

## Review Article

# Mesenchymal Stem Cells Derived Extracellular Vesicles as a New Cell Free Biological Regenerative Therapy

Christian Jorgensen<sup>1,2\*</sup>

<sup>1</sup>University of Montpellier, IRMB, INSERM, CHU Montpellier, Montpellier, France

<sup>2</sup>Clinical Immunology and Osteoarticular Diseases Therapeutic Unit, Department of Rheumatology, Lapeyronie University Hospital, Montpellier, France

**\*Corresponding author:** C Jorgensen, University of Montpellier, IRMB, INSERM, CHU Montpellier, Inserm U1183, IRMB, Hospital Saint-Eloi, 80 avenue Augustin Fliche, 34295 Montpellier cedex 5, France, E-mail: [christian.jorgensen@inserm.fr](mailto:christian.jorgensen@inserm.fr)

**Received:** 11 May 2021; **Accepted:** 25 June 2021; **Published:** 30 June 2021

**Citation:** Christian Jorgensen. Mesenchymal Stem Cells Derived Extracellular Vesicles as a New Cell Free Biological Regenerative Therapy. International Journal of Applied Biology and Pharmaceutical Technology 12 (2021): 356-361.

### Abstract

This review focus on the therapeutically potential of microvesicles. Extracellular vesicles include exosomes, microvesicles, microparticles, exosomes and apoptotic bodies processed by mesenchymal cells and/or immune cells. They are characterized by their size as well as protein expression as tetraspanins, HSP90, ALIX, TSG101 and Clathrin. These exosomes are actively secreted a long distance from the parental cell. They are highly active through the transportation as “cargo” of nucleotide materials such as micro RNAs, non-coding RNAs, messenger RNAs as well as proteins which can then be delivered to target cells. Several preclinical studies have shown the benefit of vesicles derived from mesenchymal cells due to their regenerative capacity. EV derived from MSC has been developed in ischemic heart disease, liver or kidney damage as well as in neurodegenerative damage or autoimmune diseases. Clinical trials have been conducted in infectious disease in particular lung inflammation related to SARS-Cov2. However, developing MSC derived EV, despite the advantage of a cell free product, there is still some important steps in terms of biological characterization, mode of action, access to large scale production requested for a clinical application.

Extracellular vesicles are lipid vesicles released by many cell types that play an important role in the transfer of biological information into the immediate cellular environment. These extracellular vesicles group together different populations processed by cells including exosomes, microvesicles, microparticles, exosomes and apoptotic bodies [1]. Exosomes are small extracellular vesicles of 50 to 150 nm that arise from endocytosis and belong to the group of intraluminal vesicles. They are then secreted after fusion with the plasma membrane. Microvesicles are larger, from 100 to 1000 nm. They are released into the extracellular environment directly by the plasma membrane. The secretion of these extracellular vesicles is a much conserved mode of intracellular communication between species. Extracellular vesicles are found in eukaryotes but also in plants and microorganisms. In mammals mesenchymal cells and dendritic cells are the main sources of microvesicles with therapeutic potential [2]. However, other sources such as endothelial progenitors, cardiac progenitors or IPS may be important sources to investigate. Mesenchymal cells are multipotent cells with a great capacity for differentiation in different supporting tissues including adipose, muscle or bone tissue. Mesenchymal cells have the potential to regenerate these supporting tissues and have been widely used in different therapeutic trials [3]. Along with the role of paracrine effect including the secretion of cytokines and growth factors, we now know that the expression of microvesicles and exosomes play an important role in the function of these cells.

Dendritic cells are antigen presenting cells that regulate the immune response. The extracellular vesicles derived from these dendritic cells play an important role in the immunological mechanism. Thus, dendritic cells treated with immunosuppressive drugs or cytokines give them immunosuppressive properties. The ability to regulate the inflammatory response is mediated by the expression of exosomes. They have shown their ability to regulate in experimental arthritis [4]. In addition, the genetic modification of dendritic cells to improve their immunoregulatory response leads to the expression of even more effective extracellular vesicles and can be offered as biotherapies in inflammatory and autoimmune diseases.

