Conception rate in Repeat Breeder Cows following Multiple Artificial Inseminations during Spontaneous Estrus

S. Kantharaj[1], V. Chandrashekara Murthy[2], Suresh S. Honnappagol[3], V. Girish Kumar[4], S.G. Ramachandra[5] and Lakshmikanth. T.R.[6]

[1] *Ph. D Scholar, Veterinary Gynaecology and Obstetrics, Veterinary College, Bangalore*
[2] *Professor, Veterinary Gynaecology and Obstetrics, Veterinary College, Bangalore*
[3] *Animal Husbandry Commissioner, Government of India*
[4] *Professor and Head, Veterinary Biochemistry, Veterinary College, Bangalore*
[5] *Chief Research Scientist, Central Animal Facility, Indian Institute of Science, Bangalore*
[6] *Ph. D Scholar, Veterinary Gynaecology and Obstetrics, Veterinary College*

Abstract

The present study was aimed to determine whether the application of multiple A.I with frozen-thawed semen of good fertility during spontaneous estrus would improve pregnancy rates in repeat breeder cows. Twenty two repeat breeder cows were randomly allotted to two different treatments. Group-I (control) consisted of 11 repeat breeder cows which were done single A.I. during the observed estrus. Group-II (treatment) consisted of 11 repeat breeder cows which were subjected to multiple A.I during the observed estrus until the estrus signs subside. Analysis of the data revealed that there was no significant differences (P>0.05) in the mean serum progesterone concentrations between the two groups at the time of A.I. All the repeat breeder cows in the two groups were found negative for subclinical endometritis. The conception rates obtained were 18.18 and 27.27 per cent in Group-I and Group-II repeat breeder cows, respectively. There was a significant (P<0.05) difference in the conception rate between the two groups. It can be concluded that multiple A.I could be implemented in repeat breeder cows with appreciable conception rates without incurring any financial constraints to the poor farmers.

I. INTRODUCTION

A repeat breeder cow is one that has normal or nearly normal estrous cycles and estrus periods with no palpable abnormalities of the reproductive tract and has been bred thrice or more times to a fertile bull, yet failed to conceive (Roberts, 1971). Repeat breeding syndrome is one of the frustrating gynaecological maladies of crossbred cows, leading to infertility and affects the reproductive efficiency and economy of milk production in dairy animals.

Anovulation and delayed ovulation are the major cause of repeat breeding in dairy cows and remain undiagnosed unless serious attention is given through repeated rectal examination. When delayed ovulation occurs, the poor tubal milieu and altered contractility compromises sperm survival and impairs proper gamete transport. The reproductive failure due to ovulatory disturbances is primarily due to the deficiency of LH secretion at appropriate time after estrus in cows (Khanna and Sharma, 1992). Asynchrony in timing of insemination with respect to ovulation results in low pregnancy rate due to fertilization failure (Hunter, 1994). Therefore the importance of insemination timing with respect to ovulation has repeatedly been emphasized for ensuing fertilization (Martinez, 2001). Hence, the success of insemination depends not merely on the detection of estrus, but also on the timing of ovulation related to insemination. Therefore, single insemination following AM-PM rule may lead to poor conception due

to shortage of motile/fertile spermatozoa. This problem might be alleviated if high numbers of fertilizable spermatozoa are available at ovulation.

Therefore, the present study aimed to determine whether the application of multiple A.I with frozen-thawed semen of good fertility during spontaneous estrus would improve pregnancy rates in repeat breeder cows.

II. MATERIALS AND METHODS

The study was carried out on 22 randomly selected repeat breeder cows aged 3-8 years in their first to fourth lactation with Body Condition Score (BCS) between 2.5 and 3.5. The total number of services for repeat breeder cows in the present study ranged from 3-7 with an average number of 4.5 services. The number of days these animals were not pregnant at the time of presentation for treatment ranged from 150 to 240 days.

These 22 repeat breeder cows were randomly divided into two groups with 11 cows in each group. Group-I (Control) repeat breeder cows were subjected to single A.I during the observed estrus. Group-II repeat breeder cows were subjected to multiple A.I during the observed estrus until the estrus signs subside.

Blood samples were collected from all the two groups for estimation of serum progesterone at the time of A.I. Endometrial cytology was performed in all the repeat breeder cows before A.I. by uterine lavage technique. Repeat breeder cows in the two groups were subjected to pregnancy diagnosis by trans-rectal ultrasound scanning using linear array probe with 7.5 MHz frequency on day 30 post A.I. and confirmed by rectal examination at day 45 post A.I. to evaluate the conception rate.

Mean values (± SE) for serum progesterone levels for repeat breeder cows of two groups were computed and analyzed statistically using one way analysis of variance. The conception rates for the two groups were analyzed by Chi-square test. Results were considered to be statistically significant when "P" values are less than 0.05 ($P < 0.05$).

