ML24-18 (26/06/2018)

Front Immunol. 2018 Jun 1;9:1207. doi: 10.3389/fimmu.2018.01207. eCollection 2018.

Adaptive Regulation of Osteopontin Production by Dendritic Cells Through the Bidirectional Interaction With Mesenchymal Stromal Cells.

Scutera S[1], Salvi V[2], Lorenzi L[2], Piersigilli G[1], Lonardi S[2], Alotto D[3], Casarin S[3], Castagnoli C[3], Dander E[4], D'Amico G[4], Sozzani S[2], Musso T[1].

Author information

Abstract

Mesenchymal stromal cells (MSCs) exert immunosuppressive effects on immune cells including dendritic cells (DCs). However, many details of the bidirectional interaction of MSCs with DCs are still unsolved and information on key molecules by which DCs can modulate MSC functions is limited. Here, we report that osteopontin (OPN), a cytokine involved in homeostatic and pathophysiologic responses, is constitutively expressed by DCs and regulated in the DC/MSC cocultures depending on the activation state of MSCs. Resting MSCs promoted OPN production, whereas the production of OPN was suppressed when MSCs were activated by proinflammatory cytokines (i.e., TNF-α, IL-6, and IL-1β). OPN induction required cell-to-cell contact, mediated at least in part, by β1 integrin (CD29). Conversely, activated MSCs inhibited the release of OPN *via* the production of soluble factors with a major role played by Prostaglandin E_2 (PGE_2). Accordingly, pretreatment with indomethacin significantly abrogated the MSC-mediated suppression of OPN while the direct addition of exogenous PGE_2inhibited OPN production by DCs. Furthermore, DC-conditioned medium promoted osteogenic differentiation of MSCs with a concomitant inhibition of adipogenesis. These effects were paralleled by the repression of the adipogenic markers PPARγ, adiponectin, and FABP4, and induction of the osteogenic markers alkaline phosphatase, RUNX2, and of the bone-anabolic chemokine CCL5. Notably, blocking OPN activity with RGD peptides or with an antibody against CD29, one of the OPN receptors, prevented the effects of DC-conditioned medium on MSC differentiation and CCL5 induction. Because MSCs have a key role in maintenance of bone marrow (BM) hematopoietic stem cell niche through reciprocal regulation with immune cells, we investigated the possible MSC/DC interaction in human BM by immunohistochemistry. Although DCs ($CD1c^+$) are a small percentage of BM cells, we demonstrated colocalization of $CD271^+$ MSCs with $CD1c^+$ DCs in normal and myelodysplastic BM. OPN reactivity was observed in occasional $CD1c^+$ cells in the proximity of $CD271^+$ MSCs. Altogether, these results candidate OPN as a signal modulated by MSCs according to their activation status and involved in DC regulation of MSC differentiation.

Front Immunol. 2018 May 29;9:1150. doi: 10.3389/fimmu.2018.01150. eCollection 2018.

Targeting the Epidermal Growth Factor Receptor Can Counteract the Inhibition of Natural Killer Cell Function Exerted by Colorectal Tumor-Associated Fibroblasts.

Costa D[1], Venè R[1], Benelli R[2], Romairone E[3], Scabini S[3], Catellani S[4], Rebesco B[5], Mastracci L[6], Grillo F[6], Minghelli S[7], Loiacono F[2], Zocchi MR[8], Poggi A[1].

Author information

Abstract

Mesenchymal stromal cells (MSC) present in the tumor microenvironment [usually named tumor-associated fibroblasts (TAF)] can exert immunosuppressive effects on T and natural killer (NK) lymphocytes, favoring tumor immune escape. We have analyzed this mechanism in colorectal carcinoma (CRC) and found that co-culture of NK cells with TAF can prevent the IL-2-mediated NKG2D upregulation. This leads to the impairment of NKG2D-mediated recognition of CRC cells, sparing the NK cell activation through DNAM1 or FcγRIIIA (CD16). *In situ*, TAF express detectable levels of epidermal growth factor receptor (EGFR); thus, the therapeutic anti-EGFR humanized antibody cetuximab can trigger the antibody-dependent cellular cytotoxicity of TAF, through the engagement of FcγRIIIA on NK cells. Importantly, in the tumor, we found a lymphoid infiltrate containing NKp46^{+}CD3^{-} NK cells, enriched in CD16^{+} cells. This population, sorted and cultured with IL-2, could be triggered *via* CD16 and *via* NKG2D. Of note, *ex vivo* NKp46^{+}CD3^{-} cells were able to kill autologous TAF; *in vivo*, this might represent a control mechanism to reduce TAF-mediated regulatory effect on NK cell function. Altogether, these findings suggest that MSC from the neoplastic mucosa (TAF) of CRC patients can downregulate the immune cell recognition of CRC tumor cells. This immunosuppression can be relieved by the anti-EGFR antibody used in CRC immunotherapy.

