VETERINARSKI ARHIV 92 (3), 223-232, 2022 DOI: 10.24099/vet. arhiv.1377

The relationship of negative energy balance (NEB) and energy metabolism, milk production and reproductive performance during early lactation in dairy cows in Heilongjiang, China

Zhijie Wang, Yuxi Song, Chang Zhao, Yunlong Bai, Feng Zhang, Cheng Xia*, Shixin Fu, Hongyou Zhang, Chuang Xu, Ling Wu

College of Animal Science and Veterinary Medicine, Heilongjiang Bayi Agricultural University; Heilongjiang Provincial Technology Innovation Center for Bovine Disease Control and Prevention; Heilongjiang Provincial Key Laboratory of Prevention and Control of Bovine Diseases; Heilongjiang Province Cultivating Collaborative Innovation Center for The Beidahuang Modern Agricultural Industry Technology. Daqing, China.

WANG, Z., Y. SONG, C. ZHAO, Y. BAI, F. ZHANG, C. XIA, S. FU, H. ZHANG, C. XU, L. WU: The relationship of negative energy balance (NEB) and energy metabolism, milk and reproductive performance during early lactation in dairy cows in Heilongjiang, China. Vet. arhiv 92, 223-232, 2022.

ABSTRACT

Negative energy balance (NEB) causes economic losses to dairy farms around the globe. The aim of this study was to investigate the effect of NEB on energy metabolism, reproduction, etc. during early lactation in dairy cows on intensive farms in Heilongjiang, China. According to β-hydroxybutyric acid (BHBA), glucose (GLU), Non-esterified fatty acid (NEFA) levels, and clinical manifestations 14-21 days postpartum, 118 cows were divided into a positive energy balance (PEB) group (BHBA<1.2, GLU>2.8, NEFA<0.7 mmol/L, n=67) and an NEB group (BHBA>1.2, GLU<2.8, NEFA>0.7 mmol/L, n=51). These indicators were analyzed by cross-sectional research methods combined with Pearson correlation analysis and a prospective cohort study. The results showed that at 14-21 days postpartum, compared with the PEB, the body condition score, body condition loss (BCL), milk urea nitrogen, BHBA, NEFA, the interval from calving to first estrus (ICFE), pregnancy per artificial insemination (P/AI), and calving interval were higher (P<0.05), and daily lactation (DL), milk protein and GLU, estrus rate and conception rate were lower in NEB cows. BCL was positively correlated with ICFE and P/AI (P<0.05), and negatively correlated with estrus and conception rate (P<0.05). DL was negatively correlated with the P/AI (P<0.05). There was a positive association between NEB and anestrus (χ^2 M-H=12.63, P=0.0004), the risk of anestrus caused by NEB rose by 3.67 times that of PEB. The conclusion showed that NEB is closely related to BCL, which is a factor in the decline of lactation and reproductive performance in dairy cows. NEB is a risk factor for anestrus in dairy cows.

Key words: dairy cow; negative energy balance; early lactation; reproductive performance

*Corresponding author:
Prof. Cheng Xia, College of Animal Science and Technology, Heilongjiang Bayi Agricultural University, Daqing Heilongjiang China 163319, mail: xcwlxyf@sohu.com
Zhijie Wang and Yuxi Song contributed equally as the first authors.

ISSN 0372-5480
Printed in Croatia

Introduction

In past decades, with increasing modernization and the scale of dairy farming, the rapid promotion and application of intensive feeding modes, and the continuous improvement of the level of milk production of dairy cows, the reproductive performance of high-yielding dairy cows has shown a downward trend (SZENCI et al, 2018). The specific manifestations are a prolonged interval from calving to first estrus (ICFE), calving interval (CI), more pregnancies per artificial insemination (P/AI), decreased estrus rate and conception rate, etc. (CARTHY et al., 2016; FOLNOŽIĆ et al., 2016; NEGRÓN-PÉREZ et al., 2019; LUCY, 2019). Anestrus leads to the low reproductive efficiency of high-yielding dairy cows, and thus affects the economic benefits of the herd. Therefore, anestrus has become an unavoidable problem restricting the healthy development of the dairy industry at this stage.

In the early stages of lactation, almost all dairy cows must experience a period of NEB, metabolic stress, and body condition loss (BCL) to cope with the increased energy demand during lactation (DRACKLEY, 1999; KUHLA et al., 2016; MEZZETTI et al., 2020). Delayed increase in dry matter intake, genetic selection to increase milk production, and inappropriate diet will further increase the duration and extent of NEB (OSPINA et al., 2010; VINCE et al., 2017; ALADROVIĆ et al., 2018). In recent years, metabolic signals have become a research hotspot in the regulation of food intake and energy metabolism, including nutrients, metabolites, reproductive hormones, stress hormones, leptin, insulin, insulin-like growth factors, glucagon, etc (HALAAS et al., 1995; FOLNOŽIĆ et al., 2015; TURK et al., 2016; KLECZKOWSKI et al., 2017;). This means that negative energy balance also affects changes in hormones related to energy metabolism regulation. Many studies have shown that body fat reserves or body condition score (BCS) during lactation, and changes in BCS during early lactation will affect the recovery of the estrus cycle and the reproductive success rate of dairy cows (HAILAY et al., 2019; BEZDÍČEK et al., 2020), but the impact of BCL on reproductive performance remains to be further studied. Therefore, dairy researchers from various countries have become committed to the investigation of the early lactation reproductive performance of high-yielding dairy cows, and the study of the effects of NEB on BCS, daily lactation (DL) and reproductive performance. Once a breakthrough is made, it will have a significant impact on the improvement of the reproductive performance of large scale cattle farms which is of great practical significance.

