

The Effect of Intrauterine Cephapirin on Treatment of Endometritis in Commercial Dairy Cattle

Ahmadi^{*}, M.R., Nazifi, S. and Ghaisari, H.R.

*Clinical Sciences Dept., School of Veterinary Medicine, Shiraz University,
P.O.Box.1731, Shiraz, Iran*

Received 23 Dec 2004; accepted 7 May 2005

Summary

To evaluate the effect of intrauterine cephalosporin on treatment of endometritis 39 Holstein dairy cows that affected with postpartum endometritis were selected and randomly assigned to two experimental designs. In experiment 1, 6 out of 14 cows were 25 to 30 days after parturition were treated by intrauterine oxytetracycline. Another 8 cows were treated by intrauterine cephalosporin (Metri-Care). In experiment 2, 25 cows that were 55-60 days after parturition received three types of treatment. 6 cows were received only PGF2 α , 9 cows were received PGF2 α and procaine penicillin G, and 10 cows were treated by intrauterine Metri-Care. Cervical mucosa was collected before and after each treatment. There was no significant difference between mean (\pm SD) of neutrophils before and 14 days after treatment with them. Conception rates of cows received oxytetracycline and Metri-Care were 66.66% (4/6) and 37.5% (3/8), respectively. There was significant difference between mean of neutrophils before and 14 days after treatment at all groups. Conception rate of cows received only PGF2 α was 33.33 % (2/6) and in cows received penicillin and PGF2 α was 55.55% (5/9) but in cows received Metri-Care was 90.0% (9/10). Therefore, Metri-care may be good choice for treatment of postpartum uterine infection in cow.

Keywords: endometritis, cephalosporin, neutrophils, cervical mucosa, penicillin, oxytetracycline

* Author for correspondence. Email:rahmadi@shirazu.ac.ir

Introduction

Endometritis is characterized by inflammation of the endometrium and follows parturition, copulation, AI and infusion of irritants into the endometrial cavity (Youngquist 1997). The degree of its effect on fertility varies with the severity of the inflammation, the time required for resolution of endometrial lesions, and the extent of permanent changes that impair endometrial gland functions and/or alter the uterine and/or oviductal environment. The majority of postpartum dairy cows have some degree of endometritis but resolve it by 40 to 50 days post partum (Bretzlaff 1987, Bruun *et al* 2002).

Endometritis reduces the fertility and milk production of dairy cows (Borsberry & Dobson 1989, Esslemont & Spincer 1993) and the effect of endometritis varies between herds (Sheldon & Noakes 1998). The initial uterine defence against bacterial infection is phagocytosis by uterine leukocytes (mainly neutrophils). The abnormal puerperium affects uterine defence mechanisms adversely and prolongs the time to complete uterine involution (Hussain 1989). Many cows slightly delayed uterine involution at 30 days post partum (Sheldon & Noakes 1998). When antimicrobial therapy is indicated, tetracycline is recommended for intrauterine use during the early postpartum period when mixed bacterial populations are present (Bretzlaff 1987). However, in a study most isolates of *Actinomyces pyogenes* recovered from the uterus of cows were resistant to oxytetracycline and intrauterine treatment with large doses did not affect the frequency of *A.pyogenes* isolation (Cohen *et al* 1995). The bovine uterus is an anaerobic environment, thus antibiotics chosen for intrauterine use must be active in the absence of oxygen (El-Azab 1988). In addition, most antibiotics and chemicals depress the activity of uterine neutrophils and interfere with the uterine defence mechanism; therefore, the potential benefit of their use must be carefully weighed against their deleterious effect (Vandeplassche 1981). Organisms that cause postpartum uterine infections are usually sensitive to penicillin, but bacterial contamination during the first several weeks after calving

produce penicillinase, which renders the drug ineffective if applied locally. By 30 days postpartum these organisms are usually eliminated, and intrauterine treatment with penicillin is more likely to be effective after that time (Olson *et al* 1984).

Benzatin cephapirin proposed to be use for the intrauterine treatment of endometritis in individual cattle. The aim of this study was to compare the efficiency of Metri-Care treatment of postpartum endometritis with routine treatments oxytetracycline at first month of postpartum and penicillin at next months in commercial dairy herds around Shiraz.

Materials and Methods

Drug. The drugs including were used include PGF2 α (Cloprostenol, Aboraihan, Iran) for induction of estrus; oxytetracycline 10% (Oxytetracycline, Razak, Iran), penicillin procaine G (Nasr, Iran) and Metri-Care (Metri-Care, Afarin Daroo, Iran) as intrauterine antibiotic were used. Metri-Care is an oily suspension of benzathine cephapirin that is packaged in 19g syringes for intra-uterine use. There is 500mg cephapirin/10ml syringe.

