

Relationship between postpartum ovarian cyclicity profiles and some nutritional parameters in imported dairy cows in Algeria

Relación entre los perfiles de ciclicidad ovárica posparto y algunos parámetros nutricionales en vacas lecheras importadas en Argelia

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ABSTRACT

The objective of this study was to evaluate association among certain serum nutritional parameters, body condition score, and body weight, with resumption of ovarian cyclicity postpartum in Prim'Holstein and Montbeliard dairy cows in Algeria. Milk samples were collected from 30 dairy cows twice weekly between days (d) 15 and 60 postpartum and progesterone concentration were determined in whole milk by ELISA. Body condition score and weight were determined at d 0, 30 and 60 postpartum, and blood samples were collected on d 0, 15, 30, 45 and 60 postpartum to determine serum concentration of urea, total proteins, cholesterol, triglycerides, glucose, calcium, phosphorus, aspartate transaminase and alanine transaminase. A cow was considered to have a normal ovarian cyclicity if ovulation occurs before the 35th d postpartum followed by a regular cycle. Results showed that only 40% of cows had normal resumption of ovarian cyclicity. Among abnormal resumption group, delayed first ovulation was the most recorded. Body condition score, body weight, and concentrations of cholesterol, urea and aspartate transaminase change significantly during the 60 d postpartum. Body condition score changed significantly during the first 60 d postpartum ($P<0.0001$). There was an effect of body condition score between the two groups normal resumption and abnormal resumption ($P=0.02$). There was an effect of body condition score and body condition score loss on abnormal resumption and normal resumption groups in all sampling time ($P=0.01$). There was an effect of body condition score loss ≥ 0.75 , and loss < 0.75 on resumption of cyclicity in the first 30 d postpartum. Concentrations of urea and phosphorus were higher in cows with normal resumption than abnormal resumption. Monitoring body condition score is essential to control nutritional management, in order to minimize abnormal resumption of ovarian cyclicity in dairy cows.

Key words: Body condition score; cow; metabolic parameter; ovarian cyclicity; progesterone

RESUMEN

El objetivo de estudio fue evaluar la asociación entre ciertos parámetros nutricionales séricos, la condición corporal y el peso corporal, con la reanudación de la ciclicidad ovárica posparto en vacas lecheras Prim'Holstein y Montbeliard en Argelia. Se recolectaron muestras de leche de 30 vacas lecheras dos veces por semana entre los días (d) 15 y 60 posparto, y se determinó la concentración de progesterona en la leche mediante un ELISA. La condición corporal y el peso corporal se determinaron en los d 0, 30 y 60 posparto, y se recolectaron muestras de sangre en los d 0, 15, 30, 45 y 60 posparto para determinar las concentraciones séricas de urea, proteínas totales, colesterol, triglicéridos, glucosa, calcio, fósforo, aspartato transaminasa y alanina transaminasa. Una vaca se consideró con una ciclicidad ovárica normal si la ovulación ocurrió antes del d 35 posparto, seguida de un ciclo regular. Los resultados mostraron que 40% de las vacas tuvieron una reanudación normal de la ciclicidad ovárica. Entre el grupo con reanudación anormal, la ovulación tardía fue la más registrada. La condición corporal, el peso corporal y las concentraciones de colesterol, urea y aspartato transaminasa cambiaron durante los 60 d posparto. La condición corporal cambió durante los primeros 60 d posparto ($P<0,0001$). Hubo un efecto de la condición corporal entre la reanudación normal y la reanudación anormal ($P=0,02$). La condición corporal y la pérdida de condición corporal tuvieron un efecto en la reanudación anormal y la reanudación normal en todos los momentos de muestreo ($P=0,01$). Hubo un efecto de la pérdida de condición corporal $\geq 0,75$ y pérdida $< 0,75$ en la reanudación de la ciclicidad en 30 d posparto. La concentración de urea y fósforo fueron más altas en la reanudación normal que en la reanudación anormal. Monitoreo de la condición corporal es esencial para controlar el manejo nutricional, con el fin de minimizar la reanudación anormal de la ciclicidad ovárica en vacas lecheras.

Palabras clave: Condición corporal; vaca; parámetro metabólico; ciclicidad ovárica; progesterona

INTRODUCTION

The postpartum (pp) period plays a primordial role in to control reproduction in dairy cows (*Bos taurus*). Full reproductive potential in dairy cows is related to the resumption of ovarian cyclicity in the few weeks postpartum and obtaining pregnancy within the three months after calving [1, 2].