In conclusion, the study hypothesized that there was a relationship between NEB, BCS, DL and reproductive performance. It posed the questions: Is NEB and BCL a factor in the decline of milk performance? Are NEB and BCL important factors in reproductive performance degradation? Does NEB cause estrus rates to rise? Therefore, this study discussed the influence of NEB on reproduction in early lactation in cows through blood tests, clinical pathology, biostatistics, and other aspects, to provide direction and ideas regarding the influence of NEB on early the lactation reproductive performance of cows in future.

Materials and methods

Experimental animals and grouping. According to the requirements of the Veterinary Medical Ethics Committee of the Ministry of Agriculture of China, this experiment was carried out on a large intensive cattle farm in the central region of Heilongjiang Province. All experiments on animals were carried out according to the standards approved by the Animal Welfare and Research Ethics Committee at Heilongjiang Bayi Agricultural University (No. 20200127). The total mixed ration of the tested dairy cows complied with the NRC (2001). One hundred and eighteen Holstein cows with a lactation volume of more than 9 tons (about 3 parities) were selected as the experimental animals. The total mixed ration of the dairy cows complied with the Chinese Feeding Standard of Dairy Cows and consisted of: cottonseed 1.03 kg, soybean hulls 1.50 kg, oat grass 0.50 kg, alfalfa 2.50 kg, soybean meal 1.30 kg, flake corn 2.00 kg, molasses 1.00 kg, silage 25.37 kg, corn 3.00 kg, concentrated feed 4.09 kg, and water ad libitum. The nutrient levels were Crude protein 17.70%, Starch 22.70%, Dry matter 48.00%, Dry

matter intake 23.50%, Neutral detergent fiber 31.50%, Acid detergent fiber 19.00%, Neutral detergent fiber from Roughage 18.70 %, Net milk production 0.78 Mcal/kg. On the basis of the test results relating to plasma β-hydroxybutyric acid (BHBA), glucose (GLU), Non-esterified fatty acid (NEFA) and clinical manifestations at 14 to 21 days postpartum, the 118 experimental dairy cows were divided into a positive energy balance group of 61 (PEB group, BHBA<1.2, GLU>2.8, NEFA<0.7 mmol/L), and a negative energy balance group of 57 cows (NEB group, BHBA>1.2, GLU< 2.8, NEFA>0.7 mmol/L) (ZHANG et al., 2018; ZHAO et al., 2019).

Medicaments and Instruments. Medicines for synchronous estrus, Gonarelin for injection (GnRH, Shuniu,); Cloprostenol Sodium Injection (PG, Duobaosu) were used, all purchased from China Ningbo No. 2 Hormone Factory.

Basic information collection. We used the Afimilk Ranch Management System (Afimilk®3.076, Israel) to collect basic information about the experimental cows, including background information such as. age, parity, and lactation with the previous fetus; lactation performance indicators, such as: the amount of lactation within 50 to 55 days postpartum, and the milk composition of 14 to 21 days postpartum; and reproductive performance indicators such as: ICFE, P/AI, calving to first insemination (CFI), CI, estrus rate, and conception rate. The BCS of the experimental dairy cows was evaluated twice after milking in the morning between days 14 to 21 and 50 to 55 after delivery by trained veterinarians.

Sample Collection. Blood samples were collected from the test dairy cows on days 14 to 21, 50, and 55 postpartum, and 10 mL of blood was collected from the tail vein before the morning feeding. Heparin was added for anticoagulation, the samples were centrifuged at 4000 r/min for 5 min at low speed, the supernatant aspirated, then centrifuged at 12000 r/min for 5 min at high speed, the plasma aliquoted, and stored at -80 ºC for biochemical testing.

Test Items. We tested plasma energy metabolism indicators: β-hydroxybutyric acid (BHBA, enzymatic method, mmol/L), non-esterified fatty acids (NEFA, enzymatic method, mmol/L), and glucose (GLU, glucose oxidase method, mmol/L). The kits were purchased from Shenzhen Mindray Bio-Medical Electronics Co., Ltd., and tested with a Mindray BS-830 automatic biochemical analyzer.

Presynch-Ovsynch Program. All the dairy cows in the test were subject to the Presynch-Ovsynch program. The specific steps are as follows: 40 ± 3 d DIM for the first injection of PG 0.6 μg / kg BW per cow, 14 days later (54 ± 3 d DIM) for the second injection of PG 0.6 μg / kg BW per cow, 12 days later (66 ± 3 d DIM) the first injection of GnRH 0.15μg / kg BW per cow, 7 days later (73 ± 3 d DIM) the third injection of PG 0.6 μg / kg BW per cow, 2 days later (75 ± 3 d DIM) the second injection of GnRH 0.15μg / kg BW per cow, Artificial insemination was performed after 16 to 24 hours. If the cow is found to be in heat during the execution of the Presynch-Ovsynch program, insemination should be performed and the drug injection should be stopped.

