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Prevention Of Violations Of The Functional Status Of Platelet Hemostasis In Newborn Calves With Functional Disorders Of The Digestive System.

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ABSTRACT

The newborn period in calves is the period of greatest vulnerability, when functional disorders of the digestive system can occur and platelet functions are activated with deterioration of microcirculatory processes and the frequent development of fatal thrombosis in them. For the development of approaches to the effective prevention of functional disorders of the digestive system and the simultaneous weakening of the functional activity of platelets in calves, a new highly biologically active drug, phosphopag (polyhexamethyleneguanidine phosphate), was of great scientific and practical interest. In newborn calves with a risk of functional digestive disorders, an increase in platelet aggregation functions was found in vitro and in vivo. These disorders are based on congenital changes in the lipid composition of platelet membranes, an increase in the level of middle molecules in plasma and blood plates, activation of lipid peroxidation in them, increased synthesis in the vascular wall of von Willebrand factor and intensification of thromboxane formation in blood plates. The purpose for the purpose of a prophylactic complex of phosphopag, ekos and calcium gluconate in calves with a risk of functional digestive disorders fully ensured the normalization of lipid peroxidation and the content of middle molecules in plasma and platelets. The use within 8 days of phosphopag, ekos and calcium gluconate in newborn calves with a risk of functional disorders of the digestive system, normalizes the state of the estimated indicators of primary hemostasis, optimizing platelet aggregation and intravascular activity.

Keywords: platelets, newborn calves, functional disorders of the digestive system, phosphopagus, ekos, calcium gluconate.

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INTRODUCTION

The neonatal period in calves is the most vulnerable period when functional disorders of the digestive system can occur [1-5] and platelet functions are activated [6.7] with deterioration of microcirculatory processes and frequent development of thrombosis in them [8] with a fatal outcome [9]. Activation of primary hemostasis in calves that are at risk of developing functional disorders of the digestive system [10] may be caused by increased lipid peroxidation (LPO) in animals [11,12], which can be used to predict increased intra-platelet mechanisms [13,14] with increasing aggregation blood plates [15,16]. At the same time, approaches to effective prevention of functional disorders of the digestive system with simultaneous weakening of the functional activity of platelets in calves with full normalization of microcirculation have not yet been developed [17,18].

Of great scientific and practical interest is a new highly biologically active drug - phosphopag (polyhexamethylene guanidine phosphate), which has the ability to arrest functional disorders of the digestive system in newborn calves. Extensive studies are related to the study of the properties of the economically available sorbent Ecos (hydroaluminosilicate). Calcium gluconate can be used to stimulate gastric secretion in calves. It was suggested that the effective correction of platelet dysfunction in newborn calves with functional disorders of the digestive system when using a combination of phosphopag, ekos and calcium gluconate.

The goal is set in the work: to establish the possibility of preventing the development of functional disorders of the digestive system and the activation of platelet functions in newborn calves with the simultaneous use of phosphopagus, ecos and calcium gluconate.

MATERIALS AND METHODS

Research was conducted in strict accordance with ethical principles established by the European Convent on protection of the vertebrata used for experimental and other scientific purposes (adopted in Strasbourg March 18, 1986, and confirmed in Strasbourg June 15, 2006) and approved by the local ethic committee of Russian State Social University (Record №12 dated December 3, 2015).

The study included 29 newborn calves 2-3 days of life from the herd, dysfunctional in functional digestive disorders. The control group consisted of 267 healthy newborn calves. Mandatory examinations included the determination of plasma LPO by the concentration of thiobarbituric acid-active compounds with the Agat-Med kit. Conducted a study of the antioxidant potential of the liquid part of the blood. Intra-platelet lipid peroxidation was assessed by the concentration of the basal level of malondialdehyde (MDA) in the reduction reaction with thiobarbituric acid. They found out the level of medium molecules (SM) in the plasma and washed, resuspended platelets. The platelet counts in capillary blood in the Goryaev chamber were counted. Platelet aggregation (AP) was assessed by visual micromethod with ADP inductors (0.5×10^{-4} M), collagen (dilution 1: 2 of the main suspension), thrombin (0.125 units/ml), ristomycin (0.8 mg / ml), adrenaline (5×10^{-6} M), as well as combinations of ADP and adrenaline, ADP and collagen, adrenaline and collagen, allowing to simulate the real conditions of blood flow. Intravascular platelet activity was recorded using a phase contrast microscope.

