

## EFFECT OF DIFFERENT THERAPIES IN EARLY POST PARTUM PERIOD ON FERTILITY OF HF COWS

G.K. Mishra<sup>1</sup>, H.J. Derashri, N. P. Sarvaiya and F. S. Kavani

Department of Animal Reproduction, Gynaecology and Obstetrics  
Gujarat College of Veterinary Science and Animal Husbandry  
Anand Agricultural University, Anand 388 001, India

### ABSTRACT

This study was conducted on 24 normally calved HF cows of university farm divided into four groups; each of six animals. Group I served as control- only AI; whereas Group II was treated with Oxytetracycline (2 g Uroblex oblet) intrauterine on 2<sup>nd</sup> day postpartum after expulsion of placenta + GnRH (Inj. Receptal) 50<sup>th</sup> day postpartum + AI; Group III - Oxytetracycline + GnRH + Herbal Bolus (Bioheat) orally 50<sup>th</sup> day postpartum + AI and Group IV - Oxytetracycline + GnRH + non-surgical ET 7<sup>th</sup> day post-estrus. The overall mean levels of plasma progesterone, glucose, total protein, total cholesterol and total triglycerides on 50<sup>th</sup> day postpartum were  $1.80 \pm 0.46$  ng/ml,  $32.54 \pm 5.48$  mg/dl,  $4.48 \pm 0.41$  g/dl,  $57.09 \pm 8.88$  mg/dl and  $182.96 \pm 20.56$  mg/dl, respectively. Significant differences ( $P < 0.05$ ) were observed between groups for plasma  $P_4$  and triglycerides levels. The level of progesterone in control group was significantly higher than the treatment group ( $3.25 \pm 1.31$  vs  $1.32 \pm 0.41$  ng/ml). Control group showed significant variation from the treatment groups for most metabolites, but within the treatment groups there was no significant variation. Percentage of animals conceived in control and treatment groups were 33.33, 83.33, 100.00 and 50.00, respectively. The pooled conception rate of all treatment groups [77.78% (14/18)] was higher than the control group [33.33% (2/6)]. The animals of treatment group having lower plasma progesterone concentration on 50<sup>th</sup> day postpartum responded well to the hormonal (GnRH) and herbal (Bioheat) treatments and had better conception rates than the control group. The animals having higher blood glucose and lower triglycerides concentration also conceived earlier.

**Key words:** HF cows, Plasma metabolites, Progesterone concentration, Postpartum fertility.

### INTRODUCTION

Reproduction is an important consideration in the economics of the livestock production. Successful reproduction encompasses the desire and ability to mate, the capacity to conceive and nourish the embryo, and deliver the viable young one at the end of the normal gestation period. Blood plasma profile of certain hormones and metabolites is a potential aid in characterizing nutritional status and reproductive performance of an animal. Endocrine mechanisms are the most important aspects of postpartum reproductive performance. Progesterone in cyclic animals acts as a regulator of diestrus period, because as soon as the corpus luteum fails to secrete progesterone,

<sup>1</sup>Corresponding author & present correspondence address: Dr. G. K. Mishra, Manager (SS), Animal Breeding Centre (National Dairy Development Board) Salon, Po. Box. No. 1, Salon, Dist. Raebareli, (U.P.)-229 127 India. Mob. +91-94523-37680, Email: drkodu@rediffmail.com, Fax: +91-5311-275742

development of follicles begins leading to pro-estrus phase. Moreover, cows losing body condition and undergoing negative energy balance in early postpartum period usually suffer from prolonged anestrus or sub-fertility condition leading to extended calving interval (Roche and Diskin, 2000). Cholesterol being precursor of steroid hormones is expected to vary in concentration during different reproductive phases. The levels of glucose and triglycerides reflect carbohydrate and lipid metabolism in the body and thus indicate the nutritional status of the animal. The objective of this study was to view the effect of oxytetracycline intrauterine in early postpartum with and without hormonal and herbal therapy on plasma progesterone and various metabolites concentrations and conception rates in HF cows.

### MATERIALS AND METHODS

The present study was undertaken on twenty four healthy postpartum Holstein Friesian (HF) cows of

university farm at Anand (Gujarat) during the months from August 2005 to May 2006. All animals were maintained in well-ventilated hygienic sheds and were fed green fodder, hay and compounded concentrate mixture (Amul Dan) as per the standard feeding schedule followed on the farm. The animals were randomly divided into four groups, each of six animals, and managed as under.

**Group-I:** (Control) No treatment was given but cows were inseminated on the day of estrus beyond 50 days postpartum and pregnancy was confirmed per rectum 60 days post-inseminations.

