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Correlations between vessel stiffness and biomarkers of senescent cell in elderly patients

Aim	To study the association between vascular wall stiffness and known markers for accumulation of senescent cells in blood, cells, and tissues of old patients.
Material and methods	This study included male and female patients aged 65 years and older who were referred to an elective surgical intervention, that included a surgical incision in the area of the anterior abdominal wall or large joints and met the inclusion and exclusion criteria. For all patients, traditional cardiovascular (CV) risk factors and arterial wall stiffness (pulse wave velocity, PWV) were evaluated. Also, biomaterials (peripheral blood, skin, subcutaneous adipose tissue) were collected during the surgery and were used for isolation of several cell types and subsequent histological analysis to determine various markers of senescent cells.
Results	The study included 80 patients aged 65 to 90 years. The correlation analysis identified the most significant indexes that reflected the accumulation of senescent cells at the systemic, tissue, and cellular levels ($r>0.3$, $p<0.05$) and showed positive and negative correlations with PWV. The following blood plasma factors were selected as the markers of ageing: insulin-like growth factor 1 (IGF-1), fibroblast growth factor 21 (FGF-21), and vascular endothelium adhesion molecule 1 (VCAM-1). A significant negative correlation between PWV and IGF-1 concentration was found. Among the tissue markers, P16INK, the key marker for tissue accumulation of senescent cells, predictably showed a positive correlation ($r=0.394$, $p<0.05$). A medium-strength correlation with parameters of the 96-h increment of mesenchymal stromal cells and fibroblasts and a weak correlation with IL-6 as a SASP (specific senescent-associated secretory phenotype) were noted. Results of the multifactorial linear regression analysis showed that the blood plasma marker, VCAM- 1, and the cell marker, 96-h increment of fibroblasts, were associated with PWV regardless of the patient's age.
Conclusion	Stiffness of great arteries as measured by PWV significantly correlates with a number of plasma, tissue, and cellular markers for accumulation of senescent cells. This fact suggests PWV as a candidate for inclusion in the panel of parameters for evaluation and monitoring of the biological age during the senolytic therapy.

Keywords	Pulse wave velocity; senescent cells; ageing; biomarker p16INK4a; mesenchymal stromal cell; insulin-like growth factor 1; fibroblast growth factor 21; vascular endothelium adhesion molecule 1
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Introduction

Aging is a healthy integral process of functional decline that affects and modifies all tissues, organs, and systems of the body. The risks of three or more simultaneous chronic diseases exponentially increase, physical activity significantly decreases, and mortality increases with age [1]. The conditions, for which stated age is a major risk factor, include primarily cardiovascular diseases (CVDs), strokes, and peripheral artery occlusions, it

also include cancer, diabetes, renal dysfunction, osteoporosis, arthritis, and blindness [2, 3].

CVD prevention is commonly aimed at modifiable risk factors [4]. Age is an unmodifiable parameter and is never considered a center of effort for the CVD prevention. However, a leading modern theory of aging, which attributes age-associated changes in the body to the accumulation of so-called senescent cells (Lat. senex " old man") in tissues, suggests that this process can be slowed down and actively modified by influencing the number of such cells, i.e., it translates age into a category of partially modifiable factors.

Senescence implies temporary or permanent arrest of cell growth: senescent cells linger in the G1 or G2/M phase of the cell cycle, and mechanisms of apoptosis resistance are activated, which makes it difficult to eliminate these cells. They were shown to acquire senescence-associated secretory phenotype (SASP), which is characterized by increased production of pro-inflammatory cytokines, chemokines, proteins that degrade the extracellular matrix, and other prosenescent factors that disrupt the structure and function of surrounding cells and tissue [5].

The expression of cell cycle inhibitor proteins, primarily p16INK4a, is one of the most studied biomarkers of senescent cells [6, 7]. According to some researchers, this biomarker can be identified in the systemic circulation by assessing the level p16 (as the protein or mRNA) expression in the CD3+ fraction of peripheral blood T lymphocytes. This indicator correlates statistically significantly with a patient's age and with some common risk factors for age-associated diseases, such as smoking [8].

Biomarkers of cellular senescence also include beta-galactosidase activity in cells and blood plasma [9], telomere length and telomerase activity [10], growth factors [11, 12], and the levels of certain cell-secreted factors in the culture medium, including interleukin-6 (IL-6), type 1 plasminogen activator inhibitor (PAI-1), etc. [7, 13, 14].

