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Effect of different methods of artificial insemination with chilled semen on conception rate in Mandya ewes

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Abstract

Present study was conducted to ascertain the efficiency of different methods of artificial insemination on conception rate in Mandya ewes at Livestock Research and Information Centre (Sheep), Nagamangala, between April and May 2019. A total of 48 healthy ewes were selected and were synchronized for estrus using intravaginal progesterone sponge containing 350 mg of natural progesterone (Avikesil-S®) for 12 days followed by injection of 125 µg cloprostenol sodium (Estrumate®) and 200 IU eCG (Folligon®) intramuscularly on the day of sponge removal. Ewes were divided into four groups with 12 animals in each group. Fixed time artificial insemination (FTAI) was carried out (48-56 hrs) using chilled semen with 200 million spermatozoa by vaginal, cervical and intra-cervical methods in group I, II and III, respectively. The animals of Group IV were allowed for natural mating by the rams. Conception rates of 25, 41.66, 58.33 and 66.66 per cent were recorded in groups I, II, III and IV, respectively. Significantly higher conception rates were recorded in group IV compared to Group I, II and III. However, conception rate in group II and II were not significantly different. Lower conception rate in group I compared group IV ewes might be due to handling stress, type of semen used, dose of semen used, inherent variation in ewes and rams and other factors like experience of inseminator and nutritional status. From the study it is concluded the artificial insemination in sheep can be effectively practiced using appropriate method of insemination by avoiding possible stress factors that influence the conception rate in Mandya ewes.

Keywords: Mandya ewes, artificial insemination, conception rate and spermatozoa

Introduction

Sheep with its wide variety of products like milk, meat, skin, wool and manure is an important contributor to the rural economy of India particularly in the arid, semi-arid and mountainous areas of the country. Sheep rearing is highly neglected sector owing to the low reproductive efficiency^[1], caused by unfavourable environmental and managerial conditions. Sheep population in India is approximately 74.26 million (20th livestock census, 2017) and India ranks 3rd in sheep population, 7th in mutton export and 9th in wool production. Sheep are short-day polyestrous breeders and estrus occurs in response to shortening day length. Their ovulatory season tends to be the autumn and early winter months and anovulatory season will be during late winter, spring, and early summer months with the transition season in the late summer months. Tremendous breed-to-breed variation in prolificacy and length of the ovulatory season also exist in sheep. There is an imbalance in terms of population and production owing to low genetic profile of indigenous breeds, poor nutrition, harsh climatic conditions and poor herd management. This demands a simple, feasible and economically viable assisted reproductive technique/technology (ART) like estrus synchronization and artificial insemination (AI) at field level to improve the reproductive efficiency in sheep. However, the morphological complexity of sheep cervix has made the artificial insemination procedures more difficult and resulted in limited success in sheep. Further, ram spermatozoa are more sensitive to freezing temperature than the gametes of other mammalian species and therefore, the artificial insemination in sheep is restricted to fresh semen samples only. A cheap and effective insemination technique that is easily applicable under field conditions by the farmers themselves has to be established in order to enhance the success of AI in sheep. Therefore, the present study was aimed at ascertaining the effects of different methods of artificial insemination with chilled semen on conception rate in Mandya Ewes

Materials and Methods

Healthy Mandya rams, maintained at Small Ruminant Semen Station, Hebbal, Bengaluru, were used for semen collection by artificial vagina method. Collected semen sample were subjected to spectrophotometer analysis. The initial semen sample were diluted with appropriate volume of TRIS-based extender TRIS (3.32 gm), citric acid (1.737 gm), fructose-D (0.90 gm), egg yolk (10 mL), media (100 mL) to get final concentration (200 and 100 million) of spermatozoa. Diluted semen sample was kept at 5°C for 4 hrs. Chilled semen was carried in cooling container for insemination of different groups of ewes.

The present study was conducted from April 2019 to July 2019. Total of 48 healthy Mandya ewes aged between two to five years, lambed at least once and maintained under uniform managemental conditions at Livestock Research and Information Centre (Sheep) and Instructional Livestock Form Complex, Hebbal, Bengaluru Nagamangala, were selected for the study. These ewes were subjected for 12 days progesterone treatment. On day one progesterone sponge containing 350 mg of natural progesterone (Avikesil-S¹) was introduced intravaginally in sterile condition using a plunger into the anterior vagina and left *in situ* for 12 days. On the day of sponge removal ewes were injected with 125 µg of cloprostenol sodium (Estrumate²) and 200 IU of eCG (Folligon³) intramuscularly. The ewes after estrus synchronization were divided into four groups with six animals in each group and were subjected to fixed time artificial insemination (48 to 56 hrs) using chilled semen (200 million spermatozoa, 0.50 mL) with the help of AI sheath attached to a tuberculin syringe.

