БИОХИМИЯ, ГЕНЕТИКА И МОЛЕКУЛЯРНАЯ БИОЛОГИЯ

BIOCHEMISTRY, GENETICS AND MOLECULAR BIOLOGY

DOI: 10.12731/2658-6649-2022-14-2-52-70
UDC 636.2:619:618.3:577.1

BLOOD MINERAL PROFILE IN NEWBORN CALVES WITH INTRAUTERINE GROWTH RETARDATION

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Widespread intrauterine growth retardation of embryo and fetal (IUGR) in cattle and its negative effects on postnatal growth and offspring health threaten the economic sustainability of livestock production. IUGR calves often exhibit birth weight deficits, early death, metabolic abnormalities, low growth rate, and poor feed utilization. Effective correction of these disorders is impossible without a detailed study of the causes, mechanisms of development and consequences of IUGR. In the present work, a comparative analysis of the content of macrominerals and trace elements in the blood of newborn Simmental calves with IUGR (Group I, n = 20) and the physiological course of pregnancy in their mothers (Group II, n = 20) was carried out. Blood for studies in calves was obtained at 1 day of age, before the 4th feeding of colostrum. The content of sodium, potassium, calcium and magnesium in blood serum samples and the content of iron, copper, zinc, manganese, cobalt and selenium in whole blood samples were studied by atomic absorption spectrometry (AA6300, Shimadzu, Japan). Group I calves had increased serum potassium (by 6.7%, P = 0.022) and magnesium (by 6.3%, P = 0.004) and decreased sodium (by 4.3%, P = 0. 005), reduced whole blood selenium (by 41.0%, P = 0.0001), copper (by 23.6%, P = 0.0001), and manganese (by 23.4%, P = 0.005) compared with Group II animals. The blood content of the other minerals studied did not differ *significantly between the groups. The possible causes and consequences of the identified disorders of the mineral blood profile in newborn calves with IUGR were analyzed.*

Keywords: *intrauterine growth retardation; cattle; blood analysis; macrominerals; trace elements*

For citation. *Chernitskiy A.E., Ermilova T.S., Salimzade E.A.O., Safonov V.A. Blood mineral profile in newborn calves with intrauterine growth retardation. Siberian Journal of Life Sciences and Agriculture, 2022, vol. 14, no. 2, pp. 52-70. DOI: 10.12731/2658-6649-2022-14-2-52-70*

МИНЕРАЛЬНЫЙ ПРОФИЛЬ КРОВИ У НОВОРОЖДЕННЫХ ТЕЛЯТ С ВНУТРИУТРОБНОЙ ЗАДЕРЖКОЙ РАЗВИТИЯ

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Широкое распространение внутриутробной задержки развития эмбриона и плода (ВЗРП) среди крупного рогатого скота и её негативное влияние на постнатальный рост и здоровье потомства угрожают экономической устойчивости животноводства. Телята с внутриутробной задержкой развития часто демонстрируют дефицит веса при рождении, раннюю гибель, нарушения обмена веществ, низкую интенсивность роста и плохое использование корма. Эффективная коррекция этих нарушений невозможна без детального исследования причин, механизмов развития и последствий ВЗРП. В настоящей работе проведен сравнительный анализ содержания макро- и микроэлементов в крови у новорожденных телят симментальской породы с ВЗРП (Группа I, n = 20) и физиологическим течением беременности у их матерей (Группа II, n = 20). Кровь для исследований у телят получали в 1-суточном возрасте, перед 4-м кормлением молозивом. Методом атомно-адсорбционной спектрометрии (АА6300, Shimadzu, Япония) в образцах сыворотки крови исследовали содержание натрия, калия, кальция и магния, в образцах цельной крови – содержание железа, меди, цинка, марганца, кобальта и селена. У телят Группы I установлено повышенное содержание в сыворотке крови калия (на 6.7%, P = 0.022) и магния (на 6.3%, P = 0.004) и пониженное натрия (на 4.3%, P = 0.005), пониженное содержание в цельной крови селена (на 41.0%, P = 0.0001), меди (на 23.6%, P = 0.0001) и марганца (на 23.4%, P = 0.005) по *сравнению с животными Группы II. Содержание в крови других исследованных минералов достоверно не различалось между группами. Анализируются возможные причины и последствия выявленных нарушений минерального профиля крови у новорожденных телят с ВЗРП.*