Vascular stiffness is a manifestation of vascular aging. Traditionally, it is evaluated by measuring the carotid-femoral pulse wave velocity (PWV) [15, 16]. New data have recently emerged on the correlation of PWV and its changes during treatment with other than cardiovascular mortality [17], which allows considering the stiffness of large arteries a promising marker of vascular and systemic aging (Figure 1).

Studying markers of the accumulation of senescent cells in tissues that reflect the biological age of the patient is important to determine the role of aging processes in the development of macro- and microvascular complications and the progression of CVDs and assess the body's regenerative powers. The evidence of the correlation of PWV with the markers of senescent cell accumulation will also allow using this available non-invasive parameter together with laboratory indicators to monitor the efficacy of senolytic therapy.

Objective

Investigate the correlation between vascular stiffness and the most significant biomarkers reflecting senescent cell accumulation at the organismic, tissue and cellular levels in elderly patients.

Material and methods

The study included male and female sex patients of 65 years and older who were referred for elective surgical intervention with an incision in the anterior abdominal wall or large joints and met the inclusion and exclusion criteria. All patients were assessed for common cardiovascular risk factors, arterial wall stiffness (PWV was estimated using a BPLab device (OOO "Petr Telegin", Russia) and Vasotens Office version 06.04.03 following the previously described method [18]. During surgery, biomaterials (peripheral blood, skin, subcutaneous fatty tissue) were collected, from which various cell types were isolated and tissue samples were prepared for histological analysis in order to evaluate various markers of cell senescence [19]. The local ethics committee approved the study. Inclusion and exclusion criteria are presented in Table 1.

Collecting biological samples and analyzing biomarkers reflecting senescent cell accumulation were described in detail by Sorokina et al. (2021) [19].

Statistical processing

Statistical analysis of findings was performed using the IBM SPSS Statistics software suite. All continuous values are expressed as the means (M) and standard deviations (SD) or the medians (Me) and lower (LQ) and upper (UQ) quartiles depending on the type of distribution. The hypothesis of normal distribution of an indicator was verified using the Shapiro-Wilk test. The Pearson correlation coefficient was calculated to describe the correlation between the various factors (normally distributed data). The paired Student's T-test was used when comparing different groups to determine the significance of differences between normally distributed variables. For non-normally distributed variables, the Wilcoxon test was used to investigate differences between dependent samples and the Mann-Whitney test was utilized for independent samples. A factor (principal component) analysis with Kaiser-Varimax rotation was used to detect similar patterns in the correlating variables and reduce their number. The presence of a correlation between PWV and some biomarkers of senescent cell accumulation was assessed using a regression analysis. The level of statistical significance was $p<0.05$.

Results

From 2018 to 2020, 166 people were screened. The study included 80 patients (21 male and 59 female, 65 to

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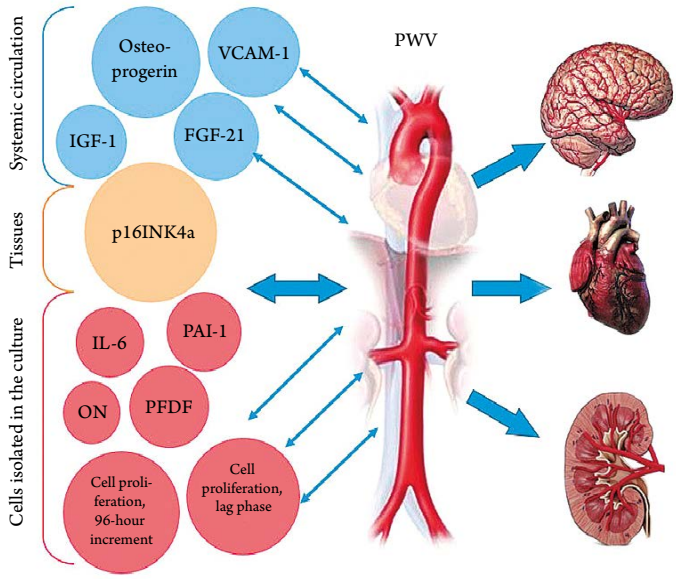
Figure 1. Investigated markers of senescent cell accumulation



Markers estimated in the systemic circulation: osteoprogerin, IGF-1 (insulin-like growth factor 1), FGF-21 (fibroblast growth factor 21), VCAM-1 (vascular cell adhesion molecule 1). Tissue markers: p16INK4a (cyclin-dependent kinase inhibitor). Markers estimated in cells: IL-6 (interleukin 6), MCP-1 (monocyte chemotactic protein 1), PAI-1 (plasminogen activator inhibitor type 1), ON (osteonectin), PEDF (pigment epithelium-derived factor).

