

Supplemental Figures

Supplemental Figure 1. Analysis of CD21 expression by A/J T cells

following immunization with HEL in the presence of CFA. Mature A/J female mice were immunized with HEL+ CFA or used as un-immunized controls. Mice were immunized on D0 and D7 and then sacrificed and cells analyzed on D10. Experimental groups consisted of three animals each. Data display results from one animal for each group but are representative of all repeated experiments. **A)** Lymph node. **B)** Spleen. The oval indicates the position of possible CD4+/CD21+ cells.

Supplemental Figure 2. Analysis of CD21 expression by memory T cells in

DO11.10 mice. Mature DO11.10 mice were immunized with OVA or equivalent volume of PBS on D0 and D7 and then sacrificed on D14. **A)** Lymph node. **B)** Spleen.

Supplemental Figure 3. Analysis of *in vitro* activated murine splenocytes for the expression of CD21 by T cells.

Total splenocytes from mature C57BL/6 males or C57BL/6 *Cr2* deficient animals (CR2^{-/-}) were collected and cultured in the presence or absence of PMA+ ionomycin for 6 or 24 hours. Cells were stained with the indicated antibodies. The oval indicates the position of possible CD4+/CD21+ cells.

Supplemental Figure 4. CD21 expression by murine T cells is coincidental with cell death.

Total splenocytes from mature C57BL/6 males were collected and cultured in the presence or absence (control) of PMA+ ionomycin for 6 hours, and then analyzed for apoptosis via Annexin V staining. Total cells (no gate) stained for CD4+ and CD21+ are shown in the left, top panel. The three gated quadrants R1, R2 and R3 obtained from the 7-AAD and Annexin V staining were then analyzed for CD4 and CD21 staining. The results from a single set of experiments are shown but are representative of multiple similar analyses.

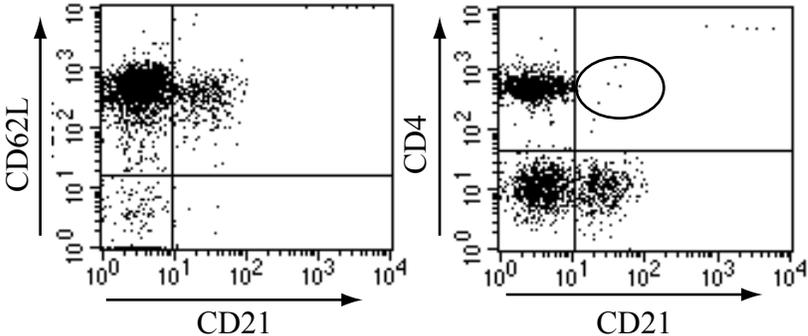
Supplemental Figure 5. Transcript analysis of CR1, CR2 and CD23 gene expression in mice lacking the MH box sequences. Total bone marrow (A), total spleen (B) and total thymus (C) RNA samples were obtained from three WT (C57BL/6) and three *Cr2iΔ* mice. Samples were analyzed for expression of CR1, CR2 and CD23 transcripts. Transcript levels are shown relative to β -actin. Data is representative of three different RNA isolations and RT-PCR analysis. Open bar, C57BL/6; black bar, *Cr2iΔ*. There is no significant statistical difference between any of the sets of samples.

Supplemental Figure 1. Roundy, et al.

