ISSN 0006-2979, Biochemistry (Moscow), 2023, Vol. 88, No. 11, pp. 1763-1777 © Pleiades Publishing, Ltd., 2023.
Russian Text © The Author(s), 2023, published in Biokhimiya, 2023, Vol. 88, No. 11, pp. 2138-2155.

REVIEW

Microgravity Effects and Aging Physiology: Similar Changes or Common Mechanisms?

Andrey Yu. Ratushnyy[1] and Ludmila B. Buravkova[1,a]*

[1]*Institute of Biomedical Problems, Russian Academy of Sciences, 123007 Moscow, Russia*
e-mail: buravkova@imbp.ru

Received July 18, 2023
Revised October 13, 2023
Accepted October 14, 2023

Abstract—Despite the use of countermeasures (including intense physical activity), cosmonauts and astronauts develop muscle atony and atrophy, cardiovascular system failure, osteopenia, etc. All these changes, reminiscent of age-related physiological changes, occur in a healthy person in microgravity quite quickly – within a few months. Adaptation to the lost of gravity leads to the symptoms of aging, which are compensated after returning to Earth. The prospect of interplanetary flights raises the question of gravity thresholds, below which the main physiological systems will decrease their functional potential, similar to aging, and affect life expectancy. An important role in the aging process belongs to the body's cellular reserve – progenitor cells, which are involved in physiological remodeling and regenerative/reparative processes of all physiological systems. With age, progenitor cell count and their regenerative potential decreases. Moreover, their paracrine profile becomes pro-inflammatory during replicative senescence, disrupting tissue homeostasis. Mesenchymal stem/stromal cells (MSCs) are mechanosensitive, and therefore deprivation of gravitational stimulus causes serious changes in their functional status. The review compares the cellular effects of microgravity and changes developing in senescent cells, including stromal precursors.

DOI: 10.1134/S0006297923110081

Keywords: microgravity, aging, cell senescence, mesenchymal stem/stromal cells (MSCs)

INTRODUCTION

Despite the careful pre-flight selection, factors related to space flight, primarily weightlessness/microgravity, increase the risk of deterioration of the space traveler's health [1-3]. In this regard, pinpointing fundamental mechanisms of adaptation to microgravity-related effects at tissue, cellular, and molecular levels is one of the most essential scientific problems for space medicine. Resemblance of the physiological changes occurring during space flight to the processes developing during human aging is of special interest that raises a crucial question about the similarity of underlying molecular and cellular mechanisms. At the same time, restoration and normalization of physiological processes in organs and tissues is observed after returning to Earth, but aging leads to unidirectional, progressive pathological changes [1, 4].

It is recognized that in many physiological systems the long-term space flights could result in emergence of signs typical to aging [4-7]. Studies carried out on the MIR orbital station and the International Space Station revealed that the long-term stay in space results in decreased bone density [8-11], dysfunction of the immune system [12], issues related to cardiovascular system functioning [13, 14] as well as lowered skeletal muscle mass and strength [15]. Furthermore, cartilage tissue in the musculoskeletal system is also noted to be affected. In addition, the size of chondrogenic pellets, synthesis of proteoglycans, and dynamic stiffness of three-dimensional cartilage constructs are reduced [7]. Finally, moderate hypothyroidism, increased stress hormone (mainly catecholamines) and decreased sex steroid levels, insulin resistance, as well as systemic pro-inflammatory state could be observed as well [6, 16]. Similar

Abbreviations: ECM, extracellular matrix; MSCs, mesenchymal stem/stromal cells; Rb, retinoblastoma protein; ROS, reactive oxygen species; RPM, Random Positioning Machine; SMG, simulated microgravity

* To whom correspondence should be addressed.

changes occur upon aging under Earth's gravity, which develop, however, much faster in spaceflights.

Degenerative changes in the musculoskeletal system pose one of the most important risks upon exposure to microgravity. In this regard, a recent meta-review summarized the data from 25 experimental papers that analyzed post-flight bone density level as well as in-flight and post-flight bone biochemical markers in 148 and 124 subjects, respectively. It turned out that compared with the pre-flight level the cosmonauts who spent more than 28 days in space were found to have lower extremity bone density decreased by 5.4% (n = 96). After landing, level of the bone resorption markers decreased, and the balance shifted toward prevailing bone formation [11].

Some of the mechanisms underlying decline in bone density during spaceflight could highly resemble those related to osteoporosis developed in aging and/or due to physical inactivity. The reduced bone mass and osteoporosis has been observed both in older people as well as in immobilized or bedridden patients of any age [6] primarily highlighting a reduced mechanical load on the musculoskeletal system. In addition, the decreased level of sex hormones such as testosterone and estrogen should be noted. Interestingly, the male astronauts demonstrate both decreased workload and lowered testosterone level (comparable to the older men) [6, 17]. It should be noted that neither daily physical tasks nor mechanical loads can fully prevent osteopenia development.

Similar data were also observed in the animal study with 16-week-old female C57BL/6J mice exposed to microgravity for 15 days during the STS-131 mission. It was shown that the bone volume fraction and bone thickness decreased by 6.2% and 11.9%, respectively. A more detailed analysis revealed elevated number of osteoclasts along with higher expression of the bone matrix metalloproteinases (MMP-1, -3, and -10). At the same time, osteoblast expression of the *CDKN1a* gene encoding p21 (one of the cell cycle inhibitors and crucial markers of cellular senescence) was upregulated by 3.3-fold. Hence, it could indicate that alterations in bone homeostasis under microgravity conditions underlie osteocyte osteolysis and p21-mediated arrest in osteoblast cell cycle [18]. Thus, phenomenology of the long-term impact of space flight factors on bone tissue physiology allows to draw some parallels with the age-related changes suggesting existence of similar cellular mechanisms.

Recent studies with astronauts identify more links between the alterations related to spaceflight and aging. Modern molecular biological methods allow to assess the state of genetic apparatus upon different damages. Thus, American researchers analyzed temporal changes in the telomere (the ends of chromosomes) length and DNA damage response (DDR) in the peripheral blood mononuclear cells from 11 astronauts on board of the ISS before, during, and after the long-term spaceflights (up to a year). In this regard, shortened telomeric regions is considered as one of the classic signs of aging, which will be discussed in more detail below. Despite that all space travelers underwent strict medical selection having no health complaints, they had shorter telomere length and lower level of telomerase activity compared to the control age-matched healthy subjects. Interestingly, the telomere length increased slightly during space flights, but rapidly decreased upon return to Earth [19]. Hence, it was unveiled that despite of the individual differences the post-flight telomere length was decreased in almost all astronauts in comparison with the pre-flight length. Furthermore, a positive correlation between the oxidative stress and dynamically fluctuating telomere length was established. Aside from this, increased prevalence of the chromosomal inversions was observed during and after the space flights. It was proposed that regardless of telomerase activity in somatic cells the in-flight chronic oxidative stress transiently activates an alternative pathway for telomere length regulation [19].

Discussing the data on the telomere length increased during space flights, it should be noted that the cosmonauts and astronauts use physical activity (about an hour daily) as a countermeasure of adverse effects related to weightlessness. Analyzing the effects of running revealed its positive effect on peripheral blood mononuclear cell telomere length [20], the telomere length positively correlated with the performance level of athletes [21].

Considering telomere length (or any other sign of aging) as one of an "integrative markers" for cumulative effect of genetic and external cues (environment and lifestyle) on human aging, it is necessary to remember that together with microgravity, a healthy person becomes affected by the elevated level of radiation, transient changes in spacecraft normoxic atmosphere (increased CO_2 level, airborne organic impurities), altered microbiome, as well as stressful situations (e.g., extravehicular activities, failures of life support system elements, etc.), which should be taken into consideration while analyzing impact of space flights on physiological processes.

Interestingly, the level of blood plasma pro-inflammatory interleukins and chemokines in astronauts significantly correlated with the telomere length and increased during the long-term space flights. For instance, the astronauts' plasma samples contained higher level of various cytokines including pro-inflammatory cues such as TNFα, IL-8, IL-1ra, Tpo, VEGF, MCP-1, CCL4, CXCL5 [12] that could be considered as a sign of chronic (sterile) inflammation, one of the signs of aging.

Microgravity is a major element affecting health of astronauts during space flight [22]. Because physiological changes at an organismal level result from the modified cellular functioning, cellular changes related to aging and microgravity were compared.

It is necessary to remember that the central regulatory processes such as fluctuated biorhythms, hormonal status etc. affecting the cell state (and aging) in the context of an integral unified system also greatly impact the gravity-related physiological effects. These effects are the subject of separate investigation. In particular, they being considered in the Dilman's neuroendocrine theory of aging and in the hypothesis suggested by A. M. Olovnikov about potential molecular mechanisms for the development and aging of multicellular organisms, including involvement of neuroendocrine cells and gravitational effects. Moreover, perception of the gravitational infradian rhythms by the animals, which could become disrupted during space flight and, probably, result in accelerated aging, has been also mentioned [23-27].

CELL SENESCENCE

Nowadays, aging is often envisioned as the loss of body physiological integrity that proceeds over time resulting in its impaired functions and elevated risk of death. Hypotheses, theories, definitions of aging at various levels of life organization have been extensively discussed in the *Biochemistry (Moscow)* journal including the Issue 12-13, 2022. In this review we will solely refer to the hallmark issues. It is also worth noting about an epigenetic "clock" of aging proposed in 2013 by Horvath [28] and in 2017 by the scientific team headed by Dr. Gladyshev [29]. However, a rather broad application of the "clock" concept with regard to space biology needs to be further assessed.

