

## EFFECT OF ANTIBIOTIC TREATMENT ON PREGNANCY RATE OF REPEAT BREEDER DAIRY CROSS BRED COWS WITH SUB-CLINICAL UTERINE INFECTION

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### ABSTRACT

The objective of this study was to investigate the effect of antibiotic treatment on pregnancy rate in repeat breeder dairy crossbred cows. Based upon culture and sensitivity test, cows were assigned into two groups. Genta (n = 10) cows were treated with gentamycin (100 mg/ml) and ENRO (n = 12) cows were treated with enrofloxacin (100 mg/ml) intramuscularly 4 ml/100 kg body weight and intra-uterine 2-4 ml diluted with 16 ml of physiological salt solution with 24 hours interval for five days in both groups (five infusions per animal on every other day basis). Control (n = 14) cows were untreated control (non-repeat breeder). After treatment, cows were inseminated approximately 12 h after the standing heat. Pregnancy diagnosis was performed by ultrasonography between days 25 and 30 post insemination. The overall pregnancy rates after three services among three groups were significantly different (P<0.05). The overall pregnancy rates after three services of Genta cows (80%) did not differ significantly (P>0.05) from Control cows (93%). However, overall pregnancy rates of Enro cows (33%) were lower (P<0.05) than Genta and Control cows. In conclusion, the pregnancy rates could be improved by treatment with gentamycin compared to enrofloxacin in repeat breeder dairy crossbred cows with sub-clinical uterine infection.

**Key words:** Repeat breeder, crossbred cows, antibiotics, pregnancy rate.

### INTRODUCTION

The repeat breeding cow is one that has clinically normal reproductive tract with normal or nearly normal oestrous cycles and oestrus periods and has been bred two or more times to a fertile bull but failed to conceive (Roberts, 1971). Incidence of repeat breeding in cows in various countries ranged from 10 to 18% (Kimura *et al.*, 1987). Potential causes of repeat breeding include sub-clinical infection of reproductive tract (Rao, 1982), age of the animal (Bartlett *et al.*, 1986), errors in detection of oestrus (DeKriuf, 1978), endocrine dysfunction (Gustafsson *et al.*, 1986), nutritional deficiencies and others (Francos *et al.*, 1977). For the repeat breeder cows suspected to have an infectious etiology, many of the treatments used have been in the form of uterine infusions with a variety of antiseptic and antibiotic solutions. The intrauterine infusions in repeat breeders has proven to be successful (Oxender and Seguin, 1976). Therapeutic use of GnRH and PGF<sub>2α</sub> for repeat breeders has been demonstrated with some improvement in pregnancy rate compared to untreated controls during the last two decades (Peters, 2005; Ahuja *et al.*, 2005).

Purebred dairy cows have been imported since long from western to the tropical or subtropical countries for enhanced milk production. Undoubtedly, their

production is high but adaptability particularly during hot and humid months is compromised. They are more prone to parasitic infestation (Afzal *et al.*, 1986). Similarly, pure or crossbred cows maintained in subtropical environment have more reproductive disorders. In Pakistan, very high incidence (47.5%) of repeat breeding due to uterine infections has been reported in crossbred dairy cows (Kakar *et al.*, 1997). This could be due to the natural service by infected bull, as a sequel of post-partum prolapse, retained fetal membranes, dystokia, abortion, handling of prolonged parturition by the unskilled person, inappropriate insemination and/or improper sterilization of insemination equipment, inflation of air and insertion of tail into vagina and other unexplained reasons.

The objective of the present study was to determine the effect of selective antibiotic treatment of either gentamycin or enrofloxacin on pregnancy rate in repeat breeder dairy crossbred cows.

### MATERIALS AND METHODS

#### Animals and management

The present study was conducted on 36 Holstein Friesian x Sahiwal crossbred cows maintained at the Rakh Dera Chahal, Lahore, Pakistan. Of these, 22 cows had the history of repeat breeding (availed six to eight

services but failed to conceive), whereas other 14 were control. The cows had moderate body condition with body weight ranged from 350 to 450 kg. Parity was from 2 to 6 and the average milk production ranged from 2500 to 3000 liters per lactation. Throughout the experimental period, the cows were maintained as a group and were housed in semi-covered sheds under similar conditions of feeding and management. Each day, cows were fed 30-40 kg of green fodder and 1-2 kg of concentrate mixture containing 15% crude protein and 65% total digestible nutrients.