Table 1. Inclusion and exclusion criteria

Inclusion criteria	Male and female patients of 65 years and older, who are able to understand the objectives of this study and follow the protocol
	Signed informed consent to participate in the study
	Indications for elective surgery involving surgical incision in the area of anterior abdominal wall or large joints
Exclusion criteria	History of myocardial infarction, intermittent claudication, or acute cerebrovascular accident
	CHF functional class III-IV
	History of cancer or systemic diseases
	Mental, physical, and other reasons that do not allow adequately assessing own behavior and correctly following the study protocol
	History of any significant condition/disease or circumstance, that, in the opinion of the researcher, prevents the inclusion in the study
	Inability/unwillingness to sign the informed consent to participate in the study
	Contraindications for surgical treatment at inclusion
	Acute and chronic infectious diseases

90 years old (median age 71 years) who met the inclusion/non-inclusion criteria and signed the informed consent to participate in the study. The main patient characteristics are provided in Table 2.

Medical history was collected from all patients. Physical examination was carried out. Before surgery, peripheral blood was tested, and arterial wall stiffness was assessed using a non-invasive method. Skin and subcutaneous fat were also sampled from 41 patients during the surgery (joint replacement, hernioplasty). We managed to isolate markers of aging in tissues and cells in 36 patients for technical reasons.

The most significant indicators of senescent cell accumulation at the systemic, tissue, and cellular levels were identified using the correlation analysis (normal distribution, Pearson correlation coefficient $r>0.3$, $p<0.05$), which showed positive and negative correlations with PWV (Table 3).

The markers are characterized provided in Table 4. The factor analysis allowed identifying the following factors that combine various indicators of senescent cell accumulation in patients of the study population (Kaiser-Meyer-Olkin (KMO) test 0.68, acceptable adequacy): IGF-1, telomeres length_PBMC, VCAM-1, cd34+, PWV, p16INK4a.

Statistically significant differences were identified when assessing a number of markers of senescent cell accumulation in the subgroups of PWV above and below the median. The stated age was similar in the subgroups (Table 5).

The results of the multivariate linear regression analysis showed that the blood plasma marker VCAM-1 and the cellular marker PB 96 h increment were correlated with PWV independently of age (Table 6).

Discussion

Our study included elderly and senile patients without decompensated diseases and confirmed that the stiffness of large arteries measured using PWV was significantly correlated with age (Pearson correlation coefficient $r=0.556$, $p<0.001$) and SBP (Pearson correlation coefficient $r=0.334$, $p<0.05$). Our findings are consistent with the literature data [15, 21].

The main hypothesis of our study was that vascular stiffness and known markers of senescent cell accumulation were correlated independently of stated age. The relevant evidence was first provided by the factor analysis. Age was not among the identified factors combining various indicators of senescent cell accumulation in the study population. The correlation was shown for IGF-1, telomere length_PBMC, VCAM-1, cd34+, PWV, p16INK4a (KMO=0.68, acceptable adequacy).

Plasma markers of senescent cell accumulation

During our study, we evaluated the levels of some factors secreted by senescent cells in the peripheral circulation. For

Table 2. Patient characteristics at inclusion (n = 80)

Parameter	Value
Age, years	71 ± 5.9
Male patients, %	26
Smoking, %	21.2
AH, %	82.5
BMI, kg/m^2	28.65 ± 5.2
IGT, %	7.5
DM type 2, %	11.2
SBP, mm Hg	136.5 ± 22.3
DBP, mm Hg	78.0 ± 9.25
TC, mmol/L	4.82 ± 1.45
LDL cholesterol, mmol/L	3.21 ± 1.03
HDL cholesterol, mmol/L	1.2 ± 0.29
TG, mmol/L	1.24 ± 0.7

Data are presented as the mean ± standard deviation (M±SD) or the percentage (%) of the total number of patients; AH, arterial hypertension, BMI, body mass index, IGT, impaired glucose tolerance; DM, diabetes mellitus; SBP, systolic blood pressure; DBP, diastolic blood pressure; TC, total cholesterol; LDL, low density lipoprotein; HDL, high density lipoprotein; TG, triglyceride.

