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Isolation and identification of putative cancer stem cells in C26 murine colon carcinoma cell line

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Background and Aims: Recently, the theory of cancer stem cells (CSCs), the existence of a distinct subpopulation of cancer cells that possesses the capacity to self-renew and to cause the heterogeneous lineages of cancer cells has presented new targets and orientations for tumor therapy. A growing body of evidence supports a role for these cells in tumor recurrence and metastasis. The major difficulties in researching CSC are isolation and purification of these cells. The present study was designed for the isolation and characterization of putative CSC populations in established C26 murine colon carcinoma cell line.
Methods: Isolation or enrichment of the putative cancer stem cell population was carried out by Magnetic Activated Cell Sorting (MACS) based on the expression of CD44 and CD133 cell surface proteins. After sorting, these populations were evaluated for CSC properties in comparison to their negative counterparts or to the parental cell line. Spheroid formation ability in serum-free medium and tumorigenicity upon injection in mice were assessed.
Results: The results showed that the percent of CD44. CD133, EpCAM were 99%, 0.5%, 0.1% respectively in this cell line by flow cytometry. After isolation of cells according to the CD133 marker by MACS , the sorted cells could form spherical clones in serum-free culture media, but the rate of clonegenesis of CD44+CD133+ and CD44+CD133_ cells was similar. In vivo, CD44+ cells showed greater ability to form tumor (about 105cells were sufficient) in comparison to the parental cell line (3×10 5 cells were needed).
Conclusions: This study indicated that C26 cell line probably contained some distinct subpopulation with stem cell properties and combination of some of these cell surface proteins could be cancer stem cell markers for colon carcinoma. These results could provide an important research tool for testing and developing novel targets for cancer therapy.

Keywords: Cancer stem cell; Magnetic activated cell sorting (MACS); Isolation, Identification; Flow cytometry