

Pharmacological correction of endothelial dysfunction using a complex of grape polyphenols

Petr R. Lebedev^{ORCID}, Tatyana V. Avtina^{ORCID}, Vladimir V. Gureev^{ORCID}, Mikhail V. Pokrovskii^{ORCID}

Belgorod State National Research University; 85 Pobedy St., Belgorod 308015 Russia

Corresponding author: Petr R. Lebedev (artkelt98@yandex.ru)

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Abstract

Introduction: Endothelial dysfunction is an important component of cardiovascular disease pathogenesis. Natural polyphenols are of interest as potential endothelioprotective agents. **The aim of this study** was to evaluate the endothelioprotective activity of a grape polyphenol complex in a mouse model of endothelial dysfunction.

Materials and Methods: The study was performed on 50 male C57BL/6 mice. Endothelial dysfunction was induced by administration of **L-NAME** in drinking water (0.5 mg/mL) for 4 weeks. The animals were divided into five groups (n = 10): intact, control(**L-NAME**), **L-NAME** + **resveratrol**, **L-NAME** + **atorvastatin**, and **L-NAME** + grape polyphenol complex. The estimated daily intake per animal was 0.812 mg of **resveratrol**, 0.052 mg of **atorvastatin**, and 0.08 mL of the grape polyphenol complex. Systolic and diastolic blood pressure and endothelium-dependent vascular responses were assessed by invasive hemodynamic monitoring. Morphometric analysis of the heart and kidneys was performed. Edn1, Nos3, and Vegfa expression was evaluated by real-time quantitative PCR.

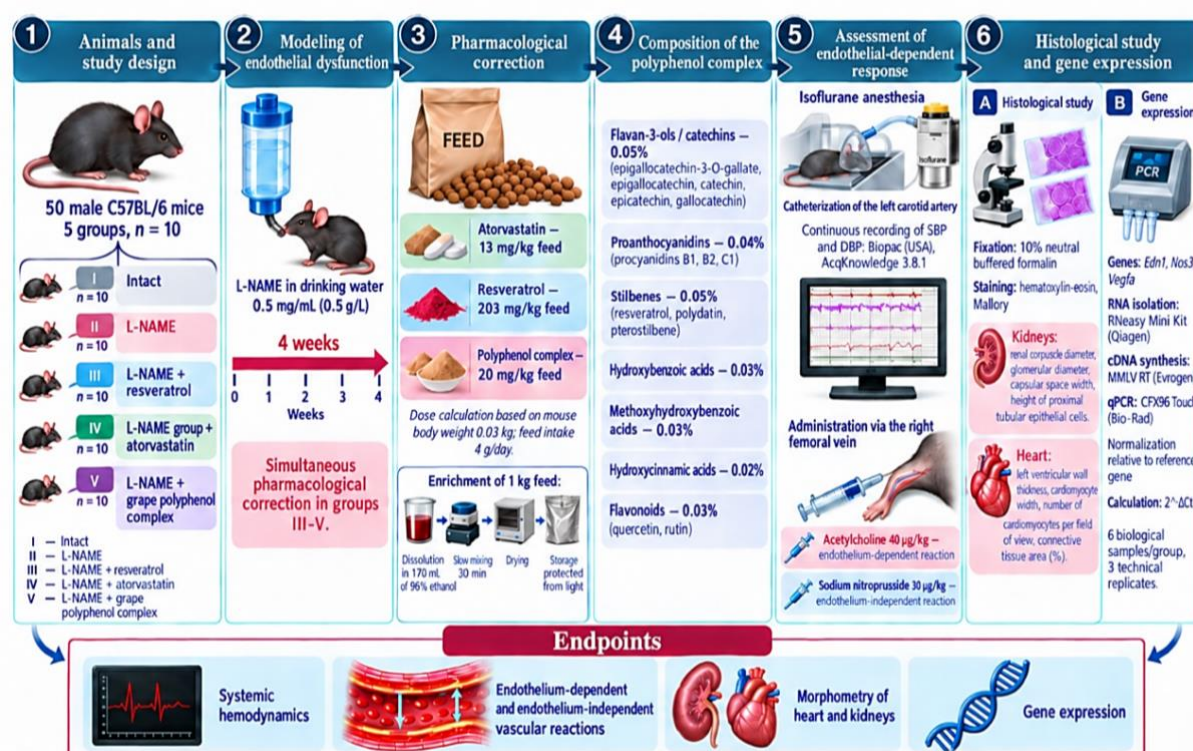
Results and Discussion: The grape polyphenol complex reduced systolic and diastolic blood pressure and the endothelial dysfunction coefficient compared with the **L-NAME** control. It also reduced morphological changes in the heart and kidneys, increased Nos3 expression, and decreased Edn1 and Vegfa expression. The observed changes approached the values of intact animals and indicate restoration of NO-dependent endothelial regulation.

