



SERUM BIOCHEMICAL PROFILE DURING EARLY, MID, LATE PREGNANCY, AND CALVING PERIODS IN DAIRY COWS IN ALGERIA

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ABSTRACT

This study aimed to assess the impact of pregnancy on biochemical indices of dairy cows raised in a semi-arid region of western Algeria. A private farm provided forty dairy cows for this study. The cows were divided into four groups based on their pregnancy stages: G1 included pregnant cows at 3 months, G2 included pregnant cows at 6 months, G3 included pregnant cows at 9 months, and G4 included lactating (calving) cows for 1–2 months. All of the cows were raised under the same environmental and management conditions, following the regional customs. Twelve biochemical parameters were measured, including cholesterol (CHO), triglycerides (TG), total protein (TP), albumin (Alb), globulin (Glob), urea (Urea), aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), calcium (Ca), phosphorus (P), and magnesium (Mg). The results of the study showed no significant difference ($P > 0.05$), in the concentrations of various biochemical constituents, among the four groups of dairy cows. The mean value of Glob slightly increased in early pregnancy, compared to late pregnancy, and non-pregnant cows. AST exhibited a significant difference, between non-pregnant, and early-pregnant cows. ALT levels significant decreased in late

pregnant cows, compared with early-pregnant cows. Additionally, Ca levels showed a slight increase, during late pregnancy, compared to non-pregnant cows. As this is the first study of its kind on dairy cows in Algeria, the data generated during this research demonstrated the good management of the herd and can serve as valuable reference values for the scientific community.

Key words: Algeria; biochemical parameters; dairy cows; pregnancy; Tiaret

INTRODUCTION

Since the 1960s, Algeria has implemented measures to promote the breeding of dairy cattle by importing pregnant heifers, with low customs duties, and subsidizing their acquisition, by farmers. In Algeria, dairy cattle breeding, play a crucial role, in the economy, as reported by ONIL [27], with the per capita milk consumption rising from 30.1 litres in 1964, to 145 litres in 2018, surpassing the global average. However, the official statistics show a much lower number of “modern” dairy cows, than expected, if the cows imported since the 1960s and their offspring, had been appropriately bred [19].

Some researchers hypothesize, that the breeding performance in this context, has been poor. The observed discrepancy, seems to be largely explained, by the low reproductive performance, which is attributed to the inadequate maintenance of the herd, in terms of food, and animal health [19]. Additionally, while importing pregnant heifers, has contributed to increased milk production, certain studies indicate, that its full potential, has not been realized [23].

Using blood biochemical characteristics as a pre-symptomatic diagnostic tool, can help in detecting, preventing, and treating infectious diseases, nutritional and metabolic health issues, as well as other disorders, like reproductive, and productivity problems in cattle [1]. However, it's important to consider that factors, such as age, breed, sex, pregnancy, lactation, diet, and health status, can all influence these parameters in cows [24]. In the same context, Turk et al. [32] indicated that all animal species undergo significant changes in blood variables during pregnancy. For dairy cows, the pre-parturient period, is associated with a high incidence of metabolic and reproductive disorders.

Managing the performance of herds in semi-arid environments during this critical period is often: challenging, particularly concerning their well-being, health, and productivity [7, 15].

Developing an animal blood metabolic profile test has proven to be a challenging, due to the variations influenced by factors such as: breed, farming practices, and environmental conditions. As a result, establishing physiological baseline values for different breeds and specific environments has become necessary.

To investigate and determine the specific biochemical parameters in dairy cows during various stages of pregnancy, and the calving period, in the semi-arid region of Tiaret, we conducted the current study. It's noteworthy that, as far as we know, this is the initial research of its kind conducted in Algeria. The primary objective was to compare the different pregnancy stages with cows that were not pregnant.

MATERIALS AND METHODS

Ethical statement

The use of animals was conducted in accordance with worldwide animal welfare standards (Terrestrial Animal

Health Code 2018, section 7, Art 7.5.1), as well as the national executive decree No. 95–363 of November 11, 1995 (Algeria).

