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# 乳腺癌残存细胞通过表达乳腺癌干细胞样特性促进裸鼠移植瘤的形成



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Title: Drug surviving cells promote formation of transplanted tumor model in nude mice through expressing breast cancer stem-like traits

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关键词: 残存细胞; 乳腺肿瘤; 肿瘤干细胞; 裸鼠移植瘤

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摘要:

目的　探讨化疗后乳腺癌残存细胞（drug surviving cells，DSCs）的干细胞样特性及其对裸鼠移植瘤的影响，阐明DSCs细胞的潜在生物学特性。　方法　以0.5 μg/mL浓度的多西紫杉醇(docetaxel，DTX)每周1次药物处理乳腺癌细胞MCF-7，分别获得处理4次后的DSCs，记为DSCs（1～4）组，未处理的MCF-7细胞为空白对照组。流式细胞仪检测空白对照组及DSCs（1～4）组中细胞周期分布。无血清悬浮培养法培养MCF-7细胞和DSCs（1～4）细胞，获得乳腺癌干细胞（breast cancer stem cells，BCSCs）微球体，荧光定量PCR和Western blot法分别检测MCF-7细胞和DSCs（1～4）细胞中乳腺癌干细胞标志物ALDH1、CD44、CD24及ABCG2 mRNA和蛋白表达水平；采用

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裸鼠移植瘤实验比较MCF-7细胞及DSCs（1～4）细胞的成瘤时间、成瘤率及瘤体大小。　　结果　　与MCF-7细胞相比，药物处理后得到的DSCs（2～4）细胞中处于静止期（$G_0/G_1$期）的细胞比例明显升高。DSCs（2～4）细胞形成乳腺癌干细胞微球体的能力更强，表达的干细胞标志物ALDH1、$CD44^+/CD24^-$和ABCG2显著增多,差异均有统计学意义（$P<0.05$）；DSCs（2～4）细胞的成瘤能力更强，瘤体体积较MCF-7细胞明显升高（$P<0.05$）。DSCs（1）细胞与MCF-7细胞相比无明显差异。　　结论　　经多西紫杉醇药物筛选后的DSCs表达乳腺癌干细胞样特性，促进裸鼠移植瘤的生成。

Abstract: Objective To determine the breast cancer stem-like traits of drug surviving cells (DSCs) and the effect on the formation of transplanted tumor in nude mice for the DSCs' biological features. Methods Breast cancer cell line MCF-7 was weekly treated with docetaxel (DTX) at the concentration of 0.5 μg/mL, and the cells at the 1st to 4th generation of DSCs were individually named as DSCs 1 to 4. The untreated MCF-7 cells served as blank control cells. The cell cycle distribution of MCF-7 and DSCs 1 to 4 were tested by flow cytometry. MCF-7 cells and DSCs 1 to 4 were cultured in serum-free suspension culture medium to acquire microspheres of breast cancer stem cells (BCSCs). The expression of BCSCs marker ALDH1, CD44, CD24 and ABCG2 at mRNA and protein levels were detected by real-time q-PCR and Western blotting. Nude mice transplantation tumor was prepared to evaluate the time, rate and volume of tumor formation of MCF-7 cells and DSCs 1 to 4, respectively. Results The cell population of stationary phase ($G_0/G_1$) was significantly increased in DTX treated groups DSCs 2 to 4 compared with MCF-7 cells. Moreover, DSCs 2 to 4 had stronger ability of forming microspheres of BCSCs than MCF-7 cells. The results of real-time qPCR and Western blotting showed that the expression of BCSCs markers ALDH1, $CD44^+/CD24^-$ and ABCG2 was obviously up-regulated in DSCs 2 to 4 ($P<0.05$). Furthermore, the growth rate, the volumes of tumors were significant elevated in DSCs 2 to 4 compared with MCF-7 cells ($P<0.05$). Whereas, DSCs 1 had no significant difference in above indexes compared with blank control MCF-7 cells. Conclusion DSCs exhibit the features of BCSCs after being treated with DTX, and promote the formation of xenograft tumor in nude mice.

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