Animal. During examination of dairy herds, which had routine visits for monitoring fertility, cows with clinical signs of endometritis were selected. The criteria for selection were that they had been calved 25-30 days (Experiment 1) and 55-60 days (Experiment 2) before time and they had a mucopurulent vulval discharge or feel abnormality at rectal palpation that confirmed by cytological evaluation of cervical mucosa. Finally 39 Holstein dairy cows were selected that were affected to postpartum endometritis. 14 cows were at 25 to 30 days after parturition and 26 cows were at 55-60 days after parturition. Endometritis was diagnosed by clinical signs using rectal palpation. Cytological smear of cervical mucosa was used for confirming of it (Ahmadi *et al* 1998, 2002). Cervical mucosa was collected by gentle suction from the cervical external Os with plastic uterine pipette covered by larger plastic tube and was aspirated by 50ml syringes.

Differential cellular counts were done on Giemsa stained smears of the mucosa. All cows at the time of diagnosis and treatment had a developed corpus luteum. They were at diestrus phase 7 ± 2 days after oestrus sign and received $\text{PGF2}\alpha$ for induction of oestrus, and were at luteal phase 14 days after treatment. Therefore we could not detect different oestrus cycle phase at time of diagnosis, sampling, treatment and resampling time.

Treatment protocol. Experiment 1). Cows were given one of the following treatments. 14 dairy cows affected to endometritis were selected at 25-30 days after parturition. They had mucopurulent discharge and were at luteal phase (oestrus sign more than 7 days ago). 6 cows were treated by 5g intrauterine oxytetracycline supplied in 50ml disposable syringes with disposable transcervical catheters. The remaining cows ($n=8$) were treated by one syringe intrauterine. The content of one Metri-Care syringe was introduced into the lumen of the uterus by using the disposable catheter. The syringe was fixed to the catheter. The cervix of the uterus was taken into a gloved hand introduced into the rectum. The catheter was introduced through the cervix into the lumen of the uterus, by gentle oscillating movements of the cervix. Experiment 2). 25 cows with mild endometritis sign that confirmed by cytological method were selected. They were at 55-60 days after parturition. 6 cows were received only $\text{PGF2}\alpha$ for induction of oestrus. 9 cows were received $\text{PGF2}\alpha$ and infused with 5million IU of procaine penicillin G in 50ml of sterile water. 10 cows were received one syringe intrauterine Metri-Care (similar to Experiment 1). The reproductive activity was monitored at next seven months after treatment, and conception rate of all groups was recorded.

Statistical analysis. Results of this study were analyzed by ANOVA, Duncan, Fisher exact and Chi-Square tests at 11.5 version of SPSS software.

Results

The results of experiment 1 are showed in table 1. The mean of epithelial cell at all groups was changed between 31-63%. Large vacuolated epithelial cells changed

2.67-6.12%. The range of mean of neutrophils was 31.50-62.62%. The percentage of lymphocyte and monocyte were very low, approximately zero. There was no significant difference between mean (\pm SD) of neutrophils before and 14 days after treatment with oxytetracycline and Metri-Care ($P>0.05$). Conception rate of cows received oxytetracycline was 66.66% (4/6) but in cows received Metri-Care was 37.5% (3/8). There was no significant difference between two conception rates ($P>0.05$).

Table 1. Comparison of cytological change percentages before and 2 weeks after treatment by oxytetracycline and Metri-Care at 20-30 after parturition

Drug	N	EPC	LVEP	Neutrophil	Lymphocyte	Monocyte
OTC (1)	6	53.67 \pm 18.03	2.67 \pm 2.08	42.67 \pm 18.66	0.50 \pm 0.34	0.17 \pm 0.17
OTC (2)	6	63.17 \pm 16.48	4.67 \pm 3.18	31.50 \pm 15.00	0.00 \pm 0.00	0.00 \pm 0.00
Metri-Care (1)	8	31.00 \pm 9.39	6.12 \pm 2.54	62.62 \pm 9.68	0.00 \pm 0.00a	0.00 \pm 0.00a
Metri-Care (2)	8	38.12 \pm 11.69	2.87 \pm 1.30	58.62 \pm 11.87	0.50 \pm 0.27b	0.25 \pm 0.25b

EPC=Epithelial Cell, LVEP=Large Vacuolated Epithelial Cell, (1)=Day of treatment, (2)=14 days after treatment

The results of experiment 2 are shown in table 2. There was significant ($P<0.05$) difference between mean (\pm SD) of some examined cells before and 14 days after treatment at all groups. Conception rate of cows received only PGF 2α was 33.33% (2/6).