Study location

This study was carried out in Tiaret. This region is located in the Centre West of the high plateau region of the country. Its space is heterogeneous, with a mountainous zone in the north, high plateaus in the centre, and semi-arid spaces in the south (35° 23' 17" nord, 1° 19' 22" east). Its morphology and geographical position give it an agro-pastoral character. The town covered around 20.086.63 km² and is located at 1,143 m (altitude of the Pass), on the flanks of Jebel Guezoul, which is part of the Tell Atlas chain. The Köppen-Geiger classification of the climate is Csa. Tiaret sees an average annual temperature of 14.7 °C (58.5 °F). Each year, there is about 529 mm (20.83 in) of precipitation.

Animals and diet

A total of 40 clinically healthy dairy cows (17 Holstein, 23 Montbeliard), were selected from a private farm. These cows were imported as pregnant heifers. The animals were comparable in age (3 and 4 years), body condition score (3.13 ± 0.70), and parity (1.34 ± 0.47). Based on their stage of pregnancy, they were divided into four groups: G1 included pregnant cows at 3 months. G2 included pregnant cows at 6 months, G3 included pregnant cows at 9 months, and G4 included lactating (calving) cows. The study spanned 8 months, from January 2020 to August 2021.

All of the cows, were raised under the same environmental, and management conditions, following regional customs. They were kept in a straw-bedded, free-moving barn, with access to the pasture allowed, only during spring and autumn for a minimum of 6 hours, during daylight (from 08:00 to 14:00 CEST). Milking occurred twice daily, at 5 a.m. and 4 p.m. The cows were fed the same diet, consisting of a total mixed ration (TMR), given twice daily at 08:00 and 14:00 CEST. The diet included a combination of 6 kilograms of bran and concentrates, 8 kilograms of maize ensilage, and 2 kilograms of dried alfalfa per cow. Additionally, the animals had continuous access to fresh water and salt lick.

Collection of blood samples

Aseptically collected blood samples were obtained from each animal in the morning using jugular vein punc-

tures. A 22-gauge needle, coupled with a 10 ml syringe (KD-JECT® III, KDM®, Germany), was inserted into the jugular vein, and the blood was collected in a vacutainer anticoagulant-containing tube (Vacutube®, Algeria). This process was repeated for each animal in each of the four groups.

Biochemical analyses

We conducted the analysis of thirteen blood biochemical metabolites (CHO, TG, TP, Glob, Alb, Urea, AST, ALT, ALP, Ca, P, and Mg) using a split-beam UV/Vis spectrophotometer (OPTIZEN 2120UV Plus, Korea). These blood tests were carried out at the laboratory of the Institute of Veterinary Medicine (Department of Animal Health), University of Tiaret.

Statistical analyses

The data obtained, was recorded in Microsoft Excel 2016. The descriptive analyses were carried out with IBM SPSS, version 21, and all results were presented as a mean $\pm$ standard deviation. Before analysing the data on pregnancy groups, the normality of the data was tested using the Kolmogorov-Smirnova test. The Kruskal-Wallis test and one-way analysis of variance test were used. To evaluate the significance of differences between groups, the Mann-Whitney and Tukey tests were used. The data from the samples of pregnant and calving cows underwent t-tests and Mann-Whitney tests, with a significance level set at $P < 0.05$.

RESULTS

Table 1 reveals that there is no significant difference, in the values, of cholesterol (CHO), triglycerides (TG), total protein (TP), albumin (Alb), Urea, phosphorus (P), and magnesium (Mg) between the groups ($P > 0.05$). However, early pregnancy showed a slightly higher mean value of globulin (Glob) compared to late pregnancy or non-pregnant cows. Aspartate aminotransferase (AST) exhibited a significant difference, between non-pregnant, and early pregnancy, whereas it decreased substantially during late pregnancy. Alanine aminotransferase (ALT) levels significantly decreased, in the late pregnant cows, compared with early pregnant ones. Additionally, the calcium (Ca) levels increased slightly during late pregnancy, compared with non-pregnant cows.

