



The influence of the preparation “Bendamin” on the morphological and biochemical indices of blood of rats in experimental modeling of heart failure

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Abstract

The purpose of this work was to study the influence of “Bendamin” on the morphological and biochemical indices of rat blood in experimental modeling of heart failure. The investigation were performed on white sexually mature young male-rats of Wistar line weighing 180–200 g, kept at the standard ration of the institute vivarium of the State Research Institute of Veterinary Preparations and Fodder Additives. To create a model of doxorubicin-induced cardiomyopathy, 24 male rats were selected. Animals were divided into 3 groups of 6 animals in each: control group – intact animals; experimental group E₁, in which animals were modeled doxorubicin-induced cardiomyopathy by intraperitoneal injected of doxorubicin at a dose of 2.5 mg/kg 3 times a week for two weeks; experimental group E₂, in which animals, after injection of doxorubicin, the preparation “Bendamin” was injected in the dose 20 mg/kg intragastrically. For experimental modeling of heart a decrease in the number of red blood cells was found in the blood of rats of the first experimental group by 34.7 % and hemoglobin level – by 24.2 %, with a simultaneous increase in the average hemoglobin content in one erythrocyte by 16.2 %. Doxorubicin-induced cardiomyopathy in rats of the first experimental group was accompanied by a decrease in leukocyte count by 22.2 %, an increase in uric acid level by almost 2 times, creatinine by 11.4 %, cholesterol by 61.1 %. In the research of the protein synthesizing function of the liver of rats in experimental modeling of heart failure, a decrease in total protein by 9.6 % was found. These changes were accompanied by a slight decrease in albumin levels and a slight increase in globulin levels. By intoxication with doxorubicin, rats of the first experimental group showed a violation of the functional state of the liver, indicating an increase in the activity of aminotransferases in their blood, namely: ALAT by 34.2 %, AsAT – by 21.7 %, accordingly. Positive action of cardio preparation “Bendamin” on rats organism under conditions of creation of a model of doxorubicin-induced cardiomyopathy was installed, manifested by the normalization of hematological and biochemical parameters, functional status and protein synthesis of the liver.

Key words: pharmacology, “Bendamin” preparation, rats, heart failure, cardiomyopathy.

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1. Introduction

Cardiology is one of the directions of veterinary medicine which is most intensively developing. Annually, a large number of new investigations are conducted in the world, based on the results of which refine the views on the treatment of cardiovascular diseases (Varkholiak & Gutyj, 2018). Recently, our ideas about the pathogenetic mechanisms of heart failure have changed significantly, and new approaches to its therapy are being developed (Tjostheim et al., 2019; Oldach et al., 2019).

Cardiac and vascular pathologies in dogs and cats are extremely diverse and fairly common in the daily practice of a veterinarian in Ukraine and beyond. According to the classification of Professor G. V. Domrachev, in domestic animals, cardiovascular pathology is divided into diseases of the myocardium, pericardium, endocardium and diseases of the

blood vessels. There is a close relationship between the above pathologies, age and animal breeds (Undhad et al., 2012; Varkholiak, 2016; Zhulikova, 2016; Yata et al., 2019).

According to foreign authors, cardiovascular pathology is about 10.0 % of all pathologies of the internal organs.

During 2014–2018, pets with the following pathologies were coming in at the “DoctorVET” in Animal Health Center of Lviv: cardiomyopathy (dilatation, hypertrophic, arrhythmogenic), MK endocardiosis, isolated cases of birth defects, and also pericarditis.

In recent years, several significant investigations have been conducted in the field of treatment of heart failure, new diagnostic techniques were introduced, the genetic features of particular pathologies were identified, and new medicines appeared, such as pimobendan, polyunsaturated fatty acids, sildenafil, levosimendan, torasemide, nebivolol, lisinopril, clopidogrel, and some others. However, data on the use of

new therapeutic means in cardiovascular pathology is not enough (Trofimjak & Slivinska, 2016; Varkholiak & Gutyj, 2019; Varcholyak & Gutyi, 2019).

Thus, there is a reasonable need to develop, conduct comprehensive investigations of the mechanisms of action of the medicines, its interaction with other preparations, clinical efficiency of treatment and safety of animals in the development of cardiovascular pathology.

The purpose of the work is to investigate the influence of cardio preparation "Bendamin" on the morphological and biochemical indices of rat blood in experimental modeling of heart failure.

