nature portfolio

Corresponding author(s): Wang Min, Jenny Huanjiao Zhou

Last updated by author(s): 07/20/2024

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	The following standard software provided by instrument suppliers was used for data collection: Microscopy: Axiovert 200 (Zeiss); P rism 6.0 and 8.0 software (GraphPad); Fluorescent Nissl dye NeuroTrace 500/525; confocal Leica SP5 microscope (Leica, Germany); confocal Zeiss Airyscan 880 microscope (Zeiss, Germany); qRT-PCR: iCycler real-time detection system (Bio-Rad Laboratories, Inc., Hercules, CA);
Data analysis	To plot the data and for statistical analyses of images and Western blots was used NIH Image J 1.52P for imaging analyses, and Prism 6.0 and 8.0 software (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

All data, including data associated with main figures and supplementary figures, are available within the article or in the online-only Data Supplement. Source data are provided with this paper.
- Accession codes, unique identifiers, or web links for publicly available datasets: The bulk RNA-seq of carotid arteries is deposited and is publicly available at NCBI site as BioProject: PRJNA1139223 (https://www.ncbi.nlm.nih.gov/bioproject/PRJNA1139223).
- A list of figures that have associated raw data: Main Figures (Fig.1-8) and all Supplementary figures (Fig.1-5, 7, 10-11) as well as bulk RNA-seq are associated with raw data.
- A description of any restrictions on data availability: No any restrictions on data availability.

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	The sex characteristics for human participants are detailed in in Supplemental Table 1. he baseline characteristics of the patients are listed in Supplementary Table 1, including sex, smoking history, hypertension, lipid profiles and diabetes.
Reporting on race, ethnicity, or other socially relevant groupings	The population characteristics of human participants are shown in Supplemental Table 1. Specifically the patients were Han Chinese.
Population characteristics	Human coronary arteries were obtained from cardiac transplant recipients (total n=9; averaged ages are 66.67±4.39) with chronic rejection undergoing no or re-transplantation (n=3), cardiomyopathy recipients undergoing first-time transplantation (n=6), and organ donors without cardiac disease (total n=6; averaged ages 50.20±4.15). The baseline characteristics of the patients are listed in Supplementary Table 1, including sex, smoking history, hypertension, lipid profiles and diabetes.
Recruitment	Human coronary arteries were obtained from cardiac transplant recipients (total n=9; averaged ages are 66.67±4.39) with chronic rejection undergoing no or re-transplantation (n=3), cardiomyopathy recipients undergoing first-time transplantation (n=6), and organ donors without cardiac disease (total n=6; averaged ages 50.20±4.15). written informed consents were obtained from all surgical recipient patients and a family member of all deceased organ donors. The baseline characteristics of the patients are listed in Supplementary Table 1, including sex, smoking history, hypertension, lipid profiles and diabetes. Our procurement techniques have been described previously50,51. Briefly, for each arterial sample procured in the operating room, disease was macroscopically diagnosed by an experienced cardiac surgeon. Half of each sample was formalin-fixed immediately for paraffin-embedding and the other half was stored at -80 °C. Aortic pathologies were examined for plaque rupture, fibrocalcific plaques and fibroatheroma.
Ethics oversight	This study was approved by the Ethics Review Board of approved by the Conjoint Health Research Ethics Board of the First People's Hospital of Changzhou and Nanjing Drum Tower Hospital, The Affiliated Hospital of Nanjing University Medical School. Informed consent was obtained from all the subjects enrolled into this study.

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	Sample sizes were chosen based on each experiment being technically feasible from a work flow standpoint while also providing a reasonable number of replicates to be confident in the results. For in vivo, group sizes were determined by an a priori power analysis for a two-sided, two-sample t-test with an α of 0.05 and power of 0.8 to detect a 10% difference in lesion size at the endpoint.
Data exclusions	None of the in vitro data were excluded. No animals were discarded.
Replication	All findings reported were reliably reproduced. All experiments were repeated at least two times.

