

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 (<i>n</i>) 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 <i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i>
☒	☐ 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. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted <i>Give P values as exact values whenever suitable.</i>
☒	☐ 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 <i>d</i> , Pearson's <i>r</i>), 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	For bulk RNA-seq data, libraries were prepared using Illumina TruSeq library preparation according to the manufacturer’s specifications. And, RNA sequencing was performed on an Illumina HiSeq2500 following the standard Illumina RNA-Seq protocol, with a read length of 2×100 bases. For proteomics data, raw data obtained from LC-MS/MS data were searched using the PEAKS Studio 10.5 (Bioinformatics Solutions, Waterloo, Canada) against the SwissProt database of Homo sapiens (human, UP00000564, downloaded 22/11/2019, 20379 entries) from Uniprot (http://www.uniprot.org/).
Data analysis	(1) Image analysis Image J Fiji (2) FACS analysis FlowJo V10 software (3) bulk RNA-seq data analysis Agilent 2100 Bioanalyzer system NGSQCToolkit Ver. 2.3.3 Cutadapt Ver. 1.18 Sickle Ver. 1.33 HISAT2 Ver. 2.0.5

StringTie Ver. 2.1.0

(4) Proteomic analysis
 NanoElute LC system
 TimsTof Pro
 PEAKS Studio Ver. 10.5

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](#)

Data are provided in the main manuscript, supplementary materials. Bulk RNA-seq data are uploaded in the Gene Expression Omnibus (GEO) (GSE261785) (<https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE261785>) and proteomic data are uploaded in ProteomeXchange (PXD050747) (<https://proteomecentral.proteomexchange.org/cgi/GetDataset?ID=PX050747>).

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

This study included a single participant, a 76-year-old female. The participant's sex and gender were not relevant to the research objectives or outcomes, and therefore no analysis was conducted regarding these factors.

Reporting on race, ethnicity, or other socially relevant groupings

Race, ethnicity, and other socially relevant groupings were not considered in this study, as they did not pertain to the research objectives or outcomes.

Population characteristics

This study involved one participant, a 76-year-old female. No additional demographic characteristics, such as race, ethnicity, or socioeconomic status, were pertinent to the study's research objectives or findings. The participant was selected based on availability and willingness to provide informed consent.

Recruitment

Participants were recruited from patients visiting the hospital. The study's purpose, procedures, and potential risks were explained through an IRB-approved explanatory statement. Participation was voluntary, and only individuals who provided informed consent were enrolled in the study.

Ethics oversight

The study was approved by the Research Review Board of Gangnam Severance Hospital, Yonsei University Health System (approval number: 3-2023-0347). All procedures followed ethical guidelines to protect participants' rights and well-being. Informed consent was obtained before the study began, and confidentiality and anonymity were rigorously maintained throughout.

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](https://www.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 statistical method was used to predetermine sample size. Sample sizes were determined based on whether data is reproducible in independent sets of experiments.

Data exclusions

No data was excluded.

Replication

Experiments were performed in triplicated unless specified.

Randomization

Samples were not randomized for this study.

Blinding

Blinding was not applicable since no comparison between sample groups were made.

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

anti-LDHB, Thermo Scientific, PA5-96736, Polyclonal, 1:200 for ICC
<https://www.thermofisher.com/antibody/product/LDHB-Antibody-Polyclonal/PA5-96736>

anti-EIF3E, NOVUS Biologicals, NBP1-84869, Polyclonal, 0.25-2 µg/mL for ICC
https://www.novusbio.com/products/eif3e-antibody_nbp1-84869

anti-SOX9, SANTA CRUZ Biotechnology, sc-166505, Monoclonal, 5 µg/mL for ICC, 1:100 for FACS
<https://www.scbt.com/p/sox-9-antibody-e-9>

anti-KI67, BD Bioscience, 556003, Monoclonal, 1:100 for ICC
<https://www.bdbiosciences.com/ko-kr/products/reagents/microscopy-imaging-reagents/immunofluorescence-reagents/purifiedmouse-anti-ki-67.556003>

anti-CD44, Abcam, ab6124, Monoclonal, 5 µg/mL for ICC, 1:100 for FACS
<https://www.abcam.com/en-us/products/primary-antibodies/cd44-antibody-f10-44-2-ab6124>

anti-KRT20, Abcam, ab76126, Monoclonal, 1:200 for ICC
<https://www.abcam.com/en-us/products/primary-antibodies/cytokeratin-20-antibody-epr1622y-cytoskeleton-marker-ab76126>

anti-Villin1, SANTA CRUZ Biotechnology, sc-7672, Polyclonal, 1:50 for ICC
<https://www.scbt.com/p/villin-antibody-c-19>

anti-Mucin2, SANTA CRUZ Biotechnology, sc-7314, Monoclonal, 1:50 for ICC
<https://www.scbt.com/p/mucin-2-antibody-ccp58>

anti-Lysozyme, Abcam, ab76784, Polyclonal, 1:200 for ICC
<https://www.abcam.com/en-us/products/primary-antibodies/lysozyme-antibody-ab76784>

anti-Chromogranin A, Thermo Scientific, MA5-14536, Monoclonal, 1:100 for ICC
<https://www.thermofisher.com/antibody/product/Chromogranin-A-Antibody-clone-SP12-Monoclonal/MA5-14536>