Some authors reported that first ovulation occurred 21 days (d) after parturition in dairy cows had better subsequent reproductive performance than those having first ovulation between 21 and 49 d after calving, which in turn, were better than those ovulating later [3]. The right time to first ovulation after parturition to have the highest reproductive performance remains unclear. Several studies showed that the incidence of abnormal ovarian cycles after calving increased in high producing dairy cows [1, 2, 4, 5].

Lower body reserves and limited energy intake were cited as factors leading to the delay of first normal ovarian cyclicity pp [6]. Nevertheless, the major factor that determine the initiation of ovarian activity and maintaining normal cyclicity is the pp energy balance [6]. Indeed, during early lactation, dairy cows fall in a negative energy balance (NEB) because nutrient demands for milk production increase faster than the intake capacity. Consequently, cows mobilize their body reserves, especially fat and protein, to compensate for the negative energy balance which results in a loss of body weight and body condition, associated with degradation of blood metabolites [7, 8].

Various metabolites have been associated with reproductive performance in dairy cows. Several studies showed the association between the resumption of ovarian cyclicity and nutritional parameters, such blood metabolites, body weight (BW) and body condition score (BCS) [3, 5, 9]. Indeed, the nutrient demand in the first weeks of lactation after calving may increase and body reserves are usually utilized. As a result, the loss of BCS resulted from the energy imbalance increases the incidence of the delayed resumption of the ovarian cyclicity pp [10, 11]. Cows with higher BCS resumed cyclicity pp earlier comparing cows with lower BCS [5]. Moreover, cows that reach nadir BCS later after parturition have lower pregnancy at first AI and longer calving interval than cows reaching nadir BCS early pp [12, 13]. BCS loss is conditioned by the state of the BCS at calving and related to the subsequent reproductive efficiency in dairy cows [14, 15].

There have been some reports on the association between a low β -hydroxybutyrate (BHB) and nonesterified fatty acid (NEFA) levels and the delay of resumption of ovarian cyclicity after calving [16]. It has been reported also that there is a strong correlation between Glucose levels, and BHB and NEFA levels [17]. Associations between delay of pp cyclicity with other blood biochemical parameters and liver function such as glucose, cholesterol, total protein, calcium (Ca), phosphorus (P), aspartate transaminase (AST), alanine transaminase (ALT) were also reported [17].

To evaluate reproductive performance of the two dairy improved cows introduced in Algeria, the present study aims to determine the occurrence of first ovulation pp and profiles of cyclicity in Prim'Holstein and Montbeliard dairy cows, and to investigate the

relationship with BCS, BW and some serum nutritional parameters in the region of Mitidja in Algeria.

MATERIALS AND METHODS

Ethical statement

The Ethical Committee (Ethic code: UNIV25-2019-001) of the Institute of Veterinary Sciences of Constantine (Algeria), approved all procedures of study and it certifies that this work was performed in accordance with local ethical guidelines.

Animals and management

The study was carried out in the Mitidja plain in Algeria, rightly considered as a dairy pond. Cows that calved during the study period were monitored for the calving day up to 60 d after calving. Thirty Prim'Holstein and Montbeliard dairy cows were studied, each cow was visited at d 0 corresponding to the d of calving, and then, twice weekly from d 15, up to d 60.

Milk sampling and progesterone evaluation

Milk samples were collected twice weekly, starting from d 15 to 60 pp before the morning milking and after cleaning the udder and eliminating the first milk jets. About 10 mL of milk was collected in plastic tubes containing 15 mg of potassium dichromate. Samples were stored at -18°C (RAYLAN HM BE1-200, Algeria) until assays were conducted.

Progesterone concentration in whole milk was determined by enzym-linked immunoabsorbent assay (Ovucheck® MILK, Biovet Inc. Canada). The commencement of luteal activity was considered if the milk progesterone concentration is $\geq 5 \text{ ng}\cdot\text{mL}^{-1}$ in at least two consecutive samples [1]. Therefore, ovulation was considered to have occurred 5 d before the first rise of milk progesterone concentration.

Definition of different types of ovarian cycles postpartum

Cows were classified into groups according to progesterone concentrations [1] recorded during the 60 d pp (FIG. 1).

