nature portfolio

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| Last updated by author(s): | 2024/23/07 |

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 <u>Editorial Policies</u> and the <u>Editorial Policy Checklist</u>.

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| 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 |
| | $oxed{x}$ 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. <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> |
| | 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 |
| | \blacksquare 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

Provide a description of all commercial, open source and custom code used to collect the data in this study, specifying the version used OR state that no software was used.

Data analysis

Provide a description of all commercial, open source and custom code used to analyse the data in this study, specifying the version used OR state that no software was used.

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 <u>data availability statement</u>. 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

Provide your data availability statement here.

Research involving human participants, their data, or biological material

Policy information about studies with <u>human participants or human data</u>. See also policy information about <u>sex, gender (identity/presentation)</u>, and sexual orientation and race, ethnicity and racism.

Reporting on sex and gender

Use the terms sex (biological attribute) and gender (shaped by social and cultural circumstances) carefully in order to avoid confusing both terms. Indicate if findings apply to only one sex or gender; describe whether sex and gender were considered in study design; whether sex and/or gender was determined based on self-reporting or assigned and methods used.

Provide in the source data disaggregated sex and gender data, where this information has been collected, and if consent has been obtained for sharing of individual-level data; provide overall numbers in this Reporting Summary. Please state if this information has not been collected.

Report sex- and gender-based analyses where performed, justify reasons for lack of sex- and gender-based analysis.

Reporting on race, ethnicity, or other socially relevant groupings

Please specify the socially constructed or socially relevant categorization variable(s) used in your manuscript and explain why they were used. Please note that such variables should not be used as proxies for other socially constructed/relevant variables (for example, race or ethnicity should not be used as a proxy for socioeconomic status).

Provide clear definitions of the relevant terms used, how they were provided (by the participants/respondents, the researchers, or third parties), and the method(s) used to classify people into the different categories (e.g. self-report, census or administrative data, social media data, etc.)

Please provide details about how you controlled for confounding variables in your analyses.

Population characteristics

Describe the covariate-relevant population characteristics of the human research participants (e.g. age, genotypic information, past and current diagnosis and treatment categories). If you filled out the behavioural & social sciences study design questions and have nothing to add here, write "See above."

Recruitment

Describe how participants were recruited. Outline any potential self-selection bias or other biases that may be present and how these are likely to impact results.

Ethics oversight

Identify the organization(s) that approved the study protocol.

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 | \prime that is the best fit for your research. I | f you are not sure, | read the appropriate sections before making your selection. |
|-----------------------------|--|---------------------|---|
| Life sciences | Behavioural & social sciences | Ecological, eve | olutionary & environmental sciences |

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Ecological, evolutionary & environmental sciences study design

All studies must disclose on these points even when the disclosure is negative.

Study description

Briefly describe the study. For quantitative data include treatment factors and interactions, design structure (e.g. factorial, nested, hierarchical), nature and number of experimental units and replicates.

Research sample

We generated a chronometric dataset of 105 new determinations (74 radiocarbon and 31 luminescence ages) from four key southern Italian sites.

Sampling strategy

Sites were visited for direct sampling (OSL dating) and bones from collections were searched for suitable samples for dating.

Data collection

Multiple fieldtrips spanning 2017-2022.

Timing and spatial scale

This is not relevant.

Data exclusions

If no data were excluded from the analyses, state so OR if data were excluded, describe the exclusions and the rationale behind them, indicating whether exclusion criteria were pre-established.

Reproducibility

1 sample in every 20 dated was analyzed twice to measure internal reproducibility. Statistical tests of agreement were run on all duplicate samples. Background samples were monitored over several years and quantified, to be later subtracted for the 14C measurements.

Randomization

Describe how samples/organisms/participants were allocated into groups. If allocation was not random, describe how covariates were controlled. If this is not relevant to your study, explain why.

