Full Length Research Paper

# Effect of hormonal and antibiotic treatment of Holstein cows with uterine discharges at 25 days in milk on reproductive performance in Mexican Plateau

M. A. Lammoglia, I. C. Daniel\* and A. Cabrera

Facultad de Ciencias Biológicas y Agropecuarias, Universidad Veracruzana, Poza Rica-Tuxpan.

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Endometritis treatment is a controversial topic among veterinary practitioners. The objective of this study was to evaluate reproductive profiles of dairy cows that presented uterine discharge at 25 days in milk and were treated with hormones and a specific antibiotic for intrauterine infusion. In a single herd, at 25 (±5) days after parturition, the reproductive tract (vulva, vagina, cervix and size of the uterine horns) of Holstein cows (n = 304) were evaluated and grouped as: 1) cows that presented normal uterine horns size, none or light uterine discharge and needed no treatment (healthy, n = 250); and, 2) cows that had placental retention and/or uterine discharge and increased size of one of the uterine horns. Cows in group 2 were treated with hormones (estradiol benzoate and prostaglandin  $F_{2\alpha}$ ) and uterine infusion (500 mg of cephapirin benzathine in 19.6 g ointment base; Metricure, Intervet; UIP group, n = 54). The statistical model included: number of treatments, days to first service, days open. number of services per conception and calving interval as affected by uterine discharge. Healthy cows had lower (P = 0.0001) number of treatments (0.0) than UIP cows (2.3 ± .31). UIP cows showed a delay (P = 0.0001) to first service (78.33  $\pm$  2.9 days) compared with healthy cows (67.46  $\pm$  0.9). Healthy cows conceived faster (P = 0.0001) and required less (P = 0.0001) services per conception (92.16 ± 2.7 days and 1.76 ± 0.08, respectively) than UIP cows (281.2 ± 31.6 days and 4.74 ± 0.47, respectively). The calving interval in healthy cows (374.05  $\pm$  2.7 days) was shorter (P = 0.0001) than in UIP cows (591  $\pm$  40.2 days). In conclusion dairy cows that had uterine discharge 25 days after calving and treated with hormones and intrauterine specific antibiotic showed no improvement in reproductive patterns compared to cows with no uterine discharges.

Key words: Uterine discharge, uterine therapy, reproductive performance.

## INTRODUCTION

Dairy cows have been genetically selected to produce more milk and these increases in milk production that had led to decrease postpartum reproduction. To improve the reproductive efficiency of dairy cows, basic care techniques have been developed, before, during and after calving. Despite the rigorous care, there are still a percentage of cows that cannot begin proper milk production and reproduction (Dubuc et al., 2011; Lucy, 2001). The high milk yield in modern cows does not explain completely the low reproductive rate and other factors have been attributed in the equation such as placental retention, metritis, ketosis etc (Galvao et al., 2009; Grohn and Rajala-Schultz, 2000). The normal physiological process of calving includes cervix dilation and also the uterus exposure to environmental bacteria resulting in 90% of cows infected after birth (Overton, 2003). To identify cows with postpartum reproductive problems, some protocols have been developed to follow up the postpartum period (LeBlanc, 2011). These protocols are based on measuring the rectal temperature during the first 10 days after calving and evaluation of daily food consumption and milk production (Overton, 2003). The last reproductive evaluation to discharge the transition and fresh cow is between 20 and 30 days after

<sup>\*</sup>Corresponding author. E-mail: idaniel@uv.mx.

Variable	Prepartum's diet	Fresh cow's diet
Dry matter	10.1 kg	15 kg
Crude protein	15.5%	17.5%
Metabolically energy Mcal/kg DM	2.52	2.66
Lactating net energy Mcal/kg DM	1.62	1.71
Macro minerals	35.0 g	209.0 g
Components anionic macro minerals	300 g	
Micro minerals	15.0 g	8.0 g
Additives	150.0 g	220.0 g
Vitamins: A, D3 and E	53.0 g	103.0 g
Mineral total	553.0 g	540.0 g
DM minerals	5.5%	3.6%
Forage	6.0 kg	8.6 kg
Concentrate	4.0 kg	6.36 kg

 Table 1. Rations fed to prepartum and fresh cows.

parturition. This assessment of the reproductive tract includes size of the horns, content of the uterus and presence of discharge in cervix, vagina and vulva. However, Leblanc (2011) reported that this time might not be best to evaluate fresh cows.

