nature research

Corresponding author(s): Xinyu Zhao

Last updated by author(s): Jan 3 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	NIS-Elements for confocal imaging (Nikon); pClamp 10.4 for whole-cell patch-clamp recordings (Molecular Devices); ImageJ for imaging analysis (RRID: SCR_003070); Quantity One for imaging regular genotyping gels (BioRad) ; Fusion Accuscan program for open field tests (Omnitech Electronics)
Data analysis	Neurolucida, StereoInvestigator, Image J, Igor Pro, STAR v2.5.3a, EBSeq v1.18.0, PANTHER, WEbGESTALT, SPSS v22, GraphPad 6

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

Submission of sequencing data to GEO with GEO # GSE117111

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	Sample size was used as dictated by literature on similar studies.
Data exclusions	No data were excluded from the analyses.
Replication	Replication of experiments was successful.
Randomization	All cells analyzed were randomly selected from in vivo and in vitro samples.
Blinding	Quantifications were performed by experimenters who were blind to the identity of the sample.

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

The primary antibodies used: chicken anti-GFP (1:500, Invitrogen, Carlsbad, CA, #A10262), rat anti-Ki67 (SolA15) (1:500, eBioscience, 14-5698, San Diego, CA, USA), rabbit anti-GFAP (1:2000, DAKO, #Z0334, Carpinteria, CA, USA), chicken anti-Nestin (1:500, Aves Labs, #NES0407, Tigard, OR, USA), rabbit anti-Doublecortin (1:500, Cell Signaling Technology, #4604S, Beverly, MA, USA), rabbit anti-cleaved caspase-3 (Asp175) (1:500, Cell Signaling, #9661, Danvers, MA, USA), mouse anti-NeuN (clone A60) (1:500, Millipore, MAB377, Billerica, MA, USA), rabbit anti-S100β (1:1000, Dako, Z0334, Carpinteria, CA, USA), mouse anti-FMRP (clone 1C3) (1:500, Millipore, MAB2160), mouse anti-Nitrotyrosine (39B6) (1:500, Santa Cruz Biotechnology, sc-32757, Texas, DA, USA), mouse anti-Huntingtin (3E10) (1:500, Santa Cruz Biotechnology, sc-47757, Texas, DA, USA), mouse anti-Opa1 ([EPR11057(B)]) (1:1000, Abcam, ab157457, Cambridge, MA), mouse anti-Mfn1 (3C9) (1:1000, Abcam, ab57602, Cambridge, MA), rabbit anti-Mfn2 (1:500, Proteintech, 12186-1-AP, Rosemont, IL), rabbit anti-Drp1 (EPR19274) (1:1000, Abcam, ab184247, Cambridge, MA) and mouse anti-FMRP (7G1-1) (1:500, DSHB, 7F1-1-C, University of Iowa, Department of Biology, IA).
Fluorescent secondary antibodies for IHC used by 1:500 dilution: goat anti-chicken-488 (A11039, Invitrogen), goat anti-mouse 568 (A11004, Invitrogen), goat anti-rabbit 647 (A21245, Invitrogen), donkey anti-goat 568 (A11057, Invitrogen), donkey anti-rabbit 647 (A31573, Invitrogen), goat anti-mouse 647 (A21235, Invitrogen), goat anti-rabbit 568 (A11011, Invitrogen), and donkey anti-mouse 647 (A31571, Invitrogen).
Fluorescent secondary antibodies for WB used by 1:5000 dilution: IRDye 800CW Goat anti-Mouse IgG (H+L) (925-32210, LiCore), IRDye 800CW Goat anti-rabbit IgG (H+L) (925-32211, LiCore), IRDye 680LT Goat anti-Mouse IgG (H+L) (925-68020), LiCore), IRDye 680LT Goat anti-Rabbit IgG (H+L) (925-68021), LiCore)

Validation

primary antibodies:
1. chicken anti-GFP (Invitrogen, Carlsbad, CA, #A10262), mouse, IHC, validation on manufacturer's website: https://www.thermofisher.com/antibody/product/GFP-Tag-Antibody-Polyclonal/A10262#/IHC-content.
2. rat anti-Ki67 (SolA15) (1:500, eBioscience, 14-5698, San Diego, CA, USA), mouse, IHC, validation on manufacturer's website: https://www.thermofisher.com/antibody/product/Ki-67-Antibody-clone-SolA15-Monoclonal/14-5698-82.
3. rabbit anti-GFAP (1:2000, DAKO, #Z0334, Carpinteria, CA, USA), mouse, IHC, validation on publication: PMID: 21516088.
4. chicken anti-Nestin (1:500, Aves Labs, #NES0407, Tigard, OR, USA), mouse, IHC, validation on publication: PMID: 28100736.

