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ESTRUS INDUCTION IN DAIRY SHEEP DURING THE ANESTROUS PERIOD

Abstract. The aim of the research was to investigate the possibility of estrus induction in dairy sheep during the anestrous period using various hormonal preparations and finding out the effectiveness of this biotechnological method.

Experiments on the estrus induction were conducted in the Nikolaev M.I. peasant farm of Krymsky region, Krasnodar Krai. The object of the research was lactating and dry sheep of the dairy breed "Lacaune" with a total number of 332 heads between the ages of 2.5 and 5 years.

It is shown that in the anestrous season (from February to August) in the climatic conditions of the Krasnodar Krai with sufficiently high effectiveness (54.3-63.2%), it is possible to induce estrus in the Lacaune dairy sheep breed. In this case, the object of stimulating estrus can be both lactating and dry females. High fertility (75.0%) in sheep with induced estrus was established in the summer period. In the winter and spring seasons, this figure was 69.3% and 60.7%, respectively. The greatest effectiveness of estrus induction was established during hormonal stimulation of sheep during the summer period when the largest number of lambs (0.71) per one ewe was received. In the spring and winter seasons, this indicator was 0.37 and 0.58, respectively.

Effective technology of hormonal estrus induction in the unsexual season allows for the planned reproduction of dairy sheep throughout the year for year-round rhythmic milk production.

Key words: dairy ewes, anestrous period, estrus cycle, hormone therapy, estrus induction, progestagens, insemination, fertility.

Introduction. Sheep are polycyclic animals with a pronounced sexual season. Outstanding scientist A.I. Lopyrin believed that in the northern hemisphere, sheep start to be tupping in 8-10 weeks after the longest day of light. Therefore, the sexual (estrus) season in almost all sheep breeds occurs in autumn, as a rule, from September to December [1]. At this time, they are inseminated, while the period of mass young stock production falls on February-May. In economic conditions, when the most important product of sheep farming was high-quality wool, which was sheared once a year, the seasonality of sheep breeding was not decisive. Currently, in connection with the transfer of sheep farming to the priority meat direction and the initiation of dairy sheep farming in the country, the biologically determined seasonality of sheep breeding has become a critical point limiting the increase in the efficiency of the industry. This is due to the fact that the market requires the receipt of meat and dairy products and their delivery throughout the year [2, 3]. Accordingly, the technology for year-round sheep reproduction is needed.

Scientists and experts all over the world try to overcome the biological barrier to the seasonality of sheep breeding. As a rule, they are divided into two directions. The first is sheep breeding improvement in

order to ensure polyestrous, i.e. the ability to be tupping and become fertile at any time of the year was a genetically determined trait. Such fairly common breeds as the Romanov, Il-de-France, Dorper, Finnish Landrace, East Frisian, Merinoland and, partly, Dorset have practically no breeding season. However, the process of breeding polyestrous sheep breeds is long and it is not always possible to get a predictable result. In addition, even in polyestrous sheep breeds, there is significant variability of cyclicity in the seasons of the year. [4].

The second direction is the use of various factors of external influence on the female's organism, based on a detailed knowledge of the mechanisms of the neurohumoral regulation of the reproductive function in sheep.

In order to induce estrus in sheep during the anestrous period, two methods are used - light and hormonal, each of which has its own advantages and disadvantages, determining the appropriateness and extent of their use in specific economic conditions.

The light method of estrus induction in sheep is based on the fact that the hypothalamic centers that regulate the release of gonadotropic hormones are adapted to light effects and are stimulated by the natural light difference when changing a long summer day to a shorter autumn one. Creating a so-called "artificial autumn", as shown by numerous experiments, stimulates estrus in some sheep [5]. However, the majority of authors agree that the number of sheep that have been tupping varies widely (from 16 to 80%), excluding the predicted and successful application of this method with a practically significant result. [1].

