Econazole 誘發人類直腸腫瘤細胞 (COLO 205) 細胞週期 G0/G1 時期停滯及細胞凋亡之分子機制研究

Studies on the Molecular Mechanisms of Econazole induced G0/G1 Cell Cycle Arrest and Apoptosis in Human Colon Adenocarcinoma Cells (COLO 205)

中文摘要

Econazole 是一種用來治療全身性黴菌感染症的抗黴菌藥物，屬於 imidazole 類之衍生物。本研究證實 Econazole 可誘使人類直腸腫瘤細胞株 (COLO 205) 發生細胞週期 G0/G1 時期停滯和細胞凋亡現象。在 COLO 205 細胞中，以不同濃度的 Econazole 處理細胞 24 小時後有明顯抑制 COLO 205 細胞生長的情形，此外，以較低劑量之 Econazole (<30uM) 處理下可觀察到 COLO 205 細胞發生細胞週期 G0/G1 時期停滯的現象，而以較高劑量之 Econazole (>40uM) 處理下則可明顯觀察到 COLO 205 細胞發生細胞凋亡的現象。在 Econazole 調控之 COLO 205 細胞細胞週期 G0/G1 時期停滯現象中，以西方墨點法觀察到 p53、p21/Cip1 和 p27/Kip1 的蛋白表現增加，CDK4 和 CDK2 的蛋白表現減少，使得 cyclin D3-CDK4 和 cyclin E-CDK2 複合物的活性受抑制，導致細胞週期停滯於 G0/G1 時期。在 Econazole 調控之 COLO 205 細胞發生細胞凋亡現象中，p53 和 Bax 的蛋白表現增加，Bcl-2 的蛋白表現有些微被抑制的情形，此外，觀察到 Bax 從細胞質間轉移至粒線體上，cytochrome c 和 AIF 從粒線體中釋出且分別轉移至細胞質間和細胞核內，以及 caspase-9 和 caspase-3 有被活化的情形，可見 Econazole 誘使 COLO 205 細胞走向粒線體細胞凋亡路徑，且此細胞凋亡現象的發生同時包含了 caspase-dependent 和 caspase-independent 之調控機制。在裸鼠實驗方面，也證實 Econazole 有抑制 COLO 205 細胞誘導之異種移植腫瘤生長的情形，所以在 in vitro 和 in vivo 實驗之結果皆顯示 Econazole 可抑制人類直腸腫瘤細胞 (COLO 205) 的生長。故本論文提出 Econazole 可誘使人類直腸腫瘤細胞株 (COLO 205) 生長停滯於細胞週期 G0/G1 時期和細胞凋亡現象的發生。

英文摘要

Econazole is an antifungal agent that has been used in the treatment of systemic fungal infection. In this study, we demonstrated that Econazole induced human colon adenocarcinoma cells cell growth arrest in the G0/G1 phase and apoptosis. Our results indicated that the growth inhibition of COLO 205 cells by Econazole with dose dependently. It is also can be observed COLO 205 cells were arrested in G0/G1 phase with low Econazole dose treatment (<30uM), however, induced apoptosis with high Econazole dosage (>40uM). In detail, Econazole (30uM)-treated G0/G1 phase arrest

in COLO 205 cells, p53, p21/Cip1 and p27/Kip1 protein levels had a significantly increase but cyclin D3, CDK4 and CDK2 expressed inhibited phenomenon. As consequence, cyclin D3-CDK4 and cyclin E-CDK2 complexes activity was inhibited and thus led to cell cycle arrest in G0/G1 phase. On induction of apoptosis by Econazole (60uM) in COLO 205 cells, the protein levels of p53 and Bax were increased but Bcl-2 was decreased, moreover, Bax proteins targeted to mitochondrial membranes. Therefore, cytochrome c and AIF were released from mitochondria into the cytoplasm and furthermore AIF was translocated to the nucleus. As result, caspase-9 and caspase-3 were activated and thus occurred to apoptosis. Clearly, Econazole-induced apoptosis in COLO 205 cells is part of mitochondrial apoptotic pathway, as well as, it includes both caspase-dependent and caspase-independent apoptotic fashion. In nude mice experiment, the growth of COLO 205 tumor xenografts was significantly reduced by Econazole treatment. Taken together, it is indicated that Econazole induced human colon adenocarcinoma cells cell growth arrest in the G0/G1 phase and apoptosis.