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Research Article

Effects of Supplementation with Rumen-Protected Choline and Methionine on Metabolic Profile and Some Reproductive Parameters in Dairy Cattle During Transition Period

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Abstract

This study conducted to determine effect of the rumen-protected choline and methionine supplementation to peripartum dairy cattle on metabolic profile, metabolic disorders and some reproductive parameters. For this, 3-6 year-old, multiparous, healthy and pregnant 32 high-yield Holstein dairy cows were divided into 4 different groups during trial as: a control (CON) group with no supplementation to the ration; a 75 gr/day rumen-protected choline chloride (CHOL) supplemented group, 42 gr/ day methionine (MET) supplemented group, 75 gr/ day choline chloride and 42 gr/ day methionine (MET + CHOL) supplemented group were during three weeks before and after parturation. Blood samples were taken at wk -3, calving, wk 3 and wk 10 and were analyzed. Health problems and reproductive data were recorded during treatment. Statistically significant differences were detected in non-esterified fatty acids (NEFA), insulin like growth factor-1 (IGF-1), glucose, total protein, albumin, direct bilirubin, total cholesterol and very low density lipoproteins (VLDL). Results of this study suggested that rumen-protected choline and methionine supplementation to the rations of dairy cattle affected some metabolic profile parameters. The statistical differences in metabolic profile tests did not affect metabolic diseases and reproductive parameters.

Keywords: Dairy cattle, metabolic profile, methionine, reproductive parameters, rumen-protected choline.

Geçiş Dönemindeki Süt Sığırlarında Korunmuş Kolin ve Metiyonin İlavesinin Metabolik Profil ve Bazı Üreme Parametreleri Üzerine Etkileri

Öz

Bu çalışma, korunmuş kolin ve metiyonin ilavesinin peripartum süt sığırlarında metabolik profil, metabolik hastalıklar ve bazı üreme parametreleri üzerine etkisini belirlemek amacıyla yapılmıştır. Araştırmada, 3-6 yaş arası, en az bir doğum yapmış, sağlıklı ve gebe 32 baş yüksek verimli Holştayn ırkı süt sığırı kullanıldı. Süt sığırları 4 farklı gruba ayrılarak, kontrol (KON) grubunda yer alan süt sığırlarının rasyonlarına deneme süresince herhangi bir katkı yapılmazken, kolin klorür (KOL) grubundaki süt sığırlarının rasyonlarına doğum öncesi 3 hafta ve doğum sonrası 3 hafta boyunca 75 gr/gün dozunda korunmuş formda kolin klorür, metiyonin (MET) grubundaki süt sığırlarının rasyonlarına ise 42 gr/gün dozunda korunmuş formda metiyonin, Metiyonin ve Kolin Klorür (MET + KOL) grubunda yer alan sığırlarada 75 gr/gün dozunda korunmuş formda kolin klorür ve 42 gr/gün dozunda korunmuş formda

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metiyonin ilave edildi. Kan örnekleri doğumdan önce 3. hafta, doğumda ve doğumdan sonra 3. ve 10. haftalarda alındı ve ticari kitlerle analiz edildi. Araştırma esnasında sağlık sorunları ve üreme verileri kaydedildi. NEFA, IGF-1, glikoz, total protein, albumin, direkt bilurubin, total kolestrol ve çok düşük dansiteli lipoproteinlerde istatistiksel olarak anlamlı farklılıklar tespit edildi. Bu araştırmanın sonucunda, süt sığırlarının rasyonlarına korunmuş kolin ve metiyonin ilavesinin bazı metabolik profil parametrelerini etkilediğini göstermiştir. Metabolik profil testlerindeki istatistiksel farklılıklar, metabolik hastalıklar ve üreme parametrelerini etkilemedi.

Anahtar Kelimeler: Süt sığırı, metabolik profil, metiyonin, üreme parametreleri, korunmuş kolin.

