Dept. of Animal Prod.,
Fac. of Agric. Al-Azhar University, Assiut Branch, Egypt.

A STUDY OF SOME BLOOD SERUM CONSTITUENTS DURING DIFFERENT REPRODUCTION STAGES IN FRIESIAN COWS.

(With 2 Tables)

By

S. FAHMY; H.M. BADR*; A.M. ASHOUR* and G.M. SOLOUMA**

* Dept. of Animal Prod., Fac.of Agric. Al-Azhar University, Nasr city, Cairo, Egypt.
** Dept. of Animal Prod., Fac.of Agric. South Valley University, Souhg, Egypt.

(Received at 23/6/2004)

دراسات على بعض مكونات مصل الدم خلال المراحل التناسل المختلفة في الأبقار الفريزيان

سالم فهمي محمد ، حسين محمد بدر ، عبد الله محمد عاشور
جمال محمود سلومه

أجريت هذه الدراسة في محطة البحوث التابعة لقسم الإنتاج الحيواني زراعة الأزهر بأسيوط. استخدم في هذه الدراسة عدد عشرة ابقار الفريزيان، ابتداء من الأسبوع التاسع قبل الولادة وعند الولادة واستمرت بعد الولادة حتى الأسبوع التاسع. واستهدفت الدراسة متابعة التغيرات في بعض مصل الدم والتي اشتملت على البروتين الكلى، الألبيومين، الجلوبيولين، والنسبه بين الألبيومين والجلوبيولين، الجلوكوز، الدهون الثلاثية والدهون الكلية. أظهرت النتائج انخفاض جميع المقاييس عند الولادة باستثناء النسبة بين الألبيومين والجلوبيولين وكان هذا الانخفاض غير معنوي في جميع القياسات باستثناء الجلوكوز كان معنويا. استمر انخفاض كلا من الجلوكو والدهون الثلاثية حتى الأسبوع الأول من الولادة ثم ارتفعا بعد ذلك حتى وصل أعلا مستوى للجلوكوز في الأسبوع السابع بعد الولادة بينما كان أعلا مستوى الدهون الثلاثية في الأسبوع التاسع سواء كان ذلك في الحيوانات المكره أو المتأخرة. على العكس ارتفع مستوى كلا من البروتين الكلى، الألبيومين، الجلوبيولين والدهون الكلية في الأسبوع الأول سواء كان ذلك في الحيوانات المبكرة أو المتأخرة ولكن الزيادة كانت معنوية في الحيوانات المبكرة. ثم بدء الانخفاض تدريجي حتى وصل إلى اقل مستوى في الأسبوع السابع ثم بدء في الارتفاع مره ثانيه

SUMMARY

Ten Friesian cows were used to study the serum changes of total protein, albumin, globulin, A/G ratio, glucose, triglycirids and total lipids in the periparturient period. During prepartum period serum total protein

showed gradual decrease from 7th prepartum week till parturition. Then it increased rapidly at one week after parturition, and then sharply declined up to 7th postpartum week in both early and later conceived cows. Albumin and globulin levels were higher at 9th week prepartum, and then steadily decreased till parturition. During postpartum period serum albumin and globulin concentrations were high at 1st week and then gradually decreased at 7th weeks in both early and late conceived. The A/G ratio increased progressively from 0.58 % to 0.78 % at 9th to 3rd week prepartum. In the postpartum period A/G ratio showed little changes among weeks. The concentration of serum glucose showed a slight increase between 9th and 7th week prepartum. it decreased from 7th weeks till parturition. The triglycerids and total lipid levels showed little changes before parturition. In the postpartum period, triglycerids and total lipid showed a special trend, where it increased gradually to reach maximum level in 9th week for both early and later conceived cows.

