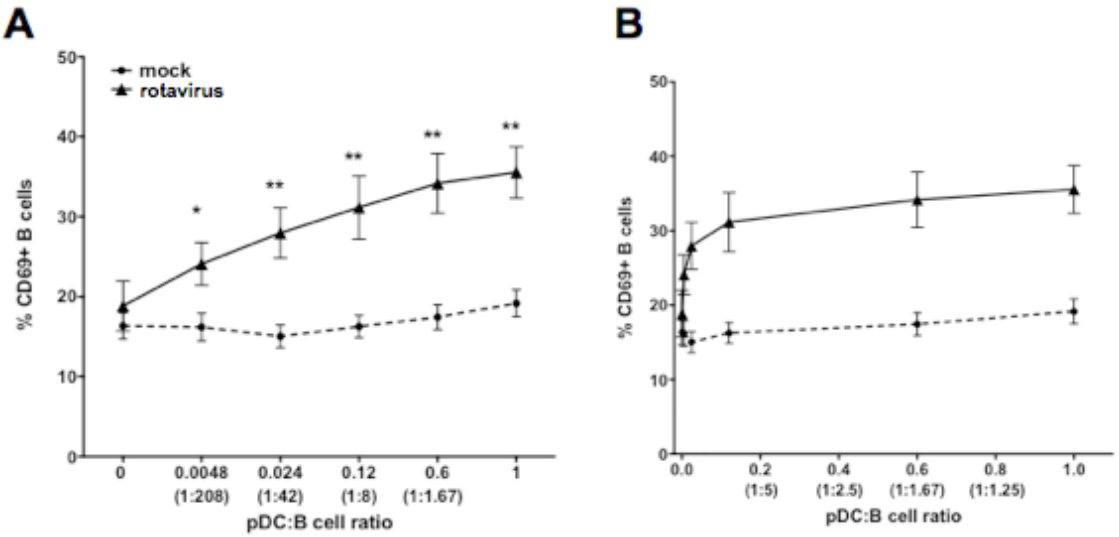
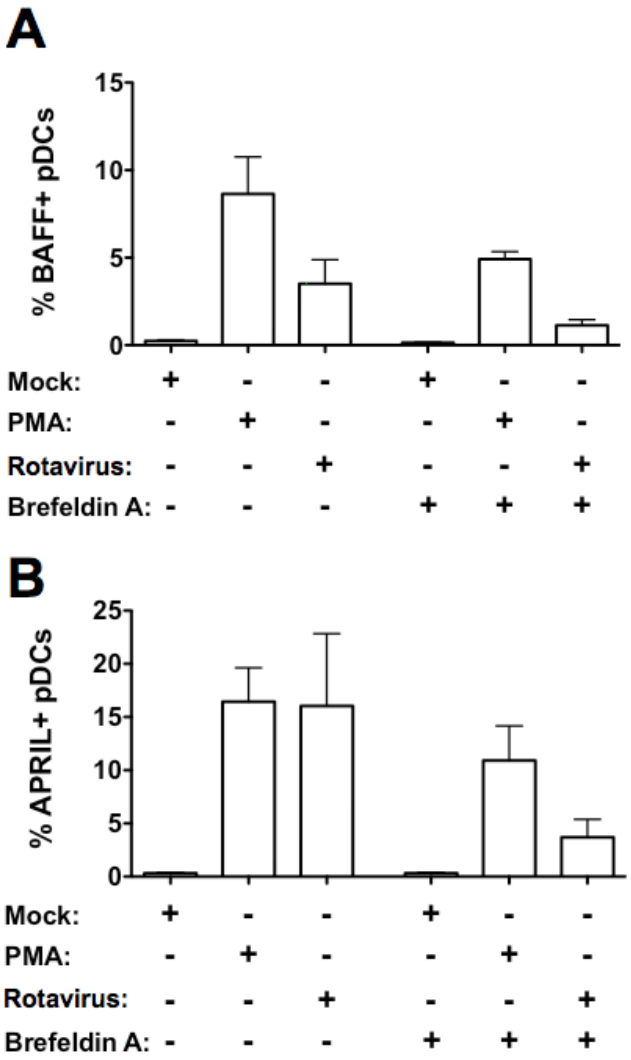
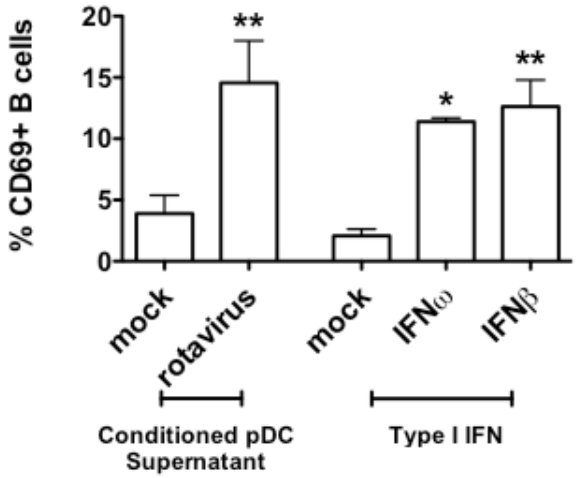
Supplemental Figure 1: Titration of the pDC:B-cell ratio required for B-cell activation by rotavirus. A: B-cell activation following coculture with pDCs at the indicated ratio was assessed by flow cytometry for expression of CD69 12h after mock (circles) or rotavirus (triangles) stimulus. **B:** Linear representation of (A). *n*=4, *: p≤0.04; **:p≤0.008; vs. mock; Mann-Whitney test.