1. Introduction

Dairy cattle with high-yield experience a dramatic physiological and metabolic adaptation during the periparturient period (transition period) attended by low dry matter intake, while the nutritional requirements for maintenance, pregnancy and milk production increase rapidly, resulting in a state of negative energy and metabolizable protein balance (Bell, 1995; Xu et al., 2006; Zhou et al., 2016). Thus, high-producing dairy cattle mobilize more body fat to meet this high nutritional requirement, which may lead to produce higher concentrations of NEFA in blood stream during this transition period (Rukkwamsuk et al., 1999; Xu et al., 2006).

The liver undertakes important role in the coordination of fatty acid metabolism (Drackley and Andersen, 2006; Zom et al., 2011). High levels of NEFA, as a result of mobilization of excess body fat, is either oxidized in the liver or circulated from the liver in the form of VLDL. VLDL is mainly synthesized in the liver and consists of a core of endogenously synthesized triacylglycerol surrounded by polar phospholipids and apolipoproteins B-100, C and E (Jorritsma et al., 2000; Sundaram and Yao, 2010). When the capacity of NEFA to be oxidized or circulated in the form of VLDL in the tricarboxylic acid cycle is exceeded, fat accumulation is shaped in the liver cells. Excessive body fat mobilization as a result of negative energy balance causes ketosis and mobilization of high amounts of NEFA, when the capacity to metabolize in the liver is exceeded as a result of fatty liver is formed. Hepatic lipidosis, known as fatty liver disease, is a metabolic disorder that potentially affects health, production and reproduction, which can affect 50% of high-production dairy cattle in the transition period (Jorritsma et al., 2000; Cooke et al., 2007).

Both choline and methionine are important methyl donors in mammals and their availability is significant for various biological functions. Methyl donors are required for synthesis of some important compounds such as phosphatidylcholine and carnitine (Pinotti et al., 2002). Methionine is enhance liver function, reducing triacylglycerol accumulation and improving the metabolic capacity of the liver to orchestrate the metabolic transition into lactation (Osorio et al., 2014; Osorio et al., 2014; Zhou et al., 2016; Zhou et al., 2016; Vailati-riboni et al., 2017) . Moreover, as a lipotropic agent, Methionine is also required for the production of VLDL, and the apolipoprotein production is necessary for the transport of VLDL molecules from the liver (Overton and Waldron, 2004; Piepenbrink et al., 2004). In the context of VLDL synthesis and liver metabolism, choline is a key component for the synthesis of phosphatidylcholine which is the most significant component of VLDL (Vance, 2002; Zom et al., 2011; Shahsavari et al., 2016; Mcguffey, 2017; Humer et al., 2019).

The objective of this study was to evaluate the effect of feeding rumen-protected choline and methionine products individually and in combination on metabolic profile, metabolic disorders and some reproductive parameters during the transitional period of high yield milk cows.

2. Material and Method

2.1. Experimental Design and Treatments

All experimental procedures were approved by The Animal Experimentation Ethics Committee of Bursa Uludag University (protocol 2013-14/4). Three to 6 years old 32 high-yield multiparous, healthy and pregnant Holstein dairy cows were used as material in this study. Four study groups (8 cows ea) were designed based on lactation number, previous lactation yield and body condition score before the close-up as Control (CON), choline (CHOL), methionine (MET) and choline plus methionine group (MET-CHOL), where cows within each group were fed a basal diet. The cows in CHOL treatment group were supplemented with 75 g of rumen protected choline (RPC). The cows in MET treatment group were supplemented with 42 g of rumen protected Methionine. The cows in treatment group MET-CHOL were supplemented with both 75 g RPC and 42 g methionine. Dosage of RPC and methionine were supplied according to the manufacturer's recommendations. All animals were fed the same close-up ration-21±2 to calving, and the same lactation ration from calving to 70 days in milk. Ingredients and chemical composition of the ration were determined according to National Research Council (NRC, 2001) recommendations as indicated by Cetin et al. (2018) (Table 1).

