

论文

胰腺干细胞与胰岛混合培养对其转化率的影响

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摘要:

目的 研究胰腺干细胞与胰岛混合培养对增加胎鼠胰腺干细胞向β前体细胞转化率的作用及机制。方法 实验分为胰腺导管干细胞单独培养组(I组)、胰岛单独培养组(II组)及干细胞-胰岛混合培养组(III组)。V型胶原酶消化大鼠胰腺获得胰岛后,使用Ficoll-400梯度离心法纯化胰岛;采用胶原酶消化法获得胎鼠胰腺干细胞进行培养,通过RT-PCR及免疫组织化学染色的方法,检测细胞角蛋白-19(CK-19)、巢蛋白(Nestin)、胰高血糖素和胰岛素等干细胞相关标志物。干细胞诱导培养基中加入纯化的胰岛进行混合培养,通过观察干细胞表达胰岛素的阳性率评价其诱导转化率。结果 V型胶原酶逆行胰管灌注继而Ficoll 400梯度离心可获得生物活性良好的胰岛;所培养的干细胞表达CK-19、Nestin、胰高血糖素,诱导后部分表达胰岛素。干细胞-胰岛混合培养组及干细胞单独培养组诱导后,细胞胰岛素阳性率分别为38.2%和23.9%,差异有统计学意义(P<0.05);CK-19阳性率分别为89.3%和81.6%,其差异亦有统计学意义(P<0.05)。结论 胰腺导管干细胞与胰岛混合培养可提高胰腺导管干细胞向β前体细胞的转化率,其作用与CK-19的表达密切相关。

关键词: 糖尿病; 胰岛移植; 干细胞; 诱导; 分化; 大鼠, Wistar

Coculture with islet to make pancreatic stem cells differentiate to insulin-producing cells

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Abstract:

Objective To observe differentiation rate of Pancreatic stem cells (PSCs) when they were cocultured with islet, and then study its mechanism. Methods Isolation of rat islets was performed with collagenase-V digestion and purification by using Ficoll-400. At the same time, PSCs were harvested from pancreatic rudiments of fetal rats by using collagenase digestion and identified by immuocytochemistry and RT-PCR after culture in the second generation. Then islet and PSCs were cultured together, and insulin expression of PSCs was observed to evaluate the rate of PSCs to differentiate to insulin-producing cells. Results Islets with biological viability were obtained by using collagenase-V and Ficoll-400 in digestion and purification. Most of the PSCs expressed CK-19, nestin and glucogon, and some of the PSCs also expressed insulin after induction. The rates of insulin-positive PSCs in islet-stem cell cocultured group and stem cell cultured group were 38.2% and 23.9% (P<0.05) respectively, and the rates of CK-19 positive PSCs in the two group were 89.3% and 81.6% respectively (P<0.05). Conclusio nCocultivation with islet can make PSCs differentiate into insulin-producing cells, which is closely related to the expression of intracellular CK-19.

Keywords: Diabetes mellitus; Islet transplantation; Stem cells; Induction; Differentiation; Rats, Wistar

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