Estrous synchronisation and artificial insemination in out of breeding season at lacaune sheep

Daniel BEREAN[1], Liviu Marian BOGDAN[1*], Anamaria Blaga PETREAN[1], George NADĂŞ[1], Mihai CENARIU[1], Sanda ANDREI[1], Ileana BOGDAN[2], Sidonia BOGDAN[1]
University of Agricultural Sciences and Veterinary Medicine from Cluj-Napoca, 400372, Calea Mănăştur, no. 3-5, Romania
[1]Faculty of Veterinary Medicine
[2]Faculty of Agriculture
[*] corresponding author, e-mail: anamaria.petrean@usamvcluj.ro

Abstract
The aim of this study was to increase fertility in sheep by applying modern biotechnology: induction and synchronization of estrous in out of breeding season. The study was conducted during May 2019- July 2019 on 132 ewes, aged between 1 and 6 years, in Covasna county, Romania. The ewes were syncronisated with Medroxyprogesterone (Ovigest, Hypra), Cloprostenol (PGF Veyx Forte, Esteve) and GnRH (Ovarelin, Ceva). After the synchronization, the artificial insemination was performed at 126 ewes by intracervical method. At 29 days after the artificial insemination was performed the diagnosis of gestation using an ultrasound. From 126 animals artificial inseminated, in 121 (91.67%) cases the diagnosis of gestation confirmed the gestation and in 22 cases was confirmed a twin gestation.
Keywords*: estrous, sheep, synchronization.*

Introduction

Sheep is seasonal anestrous, which is controlled by photoperiod, whereas; nutritional and environmental cues restrict the potentiality of attaining the global target of three lambing per ewe every two years [12]. Ewes in seasonal anestrous have lower circulating levels of LH, FSH, and estradiol with basal levels of progesterone [4]. A distinct decrease in the LH pulse frequency is the hallmark endocrine feature of ewes during seasonal anestrous which is imputed to two major inhibitory mechanisms: (1) the hypothalamus is highly sensitive to the inhibitory effects of estradiol on LH release, even at low concentrations and (2) a direct inhibitory effect of long photoperiod on the hypothalamic–pituitary–adrenal axis [6]. Estrous synchronization of sheep has been accomplished using several methods with varying degrees of success [5]. Among the different methods used are the administrations of hormones like eCG, progestogens, prostaglandins either alone or in combination. Administration of progesterone/progestogens prevents the ewe from returning to estrous and ovulating and, therefore, supplementing the ewe with progesterone for a period equal to the duration of the life of corpus luteum (CL) and then withdrawing it will synchronize the release of gonadotropins causing estrous and ovulation in groups of ewes [8]. Progestogen impregnated intravaginal sponges or implants, PGF2α alone or in combination with gonadotropins have been widely used for estrous synchronization in sheep both in and out of breeding season [1]. Saxena et al. (2015) reported a nonhormonal intervention that induced ovulation in seasonally anestrous ewes that provides proof of principle that fertile estrous can be induced in seasonally anestrous ewes using the dopamine antagonist such as sulpiride [12].

Fixed-time artificial insemination (FTAI) is an important reproductive tool in animal production system. It has a direct impact on cost-efficiency by saving time and labor for estrous detection. However, if this package is able to produce acceptable estrous synchronization and lambing percentage during non-breeding season in the field conditions, then this technology can be used for accelerated lamb production and three lambs in two year production system [7]. Whereas, the use of AI sheep declined and for this reason the farmers are more reticent towards

this technique, preferring the natural one. The advantage of this technique in sheep, is not only limited to obtain good results with the control cycles technique, but additionally facilitates the establishment of selection schemes in the offspring [10].

The aim of this study was to increase fertility in sheep by applying modern biotechnology: induction and synchronization of estrous in out of breeding season.