The RPC and methionine were top dressed (Toledo et al., 2017; Cetin et al., 2018) to basal feed once daily at the 7:00 a.m. from -21±2 to 21 days in milk. The rumen-protected choline supplement is reported to include 24% choline chloride, and is protected by spray freezing technology. The methionine used in this study includes 57% of 2-hydroxy-4-methylthio butanic acid isopropyl ester.

2.2. Animal Management and Feed

Experimental studies were conducted in a private Animal Production Training and Research Company in Bursa/Turkey between November 2013 and June 2014. Cows used in this study were housed in a semi-open free-standing stall with automatic feeders. Close-up and lactation rations were mixed daily, and fed as a total mixed ration as previously indicated (Cetin et al., 2108). The animals returned to the farm herds 70 days in milk.

Health problems, such as metritis, retained placenta, milk fever, ketosis, and displaced abomasum were recorded during the trial. Reproductive data including service period, number of services per conception and calving to first insemination period were recorded individually.

Chemical analysis (dry matter, crude protein, ether extract, ash, calcium, phosphorus) of rations were performed according

to AOAC (1990) and detection of neutral detergent fiber and acid detergent fiber was applied according to Van Soest et al. (1991).

2.3. Blood Collection and Analysis

Blood samples were obtained by venipuncture of the coccygeal vein of each cow using vacuum serum and plasma tubes before feeding on Friday in wk -3, calving, wk 3 and wk 10. Blood samples were taken 4 hours after feeding to determine BHBA. Plasma and serum samples were collected after centrifugation of the blood at 3,000 x g for 15 min. Plasma and serum were stored at -20˚C until analysis.

NEFA (Bovine Non-ester Fatty Acid Elisa Kit, MyBioSource, MBS748204), BHBA (Bovine Beta Hydoxybutyrate Elisa Kit, MyBioSource, MBS046814), IGF-1 (Bovine Insulin like Growth Factor 1 Elisa kit, MyBioSource, MBS737046), Insulin (Bovine Insulin ELISA, ALPCO, 80-INSBO-E01), Glucagon (Bovine Glucagon Elisa Kit, MyBioSource, MBS011427), PON-1 (Bovine Paraoxonase 1, MyBioSource), Apolipoprotein B100 (Bovine Apolipoprotein B100 Elisa Kit, MyBioSource), glucose (Glucose Trinder monoliquid, GL303), total protein (Total Proteins, PT371), albumin (Albumin BCG ALBG045), direct bilirubin (Bilirubin Direct Jendrassik, BDC125), total cholesterol (Cholesterol Total liquid-monocomponent, C20T5), triglycerides (Triglycerides liquid Toos, TG381), GGT (Gamma GT liquid, GT291), ALP (Alkaline Phosphatase (DEA) liquid, AP041), AST (ASAT (GOT) liquid, AS071), LDL (Cholesterol LDL direct liquid, LDL348), HDL (Cholesterol HDL direct liquid, HD320) were assayed following the manufacturer's instructions and recommendations in the commercial kits. The value of VLDL was calculated using the formula triglyceride/5 (Stein and Myers, 1994).

2.4. Statistical method

Metabolic profile parameters and reproductive data were evaluated by 'General Linear Model' where the number cows were entered as random, and duration and groups as fixed effects, respectively. The chi-square test was used to compare the proportional data for metabolic diseases and reproduction, and was selected 'Pearson chi square' or 'Fisher's exact test'. Significance was declared at $p<0.05$. Statistical analysis of data was performed using SPSS program (version 20.0, SPSS Inc, USA) (2011).