Conclusion: Administration of the grape polyphenol complex at 0.08 mL/day for 4 weeks produced an endothelioprotective effect in C57BL/6 mice with **L-NAME**-induced endothelial dysfunction. Treatment improved endothelium-dependent vascular responses, systemic hemodynamics, tissue morphology, and endothelial-related gene expression.



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Graphical Abstract



Keywords

endothelial dysfunction; blood pressure; L-NAME; polyphenol complex; C57BL/6 mice

Introduction

In healthy blood vessels, the endothelium plays a key role in maintaining vascular homeostasis. Endothelial mediators, such as nitric oxide, are of particular importance. They promote vasodilation, thereby limiting the migration of platelets and leukocytes into the vascular intima (Naderi-Meshkin and Setyaningsih 2024). However, a number of diseases adversely affect the cardiovascular system and may lead to a pathological condition known as endothelial dysfunction. Common disorders such as diabetes mellitus, arterial hypertension, chronic obstructive pulmonary disease, and others can induce endothelial dysfunction. These conditions trigger processes associated with inflammation and oxidative stress, which, in turn, initiate a cascade of subsequent reactions that further impair endothelial function. Loss of adaptive capacity may lead to degenerative changes in other organs and tissues and contribute to metabolic dysfunction (Yang et al. 2024; Marcuccio et al. 2025).

The scale of this problem makes endothelial dysfunction an important subject for further investigation. Cardiovascular diseases remain one of the leading causes not only of mortality but also of disability. Despite substantial advances in preventive and diagnostic measures, as well as progress in treatment, the number of deaths from cardiovascular diseases has increased compared with the previous decade. It should also be noted that cardiovascular diseases are increasingly observed at younger ages, and some conditions may follow a more aggressive course in younger than in older individuals (Palaniappan et al. 2026).

At present, restoration of endothelial function is largely achieved through correction of risk factors. Commonly used lipid-lowering and antihypertensive agents, as well as therapies aimed at reducing body weight, can contribute to the restoration of vascular homeostasis (Arabi et al. 2024; Ding et al. 2022). However, their effects do not always address mechanisms associated with oxidative stress and inflammation of the vascular wall. Therefore, interest remains in compounds capable of acting on multiple components of endothelial dysfunction and complementing existing approaches to its pharmacological correction.

The use of natural polyphenols represents a promising approach. Their effects are associated with several mechanisms involved in endothelial dysfunction. **Resveratrol**, catechins, anthocyanins, and proanthocyanidins can reduce the formation of reactive oxygen species. They support nitric oxide synthase activity and increase the bioavailability of nitric oxide. In addition, polyphenols may attenuate inflammatory processes in the vascular wall (Li and Zhang 2023). The effects of polyphenol complexes are of particular interest. Such combinations may be useful not only for achieving an endothelioprotective effect; the presence of several compounds may provide a broader spectrum of biological effects.

Investigation of the mechanisms of endothelial dysfunction in humans has several limitations. This also applies to the assessment of the endothelioprotective properties of natural polyphenols. Therefore, preclinical studies are of particular importance. A mouse model of L-NAME-induced endothelial dysfunction can be used for this purpose. **L-NAME** inhibits nitric oxide synthase and decreases nitric oxide production, thereby reproducing key features of impaired endothelial function.

The aim of the study was to investigate the endothelioprotective activity of a polyphenol complex in a mouse model of L-NAME-induced endothelial dysfunction by assessing hemodynamic parameters, endothelial function, and the expression of genes involved in the regulation of vascular homeostasis.

Materials and Methods

Animals

The study was approved by the Commission for the Control of the Care and Use of Laboratory Animals of Belgorod State National Research University (BelSU), expert opinion No. 01-08i/25 dated August 18, 2025. C57bl6 mice were housed under specific pathogen-free (SPF) conditions in the vivarium of Belgorod State National Research University, with an artificially regulated 12-h light/12-h dark cycle at a temperature of +22 to +26°C and free access to food and water. The study followed the ethical principles for the treatment of laboratory animals in accordance with the European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes (ETS No. 170). All painful procedures were performed in accordance with Directive 2010/63/EU of the European Parliament and of the Council of September 22, 2010 on the protection of animals used for scientific purposes.

Study design

Animal groups

The study was performed in laboratory C57bl6 mice. A total of 50 male mice were included in the experiment and randomly allocated to five equal groups (n = 10 per group).

- I – intact group;
- II – control group with L-NAME-induced endothelial dysfunction;
- III – L-NAME-induced endothelial dysfunction + **resveratrol** group;
- IV – L-NAME-induced endothelial dysfunction + **atorvastatin** group;
- V – L-NAME-induced endothelial dysfunction + grape polyphenol complex group.

