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Mesenchymal Stem Cells and Pathotropism: Regenerative Potential and Safety Concerns

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Abstract

Mesenchymal Stem cells (MSCs) are ubiquitously expressed in several organs, but the major sites in adults are bone marrow and adipose tissue. MSCs can form several cells belonging to all germ layers, such as neurons and cardiomyocytes. MSCs have the potential to be used in cell therapy for many clinical problems, e.g., tissue regeneration, replacement, and to suppress inflammatory processes. MSCs are attractive due to reduced ethical concerns, ease in expansion and ability to be used as 'off-the-shelf' cells. MSCs can be indicated for clinical disorders due to their homing to regions of high cytokines such as tissue insult. This process is generally referred as pathotropism. MSCs have been placed in numerous clinical trials. Thus far, there is no evidence of safety concerns. Besides transplantation of hematopoietic stem cells, treatments with other stem cells are relatively recent. Thus, MSC therapy requires strict monitoring for safety issues. The pathotropic effect of MSCs allows these cells to home to tumors. This property led to the use of MSCs as cellular vehicle for drugs. A major concern of using MSCs in regenerative medicine is their ability to protect and support tumor growth. This chapter focuses on the potential safety concerns of using MSCs. This issue is particularly important if the recipient of stem cells has an undiagnosed tumor or is in cancer remission.

Keywords: Stem cells, Cancer, Regeneration, Dormancy, Bone marrow, Pathotropism.

2.1 Introduction

The presence of stem cells in all organs strongly suggests that these cells may be important to protect, and perhaps replace, tissue during minimal insults. These seeming baseline properties have led scientists to investigate how stem cells can be used in tissue repair. This led to the removal of stem cells from the natural microenvironment for *ex vivo* manipulation in the absence of the natural microenvironment. There are reports in which autologous stem cells are removed and immediately transplanted (www.clinicaltrials.gov). However, in the majority of cases, the stem cells are expanded *in vitro.* This places the stem cells at risk for mutations and functional changes that could differ from their endogenous properties. As stem cells move to patients, there must be consideration on their safety and the functional alterations.

Stem cells can self-renew and differentiate into any cell type. These two major characteristics provide them with the potential to regenerate tissues and for use in organogenesis. Embryonic and induced pluripotent stem cells have scientific challenges mostly due to the ease in forming tumors. Stem cells in adults, fetus, cord, cord blood and placenta show potential in clinical application. Stem cells in the adult brain and bone marrow, such as mesenchymal stem cells (MSCs) have shown promise in regenerative medicine and in drug delivery to tumors [1].

2.2 Mesenchymal Stem Cells (MSC)

MSCs are primordial in origin and can be isolated from fetal and adult tissues such as the placenta, bone marrow and adipose tissues [2–5]. MSCs are well-characterized with regards to phenotype. There are commercially available antibodies to phenotype MSCs with antibodies such as those targeted to CD73, CD90 and CD105 [6]. MSCs do not express hematopoietic markers such as CD45. MSCs have been reported to express vimentin and fibronectin [7]. In addition to phenotype, MSCs are characterized by functionality, including assays to ensure multipotency and immune properties [6].

MSCs have significantly reduced ethical concerns, are easy to expand *in vitro*, and more importantly, can be used as 'off the shelf' sources in cell

therapy. An important consideration with MSCs as cell therapy is the changing microenvironment. Thus, a clear understanding of how the changing niche affects the functions of MSCs is key. The immune function of MSCs is particularly relevant. MSCs can be immune enhancer and suppressor cells. The immune function of MSCs depends on the milieu of the microenvironment. Specific cytokines and chemokines can be chemoattractant to facilitate the migration and homing of MSCs and other immune cells to the site of tissue injury.