Table 1. Ingredients and chemical composition of close-up and early lactation diet

Ingredient	Close-up (%DM[1])	Early lactation(%DM[1])
Wheat straw	21.75	7.26
Alfalfa hay	18.10	21.28
Corn silage	22.22	25.98
Commercial Concentrate mixture[2]	37.26	0.0
Commercial Concentrate mixture[3]	0.0	43.98
Corn gluten	0.0	0.71
Sodium bicarbonate	0.0	0.54
Magnesium oxide	0.0	0.25
Ammonium chloride	0.67	0.0
Chemical composition(%DM[1])		
Neutral detergent fibre	48.15	43.76
Acid detergent fibre	28.60	24.43
Crude protein	13.29	16.43
Ether extract	4.14	5.62
Ash	7.82	7.99
Non-fibre carbonhydrates[4]	26.6	26.2
Calcium	1.02	0.92
Phosphorus	0.28	0.63

[1]Dry Matter
[2]Proyem, Dry Period Concentrate Mixture, Matli Feed Industry, Karacabey/TURKEY
[3]Proyem, Lactation Period Concentrate Mixture, Matli Feed Industry, Karacabey/TURKEY
[4]Non-fibre carbonhydrates, 100-(% NDF + % CP + %EE + % Ash

3. Results and Discussion

The effects of MET, CHOL and MET-CHOL on metabolic parameters are reported in Table 2. Statistically significant differences were detected in NEFA, IGF-1, glucose, total protein, albumin, direct bilirubin, total cholesterol and VLDL. Rumen-protected choline and methionine supplementation had no significant effect on the plasma concentrations of BHBA, insulin, glucagon, PON-1, apo B100, triglycerides, GGT, ALP, AST, LDL and HDL.

As a result of routine control of experimental animals, 12 cows with metritis, 5 cows with retained placenta, 1 cow with milk fever and 6 cows with ketosis were diagnosed and the results are given in Table 3. No statistical differences were found between experimental groups in terms of diseases.

The treatments did not affect significantly on service period, number of services per conception and calving to first insemination period (Table 4). However, in the MET-CHOL group, the service period was shortened and the number of services per conception was decreased.

In this study, we observed significant improvements in lipid metabolism, reproductive performance and welfare of high-yield dairy cattle during transition period, when their feed was supplemented with rumen-protected choline and methionine.

There are previous studies reporting no significant effect of rumen-protected choline supplementation on BHBA, NEFA, glucose and IGF-1 concentrations (Janovick Guretzky et al.,

2006; Zahra et al., 2006; Chung et al., 2009; Leiva et al., 2015). Conversely, Cooke et al. (2007) indicated that blood NEFA concentration in cows decreased in response to rumen-protected choline supplementation. In our study, plasma NEFA concentration was reduced in experiment groups at postpartum 10 week. Moreover, Soltan et al. (2012) reported reduced NEFA in early lactation cows supplemented with rumen-protected choline or both (rumen-protected choline and methionine). On the other hand, some researchers indicated that the effect of methionine supplementation on BHBA concentration was not important (Piepenbrink et al., 2004; Osorio et al., 2013; Zhou et al., 2016) which agrees with the result of the present study. In addition, some researhers declared not important effect of rumen-protected choline or methionine supplementation on plasma BHBA and glucose concentration (Hartwell et al., 2000; Piepenbrink et al., 2004; Strzetelski et al., 2009; Chung et al., 2009; Zom et al., 2011). Besides that, in the present study, no treatments effects were detected for plasma concentration of BHBA. However, plasma glucose concentration was found statistically different compared to treatments group at postpartum 3 week.

Insulin and glucagon are the most important hormones in maintaining glucose balance. In this study, cows supplemented with rumen-protected choline, methionine or both had no significant effect on plasma insulin and glucagon concentrations during the treatments group compared with control cows. But other studies detected to higher insulin values supplemented with rumen-protected choline in cows (Leiva et al., 2015; Zhou et al., 2016).

In the present study, no differences were determined in liver function test analysis such as PON-1, Apo B100, triglycerides, GGT, ALP, AST, LDL and HDL concentrations were not different although total protein and albumin were statistically significant for postpartum 3 week and total cholesterol and VLDL were detected statistically important for postpartum 10 week. Nevertheless, FARID et al. (2013) detected reduced total cholesterol, albumin, HDL, VLDL, LDL, PON-1 and increased NEFA, BHBA, triglycerides in cows suffering from fatty liver.