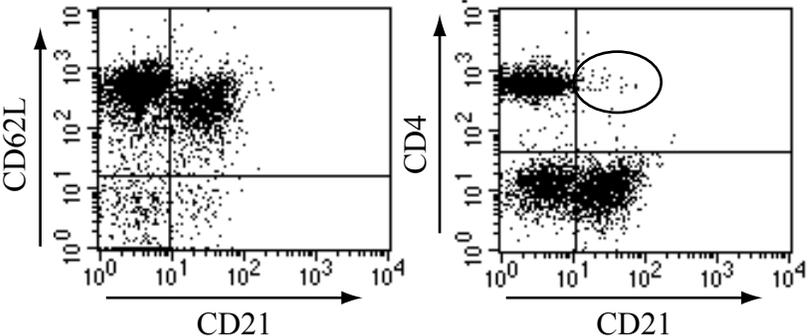
Lymph Node

A

Control



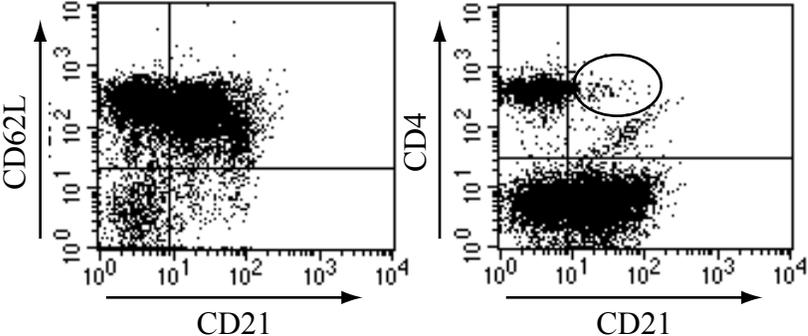
**HEL+
CFA**



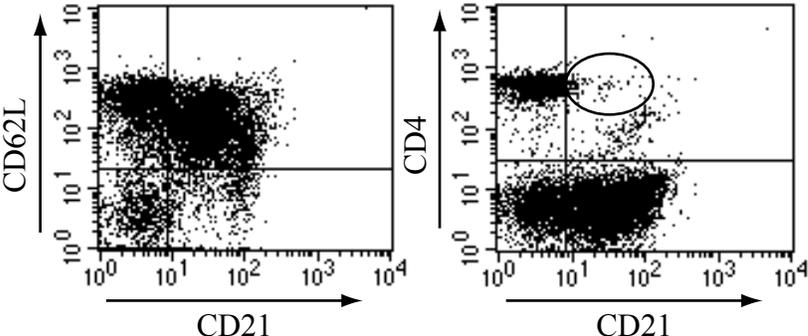
Spleen

B

