Fertility Improvement by Ovsynch Protocol in Repeat Breeder Cattle of Kathmandu Valley

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Abstract
Repeat breeders cause substantial loss in dairy herds mainly affecting the reproductive efficiency and economy of milk production in cattle and buffaloes. A study was conducted in cattle farms of various farmers of three districts of Central Region of Nepal; Kathmandu, Bhaktapur and Lalitpur during March-August 2016. A total of 20 repeat breeder cows were selected for the trial. Repeat breeder cows were identified based on the history of at least three repeated breeding at an interval of around three weeks. We applied Ovsynch protocol where two reproductive hormones; Gonadotropin Releasing Hormone (GnRH) and Prostaglandin (PGF2α) were injected. Pregnancy was initially diagnosed by Biopryn Test kit using serum samples. Rectal examination confirmed pregnancy after 2 months of artificial insemination (AI). 16 cows expressed clear estrus signs during fixed time AI. 5 (83.3%) out of 6 cows with cystic ovary expressed estrus at the time of fixed time AI. 4 (66.7%) of these cows became pregnant.

Keywords: Repeat breeders; Ovsynch protocol; GnRH; PGF2α; Biopryn test; Fixed time AI

Introduction
Repeat breeding is the most frequently encountered but poorly understood condition affecting the reproductive efficiency and economy of milk production in cattle and buffaloes. Reproductive inefficiency of cattle due to repeat breeding syndrome is an expensive hitch in profitable dairy production as the age at first calving in heifers is delayed and the inter-calving interval is extended, thus leading to lowering of calf crop (Thakur et al., 2006). This syndrome can be one of the more frustrating problems affecting reproductive management of a dairy herd. Commonly, herds with normal conception rates range between 65 and 70 percent for lactating cows in Nepal (Sankhi, 1993). Increase in inter calving interval due to repeat breeding means increase in dry period. The ultimate result is tremendous loss due to loss of milk yield as the daily milk yield of local cattle is 2.5 liters per cow, whereas that of improved cattle is 7.64 liters per cow (Sapkota, 1999). Thus, better plan of nutrition, strategic parasitic control program, improved husbandry and proper treatment regime against infectious form of infertility play key role to minimize economic loss due to infertility. About 40.1% of total causes
of repeat breeding are contributed by hormonal insufficiency and dysfunctions (Maurer et al., 1985).

Repeat breeders are likely to have delayed ovulation and an extended follicular phase which allows follicle development but postpones the Luteinizing Hormone (LH) surge (Bage 2002; Bhupender et al., 2005). The delayed LH surge produces relatively aged follicles and oocytes by the time of ovulation. Many specific and non-specific uterine infections are associated with either fertilization failure or early embryonic mortality in repeat breeder cattle. Non-specific infections of the genital tract invariably need some predisposing factor and generally involve an individual animal (Singh and Pant, 1998).

Similarly failure in fertilization may also occur due to failure in heat detection and estimation of appropriate time to perform AI. If the causes of repeat breeding are delayed ovulation, poor heat detection or improper timing of AI, the fixed time AI with suitable hormonal protocol may improve the conception rate in repeat breeder cows. In this study, we hypothesized that the Ovsynch protocol; a novel hormonal approach that follows the fixed time AI may eliminate this problem through estrus synchronization. Therefore, the objective of this study was to determine the effectiveness of Ovsynch protocol in improving fertility of repeat breeder cows in Kathmandu valley.

Methodology

Study Area
A study was conducted in cattle farms of various farmers of three districts of Central Region of Nepal; Kathmandu, Bhaktapur and Lalitpur during March-August 2016. The repeat breeders were purposively selected from Manohara-Kageswori Municipality, Kadaghari, Sanothimi, Gatthaghar, Ramkot, Sanepa areas.

Animals
From the selected areas, total 37 repeat breeder cows were examined clinically. Out of these 37 cows, a total of 20 repeat breeder cows were selected for the trial based on the following criteria. Cattle that did not meet the criteria and having poor body condition were excluded from the study. The limited time frame and financial aspect hindered taking more samples.

Selection Criteria of Repeat Breeding Cows
1. Cows with normal anatomical condition of reproductive organs.
2. Cows with second or more parity.
3. Cows which were less than 10 years of age.
4. Cows which showed negative response towards antibiotics and nutritional treatments.

Examination of Repeat Breeding Cows
Repeat breeder cows were identified based on the history of at least three repeated breeding at an interval of around three weeks. Rectal Palpation Technique was applied for clinical examination of Repeat Breeding cows. The suspected repeat breeders were examined for any abnormalities like cystic ovary, persistence of Corpus Luteum (CL). The animals were categorized on the basis of body condition score (BCS) in Holstein cows given by Ferguson et al., (1994). BCS above 3 were selected for the trial.

