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Functional Features of Hemostasis in Weakened Newborn Calves Treated with Aminosol

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ABSTRACT

With many negative changes in the body, the development of hemostatic system activity disorders with the formation of thrombophilia is possible. The high frequency of occurrence of physical weakening of newborn calves creates a risk for them to activate the hemostatic system, which requires a search for approaches to eliminate the asthenic state and optimize the activity of hemostasis. The study examined 34 weakened newborn calves that were obtained from first-born cows. To correct their condition, the calves were given aminosol in a generally accepted dose. Initially, animals had an increase in antioxidant protection of blood plasma with a decrease in the severity of lipid peroxidation processes in it. Weakened calves showed a high activity of the coagulation system of blood and platelets and decreased hemostatic capabilities of the vascular wall. As a result of the use of aminosol in weakened newborn calves, there was a significant increase in the activity of their antioxidant system, leading to the containment of lipid peroxidation processes in their blood, reduced functional readiness of platelet and plasma hemostasis and increased hemostatic capabilities of their vessels. Perhaps the continued use of this drug can lead to the normalization of the considered indicators at a longer observation time..

KEY WORDS: HEMOSTASIS, NEWBORN CALVES, AMINOSOL, PLATELETS, BLOOD VESSELS..

INTRODUCTION

Modern conditions of animal husbandry increasingly dictate the need for its intensification (Vorobyeva and Medvedev, 2020a; Glagoleva and Medvedev, 2020). However, great attention must be paid to the productive capabilities of first-calf heifers. Not in all cases, first-calf cows are capable of producing highly viable offspring. This is largely due to their insufficient body weight and sometimes early insemination (Oshurkova and Medvedev, 2018a). For this reason, calves obtained from them often have hypotrophy, negatively acting in them on metabolism, and therefore on growth in their body weight (Glagoleva and Medvedev, 2018).

It is known that with so many negative changes in the state of the animal's body, a disturbance in the activity of the components of the hemostatic system, causing the formation of thrombophilia, is possible (Vorobyeva and Medvedev, 2018; Mal et al., 2018a). A sufficiently high frequency of occurrence of weakening of the physical condition in newborn calves provides them with a risk of a high frequency of occurrence of episodes of activation of components of the hemostasis system (Vorobyeva and Medvedev, 2019; Vorobyeva and Medvedev, 2020b) with poor study of the potential of available approaches to eliminate the asthenic state with regard to the effect on hemostasis activity.

Of great scientific and practical interest is the assessment of the effect on weakened newborn calves of various

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means of increasing their viability, taking into account the severity of the effect on hemostasiopathy taking place against the background of physical weakening. Of particular interest is the assessment of the effect of the aminosol preparation, which is often used in newborn calves born to weakened, on the parameters of the hemostatic system. In the present study, the goal was set to assess the nature of the effect of the aminosol agent on the hemostatic system in weakened newborn calves.

MATERIAL AND METHODS

This work was carried out in accordance with the ethics defined by the European Convention for the Protection of Vertebrates, which is used for scientific purposes (adopted at Strasbourg on 18 March 1986 and confirmed at Strasbourg on 15 June 2006). Material for the article was obtained during a survey of 34 newborn calves born weakened, but with normal body weight. The control group consisted of 25 healthy newborn calves. The animals were examined to determine the level of lipid peroxidation in plasma, which was estimated by the amount of thiobarbituric acid-active products in their plasma using the Agat-Med kit (Russia) and the level of acylhydroperoxides when registering the plasma antioxidant potential blood (Volchegorsky et al., 2000). The platelet content in the blood of animals was determined using a Goryaev's camera. The expressiveness of the aggregation of platelets (AP) was defined, applying a visual micromethod of its registration (Shitikova, 1999) with a number of inductors: ADF (0.5×10-4 M), collagen (cultivation 1:2 main suspensions), thrombin (0.125 pieces/ml), ristomitsiny (0.8 mg/ml), adrenaline (5×10-6 M) in plasma rich with platelets after it was standardized on the maintenance of platelets to concentration 200×109 of platelets in one liter.

