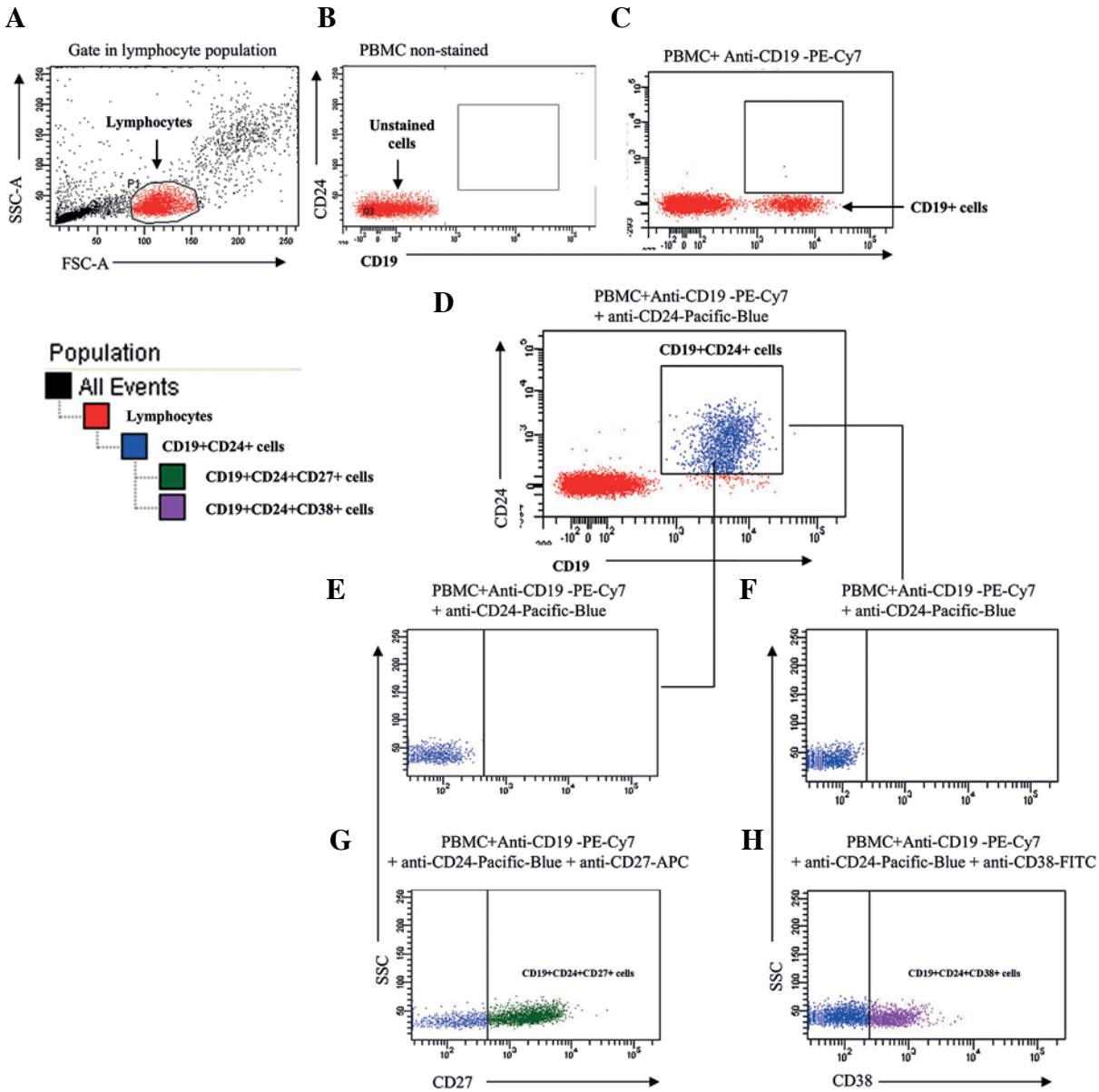
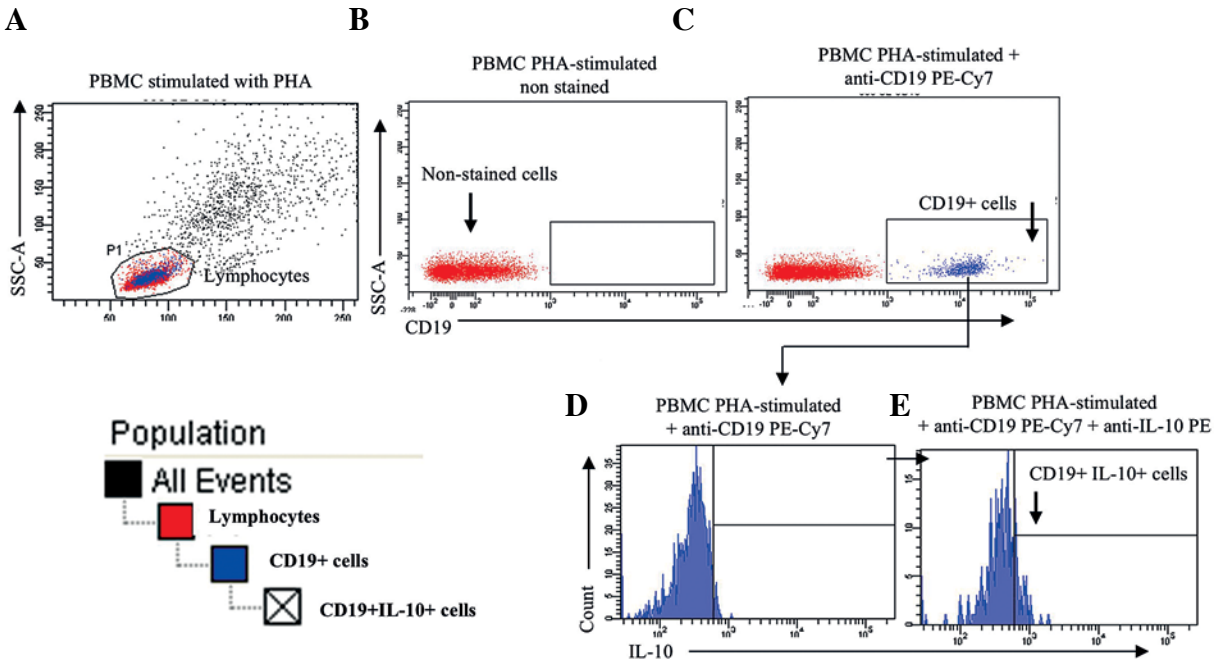


Supplementary Fig. 1. Gating strategy to assess $CD19^{+}CD24^{+}CD27^{+}$ and $CD19^{+}CD24^{+}CD38^{+}$ B cell subpopulations. **A**) Events acquired by the flow cytometer are plotted in a scatter diagram using complexity-granularity (SSC-A) versus size (FSC-A), and the lymphocyte population is selected with a gate (population in red). **B**) A dot plot gated on the lymphocyte population is generated using specific channels to read Pacific-Blue versus PE-Cy7-A fluorochromes. Because no antibodies were added here, the cells appear negative for both markers (unstained cells). A gate is defined around this population, and this gate remains fixed for subsequent analyses. **C**) After adding anti-human CD19-PE-Cy7 antibody, two populations are identified within the lymphocyte's population: one positive for CD19 (lower right) and another negative for this marker (lower left), suggesting that these are lymphocytes that do not express the CD19 antigen. **D**) Cells to which anti-human CD19 antibody and anti-human CD24 antibody are added; four different populations are observed: 1) lymphocytes positive for CD24 and negative for CD19 (upper left); 2) lymphocytes positive for both CD19 and CD24 (upper right); 3) lymphocytes negative for both CD19 and CD24 (lower left); 4) lymphocytes negative for CD24 and positive for CD19 (lower right). In this example, CD27 or CD38 expression is assessed in the $CD19^{+}CD24^{+}$ lymphocyte subpopulation. Subsequently, a dot plot gated to this population (blue) is generated using Pacific-Blue (CD24) vs. PE-Cy7 (CD19) parameters. Since these cells were not labeled with anti-human CD27 or CD38 antibodies, they are considered negative for these markers. Based on this population, a negative gate is defined for **E**) CD27 or **F**) CD38, and this gate remains fixed for subsequent analyses. **G**) When cells are stained with anti-human CD19, anti-human CD24, and anti-human CD27 antibodies, two populations are distinguished: one positive for CD19 and CD24 but negative for CD27 (blue), and another positive for all three markers CD19, CD24, and CD27 (green). **H**) Similarly, when cells are stained with anti-human CD19, anti-human CD24, and anti-human CD38 antibodies, two populations are