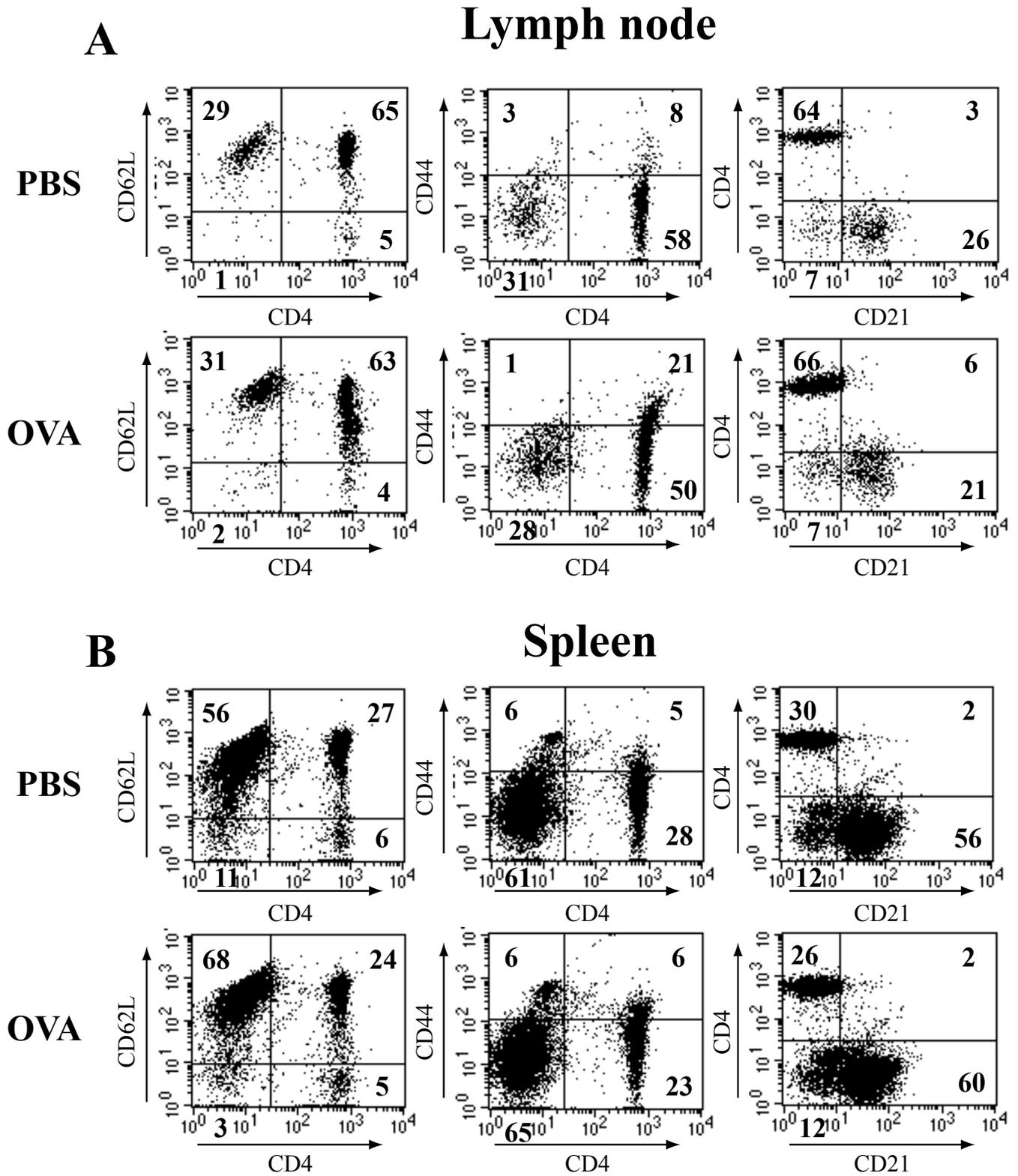
Control



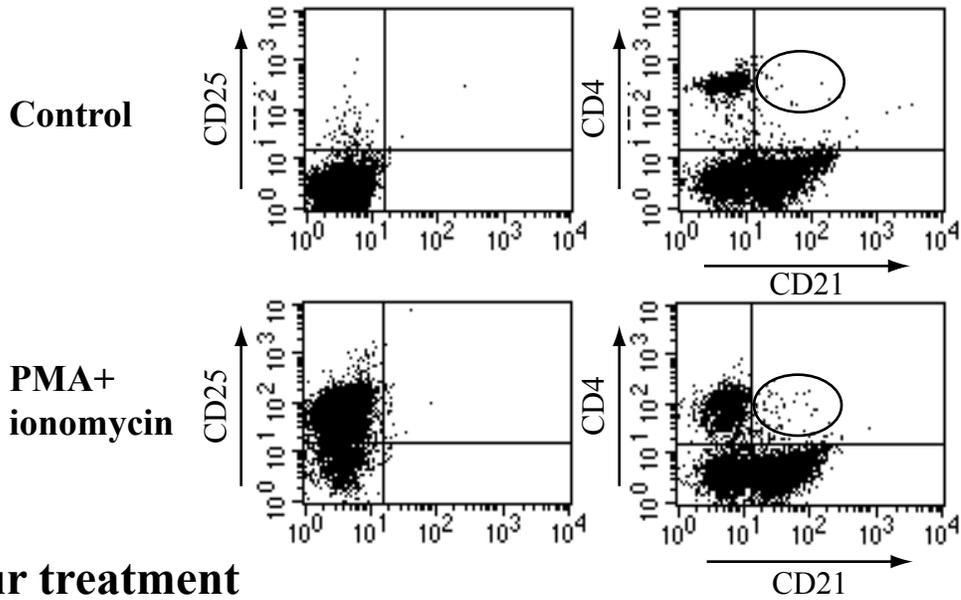
**HEL+
CFA**



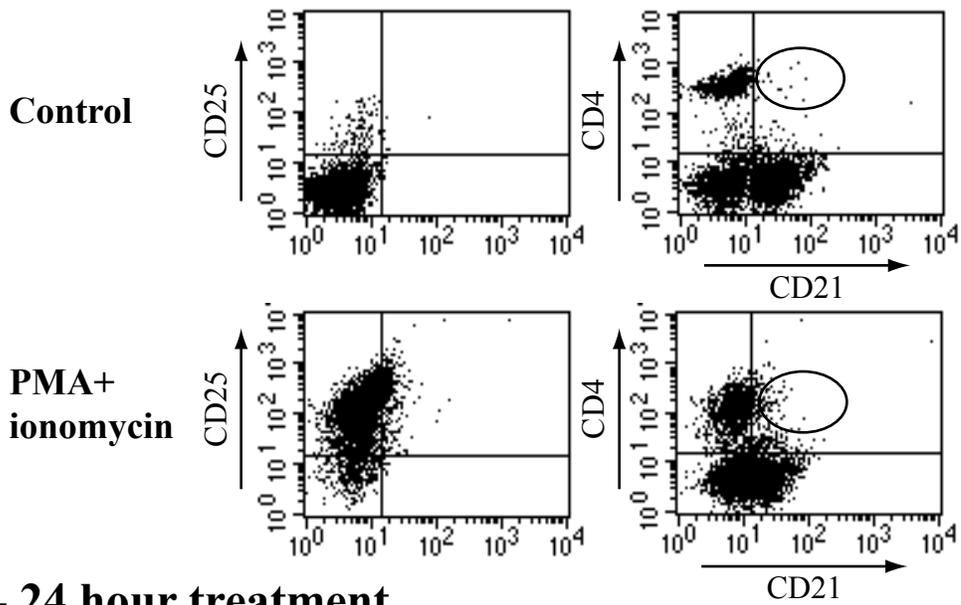
Supplemental Figure 2. Roundy, et al.