The studied compounds

Standard laboratory rodent chow was used as the basal diet. The feed was separately enriched with **atorvastatin** (Centrient Pharmaceuticals India Private Limited, India), **resveratrol** (Hunan Huacheng Biotech Inc., China), or the polyphenol complex in liquid form. Doses were calculated based on an average mouse body weight of 0.03 kg and a daily feed intake of 4 g. For each 1 kg of feed, 13 mg of **atorvastatin**, 203 mg of **resveratrol**, or 20 mL of the polyphenol complex was added. Daily intake per animal was 0.812 mg of **resveratrol**, 0.052 mg of **atorvastatin**, and 0.08 mL of the grape polyphenol complex over a period of 4 weeks. The polyphenol complex was produced at the Research Institute of Pharmacology of Living Systems, Belgorod State National Research University (BelSU), from fermented juice of red grape varieties. The composition of the polyphenol complex is presented in Table 1. The calculated amount of active substance was dissolved in 170 mL of 96% ethanol, after which the resulting solution was evenly distributed through 1 kg of feed by slow mixing for 30 min. The feed was then dried at room temperature until the ethanol had completely evaporated and stored in airtight containers protected from light.

Experimental model

Endothelial dysfunction was induced by adding **L-NAME** to the drinking water at a concentration of 0.5 mg/mL (0.5 g/L). The modeling regimen was selected based on the findings by De Moudt et al. (2022), who demonstrated a pronounced impairment of the endothelium-dependent vascular response

in C57BL/6 mice during the early stages of L-NAME exposure. The modeling period was 4 weeks. During the experimental period, animals in the corresponding groups received the assigned treatment. At the end of the modeling period, systemic hemodynamics, the endothelium-dependent vascular response, histological parameters, and the expression of genes involved in the regulation of endothelial function were assessed.

Table 1. Composition of the grape polyphenol complex

Compound group	Compounds	Content, %
Flavan-3-ols / catechins	Epigallocatechin-3-O-gallate Epigallocatechin Catechin Epicatechin Gallocatechin	0.05%
Proanthocyanidins / condensed tannins	Procyanidin B1 Procyanidin B2 Procyanidin C1	0.04%
Stilbenes / stilbenoids	Resveratrol Polydatin Pterostilbene	0.05%
Hydroxybenzoic acids	Gallic acid 4-hydroxybenzoic acid 3,4-dihydroxybenzoic acid	0.03%
Methoxyhydroxybenzoic acids	Vanillic acid Syringic acid	0.03%
Hydroxycinnamic acids	Caffeic acid trans-p-Coumaric acid	0.02%
Flavonoids	Quercetin Rutin	0.03%

Assessment of the endothelium-dependent response

Under inhalational isoflurane anesthesia, the left carotid artery was catheterized for continuous recording of systolic blood pressure (SBP) and diastolic blood pressure (DBP) using a Biopac system and AcqKnowledge 3.8.1 software (USA). Acetylcholine and sodium nitroprusside were administered as bolus injections through the right femoral vein. Endothelium-dependent and endothelium-independent vascular responses were assessed after intravenous administration of acetylcholine (40 µg/kg) and sodium nitroprusside (30 µg/kg), respectively. The endothelial dysfunction coefficient (EDC) was calculated according to the method described by Puchenkova et al. (2025).

Histological examination

After collection, the organs were fixed in 10% neutral buffered formalin, after which histological sections were prepared.

The organs were stained with hematoxylin and eosin and using Mallory's trichrome method. Kidney morphometry included measurement of the renal corpuscle diameter, vascular glomerulus diameter, width of the capsular space, and height of epithelial cells of the proximal nephron tubules. In the heart, the thickness of the left ventricular wall, cardiomyocyte width, the number of cardiomyocytes in a standard field of view, and the area of connective tissue between cardiomyocytes, expressed as a percentage, were determined.

Gene expression

To assess molecular changes associated with endothelial function, vascular tone, and vascular wall remodeling, the expression of the *Edn1*, *Nos3*, and *Vegfa* genes was examined. After euthanasia, tissue samples were rapidly excised, placed on ice, and prepared for molecular biological analysis. Total RNA was isolated from the tissues using the RNeasy Mini Kit (250) (Qiagen, Germany) according to the manufacturer's instructions. RNA quantity and purity were assessed spectrophotometrically. Complementary DNA was synthesized using the MMLV RT reverse transcription kit (Evrogen, Russia). Quantitative real-time polymerase chain reaction was performed using the CFX96 Touch Real-Time PCR Detection System (Bio-Rad Laboratories, Inc., USA). Expression levels of the genes of interest were normalized to the reference gene used in the experiment. Relative expression was calculated using the $2^{-\Delta Ct}$ method and expressed

in relative units. Six biological samples from each group were used for gene expression analysis. Each sample was analyzed in three technical replicates.

Statistical analysis

Statistical analysis was performed using GraphPad Prism 8.0 software (GraphPad Software, San Diego, CA, USA). The obtained data were normally distributed. Between-group comparisons were performed using the unpaired Student's t-test. Data are presented as mean $\pm$ standard deviation; values of $p < 0.05$ were considered statistically significant.

