

Poster

Natural inhibitors aid in studying LPS-mediated cytokine release

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Introduction. Ancient protective mechanisms are in place as our first line of defense against infections. The innate immune response rapidly eliminates the vast majority of invading microorganisms (bacteria, fungi, viruses, etc.) within minutes or hours of recognition by Toll-like receptors (TLR's). TLR's are type I transmembrane proteins found on the surface of mammalian cells. These receptors have high specificity, but limited heterogeneity i.e., little is known of their complex signaling chain reaction. Lipopolysaccharide (LPS) is a prime component of gram-negative bacteria and is used in this study to activate TLR-4 response. With the help of CD14 and MD-2, TLR-4 activation upon LPS engagement begins a series of intracellular changes in the host cell, ultimately leading to cytokine release. Kinases are proteins that transfer phosphate groups to their substrates thereby triggering intracellular signaling cascades. Their roles in cytokine release upon stimulus are as yet uncharacterized. Through a series of Enzyme-Linked Immunosorbent Assays (ELISA's), we are studying LPS-mediated cytokine release in THP-1 cells (human monocytic cell line). Employing natural inhibitors to specific intracellular proteins (kinases), we are attempting to map kinase signaling cascades that mediate this release in these cells.

Objectives. To study cytokine release using natural inhibitors.

Methods. Using Enzyme Linked Immunosorbent Assays (ELISA'S), and western blots.

Results. Polymyxin inhibits IL1beta, IL6, IL8, IL10, MCP1, and TNFalpha release. Herbamycin has a slight effect on IL10 and TNFalpha. SB203580 also has an effect on IL13 and TNFalpha.

Conclusion. A more defined model of upstream and downstream signaling pathway will be studied. Effects of inhibiting the expression of SYK at the RNA level will also be investigated.

Keywords: TLR-4, THP-1 Cells, innate immune, Enzyme Linked

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