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Research Article

# L-NAME-induced Preeclampsia: correction of functional disorders of the hemostasis system with Resveratrol and Nicorandil

Elena G. Stupakova[1], Galina A. Lazareva[1], Vladimir V. Gureev[1], Irina N. Dolzhikova[1], Lyudmila A. Zhilinkova[2], Anastasia V. Gureeva[1]

1 *Kursk State Medical University of the Ministry of Healthcare of the Russian Federation, 3 K. Marx St., Kursk 305041, Russian Federation*
2 *Kursk Academy of the Public and Municipal Service, 9 Stantsionnaya St. Kursk 305044, Russian Federation*

Corresponding author: *Elena G. Stupakova* (LazarevaGA@kursksmu.net)

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## Abstract

**Introduction.** Preeclampsia is a formidable disease of the second half of pregnancy, leading to severe complications, including disability and even death. Many authors have recognized the correlation between the severity of preeclampsia and the degree of disturbances in the hemostasis system. In this regard, the objective of this study was to assess inhibition of platelet aggregation and the possibility of its correction with resveratrol and nicorandil.

**Materials and methods.** The study was performed on 250 mature white Wistar female rats weighing 250–300 g. The platelet aggregation induced by ADP, collagen, ristomycin, adrenaline was determined, as well as PTT, TT, aPTT, fibrinogen, and the clotting time.

**Results and discussion.** Introduction of resveratrol and nicorandil resulted in a decrease in thrombocyte aggregation capacity from 53.8 ± 2.60% to 22.1 ± 1.25% and 37.1 ± 1.79%, respectively, when using ADP as an inducer. The clotting time was from 841 ± 42 s up to 1135 ± 33 s and 1034 ± 26 s, respectively. In addition, there was an increase in temporal parameters of plasma-coagulation hemostasis and a decrease in plasma fibrinogen content. The use of glibenclamide resulted in partial cancellation of the positive effects of resveratrol and nicorandil, with an increase in platelet aggregation to 28.9 ± 1.8% and 43.9 ± 1.2% when using ADP as an inducer and a decrease in the thrombosis time to 988 ± 26 s and 950 ± 22 s, respectively.

**Conclusion.** In animals with experimental preeclampsia, there were disturbances in the hemostasis system, comparable to those in the clinical situation. The use of resveratrol and nicorandil leads to a pronounced correction of hemostasis parameters. The positive effects of the studied pharmacological agents are mediated by several mechanisms, including $K^+_{ATP}$ channels.

## Keywords

experimental preeclampsia, platelet aggregation, hemostasis, nicorandil, resveratrol.

# Introduction

Preeclampsia remains an urgent problem of modern medicine and is one of the most ominous complications of pregnancy and childbirth (Gureev et al. 2014, Reznikova 2013, Shuvalova et al. 2014). Preeclampsia is among the top three causes of maternal and perinatal mortality (Ministry of Healthcare of the RF 2016, Shuvalova et al. 2014). Its incidence in recent years has tended to increase (Ministry of Healthcare of the Russian Federation 2016, Reznikova 2013, Shuvalova et al. 2014).

A large number of authors recognize endothelial dysfunction (Sánchez-Aranguren et al. 2014, Scioscia et al. 2015) and placental ischemia caused by dysangiogenesis as the main elements of gestosis pathogenesis (Pereira et al. 2015, Ducray et al. 2011, Verlohren et al. 2010). Incomplete invasion of cytotrophoblast into the spiral arteries of the mother and their remaining constriction capacity leads to trophoblast ischemia and an increased permeability of the fetoplacental barrier (Ducray et al. 2011, van Oppenraaij et al. 2011, Wang et al. 2015). At the end of complex pathogenetic processes of the response with the release of a large number of humoral factors are an increase in the content of ADMA (asymmetric dimethylargenine), endothelial dysfunction, secondary ischemic lesions, impaired hemostasis and oxidative stress (Gureev et al. 2014, Sánchez-Aranguren et al. 2014).

Disorders in the hemostasis system lead to severe complications in obstetrics: DIC-syndrome, HELLP-syndrome and uterine bleeding in various stages. The hemostasis system includes a vascular-thrombocytic unit, which consists of endothelium and platelets, and a plasma-coagulation unit, which consists of the blood coagulation system, the blood anticoagulation system and the fibrinolytic system.

In women with preeclampsia, on the surface of the platelet, the expression of collagen receptors, von Willebrand factor, P-selectin significantly increases compared with normal pregnancy. This leads to an increased platelet aggregation activity in preeclampsia (Holthe et al. 2004). In women with spontaneous premature labor, a failure of the physiological transformation of the spiral arteries and an abnormal angiogenic-antiangiogenic state of plasma were detected, comparable to such in women with preeclampsia. The use of antiplatelet agents reduced spontaneous premature delivery in pregnant women at risk for developing pre-eclampsia (Kazmi et al. 2011, van Vliet et al. 2017). The use of acetylsalicylic acid in low doses reduces the possibility of pre-eclampsia in pregnant women with high risk pregnancies (Käehne and Lundin 2017, Mone et al. 2017, Sahin et al. 2015, Sarma and Scott 2016). Simulation of preeclampsia in the experiment and its correction are also accompanied by a disturbed platelet aggregation ability and its normalization, respectively (Bariani et al. 2017, Tyurenkov et al. 2012, Tyurenkov et al. 2014).

Due to the severity of the complications caused by impaired hemostasis in preeclampsia, as well as in connection with the prevailing stereotype, abnormal platelet function has traditionally been considered within the hemostatic system (Blomqvist et al. 2018, Burke et al. 2013, Burke et al. 2016). This is clearly seen even when trying to assess the severity of preeclampsia and other pathological conditions of pregnant women by impairment of their function (Burke et al. 2016, Holthe et al 2004, Sidorenko et al. 2007). But even the available data show contradictions in the results obtained by different authors. However, most authors claim a decrease in the activity of the coagulation system, platelet aggregation ability in early pregnancy, followed by their significant increase in childbirth and in the postnatal period, which looks logical (Blomqvist et al. 2018, Burke et al. 2013, Freitas et al. 2014, Hayashi et al. 1999, Hellgren 2003). In the third trimester, the platelet aggregation sensitivity to ADP, adrenaline, ristomycin, collagen and other inducers increases (Blomqvist et al. 2018). This can be explained by increased expression of receptors for collagen, von Willebrand factor, P-selectin and other factors that contribute to the aggregation on the surface of the platelet (Holthe et al. 2004).

There have only been a small number of scientific publications written recently which discuss a certain independent role of platelets in the development of preeclampsia and the possibility of preventing and treating placental-mediated complications with drugs correcting the function of platelets (Blomqvist et al. 2018, Holthe et al. 2004, van Vliet et al. 2017).

However, at the present stage, there is enough information to consider the impaired platelet function as an independent key link in the pathogenesis of preeclampsia resulting in placental ischemia and a local endothelial dysfunction to its generalized form – systemic endotheliosis.

In this regard, the search for new drugs with a combined mechanism of action for the prevention and treatment of preeclampsia seems promising.

Earlier in our laboratory, the endothelioprotective properties of resveratrol and nicorandil were investigated (Kochkarov 2009, Stupakova et al. 2018), which can activate $K^+_{ATP}$ channels ( Novaković et al. 2015, Tanaka et al. 2010), therefore they were considered to be the most promising for carrying out the present research. In addition, when they were used, a pronounced effect was observed when correcting morphofunctional disorders under conditions of ADMA-like preeclampsia (Ivanets et al. 2015).

**Objective**: to conduct a study of platelet aggregation capacity in the condition of experimental preeclampsia and the possibility of its correction with resveratrol and nicorandil.

# Materials and methods

### Complying with the ethical and regulatory requirements when doing research

The experimental part of the study was performed at the premises of the vivarium of Kursk State Medical University.

The work was organized and carried out in compliance with the following regulatory acts and guidelines governing the conduct of experimental research in the Russian Federation:

1. Order of the Ministry of Healthcare of the Russia of April 1, 2016 No. 199N "On Approval of the Rules of Good Laboratory Practice" (Order of the Ministry of Healthcare of the Russia 2016).
2. GOST 33044-2014 "Principles of Good Laboratory Practice" (GOST 2015).
3. GOST 33217-2014 "Guidelines for the Maintenance and Care of Laboratory Animals. Rules for the Maintenance and Care of Laboratory Predatory Mammals" (GOST 2016).
4. "Guidelines for Conducting Preclinical Studies of New Drugs", Ed. Mironov A.N., - M.: Grif and Co. – 2012 (Mironov 2012).
5. The ethical principles of treating laboratory animals were in compliance with "The European Convention for the Protection of Vertebral Animals Used for Experience and Other Scientific Purposes. CETS No. 123" (European Treaty Series 1986).