The ability of vessels to inhibit platelet aggregation was recorded by applying a time venous occlusion sample (Baluda, 1987) based on an AP evaluation in visual micrometode (Shitikova, 1999) to all inductors used. The value of indices of anti-aggregation activity of vascular wall with respect to all inductors used in operation was determined. For this purpose, the value of the time of development of AP in plasma, which was obtained under conditions of temporary venous stagnation for the time of development of AP in plasma obtained outside it, was divided. The index of anticoagulation activity of the vessel wall was calculated by dividing the activity of antithrombin III in plasma (Barkagan, 1999) taken using a venous occlusion sample by the value of its activity before it in plasma obtained without applying a cuff to the vessel (Baluda, 1987). Vascular control of fibrinolysis was determined by calculating the value of the fibrinolytic activity index of the vascular wall. This was done by dividing euglobulin lysis index (Barkagan, 1999) in intact plasma by its index in plasma taken after temporary vascular occlusion (Baluda, 1987).

Coagulation hemostasis was evaluated by the values of activated partial thromboplastin time, prothrombin time and thrombin time (Barkagan, 1999). Correction of functional indices in weakened newborn calves was carried out with aminosole agent (manufactured by "Biofactors," Czech Republic) at a dose of 8 ml/day, used in the form of evaporation per head for 8 days. Determination of all taken into account parameters in control animals was carried out once, in weakened animals - twice - at the moment of taking into study and on the next day after completion of correction. Statistical processing of the obtained data in the work carried out was carried out using the Student 's t-criterion.

RESULTS

In the outcome, weakened calves showed signs of weakness, sluggishness under reduced interest in all elements of reality. In the plasma, the weakened animals had a higher content of acyl hydroperoxide (3.42±0.19 D233/1 ml) and products capable of reacting with thiobarbituric acid (5.02±0.16 μmol/l) under depression of the antioxidant possibility of their plasma (23.2±0.34%). These parameters in the control calves were 1.42±0.07 D233/1 ml, 3.48±0.10 μmol/l and 34.2±0.26%, respectively. The number of platelets in the blood of the weakened calves was within the limits of the generally accepted norm (table 1). The development time of AP in these calves was significantly reduced. Previously, AP occurred in them under the action of a collagen inductor (accelerated with respect to the control level by 69.1%), a little later when using an ADP inductor (accelerated with respect to the control level by 64.3%) and a ristomycin inductor (accelerated with respect to the control level by 44.3%). AP in response to thrombin developed even later (accelerated relative to control level by 45.9%). The most delayed in weakened calves AP developed under the influence of adrenaline (accelerated compared to control by 44.9%).

In weakened young animals, a decrease in the levels of the indices of antiaggregation activity of the vascular wall was found for all applied inducers. The index of anti-aggregation activity of the vascular wall with collagen turned out to be the lowest, the index of anti-aggregation activity of the vascular wall with adrenaline and thrombin was slightly higher, and the index of anti-aggregation activity of the vascular wall against ADP and ristomycin was even higher.

The vessels of weakened newborn calves showed a decrease in the ability to control coagulation by 19.6%, which was assessed by a decrease in their index of anticoagulation activity of the vessel wall. The weakening of their fibrinolytic activity of blood vessels amounted to 18.6%, judging by the magnitude of the index of fibrinolytic activity of the vascular wall. Physically weakened calves had high values of blood coagulation developing in external (37.5%), internal (43.0%) and final stage fibrin formation (20.3%) earlier than in the control group. Aminosol evaporation was accompanied in weakened calves by activation of their general condition and improvement of hematological indices recorded in operation. In aminosol-treated

calves, the plasma concentration of acyl hydroperoxides (to 1.65±0.25 D233/1 ml) and compounds capable of reacting with thiobarbituric acid (to 3.81±0.39 μmol/l) decreased by increasing the antioxidant properties of their plasma (to 29.8±0.07%).