Results

Hemodynamic assessment

Modeling of endothelial dysfunction was accompanied by the development of persistent arterial hypertension (Fig. 1). SBP in the control group reached 140.08 mmHg, which was 23.2% higher than in intact animals. DBP increased by 25.8%, from 96.14 to 120.91 mmHg. Accordingly, marked changes were also observed in the EDC, which increased almost six-fold on average, from 0.535 to 3.147.

Pharmacological correction attenuated these changes to varying degrees compared with the control group. After 4 weeks of treatment, SBP in the *resveratrol* and *atorvastatin* groups was 5.2% and 9% lower than in the control group, respectively. For DBP, the differences from the control group were 2.4% and 11.4%, respectively. A similar pattern was observed for the EDC: compared with the *L-NAME* group, it was reduced by 23% and 29% in the *resveratrol* and *atorvastatin* groups, respectively. However, pharmacological correction did not completely restore the parameters to the levels observed in the intact group.

Treatment with the investigated pharmacological composition reduced systolic and diastolic blood pressure by an average of 11.6% and 12.4%, respectively, compared with the control animals, although both values remained higher than those in the intact group. The EDC was lower than in all other groups, with the most pronounced difference observed relative to the control group (40.3%). Compared with the other correction approaches, SBP in the polyphenol complex group was 7% and 3% lower than in groups with *resveratrol* and *atorvastatin*, respectively, whereas DBP was lower by 10% and 1%, respectively. No statistically significant differences in SBP or DBP were found between the polyphenol complex and the statin, while the difference in EDC remained significant.

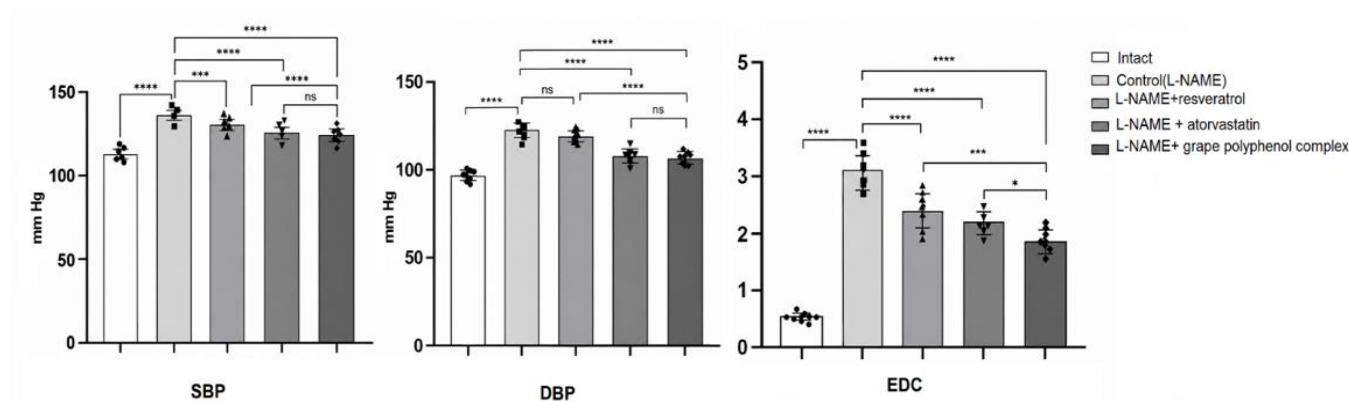


Figure 1. Hemodynamic parameters and endothelial dysfunction coefficient in *L-NAME*-treated mice. **Note:** Data are presented as mean $\pm$ SD. SBP – systolic blood pressure; DBP – diastolic blood pressure; EDC – endothelial dysfunction coefficient. Statistical significance between the indicated groups: * – $p < 0.05$; *** – $p < 0.001$; **** – $p < 0.0001$; ns – not significant.

Quantitative histological assessment

Morphometric analysis of the kidneys revealed a similar pattern of changes in the measured parameters (Fig. 2).

In particular, the diameter of the renal corpuscle in the intact group (Fig. 3) was 86.48 μm ; in the control group, this parameter decreased 1.1-fold to 78.34 μm . The diameter of the vascular glomerulus showed a similar trend and was 66.86 μm in the control group versus 79.62 μm in the intact group. The subcapsular space showed a compensatory 3.3-fold expansion compared with the intact group, in which this parameter was 5.9 μm . Analysis of proximal tubule epithelial height showed a 1.3-fold decrease in the *L-NAME* group (18.94 μm and 14.22 μm , respectively).

