实验研究报道

转染Smad 7基因的大鼠肾小球系膜细胞对Ⅰ、Ⅲ型胶原表达的改 变

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摘要

[摘要]目的 探讨Smad 7基因对大鼠肾小球系膜细胞(MsC)Ⅰ、Ⅲ型胶原(ColⅠ、ColⅢ)表达的 影响,为试图运用Smad 7对肾纤维化进行基因治疗提供实验依据。方法 经脂质体介导将含有Smad 7重 组表达质粒转染大鼠MsC,用G418筛选及Northern blot、Western blot法鉴定;又分别采用RT-PCR和Western blot法,检测转染阳性MsC克隆Col I、Col Ⅲ表达改变。结果 成功建立稳定高表达 Smad 7的阳性MsC克隆(S-22与S-26),并证实两阳性MsC克隆Col I 及Col Ⅲ mRNA及蛋白的表达 均被明显抑制, 其中S-22克隆Col I 及Col III mRNA表达分别降低47%和56%, 其蛋白表达分别降低 65 %和54 %。结论 Smad 7可能通过抑制组织内Col I 及Col Ⅲ的生成而起到减轻肾纤维化进展的作 用。

关键词 系膜细胞 Smad 7 Ⅰ型胶原 Ⅲ型胶原

分类号

Changes of Type I, III collagen expression in cultured rat glomerular mesangial cells transfected with Smad 7 cDNA

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Abstract

Objective To investigate the changes of type I, III collagen (Col I, Col III) expression on cultured rat glomerular mesangial cells (MsC) transfected with Smad 7 cDNA in order to provide experimental proofs for blocking renal fibrosis with Smad 7 gene therapy. Methods Lipofectin method was used to transfect Smad 7 cDNA into MsC and transfected cells were selected with G418, and detected Smad 7 mRNA and protein expression by Northern blot

and Western blot analysis, respectively. The expression of type I, III collagen on MsC wereetermined by reverse transcriptase-polymerase chain reaction (RT-PCR) and Western blot

analysis. Results Overexpresion Smad 7 on two MsC clones (S-22, S-26) were successfully established. Two MsC clones showed decreased expression of Col I, Col III mRNA and their proteins. The level of Col I, Col III mRNA expression on S-22 clone decreased 47 % and 56 % respectively, however the level of their protein expression decreased 65 % and 54 % respectively. Conclusion It is suggested that Smad 7 could alleviate the progression of renal fibrosis by down-regulating the expression of Col I and Col III at mRNA and protein level.

Key words mesangial cell Smad 7 gene type I collagen type III collagen

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