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Title: Effect of silencing PTP4A1 gene by lentiviral vector-mediated siRNA on biological behavior of TCA8113 cells *in vivo* and *in vitro*

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关键词: TCA8113; PTP4A1; siRNA; 生物学行为

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摘要: 目的 探讨特异性沉默人舌鳞癌细胞系TCA8113 中PTP4A1基因的表达，并研究沉默该基因表达后对TCA8113细胞体内外生物学行为的影响。 方法 采用siRNA 的慢病毒载体siRNA-PTP4A1作为阳性干扰组（KD），空病毒载体作为阴性对照组(NC)，未作任何处理的TCA8113细胞作为空白对照组(CON)。Real-time PCR和Western blot检测PTP4A1基因的干扰效果，MTT法检测3组细胞的增殖能力，流式细胞技术分析3组细胞凋亡和细胞周期情况，平板克隆形成实验检测体外克隆形成能力，Western blot分别检测3组细胞抑制凋亡蛋白Bcl-2、促凋亡蛋白Bax的表达情况，3组细胞分别裸鼠成瘤后观察移植瘤的生长情况。 结果 siRNA-PTPA1转染TCA8113细胞成功，Real-time PCR和Western blot检测PTP4A1基因的抑制率达到55%和60%以上（P<0.05）；MTT结果显示KD组细胞增殖能力小于NC、CON组；细胞凋亡率也明显增加，KD组早期凋亡为（17.48±0.70）%，而NC、CON组的早期细胞凋亡率为（7.34±0.70）%和（4.86±0.25）%(P<0.01)；KD组细胞周期阻滞在G_1/G_0和S期(P<0.05)；Western blot结果显示Bcl-2蛋白表达降低(P<0.01)，Bax蛋白表达增高(P<0.01)；KD组体外克隆形成数小于NC、CON组(P<0.01)；KD组裸鼠移植瘤的生长速度缓慢，质量和体积均小于NC、CON组(P<0.01)。 结论 siRNA PTP4A1在体内外均能有效抑制舌鳞癌TCA8113细胞

的恶性生物学行为，PTP4A1基因可能与舌鳞癌的发生、发展相关。

Abstract: Objective To study the PTP4A1 gene expression silenced by lentiviral vector-mediated siRNA and the effect on biological behavior in human tongue squamous cancer cell line TCA8113. Methods The cells were divided into three groups including a KD group (TCA8113 cells treated with siRNA-PTP4A1), a NC group (TCA8113 cells treated with empty lentivirus), and a CON group (TCA8113 cells without treatment). Real-time PCR and Western blotting were applied to detect the expression of PTP4A1. MTT assay was used to detect the inhibition of cell proliferation. Flow cytometry was performed to detect the cell apoptosis and cell cycle. Bcl-2 and Bax protein expression levels were detected by Western blotting. Clone formation was measured by plate clone forming test. Cells were subcutaneously injected into nude mice to induce tumors. Results In the KD group, the inhibitory rates of mRNA and protein of PTP4A1 were above 55% and 60% (P<0.05). The cell proliferation was significantly suppressed in the KD group compared with the NC group and CON group. The cell apoptotic rate in the KD group [(17.48$\pm$0.70)%] was significantly higher than that in the NC group [(7.34$\pm$0.70)%] and CON group [(4.86$\pm$0.25)%] (P<0.01), and the cells in the KD group were arrested at G_1/G_0 phase and S phase (P<0.05). Western blot results showed that Bcl-2 protein expression level decreased (P<0.01), but Bax protein expression level increased (P<0.01). Compared with the NC group and CON group, the clone formation ability of the cells *in vitro* (P<0.01) and the tumor growth in nude mice *in vivo* decreased significantly in the KD group (P<0.01). Conclusion Silencing PTP4A1 expression is effective in inhibiting tumor biological behavior in human tongue squamous cancer cell line TCA8113 *in vivo* and *in vitro*. PTP4A1 may play an important role in the development of human tongue squamous cell carcinoma.

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