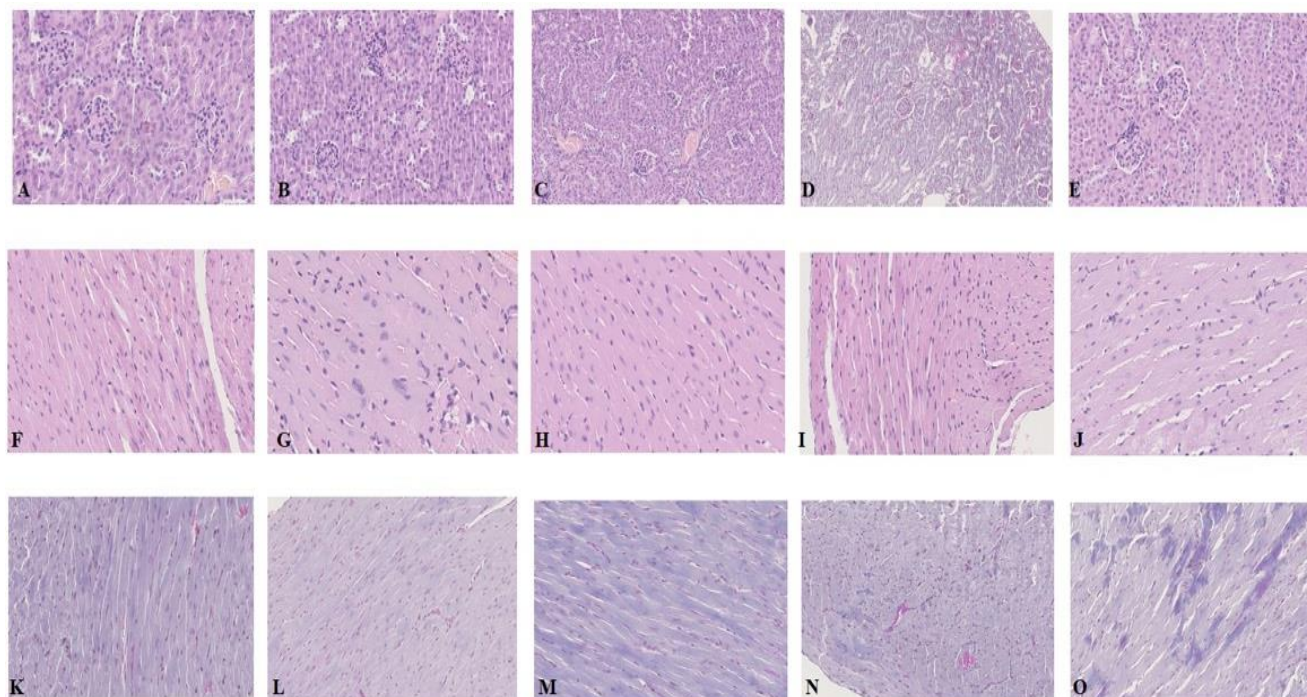


Figure 2. Histological examination of the kidneys and myocardium in experimental groups during the modeling of L-NAME-induced endothelial dysfunction in C57BL/6 mice. **Note:** A-E – kidney sections stained with hematoxylin and eosin; F-J – myocardial sections stained with hematoxylin and eosin; K-O – myocardial sections stained using Mallory's trichrome method. A, F, K – Intact group; B, G, L – Control group (L-NAME); C, H, M – L-NAME + resveratrol; D, I, N – L-NAME + atorvastatin; E, J, O – L-NAME+ grape polyphenol complex.

Under pharmacological correction, the **resveratrol** group showed a moderate therapeutic effect, morphologically manifested by a reduction in dystrophic changes, an increase in glomerular diameter compared with the control group, narrowing of the subcapsular space, and an increase in the height of the epithelium lining the proximal nephron tubules.

Mice receiving **atorvastatin** demonstrated a pronounced nephroprotective effect, morphologically manifested by renal corpuscle and vascular glomerulus diameters approaching intact values. Tubular epithelial cells retained their structure, and their height was restored to 16.94 μm .

In the grape polyphenol complex group, the diameters of the glomeruli and renal corpuscles, as well as the width of the subcapsular space, were almost completely restored toward the levels observed in intact animals, indicating restoration of an adequate filtration process. The height of proximal tubule epithelial cells was restored almost to the baseline values of the intact group, and the integrity of the brush border was observed (Table 1).

Table 2. Morphometric indicators of the kidneys in experimental groups during the modeling of L-NAME-induced endothelial dysfunction in C57BL/6 mice

Group	Diameter of the renal corpuscle, μm	Width of the subcapsular space, μm	Diameter of the vascular glomerulus, μm	Height of the proximal tubule epithelium, μm
Intact group	86.48 ± 4.44	5.90 ± 0.45	79.60 ± 3.04	18.92 ± 0.79
Control group	78.34 ± 3.63	19.62 ± 2.58	66.84 ± 1.59	14.22 ± 1.35
L-NAME + atorvastatin	82.06 ± 4.09	10.74 ± 1.11	71.32 ± 3.65	16.94 ± 1.68
L-NAME + resveratrol	70.76 ± 1.14	11.14 ± 0.62	69.58 ± 2.23	14.52 ± 0.89
L-NAME+ grape polyphenol complex	85.52 ± 2.42	5.95 ± 0.32	72.40 ± 4.10	17.34 ± 1.42

Note: Average values are presented; n=10 in each group. All morphometric indicators of the kidneys are expressed in microns.