Despite all these efforts, there are still cows that develop retained placenta and/or uterine discharges with reproductive consequences and controversy continues among practitioners. The objectives of this study were to evaluate short and long term reproductive performance of Holstein cows that had uterine discharge and received hormone and a specific antibiotic for intrauterine infusion treatment at 25 days in milk.

#### MATERIALS AND METHODS

The study followed reproduction performance of cows that had no uterine infection 25 days after calving (healthy group) and cows that had uterine discharge and increased uterine size. The observational study was conducted (June 2008 to 2010) in a dairy farm located in central Mexico plateau at 2200 m altitude. The study population consisted of lactating Holstein cows in typical Mexican plateau commercial dairy herds with modern free-stall (capacity of 90 cows/pen with no crowding). A total of 304 cows were milked 3 times a day producing 33.4 L. The cows were milked for 305 days (± 13 days) and started their reproductive program at 55 days postpartum. Pregnant cows were dried at day 233 of gestation (± 10 days) and transferred to the dry cow pen. According to the service date, as they approach 21 days to calving, dry cows were moved to a prepartum pen which basically consisted in offering a daily diet (formulated by a nutritionist) of 10.1 kg of dry matter (DM) supplemented with 0.3 kg of anionic salts (Table 1). Cows in the prepartum pen were regularly observed 24 h a day to help them at calving if assistance was needed. After parturition, cows were allowed to clean their calves and they were pulled away before nursing. Cows were milked and colostrum was fed to their calves (at least 10% of its body weight). After calving, cows started on 3 milkings a day and remained in the fresh cow pen.

Diagnosis of the reproductive tract was done in all the cows

(primiparas and multiparous) by one veterinarian practitioner at  $25 \pm 5$  days in milk and consisted in a physical assessment of the vulva, vagina, cervix and uterus as described by LeBlanc et al. (2002). If cows had normal size of uterine horns and no uterine, cervical or vaginal discharge were considered healthy and they were moved to the general population of cows. Cows were assigned to a pen according to its body size and lactation. Cows that had uterine discharge (any presence mucus with flecks of pus, muco-purulent or purulent in the cervix, vagina or vulva was considered discharge (LeBlanc et al., 2002) at 25 days in milk and had increased size of one or both of the uterine horns that were diagnosed as not healthy (UIP; n = 54). Cows not healthy were treated and evaluated weakly until they were diagnosed as healthy or clean (no discharge).

UIP treatment consisted of: 1) day one, an intramuscular injection of 5 mg of estradiol benzoate (Syba) followed by an intramuscular injection of 25 mg of prostaglandin F2 $\alpha$  (Lutalyse; Pfizer) and the second day an intramuscular injection of 25 mg of prostaglandin F2 $\alpha$  (Lutalyse; Pfizer) and the second day an intramuscular injection of 25 mg of prostaglandin F2 $\alpha$  was given. Eight days later, the reproductive tract of these cows was evaluated again. If the reproductive tract of these cows had no discharge (clean mucus) and uterine horns had reduced size, cows were diagnosed as healthy. However, if cows had uterine discharge and increased uterine horn size cows were diagnosed as not healthy and they were treated with an intrauterine infusion of 500 mg of cephapirin benzathine in 19.6 g ointment base (Metricure, Intervet). Treated cows were evaluated every eight days and treated with Metricure until they were diagnosed as healthy.

When all cows were diagnosed as healthy, they were assigned by body size and lactation to 1 of 4 pens; pen 1 primiparas n = 90; pens 2, 3 and 4 multiparous n = 214). All cows had tail paint and estrus detection was observed every day 3 to 5 times a day for 45 min periods. Pregnancy diagnose was conducted using ultrasonography (Honda 1201, 5 MHZ, real time with a rectal probe) around 28 days after artificial insemination. Pregnant cows were confirmed at 60, 90 and 200 days of gestation. If cows were open ovaries and uterus were closely evaluated, cows with follicles bigger than 12 mm and cows with a corpus luteum (CL) received an injection of 150 mcg of GnRH (Gonasyl; Syba) and 7 days latter cows were injected with 25 mg of prostaglandin  $F_{2\alpha}$  (Lutalyse; Pfizer). Cows were tail painted and observed for signs of estrus 3 times a day for 45 min periods. The statistical analysis was done using StatView ANOVA. No differences were found between primiparas and multiparous cows and data was polled. The variables were: number of treatments per cow, days to first service,

