

P014 MARCKS-lipopolysaccharide interaction in cells and its effect on LPS sequestration and signalling
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Proteins of the MARCKS (myristoylated alanine-rich C kinase substrate) family are intrinsically unfolded and contain a conserved core peptide of basic residues. Transcription of MARCKS is significantly increased on stimulation by bacterial lipopolysaccharide (LPS). We determined that MARCKS and MARCKS-related protein bind directly to LPS. Intracellular interaction between MARCKS and internalized LPS was detected by FRET in human epithelial cells. MARCKS was localized to endosomes in the presence of LPS. Production of cytokines by monocytes was inhibited by the addition of effector domain peptide to LPS. LPS binding site of MARCKS was further narrowed down to the heptapeptide KRFSFKK, which neutralized the LPS signalling. Inhibition of MARCKS expression with siRNA increased the LPS signalling and cytokine production. MEFs from MARCKS knock out mouse also had an increased response to LPS in comparison to the matched wild type MEFs. These findings suggest that direct interaction between MARCKS, which participates in several signal transduction pathways and LPS may be part of the negative regulation of cellular response and sequestration of the internalized LPS.