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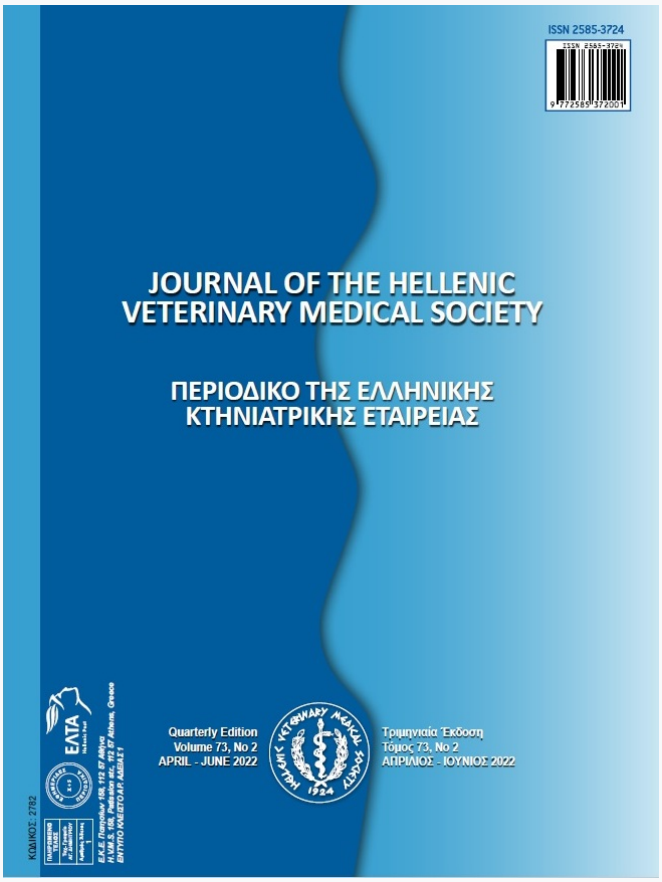

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The Effect of use L-Carnitine on Some Blood Parameters, Milk Yield and Milk Composition and Live Weight in the Transition Period of Dairy Cows

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ABSTRACT: This study aims to determine the effect of supplementing L-carnitine during the transition period on fatty liver, ketosis and milk yield of dairy cows.The control group includes ten cows fed with normal ration (n=10), while the experimental group consists of 11 cows daily supplemented with six-grams of rumen-protected L-carnitine additive orally (n=11). In the study, while there is no statistically significant difference between serum NEFA and BHBA levels between the control group and the experimental group treated with carnitine, the difference in glucose level is statistically significant ($p < 0.001$) in terms of time. While a severe decrease in glucose levels occurs in the control group after birth, the group given carnitine remains within the normal range. The carnitine-treated group's serum BUN value is significantly lower than the control group ($p < 0.05$). Accordingly, L-carnitine neither has a statistically significant effect on Ca, P, GGT, triglyceride values, nor milk yield, milk composition, and live weight. In conclusion, daily oral administration of 6 g L-carnitine increases the use of fatty acids in energy metabolism through the intake into tissues and decreases muscle protein catabolism.

Keywords: Dairy Cattle; Transition Period; L- Carnitine

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INTRODUCTION

The period from three weeks before to three weeks after parturition of dairy cattle is called the transition period (Grummer, 1995). When there is a considerable difference between the glucose amount obtained by daily ration and the body's glucose need, energy requirements are met through the mobilization of stored fat in the body. As a result of this mobilization, the plasma NEFA concentration increases, and different tissues benefit NEFAs in various ways (Drackley, 1999). When NEFAs coming to the liver exceed the existing oxidation capacity, by turning into triglycerides, they accumulate in the liver and thus cause fatty liver and ketosis (Goff and Horst, 1997). Energy is necessary for milk fat and sugar (lactose) synthesized in the udder tissue in high-yielding dairy cows. If the energy required for milk yield cannot be met with the by feed intake, a negative energy balance occurs in the cow's metabolism. As a result of the cow's body fat mobilization, NEFAs are transferred to the liver and then to the udder tissue and participate in the milk fat formation. The energy required in milk sugar is obtained by NEFA oxidation in the liver. If the amount of NEFA coming to the liver exceeds the liver's oxidation capacity, then ketosis occurs. Failure to provide the energy required for milk fat production and sugar through ration plays a significant role in the emergence of ketosis (Ospina *et al.,* 2013).

