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Physiological peculiarities of thrombocyte activity of candidates into masters of sports in athletics of preadult age



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Ilya Nikolayevich Medvedev*

ABSTRACT

Introduction: Physiologically, platelets are an important component of homeostasis of the whole body and blood system. However, not everything is clear with platelet activity of young healthy persons who have systematic exercises, particularly masters of sports' candidate. Its in vitro and in vivo should be studied further.

Aims: The aims of this investigation are to find out the abnormalities of thrombocyte activity of master of sports' candidate in athletics of young age and compare them with thrombocyte level of young people who avoided exercises during their lifetime. Through modulation of their activity, they increase capillaries' perfusion and thereby metabolism in tissues.

Materials and methods: In our investigation group we took 125 candidates into masters of sports in athletics of preadult age regularly exercising and participating in competitions of different levels. Control group was consisted of 141 healthy youths having avoided exercises during their lifetime. Biochemical, hematological and statistical methods are applied.

Results: Athlete were found to have stable normal functional platelet activity. For subjects between 18 and 22 years, their

platelets aggregation was on low level and had no reliable variations related to their stable low sensitivity to inductors. Decreased platelet activity causes conservation in athletes' blood of some quantity of circulating thrombocyte aggregates. It makes positive impact on microcirculation in organs of athlete having regular athletic exercises. Avoidance of exercise in young age promotes gradual rise of thrombocyte activity. Masters of sports' in athletics candidate who had intensive muscle activities are characterized by persistence of stable low platelet activity. It provides low quantity of active platelets and their circulating aggregates in blood. Those young persons, who avoided exercises, are noted to have elevated platelet activity with a tendency increased aggregate formation.

Conclusion: Masters of sports in athletics' candidate doing intensive muscle activity are characterized by preservation of stable not high platelet activity caused by low content of active platelets and their circulating aggregates. Avoiding exercises in young people are noted to have caused the increase of platelet activity with a tendency to cause platelet aggregation.

Keywords: junior age; candidates for master of sports; track and field athletics; aggregation; platelets

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Russian State Social University,
Moscow, Russia

INTRODUCTION

Rational physical activity is a strong stimulator of a body's adaptive mechanisms to environmental factors in any age¹ and most pathological processes² which is able to prevent the development of aggravation of well-being.³ There is no doubt that the biggest role in this process is played by blood functionally favorably changing its composition and features on the background of regular ordered muscle activity.^{4,5} Great significance in provision of haemostatic and rheological blood features, especially in microcirculation system, belongs to platelets.^{6,7} The intensity of capillary blood flow and consequently the state of tissue metabolism^{8,9} mostly depend on their activity level.¹⁰ Earlier investigations showed that exercise cause positive changes of many hematological

indices.^{11,12} In available scientific literature we couldn't find enough data about the impact of exercising regularly on haemostatic features of thrombocytes. Furthermore, not everything is clear with platelet activity of young healthy persons who have systematic exercise, particularly master of sports' candidate. Its features in vitro and in vivo should still be studied further. There were also no comparison of platelets reactions to different inductors carried out. Furthermore, morphological platelets activation in vessels' lumen of athletes and healthy young people who avoided regular exercises during lifetime are not yet studied enough.

The aim of investigation: to find out the abnormalities of thrombocyte activity of masters of sports in athletics' candidates of young age and compare them with thrombocyte level of young people who avoided exercises during lifetime.

*Corresponding to:
Ilya Nikolayevich Medvedev;
Russian State Social University,
Moscow, Russia
ilmedv1@yandex.ru

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MATERIALS AND METHODS:

The study was approved by the ethic committee of Kursk Institute of Social Education (branch of Russian State Social University) with record №5 dated 12th of May, 2014. The study was conducted in the sport complex base of the Russian State Social University in Kursk, Russia. For the experimental group we took 125 healthy youths, candidates into masters of sports in athletics regularly training and participating in competitions of different levels (25 persons of 18 years, 26 persons of 19 years, 23 persons of 20 years, 24 persons of 21 years and 27 persons of 22 years). All the athlete trained not less than 5 days a week, not less than 2 hours a day. Their average age at the beginning of training was 11.2 ± 0.7 years. Control group was consisted of 141 healthy youths having avoided exercises during their lifetime (29 persons of 18 years, 26 persons of 19 years, 27 persons of 20 years, 28 persons of 21 years and 31 persons of 22 years).

