nature portfolio

Corresponding author(s): Kory Lavine

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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	BD FACSChorus version 1.1, D FACSDiva version 8, 10x Genomics CellRanger v6.1, Zeiss Zen Blue version 3.2, Zeiss Zen Black Version 2.0, VisualSonics 770 Echocardiography System, Inveon PET/CT system (Siemens, Malvern, PA)
Data analysis	R v4.0.3, Seurat v4.0, ArchR v1.0.2, MACS v2.2, Python v3.9.2, Palantir v1.0.0, scVelo v0.2.5, DESeq2 v1.44.0, SPOTlight v1.8.0, cell2location v0.0.5, MistyR v1.8.1, nichenetr 1.1.0, dsb v1.0.3, scanpy v1.10, Velocyto CLI v0.17.17, MAGIC v3.0.0, clusterProlifer v4.12.0, progeny v1.26.0, SCTransform v0.4.1, VivoLab, Inveon Research Workplace, custom codes are available at https://github.com/jamrute/2024_Nature_IL1B_ImmuneFibroblast

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

Raw and processed sequencing files can be found on the Gene Expression Omnibus super series (GSE218392). The following additional data were used in this study E-MTAB-10324, EGAS00001006330 (https://explore.data.humancellatlas.org/projects/e9f36305-d857-44a3-93f0-df4e6007dc97), GSE183852.

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	Information Provided in Supplemental Table 1
Reporting on race, ethnicity, or other socially relevant groupings	Information Provided in Supplemental Table 1
Population characteristics	Information Provided in Supplemental Table 1
Recruitment	NA
Ethics oversight	The study is compliant with all relevant ethical regulations and has been approved by the Washington University School of Medicine Institutional Review Board (IRB #201104172). Informed consent was obtained from each patient prior to tissue collection by Washington University School of Medicine and no compensation was provided in exchange for subject participation in the study. All demographic and clinical data has been de-identified and provided in Supplementary Table 1. Patients included in this study span diverse race, age, and sex to provide an inclusive trans-ethic study population.

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	No sample size calculations were performed. Sample size was governed by tissue availability and input tissue mass was based on ability to recover sufficient cells.
Data exclusions	No samples were excluded.
Replication	In vivo experiments were replicated at-least once for a total of two batches and data was pooled for downstream analysis (all attempts at replication were successful).
Randomization	For human studies all transplants with HF, MI, and non-failing donors were processed randomized across age, sex, and race. For in vivo studies male and female mice were used and randomized to sham and treatment groups and littermate controls used in genetic studies.
Blinding	Individuals were partially blinded to the sample groups. All single cell/nuclei analysis including clustering is done using un-biased techniques. Investigators were blinded to the groups when performing initial analysis. Blinding during data collection was not necessary as nuclei isolation protocol required FACS to collect intact cells/nuclei with no exclusion of any cells/nuclei. This sorting approach does not introduce any bias into the sample collection.