Curr Protoc Stem Cell Biol. 2018 Jun 11:e55. doi: 10.1002/cpsc.55. [Epub ahead of print]

A Method for Isolating and Characterizing Mesenchymal Stromal Cell-derived Extracellular Vesicles.

Lo Sicco C[1], Reverberi D[2], Pascucci L[3], Tasso R[4].

Author information

Abstract

The unit describes protocols for isolating and characterizing extracellular vesicles (EVs) derived from human adipose tissue-derived mesenchymal stromal cells (MSCs). EVs are a mixed population of membrane-surrounded structures with overlapping composition and size. Advances made in recent years have led to a better understanding of the biological role of EVs. In particular, they can be considered key factors responsible for MSC-paracrine activity, mediating their anti-inflammatory effects towards innate immune cells, such as macrophages. The topics comprise description of the MSC-conditioned medium containing vesicles preparation, EV isolation, and characterization mainly by specifically set up flow cytometry and electron microscopy approaches, and in vitro methodologies involved in testing the EV anti-inflammatory capacity. The procedures described here can be easily reproduced and can be employed regardless of the type of progenitor cells used to secrete EVs.

Front Immunol. 2018 Feb 19;9:262. doi: 10.3389/fimmu.2018.00262. eCollection 2018.

How to Hit Mesenchymal Stromal Cells and Make the Tumor Microenvironment Immunostimulant Rather Than Immunosuppressive.

Poggi A[1], Varesano S[1], Zocchi MR[2].

Author information

Erratum in

- Corrigendum: How to Hit Mesenchymal Stromal Cells and Make the Tumor Microenvironment Immunostimulant Rather Than Immunosuppressive.[Front Immunol. 2018]

Abstract

Experimental evidence indicates that mesenchymal stromal cells (MSCs) may regulate tumor microenvironment (TME). It is conceivable that the interaction with MSC can influence neoplastic cell functional behavior, remodeling TME and generating a tumor cell niche that supports tissue neovascularization, tumor invasion and metastasization. In addition, MSC can release transforming growth factor-beta that is involved in the epithelial-mesenchymal transition of carcinoma cells; this transition is essential to give rise to aggressive tumor cells and favor cancer progression. Also, MSC can both affect the anti-tumor immune response and limit drug availability surrounding tumor cells, thus creating a sort of barrier. This mechanism, in principle, should limit tumor expansion but, on the contrary, often leads to the impairment of the immune system-mediated recognition of tumor cells. Furthermore, the cross-talk between MSC and anti-tumor lymphocytes of the innate and adaptive arms of the immune system strongly drives TME to become immunosuppressive. Indeed, MSC can trigger the generation of several types of regulatory cells which block immune response and eventually impair the elimination of tumor cells. Based on these considerations, it should be possible to favor the anti-tumor immune response acting on TME. First, we will review the molecular mechanisms involved in MSC-mediated regulation of immune response. Second, we will focus on the experimental data supporting that it is possible to convert TME from immunosuppressive to immunostimulant, specifically targeting MSC.

Bone Joint Res. 2018 May 5;7(4):263-273. doi: 10.1302/2046-3758.74.BJR-2018-0006. eCollection 2018 Apr.

Harnessing extracellular vesicles to direct endochondral repair of large bone defects.

Ferreira E[1], Porter RM[1].

Author information

Abstract

Large bone defects remain a tremendous clinical challenge. There is growing evidence in support of treatment strategies that direct defect repair through an endochondral route, involving a cartilage intermediate. While culture-expanded stem/progenitor cells are being evaluated for this purpose, these

cells would compete with endogenous repair cells for limited oxygen and nutrients within ischaemic defects. Alternatively, it may be possible to employ extracellular vesicles (EVs) secreted by culture-expanded cells for overcoming key bottlenecks to endochondral repair, such as defect vascularization, chondrogenesis, and osseous remodelling. While mesenchymal stromal/stem cells are a promising source of therapeutic EVs, other donor cells should also be considered. The efficacy of an EV-based therapeutic will likely depend on the design of companion scaffolds for controlled delivery to specific target cells. Ultimately, the knowledge gained from studies of EVs could one day inform the long-term development of synthetic, engineered nanovesicles. In the meantime, EVs harnessed from *in vitro* cell culture have near-term promise for use in bone regenerative medicine. This narrative review presents a rationale for using EVs to improve the repair of large bone defects, highlights promising cell sources and likely therapeutic targets for directing repair through an endochondral pathway, and discusses current barriers to clinical translation

Tissue Eng Part A. 2018 Jun 21. doi: 10.1089/ten.TEA.2018.0011. [Epub ahead of print]

Gelatin-based microribbon hydrogels accelerate cartilage formation by mesenchymal stem cells in 3D.