Examination of the hearts showed that cardiomyocyte width in the **L-NAME** group was 19.9 μm , which was 1.2-fold greater than in intact animals. Changes were also observed in left ventricular wall thickness, which increased 1.18-fold: 598.6 μm versus 504.0 μm in the intact group. An opposite trend was observed for cell density per unit area of the section: because of the increased cardiomyocyte size, the number of cardiomyocytes in a standard field of view decreased 1.4-fold (101.8 and 70.6, respectively). The area occupied by connective tissue increased by 13.8 percentage points, or 2.4-fold.

During treatment with the polyphenol complex, the morphohistological parameters more closely approached intact values. Cardiomyocyte thickness and left ventricular wall thickness decreased by 18.8% and 14.1%, respectively, relative to the control group. The number of cardiomyocytes increased by 41.9%, almost reaching the values of the group without correction. The area of connective tissue decreased by 53.7% compared with such in the **L-NAME** group and was only 10.2% higher than in the intact value (Table 2).

Table 3. Morphometric parameters of the heart in experimental groups during the modeling of L-NAME-induced endothelial dysfunction in C57BL/6 mice

Group	Thickness of cardiomyocytes, μm	Number of cardiomyocytes	Thickness of the left ventricular wall, μm	Area of connective tissue in the standard field of view, %
Intact group	15.55 $\pm$ 1.72	101.80 $\pm$ 10.80	504.00 $\pm$ 2.92	9.98 $\pm$ 0.31
Control group	19.90 $\pm$ 0.52	70.60 $\pm$ 4.72	598.00 $\pm$ 4.85	23.78 $\pm$ 1.90
L-NAME + atorvastatin	16.40 $\pm$ 1.08	94.60 $\pm$ 5.13	554.00 $\pm$ 25.26	12.84 $\pm$ 0.39
L-NAME + resveratrol	17.24 $\pm$ 1.20	86.40 $\pm$ 1.14	582.80 $\pm$ 22.47	22.32 $\pm$ 0.87
L-NAME + grape polyphenol complex	16.16 $\pm$ 1.21	100.20 $\pm$ 7.16	513.80 $\pm$ 2.39	11.00 $\pm$ 0.76

Note: Average values are presented; n=10 in each group. The thickness of the cardiomyocytes and the wall of the left ventricle is expressed in micrometers; the area of connective tissue is expressed as a percentage; the number of cardiomyocytes is the number of cells.

Gene expression

The most pronounced differences between the pharmacological correction approaches were observed for Nos3 expression. In the myocardium, Nos3 expression in the control group was markedly lower than in intact animals (3.43 and 9.66, respectively). The other groups had intermediate values; nevertheless, the animals receiving the pharmacological composition showed the values closest to those of mice without modeled pathology (9.16). A similar pattern was observed in the aorta: the grape polyphenol complex restored the expression value to 13.57, more than twice the result in the **L-NAME** group (6.55). In the kidneys, a significant increase in Nos3 relative to the control group was recorded with atorvastatin and the polyphenol complex.

For Edn1 and Vegfa, the general trend was increased expression in all studied organs during L-NAME-induced endothelial dysfunction. Edn1 expression in the aorta increased from 2.74 in intact animals to 6.37 in the **L-NAME** group, in the kidneys from 1.40 to 4.20, and in the heart from 0.79 to 3.64. With the polyphenol complex, the values decreased to 4.12, 2.31, and 1.44, respectively. A similar pattern was observed for Vegfa in the aorta, kidneys, and heart: expression increased from 1.38 to 3.97, from 1.77 to 3.78, and from 1.12 to 3.42, respectively. During treatment with the polyphenol complex, Vegfa expression decreased to 2.21 in the aorta, 1.68 in the kidneys, and 2.15 in the heart. The closest approximation to intact values was observed for Edn1 in the heart and kidneys and for Vegfa in the kidneys (Fig. 3).

Discussion

The model of induced endothelial dysfunction reproduced the characteristic changes associated with this pathological condition. In our study, a group of animals with pronounced impairment of endothelium-dependent regulation was obtained, allowing comparison with groups receiving pharmacological treatment. The severity of the pathology was reduced in the treatment groups, with the most pronounced effect observed in the group receiving the investigated composition.