Statistical Analysis. SPSS 22.0 (IBM, USA) was used for statistical analysis. The independent sample t test was used to analyze the significance of the differences between the two groups of experimental cows in terms of various indicators, and the data were expressed as mean ± standard error ($\bar{x} \pm$ SEM). The Pearson correlation coefficient was used to analyze the correlation between plasma energy metabolites, BCS, DL and reproductive parameters. The M-Hχ2 test for cohort studies was used to analyze the association between NEB and postpartum anestrus. The threshold of P was set to 0.05.

Results

Background Information. It may be seen from Table 1 that the plasma BHBA and NEFA levels, BCS and BCL of the cows in the NEB group were higher than those in the PEB group from 14 to 21 days postpartum (P<0.01). The plasma GLU level was lower thanhat tof the cows in the PEB group (P<0.01). However, there were no differences in the age, parity, BCS 50 to 55 days postpartum, or last lactation between the two groups of dairy cows (P>0.05). The results showed that cows with NEB occurring 14 to 21 days postpartum had higher

BCS and BCL, showing the characteristics of high plasma concentrations of BHBA, NEFA and low concentrations of GLU.

Table 1. Background information on the two groups of the cows, 14-21 days

Project	PEB (n = 61)	NEB (n = 57)	P value
Age	3.90 ± 0.12	4.14 ± 0.14	0.172
Parity	2.64 ± 0.10	2.88 ± 0.11	0.119
BCS①	3.17 ± 0.05	3.38 ± 0.04	0.003
BCS②	2.89 ± 0.06	2.78 ± 0.05	0.168
BCL	0.29 ± 0.05	0.60 ± 0.05	<0.001
Last lactation (tons)	11.04 ± 0.21	10.84 ± 0.25	0.551
BHBA (mmol/L)	0.82 ± 0.03	1.98 ± 0.08	<0.001
NEFA (mmol/L)	0.55 ± 0.01	0.85 ± 0.03	<0.001
GLU (mmol/L)	3.91 ± 0.08	2.42 ± 0.03	<0.001

Note: body condition score=BCS; body condition loss =BCL; β-hydroxybutyric acid=BHBA; non-esterified fatty acid=NEFA; glucose=GLU. ①mains body condition sore 14 to 21 days postpartum; ②mains body condition sore 50 to 55 days postpartum

Comparison of the lactation performance of two experimental groups of cows. Figure 1 shows the change trend of the DL of the experimental dairy cows in early lactation. The lactation volume increased sharply from 10 to 20 days postpartum, and the lactation volume continued to increase after 20 to 60 days. The milk production of the cows in the NEB group within 60 days after delivery was always lower than that of the PEB group cows (P<0.05); the average DL was lower than that of the PEB group cows (36.85 ± 0.70 kg/d vs 41.46 ± 0.66) kg/d) (P<0.01). The results show that the milk production of cows with NEB was significantly reduced 14 to 21 days after delivery.

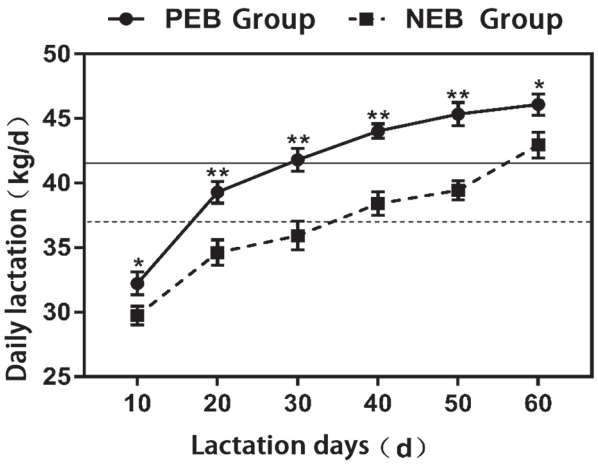

Fig. 1. Comparison of daily lactation (DL) during early lactation in the cows with positive energy balance (PEB) and negative energy balance (NEB)

As shown in Table 2, the milk protein of the cows in the NEB group was lower than that of the PEB group (P<0.05); milk urea nitrogen was higher than that of the PEB group (P<0.05). There were no differences in milk fat, lipoprotein ratio, lactose, total solids and somatic cells between the two groups of test cows (P>0.05). The results showed that the milk protein significantly decreased and milk urea nitrogen increased at 14-21 days postpartum with NEB.

Table 2. Comparison of milk composition between the two groups of cows at 14 to 21 days postpartum

Indicators	PEB (n = 61)	NEB (n = 57)	P value
Milk fat (%)	4.62 ± 0.07	4.59 ± 0.08	0.797
Milk protein (%)	3.40 ± 0.04	3.25 ± 0.05	0.015
Lactose (%)	5.23 ± 0.01	5.23 ± 0.01	0.969
Total solids (%)	13.78 ± 0.11	13.48 ± 0.11	0.052
Somatic cells ($\times 10^4$/mL)	5.15 ± 0.27	4.67 ± 0.32	0.247
Milk urea nitrogen (mg/dL)	11.32 ± 0.17	11.80 ± 0.18	0.047

Comparison of the reproductive performance of the two groups of cows. It may be seen from Table 3 that the ICFE, P/AI, and CI of the cows in NEB group were higher than those in the PEB group (P<0.01).Estrus rate and conception rate were lower than those of the PEB group (P<0.01). However, there was no difference in CFI between the two groups (P>0.05).The results show that early lactation NEB could prolong the ICFE and CI of cows, increase NSPC, and reduce the estrus rate and conception rate of cows.

