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Effect of Dietary Supplementation of Crushed Flaxseed and Soybean Oil on Ovarian Functions in Postpartum Jersey Crossbred Cows

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Abstract

The effect of dietary supplementation of crushed flaxseed and soybean oil on ovarian function in postpartum Jersey crossbred cows was studied. Total 30 postpartum normally calved cows from 2nd to 4th lactation were randomly divided into three equal groups. Cows from group T_1 and T_2 were fed with dietary supplementation of crushed flaxseed @ 15% of DM requirement from day 25 to day 70 postpartum and with soybean oil @ 3% of DM requirement of animal from day 50 to day 70 postpartum in addition to routine feed respectively, whereas cows from Group T_3 was considered as control, supplemented with routine feed. Follicular dynamic study with the help of USG was carried out at 24 hrs interval before the first injection of GnRH of Ovsynch protocol and during Ovsynch protocol (Day 0 to Day 9) of synchronization, which was implemented from day 60 postpartum in all the groups. It was observed that mean number of class I follicles was significantly increased in T_2 group, whereas the mean number of class II and total number of follicles were significantly increased in T_1 and T_2 groups on day 0. During the synchronization period the mean number of class I, class II, total numbers of follicles and preovulatory follicles size were significantly larger in T_1 and T_2 groups. It was concluded that dietary supplementation of crushed flaxseed and soybean oil helps in increasing class I, class II, total number of follicles and size of preovulatory follicle in crossbred cows.

Key words: Follicular Dynamics, Flaxseed, Soybean oil, Postpartum cows, Ovsynch protocol.

Introduction

Reproductive inefficiency is a major economic problem in dairy production, particularly during the phase of negative energy balance that occurs in early lactation. One way of improving energy status, and thereby reproductive performance is to increase the energy density of the diet with fat supplementation. Reproductive performance is enhanced by dietary fat independent of energy status (Staples *et al.,* 1998). Flaxseed and soybean oil is an excellent source of fat and polyunsaturated fatty acid (PUFA), but it is not commonly fed to cattle. PUFAs particularly the linolenic acid (omega 3) and the linoleic acid (omega 6) have gained attention owing to their classification as essential fatty acids. In cattle, dietary supplementation with various long chain

PUFAs (both n-3 and n-6) induced changes in several aspects of folliculogenesis, including both an increase in total follicular number and in the size of the dominant or pre-ovulatory follicle (Ambrose *et al.,* 2006). Higher fertility was reported in cows ovulating larger follicles even without an increase in progesterone concentration in the subsequent luteal phase (Peter and Pursley, 2003).

Some previous studies reported an increase in the number of medium-sized follicles after administration of PUFAs in the form of soybean oil or rice bran (Lammoglia *et al.,* 1996 and Thomas *et al.,* 1997). Dietary fat may enhance follicular development via metabolic hormones that act either at the ovarian level or on the central nervous system to stimulate GnRH secretion; thereby increased basal LH concentrations (Thomas and Williams, 1996). As there is minimal data on the effect of dietary supplementation of crushed flaxseed and soybean oil on bovine reproduction, the aim of this investigation was to study the influence of dietary crushed flaxseed and soybean oil on ovarian function in postpartum crossbred cows.

Materials and Methods

Selection and Treatment of Animals:

Total thirty postpartum Jersey crossbred cows (25 to 50 days in milk) were selected from the department of Dairy Science, Dr Panjabrao Deshmukh Agriculture University, Akola. All the cows which had calved normally and were from 2nd to 4th lactation were selected. Using an ultrasonic scanner, all the cows were subjected to gynaecological examination and only healthy cows were included in the study. The daily feeding of the cows was as per the routine practices along with the mineral mixture so as to meet the maintenance and production requirement. The routine feed comprised of 2/3rd roughages (2/3rd Maize Kutti + 1/3rd Jayvant grass) and 1/3rd concentrates. The dry matter consumption was estimated @ 2.5 kg/100 kg body weight. The animals were provided clean drinking water for 24 hours. All the selected cows were dewormed and vaccinated as per the routine schedule. The selected cows were divided into three groups as follows.

In Group-I, the cows were fed with crushed flaxseed at 15% of DM requirement/animal/day (Erasmus, 1993) as supplement over and above the routine feed from day 25 postpartum, while in Group-II, the cows were fed with soybean oil supplementation @ 3% of DM requirement/ animal/ day from day 50 postpartum, and this feeding was continued till synchronization period, i.e. up to day 70 postpartum in both the groups. The animals of Group-III were maintained on routine feeding practices as control and were synchronized on day 60 postpartum.

