nature portfolio

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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 *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	No software was used
Data analysis	FlowJo V10 Graphpad Prism version 9.4.1 Microsoft Excel version 16.87

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

All data supporting the findings are available in this paper and the Supplementary Information. Source data are provided with this paper.

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	not relevant
Reporting on race, ethnicity, or other socially relevant groupings	not relevant
Population characteristics	The University Medical Center Utrecht (UMCU) has a donor system in place with the specific purpose to obtain blood from healthy donors for research purposes. This system is called the 'Mini Donor Dienst' is a UMCU-wide initiative and officially approved by the local ethical committee (METC) . The collection of the samples is according to European/Dutch/ procedures that are documented within the local setting of our hospital (UMC Utrecht). No data from the volunteers will be collected.
Recruitment	Human blood from healthy volunteers was collected with informed consent in accordance with the Declaration of Helsinki. Donors enroll on a voluntary basis. Blood requested by the researcher through this service ensures pseudonymity of the donor. The amount of blood per donation will not exceed national and local regulations in The Netherlands/at the UMCU neither the amount specified in the informed consent leaflets.
Ethics oversight	Medical Ethics Committee of the University Medical Center Utrecht was obtained (METC protocol 07-125/C approved on March 1, 2010).

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	No sample size calculation was performed. We used data from at least 3 independent experiments. Based on previously published and collected data, we know that three repeats is sufficient for statistical interpretation. Furthermore, the P values resulting from statistical analyses suggest that the sample size in our study was sufficient.
Data exclusions	no data were excluded from the analyses
Replication	Cellular experiments were performed 3 times as independent experiments. For some screening experiments less repeats were performed, which is stated in the figure legends. Biochemical experiments were repeated at least once. All stated instrumentations underwent routinely maintenance and quality assessment both prior to and during the data collection period. All attempts at replication were successful.
Randomization	not relevant to the study as it would compromise ability to interpret data
Blinding	not relevant to the study as it would compromise ability to interpret data

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

Monoclonal antibodies generated from healthy donor B cells as described in this study.
goat anti-hu-IgG-AF647 (2040-31, SouthernBiotech)
goat anti-hu-IgG-AF488 (2040-3, SouthernBiotech)
goat F(ab')2 anti-hu-kappa-AF647 (2062-31, SouthernBiotech)
mouse anti-C3b-AF647 (bH6, produced inhouse,)
anti-DNP (produced in house, doi.org/10.1073/pnas.2102787118)
mouse anti-hu-CD19-BV510 (302242, BioLegend)
mouse anti-hu-CD27-PE-Cy7 (560609, BD)
mouse anti-hu-IgG-BV421 (562581, BD)
mouse anti-hu-IgG-BV421 (562581, BD)
mouse anti-hu-CD19-PE-Cy7 (560728, BD)
mouse anti hu-IgG-BV421 (562581, BD)
mouse anti hu-IgG-PE-Cy7 (561298, BD)

Validation

In-house generated antibodies were validated by SDS-PAGE, OD280 concentration measurements and Size Exclusion Chromatography as specified in the materials and methods section.

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)

EXPI293F cells obtained from Thermo Fisher, catalog number #A14527

Authentication

Visual inspection of morphology, proliferation rate monitored, protein production rates of in-house standardized antibody variants monitored.

Mycoplasma contamination

Cell line was tested negative for mycoplasma contamination.

Commonly misidentified lines
(See ICLAC register)

N/A

Plants

Seed stocks

N/A

Novel plant genotypes

N/A

Authentication

N/A

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	For B cell sorts, preparation of B cells is described in material and methods section, subheading B cell isolation. In short, PBMCs were isolated from buffy coats from healthy volunteers. B cells were isolated using untouched B cell isolation kit II (130-091-151, Miltenyi Biotec) For transposon library sort, it is described in Materials and Methods section transposon screening for antibody target identification.
Instrument	For cell sorting: Sony MA900, BD Melody , BDinflux For anaylsis: BD FACSVerse
Software	FlowJo V10
Cell population abundance	Percentages indicated in FACS plots (Fig 1b, c, SFig 1c,d, SFig 3a,f)
Gating strategy	In figure 1d, Sfig 3a,f the gating strategy is provided

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