Matherials and method

The study was carried out during May 2018 - July 2019 in a sheep flock from Covasna county Romania (lat. 44.4267674, long. 26.102538390000063). A number of 132 ewes from Lacaune breed with age between 1-6 years were synchronized with intravaginal sponges with Medroxyprogesterone (Ovigest, Hypra), Cloprostenol (PGF Veyx Forte, Esteve,) and GnRH (Ovarelin, CEVA). After that, the ewes were artificial inseminated by intracervical method at fixed time.

The sponges (60 mg Medroxyprogesterone) were inserted and kept in situ in the vagina for a period of 12 days. 50µg of GnRH and 250 µg of Cloprostenol were administered intramuscularly at the time of sponge withdrawal on 12th day. Cervical insemination was performed in ewes exhibiting estrous (restlessness, shaking of tail, slightly swollen vulva, moist and reddish cervical external os) at 53-55 hours after the removal of sponges. For artificial insemination was used fresh semen collected from 3 rams witch had higher body weight and produced a high quality sperm. The semen was collected by artificial vagina method, after that, a 1/1 dilution of the seminal material was performed. After collection, the semen samples were evaluated for volume, consistency, wave motion (0–5 scale), density and % motile spermatozoa (0–100 %). After evaluation, a second dilution of the semen samples was performed, finally the samples were 1/10 diluted. The artificial insemination was performed with 0.25 ml of semen by intracervical method. At 29 days after the insemination the ewes were diagnosed for gestation by transrectal method with a longitudinale sonde using the frequency of 5 mH.

Results and discussions

Estrous synchronization in out of season was performed in a farm from Covasna county, Romania at 132 ewes. In the first day of the experiment 132 sponges with Medroxyprogesterone were intravaginal introduced. After 12 days a number of 131(99.24%) sponges were collected, 0.76% sponges had fallen out during this period. All the ewes were intramuscularly injected with Cloprostenol and GnRH in day 12, at 53-55 hours after the administration, 126 (95.45%) cases showed estrous signs, so, this animals were artificial inseminated by intracervical method. From 126 animals inseminated in 121 (96.03%) cases the diagnosis of gestation was positive and 3.96% the diagnosis of gestation was negative. In 22 (18.18%) cases the diagnosis of twin gestation was set (Table 1).

Table 1 Estrous synchronization and fixed-time artificial insemination in out of breeding season at Lacaune sheep under field conditions of Covasna county (Romania)

Indicator	Number (percent)
Total animals	132 (100)
Sponge in	132 (100)
Sponge out	131 (99.24)
Ewes in heat	126 (95.45)
A.I ewes	126 (95.45)

Gestations of total ewes	121 (91.66)
Gestations of A.I ewes	121 (96.03)
Number of twin gestations	22 (18.18)

The percent of sponge out after the day 12 of synchronization was 98.04%, higher than in a study of 2014 [9] where the number percent of sponge out was 96.07. Estrous activity including rate, onset and duration of estrous. In our study 95.45 % of ewes responded to treatment and exhibiting estrous signs. Nasser et al. (2012) found that estrous response of ewes treated with CIDR for 12 days + eCG or 6 days + eCG was 100% for both treatments [10]. However, Kasikci et al. (2011) reported that lower estrous response of Tahirova ewes treated with whole sponge (20 mg), halved sponge (10 mg), whole sponge + 600 IU eCG, whole sponge + 300 IU eCG, halved sponge + 600 IU eCG and halved sponge + 300 IU eCG, being 91, 96, 86, 96, 98 and 94% [9]. Özbey and Tatle [11] synchronized the Awassi ewes for 14 days with sponges containing 40 mg of FGA and superovulated by 500 IU of PMSG injection and estrous and twinning rates were 100% and 46%, in our study the twinning rate as 18.18%. The use of freshly diluted semen could give the best result: 70 to 82 % [2] and 82.2% [3] of pregnancy rate, in our study the pregnancy rate was 91.66%.

Conclusions

The results of estrous synchronization and artificial cervical insemination in out breeding season with fresh diluted semen were acceptable, 95.45 % ewes showed estrous signs, gestation percent was 91,66% with 22% of twin gestation.

Estrous synchronization and artificial insemination in out of breeding season can be performed with good results in Lacaune sheep in Romania.

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