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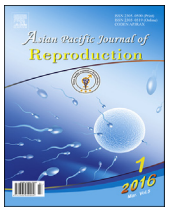

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Returning of cyclicity in infertile Corriedale sheep with natural progesterone and GnRH based strategies

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ABSTRACT

Objective: To evaluate the return of ovarian cyclicity with four hormone based protocols during non-breeding season in infertile Corriedale sheep.

Methods: The return to ovarian cyclicity was considered in terms of percent estrus response rate (ERR). The time of exhibition of estrus after either sponge removal or last $PGF_2\alpha$ injection, was considered as time to onset of estrus. Infertile Corriedale ewes were randomly selected and distributed into four groups corresponding to four hormonal protocols such as PsE (intravaginal progesterone sponges for 12 days and eCG (equine chorionic gonadotropin) at 24 h before sponge removal; $n = 6$), GP (GnRH on day 0 and $PGF_2\alpha$ on day 5; $n = 7$), GPG (GnRH on day 0, $PGF_2\alpha$ on day 5 and second injection of GnRH on day 7; $n = 7$) and PsPG (Intravaginal progesterone sponges for 12 days, $PGF_2\alpha$ and gonadotropin-releasing hormone (GnRH) respectively, at 24 h before and after sponge removal; $n = 8$).

Results: PsPG protocol produced significantly ($P < 0.05$) higher ERR (87.5%) in ewes as compared to GPG (28.57%) but non-significantly ($P > 0.05$) higher than GP (85.71%) and PsE (66%). Estrus was compact and more synchronized in PsPG group because 75% of ewes exhibited estrus during the first 48 h. The collective incidence of estrus (78.94%) was also maximum during the first 48 h.

Conclusion: PsPG estrus induction strategy has a potential to replace eCG based protocols for returning of ovarian cyclicity in infertile Corriedale sheep.

1. Introduction

In temperate climates with mid or high latitudes, as in some northern parts of India, Sheep is a seasonal breeder [1], resume their ovarian cyclicity only after a long period of ovarian quiescence during spring and summer. The fertile reproductive activity is exhibited during the autumn, which actually corresponds to the increased concentration of melatonin in the blood. During this season the length of light period becomes short and the dark period goes long. Therefore there is a transition in the photoperiod from summer to autumn [1]. It is this transition in the amount of light between different seasons that mainly controls the reproductive seasonality in sheep. There is an ample evidence that the ovarian follicular growth continues to occur during the seasonal ovarian quiescence revealed through ultrasonography [2], but the follicles do not ovulate mainly because of occurrence of less number of LH pulses (one LH pulse every 8–12 h as against one pulse every 20 min during preovulatory surge [3,4]) that are required for ovulation and with the result progesterone levels in blood become negligible [4]. However such follicles are active, synthesize and secrete estrogen, which makes them good candidates to the stimulation with exogenous gonadotropin-releasing hormone (GnRH) [5] and thus ovulation can occur. This provides an opportunity to manipulate the ewe reproductive physiology to resume ovarian cyclicity for increased lamb productivity.

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The ovarian activity in animals has been stimulated with various hormones like progesterone, prostaglandin (PGF), equine chorionic gonadotropin (eCG) and gonadotropin-releasing hormone (GnRH) [6–8]. The controlled progesterone releasing devices like sponges are very effective for estrus induction during non-breeding season in small ruminants [9,10] impregnated with either natural progesterone [11] or fluorogestone acetate or medroxyprogesterone acetate [8,12,13] The use of intravaginal sponges in combination with either eCG has boosted estrus response rate as gonadotropins stimulate ovarian follicular growth of cyclic as well as acyclic females [14–16], promoting higher levels of estrogen resulting in earlier and compact estrus response [8]. Prostaglandins cannot be used alone during non-breeding season as the animals are acyclic and do not posses corpus luteum on their ovaries. However, when used in combination with progesterone and GnRH injection prove to be efficient in controlling the life span of an active corpus luteum. A single injection of Prostaglandin 24 h before norgestomet implant removal and GnRH 24 h after implant removal has been used to induce estrus in Baladi goats [17]. Exogenous GnRH stimulates release of both FSH and LH, with former stimulates follicular growth and latter induces ovulation of dominant follicle.

