nature portfolio

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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	Light cycler 480 (Roche version 1.5.1), BZ-II viewer (Keyence v.2.1), BZ-II analyzer (Keyence v1.42), BZ-II analyzer (Keyence v1.42), Nivo (Perkin Elmer ver 3.0.2), Image J (version 10.2), Integrated genome viewer (version 2.50), ID 7000 (SONY version 1.1.0.11041), Living Image software (Perkin Elmer ver 4.5), Time OFCR1 (O'Hara & Co.,Ltd), O2/CO2 metabolic measurement system (Model MK-5000), Muromachi Kikai), FaQCs (version 1.34), TopHat analysis (version 2.0.13), Cuffdiff program (version 2.2.1), TrimGalore (v0.5.0), BWA (v0.7.17), MACS (v1.4.3), Picard (v2.18.20), TMHMM2.0 (the KEGG GENES database, May 2, 2014)
Data analysis	Data were analyzed with SPSS version 24 or 25 software.

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

RNAseq data; Gene expression data obtained in these studies were deposited in the Gene Expression Omnibus database (GSE155680 for HUVECs and GSE155596 for ApoE KO mice).

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	Sample sizes were determined based on previously published experiments where differences were observed (Aging mod; Nature. 2011 Nov 2;479(7372):232-6. Nat Med. 2018 Aug;24(8):1246-1256. Atherosclerotic assay; Science. 2016 Oct 28;354(6311):472-477. Metabolic assay; Nat Med. 2009 Sep;15(9):1082-7. Vascular assay; J Mol Cell Cardiol. 2019 Apr;129:105-117.) or were calculated by using mean and standard deviation from the preliminary experiments.
Data exclusions	All values were included for statistical analyses. Outliers were evaluated by boxplot analyses with SPSS version 24 or 25 software. Outliers in Figure 1h, 3d, and Extended data figure 3c were excluded from the analysis. Data exclusions are clearly described in each figure legend, in Excel format source data files.
Replication	All data were from different biological replicates as indicated in each figure legend and all findings were reliably reproduced in several independent cohorts.
Randomization	All mice were randomly allocated for treatments and surgical procedures under blinded condition. For cell culture experiments, wells of cultured cells were sequentially assigned to distinct treatment groups.
Blinding	Investigators were blinded to mouse genotypes during experiments. All animal models were generated and analyzed under blinded condition. Other experiments including in vitro study, investigators were blinded to allocation during experiments and outcome assessments.

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

anti-human GPNMB antibody; used in western blotting, immunoprecipitation, Cell Signaling, #13251, [lot number 1].
anti-human GPNMB antibody; used in immunohistochemistry, Abcam, ab175427, [lot number GR256360-16].
anti-human p53 antibody; used in western blotting, Santa Cruz, sc-126, [lot number F2811].
anti-human p21 antibody; used in western blotting, Abcam, ab7960, [lot number GR175274-2].
anti-human p16 antibody; used in western blotting, BD, 554079, [lot number 17926].
anti-β actin antibody (13E5); used in western blotting, Cell Signaling, #4970, [lot number 9].
anti-mouse Gpnmb antibody; used in western blotting, R&D, AF2330, [lot number ULH0211101].
anti-mouse p53 antibody (1C12); used in western blotting, Cell Signaling, #2524, [lot number 13].
anti-mouse p21 antibody; used in western blotting, Carbiochem, OP64-20UG, [lot number D00141439].
anti-α-tubulin antibody (11H10); used in western blotting, Cell Signaling, #2125, [lot number 11].
anti-GAPDH antibody; used in western blotting, Santa Cruz, sc-20357, it has been discontinued.
BV421-conjugated anti-mouse Cd31 antibody; used in flow cytometry, BioLegend, 102408, [B301862]
PE/Cy7-conjugated anti-mouse Cd45 antibody; used in flow cytometry, BioLegend, 103114, [B308464]
BB700-conjugated anti-mouse Cd11b antibody; used in flow cytometry, BD, 564454, [9093647]
BV510-conjugated anti-mouse Cd3 antibody; used in flow cytometry, BioLegend, 100234, [B320432]
BV605-conjugated anti-mouse Cd19 antibody; used in flow cytometry, BioLegend, 115539, [B324619]
APC/Fire750-conjugated anti-mouse Nk 1.1 antibody; used in flow cytometry, BioLegend, 108752, [B326399]