Blinding

Describe the extent of blinding used during data acquisition and analysis. If blinding was not possible, describe why OR explain why blinding was not relevant to your study.

| Did the study involve field work? x Yes No | | | |
|--|--|--|--|
| Field work, collection and transport | | | |
| Field conditions | Summer conditions, very hot! | | |
| Location | Castelcivita, Cala, and Oscurusciuto caves, southern Italy. The University of Siena. | | |
| Access & import/export | Describe the efforts you have made to access habitats and to collect and import/export your samples in a responsible manner and in compliance with local, national and international laws, noting any permits that were obtained (give the name of the issuing authority, the date of issue, and any identifying information). | | |
| Disturbance | Describe any disturbance caused by the study and how it was minimized. | | |
| We require information from a | r specific materials, systems and methods authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, avant 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 & experime | | | |
| n/a Involved in the study Antibodies | n/a Involved in the study ChIP-seq | | |
| | Eukaryotic cell lines Flow cytometry | | |
| Palaeontology and archaeology MRI-based neuroimaging | | | |
| Animals and other organisms | | | |
| Clinical data Dual use research o | francorn | | |
| Plants | Concern | | |
| | | | |
| Antibodies | | | |
| Antibodies used | Describe all antibodies used in the study; as applicable, provide supplier name, catalog number, clone name, and lot number. | | |
| Validation | Validation Describe the validation of each primary antibody for the species and application, noting any validation statements on the manufacturer's website, relevant citations, antibody profiles in online databases, or data provided in the manuscript. | | |
| Eukaryotic cell lin | es es | | |
| Policy information about <u>ce</u> | ell lines and Sex and Gender in Research | | |
| Cell line source(s) | Cell line source(s) State the source of each cell line used and the sex of all primary cell lines and cells derived from human participants or vertebrate models. | | |
| Authentication | Authentication Describe the authentication procedures for each cell line used OR declare that none of the cell lines used were authenticated. | | |
| Mycoplasma contaminati | Mycoplasma contamination Confirm that all cell lines tested negative for mycoplasma contamination OR describe the results of the testing for mycoplasma contamination OR declare that the cell lines were not tested for mycoplasma contamination. | | |
| Commonly misidentified (See ICLAC register) | ommonly misidentified lines (Name any commonly misidentified cell lines used in the study and provide a rationale for their use. See ICLAC register) | | |

Palaeontology and Archaeology

Specimen provenance

Permission and support for fieldwork in Apulia and Campania was obtained from the Soprintendenza Archeologia, Belle Arti e Paesaggio per le Province di Salerno e Avellino, Soprintendenza Archeologia, Belle Arti e Paesaggio per le Province di Brindisi e Lecce and Soprintendenza Nazionale per il Patrimonio Culturale Subacqueo. Excavations were conducted under permission of MiC (MIC | MIC_DG-ABAP_SERV II_UO1|07/06/2021|0019224-P| [34.61.07/1.15.1/2019]; MIC | MIC_DG-ABAP_SERVII_|30/09/2021|0032649-P| [34.61.07/1.14.1/2019] and DG-ABAP 20/06/2022 decreto 809). This included permission for all analysis and post-excavation work.