**Experimental animals**

The experiment was performed on 250 white Wistar female rats weighing 250-300 g. To form groups of pregnant animals with predetermined periods, the males (2 animals) were introduced to the females, which had been kept separately, for 24 hours. Then the animals were separated, and 10 days later, during the ether sleep, pregnancy was determined by palpation. In the experiments, pregnancy occurred in 30-40% of cases. ADMA-like agent – non-selective blocker NO-synthase N-nitro-L-arginine-methyl ether (L-NAME) – was administered intraperitoneally at a dose of 25 mg/kg/day for seven days (14-20$^{th}$ days of pregnancy) (Gureev et al. 2014).

Determination of coagulation indicators and a degree of thrombocyte aggregation was performed on the 21$^{st}$ day of gestation. Blood from the abdominal aorta was collected into a test tube with a 3.8% solution of sodium citrate in a ratio of 9:1, followed by centrifugation of 1000 rpm for 10 minutes. Antiplatelet activity was determined by G.Born's method modified by Z.A. Gabbasova et al. (1989) on a two-channel laser analyzer of platelet aggregation ALAT-2 (Biola). ADF (at a final concentration of 5 μM), collagen (50 μg/ml), ristomycin (5 μM), adrenaline (10 μM) were used as inducers (manufactured by SPD RENAM, Russia). Analysis was performed no later than 2 hours after obtaining blood. Coagulation indicators were determined on a Start 4 (Diagnostica Stago, France) analyzer using reagents for the determination of prothrombin time (PTT), thrombin time (TT), activated partial thromboplastin time (aPTT), fibrinogen test (manufactured by SIEMENS). Thrombus formation time was determined on the 20$^{th}$ day of pregnancy in anesthetized (300 mg/kg of chloral hydrate) female rats with experimental preeclampsia. Thrombus formation was caused by applying a 50% solution of iron (III) chloride, for which an area of the exposed carotid artery was isolated from the surrounding tissues, and a cotton pad moistened with a 50% solution of iron chloride (0.025 ml) was placed on it. The blood flow was recorded above the site of application, using a Doppler probe (Minimax-Doppler-K, St. Petersburg). The time of blood clot formation from the moment of application of the iron (III) chloride solution to the complete cessation of blood flow in the carotid artery was noted (Karamysheva 2014, Voronkov 2011).

Descriptive statistics were applied to all the data: the data was checked for normal distribution. The type of distribution was determined by the Shapiro-Wilk criterion. In case of normal distribution, the mean value (M) and standard error of the mean (m) were calculated. Intergroup differences were analyzed by parametric (Student's t-test) or non-parametric (Mann-Whitney test) methods, depending on the type of distribution. The statistical significance of the differences between the morphological changes after their ranking was assessed using the Mann-Whitney method for analyzing non-parametric data (Glants 1999, Sidorenko 2003). All calculations were performed using the Microsoft Excel 7.0. statistical package.

According to the objective of the study, all the animals were divided into the following groups:

1. Intact – pregnant + 0.9% NaCl from the 14$^{th}$ to the 20$^{th}$ days of pregnancy.
2. Control (L-NAME) – pregnant + ADMA-like agent (with intraperitoneal administration of L-NAME at a dose of 25 mg/kg once a day from the 14$^{th}$ to the 20$^{th}$ days of pregnancy).
3. Pregnant + L-NAME 25 mg/kg + Amlodipine (0.5 mg/kg/day orally).
4. Pregnant + L-NAME 25 mg/kg + Nicorandil (2x10 mg/kg/day orally).
5. Pregnant + L-NAME 25 mg/kg + Resveratrol (2 mg/kg/day orally).
6. Pregnant + L-NAME 25 mg/kg + Nicorandil (2x10 mg/kg/day orally) + Amlodipine (0.5 mg/kg/day orally).
7. Pregnant + L-NAME 25 mg/kg + Resveratrol (2 mg/kg/day orally) + Amlodipine (0.5 mg/kg/day orally).
8. Pregnant + L-NAME 25 mg/kg + Nicorandil (2x10 mg/kg/day orally) + Glibenclamide (50 mg/kg).
9. Pregnant + L-NAME 25 mg/kg + Resveratrol (2 mg/kg/day orally) + Glibenclamide (50 mg/kg).

## Results and discussion

Modeling ADMA-like preeclampsia by intraperitoneal administration of L-NAME at a dose of 25 mg/kg/day resulted in an increase in platelet aggregation, which is evidenced by an increase in the maximum plasma light transmission (Fig. 1). When using ADF as an inducer, the light transmission increased from 24.3±1.09% to 53.8±2.60%, when using collagen – from 50.2±1.66% to

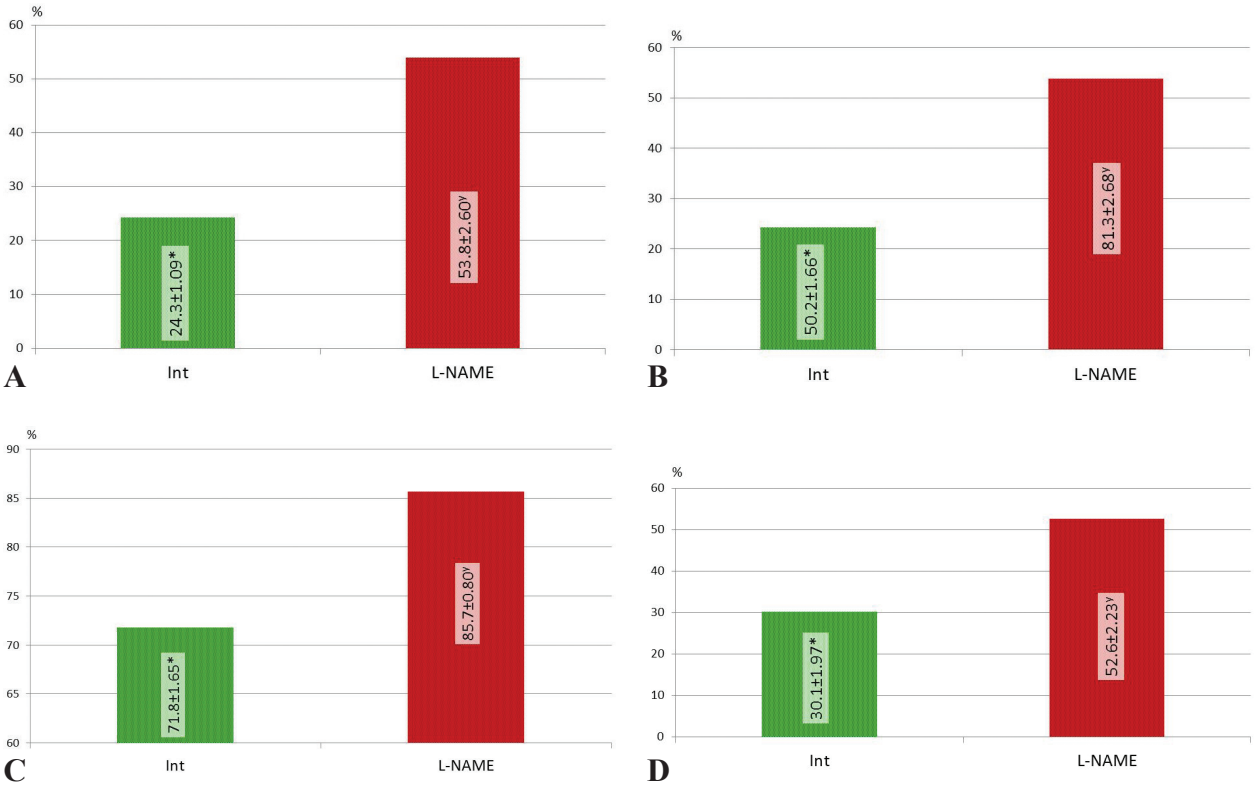


**Figure 1.** Time of induced platelet aggregation when modeling ADMA-like preeclampsia. *Note:* **A** (ADP), **B** (Collagen), **C** (Ristomycin), **D** (Adrenaline) used as inducers; * – $p<0.05$ in comparison with L-NAME animals; ʸ – $p<0.05$ in comparison with intact animals

81.3±2.68%, when using ristomycin – from 71.8±1.65% to 85.7±0.80% and when using adrenaline – from 30.1±1.97% to 52.6±2.23%.