Lima et al. (2012) showed that feeding rumen-protected choline before and after calving reduced the incidence of ketosis. Ardalan et al. (2009) reported that supplementation of rumen-protected form of choline and methionine had no retained placenta, mastitis, and dystocia even though other groups have reported that a number of metabolic problems occurred. However, in the present study, our results were conflicted with all these results.

Strzetelski et al. (2009) found that supplementation rumen-protected methionine to dairy cow rations during the transitional period had no effect on the reproductive parameters which agrees the result of current study. Ardalan et al. (2009) reported that supplementation of rumen-protected choline and methionine had decreased the open days and services per conception. However, our results from study do not seem to support the result by Ardalan et al. (2009). There were no statistical differences in service period, number of services per conception and calving to first insemination period. In addition, supplemented rumen-protected choline and methionine to the dairy cows ration caused shortening in the service period and decreased number of services per conception.

Table 2. Effect of supplementing Holstein cattle during the transition period with rumen-protected choline, methionine or both on metabolic profile parameters

Parameters	Week	CON	CHOL	MET	MET- CHOL
		$\overline{x} \pm S\,\overline{x}$	$\overline{x} \pm S\,\overline{x}$	$\overline{x} \pm S\,\overline{x}$	$\overline{x} \pm S\,\overline{x}$
NEFA (µmol/L)	-3 week	505.88±85.83	444.68±62.99	413.08±97.42	428.84±64.64
	Calving	593.90±141.44	470.03±42.71	460.15±75.84	451.39±49.30
	3 week	523.76±86.89	469.99±67.95	440.57±55.99	459.91±76.45
	10 week	607.03±111.12^{a}	525.32±62.57^{ac}	474.49±90.76^{bc}	466.37±85.54^{ac}
BHBA (µmol/ L)	-3 week	250.03±181.92	388.68±226.26	212.29±151.28	202.80±122.33
	Calving	374.07±244.96	268.91±171.32	281.27±182.67	241.28±141.89
	3 week	320.92±181.17	275.56±170.04	289.96±223.97	220.38±109.35
	10 week	297.47±202.78	201.84±124.99	186.39±56.12	239.54±111.60
IGF-1 (pg/ml)	-3 week	19011±4101^{a}	12194±6783^{b}	8899±5037^{b}	6002±2128^{b}
	Calving	17806±3241^{a}	17070±7200^{a}	8689±4680^{b}	6174±4884^{b}
	3 week	17438±5193^{a}	18856±9602^{a}	7422±4008^{b}	6477±4712^{b}
	10 week	16869±2237^{a}	13928±6233^{ac}	7964±3420^{bc}	6347±4714^{b}
Insulin (ng/ml)	-3 week	0.24±0.05	0.27±0.07	0.29±0.14	0.26±0.13
	Calving	0.21±0.03	0.20±0.01	0.21±0.04	0.23±0.09
	3 week	0.20±0.01	0.21±0.03	0.19±0.01	0.21±0.06
	10 week	0.21±0.03	0.22±0.02	0.21±0.02	0.25±0.15
Glucagon (pg/ml)	-3 week	35.80±11.98	63.27±47.14	34.51±15.84	34.45±14.19
	Calving	44.86±12.77	55.84±38.07	42.73±22.66	39.33±22.99
	3 week	52.50±11.14	51.41±32.71	41.50±23.72	38.76±19.63
	10 week	43.36±7.25	53.96±39.79	33.53±14.39	56.94±31.11
PON-1 (U/ml)	-3 week	17.11±7.89	17.72±5.33	19.68±8.82	13.76±3.89
	Calving	18.21±6.69	17.86±4.68	22.14±11.02	16.00±4.62
	3 week	15.96±4.09	16.60±6.20	22.73±9.05	16.99±5.13
	10 week	16.67±5.43	19.64±4.81	17.76±9.32	14.19±4.32