### **1. Characterization of extracellular vesicles**

Exosomes have an endosomal origin. Their secretion passes by way of endocytosis. They contain elements of intracellular content which gives them characteristics. For example, they contain senexins, tetraspanins like CD63, CD81 and CD9 as well as HSPs like 60, 70 and 90 [5]. They also express ALIX as well as TSG101 and Clathrin (CLATHRIN). These exosomes are encapsulated in a bi-lipid layer which allows them to be secreted a long distance from the parental cell. The membrane contains serine phosphatidyls as well as ceramides and sphingolipids. These exosomes make it possible to transport as “cargo” nucleotide materials such as microRNAs, non-coding RNAs, messenger RNAs as well as proteins which can then be delivered to target cells [6]. When the exosomes are released into their extracellular environment, they will interact through these numerous biological, enzymatic, protein or nucleotide factors.

### **2. What is the most suitable cellular source?**

Depending on the therapeutic target, the choice of an autologous or allogeneic parental cell should be considered. For large-scale use in universal exosome production, the consensus is to use an allogeneic source with possibly

genetic modification of the stem cell to avoid HLA class II molecule expression. The administration of exosomes will be adapted to the therapeutic demand. It can be injected locally if the site of interest is accessible and very localized. Thus in therapeutic proposals such as cardiac ischemia or the osteoarticular field, local application can be proposed. In oncology, early clinical trials of immunization using MAGE3 peptides in melanoma have been proposed [7, 8]. Exosomes derived from dendritic cells have also been proposed to stimulate the NK response and the T response in cancer immunotherapy. In this case, systemic administration is preferred.

### 3. Therapeutic applications

Several preclinical studies have shown the benefit of vesicles derived from mesenchymal cells due to their regenerative capacity. This has been proposed in ischemic heart disease, liver or kidney damage as well as in neurodegenerative damage. In models of chronic inflammation or autoimmune diseases there is also an interest in developing this strategy.

Thus, authors show that extracellular vesicles derived from mesenchymal cells have cardio-protective capacities. Vesicles derived from embryonic cells have been shown in a mouse model to reduce the size of ischemia and reduce oxidative stress [7]. Thus, the pathways of apoptosis were inhibited in myocardial tissue. At the same time, there is a reduction in local inflammation and macrophage infiltrates that contribute to early stage cardiac lesions. Finally, neoangiogenesis appears to be stimulated by injection of EV intracardiac.

In neurodegenerative conditions, injections of mesenchymal cell-derived EVs have shown intravenous administration to provide neurological recovery in cerebral ischemic models. It appears that the expression of micro RNA 133bin astrocytes and neurons has an interesting therapeutic effect [9]. Finally, EVs enriched with mRNA 17 could accelerate neurogenesis and functional recovery by suppressing the expression of PTEN.

In fibrosing hepatopathies, vesicles derived from mesenchymal cells would provide a protective effect on hepatocytes. Inactivation of the TGFB1 pathway is observed. In models of liver injury induced by CCL4 expression, extracellular microvesicles derived from mesenchymal cells accelerated liver regeneration by increasing cyclin D1 and reducing apoptosis induced by PCL [10].

In autoimmune diseases and particularly in models of lupus, extracellular vesicles derived from mesenchymal cells have a beneficial effect by inducing the expression of anti-inflammatory cytokines [11]. Thus, the FAS receptor transferred by exosomes reduced immunological damage. In an experimental arthritis model, extracellular vesicles allow the transfer of micro RNAs such as miR150 therapeutically [12]. This makes it possible to obtain a reduction in the expression of metalloproteases, inflammatory and angiogenic factors in the synovium after local injection. This leads to a reduction in pro-inflammatory joint cytokines and ultimately to an improvement in clinical scores in an experimental arthritis model.

