



Platelets' Functional Peculiarities in Persons of the Second Mature Age with Spinal Column Osteochondrosis of the Second Degree

A. A. Bikbulatova^{1*}, E. G. Andreeva² and I. N. Medvedev¹

¹*Russian State Social University, Moscow, Russia.*

²*The Kosygin State University of Russia, Moscow, Russia.*

Authors' contributions

This work was carried out in collaboration between all authors. Author AAB designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors EGA and INM managed the analyses of the study. Author INM managed the literature searches. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/ARRB/2017/37795

Editor(s):

(1) George Perry, Dean and Professor of Biology, University of Texas at San Antonio, USA.

Reviewers:

(1) Suprava Patel, All India Institutes of Medical Sciences, India.

(2) Marco Matteo Ciccone, University of Bari, Italy.

Complete Peer review History: <http://www.sciencedomain.org/review-history/22274>

Original Research Article

Received 28th October 2017
Accepted 8th December 2017
Published 13th December 2017

ABSTRACT

Introduction: Many disturbances in a body are often accompanied by some disturbance of blood parameters. Regular blood elements and, especially, platelets can play a significant role in development of dystrophic changes in any tissues. Their aggregation can influence the processes of microcirculation and metabolism in all the internals and in musculoskeletal system. Clarification of characteristics of platelet aggregation in the second mature age which very often gives clinical manifestations of osteochondrosis can help in understanding the mechanisms of its progression and the methods of its correction in having this disturbance patients.

The Aim: is to estimate platelets' aggregative activity in persons of the second mature age with spinal column osteochondrosis of the 2nd degree.

Materials and Methods of Research: In this study we enrolled 37 healthy persons of both sexes of the second mature age (men 35-60 years, women 35-55 years) and also 43 persons of both sexes of the same age with osteochondrosis of the 2nd degree. We applied biochemical, hematological and

*Corresponding author: E-mail: aabikbulatova@mail.ru;

statistical methods of investigation.

Results: Plasma of the examined persons with spinal column osteochondrosis had increased level of acylhydroperoxides by 38.4%. It was accompanied by the rise of P-selectin in their blood by 21.8% and platelet/endothelial cell adhesion molecule 1 – by 23.9%. The level of cholesterol in platelets of persons with osteochondrosis was increased by 30.5% at lowering of common phospholipids by 19.7%. The quantity of acylhydroperoxides in their platelets was increased by 32.5%. At the same time, the persons with osteochondrosis were noted to have earlier development of platelets' aggregation, than in the control: with collagen – by 37.4%, with ADP – by 32.3%, with rystomicin – by 42.2%.

Conclusion: The persons of the second mature age with spinal column osteochondrosis of the 2nd degree are noted to have strengthening of platelets' aggregation. It can affect the processes of microcirculation and promote the progression of osteochondrosis. These changes should be taken into account in the search of options for osteochondrosis deceleration and reducing the severity of its manifestations by application the tested therapeutic approaches because of their effect on aggregation properties of platelets.

Keywords: The second mature age; osteochondrosis; platelets; aggregation; microcirculation.

1. INTRODUCTION

Frequent occurrence of degenerative-dystrophic changes in spinal columns of people nowadays is mostly explained by wide range of their initiation's causes [1,2]. It's noted that heredity [3] and also negative impacts of environmental factors and way of life [4,5] play some role in their development. High prevalence of osteochondrosis among people of the second mature age, its susceptibility to chronicity, stable resistance to applied medicinal impacts dictate the necessity of detailed studying of all the changes in a body in persons of the second mature age against its background [6,7].

It is noted that development of osteochondrosis nearly always leads to worsening of the common functional state of a body [8]. It is connected with the fact that development of osteochondrosis influences negatively the processes of most internals' functioning [9]. As earlier researches showed, the presence of something unfavorable in a spinal column (even in the form of scoliosis) is accompanied by dysfunctions of regular blood elements, including platelets. It leads to worsening of microcirculation in tissues [10]. Emerging in these conditions inhibition of oxygen inflow into tissues can disturb anabolic processes in a body, weaken its vitality [11,12]. It created the basis for pathology development in the internals [13,14] and promoted the onset of vessels' persistent spasm [15,16]. It was found that degenerative changes in the spine contributed to increasing blood pressure and the gradual development of arterial hypertension [17]. Besides, the presence of osteochondrosis burdened the course of already existing cardiovascular pathology and promoted the formation

of resistance to conducted hypotensive therapy [18].