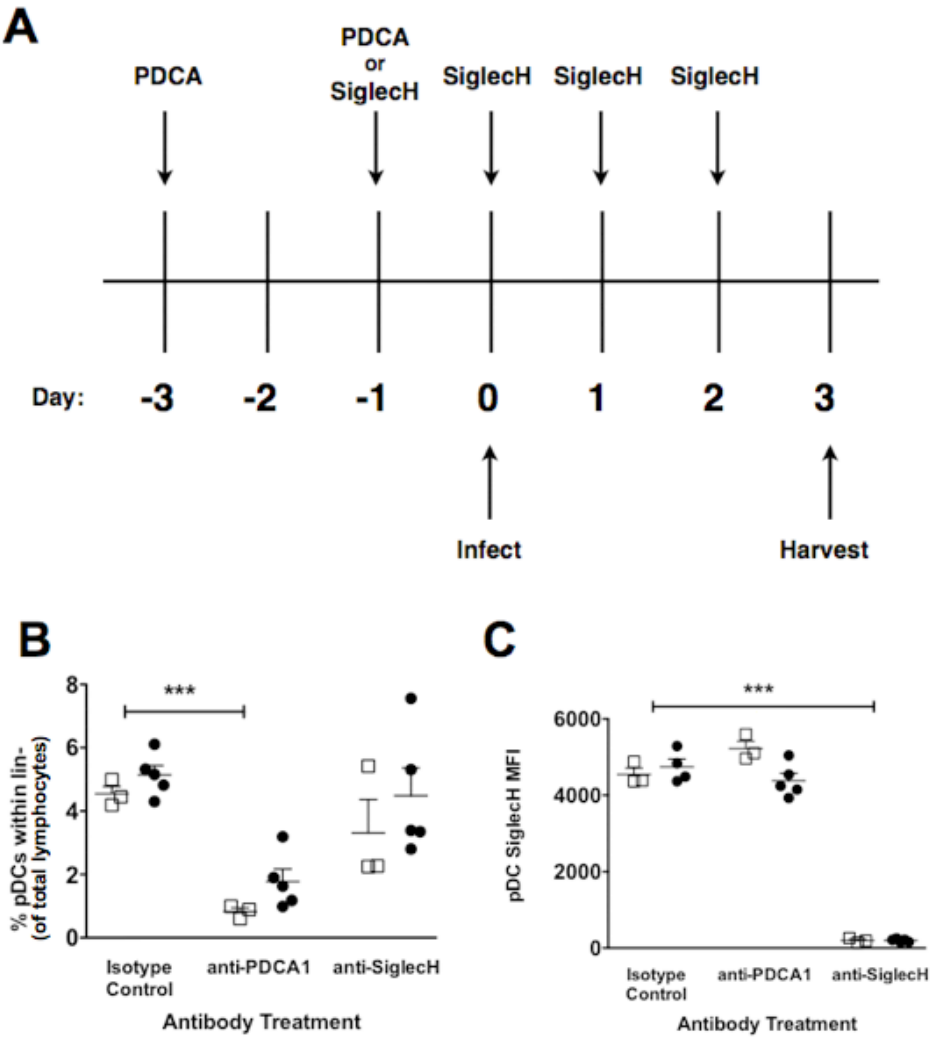
Supplemental Figure 2: Rotavirus induces pDC expression of BAFF and APRIL. BAFF (**A**) and APRIL (**B**) expression on purified pDCs 12h after the indicated stimulus in the presence or absence of brefeldin A was detected by flow cytometry. *n* = 3.