Finally, in models of induced osteoarthritis, vesicles derived from mesenchymal cells have potential therapeutic benefit. We and other were able to show that both exosomes and microvesicles derived from mesenchymal cells were able to obtain a therapeutic effect comparable to parental cells in a model of osteoarthritis induced by the injection of collagenase [13, 14]. The reduction in the histological score of osteosclerosis or osteophytosis as well as the reduction in inflammatory scores showed the beneficial interest of this strategy. In addition, the immunomodulating and chondroprotective effect on chondrocytes is confirmed in particular using MRL mouse models [15]. In this model, the beneficial character is linked to the transfer of micro RNAs allowing to obtain an effect of proliferation of chondrocytes and chondroprotectors in the experimental model. Comparative analyzes of the micro RNA content in the exosomes by comparing the MRL mice with hyper-healing compared to the control mice show an increase in particular of the micro RNA 221-3p which could explain the observed therapeutic benefit. Moreover, in disease associated with ageing like OA, EVs released by senescent cells contribute to the alteration of the tissue. We observed changes in the number and composition of EVs released by senescent cells [16]. In addition, the age-related alterations in mesenchymal stem/stromal cells (MSCs) directly contributes to aging, through alteration in EV mitochondrial content. Age-associated decline of respiring mitochondria cargo in EVs of circulating immune cells lead to immunosenescence [17].

SARS-CoV-2 pandemic has accelerated the EV clinical development, either as a biological product to prevent lung inflammation or as a vaccine strategy. Extracellular vesicles exposing ACE2 are efficient decoys for SARS-CoV-2 spike S-protein. Reduction of infectivity positively correlates with the level of ACE2 more efficient than with soluble ACE2 [18]. Recent clinical data confirmed feasibility of administration of EV as therapeutic agent [19].

#### 4. Perspectives

EVs derived from mesenchymal cells mimic the effect of parental cells and has the advantage of a biological therapy without cells and reduces the risk for patients. Moreover, EV are not classified as ATMP and therefore facilitate regulatory dossier. The use of extracellular vesicles derived from mesenchymal cells or mesenchymal cells derived from IPS has significant therapeutic potential. However, studies on the mechanism of action, pharmacokinetics and biodistribution of these extracellular EV release in the joint environment should be performed before initiating clinical trials. This will be supported through interdisciplinary work between physicians, biologists and chemists that will allow us to integrate fundamental knowledge on the functions of extracellular vesicles and the technologies to deliver them in order to optimize large-scale production and their targeted distribution. The therapeutic promises by these new biotherapy strategies are promising for the future.

#### 5. References

1. Keshtkar S, Azarpira N, Hossein Ghahremani M. Mesenchymal stem cell-derived extracellular vesicles: novel frontiers in regenerative medicine. *Stem Cell Res Ther* 9 (2018): 63.
2. Harrell, Carl Randall et al. "Mesenchymal Stem Cell-Derived Exosomes and Other Extracellular Vesicles as New Remedies in the Therapy of Inflammatory Diseases." *Cells* vol 8 (2019): 1605.
3. Alcaraz, María José et al. "Extracellular Vesicles from Mesenchymal Stem Cells as Novel Treatments for

