

Major Resources Table

In order to allow validation and replication of experiments, all essential research materials listed in the Methods should be included in the Major Resources Table below. Authors are encouraged to use public repositories for protocols, data, code, and other materials and provide persistent identifiers and/or links to repositories when available. Authors may add or delete rows as needed.

Animals (in vivo studies)

Species	Vendor or Source	Background Strain	Sex	Persistent ID / URL
Mice: <i>eNOSKO</i> B6.129P2- <i>Nos3^{tm1Unc}/J</i>	Jackson Laboratories	Strain #:002684	Female & Male	RRID:IMSR_JAX:002684
Mice: C57BL/6-Tg(<i>Pdgfra-cre</i>)1Clc/J	Jackson Laboratories	Strain #:013148	Female & Male	RRID:IMSR_JAX:013148
Mice: B6.129(Cg)- <i>Gt(ROSA)26Sor^{tm4}(ACTB-tdTomato,-EGFP)Luo/J</i>	Jackson Laboratories	Strain #:007676	Female & Male	RRID:IMSR_JAX:007676
Mice: <i>ApoE^{-/-}</i> B6.129P2- <i>ApoE^{tm1Unc}/J</i>	Jackson Laboratories	Strain #:002052	Female & Male	RRID:IMSR_JAX:002052
Mice: <i>ETBRKO</i> B6;129- <i>Ednrb^{tm1.1Nat}/J</i>	Jackson Laboratories	Strain #:011080	Female & Male	RRID:IMSR_JAX:011080
Mice: VEC-Cre B6.Cg-Tg(<i>Cdh5-cre</i>)1Spe/J	Jackson Laboratories	Strain #:033055	Female & Male	RRID:IMSR_JAX:033055

Genetically Modified Animals

Species	Vendor or Source	Background Strain	Other Information	Sex	Persistent ID / URL
<i>ECIRS1</i> transgenic mice	In house, Joslin Diabetes Center	C57BL/6J	Human IRS1 overexpression driven by <i>Cdh5</i> promoter	Female & Male	N/A

Antibodies

Target antigen	Vendor or Source	Catalog #	Working concentration	Lot # (preferred but not required)	Persistent ID / URL

p-CaMKII (Tyr231)	Cell Signaling Technology	3356	1:1,000 (WB)		https://www.cellsignal.com/products/primary-antibodies/phospho-camkii-tyr231-antibody/3356
t-CaMKII	Cell Signaling Technology	4436	1:1,000 (WB)		https://www.cellsignal.com/product/productDetail.jsp?productId=4436
p-Erk	Cell Signaling Technology	9101	1:1,000 (WB)		https://www.cellsignal.com/product/productDetail.jsp?productId=9101
t-Erk	Cell Signaling Technology	4695	1:1,000 (WB)		https://www.cellsignal.com/product/productDetail.jsp?productId=4695
p-Akt	Cell Signaling Technology	4060	1:1,000 (WB)		PRID:AB_10805010, https://www.cellsignal.com/product/productDetail.jsp?productId=4060
t-Akt	Cell Signaling Technology	9272	1:1,000 (WB)		https://www.cellsignal.com/product/productDetail.jsp?productId=9272
p-GSK3 β (Ser21/9)	Cell Signaling Technology	9331	1:1,000 (WB)		https://www.cellsignal.com/product/productDetail.jsp?productId=9331
p- β -catenin (Ser675)	Cell Signaling Technology	4176	1:1,000 (WB)		https://www.cellsignal.com/product/productDetail.jsp?productId=4176
t- β -catenin	Cell Signaling Technology	9582	1:1,000 (WB)		https://www.cellsignal.com/product/productDetail.jsp?productId=9582
iNOS	Cell Signaling Technology	2982	1:1,000 (WB)		PRID:AB_1078202, https://www.cellsignal.com/product/productDetail.jsp?productId=2982
t-PKG1 α	Cell Signaling Technology	3248	1:1,000 (WB)		https://www.cellsignal.com/product/productDetail.jsp?productId=3248
VCAM-1	R&D systems	AF643	1:1,000 (WB & Neutralization)		https://www.rndsystems.com/products/mouse-vcam-1-cd106-antibody_af643
Galectin-3	Millipore Sigma	MABT51	1:1,000 (IF)		https://www.emdmillipore.com/US/en/product/Anti-Galectin-3-Antibody-clone-M3-38,MM_NF-MABT51?ReferrerURL=https%3A%2F%2Fwww.google.com%2F
perilipin	Abcam	ab3526	1:1,000 (WB)		https://www.abcam.com/perilipin-1-antibody-ab3526.html
ICAM-1	Abcam	ab179707	1:1,000 (WB)		https://www.abcam.com/icam1-antibody-epr16608-ab179707.html
UCP1	Abcam	ab23841	1:1,000(WB and IHC)		https://www.abcam.com/ucp1-antibody-ab23841.html
p-GSK3 β (Y216/Y279)	Abcam	ab68476	1:1,000 (WB)		https://www.abcam.com/gsk3-beta-phospho-y216--gsk3-alpha-phospho-y279-antibody-epr933y-ab68476.html

