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Human mesenchymal stem cells creating an immunosuppressive environment and promote breast cancer in mice

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Human mesenchymal stem cells (hMSC) can home to tumor sites and promote tumor growth. The effects of hMSC on tumor growth are controversial and involvement of hMSC in tumor immunology has not been adequately addressed. Therefore, we investigated whether injection of hMSC affects tumor appearance, growth and metastasis, and anti-tumor immunity in an experimental animal model of metastatic breast cancer. Injection of hMSC in BALB/c mice bearing mammary carcinoma promoted tumor growth and metastasis, which was accompanied by lower cytotoxic activity of splenocytes, NK cells and $CD8^+$ T cells *in vitro*. Tumor-bearing mice that received hMSC had significantly lower percentages of $CD3^+NKp46^+$ NKT-like, higher percentages of $CD4^+Foxp3^+$ T cells, increased serum levels of Th2 and decreased serum levels of Th1 cytokines, and significantly higher number of $CD4^+$ cells expressing IL-10. These results demonstrate that immunosuppressive environment created by hMSC promoted breast tumor growth and metastasis in mice.

Mesenchymal stem cells (MSC) have been identified in the bone marrow, connective tissue and peripheral blood as multipotent cells that proliferate *in vitro* as plastic adherent cells. These cells have the capacity to differentiate into fibroblast, osteoblast, and adipocyte lineages[1,2]. It has been demonstrated that human MSC, (hMSC) maintained under standard culture conditions are non-tumorigenic *per se*: however, several reports indicate their capability to modulate tumor microenvironment thus having an impact on the tumor behaviour[3,4]. Lin *et al.*[5] demonstrated that MSC have exerted immune protection, and, in case of breast cancer, they can support cancer growth[6]. Also MSC can contribute to the tumor growth and progression through several mechanisms including their immunomodulatory effects. These effects depend on their basic characteristics, time and method of administration in relation to pathological conditions[7,8]. As far as tumor modulation is concerned, MSC can have both, stimulatory[9] and suppressive effects[10]. Most specifically, in an experimental model of breast cancer it has been shown that MSC may have both of the above effects. The intravenous administration of MSC appears to effect tumor growth indirectly by modulating the immune microenvironment before they act by direct interaction with tumor cells. It has been shown that MSC overexpressed IFN-β by affecting STAT3 signaling pathway, however, "normal" MSC stimulated rather than suppresed 4T1 breast cancer cells growth[11].

MSC are generally thought to have immunosuppressive effects, which may be an important mechanism through which MSC promote tumor growth or increase incidence of tumor formation also under *in vivo* conditions They can impair the function of a variety of immune cells directly or by secreting different growth factors possibly by inhibition of both, innate and adaptive immune cells[12,13]. However, the immunomodulatory effects of MSC, if any, are not well understood within tumors. Djouad *et al.*[14] reported that the immunosuppressive action of MSC led to a higher incidence of melanoma formation in a mouse model. Evidence for the effect of MSC on tumor immunity also comes from a clinical study[15] which could be explained by the creation of immunosuppressive milieu including shifting of immune responses from Th1 to Th2[16] or induction of generation and proliferation of regulatory T cells[17]. However, regulatory T cells (Tregs) are mostly immunosuppressive, and have been shown to suppress effector T cell functions and autoimmunity[18,19]. They suppress T-cell responses by acting directly or through the inhibition of antigen-presenting cells involving cell-to-cell contact, FasL/Fas, and PD1/B7-H1-dependent processes, or secreted factors such as IL-10, TGF-beta, IL-27, and IL-35[20–22]. As a result,