Poly-GP in cerebrospinal fluid links C9orf72-associated dipeptide repeat expression to the asymptomatic phase of ALS/FTD

Carina Lehmer, Patrick Oeckl, Jochen H. Weishaupt, Alexander E. Volk, Janine Diehl-Schmid, Matthias L. Schroeter, Martin Lauer, Johannes Kornhuber, Johannes Levin, Klaus Fassbender, Bernhard Landwehrmeyer, German Consortium for Frontotemporal lobar degeneration, Martin H. Schludi, Thomas Arzberger, Elisabeth Kremmer, Andrew Flatley, Regina Feederle, Petra Steinacker, Patrick Weydt, Albert C. Ludolph, Dieter Edbauer, and Markus Otto

Corresponding authors: Dieter Edbauer, German Center for Neurodegenerative Diseases DZNE, Munich and Marcus Otto, Ulm University Hospital, Munich

Review timeline:

<table>
<thead>
<tr>
<th>Event</th>
<th>Date</th>
</tr>
</thead>
<tbody>
<tr>
<td>Submission date</td>
<td>19 December 2016</td>
</tr>
<tr>
<td>Editorial Decision</td>
<td>30 January 2017</td>
</tr>
<tr>
<td>Revision received</td>
<td>21 February 2017</td>
</tr>
<tr>
<td>Editorial Decision</td>
<td>24 March 2017</td>
</tr>
<tr>
<td>Revision received</td>
<td>27 March 2017</td>
</tr>
<tr>
<td>Accepted</td>
<td>28 March 2017</td>
</tr>
</tbody>
</table>

Editor: Céline Carret

Transaction Report:

(Note: No Peer Review Process File is available with this article, as the authors have chosen not to make the review process public in this case.)
Corresponding Author Name: Dieder Edsbaier & Markus Otto

Manuscript Number: EMBO-2016-07486

Reporting Checklist for Life Sciences Articles (Rev. July 2015)

This checklist is used to ensure good reporting standards and to improve the reproducibility of published results. These guidelines are consistent with the Principles and Guidelines for Reporting Preclinical Research issued by the NIH in 2014. Please follow the journal’s authorship guidelines in preparing your manuscript.

A. Figures

1. Data

The data shown in figures should satisfy the following conditions:
- The data were obtained and processed according to the Klotz’s best practice and are presented to reflect the results of the experiments in an accurate and unbiased manner.
- Figure panels include only data points, measurements or observations that can be compared to each other in a scientifically meaningful way.
- Graphs include clearly labeled error bars for independent experiments and sample sizes. Unless justified, error bars should not be shown for technical replicates.
- If n = 5, the individual data points from each experiment should be plotted and any statistical test employed should be justified.
- Source Data should be included to report the data underlying graphs. Please follow the guidelines set out in the authorship guidelines on Data Presentation.

2. Captions

Each figure caption should contain the following information, for each panel where they are relevant:
- A specification of the experimental system investigated (e.g. cell line, species name).
- The assay[s] and method[s] used to carry out the reported observations and measurements.
- An explicit mention of the biological and chemical entity[ies] that are being measured.
- The exact sample size (n) for each experimental group/condition, given as a number, not a range.
- A description of the sample collection allowing the reader to understand whether the samples represent technical or biological replicates (including how many animals, litters, cultures, etc.).
- A statement of how many times the experiment shown was independently replicated in the laboratory.
- Definitions of statistical methods and measures:
  - Common tests, such as t-test (please specify whether paired or unpaired), simple t-tests, Wilcoxon and Mann-Whitney tests, can be unambiguously identified by name only, but more complex techniques should be described in the methods section;
  - Are tests one-sided or two-sided?
  - Are there adjustments for multiple comparisons?
  - Exact statistical test results, e.g. P = 0.001 vs. P < 0.05 vs. P < 0.01.
  - Definition of ‘center values’ as median or average.
  - Definition of error bars as s.d. or s.e.m.

Any descriptions too long for the figure legend should be included in the methods section and/or with the source data.

B. Statistics and general methods

1. How was the sample size chosen to ensure adequate power to detect a pre-specified effect size?

2. For animal studies, include a statement about sample size estimates even if no statistical methods were used.

3. Describe inclusion/exclusion criteria if samples or animals were excluded from the analysis. Were the criteria pre-established?