Treatment Protocol
This research followed the Ovsynch Protocol (Fig. 1) where two reproductive hormones; GnRH and PGF2α were needed. Treatment protocol has been shown in Fig. 1. All 20 cows were injected with GnRH analogue Buserelin acetate (Receptal VET, Intervet India Pvt. Ltd.) 10 µg intramuscularly on day 0 (start of the treatment protocol) and day 9. On day 7, dinoprost tromethamine (Lutalyse, Zoetis, USA) 25 mg was injected intramuscularly.

Artificial Insemination
AI was done on the 10th day or after 16-20 hours of administration of second dose of GnRH. A good quality semen of Holstein Friesian, Jersey as per the breed manufactured by National Livestock Breeding Centre, Lampatan, Pokhara was used. Skilled veterinarians performed fixed-timed AI in the hormone treated cows.

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Fig. 1: Ovysynch Protocol
Result and Discussion

Out of 20 cows treated, 16 cows (80%) expressed clear estrus signs at the time of fixed time AI. Of 20 cows, total 11 (55%) cows were positive for BioPRYN test on 30+-2 days after AI (Fig. 2). All four cows not detected in estrus at the time of fixed time AI were negative for BioPRYN test. Transrectal palpation at 60 days after fixed time AI confirmed that all 11 cows positive for BioPRYN test were pregnant, and all negative for BioPRYN test were non-pregnant. Thus, BioPRYN test (Fig. 3) had 100% accuracy in pregnancy diagnosis. Of these 20 repeat breeder cows, 6 cows (30%) had cystic ovaries. Five (83.3%) out of 6 cows with cystic ovary expressed estrus at the time of fixed time AI. Four (66.7%) of the cows having cystic ovaries became pregnant.

The hormonal treatment responds only to those repeat breeders that come in cyclic heat. Four repeat breeders of our study might not have come into cyclic heat thus not responding to the hormonal therapy. Administration of GnRH facilitates the initiation of the new estrus cycle through ovulation. Drug (hormone) response is also associated with plane of nutrition and management of animals.

Administration of prostaglandin helps in the lysis of CL present in ovary. One cow didn’t exhibit estrus response to this protocol because of cystic ovary or alteration in the hormonal metabolism might be the cause. Our study found higher conception rate than that of Sankhi (1996) which might be due to the double use of Gonadorelin and additional administration of prostaglandin. The conception rate for the cystic ovary cases were also found higher (67%) than as estimated by Sankhi (1996). The conception rate of repeat breeder cattle after treating with Gonadorelin by Sankhi (1996) was 46%.

The conception rate for the cows having cystic ovary was found to be 67% which determines the higher efficacy of this protocol for cystic ovary case of repeat breeders. The conception rate from the study is 55% which is slightly higher than that of Pursley et al., (1998) and Stevenson et al., (1999). They had estimated the conception rate of this protocol be 30-40%. This might be due to the selection of the repeat breeders that failed to show response towards other treatment.

The overall pregnancy rates of 55% in our observation was higher than that of El-Zarkouny (2010) who studied in heifers treated with Ovsynch (35.1%, n = 77). The lower ovulatory response to the first GnRH injection and other alterations in the follicular dynamics of the heifers may contribute to lower conception rate. DeJarnette et al., (2001) recorded that 20% of Ovsynch-treated cows came into premature estrus (48 hours after PGF2α) and thus estrus detection during this period enhances success of fertilization. Our observation corroborates his study as 16 out of 20 cows exhibited behavioral estrus signs. Estrus detection after PGF2α injection for 48 h is considered an acceptable practice to maximize pregnancy rates before the second GnRH injection with the Ovsynch protocol in dairy heifers (Stevenson et al., 1999).

In the experiment by Howard et al., (2007) using antiserum against PSPB for the detection of pregnancy in cattle, BioPRYN showed that 51.9% were pregnant at 30-36 days. In our study, the BioPRYN had 100% accuracy for pregnancy diagnosis. However, previous published reports on the accuracy of pregnancy diagnosis using BioPRYN are still scarce.
Conclusion

Ovsynch protocol was effective to improve fertility of repeat breeder cows resulting in 55% pregnancy rate. This protocol was also very effective for the treatment of cystic ovarian disease where 66.7% pregnancy rate was achieved. Finally, BioPRYN test had 100% accuracy in pregnancy diagnosis.

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References