Table 3. Correlation of PWV with age, SBP and indicators of senescent cell accumulation at the systemic, tissue and cellular levels

Parameter	Pearson's correlation coefficient (r)	p
Age	0.556	< 0.001
SBP	0.334	0.048
Blood plasma		
IGF-1	-0.318	0.005
FGF-21	0.326	0.004
VCAM-1	0.451	< 0.001
Osteoprogerin	0.14	0.231
Ferritin	0.15	0.932
Telomere length_PBMC, bp	-0.114	0.345
cd34+%	-0.126	-0.293
Tissue		
p16INK4a	0.394	0.042
Cells		
MSC_lag phase, h	0.320	0.057
MSC 96 h increment	-0.418	0.011
FB_lag phase, h	0.284	0.168
FB 96 h increment	-0.492	0.012
SASP		
IL-6, ng/mL_MSC	0.364	0.032
MCP-1, ng/mL_MSC	0.280	0.103
PAI-1, ng/mL_MSC	0.186	0.285
ON, ng/mL_MSC	-0.185	0.287
PEDF, ng/mL_MSC	-0.045	0.799

PWS, pulse wave velocity, SBP, systolic blood pressure; IGF-1, insulin-like growth factor 1; FGF-21, fibroblast growth factor 21; VCAM-1, vascular cell adhesion molecule 1; p16INK4a, cyclin-dependent kinase inhibitor; MSC, mesenchymal stem cells; FB, fibroblasts; IL-6, interleukin; MCP-1, monocyte chemotactic protein 1; PAI-1, plasminogen activator inhibitor 1; ON, osteonectin; PEDF, pigment epithelium-derived factor.

example, IGF-1 (insulin-like growth factor 1) that is involved in the endocrine, autocrine, and paracrine regulation of the processes of growth, development, and differentiation of cells and tissues in the body. The exact role of IGF-I in growing old and age-associated diseases has not been clearly defined. Some studies provide certain understanding of the possible contribution of this factor to growing old, but the available data are contradictory [11, 12]. Low serum IGF-I is a predictor of longevity, and it decreases with age, which is also demonstrated in our study. At the same time, treatment with IGF-I is known to improve some age-associated diseases [22].

The mean plasma level of IGF-I in our sample corresponded to the age reference. Important result of our study can be considered the identification of a significant correlation between IGF-1 and the main indicator of arterial wall stiffness in elderly patients. These results are understandable given IGF-I having a significant effect on the cardiovascular system by stimulating the growth of cardiomyocytes and smooth muscle cells in the vascular wall [23]. Higher levels of IGF-I were detected in patients with PWV<14 m/s (i.e., with less rigid vessels). If IGF-I is typical of a body of lower biological age, then elevating IGF-I reflects the natural process of growing old, i.e., increased stiffness of the vascular wall, as IGF-I decreases with age, and allows considering IGF-I as a characteristic of biological age.

It has been shown recently that FGF21 is a key element in cardiac and vascular remodeling: FGF21 expression protects from pathological cardiac hypertrophy, oxidative stress, and

Table 4. Investigated markers of systemic senescent cell accumulation

Aging marker	Value
PWV, m/s	14.15 ± 2.52
Blood plasma (n = 80)	
IGF-1	152.80 ± 46.4
FGF-21	150.80 ± 17.10
VCAM-1	32.56 [29.32; 35.8]
Tissues (n = 41)	
p16INK4a	4.61 ± 1.96
Cells (n = 41)	
MSC-lag phase, h	27.50 ± 5.39
MSC 96 h increment	63.00 ± 13.22
FB_lag phase, h	24.00 ± 5.83
FB 96h increment	78.00 ± 13.00
SASP (n = 41)	
IL-6, ng/mL_MSC	29.40 [22.64; 36.71]

The data are expressed as the mean ± standard deviation (M ± SD), the median and interquartile range (Me [Q1; Q3]); PWV, pulse wave velocity; IGF-1, insulin-like growth factor 1; FGF-21, fibroblast growth factor 21; VCAM-1, vascular cell adhesion molecule 1; p16INK4a, cyclin-dependent kinase inhibitor; MSC, mesenchymal stem cells; FB, fibroblasts; SASP, senescence-associated secretory phenotype; IL-6, interleukin-6.

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Table 5. Investigated parameters in the groups with PWV above and below the median