When administering the studied drugs, nicorandil and resveratrol, to the animals with ADMA-like preeclampsia, aggregation of platelets with individual peculiarities was corrected. The efficacy of both studied drugs exceeded the efficacy of the drug included in the standards of treatment of hypertensive states in pregnant women – amlodipine (Fig. 2).

When induced with ADP, collagen and adrenaline against the use of nicorandil, there was a statistically significant decrease in platelet aggregation relative to that in the control ($p<0.05$), but it did not reach the target level and was statistically different from that in the group of intact animals ($p<0.05$). When inducing with ristomycin, the platelet aggregation level was statistically lower ($p<0.05$) than that of the "untreated" animals and reached the target level, as evidenced by there being no statistical difference from the platelet aggregation level of the animals of the intact group (Fig. 2).

The use of resveratrol resulted in the level of platelet aggregation induced with collagen, ristomycin, adrenaline being statistically lower than the level of induced platelet aggregation of the intact group ($p<0.05$). When inducing platelet aggregation with ADP against the background of applying resveratrol, the level was comparable to the level of induced aggregation of the group of intact animals.

The use of the studied drugs in conjunction with the drug that is included in the standards of treatment of hypertensive states in pregnant women led to increased positive effects. Upon induction of platelet aggregation with collagen, ristomycin and adrenaline in animals with the combined use of nicorandil and amlodipine, there was a statistically significant ($p<0.05$) decrease in the level of aggregation compared with that in monotherapy. Upon induction of platelet aggregation with adrenaline, aggregation reached a level comparable to the level of the group of intact animals. In the animals with a combined use of resveratrol and amlodipine, there was a statistically significant ($p<0.05$) decrease, compared with that in monotherapy, in the level of platelet aggregation induced with ristomycin. At the same time, the induction level of platelet aggregation induced by ristomycin reached a statistically significant difference from the level of the group of intact animals ($p<0.05$).

The use of the studied drugs together with a $K^+_{ATP}$ blocker channel glybenclamide led to partial cancellation of the positive effects (Fig. 3). Introduction of glibenclamide to animals with correction of ADMA-like preeclampsia with resveratrol and nicorandil led to a statistically significant increase in platelet aggregation by applying all the inducers used ($p<0.05$). At the same time, the level of induced platelet aggregation remained significantly higher than that in the group of control animals.

The study of plasma-coagulation hemostasis revealed patterns that are similar in nature to changes in the previ-

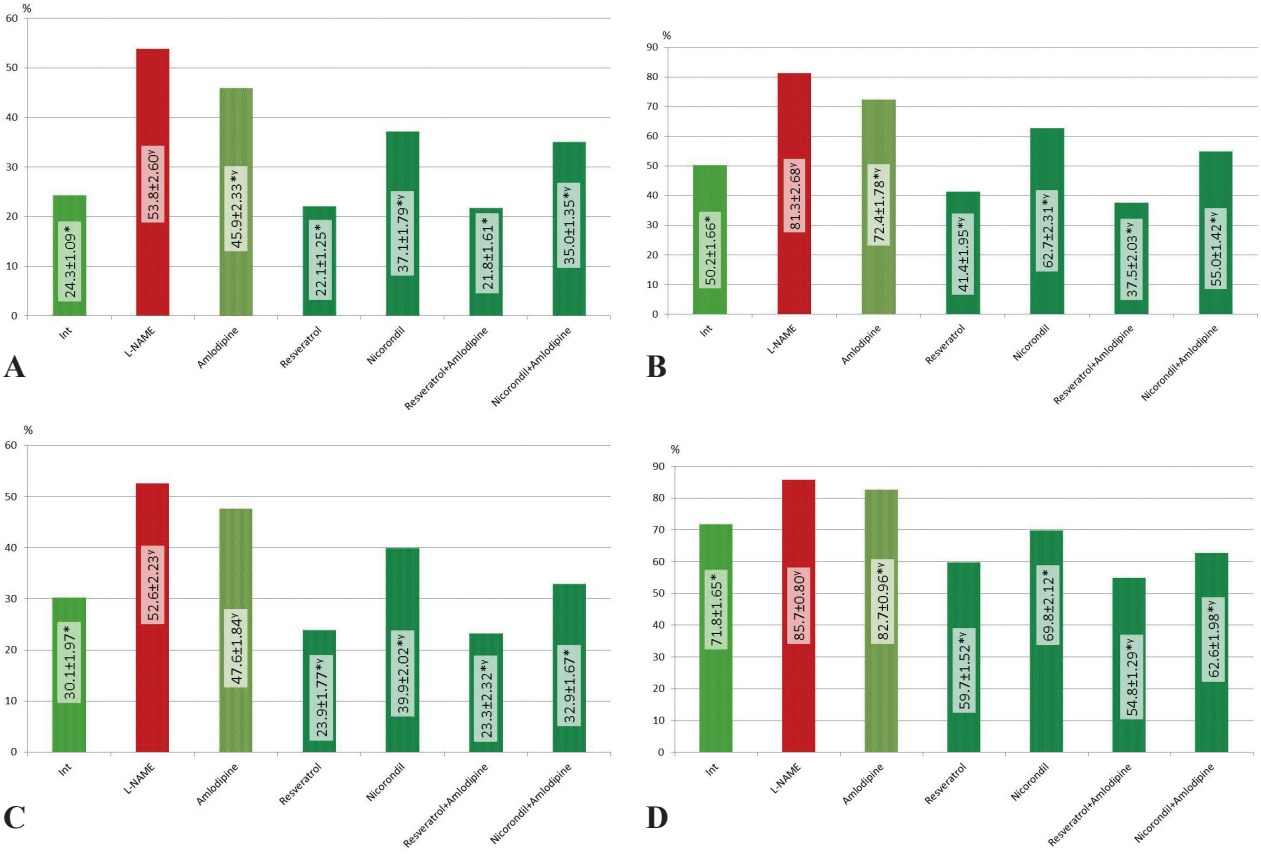


**Figure 2.** The effect of nicorandil and resveratrol on the time of induced platelet aggregation in modeling ADMA-like preeclampsia. *Note*: **A** (ADP), **B** (Collagen), **C** (Ristomycin), **D** (Adrenaline) used as inducers; * – p<0.05 in comparison with L-NAME animals; $^{\gamma}$ – p <0.05 in comparison with intact animals

ous experiments. When modeling ADMA-like preeclampsia, the hemostasis system shifted to hypercoagulation, which is evidenced by an increase in plasma fibrinogen and a decrease in TT, PTT, and aPTT (p<0.05) (Table 1).

The use of the studied pharmacological agents resulted in a statistically significant (p<0.05) correction of the parameters of plasma coagulation hemostasis, which was more pronounced than when using the reference drug amlodipine. In the groups using resveratrol and nicorandil, the level of time indicators reached that in the group of intact animals. The content of fibrinogen was at the intermediate level between the group of intact animals and the group of animals with ADMA-like preeclampsia and was statistically different from them (p<0.05).

The use of nicorandil and resveratrol together with the drug which is included in the standards of treatment of hypertensive states in pregnant women led to an enhancement of the corrective effects. Thus, in the group with the combined use of resveratrol and amlodipine, the time indicators reached a statistically higher level compared to those of the intact animals. In the group of animals with the combined use of nicorandil and amlodipine, aPTT exceeded the level of the intact animals. In both groups with the combined use of the studied pharmacological agents and amlodipine, the fibrinogen content reduced to the level of the intact animals.

When using the studied pharmacological agents with glibenclamide to elucidate the mechanisms for correcting the hemostasis system, mediated through the activation of $K^{+}_{ATP}$ channels, the data were obtained indicating a certain role of their activation in the positive effects from resveratrol and nicorandil. In the group of animals with correction of ADMA-like preeclampsia with resveratrol, glibenclamide statistically significantly increased the time characteristics of plasma-coagulation hemostasis and plasma fibrinogen content (p<0.05). In the group of animals with the correction of ADMA-like preeclampsia with nicorandil, glibenclamide statistically significantly increased PTT (p<0.05), and TT reached the level that was statistically indistinguishable from that of the group of the ”untreated” animals.

