Editorial

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Regenerative Medicine

Mesenchymal stem cells in regenerative medicine: a new paradigm for degenerative bone diseases

"It is imperative to delineate a deeper and more critical understanding of the physiology of mesenchymal stem cells, including their survival time, ability to home into organs and tissues and donor-to-donor variability."

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The mesenchymal stem cells (MSCs) have been the epicenter of regenerative medicine since their identification in the 1970s, due to their ability to differentiate into various cell types, their immunosuppressive function, and their ability to home to injury sites. Initially, MSCs were discovered from bone marrow as adherent cells that have the potential to differentiate into bone cells [1–3]. Since inception from bone marrow, analogous cells have been successfully isolated from various sources. Since then, MSCs from bone marrow have been used as a positive control for MSCs isolated from various tissues. A functional characteristic of MSCs is their ability to differentiate into ectoderm, mesoderm and endoderm tissues including bone, neurons, muscles, hepatocytes, skin, among others. MSCs may be characterized based on a panel of surface markers, distinguishing them from endothelial, hematopoietic and monocyte cells. MSCs are typically positive for CD44, CD73, CD90 (Thy-1) and CD105 (endoglin), and negative for hematopoietic ($CD45^+$ and TER119 markers) and endothelial (CD31, von Willebrand factor) markers [2,4]. A recent advancement in this field is the isolation of MSCs from dental origin including dental pulp, periodontal ligaments, apical papilla, among others; and we are achieving some positive outcomes with MSCs from dental sources [Singh SK, Saxena SK, Pers. Comm.]. Recently, it has been discovered that MSCs could be isolated from nasal polyp tissues, thus providing a potential new source of multipotent MSCs [5]. According to the Mesenchymal and Tissue Stem Cell Committee of the International Society for Cellular Therapy, the minimal criteria of standardization of human MSCs are: first, MSCs must be adherent to the surface of standard plastic culture vessels; second, MSCs must express CD105, CD73, CD90 and low levels of MHC-I and be negative for MHC-II, CD45, CD34, CD14 or CD11b surface molecules; and third, MSCs must be able to differentiate into osteoblasts, adipocytes and chondroblasts *in vitro* [4].

Biological characteristics supporting clinical use

Nowadays, MSCs are valuable sources of therapeutics for regenerative medicine. The characteristics that make them so important include their ability to differentiate into various cell lineages, secretion of unknown cellular regulators (cytokines) and site-specific migration. The differentiation potential increases interest in MSCs for clinical purpose [6]. For example, MSCs could differentiate into islet cells that express insulin and glucagon, thereby highlighting the potential

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of MSCs in diabetic treatment [7]. It has been demonstrated that MSCs could be used as an alternative in wound repair due to their ability to differentiate into multiple skin cell types [8]. MSCs have been differentiated into renal tubular epithelium and promoted tissue repair via the secretion of cytokines, which play an important role in tissue structure integrity [9]. Studies such as this have supported the idea that MSCs could repair damaged tissue and eliminate dead resistive tissue. The therapeutic potential of MSCs is however not merely dependent on their differentiation capacity, since this is only one aspect of the role of MSCs in clinical importance.

> "Since inception, mesenchymal stem cells have been associated with their potential use in bone regeneration."

Recently, various reports have documented that transplanted MSCs secrete growth factors, chemokines and cytokines, which emphasize their therapeutic potential. MSCs secrete EGF, HGF, IGF, VEGF, angiopoietin-1, macrophage inflammatory protein-1α, stromal derived factor-1, erythropoietin, nitric oxide, among others[10]. Studies have suggested that transplanted MSCs release various biological factors that affect cellular behavior and not only promote cell proliferation, but also prevent apoptosis to adjacent cells, ultimately promoting tissue regeneration [11]. Immunomodulation is another important property of MSCs that can alter the immune response through factors which interact with target cells and exert their immunomodulatory functions. These factors include TGF-β1, HGF, haemoxgenase-1, PGE-2, IL-10 and human leukocyte antigen-G5 [12]. MSCs can also downregulate T-cell proliferation, inhibit cytotoxic T-cell production and suppress the T-cell response to their cognate peptide. However, the effects of MSCs in B-cells are ambiguous. Diverse studies report that MSCs may activate/inhibit IgG secretion through B cells and enhance the proliferation and differentiation of plasma cells from memory B cells [10]. MSCs also inhibit the differentiation potential of dendritic cells from $CD34^+$ progenitors, monocytes and reduce proinflammatory cytokines [13].

