

*Original Research***Efficacy of Some Non-antibiotic Treatments in Postpartum Dairy Cows with Subclinical Endometritis**

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Abstract

Several aerobic and anaerobic bacteria contaminate the bovine uterus and depress immune function around parturition; thus, it results in clinical and subclinical endometritis (SCE) leading to reduced fertility. Efficacy of some non-antibiotic therapeutic protocols for SCE confirmed by cytobrush endometrial cytology on day 35 postpartum was evaluated in dairy cows with 15 in each group. Untreated cows in Group I served as control, Group II and III cows received single and double injections of PGF2 α , respectively whereas, once used intravaginal CIDR device was kept in situ for 14 days to Group IV cows. Single injection and double injection of PGF2 α aided the recovery rate (46.67% & 53.33%), while CIDR insert resulted in higher recovery rate (60%) and spontaneous recovery was 40%. It was concluded that once used CIDR provides a new hope for treatment of SCE as the sustained Progesterone level makes the dominant follicle to persist and secrete estrogen that promotes phagocytosis and clearance of infection.

Key words: Dairy Cows, Subclinical Endometritis, Endometrial Cytology, PGF2 α , CIDR**How to cite:** Sahadev, A., Krishnaswamy, A., Renukaradhya, G., & Rashmi, S. (2019). Efficacy of Some Non-antibiotic Treatments in Postpartum Dairy Cows with Subclinical Endometritis. International Journal of Livestock Research, 9(8), 133-139. doi: 10.5455/ijlr.20190409052840**Introduction**

Parturition and immediate postpartum period have the greatest impact on health and productivity of the dam. More than 80 per cent of dairy cows have a bacterial contamination of the uterine lumen in the first two weeks of parturition (Sheldon *et al.*, 2009a). There is a cycle of bacterial contamination, clearance and recontamination for several weeks after parturition. In many cows, the bacterial contamination is mostly resolved by uterine involution, passage of lochia and mobilization of immune defence systems. However, failure to resolve the bacterial contamination compromise the uterine defence function due to persistence of pathogenic bacterial infection of uterus in 10 to 20 per cent of dairy cows (LeBlanc *et al.*, 2002; Sheldon *et al.*, 2009a). The most common bacteria associated with uterine infections are *Escherichia coli*,

Arcanobacterium pyogenes, *Fusobacterium necrophorum* and *Prevotella sp.* (Sheldon *et al.*, 2002b) and are associated with tissue damage, delayed uterine involution, disruption of endometrial function and altered ovarian cycles (Sheldon *et al.*, 2002a; Herath *et al.*, 2006). Resultant infertility in acute cases and sub fertility in chronic cases characterized by reduced conception, increase calving to conception interval and culling rates (Herath *et al.*, 2006; Sheldon *et al.*, 2009b). Among the uterine infections, majority are either cases of CE or SCE (Sheldon *et al.*, 2009a). Clinical endometritis is characterized by purulent vaginal discharges after 21 days postpartum or a mucopurulent discharge after 26 days postpartum (Sheldon *et al.*, 2006). In the absence of clinical signs, the possibility of SCE often exists. SCE is defined according to the stage of postpartum by the presence of neutrophils greater than 10 per cent in the uterine cytological samples 35 days postpartum. Both CE and SCE significantly influence the postpartum reproductive performance in dairy cattle (Kasimanickam *et al.*, 2004; Kasimanickam *et al.*, 2005a; Barlund *et al.*, 2008; Dubuc *et al.*, 2010).

In view of the negative impact on subsequent reproductive performance due to subclinical endometritis, it obviates to identify such cows after parturition and efforts to prevent deleterious effect on fertility. Whiteside test has been applied as cow side test to detect the SCE with varying accuracies (Satheesh Kumar and Puniamurthy, 2007; Bhat *et al.*, 2014) using cervical mucus or uterine flushing. However, uterine cytology has been used as a standard technique to detect cows with SCE (Kasimanickam *et al.*, 2004). Antibiotics are used as treatment of choice for genital infections for many years. To overcome the undesirable effect of antibiotics such as the risk of bacterial resistance, treatment failures, risk of antibiotic residues in tissues and milk and moreover, depressed natural uterine defense mechanism, Prostaglandin F_{2α} is used as one of the alternative treatments for endometritis including LPS from *E. coli*, lysozyme, oyster glycogen (Palaniswamy *et al.*, 2014). Alternatively, treatment with CIDR has shown promising results in cows with clinical endometritis (Hatler *et al.* (2008). However, such study in cows with SCE has not been reported. Therefore, the present study was attempted to evaluate the efficacy of used CIDR for treatment of SCE. Since, CIDR provides sustained and low concentration of progesterone, prolong the lifespan of dominant follicle and reduce the ability of endogenous estradiol-17β to induce a pre-ovulatory surge of LH in the cow. With this background, a study was designed to evaluate the therapeutic efficacy of some non-antibiotic protocols for postpartum SCE in crossbred dairy cows.

