Review
UDC 576.3/.7+57.085.23+577.1/.2
https://doi.org/10.56871/RBR.2026.14.44.006

CC BY 4.0

Constructing products based on extracellular vesicles obtained from mesenchymal stem cells

Rodion V. Korablev[1], Andrei G. Vasiliev[1], Sarng S. Pyurveev[1, 5], Tatyana V. Brus[1], Alexandra Yu. Pyrkh[1], Anna E. Kurtukova[1], Yulian R. Ryzhov[2], Yulia A. Taminkina[3], Zaur K. Emirgaev[4]

[1] Saint Petersburg State Pediatric Medical University, 2 Litovskaya str., Saint Petersburg, 194100, Russian Federation

[2] Research Institute of Obstetrics, Gynecology and Reproductology named after D.O. Ott, 3 Mendeleevskaya line, Saint Petersburg, 199034, Russian Federation

[3] Trans-Tehnologies LTD, 40A, Zheleznodorozhny ave., Saint Petersburg, 192178, Russian Federation

[4] City Hospital of Saint Holy Martyr Elizabeth, 14 Vavilovykh str., Saint Petersburg, 195257, Russian Federation

[5] Institute of Experimental Medicine, 12 Akademika Pavlova str., Saint Petersburg, 197376, Russian Federation

For citation: Korablev R.V., Vasiliev A.G., Pyurveev S.S., Brus T.V., Pyrkh A.Yu., Kurtukova A.E., Ryzhov Yu.R., Taminkina Yu.A., Emirgaev Z.K. Constructing products based on extracellular vesicles obtained from mesenchymal stem cells. *Russian Biomedical Research.* 2026;11(1):54–66. (In Russ.). https://doi.org/10.56871/RBR.2026.14.44.006.

Received: 08.12.2025 **Revised: 28.01.2026** **Accepted: 27.03.2026**

Corresponding author:
Rodion V. Korablev.
E-mail:
rodion.korablev@gmail.com

Abstract. Mesenchymal stem cells and products based on it are an effective means of treating diseases due to their proven effect on the tissue reparation. In recent years, large amount of data has accumulated showing that the therapeutic effect of mesenchymal stem cells is realized not by the cells themselves, but by it secretome. Preclinical studies using *in vitro* and *in vivo* disease models have demonstrated promising results with small extracellular vesicles derived from stem cells, including mesenchymal stem cells: reduction of oxidative stress, increased proliferation, survival and differentiation of cells, suppression of inflammation, which promotes regeneration at the site of damage. Isolation of the vesicular fraction of mesenchymal stem cells is the starting point for the production of products based on it. However, the path from the idea to the registration of such products is associated with many difficulties. A therapeutic strategy that allows achieving the effects of cell therapy without mesenchymal stem cells transplantation offers a number of advantages. The aim of this review is to highlight the multiple factors that contribute to the high variability of the vesicular fraction obtained from mesenchymal stem cells, the challenges of developing cell-free products, and the need for standardized protocols for the production and evaluation of extracellular vesicles.

Keywords: mesenchymal stem cells, mesenchymal stem cell secretome, extracellular vesicles, exosomes, extracellular vesicle-based products

© 2026 by the authors

Funding. This study was not supported by any external sources of funding.

Обзор
https://doi.org/10.56871/RBR.2026.14.44.006

Создание продуктов на основе внеклеточных везикул из мезенхимальных стволовых клеток

Родион Владимирович Кораблев[1], Андрей Глебович Васильев[1], Сарнг Саналович Пюрвеев[1, 5], Татьяна Викторовна Брус[1], Александра Юрьевна Пырх[1], Анна Евгеньевна Куртукова[1], Юлиан Рэммович Рыжов[2], Юлия Александровна Таминкина[3], Заур Келбялиевич Эмиргаев[4]

[1] Санкт-Петербургский государственный педиатрический медицинский университет, 194100, г. Санкт-Петербург, ул. Литовская, д. 2, Российская Федерация

[2] Научно-исследовательский институт акушерства, гинекологии и репродуктологии им. Д.О. Отта, 199034, г. Санкт-Петербург, Менделеевская линия, д. 3, Российская Федерация

[3] Общество с ограниченной ответственностью «Транс-Технологии», 192148, г. Санкт-Петербург, Железнодорожный пр., д. 40, литера А, Российская Федерация

[4] Городская больница Святой преподобной мученицы Елизаветы, 195257, г. Санкт-Петербург, ул. Вавиловых, д. 14, Российская Федерация

[5] Институт экспериментальной медицины, 197376, г. Санкт-Петербург, ул. Академика Павлова, д. 12, Российская Федерация

Для цитирования: Кораблев Р.В., Васильев А.Г., Пюрвеев С.С., Брус Т.В., Пырх А.Ю., Куртукова А.Е., Рыжов Ю.Р., Таминкина Ю.А., Эмиргаев З.К. Создание продуктов на основе внеклеточных везикул из мезенхимальных стволовых клеток. *Российские биомедицинские исследования.* 2026;11(1):54–66. https://doi.org/10.56871/RBR.2026.14.44.006.

Поступила: 08.12.2025 **Одобрена: 28.01.2026** **Принята к печати: 27.03.2026**

Автор для корреспонденции:
Родион Владимирович Кораблев.
E-mail:
rodion.korablev@gmail.com

Резюме. Мезенхимальные стволовые клетки, а также продукты на их основе являются эффективным инструментом в лечении ряда заболеваний благодаря влиянию на процесс репарации тканей. В последние годы накапливается все больше данных о том, что терапевтический эффект мезенхимальных стволовых клеток реализуется не самими клетками, а их секретомом. Секретом представляет собой комбинацию везикулярной и невезикулярной фракции, которую секретируют клетки, в том числе мезенхимальные стволовые клетки. Доклинические исследования с использованием моделей заболеваний *in vitro* и *in vivo* продемонстрировали многообещающие результаты применения малых внеклеточных везикул, выделенных из стволовых клеток, включая мезенхимальные стволовые клетки: снижение окислительного стресса, усиление пролиферации, выживаемости и дифференцировки клеток, угнетение воспаления, что способствует регенерации в очаге повреждения. Изоляция везикулярной фракции мезенхимальных стволовых клеток служит отправной точкой для производства продуктов на ее основе. Однако путь от идеи до регистрации подобных продуктов сопряжен с массой сложностей. При этом терапевтическая стратегия, позволяющая добиться эффектов клеточной терапии, не прибегая к трансплантации мезенхимальных стволовых клеток, обеспечивает ряд преимуществ. Цель данного обзора — подчеркнуть множество факторов, обусловливающих высокую вариабельность везикулярной фракции, получаемую из мезенхимальных стволовых клеток, сложности разработки бесклеточных продуктов, а также необходимость создания стандартизованных протоколов получения и оценки малых внеклеточных везикул.

Ключевые слова: мезенхимальные стволовые клетки, секретом мезенхимальных стволовых клеток, внеклеточные везикулы, экзосомы, продукты на основе внеклеточных везикул

© Коллектив авторов, 2026

Источник финансирования. Авторы заявляют об отсутствии внешнего финансирования при проведении исследования.

INTRODUCTION

Multipotent stem cells (MSCs) and MSC-derived products are opening new opportunities in biotechnology and regenerative medicine. MSCs are capable of differentiating into osteogenic, adipogenic, and chondrogenic lineages, express a set of surface markers (CD105, CD73, and CD90), and adhere to specialized plastic under standard culture conditions (Fig. 1) [1].

