nature research

Corresponding author(s): Matthew Vander Heiden

Last updated by author(s): Sep 21, 2021

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 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	XCalibur Version 4.2.47 was used to collect LC/MS data on a Thermo Fisher QExactive MS and UltiMate 3000 LC.
Data analysis	LC/MS data were analyzed in XCalibur QuanBrowser (v2.2). Western blot densitometry quantification was performed in ImageJ (v1.52K). Fastq files were de-multiplexed using the Illumina software bcl2fastq (v2.20). Cell counts for proliferation assays, barcode counts for the PRISM multiplexed growth assay, metabolite levels for LC/MS data, and all other minor data were collated using Microsoft Excel (v16.53) to generate csv files from raw data and then subject to simple manipulations such as normalization, averaging, log transformation, or running statistical tests and plotted using scripts we wrote in R (v3.6.1) using jupyter notebook (ipython v5.8.0; jupyter-notebook v5.7.9) using the package collection tidyverse (v1.3.0) including ggplot (v3.3.2).

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 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

Unprocessed western blot images, integrated peak areas for metabolite level quantification, and processed data from comparative analysis of barcode counts of 489 human cancer cell lines in each condition in the PRISM multiplexed growth assay are provided as source data for Figures 1-5 and Extended Data Figures 1-5.

PCR amplicon sequencing results including raw barcode counts for each cancer cell line in the PRISM 489 cell line pool in each condition/replicate are provided in Supplementary Table 1.

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	We followed standards in the field for determining reasonable sample sizes for cell culture experiments; all experiment were performed with 3 biologically independent replicates or more (for some experiments up to 6, notated in figure legends). Further, we performed a power analysis to determine the required sample size to detect our estimated effect size in vivo (based on in vitro measurements) before beginning mouse experiments, as required by the MIT Committee on Animal Care Guidelines.
Data exclusions	No data were excluded.
Replication	We followed standards in the field for replication regarding biological and technical replicates for measurements as well as pseudo-replication methods such as showing the same results in multiple cell lines and in vitro as well as in vivo. All experiment were performed with 3 biologically independent replicates or more (for some experiments up to 6, notated in figure legends).
Randomization	Sample order was randomized for LC/MS experiments to avoid systematic bias in due to sampling/column carryover. For all experiments we avoided treating or processing samples in the same order in any systematic fashion to avoid systematic technical covariates.
Blinding	Investigators were blinded to identity of mice in all animal studies. Investigators were not blinded to allocation during all other experiments and outcome assessment.

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	Primary: MTR (Abcam, Cambridge, UK, ab66039, 1:1000), β-actin (Cell Signaling Technology, Danvers, MA, 8457 clone D6A8, 1:2500), phospho-T389 S6K (Cell Signaling Technology, Danvers, MA, 9234 clone 108D2, 1:1000) S6K (Cell Signaling Technology, Danvers, MA, 9202, 1:1000), phospho-S235/6 RPS6 (Cell Signaling Technology, Danvers, MA, 4858 clone D57.2.2E, 1:1000), RPS6 (Cell Signaling Technology, Danvers, MA, 2217 clone 5G10, 1:1000), or mono/dimethyl lysine residues (Abcam, Cambridge UK, ab23366, 1:1000). Secondary: HRP-conjugated anti-rabbit IgG antibodies (Cell Signaling Technology, Danvers, MA, 7074, 1:5000).
Validation	Antibodies were pre-validated by Abcam and Cell Signaling Technology. Validation information is supplied in the URLs below. MTR: https://www.abcam.com/mtr-antibody-ab66039.html β-actin: https://www.cellsignal.com/products/primary-antibodies/b-actin-d6a8-rabbit-mab/8457?site-search-type=Products&N=4294956287&Ntt=actin+8457&fromPage=plp&_requestid=1012231 Phospho-S6k : https://www.cellsignal.com/products/primary-antibodies/phospho-p70-s6-kinase-thr389-108d2-rabbit-mab/9234?_=1622156251960&Ntt=9234&tahead=true S6K : https://www.cellsignal.com//products/primary-antibodies/p70-s6-kinase-antibody/9202 Phospho-RPS6 :https://www.cellsignal.com//products/primary-antibodies/phospho-s6-ribosomal-protein-ser235-236-d57-2-2e-xp-rabbit-mab/4858 RPS6 : https://www.cellsignal.com//products/primary-antibodies/s6-ribosomal-protein-5g10-rabbit-mab/2217

Mono-diMethyl-Lys : https://www.abcam.com/methylated-lysine-di-methyl--mono-methyl--antibody-ab23366.html

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	All cell lines were obtained from ATCC (Manassas, VA). A549 cells were originally derived from a lung adenocarcinoma in a male patient. T.T cells were originally derived from an oral metastasis of an esophageal squamous cell carcinoma in a male patient.
Authentication	Short tandem repeat (STR) profiling authentication was performed by the supplier, ATCC, and periodically after obtaining cell lines from ATCC. The most recent date of cell line validation by STR testing was December 2016.
Mycoplasma contamination	All cell lines regularly tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	We did not use any commonly misidentified cell lines.

Animals and other organisms

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

Laboratory animals	NOD.Cg-Prkdc[scid]Il2rg[tm1Wjl]/SzJ (NSG) mice were used. Brackets [ ] denote superscript text. All experiments were performed with male mice aged 12 weeks from The Jackson Laboratory (Bar Harbor, ME, USA). The mice were housed in a controlled light cycle and temperature/humidity conditions (18-23°C, 40-60% humidity).
Wild animals	No wild animals were used.
Field-collected samples	No field-collected samples were used.
Ethics oversight	The MIT Committee on Animal Care Guidelines provided ethical oversight and approval for the experiments in our manuscript.

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