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

TotalSeq A BioLegend panel + FAP, LRRC15 for CITE-seq experiments: FAP, LRRC15, CDH11, CD45, CDH1, CD276, CD45RA, GYPA, CD235ab, CD45RB, CD27, CD45R, CD45RO, FCGR1A, KIT, TNFRSF18, CD207, CD70, TNFRSF13C, TNFSF4, LAG3, TNFRSF14, NRP1, CXCR4, MRC1, SIRPA, OLR1, SLAMF7, PDGFRA, LILRA4, PDGFRB, TNFRSF17, TNFRSF13B, CSF2RB, KLRK1, HAVCR1, CRLF2, TNFSF13B, C5AR2, CLEC4C, CD40LG, ERBB2, PDCD1LG2, CD274, CD80, IL6ST, MS4A1, LTBR, TEK, CD99, TRA, HLA-E, CD72, PRNP, FCGR3A, FCRL4, ENG, TNFRSF9, ABCB1, CSF2RA, CD36, NCAM1, CD151, FCRL5, CLEC12A, CD1D, CD34, MERTK, CD81, CD40, KLRF1, ABCG2, THY1, CCR3, TNFSF9, SIGLEC7, CEACAM8, CCR10, CD164, MICA_MICB, IL3RA, SIGLEC1, ANXA1, LRRC32, MSR1, KDR, CDH2, CLEC9A, CSF1R, FOLR2, EPCAM, FLT4, CRTAM, CD209, ICOSLG, ITGA4, TIMD4, LGALS9, IL7R, ENTPD1, SLAMF1, TIGIT, TPSAB1, NT5E, FCER1A, SELP, CD82, EGFR, CLEC1B, TCR.gamma.delta, TCRvgamma9, TCRvdelta2, DPP4, CD101, HLA-A2, IL2RA, TNFRSF4, ITGAE, CD48, PTGDR2, CTLA4, CR2, CD135, CDH5, TNFRSF8, CD244, ICOS, CD79B, ICAM2, CD47, CD28, SPN, CD7, CD84, ADGRG1, TFRC, SDC1, SELL, FAS, KLRD1, KIR2DL2, KIR3DL1, CR1, FCER2, PDCD1, HAVCR2, ITGB7, CD69, FCGR2A, Rat.IgG1.lambda, CXCR3, CCR6, CCR7, IL4R, CD163, LILRB1, IFNGR1, ITGA6, CLEC10A, TCRvbeta13.1, IL18R1, LAMP1, FCRL3, CD63, ICAM1, SELE, CD83, LY75, MME, CD1A, CD52, CD9, CD19, GP1BA, ITGA2B, CD38, KLRB1, KIR3DL2, VPREB1, TLR4, Hamster.IgG, IGHD, ITGAM, CD44, TCR.alpha.beta, CD86, CXCR5, CCR5, CD55, CX3CR1, CCR2, CXCR6, SELPLG, CCR9, L1CAM, CD1C, HLA_DR, CCR8, CCR4, CD74, CD37, IGHG1, LGALS3, CD14, THBD, TNF, NGFR, SLC3A2, CD177, IGHE, IGKC, IGLL1, IGHM, NOTCH1, NOTCH3, TNFSF11, BTLA, VTCN1, CD24, Mouse.IgG2a, Mouse.IgG1, Mouse.IgG2b, PDPN, CD96, LAIR1, FASLG, ENPP3, SLAMF6, F3, F11R, CD200, ITGA2, MCAM, CD59, NCR3, CD33, B3GAT1, PROCR, TNFSF10, CD4, BST2, Rat.IgG1.k, Rat.IgG2a, Rat.IgG2c, Rat.IgG2b, CD22, ITGAX, XCR1, PROM1, MPL, TSPAN33, KLRG1, CD8A, PVR, VCAM1, HLAII, CD46, TREM1, ITGB2, CD2, ITGB1, ITGAL, ITGA1, CD58, IL2RB. LAP, CD226. NECTIN2, CD3D, CD5, KIR2DL5A, IL6R, ITGB3, CD93, HLA-A.B.C, FUT4, CD109, ANPEP, PECAM1.

Flow Cytometry: DRAQ5 (Thermo Fisher Scientific, 564907) , DAPI (BD Biosciences, 564907), 7-AAD (Sigma; SML1633-1ML), Pdpn (BioLegend, 127423, clone 8.1.1), Cd140a (Biolegend, 135911, clone APA5), Cd31 (Biolegend, 102409, clone 390), Fap (Creative Biolabs, HPAB-0171-CN-PE), and Cd45 (Biolegend, 103132, clone 30-F11).

Histology: FAP (Abcam, ab207178, 1:250), POSTN (Abcam, ab215199, 1:250), anti-GFP (Abcam, ab13970, 1:200), and IL-1b (R&D systems, AF-401-NA, 1:50), ACTA-2 antibody (#F3777, Sigma Aldrich, 1:1000), ProLong Gold antifade reagent (#P36962, Invitrogen), Postn RNAscope mouse (#418581, mouse POSTN).

Validation

Antibodies used in this study are all from well-established commercial vendors that perform rigorous species-specific QC testing for each lot.