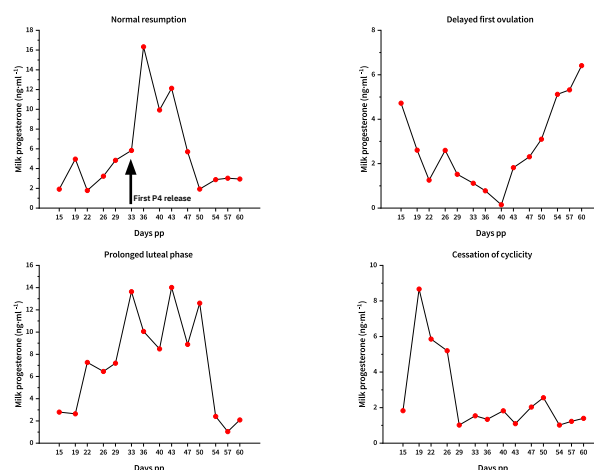


FIGURE 1. Examples of profiles of cyclicity according to milk progesterone concentrations in imported dairy cows in Algeria

- » Normal resumption (NR): Ovulation occurred ≤ 35 d pp, followed by regular ovarian cycle.
- » Abnormal resumption (AR): regroups different forms of abnormal ovarian cyclicity: delayed first ovulation (DFO) (first ovulation did not occur until > 35 d pp), cessation of activity (CA) (ovulation occurred within 35 d pp, and then, there was no luteal activity for at least 14 d between the first and second luteal phases) and prolonged luteal phase (PLP) (Ovulation occurred ≤ 35 d pp, but luteal activity being > 20 d).

Body condition score and body weight monitoring

Body condition score and body weight were monitored at d 0, 30 d and 60 d after calving. The estimation of BW of cows was done using bovimeter tape (Coburn® – The Coburn company, Inc – USA). However, the BCS were assessed using the scale 1–5 (1 = emaciated, 5 = obese) with 0.25 increments [18]. For purpose of analysis of BCS, and BCS changes between normal and abnormal resumption of ovarian cyclicity, the classification of cows according to BCS at calving was as follow: ≥ 3.75 , and < 3.75 , and according to BCS change at 30 and 60 d pp, with BCS loss of ≥ 0.75 , and < 0.75 , or no change and gain of BCS.

Blood sample collection for nutrition parameters

Blood samples were collected at 0 d, 15 d, 30 d, 45 d and 60 d after calving from coccygeal vein into vacutainer tubes. The blood samples were taken to the laboratory (ITELV, Baba Ali), and sera were separated by centrifugation (Sigma Model 3–15, Germany) at $3000 \times G$ for 10 min and frozen at -18°C (RAYLAN HM BE1–200, Algeria) until assayed.

Analytical grade chemicals (SPINREACT, Spain) were used to determine serum concentrations of urea, total proteins, cholesterol, triglycerides, glucose, calcium, P, and the liver enzyme activities: AST and ALT.

Statistical analysis

The incidences of profiles of cyclicity during the 60 d postpartum were expressed as percentages. Data for serum parameters and for BCS and BW were analyzed using GraphPad prism 8. Serum parameters, BCS and BW were presented as means \pm standard deviation (SD). The effects of ovarian resumption groups, time, and interaction group/time were determined. Because serum parameters, BCS and BW were measured over time, these data were analyzed by fitting the fixed effects of time interval, cycling and interaction of the time with cycling in a repeated measures variance analysis. The model used had the following form:

$$Y_{ijk} = m + G_i + T_j + GT_{ik} + e_{ijk}$$

Y_{ijk} = serum parameters, BCS or BW

m = general mean

G_i = effect of the group ($i = 1$ to 2)

T_j = effect of the time to calving ($j = 1$ to 5)

GT_{ik} = effect of the interaction between group and the time to calving ($k = 2 \times 5$)

e_{ijk} = residual error

The only parameter that significantly differed among groups, and in the whole time was BCS. Thus, the effect of BCS on resumption of ovarian cyclicity, and on the interval from calving to the first ovulation pp were further examined. BCS at calving was classified into two categories (≥ 3.75 and < 3.75), and the change in BCS at 30 d and 60 d pp was classified into three categories (≥ 0.75 decrease, < 0.75 decrease and no change). The proportion of cows with NR and AR, was examined with Chi-square test. The interval from calving to first ovulation pp was assessed using, ANOVA and t -test. Significant difference was considered at the level of $P < 0.05$.