That's why, studying of platelets' activity at developing osteochondrosis can help in the search of physiological and efficient approaches to inhibition of its progression and to prophylaxis of different complications' development against its background. That's why, we put the following aim in our research: to determine peculiarities of platelets' aggregative ability in persons of the second mature age with osteochondrosis of the 2nd degree.

2. MATERIALS AND METHODS

This research was approved by the local Ethics Committee of the Russian State Social University on May, 14th, 2015 (Record №5). All the examined persons gave written informed consent on participation in conducted research. The research was conducted on people living in Central Russia (Moscow City and Moscow region). Into our research we enrolled 37 healthy people of both sexes (18 men and 19 women) of the second mature age (men 35-60 years, women 35-55 years, mean age 43.5±2.5 years) who composed the control group. We also examined 43 people of both sexes (21 men and 22 women) of the same age (mean age 44.7±1.9 years) with osteochondrosis of the 2nd degree who composed the group of observation. The diagnosis of osteochondrosis of the 2nd degree was confirmed clinically (pain and discomfort in the lumbar region, intensifying with physical exertion and prolonged staying in one and the same position) and rontgenologically (decrease of the distance between the vertebrae) and the appearance of osteophytes in the lumbar spine)

[6]. Existing in some persons from the group of observation concomitant chronic diseases (chronic bronchitis, chronic tonsillitis, chronic cholecystitis) were in the state of lasting persistent remission. All the persons from the group of observation and from the control group were once observed and examined. The size of the monitoring control groups was determined according to the number of persons who expressed a desire to participate in the study and gave voluntary, informed written consent.

In our research we determined the activity of the processes of lipids' peroxidation (LPO) in blood plasma which was registered according to the content of thiobarbituric acid (TBA)-active products in it with the help of a kit produced by the firm "Agat-Med" (Russia) and to the level of acylhydroperoxides (AHP) [19]. We also registered antioxidant activity of blood [20]. Implementing this method we used prepared beforehand preparation of the combined brain homogenase obtained from 5-6 rats. The brain of each animal was homogenized in 10 ml of physiological solution and freed from large particles by centrifugation followed by filtration through gauze. The mixture of the supernatants obtained for the research was stored in the frozen state for not more than two weeks. To carry out the tests, 1.79 ml of isotonic sodium phosphate buffer was added to the test tube (pH = 7.4). Then 0.2 ml of homogenate and 0.01 ml of test plasma were added into this tube, the tube was incubated under air access conditions at 37° C for 60 minutes. After that the induced lipoperoxidation was stopped by adding 28% trichloroacetic acid with 0.1% ethylene diamine tetraacetic acid. In parallel case (control sample), the oxidation of the used homogenate was evaluated in the absence of the plasma under study (instead of it, 0.01 ml of sodium phosphate buffer was added to the tube, otherwise the determination process did not change). The antioxidant activity of blood plasma was assessed by the degree of suppression of lipoperoxidation *in vitro* in the presence of a biological fluid. The calculation was carried out according to the formula: antioxidant activity of blood plasma = $100\% \times [1 - (E_0; 60 - E_0; 0) / (E_k; 60 - E_k; 0)]$, where $E_k; 60$ and $E_k; 0$ are the optical density values after 60 minutes of incubation and at the "zero" time point for samples containing plasma under study, and $E_0; 60$ and $E_0; 0$ are analogous extinction values for samples that do not contain plasma under study.

