

Supplemental Figure 1. Gating strategy. A. Gating of surface 4-44⁺ intracellular 4-44⁺ CD19⁺ AM14 B cells, CD138⁺CD44⁺ PBs and FAS⁺GL7⁺ GC AM14 B cells in immunized and unimmunized mice, as well as ABC gating strategy (CD5^{lo}CD93^{lo}CD21^{lo}CD23^{lo}T-bet⁺CD11c^{+/-}). **B**. Gating of IgM⁻IgD⁻CS AM14 B cells. **C**. CD4⁺ T cell gating identifying GFP⁺ 13C2 T cells and KJ1-26 DO11.10 T cells, further gated as CD44⁺CD62L⁻ Tem, PD-1⁺PSGL1^{lo} Tfh, PD-1⁺PSGL1⁺ Tef, and BCL-6⁺PD-1⁺ Tfh cells. The expression of KI-67, BCL-2, and IL-2 were also assessed in these cells.



Supplemental Figure 2. Multiple glycolysis inhibitors impair the antibody production from AM14 B cells. A – F. Purified AM14 B cells were stimulated with PL2-3 or R848, without inhibitors (Ctrl), 2DG, bromopyruvate (3-BP), lanodamine (LND), koningic acid (KA) or etomoxir (Eto). Quantification of 4-44 IgM (A) and total IgM (B) in the supernatant. C. Frequency of CD138⁺ plasmablasts. D. p4E-BP1 expression. E. pAKT1 (Ser 473) expression with representative FACS plots. G-H. AM14 B cells were transferred into BALB/c mice that were immunized with PL2-3 and treated with 2DG or LND. G. Serum 4-44 IgM and total IgM. H. pAKT1 expression in AM14 B cells relative to the host polyclonal B cells. Statistical significance is indicated for paired t tests relative to controls. *P < 0.05; **P < 0.01, ****P < 0.0001.



Supplemental Figure 3. The inhibition of glycolysis impairs the cognate activation of TLR7 stimulated AM14 B cells. AM14 B cells were transferred into DO11-10 (A-D) or BALB/c (E-P) mice immunized with PL2-3. Some recipients were pre-treated with R848, then treated or not with 2DG starting with PL2-3 immunization. Control (Ctrl) were untouched mice from the recipient strain. **A**. Serum anti-dsDNA IgG. Frequency of endogenous (non-AM14) GC B cells (**B**) and PB + PC (**C**). **D**. Number of endogenous ABCs. **E**. Absolute numbers of splenocytes. **F**. Serum anti-dsDNA IgG. Frequency of AM14 PBs. **K**. Serum 4-44⁺ IgM. **L**. Frequency of IgD^{Iow}Ki-67⁺ cells among AM14 B cells. **M**. Correlation between the frequency of IgD^{Iow}Ki-67⁺ AM14 B cells and 4-44⁺ IgM serum levels. **N**. Absolute numbers of AM14 ABCs. **O**. Absolute numbers of endogenous ABCs. Data were pooled from 2 experiments for each type of recipients. Each point represents a mouse. The statistics correspond to 1-way ANOVA with multiple comparison tests or to Pearson's tests (**F** and **I**). *P < 0.05; **P < 0.01. ***P < 0.001, ***P < 0.001.