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THE EFFECT OF HORMONAL STIMULATION SCHEME AND SEASON ON THE EFFICIENCY OF ESTRUS SYNCHRONIZATION IN ROMANOV EWES (*Ovis aries* L.)

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Abstract

Estrus synchronization in sheep is an important element in the organization of reproductive programs, including those based on assisted reproductive technologies. The diversity of sheep breeds, the differences in environmental conditions of breeding zones and the specificities in hormonal status of animals not allow the finding a universal protocol for the management of reproductive cycles. The aim of our work was to identify the conditions of estrus synchronization in Romanov ewes in different seasons of the year. For the first time, a comparative analysis of two schemes of hormonal stimulation of estrus was carried out, involving either two consecutive injections of prostaglandin F2α (Scheme 1), or initially injection of gonadotropin releasing hormone followed by prostaglandin F2α treatment (Scheme 2). The study was carried out on mature Romanov ewes (n = 160). The first group (n = 121) underwent hormonal treatment using the Scheme 1, with two injections of cloprostenol (125 μg per injection) 13 and 2 days before the expected day of estrus (day 0). In the second group of animals (n = 39), the Scheme 2 was used, which included injections of 15 μg of lyuliberin acetate and 125 μg of cloprostenol at days 9 and 2 before expected day of estrus, respectively. In the first group, the results of estrus synchronization were analyzed by seasons, the autumn—winter period (n = 73) and spring—summer period (n = 48). For detail analysis, the data were studied by two-month periods: Sept-Oct (n = 24), Nov-Dec (n = 26), Jan-Feb (n = 23), Mar-Apr (n = 32), and May-June (n = 16). The efficiency of hormonal treatment was evaluated in all experimental animals based on appearance of estrus in 24 and 48 hours after the last injection. In some of the animals that showed estrus response (n = 80), visual assessment of the ovaries for the presence of corpora luteum (CL) was performed using laparotomy or laparoscopy 96 hours after estrus detection. The variance analysis showed a reliable effect of the hormonal treatment scheme (F = 5.21, p = 0.024) as well as the season (F = 13.82, p = 0.0003) on the estrus response. The average number of CL was subjected to greater variability by the year of experimental studies ($p \leq 0.05$) and the season ($p \leq 0.01$), without significant effect of the treatment scheme. Using the Scheme 1 revealed the trend of higher estrus response comparing to Scheme 2, 80.17 % vs 66.67 %. Along with that, in the second group the CL were found in all ewes with the estrus signs, while in the first group the CL were found in 90.77 % of animals ($p \leq 0.05$). Synchronization results for ewes in the autumn-winter period were better than in the spring-summer period: the estrus response was 92.50 % compared to 64.58 % ($p \leq 0.01$), the average number of CL for ewes in the estrus stage was 2.02 vs 1.28 ($p \leq 0.01$), and the average number of CL in ewes with identified CL was 2.18 vs 1.62 ($p \leq 0.05$). A more detailed analysis by the two-month periods showed that the values of the above-mentioned indices were relatively stable during the autumn-winter period, after which they decreased in Mar-Apr and reached minimum values in May-June. Thus, for stimulating the estrus in Romanov ewes we recommend two consecutive injections of cloprostenol (13 and 2 days before the expected estrus). The efficiency of the method is higher in autumn-winter season, after which a decrease occurs in the response of animals to hormonal treatment.

Keywords: ewes, Romanov breed, hormones, prostaglandin F2α, estrus stimulation, ovarian function

Synchronization of the sexual cycle (SC) in sheep is a ways to increase the efficiency of realization of the reproductive potential of females and optimize reproduction programs [1]. Since the effectiveness of synchronization is mostly assessed by the onset of heat, it is often designated as "hunt synchronization". Heat synchronization can neutralize the influence of seasonal factors on sheep reproduction to extend the breeding season, to plan lambing of ewes at a predetermined period of time throughout the year [2-4] and to obtain up to 2-3 lambings in 2 years, which increases the economic efficiency of sheep farming [1, 5].

Synchronization of the reproductive cycle is of particular relevance in connection with the development of assisted reproductive technologies [6], including artificial insemination, in vitro production of embryos, cloning, and embryo transplantation [7-9]. Biological, hormonal and combined methods provide SC synchronization.

