Acta Tecnología - International Scientific Journal about Technologies
Volume: 10 2024 Issue: 4 Pages: 179-183 ISSN 2453-675X

https://doi.org/10.22306/atec.v10i4.245 Received: 29 Oct. 2024; Revised: 17 Nov. 2024; Accepted: 02 Dec. 2024

The biotechnology of using mesenchymal stem cells in regenerative medicine

Eva Slaba
Department of Medical Biology, Faculty of Medicine, P. J. Safarik University in Kosice, Trieda SNP 1, 040 11 Košice, Slovak Republic, EU, eva.slaba@upjs.sk

Keywords: mesenchymal stem cells, regenerative medicine, biocompatibility, technology.
Abstract: Mesenchymal stem cells (MSCs) have garnered significant attention in regenerative medicine due to their multipotent capabilities and ability to differentiate into various cell types, including osteocytes, chondrocytes, and adipocytes. Sourced from bone marrow, adipose tissue, umbilical cord blood, and other tissues, MSCs possess immunomodulatory properties, making them ideal candidates for tissue repair and therapeutic applications. Their capacity to migrate to sites of injury and secrete bioactive molecules that promote tissue regeneration and inhibit inflammation is crucial in treating a range of conditions. Recent advancements have highlighted MSCs' role in the regeneration of bone, cartilage, cardiac tissue, and neural networks. They are also being explored in the treatment of degenerative diseases such as osteoarthritis, myocardial infarction, and neurodegenerative disorders. Despite the promising therapeutic potential, several challenges remain, such as optimizing cell delivery methods, understanding long-term effects, and addressing regulatory hurdles for clinical applications. This article aims to introduce mesenchymal stem cells and their main uses in regenerative medicine, offering a comprehensive overview of their biological properties, current clinical applications, and the potential challenges that need to be addressed for broader therapeutic use.

1 Introduction

Regenerative medicine aims to restore or replace damaged tissues and organs by harnessing the body's natural repair mechanisms. Mesenchymal stem cells (MSCs), due to their ability to differentiate into multiple cell types, self-renew, and modulate the immune response, are one of the most extensively studied cell types in regenerative medicine (Figure 1) [1]. Originally identified in bone marrow, MSCs can also be isolated from other tissues, including adipose tissue, umbilical cord, dental pulp, and even more recently explored sources like placenta and peripheral blood [2,3]. These alternative sources provide easier access to MSCs, often with a less invasive harvesting procedure and higher proliferative capacity, which broadens their potential for widespread clinical applications [4]. The therapeutic utility of MSCs has been explored in diverse clinical settings, including orthopedic, cardiovascular, neurodegenerative, and inflammatory conditions [5]. MSCs have shown the ability to promote tissue repair through direct differentiation into osteoblasts, chondrocytes, and myocytes in conditions like osteoarthritis, bone fractures, and myocardial infarction [6]. Furthermore, their immunomodulatory properties have made them attractive candidates for treating autoimmune diseases, graft-versus-host disease, and neurodegenerative disorders [7,8]. Their role in modulating inflammation and promoting tissue repair by secreting bioactive molecules, such as cytokines, growth factors, and extracellular vesicles, has expanded their use beyond simple tissue engineering [9].

