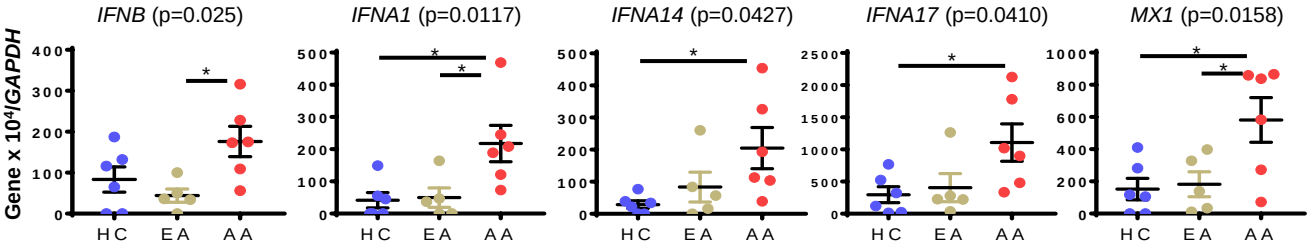
	$IFN_{P/R}$	IFN_{R}	IFN_{P}	P value	Forward Primer	Reverse Primer
TLR3	0.1412	-0.2189	0.1228	**0.0131**	ACCACCAGCAATACAACT	GAATCGTTACCAACCACATT
CCND1	0.3391	-0.1493	0.0662	**0.0235**	CACTTCCTCTCCAAAATG	TGGAAATGAACTTCACAT
CD24	0.3687	-0.1595	-0.2029	**0.0008**	TAAGAGACTCAGGCCAAGA	AATTAGTAGATTCGATGAAGACCT
IFNA21	0.0418	-0.0478	-0.0694	0.8297	CTACTTGGGAACAGAGCCTC	TCCACATTCATCAGGGG
TLR7	0.3439	-0.1888	-0.0925	**0.0026**	CAGAACTATATCTCTACAACAACA	GAGGGCAATTTCCACTTAG
IFIT1	0.4240	-0.0613	-0.1664	**0.0073**	ACAGCAACCATGAGTACAA	CAATGGATAACTCCCATGTAAAG
IFNA14	0.0363	-0.2431	0.2642	**0.0025**	GTGGTGCTCAGCTGCAAGTC	GGCTGTGGGTTTGAGACAGATT
IFNB	0.1931	-0.2734	0.1935	**0.0012**	GTCTCCTCCAAATTGCTCTC	ACAGGAGCTTCTGACACTGA
IFNA7	0.2548	-0.3836	0.2586	**<0.0001**	GCCCGGTCCTTTTCTTTACTG	TTCATGTCTGTCCTTCAAGC
IFNA16	-0.1909	-0.3500	0.5277	**<0.0001**	TATGATTTCGGATTCCCCCAGGAGGTG	GTCTCATCCCAAGCAGCAGATGAATC
IFNA10	-0.3317	-0.3818	0.6471	**<0.0001**	TAGGAGGGCCTTGATACTCCTGGG	TGCCATCAAACTCCTCCTGGGGGAT
IFNA17	-0.1350	-0.4362	0.6083	**<0.0001**	CCGTGCTGGTGCTCAGCTA	TGTGGGTCTGAGGCAGATCA
IFNA4	-0.2855	-0.4049	0.6811	**<0.0001**	ACTCCTGGCACAAATGGGAAGAATCTCTCA	GAGCCTTCTGGAACTGGTGGCCA
IFNA2	-0.1058	-0.1516	0.1605	0.0975	CTTGAAGGACAGACATGACTTTGGA	GGATGGTTTCAGCCTTTTGGA
IFNA1	0.1589	-0.2682	0.1449	**0.0020**	CTTCAACCTCTTTACCACAAAAGATTC	TGCTGGTAGAGTTCGGTGCA
IFNA8	0.1362	-0.2620	0.2347	**0.0008**	CCTTCTAGATGAATTCTACATCGAACTTG	ACTCTATCACCCCCACTTCCTG
IFNA5	-0.2513	-0.1990	0.3234	**0.0002**	CCCTGGTGGTGCTCAACTG	CTTCCCATTTGTGCCATTATC
IFNA6	-0.3010	0.0025	0.0254	0.1212	TCCATGAGGTGATTCAGCAGAC	GCTGCTGGTAAAGTTCAGTATAGAGTTT
IFIT2	-0.2386	0.3315	-0.3557	**<0.0001**	GATACGCAGGTAGAGAGGAA	TGTAACGTTGAACCAGTTGT
IRF7	0.5169	-0.1419	-0.2711	**<0.0001**	TCCTGGTGAAGCTGGAA	CATAAGGAAGCACTCGATGTC
ZBP1	0.6254	-0.1405	-0.2691	**<0.0001**	CCAACAACGGGAGGAAG	TTTGCTGTCCTCATTCCC
MX1	-0.0943	0.1510	-0.3301	**0.0018**	CCACACTGCGAGGAGATC	GTAAGCTCTAATATACGTTCCTTC
PKR	-0.0152	0.1101	-0.2613	**0.0474**	CTCCACATGATAGGAGGTTTAC	TGCTTCCTTCTTTGATCTACC
IRF9	0.1803	0.0492	-0.3078	**0.0158**	CTCCAGGACTCCCTCAATAA	CCTCAGTTGTGTCTGTAACTTC
TLR9	-0.1480	0.1240	-0.1546	0.1174	TGAGCTACAACAACATCAT	CAGAGTCTAGCATCAGGA
CCND2	-0.2741	0.1984	-0.1981	**0.0050**	GACGTGGATTGTCTCAAA	GTTCATCCTCCGACTTGG
CD69	0.0645	0.0276	-0.2690	0.0614	GATACGCAGGTAGAGAGGAA	TGTAACGTTGAACCAGTTGT
BAFFR	0.0779	0.0114	-0.1345	0.5039	CCTCACCATCTTTGACAG	AAGCGGTCTTTTACTCAT
IFNAR1	-0.1240	0.2065	-0.2415	**0.0061**	TCAGGTGTAGAAGAAAGGATTG	CCATGACGTAAGTAGTGCTG
IFNAR2	-0.0904	0.1149	-0.1176	0.2799	TCACCGTCCTAGAAGGATTCAG	TCACCGTCCTAGAAGGATTCAG
IRF3	0.2405	-0.0477	-0.1245	0.1583	TTCTGATACCCAGGAAGACA	GTGGGATTGTCCAAGCTG
RIG1	-0.0004	0.2644	-0.3873	**0.0002**	TGGTTTAGGGAGGAAGAGG	CTCCAACAGGAACTTGAGAAA