Currently, at the cellular level, altered intercellular interaction, depleted stem cell pool, cellular aging, mitochondrial dysfunction, metabolic disorders, impaired proteostasis, genome instability, telomere shortening, and epigenetic changes are referred to as signs of aging at the cellular level [30]. Cell aging (senescence) is thought to be one of the essential arms in the host aging [31]. Such phenomenon is characterized by the irreversibly arrested cell cycle and could be accompanied by the prominent phenotypic changes including increased autophagic events, modulated metabolism, chromatin remodeling, production of proinflammatory cytokines, etc. [32-36]. Morphological changes including larger cell size and flattening should be highlighted among the most recognized markers of cell senescence [37]. In addition, the senescence-associated β-galactosidase (SA-β-gal) activity also increases [38] along with the frequency of emerging of heterochromatic γ-H2AX foci [39]. To some extent, formation of a senescent cell may be considered as a consequence and as a cause of aging. On the one hand, it results from alterations at the molecular level, translated into the senescence program at the cell level, which is an elementary unit of life organization. On the other hand, the senescent cell physiology transforms dramatically leading to the impaired tissue and organ functioning in the long term.

Cell senescence is generally referred to as an antagonistic trait that has both negative and positive sides. It is generally accepted that activation of the senescence state is the most crucial barrier to tumorigenesis. Uncontrolled tumor cell division and inability of the senescent cell to divide seem to be opposite consequences resulting from the same causes primarily represented by accumulation of damages in the cell genetic material [40, 41]. Cell senescence also plays an essential role in wound healing, where, e.g., upon skin wound healing, the matricellular protein CCN1 can induce senescence of fibroblasts or myofibroblasts and thereby reduce fibrosis [42]. Another aspect of the positive senescence-related effects may also be presented as the changed intercellular crosstalk. A number of paracrine mediators typical to senescent secretome are even used for the stromal progenitor cell priming in regenerative medicine. In recent years, various approaches have been explored to extend capabilities of the stromal progenitor cells that resulted in the development of new cell products with improved potential for diverse clinical applications [43]. These studies clearly illustrate ambiguity of negative and positive biological effects, which is also applicable to senescent cells.

Today, replicative and stress-induced cellular senescence are distinguished: the former is considered as a cell condition in which proliferative activity becomes irreversibly lost following a series of mitoses. The cell cycle arrest (in this case) largely results from the shortening of telomeric regions, which can be considered as a special case of genome instability. In 1961, cellular senescence was first described as the progressive and irreversible loss of proliferative potential in human somatic cells. It was shown that even under ideal culture conditions, human embryonic fibroblasts are capable of dividing only a limited number of times (50 ± 10) [44], the phenomenon named after the author, who discovered it – the "Hayflick limit". In 1971 A. M. Olovnikov proposed a hypothesis to explain this phenomenon based on the data regarding the principles of DNA synthesis in cells [45], this hypothesis was later confirmed experimentally. It states that with each cell division chromosomes become slightly shortened due to underreplication of telomeric DNA region. Human telomeres are the terminal regions of chromosomes that contain from 4 to 15 thousand base pairs and consist of TTAGGG repetitive sequences. It is known that DNA polymerase is unable to synthesize a daughter DNA copy from the end of its chain – it can only add nucleotides to the pre-existing 3'-hydroxyl group, i.e., requires an RNA primer. Upon removal of the last primer at the 3' end, the daughter strand will inevitably be shorter leading to gradual loss of telomeric nucleotides during successive cycles of DNA synthesis [46].

The stress-induced cellular senescence is also characterized by irreversible cell cycle arrest. Unlike in the case of replicative cellular senescence, it is not associated with the number of cell divisions. The stress-induced senescence is triggered by the sublethal damages or oncogene activation [36, 47]. Most often, this condition is related to oxidative stress, i.e., an imbalance between the oxidants [usually reactive oxygen species (ROS)] and antioxidant systems. An effect of ROS on DNA including mtDNA induces formation of the products of oxidative DNA base damage such as 8-hydroxy-2'-deoxyguanosine (8OHdG) and could also lead to the strand breaks [48]. Apart from this, mitochondrial ROS could activate the c-Jun N-terminal kinase (JNK), which, in turn, facilitates release of the chromatin fragments into cytoplasm and activation of the pro-inflammatory secretome [49]. Interestingly, oxidative stress could also accelerate telomere shortening [50] probably owing to high content of guanine (G), which is most vulnerable to ROS [51].

The programs of replicative and stress-induced cellular senescence are executed via similar mechanisms. Relatively small DNA damage results in the temporary arrest of the cell cycle. After successful DNA repair, a cell can begin to divide again. By definition, this condition cannot be called cell senescence. More profound damage that cannot be repaired for a long time triggers a chronically activated DDR signaling cascade, cell response to the genetic material damage. The chronically activated DDR usually occurs upon multiple DNA damages and represents a major hallmark of the senescent state – stable cell cycle arrest. The latter is achieved via activation of the p16INK4a/Rb and p53/p21CIP1 tumor suppressor signaling cascades [47, 51] so that the cell would never be able to start division again. Suppression of activity of the relevant cyclin-dependent kinases by both inhibitors, p21 and p16 (encoded by the *CDKN1A* and *CDKN2A* genes, respectively), results in hypophosphorylation of the retinoblastoma protein (Rb). The hypophosphorylated Rb is able to bind E2F family transcription factors regulating cell cycle [52, 53]. By reversibly binding and, consequently, functionally inactivating E2F proteins, Rb controls expression of the genes, products of which are essential for regulating cell cycle as well as blocking the G1-to-S phase transition. In this scenario, the p53/p21 pathway is predominantly activated first by preventing proliferation of the cells with serious DNA damage, whereas the p16/Rb-axis becomes involved somewhat later [32]. However, based on the cellular context, one or another may become preferable.

Cellular senescence could be affected by various mechanical forces including shear stress, tension, and pressure [54, 55]. This raises the question whether microgravity could be considered as a factor causing cellular senescence or other similar changes in the cell physiology?

SENESCENCE-ASSOCIATED ALTERATIONS UNDER SIMULATED MICROGRAVITY

During the space flight, human body is exposed to multiple unfavorable stress factors noted above that can aggravate development of the aging-associated signs. At the cellular level, they may be presented by the changed radiation background able to disrupt DNA integrity and promote oxidative stress. In this review, the microgravity-related effects will be discussed in detail.

In this review, the senescence-associated changes occurring in the diverse cell types will be assessed in the simulated microgravity (SMG) settings. Due to the technical limitations posed by experiments in space missions, various ground-based models are commonly used. Usually, such models are aimed at gravitational "unloading" to simulate some of the microgravity-related effects. Ground-based experiments also allow to avoid an impact from the increased background radiation and other space flight-associated factors. To simulate microgravity effects on cell cultures, rotating wall vessels (RWV) and 2D/3D clinostats such as a device for randomizing object position relative to the gravity vector (Random Positioning Machine, RPM) are used most often. It is believed that the computer-controlled RPM consisting of two frames rotating in two perpendicular planes represents the most suitable approach for simulating microgravity effects on the adherent cell cultures. Using the RPM allows to randomize object's position relative to the gravity vector. In standard operating modes, the device simulates gravitational acceleration equivalent to 10^{-2} g [56-58].

Various cell cultures including immortalized lines, endothelium, stromal precursors, etc. have been used in the studies [59-63]. A wide range of changes demonstrating direct gravity-related effects on the cellular structures were revealed in the studies investigating *in vitro* cell morphofunctional state. A change in position of the heavy organelles such as nucleus results in redistributed load on the cytoskeleton followed by its reorganization; modifications in the adhesion molecule physical interaction between the cell and extracellular matrix also occur. Altogether, it results in the altered gene expression, changes in functioning of multiple proteins, and overall modification of the cell functional state [64-68].

Some studies using SMG attempted to identify activated senescence in the pheochromocytoma cells (PC12), erythrocytes, skeletal muscle myoblasts, and cardiomyocytes [59, 69-71]. Wang et al. [59] used SMG to analyze early (6-96 h) effects on the rat neuronal PC12 cell line. It was shown that the cell cycle was arrested in the G1 phase along with the increased SA-β-gal activity as well as activated p53 and p16 signaling cascades associated with senescence. More detailed

analysis revealed elevated level of ROS, which could induce cellular senescence. Activity of the intracellular antioxidant enzymes such as superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), and catalase (CAT) were markedly enhanced 12 h later but decreased 96 h after onset of the experiment. Moreover, the antioxidant N-acetylcysteine was able to profoundly block the ROS-associated effects that prominently abrogated the microgravity-related increase in the SA-β-gal activity. These data allowed to suggest that SMG exposure elicits cellular senescence in the PC12 cells via enhanced oxidative stress [59].

Modifications in the human erythrocyte structure and functions were assessed by using a 3D clinostat. For this, the erythrocyte structural parameters were analyzed together with the cellular senescence-specific metabolic parameters. The obtained data show that the long-term microgravity exposure results in emergence of the senescence-specific morphological patterns [69].

It was shown that SMG with the Zeromo 3D clinostat accelerates senescence in the human skeletal muscle myoblast culture. A markedly decreased proliferation, typical cytoskeletal remodeling, and hypertrophy of the cell nuclei as well as upregulated SA-β-gal expression were demonstrated. Similar changes are observed in the senescent myoblasts after several passages. It was noted that such effects were sustained even after returning to the normal gravitational conditions. Moreover, the myoblasts exposed to SMG demonstrated a reduced ability to differentiate into myotubes [70].

More recently, studies were conducted with the human iPSC (induced pluripotent stem cell)-derived cardiomyocytes. It was revealed that exposure to SMG resulted in chromosomal reorganizations, downregulated mitochondrial function, elevated ROS level, as well as other signs indicative of cellular senescence. It is worth noting that the following study limitations were specifically highlighted, which are difficult to disagree with: (i) short-term SMG exposure (48 h), (ii) further experiments assessing durability of the detected changes are required [71].

Other researchers tend to believe that ROS are involved in the DNA damage during SMG exposure. For instance, it has been noted that SMG induces DNA damage and mitochondria-mediated apoptosis via increased ROS production in the human promyelocytic leukemia cells [72]. Earlier studies already revealed that clinorotation affects mitochondria, one of the main cellular free radical producers, thereby inducing apoptosis in the thyroid cells. In particular, it was found out that 24 h after the onset of experiment 10% of the thyroid carcinoma cells (ONCO-DG1 cell line) entered the Fas-dependent apoptotic pathway. Mitochondrial destruction and redistribution, disrupted microtubules, and activated effector caspase-3 were detected. Clinorotation was also elicited apoptosis in the normal thyroid cells (HTU-5) as evidenced by the caspase-3 activation and elevated Fas and Bax protein levels [73].

