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The influence of the feed additive “Sylimevit” on the antioxidant protection of the body of dogs

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Under the influence of the pathological factor, there is a change in the intensity of lipid peroxidation, the accumulation in the blood of the concentration of products of free radical oxidation and reactive oxygen species, a decrease in the buffer capacity of the blood relative to the maintenance of optimal parameters of the intensity of free extreme reactions. The work aimed to investigate the effect of the feed additive “Sylimevit” on the antioxidant protection of the body of dogs after preventive deworming. 10 German Shepherd dogs aged 1–2 years were used for experimental research. It was established that when feeding the feed additive “Sylimevit” to dogs in the period after deworming, activation of the enzyme link of the antioxidant system occurs in the blood, which is indicated by an increase in the activity of catalase and superoxide dismutase in the blood serum of these animals and inhibition of lipid peroxidation processes (reduction of secondary and end products lipid peroxidation). On the 30th day of the experiment, the activity of catalase and superoxide dismutase in the blood of animals of the experimental group was the highest, and the level of lipid peroxidation products was the lowest. The obtained research results confirm the antioxidant properties of the Sylimevit feed additive. This may be because the composition of the drug includes milk thistle, methiphen, and vitamins, which in turn enhance the effect of each other and thereby inhibit the formation of radicals and the processes of lipid peroxidation. It is also worth noting the antioxidant properties of milk thistle, which according to the literature, also have similar properties. The complex effect of the indicated biologically active elements provided high hepatoprotective and antioxidant effects.

Key words: sylimevit, dogs, antioxidant system, peroxide oxidation, milk thistle.

Introduction

Active forms of oxygen are products of cellular metabolism. These include free radicals, products of incomplete reduction of atomic oxygen, hydrogen peroxide, singlet oxygen, etc. (Slobodian et al., 2019; Martyshchuk & Gutyi, 2019; Martyshuk & Gutyj, 2019). These highly reactive molecules can disrupt the intracellular environment's homeostasis by reacting with macromolecules such as DNA, proteins, and lipids. At low concentrations AFO (active forms of oxygen) affects physiological cellular processes: regulation of vascular tone, cell proliferation, synthesis of prostaglandins, the transmission of signals

from intercellular signaling molecules to regulatory systems that control gene expression, and microbicidal action of phagocytes (Martyshuk et al., 2016; Gutyj et al., 2016; 2017). It has been established that an increase in AFO in the body of animals leads to cell necrosis. Violation of homeostasis in the cell due to an increase in the content of reactive oxygen species is a crucial mechanism of the development of oxidative stress. Disturbances of homeostasis that lead to oxidative stress include, in particular: changes in homeostasis due to pathological factors, a change in homeostasis due to a violation of genetic information, and a defect in the regulatory system or target organ (Khariv et al., 2016; Stybel et al., 2021; 2022).

Under the influence of the pathological factor, there is a change in the intensity of lipid peroxidation, the accumulation in the blood of the concentration of products of free radical oxidation and reactive oxygen species, a decrease in the buffer capacity of the blood relative to the maintenance of optimal parameters of the intensity of free radical reactions (Hutyi, 2016; Hutyi et al., 2016; Grymak et al., 2020).

The antioxidant defense system is a system responsible for regulating the intensity of radical formation and neutralization of peroxidation products (Gutyj et al., 2018). The primary control mechanism of these reactions is connected with a chain of reverse redox reactions of metal ions, ascorbate, tocopherol, glutathione, and other substances (Lavryshyn et al., 2016). In addition, the value of these methods is significant for preserving long-existing macromolecules of nucleic acids and proteins, some components of membranes (Leskiv et al., 2022).

