

CLINICAL, CULTURAL, ENDOCRINE AND THERAPEUTIC EVALUATION OF POSTPARTUM METRITIS IN SURTI BUFFALOES*

R.D. PATIL¹, J.A. PATEL², A.J. DHAMI³ AND N.P. SARVAIYA⁴

Department of Animal Reproduction, Gynaecology and Obstetrics

College of Veterinary Science and Animal Husbandry

Anand Agricultural University, Anand-388 001

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ABSTRACT

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This study was carried out on 40 postpartum metritic Surti buffaloes divided into five groups each of eight animals. The study included use of four treatment lines and one control group, viz., Group I- Metronidazole (500 mg) I/U for 5 days; Group II- Ciprofloxacin (1500 mg) and Tinidazole (1800 mg) I/U for 5 days; Group III- Ciprofloxacin (1500 mg) and Tinidazole (1800 mg) I/U for 5 days plus PGF_{2α} 25 mg i/m once on first day; Group IV- Ceftiofur (long acting) @ 6.6 mg/kg body wt subcutaneously once and Group V (control)- Normal saline I/U for 5 days. The response to these therapeutic regimes was evaluated by comparing the clinical, cultural and endocrine changes before and after 5 days of treatment. The mean rectal temperature recorded after treatment was significantly (P<0.01) lower than the pre-treatment value in all four treatment groups. No significant change was however observed in control group. The mean values of plasma estradiol concentration were significantly (P<0.01) higher and progesterone values lower (P<0.01) on day 6 post-treatment than the corresponding pre-treatment values in all five groups. Microbial examination of uterine discharge revealed that 38 (95.00 %) animals were positive for micro-organisms and 2 (5.00 %) were sterile. Of the 38 samples, 31 (81.58%) had single and 7 (18.42%) had mixed bacterial isolates. Gram-ve bacilli were the predominant organisms (64.52 %) in metritic buffaloes. The therapeutic response achieved by various drugs and combination of drugs assessed by clinical recovery rate of the condition on 6th day post-treatment revealed that postpartum metritic buffaloes under groups I to V had a clinical cure rate of 50.00, 62.50, 87.50, 100.00 and 00.00 per cent, respectively. Clinically, Ceftiofur (long acting) was found to be the most effective drug followed by a combination of other antibiotics plus prostaglandin F_{2α} and antibiotics alone.

Key words: Postpartum metritis, Buffaloes, Antibiotics, Hormones.

INTRODUCTION

Postpartum metritis is clinically characterized by intermittent purulent or muco-purulent vulval discharge, reduced appetite, decrease in milk yield, and

incomplete/delayed uterine involution with varying amount of purulent material. Pathogenesis of metritis has not been fully explained. The endocrinological and immunological disorders, bacterial contamination, insufficient feeding, overfeeding and stress could be the causes of this disorder (Sheldon *et al.*, 2004). All these factors interfere with the contractibility resulting in uterine inertia by which lochia is retained within the uterus beyond normal period providing a good medium for the bacterial growth. Postpartum metritis is usually treated with antibiotics or hormones alone or in combination (Drillich *et al.*, 2001). Antibiotics are typically delivered systemically or can be infused

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¹Research Scholar, ²Associate Professor, ³Professor & Head, Dept. of Animal Reproduction, Gynaecology & Obstetrics, ⁴Associate Research Scientist, Reproductive Biology Research Unit, College of Veterinary Sciences, AAU, Anand 388 001.

Corresponding author, E-mail, ajdhami@aaau.in

directly into the uterine lumen. Success in the treatment of uterine infection depends on evacuation of the uterine discharge, susceptibility of the infectious agents to the drug used, concentration and frequency of drug used and the exposure of the entire endometrium to the drug (Azawi *et al.*, 2008). Despite treatment with variety of intrauterine drugs the fertility was often less than the unaffected animals (Paisley *et al.*, 1986). Perusal of literature revealed meager information on cultural, endocrine and therapeutic aspects of postpartum metritis in buffaloes. Hence, the present study was undertaken for a better understanding of the same and to manage effectively the postpartum metritis in buffaloes.

