乙型樣澱粉蛋白經由 **IKK/FKHR/Bim?徑誘導 C6** 星??經膠質瘤細胞凋亡之探討

β-Amyloid Induced C6 Glioma Cell Apoptosis via IKK/FKHR/Bim Pathway

中文摘要

乙型樣澱粉蛋白（β-Amyloid, Aβ）被認爲是引起許多?經退化性疾病的主要原因，而 Aβ和星?細胞的交互作用對?經細胞產生的傷害會?進一步促進?經的退化。此外，星?細胞又是構成血腦障壁的主要成分之一，因此星?細胞凋亡對於中樞?經系統以及腦血管退化之病?發展過程是非常重要的。在本?文中，我們將探討 Aβ調控星??經膠質瘤細胞凋亡的詳細分子機轉。在 C6 星??經膠質瘤細胞中，Aβ誘導增加 BimEL 的表現而?是 BimL 和 BimS。Aβ也可誘導增加 Bim 報告基因的活性。?用轉染 FKHR 結合序?突變型 Bim 報告基因質體可減少 Aβ誘導之 Bim 報告基因的活性。轉染野生型 FKHR oligodeoxynucleotides 能抑制 Aβ所誘導之 Bim 的表現和 C6 星??經膠質瘤細胞凋亡。Aβ誘導 FKHR 之 Ser256 的去磷酸化呈現時間相關性，並且藉由 DNA-binding affinity pull down assay 證實 Aβ可以誘導 FKHR 結合至 bim 基因起始區上。轉染野生型及持續活化型 IKKβ質體可抑制 Aβ誘導 FKHR 去磷酸化、Bim 的表現以及 C6 星??經膠質瘤細胞凋亡。Aβ也會時間相關性地誘導 IKKα/β Ser180/Ser181 的去磷酸化，並且藉由蛋白磷酸激?活性的測試顯示 Aβ可?低 IKKα/β的活性。此外，在 C6 星??經膠質瘤細胞中，Aβ可誘導 IKKα/β、FKHR 以及 14-3-3 的分?。PP2A 的抑制劑 okadaic acid 可以阻斷 Aβ誘導的 IKK 去磷酸化、FKHR 去磷酸化、Bim 的表現以及 C6 細胞的死亡。再者，我們發現 Aβ可誘導 PP2A 的活性增加。綜合以上實驗結果推測 Aβ可經由 PP2A/IKK/FKHR/Bim 訊息?徑誘導 C6 星??經膠質瘤細胞死亡。

英文摘要

β-Amyloid peptide (Aβ) has been implicated as a key molecule in the neurodegenerative diseases. The Aβ-astrocyte interaction produces a detrimental effect on neurons, which may contribute to neurodegeneration. Astrocyte is a cellular component of blood-brain barrier, thus the regulation of astrocyte apoptosis plays a causal role in pathological processes in the CNS and cerebrovascular degeneration. This study was designed to investigate the mechanism of Aβ-induced C6 glioma cell apoptosis. Aβ induced an increase in BimEL, but not BimL and BimS, expression in C6 glioma cells. Aβ also caused an increase in Bim-luciferase activity, which was reduced by transfection with the mutation of forkhead transcription factor (FKHR) site in Bim-luciferase reporter construct. Transfection with the wild type FKHR oligodeoxynucleotides inhibited Aβ-induced BimEL expression and C6 glioma cell apoptosis. Treatment of C6 glioma cells with Aβ induced FKHR dephosphorylation at Ser256 in a time-dependent manner. Aβ induced an increase in FKHR binding to the bim promoter by DNA-binding affinity pull down assay. Furthermore, transfection with the plasmids of wild type IKKβ and constitutively active IKKβ reversed Aβ-induced FKHR dephosphorylation, Bim expression, and C6 glioma cell apoptosis. Aβ also induced IKKα/β dephosphorylation at Ser180/Ser181 and reduced IKKα/β activity in a time-dependent manner. In addition, Aβ induced the dissociation among IKKα/β, FKHR, and 14-3-3 in C6 glioma cells. Okadaic acid, a potent PP2A inhibitor, inhibited Aβ-induced IKKα/β dephosphorylation, FKHR dephosphorylation, Bim expression, and C6 cell apoptosis. Furthermore, Aβ induced an increase in protein phosphatase 2A activity. Taken together, these results suggest that the mechanism of Aβ-induced C6 cell apoptosis involves PP2A activation, IKKα/β dephosphorylation, FKHR activation, and Bim expression.