Table 2. Comparison of cytological change percentages before and 2 weeks after treatment by penicillin and Metri-Care with control group at 50-60 days after parturition

Drug	N	EPC	LVEP	Neutrophil	Lymphocyte	Monocyte
Control (1)	6	53.33 \pm 20.18c	1.83 \pm 1.01c	44.67 \pm 20.11c	0.00 \pm 0.00	0.00 \pm 0.00c
Control (2)	6	71.17 \pm 13.14d	19.67 \pm 9.94d	9.00 \pm 5.11d	0.00 \pm 0.00	0.17 \pm 0.17d
Penicillin (1)	9	72.56 \pm 9.27	13.44 \pm 4.31	13.67 \pm 9.02	0.11 \pm 0.11	0.00 \pm 0.00e
Penicillin (2)	9	70.89 \pm 9.82	10.89 \pm 4.77	17.22 \pm 7.12	0.78 \pm 0.78	0.22 \pm 0.15f
Metri-Care (1)	10	49.80 \pm 13.47a	13.50 \pm 6.04a	36.20 \pm 12.29a	0.20 \pm 0.20a	0.30 \pm 0.21a
Metri-Care (2)	10	92.10 \pm 3.67b	1.80 \pm 0.69b	5.70 \pm 3.26b	0.00 \pm 0.00b	0.00 \pm 0.00b

Values with different superscripts in each column are those that differ significantly ($P<0.05$). EPC=Epithelial Cell, LVEP=Large Vacuolated Epithelial Cell, (1)=Day of treatment, (2)=14 days after treatment

Conception rate of cows received penicillin and PGF 2α were 55.55% (5/9) but in Metri-Care and PGF 2α received group it was 90.0% (9/10). There was no significant difference between three conception rates ($P>0.05$).

Discussion

A variety of antibiotics have been infused into the uterus of cows in attempts to treat postpartum infections. Some authors have found intrauterine treatment to be beneficial while others have found it to have no effect (El-Azab *et al* 1988). Many preparations of oxytetracycline are irritating and cause chemical endometritis. If oxytetracycline is selected for intrauterine therapy, doses 4 to 6g/day have been recommended (Youngquist 1997). Penicillin or one of its synthetic analogs is most commonly recommended for parenteral administration to cows with uterine infections. Ceftiofur has been suggested as an alternative if the patient does not appear to respond to penicillin administration (Youngquist 1997, Kasimanockam *et al* 2005). Oxytetracycline is probably not a good choice for systemic administration because of the difficulty in reaching the minimal inhibitory concentration required for *A.pyogenes* in the lumen of the uterus (Montes & Pugh 1993). Sheldon and Noakes (1998) reported the overall curing rate of postpartum endometritis was 68 percent. Oxytetracycline was successful in 73 percent of cases, cloprostenol in 67 percent and oestradiol in 63 percent of cases. There was no significant difference between the success rates of the treatments, except for cows with mild endometritis in which oxytetracycline was more successful than oestradiol. Conception interval was shorter at cows treated with oxytetracycline and PGF 2α than oestradiol. A prospective clinical trial was conducted on two large dairies in the San Joaquin Valley of California to determine whether a single intrauterine infusion with procaine penicillin G or oxytetracycline reduced the calving-to-conception interval in cows with endometritis (Thurmond *et al* 1993). Oxytetracycline satisfies most of the criteria for the selection of an antimicrobial substance for the treatment of

endometritis (Noakes 1991), it is poorly absorbed from the uterus after an intrauterine infusion (Bretzlaff *et al* 1983) and its endometrial concentration are higher and persist longer than after it has been administered by other parenteral routes (Masera 1980). de Kruif *et al* (1982) reported that there is no point in carrying out a single intra-uterine treatment about the fifteenth day after parturition in cattle affected with endometritis.

The result of this study showed that oxytetracycline is better for treatment at early stage of postpartum. Probably the volume of Metri-Care (10ml) was low for covering all surface of uterus at postpartum period or may be the cows at this period need more than once intrauterine treatment. Therefore injection of two tubes of Metri-Care or increase the volume and also continuing treatment for two days may be better. The result of experiment 2 showed that Metri-Care is more successful than hormonal therapy or PGF2 plus penicillin.