The therapeutic application of MSCs has been extensively investigated: as of August 2025, according to the U.S. National Institutes of Health database (https://clinicaltrials.gov), more than 1,300 clinical studies have been registered. Nevertheless, many unresolved issues remain. As a result, only 12 MSC-containing cell products have been registered to date, and only one has been approved by the U.S. Food and Drug Administration (FDA).

From the discovery of MSCs by Friedenstein in the 1970s [2] until the early 2000s, it was believed that the effects of MSCs were mediated by contact-dependent signaling to other cells, as well as by their accumulation at sites of injury, based on their capacity for multipotent differentiation [3]. This concept began to be reconsidered following the publication by Gnecchi, who demonstrated that MSCs exert their effects through the secretion and release of trophic molecules [4]. Furthermore, studies tracking MSCs after systemic administration showed that MSCs (and stem cells in general) do not become incorporated into the damaged tissue.

In recent years, increasing evidence has indicated that the primary mechanism of MSC action is their paracrine effect mediated by the release of biologically active molecules collectively known as the secretome. Over the past two decades, scientific interest in the cellular secretome, particularly its vesicular fraction, has increased dramatically. Investigation of the secretome composition has provided new insights into intercellular communication underlying numerous biological processes, both physiological and pathological. The composition of the secretome reflects the state of the secreting cell, and this property is already being utilized for disease diagnosis, prognostic assessment, and detailed studies of pathogenesis. In addition, strategies for targeted drug delivery using the vesicular fraction of the secretome are being actively developed, offering high specificity and efficiency..

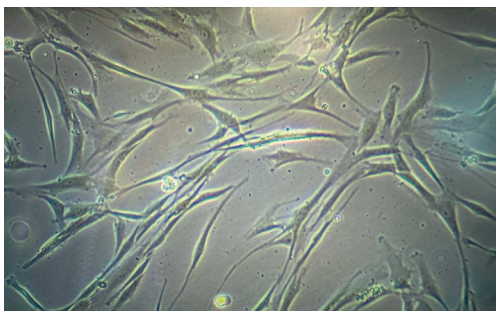


Fig. 1. **Mesenchymal stem cells (the figure is prepared by the authors)**

Рис. 1. Мезенхимальные стволовые клетки (рисунок подготовлен авторами)

COMPOSITION OF THE MESENCHYMAL STEM CELL SECRETOME

The cellular secretome has been described as a complex mixture of products released by virtually all cells of the body in the form of vesicular and non-vesicular fractions (Fig. 2) [5].

The vesicular fraction consists of vesicles that are classified according to their size. According to the recommendations of the International Society for Extracellular Vesicles (ISEV) issued on February 7, 2024, the term extracellular vesicles (EVs) refers to particles released from cells and enclosed by a lipid bilayer that are incapable of self-replication (i.e., do not contain a functional nucleus) [6]. ISEV recommends avoiding the use of the biogenesis-based terms exosomes and ectosomes, although these terms remain convenient for practical use. Instead, ISEV permits the use of the terms small extracellular vesicles (SEVs) (<200 nm) and large extracellular vesicles (LEVs) (>200 nm), with the size definition depending on the specific analytical method employed. In numerous scientific publications, extracellular vesicles are classified according to their biogenesis and release mechanisms as follows: exomeres (<50 nm), exosomes (30–150 nm), microvesicles (100–1000 nm), apoptotic bodies (1000–5000 nm), migrasomes (500–3000 nm), and oncosomes (1000–10000 nm) [7].

Non-vesicular extracellular particles (NVEPs) comprise all cell-derived particles belonging to one or more molecular classes that lack a delimiting lipid bilayer membrane [6]. NVEPs may be substantially smaller than extracellular vesicles, making their detection challenging, while MSC secretome isolation methods may result in the co-isolation of both vesicular and non-vesicular fractions [8]. The most common components of the non-vesicular fraction of the MSC secretome are presented in Table 1.

The most common components of the vesicular fraction of the MSC secretome are summarized in Table 2.

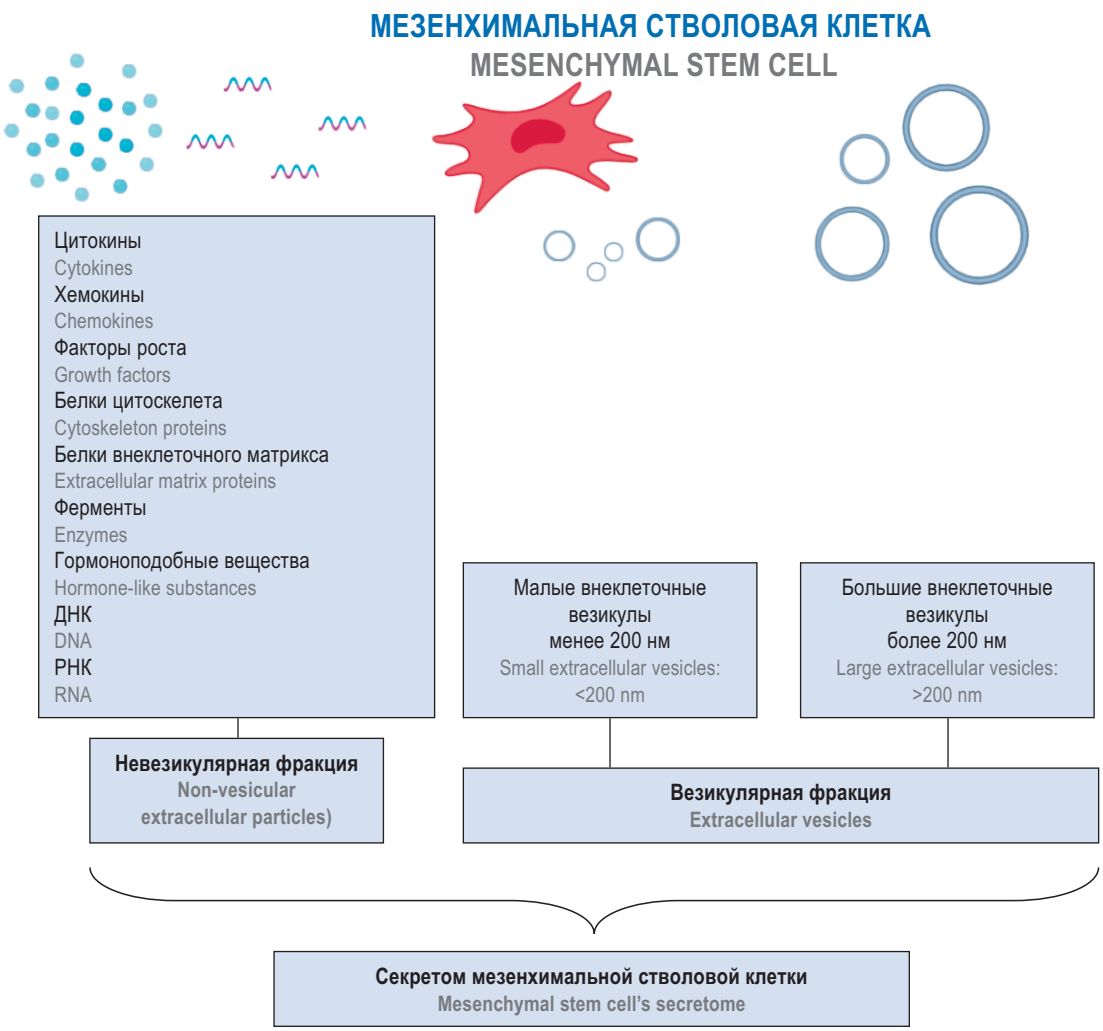


Fig. 2. **Mesenchymal stem cell`s secretome (the figure is prepared by the authors)**

