nature portfolio

Corresponding author(s): David Hackam MD PhD

Last updated by author(s): Aug 15, 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	No software was used to collect data.
Data analysis	Prism 9.0, ImageJ Fiji2, 10x Genomics Cell Ranger 6.1.1, R package Seurat 4.2, R package ComplexHeatmap 2.14.0, R package clusterProfiler 4.6.0, ggraph 2.1.0., pySCENIC 0.12.1. SCope online

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

The raw data and processed files for the single-cell RNA sequencing are available on the NCBI Gene Expression Omnibus (GEO: GSE271065).

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	Sex and gender based analyses were not conducted in this study.
Reporting on race, ethnicity, or other socially relevant groupings	Race, ethnicity and other social determinants were similarly not considered.
Population characteristics	These characteristics are published within the manuscript. Median age ~62, Male: 30.8%. Total n=26
Recruitment	Patients diagnosed with SBS (n=14) were recruited from the Gastroenterology Clinic at Washington University in St. Louis School of Medicine and Barnes-Jewish Hospital based on their diagnosis. Control subjects (n = 12) were recruited from consenting patients undergoing routine endoscopic colon cancer screening or polyp surveillance at the Center for Advanced Medicine Endoscopy Center and the general surgical/colorectal surgical service.
Ethics oversight	The Institutional Review Board of Washington University School of Medicine approved the use of human tissue in this study (IRB Number 201504100)

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 formal sample size calculation was performed. Sample sizes (4-10 animals per group) were chosen to ensure sufficient power to detect meaningful differences, considering study feasibility, available resources, and ethical guidelines. These sizes align with those used in similar studies that have demonstrated significant effects in comparable models.
Data exclusions	Some qRT-PCR data were excluded from the analyses because the qC values were not adequate, indicating potential issues with amplification efficiency or sample quality. These exclusion criteria were pre-established to ensure the accuracy and reliability of the data. All other data were included in the analysis.
Replication	All key experiments were repeated independently 3 or more times. Consistent results were obtained across these replicates, confirming the reproducibility of the findings. Additionally, controls were included in each experimental run to verify the validity of the results. No significant discrepancies were observed, and all attempts at replication were successful. Therefore, the findings reported in this study are robust and reproducible.
Randomization	Animals were allocated into experimental groups using randomization to minimize potential biases. Randomization was not feasible or relevant for our patient study due to the small sample size and the need to group by SBS versus control. However, the dataset was matched by age to control for this variable and reduce potential confounding effects
Blinding	No blinding was conducted during data collection or analysis in this study. This decision was based on the nature of the experimental setup, where blinding was not deemed necessary or feasible. All efforts were made to minimize potential biases through standardized procedures and objective measurement criteria.

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
[ ]	[x] 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: rat anti-BRDU (Novus NB500-169/ab6326 1:200) goat Sucrase-Isomaltase Antibody (Santa Cruz sc-27603 1:200), rabbit Anti-Ki67 antibody (Abcam ab-a5580 1:200), mouse anti PCNA (Santa Cruz sc-56 1:200) , goat anti-e-cadherin (R&D Biosystems AF748 1:200), rabbit anti chromogranin-A (Abcam ab15160 1:200), rhodamine phalloidin (Sigma R415 1:200)

Validation: All selected antibodies have been validated by previous use in our own lab, ensuring consistency in experimental conditions and reproducibility of results. Additionally, each of these antibodies have been verified through multiple peer-reviewed publications, which further confirm their specificity and effectiveness in detecting the target proteins under various experimental conditions. In most cases, the manufacturers have published comprehensive validation data on their websites, including performance metrics in various applications such as Western blotting, immunohistochemistry, and flow cytometry.

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s): Human iPSC (hiPSC) cell line (from Fibroblasts, Episomal, Female) ASE-9209 was obtained from Applied Stem Cell (Milpitas CA)

Authentication: Each lot of human iPS cells has been tested for growth and viability following recovery from cryopreservation, morphology, immunohistochemistry for pluripotency markers (OCT4, SOX2, SSEA4, TRA-1-60, TRA-1-81, and AP staining

Mycoplasma contamination: Each lot of human iPS cells has been tested for the absence of bacteria, fungi, mycoplasma (CoA available upon request)

Commonly misidentified lines (See ICLAC register): no commonly misidentified cell lines were used in the study.

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: WT (stock no: 000664) and Rag1 KO (stock no: 002216) C57BL/6 mice were obtained from the Jackson Laboratory. 6-week-old mice of both sexes were used in the studies. Mice were housed in a controlled environment with a 12-hour light/dark cycle. The ambient temperature was maintained at 22 ± 2°C, and the relative humidity was kept at 50 ± 10%. Mice had ad libitum access to liquid feed throughout the study.

Wild animals: NA

Reporting on sex: Sex was not considered in study design

Field-collected samples: NA

Ethics oversight: Johns Hopkins University Institutional Animal Care and Use Committee (protocol MO20M276)

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