



应用Solexa技术检测脐血CD34⁺细胞体外诱导的巨核细胞mRNA表达

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Analysis of mRNA Expression Profiles of Megakaryocytes from Human Cord Blood CD34⁺ Cells Ex Vivo Expanded Using Solexa Sequencing

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摘要

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摘要 目的 应用Solexa技术研究人脐带血CD34⁺细胞体外诱导巨核细胞的mRNA表达。**方法** 采用密度梯度离心法和免疫磁珠分选系统获取人脐带血CD34⁺细胞。100 ng/ml 促血小板生成素诱导培养12 d后,应用抗CD41⁺单克隆抗体免疫磁珠法分选获得巨核细胞和非巨核细胞。应用Solexa技术检测巨核细胞和非巨核细胞的mRNA表达差异。**结果** 获取的Tag初始数量巨核细胞和非巨核细胞分别为3-773-147和3-533-805个。整个清晰的Tag数量分别为3291132和2967947个,明显独特的tag数量分别为197769和245-318个。其中上调基因表达数量为1161个,下调为902个。上调Tag表达数量为2717个,下调为1519个。**结论** 巨核细胞和非巨核细胞 mRNA表达存在显著差异,差异基因编码产物与细胞发育、黏附、凋亡、代谢、细胞内及细胞间的信号转导以及免疫应答等功能相关,进一步深入将有助于探讨巨核细胞的表达机制、信号传导及调控机制。

关键词: 巨核细胞 mRNA表达 体外扩增 Solexa测序

Abstract: Objective To investigate the mRNA expression profiles of megakaryocytes (MKs) from human cord blood CD34⁺ cells *ex vivo* expanded using Solexa technique. Methods CD34⁺ Cells were isolated using density gradient centrifugation and magnetic activated cell sorting. Cultures were stimulated with recombinant human thrombopoietin (100 ng/ml). After 12 days, the MKs fraction was separated from the non-MKs fraction using an anti-CD41 monoclonal antibody by immunomagnetic sorting. The mRNA expression of MKs and non-MKs was detected by Solexa sequencing. Results We obtained 3773147 and 3533805 Tags from MKs and non-MKs, respectively. The amounts of unambiguous tags were 3291132 and 2967947 and those of distinct tags were 197769 and 245318. The expression of 1161 genes was up-regulated and that of 902 genes down-regulated. The expression of 2717 tags was up-regulated and that of 1519 tags down-regulated. Conclusions MKs and non-MKs have remarkably different mRNA expression profiles. The differential gene-encoded products may be involved in cellular development, adhesion, apoptosis metabolism, intra- and inter-cellular signal transduction, and immune response. Further studies on this topic may clarify the expression mechanism, signal transduction, and regulation mechanisms.

Keywords: megakaryocyte mRNA expression; *ex vivo* expansion Solexa sequencing

Received 2010-10-29;

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引用本文:

王芳,何吉,朱发明,刘晋辉,秦斐,陈舒,徐罡,吕行军,严力行.应用Solexa技术检测脐血CD34⁺细胞体外诱导的巨核细胞mRNA表达[J] 中国医学科学院学报, 2011,V33(5): 529-532

WANG Fang, HE Ji, ZHU Fa-ming, LIU Jin-hui, QIN Fei, CHEN Shu, XU Gang, Lv Xing-jun, YAN Li-xing. Analysis of mRNA Expression Profiles of Megakaryocytes from Human Cord Blood CD34⁺ Cells Ex Vivo Expanded Using Solexa Sequencing[J] CAMS. 2011.V33(5): 529-532

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