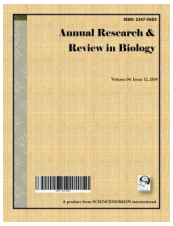

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Physiological Peculiarities of Hemocoagulation in Newborn Calves after Cresacin and Hamavit Application

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Authors' contributions

This work was carried out in collaboration between all authors. Author VDF designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors TAB and GSM managed the analyses of the study. Author ONM managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Newborn calves are still often registered to have dyspepsia or acute hypoxia. These disturbances are very often accompanied by the development of hemostasiopathy. There are no definite approaches to its correction yet.
Aim: To determine activity dynamics of coagulation hemostasis in newborn calves with dyspepsia or after acute hypoxia at birth against the background of cresacin and hamavit application.
Materials and Methods: We examined 33 newborn calves after acute hypoxia at birth (experimental group №1) and 38 calves with noninfectious dyspepsia (experimental group №2). In the course of 5 days all the experimental calves were given cresacin 4mg/kg a day in the morning for drinking and were injected hamavit intramuscularly 0.03 mg/kg once a day in the morning. We applied biochemical, hemostatic and statistical methods of investigation. The control group in our research was composed of 35 healthy newborn calves.

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Results of Research: Both groups of experimental calves were noted to have similar level rise of plasma lipids' peroxidation, strengthening of coagulation process, weakening of anticoagulation processes and fibrinolysis. Application of cresacin and hamavit combination to the examined animals provided similar positive dynamics of all the accountable indices in them. In the result of their application we noted some lowering of plasma acylhydroperoxides' level (more than in 2.2 times) on behalf of the increase of its antioxidant activity (more than by 30.0%) till the level of the norm. The newborn calves with dyspepsia or after acute hypoxia turned out to be able to reach full activity normalization of all the initially activated coagulation factors against the background of the conducted correction. The time of registered coagulation tests in the observed newborn calves from both experimental groups reached the level of the control values against the background of the correction. In the result of the conducted correction the activity of antithrombin III and protein C in the observed calves from the experimental groups increased by more than 13.0% and by more than 15.0% what provided their normalization. It was accompanied by level rise of plasminogen what provided activity normalization of fibrinolysis system in all the experimental animals.
Conclusion: Newborn calves with dyspepsia or after acute hypoxia at birth are characterized by coagulation activity and weakening of plasma anticoagulant and fibrinolytic mechanisms. The application of cresacin and hamavit combination to both categories of newborn calves provides normalization of plasma coagulation activity and mechanisms of its limitation.

Keywords: Dyspepsia; acute hypoxia; calves; newborn phase; cresacin, hamavit; blood coagulation.

1. INTRODUCTION

The system of coagulation is an important element of constancy maintenance of the body's internal environment. Its state has special physiological significance at the early stages of ontogenesis in all the productive animals including cattle [1]. Functional properties of coagulation system are mostly determined by fluid properties of blood. They provide optimal conditions for hereditary program realization of the calf's growth and development after birth [2].

On many stock-farms the calves are still registered to have acute hypoxia at birth or they may often have dyspepsia [3] in the course of the first 10 days of life. Both states lead to evident decrease of animals' resistance, their growth inhibition and often cause calves' death [4,5]. Notwithstanding the prevalence of these disturbances in newborn calves their impact on blood parameters and coagulation system are still studied rather poorly [6]. The possibility of their correction in these categories of calves (including combined application of widely used in animal husbandry stimulators of metabolism – cresacin and hamavit) is not defined yet.

In this respect we put the following aim in our research: to determine activity dynamics of coagulation hemostasis in newborn calves with dyspepsia or after acute hypoxia at birth against the background of cresacin and hamavit application.

2. MATERIALS AND METHODS

The study was conducted in strict accordance with the ethical principles established by the European Convention for the Protection of Vertebrate Animals used for experimental and other scientific purposes (adopted in Strasbourg on March, 18th, 1986, and confirmed in Strasbourg on June, 15th, 2006), approved by the local Ethics Committee of K.I. Skryabin Moscow State Academy of Veterinary Medicine and Biotechnology (record № 12, dated December, 3rd, 2015), the local Ethics Committee of Kursk State University (record № 11, December, 4th, 2015), the local Ethics Committee of Kursk State Medical University (record № 12, dated December, 3rd, 2015) and the local Ethics Committee of Samara National Research University (record № 12, dated December, 3rd, 2015).