Multisensory contact between ewes and rams, called the "male effect", is a biological method for stimulating estrus outside the breeding season [10]. Such multifactorial stimulation involves the olfactory, tactile and visual receptors of females. Stimulation of estrus is based on changing pulsations of the gonadotropic releasing hormone (GnRH) secretion and increasing the secretion of luteinizing hormone (LH). During the first ovulation which occurs 2-3 days after contact of ewes with rams, heat manifestation is often absent ("silent heat"), and the fertility of ewes remains low. The main factor limiting the use of this biological method is the reduced fertility of sheep in the first cycle and a decrease in the effectiveness of synchronization in subsequent cycles. It was noted that the effectiveness of this method increases with the artificial extension of the photoperiod during the previous two months, eventually, estrus occurred in 99% of females [11]. Another technique that increases the effectiveness of the "male effect" is treating females with 20 mg of progesterone when introducing rams into the herd [12]. Biological stimulation of the reproductive cycle in sheep are of significant interest since it does not require treatment with hormonal drugs.

Hormonal drugs, when used optimally, can provide higher efficiency in synchronizing the sexual cycle. There are various schemes based on the use of progestogen drugs, prostaglandin F2α (PGF2α) and its structural analogues. The choice of regimen depends on the breed of sheep, season, and physiological state of the animal [13]. When synchronizing during the breeding season, prostaglandins are usually applied, for which the target is the functional corpus luteum; outside the breeding season, complex progesterone-based regimens are used, followed by the administration of equine chorionic gonadotropin (eCG) and gonadotropin-releasing hormone GnRH [1, 14, 15].

Synthetic progestogens which act as prolongators of the luteal phase of the sexual cycle, can be administered orally, intravaginally, or subcutaneously as boluses. The oral method is the least labor-intensive. The drug is mixed with salt or dissolved in ethyl alcohol and added to the feed. Animals are fed the drug for 8-10 days. However, this method does not guarantee that all animals will consume the required dose [16].

Regardless of the season, progestogen intravaginal sponges impregnated with alcohol or propylene glycol solutions of drugs based on progesterone (P4), fluorogestone acetate (FGA) and medroxyprogesterone acetate (MPA) have become widespread, regardless of the season [17]. The sponge is placed in the vagina for 10-14 days followed by subcutaneous injected of eCG. Signs of heat appear within 24-48 hours in 90% of individuals [18-21]. A study of the possibility of increasing the effectiveness of intravaginal sponges containing progestogens in combination with subcutaneous melatonin implants showed [22] that in the group receiving melatonin, the fertility of females was 60.4% vs. 32.6% in the control group. The positive effect of melatonin in combination with progestogens and eCG was also noted in Awassi sheep when stimulating estrus during the aestrous period [23]. It has been established that melatonin exhibits a positive effect together with progestogens and eCG, but without other drugs it does not stimulate estrus [24].

Progestogen preparations in the form of boluses are effective and safe for animal health [25]. Using a special applicator, a bolus containing the active substance is injected subcutaneously into the ear for 8-12 days. At the same time, an injection of a synthetic analogue of progesterone - norgestamet (1.5 mg) and estradiol (1.9 mg) is given. After 8-12 days, the implant is removed and an LHC injection is given. With this scheme, within 24 hours, 95-100% of the animals showed signs of heat, and the fertilization rate with fresh sperm was 85-95% [26]. In studies by Z. Mekuriaw et al. [27] boluses were combined with an injection of 300-400 IU eCG and 50 μg cloprostenol 48 hours before implant removal. The onset of heat occurred in 65-95% of animals with a fertility rate of 50-90% [27].

To synchronize hunting during the breeding season (autumn-winter; the duration of the period depends on the breed of sheep), drugs containing prostaglandin F2 and its synthetic analogues are used. Exogenous prostaglandin F2α has no effect on cyclicity in sheep in the absence of the corpus luteum, that is, during the luteal phase of the reproductive cycle. Therefore, preparations based on prostaglandin F2α are more effective during the sexual season, during which at least some of the sheep in the herd have corpora lutea. To synchronize the sexual cycle, animals are injected with a drug based on prostaglandin F2α twice with an interval of 8-12 days [28]. Hunting occurs 48-60 hours after the last injection. The effectiveness of this method can reach 100%. In our earlier studies on a small sample of Romanov breed sheep, we showed greater effectiveness of a regimen based on duble injection of prostaglandin F2α, compared to the combined use of gonadotropic releasing hormone and prostaglandin F2α, the proportion of animals that came into heat, was 100 and 50%, respectively [29].