III. RESULTS AND DISCUSSION

Mean serum progesterone concentrations at A.I:

The mean serum progesterone concentrations at A.I. in the present study were 0.61 ± 0.06 ng/ml (Group-I) and 0.54 ± 0.07 ng/ml (Group-II) (Table 1). Analysis of the data revealed that there was no significant differences ($P>0.05$) in the mean serum progesterone concentrations between the groups at the time of A.I. This is in congruence with the findings of Ravikumar *et al*. (2014) who also reported no significant differences in serum progesterone concentrations on the day of AI in the untreated and treated repeat breeder cows. Further, the serum progesterone concentrations in every animal assigned to untreated and treated repeat breeder groups was less than 1 ng/ml in the present study confirming the complete luteolysis and the onset of estrus.

The mean serum progesterone concentrations in the repeat breeder cows of the two groups at AI were in close agreement with the reported values of 0.50 ± 0.20 ng/ml (El-Zarkouny *et al*., 2004) in dairy cows and 0.67 ± 0.08 ng/ml (Ravikumar *et al*., 2014) in repeat breeder cows at the time of AI.

Wiltbank *et al*. (2012) reported that high circulating concentrations of progesterone near AI has been shown to be detrimental to fertility in dairy cattle, but the underlying physiological mechanisms

that reduce fertility are not well understood. Near the time of AI, it is critical that progesterone concentrations are below a critical value, which appears to be about 0.4 ng/ml.

Supra basal progesterone levels around estrus have been described in repeat breeder cows (Perez-Marín and España, 2007). It may cause incomplete maturation of preovulatory follicles and subsequent final development in an elevated progesterone environment (Duchens *et al*., 1996; Moreira *et al*., 2000).

Table: 1 Mean serum progesterone concentrations and conception rate in control and treated repeat breeder cows

Groups	Mean Serum Progesterone Concentration (ng/ml) at the time of A.I	Conception rate (%)
Group-I (Single A.I) n= 11	0.61 ± 0.06	18.18[b] (2/11)
Group-II (Multiple A.I.) n=11	0.54 ± 0.07	27.27[a] (3/11)

The values with different superscript in column differs significantly (P< 0.05)

Under normal physiological situations, low progesterone concentrations during oestrus results in increased tubal spontaneous contractility, whereas a decrease in tubal spontaneous contractility is related to increasing progesterone concentrations during the luteal phase (Bennett *et al.* 1988). Moreover, suprabasal progesterone concentrations may lead to incomplete maturation of the preovulatory follicle (Duchens *et al*., 1996). It is possible that the suprabasal progesterone concentrations (0.61± 0.06 ng/ml) as recorded in repeat breeder cows in Group-I (Single A.I) at the time of AI might have led to incomplete maturation of preovulatory follicle, reduced tubal contractility, resulting in an impaired or delayed sperm transport from the sperm reservoir to the site of fertilization. Further, delay in sperm transport may contribute to substantial sperm death due to membrane and acrosome disruption impairing fertilization (Salisbury and Flerchinger, 1967). This delay could also contribute to polyspermy by ageing of the oocytes before they encounter the spermatozoa (Hunter, 1994). In either case, the resulting zygotes would have poor developing capacity and would eventually undergo early embryonic mortality. These reasons may be discounted for conception failure (Group-I) in repeat breeder cows with suprabasal concentrations of progesterone at AI in the present study.

Endometrial cytology:

None of the 22 repeat breeder cows were found positive for subclinical endometritis by lavage technique at the time of A.I.

Conception rate:

The conception rate obtained were 18.18 and 27.27 per cent for Group-I Group-II repeat breeder cows, respectively (Table: 1). Analysis of the data revealed that there was a significant difference in the conception rate (P<0.05) between the groups.

The conception rate (18.18%) obtained for untreated repeat breeder cows (Group-I) is in agreement with the value of 25 per cent (Vijayarajan and Meenakshisundaram, 2013) reported for untreated repeat breeder cows. The low conception rate of 18.18 per cent obtained for the untreated repeat breeder cows (Group-I) could be due to varying age of the cows, parity and the suprabasal levels of progesterone (0.61 ± 0.06 ng/ml) recorded in this group.

Three out of 11 repeat breeder cows in Group-II became pregnant with a conception rate of 27.27 per cent. In previous studies, Sowmya (2013) and Kumar *et al*. (2014) reported 30 % and 30 % conception rates, respectively in repeat breeder cows which are slightly higher than the observations made in the present study. However, Shukla and Misra (2008) recorded a very low conception rate of 13.33 per cent only when repeat breeding crossbred cows were re-inseminated after 8 to 16 h of the first AI.

A significant ($P<0.05$) conception rate of 27.27 per cent was obtained for repeat breeder cows subjected to multiple A.I than when compared to 18.18 per cent obtained for repeat breeder cows subjected to single A.I. These observations suggest that insemination closer to ovulation may provide more number of spermatozoa with fertilizing capacity at the fertilization site leading to improved conception rates. Improvement of conception rate following multiple AI in repeat breeder cows should therefore not be disregarded.

IV. CONCLUSIONS

It can be concluded that acceptable conception rates could be obtained following multiple artificial inseminations during the observed estrus without much financial constraints to the poor farmers. Further, such an attempt would reduce the number of days open and enable them to conceive at an earlier. This would improve the milk yield of the repeat breeder cows and enhance the economic status of the farmers.

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