5. rabbit anti-Doublecortin (1:500, Cell Signaling Technology, #4604S, Beverly, MA, USA), mouse, IHC, validation on manufacturer's website: https://www.cellsignal.com/products/primary-antibodies/doublecortin-antibody/4604.
6. rabbit anti-cleaved caspase-3 (Asp175) (1:500, Cell Signaling, #9661, Danvers, MA, USA), mouse, IHC, validation on manufacturer's website: https://www.cellsignal.com/products/primary-antibodies/cleaved-caspase-3-asp175-antibody/9661.
7. mouse anti-NeuN (clone A60) (1:500, Millipore, MAB377, Billerica, MA, USA), mouse, IHC, validation on manufacturer's website: http://www.emdmillipore.com/US/en/product/Anti-NeuN-Antibody-clone-A60,MM_NF-MAB377?ReferrerURL=https%3A%2F%2Fwww.google.com%2F&bd=1.
8. rabbit anti-S100β (1:1000, Dako, Z0334, Carpinteria, CA, USA), validation on publication: PMID: 28100736.
9. mouse anti-FMRP (clone 1C3) (1:500, Millipore, MAB2160), mouse, IHC, validation on manufacturer's website: http://www.emdmillipore.com/US/en/product/Anti-Fragile-X-Mental-Retardation-Protein-Antibody-clone-1C3,MM_NF-MAB2160.
10. mouse anti-Nitrotyrosine (39B6) (1:500, Santa Cruz Biotechnology, sc-32757, Texas, DA, USA), mouse, IHC, validation on manufacturer's website: https://www.scbt.com/scbt/product/nitrotyrosine-antibody-39b6.
11. mouse anti-Huntingtin (3E10) (1:500, Santa Cruz Biotechnology, sc-47757, Texas, DA, USA), mouse, IHC, validation on manufacturer's website: https://www.scbt.com/scbt/product/huntingtin-antibody-3e10.
12. mouse anti-Opa1 ([EPR11057(B)]) (1:1000, Abcam, ab157457, Cambridge, MA), mouse, WB, validation on manufacturer's website: https://www.abcam.com/opa1-antibody-epr11057b-ab157457.html.
13. mouse anti-Mfn1 (3C9) (1:1000, Abcam, ab57602, Cambridge, MA), mouse, WB, validation on manufacturer's website: https://www.abcam.com/mitofusin-1-antibody-3c9-ab57602.html.
14. rabbit anti-Mfn2 (1:500, Proteintech, 12186-1-AP, Rosemont, IL), mouse, WB, validation on manufacturer's website: https://www.ptglab.com/products/MFN2-Antibody-12186-1-AP.htm.
15. rabbit anti-Drp1 (EPR19274) (1:1000, Abcam, ab184247, Cambridge, MA), mouse, WB, validation on manufacturer's website: https://www.abcam.com/drp1-antibody-epr19274-ab184247.html.
16. mouse anti-FMRP (7G1-1) (1:500, DSHB, 7F1-1-C, University of Iowa, Department of Biology, IA), mouse, IP, validation on manufacturer's website: http://dshb.biology.uiowa.edu/7G1-1.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	Human FXS iPSC line (FX11-7) was published. GM1 (GM00498-4) iPSCs were generated from fibroblasts from an apparently healthy 3 year old male obtained from Coriell (GM00498) using the same Yamanaka method. More details are provided in "Supplementary Methods" subheading "Human cell culture, neural differentiation and transplantation".
Authentication	Authentication includes confirmation of correct karyotype, pluripotency, and absence of pathogen or mycoplasma contamination. Human FXS iPSC line (FX11-7) has been fully authenticated by our lab and WiCell (https://www.wicell.org/) and has been published (Doers et al 2014). GM1 (GM00498-4) iPSC line has been authenticated for pluripotency, karyotype, mycoplasma by our lab and WiCell, and the test for human pathogen is currently being performed by WiCell before distribution to scientific community.
Mycoplasma contamination	The cell lines used in this study were tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	No misidentified lines were used in this study.

Animals and other organisms

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

Laboratory animals	Mice, C57BL/6, male and female, different ages according to experiments (P0 up to 4 months old)
Wild animals	No wild animals were used in this study
Field-collected samples	No field-collected samples were used in this study
Ethics oversight	We performed all procedures involving live mice in accordance with the NIH Guide for the Care and Use of Laboratory Animals and the protocols approved by the University of Wisconsin-Madison Animal Care and Use Committee (IACUC). We performed all experiments involving human iPSCs based on the guideline of University of Wisconsin Stem Cell Research Oversight (SCRO) committee. We performed all experiments involving biohazard materials based on protocols approved by University of Wisconsin Biosafety (BIO) committee.

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

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	DG tissue were isolated from 6-7 weeks old Fmr1-/y; Dcx-DsRed mice and their WT littermates.
Instrument	Becton Dickinson FACS Aria II
Software	StereoInvestigator software (MicroBrightField)
Cell population abundance	All cell populations were isolated into single cells using a Becton Dickinson FACS Aria II contained in a Biosafety Carbinet using 20 psi pressure and 100-µm nozzle aperture. 10,000 total alive or Dcx-DsRed+ alive Cells were collected directly in Trizol.
Gating strategy	Gates were set manually by using control samples (same types of cells isolated from mice without Dcx-DsRed transgene).

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