

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 [Editorial Policies](#) 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
<i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i> |
| ☐ | ☒ | 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
<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 |
| ☒ | ☐ | 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 No software was used

Data analysis RNA sequencing data using CLC Genomics Workbench 24, HEC software for cytometry, GraphPad Prism software (v 9, GraphPad Software LLC, San Diego, CA, USA)

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 [data availability statement](#). 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](#)

The authors state that all the data is available, including the data sources.
<https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE252194>

Research involving human participants, their data, or biological material

Policy information about studies with [human participants or human data](#). See also policy information about [sex, gender \(identity/presentation\), and sexual orientation](#) and [race, ethnicity and racism](#).

Reporting on sex and gender

Reporting on race, ethnicity, or other socially relevant groupings

Population characteristics

Recruitment

Ethics oversight

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 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

Data exclusions

Replication

Randomization

Blinding

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 and archaeology
☐	☒ Animals and other organisms
☒	☐ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☐	☒ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used

Anti-mouse secondary antibodies (rabbit anti-mouse IgG H&L, Abcam, Paris, France),
CD45 PerCP-Cy5.5 Biolegend 103132
CD19 APCeF780 eBioscience 47-0193-80
TCRb AF700 Biolegend 109224 CD4 PB Biolegend 100534
CDS BV605 BD Horizon 563152
FOXP3 APC Invitrogen 17-5773-82.

All primary and secondary antibodies (with supplier name, catalogue number, clone name and batch number where applicable) are listed here in the report summary and referenced in the manuscript/supplementary table. In addition, all antibodies listed here are also provided in the manuscript (with dilution).

Validation

The validation of the antibodies was based on the manufacturer's validation on their site.

<https://www.abcam.com/products/secondary-antibodies/rabbit-mouse-igg-hl-ab6709.htm>.

Rabbit Anti-Mouse IgG H&L See all IgG secondary antibodies, Host species : Rabbit ; Target species : Mouse. Tested applications : Suitable for: WB, IP, Immunodiffusion, Conjugation, ICC/IF, ELISA, IHC-P, IHC-Fr more details. Immunogen : Mouse IgG, whole molecule

<https://www.biolegend.com/fr-ch/soluble-mhc/percp-cy5-5-anti-mouse-cd45-2-antibody-4271?GroupID=BLG1934> <https://www.thermofisher.com/antibody/product/CD19-Antibody-clone-eBio1D3-1D3-Monoclonal/47-0193-82>

Isotype Control : PerCP/Cyanine5.5 Rat IgG2b, κ Isotype Ctrl

Verified Reactivity : Mouse

Antibody Type : Monoclonal

Host Species : Rat

Immunogen : Mouse thymus or spleen

Formulation : Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide.

Preparation : The antibody was purified by affinity chromatography, and conjugated with PerCP/Cyanine5.5 under optimal conditions.

Concentration : 0.2 mg/ml

Storage & Handling : The antibody solution should be stored undiluted between 2°C and 8°C, and protected from prolonged exposure to light. Do not freeze.

Recommended Usage : Each lot of this antibody is quality control tested by immunofluorescent staining with flow cytometric analysis. For flow cytometric staining, the suggested use of this reagent is = 0.25 µg per 10⁶ cells in 100 µl volume. It is recommended that the reagent be titrated for optimal performance for each application.

<https://www.biolegend.com/en-gb/products/alexa-fluor-700-anti-mouse-tcr-beta-chain-antibody-4537?Clone=H57-597> <https://www.biolegend.com/de-at/soluble-mhc/pacific-blue-anti-mouse-cd4-antibody-2855?GroupID=BLG4211>

Alexa Fluor® 700 Armenian Hamster IgG Isotype Ctrl

Verified Reactivity : Mouse

Antibody Type : Monoclonal

Host Species : Armenian Hamster

Immunogen : Affinity purified TCR from mouse DO-11.10 cells

Formulation : Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide.

Preparation : The antibody was purified by affinity chromatography and conjugated with Alexa Fluor® 700 under optimal conditions.

Concentration : 0.5 mg/ml

Storage & Handling : The antibody solution should be stored undiluted between 2°C and 8°C, and protected from prolonged exposure to light. Do not freeze.

Recommended Usage : Each lot of this antibody is quality control tested by immunofluorescent staining with flow cytometric analysis. The suggested use of this reagent is ≤1.0 µg per million cells in 100 µl volume. It is highly recommended that the reagent be titrated for optimal performance for each application.

<https://www.bdbiosciences.com/en-ie/products/reagents/flow-cytometry-reagents/research-reagents/single-color-antibodies-ruo/bv605-rat-anti-mouse-cd8a.563152>

Alternative Name: Cd8a; CD8 alpha chain; Ly-2; Lyt2; Lyt-2; Ly-35; Ly-B

Reactivity: Mouse (QC Testing)

Isotype: Rat LOU, also known as Louvain, LOU/C, LOU/M IgG2a, κ

Immunogen: Mouse Spleen Cells or Thymocyte Membranes

Application: Flow cytometry (Routinely Tested)

Concentration: 0.2 mg/ml

RRID: AB_2738030

Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

Regulatory Status:RUO

<https://www.thermofisher.com/antibody/product/FOXP3-Antibody-clone-FJK-16s-Monoclonal-1/17-5773-82>

Species Reactivity: Bovine, Dog, Cat, Mouse, Pig, Rat

Published species: Dog, Horse, Human, Mouse, Pig, Rat

Host/Isotype: Rat / IgG2a, kappa

Recommended Isotype Control: Rat IgG2a kappa Isotype Control (eBR2a), APC, eBioscience™