Randomization	No special randomization protocol was used. Mice were randomly assigned to given treatment groups.
Blinding	The investigators were blinded to group allocation during data collection and 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
[ ]	[x] Eukaryotic cell lines
[x]	[ ] Palaeontology and archaeology
[ ]	[x] Animals and other organisms
[x]	[ ] Clinical data
[x]	[ ] Dual use research of concern
[x]	[ ] Plants

Methods

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

Antibodies

Antibodies used	Abcam: SM-22α, Abcam ab10135, MYH11Abcam ab53219, Calponin1 (CNN1) Abcam ab46794, Osteopontin (OPN) Abcam ab8448, Osteopontin (OPN) Abcam ab218237, MYH10 Abcam ab230823, MMP2 Abcam ab37150, MMP9 Abcam ab38898 Ki67, Abcam ab66155, LAMP2 Abcam Ab13524, KLF4 Abcam Ab151733, SENP1 Abcam ab236094; Cell Signaling: SENP1 Cell signaling CST11929s, SUMO1 Cell signaling CST4930s, SRF Cell signaling CST5147s, CyclinD1 Cell signaling CST2978, PCNA Cell signaling CST13110, MLK1/MRTF-A Cell signaling CST-14760, MRTF-B Cell signaling CST14613, p-MEK1/2 Cell signaling CST2338, MEK1/2 Cell signaling CST8727, p-ERK1/2 Cell signaling CST4370s, p-NF-κB Cell signaling CST3033s, NF-κB Cell signaling CST8242s, p-P38 Cell signaling CST4511s, P38 Cell signaling CST8690, HA-Tag Cell signaling CST3724, DYKDDDDK Tag (Flag) Cell signaling CST14793, Cleaved caspase-3 Cell signaling CST9579T, Caspase-9 Cell signaling CST9504, Bcl-2 Cell signaling CST15071, BAX Cell signaling CST14796, GAPDH Cell signaling CST5174s, p-SRF-S103 Cell signaling CST4261s; α-SMA Invitrogen 14-9760-82, SENP1 Invitrogen ZYMED383350, SENP1 Life science D2614000200, CD31 Millipore MAB13982, Myocardin Novus Biologicals NBP1-74113, CD31 R&D Systems AF3628; Santa Cruz: SENP1 Santa cruz sc271360, SUMO1 Santa cruz sc5308, SUMO2/3 Santa cruz sc393144, MRTF-A Santa cruz sc398675, MRTF-A Santa cruz sc390324, p-ELK1 Santa cruz sc8406, ELK1 (E-5) Santa cruz sc365876, ERK1/2 Santa cruz sc514302, α-SMA Sigma F3777-2ML, Myocardin Sigma-Aldrich SAB4200539. Alexa Fluour 594 or 488 conjugated secondary antibodies (Invitrogen); full list see Supplementary Table 3.
Validation	The information above can also be found in the methods. For all the antibodies, additional information on specificity and species cross-reactivity, with links to key publications can be found on the manufacturer's website. We validated the critical molecules by comparing WT and KO mice (e.g, SENP1) and siRNA silencing cells (e.g., SENP1). Other signaling antibodies were validated using IgG isotype as controls for parallel staining.

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)	MOVAS-1 cell line (Cat: #ATCC®CRL-2797™), derived from aortic smooth muscle cells of male C57BL/6J mice, was purchased from American type culture collection (ATCC; Manassas, VA, USA). Primary aortic VSMCs were isolated from WT and Senp1SMCKO mice by elastase/collagenase digestion protocols. the cells were resuspended in DMEM supplemented with 10% FBS and 1% penicillin-streptomycin (PenStrep, GIBCO-Invitrogen, NY, USA), and cultured in 35-mm dishes in a CO2 incubator at 37 °C. After the primary cells reached 90% confluency.
Authentication	Cell lines were not authenticated.
Mycoplasma contamination	All cell lines were tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used.

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	We used a previously generated conditional gene knockout mouse model lacking Senp1 specifically in VSMCs (Senp1SMCKO). Mice were housed in the animal care facility of Yale University under standard pathogen-free conditions with a 12 h light/dark schedule

	and provided with food and water ad libitum, temperature was between 20 and 24 °C and relative humidity between 45 and 65 rH. Wire injury was performed in Senp1lox/lox (WT) and Senp1SMCKO mice (10–12-weeks-old) as described previously.
Wild animals	No wild animal were used in this study.
Reporting on sex	Both male and female were used in the study. Data from male were presented in main figures..
Field-collected samples	No wild animal were used in this study.
Ethics oversight	All animal procedures were performed under protocols approved by Yale University Institutional Animal Care and Use Committee.

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