identified: one positive for CD19 and CD24 but negative for CD38 (blue), and another positive for all three markers CD19, CD24, and CD38 (purple). Additionally, a hierarchical gating strategy is shown. Here, we can observe the parent and child gates, which allows us to determine to which B cell subpopulation ($CD19^{+}CD24^{+}CD27^{+}$ or $CD19^{+}CD24^{+}CD38^{+}$) a given cell belongs



Supplementary Fig. 2. Gating strategy to assess CD19⁺IL-10⁺ B cells. PBMC were stimulated for 18 hours with phytohemagglutinin (PHA), as described in the Material and Methods section. **A**) The events acquired using the FACS Canto II are analyzed with a scatter plot based on complexity-granularity (SSC-A) vs. size (FSC-A). The lymphocyte population is selected using a gate (population in red). **B**) A dot plot gated on the lymphocyte population is generated using the channels for PE-Cy7-A vs. SSC-A. Since no antibodies were added to these cells, a population negative for both markers is observed (unstained cells). Based on this population, a gate is established, which remains fixed for subsequent analyses. **C**) When cells are stained which anti-human CD19-PE-Cy7 antibody, two populations are identified within the lymphocyte population: one positive for CD19 (population in blue) and another negative for this marker (population in red), suggesting that the latter consists of lymphocytes that do not express the CD19 antigen. **D**) From the CD19⁺ population, a histogram is generated using the specific channel to detect phycoerythrin (PE) fluorochrome. IL-10 expression is assessed in the CD19⁺ lymphocyte subpopulation. Since these cells were not labeled with anti-human IL-10-PE antibodies, they are considered negative for this marker (as shown in the histogram). Based on this population, a negative IL-10 gate is defined, which remains fixed for subsequent analyses. **E**) When cells are stained with both anti-human CD19 and anti-human IL-10 antibodies, two populations are distinguished: one positive for both CD19 and IL-10, and another positive for CD19 but negative for IL-10. The CD19⁺IL-10⁺ population is indicated. In addition, a hierarchical gating strategy is shown. Here, we can observe the parent and child gates, which allows us to determine to which cell subpopulation the CD19⁺IL-10⁺ B cells belong