

| | Carlos López-Otín |
|--------------------------|-------------------|
| Corresponding author(s): | Adalgisa Caccone |

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 <u>Authors & Referees</u> and the <u>Editorial Policy Checklist</u>.

When statistical analyses are reported, confirm that the following items are present in the relevant location (e.g. figure legend, table legend, main

Statistical parameters

| text | , or N | Methods section). |
|-------------|-------------|---|
| n/a | Cor | nfirmed |
| | \boxtimes | The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement |
| | \boxtimes | An indication of 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. |
| | \boxtimes | A description of all covariates tested |
| | \boxtimes | A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons |
| | | A full description of the statistics including <u>central tendency</u> (e.g. means) or other basic estimates (e.g. regression coefficient) AND <u>variation</u> (e.g. standard deviation) or associated <u>estimates of uncertainty</u> (e.g. confidence intervals) |
| | | For null hypothesis testing, the test statistic (e.g. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted <i>Give P values as exact values whenever suitable.</i> |
| | \boxtimes | For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings |
| \boxtimes | | For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes |
| | \boxtimes | Estimates of effect sizes (e.g. Cohen's d, Pearson's r), indicating how they were calculated |
| | | Clearly defined error bars State explicitly what error bars represent (e.g. SD, SE, CI) |

Our web collection on statistics for biologists may be useful.

Software and code

Data analysis

Policy information about <u>availability of computer code</u>

Data collection No software was used for data collection.

BATI is available at http://degradome.uniovi.es/downloads/vBIO-BATI-0.02.tar.gz Statistical comparisons were performed with GraphPad Prism v7.0 and R3.4.3

Signatures of positive selection were studied with PAML v4 Custom scripts are accessible from https://github.com/vqf/LG

Other standard analysis programs are described in the manuscript and supplementary information

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 upon request. 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

Data supporting the findings of this study are available within the paper and its supplementary information files. Sequencing data have been deposited at the Sequence Read Archive (SRA, https://www.ncbi.nlm.nih.gov/sra) with BioProject accession number PRJNA416050. The accession number of the assembled genomic sequence is PKMU00000000. MAKER2-predicted protein sequences can be downloaded from https://github.com/vqf/LG

| Field-specific reporting | | | | |
|--|--|--|--|--|
| Please select the b | est 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 <u>nature.com/authors/policies/ReportingSummary-flat.pdf</u> | | | |
| Life scier | nces study design | | | |
| All studies must dis | sclose on these points even when the disclosure is negative. | | | |
| Sample size | The most important hypotheses in this work actually refer to a unique individual of C. abingdonii. For comparisons with other species, we rely on inter-species conservation, which is much more stringent than intra-species conservation. For biochemical experiments, no sample size calculation was performed. | | | |
| Data exclusions | No data were excluded. | | | |
| Replication | The experiment shown in Fig. 4 was independently replicated three times, as explained in the Methods section (independent infections per replicate). The experiment shown in Suppl. Fig. S22 was performed with one clone per construct. This experiment involved two different treatments and two time points. | | | |
| Randomization | The main hypotheses in this work refer to species, and therefore randomization is not relevant to this study. For biochemical experiments, groups were established from the same cell line based on infection with different constructs. Therefore, group allocation was pre-established and no randomization was necessary. | | | |
| Blinding | For the primary results in this work, blinding was not possible, as hypotheses were tested in a single genomic sequence. For biochemical experiments, investigators were not blinded to group allocation. | | | |
| Reportin | g for specific materials, systems and methods | | | |
| Materials & experimental systems Methods | | | | |
| n/a Involved in th | | | | |
| Unique biological materials ChIP-seq | | | | |
| Antibodies | Flow cytometry | | | |

MRI-based neuroimaging

Unique biological materials

Animals and other organisms Human research participants

Eukaryotic cell lines

Palaeontology

Policy information about <u>availability of materials</u>

Obtaining unique materials

Samples from all the species of the Giant Galapagos tortoises species complex are protected and require a CITES permit (see below)

