

## EFFECT OF DIETARY SUPPLEMENTATION WITH PUFA ON BLOOD GLUCOSE, CHOLESTROL, PROGESTERONE CONCENTRATION AND CONCEPTION RATE FOLLOWING EMBRYO TRANSFER IN CROSSBRED RECIPIENT COWS

MUKESH SAHU, SHIV PRASAD, SUNIL KUMAR\* and SATISH KUMAR

Department of Veterinary Gynaecology & Obstetrics, Veterinary Clinics, College of Veterinary and Animal Sciences, G.B. Pant University of Agriculture & Technology, Pantnagar-263145, India

Received: 27.07.2019; Accepted: 27.06.2020

### ABSTRACT

The present study was designed to observe the effect of supplementation of poly unsaturated fatty acids (PUFA) enriched diet on blood glucose, cholesterol, progesterone concentration and conception in recipient cows. Animals were divided in to 02 non treated groups (Control and T-2) each containing 3 donors and 6 recipient cows and 02 treated groups (T-1 and T-3) each containing 03 donors and 6 recipient cows. Flax seeds, source of PUFA of the Omega-3 family were fed @ 300 g/100 kg body wt/day orally for 21 days to the animals as a supplement with routine diet. Blood sampling was done in recipient cows at 0, 7, 17 and 21 days of estrous cycle. A significant increase ( $p \geq 0.05$ ) at 5% LOS in values of glucose ( $65.10 \pm 3.88$  mg/dl v  $56.95 \pm 1.29$  mg/dl) and progesterone ( $8.02 \pm 0.58$  ng/ml vs  $4.80 \pm 0.51$  ng/ml) was observed in treated viz non treated recipient groups. However, a non significant ( $p > 0.05$ ) difference was found in the value of cholesterol ( $221.82 \pm 0.27$  mg/dl vs  $209.41 \pm 3.26$  mg/dl) in treated vs non treated recipients groups. Conception could occur only in the recipients in T-3 group. On the basis of our results, it can be concluded that flax seeds supplementation may enhance early embryonic survival by increasing the values of blood glucose and progesterone following embryo transfer in cattle.

**Keywords:** Cholesterol, Conception, Glucose, Progesterone, PUFA, Recipient cows

Embryo transfer technology is a potential reproductive biotechnology which has been used since decades in many countries for faster genetic improvement of inferior livestock. There are some criteria which can be used as indicators of nutritional status of animals like weight variations, body condition, nutritional balance, blood metabolites (glucose, ketone bodies, free fatty acids, cholesterol, urea and amino acids) which directly affect the reproductive functions. Management of reproductive functions by supplementation of fat source in cattle is fairly a recent development. Feeding of fat supplement affect reproduction by affecting the dominant follicle's size, decreasing the interval to first postpartum ovulation, increasing the luteal phase progesterone concentration, manipulating uterine prostaglandin synthesis, improving embryo quality and developmental competence (Santos *et al.*, 2008). Hence, the supplementation of polyunsaturated fatty acids (PUFA) of the Omega-3 family (e.g. eicosapentaenoic acid, docosahexaenoic acid) which can be obtained from flaxseed, sunflower, fish oil etc. is proved to be beneficial. For better conception, normal blood biochemical parameters are also essential. Therefore, in the present study, attempt has been made to evaluate the effect of flexed seed supplementation treatment on blood glucose, cholesterol and progesterone in crossbred embryo recipient cows.

### MATERIALS AND METHODS

The present study was conducted on Sahiwal and crossbred cattle at Instructional Dairy Farm, Nagla, G.B.

Pant University of Agriculture and Technology, Pantnagar, District – Udham Singh Nagar (Uttarakhand). Animals were divided in to 02 non treated groups as control (no treatment given to any animal) and T-2 (treatment given to donor only) each containing 3 donors and 6 recipient cows and 02 treated groups as T-1 (treatment given to recipient only) and T-3 (treatment given to both donor and recipient) each containing 03 donors and 6 recipient cows. Donor animals were used repeatedly. Flax seeds were fed @ 300g/100kg body weight/day orally for 21 days to the animals as a supplement with routine diet.

Blood sampling was done in all 24 recipient cows at 0 (day of starting of estrus), 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>st</sup> day of estrous cycle. About 15 ml blood without anticoagulant was collected in sterilized test tube and kept at room temperature as a slant for 1 hour for separation of blood serum. After centrifugation at 3000 rpm for 15 minute, serum was separated and stored at -20 °C till further analysis.