Table 3. Comparison of reproductive performance of the two groups of cows

Indicators	PEB (n = 61)	NEB (n = 57)	P value
ICFE (d)	44.56 ± 1.72	56.61 ± 2.50	<0.001
P/AI (times)	1.84 ± 0.11	2.46 ± 0.13	<0.001
CFI (d)	65.21 ± 1.23	66.30 ± 1.33	0.550
CI (d)	373.07 ± 3.19	401.89 ± 4.51	<0.001
Estrus rate (%)	85.25	57.89	0.001
Conception rate (%)	59.02	35.09	0.009

Note: interval from calving to first estrus=ICFE; pregnancy per artificial insemination=P/AI; calving to first insemination=CFI; calving interval=CI

Correlation analysis of plasma energy metabolites, body condition, milk production and reproductive performance. It may be seen from Table 4 that ICFE was positively correlated with plasma BHBA and NEFA levels and BCL on days 14 to 21 (P<0.05), and was negatively correlated with plasma GLU levels from day 14 to 21 (P<0.05); Estrus rates were positively correlated with plasma GLU levels from day 14 to 21 (P<0.05), and negatively correlated with plasma BHBA levels and BCL from day 14 to 21 (P<0.05). P/AI was positively correlated with BCS, BCL, and plasma BHBA and NEFA levels (P<0.05), and negatively correlated with GLU level and average DL from day 14 to 21 (P<0.05). The CI was positively correlated with plasma BHBA and NEFA levels from day 14 to 21 (P<0.05), and negatively correlated with plasma GLU levels from day 14 to 21 (P<0.05). The conception rate was positively correlated with plasma GLU levels from day 14 to 21 (P<0.05), and negatively correlated with plasma BHBA and NEFA levels, BCS and BCL from day 14 to 21 (P<0.05). The results showed that the decline in reproductive performance of NEB dairy cows was related to the increase in plasma BHBA and NEFA levels from day 14 to 21 postpartum, to the decrease in plasma GLU levels from day 14 to 21 and average DL, and to the increase in BCL.

Table 4. Correlation between plasma energy metabolites, body condition, milk production and reproduction parameters

Indicators		ICFE	P/AI	CI	Estrus rate	Conception rate
14-21 d BHBA	*R*	0.315	0.269	0.270	-0.245	-0.263
	P	0.001	0.003	0.003	0.008	0.004
14-21 d NEFA	*R*	0.262	0.335	0.372	-0.122	-0.327
	P	0.004	<0.001	<0.001	0.188	<0.001
14-21 d GLU	*R*	-0.315	-0.237	-0.396	0.268	0.213
	P	0.001	0.01	<0.001	0.003	0.021
14-21 d BCS	*R*	0.113	0.254	0.121	-0.09	-0.262
	P	0.223	0.006	0.191	0.33	0.004
BCL	*R*	0.186	0.257	0.148	-0.270	-0.230
	P	0.044	0.005	0.11	0.003	0.012
DL	*R*	-0.037	-0.200	-0.149	0.078	0.137
	P	0.691	0.03	0.107	0.402	0.14

Notes: β-hydroxybutyric acid=BHBA; non-esterified fatty acid=NEFA; glucose=GLU; body condition score=BCS; body condition loss =BCL; daily lactation =DL

Comparison of the two groups of dairy cows with anestrus. From the M-Hχ^2 test analysis method for cohort studies, Tables 5 and 6 show that the anestrus rate of cows in the NEB group was higher than that of the PEB group (42.10% vs. 11.48%) (P<0.01). The RR value is 3.67, which is greater than 3.0 and the 95% confidence interval (1.79-7.52) does not include 1, which is statistically significant (χ^2_{M-H} =12.63, P=0.0004). According to the correlation strength corresponding to the RR value in Table 7, the RR value between 3.0 and 9.0 is a positive correlation. The results show that there was a positive correlation between NEB and anestrus in early lactation, and the risk of anestrus in dairy cows increased 3.67 times.

Table 5. Incidence of anestrus in the two groups of test cows

Groups	Anestrus	Estrus	Total	Anestrus rate/%
PEB	7	54	61	11.48
NEB	24	33	57	42.10
Total	31	87	118	26.27

Table 6. M-Hχ^2 test for cohort studies

χ^2_{M-H}	P value	RR value	95% C.I. $_L$	95% C.I. $_U$
12.63	0.0004	3.67	1.79	7.52

Table 7. Association strength corresponding to RR value

RR value		Correlation strength
0.9-1.0	1.0-1.1	no
0.7-0.8	1.2-1.4	Weak (the former is negatively related, the latter is positively related)
0.4-0.6	1.5-2.9	Medium (same as above)
0.1-0.3	3.0-9.0	Strong (same as above)
<0.1	>10.0	Very strong (same as above)

