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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	Droplet data acquisition and sorting: - LABVIEW custom software coded by Andrew Griffiths laboratory - available on request for academic collaborations OR - HiFiBio proprietary software: The software uDROP used for droplet analyses and sorting was designed by HiFiBio, or was a custom-written Matlab script. The HiFiBiO B cell screening process is called CelliGO, which is not a commercialized product – and some aspects of it are proprietary. People can use the provided microfluidics design and purchase components separately as described in the manuscript (microscopes, compressors, pumps). Flow cytometry: BD Cell quest
Data analysis	HiFiBio proprietary software: The repertoire analyses were performed using the HiFiBio proprietary software "Absolution". The "Comat" tool is freely available under the terms of the GNU General Public License as published by the Free Software Foundation. Flow cytometry analysis: FlowJo Version 9 Graphical representation: GraphPad Prism Ver 7.0.

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

All data is provided in the manuscript supplemental information and files. All the other data that support the findings of this study are available from the corresponding authors upon request.

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	The sample size in all experiments ensure a high statistical power to support the observed effect (>95 %) based on null hypothesis testing.
Data exclusions	No data was excluded.
Replication	All experiments were replicated across the two or three replicates. The findings are consistent across the replicates.
Randomization	No method of randomization was chosen.
Blinding	Data collection and analysis were not performed in a blinded manner.

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

anti-IgM (Miltenyi, Cat#130-095-904)

anti-IgD (Miltenyi, Cat#130-101-899)

biotinylated VHH anti-mouse kappa light chain (captureselect, Thermo Fisher #7103152100)

rabbit F(ab')2 anti-mouse IgG Fc-specific (AlexaFluor647-labeled, Jackson ImmunoResearch)

anti-Tetanus Toxoid antibody clone TT7 (DeKosky, B. J., et al. (2013). "High-throughput sequencing of the paired human immunoglobulin heavy and light chain repertoire." Nat Biotechnol 31(2): 166-169.)

mouse anti-horseradish peroxidase antibody (Jackson Immuno Research, Cat#223-005-024)

anti-hTSPAN8 51D3 IgG (Pfizer Inc)

anti-hTSPAN8 43A6 IgG (Pfizer Inc)

biotinylated anti-IgG mouse mAb (Mabtech #3825-2AW-Plus)

Anti-human Fc-HRP (Bethyl)

biotin-conjugated VHH anti-IgG-Fc multi species antibodies (ThermoFisher)

goat anti-human IgG-PE (Southern Biotech #1030-09)

Validation

anti-IgM (Miltenyi, Cat#130-095-904): validated by the manufacturer

anti-IgD (Miltenyi, Cat#130-101-899): validated by the manufacturer

biotinylated VHH anti-mouse kappa light chain (captureselect, Thermo Fisher #7103152100): validated by the manufacturer and in this reference: Eyer, K., et al. (2017). "Single-cell deep phenotyping of IgG-secreting cells for high-resolution immune monitoring." Nat Biotechnol 35(10): 977-982.

rabbit F(ab')2 anti-mouse IgG Fc-specific (AlexaFluor647-labeled, Jackson ImmunoResearch): validated by the manufacturer and in this reference: Eyer, K., et al. (2017). "Single-cell deep phenotyping of IgG-secreting cells for high-resolution immune monitoring." Nat Biotechnol 35(10): 977-982.

anti-Tetanus Toxoid antibody clone TT7: validated in this reference: DeKosky, B. J., et al. (2013). "High-throughput sequencing of the paired human immunoglobulin heavy and light chain repertoire." Nat Biotechnol 31(2): 166-169.

mouse anti-horseradish peroxidase antibody (Jackson Immuno Research, Cat#223-005-024): validated by the manufacturer

anti-hTSPAN8 51D3 IgG (Pfizer Inc): validated by Pfizer Inc, and used as a positive control in this manuscript

anti-hTSPAN8 43A6 IgG (Pfizer Inc): validated by Pfizer Inc, and used as a positive control in this manuscript

biotinylated anti-IgG mouse mAb (Mabtech #3825-2AW-Plus): validated by the manufacturer

Anti-human Fc-HRP (Bethyl): validated by the manufacturer

biotin-conjugated VHH anti-IgG-Fc multi species antibodies (ThermoFisher): validated by the manufacturer

goat anti-human IgG-PE (Southern Biotech #1030-09): validated by the manufacturer

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)

- m300.19 mouse pre-B cells: LONZA (https://knowledge.lonza.com/cell?id=138)

- Chinese hamster ovary (CHO) cells: ATCC (https://www.lgcstandards-atcc.org/products/all/CCL-61.aspx?geo_country=fr)

- Expi293F suspension cells: Life Technologies (https://www.thermofisher.com/order/catalog/product/A14527?SID=srch-srp-A14527#/A14527?SID=srch-srp-A14527)

- Jurkat human T cell line: ATCC (https://www.lgcstandards-atcc.org/products/all/TIB-152.aspx)

- Jurkat reporter cells stably expressing GFP under control of the NFkB responsive promoter: SYSTEMBIO (https://systembio.com/products/imaging-and-reporter-vectors/signaling-pathway-reporters/nf-kb-jurkat-gfp-transcriptional-reporter-cell-line/)

- Ramos human B cell line: ATCC (https://www.lgcstandards-atcc.org/Products/All/CRL-1596.aspx?geo_country=fr)

Authentication

Cell lines were not independently authenticated.

Mycoplasma contamination

All cell lines tested negative for mycoplasma 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

BALB/c mice purchased from Janvier Labs.

Wild animals

N/A

Field-collected samples	N/A
Ethics oversight	Experiments using mice were validated by i) the CETEA ethics committee number 89 (Institut Pasteur, Paris, France) under #2013-0103, and by the French Ministry of Research under agreement #00513.02, and ii) the French ethics committee number 80 (CellVax) under reference APAFIS#6063-2016071213009170 v5.

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	Parental M300.19 cells and TSPAN8+ M300.19 cells were incubated with 10 µg/mL recombinant IgG identified from TSPAN8 sorts or with 5 µg/mL anti-TSPAN8 mAb clone 43A6 (positive control) in ice-cold FACS Buffer (Ca2+, Mg2+-free PBS supplemented with 3% FBS + 0.1% w/v NaN3) for 30 min on ice. Cells were washed twice, incubated with goat anti-human IgG-PE (Southern Biotech #1030-09) for 30 min on ice, and resuspended in BD Cytofix (BD Biosciences #554655) for 30 min on ice, and analyzed on a FACSCalibur flow cytometer (BD)
Instrument	FACSCalibur flow cytometer (Becton Dickinson)
Software	For data acquisition BD CellQuest; For data analysis FlowJo version 9
Cell population abundance	100% (cell lines)
Gating strategy	N/A

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