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# 兔骨髓间充质干细胞的Gd-DTPA标记及MRI体外示踪

# Gadolinium and fluorescent bi-functionally labeling and in vitro MR imaging of rabbit bone marrow mesenchymal stem cells

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中文摘要：

目的 探讨兔MSCs磁性标记及MRI体外示踪的可行性。方法 培养分离兔MSCs，以PEI-FluoR为载体，体外对MSCs进行双标记。标记后行荧光镜、电镜观察及生物学性状检测。应用1.5T MRI仪，对标记细胞进行SE序列T1WI及T2WI扫描及T1-mapping测量T1时间，并观察MRI上能显示标记MSCs的最小数目及标记后正常传代后MRI监测的持久性。结果 MSCs双标记后，标记效率为80%，细胞内可见荧光物质，电镜下Gd颗粒位于胞浆内。标记后24 h内标记细胞与未标记细胞间台盼蓝拒染率、标记后5 d内标记细胞与未标记细胞的MTT吸光度值差异无统计学意义。标记细胞凋亡指数为0.40%，未标记细胞为0.19%。未标记及标记细胞T1WI平均信号强度及T1时间分别为2166±167、(2445±21)ms,3162±350、(1404±129)ms ($t$=6.91、29.87，$P$<0.005)。体外MRI上可监测到最低$1\times10^4$个标记细胞，并可持续显示第3代标记细胞。结论 应用多聚胺载体对兔MSCs进行Gd-DTPA及荧光双标记安全、有效；MRI能示踪体外双标记的干细胞。

英文摘要：

**Objective** To determine the feasibility of magnetical labeling and tracking rabbit mesenchymal stem cells (MSCs) in vitro. **Methods** MSCs isolated from rabbit bone marrow were cultured and expanded. A bi-functional labeling reagent complex was obtained by the incubation linear polyethylenimine derivative (PEI-FluoR) with Gd-DTPA to label the MSCs bi-functionally. After labeling, the MSCs were examined under fluoroscope and electron microscope, and the biological characters were detected using trypan blue exclusion test, MTT and apoptosis detection. On a 1.5Tesla MR system, the labeled MSCs were examined with spin echo T1WI, T2WI and T1-mapping measurement. The minimal amount of labeled MSCs that could be detected by MRI was determined. The labeling duration was determined in the labeled MSCs under routine passage. **Results** With the bi-functional agents, the efficacy of labeling MSCs was 80%. After labeling, red fluorescence was found in the cytoplasm of MSCs under fluorescence microscopy and higher density electron particles of gadolinium were found around cellular apparatuses under electron microscopy. There was no statistical difference of trypan blue excusion rate between labeled cells and unlabeled cells within 24 hours of incubation ($F$=2.17-1.38, $P$>0.05), nor in the MTT proliferation test between labeled cells and unlabeled cells within 5 days after labeling ($F$=1.93-0.61, $P$>0.05). The apoptosis index for labeled cells and unlabeled cells was 0.40% and 0.19%, respectively. The signal intensity on T1WI and T1 relaxation time of unlabeled cells and labeled cells were 2166±167 and (2445±21)ms, 3162±350 and (1404±129)ms, respectively ($t$=6.91, 29.87, $P$<0.005). The minimal detectable amount of labeled cells was $1\times10^4$. After routine passage, MRI could detect labeled cells until the third passage. **Conclusion** The gadolinium and fluorescent bi-functionally labeling rabbit MSCs using the transfection agent of polyethylenimine is feasible, efficient and safe. The labeled cells can be tracked in vitro on MR imaging.

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