Conrad B[1], Han LH[2], Yang F[3].
Author information

Abstract

Hydrogels are attractive matrices for cell-based cartilage tissue regeneration given their injectability and ability to fill defects with irregular shapes. However, most hydrogels developed to date often lack cell scale macroporosity, which restrains the encapsulated cells, leading to delayed new extracellular matrix deposition restricted to pericellular regions. Further, tissue engineered cartilage using conventional hydrogels generally suffers from poor mechanical property and fails to restore the load-bearing property of articular cartilage. The goal of this study was to evaluate the potential of macroporous gelatin-based microribbon (μRB) hydrogels as novel 3D matrices for accelerating chondrogenesis and new cartilage formation by human mesenchymal stem cells (MSCs) in 3D with improved mechanical properties. Unlike conventional hydrogels, these μRB hydrogels are inherently macroporous and exhibit cartilage-mimicking shock-absorbing mechanical property. After 21 days of culture, MSC-seeded μRB scaffolds exhibit a 20-fold increase in compressive modulus to 225 kPa, a range that is approaching the level of native cartilage. In contrast, HGs only resulted in a modest increase in compressive modulus of 65 kPa. Compared to conventional hydrogels, macroporous μRB scaffolds significantly increased the total amount of neocartilage produced by MSCs in 3D, with improved interconnectivity and mechanical strength. Together, these results validate gelatin-based μRBs as promising scaffolds for enhancing and accelerating MSC-based cartilage regeneration and may be used to enhance cartilage regeneration using other cell types as well.

J Oral Sci. 2018;60(2):221-225. doi: 10.2334/josnusd.17-0187.

The effect of mesenchymal stem cells on chemotaxis of osteoclast precursor cells.

Sumi K[1], Abe T[1], Kunimatsu R[1], Oki N[1], Tsuka Y[1], Awada T[2], Nakajima K[2], Ando K[2], Tanimoto K[2].
Author information

Abstract

Regeneration of tissue, including bone, using mesenchymal stem cells (MSCs) has been progressing rapidly. Regeneration of bone requires the presence of an appropriate environment and efficient chemotaxis of cells to the target site. Differentiation of MSCs into mesenchymal cells has received considerable attention, but the effect of MSCs on chemotaxis is not well understood. In this study, we investigated the effect of MSCs on chemotaxis of RAW264 cells via C-C motif chemokine ligand 2 (CCL2). Balb/c mouse bone marrow-derived MSCs and RAW264 cells, which are osteoclast precursor cells, were co-cultured without cell contact. The gene expression of CCL2 in MSCs and CC-chemokine receptor 2 (CCR2) in RAW264 cells was determined using quantitative real-time PCR. Analysis of RAW264 cell chemotaxis was performed using the Boyden chamber assay. mRNAs for CCL2 and CCR2 were significantly upregulated upon co-culture in comparison to culture of either cell type alone, and the number of chemotactic RAW264 cells was significantly increased by co-culture. MSCs enhanced the chemotaxis of RAW264 cells, possibly via CCL2-CCR2 interaction, suggesting the potential utility of MSCs for tissue regeneration.

Int J Cancer. 2018 Jun 22. doi: 10.1002/ijc.31619. [Epub ahead of print]

The current understanding of mesenchymal stem cells as potential attenuators of chemotherapy-induced toxicity.

Rühle A[1,2], Huber PE[1,2], Saffrich R[3], Lopez Perez R[2], Nicolay NH[2,4].
Author information

Abstract

Chemotherapeutic agents are part of the standard treatment algorithms for many malignancies; however, their application and dosage are limited by their toxic effects to normal tissues. Chemotherapy-induced toxicities can be long-lasting and may be incompletely reversible; therefore, causative therapies for chemotherapy-dependent side effects are needed, especially considering the increasing survival rates of treated cancer patients. Mesenchymal stem cells (MSCs) have been shown to exhibit regenerative abilities for various forms of tissue damage. Preclinical data suggest that MSCs may also help to alleviate tissue lesions caused by chemotherapeutic agents, mainly by establishing a protective microenvironment for functional cells. Due to the systemic administration of most anticancer agents, the effects of these drugs on the MSCs themselves is of crucial importance to utilize stem cell-based approaches for the treatment of chemotherapy-induced tissue toxicities. Here, we present a concise review of the published data regarding the influence of various classes of chemotherapeutic agents on the survival, stem cell characteristics and physiological functions of MSCs. Molecular mechanisms underlying the effects are outlined, and resulting challenges of MSC-based treatments for chemotherapy-induced tissue injuries are discussed.