### Treatments

All the experimental cows were palpated per rectum and scanned by transrectal ultrasonography (Falco Vet 100; Pie Med; Holland) to make sure that the cows were not pregnant and genitalia was normal. Uterine swabs were collected from the cows having the history of repeat breeding (n = 22) for culture and antibiotic sensitivity test under sterile conditions. The antibiotics used in the test were gentamycin, kanamycin, oxytetracycline, penicillin, chloramphenicol, norfloxacin and enrofloxacin. Based upon results of culture and sensitivity test (+++ or more), cows were assigned into two groups. Genta (n = 10) cows were treated with gentamycin (100 mg/ml; Farvet, Italy) intramuscularly at the dose of 4 ml/100 kg body weight and intra-uterine 2-4 ml diluted with 16 ml of physiological saline solution. Enro (n = 12) cows were treated with enrofloxacin (100 mg/ml; Vetycare, Pakistan) intramuscularly at the dose of 4 ml/100 kg body weight and intra-uterine 2-4 ml diluted with 16 ml of physiological saline solution (five infusions per animal, on every other day basis, over a period of 10 days) with 24 hours interval for five days in both groups. Control (n = 14) cows did not receive any treatment and served as control (non-repeat breeder). After the end of antibiotic therapy, both the groups were given sexual rest of at least one month.

**Table 1: Effect of antibiotic treatment on pregnancy rate of repeat breeder dairy crossbred cows with sub-clinical uterine infection**

Group	No. of cows	Pregnancy rates (%)			
		First service	Second service	Third service	Overall
Genta <sup>†</sup>	10	(4/10) 40 <sup>a</sup>	(3/10) 30	(1/10) 10	(8/10) 80 <sup>a</sup>
Enro <sup>††</sup>	12	(1/12) 8 <sup>b</sup>	(2/12) 16	(1/12) 8	(4/12) 33 <sup>b</sup>
Control <sup>†††</sup>	14	(7/14) 50 <sup>a</sup>	(2/14) 14	(4/14) 29	(13/14) 93 <sup>a</sup>

Values within columns with different letters are significantly different (P<0.05).

<sup>†</sup> Gentamycin (100 mg/ml; Farvet) intramuscularly 4 ml/100 kg body weight and intra-uterine 2-4 ml diluted with 16 ml of physiological salt solution.

<sup>††</sup> Enrofloxacin (100 mg/ml; Vetycare) intramuscularly 4 ml/100 kg body weight and intra-uterine 2-4 ml diluted with 16 ml of physiological salt solution.

<sup>†††</sup> Control cows were untreated.

### Induction of estrus

At the end of sexual rest, cows were synchronized with two injections of 0.150 mg/2 ml of PGF<sub>2α</sub> (Dalmazin™ Fatro Co. Italy) i.m; 12 h apart after the detection of corpus luteum by transrectal ultrasonography. Cows were observed for oestrus, two times a day using a teaser bull. Cows in estrus were inseminated approximately 12 h after the standing heat using frozen thawed semen. The cows were inseminated by a single technician. Pregnancy diagnosis was performed using ultrasonography between days 25 and 30 post insemination. The cows which returned to estrus were again inseminated (second service), and checked for pregnancy. Similarly cows that returned to estrus twice were inseminated (third service) and checked for pregnancy accordingly.

### Statistical analysis

The differences in pregnancy rates among cows of three groups were compared for the first, second, third service and overall pregnancy rate using Pearson  $\chi^2$  test (SPSS version 10.0). The difference between two proportions (each of two groups) was compared using Z-test (PH Stat 2, Microsoft Excel 2002).

## RESULTS

The pregnancy rates in repeat breeder dairy crossbred cows are presented in Table 1. Pregnancy rates for first service among three groups tended to be different (P<0.05). The pregnancy rate of first service for Genta cows (40%) did not differ significantly (P>0.05) with Control cows (50%). However, pregnancy rates of first service for Enro cows (8%) was lower (P<0.05) than Control and Genta cows. The overall pregnancy rates among three groups were significantly different (P<0.05). The overall pregnancy rate of Genta cows (80%) did not differ significantly (P>0.05) with Control cows (93%). However, overall pregnancy rates of Enro cows (33%) were lower (P<0.05) than Genta and Control cows. The overall pregnancy rate of both the treated groups was 55%.

## DISCUSSION

In the present study, the overall pregnancy rates of Genta cows (80%) did not differ significantly with Control cows (93%) but were better than Enro cows (33%). The overall pregnancy rate of both the treated groups was 55%. LeBlanc *et al.* (2002) administered cephalixin benzathine by intrauterine route, and obtained improved fertility ( $P < 0.05$ ) over controls in endometritic cows. On the other hand, gentamycin sulphate administered by intrauterine infusion at the rate of 200 mg, 10 minutes following first service insemination in dairy cows did not enhance fertility (Daniels *et al.*, 1976). In the present study, higher overall pregnancy rates in Genta compared to Enro cows might be due to differences in the pharmacokinetic characteristics of two antibiotics. For example, half-life ( $t_{1/2}$ ) and bioavailability have been reported to be higher in gentamycin and were 45 hours and 92% (Haddad *et al.*, 1987) versus 6 hours and 82% respectively for enrofloxacin (Kaartinen *et al.*, 1995). Gentamycin when administered intrauterine attained maximum plasma concentration within 30 minutes (Al-Guedawy *et al.*, 1983). These findings indicate that physiochemical characteristics of gentamycin are better compared to enrofloxacin in these repeat breeder cows.