In the next series of experiments, an analysis was carried out of changes in the time of blood clot formation in the carotid artery, which in its essence was an integral sum of the states of the vascular-platelet and plasma-coagulation components of hemostasis, as well as of their interaction. Modeling ADMA-like preeclampsia resulted in a statistically significant (p<0.05) shortening of the time before cessation of blood flow in the carotid artery after applying $FeCl_3$ 1189±18 s to 841±42 s (Fig. 4).

The use of the studied pharmacological agents resulted in a statistically significant (p <0.05) lengthening of the

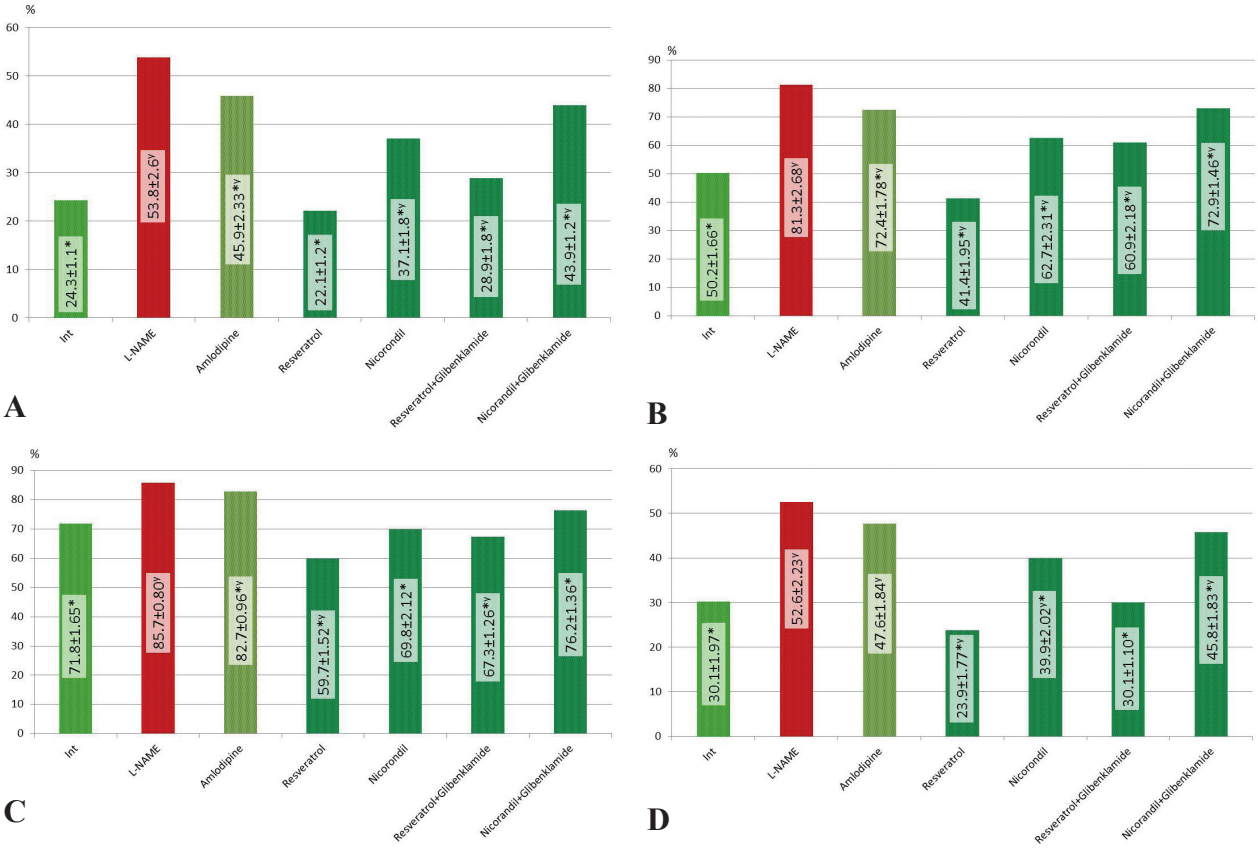


**Figure 3.** Effects of glybenclamide on the time of induced platelet aggregation with the correction of an ADMA-like preeclampsia with nicorandil and resveratrol. *Note*: **A** (ADP), **B** (Collagen), **C** (Ristomycin), **D** (Adrenaline) used as inducers; * – $p<0.05$ in comparison with L-NAME animals; ʸ – $p<0.05$ in comparison with intact animals

**Table 1.** Indicators of Plasma-coagulation hemostasis during correction of ADMA-like Preeclampsia with Nicorandil and Resveratrol (M ± m; N = 10)

| Group | TT, sec | PTT, sec | aPTT, sec | Fibrinogen, g/L |
|---|---|---|---|---|
| Intact | 24.3±1.6* | 22.7±1.6* | 17.8±0.8* | 2.34±0.16* |
| L-NAME | 15.3±1.6ʸ | 12.8±0.9ʸ | 10.3±0.6ʸ | 4.03±0.17ʸ |
| Amlodipine | 19.5±0.8*ʸ | 15.3±0.9ʸ | 11.0±0.6ʸ | 3.51±0.19*ʸ |
| Resveratrol | 26.8±1.5* | 27.0±1.7* | 19.2±0.9* | 2.98±0.13*ʸ |
| Nicorarandil | 20.8±1.4* | 20.0±1.0* | 15.1±0.9* | 3.23±0.08*ʸ |
| Resveratrol + amlodipine | 31.9±1.4*ʸ | 28.6±2.1*ʸ | 22.2±1.2*ʸ | 2.19±0.16* |
| Nicorandil + amlodipine | 25.6±1.3* | 25.8±1.3* | 21.2±1.0*ʸ | 2.70±0.17* |
| Resveratrol + glibenclamide | 20.7±1.7* | 21.0±1.8* | 15.5±1.2* | 3.39±0.12*ʸ |
| Nicorandil + glibenclamide | 17.1±1.3ʸ | 16.1±1.2*ʸ | 13.8±1.0*ʸ | 3.62±0.11*ʸ |

*Note*: * – $p<0.05$ in comparison with L-NAME animals; ʸ – $p<0.05$ in comparison with intact animals

thrombosis period to a level statistically indistinguishable from that in the group of intact animals with resveratrol administration. Their use in conjunction with the drug, included into the standard antihypertensive therapy in pregnant women, led to a greater lengthening of the time of thrombus formation with the greatest effect being observed in the group treated with nicorandil. In this group, the period before the cessation of blood flow in the carotid artery reached a level statistically indistinguishable from those in the intact group and in the groups treated with resveratrol.

As expected, based on the data from previous experiments, the use of glibenclamide shortened the time required to complete blood clot formation, but it remained to be statistically longer than the time of blood clot formation in groups of the "untreated" animals, which indicates an incomplete cancellation of the positive effects of the studied pharmacological agents.

Many aspects of the interaction of platelets and endotheliocytes in the pathogenesis of preeclampsia are still to be clarified, but joining them into a separate link within

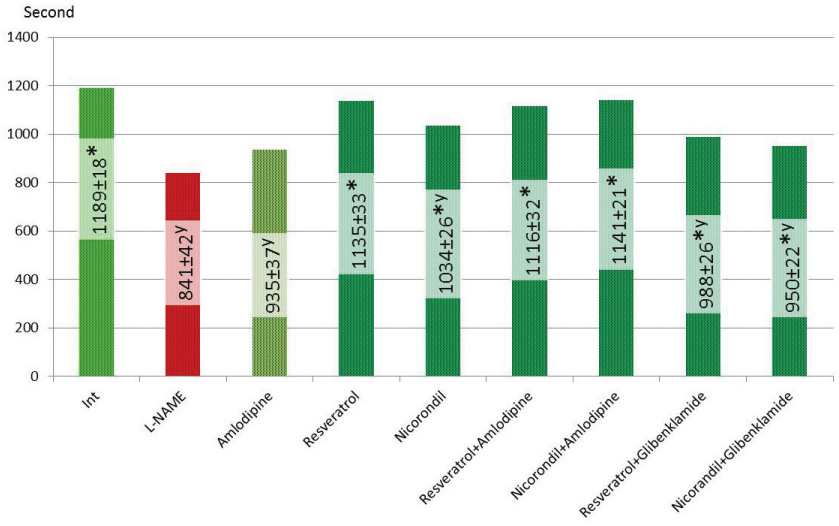


**Figure 4.** The time of formation of a blood clot in the carotid artery when correcting ADMA-like preeclampsia with nicorandil and resveratrol. *Note*: * – $p<0.05$ in comparison with L-NAME animals; $^{y}$ – $p<0.05$ in comparison with intact animals

the hemostasis emphasizes their close relationship. The triggering mechanism of the vascular-platelet hemostasis in case of damage to the vessel is the multi-stage activation of platelets when interacting with subendothelial layers. There is a change of the lipid composition of the surface layer of the membrane, an increased expression of receptors to adhesion molecules on the membrane surface, $Ca^{2+}$-dependent change in the thrombocyte shape; synthesis of thromboxane is initiated, as well as a release of humoral factors from platelet granules: ADP, serotonin, histamine, adrenalin, noradrenalin, dopamine, growth factors, factors regulating vascular wall permeability and chemotaxis of neutrophils and eosinophils and others promoting vasoconstriction, adhesion and aggregation of platelets (Kozlovsky et al. 2013, Lysenkov et al. 2004). At this stage, the reversible stage of platelet aggregation ends. The response of the endothelium is reduced formation of NO, prostacyclin and enhanced synthesis of endothelin-1. As for functioning, a vasospasm is observed, which facilitates the adhesion of platelets.

