

P019 A role for TLR4 ectodomain in controlled LPS-induced signal transduction in mammalian cells

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Innate immunity as the first line of host defense against microbial invasion relies on pattern recognition receptors (PRRs) to detect different types of pathogen associated molecular patterns.

Toll-like receptor 4 (TLR4) is a PRR that responds to bacterial lipopolysaccharide (LPS) together with its accessory proteins MD-2 and CD14. TLR4 as a type I transmembrane glycoprotein transduces the signal across the cell membrane. MD-2, bound to the extracellular domain of TLR4, is the actual LPS-binding protein and is therefore indispensable for LPS signaling, while the GPI-anchored protein CD14 augments the LPS response by presenting LPS molecules to the MD-2 or TLR4-MD-2 complex. In order to gain insight whether the role of ectodomain of TLR4 serves primarily as a scaffold for the LPS bound MD-2, we tested the effect of replacement of the TLR4 ectodomain with two LPS binding proteins MD-2 and CD14. Both fusion constructs, consisting of MD-2 or CD14 fused to the transmembrane and intracellular domains of TLR4 imparted constitutive activity and showed no LPS responsiveness. Thus, the important role of ectodomain of TLR4 is its negative regulation by inhibition of the constitutive activity of the receptor, while the precise positioning of the intracellular TIR domain is probably not essential for activation.