Estrus Synchronization & Follicular Dynamics:

On day 60th post-calving cows from all the groups were subjected to an Ovsynch synchronization protocol. Follicular study was carried out before onset (day 0) and with 24 hrs interval during Ovsynch protocol of estrus synchronization (day 0-9). Ultrasonography was carried out transrectally with linear-array 7.5 MH_Z probe. The size and number of follicles >3 mm were recorded on detail follicular maps. Follicles were grouped into three classes for analysis: class 1-small follicles (3.0 to 4.9 mm), class 2-medium follicles (5.0 to 9.9 mm) and class 3-large follicles (>10 mm). The growth rate, atresia rate, day of emergence of new follicle, day of deviation of follicle, diameter of deviated follicles, at the time of deviation and diameter of preovulatory follicles were studied. The data collected on various parameters were analysed by completely randomised design using WASP (Web Agri. Stat Package) developed by ICAR.

Result and Discussion

Ovarian response in different groups:

The data in Table 1 reveals that on the day of onset of Ovsynch protocol, the mean number of Class I follicles was significantly more in T_2 group, whereas the mean number of Class II and mean total number of follicles were at par in T_1 and T_2 groups and significantly higher as compared to T_3 group. The mean number of Class III follicles did not differ significantly between three groups.

This significant increase in total number of follicles in supplemented group may be due to dietary fat that may enhance follicular development via metabolic hormones that act on the central nervous system to stimulate GnRH secretion and thereby increased basal LH concentrations. Another way in which dietary fat may affect follicular development is through metabolic hormones acting at the ovarian level. Thomas and Williams, (1996) found that follicular development, along with plasma insulin and follicular IGF-^{2} concentrations, was enhanced by soybean oil. Supplementation of soybean oil was effective in increasing the number of medium sized follicles compared to a saturated fat or a highly unsaturated fat (fish oil) supplement and this was associated with higher serum insulin and increased granulose cell proliferation (Poretsky and Kalin, 1987).

Table 1: The total number of different class of follicles before onset of Ovsynch protocol for synchronization (on day 0) in different dietary groups of postpartum cows

Follicle Class	Total number different class of follicles			CD Value
	T_1	T_2	T_3	
Class I (Small)	1.9 ± 0.10^{ab}	2.0 ± 0.00^{a}	1.5 ± 0.22^{b}	CD (0.05)= 0.410
Class II (Medium)	3.4 ± 0.22^{a}	3.7 ± 0.21^{a}	1.8 ± 0.24^{b}	CD (0.01) = 0.896 CD (0.05) = 0.663
Class III (Large)	0.5 ± 0.22	0.4 ± 0.22	0.5 ± 0.16	NS
Total number of follicles	5.8 ± 0.2^{a}	6.1 ± 0.1^{a}	3.8 ± 0.2^{b}	CD (0.01) = 0.679 CD (0.05) = 0.503

T_1 crushed flaxseed, T_2 soyabean oil and T_3 control group. NS = non-significant.

From Table 2, it is evident that the mean number of follicles in Class I, Class II and mean total number of follicles during synchronization period were significantly higher in T_1 and T_2 groups as compared to T_3 group, whereas the values for T_1 and T_2 group were at par. The mean number of Class III follicles in group T_1, T_2 and T_3 groups did not differ significantly. These findings regarding the significant increase in Class I (small), Class II (medium), Class III (large) follicles and total number of follicles was in accordance with Ghasemzadeh *et al.* (2011) with feeding fish oil and soybean oil. Ponter *et al.* (2006) recorded increase in small size follicles whereas medium size follicles was in accordance with others (Thomas *et al.,* 1997; Robinson *et al.,* 2002; Kassa *et al.*, 2002) who reported increase in medium size follicles with feeding different source of linolenic and linoleic acid. The present findings are also in accordance with Ambrose *et al.* (2006) who reported non-significant difference for follicular development with flaxseed and sunflower seed.

Follicular dynamic during estrus synchronization in different groups:

The mean growth rate and day of emergence of follicle in T_1 and T_2 group were significantly higher as compared to T_3 group. The atresia rate was significantly different in soybean oil group as compared to crushed flaxseed and control group. The mean day of deviation, mean diameter of deviated follicles and mean diameter of subordinate follicle at the time of deviation did not differ significantly between groups. The mean preovulatory follicle diameter was significantly larger in T_1 and T_2 groups as compared to T_3 group, whereas it was recorded non-significantly different between T_1 and T_2 groups.