total OXPHOS Rodent WB Antibody Cocktail	Abcam	ab110413	1:500 (WB)		https://www.abcam.com/total-oxphos-rodent-wb-antibody-cocktail-ab110413.html
p-eNOS	BD Biosciences	612392	1:1,000 (WB)		https://www.bdbiosciences.com/en-us/search-results?searchKey=612392
t-eNOS	BD Biosciences	610297	1:1,000 (WB)		https://www.bdbiosciences.com/en-us/search-results?searchKey=610297
HDAC1	Santa Cruz Biotechnology	sc-81598	1:1,000 (WB)		PRID:AB_2118083, https://www.scbt.com/p/hdac1-antibody-10e2?requestFrom=search
goat anti-mouse HRP	Santa Cruz Biotechnology	sc-2031	1:2,500 (WB)		https://www.scbt.com/p/goat-anti-mouse-igg-hrp-cruz-marker-compatible
Goat anti-rabbit HRP	Santa Cruz Biotechnology	sc-2004	1:2,500 (WB)		https://www.scbt.com/p/goat-anti-rabbit-igg-hrp?requestFrom=search
anti-mouse lineage-biotin (Lin-1)	BioLegend	133307	1:500 (FACS)		https://www.biolegend.com/de-at/search-results/biotin-anti-mouse-lineage-panel-7611
anti-mouse CD146-PerCP-Cy5.5	BioLegend	134710	1:500 (FACS)		https://www.biolegend.com/en-us/products/percp-cyanine5-5-anti-mouse-cd146-antibody-7872
anti-mouse PE/Cy7-CD31	BioLegend	102418	1:500 (FACS)		https://www.biolegend.com/en-us/products/pe-cyanine7-anti-mouse-cd31-antibody-3942
anti-mouse PE-CD140b	BioLegend	136006	1:500 (FACS)		https://www.biolegend.com/en-us/products/pe-anti-mouse-cd140b-antibody-6256
anti-mouse Pacific-Ly-6A/E (Sca-1)	BioLegend	108120	1:500 (FACS)		https://www.biolegend.com/en-us/products/pacific-blue-anti-mouse-ly-6a-e-sca-1-antibody-3140
anti-mouse Alexa Fluor 647-CD34	BioLegend	119314	1:500 (FACS)		https://www.biolegend.com/en-us/products/alexa-fluor-647-anti-mouse-cd34-antibody-3773
anti-mouse Ter-119-Biotin	BioLegend	116204	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-ter-119-erythroid-cells-antibody-1864

anti-mouse NK-1.1-Biotin	BioLegend	108704	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-nk-1-1-antibody-428
anti-mouse CD11b-Biotin	BioLegend	101204	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-human-cd11b-antibody-346
anti-mouse Gr-1-Biotin	BioLegend	108403	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-ly-6g-ly-6c-gr-1-antibody-457
anti-mouse B20-Biotin	BioLegend	103204	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-human-cd45r-b220-antibody-444
anti-mouse CD19-Biotin	BioLegend	115504	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-cd19-antibody-1527
anti-mouse CD8a-Biotin	BioLegend	100704	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-cd8a-antibody-152
anti-mouse CD4-Biotin	BioLegend	116010	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-cd4-antibody-8121
anti-mouse CD3-Biotin	BioLegend	100243	1:500 (FACS)		https://www.biolegend.com/en-us/products/biotin-anti-mouse-cd3-antibody-10023
ICAM2 (CD102)	BD Biosciences	553326	(Isolation of ECs)		https://www.fishersci.com/shop/products/anti-cd102-clone-3c4-mic2-4-bd-0-5mg-unlabeled/bdb553326