In our research we involved 71 newborn calves (the 1st-2nd day of life) with functional disturbances: 33 newborn calves after acute hypoxia at birth (experimental group №1) and 38 calves with noninfectious dyspepsia (experimental group №2). The control was presented in our research by average values of indices which were received in 35 healthy calves at daily examination in the course of the newborn phase. All the calves taken into the study were born from crossbred cows. The cows had from 25% to 75% Holstein blood and were covered by thoroughbred Holstein bulls.

The examination of animals included activity estimation of plasma lipids' peroxidation according to the quantity of acylhydroperoxides (AHP) [7] and thiobarbituric acid (TBA)-active products with the help of a kit produced by the firm "Agat-Med" (Russia). In all the calves we registered the value of plasma antioxidant activity [8]. Functional capacities of blood coagulation system were determined in every calf under observation according to the activity of coagulation factors (I, II, V, VII, VIII, IX, X, XI, XII), duration of activated partial thromboplastin time, prothrombin and thrombin time [9].

The activity of anticoagulation capacity of blood plasma in the examined animals was determined according to the level of antithrombin III and protein C in it [9].

Fibrinolytic properties of blood plasma in calves were estimated according to the time of spontaneous euglobulin lysis, activity of plasminogen and α_2-antiplasmin [9].

State correction was conducted in all the observed calves with dyspepsia and after acute hypoxia at birth. So, in the course of 5 days they were given cresacin 4mg/kg a day in the morning for drinking and were injected hamavit intramuscularly 0.03 mg/kg once a day in the morning. Calves' state estimation was conducted at the start and in 5 days after the finish of correction.

The results were processed by Student's criterion (t). Statistical processing of received information was made with the help of a program package "Statistics for Windows v. 6.0", "Microsoft Excel". Differences in data were considered reliable in case of $p<0.05$.

3. RESULTS AND DISCUSSION

Plasma of newborn calves with dyspepsia or after acute hypoxia at birth was noted to have similar level rise of TBA-active products – approximately in 1.6 times in comparison with the control value. At the same time, the quantity of AHP in plasma in both experimental groups surpassed the level of healthy animals nearly in 2.3 times. Detected increase of lipids' peroxidation in plasma of experimental groups turned out to be possible in the result of similar weakening of plasma antioxidant activity in the animals from these groups – approximately by 30.0% (Table 1).

The newborn calves with dyspepsia and the calves after acute hypoxia at birth were found to have similar level rise of I, II, V, VII, VIII, IX, X and XI coagulation factors activity at the normal activity of XII factor. At the same time, in both groups of experimental animals the time of coagulation tests was comparably accelerated: the duration of activated partial thromboplastin time turned out to be comparably accelerated by average 31.5%, prothrombin time – by average 21.6%, thrombin time – by average 9.8% (Table 2).

At the same time, the activity of antithrombin III and protein C in the observed calves from both experimental groups turned out to be less in comparison with the control value by average 13.5% and 16.8% respectively. In both experimental groups it was accompanied by similar inhibition of spontaneous euglobulin lysis by average 27.3%, activity decrease of plasminogen by average 30.1% and activity rise of α_2-antiplasmin by average 11.2% (Table 3).

Table 1. The dynamics of biochemical indices in experimental groups of newborn calves having received cresacin and hamavit

Registered parameters	Experienced group 1		Experienced group 2		Control, n=35
	Initial values, n=33	At the end of observation, n=33	Initial values, n=38	At the end of observation, n=38	
Acyl hydroperoxides, D_{233}/1ml	3.02±0.09 p<0.01	1.32±0.14 p_1<0.01	3.10±0.18 p<0.01	1.34±0.10 p_1<0.01	1.33±0.14
TBA-active products, umol/l	4.92±0.07 p<0.01	3.06±0.05 p_1<0.01	4.88±0.08 p<0.01	3.04±0.08 p_1<0.01	3.02±0.16
Plasma antioxidant activity, %	28.2±0.06 p<0.01	37.2±0.12 p_1<0.01	27.8±0.15 p<0.01	37.3±0.17 p_1<0.01	37.0±0.10

