转录因子E2F1对急性单核细胞白血病新抗原基因MLAA-34的转录调控作用

《现代肿瘤医学》[ISSN:1672-4992/CN:61-1415/R] **期数:** 2019年20期 **页码:** 3560-3565 **栏目:** 论著（基础研究） **出版日期:** 2019-09-08

Title: Effects of transcription factor E2F1 on transcriptive regulation of acute monocytic leukemia -related gene MLAA-34

作者: 雷 博; 张王刚; 何爱丽; 陈银霞; 曹星梅; 张鹏宇; 赵万红; 王剑利; 刘 捷; 马肖容; 张彦平; 刘海玲
西安交通大学第二附属医院血液科，陕西 西安 710004

Author(s): Lei Bo; Zhang Wanggang; He Aili; Chen Yinxia; Cao Xingmei; Zhang Pengyu; Zhao Wanhong; Wang Jianli; Liu Jie; Ma Xiaorong; Zhang Yanping; Liu Hailing
Department of Hematology，The Second Affiliated Hospital，Medical School of Xi'an Jiaotong University，Shaanxi Xi'an 710004，China.

关键词: 急性单核细胞白血病; MLAA-34; E2F1; 转录调控

Keywords: acute monocytic leukemia; MLAA-34; E2F1; transcriptional regulation

分类号: R733.71

DOI: 10.3969/j.issn.1672-4992.2019.20.004

文献标识码: A

摘要: 目的：探讨转录因子E2F1对急性单核细胞白血病新抗原基因MLAA-34的转录调控作用。方法：利用双荧光素酶报告基因检测系统及定点突变技术分析E2F1 对MLAA-34基因启动子转录活性的影响。通过凝胶迁移实验（EMSA）和染色质免疫共沉淀（ChIP）实验，验证E2F1是否与MLAA-34启动子核心区直接特异性结合。构建E2F1真核表达载体和干涉载体，转染U937细胞，RT-PCR 和Western Blot检测MLAA-34基因的转录和表达变化。结果：转录因子E2F1对MLAA-34基因表达具有调控作用，E2F1结合序列点突变后，相对荧光素酶活性升高（P<0.01），绿色荧光蛋白的表达增高。EMSA和ChIP实验，从细胞内、外水平分别证明E2F1可与MLAA-34启动子直接结合而发挥调控作用。在过表达试验中，E2F1的增加可下调MLAA-34的表达（P<0.05）；在干涉试验中，E2F1的降低可上调MLAA-34的表达（P<0.05）。结论：转录因子E2F1可与MLAA-34基因启动子上的转录调控区结合，并抑制急性单核细胞白血病细胞中MLAA-34基因的转录。

Abstract: Objective：To investigate the transcriptional regulation of transcription factor E2F1 on acute monocytic leukemia-related gene MLAA-34.Methods：The effect of E2F1 on the transcriptional activity of MLAA-34 gene promoter was analyzed by luciferase reporter gene detection system and site-directed mutation technique.EMSA and ChIP assay were used to verify whether E2F1 directly and specifically binds to the core region of MLAA-34 promoter.The over-expression vector and interference vector of E2F1 were constructed to transfect U937 cells，and RT-PCR and Western Blot were used to detect the transcription and expression changes of MLAA-34 gene.Results：The transcription factor E2F1 had a regulatory effect on MLAA-34 gene expression，and the relative luciferase activity was increased after E2F1 binding point mutation (P<0.01).EMSA and ChIP experiments demonstrated that E2F1 can directly bind to MLAA-34 promoter and play a regulatory role.In the over-expression test，the increase of E2F1 can down-regulate the expression of MLAA-34 (P<0.05).In the interference test，the decrease of E2F1 can up-regulate the expression of MLAA-34 (P<0.05).Conclusion：Transcription factor E2F1 can bind to the transcriptional regulatory region on the promoter of MLAA-34 gene and inhibit the transcription of MLAA gene in acute monocytic leukemia.

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备注/Memo: National Natural Science Foundation of China(No.8153000580）；国家自然科学基金资助(编号：8153000580)

更新日期/Last Update: 1900-01-01