We carried out washing platelets off plasma for estimation of POL products' content in platelets,

level of lipids and antioxidant activity of enzymes. It was done according to the following method. Blood, mixed with 5% ethylene-diamine-tetra-acetic acid, was centrifuged at 1000 turns a minute during 10 minutes. Supernatant layer was put into dry clean test tube and centrifuged at 1500 turns a minute for 6 minutes. Then, the same layer was put into new dry clean test tube and centrifuged at 2200 turns a minute for 15 minutes. In the result, we got precipitation which was composed only of platelets. Then, supernatant liquid was removed, and there was added 1/3 of the volume of initial blood of 0.85% sodium chloride solution to the platelets' precipitation. It was prepared on 2.7% solution of ethylene-diamine-tetra-acetic acid. The precipitation was mixed carefully and centrifuged at 2200 turns a minute for 10 minutes. After that, the supernatant was removed, and there was again added physiological solution to the precipitation which was prepared on the solution of ethylene-diamine-tetra-acetic acid. The described procedure was conducted three times [21].

After platelets' washing and resuspending we estimated quantitatively the levels of cholesterol (CS) by enzymatic colorimetric method with the help of a kit produced by the firm "Vital Diagnostikum" (Russia) and common phospholipids (CPL) according to the quantity of phosphorus contents in them [22].

We determined molecules' concentrations of P-selectin and platelet/endothelial cell adhesion molecule 1 (PECAM-1) (Bender MedSystems GmbH, Austria) by enzymeimmunoassay in plasma.

The evidence of intraplatelet LPO processes was determined in washed and resuspended platelets according to concentration of malon dialdehyde (MDA) in the reaction of thiobarbituric acid reduction and quantity of AHP [19].

Platelets' quantity in children's capillary blood was calculated with the help of Gorjaev's box. Platelets' aggregation (PA) was estimated by visual micromethod with application of ADP (0.5×10^{-4} M), collagen (dilution 1:2 of the basic suspension), thrombin (0.125 un/ml), adrenaline (5.0×10^{-6} M) and hydrogen peroxide (7.3×10^{-3} M) as inductors [23]. Briefly, the micromethod of evaluating of platelet aggregation can be characterized as follows. Blood is to be taken with 3.8% sodium citrate in 9:1 ratio, centrifuged for 5 min at 1000 rpm to obtain platelet-rich plasma. Part of the plasma is taken, and the rest

is to be centrifuged at 3000 rpm for 20 minutes to obtain platelet-poor plasma. Platelet-rich plasma is standardized by the platelet number to $200 \times 10^9/L$. From final standardized plasma, 0.02 ml plasma is taken per each analyzed inductor. 0.02 ml plasma is taken on a slide from collected standardized plasma, and 0.02 ml inductor solution with different pipettes. Plasma is to be mixed with inductors with a glass rod, and then a stopwatch is to be started. The mixture is stirred so that to held the liquid within a circle of 2 cm diameter. When moving circularly the slide in transmitted beams of the illuminator, aggregates appearance is observed through a magnifying glass against a black background. Once the aggregates are clearly evident, the solution is clarified, and some aggregates are stuck to the glass, the stopwatch is stopped and the time of platelet aggregation is recorded. The reaction is repeated 2-3 times with each inductor and an

arithmetic mean is found from the obtained resultants.

Received in our research results were processes by Student's (t) criterion and correlation analysis.

3. RESULTS AND DISCUSSION

The examined persons with osteochondrosis were noted to have increase of LPO processes (Table 1). The quantity of AHP and TBA-products in their plasma surpassed the control values by 38.4% ($p < 0.01$) and 37.4% ($p < 0.01$), respectively (control values – 1.77 ± 0.23 D₂₃₃/1ml and 3.26 ± 0.29 mkmol/l, respectively). It took place against the background of weakening of plasma antioxidant activity in them which reached $23.8 \pm 0.41\%$ (in the control group – $32.6 \pm 0.49\%$, $p < 0.01$).