The serum glucose and cholesterol of crossbred recipients was analysed using standard biochemical procedures. The progesterone estimation in samples was done by using progesterone C.T. RIA kit (M/S Beckman Coulter IM 1188) at IVRI (Nuclear Research Laboratory under Division of Physiology and Climatology), Izatnagar, Bareilly, Uttar Pradesh. Total twenty four Sahiwal embryos were transferred in reproductively sound and healthy crossbred cows. Two pregnancies were confirmed in T-3 group while no pregnancy was found in rest of groups.

The examination of pregnancy was done per-

Corresponding author: drsnlvet28217@gmail.com

rectally in cows on 45<sup>th</sup> day of embryo transfer. The data obtained during experimentation were analysed for mean, standard error, and coefficient of correlation. Differences between means were compared using two sample t-test and One way ANOVA (Snedecor and Cochran, 1994).

## RESULTS AND DISCUSSION

**Mean serum glucose concentration in embryo recipient cows:** The mean serum glucose concentrations in Control, T-1, T-2, and T-3 group on day 0, 7, 14 and 21 (day of starting of estrus) are presented in Table 1. There was significant difference ( $p < 0.05$ ) between control and T-1, control and T-3, T-1 and T-2, T-2 and T-3, while in between Control and T-2, T-1 and T-3 no significant difference was found.

The mean serum concentration of glucose reported in the study was nearly similar to earlier studies (Kappel *et al.*, 1984; Lager and Jordan, 2012; Sreedhar *et al.*, 2013; Malik, 2017). Ghasemzadeh *et al.* (2011), who used fish oil and soya bean as a feed supplement and found no significant difference in mean serum concentration of glucose. The mean concentration of glucose was lower as reported by Nazir *et al.* (2013). Glucose is essential for

gonads to maintain the quality of oocytes and for development of embryo (Sutton-McDowall *et al.*, 2010; Rato *et al.*, 2012).

**Total serum cholesterol in embryo recipient cows:** There was no significant difference ( $p > 0.05$ ) between groups on day 0, while significant difference was found between groups on day 7, 14 and 21 after estrus (Table 2). The mean serum cholesterol concentration of control and T-2 group found in this experiment are in agreement with reports of Kappel *et al.* (1984), Kumar (2002), Alameen and Abdelatif (2012), Malik (2017), Maithani (2017), however these workers reported lower values of serum cholesterol than in T-1 and T-4 groups.

**Total serum progesterone concentration in recipient cows:** The mean serum progesterone concentrations in Control, T-1, T-2, and T-3 group on day 0, 7, 14 and 21 (first day of estrus) are depicted in Table 3. There was no significant difference between groups on day 0 and day 7. On day 14, there was no significant difference between group control, T-1 and T-2 but between group control and T-3, T-1 and T-3 and T-2 and T-3, significant differences

**Table 1**

**Mean ( $\pm$  SE) serum glucose concentration (mg/dl) in different groups on 0, 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>th</sup> day of estrous cycle in crossbred recipients**

Day of estrous cycle	Control	T1	T2	T3
0 day	57.03 $\pm$ 0.86 <sup>B</sup>	66.06 $\pm$ 1.96 <sup>A</sup>	51.40 $\pm$ 3.31 <sup>B</sup>	63.58 $\pm$ 4.19 <sup>AB</sup>
7day	56.01 $\pm$ 1.63 <sup>B</sup>	67.15 $\pm$ 1.12 <sup>A</sup>	53.47 $\pm$ 2.89 <sup>B</sup>	66.34 $\pm$ 3.68 <sup>A</sup>
14day	55.70 $\pm$ 0.79 <sup>B</sup>	68.08 $\pm$ 1.45 <sup>A</sup>	54.94 $\pm$ 2.55 <sup>B</sup>	66.24 $\pm$ 3.04 <sup>A</sup>
21day	56.95 $\pm$ 1.29 <sup>B</sup>	66.82 $\pm$ 1.58 <sup>A</sup>	52.07 $\pm$ 1.83 <sup>B</sup>	65.10 $\pm$ 3.88 <sup>A</sup>

Values bearing the different superscripts (<sup>A,B</sup>) across row differ significantly from each other at  $p < 0.05$

**Table 2**

**Mean ( $\pm$  SE) serum Cholesterol concentration (mg/dl) in different groups on 0, 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>th</sup> day of estrous cycle in crossbred embryo recipient cows**