BioLegend:https://www.biolegend.com/en-us/quality/quality-control
CITE-seq: TotalSeq A BioLegend panel + FAP, LRRC15 for CITE-seq experiments: FAP, LRRC15, CDH11, CD45, CDH1, CD276, CD45RA, GYPA, CD235ab, CD45RB, CD27, CD45R, CD45RO, FCGR1A, KIT, TNFRSF18, CD207, CD70, TNFRSF13C, TNFSF4, LAG3, TNFRSF14, NRP1, CXCR4, MRC1, SIRPA, OLR1, SLAMF7, PDGFRA, LILRA4, PDGFRB, TNFRSF17, TNFRSF13B, CSF2RB, KLRK1, HAVCR1, CRLF2, TNFSF13B, C5AR2, CLEC4C, CD40LG, ERBB2, PDCD1LG2, CD274, CD80, IL6ST, MS4A1, LTBR, TEK, CD99, TRA, HLA-E, CD72, PRNP, FCGR3A, FCRL4, ENG, TNFRSF9, ABCB1, CSF2RA, CD36, NCAM1, CD151, FCRL5, CLEC12A, CD1D, CD34, MERTK, CD81, CD40, KLRF1, ABCG2, THY1, CCR3, TNFSF9, SIGLEC7, CEACAM8, CCR10, CD164, MICA_MICB, IL3RA, SIGLEC1, ANXA1, LRRC32, MSR1, KDR, CDH2, CLEC9A, CSF1R, FOLR2, EPCAM, FLT4, CRTAM, CD209, ICOSLG, ITGA4, TIMD4, LGALS9, IL7R, ENTPD1, SLAMF1, TIGIT, TPSAB1, NT5E, FCER1A, SELP, CD82, EGFR, CLEC1B, TCR.gamma.delta, TCRvgamma9, TCRvdelta2, DPP4, CD101, HLA-A2, IL2RA, TNFRSF4, ITGAE, CD48, PTGDR2, CTLA4, CR2, CD135, CDH5, TNFRSF8, CD244, ICOS, CD79B, ICAM2, CD47, CD28, SPN, CD7, CD84, ADGRG1, TFRC, SDC1, SELL, FAS, KLRD1, KIR2DL2, KIR3DL1, CR1, FCER2, PDCD1, HAVCR2, ITGB7, CD69, FCGR2A, Rat.IgG1.lambda, CXCR3, CCR6, CCR7, IL4R, CD163, LILRB1, IFNGR1, ITGA6, CLEC10A, TCRvbeta13.1, IL18R1, LAMP1, FCRL3, CD63, ICAM1, SELE, CD83, LY75, MME, CD1A, CD52, CD9, CD19, GP1BA, ITGA2B, CD38, KLRB1, KIR3DL2, VPREB1, TLR4, Hamster.IgG, IGHD, ITGAM, CD44, TCR.alpha.beta, CD86, CXCR5, CCR5, CD55, CX3CR1, CCR2, CXCR6, SELPLG, CCR9, L1CAM, CD1C, HLA_DR, CCR8, CCR4, CD74, CD37, IGHG1, LGALS3, CD14, THBD, TNF, NGFR, SLC3A2, CD177, IGHE, IGKC, IGLL1, IGHM, NOTCH1, NOTCH3, TNFSF11, BTLA, VTCN1, CD24, Mouse.IgG2a, Mouse.IgG1, Mouse.IgG2b, PDPN, CD96, LAIR1, FASLG, ENPP3, SLAMF6, F3, F11R, CD200, ITGA2, MCAM, CD59, NCR3, CD33, B3GAT1, PROCR, TNFSF10, CD4, BST2, Rat.IgG1.k, Rat.IgG2a, Rat.IgG2c, Rat.IgG2b, CD22, ITGAX, XCR1, PROM1, MPL, TSPAN33, KLRG1, CD8A, PVR, VCAM1, HLAII, CD46, TREM1, ITGB2, CD2, ITGB1, ITGAL, ITGA1, CD58, IL2RB. LAP, CD226. NECTIN2, CD3D, CD5, KIR2DL5A, IL6R, ITGB3, CD93, HLA-A.B.C, FUT4, CD109, ANPEP, PECAM1.
Bulk lots are tested by PCR and sequencing to confirm the oligonucleotide barcodes. They are also tested by flow cytometry to ensure the antibodies recognize the proper cell populations. Bottled lots are tested by PCR and sequencing to confirm the oligonucleotide barcodes.
Flow cytometry: Cd140a (Biolegend, 135911, clone APA5), Cd31 (Biolegend, 102409, clone 390), and Cd45 (Biolegend, 103132, clone 30-F11).
Specificity testing of 1-3 target cell types with either single- or multi-color analysis (including positive and negative cell types). Once