Under natural conditions, when a vessel is damaged, activated platelets adhere on the subendothelial surface, in particular, collagen, and irreversible platelet aggregation occurs. Thus, activated platelets are used locally and hardly enter the systemic circulation.

With the development of preeclampsia, events occur in different conditions. It should be noted that platelet activation can be caused not only by proteins of subendothelial structures, but also by adrenaline, norepinephrine, ADP, immune complexes, peroxide radicals, hypoxia, arterial hypertension, prostaglandins PgG2 and PgH2, arachidonic acid, thromboxane $A_2$, platelet activating factor (PAF), phorbol esters, latex, lectins, A23187 ionophore, bacteria and bacterial lipopolysaccharide, tumor cells, increased shear stress, etc. (Blomqvist et al. 2018, Kozlovsky et al. 2013, Lysenkov et al. 2004, Sidorenko et al. 2007). It is ischemic placenta that excretes a large number of these humoral factors, which leads to platelet activation. Depending on the inducers and their combination, the activation of platelets, as well as the humoral factors secreted by them and their ratios are of a different nature. Unlike natural conditions, there is no exposed subendothelial layer, nor is there platelet adhesion at the site of activation. The mass of activated platelets goes into the systemic circulation, and it can be said that there is a kind of endocrine gland, located throughout the circulating blood and directly contacting with endothelium. Therefore, even if the action of each individual platelet is considered locally, their mass location determines the generalization of their effects. When passing through the ischemic placenta, more and more new platelets are activated. When a critical number is reached, the level of humoral factors secreted by platelets becomes sufficient to cause systemic endotheliosis. There is even a parallel between the DIC-syndrome, systemic inflammatory response syndrome and preeclampsia (Lysenkov et al. 2004).

One of the diagnostic criteria for the severity of preeclampsia is the sFlt-1/PlGF ratio (Baranovskaya 2018, Ivanets et al. 2012, Ivanets et al. 2013, Ivanets et al. 2015). sFlt-1 is a variant of the vascular endothelial growth factor receptor (VEGFR-1). The higher this ratio, the higher the risk of preeclampsia or its severity. The source of sFlt-1 is not only an ischemic placenta, but also a platelet-monocine complex, which is obtained as a result of aggregation of platelet-activated monocyte (Major et al. 2014).

During pregnancy, platelet sensitivity to adrenaline, collagen, ristomycin, and other factors increases. This accurately determines polyetiology and another point of application of provoking factors in the pathogenesis of preeclampsia in such conditions as: infections, psychoemotional stress, conditions accompanied by excessive formation of peroxide radicals, metabolic syndrome, con-

ditions accompanied by an increased content of ADMA and homocysteine, etc. (Karamysheva 2014, Lysenkov et al. 2004, Rudzevich 2011).

Thus, platelet activation is an intermediate pathogenetic link between the immaturity of the spiral arteries, ischemia of the placenta and systemic endothelial dysfunction. The chain of events described above looks logical in other pathological conditions as well, such as: sepsis, atherosclerosis, arterial hypertension, metabolic syndrome, varicose veins, etc. Summarizing the analyzed data, it can be said with a certain degree of accuracy that vascular-coagulation-immunological disorders in pregnant women have a common pathogenetic interaction in various diseases and, depending on the provoking factors and individual characteristics, determine the development of DIC-syndrome, preeclampsia or eclampsia.

An increase in platelet aggregation capacity and activation of the plasma-coagulation component of hemostasis in modeling ADMA-like preeclampsia repeat the patterns observed in the clinic in women with preeclampsia. L-NAME is an analogue of ADMA, which, in turn, is an eNOS inhibitor. Therefore, the introduction of L-NAME results in a reduced activity of eNOS ,, as well as a reduced formation of potent antiaggregants – NO – by endothelium. At this stage, the most sensitive are the placental vessels. The resulting endothelial dysfunction leads to ischemic events in it. When passing through the placenta, platelets get activated due to ischemia, as well as due to humoral factors released in response. Activated platelets not only have an increased aggregation capacity by themselves, but also activate intact platelets, secreting aggregation inducers into the bloodstream. Modeling ADMA-like preeclampsia leads to a pronounced persistent increase in blood pressure. There is a change in shear rate, which can also activate platelets. According to the literature, in women with preeclampsia, on the surface of the platelet, the expression of receptors to different aggregation inducers increases compared with that in normal pregnancy. This contributes to increased sensitivity of platelets in them. No concrete literature data on the number of receptors for aggregation inducers in animals with ADMA-like preeclampsia have been found, but taking into account the comparable pathogenetic mechanisms of both pathological conditions, the probability of this mechanism is rather high.

The time of the blood clot formation until the complete cessation of blood flow in the artery is an integral sum of vascular-platelet and plasma-coagulation components of hemostasis. Therefore, its shortening is a logical consequence of a shift of hemostasis towards hypercoagulation.

The pronounced positive effects of resveratrol in the correction of platelet aggregation disorders caused by the ADMA-like preeclampsia model are accounted for by several direct and indirect mechanisms. The literature describes various positive effects of resveratrol: improvement of mitochondrial respiration, improvement of metabolic processes in obesity and obesity-associated diseases, anti-inflammatory effect, suppression of the growth of cancer cells, numerous positive effects in diseases of the cardiovascular system (Bitterman and Chung 2015).

As applied to the topic touched upon in this paper, the following studies deserve special attention. It was found that resveratrol inhibits PDE (Takizawa et al. 2015), including PDE1 and PDE4 (Wang et al. 2016, Rauf et al. 2017). These features explain the neuroprotective properties of resveratrol in ischemia (Wan et al. 2016). Inhibition of PDE3 explains the enhancement of myocardial contractility without an arrhythmogenic effect. Reducing the expression of mRNA of most of the 11 PDE isoenzymes, including PDE3B, PDE8A and PDE10A, which are associated with the regulation of insulin secretion, resveratrol is capable of improving the function of β-cells of the pancreas in a dose-dependent way (Rouse et al. 2014). Resveratrol has a positive effect in erectile dysfunction in patients tolerant to PDE5 inhibitors. This effect is independent of eNOS mechanisms, is associated with the formation of hydrogen sulfide, and can also be endothelium-mediated (Dalaklioglu and Ozbey 2013, Yetik-Anacak et al. 2015). When resveratrol and sildenafil are used in animals to correct erectile dysfunction caused by streptozotocin diabetes, synergism is observed due to the effect on the NO-mediated signaling pathway (Bai and An 2015). Antiplatelet properties of resveratrol are described in (Bonechi et al. 2017, Sun et al. 2018). Resveratrol reduces the risk of thrombosis in experimental liver cirrhosis, normalizing platelet function (Xu et al. 2016). Resveratrol has pronounced positive effects in preeclampsia in humans and in experiment (Tenório et al. 2018, Bariani et al. 2017, Ivanets et al. 2015). The ability of resveratrol under experimental conditions to reduce inflammation induced by lipopolysaccharides, and to prevent preterm labor was found in (Bariani et al. 2017).