Parameters	PWV ≥ 14 m/s (n = 42)	PWV < 14 m/s (n = 39)	Statistical criterion	p
Age, years	70.17 ± 6.34	73.42 ± 5.25	Student's t-test	0.154
Blood plasma (n = 80)				
IGF-1, ng/mL	136.44 ± 38.61	161.95 ± 51.48	Student's t-test	0.015
FGF-21, ng/mL	232.23 ± 48.81	134.45 ± 50.11	Student's t-test	0.027
VCAM-1, ng/mL	50.47 ± 7.62	31.78 ± 11.24	Student's t-test	0.006
Telomere length_PBMC, bp	546.27 [177.65; 723.92]	812.18 [117.33; 1403.13]	Mann–Whitney U-test	0.077
cd34+%	0.05 [0.048; 0.066]	0.063 [0.048; 0.077]	Mann–Whitney U-test	0.217
Tissues (n = 41)				
p16INK4a, cells/mm2	5.05 ± 1.91	3.82 ± 1.87	Student's t-test	0.049
Cells (n = 41)				
MSC-lag phase, h	30.36 ± 5.75	26.14 ± 3.61	Student's t-test	0.020
MSC 96 h increment	54.33 ± 15.62	61.67 ± 6.22	Student's t-test	0.096
FB_lag phase, h	27.71 ± 6.13	21.53 ± 4.13	Student's t-test	0.045
FB 96 h increment	67.21 ± 12.53	82.64 ± 7.41	Student's t-test	0.001
SASP (n=41)				
IL-6, ng/mL_MSC	45.69 [16.99; 74.39]	28.29 [13.75; 42.82]	Mann–Whitney U-test	0.082

The data are expressed as the means ± standard deviations (M ± SD), the medians and interquartile ranges (Me [Q1; Q3]); IGF-1, insulin-like growth factor 1; FGF-21 PWV, pulse wave velocity; FGF-21, fibroblast growth factor 21; VCAM-1, vascular cell adhesion molecule 1; cd34+, positive cells with membrane protein 34; p16INK4a, cyclin-dependent kinase inhibitor; MSC, mesenchymal stem cells; FB, fibroblasts; SASP, senescence-associated secretory phenotype; IL-6, interleukin-6.

Table 6. Correlation of PWV with markers of senescent cell accumulation. Data of multivariate regression analysis

Parameter	Beta coefficient	Standard error of beta	t	p
Age	0.144	0.142	0.651	0.498
VCAM-1	0.671	0.017	3.843	0.001
FB 96 h increment	-0.411	0.047	-1.853	0.048

VCAM-1, vascular cell adhesion molecule 1; FB, fibroblasts.

myocardial infarction [24–26]. FGF21 acts as an autocrine hormone in the myocardium and controls autophagy in obesity-induced cardiomyopathy [27–29].

Attraction and adhesion of monocytes on the endothelium in crucial for the triggering of atherosclerosis. VCAM-1 indirectly reflects the extent of vascular wall damage [30], and the levels of VCAM and ICAM were shown to be correlated with PWV [31].

FGF21 as well as IGF-1 and VCAM are regarded as biomarkers of senile fragility of the body and are studied within the framework of the senescence theory [28, 32]. FGF21 is considered to be a biomarker of healthy aging, since elevated levels were observed in elderly patients without obesity and diabetes, and it was demonstrated in animal models that the overproduction of FGF21 in that case was not associated with tissue insensitivity to growth factor [33]. VCAM and ICAM are elevated in elderly patients regardless of the presence or absence of cardiovascular risk factors [34].

We also demonstrated correlations between PWV and FGF21 (r=0.326, p=0.004) and PWV and VCAM-1 (r=0.451, p<0.001), which reflects the correlation between the blood levels of these molecules and stiffness of large vessels [35]. In the PWS>14 m/s group, the levels of FGF2 and VCAM-1 were higher than in PWS<14 m/s group in the absence of age differences between the groups. These data are mainly consistent with the suggestion that stiffness of larger arteries can be considered an indicator of the systemic aging of the body.

Markers of senescent cell accumulation in tissues

Protein p16INK4a inhibits cell division and thus is involved in cell translation to senescent and is one of the most reliable markers of senescent cells. The level of p16INK4a expression is significantly correlated with age and some cardiovascular risk factors [36]. We demonstrated an independent positive correlation between the tissue levels of p16INK4a in elderly and senile patients without cancer and PWV, with levels of p16INK4a being statistically significantly different in the groups with low and high PWV. Factor analysis showed that plasma markers of growing old, p16INK4a and PWV, can be considered as a single parameter reflecting aging of the body. Thus, vascular wall stiffness and the expression of senescent cell marker p16INK4a increase as the body ages. Both indicators are interrelated, as they reflect different aspects of the same process of aging.

Markers of senescent cell accumulation in individual cell populations

During the study, we isolated FB MSC culture from the tissues and investigated the expression of senescent

cell markers in these populations. The duration of lag phase was used as a marker, which is the time to the beginning of cell division, when they are fixed on the substrate and get prepared for the division, and the number of cells were also estimated at regular intervals of 48, 72, and 96 hours.

