



# HAEMATO-BIOCHEMICAL PROFILES OF ANOESTROUS CROSSBRED HEIFERS AND COWS IN CALICUT DISTRICT, KERALA

C.P. Abdul Azeez<sup>1</sup>, J. Metilda<sup>2</sup>,  
K.N. Aravinda Ghosh<sup>3</sup>,  
N.P. Usha<sup>4</sup> and K. Ally<sup>5</sup>

Department of Animal Reproduction, Gynaecology and Obstetrics, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala-680 651.

Received- 14.08.2014  
Accepted- 06.10.2014

## Abstract

*Haemato-biochemical profiles of anoestrus heifers and cows, having a body condition score of 2.5 to 3.5, selected through field level infertility camps were estimated. The blood haemoglobin, glucose and serum protein, calcium, phosphorous, copper, cobalt, zinc and manganese levels of anoestrus heifers and cows were estimated. The values did not exhibit statistically significant difference ( $p \geq 0.05$ ) between heifers and cows and were within the normal range. Present study revealed that blood biochemical parameters of anoestrus heifers and cows with good body condition score (2.5 to 3.5) were within normal limits.*

**Key words:** Crossbred heifers and cows, body condition score, anoestrus, biochemical parameters

Anoestrus is the most common cause for infertility in cattle which is mostly manifested by delayed puberty in heifers, prolonged service period and inter calving interval in cows causing low milk and calf production resulting in great economic loss to the dairy farmers.

Minerals play an intermediate role in the action of hormones and enzymes at

cellular level in an integrated fashion. Besides working as a cofactor or activator of enzyme systems, the elements like Ca have been found to sensitize for the action of hormones. Deficiency or excess of mineral elements like P, Cu and Zn was associated with subnormal fertility and anoestrus conditions (Moddie, 1965). Dietary mineral elements were known to affect the physiological function in general and reproduction in particular (Hidiroglou, 1979).

A number of predisposing causes are involved in the etiology of anoestrus, including nutritional, managerial, environmental and hormonal, which may vary from herd to herd and animal to animal. Variations in haemato-biochemical profiles were found to be commonly associated with anoestrus in dairy cattle. Exact and early diagnosis of the etiology followed by timely intervention is a prerequisite to curtail this malady. Body condition score (BCS) is the measures of nutritional status of animals and is an important factor influencing the reproductive performance (Baruselli *et al.*, 2001). Extremes of BCS (very low and very high) at pre-calving, calving and early postpartum period delay onset of cyclicity in cattle (Butler and Smith, 1989). For optimum reproductive performance, a BCS of 2.5 (on five point scale) was required at breeding in cattle (Edmonson *et al.*, 1989).

1. Assistant Professor, Dept. of Animal Reproduction, Gynaecology and Obstetrics, CVAS, Pookode
2. Associate Professor
3. Professor and Head (Retd.)
4. Professor & Head, Dept. of Clinical Medicine, Ethics and Jurisprudence
5. Professor, Dept. of Animal Nutrition

The objective of the present study was to assess the haemato-biochemical profile of anoestrous crossbred heifers and cows with a body condition score of 2.5 to 3.5.

### Materials and Methods

Crossbred anoestrous heifers between 18 to 36 months of age and postpartum anoestrous cows between 3 to 8 years (first to fourth parity) of age having a body condition score of 2.5 to 3.5 formed the material for the present study. The animals were selected through infertility camps conducted at selected locations in Calicut district of Kerala State during the period from October 2012 to February 2014. Confirmation of anoestrous stage was done by repeated examination of reproductive tract and ovary at an interval of 10 days. Eighty apparently healthy animals comprising of 40 heifers and 40 cows were selected and their blood was collected to estimate haemato-biochemical profiles. Blood sample (20 ml) from jugular vein was collected aseptically from crossbred heifers and cows. Five ml was transferred to citrated vial for estimation of haemoglobin (Hb), 5 ml was transferred to sodium fluoride vial for estimation of glucose and 10 ml was allowed to clot for serum separation. The sera thus separated were dispensed into 2 ml aliquots, labelled and stored in serum vials at  $-20^{\circ}\text{C}$  until analysed. Later serum levels of total protein, albumin, globulin, Ca, P, Cu, Co, Zn and Mn were estimated. Blood haemoglobin level was estimated by cyan-methaemoglobin method, blood glucose level was estimated by glucose oxidase/peroxidase (GOD/POD) method, total serum protein, albumin and globulin levels were estimated by Biuret method (Dumas, 1975) using standard kits procured from Agappe diagnostics, using fully automatic haematology analyser (Mindray, BC 2800 VET). Serum calcium level was estimated by Arsenazol-III method (Bagainski, 1973), serum inorganic phosphorus level was estimated by Molybdate