2.2.1 MSC Immunology

The immune functions of MSCs are interesting since these stem cells can be stimulatory as well as suppressive, depending on the microenvironment (Figure 2.1) [8]. MSCs, like other immune cells within a niche, can produce cytokines, thereby establishing communication with the cells found within

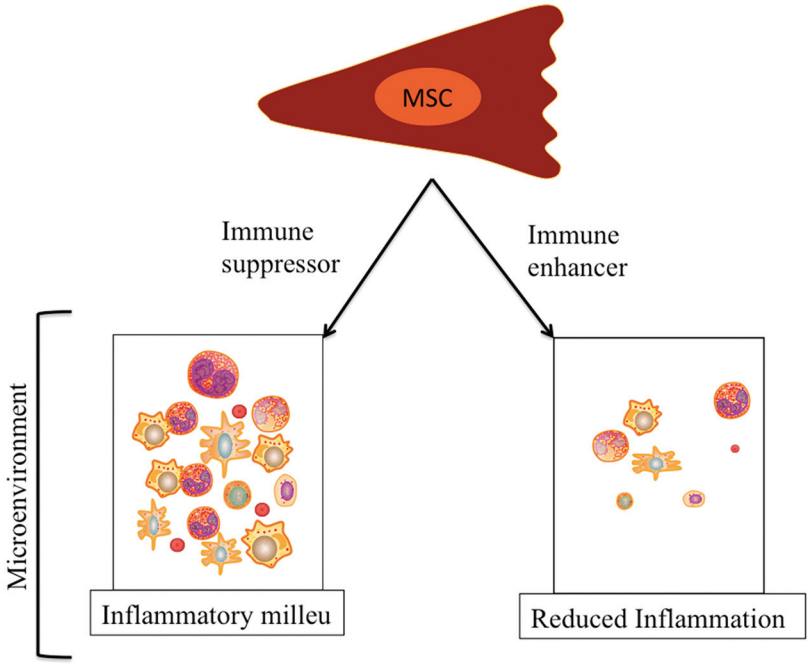

Figure 2.1 Microenvironment dictates dual role of MSCs. The inflamed microenvironment licences MSCs to immune suppressor cells whereas the reduced inflammation signal allows MSCs to be an immune enhancer.

the tissue microenvironment [9]. The expression of major histocompatibility complex-II (MHC-II) was proposed as a minimum requirement for a cell to be designated MSC [6]. There are several reports showing MSCs as negative for MHC-II, despite the correct phenotype and evidence of multipotency [10]. There are ongoing investigations to determine if the difference in MHC-II expression is due to the source from which the MSCs was derived, adipose tissue versus bone marrow, or if the difference could be explained by the type of culture condition.

MHC-II expression provides the MSCs with the ability to function as antigen presenting cells (APCs) to stimulate the immune system [11]. The method by which MSCs function as APCs is different from professional APCs. MSCs respond differently to interferon γ (IFNγ) with regards to MHC-II expression: at high levels, MHC-II expression is decreased whereas at low level, MHC-II is increased [12]. This difference is relevant for the translation of the science on MSCs because these stem cells would alter their functions with the change in the inflammatory microenvironment. Our data showed a similar re-expression of MHC-II on neurons in the presence of IFNγ [13]. This reexpression of MHC has the potential safety issues with MSC treatment.

MSCs can be licensed to become immune suppressor cells within an inflammatory milieu [14]. As immune suppressor cells, the MSCs have undetectable surface MHC-II, inhibit T-cell and B-cell stimulatory responses, and inhibit natural killer activity [6, 10, 15, 16]. These suppressive effects are generally associated with an increase in regulatory T-cells [17]. The immune properties of MSCs have been suggested to have a role in tissue repair [18].

2.3 MSC in Tumor Support

Fetal and adult MSCs can support and protect tumors [19]. It is unlikely that transplanted MSCs will form tumors since they are not likely to survive for long periods. However, they could 'waken' sleeping cancer cells since they can suppress the immune response and at the same time support tumor growth [20]. MSCs have been shown to initiate the growth and metastasis of tumor cells [21–25]. This tumor-promoting role of MSCs can occur with solid and hematologic malignancies [26–28].