BV650-conjugated anti-mouse F4/80 antibody; used in flow cytometry, BioLegend, 123149, [B309664]
Pacific Blue-conjugated anti-mouse Ly-6G antibody; used in flow cytometry, BioLegend, 127611, [B288475]
BV785-conjugated anti-mouse Cd14 antibody; used in flow cytometry, BioLegend, 123337, [B321510]
BV605-conjugated anti-mouse Cd11c antibody; used in flow cytometry, BioLegend, 117333, [B316089]
BV711-conjugated anti-mouse Icam1 (Cd54) antibody; used in flow cytometry, BioLegend, 116143, [B317417]
eFluor660-conjugated anti-mouse Gpnmb antibody; used in flow cytometry, eBioscience, 50-5708-82, [4314688]
eFluor660-conjugated anti-mouse IgG2ak Isotype control antibody; used in flow cytometry, eBioscience, 50-4321-82, [4331769]
goat anti-mouse IgG H+L (Cy5) secondary antibody; used in immunostaining, Abcam, ab6563, [GR274558-28]
biotin-XX isolectin GS-IB4 conjugate; used in immunostaining, Invitrogen, I21414
rat anti-mouse CD45; used in cell isolation, BD, BD550539, [8002547]
rat anti-mouse CD31; used in cell isolation, BD, BD553369, [0156461]
Dinabeads sheep anti-Rat IgG; used in cell isolation, Invetrogen, 11035, [00862456]
peroxidase-conjugated AffiniPure goat anti-mouse IgG (H+L); used in western blotting, Jackson ImmunoResearch, 115-035-003
peroxidase-conjugated AffiniPure goat anti-rabbit IgG (H+L); used in western blotting, Jackson ImmunoResearch, 111-035-003, [150783]
peroxidase-conjugated AffiniPure donkey anti-goat IgG (H+L); used in western blotting, Jackson ImmunoResearch, 705-035-147, [53331]

Validation

anti-human GPNMB antibody(Cell Signaling, #13251); validated to detect human GPNMB for use in western blotting stated on the manufacturer's website.
anti-human GPNMB antibody(Abcam, ab175427); validated to detect human GPNMB for use in immunohistochemistry stated on the manufacturer's website.
anti-human p53 antibody (Santa Cruz, sc-126); validated to detect human p53 for use in western blotting stated on the manufacturer's website.
anti-human p21 antibody (Abcam, ab7960); validated to detect human p21 for use in western blotting stated on the manufacturer's website.
anti-human p16 antibody (BD, 554079); validated to detect human p16 for use in western blotting stated on the manufacturer's website.
anti-β actin antibody (Cell Signaling, #4970); validated to detect human and mouse β actin for use in western blotting stated on the manufacturer's website.
anti-mouse Gpnmb antibody(R&D, AF2330); validated to detect mouse Gpnmb for use in western blotting stated on the manufacturer's website.
anti-mouse p53 antibody (1C12) (Cell Signaling, #2524); validated to detect mouse p53 for use in western blotting stated on the manufacturer's website.
anti-mouse p21 antibody (Carbiochem, OP64-20UG); validated to detect mouse p21 for use in western blotting stated on the manufacturer's website.
anti-α-tubulin antibody (11H10) (Cell Signaling, #2125); validated to detect human and mouse α-tubulin for use in western blotting stated on the manufacturer's website.
anti-GAPDH antibody (Santa Cruz, sc-20357); validated to detect human and mouse GAPDH for use in western blottingstated on the manufacturer's website.
BV421-conjugated anti-mouse Cd31 antibody (BioLegend, 102408); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
PE/Cy7-conjugated anti-mouse Cd45 antibody (BioLegend, 103114); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
BB700-conjugated anti-mouse Cd11b antibody (BD, 564454); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
BV510-conjugated anti-mouse Cd3 antibody (BioLegend, 100234); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
BV605-conjugated anti-mouse Cd19 antibody (BioLegend, 115539); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
APC/Fire750-conjugated anti-mouse Nk 1.1 antibody (BioLegend, 108752); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
BV650-conjugated anti-mouse F4/80 antibody (BioLegend, 123149); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
Pacific Blue-conjugated anti-mouse Ly-6G antibody (BioLegend, 127611); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
BV785-conjugated anti-mouse Cd14 antibody (BioLegend, 123337); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
BV605-conjugated anti-mouse Cd11c antibody (BioLegend, 117333); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
BV711-conjugated anti-mouse Icam1 (Cd54) antibody (BioLegend, 116143); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
eFluor660-conjugated anti-mouse Gpnmb antibody (eBioscience, 50-5708-82); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
eFluor660-conjugated anti-mouse IgG2ak Isotype control antibody (eBioscience, 50-4321-82); validated to detect mice I13 for use in flow cytometry stated on the manufacturer's website.
goat anti-mouse IgG H+L (Cy5) secondary antibody (Abcam, ab6563); validated to detect mouse IgG for use in immunostaining stated on the manufacture's website
biotin-XX isolectin GS-IB4 conjugate(Invitrogen, I21414); validated to detect endothelial cells for use in immunostaining stated on the manufacture's website
rat anti-mouse CD45 (BD, BD550539); validated to detect mouse CD45 for use in flow cytometory stated on the manufacture's website
rat anti-mouse CD31(BD, BD553369); validated to detect mouse CD31 for use in flow cytometory stated on the manufacture's website
Dinabeads sheep anti-Rat IgG (Invetrogen, 11035); validated to detect rat IgG for use in MACS stated on the manufacture's website
peroxidase-conjugated AffiniPure goat anti-mouse IgG (H+L) (Jackson ImmunoResearch, 115-035-003); validated to detect mouse