U.V. method (Fiske and Subbarow, 1925) using standard kits procured from Agappe diagnostics using Chemistry Analyzer (Mindray, BS 120). For mineral estimation, serum samples (1 ml each) were wet digested with 5 ml volume of di-acid mixture (Perchloric acid: Nitric acid, 1:4) on a hot plate according to the method adopted by Krishna and Ranjhan (1980). The clear transparent residues were diluted in triple glass distilled water and final volume was made to 50 ml, which was then used for the estimation of trace elements, viz., zinc, copper, cobalt and manganese on an Atomic Absorption Spectrophotometer (Model-3110, Perkin Elmer) as described by Oser (1979). The data generated were analyzed statistically using Analysis of variance (ANOVA).

### Results and Discussion

The blood haemoglobin, glucose and serum total protein, albumin and globulin values of anoestrous heifers and cows are presented in the Table 1. The values did not exhibit statistically significant difference ( $p \geq 0.05$ ) between heifers and cows and were within the normal range and were comparable with values for normal healthy cattle.

Pillai (1980) reported a haemoglobin level of 9.16 and 9.70g per cent in anoestrous cows and heifers respectively. Sharma *et al* (1983) observed that the haemoglobin value of anoestrous ( $9.05 \pm 2.05$ ) and repeat breeder ( $10 \pm 2.5$ ) were lower than that of normally cycling ( $11.1 \pm 1.90$ ) cows. However, Srivastava and Sahni (2000) reported there was no significant difference in haemoglobin per cent between normal cycling and anoestrous heifers. Satyaraj (2007) observed Hb value in crossbred heifers at puberty as  $9.83 \pm 0.25$  under improved management and feeding conditions and  $8.01 \pm 0.17$  under ordinary rural management conditions. Kumar *et al.* (2009) reported Hb value in normal cyclic and repeat breeding crossbred cows as  $10.16 \pm 0.67$  and

**Table 1.** Blood haemoglobin, glucose and serum protein profile (mean  $\pm$  SE) in anoestrous heifers and cows (n=40)

Group	Haemoglobin (g%)	Glucose(mg%)	Total protein (g%)	Albumin(g%)	Globulin(g%)
Heifers	10.01 $\pm$ 0.20	52.02 $\pm$ 0.97	6.73 $\pm$ 0.13	3.24 $\pm$ 0.08	3.27 $\pm$ 0.16
Cows	9.92 $\pm$ 0.21	51.17 $\pm$ 1.16	6.87 $\pm$ 0.14	3.53 $\pm$ 0.09	3.40 $\pm$ 0.16

**Table 2.** Serum mineral profile (mean  $\pm$  SE) in anoestrous heifers and cows (n=40)

Groups	Calcium (mg%)	Phosphorus (mg%)	Copper (ppm)	Cobalt (ppm)	Zinc (ppm)	Manganese (ppm)
Heifers	9.71 $\pm$ 0.23	5.37 $\pm$ 0.18	0.56 $\pm$ 0.03	0.20 $\pm$ 0.02	0.48 $\pm$ 0.03	0.07 $\pm$ 0.01
Cows	10.07 $\pm$ 0.24	5.80 $\pm$ 0.46	0.56 $\pm$ 0.03	0.20 $\pm$ 0.01	0.51 $\pm$ 0.03	0.07 $\pm$ 0.01

9.23  $\pm$  0.25g per cent respectively. However, Khan *et al.* (1995) reported that the value of Hb do not show any significant change between the regular breeding, repeat breeding and anoestrous cows and these values were within normal physiological range.

Diskin *et al.* (2003) stated that glucose availability influenced both tonic and surge modes of luetinising hormone (LH) secretion through its effects on gonadotropin releasing hormone (GnRH). Yadav *et al.* (2004) observed average blood glucose values of 49.44 and 62.31 g per cent in anoestrus and normal cyclic animals respectively. A similar finding was also reported by Dutta *et al.* (1998). The results obtained in the present study lies within the normal range which might be due to the selection of all the animals based on the body condition score.