Class: Monoclonal

Type: Antibody

Clone: FJK-16s

Conjugate: APCView additional formats

Excitation/Emission Max: 651/660 nmView spectraspectra

Form:Liquid; Concentration: 0.2 mg/mL

Purification: Affinity chromatography

Applications

Tested Dilution

Publications

Immunohistochemistry (IHC), Immunohistochemistry (PFA fixed) (IHC (PFA)), Immunohistochemistry (Frozen) (IHC (F)),

Immunocytochemistry (ICC/IF), Flow Cytometry (Flow), ChIP assay (ChIP), T-Cell Activation (TCA)

Eukaryotic cell lines

Policy information about [cell lines and Sex and Gender in Research](#)

Cell line source(s)

Hela epithelia I cells ATCC
<https://www.atcc.org/products/ccl-2>

Authentication

<https://www.atcc.org/products/ccl-2>. No authentication

Mycoplasma contamination

PCR detection test: This test involves the specific PCR amplification of mycoplasma DNA. The kit developed by Sigma uses pairs of primers directed against the 16S RNA sequence, which is highly conserved, enabling most commonly encountered mycoplasmas to be detected. It detects 19 different mycoplasma species. We confirm that the cell line tested negative for mycoplasma contamination

Commonly misidentified lines
(See [ICLAC](#) register)

No

Animals and other research organisms

Policy information about [studies involving animals](#); [ARRIVE guidelines](#) recommended for reporting animal research, and [Sex and Gender in Research](#)

Laboratory animals

BALB/C female mice (Supplier January Lab) 6 months old. We have described the housing conditions for the mice in the manuscript.

Wild animals

No wild animal is used

Reporting on sex

We used only females in order to look at the impact of maternal vaccination on the protection of baby mice against neonatal E. coli meningitis. Only female animal were used for the adult animal as the target of the vaccine is women with a pregnancy project. Both sex were used for baby mice

Field-collected samples

No samples were collected in the field.

Ethics oversight

The mouse experiments were conducted in accordance with local, national and European Union guidelines and had been approved by the Animal Care and Use Committee at Université Paris Cité (Paris, France; reference: 2020011519022360; approval number A75-14-08).

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Plants

Seed stocks We didn't use any plants

Novel plant genotypes We didn't use any plants

Authentication We didn't use any plants

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

Seven days after full immunization, BALB/c mice were sacrificed, and the spleens were collected for the flow cytometry analysis of lymphocytes. Red blood cells were removed from spleen cell suspensions by lysis with ammonium chloride buffer. The remaining cells were then incubated with LIVE/DEAD™ Fixable Aqua Dead Cell Stain Kit (Invitrogen L34957, Thermo Scientific Inc., Branchburg, NJ, USA). Collect spleen in HBSS+10% SVF on ice. Crush the spleen on a 40µm cell sieve. Wash 1 time with 5ml of cold HBSS + 10% SVF (300g centri 5' 4°C). Wash 1 time with 10ml HBSS (RT) (300g centri 5' 4°C). Discard the supernatant completely (empty the supernatant into a waste bin and remove the last microlitres of supernatant above the pellet with a pipette). Rub the tube on a rack to dislodge the pellet. Resuspend in 2ml of red cell lysis buffer (ACK buffer).

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Instrument A Fortessa flow cytometer (BD Biosciences, San Jose, CA, USA) was used to analyze 100000 stained cells. Doublets were excluded from the analysis by using appropriate forward scatter/side scatter gates.

Software Data were analyzed with Flowjo software

Cell population abundance

Samples	Total number of events	%living cells
C1	1.22 X 1000000	91,8
C2	1.09x 1000000	90,3
C3	1.29x 1000000	90,4
C4	1.21x 1000000	92,2
1	1.07x 1000000	89,7
2	1.22x 1000000	89,9
3	0.93x 1000000	89,4
4	1.22x 1000000	91,6

Collect spleen in HBSS+10% SVF on ice. Crush the spleen on a 40µm cell sieve. Wash 1 time with 5ml of cold HBSS + 10% SVF (300g centri 5' 4°C). Wash 1 time with 10ml HBSS (RT) (300g centri 5' 4°C). Discard the supernatant completely (empty the supernatant into a waste bin and remove the last microlitres of supernatant above the pellet with a pipette). Rub the tube on a rack to dislodge the pellet. Resuspend in 2ml of red cell lysis buffer (ACK buffer). Gently resuspend with a pipette (do not vortex) for 300ml: NH₄Cl 2.48g + EDTA 0.5M 60ul + KHCO₃ 0.3g + HCl IN pH 7.2-7.4 + H₂O MQ QSP, filtered, stored at 4°C. Start the timer as soon as the ACK is added and incubate for 5' in the dark at RT. Wash 2 times with 4ml HBSS-SVF10% (300g 5' 4°C), resuspend in 4ml HBSS-SVF10%. Pass the cells through a cell sieve (use a FACS tube cell sieve as it has a tighter mesh). Count the cells: pre-dilute to 1/20 (Sul cells, 95 µl HBSS+10%SVF) then dilute to 1/2 in trypan blue. Add 2.5x10⁶ cells per FACS tube (Make an extra tube that will not be labelled).

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Gating strategy

First of all, from all the events that pass, we select the cells. Then we select the single cells. Then I will select the living cells. After selecting the living cells I take the hematopoietic cells (so CD 45+). Then I look at the expression of CD19 and TCRb to separate B lymphocytes from T lymphocytes. In the T lymphocytes I look at the expression of CD8 and CD4 and in the CD4 I look at FOXP3 for regulatory T cells.

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