Intrathrombocyte lipids' peroxidation (LPO) was defined according to platelets concentrations of malondialdehyde (MDA) and acylhydroperoxides (AHP), taking into consideration the activity of catalase and superoxide dismutase (SOD) in them, which were measured in international units (IU) on 10^9 platelets.¹³ Platelets aggregation (AP) was registered by visual micromethod¹⁴ with ADP ($0,5 \times 10^{-4}$ M), collagen (dilution 1:2 of suspension), ristomycin (0,8 mg/ml), and adrenaline (5×10^{-6} M). Intravascular platelet activity (IPA) was registered with the help of phase-contrast microscopy.¹⁴ Statistical processing of the results was made by Student's t-criterion. Statistical processing of received data was carried out with the usage of a program packet "Statistics for Windows v. 6.0" and "Microsoft Excel". Differences in data were considered to be significant in case of $p < 0,05$.

RESULTS

Mean platelet AHP in 18-year old athlete's group was 1.71 ± 0.18 D₂₃₃/10⁹ platelets, not changing to 22 years (1.69 ± 0.16 D₂₃₃/10⁹ platelets). At the same time, the content of MDA in platelets of 18-year-old athletes reached 0.37 ± 0.12 nmol/10⁹ platelets, similar to the rest examined persons (at 22-years-old - 0.39 ± 0.28 nmol/10⁹ platelets). AHP level in platelets of 18-year-old persons who avoided exercises was higher than in athlete of the same age (2.02 ± 0.13 D₂₃₃/10⁹ platelets) and in 22 years rose to 2.24 ± 0.07 D₂₃₃/10⁹ platelets. However, MDA level in their platelets at 18 years was higher than in athletes (0.52 ± 0.012 nmol/10⁹ platelets), also rising at older age (at 22 years - 0.69 ± 0.019 nmol/10⁹ platelets).

Activity of catalase and SOD in blood platelets

of masters of sports' candidate had no reliable dynamics while aging (at 18 years - 10550.0 ± 214.5 ME/10⁹ platelets and 1990.0 ± 12.7 ME/10⁹ platelets, at 22 years - 10150.0 ± 280.3 ME/10⁹ platelets and 2060.0 ± 12.7 ME/10⁹ platelets, respectively). Activity of thrombocyte catalase and SOD in youths who avoid exercises was lower than in athlete while aging the tendency to weakening (at 18 years - 9260.0 ± 101.2 ME/10⁹ platelets and 1650.0 ± 12.3 ME/10⁹ platelets, at 22 years - 8910.0 ± 166.4 ME/10⁹ platelets and 1500.0 ± 16.6 ME/10⁹ platelets, correspondingly).

In 18-year-old athlete, AP under collagen impact wasn't high (36.4 ± 0.24 s), similar to older athletes. Analogical feature of AP activity was noted in athlete in relation to ADP and ristomycin. Adrenaline AP in them turned out to be rather slow and had also no difference between ages (table). In physically untrained 18-year-old examined persons, AP developed as a response to strong and weak inductors earlier than in athlete and increased with age (Table 1).

The content of discocytes in blood of 18-year-old athlete was $88.3 \pm 0.14\%$, not significantly different from the values of examined persons of the other age groups. Total platelets active forms also stayed stable in athletes between 18 and 22 years of life. At the same time, in athlete's blood the levels of freely circulating little and large platelets aggregates is not statistically significant between 22-year-olds 2.2 ± 0.12 and 0.05 ± 0.012 on 100 freely lying platelets, . The platelets involved into the process of aggregate-formation did not differ between 18 and 22 years of life and composed in average $4.6 \pm 0.16\%$. In physically untrained youths the content of discocytes in blood at 18 years was a bit lower than in athlete ($85.6 \pm 0.15\%$), reliably lowering to 22 years. The sum of platelets active forms in them was initially higher and gradually increased with age. In their blood the levels of freely circulating little and large aggregates was significantly increased from 2.9 ± 0.14 and 0.07 ± 0.010 in 18-year-olds to 3.6 ± 0.04 and 0.10 ± 0.007 on 100 freely lying platelets in 22-year-olds, respectively. The number of platelets involved into aggregate formation in those who avoided exercises also increased between 18 and 22 years from $6.0 \pm 0.10\%$ to $6.8 \pm 0.06\%$ ($p \square 0,05$).