Key words: *Blood, serum, reproduction, friesian, cows.*

INTRODUCTION

Attempts have been made to assess the energy balance of lactating cows by estimating blood metabolites. A relationship between reduced blood glucose levels, excessive weight loss at the time of mating and decrease pregnancy rates was demonstrated. It was found that, blood glucose values less than 30 or 23 mg/dl were associated with reduced fertility (McClure 1968; and Arthur, *et al* 1989). Moreover, Plym Forshell, *et al* (1991) found that chronic lowering of plasma glucose concentration at four and seven weeks after calving, is cause of reduced fertility.

Rowlands *et al,* (1977) reported that, serum albumen concentration can be affected by the level of protein intake and its concentration is inversely related to the number of services per conception. Although serum albumen values normally decline in cows after calving eventually returning to precalving values at seven to nine weeks postpartum (Rowlands, 1980). The same author reported that, serum albumin is significantly decreased in cows affected with sever than that affected with mild fatty liver disease, when albumin estimated in the first eight weeks after calving. Since albumin is synthesized in the liver impaired liver function will influence its production. Whilst if fat

has replaced glycogen in the liver, parenchyma, total glycogen reserves will be reduced (Arthur, *et al* 1989).

Total plasma protein as a constituent of plasma serve as indicator of amino acids pool for protein synthesis in the liver (Harper, *et al.*, 1977). Total plasma protein tended to decrease with advancement of pregnancy and increased with advancing lactation in cows (Blum, *et al*, 1983; Vukovic, *et al*, 1990; Abdel-Samee and Ibrahim, 1992 and Hassinin, *et al* 1996) and in buffalo (Abdul-Quam et, 1990 and Badr, *et al* 2002). Reduction of globulin concentration in cows during late pregnancy is a consequence of reduced alpha and gama globulins concentration with the formation of colostrums, (Vukovic, *et al* 1991). In the same time, Gadhave, *et al* (2000) found that the decreased in serum globulin concentration toward calving may be due to selective uptake of immunoglobulin by the mammary gland.

Blum, *et al*, (1983) reported that plasma triglycirędes (TG) increased slightly in association with decreasing milk yield markedly during the last 2 months of pregnancy (dry period) in cows. Then, it rapidly falls immediately before the onset of the next lactation. The author also found that blood total lipids concentration was high at 60 day prepartum. It declined steadily during the next period to reach the lowest level at the day of parturition. Within 60 days postpartum total lipid increased gradually.

This study was designed to determine the sequential serum changes in levels of total protein, albumin, globulin, triglyceride, and total lipids during late pregnancy, at parturition and during post partum period in Friesian cows.

MATERIALS and METHODS

The present study was carried out on ten Friesian cows, aged 3-6 years with body weight of 325-400 kg. The animals were kept in the experimental station of animal production department of Faculty of Agriculture, Al-Azhar university, Assiut branch, Egypt. The animals were fed according to their body weights and physiological status. All animals were fed on Egyptian clover (Trifolium alxandrium) which was offered as 20-30 kg/head/daily from December to May. In summer season animals were fed on green forage (Daraw). In addition, wheat straw and concentrate mixture consisted of yellow corn, wheat bran, molasses, cotton-seed meal, stone and salt were added. The animals were

allowed to drink fresh water along the day. The animals were kept in open free-stalls.

Two months after barturition, a fertile bull was introduced to cows for natural mating. Two months, the animals were examined ultrasonographically for pregnancy diagnosis. According to the results of diagnosis, pregnant cows were classified into: 1- Animals conceived early (< 90 days postpartum). 2- Animals conceived later (> 90 days postpartum).

Blood samples were collected from jugular vein using 10ml glass tubes at 9, 7, 5 and 3 weeks prepartum, at day of parturition, and at 1, 3, 5, 7 and 9 weeks postpartum. Samples were centrifuge at 4000 rpm for 15 min and serum stored at -20 °C until analyses. Total protein, albumin, glucose, triglysrid and total lipids levels were estimated biochemically using commerical Kits provided by Sentinel (CH) and Diamond as described by Gowan, *et al.* 1983 and Bergmeyer, (1974). Globulin was calculated by subtraction of albumin from total protein. The A/G ratio was calculated by dividing albumin by its corresponding globulin value.

