nature research

Corresponding author(s): Eran Elinav

Last updated by author(s): Jul 1, 2019

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	TargetLynx (Waters), ImageJ, DIAMOND, Qiime 2 version 2018.4.0, Metaphlan, Python, FlowJo V8, Matlab R2013B, SPM version 12
Data analysis	Graphpad prism version 7, Microsoft Excel, TargetLynx (Waters), ImageJ, DIAMOND, Qiime 2 version 2018.4.0, Metaphlan, Python, FlowJo V8 (Tree Star), Matlab R2013B, SPM version 12, bcl2fastq, RSeQC

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

The sequencing data has been deposited at the European Nucleotide Archive database with accession number PRJEB32767

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	All in vivo experiment had at least N=5 in each group and were repeated at least 3 times independently. Key experiments were repeated by more than one researcher. The exact sample size for each experiment is reported in the Results, Methods and Figure legends sections. No sample size calculation was performed. Sample sizes were determined based on previous studies and by the gold standards accepted in the field.
Data exclusions	No data were excluded from the manuscript
Replication	All the experiments in the manuscript were independently repeated at least 3 times. Key experiments were repeated by two independent researchers. All attempts of replication were successful and individual repeats are presented in ED and SI sections
Randomization	All in vivo experiments were randomized and controlled for gender, weight and cage effects
Blinding	In all in vivo experiments the researches were blinded

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	anti-CD45 clone 104, cat. 109831, dilution 1:200, Biolegend anti-CD11b clone M1/70 cat. 101229 dilution 1:200, Biolegend anti-CD3 clone 17A2, cat. 100212, dilution 1:200, Biolegend anti-B220 clone RA3-6B2 cat. 103207 dilution 1:200, Biolegend anti-Ly6G clone 1A8 cat. 127613 dilution 1:200, Biolegend anti-Ly6C clone HK1.4 cat. 128045 dilution 1:200, Biolegend anti-CD11c clone N418 cat. 117307 dilution 1:200, Biolegend anti-EpCAM clone G8.8 cat. 118227 dilution 1:200, Biolegend
Validation	All antibodies commercially available and validated by the manufacturer.

Animals and other organisms

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

Laboratory animals	Female and male C57bl6 wt and SOD1-Tg G93A mice were used between the ages of 40-180 days
Wild animals	The study did not involve wild animals
Field-collected samples	The study did not involve samples collected from the field
Ethics oversight	All experimental procedures were approved by the Weizmann Institute of Science IACUC 02660418-3

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

Human research participants

Policy information about studies involving human research participants

Population characteristics	37 ALS patients and 29 household healthy control individuals were enrolled to the study. Subjects were males and females aged 18-70 currently not following any interventional diet other than advise to avoid nutritional deficiencies and are able to provide informed consent. For detailed information please see Methods section.
Recruitment	All patients and healthy control individuals were recruited at the ALS clinic at Hadassah Medical Center, Jerusalem, Israel. Patients and their family members healthy control individuals were recruited by a clinical coordinator at the clinic, were given explanations about the study and signed an informed consent form. Every patient who met the inclusion criteria mentioned in the Methods section was included in the study. No self-selection biases were expected.
Ethics oversight	The study was approved by the Hadassah Medical Center Institutional Review Board (HMO-16-0396) and WIS IRB 365-1

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

Clinical data

Policy information about clinical studies

All manuscripts should comply with the ICMJE guidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.

Clinical trial registration	The trail was not registered online. patients and healthy control individuals were recruited at the ALS clinic at Hadassah Medical Center, Jerusalem, Israel
Study protocol	The full trail protocol appears in the manuscript, Methods section.
Data collection	Fecal and serum samples were collected for three years
Outcomes	Microbiome composition and functions (16S rDNA and shotgun sequencing) and serum metabolites (LC/MS) were measured

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	Mouse colon and small intestinal Lamina propria and spinal cord immune fractions were prepared as described in Methods.
Instrument	BD-LSRFortessa cytometer was used for the analysis
Software	FlowJo V10 (Tree Star) software
Cell population abundance	Cell populations sizes are reported in Extended Data Figure 2. Purity was validated using the appropriate single stained and isotype controls
Gating strategy	Standard Lymphoid and Myeloid panels were used as described in Methods and SI Figure 1

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

Magnetic resonance imaging

Experimental design

Design type	Resting state, block designed study was done
Design specifications	Randomized and blinded blocks were made with N=5 mice in each group, with scanning time of approximately 30 mins per mouse.
Behavioral performance measures	A resting state MRI was done with no tasks performance analyses.

Acquisition

Imaging type(s)	Structural T2 maps
Field strength	9.4 Tesla
Sequence & imaging parameters	a repetition delay (TR) of 3000 ms, 16-time echo (TE) increments (linearly from 10 to 160ms), matrix dimension of 256 x 128 (interpolated to 256 x 256) and two averages, corresponding to an image acquisition time of 12 min 48 sec. The T2 dataset consisted of 16 images per slice. Thirteen continuous slices with slice thickness of 1.00 mm were acquired with a field of view (FOV) of 2.0 x 2.0 cm2.
Area of acquisition	A whole-brain analysis was done

Diffusion MRI ☐ Used ☒ Not used

Preprocessing

Preprocessing software	All image analysis was performed using homemade scripts written in Matlab R2013B
Normalization	For optimal suitability to a mouse brain atlas (correction of head movements image artifacts), all images went through atlas registration: reslicing, realignment and smoothing.
Normalization template	SPM software (version 12, UCL, London, UK).
Noise and artifact removal	The multi-echo signal was fitted to a mono-exponential decay to extract the T2 value for each image pixel.
Volume censoring	See Methods page 14

Statistical modeling & inference

Model type and settings	A t-test was used to compare means of two groups. A p value of less than 0.01 was considered statistically significant.
Effect(s) tested	T2 relaxation time (msec) value

Specify type of analysis: ☐ Whole brain ☐ ROI-based ☒ Both

Anatomical location(s)	Anatomical locations were determined using the mouse brain atlas
Statistic type for inference (See Eklund et al. 2016)	A t-test was used to compare means of two groups. A p value of less than 0.01 was considered statistically significant.
Correction	Co-registration inter-subject and intra-subject was applied before the MRI dataset analysis.

Models & analysis

n/a	Involved in the study
☒	☐ Functional and/or effective connectivity
☒	☐ Graph analysis
☒	☐ Multivariate modeling or predictive analysis