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Functional Features Of Platelets In Newborn Calves With Iron Deficiency.

Zavalishina S Yu*.

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

ABSTRACT

Iron deficiency anemia in neonatal calves, which is still common in Russian farms of various forms of ownership, still has great urgency. It is known that it leads to inhibition of the growth and development of animals, to a depression of their resistance, contributing to the attachment of various infections and often the death of the body. It has been established that in the iron deficiency state, the hemostasis can be intensified in the calf, which leads to a tendency to thrombogenesis. The development of anemia in calves inevitably weakens their growing body, affecting the condition of the blood cells. In many respects this occurs in newborn calves with iron deficiency anemia as a result of increased peroxide oxidation of plasma lipids while weakening its antioxidant protection. All this leads in newborn calves with iron deficiency anemia to increase the platelet aggregation ability in vitro and in vivo, which inevitably leads to the formation of micro-rheological disorders and the risk of thrombotic events.

Keywords: calves, plasma, iron deficiency, platelets, aggregation.

Corresponding author

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INTRODUCTION

The development of the economy of all countries of the world always goes in parallel with the growth of the food base [1,2,3]. In many regions of the world, the further increase in the volume of food production is associated with the intensification of animal husbandry [4]. The solution of this problem is possible provided that new physiological knowledge is actively obtained [5,6], including about large cattle for their use in practice [7,8]. It seems extremely important to further study various aspects of blood, including its uniform elements [9,10]. Particular attention in this regard, many researchers pay to platelets, which play a very important role in hemostasis and microcirculation [11-15].

Their condition in newborn calves, especially with various dysfunctions, is extremely poorly understood. Platelet activity in iron deficiency anemia in newborn calves deserves a great deal of attention, which is still a frequent occurrence in Russian farms of various forms of ownership. It is known that it leads to inhibition of growth and development of animals, depression of their resistance, facilitating the attachment of various infections and often the death of the organism [16-20].

It has been established that under various conditions of the calf organism, hemostasis processes can be intensified, which leads to a tendency to thrombogenesis. The development of anemia in calves inevitably weakens their growing body, affecting the condition of the formed elements of the blood [21].

Anemia in newborn calves retains its high relevance in modern conditions as a result of its prevalence and often complicated course. At the same time, it becomes clear that this condition is accompanied by disorders in the hemostasis system with insufficient investigation of the emerging platelet dysfunction in the genesis of anemia.

In this regard, the goal of this work is to establish the state of platelet aggregation in newborn calves with iron deficiency anemia.

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).

38 newborn calves with iron deficiency anemia with erythropoiesis disorders and signs of a decrease in the level of iron in their bodies (serum iron 12.2±0.24 μmol/l, siderocytes 1.8±0.19%, hemoglobin 94,9±0,31 g/l, erythrocytes 4,4±0,16x10^{12}/l). Control is represented by 29 healthy newborn calves.

The intensity of peroxide oxidation of plasma lipids was estimated from the concentration of thiobarbituric acid-active products by the Agath-Med (Russia) kit, acyl hydroperoxides and the antioxidant potential of the liquid portion of the blood. The determination of the concentration of platelets in the capillary blood was carried out in Goryaev's chamber. Aggregation of platelets was determined by a visual micromethod with a number of inducers: ADP (0.5×10^{-4} M), collagen (dilution 1: 2 of the basic suspension), thrombin (0.125 U/ml), ristomycin (0.8 mg/ml), epinephrine (5×10^{-6} M) and hydrogen peroxide (7.3×10^{-3} M), as well as combinations of ADP with epinephrine, ADP with collagen, epinephrine and collagen. Intravascular activity of thrombocytes was assessed with phase contrast. Statistical processing of the obtained data was carried out by Student's t-test.

RESULTS OF THE STUDY

In newborn calves with iron deficiency anemia, the general condition of anemia, characteristic of anemia, has been revealed in animals: weakness, lethargy, lack of interest in the environment, pallor of the nasal mirror and visible mucous membranes.

In animals with iron deficiency anemia, a high activation of free radical lipid oxidation in the liquid part of the blood was noted (acyl hydroperoxide 3.44 ± 0.25 D_{233}/1 ml, thiobarbituric acid-active products

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5.20±0.19 µmol/l with an antioxidant activity of 22.1±0.27%). Similar values in the control were 1.44±0.09 D_{233}/1 ml, 3.46 ± 0.14 µmol/l and 33.7±0.14%, respectively.

