以核糖核酸干擾抑制作用來降低轉型生長因子β1 在老鼠結腸癌細胞的表現來抑制腫瘤細胞在老鼠體內的生長與轉移

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摘要

在罹患腫瘤的病患中，轉型生長因子β 被視為降低病患對抗腫瘤免疫反應的重要因子，本研究之目的是要以核糖核酸干擾抑制技術來降低結腸癌細胞內轉型生長因子β1 的表現，藉此達成抑制結腸癌細胞在小白鼠體內的生長與轉移。我們所設計的轉型生長因子β1 寡核苷酸能抑制結腸癌細胞（小白鼠 CT26 細胞株）轉型生長因子β1 的表現，利用核糖核酸干擾抑制技術來降低結腸癌細胞內轉型生長因子β1 的產生，可以抑制此腫瘤細胞在小白鼠皮下的生長與存活比率，而且腫瘤內有明顯的 CD4+及 CD8+T 細胞浸潤，微血管的密度明顯降低，這個發現有助於未來結腸腫瘤基因治療的應用。

關鍵字：核糖核酸干擾抑制，轉型生長因子β1，結腸癌細胞

Knockdown of Transforming Growth Factor-Beta1 Expression by RNA Interference Inhibits Colon Carcinoma growth in Immunocompetent Mice

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Abstract

Objectives: Transforming growth factor-beta (TGF-β) is the key molecule implicated in impaired immune function in human patients with colorectal carcinoma. TGF-β can promote tumor growth, invasion and metastasis in advanced stages of colorectal cancer. Blocking these tumor-promoting effects of TGF-β provides a potentially important therapeutic strategy for the treatment of colorectal cancer. We examined the effects of TGF-β1 protein knockdown by RNA interference on the growth of murine colon carcinoma cells in syngeneic Balb/c mice induced by the CT26 cell line.

Materials and Methods: The TGF-β1 hairpin oligonucleotide was cloned into retroviral vector pSM2. The resulting plasmid (TGF-β1-RNAi/pSM2) was stably introduced into murine colon carcinoma cells, CT26, and designated as CT26/TGF-β1-RNAi cells. The vector plasmid was transfected into CT26 cells and designed as CT26/vector-control cells served as a control. The growth rate of the parental cell and genetically modified murine colon carcinoma cells were compared. Balb/c mice were evaluated for survival rate in an experimental metastasis model following tail vein injection.

Results: TGF-β1 expression was reduced in CT26/TGF-β1-RNAi cells compared with CT26 cells and CT26/vector-control cells. The proliferation rate of CT26/TGF-β1-RNAi cells was similar to that of the CT26 cells and CT26/vector-control cells in vitro. The tumor sizes were 807.17 ± 139.01 mm3, 823.35 ± 113.66 mm3 and 115.28 ± 16.89 mm3 at the 21 day in the mice receiving CT26 cells, CT26/vector-control cells and CT26/TGF-β1-RNAi cells respectively. The p value was less than 0.05 by one-way ANOVA. The survival rate was prolonged significantly in mice injected with CT26/TGF-β1-RNAi cells. TGF-β1 knockdown in CT26 cells enhanced the infiltration of CD4+ and CD8+ T cells in the tumor regions. The blood vessel density of the tumors markedly reduced in CT26/TGF β1-RNAi tumors.

Conclusions: We found that TGF-β1 protein expression was significantly reduced from CT26 cells after TGF-β1 hairpin oligonucleotide transduction. Silencing of TGF-β1 expression in CT26 cells by RNA interference technology can inhibit the growth and metastasis of this tumor after being transplanted to Balb/c mice. The purpose of this work is to provide preclinical assessment of the therapeutic potential of TGF-β1 protein knockdown by RNA interference in colon carcinoma.

Keywords：RNA interference, TGF-β1, colon carcinoma