

METABOLIC PROFILES DURING LACTATION PERIOD IN COWS

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ABSTRACT

This study conducted using 40 healthy lactating mixed breed cows aged 1.8-4 years from the date of 1-10-2018 to 1-1-2019. These cows selected from Sohag Governorate; 40 cows reared on small farms feed on balanced ration; 20 cows from them were at the first time of parturition, 10 cows were at the second time of parturition and 10 cows were at the third time of parturition. 10 cows used as a control animals brought from farmers houses and feed on unbalanced food. Blood and milk samples collected during three stages of lactation (first, second and third month of lactation). On each serum sample insulin, liver enzymes (SGOT, SGPT and ALP), total bilirubin, kidney function tests (urea, creatinine and uric acid), some minerals includes (Ca, phosphorus, Cl, Na, and K), some heavy metals include (Cu, Fe, lead, cadmium and Se), some antioxidants include (GSH, CAT and SOD), some hormones include (TSH in serum and milk leptin hormone) and glucose level were measured. A significant effect of the lactation stages were observed on SGOT, SGPT, urea, creatinine, Na, Ca, phosphorus, Cl, K, Cu, Fe, leptin, TSH, GSH, CAT, SOD, glucose, insulin, while there were no significant differences during the three lactation stages on serum ALP, total bilirubin, uric acid and Se.

Key words: Dairy Cows – Transition Period – Minerals – Antioxidants - Hormones

INTRODUCTION

Dairy animals are an important source of animal protein in Egypt. It considered as main source of the food supply in the form of milk, meat (FAO, 2009) and leather (Sarwer *et al.*, 2009).

Lactation stage causes different variation in concentrations of blood metabolites in dairy cows (Yaylak *et al.*, 2009). Lactation phases affect significantly the metabolic profile and so the variation recorded during different physiological phases is expected during the dairy cow production cycle (Smith and Risco, 2005). During the first stage of lactation, mammary gland activity increased results in energy deficiency and increased lipomobilization from body reserves (Arfuso *et al.*, 2016).

During lactation, mammary gland secretory cells utilize 80% of the blood circulating metabolites for synthesis of milk, depending on the speed of infiltration of precursors of milk compounds (i.e. free amino acids, glucose and fatty acids). All animals

require minerals such as calcium (Ca), and phosphorus (P) for reproduction, growth and lactation, which often affect specific requirements, and contributes as catalytic components of enzymes or regulate several mechanism involved just in pregnancy and lactation (Samardzija *et al.*, 2011).

The level of iron at first month of lactation was less than its level at second month of lactation (Hussain *et al.*, 2001). There was non-significant change in the level of leptin hormone in the first month of lactation when compared to its level in second and third month of lactation (Accorsi *et al.*, 2005). The level of uric acid, SOD, CAT and GSH in the first month of lactation was less than its level at second month of lactation (Pintea *et al.*, 2006).

There was non-significant change in total bilirubin level at first month of lactation when compared to the second month of lactation furthermore non-significant change in calcium and creatinine level (Piccione *et al.*, 2012). The level of ALP in the first month of lactation less than its level at second and third month of lactation (Abdulkareem, 2013).

The level of chloride, potassium and phosphorus in first month of lactation not significant changed when

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compared to its level at second and third month of lactation while, the level of sodium at first month of lactation was less than its level at second month of lactation (Nozad *et al.*, 2014). The level of selenium in the first month of lactation was more than its level at second and third month of lactation (Cong and Xiao., 2016). Fiore *et al.* (2018) said that the level of SGOT at first month of lactation more than its level at second month and at third month while, the urea level at first month of lactation less than its level at second and third month of lactation and reported that the level of insulin and SGPT in lactating buffaloes at first month of lactation less than its level at second month of lactation, there was non-significant change in the level of TSH at first month of lactation when compared to its level at second and third month of lactation. The level of glucose at first month of lactation was less than its level at second month of lactation (Kalasariya *et al.*, 2017).

MATERIALS AND METHODS

This study conducted using 40 healthy lactating mixed breed cows aged 1.8-4 years from the date of 1-10-2018 to 1-1-2019. These cows selected from Sohag Governorate; 40 cows reared on small farms feed on balanced ration; 20 cows from them were at the first time of parturition, 10 cows were at the second time of parturition and 10 cows were at the third time of parturition. 10 cows used as a control animals brought from farmers houses and feed on unbalanced ration.