Carnitine functions in fatty acid metabolism and should not directly in carbohydrate metabolism. However, since carnitine is synthesized from lysine and methionine in the liver, it is associated with carbohydrate metabolism. Besides, it is suggested that a large part of carnitine consists of lysine N-trimethylation as a result of the hydrolysis of proteins. For this reason, low carnitine levels in cows with negative energy balance may be driven by either less carnitine synthesis in the liver or less hydrolysis of proteins. Also, high levels of acylcarnitine can cause low carnitine levels. Acylcarnitines are intermediates formed in lipid and amino acid oxidation and again occur in lipid and amino acid degradation processes. Carnitine plays a role in fatty acid transport in the cell. The inner mitochondrial membrane is impermeable to fatty acids, and a particular carnitine transporter system works to transport activated fatty acids from cytosol to mitochondria. Carnitine is converted into acylcarnitine to transport fatty acid (Xu *et al.,*2018).

Besides being consumed with feed, carnitine is also synthesized in the body. Most of the carnitine in the body is received with feeding. Carnitine received with nutrition is absorbed from the duodenum and jejunum with an active transport mechanism. Carnitine reaches its highest level in the blood within three hours after its absorption. Carnitine is also synthesized in the liver. Lysine and methionine amino acids make up precursors in carnitine biosynthesis. Adequate levels of vitamin B6, nicotinic acid, ascorbic acid, folic acid, and Ferrum must exist in the body as cofactors in carnitine biosynthesis (Carrol and Core, 2001). As the parturition approaches, fat mobilization increases in dairy cows due to dry matter intake decrease because of a drop in appetite. If the energy deficit is excessive, the mobilization of fat exceeds the critical limit, raises to levels that cannot be compensated by the excessive release of NEFA by the liver, and cause the accumulation of triglycerides (TG) in the environment (Drackley, 1999).

Kacar and Citil (2007) investigated the effect of L-Carnitine applications on postpartum period diseases in cows. The study was conducted on 20 pregnant cows. L-Carnitine was administered subcutaneously to the cows in the experimental group (n=10) at a dose of 1g/animal/daily in the last three weeks of gestation, on the birthday, and the 7th day after birth. The control group cows (n = 10) were administered subcutaneously at the same dose as the placebo. While the incidence of difficult births (20%) and endometritis (40%) was higher in control cows than L-Carnitine-treated cows, there was no difference in the incidence of retentio secundinarium. As a result, they concluded that L-Carnitine could prevent difficult births and endometritis cases, but new studies should be carried out on this subject.

In this study, L carnitine use in the dairy cattle's transition period is investigated and aimed to minimize metabolic diseases, especially fatty liver and ketosis, and comorbid diseases.

MATERIALS AND METHODS

This study was conducted in an experimental farm in Karacabey, Bursa, Turkey (Latitude40°14′50″N 28°14′50″E). Animal studies were carried on under the institutional committee's permission on animal use (case no. 2019/18).

Animals

The research covered 21 Holstein-Friesian cows randomly divided into two groups (carnitine n=11, control n=10), in the 3rd, 4th, and 5th lactations with

body condition scores between 3.75 and 4.25 (To investigate the effect of carnitine on lipid metabolism in cows with high body condition score). The eleven cattle in the carnitine group were administered orally 6 g of L-Carnitine to swallow easily in a piece of bread every day from the 21st day prepartum to the 21st day postpartum.

Blood samples

Blood samples were collected via vena coccygea on days -21, -14, -7, and 0 (day of calving) prepartum, and on days 7, 14, 21 postpartum, and then, NEFA, BHBA, BUN, GGT, Triglyceride, Glucose, Ca, and P analyzes were conducted on the study group's blood serums. The methods used in biochemical blood analysis are as follows. Glucose Hexokinase method, Triglyceride Colorimetric method, Blood Urea Nitrogen (BUN) Uv/Enzymatic Kinetic method, Non-esterified Fatty Acids (NEFA) Colorimetric method, Beta-hydroxy butyric acid (BHBA) Uv/ Enzymatic Kinetic method, Gamma Glutamyl Transferase (GGT) Colorimetric method, Calcium (Ca) Colorimetric method, Phosphorus (P) Uv method. Blood parameters affecting lipid and energy metabolism were evaluated. In this study, milk yield, milk composition were examined. The animal's live weights in the carnitine and control groups were measured on the 21st day postpartum.

In the study, the serum NEFA and BHBA levels were not statistically different between the control and carnitine groups, while the glucose level was found to be statistically significant ($p < 0.001$) by weeks. After birth, a severe decrease was observed in the glucose level in the control group, while the carnitine group remained within the normal range. The serum BUN value of the carnitine group was significantly lower than the control group ($p < 0.05$). There was no statistically significant difference in Ca, P, GGT, and triglyceride values ($p > 0.05$). L-carnitine had no statistically significant effect on milk yields, milk compositions and live weight.