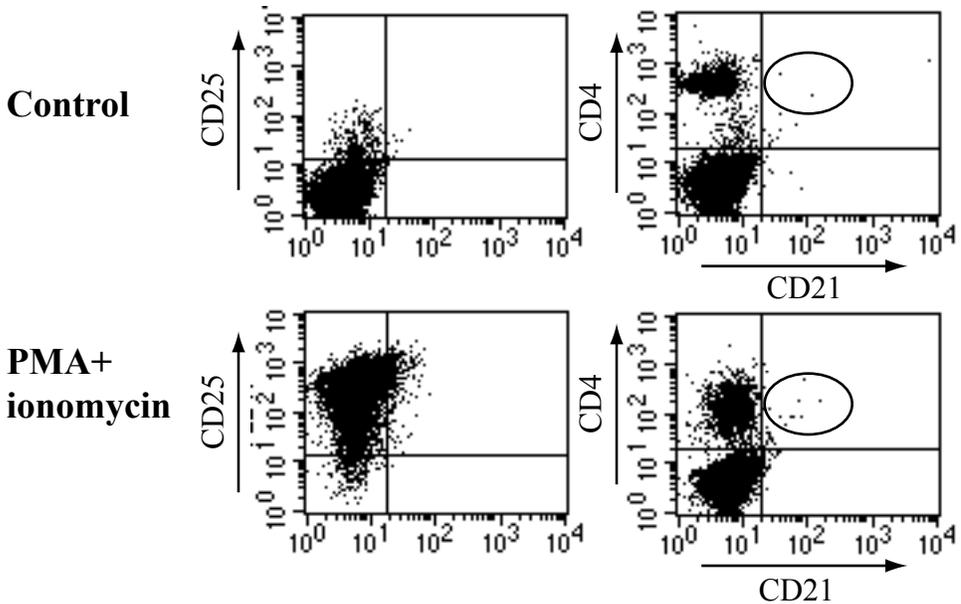
6 hour treatment



24 hour treatment

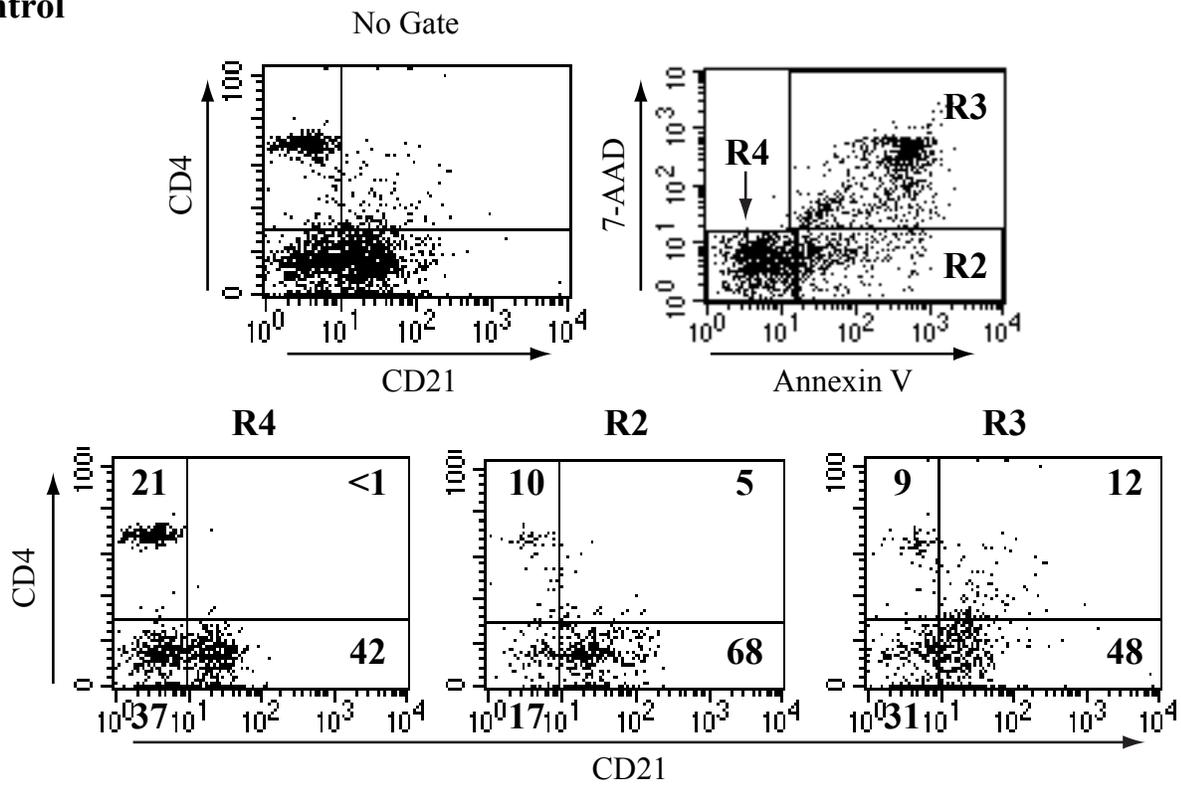