Saboorian *et al* (1997) and Amirids *et al* (2003) used the Diff-Quick (modified Wright) staining method for cytological examination of the endometrial swab. The cytological count of cells showed decrease of inflammation cells at 14 days after treatment in both experiments. The reduced inflammatory cells at experiment 1 were unacceptable for good treatment and that has agreement with result of Thurmond *et al* (1993). Therefore the cows affected to postpartum endometritis at one month after parturition need to continue treatment or hormonal therapy to help uterine defense mechanism. But at groups 55-60 days after parturition we found significant ($P < 0.05$) decrease in the mean (\pm SD) of inflammatory cells especially in the mean of neutrophil. Therefore the treatment with one tube of Metri-Care is enough for treatment of usually mild endometritis at dairy cows.

Cefquinome (fourth-generation cephalosporin) licensed for the treatment of pneumonia and mastitis in cattle (Bishop 2001). Administration of cefquinome in cows with clinical metritis resulted in treatment, as confirmed by clinical, bacteriological and cytological evidence (Amirids *et al* 2003). In a similar study,

carried out in sows, cefquinome was found to be effective in the treatment of puerperal metritis (Heinritzi & Hagn 1999). Cefquinome may be useful in the control of bovine metritis, especially in cases of reduced efficacy of older generation antimicrobial agents (Amiridis *et al* 2003).

LeBlanc *et al* (2002) reported that selected 316 cows with endometritis from 27 farms assigned randomly within herd to receive 500mg of cephalixin benzathine intrauterine (IU), 500µg of cloprostenol IM, or no treatment. Resolution rate of clinical signs 14 days after treatment was 77% and was not affected by treatment. Reproductive performance was monitored for a minimum of 7 month after treatment. Cows with endometritis treated with cephalixin IU had a significantly shorter time to pregnancy than untreated cows (hazard ratio=1.63). There was no difference in pregnancy rate between PGF2α and untreated cows, but the difference in pregnancy rate between cows treated with cephalixin IU and with PGF2α was not statistically significant. Treatment of postpartum endometritis should be reserved for cases which diagnosed after 26 DIM, based on criteria that are associated with subsequent pregnancy rate. McDougall (2002) concluded that intrauterine treatment with 0.5g cephalixin improved reproductive performance of dairy cattle, especially with RFM, a dead calf at calving or within 24h of calving and cows with a vulval discharge. Cephalixin is a first generation cephalosporin; a broad-spectrum antibiotic with bactericidal action against gram-positive and gram-negative bacteria. Cephalixin is resistant to the action of penicillinase and is active in an anaerobic environment such as encountered in an infected uterus (Adams 2001). After a single treatment with that, concentrations of cephalixin in endometrial tissue above the MIC of sensitive bacteria are maintained for at least 24h. The suspension is well tolerated, enables good diffusion of cephalixin into the endometrium and is easily infused. The residue data and use pattern supported a 2 days withholding period for meat and a NIL withholding period for milk (Adams 2001). Therefore, Metri-Care may be good choice for treatment of postpartum uterine infection in dairy cow.

References

- Adams, H.R. (2001). *Veterinary Pharmacology and Therapeutic*. (8th edn). Pp:822-825. Iowa State University Press. Ames.
- Ahmadi, M.R., Nazifi, S. and Khodakaram Tafti, A. (1998). Comparative evaluation of cytological examination of cervical mucosa and endometrial biopsy in cow with endometritis. *Pajouhesh-va-Sazandegi* 36:122-123 (in Persian).
- Ahmadi, M.R., Nazifi, S. and Gaisari, H.R. (2000). Cytology changes in heifer's cervical mucosa at different phases of the oestrus cycle. *14th ICAR*. Stockholm.
- Ahmadi, M.R., Nazifi, S. and Gaisari, H.R. (2002). A new method in diagnosing of postpartum subclinical endometritis in dairy cattle. *Sixth international symposium on reproduction in domestic ruminants*. Scotland, UK.
- Amirids, G.S., Fthenakis, G.C., Dafopoulos, J., Papanikolaou, T. and Mavrogianni, V.S. (2003). Use of cefquinome for prevention and treatment of bovine endometritis. *Journal Veterinary Pharmacology Therapy* 26:387-390.
- Bishop, Y. (2001). *The Veterinary Formulary*, (5th edn). The Pharmaceutical Press, London.
- Borsberry, S., Dobson, H. (1989). Periparturient diseases and their effect on reproductive-performance in 5 dairy herds. *Veterinary Record* 124:217-219.
- Bretzlaff, K. (1987). Rationale for treatment of endometritis in the dairy cow. *Veterinary Clinics of North America. Food Animal Practice*. 3:593-607.
- Bretzlaff, K.N., Ott, R.S., Kortiz, G.D., Bevill, R.F., Gustafsson, B, K. and Davis L.E. (1983). *American Journal of Veterinary Research*. 44:760.
- Bruun, J., Ersboll, A.K. and Alban, L. (2002). Risk factors for metritis in Danish dairy cows. *Preventive Veterinary Medicine*. 54:179-190.
- Cohen, RO., Berntein, M. and Ziv, G. (1995). Isolation and antimicrobial susceptibility of *Actinomyces pyogenes* recovered from the uterus of dairy cows with retained fetal membranes and postparturient endometritis. *Theriogenology* 43:1389.