The mechanisms by which MSCs support tumor growth would require further studies with different types of tumors. Also, since tumors are heterogeneous, the studies must be performed with various subsets of cancer cells.

2.3.1 MSC in Drug Delivery to Tumors

Based on the above discussion, pathotropism of MSCs could be deleterious for the recipient who might have an undiagnosed or is in remission from cancer (Figure 2.2). This disadvantage can be an advantage because the ability of MSCs to be attracted to the sites of tumor growth could be applied to have these stem cells deliver therapies to tumors. As an example, glioma cells secrete soluble factors, which act as chemoattractants for MSCs to the site of the tumor. MSCs with ectopic expression of anti-tumor molecules such as TNFα and IFN-β were studied as a method to inhibit tumor promotion [29–33].

The use of MSCs as a cellular vehicle for drugs to target glioblastoma multiforme (GBM) is an attractive area of research. GBM is an aggressive and invasive cancer with poor prognosis, despite aggressive treatments that include tumor resection with radio- and chemotherapy [34]. The utility of drug delivery has been demonstrated in rats in which the MSCs migrated to the region of gliomas [35]. In these studies, the MSCs were intracranially implanted into rats with GBM. The MSCs then migrated to and dispersed within the tumor mass [35]. A similar study with immunocompromised mice showed human MSCs migrating to the region of human gliomas [36]. In these studies, MSCs were injected into the ipsilateral and contralateral carotid arteries of the mice [36]. Other studies injected the MSCs intratumorally [37]. MSCs can be used to deliver RNA through gap junction or through exosomes, as demonstrated with the transfer of exosomes-derived anti-miR9 [38].

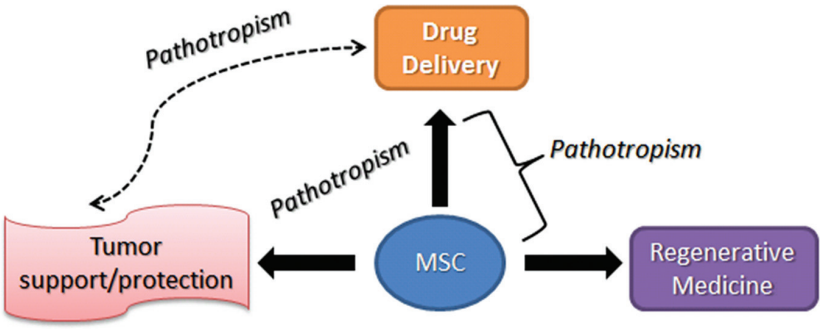

Figure 2.2 Pathotropic effects of MSCs in regenerative medicine. Shown is a MSC in the center that could be attracted to the site of tissue injury where it can be attracted to regenerate the tissue. Similarly, tumors, which can produce chemoattractants, could attract MSCs. The same pathotropic effect could be used to deliver drugs to areas of tumors.

2.4 Conclusion

A major concern with using MSCs, including the use as cellular deliverers of drugs, is the potential of MSCs to support tumor growth and to immunosuppress cells. The latter function will provide a survival advantage for the tumor while providing the MSCs with the ability to support tumor growth. At this time, there is no longitudinal study to determine if MSC treatment is safe. These are necessary studies that should be addressed. To understand how MSCs could be safely employed, it is necessary to develop experimental models of dormancy since this will recapitulate undiagnosed tumors and cancer in remission. Also, studies are needed to develop hierarchy of tumors.

A mistake noted in the literature is for scientists to presume that the properties of cancer stem cells are similar to normal stem cells. Most notable among these differences is the designation of cancer stem cells as small population SP) cells, which would indicate cells with low metabolic activity. Unlike normal stem cells, cancer stem cells are malignant cells, which would indicate relatively large cells as compared to normal stem cells. The regenerative potential of MSCs needs to be studied in conjunction with robust studies of cancer stem cells.

Regarding the drug delivery capacity of MSCs, it might be necessary to eliminate these cells after the drug is released. At present there are methods to induce suicidal genes to eliminate the MSCs [39].

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