Ключевые слова: *внутриутробная задержка развития; крупный рогатый скот; анализ крови; макроэлементы; микроэлементы*

Для цитирования. *Черницкий А.Е., Ермилова Т.С., Салимзаде Э.А.О., Сафонов В.А. Минеральный профиль крови у новорожденных телят с внутриутробной задержкой развития // Siberian Journal of Life Sciences and Agriculture. 2022. Т. 14, № 2. С. 52-70. DOI: 10.12731/2658-6649-2022-14-2-52-70*

Introduction

Embryo and fetal intrauterine growth retardation (IUGR), defined as a mismatch between the size of the developing embryo/fetus and gestational age, is a fairly common pathology among livestock [25, 51]. According to recent studies [2, 36, 46], 34.4-37.6% of pregnant cows have IUGR. The development of IUGR is caused by genetic factors [25, 51], malnutrition [23, 46, 51], endocrine and immune disorders [4, 25, 27, 36], stress [25, 26, 48], and intoxication [36, 44]. Newborns with IUGR often demonstrate body weight deficiency [20, 51], are predisposed to metabolic and infectious diseases [6, 25, 33, 51]. The economic damage from IUGR consists of the costs associated with the treatment, culling and death of such calves [6, 9, 37], reduction in their growth intensity and feed utilization efficiency [6, 20, 21, 51]. Thus, IUGR is a serious problem for beef and dairy cattle breeding, reducing the profitability of the industry.

Despite considerable interest in the problem of IUGR in cattle [11, 22, 36, 44, 45], only sporadic studies have investigated metabolic disorders in newborn calves associated with IUGR [6, 13, 25, 33, 51]. It has been shown [6, 33, 51] that newborns with IUGR are characterized by hypoglycemia, hypoxemia, lactoacidosis, insufficiency of antioxidant system and dyslipidemia. Given the important role of macrominerals and trace elements in the regulation of metabolism [19, 49], it is reasonable to expect that even minor changes in the bioelement profile of the newborn (excess, deficiency or imbalance of minerals in the body) can lead to profound disorders in its postnatal adaptation.

The aim of our study was to conduct a comparative analysis of the content of macrominerals (sodium, potassium, calcium, magnesium) and trace elements (iron, copper, zinc, manganese, cobalt, selenium) in blood of newborn calves with IUGR and the physiological course of pregnancy in their mothers.

Materials and methods

The objects of the study were 40 1-day-old calves weighing 34-55 kg obtained from Simmental cows (with productivity of 5865-9176 kg during the previous lactation) from the peasant farm "Rechnoye", Khlevensky District, Lipetsk Region, Russian Federation – two groups of 20 animals. Group I included newborn cows (12 males and 8 females) with IUGR in anamnesis; Group II included progeny of cows with a physiological course of pregnancy (9 males and 11 females).

IUGR in cows was diagnosed by transrectal palpation and echography using an ultrasound scanner "Easi-Scan-3" (BCF Technology Ltd., UK) with a 4.5-8.5 MHz linear transducer. At 38-45 days after insemination and conception, the criteria for fetuses and embryos underdevelopment were considered to be the coccyoparietal size less than 16 mm and the body diameter less than 9 mm, at 60-65 days, less than 45 mm and 16 mm, respectively; at 110-115 days, the fetal horn diameter was less than 15 cm and the placenta less than 17 mm [2, 36]. The first portion of colostrum from their mothers was given to the calves within 1.5 hours after birth, then they received two more feedings during the day with an interval of 5-6 hours – a total of 4.5-5 liters of colostrum for three feedings.

Blood for studies in animals was obtained in the morning on an empty stomach, before the 4th feeding, by puncturing the jugular vein using vacuum systems for blood sampling. Lithium heparin was used as anticoagulant. To obtain serum, blood samples without anticoagulant were incubated for 1 hour at room temperature and then centrifuged (UC-1612, ULAB, China) at 4000 × g for 10 minutes. The resulting serum and whole blood samples were frozen and stored in liquid nitrogen at -195 °C until analysis.

Quantification of sodium, potassium, calcium and magnesium in serum samples, and iron, copper, zinc, manganese, cobalt and selenium in whole calf blood samples was performed by atomic absorption spectrometry (AA6300, Shimadzu, Japan) as described previously [28].

Statistical data processing was performed using IBM SPSS Statistics 20.0 program (IBM Corp., USA). All data were expressed as mean ± standard deviation and median. The reliability of differences between calf groups was determined using the Independent-samples Mann-Whitney U test.