specificity is confirmed, each new lot must perform with similar intensity to the in-date reference lot. Brightness (MFI) is evaluated from both positive and negative populations. Each lot product is validated by QC testing with a series of titration dilutions.

Thermo Fisher Scientific: https://www.thermofisher.com/nz/en/home/life-science/antibodies/invitrogen-antibody-validation.html
Flow cytometry: DRAQ5 (Thermo Fisher Scientific, 564907)
Histology: ProLong Gold antifade reagent (#P36962, Invitrogen)
Advanced verification is additional testing that verifies that an antibody will bind to the correct target. We consider two key factors when selecting the appropriate advanced verification testing method(s): The nature of the target—this includes considering relevant target properties such as its biological function, sub-cellular localization, post-translational modifications, gene essentiality, and the rate of turnover. The nature of the sample type—the sample type can influence epitope exposure and the reactivity of an antibody in a specific application.

BD Biosciences: https://www.bdbiosciences.com/en-nz/products/reagents/flow-cytometry-reagents/research-reagents/quality-and-reproducibility
DAPI (BD Biosciences, 564907)
The specificity is confirmed by using multiple applications that may include a combination of flow cytometry, immunofluorescence, immunohistochemistry or western blot to test a combination of primary cells, cell lines or transfectant models.

Sigma: https://www.sigmaaldrich.com/US/en/technical-documents/technical-article/protein-biology/elisa/antibody-standard-validation
7-AAD (Sigma; SML1633-1ML)
ACTA-2 antibody (#F3777, Sigma Aldrich, 1:1000)
We are a global company with manufacturing facilities worldwide. Our development and manufacturing processes are subject to rigorous quality control and quality assurance measures, and each of our antibody products is supplied with a comprehensive Certificate of Analysis and Product Information Sheet.

Creative Biolabs: https://www.creative-biolabs.com/antibody-characterization.html?gclid=CjwKCAjw1emzBhB8EiwAHwZZxQvLR2WsOKnsFmy4JbbTcK8_6V1EUEy_GB6CV4M-Cxzy5TEwqthwjhoCqTAQAvD_BwE
Fap (Creative Biolabs, HPAB-0171-CN-PE)

Abcam: https://www.abcam.com/primary-antibodies/how-we-validate-our-antibodies
FAP (Abcam, ab207178, 1:250), POSTN (Abcam, ab215199, 1:250), anti-GFP (Abcam, ab13970, 1:200)
We use a variety of methods, including staining multi-normal human tissue microarrays (TMAs), multi-tumor human TMAs, and rat or mouse TMAs during antibody development. These high-throughput arrays allow us to check many tissues at the same time, providing uniformly as all tissues are exposed to the exact same conditions.

R&D: https://www.rndsystems.com/products/rd-systems-approach-antibody-quality
IL-1b (R&D systems, AF-401-NA, 1:50)
Each antibody is manufactured under controlled conditions, undergoing rigorous quality control testing to ensure lot-to-lot consistency and outstanding performance in all applications listed on our datasheets.