IgG (H+L) for use in western blotting stated on the manufacture's website
peroxidase-conjugated AffiniPure goat anti-rabbit IgG (H+L) (Jackson ImmunoResearch, 111-035-003); validated to detect rabbit IgG (H+L) for use in western blotting stated on the manufacture's website
peroxidase-conjugated AffiniPure donkey anti-goat IgG (H+L) (Jackson ImmunoResearch, 705-035-147); validated to detect goat IgG (H+L) for use in western blotting stated on the manufacture's website

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	Human Umbilical Vein Endothelial Cells (HUVEC) was purchased from Lonza Japan (CC-2517, lot 0000439577).
Authentication	HUVECs were verified by qPCR and immunostaining of CD31, and tube formation assay (Cell Biolabs, Inc.).
Mycoplasma contamination	HUVEC cell line were cirtified to be mycoplasma-negative by Lonza.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used in the study.

Animals and other organisms

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

Laboratory animals	C57BL/6 mice were purchased from SLC Japan (Shizuoka, Japan). The DBA/2J and DBA/2J-Gpnmb+ mice were obtained from the Jackson Laboratory. Mice were maintained in a pathogen-free facility at 20–26 °C, 40-60% humidity, under a 12-h light, 12-h dark regimen. We used both male and female mice for the lifespan experiment. For other experiments, we used male mice from 4-20 weeks of age. Description of research mice used for experiments were also described in the relevant Figure legends and/or Methods.
Wild animals	No wild animals were used in the study.
Field-collected samples	No field collected samples were used in the study.
Ethics oversight	All of the animal experiments were conducted in compliance with the protocol reviewed by the Institutional Animal Care and Use Committee of Niigata University and approved by the Institutional Animal Care and Use Committee of Niigata University and the President of Niigata University.