- Musculoskeletal Diseases.” *Cells* vol 9 (2019): 98.
4. Cosenza S, Toupet K, Maumus M, Luz-Crawford P, Blanc-Brude O, Jorgensen C, Noël D. Mesenchymal stem cells-derived exosomes are more immunosuppressive than microparticles in inflammatory arthritis. *Theranostics* 8 (2018): 1399-1410.
  5. Jankovičová, J., Sečová, P., Michalková, K., Antalíková, J. Tetraspanins, More than Markers of Extracellular Vesicles in Reproduction. *International journal of molecular sciences* 21 (2020): 7568.
  6. Qiu G, Zheng G, Ge M, Wang J, Huang R, Shu Q and Xu J. Mesenchymal stem cell-derived extracellular vesicles affect disease outcomes via transfer of microRNAs. *Stem Cell Research & Therapy* 9 (2018): 320.
  7. Teng X, Chen L, Chen W, Yang J, Yang Z, Shen Z. Mesenchymal Stem Cell-Derived Exosomes Improve the Microenvironment of Infarcted Myocardium Contributing to Angiogenesis and Anti-Inflammation. *Cell Physiol Biochem* 37 (2015): 2415-24.
  8. Lener T, Gimona M, Aigner L, Börger V, Buzas E, Camussi G, Chaput N, Chatterjee D, Court FA et al. Applying extracellular vesicles based therapeutics in clinical trials - an ISEV position paper. *Journal of extra cellular vesicles* 4 (2015): 30087.
  9. Xin H, Li Y, Liu Z, Wang X, Shang X, Cui Y, Zhang ZG, Chopp M. MiR-133b promotes neural plasticity and functional recovery after treatment of stroke with multipotent mesenchymal stromal cells in rats via transfer of exosome-enriched extra cellular particles, *Stem Cells* 31 (2013): 2737–46.
  10. Tan CY, Lai RC, Wong W, Dan YY, Lim S-K, Ho HK. Mesenchymal stem cell-derived exosomes promote hepatic regeneration in drug-induced liver injury models. *Stem Cell Res Ther* 5 (2014): 76.
  11. Perez-Hernandez, Javier et al. “Extracellular Vesicles as Therapeutic Agents in Systemic Lupus Erythematosus.” *International journal of molecular sciences* 18 (2017): 717.
  12. Liu H, Li R, Liu T, Yang L, Yin G, Xie Q. Immuno modulatory Effects of Mesenchymal Stem Cells and Mesenchymal Stem Cell-Derived Extracellular Vesicles in Rheumatoid Arthritis. *Front. Immunol* 11 (2020): 1912.
  13. Cosenza, Stella et al. “Pathogenic or Therapeutic Extracellular Vesicles in Rheumatic Diseases: Role of Mesenchymal Stem Cell-Derived Vesicles.” *International journal of molecular sciences* 18 (2017): 889.
  14. Li, Jiao et al. “Stem Cell-Derived Extracellular Vesicles for Treating Joint Injury and Osteoarthritis.” *Nano materials (Basel, Switzerland)* 9 (2019): 261.
  15. Wang, Rikang et al. “Intra-articular delivery of extracellular vesicles secreted by chondrogenic progenitor cells from MRL/MpJsuperhealer mice enhances articular cartilage repair in a mouse injury model.” *Stem cell research & therapy* 11 (2020): 93.
  16. Boulestreau J, Maumus M, Rozier P, Jorgensen C, Noël D. Mesenchymal Stem Cell Derived Extracellular Vesicles in Aging. *Front Cell Dev Biol* 8 (2020): 107.
  17. Zhang X, Hubal MJ, Kraus VB. Immune cell extracellular vesicles and their mitochondrial content decline with ageing. *Immunity & ageing* 17 (2020): 17-1.
  18. Federico Coccozza, Ester Piovesana, Nathalie Névo, Xavier Lahaye, Julian Buchrieser, Olivier Schwartz, Nicolas Manel, Mercedes Tkach, ClotildeThéry, Lorena Martin-Jaular. Extracellular vesicles containing ACE2 efficiently prevent infection by SARS-CoV-2 Spike protein-containing virus. *Bio Rxiv* (2020):

193672.

19. SaborniChakraborty, VamseeMallajosyula, Cristina M. Tato, Gene S. Tan, Taia T. Wang. SARS-CoV-2 vaccines in advanced clinical trials: Where do we stand?Advanced Drug Delivery Reviews 172 (2021): 314-338.



This article is an open access article distributed under the terms and conditions of the [Creative Commons Attribution \(CC-BY\) license 4.0](https://creativecommons.org/licenses/by/4.0/)