The variety of sheep breeds and differences in natural and climatic conditions in breeding areas do not allow us to select a universal synchronization protocol. In addition, the effect of hormonal drugs on the reproductive system of females can manifest itself differently depending on the initial functional state of the reproductive organs and hormonal status. Thus, the effectiveness of hormonal stimulation of reproductive cycles in sheep at the beginning of the breeding season is significantly lower than at its end. In lactating females, the effectiveness of stimulation is lower than in non-lactating animals. If hormonal drugs are used incorrectly or the dosage is poorly selected, negative consequences can be observed [30, 31]. Therefore, the choice of the optimal hormonal treatment regimen is the most important element of assisted reproductive technologies in sheep breeding.

In this work, for the first time, a comparative analysis of the effectiveness of two schemes of hormonal stimulation of oestrus, involving either two successive injections of prostaglandin F2α, or an initial injection of gonadotropic releasing hormone followed by treatment of sheep with prostaglandin F2α, was carried out. The use of the first scheme provided the best response to synchronization in relation to the proportion of animals that came into heat. A higher efficiency of synchronization of the sexual cycle was established in the autumn-winter period compared to the spring-summer period when using both schemes.

The purpose of the work was to determine the effectiveness of different schemes for synchronizing the sexual cycle in Romanov breed sheep according to the seasons of the year.

Materials and methods. The study was carried out on sexually mature Romanov breed females aged 1.5-2 years (n = 160, Federal Research Center for Animal Husbandry - Ernst VIZh, Moscow Province, from January 2021 to December 2022). Each animal was used only once during the experimental period. Young ewes were kept in pens under sheds in groups of 15-20 animals. separately from rams. In winter, young ewes received a hay-concentrated diet in accordance with the breed's requirements. In the summer, the animals were on pasture and additionally received concentrates. All animals had unlimited access to mineral salt and water.

The animals were divided into two groups. In group 1 (n = 121), the ewes were subjected to a 2-fold injection of a prostaglandin analogue cloprostenol (Estrophan, Bioveta, Czech Republic), 125 μg per injection over 13 days (–13 days) and 2 days (–2 days) before the expected date of coming into heat (0 days) (scheme 1). Animals of group 2 (n = 39) were injected with 15 μg of gonadotropin luliberin acetate (Surfagon, Mosagrogen, Russia) on day –9 and an injection of 125 μg of cloprostenol on day –2.

The effectiveness of hormonal stimulation schemes was assessed in all females based on the coming into heat, as well as the visual detection of corpora lutea (CL) in the ovaries of some individuals with signs of heat (n = 67 and n = 13 for groups 1 and 2, respectively). Coming into heat was determined using a vasectomized probe ram 24 and 48 hours after the last injection. Ovarian function (presence and number of CLs in the ovaries) was assessed by laparotomy or laparoscopy using endoscopic equipment (Karl Storz SE & Co. KG, Germany) 96 hours after the last injection.

The effectiveness of synchronization of the sexual cycle was determined by the following criteria: the proportion of females that came into heat from the total number of those subjected to hormonal treatment; the proportion of females with CLs out of the total number that showed signs of heat; average number of fatty acids per animal with signs of heat; the average number of CLs per animal with CLs. For each group of animals, these indicators were calculated for the entire test period. The influence of the seasons autumn-winter (September-February) and spring-summer (March-July) were studied in ewes of group 1 (n = 73 and n = 48, respectively). For a more detailed analysis, indicators were compared for five time periods: September-October (n = 24), November-December (n = 26), January-February (n = 23), March-April (n = 32) and May-June (n = 16).