RESULTS AND DISCUSSION

The ability to resume ovarian cyclicity in early postpartum is to the good reproductive efficiency in cow, particularly in high producing cows [19]. To obtain the ideal interval calving–calving of twelve months, pregnancy in cows should occur within 3 months pp. This requires early resumption of ovarian cyclicity [1].

First ovulation and ovarian cyclicity profiles postpartum

The current study aims to analyze reproductive performance of two cattle breeds imported for their milk production: Prim'Holstein and Montbeliard. However, their reproduction efficiency remains far less than attained in their country of origin [20]. The study showed that out of 30 cows, only 12 (40 %) had NR of ovarian cycles pp; however, 18 cows (60 %) had AR. Among the AR group, DFO was the most recorded (TABLE I).

TABLE I
Incidence of different types of ovarian cycles during the 60 days pp in Prim'Holstein and Montbeliard dairy cows in Algeria

Cyclicity profiles	No. (%)	Description
Normal resumption	18 (40)	Ovulation occurred ≤ 35 d pp, followed by regular ovarian cycle
DFO	11 (36.7)	first ovulation did not occur until > 35 d pp
PLP	6 (20)	Ovulation occurred ≤ 35 d pp, but luteal activity being > 20 d
CA	1 (3.3)	Ovulation occurred ≤ 35 d pp, but there was an absence of luteal activity for at least 14 d between the first and second luteal phases

Delayed first ovulation (DFO), prolonged luteal phase (PLP); cessation of ovarian activity (CA). (DFO, PLP and CA represent abnormal resumption of cyclicity recorded)

The results are similar to those described by other authors [2, 4, 5]. However, 20 % of cows had PLP. One cow (3.3 %) had resumed cyclicity earlier, and then, there was a cessation of activity. The incidence of abnormal sexual cycles after calving in Prim'Holstein cows was reported in Algeria [4]. It has been reported that cows presenting DFO had a low pregnancy rate and high interval calving–conception comparing to cows with NR [1, 2]. Moreover, cows with

cessation of activity presented a long interval calving–conception comparing with NR according to Gautam *et al.* [1, 2].

In the present study, the first ovulation postpartum occurred before 60 d in all cows. The average interval from calving to the first ovulation was 35.4 ± 11.7 d. The results are in agreement with Gautam *et al.* [1] who considered that the first ovulation pp occurring ≤ 35 d as normal return to ovarian function. However, in other studies, the return to the normal ovarian activity was considered as normal when the first ovulation occurs ≤ 45 d after calving [4, 5]. According to Cheong *et al.* [3], cows with DFO have more severe NEB, which resulting in lower LH pulses, and lower estradiol concentrations inhibiting thus the ovulation of the first dominant follicle.

Relationships between nutritional parameters and profiles of cyclicity pp

BCS and BW

Nutritional status plays a significant role in determining the return of ovarian cyclicity. In the early lactation, the increase in milk production surpasses the intake capacity. As a result, metabolic changes arising from NEB are activated in order to respond to the shortage in energy to cover milk production and maintenance, impairing the subsequent fertility [21]. This study revealed that the loss of ≥ 0.75 unit of BCS was responsible of the apparition of cases of abnormal cyclicity. Indeed, there was a significant association between AR and NR groups and the loss of BCS more or equal to 0.75, less than 0.75, and no change ($P=0.03$) at d 30 after calving, but no effect on the interval from calving to first ovulation pp ($P>0.05$) (TABLE II).

Several studies showed that excessive BCS loss was related with delayed ovarian cyclicity pp [3, 5, 9, 10, 11]. However, Dezetter *et al.* [14] and Manríquez *et al.* [15] showed that in Holstein cows that BCS loss was related to the BCS at calving. They indicated that higher BCS at calving and the high loss of BCS was associated with high reproductive performance. Contrariwise, a low BCS at calving and a low loss of BCS were correlated with the high risk of failure of reproductive performance.

Santos *et al.* [22] showed that cows with DFO pp had reduced dry matter intake in early lactation and greatest BCS losses post-calving. However, increased fat mobilization with an equal BCS does not affect fertility [23].