Results

The results of determining the concentration of macro- and micronutrients in the blood of calves are presented in Tables 1 and 2, respectively.

Table 1.

Content of macrominerals in blood serum of newborn calves

Element	Group I (n = 20)	Group II (n = 20)	P*
Sodium (mmol/L)	139.6 ± 6.6 (140.2)	149.6 ± 9.7 (146.5)	0.005
Potassium (mmol/L)	5.27 ± 0.24 (5.29)	5.09 ± 0.35 (4.96)	0.022
Calcium (mmol/L)	2.87 ± 0.08 (2.91)	2.97 ± 0.29 (3.02)	0.300
Magnesium (mmol/L)	1.05 ± 0.09 (1.01)	0.92 ± 0.13 (0.95)	0.004

Data are presented as mean ± standard deviation (median). * Significance level.

From Table 1 we can see that in Group I calves the median values of serum potassium and magnesium were significantly higher (by 6.7% and 6.3% respectively, P < 0.05-0.01) and sodium was lower (by 4.3%, P < 0.01) than in Group II animals. The median values of neonatal serum calcium concentrations did not differ significantly between the samples. However, Group I animals had lower serum sodium-potassium ratios (26.5 ± 0.8: 1 (median 26.2: 1) versus 29.4 ± 0.3: 1 (median 29.5: 1) in Group II) and calcium-magnesium ratios (2.76 ± 0.27: 1 (median 2.86: 1) versus 3.25 ± 0.36: 1 (median 3.22: 1) in Group II).

The data in Table 2 show that Group I calves were characterized by lower whole blood concentrations of copper (by 23.6%, P < 0.001), manganese (by 23.4%, P < 0.01) and selenium (by 41.0%, P < 0.001) compared with Group II animals. At the same time, differences between neonatal groups in the content of iron, zinc, and cobalt in whole blood were statistically insignificant.

Table 2.

Trace elements content in whole blood in newborn calves

Element	Group I (n = 20)	Group II (n = 20)	P*
Iron (mmol/L)	3.53 ± 0.26 (3.63)	3.60 ± 0.31 (3.61)	0.610
Copper (μmol/L)	9.67 ± 0.51 (9.47)	12.9 ± 2.1 (12.4)	0.0001
Zinc (μmol/L)	35.5 ± 4.5 (33.9)	39.1 ± 7.8 (40.6)	0.212
Manganese (μmol/L)	1.95 ± 0.36 (1.80)	2.37 ± 0.43 (2.35)	0.005
Cobalt (μmol/L)	1.05 ± 0.36 (1.04)	0.69 ± 0.23 (0.66)	0.066
Selenium (μmol/L)	1.39 ± 0.44 (1.44)	2.37 ± 0.29 (2.44)	0.0001

Data are presented as mean ± standard deviation (median). * Significance level.

Discussion

Comparison of our results with the data of other researchers [10, 19, 29] showed that in Group II calves, the blood content of the studied macrominerals and trace elements was generally within the reference values. Newborn calves with IUGR (Group I) had lower serum sodium content [10, 29, 47] and higher serum magnesium content [1, 10]. Serum potassium concentration in these calves, although higher (by 6.7%, $P = 0.022$) compared with Group II animals, remained within the reference interval [1, 29]. Serum calcium concentrations in neonates with IUGR were also within the reference range [10, 17, 34]. Serum sodium-potassium and calcium-magnesium ratios are the most important physiological constants regulating osmotic pressure, neural and muscular excitation, activation of many enzyme systems and metabolic adaptations of the newborn in general [18, 19, 29, 50]. It is natural that their values correlate with the level of physiological maturity [1, 33] and viability of newborn calves [7, 15]. The present study showed that in 1-day-old calves with IUGR, the serum sodium-potassium ratio ranged from 25.7: 1 to 28.3: 1 and the calcium-magnesium ratio from 2.27: 1 to 2.96: 1. In offspring of cows with physiological course of pregnancy, these values were markedly higher, (28.9-29.9): 1 and (2.64-3.99): 1, respectively. Our data confirm the results of an earlier study [33], which also found a reduced (by 11.2%, $P < 0.01$) serum calcium-magnesium ratio in animals with IUGR 24 hours after birth. The electrolyte balance abnormalities detected in 1-day-old calves with IUGR (Table 1) are risk factors for respiratory dysfunction [1, 7] and colostral immunodeficiency [1, 8], which must be considered when developing a system of treatment and preventive measures.