The correction to the weakened calves provided them with a pronounced slowdown in the AP. This was manifested by the later platelet response of these calves to the addition to platelet-rich plasma of all the aggregation inductors tested in operation. In weakened calves, as a result of the correction, an increase in the values of the indices of antiaggregatory activity of the vascular wall in response to all applied inductors occurred. The smallest was the value of the index of antiplatelet activity of the vascular wall in the case of collagen. Other indices of antiplatelet activity of the vascular wall were higher, also having a tendency to approach the control. Weakened calves given aminosol showed an increase in vascular control over plasma hemostasis, judging by an increase in the anticoagulant activity index of the vessel wall by 14.3% and an increase in vascular control over fibrinolysis, as judged by an increase in the index of fibrinolytic activity of the vascular wall by 13.5%. As a result of the use of aminosol, the activated partial thromboplastin time was inhibited by 16.5%, which was accompanied by a slowdown of the prothrombin time by 25.0% and an inhibition of the development of thrombin time by 9.4%.

Table. Hemostatic parameters in weakened newborn calves treated with aminosol

Indicators	Aminosol, n=34, M±m		Control, n=25, M±m
	exodus	after correction	
Platelet aggregation with ADP, s	25.2±0.12	36.7±0.09 p_1<0.01	41.4±0.07 p<0.01
Platelet aggregation with collagen, s	19.4±0.19	28.5±0.11 p_1<0.01	32.8±0.09 p<0.01
Platelet aggregation with thrombin, s	36.8±0.08	47.1±0.14 p_1<0.01	53.7±0.12 p<0.01
Platelet aggregation with ristomycin, s	47.2±0.10	32.7±0.17 p_1<0.01	39.4±0.12 p<0.01
Platelet aggregation with adrenaline, s	96.4±0.07	66.5±0.15 p_1<0.01	84.9±0.18 p<0.01
Vascular wall anti-aggregation index with ADP, units	1.34±0.10	1.58±0.06 p_1<0.01	1.65±0.14 p<0.01
Index of antiplatelet activity of the vascular wall with collagen, units	1.27±0.09	1.50±0.03 p_1<0.01	1.59±0.05 p<0.01
Index of antiplatelet activity of the vascular wall with thrombin, units	1.30±0.10	1.48±0.07 p_1<0.05	1.54±0.08 p<0.01
Index of antiaggregatory activity of the vascular wall	1.32±0.08	1.48±0.06 p_1<0.05	1.53±0.06 p<0.05
Index of antiplatelet activity of the vascular wall with adrenaline, units	1.35±0.05	1.57±0.04 p_1<0.05	1.66±0.05 p<0.01
Index anticoagulant activity vascular wall, units	1.12±0.06	1.28±0.07 p_1<0.05	1.34±0.04 p<0.01
Index fibrinolytic activity vascular wall, units	1.18±0.04	1.34±0.06 p_1<0.05	1.40±0.11 p<0.01
Activated partial thromboplastin time, s	27.9±0.28	32.5±0.30 p_1<0.01	39.9±0.27 p<0.01
Prothrombin time, s	12.8±0.22	16.0±0.29 p_1<0.01	17.6±0.26 p<0.01
Thrombin time, s	14.8±0.25	16.2±0.19 p_1<0.05	17.8±0.17 p<0.01

Legend: p - significance of differences in hemostatic parameters between the control group and the initial state of weakened calves, p1 - significance of the dynamics of hemostatic indicators during correction.

