[1]王永飞,赵建华,晁瑞,等.DCs和NSPCs共培养促进NSPCs增殖及其机制的初步研究[J].第三军医大学学报,2012,34(05):373-377.

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DCs和NSPCs共培养促进

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Title: Dendritic cells promote neural stem/progenitor cells proliferation *in vitro* when cocultured

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关键词: 神经干细胞; 树突状细胞; 神经营养素-3; 酪氨酸激酶受体C

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摘要: 目的　观察体外Transwell共培养系统中树突状细胞（dendritic cells，DCs）对神经干/前体细胞（neural stem/progenitor cells，NSPCs）增殖的影响，初步探讨神经营养因子-3（neurotrophin-3，NT-3）在DCs调控NSPCs中的作用机制。　方法　根据是否采用Transwell小室将DCs和NSPCs共培养分为分隔培养（DCs/NSPCs组）和混合培养（DCs+NSPCs组），ELISA法测定DCs组、NSPCs组、DCs+NSPCs组、DCs/NSPCs组共培养48 h上清中的NT-3的含量。为观察NT-3在DCs调控NSPCs中的作用及可能的机制，成球法检测NSPCs组、NSPCs/DCs+K252a组、NSPCs+DCs组、NSPCs+NT-3组的NSPCs增殖，免疫细胞荧光法和Western blot法检测各组NSPCs表面酪氨酸激酶受体C（TrkC）的表达。　结果　ELISA实验结果显示，Transwell共培养48 h上清NT-3的含量，NSPCs/DCs组较DCs组和NSPCs组明显升高（*P*<0.05）。镜下观察、测量结果显示NSPCs/DCs组和NSPCs+NT-3组神经球数量增多,平均直径明显增大（*P*<0.05）。免疫细胞荧光和Western blot检测结果表明,NSPCs+DCs组和NSPCs+NT-3组中NSPCs的TrkC表达较NSPCs组和NSPCs/DCs+K252a组高（*P*<0.05）。　结论　体外DCs与NSPCs共培养，促进NSPCs增殖、NT-3的分泌及TrkC的表达，NT-3可能是通过TrkC受体参与NSPCs增殖的调控。

Abstract: Objective　To investigate the effect of dendritic cells (DCs) coculture on the proliferation of neural stem/progenitor cells (NSPCs) *in vitro*, and explore the role of TrkC receptor and neurotrophin-3 (NT-3) in the process.　Methods　According to whether to adopt the Transwell chamber, the DCs and the NSPCs were co-cultured and divided into separate culture (DCs/NSPCs group) and mixed culture (DCs+NSPCs group, without the use of Transwell chamber). ELISA was used to detect neurotrophin-3 (NT-3) protein in the supernatant in 48 h after culture in NSPCs+DCs group (mixed culture), DCs group, NSPCs group, and NSPCs/DCs group (only cells separated by Transwell chamber). Tyrosine kinase receptors C (TrkC) proteins on the surface of NSPCs were detected by immunofluorescence staining and Western blotting analysis in NSPCs group, NSPCs/DCs+K252a group (TrKC blocker), NSPCs+DCs group, and NSPCs+NT-3 group.　Results　The levels of NT-3 in the supernatant were significantly higher in 48 h after culture in NSPCs/DCs group than in DCs group and NSPCs group (*P*<0.05). And microscopy showed significantly increases in the number and the average diameter of neurospheres in NSPCs/DCs group (*P*<0.05). Immunofluorescence staining and Western blotting indicated that the expression level of TrkC were significantly higher in NSPCs/DCs group and NSPCs+DCs group than in NSPCs group and NSPCs/DCs+K252a group (*P*<0.05).　Conclusion　DCs in coculture promotes NSPCs proliferation *in vitro*, which might be through TrkC-NT-3 signal pathway.

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