MATERIALS AND METHODS

This investigation was carried out from October 2009 to April 2010 on 40 postpartum metritic buffaloes selected at random from the villages of College Ambulatory Clinic centers and nearby villages of Anand taluka of Gujarat. The animals were screened thoroughly for their genital health and reproductive status through history and gynecological examination per rectum. The cases were selected only with supportive evidence of genital discharge on per rectal examination of the reproductive tract. Clinical symptoms such as presence of abnormal uterine discharge, decrease in milk yield and feed intake were taken into consideration. The buffaloes so selected were of first 30 days postpartum period. Buffaloes were divided into following five subgroups, each of eight animals and studied for their clinical, cultural and hormonal profile before and after various therapeutic approaches.

Blood samples were collected from all the animals by jugular vein puncture in heparinized vials on day 1 (before treatment) and on day 6th of experiment (i.e. next day after last dose). Before bleeding, rectal temperature of each animal was recorded. The blood samples were centrifuged at 3000 rpm for 10 minutes, and the plasma samples were stored with a drop of 0.01 per cent of merthiolate in 5 ml vials at -20°C till analyzed for hormonal assay.

The levels of plasma progesterone and estradiol were estimated employing standard RIA techniques. Labeled antigen (with I¹²⁵), antibody coated tubes and standards were procured from Immunotech-SA, France. The sensitivity of the progesterone and estradiol assay was 0.1 ng/ml and 4 pg/ml, respectively. Intra-assay coefficients of variation for progesterone and estradiol were 5.4 and 12.1 per cent, while inter-assay variations were 9.1 and 11.2 per cent, respectively.

Samples of uterine discharge were collected aseptically once before treatment by recto-vaginal technique using sterilized glass pipette (10 ml, capacity) inserted in the anterior vagina, the pointed end of which was connected to a syringe with rubber junction. The samples were processed immediately for cultural isolation of organisms on nutrient agar. The isolates were differentiated on the basis of Gram's staining.

The therapeutic response achieved by various drugs and combination of drugs was assessed by clinical recovery of the condition on 6th day post-treatment in view of decrease or absence of uterine discharge, improvement in feeding habit, milk yield and general body condition. Data on rectal temperature and hormonal profiles of animals before and after metritis treatment were compared using paired "t" test.

RESULTS AND DISCUSSION

The average rectal temperature (°F) recorded for different treatment groups of metritic buffaloes revealed that the mean values on day 6 post-treatment were significantly ($P < 0.01$) lower in all 4 treatment groups than the pre-treatment values. No significant difference was however observed in pre- and post-treatment values of control group-V. Among the four treatment groups, the animals of group IV (Ceftiofur) showed good response, whereas the least response was shown by the control group. These findings on clinical response are in agreement with those of Dolezel *et al.* (2005) who observed significant increase in rectal temperature in metritis animals. Pateria *et al.* (1992) also reported significant ($P < 0.05$) reduction in rectal temperature towards normalcy in metritic buffaloes following various treatments.

The mean plasma progesterone (P_4) values obtained on day 6 post-treatment in all the five groups were significantly ($P < 0.01$) lower and estradiol (E_2) higher than the pre-treatment values. Treatment of Ciprofloxacin and Tinidazole Plus Prostaglandin (group-III) gave good response, where the mean pre-treatment values of P_4 and E_2 were 0.65 ± 0.10 ng/ml and 18.11 ± 1.01 pg/ml and the post-treatment values 0.19 ± 0.05 ng/ml and 29.13 ± 1.19 pg/ml, respectively, suggesting luteolysis and follicular growth. The least response was noted in the control group-V.

Pepper and Dobson (1987) reported either unchanged or high progesterone levels in endometritic cows for 5 to 60 days postpartum. Kaczmarowski *et al.* (2006) noted higher progesterone level ($P < 0.05$) in cows with RFM as compared to control group. The present findings of low E_2 in postpartum metritic buffaloes are comparable with the observations of Lohan *et al.* (1987). Guay and Lamothe (1980) observed high serum estrogen level in cows with metritis after treatment with prostaglandin. The higher levels of plasma progesterone in postpartum metritis buffaloes may cause decrease in uterine pH allowing greater bacterial growth, because uterus is highly resistant to infection during estrogenic phase but very susceptible during the period of progesterone dominance (Paisley *et al.*, 1986). Progesterone suppresses uterine immune defense and predisposes the uterus to nonspecific infections.