Day of estrous cycle	Control	T1	T2	T3
0 day	155.95 $\pm$ 3.58	158.12 $\pm$ 2.37	144.79 $\pm$ 4.04	156.66 $\pm$ 3.28
7day	173.85 $\pm$ 2.83	188.66 $\pm$ 1.95	162.15 $\pm$ 3.52	181.77 $\pm$ 5.34
14day	165.59 $\pm$ 2.56	215.50 $\pm$ 1.93	162.79 $\pm$ 3.34	210.27 $\pm$ 4.98
21day	165.50 $\pm$ 2.63	214.89 $\pm$ 1.89	157.76 $\pm$ 4.81	208.06 $\pm$ 5.68

Statistically there is no significant difference amongst groups or days at  $p < 0.05$

**Table 3**

**Mean ( $\pm$  SE) serum progesterone concentration (ng/ml) in different groups on 0, 7<sup>th</sup>, 14<sup>th</sup> and 21<sup>th</sup> day of estrous cycle in crossbred recipients**

Day of estrus cycle	Control	T1	T2	T3
0 day	0.33 $\pm$ 0.09 <sup>A</sup>	0.48 $\pm$ 0.07 <sup>A</sup>	0.37 $\pm$ 0.08 <sup>A</sup>	0.45 $\pm$ 0.05 <sup>A</sup>
7day	2.24 $\pm$ 0.25 <sup>A</sup>	2.90 $\pm$ 0.67 <sup>A</sup>	2.01 $\pm$ 0.31 <sup>A</sup>	3.61 $\pm$ 0.31 <sup>A</sup>
14day	4.11 $\pm$ 0.45 <sup>A</sup>	5.23 $\pm$ 0.70 <sup>A</sup>	4.21 $\pm$ 0.41 <sup>A</sup>	7.51 $\pm$ 1.32 <sup>B</sup>
21day	4.92 $\pm$ 0.97 <sup>A</sup>	7.70 $\pm$ 0.90 <sup>B</sup>	4.67 $\pm$ 0.42 <sup>A</sup>	7.78 $\pm$ 0.95 <sup>B</sup>

Values bearing the different superscripts (<sup>A,B</sup>) across row differ significantly from each other at  $p < 0.05$

( $p < 0.05$ ) were found in serum progesterone concentration. Interestingly on day 21, there was significant difference ( $< 0.05$ ) between control and T-1, control and T-3, T-1 and T-2, T-2 and T-3, while in between control and T-2, T-1 and T-3, no significant difference was found ( $p > 0.05$ ).

The function of corpus luteum is directly reflected via progesterone concentration which maintains the pregnancy. Rajamahendran *et al.* (1976) have reported that progesterone level remains less than 1 ng/ml during initial days of cycle then it start to increase rapidly between 4<sup>th</sup> to 12<sup>th</sup> day and reaches a maximum level of  $5.2 \pm 1.1$  ng/ml on day 14 of the cycle. Thereafter, the level decreases to  $2.6 \pm 0.6$  ng/ml on day 16 and to  $0.40 \pm 0.1$  ng/ml on day 21 of the cycle in normal cycling animal. In two different experiments by Cordeiro *et al.* (2015), it has been reported that mean serum progesterone level found on day 0 ( $0.76 \pm 0.12$  ng/ml), day 7 ( $4.72 \pm 0.23$  ng/ml), day 19 ( $3.48 \pm 0.38$  ng/ml) and day 21 ( $4.09 \pm 0.60$  ng/ml) in control group while in treated group it was found to be  $0.88 \pm 0.14$ ,  $4.92 \pm 0.24$ ,  $5.75 \pm 0.39$  and  $5.05 \pm 0.68$  ng/ml, respectively, when samples were taken on same days as for control group in which sunflower seeds was supplemented to beef cattle. In present study, similar values were obtained except value on day 7 where it is towards lower side. Ghasemzadeh *et al.* (2011) reported higher concentration of mean progesterone concentration in Holstein cows than control (6.96 ng/ml) where cows were divided into 3 groups that were fed fish oil (6.47 ng/ml), soyabean oil (8.30 ng/ml), fish oil + soyabean oil (9.62 ng/ml). However, Nazir *et al.* (2013) found lower concentrations of mean serum progesterone in both control and flax seed supplemented group than value found in present study.

#### Conception following transfer of Sahiwal embryos:

Total twenty four Sahiwal embryos were transferred in reproductively sound and healthy crossbred cows. Two pregnancies were confirmed in T-3 group, while in other groups recipient cows returned to heat.