Statistical analysis was carried out according to SAS (1988) users Guide, tested using the Duncan's Mltiple Range Test as described, Walpole, (1974).

RESULTS

Serum total protein levels, during prepartum, at parturition and postpartum period are illustrated in table (1). During prepartum period serum total protein showed gradual decrease from 7th week prepartum till parturition. Its concentration increased rapidly at one week after parturition, and then sharply decline up to 7th week postpartum in both early and later conceived cows. The differences among weekly levels during prepartum and at parturition were not significant. Also the difference between early and later conceived cows was not significant. While, in postpartum period, the difference between 1st and 7th weeks, was significant ($P<0.05$) in both early and later conceived cows.

Serum concentration of albumin during prepartum, at parturition and postpartum periods are reported in (table 1). Albumin level was high at 9th week prepartum, and then steadily decreased till parturition. During postpartum period serum albumin concentration was high at 1st week and then gradually decreased at 7th weeks in both early and late conceived. The results of the present study showed no significant differences in albumin levels during the experiment.

Changes in serum globulin concentration during prepartum, at parturition and postpartum period are shown also in table (1). Globulin level in prepartum showed gradual decreased from 9th weeks prepartum till parturition. Then the level sharply increased to 5.71 and 4.86 g/l one week after birth in early and later conceived cows, respectively. The differences among 9th to 3rd weeks prepartum were non significant, while the difference between 9th week and parturition was significant ($P<0.05$). On the other hand in postpartum period the differences among weeks from 1st to 9th weeks were non significant in both early and later conceived, excepted, the differences between 1st and 7th weeks was significant. The difference level in globulin between early and later conceived was also non significant.

Table (1) also showed the changes in the A/G ratio of the Friesian cows during prepartum at parturition and postpartum period. The A/G ratio increased progressively from 0.58 % to 0.78 % at 9th to 3rd week prepartum. In the postpartum A/G ratio showed little changes among weeks. The maximum A/G ratios (0.91 ± 0.3 and 0.93 ± 0.22) were recorded at 9th and 5th week in both early and later conceived respectively. While the minimum A/G ratios (0.63 ± 0.14 and 0.67 ± 0.26) were recorded at 3rd and 9th in both early and later conceived cows respectively. Non significant differences were found in A/G ratio throughout the experimental period between the early and later conceived animals.

Serum concentration of glucose during prepartum, at parturition and postpartum are illustrated in (table 2). The concentration of serum glucose showed a slight increase between 9th and 7th week prepartum, it decreased from 7th week prepartum till parturition. The differences among 9, 7 and 5 weeks in prepartum were significant ($P<0.01$). Glucose levels during postpartum period gradually increased from 1st to 7th weeks in both early and later conceived cows. Early conceived cows had higher plasma glucose than later conceived, but the differences was not significant. On the other hand, the differences among 1st and 3rd to 9th weeks during postpartum period were significant in early and later conceived cows ($P<0.01$).

The changes in the triglycerides concentration during prepartum, at parturition and postpartum period are presented in table (2). Serum triglyceride decreased significantly from 57.37 ± 5.14 mg/dl before birth to 46.3 ± 5.2 mg/dl, on the day of parturition ($P<0.01$). It also sharply drop to 26.6 ± 3.19 and 29.6 ± 2.11 mg/dl at1st week after birth ($P<0.01$) in the early and later conceived cows respectively. The triglycerides

levels increased gradually to reach maximum at 9th weeks postpartum in both early and later conceived cows.

Serum concentration of total lipid during prepartum, parturition and postpartum period are presented in table (2). The total lipid levels showed little changes before parturition. during prepartum period the maximum total lipid concentration (3.72 ± 0.14 mg/dl) was recorded in 9th week, while the minimum concentration (2.34 ± 0.22 mg/dl) was recorded at parturition. In the postpartum period, total lipid take a special trend, where it increased gradually to reach maximum level in 9th week for both early and later conceived cows. No significant differences were found in total lipids throughout the experimental period.