This system combines several substances of different natures. Each component of the antioxidant system acts in a close relationship with its other structural elements, harmoniously complements, and in many cases, enhances the effect of each other (Ostapyuk et al., 2021; Gutyj et al., 2022). The glutathione system forms the functional basis of the antioxidant protection system, the constituent elements of which are glutathione and enzymes that catalyze reactions of its reverse transformation (oxidation ↔ reduction) (Martyshuk et al., 2018; 2019). These enzymes include glutathione peroxidase, glutathione reductase, and glucose-6-phosphate dehydrogenase (Lavryshyn et al., 2019). In addition to the antioxidants mentioned above, catalase, peroxidase, and superoxide dismutase are also included, which can catalyze the reactions of direct destruction of peroxide compounds in the human and animal body (Ponkalo, 2012; Vyslotska et al., 2021).

According to the mechanism of action, the antioxidant system of animals is divided into specific and non-specific. The first system of antioxidant protection is directly aimed at reducing the level of oxidants in the body of animals through the binding of reactive oxygen species, which leads to the suppression of free radical reactions. The action of the non-specific antioxidant system of the animal body is associated with a decrease in the possibility of different generations of free radicals. One of these manifestations is the elimination of the pool of metal ions of variable valence (copper, iron) due to their binding by high-molecular compounds (ceruloplasmin, lactoferrin, transferrin) and prevention of the participation of these metals in free radical oxidation reactions (Martyshuk et al., 2021; Slobodian et al., 2021).

It is also worth noting the critical role of the enzyme link of the antioxidant defense system in the pathogenesis of invasive diseases in animals. One of these enzymes is superoxide dismutase, with the participation of which the chain of free radical processes is broken at the stage of the one-electron reduction of oxygen with the formation of superoxide anion radical (Bjelenichev et al., 2002; Varkholiak & Gutyj, 2019). The mechanism of action of superoxide dismutase is based on the sequential reduction and oxidation of the enzyme's active center by superoxide anion-radicals of metal (Me) (Varkholiak et al., 2021).

Thus, this enzyme takes part in regulating free radical oxidation in the body of animals during invasive diseases at the initial stage. Superoxide dismutase is characterized by structural stability and is one of the most thermostable globular proteins (Shcherbaty et al., 2019).

It is important to note that both an increase and a decrease in superoxide dismutase activity cause pathological processes caused by invasive diseases. As a result of the strengthening of the cytotoxic effect of hydrogen peroxide, which is formed due to the dismutation of the superoxide anion radical, and insufficient protection from reactive oxygen species, the processes of lipid peroxidation increase (Lavryshyn et al., 2016; Frishtak et al., 2022).

It is worth noting that along with superoxide dismutase, catalase is also essential, which quickly splits peroxide into water and oxygen (Baglaj et al., 2011; Fedorenko, 2016). Superoxide dismutase and catalase protect the body from highly toxic oxygen radicals (Stoyanovskyy et al., 2020). Catalase is found in all tissues, containing about 10-6 M. In general, the effect of catalase is reduced to a decrease in the concentration of cytotoxic hydroxyl radicals (Duh & Vovk, 2010; Zhukova et al., 2017). The active center of this enzyme contains trivalent iron and protoporphyrin, which interacts with hydrogen peroxide (Lavryshyn et al., 2016; Kochevenko et al., 2020). The reaction proceeds in two stages: first, a complex is formed between the enzyme and one, and then with the second hydrogen peroxide molecule. The primary function of catalase in the cell is the breakdown of hydrogen peroxide (Zhukova et al., 2016). Catalase is present in the blood, bone marrow, mucous membranes, liver, and kidneys (Varkholiak et al., 2020). In many tissues, including kidneys, microbodies and peroxisomes are rich in aerobic dehydrogenases and catalase (Antonjak et al., 2000).

The primary method of combating helminthiasis in animals is pharmacotherapy with anthelmintic drugs. However, most of them in therapeutic doses are immunosuppressants and suppress the body's antioxidant status. Helminths, in turn, also cause a disturbance in the balance between radical formation and the activity of the antioxidant system (Zhuravlov et al., 2016). Therefore, the problem of determining the effectiveness of the use of anthelmintics, optimizing their doses, and using antioxidants to suppress the processes of lipid peroxidation in dewormed animals remains relevant, and the development of complex measures to combat helminthiasis is a crucial task; the solution of which will determine the effectiveness of the therapy of infected animals.