In conclusion, overall pregnancy rates in repeat breeder dairy crossbred cows with sub-clinical uterine infection were improved by the treatment of antibiotics along with sexual rest up to 55% after three services. Furthermore, pregnancy rate of gentamycin was significantly higher compared to enrofloxacin in repeat breeder dairy crossbred cows.

## REFERENCES

- Afzal, M., R. M. Nagra, M. Hussain and A. S. Akhtar, 1986. Health problems in pure-bred exotic and crossbred dairy cattle. National Workshop on Dairy Cattle Crossbreeding and Maintenance of Exotic Dairy Cattle in Pakistan. Pakistan Agri. Res. Council, Islamabad, Pakistan.
- Al-Guedawy, S. S., C. A. Neff-Davis, L. E. Davis, H. L. Whitmore and B. K. Gustafsson, 1983. Disposition of gentamycin in the genital tract of cows. *J. Vet. Pharmacol. Therap.*, 6: 85-92.
- Ahuja, C., F. Montiel, R. Canseco, E. Silva and G. Mapes, 2005. Pregnancy rate following GnRH + PGF<sub>2α</sub> treatment of low body condition, anestrous *Bos taurus* by *Bos indicus* crossbred cows during the summer months in a tropical environment. *Anim. Reprod. Sci.*, 63: 202-213.
- Bartlett, P. C., J. H. Kirk and E. C. Mather, 1986. Repeated insemination in Michigan Holstein Friesian cattle: Incidence, descriptive epidemiology and estimated economic impact. *Theriogenology*, 26: 309-322.
- Daniels, W. H., D. A. Morrow, B. W. Pickett and L. Ball, 1976. Effects of intrauterine infusion of gentamycin sulfate on bovine fertility. *Theriogenology*, 6: 61-64.
- Dekriuf, A., 1978. Factors influencing the fertility of a cattle population. *J. Reprod. Fert.*, 54: 507-518.
- Francos, G., M. Davidson and E. Mayer, 1977. The influence of some nutritional factors on the incidence of the repeat breeder syndrome in high producing herds. *Theriogenology*, 7: 105-111.
- Gustafsson, H., K. Larsson, H. Kindahl and A. Madej, 1986. Sequential endocrine changes and behaviour during estrus and metoestrus in repeat breeder and virgin estrus and metoestrus heifers. *Anim. Reprod. Sci.*, 10: 261-273.
- Haddad, N. S., W. R. Ravis, W. M. Pedersoli and R. L. J. R. Carson, 1987. Pharmacokinetics and residues of gentamycin in lactating cows after multiple intramuscular doses are administered. *Amer. J. Vet. Res.*, 48: 21-27.
- Kaartinen, L., M. Salonen, L. Älli and S. Pyörälä, 1995. Pharmacokinetics of enrofloxacin after single intravenous, intramuscular and subcutaneous injections in cows. *J. Vet. Pharmacol. Therap.*, 18: 357-362.
- Kakar, M. A., G. Rasool and F. Ahmad, 1997. Clinical incidence of reproductive disorders in crossbred cows in the province of Balochistan. *Pakistan Vet. J.*, 17: 9-12.
- Kimura, M., T. Nakao, M. Moriyoshi and K. Kawata, 1987. Luteal phase deficiency as a possible cause of repeat breeding in dairy cows. *British Vet. J.*, 143: 560-566.
- LeBlanc, S. J., T. F. Duffield, K. E. Leslie, K. G. Bateman, G. P. Keefe, J. S. Walton and W. H. Johnson, 2002. The effect of treatment of clinical endometritis on reproductive performance in dairy cows. *J. Dairy Sci.*, 85: 2237-2249.
- Oxender, W. D. and B. E. Seguin, 1976. Bovine intrauterine therapy. *J. Amer. Vet. Med. Assoc.*, 168: 217-219.
- Peters, A. R., 2005. Veterinary clinical application of GnRH: questions of efficacy. *Anim. Reprod. Sci.*, 30: 1-13.
- Rao, A. V. N., 1982. Causes and incidence of reproductive disorders among Zebu x Taurus crossbred cows in Andhra Pradesh. *Theriogenology*, 17: 189-191.
- Roberts, S. J., 1971. *Veterinary Obstetrics and Genital Diseases (Theriogenology)*. Published by the Author, Ithaca, New York, USA, pp: 496-506.