The present finding regarding mean growth rate (mm/day) of ovulatory follicle was in accordance with Ambrose *et al.* (2006) who reported no difference in growth rate with supplementation of flaxseed and sunflower seed. The atresia rate (mm/day) of reducing subordinate follicle recorded

Table 2: The total number of different classes of follicles on day 0-9 in different dietary groups of postpartum cows during Ovsynch protocol

Follicle Class	Total number of different class of follicles			CD Value
	T_1	T_2	T_3	
Class I (Small)	3.57 ± 0.08 [a]	3.82 ± 0.10 [a]	2.50 ± 0.07[b]	CD (0.01) = 0.344 CD (0.05) = 0.255
Class II (Medium)	2.62 ± 0.11[a]	2.77 ± 0.08 [a]	1.50 ± 0.07 [b]	CD (0.01) = 0.357 CD (0.05) = 0.264
Class III (Large)	1.04 ± 0.04	1.01 ± 0.03	1.00 ± 0.02	NS
Total number of follicles	7.23 ± 0.17 [a]	7.60 ± 0.16 [a]	5.00 ± 0.14 [b]	CD (0.01) = 0.635 CD (0.05) = 0.470

T_1 crushed flaxseed, T_2 soyabean oil and T_3 control group. NS = non-significant.

Table 3: The mean follicular dynamics status in different dietary groups of crossbred cows during synchronization period

Sr. No.	Parameters	T_1	T_2	T_3	CD Value
1	Mean growth rate(mm)	1.41 ± 0.05[a]	1.44 ± 0.13[a]	0.95 ±0.05[b]	CD (0.01) = 0.347 CD (0.05) = 0.257
2	Mean atresia rate (mm)	0.64 ± 0.01[a]	0.51± 0.02[b]	0.69 ± 0.02[a]	CD (0.01) = 0.082 CD (0.05) = 0.061
3	Mean day of emergence of new follicle(days)	2.0 ±0.10[b]	2.0±0.10[b]	2.5 ± 0.12[a]	CD (0.01) = 0.446 CD (0.05) = 0.330
4	Mean day of deviation (days)	3.0 ± 0.25	3.2 ± 0.32	3.3 ± 0.33	NS
5	Mean diameter of deviated follicle (mm)	8.65 ±0.05	8.54± 0.06	8.58 ±0.06	NS
6	Mean diameter of largest subordinate follicle at the time of deviation(mm)	7.32 ±0.05	7.18 ±0.04	7.32 ±0.04	NS
7	Mean diameter of pre ovulatory follicle (mm)	17.20±0.36[a]	17.00±0.36[a]	13.91±0.19[b]	CD (0.01) = 1.230 CD (0.05) = 0.911

T_1 crushed flaxseed, T_2 soyabean oil and T_3 control group. NS = non-significant.

was in close accordance with Gaur and Purohit (2007) and Henrique *et al.* (2000). The mean day of emergence of new follicle after injection of first GnRH was in close agreement with Twagiramunga *et al.* (1995). The mean day of deviation of dominant follicle after GnRH injection recorded was in agreement with Segwagwe *et al.* (2006). The mean diameter of deviated and subordinate follicle on the day of deviation recorded was in accordance with Ginther *et al.* (2000) who reported the mean diameter of deviated and subordinate follicle on the day of deviation was 8.5 and 7.7 mm, respectively. The mean diameter of preovulatory follicle recorded was in close agreement with Ghasemzadeh *et al.* (2011) in fish oil and soybean oil groups as compared to control group of cows. The non-significant difference between the mean diameter of preovulatory follicle in flaxseed and soybean oil group recorded was not in agreement with Ambrose *et al.* (2006). They found larger mean diameter of ovulatory follicle in cows fed with flaxseed as compared to sunflower seed fed group. These differences in the mean diameter of preovulatory follicle with earlier study may be due to different source of linolenic acid and linoleic acid.

Conclusion

From the study it was concluded that dietary supplementation of crushed flaxseed and soybean oil helps in increasing Class I, Class II and total number of follicles, and size of preovulatory follicle in postpartum cows.

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Conflict of Interest: All authors declare no conflict of interest.