Apo B100 (µg/ml)	-3 week	55.48±37.92	68.80±67.04	61.42±62.19	66.05±41.72
	Calving	79.80±63.25	62.30±51.01	108.17±92.79	82.55±49.91
	3 week	76.17±54.12	59.92±44.12	84.80±81.47	92.17±52.23
	10 week	65.67±42.68	103.05±29.24	81.30±63.51	68.17±44.31
Glucose (mg/dl)	-3 week	83.49±3.64	75.33±8.40	83.02±6.90	84.40±7.52
	Calving	78.34±11.32	61.56±17.12	73.69±14.68	66.65±12.99
	3 week	70.30±7.90^{ab}	60.49±11.35^{a}	75.79±12.88^{b}	61.35±9.19^{a}
	10 week	70.37±13.33	76.61±15.12	81.04±11.99	68.89±14.72
Total Protein (g/dl)	-3 week	7.91±0.94	7.89±1.03	8.44±1.04	8.91±2.08
	Calving	7.77±1.75	7.04±1.16	8.14±1.32	8.42±0.93
	3 week	8.99±1.69^{ab}	8.05±0.93^{a}	8.84±0.77^{ab}	9.63±0.57^{b}
	10 week	8.06±1.28	8.08±1.50	9.42±0.58	8.66±1.97
Albumin (g/dl)	-3 week	5.86±0.68	5.49±0.99	5.04±0.90	5.35±0.76
	Calving	5.03±1.10	5.51±0.96	5.64±1.37	5.11±0.34
	3 week	5.95±0.40^{a}	4.98±0.98^{ab}	4.78±1.11^{b}	5.07±0.56^{ab}
	10 week	5.24±0.79	5.51±0.94	6.07±0.94	5.47±0.60
Direct Bilirubin (mg/dl)	-3 week	0.15±0.17	0.13±0.06	0.22±0.15	0.18±0.10
	Calving	0.13±0.12^{a}	0.33±0.15^{b}	0.40±0.07^{b}	0.29±0.09
	3 week	0.18±0.13	0.13±0.08	0.25±0.15	0.20±0.13
	10 week	0.19±0.17	0.17±0.12	0.20±0.14	0.27±0.13
Total Cholesterol (mg/dl)	-3 week	111.28±40.32	143.74±48.14	145.55±40.38	132.55±32.36
	Calving	98.67±41.85	150.10±74.35	107.94±32.60	122.15±35.88
	3 week	131.26±42.17	188.95±69.16	186.25±32.41	177.43±42.74
	10 week	181.10±37.25^{a}	317.54±136.07^{b}	317.48±89.22^{b}	267.66±79.47^{ab}
Triglycerides (mg/dl)	-3 week	25.87±9.51	17.54±9.83	21.28±4.06	21.45±11.04
	Calving	15.44±5.58	13.41±5.26	13.63±5.86	8.60±2.95
	3 week	19.50±8.64	13.71±8.52	15.36±10.84	10.75±7.22
	10 week	37.44±19.22	27.39±16.25	30.60±13.88	29.74±16.18
GGT (U/L)	-3 week	36.56±16.28	26.33±13.86	37.54±11.30	32.13±15.15
	Calving	31.30±18.74	27.47±12.58	39.21±7.57	29.40±15.20
	3 week	26.22±16.65	24.33±16.77	27.24±12.31	33.34±15.33
	10 week	37.44±19.22	27.39±16.25	30.60±13.88	29.74±16.18
ALP (U/L)	-3 week	251.42±145.87	208.08±293.37	182.87±115.62	216.73±101.12
	Calving	239.54±149.53	165.79±101.52	197.46±101.79	221.31±61.75
	3 week	112.87±58.37	142.25±122.47	125.58±66.03	152.98±101.02
	10 week	136.24±51.18	171.56±123.34	103.98±43.51	140.69±112.23
AST (U/L)	-3 week	87.35±46.09	57.67±23.87	72.79±26.10	86.52±33.63
	Calving	47.10±6.97	51.87±16.02	61.71±13.40	63.74±27.70
	3 week	54.44±6.01	53.44±27.78	49.10±14.25	44.13±18.03
	10 week	80.75±36.07	48.55±11.40	68.66±19.93	51.42±12.81
LDL (mmol/L)	-3 week	0.55±0.31	0.83±0.68	1.72±1.19	0.79±0.46
	Calving	0.71±0.47	0.65±0.32	1.08±0.99	0.60±0.39
	3 week	1.20±0.44	1.44±1.10	2.21±1.40	1.02±0.94
	10 week	2.74±0.72	2.66±1.25	4.04±1.52	2.48±0.96
HDL (mmol/L)	-3 week	2.07±0.51	2.34±0.67	1.94±0.78	1.93±0.33
	Calving	1.80±0.36	1.93±0.56	1.79±0.73	1.85±0.55
	3 week	2.19±0.48	2.54±0.40	2.40±1.03	2.00±0.53
	10 week	2.90±0.56	2.62±0.61	3.06±0.72	2.67±0.59
VLDL (mmol/L)	-3 week	0.13±0.05	0.09±0.05	0.11±0.02	0.11±0.06
	Calving	0.08±0.03	0.07±0.03	0.07±0.03	0.04±0.02
	3 week	0.10±0.04	0.07±0.04	0.08±0.06	0.05±0.04
	10 week	0.10±0.03^{a}	0.06±0.01^{b}	0.06±0.02^{b}	0.06±0.03^{b}