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	For immunohistochemistry of human aorta, samples were obtained from patients with or without atherosclerotic diseases who admitted to the Department of Cardiovascular Surgery, Niigata University Medical and Dental Hospital, Japan, and were registered to our biobank between 2015-2019. For qPCR studies analyzing GPNMB, leukocytes were collected from patients with or without atherosclerotic diseases who admitted to the Department of Cardiovascular Medicine, Niigata University Medical and Dental Hospital, Japan, and were registered to our biobank between 2013-2015. A total of 70 unique patients (age 65.6±10.9, male 75.4%) were analyzed. For qPCR studies analyzing GPNMB in leukocytes, atherosclerotic diseases (AD) group included coronary artery disease (n=33), peripheral artery disease (n=5), and cerebrovascular disease (n=1). The patients characteristics were also shown in Extended data figure 1d.
Recruitment	All the patients hospitalized in the department of cardiovascular medicine in Niigata University medical and dental hospital were recruited into the study. There were no self-selection bias. All the patients have cardiovascular diseases needed to be hospitalized. Thus, healthy individuals were not be assessed.
Ethics oversight	All subjects provided written informed consent prior to participation. The Scientific-Ethics Committees of Niigata University approved the study protocols (protocol number H24-585, 2015-2292, 2017-0102), and the studies were performed in accordance with the Declaration of Helsinki. Donors who provided samples for experiments were anonymous.

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	N/A
Study protocol	Protocol number H24-585, 2015-2292, 2017-0102
Data collection	Data for these were collected from biobank generated with samples collected between year 2013-2019 in our department.

Outcomes	These studies (H24-585, 2015-2292 and 2017-0102) were desighed as observational study, and no outcomes were studied.

Dual use research of concern

Policy information about dual use research of concern

Hazards

Could the accidental, deliberate or reckless misuse of agents or technologies generated in the work, or the application of information presented in the manuscript, pose a threat to:

No	Yes	
☒	☐	Public health
☒	☐	National security
☒	☐	Crops and/or livestock
☒	☐	Ecosystems
☒	☐	Any other significant area

Experiments of concern

Does the work involve any of these experiments of concern:

No	Yes	
☒	☐	Demonstrate how to render a vaccine ineffective
☒	☐	Confer resistance to therapeutically useful antibiotics or antiviral agents
☒	☐	Enhance the virulence of a pathogen or render a nonpathogen virulent
☒	☐	Increase transmissibility of a pathogen
☒	☐	Alter the host range of a pathogen
☒	☐	Enable evasion of diagnostic/detection modalities
☒	☐	Enable the weaponization of a biological agent or toxin
☒	☐	Any other potentially harmful combination of experiments and agents

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	Blood samples were collected from tail vein 7 days after neutralizing antibody injection, washed with PBS, and red blood cell lysis was achieved with an ammonium chloride-based lysing buffer (Pharm Lyse, 555899, BD). Cells were resuspended in FACS buffer (PBS supplemented with 1% FBS and 5mM EDTA) and stained with the following antibodies for 30 min on ice. For isolation of the stromal vascular cell fraction (SVF) from gonadal white adipose tissue, fat was excised, minced, and digested with digesting solution (0.5U/ml collagen, 0.8U/ml dispase and 1mM CaCls in PBS) for 20 min at 37?. For isolation of cells from aorta, aorta was excised, minced, and digested with digesting solution (1.5U/ml collagen, 2.4U/ml dispase and 1mM CaCls in PBS) for 20 min at 37?. The tissue lysate was subsequently filtered through a nylon mesh (40 µm) and red blood cell lysis was achieved with an ammonium chloride-based lysing buffer (Pharm Lyse, 555899, BD). Cells were resuspended in PBS supplemented with 1% FBS and 5mM EDTA and incubated with Fcr blocker (1 µg/million cells in 100 µl, 553141, BD) at 4℃ for 5 minutes) and stained with the following antibodies and 10µM SPiDER beta Gal (Do-Jin-do, SG02) for 30 min on ice.
Instrument	ID7000, SONY
Software	ID7000 software, version 1.1.0.11041
Cell population abundance	For isolation of the stromal vascular cell fraction (SVF) from gonadal white adipose tissue, fat was excised, minced, and digested with digesting solution (0.5U/ml collagen, 0.8U/ml dispase and 1mM CaCls in PBS) for 20 min at 37?. For isolation of

	cells from aorta, aorta was excised, minced, and digested with digesting solution (1.5U/ml collagen, 2.4U/ml dispase and 1mM CaCls in PBS) for 20 min at 37?.
Gating strategy	Gating strategies are shown in Supplementary figures.

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