Рис. 2. Секретом мезенхимальной стволовой клетки (рисунок подготовлен авторами)

Table 1

The most frequently encountered components of the non-vesicular fraction of the mesenchymal stem cells secretome

Таблица 1

Наиболее часто встречаемые компоненты невезикулярной фракции секретома мезенхимальных стволовых клеток

Компоненты невезикулярной фракции секретома мезенхимальных стволовых клеток Components of the non-vesicular fraction of the mesenchymal stem cells secretome	**Примеры** Examples	**Источник данных** Data source
Цитокины Cytokines	IL-1α, IL-1β, IL-6, IL-8, IL-10, IL-13	[8, 9]
Хемокины Chemokines	• Фактор стволовых клеток (SDF-1) Stem cell factor (SDF-1);	[8]
	• белок-хемоаттрактант моноцитов (MCP1) Monocyte chemoattractant protein (MCP1);	
	• хемокиновый лиганд мотива 12 (CXCL12) Chemokine ligand 12 motif (CXCL12);	
	• субъединица 8 полипептида 1, содержащего шаперонин (CCT8) Subunit 8 of polypeptide 1 containing chaperonin (CCT8);	
	• макрофагальный воспалительный белок 1-бета (MIP-1B) Macrophage inflammatory protein 1-beta (MIP-1B)	

Ending of the Table 1 / Окончание табл. 1

Компоненты невезикулярной фракции секретома мезенхимальных стволовых клеток Components of the non-vesicular fraction of the mesenchymal stem cells secretome	**Примеры** Examples	**Источник данных** Data source
Факторы роста Growth factors	• Фактор роста фибробластов 2 (FGF-2) Fibroblast growth factor 2 (FGF-2);	[8]
	• гранулоцитарно-макрофагальный колониестимулирующий фактор (GM-CSF) Granulocyte-macrophage colony-stimulating factor (GM-CSF);	
	• фактор роста гепатоцитов (HGF) Hepatocyte growth factor (HGF);	
	• субъединица А тромбоцитарного фактора роста (PDGF-1) Platelet-derived growth factor subunit A (PDGF-1);	
	• трансформирующий фактор роста бета (TGF-β) Transforming growth factor beta (TGF-β)	
Белки цитоскелета Cytoskeletal proteins	• Актин α2 (ACTA2), актин β (ACTB), α-актин сердечной мышцы (ACTC1) Actin α2 (ACTA2), actin β (ACTB), cardiac α-actin (ACTC1)	[10]
Белки внеклеточного матрикса Extracellular matrix proteins	• Фибриллин (FBN1), бета-цепь фибриногена (FGB) Fibrillin (FBN1), fibrinogen beta chain (FGB)	[10]
Ферменты Enzymes	• Глицеральдегид-3-фосфатдегидрогеназа индоламин 2 (GAPDH); супероксиддисмутаза (SOD) Glyceraldehyde-3-phosphate dehydrogenase indoleamine 2 (GAPDH); superoxide dismutase (SOD)	[8]
Гормоноподобные вещества Hormone-like substances	• Инсулиноподобный фактор роста-1 (IGF-1) Insulin-like growth factor-1 (IGF-1)	[8]

The table is prepared by the authors
Таблица составлена авторами

SEVs derived from MSCs express the same surface markers as MSCs themselves, including CD29, CD44, CD73, CD90, and CD105 [14]. These markers can be detected using various techniques, including flow cytometry and Western blotting.

SEVs are widely present in biological fluids such as saliva, urine, breast milk, cerebrospinal fluid, plasma, and serum [15]. Although their concentration varies among different biological fluids, the concentration of SEVs in 1 mL of blood plasma may reach 10^{10} particles [16]. Identified SEV components include approximately 10,000 proteins, 3,500 RNAs, 3,000 microRNAs, and 1,000 lipids [17].

Since SEVs retain the signature of their parent cells, determining their composition in blood plasma is of considerable diagnostic and prognostic value. At the same time, the overall composition of plasma SEVs is highly heterogeneous because they originate from cells of most tissues. Investigation of SEV composition may substantially expand our understanding of disease pathogenesis and serve as a starting point for the development of novel SEV-based therapeutics and vaccines [18].

The specific protein and RNA cargo of MSC-derived SEVs varies depending on the MSC source and culture conditions [19, 20]. Culture conditions such as hypoxia and cytokine stimulation can alter SEV composition and biological effects. Hypoxia enhances the secretion of pro-angiogenic factors by MSCs and their released SEVs, thereby improving neovascularization and bone tissue regeneration [21]. Cultivation with rapamycin and growth factor restriction increases SEV release [22]. Factors such as donor age, health status, and the method used for MSC isolation also influence SEV composition [23]. For example, SEVs derived from adipose tissue MSCs obtained by surgical excision and liposuction exhibit different characteristics, and differences in microRNA profiles have been observed even when the same isolation method is used for samples collected from different anatomical sites [24]. Age-related changes in donor cells affect the quality of released SEVs, limiting their anti-inflammatory and regenerative potential [25].

Increasing attention is being paid to the nucleic acid fraction of SEVs. Several studies have shown that DNA transported within SEVs participates in horizontal gene transfer between cells, including transformed cells, contributing to genetic diversity and potentially facilitating the spread of oncogenes during cancer progression [26].

Table 2

The most common components of the vesicular fraction of the mesenchymal stem cells secretome according to different sources

Таблица 2

Наиболее часто встречаемые компоненты везикулярной фракции секретома мезенхимальных стволовых клеток согласно разным источникам

Компоненты везикулярной фракции секретома мезенхимальных стволовых клеток Components of the vesicular fraction of the mesenchymal stem cells secretome	**Примеры** Examples	**Источник данных** Data source
Белки Proteins	• Актин Actin; • β-тубулин β-tubulin; • тетраспанины (CD9, CD63, CD81, CD82) tetraspanins (CD9, CD63, CD81, CD82); • белки теплового шока (HSP770, HSP90) heat shock proteins (HSP770, HSP90); • белок опухолевой предрасположенности 101 (Tsg101) tumor predisposition protein 101 (Tsg101); • белок, взаимодействующий с запрограммированной клеточной гибелью (Alix) programmed cell death-interacting protein (Alix); • белки главного комплекса гистосовместимости I и II классов (MHC I, MHC-II) proteins of the major histocompatibility complex classes I and II (MHC I, MHC-II)	[11]
Липиды Lipids	• Церамиды Ceramides; • сфингомиелин sphingomyelin; • фосфатидилсерин phosphatidylserine;	
	• фосфатидилинозитол phosphatidylinositol; • холестерин cholesterol	[12]
Нуклеиновые кислоты Nucleic acids	• Матричная РНК messenger RNA; • микроРНК microRNA • рибосомальная РНК ribosomal RNA • ДНК DNA	[13]

The table is prepared by the authors
Таблица составлена авторами

Because the DNA of SEVs derived from certain tumors contains characteristic genetic alterations, such as mutations and gene amplifications, SEVs represent promising targets for noninvasive diagnostic applications, including liquid biopsy. Furthermore, mitochondrial DNA contained in SEVs is associated with cellular stress responses and may play a role in immune system intercellular signaling [27].