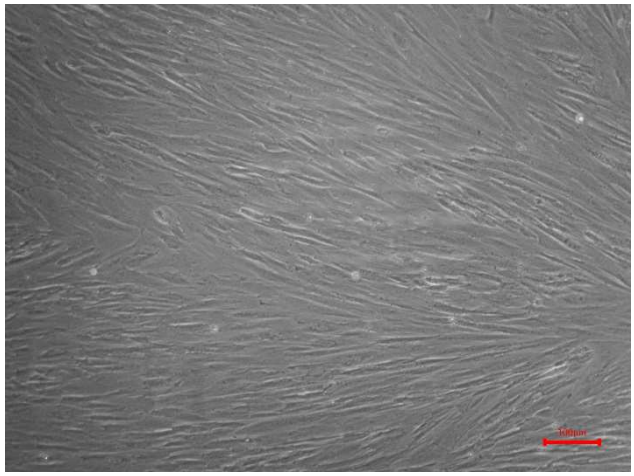

Figure 1 Typical enloged shape of mesenchymal stem cells

2 Definiton of stem cells

MSCs are loosely defined as cells that have the ability to differentiate into many different cell types while retaining the ability to self-replicate and clonogenically grow [2,10]. MSCs are characterized by their adherence to a plastic surface during in vitro culture, differentiation potential and a broadly defined phenotype. The expression of surface features typical for MSCs is as follows: CD105+, CD73+, CD90+ in the population of in vitro cultured cells and at the same time the absence of hematopoietic features CD45-, CD34-, CD14-, CD11b-, CD79-, CD19- and MHC class II molecules. In addition to the above features, MSCs express the following features: CD9+, CD29+, CD44+, CD49+, CD54+, CD61+, CD63+, CD71+, CD97+, CD98+, CD99+, CD106+, CD146+, CD155+, Stro-1+, CD166+, CD166+, CD271+, CD276+

and CD304+ [5]. Adult MSCs have been isolated from many tissues, especially bone marrow, adipose tissue, liver or muscle. In the last decade, MSCs have been isolated from perinatal tissues (Table 1). These tissues are directly related to the development of the embryo and its growth. Given the limitations of the source of stem cells, perinatal tissues represent an important and promising source of MSCs. Postpartum tissues can be used for their isolation, such as placenta, amniotic membranes, amnion or chorion [11-13].

Table 1 Sources of MSCs [5,14,15]

Source	Difficulty to Obtain	Quantity Obtainable (Relative)	Comments on Isolation
Bone Marrow	Moderate	Low to Moderate	Invasive procedure
Adipose Tissue	Low	High	Easier to collect using liposuction
Umbilical Cord	Low	Moderate	Non-invasive collection during childbirth
Dental Pulp	Moderate	Low	Requires tooth extraction
Placenta	Low	High	Non-invasive collection post-delivery
Amniotic Fluid	High	Low to Moderate	Requires amniocentesis; invasive and risky
Peripheral Blood	High	Low	Requires mobilization agents or extensive processing to yield MSCs

3 Mechanisms of mesenchymal stem cells in regenerative medicine

MSCs therapeutic potential is not limited to their ability to differentiate into various cell types; they also play a crucial role in modulating the immune system and promoting healing through the secretion of bioactive molecules. MSCs secrete a range of bioactive molecules through paracrine signaling, which promotes healing, reduces apoptosis, and stimulates angiogenesis. These mechanisms, acting in synergy, underscore the versatility and promise of MSCs in clinical applications [16].

3.1 Differentiation capacity

One of the hallmark properties of MSCs is their ability to differentiate into various cell types, including osteoblasts, chondrocytes, adipocytes, and even neurons under specific conditions. MSCs from different tissue sources exhibit varying differentiation potentials, which may influence their effectiveness in clinical applications [17]. For instance, bone marrow-derived MSCs (BM-MSCs) are known to be particularly adept at differentiating into bone cells, making them valuable for orthopedic applications [10], while adipose-derived MSCs (AD-MSCs) have shown potential in wound healing and skin regeneration [18]. Recent studies have explored how MSC differentiation is influenced by extracellular cues such as growth factors, matrix stiffness, and oxygen tension [19,20]. Understanding these factors is critical for developing effective MSC-based therapies, as they can determine the fate of MSCs in the microenvironment of injured tissues. For example, low oxygen tension (hypoxia) has been shown to enhance the differentiation of MSCs into cartilage and reduce their differentiation into adipocytes, which may benefit cartilage regeneration [21].

3.2 Immunomodulatory and anti-inflammatory effects

In addition to their differentiation abilities, MSCs exert significant immunomodulatory effects, making them useful in treating inflammatory and autoimmune diseases. MSCs can suppress the proliferation of immune cells such as T-cells, B-cells, natural killer cells, and dendritic cells through direct cell-cell contact and the secretion of anti-inflammatory cytokines like interleukin-10 (IL-10) and transforming growth factor-beta (TGF-β) [15,22]. MSCs are also known to influence the polarization of macrophages, shifting them from a pro-inflammatory (M1) to an anti-inflammatory (M2) phenotype, which can promote tissue repair and wound healing [23]. This property has been leveraged in the treatment of graft-versus-host disease (GVHD), where MSCs have been shown to reduce inflammation and promote graft acceptance in allogeneic transplants [7].