Table 1. Hematologic characteristics of the examined persons second adulthood with osteochondrosis

Indicators	Persons with osteochondrosis, n=43, M±m	Control, n=37, M±m
Acylhydroperoxides of plasma, D ₂₃₃ /l ml	2.45±0.48	1.77±0.23 p<0.01
Thiobarbituric acid-products of plasma, mkmol/l	4.48±0.52	3.26±0.29 p<0.01
Antioxidant activity of plasma, %	23.8±0.41	32.6±0.49 p<0.01
P-selectin, ng/ml	119.9±0.49	98.4±0.42 p<0.01
PECAM-1, ng/ml	58.6±0.38	47.3±0.27 p<0.01
cholesterol of platelets, mkmol/10 ⁹ platelets	1.07±0.012	0.82±0.016 p<0.01
common phospholipids of platelets, mkmol/10 ⁹ platelets	0.61±0.009	0.73±0.008 p<0.01
acylhydroperoxides of platelets, D ₂₃₃ /10 ⁹ platelets	4.08±0.017	3.08±0.012 p<0.01
malonic dialdehyde of platelets, nmol/10 ⁹ platelets	1.85±0.012	1.37±0.009 p<0.01
AP with ADP, s	31.9±0.18	42.2±0.11 p<0.01
AP with collagen, s	23.5±0.19	32.3±0.09 p<0.01
AP with thrombin, s	41.0±0.14	56.1±0.14 p<0.01
AP with ristomycin, s	33.2±0.15	45.8±0.10 p<0.01
AP with c H ₂ O ₂ , s	35.1±0.25	46.7±0.20 p<0.01
AP with epinephrine, s	72.2±0.33	93.6±0.036 p<0.01

Conventions: p – the significance of differences in the parameters of those surveyed who have osteochondrosis and control groups

Plasma of the examined persons with osteochondrosis was noted to have level increase of accountable adhesion molecules (Table 1): the levels of P-selectin and PECAM-1 turned out to be higher in them than in the control groups by 21.8% ($p<0.01$) and 23.9% ($p<0.01$), respectively.

Platelets' membranes of persons with osteochondrosis of the 2nd degree were noted to have the rise of CS level by 30.5% (1.07 ± 0.012 mkmol/ 10^9 platelets, $p<0.01$) and lowering of CPL by 19.7% (0.61 ± 0.009 mkmol/ 10^9 platelets, $p<0.01$) in comparison with the control values. It was accompanied by strengthening of LPO processes in platelets: AHP were higher by 32.5% ($p<0.01$), MDA – by 35.0% ($p<0.01$), reaching 4.08 ± 0.017 D₂₃₃/ 10^9 platelets and 1.85 ± 0.012 nmol/ 10^9 platelets, respectively.

The persons with osteochondrosis of the 2nd degree were noted to have activation of platelet activity (Table 2). The examined persons with osteochondrosis were noted to have acceleration of platelets' aggregation with all the applied in the research inductors. At the same time, the most evident platelets' reaction in them was noted in

response to collagen ($23.5\pm 0.19s$ – accelerated value by 37.4% in comparison with the control value), to ADP ($31.9\pm 0.18s$ accelerated value by 32.3% in comparison with the control value, $p<0.01$) and to ristomycin ($33.2\pm 0.15s$ accelerated value by 42.2% in comparison with the control value, $p<0.01$). PA was less active with H₂O₂ (accelerated value by 33.0% in comparison with the control value, $p<0.01$) and thrombin (accelerated value by 36.8% in comparison with the control value, $p<0.01$). Maximal duration of PA development was observed in persons with osteochondrosis in response to adrenaline which was accelerated by 29.6% in comparison with the control level ($p<0.01$).

During the evaluation of the correlation of platelet aggregation with the biochemical parameters of plasma and platelets taken into account, direct and inverse (moderate and strong) connections were revealed (Table 2).

The processes of a body's vital activity are accompanied by its continuous interaction with the environment at homeostasis maintenance [24,25]. Disturbances in different internals,

Table 2. The results of the evaluation of the correlation of platelet aggregation with LPO plasma and platelet counts, the antioxidant activity of plasma and the lipid composition of platelets in people of second adulthood with osteochondrosis of the 2nd degree