Supplemental Table 1. Genes differentially expressed in $IFN_{P/R}$ vs IFN_{R} vs IFN_{P} transitional B cell clusters derived from 3 SLE patients.

BioMark qRT-PCR analysis was used to determine the expression of the indicated gene in Fluidigm single captured transitional B cells ($CD24^{+}CD38^{+}IgD^{+}CD27^{-}$) derived from each patient. Data are mean of gene expression after normalization using the Clustvis analysis. Gene cluster analysis (shown on the left) was carried out using the Clustvis online software. The order of genes shown is based on the cluster analysis as shown in Fig. 1. Significant differences among means were analyzed using a one way ANOVA test with p value shown. Significantly differentially expressed genes are red color coded ($p<0.05$). All primers used are shown in the right.

A

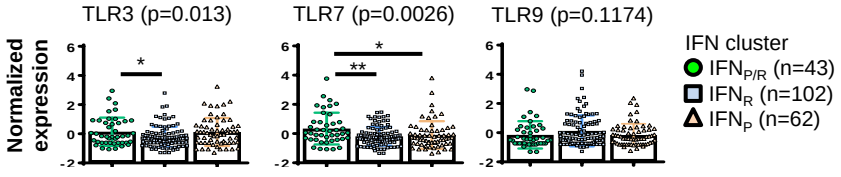


B

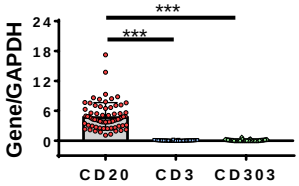
	SLEDAI	α-dsDNA	Low C3/C4	α-Sm	α-SSA	Renal	AM	IS	CS	Race	Sex	Age
SLE 1	13	Positive	Positive	Positive	Positive	Positive	Yes	None	Yes	AA	F	34
SLE 2	4	Historic pos*	Negative	Positive	ND**	Positive	Yes	None	No	AA	F	40
SLE 3	5	Negative	Negative	Positive	ND**	Positive	No	MMF	No	AA	F	50

* Historically positive; ** ND = not determined; AM = anti-malaria; IS = immune suppressant; CS = corticosteroid