It is known that the development of oxidative stress in infants with IUGR is associated with intrauterine deficiency of selenium and copper [6, 13, 26, 49] and, to a lesser extent, with zinc and manganese deficiency [6, 30, 40, 43]. The results of the current study found that 1-day-old calves with IUGR had lower blood levels of selenium (by 41.0%, $P = 0.0001$), copper (by 23.6%, $P = 0.0001$) and manganese (by 23.4%, $P = 0.005$) compared with offspring of physiologically pregnant cows; the whole blood concentrations of other trace elements (iron, zinc and cobalt) were not significantly different between groups (Table 2). Previously, lower (compared with our data) whole blood selenium levels were reported in newborn Red Holstein calves, from 0.56 to 1.49 μmol/L before the first colostrum intake and from 0.52 to 1.68 μmol/L by the end of the milk period, but comparable with the blood selenium content of their mothers (0.47 to 1.46 μmol/L) [38]. These differences, apparently, are a consequence of different biogeochemical conditions in the regions where the research was conducted, rather than breed specifics of cattle [38, 40, 41]. Lipetsk region of the Russian Federation, where our study was carried out, is considered a reference region for the content of selenium in soils and forages [3, 40], the consequence of which is the high content of the element in the blood of experimental calves (Table 2). However, even under these conditions, a marked decrease in selenium content in the blood of newborn calves with

IUGR (Table 2) may be critical for their postnatal adaptation [6, 26, 41, 49]. The biological role of selenium is not limited to its antioxidant activity, the element is involved in the regulation of thyroid function, metabolism, immune system and reproductive function in animals [6, 32, 49]. Therefore, the detected deficiency of selenium in the blood of newborn calves with IUGR should be timely corrected by the additional introduction of selenium-containing supplements in their diet or by injecting selenium preparations for the prevention of metabolic and infectious diseases [6, 42, 49].

Reduced blood levels of copper and manganese can also have serious consequences for neonatal health (Table 2). It has been shown that deficiency of these trace elements plays an important role in the pathogenesis of neonatal diarrhea [5, 42], omphalitis [39], bovine respiratory disease [6, 28, 42] and sepsis [16] in calves. Analysis of copper and manganese content in the covering hair of the tail brush in newborn calves with IUGR revealed a 28.3% ($P < 0.001$) and 9.4% ($P < 0.001$) decrease in these trace elements respectively compared to offspring of cows with physiological pregnancy [6], indicating insufficient intake of these trace elements to the developing fetus with IUGR during the last 3 months of gestation [6, 14]. Thus, our results and the sources [6, 31, 42, 46, 49] indicate that to effectively correct copper and manganese deficiency in newborn IUGR calves, additional intake of these micronutrients to their mothers during pregnancy is necessary.

Recent studies have proved the role of zinc deficiency in pregnant women in the genesis of IUGR [24, 30, 31, 46]. Zinc deficiency impairs the secretion of sex steroids, glucocorticoids, somatomedin, increases the expression of proinflammatory cytokines, inhibits cell proliferation and fetal/embryonic growth [24, 35, 46]. The in vivo experiment showed that additional zinc administration to pregnant animals effectively protects them from lipopolysaccharide-induced IUGR [12]. A lower (by 10.7%, $P < 0.001$) zinc content was also found in the hair of newborn calves with IUGR compared with offspring of cows with physiological pregnancy [6]. However, in the current experiment, the decrease in blood zinc content in calves with IUGR was not statistically significant (Table 2). Probably, to find statistically significant differences in blood zinc content between newborn calves with IUGR and offspring of cows with physiological course of pregnancy, additional studies on a larger sample of animals are required.

Conclusion

Thus, the results of the study suggest that the mineral profile of blood in 1-day-old calves with IUGR in the anamnesis has characteristic features, namely imbalance of macronutrients (increased level of potassium and magnesium and reduced – sodium in blood serum) and deficiency of essential trace elements (selenium, copper and manganese), indicating the presence of diselementosis. The latter in the absence of its adequate correction can lead to disruption of postnatal adaptation of calves, the development of metabolic and infectious diseases.

Conflict of interest. The authors declare that they have no conflict of interest.

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Поступила 09.04.2022	Received 09.04.2022
После рецензирования 25.04.2022	Revised 25.04.2022
Принята 27.04.2022	Accepted 27.04.2022