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研究报告

# 特异小干扰RNA敲除PLK1基因的表达

陈功星1, 2, 董庆华1, 张佳炜1, 符芳芳1, 许则丰1, 梁巧仪1, 郑 树1, 丁佳逸1

（1.[1]

浙江大学第二附属医院肿瘤研究所，杭州 310009[2]

2.浙江新和成股份有限公司，新昌 312500）[3]

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摘要 为研究特异小干扰RNA（siRNA）作用于大肠癌细胞株SW480中PLK1 (Polo-like kinase 1)基因表达的mRNA对该细胞分裂生长的影响，设计了对应于PLK1基因表达mRNA不同位点的10种特异siRNA，经化学合成后，用脂质体转染SW480细胞，实时定量PCR检测PLK1基因的表达，观察不同的siRNA作用强度，并计数细胞了解相应细胞的生长情况，western-blot观察PLK1表达蛋白的变化和流式细胞计数分析细胞周期改变。发现10种siRNA均可敲除PLK1基因表达的20 %以上，其中P1、P4和P9 3组敲除mRNA达80 %以上，这3种siRNA及其混合物对PLK1基因mRNA的作用具有相应浓度效应，在25 nmol/L时达到最佳作用效果，而且相同浓度的混合物作用效果更好（超过95%），PLK1表达蛋白质明显降低，细胞周期在G2期受到阻碍。72 h后的各种siRNA浓度下细胞生长变化与PLK1基因的mRNA水平变化相一致。结果表明化学合成的特异siRNA对SW480细胞中PLK1基因表达具有消除作用，混合物作用更强，在细胞水平上抑制了SW480细胞的分裂生长。

关键词 小干扰RNA； PLK1； 基因表达

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## Knockdown of PLK1 mRNA by Special siRNA

CHEN Gong-Xing1, 2, DONG Qing-Hua1, ZHANG Jia-Wei1, FU Fang-Fang1,XU Ze-Feng1, LIANG Qiao-Yi1, ZHENG Shu1, DING Jia-Yi1

(1.Cancer Institute, The 2nd Affiliated Hospital, Medical college, Zhejiang University, Hangzhou 310009, China; 2. Zhejiang NHU Limited Corporation, Xingchang 312500, China)

**Abstract**

To study knockdown effect of small interfering RNA (siRNA) to PLK1 (Polo-like kinase 1) mRNA in colorectal cancer cell line SW480 and its mitosis and growth was changed. Ten special siRNA molecules were designed targeting different sites of PLK1 mRNA sequence and chemically synthesized. The siRNA molecules were transfected into SW480 by Oligofectamine. The gene mRNA level was assayed by Real-Time PCR. The changes of PLK1 protein, SW480 cell cycle and survival percentage was checked by Western-blot, Flow cytometry and Cell counter assays respectively. All 10 siRNA molecules knocked PLK1 mRNA down in different level. Of them P1, P4 and P9 showed over 80% knockdown efficiency and the others had more than 20% knockdown efficiency to PLK1 mRNA. The best knockdown effect over 95% of all groups was at 25 nmol/L of a mixture with P1, P4 and P9 siRNA equally. In this situation the protein was very less and the cells were blocked at G2 phase of cell cycle. After 72 h cell survival percentages were consistent with PKL1 mRNA level change by siRNA gradient concentration. The results showed that siRNA targeting PLK1 mRNA had effectively knocked PLK1 mRNA down in SW480 cell line. And a blended siRNAs held the best knockdown effect. The cell was blocked on the mitosis and growth.

**Key words** small interfering RNA Polo-like kinase 1 gene expression

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通讯作者 丁佳逸 DJY@zju.edu.cn