DISCUSSION

The periparturient changes in total protein are in agreement with the results of (Rowlands, *et al*, 1980; Hassan, *et al* 1986, Vukovic, *et al* 1991; Rajora and Pachauri, 1994 and Badr, *et al*, 2002). The decreased in serum total protein in cows at parturition may be due to the decreased in albumin. This results are in agreement with that recorded by Rajorra and Pachauri (1994) who reported that, the reduction in serum total protein in late pregnancy was attributed to the decreased in both serum albumin and globulin. The drop in serum protein in cattle at parturition was caused by a loss of immune β_2 and δ_1 globulin in blood. This coincided with the colostrums formation time in the mammary gland (Singh *et al*, 1999). Moreover, El-Naggar and Abdel-Raouf (1971) found that the decreased in serum total protein in late gestation coincided with the rapid increase in the uterine weight and its contents, namely, the foetal fluids and the foetal membrane.

Total protein increased at the 1st week after birth in both early and later conceived cows, this increase may be due the increase in both albumin and globulin. This results in is agreement with Oldham *et al*, (1979) and Blum, *et al* (1983) who found that total protein was lower around parturition and increased at the onset of lactation. Because albumin levels did not change, the transient fall must have been due to the globulin fraction which is taken up by the udder during colostrums formation. Total protein had correlation with milk yield. Moreover, there were significant high levels of total protein in high as compared to low-yielding animals.

The periparturient changes in albumin concentration are in agreement with those reported by (Blum, et al, 1983 and Badr *et al*

2002). The gradual decreases during postpartum period may be due to the quick protein synthesis by the foetus. This result are in agreement with Jaindeen and Hafz, (1980) who reported that, the decrease in albumin with the advance of gestation may be due to the acceleration in protein synthesis by the foetus, this cause reduction in amino acids available for liver for albumin synthesis.

Rowlands, *et al* (1980) found that cows, which were better able to maintain stable albumin concentration, were likely to have better fertility. Atallah and Abd-Alla (1998) reported that the albumin was significant related to the rate of pregnancy and correlation between albumin concentration and fertility.

The reduction of the concentration of globulin in cows during late pregnancy is a consequence of the reduced concentration of alpha and gama globulins, which is in connection with the formation of colostrums, (Vukovic, *et al* 1991 and Rowlands et al, (1975). Gadhave, *et al* (2000) found that the decreased in serum globulin concentration toward calving may be due to selective uptake of immunoglobulin by the mammary gland.

The differences among weeks in A/G ratio may be attributed to the differences in albumin and globulin concentration. These results are in agreement with those reported by Abd-El-Bary, (1990) who reported that the decrease in A/G ratio during early pregnancy was attributed to the high globulin during period.

Periparturient changes in glucose concentration are in agreement with the results reported by (Bickerstaffe, *et al,* 1974; Rowlands, *et al,* 1980 and Rajora and Pachauri, 1994). The blood glucose levels showed decreasing trend with advancement of pregnancy. This may be attributed to the energy demands of the growing fetus. Glucose level in the present study was decreased along 60 days prepartum till parturition, which may be referred to increase of fetus size, which caused increased respiration in the dam (Singh, *et al,* 1999). The decreased of glucose levels during the first weeks after parturition may be interpreted as mainly the consequence of the high demand for the substance, primarily for lactose synthesis, (Bickerstaffe, *et al,* 1974 and Blum, *et al,* (1983).

Triglyceride was higher around prepartum and decreased in postpartum period as also found by Blum *et al* (1983) and Schwalm and Schultz (1976). Triglyceride consider an important source of long-chain fatty acids for milk fat synthesis (Bickerstaffe, *et al* 1974). It also, concentrated in the very low-density lipoprotein fraction, which is higher in dry than in lactating cows (Palmquist, 1976). At the same time,

triglyceride is taken up by the mammary gland, which could explain the negative relationship between milk yield and plasma triglyceride concentration in this study.