Investigation of the mouse embryonic stem cells demonstrated enhancement of the ROS-associated effects under SMG conditions. The authors added hydrogen peroxide to the cells and the number of DNA double-strand breaks was analyzed. It turned out that 24-hour-exposure of the treated cells to microgravity substantially elevated the degree of DNA damage [74]. In 2003, similar data were reported by Greco et al. [75] demonstrating that the frequency of chromosomal aberrations increased by around 1.2-2.8-fold in the post-flight blood samples exposed to X-ray radiation in comparison with the pre-flight blood samples. At the same time, another work shows that the early response to bleomycin-induced DNA damage (the number of γ-H2AX foci) was similar in both cells exposed to microgravity and those under control static ground conditions [76].

Thus, a set of studies indicate that exposure to SMG could result in the development of some signs of cell senescence. Moreover, a mechanism involving oxidative stress has been even suggested for this phenomenon, which, to some extent, is associated with mitochondrial dysfunction. Nonetheless, not a single study is available to confirm permanent cell cycle arrest, i.e., there is no clear evidence that after SMG exposure cells can no longer proliferate. Therefore, it is too early to conclude unequivocally that cellular senescence is initiated under exposure to SMG. We believe that the effect of SMG is hardly strong enough to cause such a profound damage at the cellular level.

SENESCENCE-ASSOCIATED STROMAL PROGENITOR FUNCTIONAL STATE IN SMG SETTINGS

Mesenchymal stromal/stem cells (MSCs) or stromal progenitors, which play a major role in renewal and regeneration, have been found in almost all body tissues. MSCs are involved in maintaining bone tissue homeostasis, hematopoiesis, regulated immunomodulation, and angiogenesis, etc. MSCs are of considerable interest both for basic science and applied regenerative medicine including age-related pathologies. To date, a consensus has been reached allowing to refer the MSC-associated beneficial effects to production of various secreted factors including extracellular matrix components and cytokines [77-80].

Some studies suggest that pathological changes in astronauts could be associated with the stromal precursor cell senescence. Further assessment of the microgravity-related effect on MSC senescence contributes to understanding of the role of senescent cells in the development of physiological and pathological changes

under spaceflight conditions [81]. Aging of an organism correlates with the decreased MSC functional activity. It decreases the rate of tissue repair typical to aging. For instance, osteoporotic bone fractures in older people heal more slowly due to the decreased MSC functions and their count [82].

As noted above, oxidative stress may be the main cause underlying cell damage. Microgravity is a stressor potentially able to induce ROS production, ultimately resulting in various damages to subcellular compartments [83]. Increased level of free radicals and mitochondrial dysfunction in MSCs have been noted in the recent study. However, addition of the antioxidant restored mitochondrial functions and reversed cell senescence. Moreover, SMG promoted expression of YAP (Yes-associated protein) and its translocation into the nucleus. YAP is an important effector in the Hippo signaling pathway that regulates development, homeostasis, and regeneration [81, 84, 85]. It is known that YAP could regulate cell senescence by acting on ATM (ataxia-telangiectasia mutated kinase), p53/p21, p16/CDK/Rb, autophagy, AMPK, mTOR, and SIRT1 signaling pathways [86-88]. Verteporfin (VP), YAP inhibitor, restored the SMG-induced MSC mitochondrial dysfunction and senescence [81].

Some other studies also suggested that SMG causes stromal progenitor cell senescence. Molecular changes associated with stemness (OCT-4, SOX2, NANOG) and cellular senescence (p19, p21, p53) in the MSCs isolated from the Wharton's jelly were analyzed. The authors supposed that the results of the study indicate cellular adaptation occurring within the first hours after exposure followed by loss of stemness and emergence of the signs of molecular senescence program [89]. Bellow the main physiological parameters of MSCs exposed to microgravity will be discuss in more detail.

Proliferation. Proliferation is one of the major cell characteristics particularly in the context of senescence. Firstly, MSC senescence is typically characterized by permanent cell cycle arrest in the G1 phase, i.e., senescent MSCs cannot proliferate and form colonies [36, 90, 91]. Several studies evidence, at least slightly decreased MSC proliferative potential upon SMG conditions. For instance, over time (from 1 h to 10 days) clinostating resulted in lowered proliferation rate and changed morphology of the cells, which became flatter and reached confluency at lower density. When exposure period prolonged up to 20 days, proliferation rate also decreased, while the number of large flat culture cells increased [92, 93]. Such changes can also be the signs of senescence. Study with the rat bone marrow MSCs confirms the findings published previously [94]. It is noted that SMG inhibits MSC growth at the G0/G1 phase of the cell cycle.

Other studies not only failed to obtain similar results, but, on the contrary, observed the opposite effect. Yuge et al. [95] showed that proliferation rate of the human MSC in the 3D clinostat increased by almost 3-fold under SMG in comparison with the control group. Hence, it was noted that SMG can be used to enhance stem cell expansion *in vitro*. We investigated functional status of the human adipose tissue-derived MSCs under SMG conditions (96 h) using RPM. It was found that the cell count increased 1.5-2-fold, whereas activity of the lysosomal compartment, cell size, and granularity decreased. No change in the ROS levels or mitochondrial transmembrane potential was observed. Therefore, this study suggests lack of the signs associated with cell stress during MSCs culturing under SMG settings [96].

Investigation of the more committed MSCs osteoblasts, showed that SMG did not affect cell growth or viability. Cells were incubated in the 3D clinostat for 12-96 h. Twenty-four hours after the onset of the experiment, the Bax/Bcl-2 mRNA level ratio (parameter of apoptosis) increased up to 136% relative to the static control. However, it was accompanied by the increase of XIAP (anti-apoptotic molecule) mRNA level to 138% of the static control. No DNA fragmentation was observed and the level of the effector caspase-3 mRNA remained unaltered [97].

Thus, it is at least premature to draw conclusions about the lowered MSC proliferation or enhanced apoptosis under SMG conditions, which requires further detailed investigated.

Differentiation. Reduced multipotency is considered as an important feature of senescent MSCs [90, 91], which may weaken their reparative potential in all tissue types. Shift in the balance between adipocyte and osteocyte lineages has been observed, although a specific vector for it remains debatable. Some studies noted decline in the osteogenic properties of MSC with increased duration of the cells culturing or upon senescence [98-100]. Other studies revealed no such changes or even reported elevated osteogenic potential [101, 102]. Such data obtained in different studies are usually accounted for by different methodological approaches and experimental models, heterogeneity of the MSC populations, as well as lack of assays accurately assessing osteogenic differentiation [98, 103]. The most informative *in vitro* marker of osteogenic potential is presented by relevant differentiation pathway followed by detection of matrix mineralization. At the same time, elevated level of cell death may result in the false-positive data due to the large calcium ion release from the dying cells and its binding to extracellular matrix [36, 90, 91]. Regarding the adipocyte potential, we are getting closer to a consensus. A somewhat wide range of data has been accumulated, however, majority of the researchers come to the conclusion that adipogenic potential wanes upon senescence [91].

Our own data clearly and unambiguously indicate reduced adipogenic potential of the MSCs isolated from

adipose tissue during replicative senescence. This is manifested by lack of pronounced formation of lipid inclusions during differentiation. A markedly downregulated expression of the gene encoding the key transcriptional regulator PPARγ was detected, which probably underlies this phenomenon. On the other hand, signs pointing at the reciprocally elevated osteogenic potential, despite the downmodulated expression of the genes coding for some positive regulators of the osteoblastic pathway (*BMP2*, *BMP6*, *IGF1*, *IL1B*) were additionally observed. In the process, intensity of the matrix calcification and osteoprotegerin concentration during differentiation increased, which may be essential in the context of atherosclerotic plaque calcification in elderly people. Transcriptional activity of the key osteogenesis regulator (RUNX2) and some analyzed marker genes (*SPARC*, *SPP1*, *COL1A1*, *BGLAP*) remained stable during the replicative senescence of MSCs [104]. Despite potentiated calcification of the matrix, its morphology differed in the "young" and senescent cell cultures that may indicate false positive data due to calcium release [36, 90, 91]. At the same time, increased osteoprotegerin production observed in the study may indicate a pro-osteogenic paracrine activity in the senescent MSCs [104].

A greater consensus was achieved on the issue regarding osteogenic differentiation under SMG setting results, with vast majority of the studies agrees that microgravity contributes to the reduced osteogenic potential in MSC [105]. In particular, such effects were demonstrated in the rat [94, 106] and human [107, 108] bone marrow MSCs, which was confirmed by Saxena et al. [109] revealing that SMG inhibits the MSC-related osteoblastogenesis and enhances adipocytogenesis under osteogenic conditions. The authors believed that this process involves decreased RhoA activity and cofilin phosphorylation, disruption of F-actin stress fibers, as well as decreased focal adhesion kinase-mediated integrin signaling. Other studies reported that the lowered osteoblastogenesis under SMG settings is, at least in part, caused by downmodulated integrin/MAPK signaling [110].

Transcriptome analysis using a genome-wide microarray showed that 882 genes were downregulated, and 505 genes were upregulated after 24-hour SMG exposure. In particular, a significantly reduced expression of the osteocytic and chondrocytic genes, as well as higher expression level of the adipocyte genes was noted [111].

More recently, attention was attracted to non-canonical pathways of cell differentiation. For instance, SMG turned out to promote MSC differentiation into neurons evidenced by the upregulated expression of several relevant markers. Moreover, secretion of neurotrophins such as nerve growth factor (NGF), brain-derived neurotrophic factor (BDNF), or ciliary neurotrophic factor (CNTF) was elevated [112]. In another study, the rat MSCs were cultured for 72 h or 10 days in a clinostat followed by expansion in various differential culture media. It was found that the short-term exposure (72 h) promoted endothelial, neuronal, and adipogenic differentiation, whereas the long-term exposure (10 days), unexpectedly, facilitated MSC differentiation into osteoblasts. In addition, the short-term SMG exposure profoundly downregulated RhoA activity, which, however, increased with prolonging SMG exposure. Hence, such data demonstrated that the duration of SMG exposure regulates MSC differentiation via the RhoA-associated pathway [113], which corroborated the above-mentioned data obtained by Saxena et al. [109].