DISCUSSION

Optimal growth and development of calves at the very beginning of ontogenesis is associated with low activity of their hemostasis (Mal et al., 2018b) [13]. The onset of asthenization in the newborn phase can lead animals to the weakening of the functioning of internal organs (Tkacheva and Medvedev, 2020) with the development of disorders in the operation of the hemostasis system (Vorobyeva and Medvedev, 2020c). They are based on a decrease in the level of antioxidant properties of plasma, leading to an increase in concentrations of lipid peroxidation products in newborn calves born weakened, damaging the structure of blood plates, vascular walls and liver, leading to hemostasiopathy (Bespalov et al., 2018a). High AP activity in weakened newborn calves indicated excessive activation of receptor and intracellular platelet activation mechanisms. The acceleration of the development of AP in response to ristomycin in animals indicated an increase in their sensitivity to Willebrand factor under astenia conditions (Karpov et al., 2018). The rapid onset of AP with ADP in weakened calves was based on the enhancement in their blood plates of the conversion of arachidonic acid into an aggregate enhancing the effect of this inductor, thromboxane (Makhov and Medvedev, 2018a).

In weakened calves, reduced anti-aggregation activity of vascular endothelium was detected in the work. Without a doubt, this situation is ensured by the depression of synthesis in the walls of prostacycline vessels and nitrogen oxide. Another important component of vasopathy development in weakened calves is depression of synthesis in vessels of substances with anticoagulant and fibrinolytic activity - antithrombin III and tissue activator plasminogen (Boldov et al., 2018). The reduction of prothrombin time in physically weakened calves was a consequence of the activation of plasma hemostasis. Apparently, at its core, this has the appearance of excess active thromboplastin in their blood. The shortening of the activated partial thromboplastin time was based on an increase in the activity of the internal clotting mechanism (Oshurkova and Medvedev, 2018b). The acceleration of fibrin formation in weakened newborn calves was evidenced by the reduction of thrombin time.

The use of aminosol in weakened newborn calves has resulted in improvement of the overall condition of observed animals. Its use reduced the intensity of lipid peroxidation in the weakened young, reducing its stimulation to free circulation platelets. The found inhibition of AP development in weakened calves with respect to all inductors in case of aminosol use was largely due to the weakening of lipid peroxidation developing against this background, facilitation of platelet receptor and post-receptor mechanisms (Stepanova et al., 2018; Bespalov et al., 2018b). The elongation of AP development time in response to ristomycin found against the background of aminosol application indicated a decrease in the blood of weakened Willebrand factor calves, which is a cofactor of adhesion (Makhov and Medvedev, 2018b).

The use of aminosol in weakened calves stimulated the possibilities of anti-aggregation, anticoagulant and fibrinolytic capabilities of their vessels, which can be clearly explained (Medvedev and Kumova, 2007; Bikbulatova, 2018a). This was apparently provided by activation in its use of synthesis in the vascular endotheliocytes of these animals prostacycline, nitric oxide, antithrombin III and tissue plasminogen activator molecules. The delay in prothrombin time revealed in the weakened animals treated with aminosol indicated optimisation of plasma hemostasis mechanisms along the external route and was caused by physiologically necessary reduction of thromboplastin synthesis triggering clotting (Medvedev and Gamolina, 2008). The inhibition of the initially accelerated activated partial thromboplastin time detected after aminosol application indicated a decrease in activity of the internal hemocoagulation mechanism in calves. Development of this in combination with inhibition of fibrin formation detected by thrombin time dynamics was a manifestation of physiologically justified reduction of hemocoagulation (Bikbulatova, 2018b; Tkacheva, 2020).

CONCLUSION

Physically weakened newborn calves have low antioxidant blood protection leading to growth of lipid peroxidation products in it. These changes contribute to increased platelet activity, hemocoagulation and reduced hemostatic properties of vessels. Under conditions of aminosol application in weakened newborn calves, the level of antioxidant potential of plasma was increased, which led to decrease of lipid peroxidation products in their blood, decrease of platelet activity level and hemocoagulation with growth of functional properties of vascular walls. It is possible that the continuation of the use of this drug can lead to normalization of the taken-into-account indicators within a longer period of observation. It is planned to test this assumption in future studies.

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