To determine the statistical significance of the influence of the studied factors (hormonal treatment regimen, season and period of the year) on the variability of PC synchronization performance indicators, two-factor analysis of variance without interaction (due to the incomplete rank of the model) was used using the STATISTICA 10 program (StatSoft, Inc., USA) according to the following equation:

$$y_{jilk} = \mu + year_j + scheme_i + season_l + animal_k + e_{jilk},$$

where $year_j$ is the fixed effect of the year of research (j is 2020-2023); $scheme_i$ is the fixed effect of the i-th hormonal treatment scheme (i is 1 and 2); $season_l$ is fixed effect of the l-th season (l is autumn—winter, spring—summer); $animal_k$ is the randomized effect of the k-th animal; e_{jilk} is the residual (randomized) effect of the equation model. To calculate the independent distribution of events according to the proportion of females that came into heat, in connection with the influence of factors, a nonparametric $\chi 2$ test according to Pearson was used. To assess the strength of the influence of factors on the synchronization performance of sheep in the experimental groups, arithmetic mean values (*M*) and the error of the mean (±SEM) were determined. The statistical significance of differences in arithmetic mean values was assessed by Student's *t*-test. The results were considered highly reliable at $p \leq 0.001$, significant at $p \leq 0.01$ and $p \leq 0.05$.

Results. Figure 1 illustrates the experimental design:

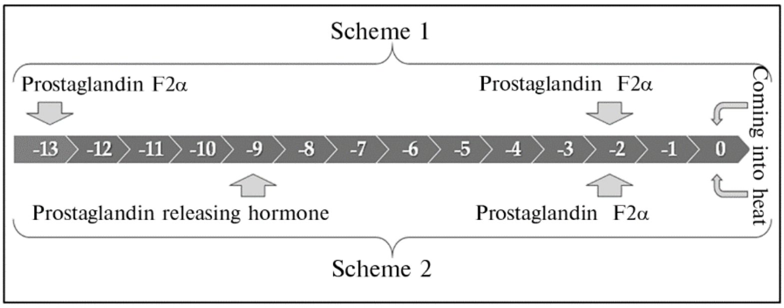

Fig. 1. Schemes of hormonal synchronization of the reproductive cycle in the Romanov 1.5-2-month-old ewes (*n* = 160, Ernst Federal Research Center of Animal Husbandry — VIZh, Moscow Province, from January 2021 to December 2022). Timeline referes to a day before going into the hunt

1. Results of a two-factor analysis of variance for synchronization of the sexual cycle in the Romanov 1.5-2-month-old ewes depending on the year, hormonal treatment scheme and season (*n* = 160, Federal Research Center of Animal Husbandry — Ernst VIZh, Moscow Province, from January 2021 to December 2022)

Parameter	Significance test	Factor			R^2, %
		year	scheme	season	
Proportion of yearling ewe that came into heat	*F*-test	0.18^{na}	5.21*	13.82***	10.3
	(p-value)	(0.910)	(0.024)	(0.0003)	
	χ^2 test	0.49^{na}	3.02^{t}	11.22**	
	((p-value)	(0.921)	(0.082)	(0.008)	
Average number of corpora lutea	*F*-test	3.49*	0.11	10.81**	23.6
	(p-value)	(0.020)	(0.736)	(0.011)	

[na] The influence of the factor is unreliable.
[t] The influence of the factor is statistically significant at $p \leq 0.10$.
*, ** and *** The influence of the factor is statistically significantat $p \leq 0,05$, $p \leq 0,01$ and $p \leq 0,001$, respectively.

For the proportion of females that came into heat (Table 1), a significant influence of the applied hormonal treatment regimen ($F = 5.21$; $p = 0.024$), as well as the season of the year ($F = 13.82$; $p = 0.0003$) was established (see Table 1). The coefficient of determination of the model, which explained the amount of variability in the proportion of bright spots between the compared groups of factors, was 0.103, or 10.3%. Pearson's $\chi 2$ test showed that the difference in this indicator also depended on the synchronization scheme ($p \leq 0.05$) and the season of the year ($p \leq 0.01$), which indicates the contingency of the distribution of events of coming into heat (yes/no) and the inapplicability of zero hypotheses about the mutual independence of the factors under study and the results of observations. The year of study factor did not influence the distribution of threshold characteristics ($F = 0.18$; $p = 0.910$). The average number of CLs was subject to greater variability due to the calendar year of the studies ($p \leq 0.05$) and the season of the year ($p \leq 0.01$), while the treatment scheme did not significantly affect this indicator. An increase in the determination coefficient R^2 (23.6% vs. 10.3%) indicated that the value characterizing the distribution of variations within groups in comparison with the overall group variability for the average number of CLs was higher than for females that came into heat (see Table 1).