Therefore, it is urgent to search for drugs to reduce the negative consequences of intoxication of the animal body with helminths and their rapid rehabilitation after deworming.

Aim of the research

To study the effect of the feed additive "Sylimevit" on the antioxidant protection of the body of dogs after preventive deworming.

Materials and Methods

The research was conducted following the “General Ethical Principles of Animal Experiments” (Ukraine, 2001), which is consistent with the provisions “On the Protection of Animals from Cruelty” and the provisions of the “European Convention on the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes” (Strasbourg, 1985).

10 German Shepherd dogs aged 1–2 years were used for experimental research. Dogs were given prophylactic deworming against mixed infestations with the drug “Brovanol D”, once at 1 g of powder per 10 kg of body weight. The anthelmintic was fed to the dogs with 1/3 of the morning ration. Blood for research was taken from experimental animals from the subcutaneous vein of the forearm before deworming and 20 and 30 days after it. Two groups of dogs with five animals each were formed. Dogs that were subjected to prophylactic deworming served as controls. The experimental group of animals was also dewormed with the drug “Brovanol D” and, starting from the first day, additionally received feed daily (once a day) for 30 days, the feed additive “Sylimevit”.

Enzyme activity was determined in the blood serum of dogs of the control and experimental groups, namely: catalase activity (CT; K.F. 1.11.1.6) – according to the method of M. A. Koroliuk (1988); activity of superoxide dismutase (SOD; K.F. 1.15.1.1) – according to the method of E. E. Dubinina et al. (1983). In addition, the content

of TBC-active products was investigated – according to the method of E. N. Korobeinikov (1989), the level of diene conjugates (DC) – according to the method of I. D. Stalna (1977) (Vlizlo, 2012).

Results and Discussion

As a result of the conducted research, the activity of the enzyme link of antioxidant protection in experimental dogs before deworming was somewhat lower than the limits of physiological indicators in healthy dogs. It was established that the activity of catalase in the blood serum of experimental animals at the beginning of the experiment ranged from 0.15 ± 0.05 mg H₂O₂ (Fig. 1). In contrast, the activity of SOD was 15.1 ± 0.62 c.u./mg protein respectively (Fig. 2). Later, in the dogs of the control and experimental groups, after deworming on the 20th and 30th day of the experiment, an increase in the activity of the indicated enzymes was observed. At the same time, it is worth noting that the use of the feed additive “Sylimevit” in the period after deworming contributed to better activation of the enzyme link of the antioxidant system, as indicated by the high activity of catalase and superoxide dismutase in the blood serum of the dogs of the experimental group on the 30th day of the experiment, where compared to the control group of dogs, catalase activity increased by 35.2 %, and superoxide dismutase activity by 24.0 %, respectively.

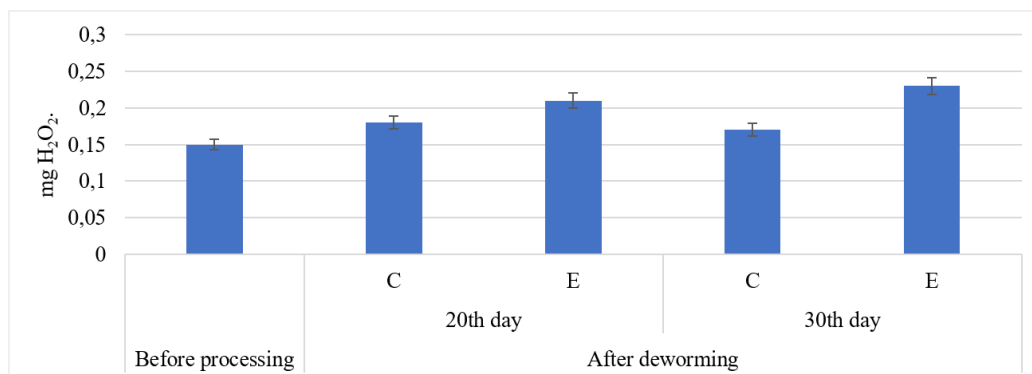


Fig. 1. Catalase activity in the blood of dogs under the influence of the feed additive “Sylimevit”

