ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Elimination of platelet dysfunctions in newborn calves with functional digestive disorders.

Zavalishina S Yu*.

Russian State Social University, st. V. Pika, 4, Moscow, Russia, 129226.

ABSTRACT

The occurrence of functional disorders of the digestive system in newborn calves often leads to deterioration of the blood rheology due to the activation of platelets against the background of the activation of lipid peroxidation and an increase in the blood content of middle molecules. Platelet dysfunctions in them contribute to the deterioration of microcirculation in the organs of animals due to active intravascular thrombus formation. In newborn calves with functional digestive disorders, an increase in platelet aggregation functions was found in vitro and in vivo. These disorders are based on deep changes in the lipid composition of platelet membranes, an increase in the content of medium molecules in the plasma and blood plates, the activation of lipid peroxidation in them, the intensification of thromboxane formation in blood plates, which is developing on this background. Activation of thromboplastin formation is the leading cause of increased blood coagulation in newborn calves with functional disorders of the digestion. It was possible to identify the possibility of correcting disorders of platelet hemostasis with the help of a complex from Phosphopag and Ekos, which can reduce the level of medium molecules in the body and correct lipid peroxidation. In addition, the use of Phosphopagus and Ekos in newborn calves with functional digestive disorders reduces the severity of platelet hemostasis disorders by approaching the rate of increased platelet aggregation and intravascular activity of platelets.

Keywords: platelets, newborn calves, functional disorders of the digestive system, Phosphopag, Ecos.

Corresponding author

ISSN: 0975-8585

INTRODUCTION

The occurrence of functional disorders of the digestive system in newborn calves often leads to deterioration of the blood rheology due to the activation of platelets against the background of the activation of lipid peroxidation (LPO) and the increase in the blood content of middle molecules [1,2,3]. At the same time, platelet dysfunctions in them contribute not only to the deterioration of microcirculation in the tissues and organs of animals [5-10], but also to intravascular thrombosis [11,12]. At the same time, the enhancement of platelet function in calves with functional disorders of the digestive system and effective approaches to their leveling are poorly understood [13,14].

Currently, the drug Phosphopag (polyhexamethylene guanidine phosphate) is being widely studied at the Institute of Ecological and Technological Problems in Moscow, which is able to effectively arrest the manifestations of functional disorders of the digestive system in newborn calves. It was also established that, in case of functional disorders of the digestion, calves are assigned sorbents, a prominent representative of which is the hydroaluminosilicate preparation “Ecos”. The authors suggested that it is possible to correct platelet dysfunctions in newborn calves with functional digestive disorders using a combination of the Phosphopag and Ecos preparations.

In this regard, the aim of the work was to assess the possibilities of correcting platelet dysfunctions in newborn calves with dyspepsia using Phosphopag and Ecos.

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 took 27 newborn calves with functional disorders of the digestive system. In animals with functional digestive disorders, all signs of indigestion with bright intoxication were noted. The control group consisted of 267 healthy newborn calves. The examination consisted in determining the activity of plasma lipid peroxidation (LPO) was determined by the content of thiobarbituric acid-active products using the Agat-Med kit. The antioxidant potential of the liquid part of the blood was determined traditionally. Intra-platelet lipid peroxidation was determined by the concentration of the basal level of malondialdehyde (MDA) in the reduction reaction with thiobarbituric acid. In plasma and washed and resuspended platelets, the level of medium molecules was assessed. Counting platelet count in capillary blood was performed in the Goryaev chamber. Platelet aggregation activity (AP) was studied by a visual micromethod using as inducers ADP (0.5×10^{-4} M), collagen (dilution 1: 2 of the main suspension), thrombin (0.125 U/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 to simulate real blood flow conditions. Intravascular platelet activity was determined visually using a phase contrast microscope. All 27 sick calves were prescribed in the morning 0.01% Phosphopag 100.0 ml each and Ekos solution 150 mg / kg in the evening for 10 days, including in the feeding scheme. Statistical processing of the results obtained was carried out using Student's t-test.

RESULTS

In experimental newborn calves with functional disorders of digestion, an increase in lipid peroxidation products - thiobarbituric acid-active compounds up to 5.06±0.11 µmol/l was established in plasma. (in the control - 3.92±0.06 µmol/l). The antioxidant activity of the plasma of animals with functional digestive disorders was reduced by (21.0±0.03%) compared with the control - 28.6±0.04% ($p < 0.01$). The level of MDA in platelets was increased to 1.65±0.002 nmol/10^9 platelets (control 0.89±0.02 nmol/10^9 platelets, $p<0.01$), which indicated an increase in their free-radical oxidation as a result of weakening intraplatelet antioxidant activity. At the same time, an increase in the level of medium molecules in a plasma was established. Medium molecules 280 - 0.54±0.02 conventional units, Medium molecules 254 - 0.33±0.06 conventional units and platelets. Medium molecules 280 - 0.063±0.06 conventional units/10^9 platelets, Average molecules 254 - 0.070±0.04 conventional units/10^9 platelets, significantly exceeding control values ($p<0.01$).

Appointment of calves with functional digestive disorders "Phosphopag" and "Ecos" reduced the activity of the LPO plasma and platelet calves. As a result of the correction, the content of thiobarbituric acid-active plasma products decreased ($p < 0.01$), which after 10 days of treatment amounted to 3.97±0.05 µmol/l. At the same time, a reduction in the level of POL in plasma was achieved with a decrease in the average molecules of 280 to 0.33±0.06 used units, the average molecules of 254 - 0.24±0.02 used units. In the platelets of calves that underwent a course of correction, a decrease in the level of MDA was also found (after 10 days of correction, 0.92±0.02 nmol/10^9 platelets). Against the background of therapy with the combination of "Phosphopag" and "Ecosome", it was also possible to achieve a decrease in platelet content. Average molecules 280 - 0.051±0.02 conventional units/10^9 platelets, Average molecules 254 - 0.056±0.03 conventional units/10^9 platelets ($p < 0.01$).