Materials and Methods

The study was conducted utilizing Holstein Friesian and Jersey crossbred cows maintained at five organized dairy farms in and around the Bangalore between July 2015 and January 2017. The herd size varied from 37 to 115, age ranged from 3.6 to 12 years with the previous lactation yield of 2440 to 7930 liters. The animals were regularly dewormed, vaccinated, allowed for grazing and housed in tie stalls on concrete floor or stone slabs. All the cows were fed with ragi straw and green fodder/silage and supplemented with concentrate feeds

and mineral mixture. Recently calved cows about 35 days were subjected to physical and vaginoscopic examination for presence of any abnormal vaginal discharges (flakey, cloudy, mucopurulent or purulent) were considered as clinical endometritis. Cows treated with antibiotics or any other treatments were excluded from the study. One hundred and ninety-four cows found negative for clinical endometritis were subjected for SCE screening by endometrial cytology using cytobrush technique as per the methods proposed by Kasimanickam *et al.* (2004). Ninety-eight cows were found positive for SCE based on whiteside test and confirmed by endometrial cytology where $> 10\%$ PMNs is considered.

A total of 60 cows with SCE at 35 days post-partum (35dpp) were randomly allotted to the four groups with 15 each. Group I cows remained untreated and served as the control. Each cow of Group II received a single intramuscular injection of 25 mg of Dinoprost tromethamine (Lutalyse®, Pfizer Animal Health, Mumbai) immediately after the diagnosis. Cows of Group III were treated with two intramuscular injections of $\text{PGF}_2\alpha$ at 12 days interval. Group IV cows received once used CIDR inserted into the vagina of SCE cows and left *in situ* for 14 days (The CIDR used in animals for the purpose of estrus synchronization or induction was collected after its removal and kept in a clean container for sterilization as stated by Eslami *et al.*, 2015).

The cows of Group I, II and III were examined on day 7 and 14 post diagnosis and/or treatment whereas the cows of Group IV were examined on 14-day post treatment (i.e., the day of CIDR removal) for persistence or clearance of the SCE based on endometrial cytology. The cut off PMN level was fixed at $>10\%$ up to 42 dpp and $>5\%$ from 49 dpp onwards. The efficacy of treatment protocols were evaluated based on the recovery rate in terms of reduction in the proportion of PMNs in all the groups after 14 days. The data generated from the study were analyzed by multiple proportion test as described by Steel and Torrie (1981).

Results and Discussion

In control group, spontaneous recovery following the initial diagnosis of SCE evaluated on day 42 and day 49 postpartum, revealed. Five cows on day 42 (33.33 %) and one cow on day 49 (6.67 %) recovered spontaneously with an overall recovery of 40.00 per cent were recorded. In Group II, 5 out of 15 cows (33.33 %) on day 42 and two cows (13.33 %) did not exhibit any evidence of SCE on day 49 postpartum treated with $\text{PGF}_2\alpha$ on 35 day with an overall recovery rate of 46.67 per cent. Cows which received double $\text{PGF}_2\alpha$ (Group III) on day 35 and day 47, six cows (13.33 %) did not show any evidence of SCE on day 54 and two cows (40.00 %) on day 61 postpartum with overall recovery rate of 53.33 per cent. While in group IV, 9 out of 15 cows (60%) showed complete recovery from SCE immediately after the removal of used CIDR (day 49 postpartum) (Table 1).

Table 1: Relative efficacy of different therapeutic protocols for SCE in crossbred dairy cows

Group (N=15)	42 dpp (7 days Post Treatment)	49 dpp (14 days Post Treatment)	Recovered (%)	Not Recovered
Group I (control)	05(33.33)	01(6.67)	06/15(40.00)	09/15(60.00)
Group II (SPG)	05(33.33)	02(13.33)	07/15(46.67)	08/15(53.33)
Group III (DPG)	06*(40.00)	02**(13.33)	08/15(53.33)	07/15(46.67)
Group IV (CIDR)	0(0.00)	09(60.00)	09/15(60.00)	06/15(40.00)

