-mouse-igg-h-l-alex-fluor-555-preadsorbed-ab150118>.
13. Anti-human CD31 (dilution 1:200, GB12064, Servicebio, 3A9C6): <https://www.service-bio.com/goodsdetail?id=4868>.
14. Anti-human/mouse GFP (dilution 1:200, ab252881, Abcam, 3H9): <https://www.abcam.cn/products/primary-antibodies/gfp-antibody-3h9-ab252881>.
15. Goat Anti-Rat IgG H&L (Alexa Fluor® 647) preadsorbed (ab150167, Abcam): <https://www.abcam.cn/products/secondary-antibodies/goat-rat-igg-h-l-alex-fluor-647-preadsorbed-ab150167>.
16. Goat Anti-Rat IgG H&L (Alexa Fluor® 488) preadsorbed (ab150165, Abcam): <https://www.abcam.cn/products/secondary-antibodies/goat-rat-igg-h-l-alex-fluor-488-preadsorbed-ab150165>.

For Western blot:

1. PDGFRA (1:1000, ab203491, Abcam, EPR22059-270): <https://www.abcam.cn/products/primary-antibodies/pdgfr-alpha-antibody-epr22059-270-ab203491>.
2. PDGFRB (1:1000, A19531, Abclonal, ARC0009): <https://abclonal.com.cn/catalog/A19531>.
3. ACTA2 (1:1000, A17910, Abclonal, ARC0298): <https://abclonal.com.cn/catalog/A17910>.
4. TAGLN/SM22 (1:1000, 60213-1-Ig, Proteintech, 2A10C2): <https://www.ptgcn.com/products/TAGLN-Antibody-60213-1-Ig.htm>.
5. β-actin (1:1000, 66009-1-Ig, Proteintech, 2D4H5): <https://www.ptgcn.com/products/Pan-Actin-Antibody-66009-1-Ig.htm>.
6. HRP-conjugated secondary antibodies goat anti-rabbit (1:10000, ZB-2301, ZSGB-BIO): <http://www.zsbio.com/product/ZB-2301>.
7. HRP-conjugated secondary antibodies goat anti-rabbitgoat anti-mouse (1:10000, ZB-2305, ZSGB-BIO): <http://www.zsbio.com/product/ZB-2305>.

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

Six-week-old male C57BL/6J mice and male BALB/cNj-Foxn1nu/Gpt nude mice were used in this study.

Wild animals

This study did not involve wild animals.

Reporting on sex

Only 6-week-old male C57 mice and male BALB/cNj-Foxn1nu/Gpt nude mice were used in this study. Males were selected to avoid the confounding effects of hormonal fluctuations in females on vascular remodeling outcomes. As the study was not designed to investigate sex-specific differences in ADSC function or vascular remodeling, sex-based analyses were not performed. A total of 15 male C57 mice and 16 male BALB/cNj-Foxn1nu/Gpt nude mice were included; no female animals were used. The potential influence of sex on the findings warrants investigation in future studies.

Field-collected samples	This study did not involve samples collected from the field.
Ethics oversight	The study protocol was approved by the Committee of Ethics on Animal Experiments at Peking Union Medical College (No. ACUC-A02-2024-006) for animal experiments.

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	NCT05629000
Study protocol	The study protocol: https://clinicaltrials.gov/study/NCT05629000?distance=50&intr=NCT05629000&rank=1
Data collection	This study collected clinical baseline data, CTA or MRI imaging data, and surgical samples—including carotid plaques, perivascular adipose tissue—from patients enrolled between May 2021 and July 2025 in Peking Union Medical College Hospital.
Outcomes	Perioperative stroke, cardiac infarction, transient ischaemic attack, cerebral hyperperfusion syndrome and death (During hospital stay, 7-30 days post operation) were defined as the primary outcomes of this clinical trial. The outcome data was not covered in the current study.

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	PVAT was cut with ophthalmic scissors into 1-mm pieces and digested with 0.05% type II collagenase (Gibco) and 0.5 µl/ml benzonase nuclease (PM204-01, Biomed, China) at 37°C with shaking for 20 min. The digested tissue was passed through a 40 µm cell filter. The remaining undigested tissue on the filter was aspirated, and the above digestion steps were repeated for another 20 min. After centrifugation, the supernatant was discarded and resuspended to obtain a single-cell suspension. Red blood cell lysis buffer was applied if necessary. The cells were resuspended in 3 ml of DPBS, and viability was measured.
Instrument	Cells were resuspended in 0.2 ml of PBS and analysed by Aurora Spectrum flow cytometry (Cytek Biosciences).
Software	Flow cytometry analyses were conducted by SpectroFlo (Cytek, V3.1.0) and FlowJo (V10.8.1).
Cell population abundance	We performed lentiviral transduction on the ADSCs to enable GFP expression. The transduction efficiency was assessed by evaluating GFP signals under a microscope to determine the sorted cell population abundance. Additionally, Western blot analysis was carried out to detect the expression of PDGFRA and PDGFRB (mesenchymal stem cell markers) in the sorted cells, in order to confirm the maintenance of their stemness.
Gating strategy	Cells were gated on SSC-A/FSC-A to remove debris. Then the cells were gated on FSC-H/FSC-A and SSC-H/SSC-A to remove cell aggregates. SSC-B-A/SSC-A were employed to remove red blood cells. Dead cells were removed by Zombie NIR dye. Flow cytometry analysis of human perivascular adipose tissue (PVAT) identified immune and stromal cell populations, including macrophages (CD45+CD14+CD163+), monocytes (CD45+CD14+S100A9+), NK cells (CD45+CD3-CD56+), B cells

(CD45+CD19+), CD4+ T cells (CD45+CD3+CD4+), CD8+ T cells (CD45+CD3+CD8+), as well as two distinct adipose-derived stromal cell (ADSC) subsets: CD55+ ADSCs (CD45-CD34+CD142+CD55+) and CXCL14+ ADSCs (CD45-CD34+CD142+GPC3+). Correspondingly, the sorting strategies for these ADSC subpopulations were defined separately for mouse and human sources. For mouse-derived ADSCs, the CD55+ subset was sorted as CD45-CD29+Sca-1+CD55+ cells, while the CXCL14+ subset was defined as CD45-CD34+CD142+CD55- cells. For human-derived ADSCs, the sorting markers mirrored the analysis panel, with CD55+ ADSCs isolated as CD45-CD34+CD142+CD55+ cells and CXCL14+ ADSCs as CD45-CD34+CD142+GPC3+ cells.

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