

## 缺氧对骨肉瘤细胞系MG-63 HIF-1 $\alpha$ 、p53、bcl-2及细胞凋亡的影响

蔡文涛<sup>1\*</sup>,陈安民<sup>1</sup>,郭风劲<sup>1</sup>,管 频<sup>2</sup>

1. 430030 华中科技大学同济医学院附属同济医院骨科(\* 现工作单位:570311 海南省人民医院骨病外科);2. 海南省人民医院

### Effect of Hypoxia on the Expression of Hypoxia Inducible Factor 1 $\alpha$ 、p53、bcl-2 and Apoptosis in Osteosarcoma Cell Line MG-63

CAI Wen-tao<sup>1\*</sup>, CHEN An-min<sup>1</sup>, GUO Feng-jin<sup>1</sup>, GUAN Pin<sup>2</sup>

1. Department of Orthopaedic, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan 430030, China (\*Present: Hainan Provincial People's Hospital, Haikou 570311); 2. Hainan Provincial People's Hospital

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管 频

**摘要** 目的 了解缺氧环境对骨肉瘤MG-63细胞缺氧诱导因子HIF-1 $\alpha$ 、p53、bcl-2的表达及细胞凋亡的影响。方法 建立骨肉瘤细胞体外缺氧模型,观察不同缺氧时间段(8、16、24h)HIF-1 $\alpha$ 、p53、bcl-2的表达和细胞凋亡的情况。半定量RT-PCR方法检测HIF-1 $\alpha$ 、p53、bcl-2的表达水平;免疫组化(SP法)和免疫印迹(WesternBlot)检测HIF-1 $\alpha$ 、p53、bcl-2蛋白表达情况;流式细胞仪检测细胞凋亡率。结果 缺氧处理后,HIF-1 $\alpha$ 转录水平未见明显改变,蛋白表达水平随缺氧时间延长明显增强;而p53、bcl-2mRNA及蛋白表达水平均显著增强,两者间存在一定的相关性;细胞凋亡率却未见明显增加。结论 在缺氧条件下,不能通过以HIF-1 $\alpha$ 为中介的p53依赖途径来诱导骨肉瘤MG-63细胞的凋亡,其机制可能与缺氧诱导野生型p53的突变或缺失使HIF-1 $\alpha$ 和bcl-2的过表达有关。

**关键词:** 骨肉瘤 缺氧诱导因子-1 $\alpha$  p53 bcl-2 细胞凋亡

**Abstract:** Objective To investigate the effect of hypoxia on the expression of hypoxia inducible factor 1 $\alpha$  (HIF-1 $\alpha$ )、p53、bcl-2 and apoptosis in osteosarcoma cell line MG-63 under hypoxia environment. Methods The hypoxia culture model was established by a hypoxia incubator. The gene productions of HIF-1 $\alpha$ 、p53、bcl-2 and the changes of the apoptosis were observed at different hypoxia culture phase. The semi-quantitative reverse transcription PCR(RT-PCR) was used to test the mRNA expression of HIF-1 $\alpha$ 、p53 and bcl-2. The protein level of HIF-1 $\alpha$ 、p53 and bcl-2 was observed by immunohistochemical staining and western blot analysis. The changes of apoptosis were analyzed using flow cytometry. Results After the hypoxia treatment, the mRNA level of HIF-1 $\alpha$  was not changed significantly, however, the protein expression of HIF-1 $\alpha$  was increased remarkably with correspondence to the hypoxia time. But the expression of p53 and bcl-2 were up-regulated in mRNA and protein level. Besides, the bcl-2 activity was markedly associated with the level of HIF-1 $\alpha$ . However, the apoptosis rate was not increased markedly. Conclusion In hypoxia environment, the apoptosis of osteosarcoma cell line MG-63 could not induce by the p53 avenue which depended on HIF-1 $\alpha$ . The mechanism is possibly related with the mutation of p53 that increases the expression of HIF-1 $\alpha$  and bcl-2.

**Key words:** Osteosarcoma Hypoxia inducible factor-1 $\alpha$  p53 bcl-2 Apoptosis

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