C

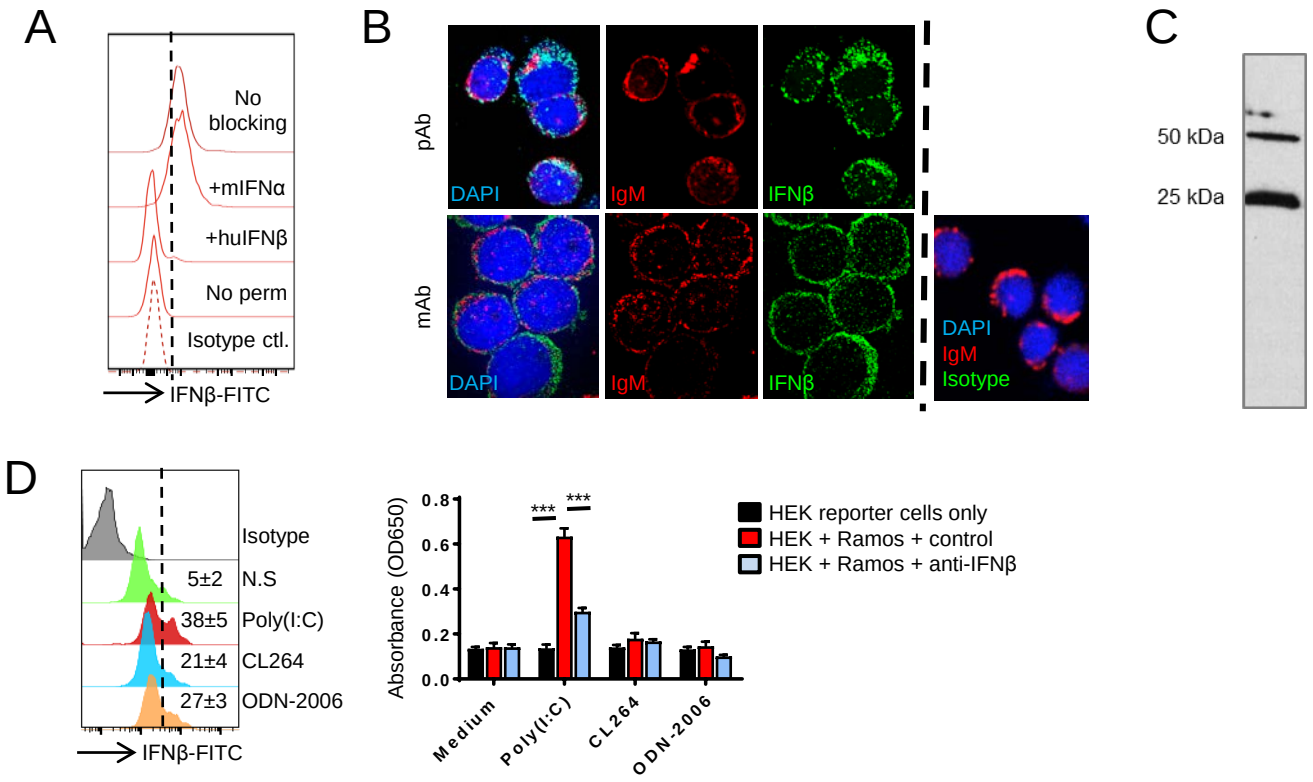


D



Supplemental Fig. 1. Type I IFN associated genes in SLE transitional B cells.

(A) qRT-PCR gene expression of the indicated gene in bulk transitional B cells ($CD24^+CD38^+IgD^+CD27^-$) from the PBMCs of healthy controls (HC), European American (EA) SLE patients, and African American (AA) SLE patients. Results are the expression of each gene after normalization with the expression of *GAPDH* (mean±s.e.m, n=6 for HC, 5 for EA SLE, and 6 for AA, SLE). **(B)** Clinical data of SLE patients whose PBMC Tr B cells were subjected for the Fluidigm/BioMark gene expression analysis shown in Figure 1. **(C)** Normalized expression values of the indicated TLR genes in in $IFN_{P/R}$ vs. IFN_R vs IFN_P transitional B cell clusters derived from 3 SLE patients. Significant differences among means were analyzed using a one way ANOVA test with p value shown on the top of each graph. **(D)** Expression of the indicated genes in single Tr B cells in SLE Patient #3 (mean±s.d.; Differences between groups were analyzed using Tukey's multiple comparisons test (n=69 single Tr B cells). Significant differences among means (A, C, and D) were analyzed using a one way ANOVA test with p value shown on the top of each graph. Differences between groups were analyzed using Tukey's multiple comparisons test (* $p<0.05$, ** $p<0.01$ or *** $p<0.005$ between the indicated comparisons).



Supplemental Fig. 2. Detection of intracellular IFNβ and its secretion by B cells.

(A) Histograms showing mode normalized counts of IFNβ staining in total CD19^{+} B cells from a representative SLE patient. Anti-IFNβ antibody was pre-incubated with either human IFNβ or mouse IFNα prior to FACS staining. In the bottom two histogram tracks, cells were either left unpermeabilized prior to anti-IFNβ antibody incubation (No-perm) or stained with an isotype control antibody (Isotype ctl.). **(B)** Super-resolution microscopy imaging (objective lens: 100x) of IFNβ in purified SLE B cells comparing two commercial Abs (*Top*: rabbit polyclonal Ab from Abcam; *Bottom*; mouse monoclonal FITC-conjugated Ab from PBL) with IgG1-k isotype control shown (right). **(C)** Immunoblot analysis of IFNβ in cytoplasmic lysates prepared from freshly isolated SLE B cells. **(D)** Left: Representative FACS plots showing the frequency of IFNβ^{+} Ramos B cells 8 hrs following stimulation with the indicted agonistic TLR ligand (5 μg/mL each); Right: HEK-blue reporter analysis of type I IFN secretion by Ramos cells under the indicated conditions of stimulation (5 μg/mL) or blocking antibody treatment (unspecific rabbit IgG or a polyclonal rabbit anti-human IFNβ,10 μg/mL each) 24 hrs post stimulation; (mean±s.e.m.; n=3, $p<0.005$ between the indicate comparison, Tukey's multiple comparisons test).