[1]陈太邦,赵建华,王永飞,等.体外树突状细胞通过NT-3上调P-ERK1/2的表达促进神经干/祖细胞分化[J].第三军医大学学报,2012,34(23):2368-2372.

Chen Taibang,Zhao Jianhua,Wang Yongfei,et al.Dendritic cells promote neural stem/progenitor cells differentiation through NT-3 up-regulating p-ERK1/2 in vitro[J].J Third Mil Med Univ,2012,34(23):2368-2372.

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# 体外树突状细胞通过NT-3上调P-ERK1/2的表达促进神经干/祖细胞分

《第三军医大学学报》[ISSN:1000-5404/CN:51-1095/R] 卷: 34 期数: 2012年第23期 页码: 2368-2372 栏目: 论著 出版日期: 2012-12-15

Title: Dendritic cells promote neural stem/progenitor cells differentiation through NT-3 up-regulating p-ERK1/2 *in vitro*

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关键词: 干细胞; 树突状细胞; 共培养; 分化; NT-3

Keywords: stem cells; dendritic cells; co-culture; differentiation; NT-3

分类号: R329.26; R329.28; R338.12

DOI: -

文献标识码: A

摘要: 目的 观察树突状细胞(dendritic cells,DCs)对神经干/祖细胞（neural stem/progenitor cells,NSPCs）分化的影响，探讨神经营养素-3（Neurotrophin-3，NT-3）在DCs调控NSPCs分化中可能的作用机制。 方法 实验共设5组，分别为NSPCs组、NSPCs/DCs组、NSPCs+NT-3组、NSPCs/DCs+抗NT-3组和DCs组。共培养24、48、72 h后，ELISA法检测各组上清液中NT-3含量；7 d后荧光免疫细胞化学法检测各组NSPCs分化情况。Western blot检测各组NSPCs磷酸化的细胞外调节蛋白激酶（extracellular regulated protein kinases,ERK1/2）表达。 结果 NSPCs/DCs组上清液中NT-3含量较NSPCs组、NSPCs/DCs+抗NT-3组和DCs显著升高（*P*<0.05）。NSPCs/DCs组和NSPCs+NT-3组β-tubulin-Ⅲ阳性细胞数较NSPCs组和NSPCs/DCs+抗NT-3组显著增高（*P*<0.05），而NSPCs组和NSPCs/DCs+抗NT-3组GFAP阳性细胞较NSPCs/DCs组和NSPCs+NT-3组显著增高（*P*<0.05）。NSPCs/DCs组和NSPCs+NT-3组NSPCs中的p-ERK1/2的表达较其余两组增高（*P*<0.05）。 结论 NSPCs与DCs共培养能显著促进NSPCs分化为神经元，可能与共培养后NT-3表达量增高，通过其特异性受体TrkC激活下游信号通路MEK-ERK有关。

Abstract: Objective To observe the effect of dendritic cells (DCs) on neural stem/progenitor cells (NSPCs) differentiation *in vitro*, and to explore the possible mechanism of neurotrophin-3 (NT-3) in NSPCs differentiation regulated by DCs. Methods Primary NSPCs and DCs generated from SD rats were separately cultured and then co-cultured by using transwell chambers, respectively. There are five experimental groups including a NSPCs group, a DCs group, a NSPCs/DCs group, a NSPCs+NT-3 group and a NSPCs/DCs+anti-NT-3 group. After incubation for 24, 48 and 72 h, ELISA was used to detect the NT-3 content in cell supernatant of each group. After 7 d, fluorescence immunocytochemistry was used to detect NSPCs differentiation, and Western blot assay was used to detect the expression of phosphorylated extracellular signal-regulated protein kinases (p-ERK1/2) of NSPCs. Results The ELISA results showed that the NT-3 content in the cell supernatant of the NSPCs/DCs group was significantly increased compared with the NSPCs group, DCs group and NSPCs/DCs+anti-NT-3 group (*P*<0.05). Seven days after induction of differentiation, the proportions of β-tubulin-Ⅲ-positive cells in the NSPCs/DCs group and NSPCs+NT-3 group were significantly higher than those of the NSPCs group and NSPCs/DCs+anti-NT-3 group (*P*<0.05), while the proportions of glial fibrillary acidic protein (GFAP)-positive cells in the NSPCs/DCs group and NSPCs+NT-3 group were significantly lower than those of the NSPCs group and NSPCs/DCs+anti-NT-3 group (*P*<0.05). The Western blotting results showed that the levels of p-ERK1/2 in the NSPCs/DCs group and NSPCs+NT-3 group were significantly higher than those in the NSPCs group, DCs group and NSPCs/DCs+anti-NT-3 group (*P*<0.05). Conclusion Co-culture of DCs and NSPCs can significantly promote NSPCs to differentiate into neurons. Activation of MEK-ERK signaling pathway may contribute to the differentiation of NSPCs into neurons, which is mediated by increased NT-3 binding to its specific receptor TrkC in co-culture system.

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备注/Memo: -