DISCUSSION

Increase of LPO level in platelets of young people, who avoided exercises, points indirectly at strengthening of the process of thromboxane formation in them.¹⁵ Evidently, it happens on behalf of simultaneous activity strengthening of metabolic enzymes of arachidonic acid of their membranes

Table 1. Platelet activity in examined persons of preadult age

Options	Age surveyed				
	18 years	19 years	20 years	21 years	22 years
Do not exercise physically, n=141, M±m					
number of examinees	n=29	n=26	n=27	n=28	n=31
AP with ADP, s	44.6±0.06	44.7±0.15	44.2±0.12	43.6±0.19	43.0±0.15
AP with collagen, s	33.9±0.27	33.8±0.26	33.3±0.12	32.7±0.17	32.0±0.04
AP with ristomicin, s	48.9±0.20	48.7±0.06	48.0±0.13	47.5±0.06	47.1±0.15
AP with epinephrine, s	106.3±0.10	105.6±0.22	98.3±0.02*	97.6±0.11*	96.8±0.11*
Thrombocytes-discocytes, %	85.6±0.15	85.4±0.10	83.0±0.04	82.1±0.03	80.0±0.04*
Sum of thrombocytes' active forms, %	14.4±0.14	14.6±0.12	17.0±0.15*	17.9±0.10*	20.0±0.09**
Candidates for the masters of sports in track and field athletics, n=125, M±m					
number of examinees	n=25	n=26	n=23	n=24	n=27
AP with ADP, s	47.9±0.12	49.4±0.14	48.5±0.13	47.1±0.22	49.2±0.16
AP with collagen, s	36.4±0.24	35.9±0.26	37.3±0.19	36.9±0.15	37.8±0.12
AP with ristomicin, s	53.2±0.20	52.8±0.23	54.3±0.28	55.0±0.26+	52.1±0.19+
AP with epinephrine, s	109.7±0.22	107.1±0.23	108.3±0.22+	110.7±0.19+	109.2±0.14++
Platelets-discocytes, %	88.3±0.14	86.9±0.26	88.1±0.17	89.4±0.09	87.3±0.05+
Sum of platelets active forms, %	11.7±0.17	13.1±0.20	11.9±0.24	10.6±0.23+	12.7±0.19++

Note: reliability of differences of platelets indices in examined with its value 18-year-olds * - $p \leq 0,05$, ** - $p \leq 0,01$.

Reliability of differences between athlete and untrained young people in one age group: + - $p < 0,05$; ++ - $p < 0,01$.

AP – aggregation of platelets.

- cyclooxygenase and thromboxane synthetase. It inevitably creates the situation for platelets activation in vivo.¹⁶

At the same time, data about platelets aggregative activity in vitro and in vivo in young people, who avoided exercises, are poorly understood.¹¹ With the result of the study, we can conclude that the absence of regular exercise promotes platelets activation. It leads to increased thromboplastin formation with the initiation of blood coagulation process and formation of one of the strongest inductors of aggregation, thrombin.

It was confirmed that young people with hypo dynamics have activation of adhesion ability of blood plates. It is realized in two ways. The first one is the increase of glycoproteins, collagen Ia-IIa and VI on the platelets membrane.¹⁷ We managed to find out with the help of collagen that cause AP acceleration. The second mechanism of activation of platelets adhesion is realized through the increase

of the content of von Willebrand Factor in blood and the number rise of places of its binding on platelets membranes (GPIB). The increase of von Willebrand Factor in blood of persons regularly avoiding exercises were caused by the rise of its synthesis in endothelium and secretion of blood plates.¹⁸ The rise of von Willebrand Factor in their blood was indirectly registered on the basis of rapid AP with ristomicin which can influence both platelets and sub endothelium of vessels. It is possible only in conditions of quantity increase of the given receptors on the surface of platelets. The first way of realization of platelets adhesion can take place in conditions of bloodstream in big vessels, the second one - in conditions of blood stream in vessels of little diameter.²⁰