The content of platelets in the bloodstream of calves with functional digestive disorders before and after correction remained within the normal range. In animals with functional disorders of the digestive system, before the correction was prescribed, the reduction of AP time was found to a greater extent for collagen (21.0±0.08 s), slower than AP developed in newborn calves with functional digestive disorders for ADP (35.0 ± 0.12 s) and ristomycin (30.0±0.06 s). Later, AP appeared with thrombin (43.5±0.20s) and adrenaline (81.0±0.02 s), occurring faster than in the control ($p<0.01$). When the combination of AP inductors developed significantly before the control ($p<0.01$) - ADP + adrenaline - 21.0±0.06 s, ADP + collagen - 21.0±0.02 s, adrenaline + collagen - 20.5±0.06c.

By the end of the course of application of "Phosphopagus" and "Ecos", AP was slowed down under the influence of all inductors, so collagen was the most active inducer of AP (29.0±0.02s), it was inferior in ADP activity (39.0±0.16s) and ristomycin (40.0±0.15s). Even more slowly, AP occurred for thrombin and adrenaline. When the inductors were combined, a significant lengthening of AP time was achieved (ADP + adrenaline - 33.0±0.02s, ADP + collagen - 26.0±0.04 s, adrenaline + collagen - 28.0±0.02 s, ($p<0.01$)) approaching control values.

The intravascular activity of platelets in calves with functional disorders of the digestive system was increased. At the same time, the discocytes in the bloodstream of calves with functional digestive disorders were reduced to 62.6±0.09% (in the control - 82.0±0.16%) with an increase in the bloodstream of disco-echinocytes in 1.63 times, spherocytes (13.0±0.04%) and sphero-echinocytes (6.2±0.05%) with the sum of active forms of platelets (37.4±0.07%), which exceeded the control 2.07 times. In newborn calves with functional digestive disorders, the level of small and large aggregates in the free circulation exceeded 4.44 times and 45.00 times control, respectively, with the level of platelet involvement in the aggregates that prevailed over control 2.76 times.

The correction in newborn calves with functional disorders of the digestive system caused a decrease in the intravascular activity of platelets. By the end of the correction by Phosphopagom and Ecosome, the number of discoid forms of platelets in the blood of animals increased to 80.1±0.2% ($p<0.01$), which was combined with a decrease in the level of disco-echinocytes, spherocytes and sphero-echinocytes significantly to 10.9±0.02%, 4.8±0.06% and 2.9±0.3%, respectively ($p<0.01$). The total content of active forms of platelets in the correction of "Phosphopag" and "Ecosome" (19.9±0.02%) also approached the values of the control. The concentration of small and large aggregates in the bloodstream of calves with functional disorders of the digestive system that underwent correction significantly decreased by 3.40 and 7.71 times, respectively ($p<0.01$) while reducing the involvement of blood plates in the aggregates from 13.8±0.06% to 5.6±0.02% ($p <0.01$).

DISCUSSION

In newborn calves with functional digestive disorders, there is an increase in POL in platelets and platelets [15,16], which is associated with a decrease in the activity of the antioxidant system in their bodies [17,18], causing an increase in average molecules levels in plasma and platelets [19]. The purpose of these animals to correct the combination of "Phosphopagus" and "Ecos" causes a decrease in peroxidation [20-25], an increase in the antioxidant potential of the plasma with a decrease in the level of middle molecules [26-29]. These results indicate a pronounced optimizing effect of the carried out correction [30] on the homeostasis of the body in newborn calves with functional disorders of the digestive system, apparently mediated by its effect on metabolism and the increased activity of antioxidant defense enzymes in their body [31-37].

Significant positive dynamics of the studied hemostasis parameters in calves with correction by the combination of "Phosphopagus" and "Ecos" indicates their positive effect on the system for the implementation of platelet functions in newborn calves [38-42]. This is due to the increased metabolic processes, the weakening of the toxic effects of POL and medium molecules in the plasma and the optimization of the interaction of exogenous signals from the outside to the platelet receptors [43-44]. The assessment of platelet hemostasis determined in calves after 10 days of correction by Phosphopagom and Ecosome established that they were closer to the values of the control group [45].

The elongation of antibodies under the action of ristomycin in newborn calves with functional disorders of the digestive system after applying Phosphopag and Ecos indicates a decrease with the approach to control of the concentration in the blood of the adhesive molecule, von Willebrand factor, while reducing the sensitivity of blood platelets to it [46,47].

The high degree of attenuation of the intravascular activity of platelets in newborn calves with functional disorders of digestion, registered during the correction by Phosphopagom and Ecosome, allows them to significantly optimize their microcirculation processes in tissues, reducing the risk of thrombotic complications [48-51]. Taking into account the completeness of the correction of platelet hemostasis disorders by the combination of "Phosphopagus" and "Ecos" in newborn calves with functional disorders of the digestive system, it can be recommended for wider use in livestock farms [52,53].

CONCLUSION

Simultaneous use of "Phosphopagus" and "Ekos" to calves with functional digestive disorders can significantly reduce the severity of peroxidation and bring the average molecules in their blood plasma and platelets closer to normal. Correction of "Phosphopagam" and "Ecosome" of newborn calves with functional digestive disorders significantly optimizes disorders of platelet hemostasis due to the approach to the rate of increased platelet aggregation and intravascular activity of blood platelets.

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