| Specimen deposition | Material from the excavations is held at the University of Siena and with the Soprintendenza Archeologia, Belle Arti e Paesaggio per le Province di Salerno e Avellino, Soprintendenza Archeologia, Belle Arti e Paesaggio per le Province di Brindisi e Lecce and Soprintendenza Nazionale per il Patrimonio Culturale Subacqueo. |
|---|---|
| Dating methods | Radiocarbon dating, OSL dating. |
| x Tick this box to confir | rm that the raw and calibrated dates are available in the paper or in Supplementary Information. |
| Ethics oversight | See specimen provenance. |
| lote that full information on t | the approval of the study protocol must also be provided in the manuscript. |
| | |
| | er research organisms |
| olicy information about <u>st</u> <u>esearch</u> | tudies involving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in |
| Laboratory animals | For laboratory animals, report species, strain and age OR state that the study did not involve laboratory animals. |
| Wild animals | Provide details on animals observed in or captured in the field; report species and age where possible. Describe how animals were caught and transported and what happened to captive animals after the study (if killed, explain why and describe method; if released, say where and when) OR state that the study did not involve wild animals. |
| Reporting on sex | Indicate if findings apply to only one sex; describe whether sex was considered in study design, methods used for assigning sex. Provide data disaggregated for sex where this information has been collected in the source data as appropriate; provide overall numbers in this Reporting Summary. Please state if this information has not been collected. Report sex-based analyses where performed, justify reasons for lack of sex-based analysis. |
| Field-collected samples | For laboratory work with field-collected samples, describe all relevant parameters such as housing, maintenance, temperature, photoperiod and end-of-experiment protocol OR state that the study did not involve samples collected from the field. |
| Ethics oversight | Identify the organization(s) that approved or provided guidance on the study protocol, OR state that no ethical approval or guidance was required and explain why not. |
| lote that full information on t | the approval of the study protocol must also be provided in the manuscript. |
| | |
| Clinical data | |
| olicy information about <u>cl</u> | inical studies with the ICMJEguidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions. |
| Clinical trial registration | Provide the trial registration number from ClinicalTrials.gov or an equivalent agency. |
| Study protocol | Note where the full trial protocol can be accessed OR if not available, explain why. |
| Data collection | Describe the settings and locales of data collection, noting the time periods of recruitment and data collection. |
| Outcomes | Describe how you pre-defined primary and secondary outcome measures and how you assessed these measures. |
| | |
| | |
| Dual use research | |
| olicy information about <u>d</u> | ual use research of concern |
| lazards | |
| Could the accidental, del in the manuscript, pose a | iberate or reckless misuse of agents or technologies generated in the work, or the application of information presented a threat to: |
| No Yes | |
| Public health | |
| National security | |

Crops and/or livestock

Any other significant area

Ecosystems

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| Doe | Does the work involve any of these experiments of concern: | | |
|-----|---|--|--|
| No | Yes | | |
| | Demonstrate how to render a vaccine ineffective | | |
| | Confer resistance to therapeutically useful antibiotics or antiviral agents | | |
| | Enhance the virulence of a pathogen or render a nonpathogen virulent | | |
| | Increase transmissibility of a pathogen | | |
| | Alter the host range of a pathogen | | |
| | Enable evasion of diagnostic/detection modalities | | |
| | Enable the weaponization of a biological agent or toxin | | |
| | Any other potentially harmful combination of experiments and agents | | |

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.

ChIP-sea

Data deposition

| Confirm that both raw and final processed data have been deposited in a public database such as <u>GEO</u> . | | |
|--|---|--|
| Confirm that you have deposited or provided access to graph files (e.g. BED files) for the called peaks. | | |
| Data access links May remain private before publication. | For "Initial submission" or "Revised version" documents, provide reviewer access links. For your "Final submission" document, provide a link to the deposited data. | |
| Files in database submission | Provide a list of all files available in the database submission. | |
| Genome browser session (e.g. <u>UCSC</u>) | Provide a link to an anonymized genome browser session for "Initial submission" and "Revised version" documents only, to enable peer review. Write "no longer applicable" for "Final submission" documents. | |

Methodology

Replicates

Describe the experimental replicates, specifying number, type and replicate agreement.

Describe the sequencing depth for each experiment, providing the total number of reads, uniquely mapped reads, length of reads and whether they were paired- or single-end.

Describe the antibodies used for the ChIP-seq experiments; as applicable, provide supplier name, catalog number, clone name, and lot number.

Peak calling parameters

Specify the command line program and parameters used for read mapping and peak calling, including the ChIP, control and index files used.

Data quality

Describe the methods used to ensure data quality in full detail, including how many peaks are at FDR 5% and above 5-fold enrichment.

Software

Describe the software used to collect and analyze the ChIP-seq data. For custom code that has been deposited into a community repository, provide accession details.

