EFFECT OF DIETARY SUPPLEMENTATION OF CRUSHED FLAXSEED AND SOYABEAN OIL ON ESTRUS ATTRIBUTES IN POSTPARTUM COWS
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Abstract: The study was undertaken to find out the effect of dietary supplementation of crushed flaxseed and soyabean oil on estrus attributes in postpartum cows. Total 48 postpartum crossbred multiparous cycling cows with normal calving history were considered and divided into three groups viz., 15% crushed flaxseed (T1), 3% soyabean oil (T2) and Control (T3- no additional fat). Cows from all the groups were subjected to an ovsynch synchronization protocol on day 60 postpartum treated with Inj. Buserelin acetate 10µg i/m on day 0, Inj. Cloprostenol sodium 500 µg on day 7 and Inj. Buserelin acetate 10µg i/m on day 9, to observe induced estrus response, time required for onset of induced estrus, intensity and duration of induced estrus respectively. The induced oestrus response was numerically highest in group fed with Soyabean oil. It was observed that the cows from soyabean oil followed by crushed flaxseed supplemented groups were numerically responded earlier to the PGF$_2\alpha$ injection as compared to the cows from control group. The percent intense intensity of estrus was numerically higher in cows supplemented with soyabean oil followed by crushed flaxseed supplemented group as compared with control group. There was no significant difference between the groups for time required for onset of induced estrus and duration of oestrus, whereas it was observed that soybean oil fed cows responded earlier.

Keywords: Crushed flaxseed, soyabean oil, oestrous response, intensity of oestrous.

Introduction
Recently, there has been a great deal of interest in feeding fat to dairy cows in order to increase energy density of the diet and improve reproduction (Oldick et al. 1997 and Staples et al. 1998). There are two main families of essential fatty acid omega-3 and omega-6, which have been linked to fertility. Animals cannot synthesize omega-6 or omega-3 fatty acids therefore these need to be supplied in the diet (Wathes et al., 2007). The effect of omega-3 and omega-6 on prostaglandin and steroid hormones also has the potential to affect the onset of oestrus and ovulation (Abayasekara and Wathes, 1999). In particular increased PGF$_2\alpha$ or PGE$_2$ associated with higher omega-6 could stimulate early luteolysis of the corpus luteum and earlier onset of oestrus (Mattos et al., 2000), while inhibition of PGF$_2\alpha$ by omega-3 has

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the potential to have the opposite effect. The times to behavioural oestrus and plasma LH and E2 peak were numerically longer when cows were supplemented with a high omega-3 linseed diet pre- and post-partum (Zachut et al., 2011). Keeping in view the above-mentioned facts, the present research work was undertaken on dietary supplementation of soybean oil and crushed linseed in postpartum cows for improving the reproductive performance in terms of estrus attributes.

Material Methods

A population of 48 normally calved postpartum crossbred multiparous cycling cows (25 to 50 days in milk or DIM) were selected from Instructional Livestock Farm Complex Dr. Punjabrao Deshmukh Krishi Vidyapeth, Akola. The cows were subjected to gynaecological examination using transrectal ultrasonography before inclusion in the study. Cows were confined for the entire period of study to a barn with access to an open shelter space. The selected cows were divided into three equal groups. Each group was fed one of the diets in a randomized design. Dietary treatments consisted of either no additional fat (control diet, T₃), 15% feed dry matter basis approximately (300 g/100 kg of body weight/animal/day) crushed flaxseed (T₁) as supplemented over and above from day 25 postpartum and was continued for 45 days and 3% soybean oil (feed dry matter basis) as supplement over and above (T₂) from day 50 postpartum and was continued for 20 days. All the cows from three different treatment groups were subjected to an ovsynch synchronization protocol on day 60 postpartum treated with Inj. Buserelin acetate 10µg i/m on day 0, Inj. Cloprostenol sodium 500 µg on day 7 and Inj. Buserelin acetate 10µg i/m on day 9 for three groups, respectively. The response in all the cows were observed for the estrus attributes like induced estrus response, time required for onset of induced estrus, intensity of induced oestrus and duration of induced estrus.

Statistical Analysis

The data was analysed by unequal completely randomized design using online software of Web Agri Stat Package 2.0.