AP registration of examined persons, who have hypo dynamics, with separate agonists allowed to establish its acceleration. Fixation of a strong agonist of aggregation – collagen in

conventional doses on receptors of platelets surface activates phospholipase C. It leads to the increase of diacylglycerol formation and strengthening of protein kinase C functioning. It also rises phospholirirovation of actomyosin. Forming in these conditions in abundance inositol triphosphate causes active release of Ca^{2+} into cytoplasm out of its depo. It also promotes acceleration and facilitation of actomyosin reduction.²¹

In case of young people avoiding exercise, weak stimulators of aggregation (ADP and adrenaline), along with corresponding receptors on platelets, causing the increase of fibrinogenic receptors (GPIIb-IIIa). At the same time, the activity of phospholipase A_2 rises and causing the release of arachidonic acid out of cells' membranes²² Cyclooxygenase and thromboxane synthetase become activated by existing biochemical situation. They provide the elevation of the most significant product (from the point of view of haemostatic) - thromboxane A_2 . It stimulates the aggregation of blood platelets through phospholipase A and C at the elevation of Ca^{2+} release into their cytoplasm.²³

To investigate the first stages of blood plates' activation in untrained young people in vivo and registration of its evidence in blood, we used IPA estimation method. It allowed to establish the increase of platelet active forms in examined person. Found evident IPA pointed indirectly at alteration of vessels' endothelium and at possibility of surplus platelets contact with sub endothelium fibers, including collagen. Besides, it pointed at the level rise of other physiological stimulators of aggregation (thrombin, ADP, adrenaline) in blood.²⁴

The elevation of platelet active forms in blood of persons with hypo dynamics leads to the increase of circulating aggregates of all sizes. While in bloodstream they can damage endotheliocytes. It promotes their destruction and desquamation, thus uncovering sub endothelium structures. These changes activate hemostasis in untrained young people and form the risk of thrombotic complications.

For athletes of 18-22 years old regularly having muscle activity it's common to have stable high platelets antioxidant activity suppressing the level of LPO in them and mostly providing conditions for constancy of blood platelets not high activity.

The masters of sports in athletics' candidate were found to have low platelet activity what is mostly connected with lowered sensitivity of their receptors to inductors. Given situation promotes support of normal activity of the whole hemostasis system.¹ It is the consequence of complicated adaptive reactions in athlete's organisms related to their adaptation to regular evident exercises.²⁵

Unexpressed AP intensity with strong agonist (collagen) has in its basis some depression in their platelets of phospholipase C, small activity of phosphoinositol and low level of proteins' phospholirirovation of contractile system.^{26,27} Forming in their platelets little quantity of inositol threephosphate promotes inhibition of Ca^{2+} outflow from its intrathrombocytedepos suppressing contractile ability of their actomyosin.^{25,28} Slow platelets reaction of athlete on weak inductors is, probably, caused by lowering of receptors' number to them on outer membranes of blood platelets, not high expression on them of fibrinogenic receptors (GPIIb-IIIa) and basal activity of phospholipase A_2 ⁵ what provides minimum necessary outcome of arachidonic acid from membrane's phospholipids and suppression of thromboxan A_2 synthesis.^{6,29}

Stable not large IPA in athletes of 18-22 years proves constancy in their blood of normal level of aggregation inductors at lowered sensitivity of platelets to them. At the same time in athlete's blood we noticed high quantity of intact discoid platelets and decreased number of their active forms additionally pointing at unexpressed activity of their aggregation mechanisms.

CONCLUSION

Masters of sports in athletics' candidate doing intensive muscle activity are characterized by preservation of stable not high platelet activity caused by low content of active platelets and their circulating aggregates. Avoiding exercises in young people are noted to have caused the increase of platelet activity with a tendency to cause platelet aggregation.

CONFLICT OF INTEREST

No Conflict of interest to declare.

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