Dependent variable	Healthy	UIP	Р
Number of treatments	0.0	2.3 ± .31	0.0001
Days to first service	67.46 ± .9	78.33 ± 2.9	0.0001
Days open	92.16 ± 2.7	281.2 ± 31.6	0.0001
Number of services per conception	1.76 ± .08	4.74 ± .47	0.0001
Calving interval	374.05 ± 2.7	591.0 ± 40.2	0.0001

**Table 2.** Comparison of reproductive performance of cows that had a healthy uterus and cows that had retained and/or uterine discharge (UIP) during the first 30 days after parturition.

days to conception, number of services per conception and calving interval as affected by uterine discharge at 25 days in milk.

## RESULTS

In this study, it was found that with all the necessary care before and after calving, 17.8% of the cows presented uterine discharge. Cows that had a healthy uterus after calving had much better reproductive performance than cows that had retained and/or uterine discharge. Cows with healthy uterus had lower (P = 0.0001) number of hormonal and uterine antibiotic treatments, less days to first service, days open, number of services per conception and shorter calving interval than cows that had uterine infectious processes (Table 2).

### DISCUSSION

In this study, it is reported that 17.8 % of the cows had uterine discharges at the 25th day in milk evaluation. LeBlanc et al. (2002) reported that 16.9% of the cows had uterine discharged when evaluated at 20 to 30 days in milk, which supports the results found in this study. Also, Chenault et al. (2004) found that 12 to 53% of the cows had uterine discharge around 28 days in milk. However, if cows would have been evaluated at latter days in milk results could have been different. LeBlanc (2011) reported that cows evaluated after 33 days in milk had less incidence of discharge that cows evaluated earlier. LeBlanc (2011) suggested that some cows with uterine discharge clean themselves without treatment; therefore, cows might need to be evaluated after 33 days in milk. Cows with UIP took several treatments to be diagnosed as healthy or uterine infection-free compared with healthy cows. High producing Holstein cows had different steroid's metabolism patterns (Wiltbank et al., 2006). Dairy cows that had retained placenta and/or UIP presented higher blood cortisol concentrations even before calving (Lyimo et al., 2000). In addition, dairy cows after calving were also immunological depressed (Goff and Horst, 1997). Cows with UIP might have compromised the immune and hormonal systems of the uterus to fight infection. Therefore, UIP cows needed an additional help such as hormonal and antibiotic treatment to alleviate the infection. Alleviation of uterine infection does not mean that fertility could be improved (Kaufmann et al., 2010; Galvao et al., 2009).

In this study, cows with UIP had longer time to present estrus behavior and to be served for the first time also, UIP cows required a greater number of services and days to conceive. This results were similar to those reported by Chenault et al. (2004) in which cows with postpartum uterine discharge had up to 20 more days open than cows that had no discharge after calving. Lewis (1997) reported that a large number of postpartum cows develop some degree of uterine infection but most of these cows heal by themselves between 21 to 30 days (Dubuc et al., 2011). Inflammation of the endometrium resulted in low fertility, reduced uterine involution and follicular dynamics depressed (Deluyker et al., 1991; Griffin et al., 1974; Lewis 1997). It had been suggested that prostaglandin is  $F_{2\alpha}$  active in the GnRH patterns and any alteration in this hormone may affect reproductive performance (Randel et al., 1996). These support the delay of UIP cows for first service and greater number of services to conceive in this study. In addition, according to LeBlanc (2011), treatment of cows that had retained placenta or had uterine discharge should be uterine treated after 33 days in milk to improve fertility and any treatment before this could have inconsistent results in reproduction. Cows with UIP that bred back had prolonged calving interval. What is interesting is that the difference between healthy cows and UIP cows in first service was only 10.87 days but difference to days open and calving interval, it is greater. This could be explained that UIP cows might had persisted subclinical endometritis and it took longer for them to breed back. In this study, most of the cows that did not breed back were induced to a new lactation with hormone treatment; therefore we did not have any data about culling cows. However, Dubuc et al. (2011) reported that the only risk for culling was that uterine discharge caused decreased fertility and open cows at the end of lactation.

In conclusion, Holstein cows in the central Mexican plateau that had uterine discharge at 25 days in milk and treated with hormones and antibiotics infused intrauterine did not have improved reproduction patterns. We suggest conducting more research to treat uterine discharge and best timing to apply these treatments in order to consistently improve reproduction in these cows.

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