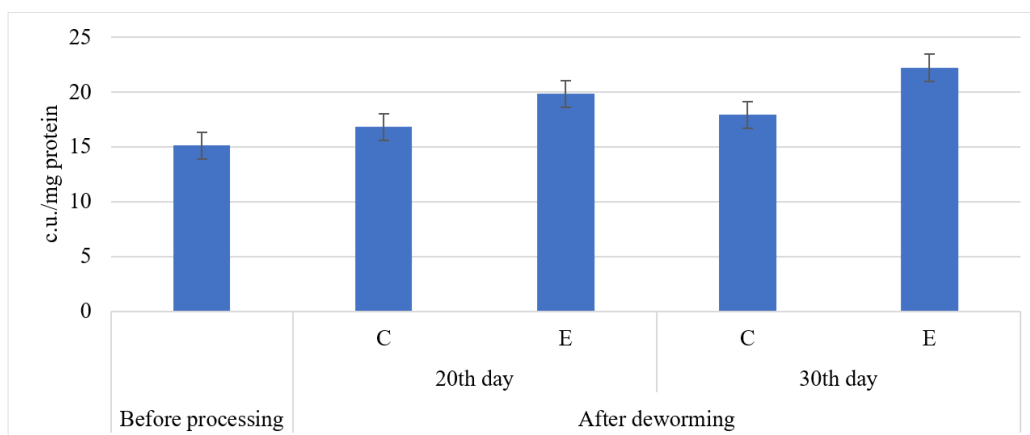


Fig. 2. The activity of superoxide dismutase in the blood of dogs under the influence of the feed additive “Sylimevit”

Peroxide oxidation of lipids in the body of animals is a normal physiological process. Mitochondrial membranes maintain a certain level of LPO, which has essential functional significance and reflects the degree of influence of molecular oxygen on mitochondrial lipids under normal physiological conditions. At the same time, the role of peroxide processes is determined by their ability to regulate the structural and functional state of membranes, which is crucial for the functioning of enzyme systems. The results in Figures 3 and 4 show that the dogs of the control group had a high level of intermediate and end products of lipid peroxidation before deworming.

After deworming the dogs of the control group on the 20th day of the experiment, a slight decrease in the content of diene conjugates and TBC-active products was established. In contrast, compared to the beginning of the investigation, their level decreased by 9.7 and 17.5 %, respectively. On the 30th day of the experiment, in dogs of the control group, the decrease in the level of secondary and end products of lipid peroxidation continued. Still, the indicators did not reach physiological values. They were only using the feed additive “Sylymevit” after

deworming contributed to the suppression of radical formation processes. Thus, on the 20th day of the experiment, a decrease in TBC-active products by 38.0 % and the level of diene conjugates by 32.3 % was noted. On the 30th day of the investigation, the level of secondary and final products of lipid peroxidation in the blood of the dogs of the experimental group, which received the supplement “Sylymevit”, was the lowest. In general, our research results indicate that the feed additive “Sylymevit” in the dogs of the research group inhibited lipid peroxidation processes and activated the antioxidant defense system, as noted in the high activity of catalase and superoxide dismutase. This may be because the drug's composition includes milk thistle, methiphen, and vitamins, which in turn enhance the effect of each other and thereby inhibit the formation of radicals and lipid peroxidation processes. It is also worth noting the antioxidant properties of milk thistle, which according to the literature, also have similar properties. The complex effect of the indicated biologically active elements provided high hepatoprotective and antioxidant effects.