As follows from the data presented in Table 2, scheme 1 (group 1) turned out to be more effective than scheme 2 (group 2) when comparing the proportion of animals that came into heat (80.17 vs. 66.67%), however, the differences found can only be considered as a trend. When using scheme 2 (group 2), CLs were detected in all females with signs of heat, while the use of scheme 1 led to the formation of CLs in 90.77% of females ($p \le 0.05$). Fot scheme 2, there was a tendency for the average number of CLs to increase compared to that for scheme 1 (+1.17) due to CLs detection in all females with signs of heat. When determining the average number of CLs in females with CLs, the differences between the groups were leveled out.

2. Effectiveness of the sexual cycle synchronization in the Romanov 1.5-2-month-old ewes as influenced by the hormonal treatment sheme (*M*±SEM, Federal Research Center of Animal Husbandry — Ernst VIZh, Moscow Province, from January 2021 to December 2022)

Parameter	Group	
	1	2
Number of animals, n_1	121	39
Proportion of females that came into heat, %	80.17±3.64	66.67±7.65
Number of animals, n_2	67	13
Proportion of females with CL[1], %	90.77±3.59*	100
Average CL number[1]	1.75±0.13	1.92±0.35
Average CL number in the responded animals[2]	1.98±0.11	1.92±0.35

N o t e. n_1 is the number of animals that have undergone synchronization of the reproductive cycle, n_2 is the number of animals that have undergone synchronization of the reproductive cycle and were tested for the presence of corpus luteum (CL); [1] is indicator based on the number of females coming into heat; [2] is an indicator based on the number of females that come into heat and have CL.

* Differences between groups are statistically significant at $p \le 0.05$.

3. Eeffectiveness of the sexual cycle synchronization scheme 1 in the Romanov 1.5-2-month-old ewes as influenced by the season (*M*±SEM, Federal Research Center of Animal Husbandry — Ernst VIZh, Moscow Province, from January 2021 to December 2022)

Parameter	Season	
	autumn-winter	spring-summer
Number of animals, n_1	73	48
Proportion of females that came into heat, %	90,41±3,45**	64,58±6,90**
Number of animals, n_2	42	25
Proportion of females with CL[1], %	92,50±3,97	80,00±10,69
Average CL number[1]	2,02±0,15**	1,28±0,19**
Average CL number in the responded animals[2]	2,18±0,16*	1,62±0,15*

N o t e. See the description of scheme 1 in Figure 1. The autumn-winter season included the period September-February, spring-summer include March-July; n_1 is the number of animals that have undergone synchronization of the reproductive cycle, n_2 is the number of animals that have undergone synchronization of the reproductive cycle and were tested for the presence of corpus luteum (CL); [1] is indicator based on the number of female ducks that came into heat; [2] is an indicator based on the number of females that come into heat and have CL.

*, ** Differences between groups are statistically significant at $p \le 0,05$ и $p \le 0,01$, resptctively.

The data in Table 3 show a significant influence of the season of the year on the effectiveness of the sexual cycle synchronization when assessed by the proportion of females that come into heat. In the autumn-winter period, the value of this indicator was 25.83% higher than in the spring-summer period (92.50 vs. 64.58%, $p \le 0.01$). A similar seasonal dependence occurred for the proportion of females with CLs from the total number of those showed signs of estrus (+12.5%), however, the differences identified were just a trend. A more detailed analysis carried out for 2-month periods showed that the proportion of animals going into heat was relatively high in the period from September to February (88.5-91.7%), followed by a decrease to 71.9% in March to April. In May-June, the proportion of females that came into heat after hormonal treatment was 50.0% (Fig. 2). An interesting pattern emerged in relation to the corpora lutea. If in September-December 100% of females that showed signs of heat had visualized CLs in the ovaries followed by a decrease in January-April to 81.3-89.5%, in May-June only up to 50 %.