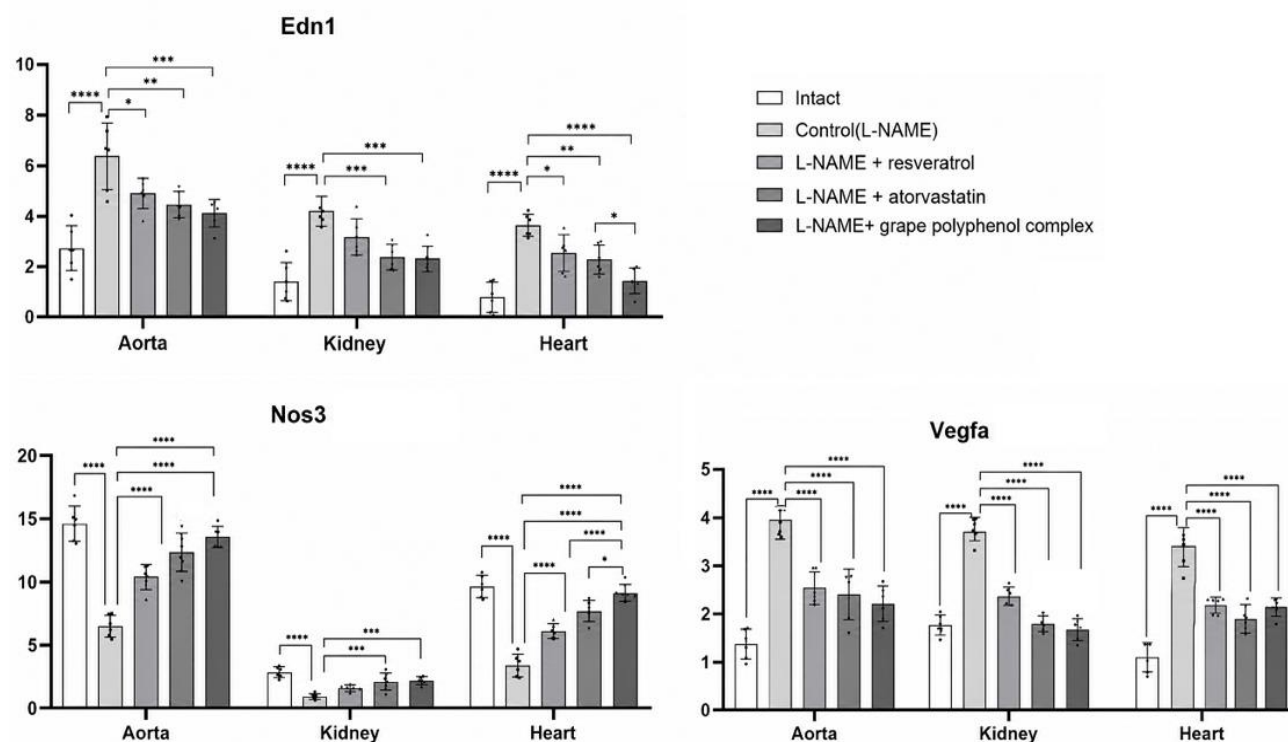


Figure 3. Expression of the *Edn1*, *Nos3*, and *Vegfa* genes in the aorta, kidneys, and heart of C57BL/6 mice in a model of endothelial dysfunction. **Note:** The data are presented as the mean value $\pm$ SD. Groups: Intact – intact animals; Control (L-NAME) – animals with L-NAME-induced endothelial dysfunction; L-NAME + resveratrol – group receiving resveratrol correction; L-NAME + atorvastatin – group receiving atorvastatin correction; L-NAME + grape polyphenol complex – group receiving grape polyphenol complex correction. Brackets indicate pairwise intergroup comparisons. Statistical significance of differences: * – $p < 0.05$; ** – $p < 0.01$; *** – $p < 0.001$; **** – $p < 0.0001$.

Based on the obtained data, the effect of the grape polyphenol complex appears to involve NO-dependent regulation of vascular tone. This is indicated by the decrease in EDC, whereas the effect of the complex on baseline blood pressure was close to that of atorvastatin. This pattern suggests that the polyphenol complex affects vascular tone regulation and that its effect is not limited to lowering arterial blood pressure. Gene expression results also showed an increase in *Nos3*: its level in the myocardium was almost indistinguishable from that in intact mice, while in the aorta it was more than twice the value observed in the control group. At the same time, *Edn1* expression decreased, indicating attenuation of endothelin-dependent vasoconstrictor regulation.

A cardioprotective effect was observed in a similar manner. Presumably, this was associated with a reduction in arterial blood pressure, thereby decreasing left ventricular afterload. This was supported by the differences in structural changes between the control group and the grape polyphenol complex group. According to the expression data, reduced activity of the endothelin pathway is of particular interest because *Edn1* is associated not only with vasoconstriction but also with the development of cardiac hypertrophy and fibrotic remodeling.

The nephroprotective effect was presumably associated with improved renal blood flow. In the experiment, administration of the polyphenol composition improved the condition of the renal vascular endothelium, contributed to preservation of perfusion, and limited a further increase in vascular resistance. Additional support for this interpretation was provided by increased *Nos3* expression and decreased *Edn1* expression in renal tissue.

An antioxidant effect may also be considered one of the proposed mechanisms of action of the grape polyphenol complex. Polyphenolic compounds can limit the formation of reactive oxygen species, thereby helping to preserve NO bioavailability and maintain endothelial function. This is indirectly consistent with restoration of *Nos3* expression and the pronounced decrease in EDC. Changes in *Vegfa* are also of interest. Its increase during induction of endothelial dysfunction, followed by a decrease during treatment with the polyphenol complex, may reflect a reduction in the hypoxic stimulus in tissues.

