nature portfolio

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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	Prism 9, Prism 10, Microsoft Office 365 for Mac, Image J/Fiji
Data analysis	All software used for data collection and image analysis was from publicly available commercial computer software vendors in our lab or university: Prism 9, Prism 10, Microsoft Office 365 for Mac, Zeiss Zen Blue, FIJI 1.52i with 3D suite plugin

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

All information has been stated in the manuscript.

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	All iPSC experiments were conducted on age, sex, and passaged matched iPSCs. Our previous analyses (Coyne et al, Science Translational Medicine, 2021 and Coyne et al, Neuron, 2020) have not identified any strong differences in Nup alterations in iPSC lines originating from male vs female patients. As a result,we have used equal numbers of male and female control and C9orf72 iPSC lines in our analyses, and have not specifically evaluated the influence of sex on our outcomes.
Reporting on race, ethnicity, or other socially relevant groupings	Race and ethnicity of patients from which iPSCs were generated are detailed in Supplemental File 1. Additional data is available through the Answer ALS data portal.
Population characteristics	Induced Pluripotent Stem Cell lines (iPSC) were available through the Answer ALS Project and obtained via the Cedars Sinai iPSC repository (https://www.cedars-sinai.edu/Research/Research-Cores/Induced-Pluripotent-Stem-Cell-Core-/Answer-ALS-Project.aspx). All patient procedures and IRB approval were previously reported in Baxi et al, Nature Neurosceince, 2021.
Recruitment	Eligible patients and/or family members are recruited from our ALS clinic for voluntary participation in blood and autopsy donations.
Ethics oversight	All blood (for iPSC generation from PBMCs) and autopsied tissue collections are Johns Hopkins IRB approved with Johns Hopkins ethics oversight. All patient information is HIPPA compliant. All iPS cell lines have been previously reported including all regulatory oversight (Baxi et al, Nat. Neuroscience 2021)

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	Samples sizes were chosen based on previous expertise, knowledge from past experiments and statistical considerations using similar iPSC derived motor neuron model systems and current accepted standards based on journal policies and a review of the literature. No statistic analysis was used to pre-determine sample size. Sample sizes were chosen to be similar or exceed those previously reported in the literature. Each experiment was repeated with multiple biological replicates as stated and the statistical analysis demonstrates that our sample sizes revealed significant differences between groups.
Data exclusions	No data were excluded from the study.
Replication	All experiments were done with a minimum number of replicates based on previous expertise in statistical analyses of similar experimental datasets. Samples sizes were chosen based on previous expertise and knowledge from past experiments using similar iPSC-derived model systems and current accepted standards based on literature review. Replicates are detailed within the manuscript.
Randomization	Not relevant to this study.
Blinding	Investigators were blinded to iPSC line and genotypic and treatment information for all experiments presented in this study.

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

Primary Antibodies:
Rabbit Anti-N terminal TDP-43 (ProteinTech 10782-2-AP)
Mouse Anti-N terminal TDP-43 (Abcam ab104223)
Rabbit Anti-C terminal TDP-43 (ProteinTech 12892-1-AP)
Rat Anti-Phospho TDP-43 (Ser409/410) (BioLegend 829901)
Guinea Pig Anti-Map2 (Synaptic Systems 188 004)
Mouse Anti-CHMP7 (Santa Cruz Biotechnology sc-271805)

Validation

The use of the primary antibodies and their protein specificity was determined by each commercial vendor as provided by their websites:
N terminal TDP-43 (ProteinTech) Species Reactivity: Human, Mouse, Rat, Zebrafish; Applications: WB, RIP, IHC, IF, IEM, FC, CoIP, ChIP, ELISA
N terminal TDP-43 (Abcam) Species Reactivity: Human, Mouse, Rat; Applications: WB, ICC/IF, Flow Cyt
C terminal TDP-43 (ProteinTech) Species Reactivity: Human, Mouse, Rat, Monkey, Zebrafish, Chicken, Drosophila; Applications: WB, IHC, IF/ICC, IP, CoIP, ChIP, RIP, ELISA
Phospho TDP-43 (BioLegend) Species Reactivity: Human, Rat; Applications: WB, IHD, ICC, ELISA
Map2 (Synaptic Systems) Species Reactivity: Human, Mouse, Rat; Applications: WB, ICC, IHC
CHMP7 (Santa Cruz Biotechnology) Species Reactivity: Human, Mouse, Rat; Applications: WB, IP, IF, ELISA

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)

iPSC lines were obtained from Cedars Sinai Answer ALS Repository. Cell line demographic information provided in Supplemental File 1. Description of the original lines acquisition in Baxi et al, Nature Neurosceince 2021.

Authentication

iPSCs were tested for pluripotency by Cedars Sinai iPSC Core. iPSC derived spinal neurons were validated with known neuronal markers.

Mycoplasma contamination

All cell lines tested negative for mycoplasma contamination.

Commonly misidentified lines
(See ICLAC register)

No commonly misidentified cell lines were used.

Plants

Seed stocks

Report on the source of all seed stocks or other plant material used. If applicable, state the seed stock centre and catalogue number. If plant specimens were collected from the field, describe the collection location, date and sampling procedures.

Novel plant genotypes

Describe the methods by which all novel plant genotypes were produced. This includes those generated by transgenic approaches, gene editing, chemical/radiation-based mutagenesis and hybridization. For transgenic lines, describe the transformation method, the number of independent lines analyzed and the generation upon which experiments were performed. For gene-edited lines, describe the editor used, the endogenous sequence targeted for editing, the targeting guide RNA sequence (if applicable) and how the editor was applied.

Authentication

Describe any authentication procedures for each seed stock used or novel genotype generated. Describe any experiments used to assess the effect of a mutation and, where applicable, how potential secondary effects (e.g. second site T-DNA insertions, mosiacism, off-target gene editing) were examined.