The studied cows divided into 3 groups that fed on balanced ration (B at first month of lactation, C at second month of lactation and D at third month of lactation. While, the control group A fed on unbalanced ration.

The main components of the ration of healthy dairy cows and hazard control animals illustrated in (table 1 and 2).

Table 1: The main components of balanced ration.

Component	Amount (kg) in 1 ton of feed
Corn	625
Bran	250
Soya meal	100
Mineral mixture	5
Vitamin	2.5
NaCl	12.5
Lime stone	5

Table 2: The main components of unbalanced ration.

Component	Amount (kg) in 1 ton of feed
Bran	550
Rice hay	350
Alfa alfa	100

Jugular blood samples collected in plain tubes for blood serum biochemistry. The blood was allowed to clot and centrifuged, then clear blood serum was separated and stored at -20 °C until analyzed and whey milk sample for leptin hormone analysis.

The separated sera used for detection of some liver enzymes, kidney function tests, glucose and antioxidants by using of test kits supplied by Spectrum Egyptian company of biotechnology, Diamound diagnostic company and Biodiagnostic company, Egypt and by the using of (CECIL 3000 SERIES scanning spectrophotometer).

Serum minerals and some heavy metals by using of atomic absorption (SensAA GBC Scientific Equipment) according to (A.O.A.C., 2015).

Insulin, TSH and leptin hormone by using of ELISA according to (A.O.A.C., 2015) by coating the plate with a capture antibody, then sample is added, and any antigen present binds to capture antibody. Detecting antibody is added, and binds to antigen, and then enzyme-linked secondary antibody which is specific to the first antibody is added, and binds to the detecting antibody. Finally substrate is added, and is

converted by enzyme to detectable colored form which could be measured by ELISA plate reader.

Statistical analysis

The obtained data statistically analyzed after the methods described by (Snedecor and Cochran, 1980) analysis of variance, least significant difference and correlation matrix also done by statistical package for social science (SPSS) computer program and ANOVA test.

RESULTS

In the present study we measured some liver function tests include (SGOT, SGPT, ALP and Total bilirubin) by using spectrophotometer, the result revealed that the mean values were (59.2 u/ml, 36 u/ml, 125.9 u/ml, 1.02 mg/dl) respectively in animals feed on unbalanced ration , while in animals feed on balanced ration in the 1st month of lactation were (41.3 u/ml, 38.6 u/ml, 121.9 u/ml, 0.83 mg/dl) respectively ,in the 2nd month of lactation were (48.4 u/ml, 36.3 u/ml, 123.2 u/ml, 0.92 mg/dl) respectively, in the 3rd month of lactation were (48 u/ml, 36.5 u/ml, 123.3 u/ml, 0.84 mg/dl) respectively.

In addition to ,we measured some kidney function tests include (Urea, Creatinine and Uric acid) by using spectrophotometer, the result revealed that the mean values were (35.2 mg/dl, 1.19 mg/dl, 4.99 mg/dl) respectively in animals feed on unbalanced ration, while in animals feed on balanced ration in the 1st month of lactation were (21.1mg/dl, 0.88 mg/dl, 4.7 mg/dl) respectively ,in the 2nd month of lactation were (27.8 mg/dl, 1.01 mg/dl, 4.99 mg/dl) respectively, in the 3rd month of lactation were (29 mg/dl, 1.1 mg/dl, 4.99 mg/dl) respectively.

Some antioxidants include (GSH, SOD and CAT) were measured by using spectrophotometer, the result revealed that the mean values were (18.4 mu/ml, 1.28 u/ml, 4.02 u/I) respectively in animals feed on unbalanced ration , while in animals feed on balanced ration in the 1st month of lactation were (25.2mu/ml, 3 u/ml, 5.23 u/I) respectively, in the 2nd month of lactation were (24.2 mu/ml, 2.2 u/ml, 4.7 u/I) respectively , in the 3rd month of lactation were (20.9 mu/ml, 1.6 u/ml, 4.19 u/I) respectively.