Various forms of RNA have also been identified within SEVs, including messenger RNA (mRNA), microRNA, ribosomal RNA, circular RNA, long non-coding RNA and DNA. SEV-associated RNAs undergo horizontal transfer between cells, resulting in phenotypic changes in recipient cells due to their important role in gene regulation and participation in diverse physiological processes [28]. For example, SEVs derived from embryonic stem cells can reprogram hematopoietic progenitor cells through the delivery of mRNA [29]. During the immune response, unidirectional transfer of mRNA occurs from T lymphocytes to antigen-presenting cells, a process that may be antigen-dependent. Viral microRNAs contained in SEVs released by infected cells can be taken up by uninfected cells [30].

MicroRNAs are the most abundant RNA species found in human plasma SEVs, accounting for approximately 40% of the total RNA content, which makes them particularly advantageous for diagnostic applications.

FORMATION AND BIOLOGICAL EFFECTS OF EXTRACELLULAR VESICLES

The secretion of small extracellular vesicles by cells is a fundamental process required for intercellular communication under both physiological and pathological conditions [31] and has likely existed throughout evolution [32]. Under in vitro conditions, SEVs are released by cultured cells into the culture medium.

The process of vesicle formation begins with the invagination of the plasma membrane into the cell, leading to the formation of early endosomes, followed by the development of late endosomes or multivesicular bodies (MVBs) (Fig. 3) [33].

At this stage, MVBs contain proteins that were present on the plasma membrane, as well as RNA and proteins derived from the cytoplasm. Subsequently, proteins from the endoplasmic reticulum and Golgi apparatus are incorporated into MVBs. The *de novo* incorporation of various components into MVBs is regulated by the ESCRT protein complex (Endosomal Sorting Complexes Required for Transport) or mediated through ceramide-/tetraspanin-dependent pathways [34].

Some of the formed MVBs fuse with lysosomes, resulting in degradation of their contents. Another fraction of MVBs is “tagged” by ceramides; the outer membrane of the MVB fuses with the plasma membrane, and SEVs are released into the extracellular space [35].

The biogenesis and secretion of SEVs are controlled by specific GTPase proteins. Small GTPases of the Rab family regulate intracellular vesicular transport. For example, Rab5 participates in endosome formation, whereas Rab7 promotes degradation of MVB contents within lysosomes. Several Rab proteins (Rab35, Rab11, Rab27a, and Rab27b) regulate the secretion of SEVs into the extracellular space [31]. The formation and secretion of SEVs may be altered by intracellular processes or external

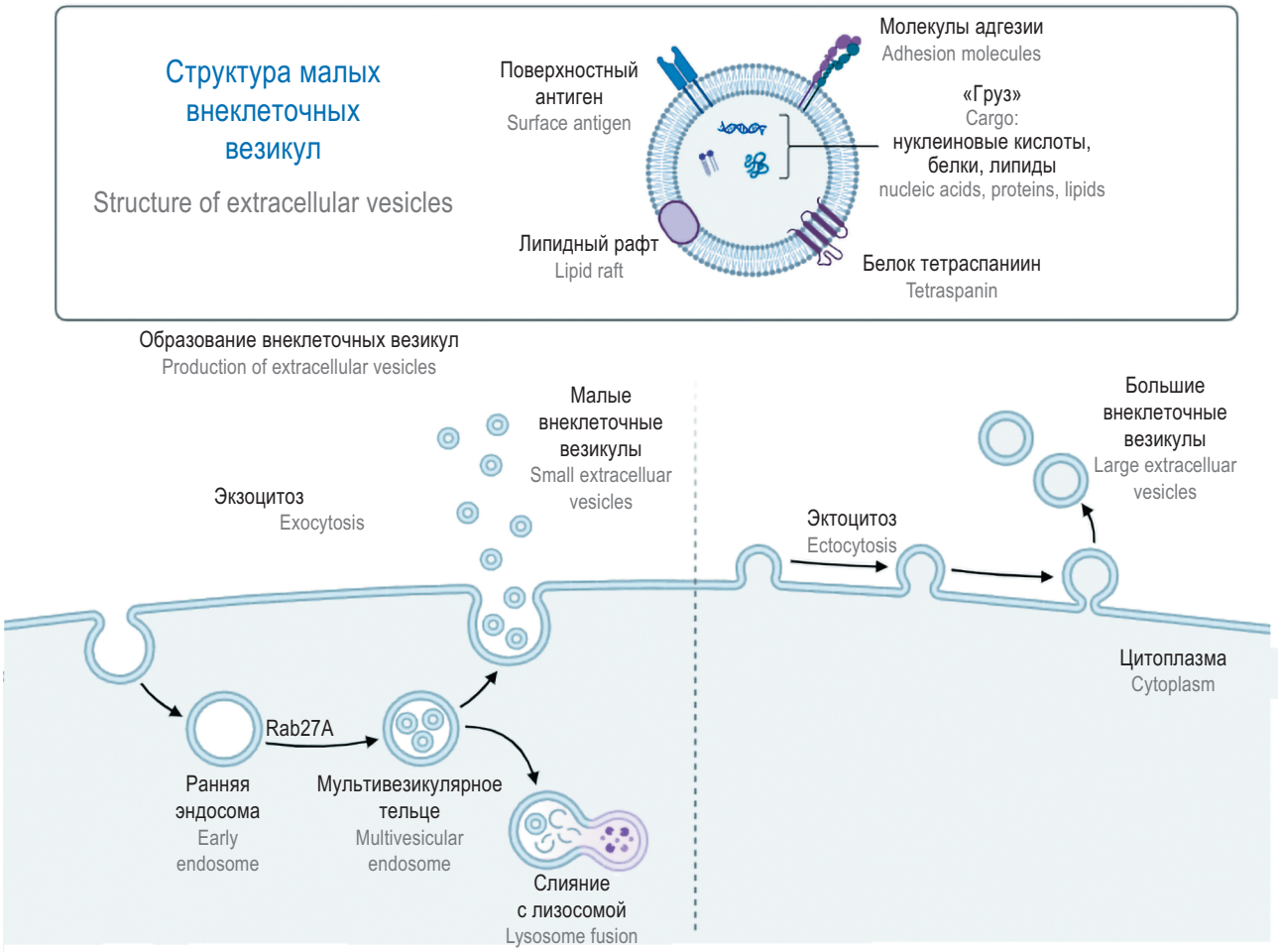


Fig. 3. **Structure and biogenesis of extracellular vesicles (the figure is prepared by the authors)**

Рис. 3. Структура и биогенез внеклеточных везикул (рисунок подготовлен авторами)

stimuli. For example, transformed tumor cells secrete significantly greater numbers of SEVs than non-transformed healthy cells [36]. Cellular stress is considered a possible factor that increases SEV release [37].

