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# Efficacy of the application of estrus synchronization in sheep during non-breeding season

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## Abstract

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The present study examines the success rate of the estrus synchronization in 289 sheep depending on their age group. All animals were subject to the same synchronization scheme. It involved the insertion of vaginal progesterone-containing sponges for sheep impregnated with fluorogestone acetate (FGA) 30 mg which remained in the vagina for 14 days. Lyophilized serum gonadotropin hormone (PMSG) 500 IU/ewe was applied after the sponge removal. The first artificial insemination was performed 48 hours later, and the second one – after around 5 hours. The average pregnancy rate for all groups was  $61.6\% \pm 3.486$  (p < 0.001). Upon examining the percentage of pregnant ewes depending on the age group, we ascertained that the second age group comprising the ewes from 2 to 4 years had the best pregnancy rate – 72.9%. Predictably, the lowest pregnancy rate-42.6% was that in the first age group comprising the one-year-old animals. The third age group comprised all animals above 5 years. Its pregnancy rate was 54.9% at p < 0.001.

Significant differences were reported with reference to group one and two at p < 0.001 and group two and three -p < 0.05. The differences between age group one and three were insignificant p = 0.238.

Keywords: sheep; estrus synchronization; FGA sponges; ewes

# Introduction

The non-breeding season hinders the ability of sheep to give birth and provide milk year-round. A key element of the effective reproductive management in sheep is the estrus synchronization. It allows the breeding and lambing campaigns to be performed within a short period, enhances the technological processes organization through the simultaneous weaning of large groups of lambs and levelled sheep milking groups, etc. (Metodiev, 2018). A range of authors examine the opportunities for the application of different methods for intensification of the breeding process in small ruminants during (Pampukidou et al., 2011a; Pampukidou et al., 2011b; Meslev et al., 2010) and out of the breeding season (Bonev & Kostadinova, 2011; Luridiana et al., 2015; Suocheng Wei et al., 2016; Hatem, 2018). The hormonal methods are main in estrus induction during the non-breeding season. According to Silveira et al. (2021) FSH treatment upon estrus synchronization in non-breeding season leads to follicular growth but cannot induce ovulation. Upon application of fixed-time artificial insemination synchronization regimes eCG proves to be more effective in estrus induction than FSH combined with GnRH and estradiol esters.

Maslev et al., (2010) ascertain that if the vaginal sponges remain inserted for a longer period (12 days) in sheep mark better results than if the sponges are used for a shorter period (6 days).

Despite all the advantages and benefits of the estrus synchronization application, it also has a range of disadvantages, which seriously affect its adoption as a routine procedure, especially upon its application during the non-breeding season. Therefore, we set ourselves the aim to study the possibility for estrus synchronization and artificial insemination during the non-breeding season in Assaf crossbred sheep.

## **Materials and Methods**

The estrus synchronization was applied in 289 ewes. During the data analysis, the animals were grouped in three age groups as follows: 1 age group – one-year-old animals, 2 age group – from 2 to 4 year-old and 3 age group comprised all animals above 5 years of age. The rams were reared in a group in a different place separately from the ewes. The daily physical activities of the breeding animals were ensured by morning and evening access to the open air. The ram preparation started 40 days prior the breeding campaign and the animals were given an additional 0.500 kg concentrated feed, carrots, vitamins et al in addition to the main ration.

Training of rams for semen collection was carried out as of the beginning of the preparation period. The animals were trained to provide ejaculate by the artificial vagina method. Prior the ram inclusion as sperm donors, their sperm production was thoroughly examined. Subcutaneous melatonin implants were inserted in the rams thirty days prior the planned estrus synchronization.

During the preparation of the sheep, all breeding female animals were subject to anti-parasite treatment one month prior the synchronization. Their ration was complemented by 0.300-0.400 kg concentrated feed (corn, barley) and vitamin lick blocks. The synchronization was performed during March and April – non-breeding season. Due to organizational reasons of the farm, the animals were subdivided into 5 groups and were subject to synchronization in stages in threeday intervals.

The pregnancy testing was performed with a veterinary ultrasound Portable Veterinary Ultrasound.

The data were statistically processed with the program SPSS 21 IBM and MS Excel 2016. The group variation was reported by means of Tukey HSD multiple comparison test.

#### **Results and Discussion**

During the estrus synchronization, all 289 female animals were subject to one synchronization scheme (Figure 1). The day of insertion of the vaginal sponges (30 mg FGA Synchropart®, Ceva Sante Animal) which remained in the animals vaginas for 14 days was considered day 0. Lyophilized serum gonadotropin hormone (PMSG) (Gonaser, HIPRA), 500 IU/ewe was applied in sponge removal. The first artificial insemination was performed 48 hours later, and the second one- after around 5 hours. The insemination



Fig. 1. Sheep synchronization scheme at non-breeding season

was performed with a previously evaluated and diluted semen following a breeding plan elaborated beforehand. An ultrasound pregnancy test was conducted two months after the synchronization.