Feed Analysis

TMR (Total Mixed Ration) analysis was performed in the Matlı feed factory. NDF, ADF and ADL analysis (in Ankom 200 Fiber Analyzer) was performed according to the procedures (Van Soest et al., 1991). Crude protein, crude cellulose, ether extract (with Soxhlet method by using Ankom XT15 device), ash, dry matter, and starch analysis (by using Atago AP300 device) were performed according to the method of the Association of Official Analytical Chemists (AOAC, 1990).

Milk Sampling and Analysis

Milk samples were taken from the animals in the study on the 21st day postpartum and analyzed in the Matlı feed factory laboratory on the same day. The enterprise uses the GEA herd management & milking system. Milk data are taken from this program.

Statistical Analysis

Descriptive statistics were performed as mean plus/minus standard deviation and standard error. Blood parameters were evaluated by variance analysis in repeated measurements on the -21st, - 14th, -7th, 0th (day of calving), 7th, 14th, and 21st days. When the sphericity assumption could not be achieved, Greenhouse-Geisser correction was used. Differences between groups were evaluated by the Tukey HSD post hoc test. IBM SPSS 23 package program was used for statistical analysis.

$$Y_{ij}=\mu+\alpha_i+\beta_j+(\alpha\beta)_{ij}+\varepsilon_{ij},$$

where Y_{ij} is the dependent variable, μ is the overall mean, α_i is the systemic effect of treatment (group), and β_j is the systemic effect of time.

RESULTS AND DISCUSSION

During the transition period, blood samples were taken from carnitine and control animals at one-week intervals. The difference between the carnitine and control groups was found statistically insignificant in the NEFA, which was examined from blood serum taken on the -21st, -14th, and -7th days prepartum. The difference between the carnitine and control groups in the BHBA values obtained from the blood serum collected on the postpartum 0th, 7th, 14th, and 21st days was statistically insignificant. The time-dependent change was found statistically negligible in both groups (Table 1).

The difference between carnitine and control groups was statistically significant ($p < 0.05$) in glucose measured from blood serum taken on the -21st, -14th, -7th, 0th, 7th, 14th, 21st days. There was a statistically significant difference ($p < 0.001$) on days -7, 0, and +7 in either group, while the difference between other days was insignificant (Table 1). The difference between carnitine and control groups in the triglyceride values measured from blood serum taken on the -21st, -14th, -7th, 0th, 7th, 14th, and 21st days was

not statistically significant. In the carnitine group, the difference due to the decrease between day -7 and day 0 was found statistically significant (p <0.001), while the difference between other days was insignificant. No statistically significant difference was found in the control group (Table 1).

The difference between carnitine and control groups was found statistically significant (P <0.05) in the BUN values acquired from blood serum taken on the -21st, -14th, -7th, 0th, 7th, 14th, and 21st days. In the carnitine group, the difference between the -21st and -7th days was observed statistically significant (p <0.001), while the difference between other days was insignificant. In the control group the increase between the -21st and-14th days and the increase between the -21st and +21st days were determined statistically significant (p<0.001), while the difference between other days was insignificant (Table 2).

The difference between carnitine and control groups was seen statistically insignificant in the calcium values measured in blood serum on the -21st, -14th, -7th, 0th, 7th, 14th, and 21st days. In the carnitine group there was a significant difference between the -7th, 0th, and 7th days (p<0.001), while there was

Table 1. Serum NEFA, BHBA, Glucose and Triglyceride values in the carnitine and control groups in dairy cattle

Parameter	Group	-21.day (X̄± sd)	-14.day (X̄± sd)	-7.day (X̄± sd)	0.day (X̄± sd)	7.day (X̄± sd)	14.day (X̄± sd)	21.day (X̄± sd)	p Group	p Time
NEFA (Non Esterified Fatty Acids)	Carnitine	0.303 ± 0.206	0.207 ± 0.110	0.304 ± 0.254	-	-	-	-	0.460	0,2200
	Control	0.270 ± 0.151	0.250 ± 0.098	0.306 ± 0.237	-	-	-	-		
BHBA (Beta-hydroxybutyrate)	Carnitine	-	-	-	0.391 ± 0.094	0.513 ± 0.182	0.560 ± 0.176	0.546 ± 0,200	0.180	0.0800
	Control	-	-	-	0.473 ± 0.166	0.717 ± 0.329	0.528 ± 0.200	0.652 ± 0.166		
Glucose	Carnitine	33,000 ± 14.241^{a}	50.600 ± 14.623^{a}	50.429 ± 11.356^{a}	88.364 ± 33.699^{b}	48.273 ± 12.515^{a}	41.909 ± 11.059^{a}	40.546 ± 13.382^{a}	0.049	0.0010*
	Control	34.800 ± 11.302^{a}	48.800 ± 10,871^{a}	38.000 ± 10.055^{a}	102.700 ± 34.596^{b}	31.800 ± 13.847^{a}	38.400 ± 19.523^{a}	38.000 ± 14.239^{a}		
Triglyceride	Carnitine	15.000 ± 5.933^{c}	11.700 ± 3.020^{bc}	14.714 ± 4.192^{c}	6.364 ± 2.618^{a}	6.909 ± 3.360^{a}	8.091 ± 3.208 ^{ab}	8.182 ± 1.834^{ab}	0.050	0.0010*
	Control	17.200 ± 5.371	13.900 ± 3.446	18.200 ± 9.908	6.600 ± 3.098	5.700 ± 1.703	16.200 ± 24.908	8.889 ± 2.421		