Tegegne *et al.* (1993) did not observe consistent trend in plasma protein levels during first 33 weeks postpartum and the levels were neither influenced by feeding or suckling regimen, nor by cyclic and acyclic nature of cows. Patel and Dhami (2006) also failed to see any significant difference in total protein concentrations of anestrous and subestrous HF cows. Vohra *et al.* (1995) and Joe *et al.* (1998) reported significantly higher serum total protein levels in normal cyclic than in anoestrus cows (8.62  $\pm$  0.13 vs 6.82  $\pm$  0.40 g/dl) and (7.45  $\pm$  0.39 vs 4.80  $\pm$  0.53 g%) respectively. Peter *et al.* (2009) opined that diets high in crude protein, in excess of 16 per cent support high milk yield but may be detrimental to reproductive performance due to elevated blood urea concentration.

The serum calcium, phosphorous, copper, cobalt, zinc and manganese in anoestrus heifers and cows are summarised in Table 2. Analysis of data revealed no statistically significant difference between heifers ad cows ( $p \geq 0.05$ ).

Dutta *et al.* (2001) observed that the circulatory levels of serum calcium and inorganic phosphorus were significantly higher ( $P < 0.01$ ) in normal cyclic (10.73  $\pm$  0.08, 4.22  $\pm$  0.07 mg%) than in anoestrus (9.54  $\pm$  0.22, 3.48  $\pm$  0.12 mg%) or repeat breeding nondescript cows of Assam (9.95  $\pm$  0.18, 3.62  $\pm$  0.14 mg%). Similar results were observed by Yadav *et al.* (2004) and Mishra *et al.* (2007). Whereas, Patel and Dhami (2005) and Raj *et al.* (2006) recorded no significant difference in the mean values of calcium and phosphorus between conceived and non conceived crossbred cows. Trace element deficiencies (notably Mg, P, Cu, Co and Mn) are commonly associated with anoestrus; sometimes these are causal; others may be reflective of, or additive with an inadequate supply of energy (Mc Clure, 1994).

From the present study it could be concluded that the blood biochemical parameters of anoestrus heifers and cows with good body condition score (2.5 to 3.5) was within the normal limits and such animals can be selected for hormonal induction of oestrus.

## References

- Bagainski, E.S. 1973. Product profile of diagnostic kit for total protein from Crest Biosystems Pvt. Ltd. *Clin. Chem. Acta.* **46**: 46.
- Baruselli, P.S., Barnabe, V.H., Barnabe, R.C., Visintin, J.A., Molero-Filho, J.R. and Porto, R. 2001. Effect of body condition score at calving on postpartum reproductive performance in buffalo. *Buffalo J.* **17**: 53-65.
- Butler, W.R. and Smith, R.D. 1989. Interrelationships between energy balance and postpartum reproductive function in dairy cattle. *J. Dairy Sci.* **72**: 767-772
- Diskin, M.G., Mackey, D.R. and Sreenan, J.M. 2003. Effects of nutrition and metabolic status on circulating hormones and ovarian follicle development in cattle. *Anim. Reprod. Sci.* **78**: 345-370.