Periparturient changes in total lipids concentration is in agreement with the results of Badr, *et al* (2002) and Sahukar *et al*, (1985). The gradual decrease in serum total lipids during postpartum and at parturition. It increased gradually from 1st to 9th week in both early and later conceived cows during postpartum period. This result is in agreement with that of Schmidt (1971) who reported that, the decline in serum total lipids shortly before parturition and at the postpartum period may attributed to the increased demands of mammary glands for fatty acids for the synthesis of triglycired since 50% butter fat in the cows are received from blood lipids.

The increased of total lipids in early conceived than later conceived cows may be associated with development of the corpus luteum. Bard, *et al* (2002); and Thorpe, *et al* (1964).

Table 1: Changes in serum concentration of total protein, albumin, globulin and A/G ratio during prepartum, at parturition and during postpartum period in Friesian cows.

Weeks	Total. Protein g/l		Albumin g/l		Globulin g/l		A/G ratio	
-9	90.23 ± 0.72ab		30.7± 0.31 ^{a}		60.35± 0.88^{a}		0.58 ±0.12 ^{a}	
-7	90.82 ± 0.77^{ab}		30.61± 0.15^{a}		50.61±0.86^{ab}		0.63 ±0.12^{a}	
-5	80.28 ± 0.83^{ab}		30.47± 0.28 ^{a}		40.66± 0.78^{ab}		0.74± 0.4^{a}	
-3	70.71± 0.93 ^{bc}		30.34± 0.18 ^{a}		40.42 ±1.1^{ab}		0.78± 0.38^{a}	
Parturition	60.94 ± 0.61^{bc}		30.0 ± 0.21 ^{a}		40.13 ±0.65^{b}		0.71±0.23^{a}	
Postpartum	Early. Conceived	Later conceived	Early. conceived	Later conceived	Early. Conceived	Later conceived	Early. conceived	Later conceive
1	100.28 ±1.2 ^{a}	80.8 ±1.3^{ab}	40.32 ±0.3^{a}	40.11 ±0.38^{a}	50.31 ±0.67^{ab}	40.86 ±0.57^{ab}	0.76 ±0.36^{a}	0.82 ±0.2^{a}
3	80.56 ±1.3^{ab}	80.54 ±1.1^{ab}	30.3 ±0.49^{a}	30.88 ±0.49^{a}	50.26 ±0.72^{ab}	40.82 ±0.45 ^{ab}	0.63 ±0.14^{a}	0.80 ±0.2^{a}
5	80.42 ±0.72^{ab}	70.75 ±0.74 ^{b}	30.82 ±0.56^{a}	30.49 ±0.47^{a}	40.6 ±0.45^{abc}	30.75 ±0.82^{bc}	0.83 ±0.06^{a}	0.93 ±0.22^{a}
7	60.66 ± 1.1^{bc}	50.1 ±0.71^{c}	20.84 ±0.3 ^{a}	20.34 ±0.1 ^{a}	30.82 ±0.67^{bc}	20.76 ±0.66^{c}	0.75 ±0.18^{a}	0.85 ±0.26^{a}
9	80.12 ±0.12^{ab}	60.21 ±0.73^{bc}	30.82 ±0.21^{a}	20.42 ±0.18 ^{a}	40.11 ±0.42^{bc}	30.71 ±0.32^{bc}	0.91 ±0.3^{a}	0.67 ±0.26^{a}

a, b, c,d and e : Values in the same rows and clons with different superscripte are different (P<0.05).

Table 2: Changes in serum concentration of glucose, triglyceride and total lipids, during prepartum, at parturition and postpartum period in Friesian cows.