CR2^{-/-} 24 hour treatment

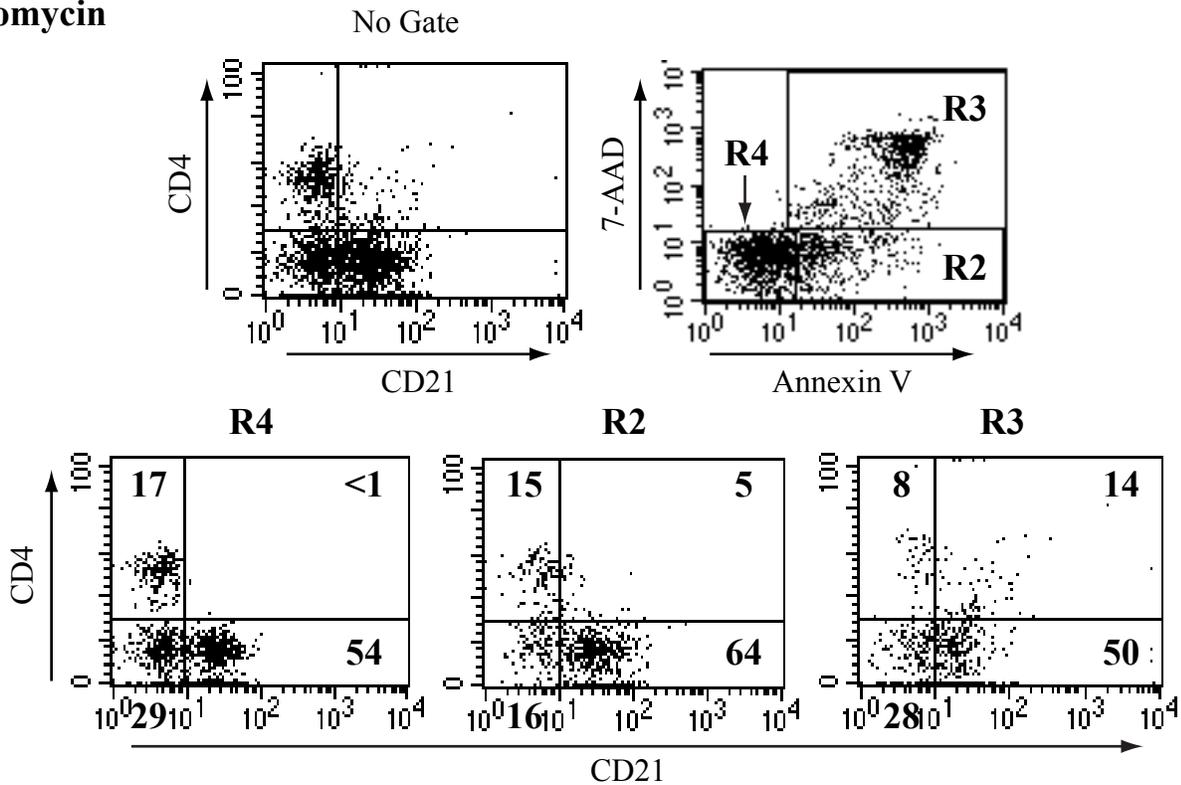


Supplemental Figure 4. Roundy, et al.