Thus, SMG can largely affect MSC differentiation potential. Such effect is of inhibitory nature, at least with regard to osteogenic differentiation, which may explain similar physiological consequences related to the impaired bone metabolism.

Secretory activity. It is assumed that the senescent cells could contribute to maintenance of chronic inflammation and development of aging-associated diseases, hence, investigating the issue of paracrine regulation is of great importance. Senescent cells continue to interact with the surrounding environment and exert local and systemic effects primarily through paracrine regulation. The secretory activity changes dramatically, and this phenotype was even named accordingly – SASP (senescence-associated secretory phenotype). First of all, the SASP is characterized by increased proinflammatory secretome, although individual cues vary widely depending on the cell type and the way to induce senescence [36, 90, 91, 114].

It was discovered that the level of one of the main pro-inflammatory cytokines, IL-8, in the human bone marrow-derived MSCs increased in the culture medium during culturing in 2D clinostat by 1.4-3.2-fold (20-d exposure), as well as 1.5-6-fold (10-d exposure) and 1.6-2.1-fold (20-d exposure) on average in the culture subjected to RPM [115]. Later, studies with the human adipose tissue-derived MSCs complemented these findings. Assessing the changes in paracrine activity upon 96-hour exposure to RPM showed elevated production of IL-8 accompanied by the decreased IL-6 level. At the same time, higher production of VEGF, a key positive angiogenesis regulator, was noted [96].

Our study demonstrated that the post-RPM MSC-conditioned medium stimulated formation of the vascular network *in ovo*, of the capillary-like network of endothelial cells (ECs) in Matrigel, and the EC non-directional migration *in vitro*. Such effects were explained by the changed expression of the genes and proteins associated with angiogenesis, primarily by the higher level of expression of the angiogenesis regulators Serpin E1, Serpin F1, IGFBP, VEGF, and IL-8 along with upregulated transcription of the genes encoding proangiogenic growth factors such as *VEGF-c* and *VEGF-a*. Hence, such data demonstrate that microgravity may exert an MSC-mediated effect on the EC functional state [63].

In addition, we found higher transcriptional activity of the brain-derived neurotrophic factor, BDNF [63], corroborating the findings reported by Chen et al. [112] about potentiated MSC differentiation into neuron-like cells under SMG. Additionally, therapeutic efficacy of the microgravity-cultured MSCs after spinal cord ischemia-reperfusion injury was assessed revealing higher count of the BDNF-positive and lower count of the caspase-3-positive apoptotic astrocytes, as well as restored cell motility, which implies existence of positive effect of the post-SMG MSCs on regenerative process [116].

From the viewpoint of assessing the senescent state in progenitor cells, it is important to study the proinflammatory secretome. While analyzing the SMG-related effect of the adipose tissue-derived MSCs on the TNFα-mediated priming, it was shown that SMG *per se* results in no changes in the surface expression of ICAM-1 and HLA-ABC, which can be considered as markers of pro-inflammatory cell activation. A weakened MSC response to the TNFα priming upon SMG exposure was documented in the process that was manifested as downmodulated production of the TNFα-dependent pleiotropic cytokines (IL-8 and MCP-1), matrix remodeling proteases, and suppression of some genes encoding growth factors and cytokines [117].

Along with the proinflammatory cytokines, a stimulatory effect related to the 10-day exposure to microgravity on paracrine activity of the osteocommitted and intact MSCs was evaluated. It was found that the MSC response to SMG depended on the degree of cell commitment so that for the osteocommitted MSCs it was less pronounced and manifested by higher production of sclerostin, negative regulator of osteoblastogenesis. In contrast, the intact MSCs were distinguished by low osteoprotegerin production. Such SMG-related changes may underlie shift of the bone homeostasis towards bone resorption [118]. In this regard, it is worth reminding that the osteoprotegerin level becomes increased during senescence [104].

Thus, it may be noted that exposure to SMG leads to elevated secretion of the pro-inflammatory cytokine IL-8, which, in turn, could promote production of the downstream cues such as VEGF. Experiments show that the changes in the secretome could be considered for application in regenerative medicine to enhance neuro- and angiogenesis. It is known that the pro-inflammatory SASP in the senescent cells could also positively affect tissue regeneration, whereas potential negative consequences could be realized only upon chronic exposure. Nonetheless, it is still premature to suggest the existence of similarities between the SMG exposure and senescence in the context of secretome.

Extracellular matrix. In addition to cells *per se* and relevant paracrine cues, extracellular matrix (ECM) plays an important role in the body tissue functioning. It varies greatly depending on localization and mediates intercellular interactions. The cell-ECM crosstalk is necessary for normal cell functioning including cell proliferation and differentiation [119, 120]. Various ECM components perform specific functions. Proteoglycans retain water, deposit metabolites and growth factors due to their molecular structure and large number of charged groups [121]. On the other hand, collagen and fibronectin as protein constituents ensure tissue mechanical properties that cells rely on to maintain their own shape as well as to migrate. Together with other ECM proteins such as elastin and laminin, they ensure matrix elasticity.

Available publications point at changes of ECM in the senescent cells associated with their catabolic phenotype. In particular, upregulated expression of proteolytic enzymes (matrix metalloproteinases, adamalysins (ADAMs), urokinases and cathepsins) and lowered production of ECM structural components (collagens, glycoproteins, proteoglycans) was demonstrated. Eventually, it results in the reduced tissue elasticity, basement membrane damage, and increased ECM stiffness [122].

At the moment, studies are mainly focused on assessing effects of the ECM produced by young and senescent cells on functional activity of the cellular elements in the tissue niche. The study by Choi et al. [123] showed that the senescent fibroblasts seeded on the ECM derived from the early passage fibroblasts had reduced SA-β-gal expression along with the decreased free radical levels, restored mitochondrial potential, as well as telomere elongation. And *vice versa*, decline in proliferation of the "young" fibroblasts cultured on the senescent cell-derived ECM was observed [123].

The study of bone marrow MSCs obtained from the young (3 weeks) and old (18 weeks) animals described altered ECM properties. For this, the MSCs from young and old mice were seeded onto decellularized matrix from the relevant cell groups. It turned out that the ECM released from the young MSCs produced lowered ROS level that decreased by 30-50% in both young and senescent MSCs compared to the ECM derived from the old MSCs or cells grown on plastic [124]. It was also shown that cultivation of the synovial fluid-derived senescent MSCs on the decellularized fetal matrix enhances the MSC potential to undergo chondro- and adipodifferentiation [125].

From the viewpoint of gravireception, the ECM–integrin–cytoskeleton complex represents a mechanosensitive platform that coordinates cell and tissue functional state in the gravitational field [68]. The Myoui's group assessed cell differentiation [106] while the rat bone marrow MSCs were cultured for 2 weeks inside the pores of calcium hydroxyapatite on a 3D clinostat. It was discovered that compared with the control group, alkaline phosphatase activity (a marker of osteoblastic differentiation) reduced by 40%. The MSC-containing composites implanted into the syngeneic rats revealed

that bone formation was markedly lowered upon SMG exposure. It is important to note that the lower amount extracellular matrix was observed in the culture cultivated in clinostat, so these two events might be related [106]. More recent work noted that exposure to SMG resulted in elevated expression of the adhesion molecules (ITGB1, CD44), MMP1 protease, as well as one of the collagen types (ColIII) in the MSCs. MMP1 is known to degrade interstitial collagens including ColIII. It is likely that the authors observed a compensatory reaction in this case. Expression of the *FBN1* and *VIM* genes was downregulated. FBN1 is an extracellular matrix glycoprotein required for elastic fiber formation, whereas VIM is the major intermediate filament in the stromal progenitor cell cytoskeleton [126].

Recent studies in our laboratory complemented the results obtained previously. It was shown that the 10-day exposure to RPM results in the decreased level of collagen components in ECM likely due to the decreased collagen synthesis and protease activation. The presented data demonstrate that the ECM-associated molecules from both native and osteocommitted MSCs could be involved in the bone matrix reorganization during spaceflight [127].

Flight experiments on the Foton 10 spacecraft demonstrated downregulated expression of the major structural protein COL1A in the MG-63 osteoblast cell line. In addition, the SJ-10 experiment showed that the 2-day-spaceflight was associated with the lowered level of several genes encoding matrisome structural proteins and elevated MMP1 expression in the bone marrow MSCs. An inhibitory effect on the COL1A2 level was observed additionally after 5 days of spaceflight. Based on the results of onboard experiments, it can be concluded that the matrisome structural proteins are negatively affected by microgravity at the transcriptional level [128, 129].

It is easy to recognize certain similarities between the changes occurring during activation of cellular senescence and upon exposure to SMG. In both cases, less amounts of the ECM constituents are produced, which is accompanied by the higher protease activity. It is likely that the negative link between the matrix degradation and osteogenic differentiation really exists.

CONCLUSION

Development of physiological/pathophysiological alterations in humans during long-term space flights may be a sign of the "atrophic syndrome" described repeatedly by G. Libertini [130]. This syndrome is characterized by the lowered cell duplication capacity, decreased number of cells, substitution of specific cells with nonspecific cells, hypertrophy of the remaining specific cells, altered functioning of the cells with shortened telomeres, and altered cellular microenvironment depending on the state of senescent cells [130]. The atrophic syndrome resulting from the unloading is considered reversible in the spaceflights by duration no more than a year under condition when a fairly wide range of preventive measures is performed on board of the orbital space stations. In our opinion, it is impossible to determine whether there are some threshold values for decline in gravity or a maximum permissible time spent in the unloading environment without countermeasures, below which the essential physiological systems would lose own functional potential due to the processes similar to those occurring in aging. Moreover, almost nothing is known about the microgravity-related effect on the lifespan of different organisms including mammals.