Table 1 displays the results of the performed synchronizations. The average pregnancy rate for all groups was 61.6%(p < 0.001). Upon studying the percentage of pregnant ewes depending on the age group, we found that the best pregnancy rate – 72.9% was that in the second age group including the sheep from 2 to 4 years. In the first age group including the one-year-old animals, the pregnancy rate was predictably the lowest – 42.6%. The third age group include all animals above 5 years. Its pregnancy rate was 54.9%.

The reported results were considerably lower than those reported in most studies (Barean et al., 2021; Suocheng Wei et al., 2016; Martinez-Ros et al., 2018). Berean et al. (2021) report a significantly higher pregnancy rate of 91,67% in one of their studies which followed a similar synchronization protocol, season and natural and climatic conditions (March, Romania). Upon application of similar synchronization protocols, another study (Garoussi et al., 2020) of the fat-tailed sheep in Iran marked pregnancy rates from 28% to 44% in the different groups. Maslev et al. (2010) establish a pregnancy rate of 71.4% in sheep of the Karakachan breed after applying a similar synchronization protocol at the beginning of the breeding season. During a study of Awassi sheep carried out at the end of March, Kridli et al. (2006) report a pregnancy rate of 50%.

After the end of the period of lambing we ascertained that the fecundity of the flock examined was 52.6% in total. It was the lowest in the first age group – 41.1% i.e. 54 inseminated ewes provided 22 lambs. The prolificacy was 116%, the multiple births were 15.8%. The second age group had the highest fecundity, prolificacy and multiple births rates- respectively 63.9%, 123% and 22.7%. Of 91 inseminated ewes were born out 38 lambs in the third age group. When analyzing the prolificacy, we ascertained that the percentage of the lambs born and the lambed ewes was 115. Of 33 lambings, only in 5 there was a twin birth. The total prolificacy and the twin-birth percentages were respectively 120% and 19.7%.

Age group	Pregnancy rate (n/N), %	Fecundity (lambs born per mated ewe) (n/N), %	Prolificacy (lambs born per lambed ewe) (n/N), %	Multiple births (n/N), %
1	(23/54)	(22/54)	(22/19)	(3/19)
	42.59	41.07	115.8	15.79
2	(105/144)	(92/144)	(92/75)	(17/75)
	72.92	63.89	122.7	22.67
3	(50/91)	(38/91)	(38/33)	(5/33)
	54.95	41.76	115.2	15.15
Total	(178/298)	(152/289)	(152/127)	(25/127)
	61.59	52.60	119.7	19.69

Table 1. Percentage of pregnant sheep within the age group

Table 2. Multiple comparison (Age group) depending on the pregnancy rate using Tukey HSD

Age group (I)	Age group (J)	Mean Difference (I-J)	$\pm$ SE	Sig.
1	2	-31.10***	7.506	0.000
	3	-13.13	8.087	0.238
2	1	31.10***	7.506	0.000
	3	17.97*	6.341	0.014
3	1	13.13	8.087	0.238
	2	-17.97*	6.341	0.014

\*\*\*p < 0.001; \*\* p < 0.01; \* p < 0.05

The differences between the groups compared following Tukey are displayed in Table 2. There were significant differences between group one and group two -p < 0.001, and between group two and group three -p < 0.05. The differences between age groups one and three were insignificant p = 0.238.

## Conclusions

The average pregnancy rate during the study carried out was 61.6%, p < 0.001. When analyzing the influence of the age group, we ascertained that the best pregnancy rates of 72.9% were achieved by the second age group including the ewes from 2 to 4 years. The pregnancy rate in the first group including the one-year-old animals was the lowest – 42.6%. The pregnancy rate reported for the third age group was 54.9% at p < 0.001.

The total fecundity in the flock examined was 52.6%. It was the lowest in the first age group -41.1% i.e. 54 inseminated ewes gave birth to 22 lambs. The prolificacy was 116% and the multiple births -15.8%. The fecundity, prolificacy and multiple births values of the second age group were the highest -63.9%, 123% and 22.7% respectively. In age group three, only 38 lambs were born from a total of 91 inseminated ewes. After analyzing the prolificacy, we ascertained that the percentage ratio between the lambs born and the lambing ewes was 115. Of 33 lambings, only in 5 there was a twinbirth. The total prolificacy and the twin-birth percentage were respectively 120% and 19.7%.

The differences between the groups compared under Tukey were significant between group one and group two at p < 0.001, and between group two and group three -p < 0.05. The differences between age groups one and three were insignificant p = 0.238.

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