Efficacy of herbal plants to cure repeat breeding due to sub-clinical endometritis in cattle

Brahmanand, Omprakash Shrivastava, Satyanidhi Shukla, Akshay Kumar and Rajesh Kumar

Abstract
The present study was conducted to describe the effects of intrauterine infusion of methanol fraction of neem oil and extract of neem bark in the treatment of sub-clinical endometritis in repeat breeder cattle. A total of 100 repeat breeder cattle were screened from college livestock farm, organized dairy farm and farmers’ door in and around Jabalpur. After recording history, all the animals were subjected to gynaecological examination and White side test. A total of thirty repeat breeder cattle with sub-clinical endometritis were selected and randomly divided into five groups. The group I cattle i.e. control group were administered with 25 ml normal saline at 24 hrs. Interval for three days. Group II and III were administered with methanol fraction of neem oil for the different time interval. Group IV and V were administered with neem bark extract. Efficacy of both preparations was assessed by Fern pattern, pH of CVM, white side test and conception rate. The results indicate that administration of neem oil at 24 hrs. Interval for two days retrieved the cattle from sub-clinical endometritis. All cattle (100% in group II) showed negative to white side test. Further, the conception rate (83.33%) was obtained in neem oil fraction treated cattle at 24 hrs. interval than other treatment groups. On the basis of the present investigation, it is concluded that infusion of neem oil fraction at 24 hrs. interval for two days was found as the most effective treatment among all treated groups and, thus, can replace conventional antibiotic in the future for sub-clinical endometritis leading to repeat breeding condition in cattle.

Keywords: Conception rate, neem oil, repeat breeding, sub-clinical endometritis

1. Introduction
Repeat breeding is one of the major functional causes (40%) of infertility in dairy cows [1] posing considerable economic loss to dairy farmers. Endometritis, particularly subclinical endometritis has been the major cause of repeat breeding [2]. Subclinical endometritis alter the physico-chemical properties of cervical mucus and, therefore, examination of cervical mucus for appearance, consistency and pH may be valuable in its diagnosis. As far as therapeutic approach to repeat breeder cow is concerned, it involves either antibiotics and antiseptics or hormonal therapy [3, 4]. However, these therapies involve high cost of treatment, inconsistent results, milk disposal and several other side effects. Therefore, herbal preparations called as phytotherapy were used for treatment of subclinical endometritis and repeat breeding condition in cows in the present study.

2. Materials and Methods
A Total of 100 repeat breeding cattle were selected on the basis of history, breeding records and per rectal examination. Further, Thirty repeat breeder cattle due to sub-clinical endometritis was selected on the basis of white side test. The repeat breeding animals were divided into five treatment groups containing six animals in each group, viz. group I, II, III, IV and V. Different regimes of drugs were followed for different time interval and for different period. In group I, 25 ml normal saline was administered, once a day (OD), intrauterine (I/U) for three days. In group II, 25 ml methanol fraction of neem oil was administered, OD, I/U for two days. In group III, 25 ml methanol fraction of neem oil was administered, at 48 hrs. Interval, I/U for two days. In group IV, 25 ml neem bark extract was administered, OD, I/U for three days. In group V, 25 ml neem bark extract was administered at 48 hrs. Interval, I/U for three days. Estrual cervical mucus samples were collected on the day of estrum before and after treatment (on next estrus) and tested for appearance, pH, Fern pattern and white side test. At subsequent estrus after treatment, all the animals treated were inseminated. Cattle,
which returned to heat after first insemination were again inseminated at second estrus after treatment. Pregnancy was confirmed per rectally after 45-60 days of insemination. The data so generated were analysed using analysis of variance (ANOVA) to test the significant differences of means [3].

3. Result
The data regarding Fern pattern, Whiteside test, pH of cervical mucus and Conception Rate in sub-clinical endometritic repeat breeder cattle is presented in Table 1. The sub-clinical endometritic cows exhibited typical fern pattern as 16.67, 16.67, 0.00, 0.00 and 16.67 per cent, atypical fern pattern as 50.00, 66.67, 83.33, 66.67 and 66.67 per cent, nil fern pattern as 33.33, 16.67, 16.67, 33.33 and 16.67 per cent before treatment in group I, II, III, IV and V, respectively. After treatment the cows exhibited typical fern pattern as 33.33, 83.33, 66.67, 66.67 and 50.00 per cent, atypical fern pattern as 50.00, 16.67, 33.33, 16.67 and 33.33 per cent, nil fern pattern as 16.67, 0.00, 0.00, 16.67 and 16.67 per cent in group I, II, III, IV and V, respectively. After treatment the cows exhibited typical fern pattern as 33.33, 83.33, 66.67, 66.67 and 50.00 per cent, atypical fern pattern as 50.00, 16.67, 33.33, 16.67 and 33.33 per cent, nil fern pattern as 16.67, 0.00, 0.00, 16.67 and 16.67 per cent in group I, II, III, IV and V, respectively. Results revealed maximum number of animals exhibiting typical fern pattern in group II after treatment, followed by group III (66.67%), group IV (66.67%), group V (50.00%) and least number of animals exhibited typical fern pattern in group I (33.33%).