2. Materials and methods

The investigations were performed on white sexually mature young rats-males of the Wistar line weighing 180–200 g, which were kept on a standard ration of the institute vivarium of the State Research Institute of Veterinary Preparations and Feed Additives. Throughout the experiment, rats were kept in a balanced ration containing all the necessary components, and animals were provided with drinking water without restrictions from 0.2 liter glass bowls.

The experimental investigations were conducted in accordance with the requirements of the biological-biological experiment for the selection of analogues, setting control, compliance with the same feeding conditions and retention during the experiment and the results.

To create a model of doxorubicin-induced cardiomyopathy, 24 male rats were selected. Animals were divided into 3 groups of 6 animals in each: control group – intact animals; experimental group E₁, in which animals were modeled doxorubicin-induced cardiomyopathy by intraperitoneal introduction of doxorubicin at a dose of 2.5 mg/kg 3 times a week for two weeks; experimental group E₂, in which animals were injected intragastrically with "Bendamin" at a dose of 20 mg/kg.

Morphological and biochemical indices of blood were investigated by the method (Vlizlo et al., 2012).

All animal manipulations were carried out in accordance with the European Convention for the Protection of Vertebrate Animals, which is used for experimental and scientific purposes (Strasbourg, 1986).

Analysis of the research results was performed using the Statistica 6.0 software package. The probability of differences was evaluated by Student's t-test. The results were considered probable at $P \leq 0.05$.

3. Results and discussion

In experimental modeling of heart failure by intraperitoneal injection of doxorubicin at a dose of 2.5 mg/kg 3 times a week for two weeks, was found reducing the number of erythrocytes in the blood of rats of the first experimental group by 34.7 % and hemoglobin level – by 24.2 %, with a simultaneous increase in the average hemoglobin content in one erythrocyte by 16.2 % (Table. 1). These changes may be related to the negative action of oxidative stress on cardiomyocytes.

In the search of the above mentioned indices in the blood of the experimental group E₂ an increase in the number of erythrocytes and hemoglobin level compared with sick animals was observed. It should be noted that the number of erythrocytes in the blood of rats treated with the preparation "Bendamin" ranged from 7.14 ± 0.547 T/l, whereas in rats of the first experimental group this indicator was 4.94 ± 0.125 T/l. In the research of hemoglobin in the blood of rats in the experimental groups, their increase is found in the second experimental group of rats compared to the first experimental group, however, when compared with the control group, this indicator was lower by 5.7 %, respectively.

The average hemoglobin content in one erythrocyte was higher in the first experimental group, where it was respectively 17.71 ± 1.10 pg, slightly lower was in the second experimental group.

Doxorubicin-induced cardiomyopathy in rats of the first experimental group was accompanied by a decrease in leukocyte number by 22.2 %, which may be related to a decrease in immunity and inhibition of leukocyte formation in the bone marrow against the background of oxidative stress caused by the injection of doxorubicin.

When using the preparation "Bendamin" to experimental rats under the conditions of experimental modeling of heart failure was found an increase of their number to 6.58 ± 0.55 G/l, which is 26.3 % higher than the indices of the first experimental group.

In order to evaluate the influence of preparation "Bendamin" in simulated doxorubicin-induced cardiomyopathy, on kidney condition was determined by such indicators as uric acid and creatinine levels. It was installed, that in the first experimental group of rats uric acid levels were increased almost in 2-times, whereas creatinine levels were increased by 11.4 %. The lowest level of uric acid was in the experimental group E₂, which along with the intoxication of doxorubicin were used the preparation "Bendamin". Similar changes are observed in the research of creatinine level, which in the experimental group E₂ was fluctuated respectively within 50.4 ± 1.54 μ mol/l, whereas in the control group of rats this indicator was 49.0 ± 1.07 μ mol/l. These changes in the blood of the second experimental group indicate a partial normalization of renal function.

We found that in modeling heart failure in the experimental animals of the first experimental group there was an increase in cholesterol level by 61.1 % compared with the control group. It should be noted that when using the preparation "Bendamin" cholesterol level was decreased by 36.3 % and reached physiological values.

Reorientation of oxidative metabolism in the myocardium to the anaerobic pathway as a result of rats intoxication with doxorubicin and damage to their heart muscle was reflected by a 59 % increase in the creatine phosphokinase MB activity in the rats serum of the first experimental group compared with indicators taken from the control group of rats. This enzyme is a specific and sensitive marker of myocardial damage.