Table. Platelet activity in newborn calves with iron deficiency anemia

Recorded indicators	Calves with anemia, n=38, M±m	Control, n=29, M±m
Aggregation with ADP, s	25.6±0.11	40.2±0.08 p<0.01
Aggregation with collagen, s	19.8±0.15	31.4±0.08 p<0.01
Aggregation with thrombin, s	37.5±0.07	53.8±0.07 p<0.01
Aggregation with ristomycin, s	23.0±0.12	48.0±0.12 p<0.01
Aggregation with H_2O_2, s	24.7±0.05	41.1±0.06 p<0.01
Aggregation with epinephrine, s	70.6±0.14	97.6±0.06 p<0.01
Aggregation with ADP and epinephrine, s	24.2±0.06	38.0±0.09 p<0.01
Aggregation with ADP and collagen, s	17.5±0.15	27.9±0.06 p<0.01
Aggregation with epinephrine and collagen, s	16.3±0.09	30.8±0.07 p<0.01
Discolets, %	57.8±0.18	77.7±0.11 p<0.01
Disco-echinocytes, %	29.0±0.15	13.9±0.13 p<0.01
Spherocytes, %	7.9±0.16	4.7±0.06 p<0.01
Sphero-echinocytes, %	4.3±0.08	2.7±0.05 p<0.01
Bipolar forms, %	1.0±0.03	1.0±0.05 p<0.01
Sum of active forms, %	42.2±0.20	22.3±0.11 p<0.01
The number of platelets in the aggregates, %	10.6±0.08	4.9±0.07 p<0.01
Number of small units by 2-3 thrombocytes per 100 free-standing platelets	14.8±0.11	3.5±0.06 p<0.01
Number of medium and large aggregates, 4 and more platelet per 100 free-standing platelets	3.26±0.23	0.14±0.07 p<0.01

Legend: p - reliability of the differences in the parameters between the control of healthy and sick.

The number of platelets in the blood of newborn calves with anemia corresponded to the norm. Aggregation of thrombocytes in animals with anemia was accelerated. The earliest was the aggregation of platelets under the action of collagen (19.8±0.15 s), somewhat later with ADP and ristomycin, even later with H_2O_2 (24.7±0.05 s) and thrombin (37.5±0.07 from). The latest aggregation of platelets in calves with iron deficiency came under the influence of epinephrine a (70.6±0.14 s). The combination of inducers caused their mutual potentiation, accelerating the aggregation of platelets in sick animals almost in two (Table).

The quantitative content of discoid platelets in the blood of anemized calves reached 57.8 ± 0.18% (in control - 77.7±0.11%). In the total number of platelets of disco-echinocytes was doubled (29.0±0.15%). The levels of spherocytes, sphero-echinocytes and bipolar platelet forms also significantly exceeded those of calves in the control and reached 7.9±0.16%, 4.3±0.08% and 1.0±0.03%, respectively. The sum of active forms of platelets in calves with iron deficiency anemia was 42.2±0.20%, (in control - 22.3±0.11%). Small and large aggregates in the bloodstream of sick animals contained - 14.8±0.11 and 3.26±0.23 per 100 free-standing platelets, in control - 3.5±0.06 and 0.14±0.07, respectively. The content of platelets in aggregates in newborn calves with anemia was 10.6±0.08%, in control 4.9±0.07% (Table).

DISCUSSION

The development of iron deficiency anemia is accompanied by impaired functioning of many organs and systems [22-26], including platelet hemostasis due to the occurrence of thrombocytopathy [27]. Depression of antioxidant protection of plasma of newborn calves with anemia against the background of hypoxia developing in them [28-31] promotes activation of lipid peroxidation in plasma, disrupting platelet structures and causing stimulation of their functions [32,33]. An increase in newborn calves with anemia of platelet aggregation into individual inducers in vitro indicates an increase in their sensitivity to stimulating influences [34,35]. Taking into account the growth of platelet aggregation with ristomycin in calves with anemia, one can speak of an increase in their sensitivity to the von Willebrand factor [36,37]. Acceleration of platelet aggregation with ADP indicates in these animals the amplification of arachidonic acid in their blood plates with the formation of a powerful platelet aggregation stimulator-thromboxane [38]. Acceleration of platelet aggregation with combinations of aggregation inducers largely reflects the real process of platelet interaction against the background of their activation in vivo in neonatal calves with anemia [39]. Intensive vascular activity of thrombocytes in newborn calves with anemia indicates a significant increase in aggregation activity of blood platelets in the bloodstream and high availability of subendothelial structures for them [40], probably due to the amplification of endotheliocyte peeling under these conditions.

CONCLUSION

In newborn calves with iron deficiency anemia, there is an increase in the peroxidation of plasma lipids due to the weakening of its antioxidant protection. In addition, neonatal calves with iron deficiency anemia have an increase in the platelet aggregate ability to register this process in vitro and in vivo, which inevitably leads to the formation of micro-rheological disorders and the risk of thrombotic events.

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