miR-106a负调控PTEN促进骨肉瘤细胞增殖并抑制凋亡

《现代肿瘤医学》[ISSN:1672-4992/CN:61-1415/R] 期数: 2019年18期 **页码:** 3186-3190 **栏目:** 论著（基础研究） **出版日期:** 2019-08-08

Title: **miR-106a promoted osteosarcoma cell proliferation and inhibited apoptosis via negatively regulating the expression of PTEN**

作者: 曾桂平[1]; 文昌明[1]; 王 聪[2]; 方 忠[3]

1湖北省阳新县人民医院骨二科；2手术室，湖北 阳新 435200;3华中科技大学同济医学院附属同济医院骨科,湖北 武汉 430030

Author(s): Zeng Guiping[1]; Wen Changming[1]; Wang Cong[2]; Fang Zhong[3]

1.Second Department of Orthopaedics;2.Operation room,Yangxin People's Hospital,Hubei Yangxin 435200,China;3.Department of Orthopaedics,Tongji Hospital Affiliated to Tongji Medical College,Huazhong University of Science and Technology,Hubei Wuhan 430030，China.

关键词: miR-106a; 同源性磷酸酶-张力蛋白; 骨肉瘤; 治疗靶点

Keywords: miR-106a; phosphataseand tensin homolog; osteosarcoma; therapeutic target

分类号: R738.1

DOI: 10.3969/j.issn.1672-4992.2019.18.005

文献标识码: A

摘要: 目的:研究miR-106a在骨肉瘤组织和MG-63细胞中的表达水平及其对MG-63细胞增殖和凋亡的影响及机制。方法:用荧光实时定量PCR法检测20对骨肉瘤和相邻正常组织及MG-63和成骨细胞hFOB 1.19中miR-106a的表达。用miR-106a mimics、miR-106a antagomir 及两者相应的对照物转染MG-63细胞，然后分别用CCK-8法检测四组细胞增殖活性和FCM法检测细胞凋亡率。miR-106a mimics和mimics control与野生型或突变型PTEN 3'-UTR 重组载体共转染后，应用荧光素酶基因报告系统检测miR-106a是否与PTEN基因3'-UTR 结合。利用Western blot技术检测PTEN蛋白在上述四组转染MG-63细胞和骨肉瘤标本中的表达水平。结果:与相邻正常组织（1.19±0.15）相比，肿瘤组织miR-106a的表达水平（2.60±0.86）显著升高；同时，miR-106a在MG-63中的表达水平（2.60±0.92）明显高于hFOB 1.19（1.19±0.39），以上差异均有统计学意义（P < 0.05）。CCK-8和FCM检测结果显示，与mimics control组相比，miR-106a mimics组的增殖率明显增加，而细胞凋亡率下降；反之，miR-106a antagomir组与antagomir control组相比，增殖率前者低于后者，而凋亡率前者高于后者，上述差异均有统计学意义（P < 0.05）。荧光素酶报告实验显示，miR-106a mimics和wt PTEN 3'-UTR共转染组的荧光强度值明显低于mimics control和wt PTEN 3'-UTR组（P < 0.05）。Western blot发现，与对照组相比，miR-106a mimics组PTEN表达下调，而miR-106a antagomir表达上调；临床标本，肿瘤组织PTEN表达明显低于正常组织，差异均有统计学意义（P < 0.05）。结论:miR-106a在骨肉瘤组织及细胞中过表达，并靶向负调控PTEN表达，促进骨肉瘤细胞增殖并抑制其凋亡，从而发挥促癌作用。因此，miR-106a可为骨肉瘤的诊治提供新的潜在分子靶点。

Abstract: Objective:To study the expression level of miR-106a in osteosarcoma and MG-63 cells and its effect on proliferation and apoptosis of MG-63 cells and the underlying mechanism.Methods:The expression level of miR-106a in 20 pairs of osteosarcoma and adjacent normal tissues,MG-63 and osteoblasts hFOB 1.19 was detected by real-time quantitative PCR.MG-63 cells were transfected with miR-106a mimics,mimics control,miR-106a antagomir and antagomir control,respectively.Then the cell viability of four groups was detected by CCK-8 and their apoptosis rate was detected by FCM.After co-transfection of miR-106a mimics and mimics control with wild or mutant PTEN 3'-UTR recombinant vectors,the luciferase gene report system was used to detect whether miR-106a was combined with the 3'-UTR of gene PTEN.The expression level of PTEN protein in the four groups of MG-63 cells transfected and osteosarcoma samples was detected by Western blot.Results:Compared with the adjacent normal tissues （1.19±0.15）,the relative expression level of miR-106a in tumor tissues （2.60±0.86） was significantly higher.Meanwhile,the expression level of miR-106a in MG-63（2.60±0.92） was significantly higher than that of hFOB 1.19 （1.19±0.39）,and the above differences were statistically significant (P <