NEFA: non-esterified fatty acids, BHBA: beta-hydroxy butyric acid, IGF-1: insulin like growth factor-1 PON-1: paraoxonase-1, Apo B100: apolipoprotein B100, GGT: gamma-glutamyl transferase, ALP: alkaline phosphatase, AST: aspartate aminotransferase, LDL : low density lipoprotein, HDL : high density lipoprotein, VLDL : very low density lipoprotein, Different superscripts indicate stastical differences ^{a,c}: $P< 0.05$

Table 3. Effect of supplementing Holstein cattle during the transition period with rumen-protected choline, methionine or both on some diseases

Parameter	Group	Diseases	
		Yes (%)	No (%)
Metritis	CON	50	50
	CHOL	50	50
	MET	25	75
	MET- CHOL	25	75
Retained placenta	CON	37.5	67.5
	CHOL	0	100
	MET	12.5	87.5
	MET- CHOL	12.5	87.5
Milk fever	CON	0	100
	KOL	0	100
	MET	0	100
	MET- CHOL	12.5	87.5
Ketosis	KON	12.5	87.5
	CHOL	25	75
	MET	12.5	87.5
	MET- CHOL	25	75
Displaced abomasum	CON	0	100
	CHOL	0	100
	MET	0	100
	MET- CHOL	0	100

There were no difference between the groups ($p>0.05$)

Table 4. Effect of supplementing Holstein cattle during the transition period with rumen-protected choline, methionine or both on reproductive parameters

Parameters	CON	CHOL	MET	MET-CHOL
	$\bar{x} \pm S\,\bar{x}$	$\bar{x} \pm S\,\bar{x}$	$\bar{x} \pm S\,\bar{x}$	$\bar{x} \pm S\,\bar{x}$
Service period (days)	109.50±55.28	114.50±47.50	108.75±37.81	94.87±58.38
Number of services per conception	2.37±1.51	2.62±1.19	2.50±1.07	1.75±1.16
Calving to first insemination period (days)	66.12±17.42	64.62±7.41	60.50±15.12	61.50±3.89

There were no difference between the groups ($p>0.05$)

4. Conclusions and Recommendations

In conclusion, current study results suggested that supplementation of rumen-protected choline and methionine to the rations of dairy cows affected some metabolic profile parameters. However, the statistical differences in metabolic profile tests did not change for metabolic diseases and reproductive parameters. More studies must be conducted to evaluate the effects of supplemented with methionine, rumen-protected choline or both during the transition period on metabolic profile and reproductive parameters on dairy cows.

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