ACD Bio: https://acdbio.com/control-slides-and-control-probes-rnascope
Postn RNAscope mouse (#418581, mouse POSTN)
Success with any assay begins with good and consistent quality control practices. RNAscope™ in situ hybridization is a nucleic acids-based method in which rigorous controls can be easily incorporated into every assay. ACD recommends two levels of quality control practice:

Technical assay control check: The RNAscope™ Assay is an easy single-day assay, similar in workflow to the immunohistochemistry (IHC) workflow. The highly specific and sensitive nucleic acid hybridization is made possible with RNAscope™ technology, which requires close attention to the protocol. To ensure first-time success with specific detection of your intended target using the RNAscope™ assay, ACD strongly encourages all users to perform a technical assay control to ensure that the assay is being performed appropriately. This technical control is easily performed using a cell pellet control sample, tested on two slides with a low-copy housekeeping gene positive control probe and a non-specific bacterial negative control probe. When the assay is run properly, this technical control will show strong positive control probe staining and clean negative control probe staining. We do this with every assay to verify that it is being run with the correct technique.

Sample/RNA quality control check: The RNAscope™ Assay has universal assay conditions. Every target-specific probe uses identical hybridization conditions. However, tissue RNA quality and fixation conditions often vary. As a result, it is occasionally necessary to adjust the pretreatment conditions for optimal RNA exposure and detection by RNAscope™ ISH. Empirically, we have observed that the vast majority of properly fixed tissues are suitable for RNA detection with RNAscope™ ISH. ACD provides guidelines on pretreatment conditions for certain tissue types. However, we encourage you to optimize the specific pretreatment conditions for your tissue samples before performing your experiments. Do this by first using the positive and negative control probes on your tissues to verify that you have high positive control signal and no negative control background. If signal is low or there is any background, this is most often improved with adjustment to pretreatment times. Once you have established that sample and RNA quality are good, you can be sure that the RNAscope™ ISH experiments with target-specific probes will be successful.

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	Mice were approximately 8 weeks of age at the time of experiments. Mice strains used: C57BL/6J (strain #000664; https://www.jax.org/strain/000664), CCR2 GFP (strain #027619; https://www.jax.org/strain/027619), Il1r f/f Dermo1Cre (generated by crossing Dermo1-Cre (strain #008712; https://www.jax.org/strain/008712) to Il1r loxP (strain #028398; https://www.jax.org/

	strain/028398)). Il1b f/f (PMID 28092375) were crossed to Ccrt-ert2Cre (PMID 26341401). A Fap-ert2Cre was made as described in the Supplementary Methods. All animals were maintained on 12-hour light cycles and housed at 70F and 50% humidity. Experiments were performed with mice 6-12 weeks of age, with sex-matched littermates whenever possible.
Wild animals	No wild animals were used in this study.
Reporting on sex	Male and female mice were used.
Field-collected samples	No field collected samples were used in this study.
Ethics oversight	All mice were maintained in our specific-pathogen free facility following institutional guidelines and with protocols approved by the AAALAC-accredited Animal Studies Committee at Washington University in St. Louis. All protocols and animal studies were approved by Washington University School of Medicine Institutional Animal Care and Use Committee (IACUC) protocol #20-0436.