According to the literature, senescent cells are characterized by longer lag phase due to decreased replicative and adaptive properties, and reduced number of divisions, which is fully consistent with our findings [37, 38]. Both MSC and FB showed a significant difference in these parameters in patients with PWV>14 m/s and PWV<14 m/s. The elongation of lag phase and a smaller number of cells at 96 h were characteristic of patients with PWV>14 m/s (higher stiffness of arteries), i.e., the age of cells in patients with PWV >14 m/s is higher the age of cells in patients with lower stiffness of arteries. Multiple regression analysis demonstrated an age-independent correlation between PB 96 h increment and PWV.

We also evaluated the levels of SASP components in the secretion of isolated cells. Level of IL-6 is statistically significantly correlated with PWV (r=0.364, p=0.032). There are no trends in the nature of changes in the levels of SASP components depending on high or low PWV, which may be due to insufficient sampling and difficulties in determining SASP poorly secreted by cells [7].

The study was limited by small sample size and the lack of analysis of the parameters depending on the drug therapy administered.

Conclusion

Stiffness of large arteries, as measured by the PWV, is significantly correlated with numbers of plasma, tissue, and cell markers (accumulation of senescent cells). Factor and multiple regression analysis data suggest that this correlation is not attributable solely to stated age. The technical difficulties in collecting and processing biomaterial samples for this study limited the sample size and did not allow us to assess the independent contribution of each indicator of interest. However, the data obtained make it possible to consider the inclusion of pulse wave velocity in the panel of parameters for assessing and monitoring the biological age of the body during senolytic therapy.

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REFERENCES

1. St Sauver JL, Boyd CM, Grossardt BR, Bobo WV, Finney Rutten LJ, Roger VL et al. Risk of developing multimorbidity across all ages in an historical cohort study: differences by sex and ethnicity. BMJ Open. 2015;5(2):e006413. DOI: 10.1136/bmjopen-2014-006413
2. Kirkland JL. Translating the Science of Aging into Therapeutic Interventions. Cold Spring Harbor Perspectives in Medicine. 2016;6(3):a025908. DOI: 10.1101/cshperspect.a025908
3. Kirkland JL, Tchkonia T. Senolytic drugs: from discovery to translation. Journal of Internal Medicine. 2020;288(5):518–36. DOI: 10.1111/joim.13141
4. World Health Organization. A global brief of hypertension. Silent killer, global public health crisis: World Health Day 2013. 2013. [Av. at: https://www.who.int/publications/i/item/a-global-brief-on-hypertension-silent-killer-global-public-health-crisis-world-health-day-2013]
5. Özcan S, Alessio N, Acar MB, Mert E, Omerli F, Peluso G et al. Unbiased analysis of senescence associated secretory phenotype (SASP) to identify common components following different genotoxic stresses. Aging. 2016;8(7):1316–29. DOI: 10.18632/aging.100971
6. Baker DJ, Childs BG, Durik M, Wijers ME, Sieben CJ, Zhong J et al. Naturally occurring p16Ink4a-positive cells shorten healthy lifespan. Nature. 2016;530(7589):184–9. DOI: 10.1038/nature16932
7. Kirkland JL, Tchkonia T. Cellular Senescence: A Translational Perspective. EBioMedicine. 2017;21:21–8. DOI: 10.1016/j.ebiom.2017.04.013
8. Liu Y, Sanoff HK, Cho H, Burd CE, Torrice C, Ibrahim JG et al. Expression of p16INK4a in peripheral blood T-cells is a biomarker of human aging. Aging Cell. 2009;8(4):439–48. DOI: 10.1111/j.1474-9726.2009.00489.x
9. Spazzafumo L, Mensà E, Matacchione G, Galeazzi T, Zampini L, Recchioni R et al. Age-related modulation of plasmatic beta-Galactosidase activity in healthy subjects and in patients affected by T2DM. Oncotarget. 2017;8(55):93338–48. DOI: 10.18632/oncotarget.21848
10. Moslehi J, DePinho RA, Sahin E. Telomeres and Mitochondria in the Aging Heart. Circulation Research. 2012;110(9):1226–37. DOI: 10.1161/CIRCRESAHA.111.246868
11. Salminen A, Kaarniranta K, Kauppinen A. Insulin/IGF-1 signaling promotes immunosuppression via the STAT3 pathway: impact on the aging process and age-related diseases. Inflammation Research. 2021;70(10–12):1043–61. DOI: 10.1007/s00011-021-01498-3
12. Vitale G, Pellegrino G, Vollery M, Hofland LJ. ROLE of IGF-1 System in the Modulation of Longevity: Controversies and New Insights from a Centenarians' Perspective. Frontiers in Endocrinology. 2019; 10:27. DOI: 10.3389/fendo.2019.00027
13. Matjusaitis M, Chin G, Sarnoski EA, Stolzing A. Biomarkers to identify and isolate senescent cells. Ageing Research Reviews. 2016;29:1–12. DOI: 10.1016/j.arr.2016.05.003
14. Birch J, Gil J. Senescence and the SASP: many therapeutic avenues. Genes & Development. 2020;34(23–24):1565–76. DOI: 10.1101/gad.343129.120
15. Mikael L de R, Paiva AMG de, Gomes MM, Sousa ALL, Jardim PCBV, Vitorino PV de O et al. Vascular Aging and Arterial Stiffness. Arquivos Brasileiros de Cardiologia. 2017;109(3):253–8. DOI: 10.5935/abc.20170091
16. Kobalava Zh.D., Konradi A.O., Nedogoda S.V., Shlyakhto E.V., Arutyunov G.P., Baranova E.I. et al. Arterial hypertension in adults. Clinical guidelines 2020. Russian Journal of Cardiology. 2020;25(3):149–218. [Russian: Кобалава Ж.Д., Конради А.О., Недогода С.В., Шляхто Е.В., Арутюнов Г.П., Баранова Е.И. и др. Артериальная гипертензия у взрослых. Клинические рекомендации 2020. Рос-

