

13.8% ± 5.2%

4.8% ± 2.8% n=16

P<0.01

GC% (CD95+/PNAhigh)



Supplementary Figure 1. Defective germinal center (GC) B cell formation in miR-155 null mice. A) Mature B cells isolated from miR-155 +/+ and -/- mice were grown in the presence of LPS/IL4 for 96h and examined by FACS for the presence of GC B cell subpopulation (B220+, CD95+/PNAhigh). Significantly fewer GC cells were generated from a miR-155 null splenic mature B cell population than from WT counterpart (p< 0.01 two-sided Student's t-test). Data shown are mean and SD of 8 pairs of mice. A representative FACS display is shown at the top. **B)** Mature B cells isolated from miR-155 +/+ and -/- mice immunized with NP-CGG and examined by FACS for the presence of GC B cell subpopulation (B220+, CD95+/PNAhigh). Significantly fewer GC cells were generated in miR-155 null than in WT littermates (p< 0.01 two-sided Student's t-test). Data shown are mean and SD of 8 pairs of mice. A representative FACS display is shown at the top.

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Supplementary Figure 2. Ectopic miR-155 expression in mature B cells. Stem-loop RT-PCR was used to quantify miR-155 levels in mature B cells from four unique miR-155 null mice transduced with an empty MSCV or MSCV-miR-155 retrovirus. A robust reconstitution of miR-155 expression was detected in all cases. Data shown are mean ±SD of the delta CT values for each assay (normalized by the expression of the small nucleolar RNA 202). Note that lower the CT value, higher the expression of the target gene is.



Supplementary Figure 3. Expression of p53 transcriptional targets. Mature B cells (A), thymocytes (B), and bone marrow cells (C) from miR-155+/+ and -/- mice (n=4) were isolated and exposed to ionizing irradiation (IR, 5Gy), etoposide (4 μ M for 20h), or were vehicle control exposed and not irradiated (ctrl). Real-time RT-PCR quantification of four p53 transcriptional targets (*p21, Cdc25c, Gaddd45a, Pcna*) defined their marked modulation by IR or upon etoposide exposure (normalized by ctrl cells levels). However, this p53-mediated transcriptional activity was not significantly different between miR-155+/+ or -/- cells. Data represent the mean ± SD of independent RT-PCR assays performed in triplicate. As a control, the mature B cells from these four mice were also exposed to LPS/IL4, and the expected significantly higher DSB foci accumulation was detected in the miR-155 -/- model (**P*<0.01 two-sided Student's t-test). Data shown are mean and SD of 50 nuclei scored/mouse.



Supplementary Figure 4. Socs1 and p53 form a complex in mature murine B cells. p53 was immunoprecipitated (IP) in whole cell lysates of miR-155 +/+ and -/- mature B cells (n=4 mice). In all instances, Socs1 was readily co-immunoprecipitated with p53. IgG control IP (left most lane) and immunoblotting with p53 confirms the specificity of this assay. The panels on the bottom depict the IP input protein, which as expected show a higher expression of Socs1 in miR-155-/- cells. Repeated attempts to IP Socs1 were unsuccessful indicating that the tested antibodies are not capable of precipitating this protein.