References:

Ambrose, J.D., Kastelic, J.P., Corbett, R., Pitney, P.A., Petit, H.V., Small, J.A., Zalkovic, P., (2006). Lower pregnancy losses in lactating dairy cows fed a diet enriched in alpha-linolenic acid. *J. Dairy Sci.*, 89: 3066–3074.

Erasmus, U. (1993). *Fats that Heal Fats that Kill*. 9th Ed. Alive books, Burnaby, BC. pp.1-456.

Gaur, Mitesh and Purohit G.N. (2007). Follicular dynamics in Rathi cattle. *Veterinarski Arciv,* 77(2): 177-186.

Ghasemzadeh-Nava**,** H; Fatahnia, F., Nikkhah,A. and Zamiri, M.J. (2011). Effects of dietary polyunsaturated fatty acids on ovarian function and prostaglandin secretion in lactating dairy cows. *Int. J. Vet .Res.,* 5(2): 129-135.

Ginther, O.J., Bergfelt, D.R., Kulick, L.J. and Kot, K. (2000). Selection of the dominant follicle in cattle: Role of estradiol. *Biol.Reprod*., 63: 383-389.

Henrique, J.M.V., Ademir, De.M.F., Wanderlei, F.D.S. and Luis Sergio D.A.C. (2000). Follicular dynamics in zebu cattle. *pesq. agropec. bras. Brasilia* 35(12): 2501-2509.

Kassa, T., Ambrose, J.D., Adams, A.L., Risco, C., Staples, C.R., Thatcher, M.J., Van Horn, H.H., Garcia, A., Head, H.H. and Thatcher, W.W. (2002). Effects of whole cottonseed diet and recombinant bovine somatotropin on ovarian follicular dynamics in lactating dairy cows. *J. Dairy Sci.*,85: 2823-2830.

Lammoglia, M.A., Willard, S.T., Oldham, J.R. and Randel, R.D. (1996). Effects of dietary fat and season on steroid hormonal profiles before parturition and on hormonal, cholesterol, triglycerides, follicular patterns, and postpartum reproduction in Brahman cows. *J. Anim. Sci.,* 74: 2253-2262.

Peters, M.W. and Pursley, J.R. (2003). Timing of final GnRH of the Ovsynch protocol affects ovulatory follicle size, subsequent luteal function, and fertility in dairy cows. *Theriogenology*, 60: 1197-1204.

Ponter, A.A., Parsy. A.F., Sadeeb, M. and Grimardia. B. (2006). Effect of supplement rich in linolenic acid added to the diet of postpartum dairy cows on ovarian follicle growth, and milk and plasma fatty acid composition. *Reprod. Nutr. Dev*., 46: 19-29.

Poresty, L. and Kalin, M.F. (1987). The gonadotropic function of insulin. *Endocr. Rev*., 8: 132-139.

Robinson, R.S., Pushpakumara, P.G.A., Cheng, Z.., Peters, A.R., Abayasekara, D.R.E. and Wathes, D.C. (2002). Effects of dietary polyunsaturated fatty acids on ovarian and uterine function in lactating dairy cows. *Reproduction* 124: 119-131.

Segwagwe, B.V.E., Macmillan, K.L. and Mansell, P.D. (2006). The effect of GnRH or oestradiol injected at pro-oestrus on luteal function and follicular dynamics of the subsequent oestrous cycle in non-lactating cyclic Holstein cows. *J. Vet. Res.,* 73: 61-70.

Staples, C.R., Burke, J.M. and Thatcher, W.W. (1998). Influence of supplemental fats on reproductive tissues and performance of lactating cows. *J. Dairy Sci.,* 81: 856-871.

Thomas, M.G. and Williams, G.L. (1996). Metabolic hormone secretion and FSH – induced superovulatory response of beef heifers fed dietary fat supplements containing predominantly saturated or polyunsaturated fatty acids. *Theriogenology*, 45: 451-458.

Thomas, M.G., Bao, B. and Williams, G.L. (1997). Dietary fats varying in their fatty acid composition differentially influence follicular growth in cows fed isoenergetic diets. *J. Anim. Sci.*, 75: 2512-2519.

Twagiramunga, H., Guiltbault, L.A. and Dufour, J.J (1995). Synchronization of ovarian follicular waves with a gonadotropin-releasing hormone agonist to increase the precision of estrus in cattle: a review. *J. Anim. Sci.,* 73: 3141-3151.

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