4 Applications of mesenchymal stem cells in regenerative medicine

4.1 Orthopedic applications

The use of MSCs in orthopedic applications has shown promise in treating bone defects, cartilage injuries, and osteoarthritis. Several studies have demonstrated that MSCs can enhance bone regeneration when combined with scaffolds or biomaterials, allowing for the formation of new bone tissue in critical-size defects [11-13]. In cartilage repair, MSCs have been shown to differentiate into chondrocytes under appropriate conditions, leading to the restoration of damaged cartilage in osteoarthritic joints. A significant body of research supports the use of MSCs in intra-articular injections for osteoarthritis, where they reduce inflammation and promote cartilage regeneration [6] (Table 2).

Table 2 Examples of orthopedic applications of MSCs [6,24,25]

Disease/Condition	MSC Source	Therapeutic Outcome
Osteoarthritis	Bone marrow	Enhanced cartilage regeneration and reduced inflammation
Non-union fractures	Bone marrow	Accelerated bone healing
Spinal disc degeneration	Adipose tissue	Reduced disc degeneration and improved mobility

4.2 Cardiovascular applications

MSCs have also shown significant potential in treating cardiovascular diseases, particularly myocardial infarction (MI). MSCs can contribute to cardiac repair by differentiating into cardiomyocytes, secreting pro-angiogenic factors that promote neovascularization, and reducing fibrosis in damaged cardiac tissue. Several preclinical and clinical studies have demonstrated that MSCs improve cardiac function and reduce scar size in patients following MI [26]. However, challenges remain in enhancing MSC engraftment and survival in the hostile environment of ischemic cardiac tissue. Strategies such as preconditioning MSCs with hypoxia or using biomaterial scaffolds to deliver MSCs have shown promise in improving their therapeutic efficacy [27].

4.3 Neurological applications

MSC-based therapies are being explored for a range of neurodegenerative and neuroinflammatory conditions, including multiple sclerosis (MS), Parkinson's disease, and spinal cord injury. In the case of MS, MSCs have been shown to reduce inflammation, promote remyelination, and support the survival of neurons in preclinical models [28]. Their ability to cross the blood-brain barrier and their immunomodulatory properties make them attractive candidates for treating central nervous system (CNS) disorders. Recent advances in the use of MSCs for spinal cord injury (SCI) repair highlight their ability to promote axonal regeneration, reduce glial scar formation, and improve functional recovery. While clinical trials are ongoing, the preclinical data support the potential of MSCs to provide significant benefits for patients with SCI [29].

5 Future directions

The field of mesenchymal stem cells in regenerative medicine has made significant strides, but there is still much to be explored in enhancing their therapeutic efficacy and expanding their clinical applications. Ongoing research is focusing on optimizing MSC delivery methods [4], improving their engraftment and survival [8], and overcoming challenges related to heterogeneity and scaling up for clinical use. Advances in biotechnology, such as 3D printing and biocompatibility analysis using stem cells, hold great potential for pushing the boundaries of MSC-based therapies.

5.1 Use of stem cells in biocompatibility analysis

As the field of biomaterials grows, the need for accurate and reliable methods to assess biocompatibility is critical. Traditionally, biocompatibility testing involved the use of animal models or isolated cell lines, but the advent of MSC-based biocompatibility analysis offers a more sophisticated and physiologically relevant approach. MSCs, due to their ability to mimic various tissue types, can be used as a model system to assess how biomaterials interact with living cells [11,30]. Mesenchymal stem cells are ideal candidates for biocompatibility testing because of their sensitivity to environmental cues and their ability to differentiate into various cell types. This makes MSCs highly suitable for assessing the cytotoxicity, cell adhesion, proliferation, and differentiation capabilities of novel biomaterials. For example, a biocompatibility study using MSCs can evaluate whether a material promotes or inhibits MSC adhesion and proliferation, which are crucial factors in determining its suitability for tissue engineering applications [12,13]. Recent studies have used MSCs to test the biocompatibility of various scaffolds, hydrogels, and implants designed for tissue regeneration. The outcomes of these tests not only determine the material's safety but also provide insights into how well the material supports MSC-mediated tissue repair [11,31].

5.2 3D Printing using MSCs

One of the most exciting frontiers in regenerative medicine is the integration of mesenchymal stem cells with 3D printing technologies. This approach, also known as "bioprinting," involves the use of stem cells to create three-dimensional, tissue-like structures that mimic the natural architecture of human tissues. MSCs, due to their ability to differentiate into multiple cell types, are particularly well-suited for this technology, allowing researchers to engineer complex tissues such as bone, cartilage, and even organs [32]. Bioprinting using offers several advantages over traditional tissue engineering approaches. First, it allows for precise control over the spatial arrangement of cells, which is critical for replicating the intricate microenvironment of tissues. Second, bioprinting enables the incorporation of bioactive materials, such as growth factors and scaffolds, which can enhance MSCs differentiation and promote tissue maturation. These advancements have significant implications for regenerative medicine, particularly in developing personalized therapies tailored to individual patients [30]. Recent studies have demonstrated the potential of 3D-printed MSC constructs for cartilage regeneration, where printed scaffolds seeded with MSCs were shown to promote cartilage tissue formation and repair in osteoarthritic models [33-35].