Discussion

Many studies have reported that the poor reproductive performance of high-yield dairy cows is related to NEB in early lactation (RABOISSON et al., 2014; RUTHERFORD et al., 2016). NEB is usually related to biochemical indicators in the blood, such as high concentrations of BHBA and NEFA, and a low concentration of GLU (HADŽIMUSIĆ and POROBIJA, 2018; GÄRTNER et al., 2019; ĐURIČIĆ et al., 2020). In this study, the plasma BHBA and NEFA levels of the NEB cows from 14 to 21 days postpartum were significantly higher than those of the PEB cows, and plasma GLU levels were significantly lower than those of the PEB cows, which was consistent with the biochemical changes in the blood of the NEB cows. However, previous studies have reported the relationship between plasma BHBA, NEFA, and GLU levels, and various reproductive parameters. For example, in the second week postpartum, the pregnancy rate of cows with plasma BHBA levels higher than 1.4 mmol/L after the first artificial insemination decreased by 20% (WALSH et al., 2007). Higher postpartum plasma NEFA levels and decreased GLU levels are related to increased reproductive disorders and decreased reproductive ability (SHIN et al., 2015). In addition, Walsh, Wathes, Hill et al. also reported the negative relationship between high levels of BHBA and NEFA, and reproduction (WALSH et al., 2007; WATHES et al., 2007; HILL et al., 2018). In this study, the correlation between plasma BHBA, NEFA, GLU levels and reproductive parameters 14 to 21 days postpartum all indicated that the abnormal energy metabolism caused by NEB in early lactation had an adverse effect on the reproductive performance of dairy cows.

It is well known that the loss of body condition in the early lactation period in a herd differs and is related to its future health and reproductive status (ROCHE et al., 2009; LÜTTGENAU et al., 2016). If the BCS of the cow is too high during calving, it will limit postpartum feed intake, resulting in a large loss of postpartum body condition, and making it prone to metabolic diseases and reproductive disorders. On the contrary, if the BCS is too low when the cow is calving, it will affect postpartum lactation due to insufficient nutrient reserves. However, a suitable BCS during calving will reduce postpartum BCL, and can maintain higher lactation and less reproductive disorders (WATHES et al., 2007; BEZDÍČEK et al., 2020). Westwood, Lüttgenau, Goselink, et al. reported that a higher BCL after calving will prolong the ICFE and increase P/AI,

and reduce the estrus and conception rates of cows (WESTWOOD et al., 2002; LÜTTGENAU et al., 2016; GOSELINK et al., 2020). In this study, the BCS and BCL of NEB dairy cows at 14 to 21 days was significantly higher than that of the PEB dairy cows, indicating that postpartum NEB is related to body condition during delivery and postpartum BCL. Since BCS is too high during calving, it will limit the postpartum feed intake of the dairy cows, and the increase in postpartum BCL is due to the loss of fat and protein, which is related to a lack of energy required for lactation (WATHES et al., 2007; ĐURIČIĆ., et al, 2020.). However, BCL is significantly positively correlated with ICFE and P/AI, and significantly negatively correlated with estrus rate, and conception rate, which is consistent with previous studies. Therefore, the adverse effects of NEB in early lactation on reproductive performance are manifested in the prolonged ICFE, increased P/AI, decreased estrus, and conception rates.

The effect of the increase in milk production in early lactation on the reproductive performance of dairy cows is controversial. It is generally believed that the fertility rate of modern dairy cows is declining, especially Holsteins, asa result of genetic selection of offspring with high milk production. However, Leblanc, Bello, and others, have different views on this (LEBLANC, 2010; BELLO et al., 2012). The reproductive success rate within a herd and between herds is widely distributed. A study of 5 California cattle herds, including 6,396 cows, showed that within 90 days postpartum cows with an average DL of 32.1 kg/d were less likely to resume the estrus cycle at 65 days postpartum than cows with 39.1 kg/d, 43.6 kg/d and 50.0 kg/d. Research by Santos et al. showed that milk production does not increase the risk of pregnancy (SANTOS et al., 2009). In this study, the average DL of the NEB cows within 60 days postpartum was extremely significantly lower than that of the PEB cows (36.85±5.31 vs 41.46±5.16 kg/d). The average DL was significantly negatively correlated with the P/AI, which is consistent with the former view. However, the initiation of lactation and high lactation induce NEB in early lactation. Once NEB occurs, it will limit the genetic fertility of modern dairy cows.

Due to the increase in milk production, the nutritional requirements of high-yielding dairy cows increase rapidly within a few weeks after delivery, and most dairy cows enter NEB status. In this case, the cow may preferentially transfer nutrients so that it cannot reproduce, and thus experience a period of anestrus, which may last for several weeks. This state is believed to be the result of the insufficient hormone balance caused by NEB in the early lactation period, and is characterized by decreased blood levels of LH and IGF-1. This can lead to impaired follicular development, restricted estrus signs, reduced LH fluctuations, and delayed ovulation (GUPTA et al., 2014). Many studies using milk progesterone to monitor cattle have shown that NEB in early lactation is not only associated with a higher incidence of anestrous, but also increases the number of days to the first mating, reduces the conception rate, and affects the development of follicles and luteum, and the quality of oocytes (PETER et al., 2009). In this study, the incidence of postpartum fatigue in NEB dairy cows was significantly higher than in the PEB dairy cows, which showed that there was a strong positive correlation between NEB and anestrus in early lactation. The risk of anestrus in cows that experienced NEB 14 to 21 days after delivery was 3.67 times than that of PEB cows. This suggests that NEB cows have a greatly increased risk of postpartum anestrus. Therefore, NEB is an important risk factor for an increase in the postpartum anestrus rate in dairy cows.