According to Guáqueta *et al.* [24], the decrease in BCS of cows during the first 60 d pp does not affect the occurrence of first follicular wave in the first 20 d pp. However, the return to a normal cyclicity after first ovulation pp was impaired. Moreover, cows having a value equal or less than 2.5 are the most exposed to the risk of delayed ovulation and abnormal cyclicity [4, 24]. The loss of ≥ 1 unit in early lactation from calving to 30 d pp delays resumption of ovarian cyclicity and increases 2.4 fold the risk of embryonic mortality [22], and cows are unlikely to respond to hormonal treatments [6].

Biochemical parameters

Biochemical parameters are usually used to determine energy status and their influence on the subsequent reproduction in the dairy cows. Indeed, Chen *et al.* [7] demonstrated that cows presenting normal cycle length (between 18 and 24 d) had greater plasma glucose levels and higher energy balance than those with long cycle (more than 24 d) [7]. In the present study, There was no significant change in glucose and triglycerides neither between groups nor during the sampling time and there was no interaction between groups and time ($P>0.05$) (TABLE III).

The results concord with reports of Kalem *et al.* [20]. But Cheong *et al.* [3] indicated that in cows with delayed ovarian cyclicity postpartum, the glucose tolerance was higher; this means a greater insulin resistance. Likewise, in a previous study, high plasma insulin was associated with early resumption of cyclicity [25]. Fat and protein mobilization that provide substrates for gluconeogenesis in the liver maintaining thereby, a normal level of glucose [8], this may explain the maintenance of glucose levels in the two groups in the whole period in the present study. However, in a recent study, glucose levels can reflect the concentrations of NEFA and BHB, since, there was a negative correlation between BHB and glucose, and positive correlation between glucose and NEFA, highlighting

TABLE II
Association between BCS at calving and BCS change, and NR and AR groups, and calving–first ovulation postpartum in Prim'Holstein and Montbeliard dairy cows in Algeria

	Number of cows (%)	Normal resumption (%)	Abnormal resumption (%)	P-value	Calving – First ovulation (days) (mean \pm S.D)	P-value
BCS at calving						
≥ 3.75	7 (23.3)	4 (57.1)	3 (42.9)	$P>0.05$	32.1 ± 11.6	0.4
< 3.75	23 (76.7)	8 (34.7)	15 (65.2)		36.4 ± 11.8	
BCS at d 30 pp						
Loss ≥ 0.75	12 (40)	2 (16.7)	10 (83.3)	$P=0.03$	35.1 ± 14.2	0.9
Loss < 0.75	13 (43.3)	6 (46.2)	7 (53.9)		35.1 ± 10.8	
No change	5 (16.7)	4 (80)	1 (20)		36.8 ± 9.40	
BCS at d 60 pp						
Loss ≥ 0.75	9 (30)	2 (16.7)	7 (38.9)	0.10	30.7 ± 14.3	0.36
Loss < 0.75	16 (53.3)	6 (50)	10 (55.6)		37.6 ± 10.6	
No change	5 (16.7)	4 (33.3)	1 (8.33)		36.8 ± 9.40	

Effects of BCS at calving and BCS loss at d 30 and 60 pp between groups NR and AR, and on the calving – first ovulation interval. Statistical differences are indicated in bold. Body condition score: (BCS), normal resumption: (NR), abnormal resumption: (AR), postpartum: (pp)

TABLE III
Comparison of means and SD of biochemical parameters, BCS and BW between NR and AR groups in Prim'Holstein and Montbeliard dairy cows

