

基础研究

体外诱导人骨髓间充质干细胞定向分化为心肌细胞实验方法的建立

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摘要:

目的:探讨体外培养并诱导人骨髓间充质干细胞(hMSCs)分化为心肌细胞(CMs)的实验方法。方法:采用密度梯度离心法联合贴壁培养法分离、纯化hMSCs,采用5-氮杂胞苷(5-Aza)和碱性成纤维细胞生长因子(bFGF)对hMSCs进行联合诱导,诱导3~4周,观察细胞形态学的变化,免疫组织化学方法鉴定肌细胞及心肌细胞特异蛋白的表达。结果:hMSCs体外经5-Aza和bFGF联合诱导1~2周后,细胞体积较诱导前增大,呈长梭形。诱导2~3周后细胞伸出伪足呈多爪形;诱导3~4周后细胞伪足间相互连接形成肌管样结构。联合诱导3~4周后免疫组织化学鉴定结蛋白(Desmin)、肌钙蛋白(cTn I)和横纹肌肌动蛋白( $\alpha$ -sarcomeric actin)表达均呈阳性。结论:hMSCs在体外经5-Aza和bFGF联合诱导可以分化为CMs。

关键词: 间质干细胞;肌细胞 心脏;5-氮胞苷

Establishment of |method to induce human bone mesenchymal stem |cells to differentiate into cardiomyocytes in vitro

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Abstract:

Abstract: Objective

To establish the method to induce human bone mesenchymal stem cells(hMSCs) to differentiate into cardiomyocytes in vitro. Methods hMSCs were isolated and purified from the bone marrow of human by density gradient centrifugation and adhering to the culture dishes. The fourth passage of MSCs was induced by 5-azacytidine(5-Aza) and bFGF for four weeks. The expression of Desmin, Troponin I and  $\alpha$ -sarcomeric actin were detected by immunocytochemistry 3-4 weeks after induction. Results hMSCs treated with 5-Aza and bFGF had long cytoplasmic process one week after induction, and had multiple branches two weeks after induction, and they connected with adjoining cells forming myotube-like structures four weeks after induction. The induced hMSCs were stained positively for Desmin, Troponin I and  $\alpha$ -sarcomeric actin. Conclusion 5-Aza and bFGF can induce hMSCs to differentiate into cardiomyocytes in vitro.

Keywords: mesenchymal stem cells; myocytes, cardiac; 5-azacytidine

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