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# ORP5/ORP8 localize to endoplasmic reticulum-mitochondria contacts and are involved in mitochondrial function

Romain Galmes, Audrey Houcine, Alexander R. van Vliet, Patrizia Agostinis, Cathy Jackson, Francesca Giordano

Corresponding author: Francesca Giordano, Institut Jacques Monod

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Corresponding Author Name: Francesca Giordano

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#### **Reporting Checklist For Life Sciences Articles**

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 (see link list at top rig

#### A- Figures

#### 1. Data

- The data shown in figures should satisfy the following conditions:

  → the data were obtained and processed according to the field'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 only for independent experiments and sample sizes where the
  - application of statistical tests is warranted (error bars should not be shown for technical replicates)
  - when n is small (n < 5), the individual data points from each experiment should be plotted alongside an error
  - → Source Data should be included to report the data underlying graphs. Please follow the guidelines set out in the author ship guidelines on Data Presentation (so

#### 2. Captions

#### Each figure caption should contain the following information, for each panel where they are relevant:

- a specification of the experimental system investigated (eg 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.
   an explicit mention of the biological and chemical entity(ies) that are altered/varied/perturbed in a controlled manner.
- 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 vs. unpaired), simple x2 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 values = x but not P values < x;
  - 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

se ensure that the answers to the following questions are reported in the ma to include a specific subsection in the methods section for statistics, reagents, animal models and human subjects.

In the pink boxes below, provide the page number(s) of the manuscript draft or figure legend(s) where he information can be located. Every question should be answered. If the question is not relevant to rour research, please write NA (non applicable).

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# B- Statistics and general methods

cs and general methods	Please fill out these boxes $lacksquare$	
1.a. How was the sample size chosen to ensure adequate power to detect a pre-specified effect size?	The size of biological samples (cells) was chosen in order to compare the specific analyzed conditions and obtain a significant data (Student's T-test). (Fig 1E, 1H,2C,2F,4D,4E, EV1D, EV4D, EV4E).	
1.b. For animal studies, include a statement about sample size estimate even if no statistical methods were used.	NA	
2. Describe inclusion/exclusion criteria if samples or animals were excluded from the analysis. Were the criteria pre-established?	We have analyzed random samples (Fig 1E, 1H,2C,2F,4D,4E, EV1D, EV4D, EV4E).	
3. Were any steps taken to minimize the effects of subjective bias when allocating animals/samples to treatment (e.g. randomization procedure)? If yes, please describe.	Yes. The samples were analyzed multiple times in parallel experiments by different investigators to be sure that the analysis was unbiased. The cells analyzed were choosen randomly. (Fig 1E, 1H,2C,2F,4D,4E, EV1D, EV4D, EV4E).	
For animal studies, include a statement about randomization even if no randomization was used.	NA	
4.a. 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.	Yes. We applied the blinding of the investigator (Fig 1H, 2C,2F and EV1D).	
4.b. For animal studies, include a statement about blinding even if no blinding was done	NA	
5. For every figure, are statistical tests justified as appropriate?	Yes	
Do the data meet the assumptions of the tests (e.g., normal distribution)? Describe any methods used to assess it.	We used the Student's T-Test (unpaired) for indipendent samples. (Fig 1E, 1H,2C,2F,4D,4E, EV1D, EV4D, EV4E).	
Is there an estimate of variation within each group of data?	Yes. It can be represented as standard deviation or standard error of the mean. (Fig. 1E, 1H,2C,2F,4D,4E, EV1D, EV4D, EV4E).	
Is the variance similar between the groups that are being statistically compared?	Yes	

#### C- Reagents

6. To show that antibodies were profiled for use in the system under study (assay and species), provide a	We gave the species of antibodies used as well as the compagny we bought
citation, catalog number and/or clone number, supplementary information or reference to an antibody	them. Some antibodies were used in other studies which can be found in the
validation profile. e.g., Antibodypedia (see link list at top right), 1DegreeBio (see link list at top right).	material and method section
7. Identify the source of cell lines and report if they were recently authenticated (e.g., by STR profiling) and	HeLa cells
tested for mycoplasma contamination.	

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#### **D- Animal Models**

<ol><li>Report species, strain, gender, age of animals and genetic modification status where applicable. Please detail housing and husbandry conditions and the source of animals.</li></ol>	NA .
9. For experiments involving live vertebrates, include a statement of compliance with ethical regulations	NA
and identify the committee(s) approving the experiments.	
10. We recommend consulting the ARRIVE guidelines (see link list at top right) (PLoS Biol. 8(6), e1000412,	NA
2010) to ensure that other relevant aspects of animal studies are adequately reported. See author	
guidelines, under 'Reporting Guidelines' (see link list at top right). See also: NIH (see link list at top right)	
and MRC (see link list at top right) recommendations. Please confirm compliance.	

# E- Human Subjects

11. Identify the committee(s) approving the study protocol.	NA
12. Include a statement confirming that informed consent was obtained from all subjects and that the	NA
experiments conformed to the principles set out in the WMA Declaration of Helsinki and the Department	
of Health and Human Services Belmont Report.	
13. For publication of patient photos, include a statement confirming that consent to publish was	NA
obtained.	
<ol> <li>Report any restrictions on the availability (and/or on the use) of human data or samples.</li> </ol>	NA
<ol> <li>Report the clinical trial registration number (at ClinicalTrials.gov or equivalent), where applicable.</li> </ol>	NA
16. For phase II and III randomized controlled trials, please refer to the CONSORT flow diagram (see link list	NA
at top right) and submit the CONSORT checklist (see link list at top right) with your submission. See author	
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17. For tumor marker prognostic studies, we recommend that you follow the REMARK reporting guidelines	NA
(see link list at top right). See author guidelines, under 'Reporting Guidelines' (see link list at top right).	

# F- Data Accessibility

18. Provide accession codes for deposited data. See author guidelines, under 'Data Deposition' (see link list	NA NA
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Data deposition in a public repository is mandatory for:	
a. Protein, DNA and RNA sequences	
b. Macromolecular structures	
c. Crystallographic data for small molecules	
d. Functional genomics data	
e. Proteomics and molecular interactions	
19. Deposition is strongly recommended for any datasets that are central and integral to the study; please	NA
consider the journal's data policy. If no structured public repository exists for a given data type, we	
encourage the provision of datasets in the manuscript as a Supplementary Document (see author	
guidelines under 'Expanded View' or in unstructured repositories such as Dryad (see link list at top right)	
or Figshare (see link list at top right).	
20. Access to human clinical and genomic datasets should be provided with as few restrictions as possible	NA .
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).	
21. As far as possible, primary and referenced data should be formally cited in a Data Availability section:	NA
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Wetmore KM, Deutschbauer AM, Price MN, Arkin AP (2012). Comparison of gene expression and mutant	
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AP-MS analysis of human histone deacetylase interactions in CEM-T cells (2013). PRIDE PXD000208	
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When possible, standardized format (SBML, CellML) should be used instead of scripts (e.g. MATLAB).	
Authors are strongly encouraged to follow the MIRIAM guidelines (see link list at top right) and deposit	
their model in a public database such as Biomodels (see link list at top right) or JWS Online (see link list at	
top right). If computer source code is provided with the paper, it should be deposited in a public repository	
or included in supplementary information.	
or medaca in supplementary information.	

# G- Dual use research of concern

23. Could your study fall under dual use research restrictions? Please check biosecurity documents (see	NA
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