When $K^+_{ATP}$ channels are activated by resveratrol, cytoprotective mechanisms of pharmacological preconditioning are triggered. In addition, activation of $K^+_{ATP}$ channels leads to a decrease in myometrial tone and improvement of microcirculation in the tissues of the placenta. Triggering pharmacological preconditioning and improving the microcirculation in the placenta lead to a decrease in the number of ischemic events in it, and consequently, the activation of platelets passing through it decreases. The fact that resveratrol has endothelium protective properties encourages the normalization of endothelium function and leads to the restoration of NO synthesis, lower blood pressure and also contributes to the improvement of microcirculation in the placenta with all logical consequences. Due to its direct action, resveratrol can inhibit PDE3 and PDE5 in the platelet (Kleppe et al. 2018, Rauf et al. 2017, Takizawa et al. 2015, Wang et al. 2016). This leads to the accumulation of cyclic monophosphates inside the platelet, prevents the release of calcium from the depot and prevents the activation of the platelet or puts it in an inactive state.

The pronounced positive effects of nicoradil in the correction of platelet aggregation disorders caused by an ADMA-like model of preeclampsia are also associated with direct and mediated mechanisms. The literature also describes the positive effects of nicorandil in preg-

nant women (Rezk et al. 2015), in particular, its ability to reduce platelet aggregation, including not through the $Ca^{2+}$-dependent mechanism (Ivanova et al 1993).

By activating the $K^+_{ATP}$-channels, nicorandil, like resveratrol, reduces ischemic effects in the placenta, triggering pharmacological preconditioning mechanisms and improving microcirculation in it. Endothelioprotective effects of nicorandil help reduce the factors that bring platelets into an activated form, reducing blood pressure and restoring the NO-synthesizing function of the endothelium similar to preeclampsia in ADMA. The direct mode of action of nicorandil shows in its ability to be an NO-donator, which in itself is a potnet factor preventing platelet aggregation, as well as to reduce platelet aggregation, including not through a $Ca^{2+}$-dependent mechanism (Ivanova et al. 1993).

The enhancement of the positive effects of resveratrol and nicorandil by means of a drug that is included in the standards of treatment of hypertensive states in pregnant women, amlodipine, is explained by its ability to enhance the positive effects of the studied pharmacological agents in correcting morphofunctional disorders in animals with ADMA-like preeclampsia and, thus, to a greater extent, lead to a decrease in the number of factors promoting the transition of platelets into the activated form.

Lengthening the time of thrombosis when using the studied pharmacological agents, as well as their combined use with the drug which is included in the standards of treatment of hypertensive states in pregnant women, is a logical consequence of the protective effects and reflects the patterns of changes in vascular-platelet and plasma-coagulation components of hemostasis.

The incomplete cancellation of the positive effects of resveratrol and nicorandil when they are used against the background of a $K^+_{ATP}$-channel blocker, glibenclamide, indicates their significant role in realizing the positive effects of the studied pharmacological agents. On the other hand, this indicates that resveratrol and nicorandil have a complex mode of action, in which of great importance are the mechanisms that are not mediated by $K^+_{ATP}$-channels. This is also proved by the dynamics of the time of blood clot formation until the complete cessation of blood flow in the arteries.

## Conclusion

When modeling ADMA-like preeclampsia, there is an increase in platelet aggregation from about 19% to 121% when the inducers used are ADP, collagen, ristomycin and adrenaline, as well as a shift in the plasma-coagulation component of hemostasis towards hypercoagulation. The time of blood clot formation was reduced to 70% of the original. The use of resveratrol and nicorandil resulted in a pronounced correction of the disturbance of induced platelet aggregation, indicators of the plasma-coagulation component of hemostasis and the time of thrombus formation. A significant part of the positive effects of resveratrol and nicorandil in the correction of hemostatic disorders in animals with ADMA-like preeclampsia are mediated by $K^+_{ATP}$-channels.

## Conflict of interests

The authors state no conflict of interest concerning with the present submitted manuscript.

## References


- Bai Y, An R (2015) Resveratrol and sildenafil synergistically improve diabetes-associated erectile dysfunction in streptozotocin-induced diabetic rats. Life Sciences 135: 43–48. https://doi.org/10.1016/j.lfs.2015.04.020 [PubMed]
- Baranovskaya EI (2018) Preeclampsia in modern conditions. Obstetrics and Gynecology [Akusherstvo i ginekologiya] (11): 5–9. [in Russian] https://doi.org/10.18565/aig.2018.11.5-9
- Bariani MV, Correa F, Leishman E, Domínguez Rubio AP, Arias A, Stern A, Bradshaw HB, Franchi AM (2017) Resveratrol protects from lipopolysaccharide-induced inflammation in the uterus and prevents experimental preterm birth. Molecular Human Reproduction: Basic Science of Reproductive Medicine 23(8): 571–581. https://doi.org/10.1093/molehr/gax036 [PubMed] [PMC]
- Bitterman JL, Chung JH (2015) Metabolic effects of resveratrol: addressing the controversies. Cellular and Molecular Life Sciences 72(8): 1473–1488. https://doi.org/10.1007/s00018-014-1808-8 [PubMed] [PMC]
- Blomqvist LRF, Strandell AM, Baghaei F, Hellgren MSE (2018) Platelet aggregation in healthy women during normal pregnancy – a longitudinal study. Platelets 16: 1–7. https://doi.org/10.1080/09537104.2018.1492106 [PubMed]
- Bonechi C, Lamponi S, Donati A, Tamasi G, Consumi M, Leone G, Rossi C, Magnani A (2017) Effect of resveratrol on platelet aggregation by fibrinogen protection. Biophysical Chemistry 222: 41–48. https://doi.org/10.1016/j.bpc.2016.12.004 [PubMed]
- Burke N, Flood K, Muellers S, Murray A, Dunne E, Cotter B, Dempsey M, Dicker P, Geary MP, Kenny D, Malone FD (2016) Reduced spontaneous platelet aggregation: a novel risk factor for adverse pregnancy outcome. European Journal of Obstetrics & Gynecology and Reproductive Biology 199: 132–136. https://doi.org/10.1016/j.ejogrb.2016.02.010 [PubMed]
- Burke N, Flood K, Murray A, Cotter B, Dempsey M, Fay L, Dicker P, Geary MP, Kenny D, Malone FD (2013) Platelet reactivity changes significantly throughout all trimesters of pregnancy compared with the nonpregnant state: a prospective study. BJOG: An International Journal of Obstetrics & Gynaecology 120(13): 1599–1604. https://doi.org/10.1111/1471-0528.12394 [PubMed]
- Dalaklioglu S, Ozbey G (2013) The potent relaxant effect of resveratrol in rat corpus cavernosum and its underlying mechanisms. International Journal of Impotence Research 25(5): 183–193. https://doi.org/10.1038/ijir.2013.6 [PubMed]