Whiteside test was performed on CVM collected from the animals of all the groups before and after treatment. All the animals selected for the study were positive to white side test. The data presented revealed that after treatment, at subsequent estrus, 83.33 per cent cattle in group I, 0.00 per cent in group II, 33.33 per cent in group III & IV and 50 per cent in group V became positive for white side test. The analysis of data reveals that WST was negative in group II as compared to other treatment groups with treatment of neem oil administered at 24 hrs. Interval for two days showing better effect of treatment.

The pH value of CVM before treatment from different groups varied from 7.40±0.05 to 7.67±0.05. The pH values before treatment varied significantly (p<0.05) between different groups. The difference was found non-significant (p>0.05) between treatment groups I and IV, II and III, II and V, III and V whereas, it was significant (p<0.05) between groups I and II, I and III, I and V.

The pH value of CVM after treatment in different groups varied from 7.00±0.05 to 7.50±0.04. The pH value after treatment was significantly decreased in all the groups (p<0.05). However, the post treatment difference was found statistically non-significant (p>0.05) between treatment groups II and III, II and IV, III and IV, whereas, it was significant (p<0.05) between groups I and II, I and III, I and IV, I and V. Fertility response was recorded in terms of conception rate in different treatment groups of sub-clinical endometritic repeat breeder cattle. The first service conception rate in groups I, II, III, IV and V were 0.00, 66.67, 33.33, 16.67 and 16.67 per cent, respectively. The overall conception rate was seen to be higher (83.33%) in group II as compared to group III (66.67%), IV (50.00%), V (33.33%) and group I (33.33%).

4. Discussion
Clear estrual mucus is conducive for sperm penetration and conception, whereas, turbidity retards sperm motility in estrual mucus [6]. No reference could be traced about the increase in clarity of mucus after treatment with herbal extract used. However, percentage of cows with clear discharge after treatment with antibiotic was found to be higher that reported by [7]. This may be attributed to the use of the different antibiotic. Thick cervical mucus may retard sperm motility and thus, may cause fertilization failure [8] In this study, mean pH of estrual cervical mucus in all the groups before treatment was towards the alkaline side (more than 7.4) indicating infection [9, 10]. This increase in pH may be caused due to metabolites of bacteria and inflammatory exudates in estrual cervical mucus [11]. Significant decline (p<0.05) in pH was observed in all the groups after treatment. A significant decrease (p<0.05) in pH of group II (7.00±0.05), group III (7.10±0.04), group IV (7.10±0.04), group V (7.30±0.04) was observed as compared to group I (7.50±0.04). This reduction in pH may be due to the decline in bacterial load and inflammatory process in uterus after treatment [12]. All the cattle taken in this study were positive (100%) for white side test before treatment indicating positive for sub-clinical endometritis. At subsequent estrus after treatment, 83.33% cattle in group I followed by 0.00% in group II, 33.33% cattle in group III, IV and 50% cattle in group V became positive to white side test. Positive reaction to white side test could be explained on the basis of number of leukocytes present in the uterine discharge [13]. The normal discharge has less number of leukocytes to cause any change of colour, whereas in clinical and sub-clinical cases of endometritis, discharge contains increased number of leukocytes causing a colour reaction [14]. The absence of colour development to white side test in higher number of cattle treated with herbal extracts revealed their efficacy for combating infection.

Better conception rate (83.33%) was found in methanol fractioned neem oil at 24 hrs. interval for two days treated group as compared to other treatment groups. Conception rate of 66.67% was achieved in group III when treated with methanol fractioned neem oil at 48 hrs. For two days followed by group IV (50%) which is treated with neem bark extract at 24 hrs. interval for three days, group V and I (33.33%). Maximum conception rate of 83.33% was achieved in group II.

Table 1: Fern pattern, white side test, pH of cervical mucus and conception rate in sub-clinical endometritic repeat breeder cattle. (Figures in parenthesis in Fern pattern are No. of animals and in conception rate are per cent of animals.)

<table>
<thead>
<tr>
<th>Groups</th>
<th>Fern Pattern</th>
<th>Whitens Test</th>
<th>pH of CVM</th>
<th>Conception Rate</th>
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<tbody>
<tr>
<td></td>
<td>Typical (%)</td>
<td>Atypical (%)</td>
<td>Nil (%)</td>
<td>Positive (%)</td>
</tr>
<tr>
<td>Group  I</td>
<td>16.67 (1)</td>
<td>50.00 (3)</td>
<td>33.33 (2)</td>
<td>33.33 (2)</td>
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<tr>
<td>Group II</td>
<td>16.67 (1)</td>
<td>66.67 (4)</td>
<td>16.67 (1)</td>
<td>83.33 (5)</td>
</tr>
<tr>
<td>Group III</td>
<td>0.00 (0)</td>
<td>83.33 (5)</td>
<td>16.67 (1)</td>
<td>66.67 (4)</td>
</tr>
</tbody>
</table>
5. Conclusion
It is concluded from this study that the herbal treatment of sub-clinical endometritis and thus repeat breeding cattle with intrauterine infusion of methanol fractioned neem oil at 24 hrs. Interval for two days post estrus, cured the condition and subsequently improved conception rate.

6. References