DISCUSSION

The average total plasma cholesterol recorded in our study was similar to the findings reported by Brugère-Picoux [5]. However, our results did not align, with those obtained by Al-Miahy and Saleh [1], where cholesterol concentrations in the serum of lactating cows were significantly decreased compared to pregnant cows. Other researchers, such as Hagawa et al. [14], found that the cholesterol level increased in cows during lactation. According to these authors, the rise in plasma cholesterol in lactating cows

Table 1. Results of biochemical analyses (Mean $\pm$ SD) at different stages of pregnancy

Parameter	Mean $\pm$ SD	Max-Min	Mean $\pm$ SD	Max-Min	Mean $\pm$ SD	Max-Min	Mean $\pm$ SD	Max-Min
CHO g.l ⁻¹	1.17 $\pm$ 0.39	1.86–0.30	1.05 $\pm$ 0.18	1.330.84	1.10 $\pm$ 0.32	1.66–0.61	1.02 $\pm$ 0.38	1.58–0.37
TG g.l ⁻¹	0.23 $\pm$ 0.09	0.38–0.12	0.26 $\pm$ 0.08	0.36–0.16	0.20 $\pm$ 0.04	0.29–0.14	0.27 $\pm$ 0.08	0.39–0.16
TP g.l ⁻¹	68.03 $\pm$ 9.66	80.40–53.57	63.65 $\pm$ 13.24	86.77–40.76	61.87 $\pm$ 14.03	72.92–29	61.34 $\pm$ 10.178	80.57–46.72
Alb g.l ⁻¹	13.56 $\pm$ 2.32	16.10–9.10	14.05 $\pm$ 3.41	19.60–8.85	14.60 $\pm$ 2.27	18.05–10.35	13.81 $\pm$ 1.79	17.60–9.80
Glob g.l ⁻¹	54.47 $\pm$ 10.79	69.70–40.07	49.59 $\pm$ 15.49	78.12–22.39	47.27 $\pm$ 13.84	58.30–15.40	47.53 $\pm$ 10.67	67.02–29.12
Urea g.l ⁻¹	0.07 $\pm$ 0.01	0.10–0.05	0.08 $\pm$ 0.01	0.09–0.05	0.08 $\pm$ 0.01	0.10–0.06	0.06 $\pm$ 0.02	0.10–0.04
AST IU.l ⁻¹	59.50 $\pm$ 34.02*	117.8–19.83	43.17 $\pm$ 2.77	79.92–1.17	40.19 $\pm$ 32.55	97.42–2.33	38.01 $\pm$ 30.88*	86.33–4.08
ALT IU.l ⁻¹	41.80 $\pm$ 16.78**	73.50–9.33	29.82 $\pm$ 7.29	47.83–24.50	22.63 $\pm$ 10.92**	36.75–8.75	32.26 $\pm$ 11.51	55.42–15.17
ALP IU.l ⁻¹	129.31 $\pm$ 31.52	172.7–86.9	124.44 $\pm$ 42.77	191.4–68.02	112.64 $\pm$ 59.42	279.4–53.9	113.05 $\pm$ 76.67	361.9–55
Ca mg.l ⁻¹	87.93 $\pm$ 10.17	104.67–79.10	97.51 $\pm$ 26.65	165.03–74.70	99.94 $\pm$ 12.74*	120.6–72.48	82.21 $\pm$ 16.86*	117.9–54.84
P mg.l ⁻¹	87.72 $\pm$ 16.82	126.14 75.24	84.51 $\pm$ 15.25	98.52–50.65	79.48 $\pm$ 13.69	107.01–1.53	79.98 $\pm$ 14.99	99.25–55.93
Mg mg.l ⁻¹	52.97 $\pm$ 25.81	108.67–29.01	53.48 $\pm$ 28.51	98.12–13.14	43.19 $\pm$ 12.87	57.03–20.18	45.63 $\pm$ 29.36	102.58–7.62

Mean $\pm$ Standard deviation (SD), and Maximum – Minimum (Max-Min) values are reported.

Significance of differences: * $P < 0.05$; ** $P < 0.001$;

might be a physiological adjustment to meet the demands of lactation. The fluctuations in serum cholesterol levels in cows during the peripartum period could be attributed to a physiological adaptation to meet the requirements of lactation, similar to animals on a restricted diet [11].

In our study, the average triglycerides (TG) obtained was higher, compared to the reference values of Radoštić et al. [29]. Despite this difference, we found no noticeable variation in plasma triglyceride levels among the different cow groups, which may be attributed to the feed consumed by the cattle.