In Group I ewes, the semen was deposited in anterior vagina per the procedure of Young Quist and Threlfall (2007)^[2], in Group II ewes the semen was deposited on the external os of cervix as per the procedure described by Donovan *et al.* (2004)^[3] and in Group III ewes the semen was deposited in the body of cervix as per the procedure described by Faigl *et al.* (2012)^[4]. Group IV Ewes were allowed for natural pen / hand mating after synchronisation and they constituted control group. Ewes in all the groups were subjected for pregnancy diagnosis 30 days post insemination and further confirmed on 45th day by using real time B-mode (Easi-scan⁴). The sponge retention rate and estrus response were expressed as percentage. The intensity of estrus and conception rates for different methods of AI and different doses of semen were expressed as percentage and was subjected to chi-square test and the level of significance was considered at $p \leq 0.05$ ^[5].

Results and Discussion

Significantly ($P < 0.05$) higher conception rate was recorded in group IV ewes compared to group I ewes, however, there was

no significant ($P > 0.05$) difference in conception rate among group II and Group IV ewes (Table 1. and Figure 1).

In the present study, vaginal insemination resulted in 25 per cent of conception rate. Almost similar conception rates of 31 and 35 per cent were reported by Anel *et al.* (2005)^[6] and Olivera-Muzantea *et al.* (2011)^[7], respectively using chilled semen. Cervical insemination resulting in 41.66 per cent of conception rate in the study was in accordance with the reports of Menchaca and Rubianes (2004)^[8], and Vishal *et al.* (2015)^[9] who also recorded conception rates of 49.50 and 47.62 per cent, respectively. The low conception rate in the present study in group II and II might be due to heat stress^[6] and nutritional or handling stress^[10] to the ewes as the study was conducted in the between April and May months. In India, nutritionally stressed ewes experience more heat stress than ewes maintained under nutritionally adequate diets^[11]. In tropical sheep, although ewes are year-round breeders, they experience a reduction in sexual activity during summer periods, irrespective of nutritional status of the animal^[12]. Intra-cervical insemination in the present study yielded 58.33 per cent conception rate and was in agreement with Shackell *et al.* (1990)^[13], Aguer *et al.* (1992)^[14] and Maclas *et al.* (2017)^[15] who reported conception rate of 61, 62 and 59.40 per cent, respectively. Andersen *et al.* (1973)^[16] reported conception rate of 70 to 75 per cent and Meghan *et al.* (2002)^[17] reported pregnancy rate of 66.60 per cent using fresh diluted semen containing 200 to 300 million spermatozoa via intra cervical method of insemination. In ewes of IV group the resultant conception rate was 66.66 per cent. Similar conception rate was also reported by Vinales *et al.*, 2001^[18] (63 per cent), Menchaca and Rubianes 2004^[8] (68.80 per cent) and Amer and Hazzaa 2009 (66.70 per cent)^[19]. However, Ataman *et al.* (2006)^[20] has recorded conception rate of 86 per cent and 76 per cent in breeding and non-breeding season using long term FGA, which is higher than the present study (Table 1 and Fig. 1). The variations in the conception rates obtained by various authors in relation to the results of the present study may be due to differences in method of estrus synchronization/natural estrus, season of the study^[21], the breed of the ewes^[22] and type of semen used^[23], which are the factors affecting the fertility rate in sheep. The low conception rates were recorded in vaginal insemination may be attributed to a great number of the spermatozoa being lost by drainage and phagocytosis and disintegration to a certain extent^[16]. The improved fertility in cervical and intra-cervical insemination compared to vaginal insemination might be due to an increased number of sperms reaching the internal genitalia and in turn the site of fertilization due to deposition of semen in external cervical os and in the folds of cervix^[16].

Table 1: Relative conception rates in Mandya ewes inseminated with 200 million spermatozoa using different methods of artificial insemination

Sl. No.	Method of insemination	No. of animals inseminated	No. of animals conceived	Conception rate (%)
1	Group I (Vaginal)	12	3	25.00 ^a
2	Group II (Cervical)	12	5	41.66 ^{ab}
3	Group III (Intra-cervical)	12	7	58.33 ^{ab}
4	Group IV (Control)	12	8	66.66 ^b

The values with in a column with different superscripts differ significantly ($P < 0.05$)

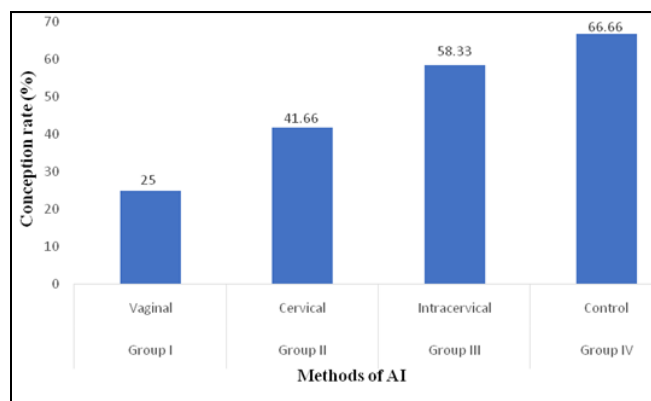


Fig 1: Relative conception rates in Mandya ewes inseminated using different methods of AI

Conclusion

From the study, it is concluded that, in spite of significantly lower conception rate in ewes of group I (Inseminated intravaginally), group II (Inseminated on the external os of the cervix) and group III (Inseminated in the body of the cervix) compared to group IV (Allowed for natural mating), the artificial insemination provides an effective alternative method of breeding in small ruminants like Mandya sheep.

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