Parameters	Group	Day 0	Day 15	Day 30	Day 45	Day 60	Group	P Time	Group×time
Glucose	NR	0.72 ± 0.12	0.64 ± 0.28	0.67 ± 0.28	0.66 ± 0.14	0.68 ± 0.21	0.66	0.75	0.55
	AR	0.67 ± 0.23	0.68 ± 0.24	0.66 ± 0.20	0.65 ± 0.16	0.60 ± 0.19			
Cholesterol	NR	0.67 ± 0.20 ^c	0.85 ± 0.42	0.98 ± 0.39 ^c	1.27 ± 0.66 ^d	1.21 ± 0.68 ^d	0.75	<0.0001	0.7
	AR	0.65 ± 0.24 ^c	0.85 ± 0.35	0.98 ± 0.48 ^d	1.11 ± 0.63 ^d	1.11 ± 0.53 ^d			
Triglycerids	NR	0.21 ± 0.12	0.25 ± 0.11	0.19 ± 0.12	0.21 ± 0.13	0.21 ± 0.07	0.06	0.37	0.09
	AR	0.16 ± 0.12	0.13 ± 0.10	0.14 ± 0.09	0.20 ± 0.16	0.21 ± 0.16			
Urea	NR	0.40 ± 0.17 ^c	0.31 ± 0.13	0.28 ± 0.15 ^d	0.36 ± 0.17	0.38 ± 0.16 ^{c*}	0.03	0.02	0.21
	AR	0.32 ± 0.10 ^c	0.25 ± 0.09	0.23 ± 0.15 ^d	0.24 ± 0.17	0.23 ± 0.17 [*]			
Total proteins	NR	91.7 ± 15.3	89.0 ± 9.60	85.5 ± 24.6	86.5 ± 15.6	78.3 ± 20.3	0.87	0.77	0.05
	AR	81.5 ± 11.6	85.7 ± 14.4	87.2 ± 18.3	90.7 ± 14.0	88.5 ± 12.2			
Calcium	NR	86.6 ± 12.1	89.1 ± 13.9	92.7 ± 15.6	83.7 ± 10.8	89.1 ± 14.5	0.54	0.57	0.36
	AR	85.5 ± 14.6	94.8 ± 13.6	85.7 ± 23.6	90.9 ± 20.5	95.4 ± 20.5			
Phosphorus	NR	90.3 ± 21.8 ^a	81.1 ± 30.7 ^b	81.6 ± 17.5 ^b	77.3 ± 17.2 ^b	73.3 ± 19.6 ^{ab}	0.008	0.14	0.002
	AR	60.8 ± 15.5	63.0 ± 9.94	73.0 ± 14.8	66.2 ± 20.0	62.9 ± 18.8			
AST	NR	53.7 ± 18.5 ^c	43.5 ± 14.1	41.6 ± 17.2	44.7 ± 19.0	39.8 ± 9.10 ^d	0.58	0.006	0.78
	AR	54.1 ± 29.6 ^c	47.3 ± 19.1	38.9 ± 21.4 ^d	39.1 ± 23.4 ^d	32.6 ± 9.60 ^d			
ALT	NR	26.4 ± 16.0	26.0 ± 21.4	19.7 ± 13.0	24.9 ± 10.8	25.0 ± 11.0	0.73	0.91	0.10
	AR	18.5 ± 12.1	21.3 ± 20.7	36.2 ± 53.9	17.7 ± 6.0	20.6 ± 9.30			
BCS	NR	3.42 ± 0.64 ^a		3.08 ± 0.63 ^b		3.06 ± 0.66 ^b	0.02	<0.0001	0.01
	AR	3.25 ± 0.41		2.64 ± 0.33		2.64 ± 0.30			
BW	NR	584 ± 92.5 ^c		556.8 ± 61.3		553.2 ± 79.0 ^d	0.46	<0.0001	0.16
	AR	618.6 ± 76.8 ^c		570.0 ± 76.3		562.9 ± 59.4 ^d			

Parameters that showed significant difference ($P < 0.05$) of interaction group×time are indicated with different superscript (^{a,b,ab}). Different superscript (^{c,d}) indicates a significant difference in sampling time in each parameter in each group. Similar symbol (*) indicates a significant difference ($P < 0.05$) between NR and AR groups. Values without superscripts are not statistically significant. Aspartate transaminase: (AST), Alanine transaminase: (ALT), Body condition score: (BCS), normal resumption: (NR), abnormal resumption: (AR), postpartum: (pp)

the status of NEB [17, 26]. This means that determining the glucose levels can be useful to predict BHB and NEFA levels, and thus, the status of NEB.

A significant change in cholesterol was recorded in the 60 d pp ($P < 0.0001$). Similar findings were reported [27]. However, there was no difference between the two groups NR and AR. Fat mobilization that occurs during the first weeks pp to respond to the high demand for milk production may explain the progressive augmentation of cholesterol. Additionally, the needs of the ovaries for synthesis of steroid hormones confirm the importance of cholesterol. This may explain the fact that all the cows had ovulated before 60 d pp.