Interestingly, the plasma triglyceride (TG) concentration in the pregnant group did not show any significant difference, compared to that of the lactating group, contrary to the findings of Kristanto et al. [20]. In a similar vein, Ashmawy [2] observed higher TG concentrations in pregnant buffaloes than during lactation, and [10] noted that TG concentrations were elevated in the three weeks prior to parturition compared to the lactation phase. These authors associated these noteworthy fluctuations in TG concentration in cows with increased energy requirements during lactation. Conversely, the study by Chillar et al. [8] suggested that TG concentrations were influenced by the cows' body condition score (BCS), with higher levels recorded in cows with a BCS above 3.5.

Based on the protein content of the animal's feed, the total protein (TP) content in cows' blood plasma can serve as a hidden indicator of the animal's nutritional status, as it reflects nitrogen metabolism in the organism [21].

The TP levels observed in the studied cows were similar to the values reported for healthy cattle, ranging from 6.2 to 8.2 dg.dl⁻¹, as documented by Kham and Line [18]. Throughout the research period, no significant changes in total protein concentration were observed in these cows. However, the mean plasma TP was slightly decreased in late pregnancy, non-pregnant, and lactating cows, which is consistent with the findings of Al-Miahy and Saleh [1], who also reported no variation in serum total protein, between pregnant, and non-pregnant cows.

For Holstein cows, Yalak et al. [34] recorded lower protein values during the dry and early stages of lactation. Conversely, the study by Pouroucholtamane et al. [28] showed an increase in serum total protein in non-lactating yaks.

Regarding the mean concentration of serum albumin (Alb) in the cow groups of our study, no significant differ-

ence was observed, but the mean concentration of serum Alb was lower than the physiological level for cows, as reported by Radoštić et al. [29]. This finding contrasts with previous studies on dairy cows [10, 24]. The lower serum Alb levels found in our study could indicate that the animals in these groups had a suboptimal digestible crude protein status. According to Herdt [16], the long-term circulation levels of Alb can serve as an indicator of the quantity of digestible crude protein.

In the first trimester of pregnancy, the plasma globulin (Glob) concentration in our cows was higher than the physiologically normal amount described by Radoštić et al. [29]. These findings differ from those obtained by Giansella et al. [13] and Mohammied [26], who found that the breastfeeding stage significantly affects Glob concentrations. Our findings align with those reported by Aziz and Mujalli [4], who discovered that in dairy cows, lactation was accompanied by a decrease in Glob concentration compared to the non-lactation phase.

In our study, no statistically significant differences in blood urea nitrogen concentration were observed in the cows, and the urea values recorded fell within the normal limits as reported by Radoštić et al. [29]. This indicates that the feed protein was efficiently utilized by the rumen microflora [3].

These results are in line with those of Čelėskā et al. [6], who found no variations in urea levels between prepartum and postpartum Frisian-Holland cows. Additionally, our findings differ from those of Djoković et al. [10], who reported that urea levels in pre-partum and late pregnancy were higher than during the lactation period.

Urea is a valuable and accurate measure of protein or energy imbalance and protein utilization efficiency. Blood or milk sera can be used to assess the relationship between dietary protein levels and reproduction [31].

Liver function significantly influences the levels of numerous blood parameters and various physiological systems. Liver enzymes, such as AST and ALT, are commonly used in medicine as indicators of liver function [10].

In our study, the enzyme activity of AST was found to be lower than the reference range, with a significant difference between non-pregnant and pregnant cows in the first trimester ($P < 0.05$). On the other hand, the mean activity of ALT in our research remained within the limits of normal values, as reported by Radoštić et al. [29], with a significant difference between early and late pregnancy ($P < 0.001$).

D a s et al. [9] observed no significant alteration in the concentration of AST and ALT among the three groups at different stages of lactation. In contrast, K h a d i m and A l i [17] reported that ALT activity decreased during the latter three months of pregnancy until delivery, and increased during the lactation period. Higher levels of AST during cows' lactation may indicate the presence of fatty liver syndrome, ketosis, or a lower dry matter intake. Additionally, AST activity in dairy cows can change throughout early lactation and gestation due to metabolic events [17].