This response to the polyphenol complex suggests that its endothelioprotective effect may be produced by compounds with overlapping molecular targets. Flavan-3-ols and proanthocyanidins are primarily capable of supporting NO-dependent regulation of vascular tone. For example, epicatechin has been shown to activate eNOS through the PI3K/Akt-dependent pathway, followed by increased NO production (Ramirez-Sanchez et al. 2010),

whereas **epigallocatechin-3-O-gallate (EGCG)** promotes phosphorylation of Akt and eNOS with the development of endothelium-dependent vasodilation (Lorenz et al. 2004). A similar mechanism has been described for grape proanthocyanidins. Their administration increased eNOS expression through the AMPK/SIRT1/KLF2 pathway and enhanced NO production, which in experimental conditions was accompanied by attenuation of the increase in arterial blood pressure (Cui et al. 2012). Direct effects on NO production have also been reported for procyanidin C1 (Byun 2012). Stilbenoids contained in the complex act in the same general direction. **Resveratrol** has been shown to increase eNOS expression and activity, prevent eNOS uncoupling, and reduce endothelin-1 production, with SIRT1 and AMPK playing key roles in these effects (Li et al. 2019). **Pterostilbene** also stimulates eNOS through PI3K/Akt (Park et al. 2015), and flavonoids (Naletova et al. 2025) can stimulate eNOS expression and NO production in endothelial cells (Ugusman et al. 2014).

Another component of the effect of the grape polyphenol complex is probably related to modulation of oxidative stress and may contribute to its nephroprotective and cardioprotective effects. Proanthocyanidins, protocatechuic acid, and rutin can reduce the formation of reactive oxygen species and thereby limit endothelial injury. Proanthocyanidins have also been shown to increase SOD activity, which may strengthen the cell's intrinsic antioxidant defense (Okudan et al. 2011). **Polydatin** acts through a SIRT1/Nrf2-dependent pathway (Huang et al. 2015), while rutin additionally suppresses Nox4 and NLRP3 signaling (Wang et al. 2017). Caffeic acid supports NO-dependent endothelial function under conditions of hypoxia and oxidative stress (Migliori et al. 2015). This combination of effects may be one reason for the less pronounced changes in the heart and kidneys during treatment with the grape polyphenol complex.

It should be taken into account that not all data obtained in this study allow unambiguous conclusions regarding the presence of the proposed mechanisms. In some cases, the changes observed during administration of the composition should be regarded primarily as indirect evidence of the corresponding mechanism of action. More rigorous assessment will require further experimental studies with a broader range of diagnostic methods, for example blood rheology, as well as the inclusion of direct molecular and biochemical markers.

Conclusion

The grape polyphenol complex demonstrated endothelioprotective activity in L-NAME-induced endothelial dysfunction in C57BL/6 mice at a daily dosage of 0.08 mL of the grape polyphenol complex over a period of 4 weeks. Its administration improved endothelial-dependent vascular responses, normalized hemodynamic and gene expression parameters, and reduced structural changes in the heart and kidneys.

Additional Information

Conflict of interest

The authors declare that they have no conflicts of interest.

Funding

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Ethics statement

The study was approved by the Animal Ethics Committee of Belgorod State National Research University (BelSU), approval No. 01-08i/25 dated 18 August 2025.

Acknowledgments

The figures were created with BioRender.com.

Data availability

All relevant data generated and analyzed during this study are included in this article.

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Author Contributions

- **Petr R. Lebedev**, Junior researcher, Belgorod State National Research University, Belgorod, Russia; e-mail: artkelt98@yandex.ru; **ORCID ID**: <https://orcid.org/0000-0001-9102-3360>. Developing the concept, methodology and investigation.
- **Tatyana V. Avtina**, PhD in Pharmaceutical Sciences, Associate Professor of the Department of Pharmacology and Clinical Pharmacology, Belgorod State National Research University, Belgorod, Russia; e-mail: tatyanavtina@yandex.ru; **ORCID ID**: <https://orcid.org/0000-0003-0509-5996>. Developing a research design, processing the results obtained, and writing the article.
- **Vladimir V. Gureev**, Belgorod State National Research University, Doctor Habil. of Medical Sciences, Associate Professor, Professor of the Department of Pharmacology and Clinical Pharmacology, Belgorod State National Research University, Belgorod, Russia; e-mail: produmen@mail.ru; **ORCID ID**: <https://orcid.org/0000-0003-1433-1225>. Developing a research design, processing the results obtained, and writing the article.
- **Mikhail V. Pokrovskii**, Doctor Habil. of Medical Sciences, Professor, Department of Pharmacology and Clinical Pharmacology, Belgorod State National Research University, Belgorod, Russia; e-mail: mpokrovsky@yandex.ru; **ORCID ID**: <https://orcid.org/0000-0002-1493-3376>. Setting the research objectives, motivating the team, conducting a critical analysis of the material, and giving the final approval of the manuscript.