4. For animal studies, include a statement about randomization even if no randomization was used.

5. Were any steps taken to minimize the effects of subjective bias during group allocation or and when assessing results (e.g. blinding of the investigator)? If yes please describe.

6. For animal studies, include a statement about blinding even if no blinding was done.

7. For every figure, are statistical tests justified as appropriate?

8. Do the data meet the assumptions of the tests (e.g. normal distribution)? Describe any method used to assess this.

9. Have an estimate of variation within each group of data?

10. Describe the variance within the groups that are being statistically compared?

C. Reagents

USEFUL LINKS FOR COMPLETING THIS FORM

- http://clinicaltrials.gov
- http://www NIH.gov/policies/clinicaltrials.html
- http://www.mrc.ac.uk/TermsofUseResearchGuidanceUseofanimals/index.htm
- http://www.plosone.org
- http://www.compet-statement.org
- http://www.embopress.org
- http://bioinformatics.net/bertram/
- http://figshare.com
- http://www.embob.info
- http://biomodels.net/
- http://www.verbose.org
- http://www.ebi.ac.uk/eye
- http://biomodels.net/
D. Animal Models

4. Report species, strain, gender, age of animals and genetic modification status where applicable. Please detail housing and husbandry conditions and the source of animals.

5. For experiments involving invertebrates, include a statement of compliance with ethical regulations and identify the committee(ies) approving the experiments.

6. We recommend consulting the ARRIVE guidelines (see link list at top right) (Elmore, A.J., et al., PLoS Biol. 8(2), e1000362, 2010) to ensure that other relevant aspects of animal studies are adequately reported. See author guidelines, under ‘Reporting Guidelines’. See also: NIH (see link list at top right) and MRC (see link list at top right) recommendations. Please confirm compliance.

E. Human Subjects

1. Identify the committee(s) approving the study protocol.

2. Include a statement confirming that informed consent was obtained from all subjects and that the experiments conformed to the principles set out in the WMA Declaration of Helsinki and the Department of Health and Human Services Belmont Report.

3. For publication of patient photos, include a statement confirming that consent to publish was obtained.

4. Report any restrictions on the availability (e.g., on the use of human data or samples).

5. Report the clinical trial registration number (at ClinicalTrials.gov or equivalent), where applicable.

6. For phase II and III randomized controlled trials, please refer to the CONSORT flow diagram (see link list at top right) and submit the CONSORT checklist (see link list at top right) with your submission. See author guidelines, under ‘Reporting Guidelines’. Please confirm you have submitted this list.

7. For tumor marker prognostic studies, we recommend that you follow the REMARK reporting guidelines (see link list at top right). See author guidelines, under ‘Reporting Guidelines’. Please confirm you have followed these guidelines.

F. Data Accessibility

8. Provide accession codes for deposited data. See author guidelines, under ‘Data Availability’.

Data deposition in a public repository is mandatory for:

a. Proteins, DNA and RNA sequences
b. Microarrays and microarray raw data
c. Crystallographic data for small molecules
d. Functional genomics data

9. Deposition is strongly recommended for any datasets that are central and integral to the study, please consider the journal’s data policy. If no structured public repository exists for a given data type, we encourage the provision of data in the manuscript as a Supplementary Document (see author guidelines under ‘Expanded View’ or in the supplementary repositories such as Dryad (see link list at top right) or FigShare (see link list at top right)).

10. Access to human clinical and genomic datasets should be provided with as few restrictions as possible while respecting ethical obligations to the patients and relevant medical and legal issues. If practically possible and compatible with the individual consent agreement used in the study, such data should be deposited in one of the major public access controlled repositories such as dbGAP (see link list at top right) or EGA (see link list at top right), or equivalent, where applicable.

11. If an in silico or computational model is provided, this should be deposited in a public database such as BioModels (see link list at top right) or KEGG (see link list at top right) or equivalent, where applicable. If computer source code is provided with the paper, it should be deposited in a public repository or included in supplementary information.

G. Dual use research of concern

12. To show that antibodies were profiled for use in the system under study (assay and species), provide a citation, catalog number and/or clone number, supplementary information or reference to an antibody validation profile, e.g., Antibodypedia (see link list at top right), 12Tagger (see link list at top right).

13. Identify the source of cell lines and report if they were recently authenticated (e.g., by STR profiling) and tested for mycoplasma contamination.

For all hyperlinks, please see the table at the top right of the document.