Summarizing some experimental data of the microgravity effects on the cells and comparing the identified effects to the senescence-associated changes, it should be noted that microgravity may probably trigger initial stages of the stress-induced reactions. Some of the studies discussed here directly suggest similar mechanisms underlying the analogy response to microgravity and senescence. Even upon the short-term exposure, such shifts can cause lowered proliferation, skewed differentiation pathway, altered secretory profile including paracrine mediators and ECM-associated molecules. At the same time, the question regarding reversibility of the changes discovered during SMG remains open, because such reversibility does not allow to infer senescence directly. And, secondly, it should be noted that the vast majority of experimental studies proving that the cellular senescent state could be activated *in vitro* rely on the short-term microgravity exposures (24-72 h), which is clearly insufficient to enable the cell senescence program.

Contributions. L.B.B. proposed initial concept of the study; L.B.B. and A.Yu.R. wrote the manuscript, comparatively analyzed microgravity- and aging-related effects discussed in the review.

Funding. The study was financially supported equally by the program of fundamental research of the State Scientific Center of the Russian Federation – Institute of Biomedical Problems of the Russian Academy of Sciences (Topic 65.3) and by the Russian Science Foundation (grant no. 21-75-10117).

Ethics declarations. The authors declare no conflict of interest in financial or any other sphere. The article contains no description of studies with human subjects or animals performed by any of the authors.

REFERENCES

1. Demontis, G. C., Germani, M. M., Caiani, E. G., Barravecchia, I., Passino, C., and Angeloni, D. (2017) Human pathophysiological adaptations to the space environment, *Front. Physiol.*, **8**, 547, doi: 10.3389/fphys.2017.00547.

2. Tran, K. N., and Choi, J. I. (2022) Mimic microgravity effect on muscle transcriptome under ionizing radiation, *Life Sci. Space Res.*, **32**, 96-104, doi: 10.1016/j.lssr.2021.12.002.
3. Patel, S. (2020) The effects of microgravity and space radiation on cardiovascular health: from low-Earth orbit and beyond, *Int. J. Cardiol. Heart Vasc.*, **30**, 100595, doi: 10.1016/j.ijcha.2020.100595.
4. Vernikos, J., and Schneider, V. S. (2010) Space, gravity and the physiology of aging: parallel or convergent disciplines? A mini-review, *Gerontology*, **56**, 157-166, doi: 10.1159/000252852.
5. Wang, E. (1999) Age-dependent atrophy and microgravity travel: what do they have in common? *FASEB J.*, **13**, S167-S174, doi: 10.1096/fasebj.13.9001.s167.
6. Biolo, G., Heer, M., Narici, M., and Strollo, F. (2003) Microgravity as a model of ageing, *Curr. Opin. Clin. Nutr. Metab. Care*, **6**, 31-40, doi: 10.1097/00075197-200301000-00006.
7. Strollo, F., Gentile, S., Strollo, G., Mambro, A., and Vernikos, J. (2018) Recent progress in space physiology and aging, *Front. Physiol.*, **9**, 1551, doi: 10.3389/fphys.2018.01551.
8. Burger, E. H., and Klein-Nulend, J. (1998) Microgravity and bone cell mechanosensitivity, *Bone*, **22**, 127S-130S, doi: 10.1016/s8756-3282(98)00010-6.
9. Keune, J. A., Branscum, A. J., Iwaniec, U. T., and Turner, R. T. (2015) Effects of spaceflight on bone microarchitecture in the axial and appendicular skeleton in growing ovariectomized rats, *Sci. Rep.*, **5**, 18671, doi: 10.1038/srep18671.
10. Gerbaix, M., Gnyubkin, V., Farlay, D., Olivier, C., Ammann, P., Courbon, G., Laroche, N., Genthial, R., Follet, H., Peyrin, F., Shenkman, B., Gauquelin-Koch, G., and Vico, L. (2017) One-month spaceflight compromises the bone microstructure, tissue-level mechanical properties, osteocyte survival and lacunae volume in mature mice skeletons, *Sci. Rep.*, **7**, 2659, doi: 10.1038/s41598-017-03014-2.
11. Stavnichuk, M., Mikolajewicz, N., Corlett, T., Morris, M., and Komarova, S. V. (2020) A systematic review and meta-analysis of bone loss in space travelers, *NPJ Microgravity*, **6**, 13, doi: 10.1038/s41526-020-0103-2.
12. Crucian, B., Simpson, R. J., Mehta, S., Stowe, R., Chouker, A., Hwang, S. A., Actor, J. K., Salam, A. P., Pierson, D., and Sams, C. (2014) Terrestrial stress analogs for spaceflight associated immune system dysregulation, *Brain Behav. Immun.*, **39**, 23-32, doi: 10.1016/j.bbi.2014.01.011.
13. Sofronova, S. I., Tarasova, O. S., Gaynullina, D., Borzykh, A. A., Behnke, B. J., Stabley, J. N., McCullough, D. J., Maraj, J. J., Hanna, M., Muller-Delp, J. M., Vinogradova, O. L., and Delp, M. D. (2015) Spaceflight on the Bion-M1 biosatellite alters cerebral artery vasomotor and mechanical properties in mice, *J. Appl. Physiol.*, **118**, 830-838, doi: 10.1152/japplphysiol.00976.2014.
14. Hughson, R. L., Yee, N. J., and Greaves, D. K. (2016) Elevated end-tidal Pco_2 during long-duration spaceflight, *Aerospace Med. Hum. Perform.*, **87**, 894-897, doi: 10.3357/AMHP.4598.2016.
15. Shen, H., Lim, C., Schwartz, A. G., Andreev-Andrievskiy, A., Deymier, A. C., and Thomopoulos, S. (2017) Effects of spaceflight on the muscles of the murine shoulder, *FASEB J.*, **31**, 5466-5477, doi: 10.1096/fj.201700320R.
16. Novikov, V. E., and Ilyin, E. A. (1981) Age-related reactions of rat bones to their unloading, *Aviat. Space Environ. Med.*, **52**, 551-553.
17. Strollo, F., Riondino, G., Harris, B., Strollo, G., Casarosa, E., Mangrossa, N., Ferretti, C., and Luisi, M. (1998) The effect of microgravity on testicular androgen secretion, *Aviat. Space Environ. Med.*, **69**, 133-136.
18. Blaber, E. A., Dvorochkin, N., Lee, C., Alwood, J. S., Yousuf, R., Pianetta, P., Globus, R. K., Burns, B. P., and Almeida, E. A. (2013) Microgravity induces pelvic bone loss through osteoclastic activity, osteocytic osteolysis, and osteoblastic cell cycle inhibition by CDKN1a/p21, *PLoS One*, **8**, e61372, doi: 10.1371/journal.pone.0061372.
19. Luxton, J. J., McKenna, M. J., Lewis, A., Taylor, L. E., George, K. A., Dixit, S. M., Moniz, M., Benegas, W., Mackay, M. J., Mozsary, C., Butler, D., Bezdan, D., Meydan, C., Crucian, B. E., Zwart, S. R., Smith, S. M., Mason, C. E., and Bailey, S. M. (2020) Telomere Length dynamics and DNA damage responses associated with long-duration spaceflight, *Cell Rep.*, **33**, 108457, doi: 10.1016/j.celrep.2020.108457.
20. Zhuikova, S. E., and Nagovitsyn, R. S. (2021) Influence of running on some physiological and molecular biological markers of human aging, *Hum. Physiol.*, **47**, 587-594, doi: 10.1134/S0362119721050133.
21. Simoes, H. G., Sousa, C. V., Dos Santos Rosa, T., da Silva Aguiar, S., Deus, L. A., Rosa, E. C. C. C., Amato, A. A., and Andrade, R. V. (2017) Longer telomere length in elite master sprinters: relationship to performance and body composition, *Int. J. Sports Med.*, **38**, 1111-1116, doi: 10.1055/s-0043-120345.
22. Prasad, B., Grimm, D., Strauch, S. M., Erzinger, G. S., Corydon, T. J., Lebert, M., Magnusson, N. E., Infanger, M., Richter, P., and Krüger, M. (2020) Influence of microgravity on apoptosis in cells, tissues, and other systems *in vivo* and *in vitro*, *Int. J. Mol. Sci.*, **21**, 9373, doi: 10.3390/ijms21249373.
23. Fernando, H. J., and Bowers, D. (2021) Neuroendocrine Theory of Aging, in *Encyclopedia of Gerontology and Population Aging* (Gu, D., and Dupre, M. E., eds) Springer, Cham, doi: 10.1007/978-3-030-22009-9_673.
24. Dilman, V. M., Revskoy, S. Y., and Golubev, A. G. (1986) Neuroendocrine-ontogenetic mechanism of aging: toward an integrated theory of aging, *Int. Rev. Neurobiol.*, **28**, 89-156, doi: 10.1016/s0074-7742(08)60107-5.
25. Olovnikov, A. (2005) Lunasensor, infradian rhythms, telomeres, and the chronomere program of aging, *Ann. NY Acad. Sci.*, **1057**, 112-132, doi: 10.1196/annals.1356.006.

