[1]陈娟娟,艾剑刚,黄世峰,等.HBx介导miR-221上调-ERα下调致HepG2细胞恶性增殖[J].第三军医大学学报,2013,35(20):2191-2194.

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Title: HBx protein-induced up-regulation of miR-221 promotes HepG2 cell malignant proliferation by targeting ERα

作者: 陈娟娟; 艾剑刚; 黄世峰; 曹炬; 张莉萍
重庆医科大学附属第一医院检验科

Author(s): Chen Juanjuan; Ai Jiangang; Huang Shifeng; Cao Ju ; Zhang Liping
Department of Clinical Laboratory, First Affiliated Hospital, Chongqing Medical University, Chongqing, 400016, China

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摘要: 目的 探讨microRNA221(miR-221)在乙型肝炎病毒（hepatitis B virus, HBV）感染相关性肝细胞癌（hepatocellular carcinoma, HCC）中的促癌功能以及HBV基因编码的X蛋白（HBx）诱导miR-221上调促进HepG2细胞异常增殖的分子机制。 方法 重组HBx腺病毒（Ad-HBx）感染人肝癌HepG2细胞，荧光定量PCR检测HepG2-HBx细胞中miR-221和ERα mRNA表达的变化；流式细胞术检测细胞周期，Western blot检测雌激素受体α（estrogen receptor α, ERα)蛋白表达水平变化，分别用miR-221 mimic和miR-221 inhibitor 转染HepG2细胞后，荧光定量PCR检测HepG2-HBx细胞中miR-221和ERα mRNA表达变化；Western blot检测ERα蛋白水平表达变化。 结果 RT-PCR实验证实，adv-HBx感染HepG2细胞后，HBx在HepG2细胞中高效表达；感染48 h后，HBx蛋白可显著上调miR-221[（495.84±61.16）*vs*（239.25±

21.15），*P*<0.05]并抑制ERα蛋白[（0.24±0.01）*vs*（0.61±0.02），*P*<0.05]的表达水平，同时促进HepG2细胞异常增殖[（31.73±3.53）% *vs*（56.08±1.56）%，*P*=0.01]。miRNA转染实验及Western blot证实：miR-221抑制ERα蛋白的表达（*P*<0.05)，miR-221抑制剂促进ERα蛋白的表达（*P*<0.05)。　　结论　　HBx可能通过上调miR-221进而下调ERα对肝癌的保护性效应而促进肝癌细胞异常增殖，靶向miR-221的策略具有抑制肝癌细胞增殖的治疗潜能。

Abstract: Objective To investigate the mechanisms that microRNA221 (miR-221) promotes cancer development in hepatitis B virus (HBV)-related hepatocellular carcinoma (HCC) as well as HBx protein promotes HepG2 cells aberrant proliferation through up-regulation of miR-221. Methods Recombinant adenovirus with HBx gene (adv-HBx) was used to transfect HepG2 cells. The changes of miR-221 and estrogen receptor α (ERα) mRNA were validated by real-time fluorescence quantitative PCR, and the protein levels of ERα were quantified by Western blotting. Cell cycle was detected by flow cytometry. miR-221 mimic and miR-221 inhibitor were used to transfect HepG2 cells respectively. The changes of miR-221 and ERα mRNA in HBx-HepG2 cells were determined by real-time fluorescence quantitative PCR, and ERα protein levels were quantified by Western blotting. Results RT-PCR results