**Group-II:** The animals were treated with oxytetracycline (2g Oblet Uroblex, Pfizer India Ltd.) intrauterine after expulsion of placenta and Gonadotropin Releasing Hormone i. e. GnRH 5 ml (Receptal, Intervet International GmbH, Germany) was given i/m on 50<sup>th</sup> day postpartum and inseminated on observed estrum.

**Group-III:** The same treatment was given which was given to Group-II but the animals were also given herbal (Bolus Bioheat, Kids Herbal Care, India) treatment orally on 50<sup>th</sup> day over and above GnRH and inseminated on observed estrum.

**Group-IV:** The treatment was given as per the Group-II but the animals served as recipients for fresh poor quality 2<sup>nd</sup> or 3<sup>rd</sup> grade fresh embryos non-surgically 7<sup>th</sup> day post-estrus (instead of AI).

All the animals were followed till 150 days postpartum and pregnancy was confirmed per rectum 60 days post-AI or Embryo Transfer (ET). On 50<sup>th</sup> day postpartum, about 15 ml of jugular blood samples were collected in vacutaneous vials containing Ethylene Diamine Tetra-Acetic acid (EDTA) as an anticoagulant from all animals and plasma was separated out immediately by centrifugation of samples for 15 minutes @ 3000 rpm and stored at -20°C with a drop 0.001 % Merthiolate as preservative till analysis.

Plasma progesterone ( $P_4$ ) concentration was estimated by employing standard Radio-Immuno-Assay (RIA) technique of Kubasic *et al.* (1984). The sensitivity of assay was 30 pg/ ml. Intra-assay coefficient variation was 5.4 %, while inter-assay variation was 9.1 %. Cross reactivity of the antibody with progesterone, 20 $\alpha$ -dihydroprogesterone and 17 $\alpha$ -hydroxy-progesterone was 100, 1.00 and 0.13 %, respectively.

The plasma concentration of certain metabolites, viz. plasma total protein (Biuret method; Doumas, 1975), total cholesterol (CHOD/ PAP method; Trinder, 1969), triglyceride (GPO/ PAP method; Trinder, 1969) and glucose (GOD/ POD method; Trinder, 1969) were estimated using standard assay kits procured from Crest Biosystems, with the help of autoanalyzer (Photometer, 8T 224 Biotechnica Instrument, Rome).

The results were tabulated and analysed using Completely Randomized Design and critical difference test for plasma  $P_4$  and various metabolites, and conception rates were compared by  $\chi^2$  test (Snedecor and Cochran, 1994).

## RESULTS AND DISCUSSION

The mean level of plasma progesterone on 50<sup>th</sup> day postpartum was  $1.80 \pm 0.46$  (range <0.1 to 7.6) ng/ml in HF cows. The level of progesterone in control group of animals was  $3.25 \pm 1.31$  ng/ml, which was significantly ( $P < 0.01$ ) higher than the values of all treatment groups ( $1.58 \pm 0.85$ ,  $1.41 \pm 0.72$  and  $0.96 \pm 0.63$  ng/ml, for groups- II, III and IV, respectively), however the conception rate in control group of animals was lower. Pooled service period in the treatment groups was less ( $71.5 \pm 4.83$  days) as compared to control group ( $144.83 \pm 3.39$  days). Higher conception rates were observed in all treatment groups [77.78% {14/18}, (83.33, 100.00 and 50% for groups- II, III and IV, respectively)] as compared to control group [33.33%] (Table).

These findings of higher  $P_4$  concentration with lower pregnancy rate in control group and vis-à-vis in treatment groups coincided with the results of Thibier and Rakotonanahary (1979).

The mean plasma glucose level on 50<sup>th</sup> day postpartum was  $32.54 \pm 5.48$  (range 23.92 to 43.19) mg/dl in HF cows. The level in control group of animals ( $28.19 \pm 1.00$  mg/dl) was significantly ( $P < 0.05$ ) lower than the values of treatment group ( $31.65 \pm 2.65$ ,  $35.51 \pm 2.36$  and  $34.80 \pm 1.64$  mg/dl for groups- II, III and IV, respectively). Conception rates also followed the same trend (Table).

The present values of mean plasma glucose levels observed in treatment and control groups were below the normal physiological range. The mean plasma glucose level was higher at parturition than at any stage of lactation (Ghosh *et al.*, 1991) and increasing trends of plasma glucose level from calving to different weeks postpartum has been reported Setia *et al.* (1992) and

higher plasma glucose level in normal cycling cows than anestrus and repeat breeder cows (Chandrakar *et al.*, 2003). The blood glucose level gives an indication of the energy status of an animal. Present findings of higher plasma glucose in treatment groups suggested that increasing trend of glucose concentration helps in postpartum ovarian activity and early exhibition of postpartum estrus. Oxenreider and Wagner (1971) found that both energy intake and lactation had a significant effect on plasma glucose levels during the first 8 weeks postpartum in cows.