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сийский кардиологический журнал. 2020;25(3):149-218]. DOI: 10.15829/1560-4071-2020-3-3786
17. Vlachopoulos C, Terentes-Printzios D, Laurent S, Nilsson PM, Protogerou AD, Aznaouridis K et al. Association of Estimated Pulse Wave Velocity with Survival: A Secondary Analysis of SPRINT. JAMA Network Open. 2019;2(10):e1912831. DOI: 10.1001/jamanetworkopen.2019.12831
18. Tkachenko Yu.V., Strazhesko I.D., Borisov E.N., Plisiuk A.G., Orlova Ya.A. Adaptation of the method of pulse wave velocity measurement for screening examinations in outpatient practice. Journal of Clinical Practice. 2019;10(1):48–56. [Russian: Ткаченко Ю.В., Стражеско И.Д., Борисов Е.Н., Плисюк А.Г., Орлова Я.А. Адаптация методики измерения скорости пульсовой волны для скрининговых обследований в амбулаторной практике. Клиническая практика. 2019;10(1):48–56]. DOI: 10.17816/clinpract10148-56
19. Sorokina A.G., Orlova Ya.A., Grigorieva O.A., Novoseletskaya E.S., Basalova N.A., Alexandrushkina N.A. et al. Creation of a collection of different biological sample types from elderly patients to study the relationship of clinical, systemic, tissue and cellular biomarkers of accumulation of senescent cells during aging. Cardiovascular Therapy and Prevention. 2021;20(8):164–75. [Russian: Сорокина А.Г., Орлова Я.А., Григорьева О.А., Новоселецкая Е.С., Басалова Н.А., Александрушкина Н.А. и др. Создание коллекции биологических образцов разного типа, полученных от пожилых пациентов, для изучения взаимосвязей клинических, системных, тканевых и клеточных биомаркеров накопления сенесцентных клеток при старении. Кардиоваскулярная терапия и профилактика. 2021;20(8):164-75]. DOI: 10.15829/1728-8800-2021-3051
20. Avolio AP, Kuznetsova T, Heyndrickx GR, Kerkhof PLM, Li JK-J. Arterial Flow, Pulse Pressure and Pulse Wave Velocity in Men and Women at Various Ages. Advances in Experimental Medicine and Biology. 2018;1065:153–68. DOI: 10.1007/978-3-319-77932-4_10
21. Battistoni A, Michielon A, Marino G, Savoia C. Vascular Aging and Central Aortic Blood Pressure: From Pathophysiology to Treatment. High Blood Pressure & Cardiovascular Prevention. 2020;27(4):299–308. DOI: 10.1007/s40292-020-00395-w
22. Johnson SC. Nutrient Sensing, Signaling and Ageing: The Role of IGF-1 and mTOR in Ageing and Age-Related Disease. Subcell Biochemistry. 2018;90:49–97. DOI: 10.1007/978-981-13-2835-0_3
23. Chisalita SI, Johansson GS, Liefvendahl E, Bäck K, Arnqvist HJ. Human aortic smooth muscle cells are insulin resistant at the receptor level but sensitive to IGF1 and IGF2. Journal of Molecular Endocrinology. 2009;43(6):231–9. DOI: 10.1677/JME-09-0021
24. Planavila A, Redondo-Angulo I, Villarroya F. FGF21 and Cardiac Physiopathology. Frontiers in Endocrinology. 2015;6:133. DOI: 10.3389/fendo.2015.00133
25. Planavila A, Redondo-Angulo I, Ribas F, Garrabou G, Casademont J, Giralt M et al. Fibroblast growth factor 21 protects the heart from oxidative stress. Cardiovascular Research. 2015;106(1):19–31. DOI: 10.1093/cvr/cvu263
26. Joki Y, Ohashi K, Yuasa D, Shibata R, Ito M, Matsuo K et al. FGF21 attenuates pathological myocardial remodeling following myocardial infarction through the adiponectin-dependent mechanism. Biochemical and Biophysical Research Communications. 2015;459(1):124–30. DOI: 10.1016/j.bbrc.2015.02.081
27. Rupérez C, Lerin C, Ferrer-Curriu G, Cairo M, Mas-Stachurska A, Sitges M et al. Autophagic control of cardiac steatosis through FGF21 in obesity-associated cardiomyopathy. International Journal of Cardiology. 2018;260:163–70. DOI: 10.1016/j.ijcard.2018.02.109
28. Cardoso AL, Fernandes A, Aguilar-Pimentel JA, de Angelis MH, Guedes JR, Brito MA et al. Towards frailty biomarkers: Candidates from genes and pathways regulated in aging and age-related diseases. Ageing Research Reviews. 2018;47:214–77. DOI: 10.1016/j.arr.2018.07.004
29. Tezze C, Romanello V, Sandri M. FGF21 as Modulator of Metabolism in Health and Disease. Frontiers in Physiology. 2019;10:419. DOI: 10.3389/fphys.2019.00419