- Ducray JF, Naicker T, Moodley J (2011) Pilot study of comparative placental morphometry in pre-eclamptic and normotensive pregnancies suggests possible maladaptations of the fetal component of the placenta. European Journal of Obstetrics & Gynecology and Reproductive Biology 156(1): 29–34. https://doi.org/10.1016/j.ejogrb.2010.12.038 [PubMed]
- Freitas LG, Sathler-Avelar R, Vitelli-Avelar DM, Bela SR, Teixeira-Carvalho A, Carvalho Md, Martins-Filho OA, Dusse LM (2014) Preeclampsia: integrated network model of platelet biomarkers interaction as a tool to evaluate the hemostatic/immunological interface. Clinica Chimica Acta 436: 193–201. https://doi.org/10.1016/j.cca.2014.05.020 [PubMed]
- Glants S (1999) Biomedical statistics. Practice Publishers, Moscow, 459 pp. [in Russian]
- GOST 33044-2014 (2015) Principles of Good Laboratory Practice. Standartinform, Moscow, 12 pp.
- GOST 33217-2014 (2016) Guidelines for the Maintenance and Care of Laboratory Animals. Rules for the Maintenance and Care of Laboratory Predatory Mammals. Standartinform, Moscow, 12 pp.
- Gureev VV, Pokrovsky MV, Korokin MV (2014) ADMA – eNOS – deterministic ways of pharmacological correction of preeclampsia. The Publishing House "Belgorod" of Belgorod State National Research University, Belgorod, 260 pp. [in Russian]
- Hayashi M, Kiumi F, Mitsuya K (1999) Changes in platelet ATP secretion and aggregation during pregnancy and in preeclampsia. The American Journal of the Medical Sciences 318(2): 115–121. https://doi.org/10.1016/S0002-9629(15)40593-2 [PubMed]
- Hellgren M (2003) Hemostasis during normal pregnancy and puerperium. Seminars in Thrombosis and Hemostasis 29(2): 125–130. https://doi.org/10.1055/s-2003-38897 [PubMed]
- Holthe MR, Staff AC, Berge LN, Lyberg T (2004) Different levels of platelet activation in preeclamptic, normotensive pregnant, and nonpregnant women. American Journal of Obstetrics and Gynecology 190(4): 1128–1134. https://doi.org/10.1016/j.ajog.2003.10.699 [PubMed]
- Ivanets TJ, Alekseeva ML, Goncharova EA, Khodzhaeva ZS, Vavina OV (2012) Markers of preeclampsia in I and II trimesters of pregnancy. Problems of Reproduction [Problemy reproduktsii] (3): 83–87. [in Russian]
- Ivanets TJ, Alekseeva ML, Kan NE, Tyutyunnik VL, Amiraslanov EY, Nasonova DM, Fanchenko ND (2015) Diagnostic value of determination of placental growth factor and soluble fms-like tyrosine kinase-1 as markers of pre-eclampsia. Problems of Reproduction [Problemy reproduktsii] (4): 129–133. [in Russian] https://doi.org/10.17116/repro2015214129-133
- Ivanets TJ, Alekseeva ML, Loginov NS, Kolodko VG, Nasonova DM, Khasyanova ZS (2013) Placental growth factor and fmf-like tyrosine kinase-1 as markers of preeclampsia in pregnancy dynamics. Clinical Laboratory Diagnostics [Klinicheskaya laboratornaya diagnostika] (8): 14–17. [in Russian]
- Ivanova K, Schaefer M, Drummer C, Gerzer R (1993) Effects of nitric oxide-containing compounds on increases in cytosolic ionized $Ca^{2+}$ and on aggregation of human platelets. European Journal of Pharmacology: Molecular Pharmacology. 244(1): 37–47. https://doi.org/10.1016/0922-4106(93)90057-G [PubMed]
- Käehne LV, Lundin ICR (2017) Treatment with low-dose acetylsalicylic acid can reduce risk of pre-eclampsia in high-risk pregnant women. Ugeskrift for Laeger 179 (14). [PubMed]
- Karamysheva VI (2014) Influence of GABA derivatives on blood flow in uteroplacental complex in conditions of normal and experimental preeclampsia. PhD thesis, Volgograd, Russia: Volgograd State Medical University. [in Russian]
- Kazmi RS, Cooper AJ, Lwaleed BA (2011) Platelet function in pre-eclampsia. Seminars in thrombosis and hemostasis 37(2): 311–316. https://doi.org/10.1055/s-0030-1270339 [PubMed]
- Kleppe R, Jonassen I, Døskeland SO, Selheim F (2018) Mathematical Modelling of Nitric Oxide/Cyclic GMP/Cyclic AMP Signalling in Platelets. International Journal of Molecular Sciences 19(2). ttps://doi.org/10.3390/ijms19020612 [PubMed] [PMC]
- Kochkarov VI (2009) Resveratrol and its combinations with the main antihypertensive drugs in pharmacological correction of endothelial dysfunction. Doctoral thesis, Smolensk, Russia: Smolensk State Medical University. [in Russian]
- Kozlovsky VI, Kovtun OM, Seroukhova OP, Detkovskaya IN, Kozlovsky IV (2013) Research methods and clinical significance of platelet aggregation. Focus on spontaneous aggregation. Bulletin of Vitebsk State Medical University [Vestnik Vitebskogo gosudarstvennogo medtsinskogo universiteta] 12(4): 79–91. [in Russian]
- Lysenkov SP, Myasnikova VV, Ponomarev VV (2004) Emergency conditions and anesthesia in obstetrics. Clinical pathophysiology and pharmacotherapy. ELBY-SPb Publishers, Saint-Petersburg, 600 pp. [in Russian]
- Major HD, Campbell RA, Silver RM, Branch DW, Weyrich AS (2014) Synthesis of sFlt-1 by platelet-monocyte aggregates contributes to the pathogenesis of preeclampsia. American Journal of Obstetrics and Gynecology 210: 547. https://doi.org/10.1016/j.ajog.2014.01.024 [PubMed] [PMC]
- Ministry of Healthcare of the Russian Federation (2016) Methodical letter of the Ministry of Healthcare of the Russian Federation of 14.12.2016 No. 15-4/10/2-7955 "Results of confidential audit of maternal mortality in the Russian Federation in 2015", Moscow, 61 pp. [in Russian]
- Mironov AN (2012) Guidelines for conducting preclinical studies of new drugs. Grif and Co. Publishers, Moscow, 944 pp. [in Russian]
- Mone F, Mulcahy C, McParland P, McAuliffe FM (2017) Should we recommend universal aspirin for all pregnant women? American Journal of Obstetrics and Gynecology 216(2): 141. https://doi.org/10.1016/j.ajog.2016.09.086 [PubMed]
- Novaković R, Radunović N, Marković-Lipkovski J, Ćirović S, Beleslin-Čokić B, Ilić B, Ivković B, Heinle H, Živanović V, Gojković-Bukarica LJ (2015) Effects of the polyphenol resveratrol on contractility of human term pregnant myometrium. Molecular Human Reproduction: Basic science of reproductive medicine 21(6): 545–551. https://doi.org/10.1016/j.ajog.2016.09.086 [PubMed]
- Order of the Ministry of Healthcare of the Russia 199N (2016) On Approval of the Rules of Good Laboratory Practice. Moscow, 8 pp.
- Pereira RD, De Long NE, Wang RC, Yazdi FT, Holloway AC, Raha S (2015) Angiogenesis in the placenta: The role of reactive oxygen species signaling. Biomed Research International 2015: 814543, 12 pages. http://dx.doi.org/10.1155/2015/814543 [PubMed] [PMC]
- Rauf A, Orhan IE, Ertas A, Temel H, Hadda TB, Saleem M, Raza M, Khan H (2017) Elucidation of phosphodiesterase-1 inhibitory effect of some selected natural polyphenolics using in vitro and in silico methods. Current Topics in Medicinal Chemistry 17(4): 412–417. https://doi.org/10.2174/1568026616666160824103615 [PubMed]
- Rezk M, Sayyed T, Masood A, Dawood R (2015) Nicorandil vs nifedipine for the treatment of preterm labour: a randomized clinical trial. European Journal of Obstetrics & Gynecology and Reproductive Biology 195: 27–30. https://doi.org/10.1016/j.ejogrb.2015.09.038 [PubMed]

