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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, seeAuthors & Referees and theEditorial Policy Checklist.

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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
	$\blacksquare$ 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.
	X 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
	Estimates of effect sizes (e.g. Cohen's <i>d</i> , Pearson's <i>r</i> ), indicating how they were calculated

Our web collection on  $\underline{statistics\ for\ biologists}$  contains articles on many of the points above.

#### Software and code

Policy information about availability of computer code

Data collection

Standard Illumina NextSeq 500 procedures and software. Base calling and quality scoring were determined using Real-Time Analysis on board.

BD Aria III, BD LSRFortessa

Data analysis

In no particular order: diffHic v1.16.0 (Lun and Smyth, 2015), bowtie2 v2.2.5 (Langmead and Salzberg, 2012), Picard suite v1.117 (https://broadinstitute.github.io/picard/), quasi-likelihood (QL) framework (Lund et al., 2012) of the edgeR package v3.26.5, csaw package v1.18.0 (Lun and Smyth, 2016), TADbit v0.2.0.5 python based software (Serra et al., 2016), limma package v3.40.2 (Ritchie et al., 2015), Sushi R package v1.22.0 (Phanstiel, 2015), rtracklayer package v1.32.2 (Lawrence et al., 2009), Rsubread v1.28.0 (Liao et al., 2013), MACS2 v2.1.0 (Zhang et al., 2008), HOMER v4.11 (Heinz et al., 2010), MEME v5.0.5 (McLeay and Bailey, 2010), cutadapt v0.9.5 (Martin, 2011), HiCRep v1.14.0 (Yang et al. 2017), viridisLite v0.3.0 (Garnier, 2018), IRanges v2.20.2 (Lawrence et al. 2013) FlowJo 10.4.1

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 <u>availability of data</u>

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

Sequence data that support the findings of this study are tabulated in the supplementary tables and are available in the GEO database under accession numbers GSE147497 and GSE99151.

	hment was performed with the program ame of the MEME v5.0.5 89 software package on each pattern. The motif database used was II_MOUSE from MEME.		
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	one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.		
<b>x</b> Life sciences	Behavioural & social sciences Ecological, evolutionary & environmental sciences		
or a reference copy of	the document with all sections, see <a href="mailto:nature.com/documents/nr-reporting-summary-flat.pdf">nature.com/documents/nr-reporting-summary-flat.pdf</a>		
_ife scier	nces study design		
	isclose on these points even when the disclosure is negative.		
Sample size	Sequence experiments were performed in duplicate. From previous previous experience, and as demonstrated in Figures 1b, 1g and Supplementary Figure 1g, the Hi-C and RNA-seq experiments are highly reproducible with duplicate libraries showing minimal variation. These were sufficient to determine differences between samples.  In mouse experiments, data was generated from 4 independent experiments. The reproducible experimental data suggests this sample size is sufficient and allows rigorous statistical testing. Sample size is stated in each figure legend.		
Data exclusions	No exclusions		
Replication	Hi-C and RNA-seq have been replicated twice, whereas mouse experimental data have been obtained from 4 independent experiments.		
Randomization	All males were randomly chosen from the relevant pool.		
Blinding	Results were analysed without blinding of grouping.  Analysis is all computational, no blinding required.		
Reportin	ng for specific materials, systems and methods		
Ve require informati	tion from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material sted 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.		
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Antibodies			
<b>x</b> Eukaryotic	c cell lines Flow cytometry		
<b>x</b> Palaeontol			
📕 🗶 Animals ar	nd other organisms		

# **Antibodies**

Antibodies used

Clinical data

Human research participants

Target-Fluorophore-Clone-Vendor TCRβ-PE-H57-597-BD 562841 CD19-PE/Cy7-ID3-BD 561739 B220-PacB-RA3-6B2-WEHI in house IgM-FITC-X-54-Miltenyi 130-095-906 IgD-APC-11-26c.2a-Miltenyi 130-102-445 CD69-APC-H1.2F3-Miltenyi 130-103-980 CD138-PE/Cy7-281-2-Biolegend 142513 CD22-PE-OX-97-Biolegend 126111 CD22-APC-OX-97-Biolegend 126109

Dilutions used for each Ab have been indicated in Supplementary Table 13.

Validation

Antibody validations were performed by antibody suppliers per quality assurance literature provided by each supplier, and were used as guidelines. Also the antibodies were validated and titrated by using positive/negative control cells from C57BL/6 mouse before the experiments.

## Animals and other organisms

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

Laboratory animals

Experiments were performed using male C57BL/6, FUCCI (Sakaue-Sawano et al, 2008) or Blimp1-transgenic mice (Kallies et al, 2004) at age 6-12 w. Mice were noused in a specific pathogen-free environment with ambient temperature around 23°C,

humidity 40-60% and light/dark cycle of 12h/12h.

Wild animals No wild animals were used in this study.

Field-collected samples No field-collected samples were used in this study.

Walter and Eliza Hall Institute Animal Ethics Committee (#2016-003) Ethics oversight

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 Splenocytes were obtained from mechanically homogenized organs

Instrument Flow cytometric analyses were performed on BD FACSCanto or BD LSRFortessa with sorting performed on the BD FACS Aria III.

FlowJo v10.4.1 Software

Cell population abundance Purity was checked and always exceeded 97%

All cells were initially gated on FSC-A/SSC-A, followed by doublet exclusion by FSC-H/FSC-A, then dead cells were gated out by Gating strategy using PI as viability dye. For naive B cells, they were first gated as B220-PB and CD19-PE/Cy7, then on IgM-FITC and IgD-APC. For 3h, 10h and 33h activated B cells, they were first gated on TCRβ-PE and CD19-PE/Cy7, then on CD69-APC and CTV. For Expanded

activated B cells and plasmablasts, the live cells were gated straight on CD22-PE and CD138-PE/Cy7.

For FUCCI cell cycle analyses, cells were first gated on CD22-APC and CD138-PE/Cy7, then on CTV, followed by mKO2 and mAG.

For Blimp1 mice analyses, live cells were gated on Blimp1-GFP vs SSC-A.

All the gating strategies are shown in Data figures

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