Flow Cytometry

| PΙ | ots |
|----|-----|
|----|-----|

| Plots | |
|-----------------------------------|---|
| Confirm that: | |
| The axis labels state the marke | er and fluorochrome used (e.g. CD4-FITC). |
| The axis scales are clearly visib | le. 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 | Describe the sample preparation, detailing the biological source of the cells and any tissue processing steps used. |
| Instrument | dentify the instrument used for data collection, specifying make and model number. |
| | Describe the software used to collect and analyze the flow cytometry data. For custom code that has been deposited into a community repository, provide accession details. |
| | Describe the abundance of the relevant cell populations within post-sort fractions, providing details on the purity of the samples and how it was determined. |
| | Describe the gating strategy used for all relevant experiments, specifying the preliminary FSC/SSC gates of the starting cell population, indicating where boundaries between "positive" and "negative" staining cell populations are defined. |
| Tick this box to confirm that a | figure exemplifying the gating strategy is provided in the Supplementary Information. |
| | |
| Magnetic resonance im | laging |
| experimental design | |
| Design type | Indicate task or resting state; event-related or block design. |
| Design specifications | Specify the number of blocks, trials or experimental units per session and/or subject, and specify the length of each trial or block (if trials are blocked) and interval between trials. |
| Behavioral performance measures | State number and/or type of variables recorded (e.g. correct button press, response time) and what statistics were used to establish that the subjects were performing the task as expected (e.g. mean, range, and/or standard deviation across |

Acquisition

| Imaging type(s) | Specify: functional, structural, diffusion, perfusion. |
|-------------------------------|--|
| Field strength | Specify in Tesla |
| Sequence & imaging parameters | Specify the pulse sequence type (gradient echo, spin echo, etc.), imaging type (EPI, spiral, etc.), field of view, matrix size, slice thickness, orientation and TE/TR/flip angle. |
| Area of acquisition | State whether a whole brain scan was used OR define the area of acquisition, describing how the region was determined. |
| Diffusion MRI Used | Not used |

subjects).

Noise and artifact removal

| Preprocessing | |
|------------------------|---|
| Preprocessing software | Provide detail on software version and revision number and on specific parameters (model/functions, brain extraction, segmentation, smoothing kernel size, etc.). |
| Normalization | If data were normalized/standardized, describe the approach(es): specify linear or non-linear and define image types used for transformation OR indicate that data were not normalized and explain rationale for lack of normalization. |

Describe the template used for normalization/transformation, specifying subject space or group standardized space (e.g. Normalization template original Talairach, MNI305, ICBM152) OR indicate that the data were not normalized.

> Describe your procedure(s) for artifact and structured noise removal, specifying motion parameters, tissue signals and physiological signals (heart rate, respiration).

| Volume censoring Dep | fine your software and/or method and criteria for volume censoring, and state the extent of such censoring. |
|-------------------------------------|---|
| Statistical modeling & inference | e 2 |
| 7,1 | ecify type (mass univariate, multivariate, RSA, predictive, etc.) and describe essential details of the model at the first and cond levels (e.g. fixed, random or mixed effects; drift or auto-correlation). |
| | fine precise effect in terms of the task or stimulus conditions instead of psychological concepts and indicate whether ANOVA factorial designs were used. |
| Specify type of analysis: Whole | e brain ROI-based Both |
| Statistic type for inference Spe | ecify voxel-wise or cluster-wise and report all relevant parameters for cluster-wise methods. |
| (See Eklund et al. 2016) | |
| Correction | scribe the type of correction and how it is obtained for multiple comparisons (e.g. FWE, FDR, permutation or Monte Carlo). |
| Models & analysis | |
| n/a Involved in the study | |
| Functional and/or effective connect | Report the measures of dependence used and the model details (e.g. Pearson correlation, partial correlation, mutual information). |
| Graph analysis | Report the dependent variable and connectivity measure, specifying weighted graph or binarized graph, subject- or group-level, and the global and/or node summaries used (e.g. clustering coefficient, efficiency, etc.). |

Specify independent variables, features extraction and dimension reduction, model, training and evaluation

Multivariate modeling and predictive analysis

metrics.