Translational potential of MSCs for bone & cartilage disease

Currently, MSCs serve as a putative therapeutics for several diseases. Numerous studies have already demonstrated beneficial uses of MSCs during the last decade and more, in both preclinical research and clinical trials. In fact, several clinical trials have highlighted the promise of MSCs in a number of disorders including lymphocyte recovery, multiple sclerosis, Type II diabetes, tendinopathy, osteoarthritis (OA), chronic spinal cord injury and feline chronic kidney disease [14,15]. Phase III clinical trials have also been carried out in stroke patients [16]. Bone cartilage defects are one of the most promising health problems suggesting favorable role of tissue regeneration therapies.

Since inception, MSCs have been associated with their potential use in bone regeneration. Their ability to differentiate into osteocytes and chondrocytes highlights their promise in bone regenerative therapeutics. OA is the most common chronic disease of the joint and the most common chronic disease in the aged population [17]. The characteristic feature of this disease is the degeneration of bone and synovial inflammation. Current treatment strategies do not adequately prevent destruction of the OA joint and options are relatively limited to treating symptoms or managing pain control using various inflammatory drugs or steroids. Ultimately, surgery is the option including autologous chondrocyte implantation, osteochondral graft or total joint replacement [18]. It has been evidenced that the OA reduced the adipo- and chond-rogenic potential of resident MSCs [19].

In a recent study, bone marrow derived MSCs were exposed to low MW fraction (<5 kDa) of commercial human serum albumin (LMWF5A), which resulted in both mobilization and chondrogenic differentiation of the MSCs [20]. A clinical trial involving a preliminary set of patients indicated that at 12 weeks following administration of LMWF5A, there was an improvement in stem cell infiltration, self-renewal, differentiation and a reduction in inflammation in the knee [20]. Autologous adipose-derived MSCs have been shown to be safe and well tolerated in patients with knee OA [21].

Osteogenesis imperfecta (OI), colloquially known as brittle bone disease, is a genetic disorder caused by mutation in type I collagen genes (*COL1A1* and *COL1A2*). It is one of the most challenging diseases that were characterized by severe atypical skeletal development, shortened stature and osteopenia. Currently, treatment may include pain control, prevention of bone fracture and a surgery correction that puts metal rods through long bones. Bisphosphonate is the only available medication, which has symptomatic, but not curative therapeutics [22]. During transplantation of allogenic MSCs in an OI mouse model, donor MSCs homed to the bones, where they contributed to increase in both collagen and mineral content [23]. Clinical trials involving *in utero* MSCs transplantation in the female fetus with severe OI have exhibited only three fractures at 2-year follow-up. At 8 years of age, further secondary transplantation of same-donor MSCs resulted in improved linear growth and mobility of bone with low fracture incidence [22]. The bone

marrow MSCs have also exhibited improved bone mineral content of severe OI children compared with healthy ones [24].

Osteoporosis is a disease of reduced bone mass, leading to bone fragility and increased risk of fractures [25,26]. This disease is due to imbalance in osteoblast formation and resorption of osteoclasts [27]. It has been evidenced that a defect in MSCs proliferation and differentiation into osteoblasts is linked to osteoporosis [28]. Data suggest that the impaired osteoblast differentiation participates in the pathology of osteoporosis [29]. Various studies have demonstrated the role of MSCs in animal models of osteoporosis [28,30]. More clinical studies are required to further evaluate the significance of these cells in medical practice.

Conclusion & future perspective

The human body is fortified with a unique cell population, namely MSCs, which has the ability to regenerate and differentiate into various other cell types. Moreover, MSCs have the immunoregulatory properties. Potentially, they can be easily isolated and safely transplanted to injured sites that make them beneficial in biomedical research. Numerous *in vitro* and *in vivo* studies in animal models have successfully demonstrated the potential of MSCs for various diseases however the clinical outcomes are not very encouraging. The communication between basic scientists and clinicians are one of the reasons that the various important outcomes could not be translated for clinical purpose. So, more work is required before expanding the clinical application of MSCs to complete research and development. It is imperative to delineate a deeper and more critical understanding of the physiology of MSCs, including their survival time, ability to home into organs and tissues and donor-to-donor variability. It requires the collective and global standardization of culture techniques and determination of the ideal source (bone marrow MSCs, dental pulp MSCs, among others), optimal dosage, treatment frequency, route of administration, correct algorithms and methods for biobanking of MSCs. In a nutshell, we need to solve these issues in order to make MSCs a useful resource in clinical therapeutics in the future. Basic and clinical researchers have to come together to develop comparative and internationally accepted standard operating protocols for the clinical grade MSCs. Nevertheless, cooperative efforts may resolve these issues and make clinical application of MSCs affordable to common man in the near future by developing novel and easy protocols without altering their basic characteristics, and we need to proceed with a sense of urgency in this matter.

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