6 Conclusion

The future of mesenchymal stem cell-based therapies lies in advancing both the technologies used to harness their potential and the strategies to overcome the current limitations [30]. Innovations such as 3D bioprinting and the use of MSCs in biocompatibility testing represent critical steps forward in optimizing MSC-based therapies for clinical use. As research continues, the combination of MSCs with emerging technologies will undoubtedly expand the therapeutic applications of stem cells, bringing regenerative medicine closer to fully restoring damaged tissues and organs in a range of medical conditions [5]. These future directions hold the key to unlocking the full regenerative potential of MSCs, making them a cornerstone in the evolution of modern medicine.

References

[1] ORYAN, A., KAMALI, A., MOSHIRI, A., ESLAMINEJAD, M.B.: Role of mesenchymal stem cells in bone regenerative medicine: what is the evidence?, *Cells Tissues Organs*, Vol. 204, No. 2, pp. 59-83, 2017. https://doi.org/10.1159/000469704

[2] ZACHAR, L., BAČENKOVÁ, D., ROSOCHA, J.: Activation, homing, and role of the mesenchymal stem cells in the inflammatory environment, *Journal of Inflammation Research*, Vol. 15, No. 9, pp. 231-240, 2016. https://doi.org/10.2147/JIR.S121994

[3] CAPLAN, A.I.: *Chapter 15 - Mesenchymal Stem Cells in Regenerative Medicine*, Principles of Regenerative Medicine, Elsevier, pp. 219-227, 2019. https://doi.org/10.1016/B978-0-12-809880-6.00015-1

[4] ZHAO, W., LI, K., LI, L., WANG, R., LEI, Y., YANG, H., SUN, L.: Mesenchymal Stem Cell-Derived Exosomes as Drug Delivery Vehicles in Disease Therapy, *International Journal of Molecular Sciences*, Vol. 25, No. 14, 7715, pp. 1-26, 2024. https://doi.org/10.3390/ijms25147715

[5] PATEL, D.M., SHAH, J., SRIVASTAVA, A.S.: Therapeutic potential of mesenchymal stem cells in regenerative medicine, *Stem Cells International*, Vol. 2013, No. 1, 496218, 2013. https://doi.org/10.1155/2013/496218

[6] BAČENKOVÁ, D., TREBUŇOVÁ, M., DEMETEROVÁ, J., ŽIVČÁK, J.: Human chondrocytes, metabolism of articular cartilage, and strategies for application to tissue engineering, *International Journal of Molecular Sciences*, Vol. 24, No. 23, 17096, pp. 1-25, 2023. https://doi.org/10.3390/ijms242317096

[7] KADRI, N., AMU, S., IACOBAEUS, E., BOBERG, E., LE BLANC, K.: Current perspectives on mesenchymal stromal cell therapy for graft versus host disease, *Cellular & Molecular Immunology*, Vol. 20, No. 6, pp. 613-625, 2023.

[8] GARG, A., KHAN, S., LUU, N., NICHOLAS, D.J., DAY, V., KING, A.L., FEAR, J., LALOR, P.F., NEWSOME, P.N.: TGFβ1 priming enhances CXCR3-mediated mesenchymal stromal cell engraftment to the liver and enhances anti-inflammatory efficacy, *Journal of Cellular and Molecular Medicine*, Vol. 27, No. 6, pp. 864-878, 2023. https://doi.org/10.1111/jcmm.17698

[9] BAGNO, L., HATZISTERGOS, K.E., BALKAN, W., HARE, J.M.: Mesenchymal stem cell-based therapy for cardiovascular disease: progress and challenges, *Molecular Therapy*, Vol. 26, No. 7, pp. 1610-1623, 2018.