There are two categories of ewes which can be utilized for ovarian manipulation during the period of acyclicity. The first category includes the normal ewes which will conceive during the breeding season but become acyclic after lambing in late winter or early spring and the second includes the ewes that fail to conceive during the previous breeding season are considered as infertile. The percentage of dry ewes (infertile) has been reported to be 23.62% [18] and if such ewes are utilized for ovarian stimulation during the following non-breeding season can substantially boost the economy of a sheep farm. Therefore, the main objective of our study was to record the effectiveness of various hormonal protocols in inducing estrus in infertile ewes during non-breeding season. It is worth to mention here that the non-breeding season besides infertile condition of ewes was also a limiting factor for returning of cyclicity.

2. Materials and methods

2.1. Animals and experimental design

The study was carried out at Mountain Research Centre for Sheep and Goat, Faculty of Veterinary Sciences and Animal Husbandry, Shuhama, located at the foot hills of greater Himalayan mountainous region of Kashmir at the outskirt of Srinagar city, which is in latitude 34°5′ North and longitude 74°48′ East and lies above the centre of the Kashmir valley at an altitude of 1585 m above mean sea level. A total of 28 infertile (acyclic) sexually mature Corriedale ewes (2–5 years of age) were selected and randomly distributed among four treatment groups. The body condition score and mean body weight of the animals were respectively 3 and 32.50 ± 5.00 kg. As the ewes did not conceive during the previous breeding season and were considered as infertile. However, on clinico-gynecological examination, the ewes were found healthy and routine deworming was done before the trial. The trial was conducted during April to May, which corresponds to non-breeding season for this breed under temperate climate and the ewes were grazed on lush green pastures from 9:00 am to 3:00 pm. Water and salt lick was provided at *ad libitum*. The ewes were not allowed to come in contact with the rams during the entire study. The four treatment groups are as under:

1) Group *PsE (n = 6) – on day 0, the ewes in this group received intravaginal progesterone sponges (350 mg progesterone impregnated, CSWRI, India) for a period of 12 days followed by an intramuscular injection of 400 IU of eCG (Folligon, Intervet India Pvt. Ltd.) at the time of sponge removal.
2) Group *GP (n = 7) – on day 0, ewes were injected with 5 μg of Buserelin acetate, a GnRH analogue (Receptal, Intervet India Pvt. Ltd.) followed by an intramuscular injection on day 5 with 263 μg of Cloprostenol, a $PGF_2\alpha$ analogue (Cyclix, Intervet India Pvt. Ltd.).
3) Group GPG (n = 7) – on day 0, ewes were injected with 5 μg of Buserelin acetate followed by an intramuscular injection on day 5 with 263 μg of Cloprostenol and finally second injection of 5 μg of Buserelin acetate on day 7.
4) Group PsPG (n = 8) – on day 0, the ewes in this group received intravaginal progesterone sponges (350 mg progesterone impregnated, CSWRI, India) for a period of 12 days, followed by an intramuscular injection of 263 μg of Cloprostenol at 24 h before and 5 μg of Buserelin acetate at 24 h after sponge removal.

Ps is abbreviation of Progesterone sponges, G is Gonadotropin, P is Prostaglandin, and E is ecG.

2.2. Parameters recorded

The parameters that were recorded include estrus response rate and time to onset of estrus. After the sponge removal or $PGF_2\alpha$ injection, two healthy normal teaser rams with colored mark on the brisket region were kept with treated ewes from 5:00 pm to 9:00 am for a period of five days. The estrus was detected on the basis of tupping mark on rump, hyperactivity of the ewes and edema of vulva. The estrus detection was done twice daily i.e. morning and evening. The ewes with color mark on croup region and vulvar edema in the morning time were considered to have exhibited estrus from midnight and those who had color mark on the croup region in the evening time were considered to have exhibited estrus from mid-day [8]. The calculations were done as:

Estrous response rate = [no. of ewes exhibited estrus/total no. of ewes treated] × 100

Time to onset of estrus = time elapsed from the sponge removal or $PGF_2\alpha$ injection to occurrence of estrus

2.3. Statistical analysis

The data obtained in respect of estrus response rate (ERR %) under different hormonal treatments was analyzed with Z test [19]. P value ≤ 0.05 was considered as significant.

3. Results

The distribution of the estrus response rate (ERR %) and time to onset of estrous is set out in Table 1. The highest estrus response rate (87.50%) was recorded in group PsPG followed by