0.05).The results of CCK-8 and FCM assay showed that compared with the mimics control group,the proliferation rate of miR-106a mimics group increased significantly,while its apoptosis rate decreased.On the contrary,the proliferation rate in miR-106a antagomir groupwas obviously lower than that of antagomir control group,and its apoptosis rate was higher than that of the latter,and the all difference was statistically significant (P < 0.05).The fluorescence intensity of miR-106a mimics and wt PTEN 3'-UTR co-transfection group was significantly lower than that of mimics control and wt PTEN 3'-UTR group (P < 0.05).Western blot found that compared with the corresponding control group,the protein expression of PTEN in the miR-106a mimics group was significantly down-regulated,but the expression of PTEN in miR-106a antagomir was up-regulated,and the expression of PTEN in the tumor tissue was significantly lower than that of the adjacent normal tissue,and all difference was statistically significant (P < 0.05).Conclusion:miR-106a was overexpressed in the osteosarcoma tissues and MG-63 cells and negatively regulated the expression of PTEN,which promotes the proliferation of osteosarcoma cells and inhibits its apoptosis,and thus may play an important role in promotingtumorigenesis.Therefore,miR-106a likely becomes a new potential molecular target for the diagnosis and treatment of osteosarcoma.

参考文献/REFERENCES

[1] Mens MMJ,Ghanbari M.Cell cycle regulation of stem cells by microRNAs [J] .Stem Cell Rev,2018,3(14):309-322.
[2] Qin Y,Huo Z,Song X,et al.miR-106a regulates cell proliferation and apoptosis of colon cancer cells through targeting the PTEN/PI3K/AKT signaling pathway [J] .Oncology Letters,2018,3(15):3197-3201.
[3] Xie X,Liu HT,Mei J,et al.miR-106a promotes growth and metastasis of non-small cell lung cancer by targeting PTEN [J] .International Journal of Clinical and Experimental Pathology,2015,4(8):3827-3834.
[4] Luo B,Kang N,Chen Y,et al.Oncogene miR-106a promotes proliferation and metastasis of prostate cancer cells by directly targeting PTEN in vivo and in vitro [J] .Minerva Medica,2018,1(109):24-30.
[5] Li A,Qiu M,Zhou H,et al.PTEN,insulin resistance and cancer [J] .Current Pharmaceutical Design,2017,25(23):3667-3676.
[6] Eichelmann AK,Matuszcak C,Hummel R,et al.Role of miRNAs in cell signaling of cancer associated fibroblasts [J] .International Journal of Biochemistry and Cell Biology,2018 (101) :94-102.
[7] Wang Z,Zhao Z,Yang Y,et al.miR-99b-5p and miR-203a-3p function as tumor suppressors by targeting IGF-1R in gastric cancer [J] .Scientific Reports,2018,1(8):10119.
[8] Zheng Y,Sun Y,Liu Y,et al.The miR-31-SOX10 axis regulates tumor growth and chemotherapy resistance of melanoma via PI3K/AKT pathway [J] .Biochemical and Biophysical Research Communications,2018,6(15):175-182.
[9] Zhang W,Chen JH,Shan T,et al.miR-137 is a tumor suppressor in endometrial cancer and is repressed by DNA hypermethylation [J] .Laboratory Investigation,2018,6(28):1-11.
[10] Group E.SMO/European Sarcoma Network Working.Bone sarcomas:ESMO Clinical Practice Guidelines for diagnosis,treatment and follow-up [J] .Annals of Oncology,2014(25 Suppl 3):iii113-123.
[11] Zhu M,Zhang N,Lu X,et al.Negative regulation of kruppel-Like factor 4 on microRNA-106a at upstream transcriptional level and the role in gastric cancer metastasis [J] .Digestive Diseases and Sciences,2018,31(5):172-179.
[12] Hao H,Xia G,Wang C,et al.miR-106a suppresses tumor cells death in colorectal cancer through targeting ATG7 [J] .Medical Molecular Morphology,2017,2(50):76-85.
[13] Namlos HM,Meza-Zepeda LA,Baroy T,et al.Modulation of the osteosarcoma expression phenotype by microRNAs [J] .PloS One,2012,10(7):e48086.
[14] Zhu M,Zhang N,He S,et al.MicroRNA-106a functions as an oncogene in human gastric cancer and contributes to proliferation and metastasis in vitro and in vivo [J] .Clinical and Experimental Metastasis,2016,5(33):509-519.
[15] Pan YJ,Zhuang Y,Zheng JN,et al.miR-106a:Promising biomarker for cancer [J] .Bioorganic and Medicinal Chemistry Letters,2016,22(26):5373-5377.
[16] Huang Q,Ma Q.MicroRNA-106a inhibits cell proliferation and induces apoptosis in colorectal cancer cells [J] .Oncology Letters,2018,6(15):8941-8944.
[17] Haddadi N,Lin Y,Travis G,et al.PTEN/PTENP1:' Regulating the regulator of RTK-dependent PI3K/Akt signalling',new targets for cancer therapy [J] .Molecular Cancer,2018,1(17):37-46.

备注/Memo: -

更新日期/Last Update: 1900-01-01