[10] BAČENKOVÁ, D., TREBUŇOVÁ, M., MOROCHOVIČ, R., DOSEDLA, E., BALOGOVÁ, A., GAŠPAROVÁ, P., ŽIVČÁK, J.: Interaction between mesenchymal stem cells and the immune system in rheumatoid arthritis, *Pharmaceuticals*, Vol. 15, No. 8, p. 941, 2022. https://doi.org/10.3390/ph15080941

[11] BAČENKOVÁ, D., DEMETEROVÁ, J., DOSEDLA, E., GAŠPAROVÁ, P., TREBUŇOVÁ, M.: The use of alginate in the cultivation of mesenchymal cells in biomedical engineering, *Acta Technologia*, Vol. 10, No. 2, pp. 45-49, 2024. https://doi.org/10.22306/atec.v10i2.198

[12] TREBUŇOVÁ, M., PETROUŠKOVÁ, P., BALOGOVÁ, A., IŽARÍKOVÁ, G., HORŇÁK, P., BAČENKOVÁ, D., DEMETEROVÁ, J., ŽIVČÁK, J.: Evaluation of biocompatibility of PLA/PHB/TPS polymer scaffolds with different additives of ATBC and OLA plasticizers, *Journal of Functional Biomaterials*, Vol. 14, No. 8, 412, pp. 1-22, 2023. https://doi.org/10.3390/jfb14080412

[13] TREBUŇOVÁ, M., GROMOŠOVÁ, S., BAČENKOVÁ, D., ROSOCHA, J., ŽIVČÁK, J.: Biocompatibility of the human mesenchymal stem cells with bovine bone tissue at the cellular level in vitro, *Clinican and Technology*, Vol. 48, No. 2, pp. 59-65, 2018.

[14] TROHATOU, O., ROUBELAKIS, M.G.: Mesenchymal stem/stromal cells in regenerative medicine: past, present, and future, *Cellular reprogramming*, Vol. 19, No. 4, pp. 217-224, 2017.

[15] BAČENKOVÁ, D., TREBUŇOVÁ, M., ČIŽKOVÁ, D., HUDÁK, R., DOSEDLA, E., BALOGOVÁ, A., ŽIVČÁK, J.: In vitro model of human trophoblast in early placentation, *Biomedicines*, Vol. 10, No. 4, 904, 2022. https://doi.org/10.3390/biomedicines10040904

[16] AUGELLO, A., DE BARI, C.: The regulation of differentiation in mesenchymal stem cells, *Human Gene Therapy*, Vol. 21, No. 10, pp. 1226-1238, 2010. https://doi.org/10.1089/hum.2010.173

[17] BEREBICHEZ-FRIDMAN, R., MONTERO-OLVERA, P.R.: Sources and clinical applications of mesenchymal stem cells: state-of-the-art review, *Sultan Qaboos University Medical Journal*, Vol. 18, No. 3, p. e264, 2018.

[18] MAZINI, L., ROCHETTE, L., AMINE, M., MALKA, G.: Regenerative capacity of adipose derived stem cells (ADSCs), comparison with mesenchymal stem cells (MSCs), *International Journal of Molecular Sciences*, Vol. 20, No. 10, p. 2523, 2019.

[19] ZHANG, X., ZHANG, S., WANG, T.: How the mechanical microenvironment of stem cell growth affects their differentiation: A review, *Stem cell research & therapy*, Vol. 13, No. 1, p. 415, 2022.

[20] KUSUMA, G.D., CARTHEW, J., LIM, R., FRITH, J.E.: Effect of the microenvironment on mesenchymal stem cell paracrine signaling: opportunities to engineer the therapeutic effect, *Stem cells and development*, Vol. 26, No. 9, pp. 617-631, 2017.

[21] LAMMI, M.J., QU, C.: Regulation of oxygen tension as a strategy to control chondrocytic phenotype for cartilage tissue engineering and regeneration, *Bioengineering*, Vol. 11, No. 3, p. 211, 2024.

[22] GAŠPAROVÁ, P., BALLOVÁ, Z., BAČENKOVÁ, D., TREBUŇOVÁ, M., DOSEDLA, E.: Trophoblast stem cells, trophoblast invasion, and organoids – advancements in gynecology, *Czech gynecology = Journal of the Czech gynecology and obstetrics society*, Vol. 89, No. 2, pp. 151-155, 2024. https://doi.org/10.48095/cccg2024151

[23] YANG, G., FAN, X., LIU, Y., JIE, P., MAZHAR, M., LIU, Y., DECHSUPA, N., WANG, L.: Immunomodulatory mechanisms and therapeutic potential of mesenchymal stem cells, *Stem Cell Reviews and Reports*, Vol. 19, No. 5, pp. 1214-1231, 2023.