Following their release, SEVs adhere to the surface of target cells through the action of integrins and tetraspanins. For example, integrins αv (CD51), α3 (CD49c), β1 (CD29), as well as lactadherin and vitronectin, are involved in the adhesion of SEVs to leukocytes [33]. Tetraspanins present on the plasma membrane are also highly expressed on SEVs (CD9, CD81, CD63, and CD82), allowing their use as specific SEV markers. Tetraspanins play a key role in the adhesion, fusion, and selective uptake of SEVs by recipient cells [38]. Changes in the expression of specific tetraspanins may modulate the selective targeting and uptake of SEVs, thereby influencing the cellular response. The specificity of SEVs for target cells may be determined by the combination of antigens with MHC class I and II molecules [39].

The interaction of SEVs with recipient cells may lead to several outcomes: (1) signal transmission through intracellular signaling pathways (juxtacrine effect); (2) fusion with the cell membrane and release of vesicular contents into the cytoplasm of the recipient cell; and (3) uptake of SEVs through phagocytosis, macropinocytosis, or receptor-mediated endocytosis. Within the recipient cell, the contents of SEVs are either degraded or induce a cellular response. Although SEVs possess a wide range of biological activities, their principal significance lies in their ability to serve as carriers of information, facilitating long-distance intercellular communication. Compared with the diffusion of soluble signaling molecules, SEV-mediated intercellular communication is more effective both in terms of biological impact and specificity.

ADVANTAGES OF USING CELL-FREE PRODUCTS BASED ON COMPONENTS OF THE MESENCHYMAL STEM CELL SECRETOME

Despite numerous clinical studies, only 12 mesenchymal stem cell (MSC)-containing products have been approved to date, and only one of them has received FDA approval — Prochymal (Remestemcel-L) (Osiris Therapeutics Inc., Mesoblast Ltd.), which is used for the treatment of graft-versus-host disease. In this regard, the potential use of cell-free products based on the MSC secretome or its vesicular fraction offers several key advantages over MSC-containing cellular products, namely:

1) improved patient safety and avoidance of the low but potentially existing risks of tumorigenesis and transmission of infectious agents associated with the transplantation of living stem cell populations;
2) reduction in the number of invasive material collection procedures or their complete avoidance (depending on the MSC source);
3) assessment of safety and efficacy using approaches similar to those applied to pharmaceutical agents;
4) substantially prolonged storage life through cryopreservation or lyophilization without loss of biological activity; products containing viable cells are generally short-lived (from several hours to one day), requiring complex logistics for rapid delivery to patients;
5) large-scale manufacturing of cell-free products using specially established cell lines under controlled laboratory conditions (e.g., in bioreactors) to obtain MSC secretome components at desired concentrations and to tailor the secretome composition according to specific therapeutic goals;
6) reduced production time and cost, with immediate availability of ready-to-use products, which is particularly important for the treatment of acute conditions such as cerebral ischemia, myocardial infarction, and combat-related injuries.

CHALLENGES IN THE DEVELOPMENT OF CELL-FREE PRODUCTS BASED ON COMPONENTS OF THE MESENCHYMAL STEM CELL SECRETOME

Although cell-free products based on MSC secretome components offer potential advantages over MSC-containing cellular products, their production and regulatory approval are associated with numerous challenges.

1. Heterogeneity of SEVs derived from MSCs. The biological variability of SEV cargo, discussed above, poses a challenge for the standardization and predictability of therapeutic outcomes based on SEVs. Strict quality control measures, including the use of reference samples, development of calibration standards, and consensus guidelines for SEV research, may improve the reproducibility of SEV-related studies [40].

2. Use of different methods for SEV isolation and characterization.

To date, no standardized methods for SEV isolation, purification, and characterization have been

established [41]. This creates uncertainty regarding their expected biological effects and potential clinical outcomes, significantly complicating the assessment of efficacy and safety in both clinical and experimental studies. Commonly used methods for SEV isolation include ultrafiltration, ultracentrifugation, size-exclusion chromatography, and immunoaffinity-based approaches [42].

The small size of SEVs (30–150 nm), their heterogeneity within source samples, the presence of protein contaminants, and high economic costs hinder the selection of an optimal method for obtaining SEVs with predefined characteristics. Ultrafiltration, based on size separation, does not require expensive equipment but is time-consuming and associated with a high risk of membrane fouling by larger particles [43]. Ultracentrifugation, which relies on differences in density and size, is labor-intensive, costly, technically demanding, and may result in high levels of contaminants (RNA, proteins) as well as damage to SEV integrity [44]. Size-exclusion chromatography is used to obtain highly purified SEVs but requires expensive equipment and complex procedures. Immunoaffinity-based methods, which identify SEVs according to unique surface protein profiles using biotinylated antibodies, enable the isolation of tissue-specific SEVs with high purity; however, these methods are costly, low-throughput, and require extensive preparation [42].

Following isolation, SEVs must be characterized. Since SEVs contain nucleic acids, proteins, lipids, and a range of characteristic surface markers, each of these components may be measured as a surrogate indicator of SEV abundance. However, none of these parameters perfectly correlate with the actual number of SEVs. SEV purity and yield may be assessed using transmission or scanning electron microscopy, flow cytometry, Western blotting, and liquid chromatography–mass spectrometry [44]. A detailed description of these methods is beyond the scope of this review.

3. Low SEV yield. Although SEVs represent promising therapeutic agents, they are typically not secreted in quantities sufficient for clinical application, which limits the full realization of their therapeutic potential. Insufficient SEV production remains one of the major obstacles to their effective use [45]. For example, 1 mL of conditioned culture medium can produce approximately 10^9 SEVs, whereas an effective therapeutic dose is generally considered to be around 10^{10} SEV particles per milliliter [46]. Furthermore, no recommendations have yet been developed regarding the required in vivo dose of SEVs, taking into account their pharmacokinetics and pharmacodynamics [47].

Strategies to increase SEV release from cultured cells include three-dimensional (3D) culture systems using bioreactors and spheroids, physical stimulation (mechanical, electrical, or magnetic field-based), chemical stimulation, physiological modification, and genetic manipulation of the parent cell population producing SEVs [48]. In addition to difficulties associated with obtaining high SEV yields from cell cultures, it should also be considered that interactions between SEVs and target cells are accompanied by partial lysosomal degradation, ultimately reducing their therapeutic potential.

4. Variable stability of SEVs. Currently, there are no standardized requirements or conditions for SEV storage, despite their importance for planning the manufacture of SEV-containing products. Three principal methods are commonly used for SEV preservation: cryopreservation, spray drying, and freeze-drying (lyophilization).

Cryopreservation is typically performed at −80 °C or −196 °C. This process involves freezing, thawing, and potentially refreezing SEVs following partial use. To preserve SEV integrity during freeze–thaw cycles, various cryoprotective agents are employed; these should ideally possess low toxicity, water solubility, and low cost.

Factors such as the number of freeze–thaw cycles, pH, storage medium composition, and osmotic pressure influence the stability of SEV cargo, particularly proteins and microRNAs that mediate therapeutic effects, in MSC-derived SEVs [49]. For example, storage of SEVs in phosphate-buffered saline results in a rapid decline in SEV recovery within several days, whereas dilution of the buffer may reduce recovery within minutes [50]. The addition of human serum albumin and trehalose to the storage medium at −80 °C appears to be the most promising approach for long-term storage and preservation of stability through multiple freeze–thaw cycles.

Spray drying converts a liquid suspension containing SEVs into a dry powder through rapid exposure to hot gas. However, this process may denature many surface proteins and is therefore less suitable for SEVs containing thermosensitive components. During freeze-drying (lyophilization), a two-step preservation process involving freezing followed by water removal under reduced pressure, water is removed from SEVs in a manner that stabilizes them [51]. After lyophili-

zation, SEVs may be stored at room temperature for extended periods. Nevertheless, the choice of cryoprotectants remains crucial for preventing aggregation and maintaining functional integrity following reconstitution [52].