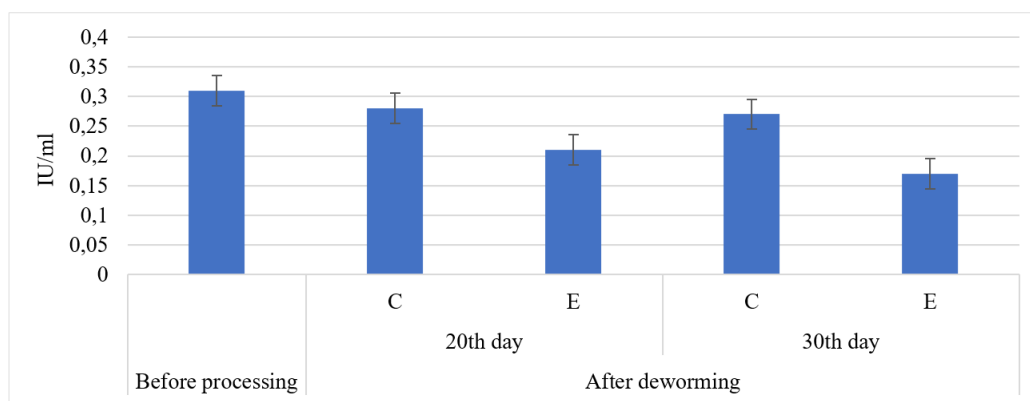


Fig. 3. The level of diene conjugates in the blood of dogs under the influence of the feed additive “Sylymevit”

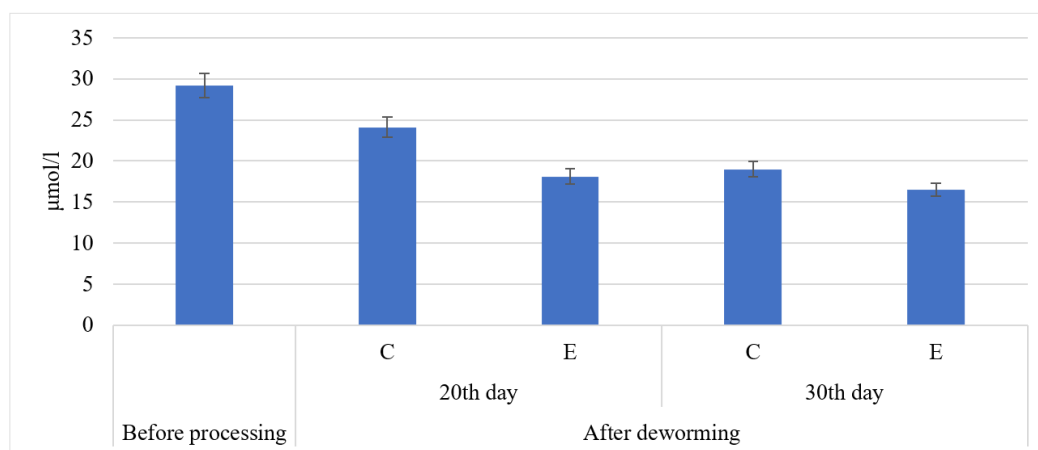


Fig. 4. The level of TBC-active products in the blood of dogs under the influence of the feed additive “Sylymevit”

Conclusions

It was established that when the supplement “Sylymevit” is used in dogs after deworming, the enzyme link of the antioxidant system is activated in the blood, as indicated by the increase in the activity of catalase and superoxide dismutase in the blood serum of these animals and

inhibition of the processes of lipid peroxidation (reduction of secondary and end products of peroxide lipid oxidation). On the 30th day of the experiment, the activity of catalase and superoxide dismutase in the blood of animals of the experimental group was the highest, and the level of lipid peroxidation products was the lowest. The ob-

tained research results confirm the antioxidant properties of the Sylimevit feed additive.

Conflict of interest

The authors declare that there is no conflict of interest.

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