X̄: mean, sd: standard deviation, * (p <0.05) a, b,c Values with different letters in the same column were found different from each other.

Table 2. Serum BUN, Calcium, GGT and Phosphorus values in carnitine and control groups in dairy cattle

Parameter	Group	-21.day (X̄± sd)	-14.day (X̄± sd)	-7.day (X̄± sd)	0.day (X̄± sd)	7.day (X̄± sd)	14.day (X̄± sd)	21.day (X̄± sd)	p Group	p Time
BUN (Blood Urea Nitrogen)	Carnitine	9.93 ± 3.37^{b}	12.15 ± 2.11^{ba}	13.82 ± 1.72^{a}	11.56 ± 1.87^{ba}	11.16 ± 2.25^{ba}	12.39 ± 1.96^{ba}	12.88 ± 1.70^{ba}	0.049	0.001*
	Control	10.24 ± 0.93^{b}	14.81 ± 3.16^{a}	15.77 ± 3.19^{a}	21.69 ± 3.72^{ba}	13.17 ± 3.88^{ba}	13.64 ± 3.56^{ba}	15.94 ± 4.01^{a}		
Calcium	Carnitine	10.512 ± 1.241^{b}	10.828 ± 0.946^{b}	11.299 ± 0.626^{b}	9.053 ± 0.795^{a}	10.987 ± 0.677^{b}	11.037 ± 0.667^{b}	11.417 ± 0.603^{b}	0.889	0.0010*
	Control	10.542 ± 0.621^{a}	10.916 ± 0.323^{b}	11,323 ± 0,545^{b}	8.993 ± 1.205^{b}	11.075 ± 1.383^{b}	10.981 ± 1.071^{b}	11.536 ± 0.763^{b}		
GGT (Gamma Glutamyl Transferase)	Carnitine	17.64 ± 12.35	18.67 ± 5.24	17.14 ± 5.34	15.82 ± 14.69	17.00 ± 7.98	19.09 ± 7.89	16.36 ± 7.54	0.070	0.06
	Control	11.40 ± 5.27	11.44 ± 5.66	9.20 ± 5.53	10.60 ± 7.89	12.40 ± 8.20	15.00 ± 3.86	16.56 ± 7.40		
Phosphorus	Carnitine	6.761 ± 0.875^{b}	6.720 ± 0.640^{b}	6.866 ± 0.740^{b}	4,258 ± 1,800^{a}	6.248 ± 1.147 ^{b}	7.067 ± 0.644 ^{b}	7.215 ± 0.614^{b}	0.803	0.0001*
	Control	6.799 ± 0.721^{b}	6.258 ± 0.573^{b}	6.478 ± 0.835^{b}	4,137 ± 0,832^{a}	6.428 ± 1.029^{b}	6.763 ± 0.747^{b}	6.702 ± 0.628^{b}		

X̄: mean, sd: standard deviation * (p <0.05) ^{a,b} Values with different letters in the same column were found different from each other.

Table 3. Average milk yield of the experimental groups in the first four weeks after calving (liter)

Parameter	Group	7th day ($\bar{X}$± s)	14th day ($\bar{X}$±s)	21st day ($\bar{X}$ ± s)	28th day ($\bar{X}$± s)	p Group	p Time
Milk Yield (liter)	Carnitine (n=11)	30.55 ± 11.74[a]	41.04 ± 13.76[ab]	45.64 ±14.60[ab]	49.55 ± 16.86[b]	0.83	0.001*
	Control (n=10)	28,35 ± 8,01[a]	40.81 ± 9.85[b]	48.29 ± 10.74[b]	52.00 ± 11.15[b]		

$\bar{X}$: average, s: standard deviation* ($p<0.05$) [a,b] Values with different letters in the same row were found different from each other.

