

**Induction of inducible nitric oxide synthase in RAW  
264.7 macrophages by lipoteichoic acid derived from  
Staphylococcus aureus: involvement of protein kinase  
C- and nuclear factor-kB-dependent mechanisms.**

林建煌,許準榕

Kuo CS;Chiang LL;Lee CN;Yu MT;Pei KZ;Lee HM;Lee WS;Sheu JR and Lin CH

**Abstract**

This study investigates the signaling pathway involved in inducible nitric oxide synthase (iNOS) expression and nitric oxide (NO) release caused by *Staphylococcus aureus* lipoteichoic acid (LTA) in RAW 264.7 macrophages. A phosphatidylcholine-phospholipase C (PC-PLC) inhibitor (D-609) and a phosphatidylinositol-phospholipase C (PI-PLC) inhibitor (U-73122) attenuated LTA-induced iNOS expression and NO release. Two PKC inhibitors (Go 6976 and Ro 31-8220), an NF- $\kappa$ B inhibitor (pyrrolidine dithiocarbamate; PDTC), and long-term (24 h) 12-phorbol-13-myristate acetate (PMA) treatment each also inhibited LTA-induced iNOS expression and NO release. Treatment of cells with LTA caused an increase in PKC activity; this stimulatory effect was inhibited by D-609, U-73122, or Ro 31-8220. Stimulation of cells with LTA caused IB- phosphorylation and IB- degradation in the cytosol, and translocation of p65 and p50 NF- $\kappa$ B from the cytosol to the nucleus. Treatment of cells with LTA caused NF- $\kappa$ B activation by detecting the formation of NF- $\kappa$ B-specific DNA-protein complexes in the nucleus; this effect was inhibited by Go 6976, Ro 31-8220, long-term PMA treatment, PDTC, L-1-tosylamido-2-phenylethyl chloromethyl ketone (TPCK), and calpain inhibitor I. These results suggest that LTA might activate PC-PLC and PI-PLC to induce PKC activation, which in turn initiates NF- $\kappa$ B activation, and finally induces iNOS expression and NO release in RAW 264.7 macrophages.