

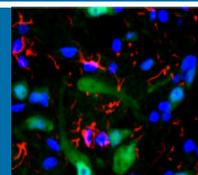
Recognition of neural stem cells derived from hippocampus of tree shrew in vitro

Gui-Qin Huang*

Department of anatomy, kunming medical university, kunming 650500

[Abstract] Objective: To explore and establish a new strategy to culture neural stem cells (NSCs) of hippocampus derived from tree shrew, and lay a theoretical foundation for the treatment of nerve diseases by using tree shrew hippocampal NSCs transplantation. **Methods:** The hippocampal tissues were harvested from embryonic tree shrew, then the tissues were made into monocellular suspension by cell screener. The density of cell suspension was adjusted at optional value, then cells were cultured under suitable condition. The specific antibody was used to identify the NSCs. Moreover, 1, 3, 5, 7d in the culture stage were selected to observe the NSCs morphology in P1, P2 and P3 passage, respectively. Enzyme histochemical staining was employed to identify the capacity of differentiation of NSCs. And immunofluorescence staining was used to identify the differentiation ability of NSC. **Results:** The morphological observation showed that cells exhibited monocellular suspension in 24-hole culture plate, and parts of them began to be adherence and showed ball in shape at 1h after incubation. NSCs became to adherence, and some small cell spheres formed by 3 to 5 cell mass at 1d. More neurospheres were found with increased volume at 3d. The number of neurospheres was getting more with more larger volume at 5d. On 7d, the volume of neurospheres reaches the peak, and NSCs began to migrate from the neurospheres and erupted the processes. The results showed that parts of NSCs showed Neun positive, with coloring in neuclei, and part of them showed GFAP labeled, with coloring in cytoplasm, and part of them showed O4 positive, with coloring in neuclei, which confirmed the differentiation of NSC. To verify the differentiation ability of NSC, neuronal early marker-Tuj-1 and GFAP were utilized, and the results showed that cultured NSC had the capacity to differentiate into immature neurons and mature astrocyte. **Conclusion:** The results suggested that tree shrew NSCs have strong proliferative and differentiation capacity, which provided useful morphological evidence for subsequent basic research and transformation application.

Key words: Neural StemCells; Hippocampus; Tree shrew; Morphology



树鼩海马神经干细胞的体外识别

黄桂琴*

昆明医科大学解剖教研室，昆明 650500

【摘要】目的：探索和建立树鼩海马神经干细胞培养的新策略，为树鼩海马神经干细胞移植治疗神经疾病奠定理论基础。**方法：**取树鼩胚胎海马组织，经细胞筛选制成单细胞悬液。将细胞悬液的浓度调整到可选值，然后在合适的条件下培养细胞。用特异性抗体对 NSCs 进行鉴定。选择培养期 1、3、5、7d 分别观察 P1、P2 和 P3 传代的 NSCs 形态。酶组织化学染色鉴定 NSCs 的分化能力。免疫荧光染色检测 NSC 的分化能力。**结果：**形态学观察显示，细胞在 24 孔培养板上呈单细胞悬液状，培养 1h 后部分细胞开始粘附，呈球状。1d 后，NSCs 开始粘附，3~5 个细胞团形成小细胞球。在 3d 时发现更多的神经球体积增加。5d 时神经球数量增多，体积增大。7d 时，神经球体积达到峰值，NSCs 开始从神经球中迁移并爆发突起。结果显示，部分 NSCs 表现为 Neun 阳性，细胞核染色，部分 NSCs 表现为 GFAP 标记，细胞质染色，部分 NSCs 表现为阳性，细胞核染色，证实 NSC 分化。利用神经元早期标记物 tuj-1 和 GFAP 验证 NSC 的分化能力，结果表明培养的 NSC 具有向未成熟神经元和成熟星形细胞分化的能力。**结论：**树鼩 NSCs 具有较强的增殖和分化能力，为后续的基础研究和转化应用提供了形态学依据。

关键词：神经干细胞，海马，树鼩，形态学