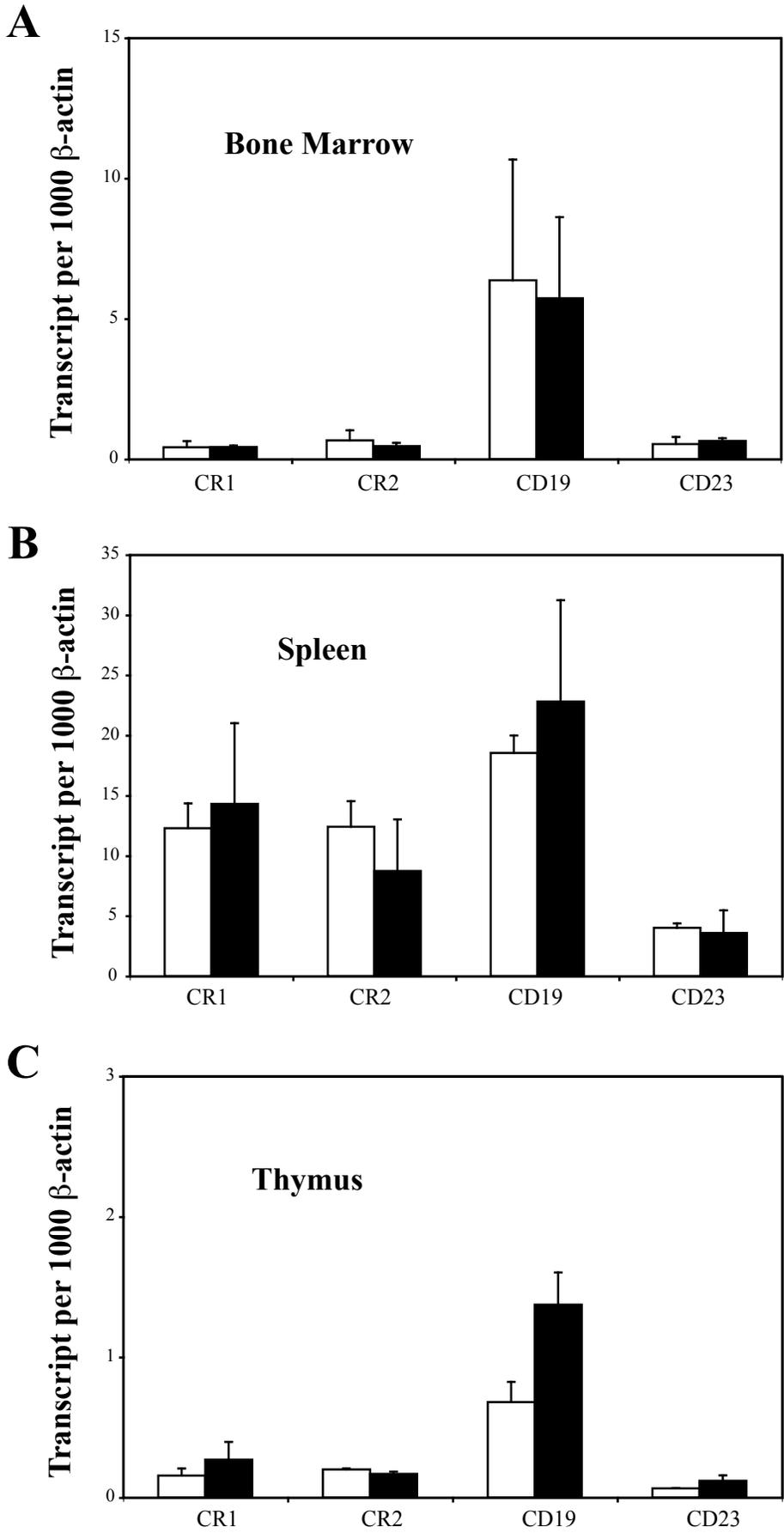
Control



PMA + ionomycin



Supplemental Figure 5. Roundy, et al.



| | Wildtype | Cr2iΔ |
|---------------------------|------------|------------|
| <i>Spleen</i> | | |
| CD21+ | 53.7 ± 1.9 | 53.7 ± 2.4 |
| CD35+ | 52.7 ± 1.3 | 54.4 ± 6.4 |
| B220+ | 60.2 ± 2.4 | 60.6 ± 0.6 |
| B-2 | 71.9 ± 4.2 | 73.3 ± 4.6 |
| MZ | 6.6 ± 3.1 | 6.7 ± 2.5 |
| B-1a | 0.9 ± 0.5 | 0.7 ± 0.5 |
| <i>Bone Marrow</i> | | |
| CD21+ | 6.1 ± 3.5 | 4.6 ± 1 |
| CD35+ | 7.4 ± 3.6 | 4.0 ± 0.3 |
| B220+ | 27 ± 2.1 | 29.7 ± 6.5 |
| <i>Thymus</i> | | |
| CD21+ | 0.3 ± 0.1 | 0.2 ± 0.2 |
| CD35+ | 0.4 ± 0.2 | 0.4 ± 0.2 |
| CD3+ | 15 ± 4.4 | 13.2 ± 4.9 |