- de Kruif, A., Gunnink, J.W. and de Bois, C.H. (1982). Diagnosis and treatment of postpartum endometritis in cattle. *Tijdschr Diergeneeskd* 107:717-725.
- El-Azab, M.A., Whitmore, H.L. and Kakoma, I. (1988). Evaluation of the uterine environment in experimental and spontaneous bovine metritis. *Theriogenology* 29:1327-1334.
- Esslemont, R J., Spincer, I. (1993). The incidence and cost of disease in dairy herds. *Dairy report* No 2. Department of Agriculture, University of Reading.
- Heinritz, K., Hagn, J. (1999). Comparison of therapeutic Performance of the new cephalosporin cefquinome with other treatment regimes in gilts with puerperal septicemia and toxemia syndrome. *Tierärztliche Praxis Ausgabe Grosstiere Nutztiere* 27:114-121.
- Hussain, AM. (1989). Bovine uterine defense mechanisms: a review. *Zentralbl Veterinärmedizin B* 36:641-651.
- Kasimanickam, R., Duffield, T.F., Foster, R.A., Gartley, C.J., Laslie, K.E., Walton, J.S. and Johnson, W.H. (2005). The affect of a single administration of cephalosporin of cloprostonol on the reproductive performance of dairy cows with subclinical endometrities. *Theriogenology* 63:818-830.
- LeBlanc, S.J., Duffield, T.F., Leslie, K.E., Bateman, K.G., Keefe, G.P., Walton, J.S. and Johnson, W.H. (2002). The effect of treatment of clinical endometritis on reproductive performance in dairy cows. *Journal Dairy Science* 85:2237-2249.
- Masera, J., Gustafsson, B.K., Afiefy, M.M., Stowe, C.M. and Bergt, G.P. (1980). *Journal of the American Veterinary Medical Association*. 176:1099.
- McDougall, S. (2002). Effect of intrauterine infusion of Cephapirin on reproduction and production of cows with peripartum disease. *XXII World Buiatrics Congress*. 18-23 August, Hanover, Germany.
- Montes, A.J., Pugh, D.G. (1993). Clinical approach to postpartum metritis. *Compendium continue Education. Practice Veterinary* 15:1151-1153.

- Noakes, D.E. (1991). Cattle fertility. *Proceeding of the British cattle veterinary association*. P:47.
- Olson, J.D., Ball, L. and Mortimer, R.G. (1984). Therapy of postpartum uterine infections. *Proceeding. Annual. Mtg. Soc. Theriogenology*. P:170.
- Saboorian, M.H., Ashfaq, R., Vandersteenhoven, J.J. and Schneider, N. (1997). Cytogenetics as an adjunct in establishing a definitive diagnosis of synovial sarcoma by fine-needle aspiration. *Cancer Cytopathology* 81:187–192.
- Sheldon, I.M., Noakes, D.E. (1998). Comparison of three treatments for bovine endometritis. *Veterinary Record* 142:575-579.
- Stevenson, J.S., Call, E.P. (1998). Reproductive disorders in the periparturient dairy cow. *Journal of Dairy Science* 71:2572.
- Thurmond, M.C., Jameson, C.M. and Picanso, J.P. (1993). Effect of intrauterine antimicrobial treatment in reducing calving-to-conception interval in cows with endometritis. *Journal of American Veterinary Medicine Association* 203:1576-1578.
- Vandeplassche, M. (1981). New comparative aspects of involution and puerperal metritis in the mare, cow and sow. *Montash Veterinaermedizin* 36:804.
- Youngquist, R.S. (1997). Current therapy in large animal. In: *Theriogenology*. Pp:335-339. W.B. Saunders Company, Philadelphia.