Biochemical parameters of plasma and platelets	Platelet aggregation					
	With ADP	With collagen	With thrombin	With ristomycin	With c H ₂ O ₂	With epinephrine
Acyhydroperoxides of plasma	0.73 $p<0.01$	0.78 $p<0.01$	0.79 $p<0.01$	0.72 $p<0.01$	0.84 $p<0.01$	0.72 $p<0.01$
Thiobarbituric acid-products of plasma	0.65 $p<0.05$	0.62 $p<0.05$	0.67 $p<0.01$	0.57 $p<0.05$	0.54 $p<0.05$	0.51 $p<0.05$
Antioxidant activity of plasma	-0.57 $p<0.05$	-0.58 $p<0.05$	-0.62 $p<0.05$	-0.54 $p<0.05$	-0.58 $p<0.05$	-0.51 $p<0.05$
P-selectin	0.78 $p<0.01$	0.84 $p<0.01$	0.88 $p<0.01$	0.85 $p<0.01$	0.85 $p<0.01$	0.84 $p<0.01$
PECAM-1	0.82 $p<0.01$	0.86 $p<0.01$	0.85 $p<0.01$	0.89 $p<0.01$	0.86 $p<0.01$	0.84 $p<0.01$
cholesterol of platelets	0.69 $p<0.01$	0.71 $p<0.01$	0.73 $p<0.01$	0.68 $p<0.01$	0.65 $p<0.01$	0.63 $p<0.01$
common phospholipids of platelets	-0.63 $p<0.01$	-0.70 $p<0.01$	-0.68 $p<0.01$	-0.64 $p<0.01$	-0.61 $p<0.01$	-0.60 $p<0.01$
acyhydroperoxides of platelets	0.66 $p<0.01$	0.71 $p<0.01$	0.73 $p<0.01$	0.70 $p<0.01$	0.68 $p<0.01$	0.69 $p<0.01$
malonic dialdehyde of platelets	0.60 $p<0.05$	0.54 $p<0.05$	0.57 $p<0.05$	0.52 $p<0.05$	0.53 $p<0.05$	0.55 $p<0.05$

Conventions: p - reliability of the identified correlation

Including spinal column, can often develop against this background. Degenerative-dystrophic changes develop in it most often. It was established that osteochondrosis occurred in approximately 70% of patients with spinal pathology, causing disability in 41.1% of cases [26]. It's noted long ago that development of osteochondrosis and its progression influence very often negatively functioning of all the internals and metabolism [27]. Activated aseptic inflammatory process in a spinal column at osteochondrosis often influences blood and vessels. It is accompanied by permeability rise of a basal membrane of vessels' wall what leads to lymph exudation, yield of leucocytes, macrophages and fibrinogen. Released thromboplastic substances accelerate transformation of fibrinogen into fibrin [28]. Rather important role here, as in any pathological process, is played by worsening of microcirculation process [29,30] which is connected with properties' changing of regular blood elements [31,32]. Notwithstanding the significant quantity of publications and all-round understanding of changes in a body in conditions of osteochondrosis development, the problems of platelet activity in conditions of the given state still remain poorly investigated and need additional studying.

The performed research was conducted on not very large number of participants. At the same time, the received data are reliable what allows us drawing conclusions based on them.

It was found out in the conducted research that there was some weakening of a body's antioxidant protection at osteochondrosis together with the rise of LPO intensity in plasma and its cells. The products of plasma and platelets' lipid peroxidation caused rearrangements in platelet membranes and worsened their functioning. It was aggravated by CS growth in platelets' membranes at osteochondrosis and by CPL decrease promoting the formation of membrane-pathy [28]. The study found out that increased platelet aggregation in persons with osteochondrosis strongly correlates with biochemical disorders in plasma and platelets. Consequently, microcirculation was inevitably worsened in all the internals of their bodies what created preconditions for metabolism weakening and risk formation of vascular disturbances.

Concentrations of molecules of cellular adhesion P-selectin and PECAM-1 in blood are very sensitive indices of platelet activity. These

molecules have platelet and endothelial origin. Their concentrations point at the level of their expression and, thus, at the potential of interaction between platelets and endothelium. It allows considering them markers of platelets' ability to endothelial adhesion. The strong correlation between the increased levels of concentrations of P-selectin and PECAM-1 in plasma and strengthened PA indicates a significant contribution to the development of physiologically unprofitable changes in platelet aggregation and disaggregation. So, the rise of these molecules' plasma level can be considered one of the mechanisms of platelet activity strengthening in vivo.

