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Reporting Summary

Nature Research 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 Research policies, see Authors & Referees 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	XCalibur 4.0 software for LCMS data; Gen5 TS 2.09 software for Biotek microplate reader data; ImageStudio Lite Ver 5.2 (Licor) for western blot imaging; LightCycler 480 Software (V1.5) for qPCR data collection and Ct calling; Agilent XF96 Seahorse 1.4.2.3 for OCR data
Data analysis	Graph Pad Prism 6.0 were used to perform statistical analyses; Maven was used to analyze LCMS data (build 682); ImageStudio Lite Ver 5.2 was used for western blot analysis; FlowJo 10.4 (TreeStar) was used for flow cytometry analysis

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/reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research 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 list of figures that have associated raw data
- A description of any restrictions on data availability

All data sets generated during the current study are available upon request from the corresponding author.

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

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	no sample size calculation was performed prior to experiments; sample sizes were chosen based on experience.
Data exclusions	No data was excluded from successfully executed experiments with one exception (Supplemental Figure 7e, Hk2 expression in itaconate treated group-- one outlier was tested out based on Grubbs' test (alpha = 0.05))
Replication	Many of these experiments were repeated multiple times in both systems used.
Randomization	Plated cells were randomly allocated to each treatment group.
Blinding	Experiments were not conducted with blinding. Blinding was not possible as frequently one person was responsible for carrying out each experiment from beginning to data 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
☐	☒ Eukaryotic cell lines
☒	☐ Palaeontology
☐	☒ Animals and other organisms
☒	☐ Human research participants
☒	☐ Clinical data

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☐	☒ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used

Primary Antibodies:
anti-HIF-1α: CST, cat #14179, clone #D2U3T, lot 1, dilution 1:1000
anti-H3: CST, cat #4499, clone #D1H2, lot GR31757189-2, dilution 1:1000
anti- Tri-methyl H3K27: CST, cat #9733, clone #C36B11, lot 8, dilution 1:1000
anti-Tri-methyl H3K4: CST, cat #9751, clone #C42D8, lot 10, dilution 1:1000
anti- Tri-methyl H3K9: CST, cat #13969, lot 3, clone #D4W1U
anti- PDH E1: CST, cat #2784S, lot 2, dilution 1:1000
anti-p-PDH Ser 293: Novus, cat #NB110-93479, lot 0-1, dilution 1:1000
anti-p-PDH Ser 300: Calbiochem, cat#AP1064, , lot 2733462, dilution 1:1000
anti-α-tubulin: Abcam, cat #ab7291, lots GR3157113-5, GR310199-11, GR310199-6, dilution 1:1000
anti-IRG1: Abcam, cat#222411, lot GR3237083-1, clone EPR22066, dilution 1:1000
anti PDH E2: Abcam, cat #ab66511, lot GR257784-15, dilution 1:1000
anti lipoic acid: Calbiochem, cat# 467695, batch 3074980 , dilution 1:1000
anti-OGDH: Proteintech, cat#15212-1-AP, lot 27, dilution 1:1000
anti OGDH: Abcam, cat#ab87057, lot GR239911-18, dilution 1:1000
anti-B-actin: CST, cat#4967, lot 8, dilution 1:1000
Secondary Antibodies:
Licor IRDye 800CW Goat anti-rabbit, dilution 1:10000
Licor IRDye 680RD Goat anti-mouse, dilution 1:10000

Validation

all had previously been validated in mouse by providers or previous publications HIF-1α: https://www.cellsignal.com/products/primary-antibodies/hif-1a-d2u3t-rabbit-mab/14179; H3: https://www.cellsignal.com/products/primary-antibodies/histone-h3-d1h2-xp-rabbit-mab/4499; Tri-methyl H3K27: https://www.cellsignal.com/products/primary-antibodies/tri-methyl-histone-h3-lys27-c36b11-rabbit-mab/9733; Tri-methyl H3K4: https://www.cellsignal.com/products/primary-antibodies/tri-methyl-histone-h3-lys4-c42d8-rabbit-mab/9751 Tri-methyl H3K9: https://www.cellsignal.com/products/primary-antibodies/tri-methyl-histone-h3-lys9-d4w1u-rabbit-mab/13969 PDH E1: https://www.cellsignal.com/products/primary-antibodies/pyruvate-dehydrogenase-antibody/2784 p-PDH Ser 293: https://www.novusbio.com/products/pyruvate-dehydrogenase-e1-alpha-subunit-antibody_nb110-93479 p-PDH Ser 300: Rardin M.J., et. al. 2009. Anal. Biochem. 2, 157. α-tubulin: https://www.abcam.com/alpha-tubulin-antibody-dm1a-loading-control-ab7291.html IRG1: https://www.abcam.com/irg1-antibody-epr22066-ab222411.html PDH E2: https://www.abcam.com/pyruvate-dehydrogenase-e2-antibody-ab66511.html lipoic acid: http://

www.emdmillipore.com/US/en/product/Anti-Lipoic-Acid-Rabbit-pAb,EMD_BIO-437695#anchor_REF OGDH 116kD: https://www.ptglab.com/products/OGDH-Antibody-15212-1-

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	RAW 264.7 (ATCC® TIB-71™)
Authentication	RAW 264.7, which is a murine cell line, was not authenticated
Mycoplasma contamination	RAW 264.7 cell line tested negative for mycoplasma
Commonly misidentified lines (See ICLAC register)	no commonly misidentified cell lines were used

Animals and other organisms

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

Laboratory animals	For isolation of BMDM the following animals were used: Species: Mus musculus Strains: C57BL/6J and C57BL/6NJ FVB.129S6-Gt(ROSA)26Sortm2(HIF1A/luc)Kael/J crossed 8+ generations into C57BL/6 background (heterozygotes used) C57BL/6NJ-Acod1em1(IMPC)J/J (Irg1-/-) (homozygous) Age: 6-10 weeks of age Sex: male and females
Wild animals	study did not involve wild animals
Field-collected samples	study did not involve field-collected samples
Ethics oversight	Mice were bred and maintained according to protocols approved by the University of Wisconsin-Madison Institutional Animal Care and Use Committee

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

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	RAW 264.7 cells were incubated with 500nM CellRox Green in media for 1 hour. Following incubation, media was aspirated and cells were washed twice with PBS, and then detached from plates by Accutase (Innovative Cell Technologies) treatment for 15 minutes. Cells were then spun down (1000x g , 3 min) and resuspended in 2% FBS in PBS and stained with 5nM SyTOX Red (ThermoFisher) to measure cell viability
Instrument	MACs Quant 10 Analyzer (Miltenyi Biotec)
Software	FlowJo10.4 (Tree Star)
Cell population abundance	All live cells (>90% , > 50000 cells in each sample) were analyzed for CellRox Green Signal.
Gating strategy	Cells with very low FSC and SSC were gated out. Dead cells (SyTOX red positive) were then gated out. The median CellRox Green signal of the remaining population was determined. Single color controls of SyToX red and CellRox green were used for compensation. A control dyed with CellRox Green but without StTOX Red was used to determine gating boundary for live and dead cells.

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