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桔梗皂苷D诱导人肺癌细胞A549的凋亡及机制

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中文摘要:目的:研究桔梗皂苷D(platycodin,PD)抑制人肺癌A549细胞株增殖和诱导凋亡的分子机制。 方法:体外培养人肺癌细胞株A549,PD作用终浓度分别为5~20 μmol·L^{-1}。MTT法测定PD对细胞的增殖抑制作用,显微镜观察细胞的形态学变化,Annexin V-FITC/PI双标法检测细胞凋亡率,JC-1检测线粒体膜电位的变化,Western blot方法检测PD对Caspase-3,Caspase-9,PARP的剪切片段和Bax,Bcl-2,Bak,Bcl-xl蛋白表达的影响。 结果:PD抑制A549细胞的增殖,并随药物浓度的增加和作用时间的延长,作用更显著;与对照组相比,不同浓度的PD作用24 h后,细胞凋亡率增加,线粒体膜电位降低,且差异显著。蛋白电泳检测结果显示蛋白Caspase-3,Caspase-9均出现剪切片断,并随着时间的增加断裂更明显。PD处理A549细胞后,Bax和Bak蛋白表达升高,Bcl-2和Bcl-xl蛋白表达下降。 结论:PD具有明显的细胞毒作用,能诱导A549细胞凋亡,通过对Bax,Bak和Bcl-2,Bcl-xl表达的调控,导致线粒体膜电位的下降,进而激活Caspase最终导致肺癌细胞死亡。

中文关键词:桔梗皂苷D A549 线粒体膜电位 细胞凋亡

Mechanism of platycodin D-induced humane long cancer cells A549 apoptosis

Abstract:Objective: To investigate the molecular mechanism of platycodin D showing the inhibitory effect on proliferation and induced apoptosis of humane long cancer cells A549. **Method:** Humane long cancer cells A549 were cultured *in vitro*, with the final PD concentration of 5-20 μmol·L^{-1}. PD's inhibitory effect on cell proliferation was examined by MTT assay. Morphological changes in cells were observed with microscope. The cell apoptosis rate was detected by Annexin V-FITC/PI double staining. The change of mitochondrial membrane potential was detected by JC-1. The protein expressing of leaved Caspase-3, cleaved Caspase-9, cleaved PARP, Bcl-2, Bcl-xl, Bak and Bax were detected by Western blot analysis. **Result:** PD could inhibit the proliferation of A549 cells and show stronger effect with the increase of concentration and over time. Compared with the control group, PDs of different concentration showed significant increase in the cell apoptosis rate, decrease in mitochondrial membrane potential after 24 h. Protein electrophoresis inspection showed cut segments in both protein Caspase-3 and Caspase-9 and notable fractures with time. Further study found that PD decreased Bcl-2, Bcl-xl proteins and increased Bax, Bak proteins after processing A549 cells. **Conclusion:** PD shows notable effect on cytotoxicity and can induce A549 cell apoptosis. It causes decrease in mitochondrial membrane potential by regulating Bax, Bak, Bcl-2 and Bcl-xl expressions, and thus activating caspase and finally causing long cancer cell apoptosis.

keywords:platycodin D A549 mitochondrial membrane potential apoptosis

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