Weeks	Glucose mmol/l		Triglycerid. mmol/l		T. Lipids mmol/dl	
-9	71.18±3.16^{a}		57.37±5.14^{a}		3.72±0.14^{a}	
-7	72.09±4.31^{a}		48.81±1.1^{a}		3.11±0.13^{a}	
-5	63.27±4.72^{b}		49.11±1.3^{a}		3.12±0.13^{a}	
-3	56.03±7.62^{c}		47.15±2.1^{a}		2.87±0.05^{a}	
Parturitio	54.4±4.38^{c}		46.32±5.2^{a}		2.34±0.22^{a}	
Postpartu	Early. conceived	Later conceived	Early. Conceived	Later conceived	Early. conceived	Later conceived
1	49.61 ±9.25^{cd}	44.26 ±3.2^{d}	26.6 ±3.27^{a}	29.6 ±2.11^{a}	3.33 ±0.3^{a}	3.28 ±0.42^{a}
3	66.16 ±5.46^{ab}	63.9 ±3.21^{b}	32.8 ±3.19^{a}	32.6 ±3.18^{a}	3.91 ±0.4^{a}	4.13 ±0.32^{a}
5	70.41 ±10.5^{a}	69.54 ±7.03^{a}	35.4 ±3.38^{a}	33.21 ±2.32^{a}	4.25 ±0.43^{a}	4.18 ±0.18^{a}
7	72.26 ±2.59^{a}	71.58 ± 2.9^{a}	35.52 ±3.58^{a}	30.5 ±2.56^{a}	4.88 ±0.25^{a}	4.28 ±0.27^{a}
9	69.52 ±5.59^{a}	60.81 ±6.8^{b}	41.12 ±2.12^{a}	38.08 ±3.3^{a}	4.93 ±0.69^{a}	4.34 ±0.32^{a}

a, b, c,d and e: Values in the same rows and clomns with different superscripte are different (P<0.01).

REFERENCES

Abdel-Samee, A.M. and H. Ibrahim (1992): Triiodothyronine and blood metabolites relation to milk yield and spray cooling of heat stressed lactating Friesian and Holsteins in Egypt. J. Anim. Prod. 29, No 2, PP. 215-227.

Abdul Quayam, S.; T.G. Devanathan and S.R. Pattabiraman (1990): Serum total protein and blood glucose levels during prepartum and postpartum Murrah buffaloes. Indian J.Anim. Sci. 60: 140-142.

Arthur, G.H.; Noakes, D.E. and Pearson, H. (1989): Veterinary Reproduction and Obstetrics 6th Ed. Parturition and the care of Parturient Animals. PP. 132-160.

Atallah, S.A. and O.A. Abd-Alla (1998): Relationship between fertility and some blood metabolites in buffaoes. Assiut. Vet. Med. J.: 39: 175-183.

Badr, H.M.; Ashour, A.M. and G.M. solouma (2002): Changes in some blood constituents and their relation to reproductive performance in pre and postpartum periods in Egyptian Buffaloes. J. Agric. Sci. Mansoura Univ. 27 (5): 2909-2923.

Bickerstaffe, R.; Annison, E.F. and J.I. Linzell (1974): The metabolism of glucose, acetate, lipids and amino acids in lactating dairy cows. J.Agric. Sci.., Comb. 28:71-85.

Blum, J.W.; Kunz, P. and Leuenberge H. (1983): Thyriod hormones, blood plasma metabolires and haematological parameters in relationship to milk hield in dairy cows. Anim. Prod.; 36: 93-104.

El-Naggar, M.A. and Abdel Raouf, M. (1971): The foetal membranes and fluids in the Egyptian buffaloes. Assiut Vet. J., 20: 149-155.

Harper, U.A.; Roddwell, V.W. and Mayes, P.A. (1977): Review of physiological chemistry 16th Longe Meedicaal publications California, USA, PP. 76-79.

Gadhave. L.R.; Mantri, A.M.; Talvelkar, B.A. and Deshmuka, B.T. (2000): Serum protein profile during gestation and early postpartum period in Girsay and crossbred cows. Indian Vet. J., 77: 114- 116.

Hassan, G.A.; El-Nouty, F.D.; Samak, M.A. and Salem, M.H. (1986): Relationship between milk production and some blood constituents in Egyptian Baladi goats. J. Tropical Agric. Vet. Med., 21:213-219.