26. Olovnikov, A. M. (2015) Chronographic theory of development, aging, and origin of cancer: role of chronomeres and printomeres, *Curr. Aging Sci.*, **8**, 76-88, doi: 10.2174/1874609808666150422114916.
27. Olovnikov, A. M. (2022) Planetary metronome as a regulator of lifespan and aging rate: the metronomic hypothesis, *Biochemistry (Moscow)*, **87**, 1640-1650, doi: 10.1134/S0006297922120197.
28. Horvath, S. (2013) DNA methylation age of human tissues and cell types, *Genome Biol.*, **14**, R115, doi: 10.1186/gb-2013-14-10-r115.
29. Petkovich, D. A., Podolskiy, D. I., Lobanov, A. V., Lee, S. G., Miller, R. A., and Gladyshev, V. N. (2017) Using DNA methylation profiling to evaluate biological age and longevity interventions, *Cell Metab.*, **25**, 954-960.e6, doi: 10.1016/j.cmet.2017.03.016.
30. López-Otín, C., Blasco, M. A., Partridge, L., Serrano, M., and Kroemer, G. (2013) The hallmarks of aging, *Cell*, **153**, 1194-1217, doi: 10.1016/j.cell.2013.05.039.
31. McHugh, D., and Gil, J. (2018) Senescence and aging: causes, consequences, and therapeutic avenues, *J. Cell Biol.*, **217**, 65-77, doi: 10.1083/jcb.201708092.
32. Campisi, J., and d'Adda di Fagagna, F. (2007) Cellular senescence: when bad things happen to good cells, *Nat. Rev. Mol. Cell Biol.*, **8**, 729-740, doi: 10.1038/nrm2233.
33. Ratushnyy, A. Y., Rudimova, Y. V., and Buravkova, L. B. (2020) Replicative senescence and expression of autophagy genes in mesenchymal stromal cells, *Biochemistry (Moscow)*, **85**, 1169-1177, doi: 10.1134/S0006297920100053.
34. Kuilman, T., Michaloglou, C., Mooi, W. J., and Peeper, D. S. (2010) The essence of senescence, *Genes Dev.*, **24**, 2463-2479, doi: 10.1101/gad.1971610.
35. Salama, R., Sadaie, M., Hoare, M., and Narita, M. (2014) Cellular senescence and its effector programs, *Genes Dev.*, **28**, 99-114, doi: 10.1101/gad.235184.113.
36. Ratushnyy, A. Y., and Buravkova, L. B. (2020) Cell senescence and mesenchymal stromal cells, *Hum. Physiol.*, **46**, 85-93, doi: 10.1134/S0362119720010132.
37. Imai, S., and Guarente, L. (2014) NAD^+ and sirtuins in aging and disease, *Trends Cell Biol.*, **24**, 464-471, doi: 10.1016/j.tcb.2014.04.002.
38. Dimri, G. P., Lee, X., Basile, G., Acosta, M., Scott, G., Roskelley, C., Medrano, E. E., Linskens, M., Rubelj, I., and Pereira-Smith, O. (1995) A biomarker that identifies senescent human cells in culture and in aging skin *in vivo*, *Proc. Natl. Acad. Sci. USA*, **92**, 9363-9367, doi: 10.1073/pnas.92.20.9363.
39. Narita, M., Nũnez, S., Heard, E., Narita, M., Lin, A. W., Hearn, S. A., Spector, D. L., Hannon, G. J., and Lowe, S. W. (2003) Rb-mediated heterochromatin formation and silencing of E2F target genes during cellular senescence, *Cell*, **113**, 703-716, doi: 10.1016/s0092-8674(03)00401-x.
40. Collado, M., Blasco, M. A., and Serrano, M. (2007) Cellular senescence in cancer and aging, *Cell*, **130**, 223-233, doi: 10.1016/j.cell.2007.07.003.
41. Hanahan, D., and Weinberg, R. A. (2011) Hallmarks of cancer: the next generation, *Cell*, **144**, 646-674, doi: 10.1016/j.cell.2011.02.013.
42. Jun, J. I., and Lau, L. F. (2010) The matricellular protein CCN1 induces fibroblast senescence and restricts fibrosis in cutaneous wound healing, *Nat. Cell Biol.*, **12**, 676-685, doi: 10.1038/ncb2070.
43. Noronha, N. C., Mizukami, A., Caliári-Oliveira, C., Cominal, J. G., Rocha, J. L. M., Covas, D. T., Swiech, K., and Malmegrim, K. C. R. (2019) Priming approaches to improve the efficacy of mesenchymal stromal cell-based therapies, *Stem Cell Res. Ther.*, **10**, 131, doi: 10.1186/s13287-019-1224-y.
44. Hayflick, L., and Moorhead, P. S. (1961) The serial cultivation of human diploid cell strains, *Exp. Cell Res.*, **25**, 585-621, doi: 10.1016/0014-4827(61)90192-6.
45. Olovnikov, A. M. (1971) Principle of marginotomy in template synthesis of polynucleotides, *Dokl. Akad. Nauk SSSR*, **201**, 1496-1499.
46. Victorelli, S., and Passos, J. F. (2017) Telomeres and cell senescence – size matters not, *EBioMedicine*, **21**, 14-20, doi: 10.1016/j.ebiom.2017.03.027.
47. De Magalhães, J. P., and Passos, J. F. (2018) Stress, cell senescence and organismal ageing, *Mech. Ageing Dev.*, **170**, 2-9, doi: 10.1016/j.mad.2017.07.001.
48. Gruber, J., Schaffer, S., and Halliwell, B. (2008) The mitochondrial free radical theory of ageing – where do we stand? *Front. Biosci.*, **13**, 6554-6579, doi: 10.2741/3174.
49. Vizioli, M. G., Liu, T., Miller, K. N., Robertson, N. A., Gilroy, K., Lagnado, A. B., Perez-Garcia, A., Kiourtis, C., Dasgupta, N., Lei, X., Kruger, P. J., Nixon, C., Clark, W., Jurk, D., Bird, T. G., Passos, J. F., Berger, S. L., Dou, Z., and Adams, P. D. (2020) Mitochondria-to-nucleus retrograde signaling drives formation of cytoplasmic chromatin and inflammation in senescence, *Genes Dev.*, **34**, 428-445, doi: 10.1101/gad.331272.119.
50. Von Zglinicki, T. (2002) Oxidative stress shortens telomeres, *Trends Biochem. Sci.*, **27**, 339-344, doi: 10.1016/s0968-0004(02)02110-2.
51. Campisi, J. (2013) Aging, cellular senescence, and cancer, *Annu. Rev. Physiol.*, **75**, 685-705, doi: 10.1146/annurev-physiol-030212-183653.
52. Sherr, C. J., and Roberts, J. M. (1999) CDK inhibitors: positive and negative regulators of G1-phase progression, *Genes Dev.*, **13**, 1501-1512, doi: 10.1101/gad.13.12.1501.
53. Sherr, C. J., and McCormick, F. (2002) The RB and p53 pathways in cancer, *Cancer Cell*, **2**, 103-112, doi: 10.1016/s1535-6108(02)00102-2.
54. Tharp, K. M., Higuchi-Sanabria, R., Timblin, G. A., Ford, B., Garzon-Coral, C., Schneider, C., Muncie, J. M., Stashko, C., Daniele, J. R., Moore, A. S., Frankino, P. A., Homentcovschi, S., Manoli, S. S., Shao, H., Richards, A. L., Chen, K. H., Hoeve, J. T., Ku, G. M., Hellerstein, M., Nomura, D. K., et al. (2021) Adhesion-mediated mechanosignaling forces mitohormesis, *Cell Metab.*, **33**, 1322-1341.e13, doi: 10.1016/j.cmet.2021.04.017.

