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## Functional Properties Of Hemocoagulation In Calves Of Dairy Nutrition.

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#### ABSTRACT

Age-related changes in the activity and interaction of coagulation, anticoagulant and fibrinolytic systems of blood plasma in the phase of milk nutrition in calves is one of the most important physiological elements in providing homeostasis at this stage of development. The formation of the functional activity of these systems largely ensures adaptation to the external environment of their organism, controlling the liquid properties of blood in all the calf vessels, thereby contributing to the optimal deployment of an individual program for its development. The study found that in young cattle in the phase of dairy nutrition there is a regular dynamics of the functional state of the activity of plasma hemostasis and the mechanisms of its limiting, which provides the optimal rheology of blood necessary for the growth and development of the animal. Increased activity of the mechanisms of blood clotting, anticoagulation and fibrinolysis helps the calf to adapt to the phase of dairy nutrition, ensuring its preparation for nutrition with plant foods. **Keywords:** coagulation, anticoagulation and fibrinolysis, calves, dairy nutrition phase.



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#### Introduction

It becomes clear that a serious request of the society to increase food production [1,2,3] can be solved only with reliance on modern science [4,5]. Of great importance here is the study of bioregulatory processes in the blood of productive animals and mechanisms that ensure its preservation in the liquid state [6,7]. This is able to provide optimum tissue perfusion and trophic tissue [8], and, hence, the maximum growth rate of animals under optimal conditions of keeping and feeding [9,10].

In this regard, hemocoagulation is of particular interest in young cattle during its early ontogeny [11,12].

It becomes clear that age changes in the activity and interaction of coagulation, anticoagulant and fibrinolytic systems of blood plasma in the phase of milk nutrition in calves is one of the most important physiological elements in providing homeostasis at this stage of development [13]. The formation of the functional activity of these systems largely provides adaptation to the external environment of the whole organism, controlling the liquid properties of blood in all vascular regions of the animal, thus contributing to the optimal deployment of an individual calf development program [14,15]. At the same time, many aspects of age-related changes in the activity of coagulating, anticoagulant and fibrinolytic systems in healthy calves in the phase of dairy nutrition have not been adequately studied.

In this regard, the goal of the study was formulated: to establish the dynamics of the physiological state of coagulation, anticoagulant and fibrinolytic systems of blood plasma in healthy calves in the phase of milk nutrition.

#### 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 observation group included 32 healthy calves of milk nutrition at the age of 11 days. The complex of examinations consisted of determining the activity of peroxide oxidation of plasma lipids by the content of acyl hydroperoxides, thiobarbituric acid-active products by the Agat-Med company (Russia) and the antioxidant activity of the liquid part of the blood. Each activity of the calf in the plasma was evaluated for the activity of coagulation factors (I, II, V, VII, VIII, IX, X, XI, XII), duration of activated partial thromboplastin time, prothrombin and thrombin time. Each calf included in the study evaluated the activity of the anticoagulant system of blood plasma by determining the activity of antithrombin III and protein C. To determine the fibrinolytic ability of plasma in calves, a method was used to determine the time of spontaneous euglobulin lysis, the level of plasminogen,  $\alpha$ 2 antiplasmin and the content of fibrin degradation products by phenanthroline method.

Healthy calves were examined during the milk feeding phase 5 times: on 11, 15, 20, 25 and 30 days of life.

Statistical processing of the results was carried out using Student's t-test.

#### **RESULTS OF THE STUDY**

During the phase of dairy nutrition in healthy calves, the level of antioxidant plasma activity was stable (on average 32.6% -0.19%) and lipid peroxidation activity. Thus, the level of acyl hydroperoxides in it averaged  $1.51\pm0.14 D_{233}/1$  ml with a low content of thiobarbituric acid-active compounds (on average  $3.53\pm0.15 \mu$ mol/l), not significantly differing from the study to the study in the entire phase of the dairy diet.

In calves 11 days of life, the low functional capacity of all clotting factors was found. By 15 days of life in animals, an increase in the content of factors II and VII was observed with a tendency to increase in the level of fibrinogen in the blood and factors X and XII. At the same time, the activity of V, VIII, X and XI factors did



not experience statistically significant dynamics. The twenty-second day of life of healthy calves was marked by a further increase in the level of fibrinogen, Factors II, VII, IX, XI, XII with the invariance of V, VII and X factors. On 25 and 30 days, the calves showed an additional significant increase in the level of fibrinogen and factors II, VII, IX, XI, XII in the absence of statistically significant dynamics of V, VIII and X factors (Table 1).