The result of Glucose by using spectrophotometer revealed that the mean value was (40.6 mg/dl) in animals feed on unbalanced ration, while in animals feed on balanced ration in the 1st month of lactation was (48 mg/dl), in the 2nd month of lactation was (43.8 mg/dl), in the 3rd month of lactation was (42.1 mg/dl).

Minerals include (Calcium, Phosphorus, Potassium, Chloride and Sodium) measured by using atomic absorption, the result revealed that the mean values were (8.9 mg/dl, 4.75 mg/dl, 3.24 m mol/ ml, 96.5m mol/I, 119.6 m mol/I) respectively in animals feed on unbalanced ration , while in animals feed on balanced ration in the 1st month of lactation were (10.56 mg/dl, 5.8 mg/dl, 4.07 m mol/ ml, 134.3 m mol/I, 141 m mol/I) respectively, in the 2nd month of lactation were (9.84 mg/dl, 5.44 mg/dl, 3.43 m mol/ ml, 125.7 m mol/I, 129.3 m mol/I) respectively, in the 3rd month of lactation were (9.05 mg/dl, 4.87 mg/dl, 3.05 m mol/ ml, 117.9m mol/I, 114.2 m mol/I) respectively.

The result of Cupper, Iron and Selenium revealed that the mean values were (63.5 mg/dl, 160.6 mg/dl, 0 ug/dl) respectively in animals feed on unbalanced ration , while in animals feed on balanced ration in the 1st month of lactation were (92 mg/dl, 196.8 mg/dl, 0.58 ug/dl) respectively, in the 2nd month of lactation were (96.7 mg/dl, 214.2 mg/dl, 0.196 ug/dl) respectively, in the 3rd month of lactation were (104.2 mg/dl, 186.5 mg/dl, 0.157 ug/dl) respectively, also we measured (lead and cadmium) and obtained no data in both unbalanced and balanced rations.

The result of TSH in serum and Leptin in milk by using ELISA revealed that the mean values were (2.4, 7.6) respectively in animals feed on unbalanced ration, while in animals feed on balanced ration in the 1st month of lactation were (2.8, 9.6) respectively, in the 2nd month of lactation were (2.7, 12.7) respectively, in the 3rd month of lactation were (3.4, 13.4) respectively.

Finally, Insulin measured by using ELISA, the result revealed that the mean value was (13.8 mg/dl) in animals feed on unbalanced ration, while in animals feed on balanced ration in the 1st month of lactation was (10 mg/dl), in the 2nd month of lactation was (12.7 mg/dl), in the 3rd month of lactation was (13.77 mg/dl).

All these results showed in table (3).

Table 3: Effect of lactation stages on different parameters.

Parameters (X±SD)	Animals feed on on unbalanced ration (A)	Animals feed on balanced ration		
		1 st month (B)	2 nd month (C)	3 rd month (D)
Insulin (mg/dl)	13.8±0.53	10±0.25 ^{AB*}	12.7±0.38*	13.77±0.47*
SGOT (u/ml)	59.2±3.01	41.3±2.11 ^{AB*}	48.4±3.17*	48±1.89*
SGPT (u/ml)	36±2.5	38.6±2.46 ^{AB*}	36.3±2.7*	36.5±2.32*
ALP (u/ml)	125.9±4.41	121.9±7.6	123.2±6.3	123.3±5.4
Total bilirubin(mg/dl)	1.02±0.056	0.83±0.099 ^{AB*}	0.92±0.083*	0.84±0.089
Urea (mg/dl)	35.2±4.9	21.1±3.57 ^{AB*}	27.8±2.57*	29±3.16*
Creatinine (mg/dl)	1.19±0.12	0.88±0.11 ^{AB*}	1.01±0.99*	1.1±0.062*
Uric acid (mg/dl)	4.99±0.63	4.7±0.51	4.99±0.17	4.99±0.36
Calcium (mg/dl)	8.9±0.2	10.56±0.53 ^{AB*}	9.84±0.28*	9.05±0.34*
Phosphorus (mg/dl)	4.75±0.26	5.8±0.15 ^{AB*}	5.44±0.36*	4.87±0.12*
Potassium (mmol/ml)	3.24±0.18	4.07±0.37 ^{AB*}	3.43±0.25*	3.05±0.09*
Chloride (mmol/l)	96.5±4.43	134.3±5.7 ^{AB*}	125.7±3.8*	117.9±5.49*
Sodium (mmol/l)	119.6±1.96	141±2.44 ^{AB*}	129.3±1.8*	114.2±4.19*
Copper (mg/dl)	63.5±4.5	92±5.2 ^{AB*}	96.7±2.2*	104.2±4.5*
Iron (mg/dl)	160.6±12.7	196.8±7.8 ^{AB*}	214.2±6.68*	186.5±10.3*
Selenium (ug/dl)	-----	0.58±0.32 ^{AB*}	0.196±0.07*	0.157±0.04*
GSH(mu/ml)	18.4±1.3	25.2±2.6 ^{AB*}	24.2±2.5*	20.9±1.52*
SOD (u/ml)	1.28±0.13	3±0.4 ^{AB*}	2.2±0.14*	1.6±0.32*
CAT(u/l)	4.02±0.14	5.23±0.20 ^{AB*}	4.7±0.19*	4.19±0.18*
Leptin	7.6±0.5	9.6±0.72 ^{AB*}	12.7±0.8*	13.4±0.53*
TSH	2.4±0.35	2.8±0.7	2.7±0.5	3.4±0.4*
Glucose (mg/dl)	40.6±1.5	48±2.5 ^{AB*}	43.8±1.9*	42.1±2.13*