ACKNOWLEDGEMENTS

This study was supported by the Key Project of Natural Science Foundation of Heilongjiang Province (ZD2021C006); National Natural Science Foundation of China (31873028).

OFF-LABEL ANTIMICROBIAL DECLARATION

Authors declare no off-label use of antimicrobials.

CONFLICT OF INTEREST DECLARATION

The authors declare that they have no conflict of interest.

References

ALADROVIĆ, J., M. PAVKOVIĆ, B. BEER-LJUBIĆ, L. VRANKOVIĆ, Z. STOJEVIĆ (2018): Metabolic profile in Holstein dairy cow herd. Vet. stn. 49, 9-18.

BELLO, NM., J. S. STEVENSON, R. J. TEMPELMAN (2012): Invited review: milk production and reproductive performance: modern interdisciplinary insights into an enduring axiom. J Dairy Sci. 95,5461-5475.
DOI: 10.3168/jds.2012-5564

BEZDÍČEK, J., A. NESVADBOVÁ, A. MAKAREVICH, E. KUBOVIČOVÁ (2020): Relationship between the animal body condition and reproduction: the biotechnological aspects. Arch Anim Breed. 63,203-209.
DOI: 10.5194/aab-63-203-2020

CARTHY, T. R., D. P. RYAN, A. M. FITZGERALD, R. D. EVANS, D. P. BERRY (2016): Genetic relationships between detailed reproductive traits and performance traits in Holstein-Friesian dairy cattle. J Dairy Sci. 99, 1286-1297.
DOI: 10.3168/jds.2015-9825

DRACKLEY, J. K. (1999): Biology of dairy cows during the transition period: the final frontier? J Dairy Sci. 82, 2259-2273.
DOI: 10.3168/jds.s0022-0302(99)75474-3

ĐURIČIĆ, D., B. B. LJUBIĆ, S. VINCE, R. TURK, H. VALPOTIĆ, I. Ž. ŽAJA, N. MAĆEŠIĆ, M. BENIĆ, I. GETZ, M. SAMARDŽIJA (2020): Effects of dietary clinoptilolite supplementation on β-hydroxybutirate serum level and milk fat to protein ratio during early lactation in Holstein-Friesian cows. Microporous and mesoporous materials, 292, 109766, 4.
DOI:10.1016/j.micromeso

FOLNOŽIĆ, I., R. TURK, D. ĐURIČIĆ, S. VINCE, J. PLEADIN, Z. FLEGAR-MEŠTRIĆ, H. VALPOTIĆ, T. DOBRANIĆ, D. GRAČNER, M. SAMARDŽIJA (2015): Influence of Body Condition on Serum Metabolic Indicators of Lipid Mobilization and Oxidative Stress in Dairy Cows During the Transition Period. Reprod Domest Anim. 50,910-917. DOI: 10.1111/rda.12608

FOLNOŽIĆ, I., R. TURK, D. ĐURIČIĆ, S. VINCE, Z. FLEGAR-MEŠTRIĆ, P. SOBIECH, M. LOJKIĆ, H. VALPOTIĆ, M. SAMARDŽIJA (2016): The effect of parity on metabolic profile and resumption of ovarian cyclicity in dairy cows. Vet. arhiv 86, 641-653.

GÄRTNER, T., E. GERNAND, J. GOTTSCHALK, K. DONAT (2019): Relationships between body condition, body condition loss, and serum metabolites during the transition period in primiparous and multiparous cows. J Dairy Sci. 102, 9187-9199.
DOI: 10.3168/jds.2018-15762

GOSELINK, R. M. A., J. T. SCHONEWILLE, G. DUINKERKEN. VAN, W. H. HENDRIKS (2020): Physical exercise prepartum to support metabolic adaptation in the transition period of dairy cattle: A proof of concept. J Anim Physiol Anim Nutr (Berl). 104, 790-801.
DOI: 10.1111/jpn.13330

GUPTA, P. S., J. K. FOLGER, S. K. RAJPUT, L. L. V, J. YAO, J. J. IRELAND, G. W. SMITH (2014): Regulation and regulatory role of WNT signaling in potentiating FSH action during bovine dominant follicle selection. PLoS One. 9, e100201.
DOI: 10.1371/journal.pone.0100201

HADŽIMUSIĆ, N., A. H. POROBIJA (2018): Effects of seasonal and geographical variation on blood biochemistry parameters of dairy cows. Vet stn. 49, 229-237.