Table 4. The milk values of the experimental groups on the 21st day after calving

Parameter	Group	Measurement on the 21st day after giving birth ($\bar{X}$± s)	p Group
Milk Fat	Carnitine	4.075 ± 0.931	0.21
	Control	3.501 ± 1.026	
Milk protein	Carnitine	3.151 ± 0.372	0.24
	Control	2.992 ± 0.202	
Dry Matter	Carnitine	12.912 ± 1.123	0.10
	Control	12.122 ± 0.940	

$\bar{X}$: average, s: standard deviation ($p>0.05$).

Table 5. Live weight data of experimental groups at parturition and on 21st day after calving

Parameter	Group	Live weight at birth ($\bar{X}$± s)	Live weight on the 21st postnatal day ($\bar{X}$± s)	p Group	p Time
Live Weight	Carnitine	714.545±58.713	712.7273± 30.0303	0.86	1.00
	Control	736.000±59.479	702.2222± 42.9470		

$\bar{X}$: average, s: standard deviation ($p>0.05$)

a significant difference between the -21st and -14th days ($p<0.001$) in the control group (Table 2). The difference between the carnitine and control groups in the phosphorus values obtained from the blood serum taken on the -21st, -14th, -7th, 0th, 7th, 14th, and 21st days were statistically insignificant. .

The data showing the milk yield of the animals in the carnitine and control groups in the postpartum 1st, 2nd, 3rd, and 4th weeks are given (Table 3). When the table was examined, the difference between carnitine and control groups was found to be statistically insignificant. In the carnitine group, there was a significant difference between the 7th and 28th-day milk yields ($p <0.001$), while the difference between the other days was insignificant; in the control group, the difference between days ($p <0.001$) was due to the low milk yield on the 7th day.

A homogeneous sample was taken from the milk of the animals in the carnitine and control groups on the 21st day after giving birth and the data are given (Table 4). As the study was terminated on day 21 postpartum, the samples were taken on day 21. Fat, protein and, dry matter analysis were performed in milk, and there was no statistically significant difference between the carnitine and control groups.

The animal's live weights in the carnitine and control groups were measured on the day of delivery and on the 21st day postpartum when the study ended, and the difference between the two groups was statistically insignificant. In neither group, the time-dependent change was found statistically significant (Table 5).

Values regarding metabolic diseases seen in the carnitine and control group animals during the study are given (Table 6). Values regarding metabolic illnesses observed in the carnitine and control group animals during the research are shown (Table 6). While Clinical ketosis and left displacement of the abomasum (LDA) were observed in the control group animals, no such case was determined in the carnitine group. The chemical composition of experimental rations is in Table 7.

NEFA and BHBA values in blood in transitional dairy cows are parameters that provide crucial in-

Table 6. The Number of metabolic diseases seen in carnitine and control groups

	Hypocalcemia	Retensio secundinarum	Metritis	Clinical ketosis	Left Displacement of the Abomasum	Acidosis
Carnitine group (n=11)	-	-	3	-	-	1
Control group (n=10)	-	1	4	2	2	4

Table 7. Chemical composition of TMR

Chemical composition DM (%)	Nutritive value of TMR for close-up period (% DM)	Nutritive value of TMR for early lactation period (% DM)
DM (Dry matter) %	57.25	49.27
CP (Crude protein) %	14.83	17.72
CA (Crude ash) %	8.03	8.83
CC (Crude cellulose)%	21.31	22.12
EE (Ether extract) %	3.32	4.16
Starch %	20.02	22.75
NEL (Net Energy Lactation) (kcal/kg)	1.48	1.60
NDF (Neutral Detergent Fiber)	38.42	40.79
ADF (Acid Detergent Fiber)	24.10	25.57
ADL (Acid Detergent Lignin)	4.98	5.56

formation about the level of body fat mobilization, state of carbohydrate metabolism, and susceptibility to metabolic diseases (ketosis, fatty liver syndrome, etc.) (Grummer, 1993; Overton and Waldron, 2004). In case the NEFA value is higher than 0.3 mmol/L in prepartum and higher than 0.6 mmol/L in postpartum, and the BHBA value is higher than 1.2 mmol/L in the early lactation period, this situation can cause some problems such as abomasum displacement, ketosis, retention secundinarium, metritis, decrease in pregnancy rate at first artificial insemination and mastitis (Esposito *et al.,* 2014).

Although there was no statistically significant difference in serum BHBA values between the two groups in our study, an increase was observed in the control group compared to the carnitine group on the 7th day after postpartum. Serum BHBA values did not exceed typical values in both groups. There was no significant difference in serum NEFA values measured in the prenatal period. In the study, daily 20g rumen-protected L carnitine was used during the transition period, and it was observed that while serum BHB concentration increased, NEFA concentration was not affected by this situation (Carlson *et al.,* 2006).