Table 1

Morphological and biochemical indices of blood of rats in experimental modeling of heart failure and action of the preparation "Bendamin" ($M \pm m$, $n = 6$)

| Indicators | Groups of animals | | |
|---|-------------------|----------------|----------------|
| | Control | Experimental 1 | Experimental 2 |
| Erythrocytes, T/l | 7.57 ± 0.349 | 4.94 ± 0.125* | 7.14 ± 0.547 |
| Hemoglobin, g/l | 115.4 ± 2.9 | 87.5 ± 2.0* | 110.6 ± 3.2 |
| The average hemoglobin content in one erythrocyte, pg | 15.24 ± 1.14 | 17.71 ± 1.10* | 15.49 ± 1.15 |
| Leukocytes, G/l | 6.70 ± 0.62 | 5.21 ± 0.75* | 6.58 ± 0.55 |
| Creatinine, μmol/l | 49.0 ± 1.07 | 54.6 ± 2.27* | 50.4 ± 1.54 |
| Cholesterol, mmol/l | 1.90 ± 0.072 | 3.06 ± 0.057* | 1.95 ± 0.072 |
| Uric acid, μmol/l | 83.4 ± 3.60 | 164.2 ± 4.11* | 91.3 ± 5.35 |
| LDH, mmk/l | 1.93 ± 0.20 | 1.85 ± 0.15 | 2.10 ± 0.30 |
| Creatine phosphokinase MB, in units/l | 78 ± 3.1 | 124 ± 4.3* | 105 ± 3.9 |

The injection of the cardio preparation to the experimental rats contributed to a slight decrease in creatine phosphokinase MB activity by 15.3 % compared with the first experimental group.

One of the major enzymes involved in glycolysis reactions is lactate dehydrogenase. Based on the conducted research, it was found that LDH activity in the blood of rats of the first experimental group decreased by 4 % due to doxorubicin intoxication. When using the "Bendamin" cardio preparation, to rats of the E₂ experimental group were found

the increase of this enzyme compared to the control and the first experimental group, here, accordingly, LDH activity ranged from 2.10 ± 0.30 mmk/l.

In the research of the protein synthesizing function of rat liver in experimental modeling of heart failure and the action of the "Bendamin" cardio preparation reduced the total protein in the first experimental group by 9.6 %, of the second experimental group – by 2.3 % compared with the control group (Table 2).

Table 2

Protein synthesizing function of rat liver in experimental modeling of heart failure and action of "Bendamin" preparation ($M \pm m$, $n = 6$)

| Indices | Groups of animals | | |
|--------------------|-------------------|----------------|----------------|
| | Control | Experimental 1 | Experimental 2 |
| Total protein, g/l | 62.31 ± 3.12 | 56.32 ± 3.57* | 60.84 ± 2.78 |
| Albumin, g/l | 24.12 ± 2.18 | 17.57 ± 3.34* | 22.67 ± 2.45 |
| Globulins, g/l | 38.19 ± 3.41 | 38.75 ± 4.10 | 38.17 ± 3.11 |

These changes were accompanied by a slight decrease in albumin levels and a slight increase in globulin levels, the level of albumins and globulins was thus set at 17.57 ± 3.34 and 38.75 ± 4.10 g/l in the first experimental group versus 24.12 ± 2.18 and 38.19 ± 3.41 in the control group of rats.

By intoxication with doxorubicin in rats of the first experimental group was found violations of the functional state of the liver which is indicated by increased activity of aminotransferases in their blood, namely: AlAT by 34.2 %, AsAT – by 21.7 %, respectively (Table 3).

Table 3

Functional state of rat liver in experimental modeling of heart failure and action of the preparation "Bendamin" ($M \pm m$, $n = 6$)

| Indices | Groups of animals | | |
|------------|-------------------|----------------|----------------|
| | Control | Experimental 1 | Experimental 2 |
| AlAT, un/l | 38.47 ± 2.10 | 51.62 ± 3.57* | 38.50 ± 2.10 |
| AsAT, un/l | 188.5 ± 15.86 | 229.4 ± 10.30* | 189.2 ± 11.25 |

The use of the preparation "Bendamin" in experimental rats under experimental modeling of heart failure, was set up a decrease in the activity of aminotransferases in serum of experimental group E₂. Thus, the activity of AlAT and AsAT in the serum of the second experimental group was decreased by 25.4 and 17.5 %, respectively, compared to the intoxicated rats, who were not given a cardiac preparation.

conditions of creation of a doxorubicin-induced cardiomyopathy model, that appears itself by the normalization of hematological and biochemical indices, functional status and protein synthesis function of the liver.

4. Conclusions

Based on our research, positive action of "Bendamin" cardio preparation on rats organism was set up under the

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