All 29 calves entered the study were prescribed with a prophylactic purpose 0.01% phosphopagus 100.0 ml each in the morning, 10% 10.0 calcium gluconate at lunch and ekos 150 mg / kg body weight in the evening for 8 days, included in the feeding scheme. Statistical processing of the results obtained by the t-student criterion.

RESULTS

In the examined newborn calves, predisposed to the development of functional disorders of the digestive system, an increase in the LPO was noted. Thus, the level of thiobarbituric acid-active products in plasma reached 4.36±0.03 µmol/l (in the control - 3.92±0.06 µmol/l). The antioxidant ability of plasma calves at risk of developing functional disorders of the digestive system was reduced (23.7±0.09%), against control - 28.6±0.04%. The content of MDA in the bloodstream was increased (1.22±0.006 nmol/10^9 platelets) at a control level of 0.89±0.02 nmol/10^9 platelets), which indicated the activation of free-radical oxidation in them due to depression of intraplatelet antioxidant activity. At the same time, the content of medium molecules in

plasma and platelets was increased - Medium molecules 280 - 0.43±0.08 conventional units, Medium molecules 254 - 0.29±0.02 conventional units, and Medium molecules 280 - 0.055±0.03 conventional units/10^9 platelets, Medium molecules 254 - 0.061±0.04 conventional units/10^9 platelets, respectively, significantly exceeding the level of control.

The prophylactic use of calves with the possibility of functional digestive disorders of the combination of phosphopagus, ekos and calcium gluconate made it possible to eliminate the manephistination of this condition in them, reducing the plasma LPO and platelets to their normal level. At the same time, the level of thiobarbituric acid-active plasma products decreased ($p<0.01$); on the 8th day of correction, their concentration reached 3.92±0.06 µmol/l. Simultaneously with a decrease in the activity of lipid peroxidation in plasma, normalization of the average molecules of 280 to 0.32±0.02 conventional units was achieved, the average molecules of 254 - 0.23±0.04 conventional units. The weakening of the LPO plasma proceeded in parallel with the decrease in the basal level of MDA in platelets after the 8th day of treatment (0.91±0.02 nmol/10^9 platelets). With the combined appointment of calves phosphopagus, ekos and calcium gluconate normalized in platelets level Average molecules 280 - 0.050±0.06 conventional units/10^9 platelets, Average molecules 254 - 0.053±0.02 conventional units/10^9 platelets.

The content of platelets in the blood of calves predisposed to functional disorders of the digestive system before and after treatment was within the normal range. In calves with a risk of functional disorders of the digestive system, acceleration of antibodies was recorded before the start of correction, mainly under the influence of collagen (27.8±0.06 s). Slower AP occurred in calves with functional digestive disorders under the influence of ADP (34.3±0.02 s) and ristomycin (32.6±0.07 s). Thrombin (48.6±0.03 s) and adrenaline (85.2±0.06 s) AP developed later, but faster than in the control ($p<0.01$). The time of AP development using physiological combinations of inductors was also shortened (ADP + adrenaline - 26.2±0.03 s, ADP + collagen - 23.9±0.06 s, adrenaline + collagen –25.3±0.02 s).

By the end of the prophylactic use of phosphopag, ekos and calcium gluconate, the time of AP increased under the action of all inductors and their combinations. By the 8th day of correction, the most active inducer of AP remained collagen (29.6±0.03 s). Later, AP developed on ADP (38.8±0.02 s), ristomycin (39.9±0.03 s), and even later developed AP under the influence of thrombin and adrenaline. The time of AP was prolonged when the inductors were combined (ADP + adrenaline - 35.8±0.02 s, ADP + collagen - 27.3±0.08 s, adrenaline + collagen - 29.8±0.02 s).