[24] HARRELL, C.R., MARKOVIC, B.S., FELLABAUM, C., ARSENIJEVIC, A., VOLAREVIC, V.: Mesenchymal stem cell-based therapy of osteoarthritis: Current knowledge and future perspectives', *Biomedicine & pharmacotherapy*, Vol. 109, pp. 2318-2326, 2019.

[25] GRANERO-MOLTO, F., WEIS, J.A., LONGOBARDI, L., SPAGNOLI, A.: Role of mesenchymal stem cells in regenerative medicine: application to bone and cartilage repair, *Expert opinion on biological therapy*, Vol. 8, No. 3, pp. 255-268, 2008.

[26] ALA, M.: The beneficial effects of mesenchymal stem cells and their exosomes on myocardial infarction and critical considerations for enhancing their efficacy, *Ageing Research Reviews*, Vol. 89, 101980, 2023.

[27] YUCE, K.: The Application of Mesenchymal Stem Cells in Different Cardiovascular Disorders: ways of Administration, and the Effectors, *Stem Cell Reviews and Reports*, Vol. 20, No. 7, pp. 1671-1691, 2024.

[28] GHASEMI, M., ROSHANDEL, E., MOHAMMADIAN, M., FARHADIHOSSEINABADI, B., AKBARZADEHLALEH, P., SHAMSASENJAN, K.: Mesenchymal stromal cell-derived secretome-based therapy for neurodegenerative diseases: overview of clinical trials, *Stem cell research & therapy*, Vol. 14, No. 1, p. 122, 2023.

[29] MONTOTO-MEIJIDE, R., MEIJIDE-FAÍLDE, R., DÍAZ-PRADO, S.M., MONTOTO-MARQUÉS, A.: Mesenchymal stem cell therapy in traumatic spinal cord injury: a systematic review, *International Journal of Molecular Sciences*, Vol. 24, No. 14, p. 11719, 2023.

[30] BALOGOVÁ, A.F., TREBUŇOVÁ, M., RAJŤUKOVÁ, V., HUDÁK, R.: Fresh method: 3D bioprinting as a new approach for tissue and organ regeneration, *Acta Tecnologia*, Vol. 7, No. 3, pp. 79-82, 2021. https://doi.org/10.22306/atec.v7i3.112

[31] TREBUŇOVÁ, M., DEMETEROVÁ, J., BAČENKOVÁ, D., RAJŤUKOVÁ, V., ŽIVČÁK, J.: Surface biocompatibility of porous titanium structures with stem cells, *Lékař a technika Clinician and Technology*, Vol. 53, No. 1, pp. 15-18, 2023. https://doi.org/10.14311/CTJ.2023.1.03

[32] BARI, E., DI GRAVINA, G.M., SCOCOZZA, F., PERTEGHELLA, S., FRONGIA, B., TENGATTINI, S., SEGALE, L., TORRE, M.L., CONTI, M.: Silk Fibroin Bioink for 3D Printing in tissue regeneration: controlled release of MSC extracellular vesicles, *Pharmaceutics*, Vol. 15, No. 2, 383, pp. 1-19, 2023. https://doi.org/10.3390/pharmaceutics15020383

[33] SUN, T., FENG, Z., HE, W., LI, C., HAN, s., LI, Z., GUO, R.: Novel 3D-printing bilayer GelMA-based hydrogel containing BP, β-TCP and exosomes for cartilage–bone integrated repair, *Biofabrication*, Vol. 16, No. 1, p. 015008, 2023.

[34] MESSAOUDI, O., HENRIONNET, C., BOURGE, K., LOEUILLE, D., GILLET, P., PINZANO, A.: Stem cells and extrusion 3D printing for hyaline cartilage engineering, *Cells*, Vol. 10, No. 1, 2, pp. 1-24, 2020. https://dx.doi.org/10.3390/cells10010002

[35] COSTANTINI, M., IDASZEK, J., SZÖKE, K., JAROSZEWICZ, J., DENTINI, M., BARBETTA, A., BRINCHMANN, J.E., ŚWIĘSZKOWSKI, W.: 3D bioprinting of BM-MSCs-loaded ECM biomimetic hydrogels for in vitro neocartilage formation, *Biofabrication*, Vol. 8, No. 3, p. 035002, 2016.

Review process
Single-blind peer review process.