Results and discussion

Induced estrus response

The percent induced estrus response recorded in present study in T₁, T₂ and T₃ groups was 87.5, 100 and 81.25 percent, respectively (Table 1).
Table 1: The induced estrus response (%) in crushed flaxseed (T₁) soyabean oil (T₂) and control (T₃) groups

<table>
<thead>
<tr>
<th>Sr.No</th>
<th>Parameters</th>
<th>Treatment groups</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>T₁ (n=16)</td>
</tr>
<tr>
<td>1</td>
<td>Induced estrus response (%)</td>
<td>14/16=87.5</td>
</tr>
</tbody>
</table>

The percent induced estrus response was numerically higher in cows supplemented with soyabean oil followed by crushed flaxseed supplemented group as compared to control. The 100 percent induced estrus response was observed in soyabean oil fed cows which might be associated with higher n-6 could stimulate early luteolysis of the CL and earlier onset of estrus (Mattos et al., 2000). The present finding regarding induced estrus response in T₁ and T₂ groups is in close agreement with Dirandeh et al. (2013) reported 80 and 95 % induced estrus response in cows fed with flaxseed and soyabean, respectively. However, Fillery et al. (2000) reported no difference observed in induced estrus in cows supplemented with omega-6 compared without supplementation which is in accordance with findings of present study in T₁ and T₃ groups. In contrast, the lower estrus response was reported by Grant et al. (2003) reported 50% induced estrus response synchronized on day 40 postpartum in cows supplemented with safflower seed. Wuenschel (2006) reported 73.3% induced estrus response in heifers supplemented with rumen protected unsaturated fatty acid which is lower than the present findings.

The results of present findings for the induced estrus response in T₃ groups is in accordance with Satriomoorthy and Subramanium (2003) reported 80% induced estrus response with ovsynch protocol in postpartum dairy cows. Dirandeh et al. (2014) reported 82.5% induced estrus response with Ovsynch protocol in postpartum dairy cows which is in accordance with the present findings.

The higher induced estrus response than the present findings of T₃ group recorded by Velladuri et al. (2014) reported 100% induced estrus response with ovsynch protocol in postpartum Kankrej cows. The lower induced estrus response than the present findings of T₃ group recorded by Colazo and Mapletoft (2014) reported 68 % induced estrus response with ovsynch protocol in postpartum dairy cows and similarly Cabara and Serafi (2013) reported 63% induced estrus response with ovsynch protocol in postpartum dairy cows. The discrepancy in induced estrus response of present study with the earlier findings of various...
research workers might be due to the different source of linolenic and linoleic acid fed to the cows, duration of feeding which leads to variation in amount of fatty acid reaching to the specific tissue. (Scholljegerdes et al., 2011), different breeds, season, different feeding patterns and variation in reproductive status of animals.

**Time required for onset of induced estrus.**

The mean time required for onset of induced estrus observed in T1, T2 and T3 groups were 46.00 ± 2.65, 42.12 ± 1.61 and 45.53 ± 2.90 hrs, respectively. It was observed that the cows from soyabean oil followed by crushed flaxseed supplemented groups were numerically responded earlier to the PGF2α injection as compared to the cows from control group. There was no significant difference between the groups for time required for onset of induced estrus (Table 2).

**Table 2: The mean time required for onset of estrus in crushed flaxseed (T1) soyabean oil (T2) and control (T3) groups**

<table>
<thead>
<tr>
<th>Sr. No</th>
<th>Parameters</th>
<th>T1 (n=14)</th>
<th>T2 (n=16)</th>
<th>T3 (n=13)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Time required for onset of induced estrus (hrs)</td>
<td>46.00 ± 2.65</td>
<td>42.12 ± 1.61</td>
<td>45.53 ± 2.90</td>
</tr>
</tbody>
</table>

(Differences between means were found statistically non-significant)

Earlier estrus response in soyabean oil fed cows might be associated with higher (n-6) could stimulate early luteolysis of the CL and earlier onset of estrus (Mattos et.al., 2000), while inhibition of PGF2α by (n-3) delayed the luteolysis of the CL (Burke et.al., 1997) which is in accordance with present findings.

The present findings for the numerically longer time required for onset of estrus in T1 group as compared with T2 group is in agreement with Zachut et al.(2010) reported numerically longer time required for onset of estrus with extruded flaxseed fed cows as compared to sunflower fed cows. However, Wuenchel (2006) observed non significant difference for the time required for onset of estrus with fish meal fed and no fat fed cows which are in concurrence with the present findings in T1 and T2 groups compared with T3 group.

The results of present findings for the time required for onset of induced estrus in T1 and T2 groups is not in accordance with Zachut et al.(2011) reported longer 60.4 ± 2.3 hrs time required for the onset of induced estrus in cows supplemented with extruded flaxseed from prepartum to postpartum period.
The results of present findings for the time required for onset of induced estrus in T₃ groups is in accordance with Ahmed et al. (2016) and Velladuri et al. (2014) reported 48.75 ± 0.71 and 47.97 ± 2.65 hrs, respectively with ovsynch protocol in postpartum dairy cows. The results of present findings for the time required for onset of induced estrus in T₃ group is not in accordance with Sathiomoorthy et al. (2007) reported higher 56.40 ± 8.40 hrs in non descript cows with ovsynch protocol. The discrepancy in the time required for the onset of induced estrus of present study with the earlier findings of various research workers might be due to the different source of PUFA fed to the cows, duration of feeding PUFA which leads to variation in amount of fatty acid reaching to the specific tissue. (Scholljegerdes et al., 2011), different breeds, season, different feeding patterns and variation in reproductive status of animals.