- Reznikova LB (2013) Endothelioprotective activity of GABA derivatives with experimental gestosis. PhD thesis, Volgograd, Russia: Volgograd State Medical University. [in Russian]
- Rouse M, Younès A, Egan JM (2014) Resveratrol and curcumin enhance pancreatic β-cell function by inhibiting phosphodiesteraseactivity. The Journal of Endocrinology 223(2): 107–117. http://dx.doi.org/10.1530/JOE-14-0335 [PubMed] [PMC]
- Rudzevich AY (2011) Violations of the processes of aggregation and release reaction of platelets, continuous intravascular coagulation and their correction with antioxidants in arterial hypertension in pregnant women. Doctoral thesis, Tyumen, Russia: Tyumen State Medical Academy. [in Russian]
- Sahin S, Ozakpinar OB, Eroglu M, Tulunay A, Ciraci E, Uras F, Tetik S (2015) The impact of platelet functions and inflammatory status on the severity of preeclampsia. The Journal of Maternal-Fetal & Neonatal Medicine 28(6): 643–648. https://doi.org/10.3109/14767058.2014.927860 [PubMed]
- Sánchez-Aranguren LC, Prada CE, Riaño-Medina CE, Lopez M (2014) Endothelial dysfunction and preeclampsia: role of oxidative stress. Frontiers in Physiology 5: 372–405. https://doi.org/10.3389/fphys.2014.00372 [PubMed] [PMC]
- Sarma A, Scott NS (2016) Aspirin use in women: current perspectives and future directions. Current Atherosclerosis Reports 18(12): 74. https://doi.org/10.1007/s11883-016-0630-1 [PubMed]
- Scioscia M, Karumanchi SA, Goldman-Wohl D, Robillard PY (2015) Endothelial dysfunction and metabolic syndrome in preeclampsia: an alternative viewpoint. Journal of Reproductive Immunology 108: 42–47. https://doi.org/10.1016/j.jri.2015.01.009 [PubMed]
- Shuvalova MP, Frolova OG, Ratushnyak SS (2014) Preeclampsia and eclampsia as the cause of maternal mortality. Obstetrics and Gynecology [Akusherstvo i ginekologiya] (1): 81–87. [in Russian]
- Sidorenko EV (2003) Methods of mathematical processing in psychology. Speech Publishers, Saint-Petersburg, 350 pp. [in Russian]
- Sidorenko VN, Mazhul V M, Chernovets TS (2007) Molecular-membrane mechanisms of dysfunction of activity and protease-induced platelet aggregation in pregnancy complicated by gestosis. Journal of Grodno State Medical University [Zhurnal Grodnenskogo gosudarstvennogo meditsinskogo universiteta] 2: 35–38. [in Russian]
- Stupakova EG, Lazareva GA, Gureev VV (2018) Correction of morphofunctional disturbances arising when modelling Preeclampsia with resveratrol and nicorandil. Research Results in Pharmacology 4(1): 59–71. https://doi.org/10.3897/rrpharmacology.4.25528
- Sun J, Zhang M, Chen K, Chen B, Zhao Y, Gong H, Zhao X, Qi R (2018) Suppression of TLR4 activation by resveratrol is associated with STAT3 and Akt inhibition in oxidized low-density lipoprotein-activated platelets. European Journal of Pharmacology 836: 1–10. https://doi.org/10.1016/j.ejphar.2018.08.014 [PubMed]
- Takizawa Y, Nakata R, Fukuhara K, Yamashita H, Kubodera H, Inoue H (2015) The 4'-hydroxyl group of resveratrol is functionally important for direct activation of PPARα. PLoS One 10(3): e0120865. https://doi.org/10.1371/journal.pone.0120865 [PubMed] [PMC]
- Tanaka K, Kato K, Takano T, Katagiri T, Asanoi H, Nejima J, Nakashima M, Kamijo T, Sakanashi M (2010) Acute effects of intravenous nicorandil on hemodynamics in patients hospitalized with acute decompensated heart failure. Journal of Cardiology 56(3): 291–299. https://doi.org/10.1016/j.jjcc.2010.06.009 [PubMed]
- Tenório MB, Ferreira RC, Moura FA, Bueno NB, Goulart MOF, Oliveira ACM (2018) Oral antioxidant therapy for prevention and treatment of preeclampsia: Meta-analysis of randomized controlled trials. Nutrition, Metabolism and Cardiovascular Diseases 28(9): 865–876. https://doi.org/10.1016/j.numecd.2018.06.002 [PubMed]
- European Treaty Series (1986) No. 123. The European Convention for the Protection of Vertebral Animals Used for Experience and Other Scientific Purposes. Strasbourg, 11 pp.
- Tyurenkov IN, Ivanova LB, Karamysheva VI, Lebedeva SA (2012) Correction of hemostatic disorders in females with experimental gestosis using GABA derivatives. Journal of Obstetrics and Women's Diseases [Zhurnal akusherstva i zhenskikh bolezney] 61(4): 119–123. [in Russian] https://doi.org/10.17816/JOWD614119-123
- Tyurenkov IN, Perfilova VN, Karamysheva VI, Reznikova LB, Mokrousov IS, Mikhailova LI, Berestovitskaya VM, Vasil'eva OS (2014) Effect of GABA derivatives on the rate of thrombus formation, platelet aggregation, and plasma coagulation capacity in rats with experimental gestosis. Bulletin of Experimental Biology and Medicine 158(2): 219–221. https://doi.org/10.1007/s10517-014-2726-3 [PubMed]
- van Oppenraaij RH, Bergen NE, Duvekot JJ, Krijger de RR, Ir WH, Steegers EAP, Exalto N (2011) Placental vascularization in early onset small for gestational age and preeclampsia. Reprodctive Sciences 18(6): 586–593. https://doi.org/10.1177/1933719110396231 [PubMed]
- van Vliet EO, Askie LA, Mol BW, Oudijk MA (2017) Antiplatelet agents and the prevention of spontaneous preterm birth: a systematic review and meta-analysis. Obstetrics and Gynecology 129(2): 327–336. https://doi.org/10.1097/AOG.0000000000001848 [PubMed]
- Verlohren S, Geusens N, Morton J, Verhaegen I, Hering L, Herse F, Dudenhausen JW, Muller DN, Luft FC, Cartwright JE, Davidge ST, Pijnenborg R, Dechend R (2010) Inhibition of trophoblast-induced spiral artery remodeling reduces placental perfusion in rat pregnancy. Hypertension 56(2): 304–310. https://doi.org/10.1161/Hypertensionaha.110.153163 [PubMed]
- Voronkov AV (2011) Endothelial dysfunction and ways of its pharmacological correction. Doctoral thesis, Volgograd, Russia: Volgograd State Medical University. [in Russian]
- Wan D, Zhou Y, Wang K, Hou Y, Hou R, Ye X (2016) Resveratrol provides neuroprotection by inhibiting phosphodiesterases and regulating the cAMP/AMPK/SIRT1 pathway after stroke in rats. Brain Research Bulletin 121: 255–262. https://doi.org/10.1016/j.brainresbull.2016.02.011 [PubMed]
- Wang G, Chen L, Pan X, Chen J, Wang L, Wang W, Cheng R, Wu F, Feng X, Yu Y, Zhang HT, O'Donnell JM, Xu Y (2016) The effect of resveratrol on beta amyloid-induced memory impairment involves inhibition of phosphodiesterase-4 related signaling. Oncotarget 7(14): 17380. https://doi.org/10.18632/oncotarget.8041 [PubMed] [PMC]
- Wang QJ, Song BF, Zhang YH, Ma YY, Shao QQ, Liu J, Qu X (2015) Expression of RGC32 in human cells and migration. Placenta 36(4): 350–356. https://doi.org/10.1016/j.placenta.2014.12.012 [PubMed]
- Xu M, Xue W, Ma Z, Bai J, Wu S (2016) Resveratrol reduces the incidence of portal vein system thrombosis after splenectomy in a rat fibrosis model. Oxidative Medicine and Cellular Longevity. http://dx.doi.org/10.1155/2016/7453849 [PubMed]
- Yetik-Anacak G, Dereli MV, Sevin G, Ozzayım O, Erac Y, Ahmed A (2015) Resveratrol stimulates hydrogen sulfide (H2 S) formation to relax murine corpus cavernosum. The Journal of Sexual Medicine 12(10): 2004–2012. https://doi.org/10.1111/jsm.12993 [PubMed]

## Author contributions


- **Elena G. Stupakova**, Assistant, Department of Obstetrics and Gynecology, Faculty of Postgraduate Education, e-mail: LazarevaGA@kursksmu.net, **ORCID:** 0000-0002-4591-0417. The author had a leading role in planning and performing the experiment, analyzing the data and literature and writing the article.

- **Galina A. Lazareva**, Doctor of Medical Sciences, Professor, Head of the Department of Obstetrics and Gynaecology, Faculty of Postgraduate Education, e-mail: akush.fpo@gmail.com, **ORCID:** 0000-0002-1225-8039. The author took part in planning experiments, analyzed the literature and participated in interpreting the data.

- **Vladimir V. Gureev**, Doctor of Medical Sciences, Professor of the Department of Pharmacology and Clinical Pharmacology, e-mail: produmen@yandex.ru, **ORCID:** 0000-0003-1433-1225. The author participated in planning the experiments, analyzed the literature and participated in interpreting the data.

- **Irina N. Dolzhikova**, PhD of Biological Sciences, Associate Professor of the Department of Pharmacology and Clinical Pharmacology, e-mail: ihcdaa@mail.ru, **ORCID:** 0000-0001-9534-2959. The author participated in planning the experiments, analysedthe literature and participated in interpreting the data.

- **Lyudmila A. Zhilinkova**, PhD in Technical Sciences, Associate Professor, Department of Philosophy, Social, Legal and Natural Sciences, e-mail: l_zhilinkova@mail.ru**, ORCID:** 0000-0003-0443-7130. The author took part in the development of the article design and in the statistical processing of the research results.

- **Anastasia Gureeva**, 2nd-year student, Medical Faculty, e-mail: nastasyi.207@gmail.com, **ORCID:** 0000-0003-1719-7316. The author had a leading role in planning and performing the experiment, analyzing the data and literature and writing the article.