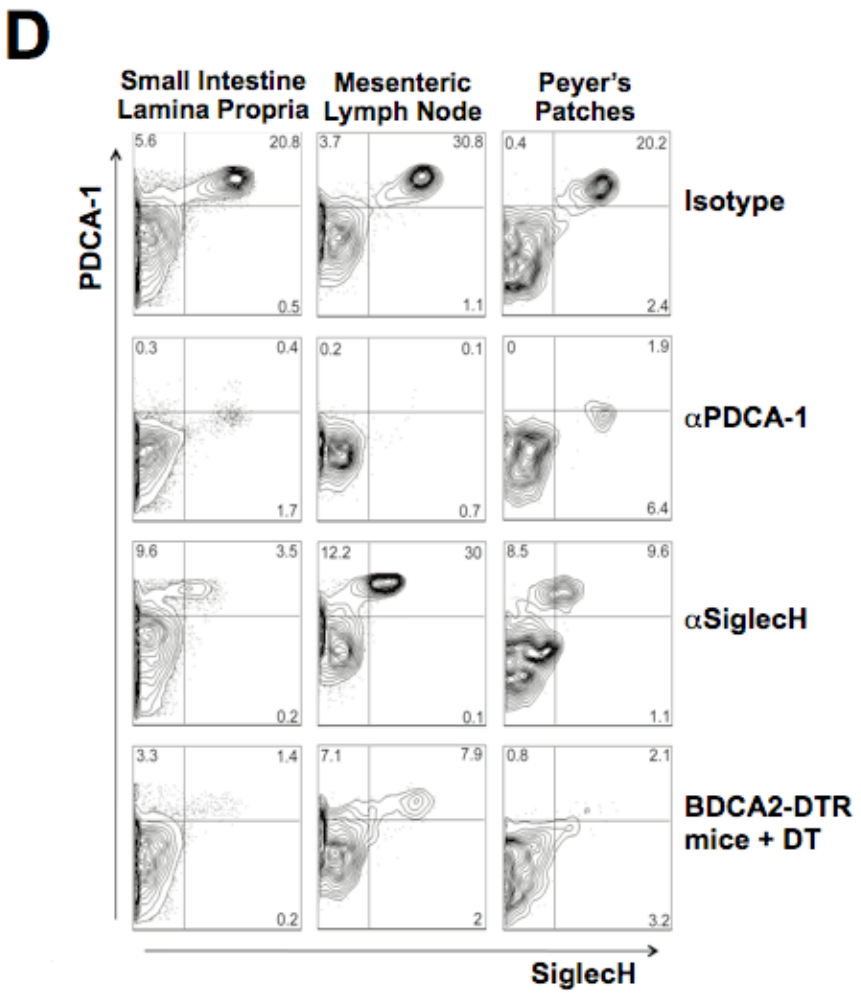


Supplemental Figure 3: IFNω and IFNβ induce human B-cell activation. B-cell activation was assessed by flow cytometry analysis of CD69 expression by purified B cells cultured for 12h after stimulus with supernatants from mock- or rotavirus-stimulated pDCs or mock, IFNω, or IFNβ, as indicated. * : $p \leq 0.05$; ** : $p \leq 0.01$; vs. appropriate mock; repeated measures ANOVA with Tukey's multiple comparison test. *n* = 3.