Cultured Cells

Name	Vendor or Source	Sex (F, M, or unknown)	Persistent ID / URL
Bovine aortic endothelial cells	Bovine aorta (isolated in the lab)	Female & Male	N/A
Mouse lung endothelial cells	Mouse lung (isolated in the lab)	Female & Male	N/A
Perivascular progenitor cells	Mouse subcutaneous white adipose tissue (isolated in the lab)	Female & Male	N/A

Oligonucleotides

Gene ID	siRNA target sequence (5'→3')	Use	Source
<i>PKG1α-1</i>	F: GGAUUGACAUGAUAGAAUUUCCAAA	siRNA transfection	Integrated DNA Technologies
	R: UUUGGAAAUUCTAUCAUGTCAAUCCCC		
<i>PKG1α-2</i>	F: GACAUGAUAGAAUUUCCAAAGAAA	siRNA transfection	Integrated DNA Technologies
	R: UCUUCUUUGGAAAUUCUAUCAUGUCA		
<i>PKG1α-3</i>	F: AUGAUAGAAUUUCCAAAGAAGAUTG	siRNA transfection	Integrated DNA Technologies
	R: CAAUCUUCUUUGGAAAUUCUAUCAUGU		
<i>Mcad</i>	F: ACTGACGCCGTTTCAGATTTT R: GCTTAGTTACACGAGGGTGATG	qRT-PCR	Integrated DNA Technologies
<i>Lcad</i>	F: ATGGCAAATACTGGGCATC R: TCTTGCGATCAGCTCTTTCA	qRT-PCR	Integrated DNA Technologies
<i>Ppara</i>	F: GAGAATCCACGAAGCCTACC R: AATCGGACCTCTGCCTCTTT	qRT-PCR	Integrated DNA Technologies
<i>Pgc1a</i>	F: GTAAATCTGCGGGATGATGG R: AGCAGGGTCAAATCGTCTG	qRT-PCR	Integrated DNA Technologies
<i>Hdha</i>	F: TCAGGAGGGCTCAAAGAATAA R: GAAAGCCAAGCCCAAAGAC	qRT-PCR	Integrated DNA Technologies
<i>Hdhb</i>	F: GCCAACAGACTGAGGAAGGA R: AACTGGCAAGGCTGGATT	qRT-PCR	Integrated DNA Technologies
<i>Cd36</i>	F: CCATTGGTGATGAAAAAGCA R: GATCGGCTTTACCAAAGATGTAG	qRT-PCR	Integrated DNA Technologies

<i>Etf</i>	F: TGACAAAAAGTGACCCACCA R: GAACAAAGCCAGCATCAACA	qRT-PCR	Integrated DNA Technologies
<i>Etfb</i>	F: GGTAAGAGTGGAACGGGAAA R: TCTCCACCTTGACTCCTGCT	qRT-PCR	Integrated DNA Technologies
<i>Etfq</i>	F: CCTCTGTGGCTTTGAGTGGT R: TCGAAATCCATCACCTTGTTT	qRT-PCR	Integrated DNA Technologies
<i>Cox4i1</i>	F: CGCTGAAGGAGAAGGAGAAG R: GCAGTGAAGCCAATGAAGAA	qRT-PCR	Integrated DNA Technologies
<i>Uqcrc1</i>	F: TGCCAGAGTTTCCAGACCTT R: CCAAATGAGACACCAAAGCA	qRT-PCR	Integrated DNA Technologies
<i>Ndufv1</i>	F: TGTGAGACCGTGCTAATGGA R: CATCTCCCTTCACAAATCGG	qRT-PCR	Integrated DNA Technologies
<i>Ndufa9</i>	F: ATCCCTTACCCTTTGCCACT R: CCGTAGCACCTCAATGGACT	qRT-PCR	Integrated DNA Technologies
<i>Tfam</i>	F: CAAAAAGACCTCGTTCAGCA R: CTTCAGCCATCTGCTCTTCC	qRT-PCR	Integrated DNA Technologies
<i>Nrf1</i>	F: CTTCAGAACTGCCAACCACA R: GCTTCTGCCAGTGATGCTAC	qRT-PCR	Integrated DNA Technologies
<i>Ucp1</i>	F: CTGCCAGGACAGTACCCAAG R: TCAGCTGTTCAAAGCACACA	qRT-PCR	Integrated DNA Technologies
<i>18S</i>	F: GTAACCCGTTGAACCCATT R: CCATCCAATCGGTAGTAGCG	qRT-PCR	Integrated DNA Technologies
<i>Vegfa</i>	F: GGCAGCTTGAGTTAAACGAAC	qRT-PCR	Integrated DNA Technologies