HAILAY, T., M. HOELKER, M. POIRIER, S. GEBREMEDHN, F. RINGS, M. SAEED-ZIDANE, D. SALILEW-WONDIM, C. DAUBEN, E. THOLEN, C. NEUHOFF, K. SCHELLANDER, D. TESFAYE (2019): Extracellular vesicle-coupled miRNA profiles in follicular fluid of cows with divergent post-calving metabolic status. Sci Rep. 9, 12851.
DOI: 10.1038/s41598-019-49029-9

HALAAS, J. L., K. S. GAJIWALA, M. MAFFEI, S. L. COHEN, B. T. CHAIT, D. RABINOWITZ, R. L. LALLONE, S. K. BURLEY, J. M. FRIEDMAN (1995): Weight-reducing effects of the plasma protein encoded by the obese gene. Science. 269,543-546.
DOI: 10.1126/science.7624777

HILL, S. L., K. C. OLSON, J. R. JAEGER, J. S. STEVENSON (2018): Serum and plasma metabolites associated with postpartum ovulation and pregnancy risks in suckled beef cows subjected to artificial insemination. J Anim Sci. 96, 258-272.
DOI: 10.1093/jas/skx033

KLECZKOWSKI, M., W. KLUCIŃSKI, M. CZERSKI, E. KUDYBA (2017): Association between acute phase response, oxidative status and mastitis in cows. Vet. stn. 48, 177-186.

KUHLA, B., C. C. METGES, H. M. HAMMON (2016): Endogenous and dietary lipids influencing feed intake and energy metabolism of periparturient dairy cows. Domest Anim Endocrinol. 56 Suppl, S2-S10.
DOI: 10.1016/j.domaniend.2015.12.002

LEBLANC, S. (2010): Assessing the association of the level of milk production with reproductive performance in dairy cattle. J Reprod Dev. 56 Suppl, S1-S7.
DOI: 10.1262/jrd.1056s01

LUCY, M. C. (2019): Symposium review: Selection for fertility in the modern dairy cow-Current status and future direction for genetic selection. J Dairy Sci. 102, 3706-3721. DOI: 10.3168/jds.2018-15544

LÜTTGENAU, J., S. PURSCHKE, G. TSOUSIS, R. M. BRUCKMAIER, H. BOLLWEIN (2016): Body condition loss and increased serum levels of non-esterified fatty

acids enhance progesterone levels at estrus and reduce estrous activity and insemination rates in postpartum dairy cows. Theriogenology. 85, 656-663.
DOI: 10.1016/j.theriogenology.2015.10.003

MEZZETTI, M., M. BIONAZ, E. TREVISI (2020): Interaction between inflammation and metabolism in periparturient dairy cows. J Anim Sci. 98(Suppl 1), S155-S174.
DOI: 10.1093/jas/skaa134

NEGRÓN-PÉREZ, V. M., D. W. FAUSNACHT, M. L. RHOADS (2019): Invited review: Management strategies capable of improving the reproductive performance of heat-stressed dairy cattle. J Dairy Sci. 102, 10695-10710.
DOI: 10.3168/jds.2019-16718

OSPINA, P. A., D. V. NYDAM, T. STOKOL, T. R. OVERTON (2010): Associations of elevated nonesterified fatty acids and beta-hydroxybutyrate concentrations with early lactation reproductive performance and milk production in transition dairy cattle in the northeastern United States. J Dairy Sci. 93, 1596-1603.
DOI: 10.3168/jds.2009-2852

PETER, A. T., P. L. VOS, D. J. AMBROSE (2009). Postpartum anestrus in dairy cattle. Theriogenology. 71, 1333-42.
DOI: 10.1016/j.theriogenology.2008.11.012.

RABOISSON, D., M. MOUNIÉ, E. MAIGNÉ (2014): Diseases, reproductive performance, and changes in milk production associated with subclinical ketosis in dairy cows: a meta-analysis and review. J Dairy Sci. 97: 7547-7563.
DOI: 10.3168/jds.2014-8237

ROCHE, J. R., N. C. FRIGGENS, J. K. KAY, M. W. FISHER, K. J. STAFFORD, D. P. BERRY (2009): Invited review: Body condition score and its association with dairy cow productivity, health, and welfare. J Dairy Sci. 92, 5769-5801.
DOI: 10.3168/jds.2009-2431

RUTHERFORD, A. J., G. OIKONOMOU, R. F. SMITH (2016): The effect of subclinical ketosis on activity at estrus and reproductive performance in dairy cattle. J Dairy Sci. 99, 4808-4815.
DOI: 10.3168/jds.2015-10154

SANTOS, JE., HM. RUTIGLIANO, MF. SÁ. FILHO (2009): Risk factors for resumption of postpartum estrous cycles and embryonic survival in lactating dairy cows. Anim Reprod Sci. 110, 207-221.
DOI: 10.1016/j.anireprosci.2008.01.014

SHIN, E. K., J. K. JEONG, I. S. CHOI, H. G. KANG, T. Y. HUR, Y. H. JUNG, I. H. KIM (2015): Relationships among ketosis, serum metabolites, body condition, and reproductive outcomes in dairy cows. Theriogenology. 84, 252-260.
DOI: 10.1016/j.theriogenology.2015.03.014

SZENCI, O., Z. SZELÉNYI, L. LÉNÁRT, D. BUJÁK, L. KOVÁCS, L. F. KÉZÉR, A. HORVÁTH (2018): Importance of monitoring the peripartal period to increase reproductive performance in dairy cattle. Vet. stn. 49, 297-307.

TURK, R., I. FOLNOŽIĆ, D. ĐURIČIĆ, S. VINCE, Z. FLEGAR-MEŠTRIĆ, T. DOBRANIĆ, H. VALPOTIĆ, M. SAMARDŽIJA (2016): Relationship between paraoxonase-1 activity and lipid mobilisation in transition dairy cows. Vet. arhiv 86, 601-612.