The average level of plasma total protein on 50<sup>th</sup> day postpartum was  $4.48 \pm 0.41$  (range 2.345 to 8.922) g/dl. There was no significant difference between different groups. The level of plasma total protein in control group of animals was  $5.95 \pm 1.14$  g/dl, which was comparatively higher than the treatment group,  $4.06 \pm 0.35$ ,  $4.75 \pm 0.88$  and  $3.16 \pm 0.20$  g/dl for groups-II, III and IV, respectively (Table).

The present findings of lower protein levels observed in treatment groups than the control group were similar to that of Ahlawat (2003), but were in contrast to result of Chandrakar *et al.* (2003). Higher levels of total plasma protein were observed in control group; even though poor ovarian rebound was recorded as compared to treatment groups with better ovarian activity and pregnancy rates.

The mean plasma total cholesterol on 50<sup>th</sup> day postpartum in HF cows was  $57.09 \pm 8.88$  (range 18.62 to 167.6) mg/dl and it did not vary significantly between groups. The level of cholesterol in control group of animals was  $34.80 \pm 3.65$  mg/dl, which was comparatively lower than the values of treatment group,  $50.56 \pm 23.57$ ,  $92.87 \pm 21.31$  and  $50.16 \pm 7.48$  mg/dl for groups- II, III and IV, respectively (Table).

The increasing trend of plasma total cholesterol concentration seen from the day of calving to subsequent weeks postpartum has been reported to be associated with the initiation of ovarian activity and establishment of estrus cyclicity postpartum (Chandrakar *et al.*, 2003). Rowlands *et al.* (1980) observed 2.5 fold increases in cholesterol levels during first 8 weeks postpartum in non-suckled HF cows, but it had no relation with conception rate. Plasma total

cholesterol, being precursor of steroid hormones, is closely associated with physiological status of animal reproduction. During the present study, higher levels of plasma cholesterol were observed and so the ovarian activity and better pregnancy rates in different treatment group than the control group. It suggests that increasing trends of cholesterol concentration post partum helps in gonadal steroidogenesis, which subsequently increases the ovarian rebound and pregnancy.

The mean level of plasma triglycerides on 50<sup>th</sup> day postpartum was  $182.96 \pm 20.56$  (range 30.67 to 446.1) mg/dl in animals. The level of plasma triglycerides in control group of animals was  $289.68 \pm 47.23$  mg/dl, which was significantly higher than the values of treatment group,  $142.29 \pm 25.26$ ,  $141.18 \pm 28.47$  and  $158.69 \pm 32.07$  mg/dl for groups- II, III and IV, respectively, although the conception rate in control group of animals was lower (Table).

The present findings of triglycerides coincided with the report of Dhami *et al.* (2005). Guedon *et al.* (1999) observed that the plasma triglycerides levels were influenced by physiological status of animal and were higher during last 10 weeks of pregnancy than at or after calving. Prakash and Tandon (1979) found that the rapid increase in the triglycerides during early postpartum/ lactation was attributed to increased demand of the udder for fatty acids synthesis for milk fat, and also to lowest level of circulatory oestrogen and thyroxine concentration, which influence the lipid metabolism.

It was thus concluded that the intrauterine oxytetracycline treatment during early postpartum period and hormonal (GnRH) and herbal (Bioheat) treatments at the 50<sup>th</sup> day post partum increases fertility in the animals.

#### ACKNOWLEDGEMENTS

Authors thank Principal and Dean of the College, and Unit Officer, HF Project, AAU, Anand for extending co-operation and help during the experiment. The RIA and autoanalyser facilities availed by Research Scientist & Head, RBRU, Veterinary College, Anand, and by Deputy Director, FMD Typing Scheme Laboratory, ADIO, Ahmedabad is also gratefully acknowledged.



**Table : Plasma concentrations of progesterone and certain metabolites on 50<sup>th</sup> day postpartum in HF cows under different treatment regimes (Mean  $\pm$  SE)**