- Doumas, B.T. 1975. Product profile of diagnostic kit for total protein from Crest Biosystems, Pvt, Ltd. *Clin. Chem.* **21**: 1159.
- Dutta, A., Baruah, B., Sarmah, B.C., Baruah, K.K. and Goswami, R.N. 2001. Macro mineral levels in cyclic, postpartum anoestrus and repeat breeding local cows in lower Brahmaputra Valley of Assam. *Indian J. Anim. Reprod.* **22**: 41-44.
- Dutta, A., Baruah, B., Sarmah, B.C., Baruah, K.K., Sarma, D.N. and Dutta, J.C. 1998. Micro mineral profiles during cyclic, postpartum anoestrus and repeat breeding cows in lower Brahmaputra Valley of Assam. *Indian J. Anim. Sci.* **70**: 712-713.
- Edmonson, A.J., Lean, I.J., Weaver, L.D., Farver, T., and Webster, G. 1989. A body conditioning chart for Holstein dairy cows. *J. Dairy Sci.* **72**: 68-78.
- Fiske, C.H. and Subbarow, Y. 1925. The colorimetric determination of phosphorus. *J. Biol. Chem.* **66**: 375.
- Hidiroglou, M. 1979. Trace element deficiency and fertility in ruminants. A review. *J. Dairy Sci.*, **62**: 1195-1206.
- Joe, A.J., Kathiresan, D., Devanathan, T.G., Rajasundaram, R.C. and Rajasekavan, J. 1998. Blood biochemical profile in normal cyclical and anoestrus cows. *Indian J. Anim. Sci.* **68**: 1154-1156.
- Khan, L.C., Ingraham, R.H., Morgan, E.B., Zeringue, L., Wilson, D. and Babcock, D.K. 1995. Relationship between fertility and haemoglobin, glucose and cholesterol concentrations in Holstein cows. *Am. J. Vet. Res.* **45**: 2607-2612.
- Krishna, O.P. and Ranjhan, S.K. 1980. *Laboratory Manual for Nutrition Research*. Vikash Publishing House Pvt. Ltd., New Delhi, India, 83-84.
- Kumar, P.R., Sanjay, K., Suresh, D., Chethan, S., Bijay, K., Satya, N., Harendra, K., Sudhir Kumar, R., Butani, M.J. and Dharni, A.J. 2009. Progesterone, metabolites and minerals in anoestrus, suboestrus, repeat breeding and cyclic cows. *Indian J. Anim. Reprod.* **30**: 19-22.
- Mc Clure, T.J. 1994. Nutritional and metabolic infertility in the cow, CAB international, Wallingford, Oxon.
- Mishra, S., Vashishta, N.K. and Singh, M. 2007. Incidence of infertility in the cows of Kangra valley of Himachal Pradesh. *Proc. XXII Annual Convention and National Symposium on "Challenges in Improving Reproductive Efficiency of Farm and Pet Animals"*, 7<sup>th</sup>-9<sup>th</sup> December, 2007, Bhubaneswar. P. 156.
- Moddie, E.W. 1965. Hypocalcaemia and hypomagnesaemia. *Br. Vet. J.* **121**: 338-342.
- Oser, B.L. 1979. *Hawk's Physiological Chemistry*. (14<sup>th</sup> Ed.) McGraw Hill Books Co., Bombay, India.
- Pillai, G.P.V. 1980. Studies on anoestrus in crossbred cattle. *M.V.Sc. Theses submitted to Kerala Agricultural University*. pp 35.
- Raj, S., Tiwari, R.P., Tiwari, S.P. and Poyam, M.R. 2006. Plasma mineral profile of anoestrus and normal cyclic Sahiwal heifers. In *proc: National symposium on "Recent trend and future strategies for improved reproduction of livestock companion and wild animals" and XXII Annual convention of ISSAR*, Mhow, (MP), 10-12 November, p 112.
- Peter, A. T., Vos, P. L. A. and Ambrose, D. J. 2009. Postpartum anoestrus in dairy cattle. *Theriogenology* **71**: 1333-1342.
- Patel, P.M. and Dharni, A.J. 2006. Postpartum plasma profile of glucose and total protein in Holstein Friesian cows with and without hormone therapy under tropical climate. *Indian J. Anim. Sci.*, **76**: 118-123.
- Satyraj, N. 2007. Reproductive performance of crossbred heifers under special livestock breeding programme of Kerala. *M.V.Sc. thesis*, Kerala Agricultural University, Mannuthy. pp.28-29.
- Srivastava, S.K. and Sahni, K.L. 2000. Blood mineral level affecting pregnancy rates in cows and buffaloes. *Indian J. Anim. Sci.*, **70**: 33-34.
- Tegegne, A., Entwistle, K.W. and Mukasa-Mugerwa, E. 1993. Plasma progesterone and blood metabolite profile in postpartum small East African Zebu cows. *Trop. Anim. Health Prod.*, **25**: 101-110.
- Vohra, S.C., Dindorkar, C.V. and Kaikini, A.S. 1995. Studies on blood serum levels of certain biochemical constituents in normal cycling and anoestrus crossbred cows. *Indian J. Anim. Reprod.*, **16**: 85-87.
- Yadav, Y.P., Singh, A.P., Kunj, V., Akhtar, M.H., Roy, G.P. and Singh, C. 2004. Study on incidence of anoestrus and blood biochemical constituents in non-cyclic and cyclic crossbred cows. *Indian J. Anim. Reprod.*, **25**: 116-119. ■