The rise of the given index also pointed at the presence of risk episodes of capillary course blocking by platelet aggregates and the formation of conditions for metabolism weakening in tissues [28,32].

The strong and moderate correlation between the acceleration of PA and LPO increase in plasma and platelets revealed in individuals with osteochondrosis indicates the existence of its serious contribution to the development of hyperaggregation in this state. Membranopathy is inevitably formed in persons with osteochondrosis of the 2nd degree against the background of excessive lipid peroxidation and lipid abnormalities in platelet membranes which are accompanied by physiologically unfavorable changes of receptor and post-receptor mechanisms in platelets [33]. The authors connected the reduction of AP development period under the impact of rystomicin in persons with osteochondrosis with the content rise of von Willebrand's Factor in patients' blood [34]. The lowering of platelets' resistance to hydrogen peroxide in the test on PA with H₂O₂ pointed at activity weakening of platelets' antioxidation system. Reached significant PA acceleration against the background of osteochondrosis can be connected with not only high LPO in platelets' membranes but also with activity rise of platelet enzymes of thromboxane-formation. It is indirectly pointed at by acceleration of PA process with weak inductors which is always realized through the mechanism of thromboxane-formation [35].

Found high platelets' aggregative activity in persons with osteochondrosis allows looking at the given state in a new way. It becomes clear that the rise of platelets' aggregation at osteochondrosis inevitably disturbs

microcirculation in tissues, including bones, cartilages and muscles. It promotes pathology progression in spinal column. That's why, there are all grounds to consider that it's quite possible to inhibit osteochondrosis manifestations in case of planned weakening of platelets' activity. Not only medicines should be used for reaching this aim. Apparently, care and non-drug impacts on a body are very important for recovery. In some cases, it is possible to get better results than after taking medications [36]. The hope on greater perspectiveness of the given approach is inspired by earlier found in children with scoliosis possibility of normalization of platelet activity and spinal column's morpho-functional characteristics with the help of non-pharmacological impact [37]. Taking these facts into consideration the authors plan to test the capabilities of designed earlier by them medicinal-prophylactic clothes [38,39] in relation to the evidence of clinical manifestations of osteochondrosis and platelet activity in persons of mature age.

4. CONCLUSION

Development of spinal column osteochondrosis is rather often accompanied by worsening of regular blood elements' functional properties what can negatively influence microcirculation. In the conducted research it was detected that persons of the second mature age with osteochondrosis of the 2nd degree were characterized by strengthening of lipids' peroxidation processes in plasma and platelets. It was accompanied by acceleration of platelet aggregation in them. Found disturbances can inevitably worsen the processes of blood rheology in capillaries. It leads to negative changes in tissue trophism, including spinal column, promoting progression of osteochondrosis. Taking negative consequences of microrheological disturbances at osteochondrosis into account, it seems to be very important to conduct further search of correction variants of the given state with account of their ability to influence platelets' aggregation.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. van Weeren PR, Jeffcott LB. Problems and pointers in osteochondrosis: Twenty years on *Veterinary Journal*. 2013;197(1):96-102.
2. Wang S, Guo X, Wang W, Wang S. Genome-wide study identifies the regulatory gene networks and signaling pathways from chondrocyte and peripheral blood monocyte of Kashin-Beck disease. *Genes to Cells*. 2012;17(8):619-632.
3. Riddick TL, Duesterdieck-Zellmer K, Semevolos SA. Gene and protein expression of cartilage canal and osteochondral junction chondrocytes and full-thickness cartilage in early equine osteochondrosis. *Veterinary Journal*. 2012;194(3):319-325.
4. Serteyn D, Piquemal D, Vanderheyden L, Verwilghen D, Sandersen C. Gene expression profiling from leukocytes of horses affected by osteochondrosis. *Journal of Orthopaedic Research*. 2010;28(7):965-970.
5. van Weeren PR, Olstad K. Pathogenesis of osteochondrosis dissecans: How does this translate to management of the clinical case? *Equine Veterinary Education*. 2016;28(3):155-166.
6. Sviatskaya EF. Lumbar osteochondrosis: diagnosis, treatment, medical rehabilitation. *Problems of Health and Ecology*. 2012;1(31):85-92.
7. Cociug A, Nacu V, Macagonova O. The modality of the regeneration of the intervertebral lomber disc in osteochondrosis. *IFMBE Proceedings*. 2016;55:454-457.
8. Del Grande F, Maus TP, Carrino JA. Imaging the intervertebral disk. age-related changes, herniations, and radicular pain. *Radiologic Clinics of North America*. 2012;50(4):629-649.
9. Glagoleva TI, Zavalishina SYu. Physiological peculiarities of vessels' disaggregating control over new-born calves' erythrocytes. *Annual Research & Review in Biology*. 2017;19(1):1-9. DOI: 10.9734/ARRB/2017/37232
10. Zavalishina SY, Nagibina EV. Dynamics of microrheology characteristics of erythrocyte in children 7-8 years with scoliosis with therapeutic physical training and massage. *Technologies of Living Systems*. 2012;9(4):29-34.
11. Zavalishina SYu, Kutafina NV, Vatnikov YuA, Makurina ON, Kulikov EV. Platelet-activity dependence on the age of rats with experimental dyslipidemia. *Biol Med (Aligarh)*. 2016;8:326. DOI: 10.4172/0974-8369.1000326