Antibodies

Antibodies used

The primary antibodies used were: anti-phospho-Histone H2AX (Ser139) (EMD Millipore, 05-636, clone JBW301, lot 2854120), anti-PARP (Cell Signalling, 9542S, rabbit polyclonal, lot 15), anti-FLAG (Cell Signalling, 2368S, rabbit polyclonal, lot 12), anti-IGF1R (Abcam, ab182408, clone EPR19322, lot GR312678-8), anti-IGF1R (p Tyr1161) (Novus Biologicals, NB100-92555, rabbit polyclonal, lot CJ36131), anti-β-actin (Sigma, A5441, clone AC-15, lot 014M4759) and anti-α-tubulin (Sigma, T6074, clone B-5-1-2, lot 075M4823V).

The secondary antibodies used were:

LI-COR, IRDye 680RD, 926-68071, polyclonal goat-anti-rabbit, lot C41217-03

LI-COR, IRDye 680, 926-32220, polyclonal goat-anti-mouse, lot C00727-03

LI-COR, IRDye 800CW, 926-32211, polyclonal goat-anti-rabbit, lot C60113-05

LI-COR, IRDye 800CW, 926-32210, polyclonal goat-anti-mouse, lot C50316-03

Validation

All antibodies used in this study were purchased from commercial companies, and they had been verified by the manufacturer. As stated in their websites:

-Anti-phospho-Histone H2A.X (Ser139), clone JBW301 is a well published Mouse Monoclonal Antibody validated in ChIP, ICC, IF, WB. This purified mAb is highly specific for phospho-Histone H2A.X (Ser139) also known as H2AXS139p.

-PARP Antibody detects endogenous levels of full length PARP1 (116 kDa), as well as the large fragment (89 kDa) of PARP1 resulting from caspase cleavage. The antibody does not cross-react with related proteins or other PARP isoforms.

-DYKDDDDK Tag Antibody (Anti-Flag) detects exogenously expressed DYKDDDDK proteins in cells. The antibody recognizes the DYKDDDDK peptide (the same epitope recognized by Sigma's Anti-FLAG® antibodies) fused to either the amino- or carboxyterminus of targeted proteins. The binding specificity of this antibody is NOT dependent on the presence of divalent metal cations.

-Our Abpromise guarantee covers the use of ab182408 in the following tested applications: WB (1/1000 dilution). Detects a band of approximately 100,200 kDa (predicted molecular weight: 156 kDa)...

-Anti-IGF1R (p Tyr1161): Validated by Western blot (WB) analysis of p-IGF-1R (Y1161) pAb in extracts from Hela cells.

-Anti-β-actin western blot validation: 1:5,000-1:10,000 using cultured human or chicken fibroblast cell extracts. Reacts against guinea pig, canine, Hirudo medicinalis, feline, pig, carp, mouse, chicken, rabbit, sheep, rat, human and bovine orthologs. Does not react against Dictyostelium discoideum.

-Anti-α-tubulin western blot validation: 0.25-0.5 μg/mL using total cell extract of human foreskin fibroblast cell line (FS11). Species reactivity: human, Chlamydomonas, African green monkey, chicken, kangaroo rat, bovine, mouse, rat, sea urchin.

Eukaryotic cell lines

Authentication

Policy information about cell lines

Cell line source(s) ATCC

Cell lines were not tested for mycoplasma contamination Mycoplasma contamination

Commonly misidentified lines (See ICLAC register)

HEK-293T cells are widely used for infection experiments. The identity of these cells was assessed by PCR-based microsatellite characterization

Animals and other organisms

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

Laboratory animals The study did not involve laboratory animals.

Wild animals The study did not involve observations but did involve temporary captures of wild animals to extract blood samples.

PCR-based microsatellite characterization was performed at the University of Oviedo.

Field-collected samples All work on field samples was conducted at Yale University under IACUC permit number 2016-10825, Galapagos Park Permit

PC-75-16 and CITES number 15US209142/9