anti-ECAD, R&D Biosystems, AF648, Polyclonal, 1:200 for ICC
https://www.rndsystems.com/products/human-mouse-e-cadherin-antibody_af648

anti-ECAD, abcam, ab1416, Monoclonal, 1:100 for IHC
<https://www.citeab.com/antibodies/706564-ab1416-anti-e-cadherin-antibody-hecd-1-intercellu>

anti-ECAD, abcam, ab40772, Monoclonal, 1:100 for IHC
<https://www.abcam.com/en-us/products/primary-antibodies/e-cadherin-antibody-ep700y-intercellular-junction-marker-ab40772>

anti-FABP1, Cell signaling Technology, 13368, Monoclonal, 1:50 for ICC
<https://www.cellsignal.com/products/primary-antibodies/fabp1-d2a3x-xp-rabbit-mab/13368>

anti-CD34, SANTA CRUZ Biotechnology, sc-74499, Monoclonal, 1:50 for FACS
https://www.scbt.com/p/cd34-antibody-b-6?srltid=AfmBOoq_dcv0AFi6zyVHhXLCFXPO1V8_9ddaA1WNd1-Dy_3iRUjcpXkd

	APC anti-human CD3 (CYTEK), 20-0038, 1:100 for FACS https://cytekbio.com/products/apc-anti-human-cd3-ucht1?variant=40581241077796 Alexa Fluor 488 donkey anti-mouse IgG antibody, Invitrogen, A21202, 1:1000 https://www.thermofisher.com/antibody/product/Donkey-anti-Mouse-IgG-H-L-Highly-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-21202 Alexa Fluor 488 donkey anti-goat IgG antibody, Invitrogen, A11055, 1:1000 https://www.thermofisher.com/antibody/product/Donkey-anti-Goat-IgG-H-L-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-11055 Alexa Fluor 594 donkey anti-mouse IgG antibody, Invitrogen, A21203, 1:1000 https://www.thermofisher.com/antibody/product/Donkey-anti-Mouse-IgG-H-L-Highly-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-21203 Alexa Fluor 594 donkey anti-rabbit IgG antibody, Invitrogen, A21207, 1:1000 https://www.thermofisher.com/antibody/product/Donkey-anti-Rabbit-IgG-H-L-Highly-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-21207 goat anti-mouse IgG Alexa Fluor 488, Abcam, ab150113, 1:1000 https://www.abcam.com/en-us/products/secondary-antibodies/goat-mouse-igg-h-l-alex-fluor-488-ab150113 goat anti-rabbit IgG Alexa Fluor 594, Abcam, ab150080, 1:1000 https://www.abcam.com/en-us/products/secondary-antibodies/goat-rabbit-igg-h-l-alex-fluor-594-ab150080
Validation	We used only commercial antibodies which have been validated by the selling companies. Furthermore, the antibodies have been further validated in numerous published research papers citing the antibodies.

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research	
Cell line source(s)	H9 hESCs were purchased from WiCell. Human iPSCs were generated by our previous work.
Authentication	Cell lines were authenticated by short tandem repeat (STR) analysis, karyotyping, marker gene expression, Alkaline phosphatase (AP) staining, and in vitro differentiation.
Mycoplasma contamination	Mycoplasma contamination was routinely checked and negative results were obtained.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used in this 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	The male NIG (NOD/SCID deleted IL2Rg gene) aged 6-12 weeks was purchased from GHBIO, Daejeon, South Korea.
Wild animals	No wild animals were used in this study.
Reporting on sex	Gender and sex-based analyses were not conducted in this study. Following the models of intestinal epithelial injury described by Sugimoto et al., 2018 (Cell Stem Cell) and Kwon et al., 2024 (Nat Commun), which used male mice, we observed only the effects of transplanted cells, independent of sex. The sex of the animals used in the experiments is detailed in the Methods section.
Field-collected samples	No field-collected samples were used in this study.
Ethics oversight	The mouse experiments were performed with the approval of the Institutional Animal Care and Use Committee (IACUC) of KRIBB, under approval number KRIBB-AEC-22233.

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

Plants

Seed stocks No plant samples were used in this study.

Novel plant genotypes No plant samples were used in this study.

Authentication No plant samples were used in this study.

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 Cells were dissociated into single cells, fixed with 4% paraformaldehyde, and permeabilized with PBS containing 0.1% Triton X-100, followed by incubation with human fluorochrome block reagent (BD Pharmingen) in FACS buffer consisting of 1% FBS and 0.1% sodium azide (Sigma-Aldrich) in PBS at 4°C for 10 min. Then, cells were incubated with primary antibodies in FACS buffer at 4°C for 10 min. After the cells were washed with PBS, they were incubated with secondary antibodies in FACS buffer at 4°C for 20 min.

Instrument FACS analysis was performed with FACSVerse flow cytometer (BD Biosciences)

Software Data were analyzed using FlowJo V10 software (TreeStar, USA)

Cell population abundance 5000 cells were used for FACS analysis.

Gating strategy For Fig. 4c, cells were initially gated by FSC-A vs SSC-A for the exclusion of debris. For single cells, samples were further gated by FSC-H vs FSC-A.

In lymphocyte reaction assay (Supplementary Fig. 5a, 9), lymphocytes were gated in FSC-A vs SSC-A plot, followed by single cell gating in a FSC-H vs FSC-A plot and CD3 positive T cell gating.

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