Despite the evident advantages of cell-free products, numerous challenges must be addressed before the first regulatory approval can be granted for a product containing the MSC secretome or its components. The most pressing issues in the development and registration of such products are the standardization and improvement of methods for the isolation, storage, and characterization of the vesicular fraction of the secretome. Therefore, the establishment of a reliable therapeutic platform for SEV-based products requires scalable manufacturing processes with clearly defined timelines and economic feasibility. Equally important are further studies on the composition, biogenesis, and biological effects of SEVs on target cells, which will enable a more detailed assessment of the efficacy and safety of SEV-based therapeutic products and facilitate modification of their composition according to specific clinical applications.

ADDITIONAL INFORMATION

Author contribution. R.V. Korablev — writing and editing, review`s concept; A.G. Vasiliev — writing and editing, review`s concept, supervision; S.S. Purveev, T.V. Brus, A.Yu. Pyrkh, Z.K. Emirgaev, A.E. Kurtukova, Yu.R. Ryzhov, Yu.A. Taminkina — writing and editing.

All authors made a substantial contribution to the conception of the study, acquisition, analysis, interpretation of data for the work, drafting and revising the article, final approval of the version to be published and agree to be accountable for all aspects of the study.

Competing interests. The authors declare that they have no competing interests.

ДОПОЛНИТЕЛЬНАЯ ИНФОРМАЦИЯ

Вклад авторов. Р.В. Кораблев — написание и редактирование текста, концепция обзора; А.Г. Васильев — написание и редактирование текста, концепция обзора, руководство; С.С. Пюрвеев, Т.В. Брус, А.Ю. Пырх, З.К. Эмиргаев, А.Е. Куртукова, Ю.Р. Рыжов, Ю.А. Таминкина — написание и редактирование текста.

Все авторы внесли существенный вклад в разработку концепции, проведение исследования и подготовку статьи, прочли и одобрили финальную версию перед публикацией.

Конфликт интересов. Авторы декларируют отсутствие явных и потенциальных конфликтов интересов, связанных с публикацией настоящей статьи.

ЛИТЕРАТУРА / REFERENCES

1. Viswanathan S., Shi Y., Galipeau J., Krampera M., Leblanc K., Martin I., Nolta J., Phinney D.G., Sensebe L. Mesenchymal stem versus stromal cells: International Society for Cell & Gene Therapy (ISCT®) Mesenchymal Stromal Cell committee position statement on nomenclature. *Cytotherapy*. 2019;21:1019–1024. https://doi.org/10.1016/j.jcyt.2019.08.002.
2. Friedenstein A.J., Deriglasova U.F., Kulagina N.N., Panasuk A.F., Rudakowa S.F., Luriá E.A., Ruadkow I.A. Precursors for fibroblasts in different populations of hematopoietic cells as detected by the in vitro colony assay method. *Exp Hematol*. 1974;2:83–92.
3. Ankrum J.A., Ong J.F., Karp J.M. Mesenchymal stem cells: immune evasive, not immune privileged. *Nat Biotechnol*. 2014;32(3):252–260. https://doi.org/10.1038/nbt.2816.
4. Gnecchi M., He H., Liang O.D., Melo L.G., Morello F., Mu H. et al. Paracrine action accounts for marked protection of ischemic heart by Akt-modified mesenchymal stem cells. *Nat Med*. 2005;11(4):367–368. https://doi.org/10.1038/nm0405-367.
5. Teixeira F.G., Salgado A.J. Mesenchymal stem cells secretome: current trends and future challenges. *Neural Regen Res*. 2019;15(1):75–77. https://doi.org/10.4103/1673-5374.264455.
6. Welsh J.A., Goberdhan D.C.I., O'Driscoll L., Buzas E.I., Blenkiron C., Bussolati B. et al. Minimal information for studies of extracellular vesicles (MISEV2023): From basic to advanced approaches. *J Extracell Vesicles*. 2024;13(2):e12404. https://doi.org/10.1002/jev2.12404.
7. Wang W., Li M., Chen Z., Xu L., Chang M., Wang K. et al. Biogenesis and function of extracellular vesicles in pathophysiological processes of skeletal muscle atrophy. *Biochem Pharmacol*. 2022;198:114954. https://doi.org/10.1016/j.bcp.2022.114954.
8. da Silva A.V., Serrenho I., Araújo B., Carvalho A.M., Baltazar G. Secretome as a Tool to Treat Neurological Conditions: Are We

