19. The role of toll-like receptor-4 (TLR-4) activation in a murine model of systemic lupus erythematosus (SLE).

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Systemic lupus erythematosus (SLE) is an autoimmune disease in which individuals develop autoantibodies to multiple self-antigens, including phospholipid-binding proteins (e.g., □2-glycoprotein I [□2GPI]). Anti-□2GPI autoantibodies are among the earliest autoantibodies detected in SLE.

Our laboratory has developed a murine model of SLE in which mice immunized with □2GPI and lipopolysaccharide (LPS) produce high levels of SLE autoantibodies and develop SLE-like glomerulonephritis.

We hypothesize that LPS (a toll-like receptor 4 [TLR4] agonist) activates antigen-presenting cells (APCs), particularly B cells and dendritic cells (DCs), resulting in enhanced presentation of □2GPI to T cells and a strong □2GPI-specific T cell response. Using an in vitro antigen presentation assay, we found that □2GPI presentation by C57BL/6 (wild type [WT]) splenocytes increased in a dose-dependent manner with LPS stimulation. In contrast, TLR-4-deficient splenocytes were not able to present □2GPI, even in the presence of LPS. Splenocytes from mice deficient in either MyD88 or TRIF, the signaling molecules triggered by TLR-4, were also defective in □2GPI presentation. To investigate the effect of LPS on APCs, we evaluated the expression of co-stimulatory (CD80, CD86, and CD40) and MHC II molecules on LPS-treated and untreated splenocytes from WT and deficient mice by flow cytometry. Co-stimulatory molecule and MHC II expression was increased on both CD19+ (B cells) and CD11c+ (dendritic cells [DCs]) in LPS-stimulated splenocytes from WT mice, compared to unstimulated controls. In contrast, LPS-stimulated CD19+ and CD11c+ cells from TLR-4-deficient mice showed no increase in co-stimulatory molecule and MHC II expression, while MyD88- and TRIF-deficient cells were intermediate between WT and TLR-4 deficient cells. To determine which APC was most important in □2GPI presentation, we isolated splenic CD19+ (B cells) and CD11c+ (DCs) using antibody-coated magnetic beads. Although isolated CD19+ and CD11c+ cells showed similar increases in co-stimulatory molecule and MHC II expression, they differed significantly in their ability to present □2GPI. Naive B cells could not present □2GPI to T cells, while DCs were highly effective APCs.

We propose that initial presentation of □2GPI is enhanced by TLR-4 activation of DCs, resulting in a strong T cell response to □2GPI and the subsequent emergence of multiple SLE autoantibodies.