Serum urea changed significantly between NR and AR ($P = 0.03$) and in sampling time ($P = 0.02$). Urea level in NR group exceeded the threshold. Results can be explained by the mobilization of a little amount of body proteins after the mobilization of body fat, which increased serum urea concentrations [21]. Kim *et al.* [2] reports that higher or lower concentrations of urea was associated with abnormal cyclicity pp in dairy cows. The concentration of total protein was not different between NR and AR, and did not

statistically change during the 60 d pp. Similar findings reported by Shrestha *et al.* [5] who indicated that the supplementation of crude protein in the diet did not affect the resumption of ovarian activity. Nevertheless, detrimental effect of urea may occur during the pre (antral) stages of the follicle, affecting thus, the oocyte development [21].

In the current study, Calcium did not change significantly in the 60 d pp, and was not different between NR and AR. These findings were in agreement with previous studies [20]. However, there was a significant effect of group NR /AR ($P = 0.008$) and interaction group / time ($P = 0.002$) on P (TABLE III). Kusza *et al.* [28] showed that high reproductive function was correlated with optimum concentrations of calcium. P was higher in NR, but P levels exceeded the threshold only on d 0. Relationship between low P concentrations and infertility were reported [29]. The decrease of P may affect ovarian activity [11]. However, in previous studies, supplementation of P had no effect on the interval calving–first ovulation or the diameter of dominant follicles [29]. Zhou *et al.* [30] showed that supplementation of both Ca and P improve significantly reproductive performance in heifers.

Fat mobilization in early lactation may causing lipid infiltration and its deposition in the liver which can lead to the damage of hepatocytes [31] changing thus, plasma concentrations of AST and ALT. The good liver function is essential to maintain in a good reproductive performance. The augmentation of lipomobilisation in the pp causes the damage of the mitochondria of hepatocytes by fatty infiltration leading to the increase of AST and ALT [31, 32]. In the current study, ALT did not significantly change, but there was a significant change in AST during the first 60 d pp ($P=0.006$). However, there was no effect of group NR / AR or interaction between group and time ($P>0.05$). The results are not in agreement with reports of an association between AST concentrations and delay of first ovulation pp [20, 33, 34, 35]. However, the significant decrease of AST concentrations after four weeks pp was noticed by Rafa *et al.* [36].

The change of BCS and BW was significant in the first 60 d pp in the two groups NR and AR ($P<0.0001$). A significant difference of the BCS between groups NR and AR ($P=0.02$) were noticed, and there was a significant BCS decline in AR group than in RN in all sampling time ($P=0.01$) (TABLE III). Adjorlolo *et al.* [37] indicated that the interval from parturition to resumption of ovarian activity did not differ between cows with and without supplementary nutrients during the pp period. However, lower body reserves can delay return to cyclicity [6]. Saqib *et al.* [10] indicated that marked loss in BW during the pp period was associated with delay in ovarian resumption. The finding of this study showed that despite the significant different change in BW in the first 60 d pp, there was no difference in BW between NR and AR groups. Higher NEB is associated with high rates of tissue mobilization. This can lead to metabolic disorders that increases the risk of ketosis, fatty liver and hypocalcaemia. This metabolic status is more prone delayed of the resumption of normal cyclicity [38].

CONCLUSION

The present study demonstrated that all the cows had resumed their first ovulation before the 60th day postpartum. The calving to first ovulation interval was considered as normal return of the ovarian activity postpartum. However, this resumption was followed by abnormal cycles in 60% of cows. Delayed first ovulation was the most frequent among the abnormal patterns of cyclicity postpartum. The study showed that cholesterol, urea, aspartate transaminase, phosphorus, body condition score and body weight changed significantly during the first 60 days postpartum. Urea and phosphorus were higher in normal resumption group than abnormal resumption. Greater loss of body condition score (≥ 0.75 unit) in the first 30 days postpartum were associated with the increase the risk of abnormal resumption. Marked loss of body condition score in high dairy cows in the postpartum may inhibit timing to the return to a normal cyclicity. Thus, monitoring body condition score of dairy cows in the peripartum is practical and inexpensive. It provides a reliable estimation of body energy reserves In Prim'Holstein and Montbeliard cows. It may be useful in identifying cows with possible delay resumption of ovarian cycles. In addition, monitoring serum metabolites in the peripartum could help in preventing metabolic disorders and enhancing the reproductive efficiency in dairy cows. Additional to this study, nutritional parameters need to be investigated in the dry period to limit higher change of metabolic status and the greater body condition score loss in the early lactation.

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Conflict of interest

The authors declare no conflicts of interest regarding this article's research, authorship, and/or publication.

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