### CONCLUSION

On the basis of above results, it may be concluded that supplementation of flax seed for 21 days during estrous cycle in recipient cows results into increase in glucose, cholesterol and progesterone concentration which may improve conception following embryo transfer.

### ACKNOWLEDGEMENT

Thanks are due to Director Experiment Station of the University, Dean, College of Veterinary and Animal Sciences and Joint Director, Instructional Dairy Farm, Pantnagar for providing necessary funds and facilities

during the investigation.

### REFERENCES

- Alameen, A.O. and Abdelatif, A.M. (2012). Metabolic and endocrine responses of crossbred dairy cows in relation to pregnancy and season under tropical conditions. *Am. Euras. J. Agric. Environ. Sci.* **12**(8): 1065-1074.
- Cordeiro, M.B., Peres, M.S., de Souza, J.M., Gaspar, P., Barbieri, F., Sá Filho, M.F., Maturana Filho, M., Dinardi, R.N., Nogueira, G.P., Mesquita, F.S. and Pugliesi, G. (2015). Supplementation with sunflower seed increases circulating cholesterol concentrations and potentially impacts on the pregnancy rates in *Bos indicus* beef cattle. *Theriogenology*. **83**(9): 1461-1468.
- Ghasemzadeh-Nava, H., Fatahnia, F. and Nikkhah, A. (2011). Effects of dietary polyunsaturated fatty acids on ovarian function and prostaglandin secretion in lactating dairy cows. *Iranian J. Vet. Med.* **5**(2): 129-135.
- Kappel, L.C., Ingraham, R.H., Morgan, E.B., Zeringue, L., Wilson, D. and Babcock, D.K. (1984). Relationship between fertility and blood glucose and cholesterol concentrations in Holstein cows. *Am. J. Vet. Res.* **45**(12): 2607-2612.
- Kumar, N. (2002). Studies on superovulatory response in Sahiwal and crossbred cows following human chorionic Gonadotropin (hCG) and estradiol pretreatment. M.V.Sc thesis submitted to G.B. Pant University of Agriculture and Technology, Pantnagar. U.S Nagar, Uttarakhand, India.
- Lager, K. and Jordan, E. (2012). The metabolic profile for the modern transition dairy cow. In: Proceedings of *Mid-South Ruminant Nutrition Conference*. Grapevine, TX, USA. pp. 9-16.
- Maithani, M. (2017). Study on tolafenamic acid to improve conception following embryo transfer in cattle. M.V.Sc. thesis submitted to G.B. Pant University of Agriculture and Technology, Pantnagar. U.S Nagar, Uttarakhand, India.
- Malik, A. (2017). Study on Flunixin meglumine to improve conception following embryo transfer in cattle. M.V.Sc thesis submitted to G.B. Pant University of Agriculture and Technology, Pantnagar. U.S Nagar, Uttarakhand, India.
- Nazir, G., Ghuman, S.P.S., Singh, J., Honparkhe, M., Ahuja, C.S., Dhaliwal, G.S., Sangha, M.K., Saijapaul, S. and Agarwal, S.K. (2013). Improvement of conception rate in postpartum flax seed supplemented buffalo with Ovsynch+ CIDR protocol. *Anim. Reprod. Sci.* **137**(1): 15-22.
- Rajamahendran, R., Lague, P.C. and Baker, R.D. (1976). Plasma progesterone levels in cycling and gonadotrophin- prostaglandin treated heifers. *Canadian J. Anim. Sci.* **56**: 37-42.
- Rato, L., Alves, M.G., Socorro, S., Duarte, A.I., Cavaco, J.E. and Oliveira, P.F. (2012). Metabolic regulation is important for spermatogenesis. *Nat. Rev. Urol.* **9**: 330-338.
- Santos, J.E.P., Cerri, R.L.A. and Sartori, R. (2008). Nutritional management of the donor cow. *Theriogenology*. **69**(1): 88-97.
- Snedecor, G.W. and Cochran, W.G. (1994). Statistical Methods (8<sup>th</sup> Edn.) Affiliated East-West Press, New Delhi.
- Sreedhar, S., Rao, K.S., Suresh, J., Moorthy, P.R.S. and Reddy, V.P. (2013). Changes in haematocrit and some serum biochemical profile of Sahiwal and Jersey×Sahiwal cows in tropical environments. *Veterinarski Arhiv.* **83** (2): 171-187.
- Sutton-McDowall, M.L., Gilchrist, R.B. and Thompson, J.G. (2010). The pivotal role of glucose metabolism in determining oocyte developmental competence. *Reproduction*. **139**(4): 685-695.