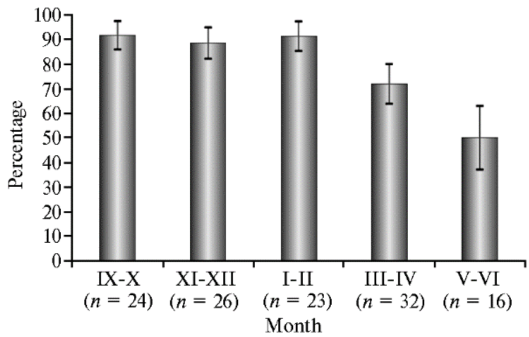

Fig. 2. Effectiveness of the sexual cycle synchronization in Romanov 1.5-2-month-old ewes under hormonal treatment scheme 1: the proportion of females that came into heat from the total number of those subjected to hormonal treatment, % (M±SEM; data recorded at a 2-month intervals, Ernst Federal Research Center of Animal Husbandry — VIZh, Moscow Province, from January 2021 to December 2022). See Figure 1 for the description of scheme 1.

We revealed a significantly higher number of CLs in females that underwent synchronization of the reproductive cycle in the autumn-winter period, compared to that in the summer-spring period (2.02 vs. 1.28, $p \leq 0.01$), and the identified differences remained the same when calculating this indicator for females with CLs (2.18 vs. 1.62, $p \leq 0.05$) (see Table 3). Analysis of changes over 2-month periods showed that the mean number of CLs per female that came into heat was greater from September to February, 1.83-2.17. In March-April, the number of identified CLs decreased to 1.47, in May-June it dropped to a minimum for the entire period of the experiment and amounted to 0.67 (Fig. 3). In females that came into heat and had yellow bodies for the above periods of the year the number of CLs was 1.83-2.38, 1.65 and 1.33, respectively.

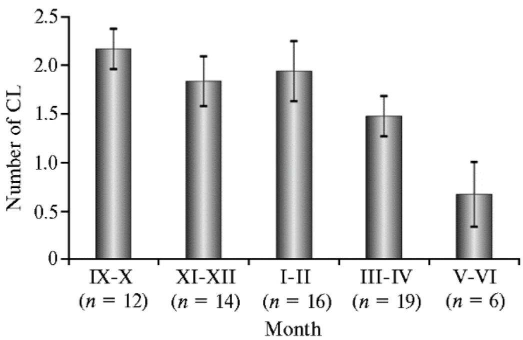

Fig. 3. Effectiveness of the sexual cycle synchronization in Romanov 1.5-2-month-old ewes under hormonal treatment scheme 1 when counting CLs per female: average CL number per all females that came into heat (M±SEM; data recorded at a 2-month intervals, Federal Research Center of Animal Husbandry — Ernst VIZh, Moscow Province, from January 2021 to December 2022). See Figure 1 for the description of scheme 1.

Classic protocols for synchronizing SC in sheep include intravaginal insertion of a progesterone-containing sponge or intravaginal device (CIDR) impregnated with fluorogestone acetate or medroxyprogesterone, combined with an intramuscular injection of pregnant mare serum gonadotropin (PMSG) at the date of estrus [20, 32]. Various variants of this protocol are also used (Table 4), but in the vast majority of cases, hormonal synchronization of estrus does not provide 100% effectiveness in terms of the number of animals that come into heat. With only one scheme out of 25 analyzed by us (see Table 4), all treated animals showed signs of heat [20]. In other variants, the effectiveness of synchronization, depending on the scheme and breed, varied from 59.1 to 96.0%. Overcoming seasonality through hormonal stimulation of estrus in this species also remains an unresolved problem [26].

In contrast to the approaches described above, in the present study, to stimulate the estrus, either a 2-fold injection of PGF2α was used 13 and 2 days before the expected date of coming into heat (scheme 1), or injections of GnRH 9 days before and PGF2α 2 days days before the estrus (scheme 2). Both hormonal treatment regimens (albeit to varying degrees) showed effectiveness in stimulating estrus in mature females (see Tables 2, 3). However, during the year (except for the period July-August), the average performance for scheme 1 was higher than for scheme 2, amounting to 80.17%, which is comparable to traditional protocols (see Table 4). In addition, from September to December, 2-fold administration of PGF2α provided stimulation of estrus in 100% of treated mature females, which was consistent with the result for CIDR in combination with PGF2α, PMSG and estradiol benzoate [20]. It should be noted that the effectiveness of scheme 1 decreased only in the summer.