VINCE, S., D. ĐURIČIĆ, H. VALPOTIĆ, D. GRAČNER, I. FOLNOŽIĆ, B. ŠPOLJARIĆ, P. SOBIECH, M. SAMARDŽIJA (2017): Risk factors and prevalence of subclinical ketosis in dairy cows in Croatia. Vet. arhiv 87, 13-24.

WALSH, R. B., J. S. WALTON, D. F. KELTON, S. J. LEBLANC, K. E. LESLIE, T. F. DUFFIELD (2007): The effect of subclinical ketosis in early lactation on reproductive performance of postpartum dairy cows. J Dairy Sci. 90, 2788-2796.
DOI: 10.3168/jds.2006-560

WATHES, D. C., N. BOURNE, Z. CHENG, G. E. MANN, V. J. TAYLOR, M. P. COFFEY (2007): Multiple correlation analyses of metabolic and endocrine profiles with fertility in primiparous and multiparous cows. J Dairy Sci. 90, 1310-1325.
DOI: 10.3168/jds.S0022-0302(07)71619-3

WESTWOOD, C. T., I. J. LEAN, J. K. GARVIN (2002): Factors influencing fertility of Holstein dairy cows: a multivariate description. J Dairy Sci. 85, 3225-3237.
DOI: 10.3168/jds.S0022-0302(02)74411-1

ZHANG, J., G. WANG, C. ZHAO, Y. BAI, S. SHU, Z. FAN, C. XIA (2018): 1H NMR plasma metabolomic profiling of ovarian quiescence in energy balanced postpartum dairy cows. Vet Q. 38, 47-52.
DOI: 10.1080/01652176.2018.1473660

ZHAO, C., S. SHU, Y. BAI, D. WANG, C. XIA, C. XU (2019): Plasma Protein Comparison between Dairy Cows with Inactive Ovaries and Estrus. Sci Rep. 9, 13709.
DOI: 10.1038/s41598-019-49785-8

Received: 19 November 2020
Accepted: 30 November 2020

WANG, Z., Y. SONG, C. ZHAO, Y. BAI, F. ZHANG, C. XIA, S. FU, H. ZHANG, C. XU, L. WU: Povezanost negativne energetske bilance (NEB) s energetskim metabolizmom, proizvodnjom mlijeka i reprodukcijskom sposobnošću mliječnih krava tijekom rane laktacije u pokrajini Heilongjiang, Kina. Vet. arhiv 92, 223-232, 2022.

SAŽETAK

Negativna energetska bilanca (NEB) uzrokuje ekonomske gubitke na farmama za proizvodnju mlijeka diljem svijeta. Cilj ovog rada bio je istražiti utjecaj NEB-e na energetski metabolizam i reprodukciju tijekom rane laktacije krava na farmama za intenzivnu proizvodnju mlijeka pokrajine Heilongjiangu, u Kini. Uzimajući u obzir razine β-hidroksibutrične kiseline (BHBA), glukoze (GLU) i neesterificiranih masnih kiselina (NEFA) te kliničke manifestacije 14-21 dan nakon porođaja, 118 krava podijeljeno je u skupinu s PEB (pozitivna energetska bilanca; BHBA<1,2, GLU>2,8, NEFA<0,7 mmol/l; n=67) i skupinu s NEB (negativna energetska bilanca; BHBA>1,2, GLU<2,8, NEFA>0,7 mmol/L; n=51). Navedeni pokazatelji analizirani su kombinacijom metoda presječnog istraživanja, Pearson-ovog koeficijenta korelacije i prospektivnog kohortnog istraživanja. Rezultati su pokazali da je 14-21 dan nakon porođaja skupina NEB u usporedbi sa skupinom PEB imala višu ocjenu tjelesne kondicije, veći gubitak tjelesne kondicije (BCL), te veće vrijednosti za dušik iz ureje mlijeka, BHBA, NEFA, interval od teljenja do prvog estrusa (ICFE), graviditete po umjetnoj oplodnji (P/AI) i interval između teljenja (P<0,05), a niže vrijednosti za dnevnu proizvodnju mlijeka (DL), mliječni protein, GLU, stopu pojave estrusa i stopu koncepcije. Gubitak tjelesne kondicije (BCL) je bio pozitivno povezan s ICFE i P/AI (P<0,05) i negativno povezan sa stopom pojave estrusa te stopom koncepcije (P<0,05). Dnevna proizvodnja mlijeka (DL) pokazala je negativnu povezanost s P/AI (P<0,05). Uočena je pozitivna povezanost NEB-a i anestrusa (2M-H = 12,63, P = 0,0004), a rizik od anestrusa uzrokovanih NEB povećan je 3,67 puta u odnosu na PEB. Zaključci su pokazali da je NEB usko povezan s BCL, što je čimbenik koji utječe na snižavanje proizvodnje mlijeka i reprodukciju mliječnih krava. Osim toga, NEB se pokazao i kao čimbenik rizika za anestrus u mliječnih krava.

Ključne riječi: mliječna krava; negativna energetska bilanca; rana laktacija; reprodukcijski pokazatelji