Ready? *Int J Mol Sci*. 2023;24(22):16544. https://doi.org/10.3390/ijms242216544.
9. Vizoso F.J., Eiro N., Cid S., Schneider J., Perez-Fernandez R. Mesenchymal stem cell secretome: toward cell-free therapeutic strategies in regenerative medicine. *Int J Mol Sci*. 2017;18(9):1852. https://doi.org/10.3390/ijms18091852.
10. Advani D., Farid N., Tariq M.H., Kohli N. A systematic review of mesenchymal stem cell secretome: Functional annotations, gene clusters and proteomics analyses for bone formation. *Bone*. 2025;190:117269. https://doi.org/10.1016/j.bone.2024.117269.
11. Doyle L.M., Wang M.Z. Overview of extracellular vesicles, their origin, composition, purpose, and methods for exosome isolation and analysis. *Cells*. 2019;8(7):727. https://doi.org/10.3390/cells8070727.
12. Ding J.-Y., Chen M.-J., Wu L-F, Shu G.-F., Fang S.-J., Li Z.-Y. et al. Mesenchymal stem cell-derived extracellular vesicles in skin wound healing: roles, opportunities and challenges. *Mil Med Res*. 2023;10(1):36. https://doi.org/10.1186/s40779-023-00472-w.
13. Dong R., Liu Y., Yang Y., Wang H., Xu Y., Zhang Z. MSC-Derived exosomes-based therapy for peripheral nerve injury: a novel therapeutic strategy. *Biomed Res Int*. 2019;2019:6458237. https://doi.org/10.1155/2019/6458237.
14. Shin S., Lee J., Kwon Y., Park K.-S., Jeong J.-H., Choi S.-J. et al. comparative proteomic analysis of the mesenchymal stem cells secretome from adipose, bone marrow, placenta and Wharton's jelly. *Int J Mol Sci*. 2021;22(2):845. https://doi.org/10.3390/ijms22020845.
15. Li X., Corbett A.L., Taatizadeh E., Tasnim N., Little J.P., Garnis C. et al. Challenges and opportunities in exosome research — Perspectives from biology, engineering, and cancer therapy. *APL Bioeng*. 2019;3(1):011503. https://doi.org/10.1063/1.5087122.
16. Johnsen K.B., Gudbergsson J.M., Andresen T.L., Simonsen J.B. What is the blood concentration of extracellular vesicles? Implications for the use of extracellular vesicles as blood-borne biomarkers of cancer. *Biochim Biophys Acta Rev Cancer*. 2019;1871(1):109–16. https://doi.org/10.1016/j.bbcan.2018.11.006.
17. Huang C., Li J., Xie Z., Hu X., Huang Y. Relationship between exosomes and cancer: formation, diagnosis, and treatment. *Int J Biol Sci*. 2025;21(1):40–62. https://doi.org/10.7150/ijbs.95763.
18. Ng C.Y., Kee L.T., Al-Masawa M.E., Lee Q.H., Subramaniam T., Kok D. et al. Scalable Production of extracellular vesicles and its therapeutic values: a review. *Int J Mol Sci*. 2022;23(14):7986. https://doi.org/10.3390/ijms23147986.
19. Ramos T.L., Sánchez-Abarca L.I., Muntión S., Preciado S., Puig N., López-Ruano G. et al. MSC surface markers (CD44, CD73, and CD90) can identify human MSC-derived extracellular vesicles by conventional flow cytometry. *Cell Commun Signal*. 2016;14:2. https://doi.org/10.1186/s12964-015-0124-8.
20. Zhang Y., Liu Y., Liu H., Tang W.H. Exosomes: biogenesis, biologic function and clinical potential. *Cell Biosci*. 2019;9:19. https://doi.org/10.1186/s13578-019-0282-2.
21. Liu W., Li L., Rong Y., Qian D., Chen J., Zhou Z. et al. Hypoxic mesenchymal stem cell-derived exosomes promote bone fracture healing by the transfer of miR-126. *Acta Biomater*. 2020;103:196–212. https://doi.org/10.1016/j.actbio.2019.12.020.
22. Zou W., Lai M., Zhang Y., Zheng L., Xing Z., Li T. et al. Exosome Release is regulated by mTORC1. *Adv Sci (Weinh)*. 2018;6(3):1801313. https://doi.org/10.1002/advs.201801313.
23. Chen Y., Qi W., Wang Z., Niu F. Exosome Source matters: a comprehensive review from the perspective of diverse cellular origins. *Pharmaceutics*. 2025;17(2):147. https://doi.org/10.3390/pharmaceutics17020147.
24. Visconte C., Taiana M.M., Colombini A., De Luca P., Ragni E., de Girolamo L. Donor sites and harvesting techniques affect miRNA cargos of extracellular vesicles released by human adipose-derived mesenchymal stromal cells. *Int J Mol Sci*. 2024;25(12):6450. https://doi.org/10.3390/ijms25126450.
25. Zhang J., Cai Z., Feng F., Peng Y., Cui Y., Xu Y. Age-different BMSCs-derived exosomes accelerate tendon-bone interface healing in rotator cuff tears model. *Gene*. 2024;895:148002. https://doi.org/10.1016/j.gene.2023.148002.
26. Collino F., Deregibus M.C., Bruno S., Sterpone L., Aghemo G., Viltono L. et al. Microvesicles Derived from adult human bone marrow and tissue specific mesenchymal stem cells shuttle selected pattern of miRNAs. *PLoS One*. 2010;5(7):e11803. https://doi.org/10.1371/journal.pone.0011803.
27. Sansone P., Savini C., Kurelac I., Chang Q., Amato L.B., Strillacci A. et al. Packaging and transfer of mitochondrial DNA via exosomes regulate escape from dormancy in hormonal therapy-resistant breast cancer. *Proc Natl Acad Sci U S A*. 2017;114(43):E9066–E9075. https://doi.org/10.1073/pnas.1704862114.
28. Mittelbrunn M., Gutiérrez-Vázquez C., Villarroya-Beltri C., González S., Sánchez-Cabo F., González M.Á. et al. Unidirectional transfer of microRNA-loaded exosomes from T cells to antigen-presenting cells. *Nat Commun*. 2011;2:282. https://doi.org/10.1038/ncomms1285.
29. Ratajczak J., Miekus K., Kucia M., Zhang J., Reca R., Dvorak P., Ratajczak M.Z. Embryonic stem cell-derived microvesicles reprogram hematopoietic progenitors: evidence for horizontal transfer of mRNA and protein delivery. *Leukemia*. 2006;20(5):847–856. https://doi.org/10.1038/sj.leu.2404132.
30. Pegtel D.M., Cosmopoulos K., Thorley-Lawson D.A., van Eijndhoven M.A.J., Hopmans E.S., Lindenberg J.L. et al. Functional delivery of viral miRNAs via exosomes. *Proc Natl Acad Sci U S A*. 2010;107(14):6328–6333. https://doi.org/10.1073/pnas.0914843107.
31. Gurunathan S., Kang M.-H., Qasim M., Khan K., Kim J-H. Biogenesis, membrane trafficking, functions, and next generation nanotherapeutics medicine of extracellular vesicles. *Int J Nanomedicine*. 2021;16:3357–3383. https://doi.org/10.2147/IJN.S310357.
32. Raposo G., Stoorvogel W. Extracellular vesicles: exosomes, microvesicles, and friends. *J Cell Biol*. 2013;200(4):373–383. https://doi.org/10.1083/jcb.201211138.

