Dipyridamole 抑制 RAW 264.7 巨噬細胞中脂多醣體誘導的一氧化氮合成酶;與環氧酵素-2 的表現

Dipyridamole Inhibits Lipopolysaccharide-Induced Inducible Nitric Synthase and Cyclooxygenase-2 Expression in RAW 264.7 Cells

中文摘要

Dipyridamole 是一個核苷運送的抑制劑，也是一個非選擇性的 phosphodiesterase 的抑制劑，因此能夠藉由抑制 phosphodiesterase 的機制來增加細胞內 cAMP 以及 cGMP 的濃度。第四型的 phosphodiesterase 抑制劑已經在許多實驗中被證實具有抗發炎的功能，本研究所要探討的主題就是，在 RAW 264.7 巨噬細胞中 Dipyridamole 是否可以抑制 Lipopolysaccharide (LPS) 誘導的 iNOS 以及 COX-2 的表現。以 LPS 處理 RAW 264.7 巨噬細胞會造成 iNOS 以及 COX-2 以劑量依存性及時間依存性表現。若以 Dipyridamole 前處理細胞則可以阻斷 LPS 所誘導的 iNOS 及 COX-2 表現。藉由抑制 I B phosphorylation、degradation、p65 NF- B translocation 以及 reporter gene 的轉錄作用的方式來證明 Dipyridamole 會抑制 NF- B 路徑的活化。另外，Dipyridamole 也可以抑制 LPS 在 RAW 264.7 細胞中所造成的 p38 MAPK 以及 IKK- 的活化。若進一步以 p38 MAPK 的抑制劑 SB203580 前處理細胞，則能抑制 LPS 誘導的 iNOS 表現以及 IKK- 活化，所以 LPS 是先活化了 p38 MAPK，再活化 NF- B 的訊息傳遞路徑。另外，Dipyridamole 能夠刺激 mitogen-activated protein kinase phosphatase 1 (MKP-1) 的磷酸化及活化而使得 p38 MAPK 去磷酸化及去活化而失去功能。總而言之，本研究證明在 RAW 264.7 巨噬細胞中，Dipyridamole 會先藉由活化 MKP-1 的方式使得 p38 MAPK 去磷酸化而失去功能。然而 p38 MAPK 去活化後，接著就會抑制 IKK- 的活化以及後續由 NF- B 所調控的訊息傳遞路徑，因而抑制 LPS 所誘導的 iNOS 及 COX-2 表現。本研究的結果支持 dipyridamole 具抗發炎作用的假說。

英文摘要

Dipyridamole is a nucleoside transport inhibitor and a non-selective phosphodiesterase inhibitor that increases intracellular level of cAMP and cGMP through phosphodiesterase inhibition. Type 4 phosphodiesterase has been demonstrated to have anti-inflammatory effects in many experimental systems. This study investigates whether dipyridamole inhibits lipopolysaccharide (LPS)-induced inducible nitric oxide (iNOS) and cyclooxygenase (COX-2) expression in RAW 264.7 macrophages. Treatment of RAW 264.7 macrophages with LPS caused dose- and time-dependent increases in iNOS and COX-2 expression. Treatment of cells with dipyridamole blocked the LPS-induced iNOS and COX-2 expression. Dipyridamole

inhibited NF- B activation as demonstrated by inhibition of I B phosphorylation, I B degradation, p65 NF- B translocation and the transcription of reporter gene. Dipyridamole also inhibited LPS-stimulated p38 MAPK and IKK- activities in RAW 264.7 cells. A p38 mitogen-activated protein kinase (MAPK) inhibitor, SB203580, inhibited LPS-stimulated iNOS expression and IKK- activation, suggesting LPS may activate NF- B signaling pathway via upstream p38 MAPK activation. Furthermore, dipyridamole stimulated a transient activation of mitogen-activated protein kinase phosphatase 1 (MKP-1), a potent inhibitor of p38 MAPK. Taken together, these data suggest that dipyridamole exerts anti-inflammatory effect via activation of MKP-1, which dephosphorylates and inactivates p38 MAPK. Inactivation of p38 MAPK in turn inhibits IKK- activation and subsequent NF- B signaling pathway that mediates LPS-induced iNOS and COX-2 expression in RAW 264.7 cells. These results support the notion that dipyridamole may have anti-inflammatory effects.