S.D= Standard deviation \bar{x} = mean values * means *P* values <0.05 was significant.

DISCUSSION

The lactation phase affects significantly the metabolic profile and so the variation recorded during this phase is expected. The lactation phase is a period of great metabolic stress for dairy cows (Rollin *et al.*, 2010).

There was a significant increment in insulin level of second and third month of lactation groups when compared to first month of lactation group, this observation may be attributed to the effect of high concentrate diet is at long term and very small in the early lactation (Campanile *et al.*, 1995). These results agreed with that reported by (Fiore *et al.*, 2014 and 2018) and disagreed with that reported by (Accorsi *et al.*, 2005).

There was a significant increment in SGOT and ALP levels of second and third months of lactating animals when compared to its level in the first month of lactating group while, there was a decrement in the level of SGPT in second and third months when compared to the first month of lactation, this change following physiological or pathological conditions. These results agreed with that reported by (Nozed *et al.*, 2014 and Khan *et al.*, 2016) and disagreed with (Jozwik *et al.*, 2012).

There was non-significant change in total bilirubin level of the third month of lactation groups when

compared to first month of lactation group while, in the second month its level increased, this observation may be due to physical stress. These results agreed with that reported by (Piccione *et al.*, 2012).

There was a significant increment in urea level of second and third month of lactation groups when compared to first month of lactation group, this observation may be attributed to the fact that increased deamination or increased protein intake in dairy animals (Roubies *et al.*, 2006). These results agreed with that reported by (Eman *et al.*, 2014 and Fiore *et al.*, 2018) and disagreed with that recorded by (Accrosi *et al.*, 2005 and Piccione *et al.*, 2012).

There was a significant increment in creatinine level of second and third month of lactation groups when compared to first month of lactation group. These results disagreed with that reported by (Piccione *et al.*, 2012).

There was non-significant decrement in uric acid level of second and third month of lactation groups when compared to first month of lactation group, these results may attributed to oxidative stress in dairy cows. These results agreed with that reported by (Pienta *et al.*, 2006).

There was a significant decrement in calcium, chloride, sodium, potassium and phosphorus levels of

second and third month of lactation groups when compared to first month of lactation group. These results may attributed to physiological status of lactating animals and the high production of milk in the second and third month of lactation in compared to the first month as reported by (Cozzi *et al.*, 2011). The results of chloride, sodium, potassium and phosphorus not agreed with that reported by (Nozad *et al.*, 2014) and the results of both chloride and calcium agreed with that recorded by (Hussien *et al.*, 2001).

There was a significant increment in copper level of second and third month of lactation groups when compared to first month of lactation group.

There was a significant increment in iron level of second month of lactation groups when compared to first month of lactation group while, there was a decrement in iron level in third month of lactation when compared to the first and second month of lactation. These results not agreed with that reported by (Hussien *et al.*, 2001).

There were no data obtained for lead and cadmium in our balanced and un balanced rations. These results may attributed to the healthy environment in which the both healthy and control groups reared.