55. Duan, J. L., Ruan, B., Song, P., Fang, Z. Q., Yue, Z. S., Liu, J. J., Dou, G. R., Han, H., and Wang, L. (2022) Shear stress-induced cellular senescence blunts liver regeneration through Notch-sirtuin 1-P21/P16 axis, *Hepatology*, **75**, 584-599, doi: 10.1002/hep.32209.
56. Van Loon, J. W. A. (2007) Some history and use of the random positioning machine, RPM, in gravity related research, *Adv. Space Res.*, **39**, 1161-1165, doi: 10.1016/j.asr.2007.02.016.
57. Kopp, S., Warnke, E., Wehland, M., Aleshcheva, G., Magnusson, N. E., Hemmersbach, R., Corydon, T. J., Bauer, J., Infanger, M., and Grimm, D. (2015) Mechanisms of three-dimensional growth of thyroid cells during long-term simulated microgravity, *Sci. Rep.*, **5**, 16691, doi: 10.1038/srep16691.
58. Wuest, S. L., Richard, S., Kopp, S., Grimm, D., and Egli, M. (2015) Simulated microgravity: critical review on the use of random positioning machines for mammalian cell culture, *BioMed Res. Int.*, **2015**, 971474, doi: 10.1155/2015/971474.
59. Wang, J., Zhang, J., Bai, S., Wang, G., Mu, L., Sun, B., Wang, D., Kong, Q., Liu, Y., Yao, X., Xu, Y., and Li, H. (2009) Simulated microgravity promotes cellular senescence via oxidant stress in rat PC12 cells, *Neurochem. Int.*, **55**, 710-716, doi: 10.1016/j.neuint.2009.07.002.
60. Kapitonova, M. Y., Muid, S., Froemming, G. R., Yusoff, W. N., Othman, S., Ali, A. M., and Nawawi, H. M. (2012) Real space flight travel is associated with ultrastructural changes, cytoskeletal disruption and premature senescence of HUVEC, *Malays. J. Pathol.*, **34**, 103-113.
61. Ulbrich, C., Wehland, M., Pietsch, J., Aleshcheva, G., Wise, P., van Loon, J., Magnusson, N., Infanger, M., Grosse, J., Eilles, C., Sundaresan, A., and Grimm, D. (2014) The impact of simulated and real microgravity on bone cells and mesenchymal stem cells, *BioMed Res. Int.*, **2014**, 928507, doi: 10.1155/2014/928507.
62. Winkelmaier, G., Jabbari, K., Chien, L. C., Grabham, P., Parvin, B., and Pluth, J. (2023) Influence of simulated microgravity on mammary epithelial cells grown as 2D and 3D cultures, *Int. J. Mol. Sci.*, **24**, 7615, doi: 10.3390/ijms24087615.
63. Ratushnyy, A., Ezdakova, M., Yakubets, D., and Buravkova, L. (2018) Angiogenic activity of human adipose-derived mesenchymal stem cells under simulated microgravity, *Stem Cells Dev.*, **27**, 831-837, doi: 10.1089/scd.2017.0262.
64. Ingber, D. E. (2003) Mechanobiology and diseases of mechanotransduction, *Ann. Med.*, **35**, 564-577, doi: 10.1080/07853890310016333.
65. Louis, F., Deroanne, C., Nusgens, B., Vico, L., and Guignandon, A. (2015) RhoGTPases as key players in mammalian cell adaptation to microgravity, *BioMed Res. Int.*, **2015**, 747693, doi: 10.1155/2015/747693.
66. Mao, X., Chen, Z., Luo, Q., Zhang, B., and Song, G. (2016) Simulated microgravity inhibits the migration of mesenchymal stem cells by remodeling actin cytoskeleton and increasing cell stiffness, *Cytotechnology*, **68**, 2235-2243, doi: 10.1007/s10616-016-0007-x.
67. Ratushnyy, A. Y., and Buravkova, L. B. (2017) Expression of focal adhesion genes in mesenchymal stem cells under simulated microgravity, *Dokl. Biochem. Biophys.*, **477**, 354-356, doi: 10.1134/S1607672917060035.
68. Buravkova, L., Larina, I., Andreeva, E., and Grigoriev, A. (2021) Microgravity effects on the matrisome, *Cells*, **10**, 2226, doi: 10.3390/cells10092226.
69. Dinarelli, S., Longo, G., Dietler, G., Francioso, A., Mosca, L., Pannitteri, G., Boumis, G., Bellelli, A., and Girasole, M. (2018) Erythrocyte's aging in microgravity highlights how environmental stimuli shape metabolism and morphology, *Sci. Rep.*, **8**, 5277, doi: 10.1038/s41598-018-22870-0.
70. Takahashi, H., Nakamura, A., and Shimizu, T. (2021) Simulated microgravity accelerates aging of human skeletal muscle myoblasts at the single cell level, *Biochem. Biophys. Res. Commun.*, **578**, 115-121, doi: 10.1016/j.bbrc.2021.09.037.
71. Acharya, A., Nemade, H., Papadopoulos, S., Hescheler, J., Neumaier, F., Schneider, T., Rajendra Prasad, K., Khan, K., Hemmersbach, R., Gusmao, E. G., Mizi, A., Papantonis, A., and Sachinidis, A. (2022) Microgravity-induced stress mechanisms in human stem cell-derived cardiomyocytes, *iScience*, **25**, 104577, doi: 10.1016/j.isci.2022.104577.
72. Singh, R., Rajput, M., and Singh, R. P. (2021) Simulated microgravity triggers DNA damage and mitochondria-mediated apoptosis through ROS generation in human promyelocytic leukemic cells, *Mitochondrion*, **61**, 114-124, doi: 10.1016/j.mito.2021.09.006.
73. Kossmehl, P., Shakibaei, M., Cogoli, A., Infanger, M., Curcio, F., Schönberger, J., Eilles, C., Bauer, J., Pickenhahn, H., Schulze-Tanzil, G., Paul, M., and Grimm, D. (2003) Weightlessness induced apoptosis in normal thyroid cells and papillary thyroid carcinoma cells via extrinsic and intrinsic pathways, *Endocrinology*, **144**, 4172-4179, doi: 10.1210/en.2002-0171.
74. Ran, F., An, L., Fan, Y., Hang, H., and Wang, S. (2016) Simulated microgravity potentiates generation of reactive oxygen species in cells, *Biophys. Rep.*, **2**, 100-105, doi: 10.1007/s41048-016-0029-0.
75. Greco, O., Durante, M., Gialanella, G., Grossi, G., Pugliese, M., Scampoli, P., Snigiryova, G., and Obe, G. (2003) Biological dosimetry in Russian and Italian astronauts, *Adv. Space Res.*, **31**, 1495-1503, doi: 10.1016/s0273-1177(03)00087-5.
76. Lu, T., Zhang, Y., Kidane, Y., Feiveson, A., Stodieck, L., Karouia, F., Ramesh, G., Rohde, L., and Wu, H. (2017) Cellular responses and gene expression profile changes due to bleomycin-induced DNA damage in human fibroblasts in space, *PLoS One*, **12**, e0170358, doi: 10.1371/journal.pone.0170358.
77. Murphy, M. B., Moncivais, K., and Caplan, A. I. (2013) Mesenchymal stem cells: environmentally responsive

therapeutics for regenerative medicine, *Exp. Mol. Med.*, **45**, e54, doi: 10.1038/emm.2013.94.

78. Spees, J. L., Lee, R. H., and Gregory, C. A. (2016) Mechanisms of mesenchymal stem/stromal cell function, *Stem Cell Res. Ther.*, **7**, 125, doi: 10.1186/s13287-016-0363-7.
79. Andreeva, E., Bobyleva, P., Gornostaeva, A., and Buravkova, L. (2017) Interaction of multipotent mesenchymal stromal and immune cells: bidirectional effects, *Cytotherapy*, **19**, 1152-1166, doi: 10.1016/j.jcyt.2017.07.001.
80. Fujii, S., and Miura, Y. (2022) Immunomodulatory and regenerative effects of MSC-derived extracellular vesicles to treat acute GVHD, *Stem Cells*, **40**, 977-990, doi: 10.1093/stmcls/sxac057.
81. Lv, W., Peng, X., Tu, Y., Shi, Y., Song, G., and Luo, Q. (2023) YAP inhibition alleviates simulated microgravity-induced mesenchymal stem cell senescence via targeting mitochondrial dysfunction, *Antioxidants*, **12**, 990, doi: 10.3390/antiox12050990.
82. Moretta, L., Uccelli, A., and Pistoia, V. (2015) Mesenchymal stromal cells and immunity: Introductory overview, *Immunol. Lett.*, **168**, 127-128, doi: 10.1016/j.imlet.2015.08.010.
83. Yatagai, F., Honma, M., Dohmae, N., and Ishioka, N. (2019) Biological effects of space environmental factors: A possible interaction between space radiation and microgravity, *Life Sci. Space Res.*, **20**, 113-123, doi: 10.1016/j.lssr.2018.10.004.
84. Moya, I. M., and Halder, G. (2019) Hippo-YAP/TAZ signalling in organ regeneration and regenerative medicine, *Nat. Rev. Mol. Cell Biol.*, **20**, 211-226, doi: 10.1038/s41580-018-0086-y.
85. Pobbati, A. V., and Hong, W. (2020) A combat with the YAP/TAZ-TEAD oncoproteins for cancer therapy, *Theranostics*, **10**, 3622-3635, doi: 10.7150/thno.40889.
86. Childs, B. G., Gluscevic, M., Baker, D. J., Laberge, R. M., Marquess, D., Dananberg, J., and van Deursen, J. M. (2017) Senescent cells: an emerging target for diseases of ageing, *Nat. Rev. Drug Discov.*, **16**, 718-735, doi: 10.1038/nrd.2017.116.
87. Xie, Q., Chen, J., Feng, H., Peng, S., Adams, U., Bai, Y., Huang, L., Li, J., Huang, J., Meng, S., and Yuan, Z. (2013) YAP/TEAD-mediated transcription controls cellular senescence, *Cancer Res.*, **73**, 3615-3624, doi: 10.1158/0008-5472.CAN-12-3793.
88. Yeung, Y. T., Guerrero-Castilla, A., Cano, M., Muñoz, M. F., Ayala, A., and Argüelles, S. (2019) Dysregulation of the hippo pathway signaling in aging and cancer, *Pharmacol. Res.*, **143**, 151-165, doi: 10.1016/j.phrs.2019.03.018.
89. Pala, R., Cruciani, S., Manca, A., Garroni, G., El Faqir, M. A., Lentini, V., Capobianco, G., Pantaleo, A., and Maioli, M. (2023) Mesenchymal stem cell behavior under microgravity: from stress response to a premature senescence, *Int. J. Mol. Sci.*, **24**, 7753, doi: 10.3390/ijms24097753.
90. Turinetto, V., Vitale, E., and Giachino, C. (2016) Senescence in human mesenchymal stem cells: functional changes and implications in stem cell-based therapy, *Int. J. Mol. Sci.*, **17**, 1164, doi: 10.3390/ijms17071164.
91. Li, Y., Wu, Q., Wang, Y., Li, L., Bu, H., and Bao, J. (2017) Senescence of mesenchymal stem cells (Review), *Int. J. Mol. Sci.*, **39**, 775-782, doi: 10.3892/ijmm.2017.2912.
92. Merzlikina, N. V., Buravkova, L. B., and Romanov, Y. A. (2004) The primary effects of clinorotation on cultured human mesenchymal stem cells, *J. Gravitat. Physiol.*, **11**, P193-P194.
93. Gershovich, J. G., and Buravkova, L. B. (2007) Morphofunctional status and osteogenic differentiation potential of human mesenchymal stromal precursor cells during in vitro modeling of microgravity effects, *Bull. Exp. Biol. Med.*, **144**, 608-613, doi: 10.1007/s10517-007-0387-1.
94. Dai, Z. Q., Wang, R., Ling, S. K., Wan, Y. M., and Li, Y. H. (2007) Simulated microgravity inhibits the proliferation and osteogenesis of rat bone marrow mesenchymal stem cells, *Cell Prolif.*, **40**, 671-684, doi: 10.1111/j.1365-2184.2007.00461.x.
95. Yuge, L., Kajiume, T., Tahara, H., Kawahara, Y., Umeda, C., Yoshimoto, R., Wu, S. L., Yamaoka, K., Asashima, M., Kataoka, K., and Ide, T. (2006) Microgravity potentiates stem cell proliferation while sustaining the capability of differentiation, *Stem Cells Dev.*, **15**, 921-929, doi: 10.1089/scd.2006.15.921.
96. Ratushnyy, A. Yu., and Buravkova, L. B. (2016) Functional state of multipotent mesenchymal stromal cells during modeling the effects of microgravity [in Russian], *Aviakosm. Ekol. Med.*, **50**, 24-29.
97. Nakamura, H., Kumei, Y., Morita, S., Shimokawa, H., Ohya, K., and Shinomiya, K. (2003) Antagonism between apoptotic (Bax/Bcl-2) and anti-apoptotic (IAP) signals in human osteoblastic cells under vector-averaged gravity condition, *Ann. NY Acad. Sci.*, **1010**, 143-147, doi: 10.1196/annals.1299.023.
98. Kim, M., Kim, C., Choi, Y. S., Kim, M., Park, C., and Suh, Y. (2012) Age-related alterations in mesenchymal stem cells related to shift in differentiation from osteogenic to adipogenic potential: implication to age-associated bone diseases and defects, *Mech. Ageing Dev.*, **133**, 215-225, doi: 10.1016/j.mad.2012.03.014.
99. Despars, G., Carbonneau, C. L., Bardeau, P., Coutu, D. L., and Beauséjour, C. M. (2013) Loss of the osteogenic differentiation potential during senescence is limited to bone progenitor cells and is dependent on p53, *PLoS One*, **8**, e73206, doi: 10.1371/journal.pone.0073206.
100. Yang, Y. K., Ogando, C. R., Wang See, C., Chang, T. Y., and Barabino, G. A. (2018) Changes in phenotype and differentiation potential of human mesenchymal stem cells aging *in vitro*, *Stem Cell Res. Ther.*, **9**, 131, doi: 10.1186/s13287-018-0876-3.
101. Wagner, W., Horn, P., Castoldi, M., Diehlmann, A., Bork, S., Saffrich, R., Benes, V., Blake, J., Pfister, S., Eckstein, V., and Ho, A. D. (2008) Replicative senescence of mesenchymal stem cells: a continuous and organized process, *PLoS One*, **3**, e2213, doi: 10.1371/journal.pone.0002213.