Conventional signs: p – reliability of initial indices' differences in experimental groups and the control group; p_1 – reliability of indices' dynamics in experimental groups. In the following tables the notation is similar

Table 2. The dynamics of coagulation indices in experimental groups of newborn calves having received cresacin and hamavit

Registered parameters	Experienced group 1		Experienced group 2		Control, n=35
	Initial values, n=33	At the end of observation, n=33	Initial values, n=38	At the end of observation, n=38	
Factors I, г/л	2.0±0.12	1.5±0.10	2.1±0.09	1.4±0.24	1.4±0.08
	p<0.01	p_1<0.01	p<0.01	p_1<0.01	
Factors II, %	67.2±0.22	63.8±0.28	67.4±0.20	64.0±0.26	64.1±0.15
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Factors V, %	119.5±0.26	89.0±0.46	119.2±0.45	89.8±0.35	89.2±0.12
	p<0.01	p_1<0.01	p<0.01	p_1<0.01	
Factors VII, %	78.6±0.37	72.0±0.28	78.6±0.32	72.5±0.31	72.3±0.08
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Factors VIII, %	132.3±0.45	98.0±0.38	133.0±0.22	97.5±0.36	97.6±0.12
	p<0.01	p_1<0.01	p<0.01	p_1<0.01	
Factors IX, %	97.1±0.38	88.3±0.32	96.7±0.36	88.5±0.32	88.7±0.15
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Factors X, %	65.4±0.32	61.9±0.17	65.7±0.21	61.8±0.28	62.1±0.14
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Factors XI, %	94.3±0.34	90.3±0.23	95.0±0.18	89.8±0.21	90.2±0.12
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Factors XII, %	90.9±0.30	90.8±0.18	90.2±0.24	91.1±0.14	91.3±0.20
Activated partial tromboplastin time, s.	27.2±0.29	36.2±0.22	27.7±0.19	36.0±0.16	36.1±0.18
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Prothrombin time, s.	13.1±0.26	16.1±0.22	13.2±0.27	16.2±0.32	16.0±0.15
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Thrombin time, s.	15.8±0.22	17.4±0.19	15.7±0.16	17.3±0.26	17.3±0.12

Table 3. Dynamics of anticoagulation and fibrinolysis in experimental groups of newborn calves receiving cresacin and hamavit

Registered parameters	Experienced group 1		Experienced group 2		Control, n=35
	Initial values, n=33	At the end of observation, n=33	Initial values, n=38	At the end of observation, n=38	
Activity of antithrombin III in plasma, %	81.3±0.13	91.9±0.20	80.8±0.26	92.2±0.24	92.0±0.16
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Protein C, %	42.8±0.10	50.4±0.11	43.3±0.15	50.2±0.33	50.3±0.18
	p<0.05	p_1<0.05	p<0.05	p_1<0.05	
Time of spontaneous euglobulin lysis, min.	241.2±0.49	188.1±0.37	238.9±0.29	188.3±0.45	188.5±0.38
	p<0.01	p_1<0.01	p<0.01	p_1<0.01	
Plasminogen, %	84.2±0.25	111.2±0.32	85.1±0.30	110.6±0.24	110.2±0.24
	p<0.01	p_1<0.01	p<0.01	p_1<0.01	
α_2-antiplasmin, %	143.0±0.37	127.9±0.22	142.1±0.17	128.0±0.22	128.1±0.29
	p<0.01	p_1<0.01	p<0.01	p_1<0.01	

The application of cresacin and hamavit combination to the examined animals as a kind of correction impact provided similar positive dynamics of all the accountable indices in them. Initially increased levels of AHP and TBA-active compounds in plasma of calves from both groups fell in the result of cresacin and hamavit application and reached the control values on

behalf of normalization of plasma antioxidant activity (Table 1).

Against the background of the conducted correction the newborn calves with dyspepsia or after acute hypoxia turned out to be able to reach full activity normalization of all the initially activated enzymatic (I, II, VII, IX, X and XI) and nonenzymatic (V and VIII) coagulation factors at preservation of XII factor activity on the normal level (Table 2).