Out of 40 cervico-uterine discharge samples of metritic buffaloes cultured pre-treatment, 38 (95.00 %) were positive for microorganisms and 2 (5.00 %) were found sterile. The samples contained single and mixed isolates. Out of 40 buffaloes, 31 (81.58 %) had single and 7 (18.42 %) had mixed bacterial isolates. The study showed that gram-ve bacilli were the predominant organisms with 64.52 per cent prevalence.

These findings are in agreement with those of Singh *et al.* (1994) who reported 93.33 % positive samples from postpartum metritic buffaloes. Pateria *et al.* (1989) reported the highest incidence of gram -ve organisms in clinical metritic buffaloes. Sharma *et al.* (2008) reported the incidence of gram +ve and gram -ve isolates as 38.70 and 61.29 % in repeat breeding buffaloes. The development of infection depends upon

the resistance of the individual animal against particular organism and the virulence of the organism. These pathogenic micro-organisms are associated with wide variety of lesions in the reproductive tract causing infertility in dairy animals (Sharma *et al.*, 2008).

The response of postpartum metritic buffaloes to various treatments is presented in Table 3. The animals under treatment groups I, II, III and IV showed a clinical cure rate of 50.00, 62.50, 87.50, and 100.00 per cent, respectively, as evident from cessation of uterine discharge with improvement in feed intake and milk yield, although it was not authenticated by cultural examination of uterine content post-treatment. The animals under control group-V did not show any improvement. Dhama *et al.* (1986) reported that metronidazole was effective in 40.00 per cent of buffaloes with endometritis, probably of anaerobic infections. Sandhu (2006) reported higher cure rate when metronidazole combined with other antibiotics was used for treating various reproductive disorders. The effectiveness of metronidazole in the present study might be due to its effect against anaerobic bacteria and ability to diffuse through deeper tissues.

Dhillon *et al.* (2005) reported that ciprofloxacin and tinidazole combination was effective in 60.00 per cent of repeat breeding crossbred cows, may be due to its efficiency of eliminating both aerobic and anaerobic infection. Azawi *et al.* (2008) and Prabhakar *et al.* (2010) revealed that addition of prostaglandin in the treatment protocol of buffaloes with postpartum metritis proved highly beneficial to clear uterine infections. The beneficial effect of prostaglandin $F_{2\alpha}$ may be attributed to myometrial contraction aiding expulsion of large amount of lochia, the induction of luteolysis if a functional CL is present, direct uterotonic effect, stimulation of phagocytosis by uterine leucocytes, and stimulation of uterine defense mechanism consequent to a fall in the circulating progesterone and a rise in circulating oestrogen (Paisely *et al.*, 1986). The better response (87.5 %) with a combination of $PGF_{2\alpha}$ and ciprofloxacin and tinidazole achieved in the present study (group III) was probably due to synergistic effect of parenteral $PGF_2\alpha$ and local antibiotics therapy.

The present finding of 100 % clinical cure rate of animals under Ceftiofur treatment (group IV) was in agreement with that of Drillich *et al.* (2001) and Chenault *et al.* (2004). They all recorded beneficial effect of ceftiofur HCl in treatment of postpartum metritis. Ceftiofur is third-generation cephalosporin developed exclusively for Veterinary use and is approved for treatment of foot rot and postpartum metritis in cattle (Drillich *et al.*, 2006). The cent per cent recovery observed in present study with Ceftiofur in group IV may be due to maintenance of its prolonged (7 days) therapeutic blood concentration on subcutaneous administration, and that the mean concentration of ceftiofur derivatives exceeds the minimum drug concentrations required to inhibit the growth of 90 per cent of aerobic and anaerobic isolates such as *E. coli*, *Fusobacterium necrophorum*, and *Arcanobacterium pyogenes* (Drillich *et al.*, 2006).

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