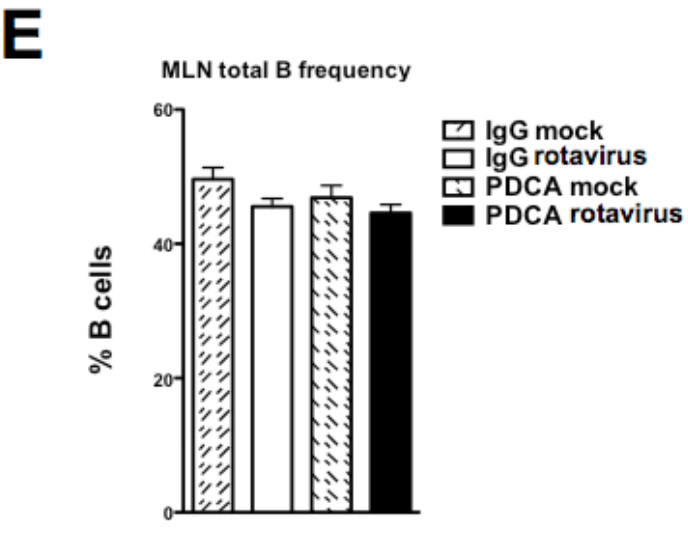


Supplemental Figure 4: Antibody administration and pDC kinetics. A: SiglecH, PDCA-1, or IgG2b isotype control antibody was administered i.p. at the indicated times. **B, C**: Antibody administration affects the percent of PDCA-1^{+}lineage^{-} cells at 3dpi (**B**) and SiglecH MFI at 7dpi (**C**) within the spleens of mock (white) or rotavirus (black) inoculated mice. **D**: Antibody or diphtheria toxin administration affects the percent of PDCA-1^{+}SiglecH^{+}lineage^{-} cells within the indicated organs at the time of infection. **E**: The B cell population within the MLN is not affected by PDCA-1 administration. ***: p ″ 0.002; n=3-5 per group; t test.



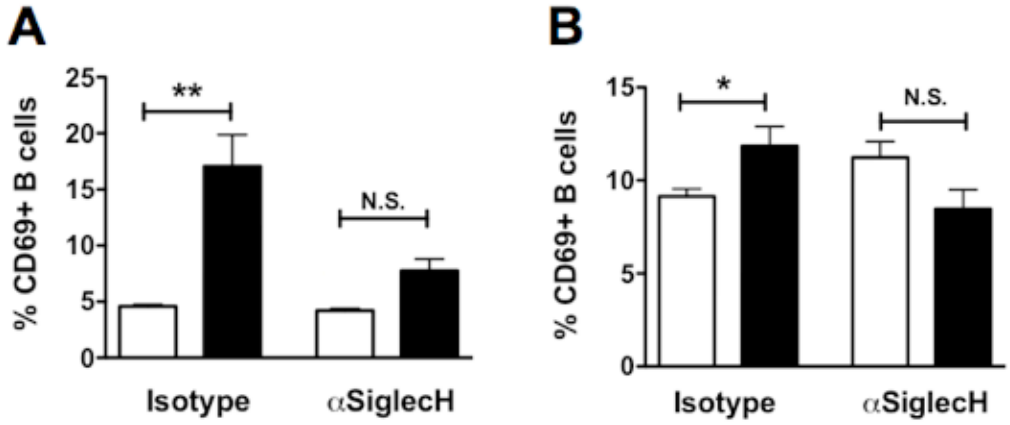

D
Small Intestine Lamina Propria
Mesenteric Lymph Node
Peyer's Patches
PDCA-1
SiglecH
Isotype
αPDCA-1
αSiglecH
BDCA2-DTR mice + DT
5.6
20.8
0.5
3.7
30.8
1.1
0.4
20.2
2.4
0.3
0.4
1.7
0.2
0.1
0.7
0
1.9
6.4
9.6
3.5
0.2
12.2
30
0.1
8.5
9.6
1.1
3.3
1.4
0.2
7.1
7.9
2
0.8
2.1
3.2

1002


E
MLN total B frequency
% B cells
60
40
20
0
IgG mock
IgG rotavirus
PDCA mock
PDCA rotavirus

1003

Supplemental Figure 5: anti-SiglecH antibody prevents B-cell activation following in vivo rotavirus infection. CD69 expression was assessed on B cells isolated from the Peyer's Patches (**A**) and mesenteric lymph node (**B**) of isotype-control or anti-SiglecH-treated mice 3 days following in vivo rotavirus infection. *: p ″ 0.05; **: p ″ 0.01; n=3-5 per group; t test.



Supplemental Figure 6: anti-SiglecH antibody inhibits type I IFN secretion by pDCs. Murine pDCs were purified from FLT3L-treated bone marrow and stimulated with CpG 1585 or mock in the presence or absence of 5 µg/mL anti-SiglecH antibody. Resulting IFNα secretion was assessed by ELISA.