Intravascular platelet activity of calves with a risk of functional disorders of the digestive system was increased. The content of discocytes in the bloodstream predisposed to functional disorders of the digestive system of calves was 70.9±0.3% (in the control - 82.0±0.16%). The level of disco-echinocytes was increased to 12.9±0.06%. The content of spherocytes and sphero-echinocytes significantly exceeded the control level (9.6±0.05% and 5.5±0.07%, respectively). The sum of active forms of blood platelets (29.1±0.03%) in calves with a predisposition to functional disorders of the digestive system exceeded control by 1.62 times. The level of small and large aggregates exceeded the standard values 2.25 and 20.6 times, respectively, and the content of platelets in the aggregates in animals with functional digestive disorders exceeded the control 2.3 times.

Prophylactic correction of functional digestive disorders and thrombocytopathy with a combination of phosphopag, ekos and calcium gluconate calves provided a normalization of the level of intravascular platelet activity, so by the end of the 8 day use of the used means a significant improvement in the intravascular platelet activity was detected. The level of discoid forms of platelets in the blood of newborn calves as a result of the use of phosphopag, ekos and calcium gluconate reached 81.2±0.6%. The content of disco-echinocytes, spherocytes and sphero-echinocytes in the blood of animals significantly decreased under the influence of the chosen correction (10.5±0.02%, 4.2±0.06% and 2.6±0.3%, respectively). At the same time, the sum of the active forms of platelets against the background of the use of a combination of phosphopagus, ekos and calcium gluconate (18.8±0.06%) reached the level of control. Under these conditions, the number of small and large units by the 8th day of correction significantly decreased 2.25 and 20.6 times, correspondingly equaling the same control indicators with platelet involvement in the aggregates 4.5±0.02%.

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DISCUSSION

The increase in LPO in plasma and platelet calves with the risk of functional disorders of the digestive system indicates a congenital decrease in the antioxidant system of their body [17–20], which suggests an increase in the level of medium molecules in plasma [21] and platelets [22]. Normalization of the level of peroxidation and antioxidant plasma activity with a simultaneous decrease in the average molecules as a result of prophylactic correction indicates a pronounced normalizing effect of combined use of phosphopagus, ecos and calcium gluconate on homeostasis in newborn calves with the likelihood of functional digestive disorders [23-26]. The effects of this complex are mediated by the effect of each of the drugs on the metabolism and the expression level of the enzymes of the antioxidant system of the body [27-30].

Normalization of all estimated hemostasis indicators in calves against the background of prophylactic use of phosphopagus, ecos and calcium gluconate indicates its normalizing effect on the mechanisms of platelet hemostasis in newborn calves with the risk of functional disorders of digestion [31-35]. Obviously, this is due to the optimization of metabolic processes, depression of the toxic effect of POL and medium molecules in plasma and platelets with normalization of the reception of exogenous signals by blood plates [36]. The aggregation of platelets in newborn calves with functional disorders of the digestive system at the appointment of an 8-day period of phosphopag, ekos and calcium gluconate has reached the level of control [37-39].

The normal duration of AP when using ristomycin in newborn calves in the prophylactic use of phosphopag, ecos and calcium gluconate indicates an optimization of the concentration of Willebrand factor in the blood of the adhesive molecule [40-45].

Reaching the normal level of intravascular platelet activity by correcting phosphopagous, eco-ecos and calcium gluconate in newborn calves with a risk of functional disorders of the digestive system minimizes microcirculatory disorders and the risk of thrombotic complications [46-50]. Taking into account the high efficiency of the correction of platelet hemostasis in newborn calves with a risk of functional disorders of the digestive system with the use of a prophylactic approach, it can be recommended for widespread use in livestock farms [51-53].

CONCLUSION

Purpose for the purpose of a prophylactic complex of phosphopag, ekos and calcium gluconate in calves with a risk of functional disorders of the digestive system fully normalizes lipid peroxidation and the content of medium molecules in plasma and platelets. The use within 8 days of phosphopag, ekos and calcium gluconate in newborn calves with the possibility of functional digestive disorders normalizes the state of the estimated indicators of primary hemostasis, optimizing platelet aggregation and intravascular activity.

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