It was determined that there was a significant decrease in BHBA values on the 10th and 20th days after parturition in L-Carnitine groups (Rubén *et al.,* 2019). Another study stated that while L carnitine supplementation did not affect NEFA and BHBA concentrations, the triglyceride concentration of L-carnitine-supplemented cows was found significantly higher shortly before calving (Buttchereit *et al.,* 2011).

Differently from our study, serum BHBA concentration was found lower in cattle injected with L-carnitine at a dose of 1gr/animal/day from the last three weeks of gestation until the 7th postnatal day. It was determined that serum BHBA concentration was lower on the 7th day of postpartum in L-carnitine-treated cattle than control cattle, and a statistical difference was observed (p=0.15). However, in parallel to our study, there was no significant difference in NEFA values. Serum glucose level was significantly higher in the carnitine group compared to the control group. In both groups, glucose levels increased on the day of parturition. The carnitine group's serum glucose level did not fall below 40 mg/dl in any week after starting L-carnitine (Kaçar *et al.,* 2013).

Changing parameters in carnitine metabolism are associated with glucose metabolism. Serum glucose concentration was determined to increase in cows treated with carnitine. Fatty acid oxidation in β-cells can be increased in cows treated with carnitine infusion, and decreased use of fatty acids may increase insulin secretion. Conversely, carnitine infusion may affect glucose metabolism by altering insulin sensitivity (Carlson *et al.,* 2006). It is thought the decrease in insulin concentration causes a change in serum glu-

cose. Low serum insulin concentration can increase the oxidation of fatty acids in the pancreas as a result of carnitine administration. The ability of glucose to stimulate insulin secretion from pancreatic β-cells depends on the availability of fatty acids (Stein *et al.,* 1996).

Meyer et al. stated that 25 g/daily rumen-protected L carnitine was used during the transition period, and the triglyceride value in blood plasma increased (Meyer et al., 2020). In another study, daily 20 grams of L carnitine was used during the transition period, and serum triglyceride levels increased. It was thought the increased serum triglyceride level reduced triglyceride accumulation in the liver (Carlson *et al.,* 2006).

In a study, 200 grams of L-Carnitine, which was not bypassed, was given daily during the transition period. It was observed that triglyceride accumulation reduced significantly as to biopsy taken from the liver. In the same study, 100g and 200g of non-bypass L Carnitine were used. And, differently from our research, it was observed that there was a significant increase in blood urea concentrations in either group that was supplemented with L-Carnitine ($p < 0.05$) (Rubén *et al.,* 2019). During calving, plasma urea values are high due to the metabolism of amino acids as a result of cortisol-induced glycogenesis (Hammon *et al.,* 2005). It was observed that the liver increases carnitine synthesis in dairy cows in the early lactation period. It was stated that it is physiologically acceptable for a cow having a negative energy balance should have a sufficient amount of carnitine in its liver in order to carry an excessive amount of NEFA (Schlegel *et al.,* 2012). It was determined that subcutaneous L-carnitine administration in cows in the peripartum period changes BHBA, glucose, and urea concentrations that affect energy metabolism in the early postpartum period but do not affect other biochemical parameters investigated (Kaçar *et al.,* 2013).

Unlike the study we conducted, it was found that when carnitine was given alone or in combination with niacin, it increased milk yield in the first period of lactation in Holstein breeds by 1.5 liters per animal/day. Animals with low milk yield were applied 20 g carnitine support for ten days, and parallel to our study, no positive effect on milk yield was detected, but it was observed that the amount of carnitine in milk increased (Babai and Mezes, 1996).

In Holstein breed cattle with high milk yield (40 kg/day), daily 6g carnitine was given directly to the rumen and abomasum by a cannula. It was observed that direct administration of carnitine to the rumen increased milk yield and the numbers of volatile fatty acids in the rumen. Moreover, it was determined that when carnitine was given directly to the abomasum, it decreased milk yield and increased the amount of protein and dry matter in milk. In both studies, it was found that the amount of free carnitine in milk increased by 25-30% (Drackley and La Count, 1994).

Carlson *et al.* (2006) stated that, since L carnitine supplemented animals have higher serum triglyceride concentrations, their milk-fat percentages can be higher. Unlike our study, Meyer *et al.* (2020) stated that since L carnitine-supplemented cows may have a higher capacity to oxidize NEFA to BHB, this may increase milk-fat synthesis. The daily abomasum infusion of 12g L-carnitine to the dairy cows in lactation did not affect dry matter consumption, milk yield, and milk composition (La Count *et al.,* 1996). It was determined that direct administration of carnitine to the rumen increases the number of volatile fatty acids in the rumen, decreases the amount of free fatty acids in the blood, and increases the amount of glucose (Drakley and La Count, 1993).