Hassanin, S.H.; Abdalla, E.B.; Thaarwat, A.A.; El-Sharbiny and Kotby, E.A. (1996): Effect of kidding season on some blood constituents, milk yield and milk composition of Egyptian Zaraibi goats. Menofiya J. Agric Res.; 21: 329-341.

Jainudeen, M.R. and Ehafez, E.S. (1980): Gestation, perinatal physiology and parturition. In: Reproduction in Farm Animals. Fourth Ed., E.S.E. Hafez (Edt). Lea and Febiger, philadelphia.

Oldham, J.D., Broster, W.H., Napper, D.J. and Sivitter, J.W. (1979): The effect of low protein ration on milk yield and plasma metabolites in Freasian heifers during early lactation. Br. J. Nutr. 42: 149-162.

McClure, T.J. (1968): Hypoglycaemia, an an apparent cause ofinfertilit of lactating cows. Brit. Vet. J. 124, 126-130.

Gowan, M.C.; Artiss, M.W.; Strandbergh, J.D. and Zak, D.R. (1983): B-Clin. Chem. 29, 538.

Palmquist, D.L. (1976): A kinetic concept of lipid transport in ruminants. A review. J. Dairy Sci. 59: 355-363.

Plym Forshell, K.; Andersson, L. and Behrson, B. (1991): The relationships between the fertility of dairy cows and clinical and biochemical measurements, with special referance to plasma glucose and milk acetone. J. vet. Med. A38, 608-616.

Rajora, V.S. and Pachauri, S.P. (1994): Blood profile evaluation in crossbred cows under different stages of lactation. Indian J. Anim. Sci..; 64: 1351-1353.

Rowlands, G.J. (1980): A review of variattions in the concentration of metabolites in the blood of beef and dairy cattle associated with physiology, nutrition and disease, with particular reference to the interpretaation of metabolic profiles. Wld. Rev. Nutr. Diet. 35, 172-235.

Rowlands, G.J.; Little, W. and Kitchenham, B.A. (1977): relationships between blood composition and fertility in dairy cows a field study. J. Dairy Res. 44: 1-7.

Rowlands, G.J.; Manston, R. and Sally, M.D. (1975): relationships between stages of lactation and pregnancy and blood composition in a herd of dairy cows and the influence off seasonal changes in management on these relationship. J. Dairy Res., 42: 349-362.

Sahukar, C.S.; Pandit, R.K.; Chuhan, R.A.S. and Powar, M.L. (1985): Cho;esterol and alkaline phosphatase in crosbred cows. Ind. J. Anim Sci., 55: 421-423.

SAS. (1988): SAS Users Guide : Statistics. SAS Inst ., Inc., Cary, NC. Schams, D.

Schmidt, G.H. (1971): Biology of lactation 1st Ed. Freman and Co. San Fransisco.

Schwalm, J.W. and Schultz, L.H. (1976): Relashinship of insulin concentration to blood metabolites in dairy cow. J. Dairy Sci. 59: 255-261.

Singh, M.J.M.; Nigam, M.; Singh and Sharma, K.B. (1999): Blood plasma biochemical profile of pregnant and non-pregnant Yaks in comparison with reproductive values in cattle. Indian Vet. J. 76: 568-570.

Thorpe, W.V.; Bray, H.G. and James, S.P. (1964) Biochemistry for Medcal students. 8th Ed. J. and Churchill LTD. London.

Vukovic, D.; Samanc, H.; Damnjanovic, Z. and Petrujkic, T. (1990): Study of the influence of metabolic disorder in high-milk cows on their reproductive abilites. Proceeding of the 12th Mt. Cong. Drim. Rep., the Netherlands, PP. 144.

Walpole, R.E. (1974): Introduction Statistics. 2nd Ed. Macmillar Publishing Co. Inc. New York, Collier Macmillan Pub. Landon PP. 277-278.