

[1]黄帆,刘映孜,杨秋珺,等.全反式维甲酸增强骨形态蛋白9诱导间充质干细胞成骨分化作用[J].第三军医大学学报,2013,35(04):328-331.

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## 全反式维甲酸增强骨形态蛋白9诱导间充质干细胞成骨分化作用

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**Title:** All-trans retinoic acid enhances BMP9-induced osteogenic differentiation in mesenchymal stem cells

**作者:** 黄帆; 刘映孜; 杨秋珺; 周龙洋; 周岐新; 罗进勇; 何百成; 邓忠良  
重庆医科大学附属第二医院骨科; 重庆医科大学:药理教研室, 检验学院

**Author(s):** Huang Fan; Liu Yingzi; Yang Qiujun; Zhou Longyang; Zhou Qixin; Luo Jinyong; He Baicheng; Deng Zhongliang

Department of Orthopaedics, Second Affiliated Hospital, Chongqing Medical University, Chongqing, 400010; Department of Pharmacology, College of Laboratory Medicine, Chongqing Medical University, Chongqing, 400016, China

**关键词:** 全反式维甲酸; 骨形态蛋白9; 间充质干细胞; Smad信号

**Keywords:** all-trans retinoic acid; bone morphogenetic protein; mesenchymal stem cell; Smad signaling

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**摘要:** 目的 研究全反式维甲酸 (all-trans retinoic acid, ATRA) 对骨形态蛋白9 (bone morphogenetic protein 9, BMP9) 诱导间充质干细胞成骨分化的影响及可能机制。 方法 采用组织化学染色检测各组第5、7天碱性磷酸酶活性变化。Western blot检测各组第9、11天骨桥素蛋白表达水平。茜素红染色检测各组第14、20天钙盐沉积水平。采用Western blot检测各组Smad-1/5/8蛋白磷酸化水平, 以及用荧光素酶报告质粒检测BMPR-Smad信号通路的活化程度。 结果 ATRA及BMP9均诱导C3H10T1/2细胞碱性磷酸酶活性升高, 但ATRA合并BMP9组碱性磷酸酶活性明显强于单用ATRA或BMP9组; BMP9合并ATRA组骨桥素蛋白表达水平高于BMP9组; BMP9合并ATRA较单用BMP9能明显促进钙盐沉积。BMP9合并ATRA组Smad1/5/8磷酸化水平明显强于单用BMP9组, BMPR-Smad报告质粒荧光素酶活性也呈相同变化趋势 ( $P<0.01$ )。 结论 ATRA能增强BMP9诱导间充质干细胞成骨分化的作用, 其机制可能与促进Smads信号转导活性有关。

**Abstract:** Objective To investigate the effect of all-trans retinoic acid (ATRA) on the osteogenesis induced by bone morphogenetic protein 9 (BMP9) in mesenchymal stem cells (MSCs). Methods Histochemical staining was used to test the

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alkaline phosphatase (ALP) activities when C3H10T1/2 cells were treated with BMP9 and/or ATRA for 5 or 7 d. Then, the protein level of osteopontin in the C3H10T1/2 cells after treatment of BMP9 and/or ATRA for 9 or 11 d was detected by Western blotting. The mineralization in the cells were tested with Alizarin red staining after the treatment for 14 or 20 d. Finally, the activation of BMP-Smad signaling was studied with BMPR-Smad binding site luciferase reporter assay and Western blotting to determine the phosphorylation level of Smad1/5/8.

**Results** The activities of ALP were increased by BMP9 and/or ATRA, but the ALP activities in the cells treated by BMP9 and ATRA are more pronounced than that the cells by BMP9 or ATRA alone. ATRA potentiated the osteopontin expression and mineralization induced by BMP9 in C3H10T1/2 cells. ATRA promoted the phosphorylation level of Smad-1/5/8 and increased the luciferase activity of BMPR-Smad luciferase reporter. **Conclusion** ATRA potentiates the osteogenesis induced by BMP9 in MSCs, which may be mediated by increase of the BMP-Smad signaling transduction.

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