| CD19+ | 0.3 | 0.4 ± 0.3 |

% of live cells

Immature animal

| | Wildtype | Cr2iΔ |
|---------------------------|------------|------------|
| <i>Spleen</i> | | |
| CD21+ | 28.6 ± 2.2 | 24.1 ± 2.8 |
| CD35+ | 29.1 ± 7.1 | 29.4 ± 3.2 |
| B220+ | 73.2 ± 5.1 | 72.7 ± 5.8 |
| B-2 | 22.2 ± 3.3 | 21.3 ± 4.6 |
| <i>Bone Marrow</i> | | |
| CD21+ | 1.7 ± 0.7 | 1.0 ± 0.7 |
| CD35+ | 1.7 ± 0.5 | 1.4 ± 0.7 |
| B220+ | 59.8 ± 0.4 | 51.8 ± 8.2 |
| <i>Thymus</i> | | |
| CD21+ | 0.5 ± 0.6 | 0.1 ± 0.1 |
| CD35+ | 1.0 ± 0.8 | 0.7 ± 0.7 |
| CD3+ | 13.9 ± 1.3 | 14.2 ± 1.4 |
| CD19+ | 0.5 ± 0.4 | 0.1 ± 0.1 |

Supplemental Table 1. B cell subsets comparing Cr2iΔ and wild type (C57BL/6) mice.