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小鼠乳腺癌4T1细胞中肿瘤干细胞样细胞的富集和鉴定 点此下载全文

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摘要:

目的:无血清培养基(serum-free medium, SFM)悬浮培养小鼠乳腺癌细胞株4T1,富集并鉴定4T1细胞株中肿瘤干细胞样细胞。方法:通过含EGF、bFGF和B2 7等细胞因子的SFM培养富集4T1细胞中肿瘤干细胞样细胞,将其接种于含血清培养基(serum-supplemented medium, SSM),观察4T1肿瘤干细胞样细胞分化情况。应用细胞表面标志CD44 +CD24-/low和Hoechst 33342染色法检测4T1细胞中肿瘤干细胞样细胞的比例,小鼠致瘤实验验证不同培养条件下4T1肿瘤干细胞样细胞的致瘤能力。结果:4T1细胞在SFM中能够存活、增殖,并形成细胞球,细胞球可连续传代,若重新接种于SSM中可贴壁分化。4T1细胞球中CD44 +CD24-/low细胞比例为6.4%-68.9%,侧群(side population,SP)细胞比例为7.3%-61.2%,均显著高于SSM中培养的4T1细胞(P<0.05);随着SFM中细胞球传代次数增加,CD44 +CD24-/low细胞和SP细胞的比例逐渐升高。小鼠致瘤实验结果显示,富集了肿瘤干细胞的细胞球比常规培养4T1细胞的致瘤性更强。结论:乳腺癌4T1细胞可在含多种生长因子的SFM中悬浮生长并形成细胞球,4T1细胞中含有的乳腺癌干细胞样细胞可通过SFM培养法富集。

关键词: 无血清悬浮培养 乳腺癌4T1细胞 肿瘤干细胞 CD44 +CD24-/low 侧群细胞

Enrichment and identification of cancer stem cell-like cells in mouse breast cancer cell line 4T1 <u>Download Fulltext</u>

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Abstract:

Objective: To culture mouse breast cancer 4T1 cells in serum-free medium(SFM), and to screen for and identify the cancer stem-like cells in 4T1 cells. Methods: Breast cancer stem-like cells were enriched from 4T1 cells cultured in SFM containing EGF, bFEGF, and B27, etc. The suspension spheres were seeded in serum-supplement medium (SSM) and cell differentiation was observed. The proportion of cancer stem-like cells in 4T1 cells was determined through cell surface markers CD44 +CD24-/low and Hoechst 33342 staining. Tumorigenic abilities of 4T1 cells in different culture conditions were detected by mouse tumorgenesis experiment. Results: 4T1 cells could survive, proliferate and form breast cancer suspension spheres in SFM for prolonged time period. 4T1 spheres seeded into SSM could differentiate and adhere to the culture plates. There were 6.4%-68.9% CD44 +CD24-/low cells and 7.3%-61.2% side population (SP) cells in 4T1 spheres, which were significantly higher than those in 4T1 cells cultured in SSM (P<0.05). The ratios of CD44 _CD24-/low and SP cells were gradually increased with the passage of 4T1 spheres in SFM. 4T1 spheres with enriched cancer stem cell were more tumorigenic than 4T1 cells cultured in the SSM. Conclusion: 4T1 cells can grow and form spheres in serum-free suspension medium containing growth factor, and they contain breast cancer stem-like cells, which can be enriched when cultured in SFM.

Keywords:serum-free suspension culture breast cancer 4T1 cell cancer stem cell CD44 +CD24-/low side population cell

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