Iran J Basic Med Sci. 2018 May;21(5):465-468. doi: 10.22038/IJBMS.2018.26152.6420.

An answer to colon cancer treatment by mesenchymal stem cell originated from adipose tissue.

Iplik ES[1], Ertugrul B[2], Kozanoglu I[3], Baran Y[4,5], Cakmakoglu B[2].

Author information

Abstract

OBJECTIVES:

Colon cancer is risen up with its complex mechanism that directly impacts on its treatment as well as its common prevalence. Mesenchymal stem cells (MSCs) have been considered as a therapeutic candidate for conventional disease including cancer. In this research, we have focused on apoptotic effects of adipose tissue-derived MSCs in colon cancer.

MATERIALS AND METHODS:

MSCs were obtained from adipose tissue and characterized by Flowcytometer using suitable antibodies. MSCs, HT-29, HCT-116, RKO and healthy cell line MRC5 were cultured by different seeding procedure. After cell viability assay, changes in caspase 3 enzyme activity and the level of phosphatidylserine were measured.

RESULTS:

For cell viability assay, a 48 hr incubation period was chosen to seed all cells together. There was a 1.36-fold decrease in caspase 3 enzyme activity by co-treatment of RKO and MSCs in addition to 2.02-fold decrease in HT-29 and MSCs co-treatment, and 1.103-fold increase in HCT-116 and MSCs. The results demonstrated that HCT-116 led to the highest rate of apoptotic cell death (7.5%) compared with other cells.

CONCLUSION:

We suggest that MSCs might remain a new treatment option for cancer by its differentiation and repair capacity.

Bioengineering (Basel). 2018 Jun 19;5(2). pii: E48. doi: 10.3390/bioengineering5020048.

Dynamic Cultivation of Mesenchymal Stem Cell Aggregates.

Egger D[1], Tripisciano C[2], Weber V[3], Dominici M[4,5], Kasper C[6].

Author information

Abstract

Mesenchymal stem cells (MSCs) are considered as primary candidates for cell-based therapies due to their multiple effects in regenerative medicine. Pre-conditioning of MSCs under physiological conditions—such as hypoxia, three-dimensional environments, and dynamic cultivation—prior to transplantation proved to optimize their therapeutic efficiency. When cultivated as three-dimensional aggregates or spheroids, MSCs display increased angiogenic, anti-

inflammatory, and immunomodulatory effects as well as improved stemness and survival rates after transplantation, and cultivation under dynamic conditions can increase their viability, proliferation, and paracrine effects, alike. Only few studies reported to date, however, have utilized dynamic conditions for three-dimensional aggregate cultivation of MSCs. Still, the integration of dynamic bioreactor systems, such as spinner flasks or stirred tank reactors might pave the way for a robust, scalable bulk expansion of MSC aggregates or MSC-derived extracellular vesicles. This review summarizes recent insights into the therapeutic potential of MSC aggregate cultivation and focuses on dynamic generation and cultivation techniques of MSC aggregates.

Bone Joint Res. 2018 May 5;7(4):263-273. doi: 10.1302/2046-3758.74.BJR-2018-0006. eCollection 2018 Apr.

Harnessing extracellular vesicles to direct endochondral repair of large bone defects.

Ferreira E[1], Porter RM[1].

Author information

Abstract

Large bone defects remain a tremendous clinical challenge. There is growing evidence in support of treatment strategies that direct defect repair through an endochondral route, involving a cartilage intermediate. While culture-expanded stem/progenitor cells are being evaluated for this purpose, these cells would compete with endogenous repair cells for limited oxygen and nutrients within ischaemic defects. Alternatively, it may be possible to employ extracellular vesicles (EVs) secreted by culture-expanded cells for overcoming key bottlenecks to endochondral repair, such as defect vascularization, chondrogenesis, and osseous remodelling. While mesenchymal stromal/stem cells are a promising source of therapeutic EVs, other donor cells should also be considered. The efficacy of an EV-based therapeutic will likely depend on the design of companion scaffolds for controlled delivery to specific target cells. Ultimately, the knowledge gained from studies of EVs could one day inform the long-term development of synthetic, engineered nanovesicles. In the meantime, EVs harnessed from *in vitro* cell culture have near-term promise for use in bone regenerative medicine. This narrative review presents a rationale for using EVs to improve the repair of large bone defects, highlights promising cell sources and likely therapeutic targets for directing repair through an endochondral pathway, and discusses current barriers to clinical translation