The time of coagulation tests in the observed newborn calves from both experimental groups reached the level of the control values in the result of correction. It took place in these calves in the result of prolongation of activated partial thromboplastin time approximately on 1/3, duration increase of prothrombin time approximately on ¼ and the increase of thrombin time approximately by 10.0%.

In the result of the conducted correction the activity of antithrombin III and protein C in the observed calves from both experimental groups increased in comparison with the start by more than 13.0% and by more than 15.0% what provided their normalization. Found at that evident time decrease of spontaneous euglobulin lysis pointed at activity normalization of fibrinolysis system in all the experimental animals. It was mostly reached in the result of weakening of initially surplus α_2-antiplasmin activity against the background of correction and the increase of plasminogen in their blood till the level of healthy animals (Table 3).

Various dysfunctions can be often registered already at the very start of ontogenesis. Acute hypoxia and dyspepsia are very often met. They significantly weaken the body and can cause its death [10,11]. In the present research the newborn calves with dyspepsia and calves after acute hypoxia at birth were noted to have similar weakening of plasma antioxidant protection with comparable level increase of AHP and TBA-active compounds in it. It inevitably caused increase of blood cells' aggregation in them [12,13], alteration of endotheliocytes and liver structures [14]. Given changes could strongly disturb the balance of pro- and anticoagulants in blood plasma [15]. In both experimental groups of animals it manifested itself by comparable acceleration of coagulation in its both ways. Given disturbances strengthened hypoxia and formed the risk of intraorganic thrombosis [16,17].

Coming after the episode of acute hypoxia or against the background of dyspepsia surplus thrombin-formation in newborn calves was repressed by their weakened system of natural coagulants, including antithrombin III and protein C [18,19]. Besides, dystrophic manifestations in the endothelium were caused by activation of lipids' peroxidation and/or hypoxia. They mostly promoted the disturbance of fixation process of antithrombin III with heparitin sulfate and glycosamine-glycans of endothelium. It strongly decreased vessels' thrombo-resistance [20,21]. Found in blood of newborn calves with dyspepsia or after acute hypoxia decrease of protein C activity pointed at the weakness of inhibitory control over the activity of V and VIII factors in these animals [22]. Found in all the experimental calves activity rise of α_2-antiplasmin and activity decrease of plasminogen caused weakening of their blood fibrinolytic properties.

Detected in both experimental groups of animals coagulopathy stressed the necessity to conduct effective correction directed at optimization of hemocoagulation mechanisms [23,24]. For solving of this task there were applied two means which strengthened metabolism.

The combination of cresacin and hamavit turned out to be able to normalize the level of plasma lipids' peroxidation in all the observed calves. It positively influenced metabolism in marrow, vascular walls and liver. Reached optimization of hemocoagulation activity turned out to be possible in the result of activity normalization of all the coagulation factors. Detected dynamics of their activity (against the background of cresacin and hamavit application to newborn calves with dyspepsia or after acute hypoxia) is the consequence of positive changes of liver metabolism in response to the introduction of the given preparations into the body [25]. Reached results in both experimental groups of animals were followed by the increase of plasma anticoagulation capacity. The rise of initially low antithrombin III activity till the level of the norm optimized generation of thrombin. It also controlled the quantity of active VII, IX, X, XI and XII coagulation factors in plasma [26,27]. The applied correction also normalized the activity of protein C in blood of all the experimental calves and provided optimal plasma inhibitory control over V and VIII coagulation factors in them [28,29]. At the same time, against the background of cresacin and hamavit the observed newborn calves turned out to be able to reach intensity rise of plasminogen synthesis

[30] with suppression of surplus antiplasmin activity of their plasma. All this normalized the process of fibrinolysis [31,32]. Given situation provided full normalization of the whole hemocoagulation system, optimum of fluid blood properties and functionally favorable conditions for the realization of their genetic potential [33,34].

4. CONCLUSION

The newborn calves with dyspepsia or after acute hypoxia at birth are noted to have surplus increase of plasma coagulation activity and decrease of anticoagulation and fibrinolytic indices. In the result of application of cresacin and hamavit combination to these categories of newborn calves we reached normalization of plasma coagulation activity and its mechanisms of anticoagulation and fibrinolysis.

CONSENT

As per international standard or university standard written patient consent has been collected and preserved by the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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