In their study, Kacar and Citil (2007) found that the rate of endometritis was higher in the control group cows in the prepartum period compared to L-Carnitine supplemented cows. However, there was no difference in the ratio of retentio secundinarium formation in parallel to our study. They concluded that L-Carnitine could be used to protect against difficult birth and endometritis cases, but this subject is required new investigations. The amount of total and ester carnitine in the blood serum of cows with metritis and retentio-secundinarium was higher than in healthy animals. Sick animals' ester carnitine concentrations in blood serum were detected five times higher than free carnitine concentrations. A high ester carnitine concentration was reported to indicate insufficient energy in the animal (Citil *et al.,* 2003).

CONCLUSIONS

L-carnitine, which was used orally 6g in the transition period, has caused a difference in serum BUN and glucose values but has not demonstrated a significant difference in other parameters. The obtained parameters have shown that L-carnitine has a positive effect on energy metabolism. The transportation amount of unesterified fatty acids to the liver depends on the negative amount of energy. In other words, the

higher the cow's negative energy balance is, the more unesterified fatty acid is transferred to the liver.

The amount of the carnitine carrying fatty acids in the cytosol to the mitochondrial matrix directly determines the number of fatty acids transported from cytosol to the liver mitochondrial matrix, so the carnitine amount is a speed determinant in the oxidation of fatty acids. The more fatty acids come into the mitochondrial matrix, the more acetyl CoA will be formed because of their β-oxidation. If there is not enough oxaloacetate in the liver, the resulting acetyl CoA can not enter the TCA cycle and turns into ketone bodies. As a result, Carnitine is an enzyme enabling the transport of NEFAs resulting from negative energy balance, from the cytosol to the mitochondrial matrix, for gluconeogenesis and ketogenesis in the liver. Therefore, it is recommended in the treatment of ketosis.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

AOAC (1990). Association of Official Analytical Chemists, 16th Ed. Washington, D.C., USA.

Babai K, and Mezes M (1996) Investigation Concerning the Effect of L Carnitine Alone and with Combination of Niacin on the Milk Production of High-Lactating Dairy Cows. Sci Report Budapest: Gödöllö: 2-6.

Buttchereit N, Stamer E, Junge W, Thaller G (2011) Short Communication: Genetic Relationships among Daily Energy Balance, Feed Intake, Body Condition Score, and Fat to Protein Ratio of Milk in Dairy Cows. J. Dairy Sci, 94, 1586-1591. Doi: 10.3168/Jds.2010-3396

Carlson DB, Litherland NB, Dann HM, Woodworth JC, Drackley JK (2006) Metabolic Effects of Abomasal Lkarnitin Infusion and Feed Restriction in Lactating Holstein Cows. J Dairy Sci, 89, 4819-4834. Doi: 10.3168/Jds.S0022-0302 (06)72531-0

Carrol MC, Core E (2001) Carnitine: A Review. Comp. Cont. Educ. Pract. Vet, 23, 45-52.

Citil M, Harmeyer J, Fürrl M (2003) Carnitin Konzentrationen und Weitere Biochemische Parameter im Blutserum Von Gesunden Milchküken Und Kühen Mit Dislocatio Abomasi Sowie Puerperalstörungen. Berl Münch Tierärztl Wschr, 116, 322-327.

Drakley, J. K. and La Count D. W. 1993. Carnitine Suplementation to High Producing Dairy Cows. Progress Report August 2 Dpt. Anim Sci. Unif. Of Illinois.

Drackley JK, La Count DW (1994) Carnitine and A Nutritional Supplement of Dairy Cows. Proc Dairy Seminar Lonza Inc, Fair Lawn, Nj, Premierre Agri Technologies, Inc.