ISSN 0022-9040. Kardiologiia. 2022;62(6). DOI: 10.18087/cardio.2022.6.n2033

30. Chen W, Tian B, Liang J, Yu S, Zhou Y, Li S. Matrix stiffness regulates the interactions between endothelial cells and monocytes. Biomaterials. 2019;221:119362. DOI: 10.1016/j.biomaterials.2019.119362
31. Srivastava P, Badhwar S, Chandran DS, Jaryal AK, Jyotsna VP, Deepak KK. Imbalance between Angiotensin II - Angiotensin (1-7) system is associated with vascular endothelial dysfunction and inflammation in type 2 diabetes with newly diagnosed hypertension. Diabetes & Metabolic Syndrome: Clinical Research & Reviews. 2019;13(3):2061–8. DOI: 10.1016/j.dsx.2019.04.042
32. Stanifer JW, Landerman L, Pieper CF, Huffman KM, Kraus WE. Relations of established aging biomarkers (IL-6, D-dimer, s-VCAM) to glomerular filtration rate and mortality in community-dwelling elderly adults. Clinical Kidney Journal. 2018;11(3):377–82. DOI: 10.1093/ckj/sfx097
33. Villarroya J, Gallego-Escuredo JM, Delgado-Anglés A, Cairó M, Moure R, Gracia Mateo M et al. Aging is associated with increased FGF21 levels but unaltered FGF21 responsiveness in adipose tissue. Aging Cell. 2018;17(5):e12822. DOI: 10.1111/acel.12822
34. Richter V, Rassoul F, Purschwitz K, Hentschel B, Reuter W, Kuntze T. Circulating Vascular Cell Adhesion Molecules VCAM-1, ICAM-1 and E-Selectin in Dependence on Aging. Gerontology. 2003;49(5):293–300. DOI: 10.1159/000071710
35. Jia G, Aroor AR, Jia C, Sowers JR. Endothelial cell senescence in aging-related vascular dysfunction. Biochimica et Biophysica Acta (BBA) - Molecular Basis of Disease. 2019;1865(7):1802–9. DOI: 10.1016/j.bbadis.2018.08.008
36. Shimizu I, Minamino T. Cellular senescence in cardiac diseases. Journal of Cardiology. 2019;74(4):313–9. DOI: 10.1016/j.jjcc.2019.05.002
37. Bruder SP, Jaiswal N, Haynesworth SE. Growth kinetics, self-renewal, and the osteogenic potential of purified human mesenchymal stem cells during extensive subcultivation and following cryopreservation. Journal of Cellular Biochemistry. 1997;64(2):278–94. DOI: 10.1002/(SICI)1097-4644(199702)64:2<278::AID-JCB11>3.0.CO;2-F
38. Dodig S, Čepelak I, Pavić I. Hallmarks of senescence and aging. Biochemia medica. 2019;29(3):483–97. DOI: 10.11613/BM.2019.030501

ISSN 0022-9040. Kardiologiia. 2022;62(6). DOI: 10.18087/cardio.2022.6.n2033