	R: TGGTGACATGGTTAATCGGTC		
FOXA2	F: CCGACTGGAGCAGCTACTATG R: TACGTGTTTCATGCCGTTTCAT	qRT-PCR	Integrated DNA Technologies
C/EBPα	F: AAACAACGCAACGTGGAGAC	qRT-PCR	Integrated DNA Technologies
C/EBPβ	R: TGTCCAGTTCACGGCTCAG F: GGGTTGTTGATGTTTTTGTTT R: GAAACGGAAAAGGTTCTCAAAA	qRT-PCR	Integrated DNA Technologies
Creb	F: TGCCAACTCCAATTTACCAA R: ACCCCATCGGTACCATTGTT	qRT-PCR	Integrated DNA Technologies
Mfn1	F: CATTGCGTTTCGGTTTTTCCC R: GAAGGAGCAGTAGGAGTTGAAG	qRT-PCR	Integrated DNA Technologies
Mfn2	F: TGAATGCTTCCCCTCTCAAG R: TCCAGTTCTGTGTTCCCTGTG	qRT-PCR	Integrated DNA Technologies
Opa1	F: GTGTGCTGGAAATGATTGCTC R: TGGTGAGATCAAATCCCGAG	qRT-PCR	Integrated DNA Technologies
Ephx1	F: GAGTGGAGGAACTGCACACC R: GAAGCCAGGATGAGTTCCAG	qRT-PCR	Integrated DNA Technologies
Ephx2	F: GTTCGACCTTGACGGAGTG R: GCACCAAGCAGGAAGTCTCT	qRT-PCR	Integrated DNA Technologies
Cidea	F: GAATAGCCAGAGTCACCTTCG R: AGCAGATTCCCTTAACACGGC	qRT-PCR	Integrated DNA Technologies
Adipsin	F: CCATTGTTGAATTCTCCTCCA R: TTCCACTCTCTTGGGTTTCCTT	qRT-PCR	Integrated DNA Technologies

ARRIVE GUIDELINES

The ARRIVE guidelines (<https://arriveguidelines.org/>) are a checklist of recommendations to improve the reporting of research involving animals. Key elements of the study design should be included below to better enable readers to scrutinize the research adequately, evaluate its methodological rigor, and reproduce the methods or findings.

Study Design

Groups	Sex	Age	Number (prior to experiment)	Number (after termination)	Littermates (Yes/No)	Other description
(ApoE ^{-/-} -Vehicle, Control)	F	22 weeks (WD-16 weeks)	12	8	No	
(ApoE ^{-/-} -12,13-diHOME)	F	22 weeks (WD-16 weeks)	12	8	No	

Sample Size: Please explain how the sample size was decided. Please provide details of any a *prior* sample size calculation, if done.

The mean *en face* plaque area and complexity of atherosclerosis in ApoE^{-/-} mice treated with 12,13-diHOME or exposure at 30°C at 16-20 weeks of age, and fed a Western diet for 12-16 weeks, was 8.5% (standard deviation 4.4%). With a similar variability, a sample size of mice in each group would rely on the 80% power to detect a 50% change with a significance level of 0.05.

Inclusion Criteria

We have two main reasons for the choosing male mouse models in this study. First is to exclude the complex effects of the estrus cycle and other hormonal changes in female mice, which could potentially affect study results. Moreover, including female mice to study sex differences within our existing mouse models would also be cost- and time-prohibitive, given that our current study design already included 4 different genetically-modified mouse models with their respective controls (namely, ECIRS1 mice with C57BL6 mice, ETBRKO mice with ETBR(fl/fl) mice, eNOSKO mice with C57BL6 mice and VEIRKO mice with VEIRKO(fl/fl) mice), as well as 3 pharmacologically-treated ApoE^{-/-} mouse models and their respective controls, including AngII-, L-NAME- and 12,13-diHOME-treated mice.

Exclusion Criteria

Deaths or impaired behavior activity of mice during experiments were excluded since they have influenced the accuracy and validity of data in metabolic and cardiovascular research.