12. Zavalishina SYu. Physiological dynamics of spontaneous erythrocytes' aggregation of rats at last ontogenesis. Annual Research & Review in Biology. 2017; 13(1):1-7.
DOI: 10.9734/ARRB/2017/33616
13. Zavalishina SYu, Vatnikov YuA, Makurina ON, Kulikov EV, Sotnikova ED, Parshina VI, Rystsova EO, Kochneva MV, Sturov NV. Diagnostical appreciation of physiological reaction of intravascular thrombocytes activity of two-years-old mice to regular physical loads. Biomedical & Pharmacology Journal. 2017;10(1):129-136.
14. Zavalishina SY. Restoration of physiological activity of platelets in newborn calves with iron deficiency. Biomed Pharmacol J. 2017;10(2):711-716.
DOI: <http://dx.doi.org/10.13005/bpj/1160>
15. Skoryatina IA, Zavalishina SYu. Impact of experimental development of arterial hypertension and dyslipidemia on intravascular activity of rats' platelets. Annual Research & Review in Biology. 2017;14(5):1-9.
DOI: 10.9734/ARRB/2017/33758
16. Glagoleva TI, Zavalishina SYu. Aggregative activity of basic regular blood elements and vascular disaggregating control over it in calves of milk-vegetable nutrition. Annual Research & Review in Biology. 2017;12(6):1-7.
DOI: 10.9734/ARRB/2017/33767
17. Skoryatina IA, Zavalishina SYu. A Study of the Early Disturbances in Vascular Hemostasis in Experimentally Induced Metabolic Syndrome. Annual Research & Review in Biology. 2017;15(6):1-9.
DOI: 10.9734/ARRB/2017/34936
18. Skoryatina IA, Zavalishina SYu, Makurina ON, Mal GS, Gamolina OV. Some aspects of treatment of patients having dislipidemia on the background of hypertension. Prensa Med Argent 103:3.
DOI: 10.4172/lpma.1000250
19. Gavrillov VB, Mishkorudnaya MI. Spectrophotometric determination of the content of lipid hydroperoxides in blood plasma. Laboratory Work. 1983;3:33-36.
20. Volchegorskij IA, Dolgushin II, Kolesnikov OL, Cejlikman VJe. Experimental modeling and laboratory assessment of adaptive reactions of the organism. Cheljabinsk. 2000;167.
21. Yastrebov GN. Method of platelets' isolation to explore their lipid composition. Laboratory Work. 1985;2:93-94.
22. Kolb VG, Kamyshnikov VS. Handbook of clinical chemistry. Minsk: "Belarus". 1982;367.
23. Vorobyeva NV. Physiological reaction of erythrocytes' microrheological properties on hypodynamia in persons of the second mature age. Annual Research & Review in Biology. 2017;20(2):1-9.
DOI: 10.9734/ARRB/2017/37718
24. Zaytsev GS, Bikbulatova AA, Egorova NA, Mozdykov AV, Kalashkova DO. Liminal aspects of dreams. Man In India. 2016; 96(12):5719-5734.
25. Bikbulatova AA. Determining the thickness of materials in therapeutic and preventive heat-saving garments. Proceedings of Higher Education Institutes. Textile Industry Technology. 2014;1(349):119-123.
26. Ma XL. A new pathological classification of lumbar disc protrusion and its clinical significance. Orthopaedic Surgery. 2015; 7(1):1-12.
27. Petraevskii AV, Gndoian IA. Pseudoexfoliation syndrome: Pathogenesis of impairment of vegetative sympathetic innervation associated with cervical spine disorder. Vestnik Oftalmologii. 2012;128(4):42-47.
28. Szponder T, Wessely-Szponder J. Plasma level of protein c, fibrinogen concentration, and platelet number in dogs with legg-calve-perthes disease and osteochondrosis. preliminary studies. Bulletin of the Veterinary Institute in Pulawy. 2010;54(3):433-436.
29. Maksimov VI, Parakhnevich AV, Parakhnevich AA, Glagoleva TI, Kutafina NV. Physiological reaction of erythrocytes' micro rheological peculiarities in milk fed piglets after the negative impact of the environment. Annual Research & Review in Biology. 2017;17(1):1-8.
DOI: 10.9734/ARRB/2017/35867
30. Maksimov VI, Parakhnevich AV, Parakhnevich AA, Glagoleva TI, Kutafina NV. Rheological properties of erythrocytes of healthy piglets during the transition from dairy to vegetable nutrition. Annual Research & Review in Biology. 2017; 16(4):1-7.
DOI: 10.9734/ARRB/2017/35865
31. Maksimov VI, Parakhnevich AV, Parakhnevich AA, Glagoleva TI, Kutafina