According to K u p c z y n s k i and C h u d o b a – D r o w d o w s k a [21], variations in ALT activity do not provide enough information to assess the nature, scope, or reversibility of hepatic alterations. An increase in the activity of these enzymes is often associated with disorders of energy metabolism.

According to R a d o s t i t s et al. [29], the alkaline phosphatase (ALP) activity observed in the cows under investigation remained within appropriate limits, with no statistically significant variations in blood levels between the groups as reported by K a d h i m et al. [17]. These researchers observed heightened ALP levels in pregnant cows, suggesting a potential association with the presence of the corpus luteum, responsible for progesterone production and the establishment and maintenance of pregnancy. Conversely, S h a r e f et al. [30] noted a significant increase ($P < 0.05$) in ALP activity during late pregnancy compared to other periods. In contrast, D a s et al. [9] reported no significant alteration in ALP concentration among the groups at different stages of pregnancy. Regarding calcium levels, our results were within the normal range recorded for healthy cattle by K a h n and L i n e [18], with an increase in late pregnant cows, a significant difference was observed between mid and late pregnancy ($P < 0.05$), which is in agreement with M o h a m m e d et al. [26] in crossbred dairy cows. They demonstrated that there was no significant variation in blood calcium levels throughout pregnancy. The study of F a d l a l l a et al. [12] also reported no appreciable differences in calcium content in dairy cows at various physiological phases. On the other hand, D j o k o v i c et al. [10] found that the concentration of calcium in female cattle in the lactation group was significantly higher than in pregnant cows.

The mean value of phosphorus (P) did not show a significant difference between the groups of pregnant and non-pregnant cows. Our investigation's findings demon-

strate that P levels were within the acceptable ranges reported by L a t i m e r et al. [22]. These results are consistent with those of D j o k o v i c et al. [10] and M o h a m m e d et al. [26], who found no variations in P concentration between pregnant and lactating cows. The amount of phosphorus in the diet is believed to be reflected in the blood phosphorus levels [33], and the analysis of our study's results suggests that the cows' diet strongly influenced blood phosphorus concentration.

Regarding blood serum magnesium, the mean concentration in all groups was higher than the normal range, indicating an adequate daily supply of magnesium and good absorption of magnesium in the cattle's digestive tract. As mentioned in the literature, the concentration of magnesium in livestock blood is influenced by the magnesium content of the feed or magnesium intake and its absorption in the rumen [10].

However, our results contradict the findings of M o h a m m e d et al. [25], who showed that magnesium levels in lactating animals were higher than those in pregnant animals. F e d l a l l a et al. [12] also stated that the concentration of magnesium in the prepartum period was higher than in the postpartum period, with no significant change during lactation.

Based on the study's findings, all groups of cattle exhibited mean blood serum magnesium concentrations that were higher than the considered normal range. This suggests that the cattle have a sufficient daily intake of magnesium and that their digestive systems absorb magnesium well. As supported by the literature, the amount of magnesium present in the feed or the amount consumed and absorbed in the rumen can influence the concentration of magnesium in the blood of cattle [10].

However, our results are in contrast to those of M o h a m m e d et al. [26], who demonstrated that magnesium levels are higher in lactating animals compared to pregnant ones. Additionally, F a d l a l l a et al. [12] reported a higher concentration of magnesium during the prepartum phase.

CONCLUSIONS

In Algeria, dairy cattle breeding play a crucial role in the agricultural economy. To improve this sector, the government has recommended importing heifers with high genetic poten-

tial. However, production and reproductive performance in the country are currently below their true potential. Moreover, genetic merit is significantly influenced by non-genetic factors and can vary across different environments.

Western Algeria, with its focus on agriculture and cattle breeding, has chosen to prioritize dairy farming. To promote successful breeding, it is essential to manage and control reproduction and production according to local conditions. In our study, we did not find significant variations among the biochemical profiles during different stages of pregnancy, and most values fell within the reference intervals. These findings are indicative of the herd's good management in terms of feeding and breeding conditions.

Nutritionists may find value in this study as blood biochemical analysis is commonly used to identify dietary factors affecting productivity in dairy cows. This research contributes valuable insights that can aid in optimizing feeding practices and overall herd management.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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