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

Plants

Seed stocks	NA
Novel plant genotypes	NA
Authentication	NA

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	Cardiac tissues from LVAD cores at the time of LVAD implant (U/RR-pre) and adjacent to core samples at the time of explant (U/RR-post) from paired patient were flash frozen using liquid nitrogen. Identical regions from the apex of LV from explanted donors were used. Single nuclei suspensions were generated as previously described. In brief: flash frozen sections were minced with a razor blade, transferred to a Dounce Homogenizer containing 1 mL of lysis buffer (10mM Tris-HCl, pH 7.4, 10 mM NaCl, 3mM MgCl2 and 0.1% NP-40 in nuclease-free water) on ice. Samples were homogenized using five strokes, an additional 1 mL of lysis buffer added, and incubated on ice for 15 mins. Samples were then filtered with a 40µm filter and filter was rinsed with 1mL of lysis buffer. The mixture was then centrifuged at 500g for 5 min 4°C, resuspended in 1mL nuclei wash buffer (2% BSA and 0.2 U µl−1 RNase inhibitor (Thermo Fisher, cat. no. AM2694) in 1× PBS) and, filtered using a 20µm pluristrainer (Pluriselect, cat. No. SKU43-50020-03). Filtered solution as centrifuged using the above criteria and resuspended in 300 µL Nuclei Wash Buffer and transferred into a 5mL tube for flow cytometry. Subsequently, 1µl DRAQ5 (5mM solution; Thermo Fisher, cat. no. 62251) was added, sample gently vortexed, and allowed to incubate for 5min prior to sorting. DRAQ5 +nuclei were sorted into 300 µL Nuclei Wash Buffer using a BD FACS Melody (BD Biosciences) with a 100 µM nozzle. Sorted nuclei were then centrifuged using the above conditions and resuspended in Nuclei Wash Buffer for a final target concentration of 1,000 nuclei/µL – nuclei were counted on a hemocytometer. Nuclei were isolated according to 10x Genomics protocol (CG00375; Nuclei Isolation Complex Sample for ATAC GEX Sequencing RevB) and flow cytometry for 7-AAD (Sigma; SML1633-1ML) positive nuclei was used for sorting. Protocol CG000338 from 10x Genomics was used for Chromium Next GEM Single Cell Multiome ATAC + Gene Expression. Briefly, following nuclei isolation, permeabilization was performed, followed by transposition, GEM generation and barcoding using ChipJ (10x Genomics; PN1000234), post-GEM clean up, pre-amplification PCR, cDNA amplification, library construction, and sequencing. Gene expression and ATAC libraires were sequenced to a read depth of 50,000 and 25,000 respectively. Fresh cardiac left ventricular tissue from ex-planted hearts at the time of transplantation, LVAD cores, or donors declared DCD at Washington University School of Medicine and Mid America Transplant Service were harvested, perfused with cardioplegia, and transported on ice. In donor, ICM, and NICM hearts a section of the LV apex was dissected out; in acute MI hearts a section of the infarct region (identified as a white discoloration) was dissected. Tissues were minced using a razor blade on ice and transferred to a 15 mL conical tube containing 3mL DMEM with 170uL Collagenase IV (250U/mL final

concentration), 35uL DNAse1 (60U/mL), and 75uL Hyaluronidase (60U/mL) and incubated at 37 °C for 45 min with agitation. After 45 min, the digestion reaction was quenched with 6 mL of HBB buffer (2% FBS and 0.2% BSA in HBSS), filtered through 40 µm filters into a 50 mL conical tube and transferred back into a 15 mL conical tube to obtain tighter pellets. Samples were then spun down at 4 °C, 1200rpm for 5 min and the supernatant was discarded. Pellet resuspended in 1 mL ACK Lysis buffer (Gibco A10492-01) and incubated at room temperature for 5 min followed by the addition of 9 mL DMEM and centrifugation (4 °C, 5 min, 1200 rpm). Supernatant was discarded and the pellet was resuspended in 2 mL FACS buffer (2% FBS and 2mM EDTA in calcium/magnesium free PBS) and centrifugation was repeated in above conditions and supernatant aspirated. The TotalSeq A 277 panel (BioLegend) antibody cocktail was resuspended in 100 uL of FACS buffer and 1 uL each of custom oligo-labeled FAP (Amgen) and LRRC15 (Amgen) were added. The combined 102 uL were used to resuspend the pellet with the addition of 1 uL of DRAQ5 (Thermo Fisher Scientific, 564907) and incubated on ice for 30 min. Solution was washed 3x with FACS buffer following same centrifugation as above and then resuspended in 500 uL of FACS buffer and 1 uL DAPI (BD Biosciences, 564907) and filtered into filter-top FACS tubes. First singlets were gated and subsequent DRAQ5+/DAPI- events were collected in 300 uL cell resuspension buffer (0.04% BSA in PBS) – collected cells were centrifuged as above and resuspended in collection buffer to a target concentration of 1,000 cells/uL. Cells were counted on a hemocytometer before proceeding with the 10x protocol.

Instrument	BD FACSAria II and FACSMelody
Software	BD FACSDiva and FACSChorus
Cell population abundance	10,000 nuclei or cells
Gating strategy	CITE-seq: DRAQ5+, DAPI- (Supplementary Figure 1) Multiome: 7-AAD+ (Supplementary Figure 1) In vivo experiments: (Supplementary Figure 6)

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