Plasma constituent	Control Group-I	Treatment groups			Overall values	'F' test
		Group-II	Group-III	Group-IV		
Progesterone (ng/ml)	3.25 $\pm$ 1.31	1.58 $\pm$ 0.85	1.41 $\pm$ 0.72	0.96 $\pm$ 0.63	1.80 $\pm$ 0.46	P<0.01
Glucose (mg/dl)	28.19 $\pm$ 1.00	31.65 $\pm$ 2.65	35.51 $\pm$ 2.36	34.80 $\pm$ 1.84	32.54 $\pm$ 5.48	P<0.05
Total protein(g/dl)	5.95 $\pm$ 1.14	4.06 $\pm$ 0.35	4.75 $\pm$ 0.88	3.16 $\pm$ 0.20	4.48 $\pm$ 0.41	NS
Cholesterol (mg/dl)	34.80 $\pm$ 3.65	50.56 $\pm$ 23.57	92.87 $\pm$ 21.31	80.16 $\pm$ 7.48	57.09 $\pm$ 8.88	NS
Triglycerides (mg/dl)	289.68 $\pm$ 47.23	142.29 $\pm$ 25.26	141.18 $\pm$ 28.47	158.69 $\pm$ 32.07	182.96 $\pm$ 20.56	P<0.01
Service period (days)	144.83 $\pm$ 3.39	83.33 $\pm$ 13.47	64.33 $\pm$ 1.43	65.33 $\pm$ 0.95	-	NS
Conception Rate (%)	33.33	83.33	100.0	50.00	-	NS

ng/ml = Nanogram per milliliter, mg/dl = Milligram per decilitre, g/dl = Gram per deciliter

#### REFERENCES

- Ahlawat, A. R. (2003). Clinical, Biochemical and Endocrinological Diagnosis of Patho-physiological Conditions in Cattle. M.V.Sc. Thesis, Gujarat Agricultural University, Anand, India.
- Chandrakar, D., Tiwari, R. P., Awasthi, M. K. and Dutta, G. K. (2003). Serum biochemical profile of repeat breeder crossbred cows. *Indian J. Anim. Reprod.*, **24**(2): 125-127.
- Dhami, A. J., Lakum, P. D., Patel, P. M., Panchal, M. T and Kavani, F. S. (2005). Blood biochemical profile in relation to age and reproductive status of Holstein Friesian cattle reared under tropical climate. *Indian J. Anim. Reprod.*, **26**(1): 34-38.
- Doumas, B. T. (1975). *Clin. Chem.*, **21**: 1159 (c. f. product profile of diagnostic kit for total protein from Crest Biosystems, Pvt. Ltd.).
- Ghosh, P. P., Roy Choudhury, R., Bandhopadhyay, S. K., Sanyal, S., Haidar, S. and Ghosh, B. B. (1991). Blood glucose and protein levels in crossbred cows during pregnancy. *Indian J. Anim. Reprod.*, **12**(2): 165-166.
- Guedon, L., Saumande, J., Dupron, F., Couquel, C. and Desbals, B. (1999). Serum cholesterol and triglycerides in postpartum beef cows and their relationship to the resumption of ovulation. *Theriogenology*, **51**(7): 1405-1415.
- Kubasic, N. P., Hallauer, G. D. and Brodows, R. G. (1984). Evaluation of direct solid phase RIA for progesterone, useful for monitoring luteal function. *Clin. Chem.*, **30**(2): 284-286.
- Oxenreider, S. L. and Wagner, W. C. (1971). Effect of lactation and energy intake on postpartum activity in the cows. *J Anim. Sci.*, **33**: 1026-1031.
- Prakash, B.S. and Tandon, R. N. (1979). A note on the late pregnancy and early lactation on blood serum cholesterol and total lipids of Holstein x Tharparkar first lactation cows. *Indian J. Anim. Sci.*, **49**: 308-309.
- Roche, J. F. and Diskin, M. G. (2000). Resumption of reproductive activity in the early postpartum period of cows. *Anim. Breed. Abstr.*, **69**: 880.
- Rowlands, G. J., Manston, R., Stark, A. J., Russel, A. M., Collis, K. A. and Collis, S. C. (1980). Changes in albumin, globulin, glucose and cholesterol concentration in the blood of dairy cows in late pregnancy and early lactation and relationship with subsequent fertility. *J. Agril. Sci.*, **94**: 517-527.
- Setia, M. S., Duggal, R. S. and Singh, R. (1992). Biochemical constituents of blood in buffaloes and cows during late pregnancy and different stages of lactation-A longitudinal study. *Buffalo J.*, **8**(2): 123-129.
- Snedecor, G. W. and Cochran, W. G. (1994). *Statistical Methods*. 8<sup>th</sup> Edn. Iowa State University Press, Ames, Iowa, USA.
- Thibier, M. and Rakotonanahary, A. (1979). Levels of plasma progesterone at the time of artificial insemination in relation to fertility in the dairy cow. *Anim. Breed. Abstr.*, **47**(2): 708.
- Trinder, P. (1969). *Ann. Clin. Biochem.*, **6**: 24 (c. f. product profile of diagnostic kit for blood glucose, total cholesterol and triglycerides from Qualigens <sup>TM</sup> Diagnostic, Pvt., Ltd.).