**Intensity of estrus**

The 42.86, 42.86 and 14.28 percent cows from crushed flaxseed supplemented group (T₁) showed intense, intermediate and weak type of estrus, respectively. The cows from soyabean oil supplemented group (T₂) showed 56.25, 31.25 and 12.50 percent intense, intermediate and weak type of estrus, respectively, where as 38.46, 46.15 and 15.39 percent cows from control group (T₃) showed intense, intermediate and weak type of estrus, respectively (Table 3).

<table>
<thead>
<tr>
<th>Sr.No.</th>
<th>Groups</th>
<th>Intensity of estrus (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>T₁(n=14)</td>
<td>6/14 (42.86%) 6/14 (42.86%) 2/14 (14.28 %)</td>
</tr>
<tr>
<td>2</td>
<td>T₂(n=16)</td>
<td>9/16 (56.25%) 5/16 (31.25%) 2/16 (12.50%)</td>
</tr>
<tr>
<td>3</td>
<td>T₃(n=13)</td>
<td>5/13(38.46%) 6/13 (46.15%) 2/13 (15.39%)</td>
</tr>
</tbody>
</table>

The percent intense intensity of estrus was numerically higher in cows supplemented with soyabean oil followed by crushed flaxseed supplemented group as compared with control group. It showed the potential of polyunsaturated fatty acid to influence the greater behavioural estrus characteristics. This might be due to longer duration of preovulatory estrodiol surge which might help in greater intensity of behavioural estrus (Zachut et al., 2010).

The present findings for numerically greater intensity of induced estrus in soyabean oil followed by crushed flaxseed as compared to control group is in close accordance with Boken
et al. (2005) observed that feeding cows in pasture with soyabean oil refining by-product increased the number of mount during estrus compared with control. Similarly Zachut et al. (2010) reported behavioural estrus intensity was higher in extruded flaxseed fed cows as compared to no fat fed cows.

The higher intense type of intensity than the present findings of T3 group recorded by Bhoraniya et al. (2012) and Velladuri et al. (2014) reported 50%, respectively with ovsynch protocol in postpartum dairy cows. Whereas the lower intense type of intensity than the present findings of T3 group recorded by Ahmed et al. (2016) reported 16.67 percent with ovsynch protocol in postpartum dairy cows. The discrepancy in the intense type of intensity of present study for T3 group with the earlier findings of various research workers might be due to the variation in reproductive status of animals, breed, season and different feeding pattern.

**Duration of estrus**

The mean duration of estrus recorded in T1, T2 and T3 groups were 21.42 ± 0.57, 22.25 ± 0.54 and 21.84 ± 0.65 hrs, respectively (Table 4). There was no significant difference between the groups and the mean duration of estrus was not affected by the diet containing crushed flaxseed and soyabean oil.

**Table 4: The mean duration of estrus in crushed flaxseed (T1) soyabean oil (T2) and control (T3) groups**

<table>
<thead>
<tr>
<th>Sr.No</th>
<th>Parameters</th>
<th>T1 (n=14)</th>
<th>T2 (n=16)</th>
<th>T3 (n=13)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Duration of estrus (hrs)</td>
<td>21.42 ± 0.57</td>
<td>22.25 ± 0.54</td>
<td>21.84 ± 0.65</td>
</tr>
</tbody>
</table>

(Differences between means were found statistically non-significant)

The mean duration of induced estrus observed in crushed flaxseed groups in present study is slightly higher than Zachut et al. (2010) reported the duration of induced estrus was 18.60 ± 0.8 hrs by feeding flaxseed supplementation. Ahmad et al. (2016) recorded the duration of induced estrus with ovsynch protocol was 21.08 ± 0.78 hrs which is in accordance with results of T3 groups and similarly Krishnakumar and Chandrahasan (2012) reported 21.80 hrs duration of estrus in cows synchronized with ovsynch protocol.

The higher duration of induced estrus than the results of present findings of T3 group recorded by Velladuri et al. (2014) reported 29.84 ± 0.67 hrs in postpartum dairy cows synchronized with ovsynch protocol. The lower duration of induced estrus than the results of present findings of T3 group recorded by Sathiamoorthy et al. (2007) reported 18.40 ± 2.60 hrs in postpartum non descript cows synchronized with ovsynch protocol. The discrepancy in the duration of induced estrus in present study for T3 group with the earlier findings of
various research workers might be due to the variation in reproductive status of animals, breed, season and different feeding pattern.

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