*52 dpp ** 59 dpp. Values in the parentheses indicate percentage

In the present study, the recovery percentage of SCE 14 days after single, double PGF₂ α and used CIDR treatment (excluding spontaneous recovery) were respectively 46.67, 53.33 and 60.00 per cent (Table 1). CIDR treatment resulted in higher recovery rate (60%); whereas in single and double PGF₂ α treatment groups yielded marginally higher recovery rate (46.67% and 53.33%) as compared to the control group (40%). It is difficult to precisely predict spontaneous recovery of SCE. Nonetheless, early diagnosis of SCE therefore becomes a necessity. Therefore, endometrial cytology provides simple and efficient tool to detect SCE in cows. Previous studies have demonstrated decrease in the percentage of uterine lumen PMNs with lapse of time (Kasimanickam *et al.*, 2005a; Gabler *et al.*, 2009). The decrease in PMNs was due to decrease in the uterine inflammatory process with increased postpartum period as the result of involution process (Gilbert *et al.*, 2005; Senosy *et al.*, 2009).

The post-partum uterus is never sterile and 85 to 90 percent of the uteri of cows are infected during the first two weeks of parturition (Dekruif, 1978; Bostedt, 1984). The infection spontaneously decreases within the next few weeks (Steffan *et al.*, 1984) and sterile by 4 to 5th week after parturition. In the present study, uterine infection persisted without any clinical signs in up to 50 per cent cows with normal calving at day 35 postpartum. However, it was observed that the uterine defence mechanism of the cow continues to make efforts to cleanse the uterus even beyond 35 dpp. Although 33.33 per cent of cows recovered spontaneously between 35 and 42 dpp in the present study, nevertheless, the number of cows which recovered from SCE thereafter are very few. Similar findings were reported by Kasimanickam *et al.* (2004, 2005); Gilbert *et al.* (2005), Back *et al.* (2009) and Plontzke *et al.* (2010) in dairy cows and in crossbred cows (Bacha and Regassa, 2009). The spontaneous recovery observed in six cows which did not receive any treatment may be ascribed to cyclical activity and the estrus phase which enhance the uterine immunity besides expulsion of uterine contents due to myometrial contractions which aided in mechanical cleansing of the uterus.

In the present study, cows which received PGF₂ α treatment, 12 cows did not have palpable corpus luteum (CL) prior to treatment. Among the cows responded to treatment (7 cows), four cows although they did not have a palpable CL recovered from SCE. Galvao *et al.* (2009) and Dubuc *et al.* (2010) found no beneficial effect of PGF₂ α treatment for the SCE in their studies. Britt *et al.* (1964) and Tennant and Peddicord (1968) reported uterotonic effect of PGF₂ α which is independent of progesterone concentration at the time of treatment

(Martinez and Theiber, 1984). Therefore, the beneficial effect of $\text{PGF}_{2\alpha}$ in cows without CL, as observed in the present study may be subscribed to uterotonic effect of $\text{PGF}_{2\alpha}$ and promoting phagocytosis by enhancing neutrophil function due to its pro-inflammatory effect (Lewis, 2004). The treatment of SCE with two injections of $\text{PGF}_{2\alpha}$ was attempted to bring about repeated uterotonic effects in case where the cyclical activity was yet to be initiated. The repeated $\text{PGF}_{2\alpha}$ may promote phagocytosis and cleansing of uterus. However, in the present study, following double injection, 13 and 7 per cent higher recovery rate than single $\text{PGF}_{2\alpha}$ and the control cows was recorded, respectively. Therefore, repeated $\text{PGF}_{2\alpha}$ treatment may be beneficial in cases of postpartum SCE. However, this needs further studies utilizing larger sample size to draw any conclusions. The CIDR that has been used for 14-day vaginal insertion to induce or synchronize estrus in cows which maintain the dominant follicle and causes sustained production of estradiol 17β that up-regulate the uterine defence mechanism (Hatler *et al.*, 2008). This concept was exploited by Eslami *et al.* (2015) utilizing the once used CIDR as an alternate treatment for CE in cows. The present study strengthens the above concept in which used CIDR was used for treatment of SCE. The results revealed higher recovery in SCE cows treated with CIDR than single or double injection of $\text{PGF}_{2\alpha}$ treatment, suggesting the used CIDR could be effectively utilized instead of being discarded.

Conclusion

It is concluded that SCE could be present up to 50 per cent of crossbred cows on day 35 postpartum. About 40 per cent of these cows recover spontaneously. However, not ignoring the adverse effect of SCE on subsequent reproductive performance, an early diagnosis of SCE and treatment measures becomes necessary in cows. Though single and double $\text{PGF}_{2\alpha}$ injections considerably alter the course of the disease, however, CIDR treatment significantly improved the recovery rate in cows with SCE and offers a new hope for the treatment of SCE.

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