Evaluation of coagulation tests in healthy calves during the milk feeding phase has highlighted a certain dynamics of activity of the coagulation system, reflecting changes in the plasma content of individual clotting factors in this phase of early ontogeny (Table 1). Thus, in assessing the age-related dynamics of activated partial thromboplastin time, its acceleration was established from 15.0 days from  $48.0\pm0.22c$  at the beginning of the phase of dairy nutrition to  $39.6\pm0.34$  s at its end. Prothrombin time, slowed down at the beginning of the phase, already reached  $15.7\pm0.13$  s by the 15th day, with a subsequent steady reduction to the end of the phase. Thrombin time, reflecting the intensity of the transition of fibrinogen to fibrin from 11 to 30 days of life in calves, was overall accelerated by 8.7%.

The included calves included an evaluation of the activity of anticoagulant and fibrinolytic activity of blood throughout the whole phase of the dairy diet. In the dynamics of activity of the components being evaluated, a statistically significant regularity was found in animals (Table 1).

Registered parameters	Milk phase, n=32, M±m					
	11	15	20	25	30	
	day of life	day of life	day of life	day of life	day of life	
Coagulation factor I, g/l	2.2±0.14	2.3±0.14	2.6±0.18	2.8±0.09	3.0±0.07	
			p<0.05	p<0.01	p<0.01	
Coagulation factor II, %	84.5±0.11	85.0±0.02	85.8±0.10	86.7±0.03	87.4±0.09	
		p<0.05	p<0.05	p<0.05	p<0.05	
Coagulation factor V, %	82.7±0.12	82.6±0.10	82.9±0.08	83.1±0.16	82.8±0.17	
Coagulation factor VII,	68.0±0.04	69.5±0.07	69.9±0.04	71.6±0.07	72.7±0.05	
%		p<0.01	p<0.01	p<0.01	p<0.01	
Coagulation factor VIII,	85.3±0.16	84.9±0.20	85.7±0.26	85.3±0.29	86.0±0.21	
%						
Coagulation factor IX,	83.0±0.14	82.8±0.15	84.6±0.03	85.6±0.08	86.5±0.11	
%			p<0.05	p<0.05	p<0.05	
Coagulation factor X, %	61.3±0.19	61.4±0.26	62.6±0.25	62.9±0.23	63.3±0.16	
Coogulation factor VI	00.1+0.10	00 6±0 22	02 2+0 15	02 4+0 20	04 6+0 12	
	90.1±0.10	90.0±0.22	92.3±0.15	93.4±0.20	94.0±0.12	
70 Coogulation factor VII	96.2+0.17	96.0+0.14	p<0.03	00.1+0.14	02 2+0 17	
	80.3±0.17	80.9±0.14	88.3±0.10	90.1±0.14	92.2±0.17	
Activated partial	48.0+0.22	46 1+0 12	μ<0.03	μ<0.01 41 7+0 10	20 6+0 24	
thrombonlastin time s	40.0±0.22	40.110.12 n<0.05	n<0.01	$+1.7\pm0.10$	n<0.01	
Prothrombin time s	19 0+0 17	18 7+0 13	17 0+0 08	16 5+0 07	16.0+0.05	
riounomoniene, s	19.010.17	p<0.05	p<0.01	p<0.01	p<0.05	
Thrombin time, s	16.2±0.12	15.9±0.19	15.6±0.10	15.2±0.04	14.9±0.02	
			p<0.05	p<0.05	p<0.05	
The activity of	101.7±0.07	103.6±0.14	105.4±0.12	106.9±0.10	108.2±0.16	
antithrombin III in		p<0.05	p<0.05	p<0.05	p<0.05	
plasma, %		•				
Protein C, %	76.0±0.10	78.2±0.16	79.4±0.04	81.6±0.06	83.5±0.08	
		p<0.05	p<0.05	p<0.05	p<0.05	
Time of spontaneous	178.2±0.34	175.4±0.15	173.0±0.22	172.0±0.18	170.3±0.15	
euglobulin lysis,		p<0.05	p<0.05		p<0.05	

Table. Coagulation, anticoagulation and fibrinolysis activity in healthy calves of dairy nutrition.

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minutes					
Plasminogen, %	122.0±0.05	123.8±0.08	124.6±0.06	126.0±0.05	128.6±0.10
		p<0.05	p<0.05	p<0.05	p<0.05
$\alpha_2$ antiplasmin, %	101.3±0.19	100.0±0.14	98.7±0.06 p<0.05	97.3±0.05 p<0.05	96.4±0.09 p<0.05
Fibrin degradation products, μg / ml	40.2±0.25	41.3±0.19	41.9±0.20	42.2±0.09	42.8±0.16

Legend: p - reliability of ontogenetic dynamics of the indicators being evaluated.