33. McKelvey K.J., Powell K.L., Ashton A.W., Morris J.M., McCracken S.A. Exosomes: mechanisms of uptake. *J Circ Biomark*. 2015;4:7. https://doi.org/10.5772/61186.
34. Larios J., Mercier V., Roux A., Gruenberg J. ALIX- and ESCRT-III-dependent sorting of tetraspanins to exosomes. *J Cell Biol*. 2020;219(3):e201904113. https://doi.org/10.1083/jcb.201904113.
35. Tenchov R., Sasso J.M., Wang X., Liaw W.-S., Chen C.-A., Zhou Q.A. Exosomes — nature's lipid nanoparticles, a rising star in drug delivery and diagnostics. *ACS Nano*. 2022;16(11):17802–17846. https://doi.org/10.1021/acsnano.2c08774.
36. Graves L.E., Ariztia E.V., Navari J.R., Matzel H.J., Stack M.S., Fishman D.A. Proinvasive properties of ovarian cancer ascites-derived membrane vesicles. *Cancer Res*. 2004;64(19):7045–7049. https://doi.org/10.1158/0008-5472.CAN-04-1800.
37. Lai J.J., Chau Z.L., Chen S., Hill J.J., Korpany K.V., Liang N. et al. Exosome Processing and characterization approaches for research and technology development. *Adv Sci (Weinh)*. 2022;9(15):2103222. https://doi.org/10.1002/advs.202103222.
38. Jankovičová J., Sečová P., Michalková K., Antalíková J. Tetraspanins, more than markers of extracellular vesicles in reproduction. *Int J Mol Sci*. 2020;21(20):7568. https://doi.org/10.3390/ijms21207568.
39. Huda M.N., Nafiujjaman M., Deaguero I.G., Okonkwo J., Hill M.L., Kim T., Nurunnabi M. Potential use of exosomes as diagnostic biomarker and in targeted drug delivery: progress in clinical and preclinical application. ACS *Biomater Sci Eng*. 2021;7(6):2106–2149. https://doi.org/10.1021/acsbiomaterials.1c00217.
40. Chen X., Tian B., Wang Y., Zheng J., Kang X. Potential and challenges of utilizing exosomes in osteoarthritis therapy (Review). *Int J Mol Med*. 2025;55(3):43. https://doi.org/10.3892/ijmm.2025.5484.
41. Mei R., Wan Z., Yang C., Shen X., Wang R., Zhang H. et al. Advances and clinical challenges of mesenchymal stem cell therapy. *Front Immunol*. 2024;15:1421854. https://doi.org/10.3389/fimmu.2024.1421854.
42. Abdulmalek O.A.A.Y., Husain K.H., AlKhalifa H.K.A.A., Alturani M.M.A.B., Butler A.E., Moin A.S.M. Therapeutic Applications of stem cell-derived exosomes. *Int J Mol Sci*. 2024;25(6):3562. https://doi.org/10.3390/ijms25063562.
43. Théry C., Amigorena S., Raposo G., Clayton A. Isolation and characterization of exosomes from cell culture supernatants and biological fluids. *Curr Protoc Cell Biol*. 2006;Chapter 3:Unit 3.22. https://doi.org/10.1002/0471143030.cb0322s30.
44. Sani F., Shafiei F., Dehghani F., Mohammadi Y., Khorraminejad-Shirazi M., Anvari-Yazdi A.F. et al. Unveiling exosomes: Cuttingedge isolation techniques and their therapeutic potential. *J Cell Mol Med*. 2024;28(20):e70139. https://doi.org/10.1111/jcmm.70139.
45. Wang J., Bonacquisti E.E., Brown A.D., Nguyen J. Boosting the biogenesis and secretion of mesenchymal stem cell-derived exosomes. *Cells*. 2020;9(3):660. https://doi.org/10.3390/cells9030660.
46. Luo L., Wu Z., Wang Y., Li H. Regulating the production and biological function of small extracellular vesicles: current strategies, applications and prospects. *J Nanobiotechnology*. 2021;19(1):422. https://doi.org/10.1186/s12951-021-01171-1.
47. Erwin N., Serafim M.F., He M. Enhancing the cellular production of extracellular vesicles for developing therapeutic applications. *Pharm Res*. 2023;40(4):833–853. https://doi.org/10.1007/s11095-022-03420-w.
48. de Almeida Fuzeta M., Bernardes N., Oliveira F.D., Costa A.C., Fernandes-Platzgummer A., Farinha J.P. et al. Scalable production of human mesenchymal stromal cell-derived extracellular vesicles under Serum-/Xeno-Free conditions in a microcarrier-based bioreactor culture system. *Front Cell Dev Biol*. 2020;8:553444. https://doi.org/10.3389/fcell.2020.553444.
49. Yadav S., Maity P., Kapat K. The Opportunities and challenges of mesenchymal stem cells-derived exosomes in theranostics and regenerative medicine. *Cells*. 2024;13(23):1956. https://doi.org/10.3390/cells13231956.
50. Luo T., von der Ohe J., Hass R. MSC-Derived extracellular vesicles in tumors and therapy. *Cancers (Basel)*. 2021;13(20):5212. https://doi.org/10.3390/cancers13205212.
51. Trenkenschuh E., Richter M., Heinrich E., Koch M., Fuhrmann G., Friess W. Enhancing the Stabilization potential of lyophilization for extracellular vesicles. *Adv Healthc Mater*. 2022;11(5):2100538. https://doi.org/10.1002/adhm.202100538.
52. Bahr M.M., Amer M.S., Abo-El-Sooud K., Abdallah A.N., El-Tookhy O.S. Preservation techniques of stem cells extracellular vesicles: a gate for manufacturing of clinical grade therapeutic extracellular vesicles and long-term clinical trials. *Int J Vet Sci Med*. 2020;8(1):1–8. https://doi.org/10.1080/23144599.2019.1704992.

Authors' info

*** Rodion V. Korablev** — Cand. Sci. (Med.), Associate Professor, Department of Pathological Physiology with a Course of Immunopathology. E-mail: rodion.korablev@gmail.com; https://orcid.org/0009-0004-5754-8437; SPIN: 4969-6038
Andrei G. Vasiliev — Dr. Sci. (Med.), Professor, Head of the Department of Pathological Physiology with a Course of Immunopathology. E-mail: avas7@mail.ru: https://orcid.org/0000-0002-8539-7128; SPIN: 1985-4025

Об авторах

*** Родион Владимирович Кораблев** — к.м.н., доцент, кафедра патологической физиологии с курсом иммунопатологии. E-mail: rodion.korablev@gmail.com; https://orcid.org/0009-0004-5754-8437; SPIN: 4969-6038
Андрей Глебович Васильев — д.м.н., профессор, заведующий кафедрой паталогической физиологии с курсом иммунологии. E-mail: avas7@mail.ru; https://orcid.org/0000-0002-8539-7128; SPIN: 1985-4025

Об авторах

Sarng S. Pyurveev — Cand. Sci. (Med.), Associate Professor of the Department of Pathological Physiology with a Course of Immunopathology, Saint Petersburg State Pediatric Medical University;Junior Research, Department of Neuropharmacology named after S.V. Anichkov, Institute of Experimental Medicine. E-mail: dr.purveev@gmail.com; https://orcid.org/0000-0002-4467-2269; SPIN: 5915-9767
Tatyana V. Brus — Cand. Sci. (Med.), Associate Professor, Department of Pathological Physiology with a Course of Immunopathology. E-mail: bant.90@mail.ru; https://orcid.org/0000-0001-7468-8563; SPIN: 9597-4953
Alexandra Yu. Pyrkh — Assistant, Department of Pathological Physiology with a course in Immunopathology. E-mail: apyrkh@yandex.ru; https://orcid.org/0009-0001-4761-5076; SPIN: 3117-3503
Anna E. Kurtukova — 4th-year student at the Faculty of Pediatrics. https://orcid.org/0009-0008-7985-782X; SPIN: 2674-3230
Yulian R. Ryzhov — Cand. Sci. (Med.), Head of Assisted Reproductive Technologies. https://orcid.org/0000-0002-5073-8279; SPIN: 8320-1234
Yulia A. Taminkina — Biophysicist. https://orcid.org/0000-0001-5256-9360; SPIN: 4095-7301
Zaur K. Emirgaev — Urologist of the Urology Department. https://orcid.org/0000-0002-8143-0461; SPIN: 6771-7532

* Corresponding author.
Автор, ответственный за переписку.

Authors' info

Сарнг Саналович Пюрвеев — к.м.н., доцент кафедры патологической физиологии с курсом иммунопатологии, Санкт-Петербургский государственный педиатрический медицинский университет; научный сотрудник отдела нейрофармакологии им. С.В. Аничкова, Институт экспериментальной медицины. E-mail: dr.purveev@gmail.com; https://orcid.org/0000-0002-4467-2269; SPIN: 5915-9767
Татьяна Викторовна Брус — к.м.н., доцент, кафедра патологической физиологии с курсом иммунопатологии. E-mail: bant.90@mail.ru; https://orcid.org/0000-0001-7468-8563; SPIN: 9597-4953
Александра Юрьевна Пырх — ассистент, кафедра патологической физиологии с курсом иммунопатологии. E-mail: apyrkh@yandex.ru; https://orcid.org/0009-0001-4761-5076; SPIN: 3117-3503
Анна Евгеньевна Куртукова — студентка 4-го курса педиатрического факультета. https://orcid.org/0009-0008-7985-782X; SPIN: 2674-3230
Юлиан Рэммович Рыжов — к.м.н., заведующий отделением вспомогательных репродуктивных технологий. https://orcid.org/0000-0002-5073-8279; SPIN: 8320-1234
Юлия Александровна Таминкина — биофизик. https://orcid.org/0000-0001-5256-9360; SPIN: 4095-7301
Заур Келбялиевич Эмиргаев — врач-уролог урологического отделения. https://orcid.org/0000-0002-8143-0461; SPIN: 6771-7532