4. Effectiveness of the reproductive cycle synchronization in different sheep breeds depending on the hormonal treatment scheme

Scheme — drug/device (days to the predicted date of entry into heat)	Animals that came into heat, % of total treated	Breed	References
PGF2α (–13, –2)	80.17	Romanovskaya	This work
GnRH (–9), PGF2α (–2)	66.67		
CIDR (c –7 to 0)	89	Southdown, Ramboulier, Columbian, Suffolk × Hampshire, Romanovskaya × White Dorper, Romanovskaya × Katahdin	[19]
CIDR (c –7 to 0), PGF2α (0)	93		
CIDR (c –14 to 0)	93		
CIDR (from –10 to 0)	68	Burgskaya	[19]
CIDR (from –19 to 0)	72		
CIDR (from –6 to 0), PGF2α (–6), PMSG (0), EB (+1)	100	Dorper	[20]
PG-sponge (from –14 to 0), PMSG (0)	76.7	Lakon, segureno, segureno × Romanovskaya	[21]
PG-sponge (c from –7 to 0), PGF2α (–7), PMSG (0)	80		
PG-sponge (from –7 to 0), PMSG + PGF2α (0)	90		
PG-sponge (from –14 to 0), PMSG (0)	80		
PG-sponge (from –7 to 0) + PGF2α (–7), PMSG (0)	79.2		
PG-sponge (from –7 to 0), PMSG (0), PGF2α (0)	59.1		
PG-sponge (from –13 to –2), PGF2α (–4), GnRH (0)	70	Chinese Hu Sheep	[15]
PG-sponge (from –13 to –2), PGF2α + PMSG) (–4)	83.3		
PG-sponge (from –13 to –2), PGF2α + PMSG (–4), GnRH (0)	86.7		
PG-sponge (from –13 to 0), PGF2α (–1), GnRH (0)	80		
PG-sponge (from –13 to 0), GnRH (0)	76.7		
CIDR (from –5 to 0)	89	Columbian, Hampshire	[33]
CIDR (from –5 to 0), PGF2α (0)	77		
GnRH (–5), CIDR (from –5 to 0), PGF2α (0)	75		
CIDR (from –5 to 0)	78	Horned Dorset, Katahdin	
CIDR (from –5 to 0), PGF2α (0)	90		
GnRH (–5), CIDR (from –5 to 0), PGF2α (0)	96		

N o t e. GnRH — gonadotropin releasing hormone, PMSG — pregnant mare serum gonadotropin, EB — estradiol benzoate, PGF2α — prostaglandin F2α, PG-sponge — progestogen intravaginal sponge, CIDR — device for controlled internal release of the drug.

Thus, our studies on the sexual cycle synchronization in Romanov young ewes revealed a significant influence of the hormonal treatment scheme ($F = 5.21$; $p = 0.024$) and the season of the year ($F = 13.82$; $p = 0.0003$) on the proportion of females who came into heat. A 2-fold injection of prostaglandin F2α on days –13 and –2 before the expected date of estrus (day 0) ensures higher performance compared to the administration of gonadotropin releasing hormone on day –9 and prostaglandin F2α on day –2, 80.17 vs. 66.67%. For the second scheme, all the females that showed signs of heat had corpus luteum (CLs) in their ovaries, whereas with the first scheme their share was 90.77% ($p \leq 0.05$). The best response to hormonal treatment occurs during the sexual season. In September-February, the proportion of females that came into heat, the average number of CLs in females that came into heat, and the average number of CLs in females that had CLs were significantly higher than in March-July, 90.41 vs. 64.58% ($p \leq 0.01$); 2.02 vs. 1.28 ($p \leq 0.01$), and 2.18 vs.1.62 ($p \leq 0.05$) , respectively. To synchronize the sexual cycle in Romanov ewea, we recommend a 2-fold injection of prostaglandin F2α on days –13 and –2 before the expected date of estrus. In terms of the proportion of females that come into heat, the effectiveness of the scheme we propose is comparable to that described by other reserachers, and in some cases exceeds it.

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