- NV. Erythrocytes' microrheological features of piglets during the phase of dairy-vegetable nutrition after damage or common supercooling. Annual Research & Review in Biology. 2017;16(3):1-8.
DOI: 10.9734/ARRB/2017/35864
32. Maksimov VI, Parakhnevich AV, Parakhnevich AA, Glagoleva TI, Kutafina NV. Physiological reaction of erythrocytes' micro rheological features in newborn piglets on unfavourable environmental factors. Annual Research & Review in Biology. 2017;16(1):1-8.
DOI: 10.9734/ARRB/2017/35866
33. Kutafina NV. Platelet parameters of holstein newborn calves. Annual Research & Review in Biology. 2017;15(2):1-8.
DOI: 10.9734/ARRB/2017/35214
34. Zavalishina SYu. Physiological features of hemostasis in newborn calves receiving ferroglukin, fosprenil and hamavit, for iron deficiency. Annual Research & Review in Biology. 2017;14(2):1-8.
DOI: 10.9734/ARRB/2017/33617
35. Glagoleva TI, Zavalishina SYu. Aggregation of basic regular blood elements in calves during the milk-feeding phase. Annual Research & Review in Biology. 2017;17(1):1-7.
DOI: 10.9734/ARRB/2017/34380
36. Ciccone Marco Matteo, Aquilino Ambrogio, Cortese Francesca, Scicchitano Pietro. Feasibility and effectiveness of a disease and care management model in the primary health care system for patients with heart failure and diabetes (Project Leonardo). Vasc Health Risk Manag. 2010;6(6):297-305.
37. Lapshina EV. State of locomotor function and activity of thrombocytes in children with scoliosis: Dissertation of candidate of biological Sciences. Kursk. 2011;152.
38. Kayumova RF, Bikbulatova AA. The outerwear correcting a bearing. Patent for the invention RU 2211651,16.11; 2001.
39. Bikbulatova AA, Khamatshina DA. Belt-corset with a warming effect. Patent. for the utility model RU 100719, 01.07.2010.

© 2017 Bikbulatova et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:

*The peer review history for this paper can be accessed here:
<http://sciedomain.org/review-history/22274>*