102. Digirolamo, C. M., Stokes, D., Colter, D., Phinney, D. G., Class, R., and Prockop, D. J. (1999) Propagation and senescence of human marrow stromal cells in culture: a simple colony-forming assay identifies samples with the greatest potential to propagate and differentiate, *Br. J. Haematol.*, **107**, 275-281, doi: 10.1046/j.1365-2141.1999.01715.x.
103. Cheng, H., Qiu, L., Ma, J., Zhang, H., Cheng, M., Li, W., Zhao, X., and Liu, K. (2011) Replicative senescence of human bone marrow and umbilical cord derived mesenchymal stem cells and their differentiation to adipocytes and osteoblasts, *Mol. Biol. Rep.*, **38**, 5161-5168, doi: 10.1007/s11033-010-0665-2.
104. Ratushnyy, A. Yu., and Buravkova, L. B. (2022) Differentiation potential of the mesenchymal stromal cells during replicative senescence [in Russian], *Aviakosm. Ekol. Med.*, **56**, 64-69.
105. Buravkova, L. B., Gershovich, P. M., Gershovich, J. G., and Grigor'ev, A. I. (2010) Mechanisms of gravitational sensitivity of osteogenic precursor cells, *Acta Naturae*, **2**, 28-36, doi: 10.32607/actanaturae.10734.
106. Nishikawa, M., Ohgushi, H., Tamai, N., Osuga, K., Uemura, M., Yoshikawa, H., and Myoui, A. (2005) The effect of simulated microgravity by three-dimensional clinostat on bone tissue engineering, *Cell Transplant.*, **14**, 829-835, doi: 10.3727/000000005783982477.
107. Zayzafoon, M., Gathings, W. E., and McDonald, J. M. (2004) Modeled microgravity inhibits osteogenic differentiation of human mesenchymal stem cells and increases adipogenesis, *Endocrinology*, **145**, 2421-2432, doi: 10.1210/en.2003-1156.
108. Buravkova, L. B., Gershovich, P. M., Gershovich, J. G., and Grigoriev, A. I. (2013) Microgravity and mesenchymal stem cell response, *Curr. Biotechnol.*, **2**, 217-225, doi: 10.2174/22115501113029990016.
109. Saxena, R., Pan, G., and McDonald, J. M. (2007) Osteoblast and osteoclast differentiation in modeled microgravity, *Ann. NY Acad. Sci.*, **1116**, 494-498, doi: 10.1196/annals.1402.033.
110. Meyers, V. E., Zayzafoon, M., Gonda, S. R., Gathings, W. E., and McDonald, J. M. (2004) Modeled microgravity disrupts collagen I/integrin signaling during osteoblastic differentiation of human mesenchymal stem cells, *J. Cell. Biochem.*, **93**, 697-707, doi: 10.1002/jcb.20229.
111. Sheyn, D., Pelled, G., Netanely, D., Domany, E., and Gazit, D. (2010) The effect of simulated microgravity on human mesenchymal stem cells cultured in an osteogenic differentiation system: a bioinformatics study, *Tissue Eng. Part A*, **16**, 3403-3412, doi: 10.1089/ten.tea.2009.0834.
112. Chen, J., Liu, R., Yang, Y., Li, J., Zhang, X., Li, J., Wang, Z., and Ma, J. (2011) The simulated microgravity enhances the differentiation of mesenchymal stem cells into neurons, *Neurosci. Lett.*, **505**, 171-175, doi: 10.1016/j.neulet.2011.10.014.
113. Xue, L., Li, Y., and Chen, J. (2017) Duration of simulated microgravity affects the differentiation of mesenchymal stem cells, *Mol. Med. Rep.*, **15**, 3011-3018, doi: 10.3892/mmr.2017.6357.
114. Coppé, J. P., Desprez, P. Y., Krtolica, A., and Campisi, J. (2010) The senescence-associated secretory phenotype: the dark side of tumor suppression, *Annu. Rev. Pathol.*, **5**, 99-118, doi: 10.1146/annurev-pathol-121808-102144.
115. Gershovich, Iu. G., and Buravkova, L. B. (2009) Interleukine production in culture of mesenchymal stromal cells of humans during simulation of the microgravity effects [in Russian], *Aviakosm. Ecolog. Med.*, **43**, 44-50.
116. Kurose, T., Takahashi, S., Otsuka, T., Nakagawa, K., Imura, T., Sueda, T., and Yuge, L. (2019) Simulated microgravity-cultured mesenchymal stem cells improve recovery following spinal cord ischemia in rats, *Stem Cell Res.*, **41**, 101601, doi: 10.1016/j.scr.2019.101601.
117. Ratushnyy, A., Yakubets, D., Andreeva, E., and Buravkova, L. (2019) Simulated microgravity modulates the mesenchymal stromal cell response to inflammatory stimulation, *Sci. Rep.*, **9**, 9279, doi: 10.1038/s41598-019-45741-8.
118. Zhivodernikov, I. V., Ratushnyy, A. Y., and Buravkova, L. B. (2021) Secretory activity of mesenchymal stromal cells with different degree of commitment under conditions of simulated microgravity, *Bull. Exp. Biol. Med.*, **170**, 560-564, doi: 10.1007/s10517-021-05106-6.
119. Aamodt, J. M., and Grainger, D. W. (2016) Extracellular matrix-based biomaterial scaffolds and the host response, *Biomaterials*, **86**, 68-82, doi: 10.1016/j.biomaterials.2016.02.003.
120. Choudhury, D., Tun, H. W., Wang, T., and Naing, M. W. (2018) Organ-derived decellularized extracellular matrix: a game changer for bioink manufacturing? *Trends Biotechnol.*, **36**, 787-805, doi: 10.1016/j.tibtech.2018.03.003.
121. Hospodiuk, M., Dey, M., Sosnoski, D., and Ozbolat, I. T. (2017) The bioink: A comprehensive review on bioprintable materials, *Biotechnol. Adv.*, **35**, 217-239, doi: 10.1016/j.biotechadv.2016.12.006.
122. Mavrogonatou, E., Pratsinis, H., Papadopoulou, A., Karamanos, N. K., and Kletsas, D. (2019) Extracellular matrix alterations in senescent cells and their significance in tissue homeostasis, *Matrix Biol.*, **75-76**, 27-42, doi: 10.1016/j.matbio.2017.10.004.
123. Choi, H. R., Cho, K. A., Kang, H. T., Lee, J. B., Kaeberlein, M., Suh, Y., Chung, I. K., and Park, S. C. (2011) Restoration of senescent human diploid fibroblasts by modulation of the extracellular matrix, *Aging Cell*, **10**, 148-157, doi: 10.1111/j.1474-9726.2010.00654.x.
124. Sun, Y., Li, W., Lu, Z., Chen, R., Ling, J., Ran, Q., Jilka, R. L., and Chen, X. D. (2011) Rescuing replication and osteogenesis of aged mesenchymal stem cells by exposure to a young extracellular matrix, *FASEB J.*, **25**, 1474-1485, doi: 10.1096/fj.10-161497.
125. Lynch, K., and Pei, M. (2014) Age associated communication between cells and matrix: a potential impact on stem cell-based tissue regeneration strategies, *Organogenesis*, **10**, 289-298, doi: 10.4161/15476278.2014.970089.

126. Ebnerasuly, F., Hajebrahimi, Z., Tabaie, S. M., and Darbouy, M. (2018) Simulated microgravity condition alters the gene expression of some ECM and adhesion molecules in adipose derived stem cells, *Int. J. Mol. Cell. Med.*, **7**, 146-157, doi: 10.22088/IJMCM.BUMS.7.3.146.
127. Zhivodernikov, I., Ratushnyy, A., and Buravkova, L. (2021) Simulated microgravity remodels extracellular matrix of osteocommitted mesenchymal stromal cells, *Int. J. Mol. Sci.*, **22**, 5428, doi: 10.3390/ijms22115428.
128. Carmeliet, G., Nys, G., and Bouillon, R. (1997) Microgravity reduces the differentiation of human osteoblastic MG-63 cells, *J. Bone Min. Res.*, **12**, 786-794, doi: 10.1359/jbmr.1997.12.5.786.
129. Zhang, C., Li, L., Jiang, Y., Wang, C., Geng, B., Wang, Y., Chen, J., Liu, F., Qiu, P., Zhai, G., Chen, P., Quan, R., and Wang, J. (2018) Space microgravity drives transdifferentiation of human bone marrow-derived mesenchymal stem cells from osteogenesis to adipogenesis, *FASEB J.*, **32**, 4444-4458, doi: 10.1096/fj.201700208RR.
130. Libertini, G. (2014) The programmed aging paradigm: how we get old, *Biochemistry (Moscow)*, **79**, 1004-1016, doi: 10.1134/S0006297914100034.