Drackley JK (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

Esposito G, Irons PC, Webb EC, Chapwanya A (2014) Interactions between Negative Energy Balance, Metabolic Diseases, Uterine Health and Immune Response in Transition Dairy Cows. Animaln Reproduction Science, 144 (3), 60-71.Doi: 10.1016/J.Anireprosci.2013.11.007

Goff JP, Horst RL (1997) Physiological Changes at Parturition and Their Relationship to Metabolic Disorders1, 2. Journal of Dairy Science, 80 (7), 1263. Doi: 10.3168/Jds.S0022-0302 (97)76055-7

Grummer RR (1993) Etiology Of Lipit Related Metabolic Disorder in Periparturient Dairy Cows. J Dairy Sci, 76, 3882-3896. Doi: 10.3168/Jds.S0022-0302 (93)77729-2

Grummer RR (1995) İmpact in Changes in Organic Nutrient Metabolizm on Feding the Transition Cow. J. Anim. Sci, 73 (9), 2830-2833.Doi: 10.2527/1995.7392820x

Hammon HM, Philipona C, ZbindenY, Blum JW, Donkin SS (2005) Effects of Dexamethasone and Growth Hormone Treatment on Hepatic Gluconeogenic Enzymes in Calves. J Dairy Sci, 88, 2107-2116. Doi: 10.3168/Jds.S0022-0302 (05)72887-3

Kacar C, Citil M (2007) Gebe İneklerde L-Carnitin Uygulamalarının Güç Doğum, Retentio Secundinarium, Endometritis ve Mastitis Üzerine Etkilerinin Araştırılması, Kafkas Üniv Vet Fak Dergisi, 13 (1): 1-4.

Kacar C, Pancarci ŞM, Karapehlivan M, Kaya S, Kuru M, Çitil M, Gürbulak K (2013) Peripartum Dönemdeki Ineklerde Subkutan L-Karnitin Uygulamalarının Enerji Metabolizmasının Bazı Biyokimyasal Parametrelerine Etkisi, Harran Üniv Vet Fak Dergisi, 2 (2), 67-74.

Lacount DW, Emmert LS, Drackley JK (1996) Dose Response of Dairy Cows to Abomasal Administration of Four Amounts of L-Carnitine. Journal of Dairy Science, *79* (4):591 602, 1996. Doi: 10.3168/Jds.S0022-0302 (96)76404-4

Meyer J, Daniels SU, Grindler S, Tröscher-Mußotter J, Alaedin M, Frahm J, Hüther L, Kluess J, Kersten S, Soosten D, Meyer U, Most E, Eder K, Sauerwein H, Seifert J, Huber K, Rehage J, Dänicke S (2020) Effects of A Dietary L-Carnitine Supplementation on Performance, Energy Metabolism and Recovery From Calving in Dairy Cows. Animals, 10 (2), 1-28. Doi: 10.3390/Ani10020342

Ospina PA, Mcart JA, Overton TR, Stokol T, Nydam DV (2013) Using Nonesterified Fatty Acids and B-Hydroxybutyrate Concentrations During the Transition Period for Herd-Level Monitoring of Increased Risk of Disease and Decreased Reproductive and Milking Performance. Veterinary Clinics of North America: Food Animal Practice, 2 (2), 387-412. Doi: 10.1016/J.Cvfa.2013.04.003

Overton TR, Waldron MR (2004) Nutritional Management of Transition Dairy Cows: Strategies to Optimize Metabolic Health. J Dairy Sci, 87, 105- 119. Doi: 10.3168/Jds.S0022-0302 (04)70066-1

Rubén DG, Liset VM, Nicolás R (2019) L-Carnitine Supplementation Descreases Hepatic Triglyceride Accumulation in Holstein Cows During the Transition Period, Rev Colomb Cienc Pecu, 32 (3), 166-174. Doi: 10.17533/Udea.Rccp.V32n3a01

Schlegel G, Keller J, Hirche F, Geißler S, Schwarz FJ, Ringseis R, Stangl GI, Eder K (2012) Expression of Genes Involved in Hepatic Carnitine Synthesis and Uptake in Dairy Cows in the Transition Period and At Di_Erent Stages of Lactation. Bmc Vet. Res, 8 (28), 1-12. Doi: 10.1186/1746-6148-8-28

Stein DT, Esser V, Stevenson B, Lane KE, Whiteside JH, Daniels MB, Chen S, Mcgarry JD (1996) Essentiality of Circulating Fatty Acids for Glucose-Stimulated Insulin Secretion in the Fasted Rat. J Clin Invest, 97, 2728-2735. Doi: 10.1172/Jci118727

Van Soest PJ, Robertson JB, Lewis BA (1991) Methods for Dietery Fiber, Neıtral Detergent Fiber and Nonstarch Polysaccharides in Relation to Animal Nutrition. J Dairy Sci, 74: 3583-3597. Doi: 10.3168/Jds.S0022-0302 (91)78551-2.

Xu W, Vervoort J, Saccenti E, Van Hoeij R, Kemp B, Van Knegsel A (2018) Milk Metabolomics Data Reveal the Energy Balance of Individual Dairy Cows in Early Lactation. Scientific Reports, 8 (1),1-11. Doi: 10.1038/S41598-018-34190-4