In their blood, a small but significant increase in the level of antithrombin III, an average of 105.2±0.13%, was established. Simultaneously, there was a significant increase in the level of protein C from 76.0 ± 0.10% to 83.5±0.08% from 11 to 30 days of life. In this case, a significant increase in the level of plasminogen was established in animals with a significant decrease in the inhibitor of its active form -  $\alpha_2$  antiplasmin by 5.1% for this phase of early ontogeny. This provided a small but steady decrease in the time of spontaneous euglobulin lysis with a constant level of fibrin degradation products during the dairy phase.

Thus, during the entire phase of milk nutrition in calves, the interdependent dynamics of the process of coagulation, anticoagulation and fibrinolysis, which facilitates the transition of their hemostasis to the level required for further growth and development of the organism and preparation for plant nutrition, is noted.

#### DISCUSSION

Calves 11-30 days life showed the lack of significant fluctuations in the level of lipid peroxidation and antioxidant protection of the Plaza at a certain dynamics and activity of plasma hemostasis, which, undoubtedly, allows to fix an adaptation of a calf to the conditions of extrauterine existence, ensuring the normal rheological state of blood [16], and thus the required supply of nutrients and oxygen [17,18] developing tissues of the animal body [19]. This is an important element in the protection of calves against possible adverse environmental factors [20], influencing the organism in the phase of milk feeding [21]. Dynamics of the coagulation system that controls the aggregate state of blood, is largely facilitated by the stability of the lipid peroxidation at an optimal level while increasing the influence of environmental factors [22,23]. The acceleration of the prothrombin clotting time of the blood [24,25], reflecting the strengthening of the mechanisms of activation and activity that triggers the clotting thromboplastin [26,27]. The summation of these phenomena provides the necessary for this stage of ontogenesis the level of the liquid properties of blood and the optimal degree of perfusion of internal organs [28,29], which greatly supports the necessary level of metabolism in tissues of the calf, contributing to its growth and development [30].

In calves of dairy nutrition, the content of V, VII and X factors in the blood is constant, with the activity of the other clotting factors increasing [31]. As a result, activated partial thromboplastin time, reflecting the activity of the internal coagulation pathway and prothrombin time, revealing the activity of its external pathway and its terminal stage, estimated by thrombin time, are accelerated [32,33]. Obviously, the revealed dynamics of the intensity of blood coagulation is an indispensable element of the organism's preparation for a new diet - the beginning of consumption of plant foods [34].

The age dynamics of the anticoagulation system that controls the aggregate state of blood and the fibrinolysis system that dissolves excess fibrin is largely due to the stable optimality of lipid peroxidation despite the increase in the influence of environmental factors [35]. Thus, during the phase of milk nutrition, the activity of inhibitors of coagulation and the level of fibrinolytic agents significantly increase: antithrombin III, protein C and plasminogen increase and the activity of the fibrinolysis inhibitor- $\alpha_2$  antiplasmin decreases [36]. Obviously, this is a physiological response to the adaptation of the organism, which, upon completion of the neonatal phase, requires an increase in fibrinolysis activity [37]. In view of the fact that the general inhibitor of the contact activation of plasma proteases, plasminogen gradually increases with the level of fibrin degradation products remaining in the blood, one can think about the optimal functioning of the mechanisms of hemostasis adaptation in these conditions without signs of hypocoagulant orientation of the hemostasis at these times, providing optimal microcirculation conditions [38].

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Summation of the dynamics of the activity of coagulation, anticoagulation and fibrinolysis provides the level of liquid blood properties necessary for this stage of ontogeny and the optimal degree of perfusion of internal organs, which largely maintains the necessary level of metabolism in calf tissues, contributing to its growth and development [39, 40].

Thus, calves have a small but significant increase in plasma hemostasis activity, adequately restrained by anticoagulation and fibrinolysis systems, which is probably an element of the general adaptation process of the organism in early ontogeny.

#### CONCLUSION

In young cattle in the phase of dairy nutrition there is a regular dynamics of the functional state of the activity of plasma hemostasis and the mechanisms of its limiting, which provides the optimal rheology of blood necessary for the growth and development of the animal. Increased activity of the mechanisms of blood clotting, anticoagulation and fibrinolysis helps the calf to adapt to the phase of dairy nutrition, ensuring its preparation for nutrition with plant foods.

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