There was a significant decrement in selenium level of second and third month of lactation groups when compared to first month of lactation group. These results may attributed to the transfer of Se from the blood to the milk produced largely in peak lactation, these results not agreed with that reported by (Jana *et al.*, 2015) and agreed with that recorded by (Cong and Xiao, 2016).

There was a significant decrement in SOD, GSH and CAT levels of second and third month of lactation groups when compared to first month of lactation group, these results may be due to oxidative stress. These results agreed with that reported by (Bernabucci *et al.*, 2002 and Jana *et al.*, 2015) and disagreed with that reported by (Iulia *et al.*, 2012).

There was a significant increment in leptin hormone level of second and third month of lactation groups when compared to first month of lactation group, increasing leptin concentration in milk with time the advancement of lactation. These results agreed with that reported by (Ashmawy, 2015) and disagreed with that reported by (Accrosi, 2005).

There was a significant decrement in TSH level of first and second month of lactation groups when compared to third month of lactation group, these results suggesting a possible role of thyroid gland on the start of lactogenesis. These results agreed with that reported by (Fiore *et al.*, 2018).

There was a significant decrement in glucose level of second and third month of lactation groups when compared to first month of lactation group, these results may lead to over production of ketones. These results disagreed with that reported by (Hagwane *et al.*, 2009 and Kalasariya *et al.*, 2017).

In addition, there were variations between the levels of all parameters except ALP, uric acid and TSH in healthy animals when compared to the hazard control ones that may attributed to the differences in ration.

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صورة المؤشرات الكيموحيوية في مصل دم الأبقار اثناء فترة الحليب

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اشتملت هذه الدراسة على عدد ٤٠ من الأبقار الخليط الحلابة التابعة لمحافظة سوهاج والتي تتراوح أعمارهم من ٢٠ شهر حتى ٤ سنوات وذلك في الفترة من ٢٠١٨/١٠/١ حتى ٢٠١٩/١/١ والتي تم أخذها من مزارع صغيرة تتغذي علي عليقه متزنة من بينهم ٢٠ بقرة بعد الولادة الأولي و ١٠ أبقار بعد الولادة الثانية و ١٠ أبقار بعد الولادة الثالثة. وتم أيضا أخذ ١٠ أبقار من منازل بعض المربين والتي تتغذي علي عليقه غير متزنة كمجموعة ضابطة. وقد تم تجميع عينات الدم واللبن من الأبقار خلال شهر الحليب الثلاثة الأولي. وقد تم قياس نسبة بعض إنزيمات الكبد من بينها (إنزيم ناقل أمين الالانين وإنزيم ناقل أمين الاسبرتات وإنزيم الكالين فوسفاتيز) وأملاح الصفراء ، كما تم قياس نسبة اليوريا والكرياتينين وحامض اليوريك ، كما تم قياس نسب بعض المعادن ومنها (الكالسيوم والفسفور والكلور والصوديوم والبوتاسيوم) ، وأيضا تم قياس نسب بعض العناصر الثقيلة والنادرة ومنها (النحاس والحديد والرصاص والكاميوم والسلينيوم) ، وقياس مستوي بعض مضادات الأكسدة ومنها (إنزيم فوق أكسيد الديسموتيز والكاتاليز والجلوتاثيون) ، كما تم قياس نسب بعض الهرمونات منها (هرمون الغدة الدرقية في مصل الدم وهرمون اللبتين في اللبن) ، وقياس معدل كلا من الجلوكوز والأنسولين في مصل الدم. وقد أوضحت الدراسة وجود فروق معنوية في كلا من (إنزيم ناقل أمين الالانين وإنزيم ناقل أمين الاسبرتات وأملاح اليوريا والكرياتينين والصوديوم والكالسيوم والفسفور والكلور والبوتاسيوم والنحاس والحديد وهرمون اللبتين وهرمون الغدة الدرقية وإنزيم فوق أكسيد الديسموتيز والكاتاليز والجلوتاثيون والجلوكوز والأنسولين) ، بينما لا توجد فروق معنوية في كلا من (إنزيم الكالين فوسفاتيز وأملاح الصفراء وحامض اليوريك والسلينيوم) في شهر الحليب الثلاثة الأولي عند مقارنتها بالمجموعة الضابطة والمأخوذة من منازل المربين .