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Corresponding author(s): Philip Santangelo

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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	Volocity (Microscopy acquisiton; PerkinElmer); StepOnePlus (ABI); BD FACSDiva (Flow acquisition); Fluobeam aquisition software (Fluoptics).
Data analysis	Volocity (Microscopy analysis; PerkinElmer); StepOnePlus (ABI); FlowJo (Flow analysis); MIM (PET/CT analysis); Amira (PET/CT 3D reconstructions); Graphpad Prism 7 (plotting and statistical analyses).

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

The authors declare that all data supporting the findings of this study are available within the paper and its Supplementary Information. PET/CT-scanner acquisition settings are available upon request from the corresponding author.

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	This study did not require pre-specified effect sizes and therefore no sample-size calculations were needed. All in vitro experiments (immunofluorescence in cells and tissues, qPCR) were performed at least twice in triplicates. PET-CT studies were performed in four animals. Probe validation and flow cytometry experiments were performed using samples from one animal.
Data exclusions	No data were excluded.
Replication	Probe validation and flow-cytometry experiments were not repeated. All other experiments were replicated.
Randomization	This study did not require randomization. All animals received the same treatment.
Blinding	This study did not require blinding. All animals received the same treatment.

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	All information about the antibodies used in the studies for flow cytometry and immunofluorescence are listed in Supplementary tables 2–4, including clone, catalogue number and vendor.
Validation	Antibodies for immunofluorescence were validated using mock controls or no primary controls. All antibodies for flow cytometry were validated by the manufacturer.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	HeLA ATCC CCL-2.
Authentication	ATCC cell-line authentication service.
Mycoplasma contamination	Cells were routinely inspected for contamination.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used.

Animals and other organisms

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

Laboratory animals	Five Cynomolgus monkeys were used: AD036 8.4 kg, AD031 6.6kg, AF032 7.65 kg, AF093 6.35 kg (all age 9 and males), and CM653 9.6kg, male, age 12.
Wild animals	The study did not involve wild animals.
Field-collected samples	The study did not involve samples collected from the field.
Ethics oversight	Macaques were maintained in accordance with the regulations of the Guide for the Care and Use of Laboratory Animal at New Iberia Research Center, University of Louisiana.

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	One animal was euthanized at 16 hours post vaccine administration. During necropsy, muscle tissues and lymph nodes were screened for near-IR signal using the Fluobeam near-IR portable camera (Fluoptics Imaging Inc, Cambridge MA), and the radioactive signal was measured using a well counter. Contralateral tissues were also collected as negative controls. Excised LNs were mechanically dissociated by pressing and grinding the tissue on top of a 70 µm filter and a 40 µm filter (BD Biosciences), fitted on top of a 50-mL conical tube. During mechanical disruption, the tissue was systematically washed with RPMI1640 media with 10% Pen/Step, under sterile conditions. Muscle tissues were cut into small pieces and digested with 0.2% Type IV collagenase (Worthington) and 0.01% RQ1 DNase (Promega) for 90 minutes at 37ºC, with gentle shaking. Digested muscles were passed through a 15G blunt followed by an 18G blunt needle several times. Tissues were subsequently filtered through a 70-µm and a 40-µm strainer. The cells were centrifuged at 1000xg for 10 minutes at 4°C; the resultant cell pellet was treated with ACK solution for 5 minutes for red-blood-cells lysis and washed with RPMI. Cells were resuspended in 10 ml of complete RPMI, counted with a hemocytometer, and immediately processed for flow cytometry. One million cells were chosen and stained with the cell type markers indicated in Supplementary Tables 2 and 3. Surface cell staining was performed in FACS Wash buffer (2% FBS in 1x PBS). Cells were fixed with 1% PFA in FACS Wash at 4°C until data acquirement. Cells were gated as indicated in Supplemenary Figs. 2–5. All samples were acquired using a FACSAria Fusion (BD Biosciences). All analyses were performed using the FlowJo software package (Tree Star, Ashland, OR, USA). A total of at least 300,000 events were recorded for each organ.
Instrument	FACSAria Fusion (BD Biosciences)
Software	Acquisition - BD FACSDiva; Analysis - FlowJo software package (Tree Star, Ashland, OR, USA).
Cell population abundance	A total of at least 300,000 events were recorded for each experimental condition.
Gating strategy	The gating strategy for the main cell types is illustrated and explained in Supplementary figures 3, 4 and 5.

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