The classic method of estrus induction in the off-season is the induction of estrus with the help of hormonal drugs, based on the prolongation of the luteal phase of the estrus by progestagens [1, 6]. Some authors mistakenly believe that "progesterone, affecting the pituitary gonadotropic cells, stimulates the synthesis and release of follicle-stimulating hormone (FSH) and luteinizing hormone (LH), which are involved in the regulation of ovarian function and the formation of follicles" [4]. On the contrary, progestagens inhibit the secretory function of the hypothalamus and the release of releasing hormone, which leads to suppression of the function of the posterior pituitary and release of follicle-stimulating hormone. Low concentration of FSH and LH in the peripheral blood leads to inhibition of the generative function of the ovaries, respectively, the growth and development of follicles are suppressed. As a result of hormonal treatment with progestagens, in animals, the stages of the estrus cycle are equalized, and after their action ceases, the physiological status of the generative apparatus of the females comes close to the post-luteal (or preovulatory) phase of the estrus cycle, the gonadotropic cells of the posterior lobe of hypophysis are activated, and the growth and maturation of the follicles in the ovaries are enhanced in all treated animals and, accordingly, animals are simultaneously tupping [7-9].

As noted above, year-round rhythmic milk production is the main technological aspect of effective dairy sheep farming. This requires the breeding of dairy sheep in both the estrous and anestrous periods. However, for different breeds of sheep, their age and physiological state, the adopted technology of production in different climatic conditions, hormonal treatment to induce estrus and its effectiveness may vary significantly [8,9,10]. In this regard, the study of the possibility of estrus induction in dairy sheep and finding out the effectiveness of this biologically-inspired technique is relevant, which determined the aim of this investigation.

Materials and research methods. Own experiments on the estrus induction were conducted in the Nikolaev M.I. peasant farm of the Krymsky district of Krasnodar Krai (figure 1). The object of the research was lactating and dry sheep of the dairy breed "Lacaune". For the induction of estrus, several drugs were used, injected and intravaginally. For the treatment of one group of sheep, the common drug “progesterone” was used, which was administered intramuscularly at a dose of 0.7 ml for 11 days. For hormonal treatment of two other groups of sheep, intravaginal pessaries of domestic production were used, impregnated with AMOL, the active ingredient of which is the synthetic analogue of progesterone 17a-mepregenol acetate and imported Syncro-part pessaries (France), containing 30 mg of active ingredient fluogestone acetate (Flugestone acetatum).

In all groups of sheep, to stimulate the growth of follicles in the ovaries after the removal of the progestogenic preparation, pregnant-mare serum (PMS) (Syncro-part Pmsg 6000me France) and the Follimag drug were used in different doses. Studies were conducted from February to August. In order to determine the impact of the season on the effectiveness of estrus induction, experiments with a certain degree of conditionality were divided into winter (February-March), spring (April-May) and summer (June-August).

Figure 1 – Interior design of the premises for the maintenance and milking of the Lacaune breed sheep in Nikolaev M.I. peasant farm of Krasnodar Krai

The methodological features of individual experiments are given in the description of experimental results.

Results and discussion.

Experiment 1. The first experiment was conducted in February-March 2017. 3 groups of animals were formed.

In the first group of sheep (lactating ewes in the amount of 85 heads), pessaries impregnated with the AMOL preparation at a dose of 30 mg AS were intravaginally administered for 12 days. After sponge removal, all the animals were intramuscularly injected with PMS at a dose of 600 units.

The second group of sheep was formed from dry ewes in the number of 51 heads, which were treated similarly to group 1.

For the treatment of the third group sheep, also consisting of dry females (n = 17), the "progesterone" drug was used, which was administered intramuscularly at a dose of 0.7 ml for 11 days. After treatment with progestagens in order to stimulate folliculogenesis, the Follimag drug was administered once intramuscularly at a dose of 500 units.

In all groups of sheep, sampling in estrus was not carried out, insemination of sheep was conducted after 55 hours, using freshly obtained semen for quality that meets the minimum requirements of the "Manual on the operating procedures of organizations of artificial insemination and transplantation of embryos of farm animals" (M., 2000).

The following results were attained. In the first group, 61 sheep were fertilized (71.2%), 62 lambs were born (fertility - 101.6%). In the second group, 34 sheep (66.6%) were fertilized, from which 36 lambs were obtained (fertility - 105.8%). In the third group, insemination performance was 64.7% (11 out of 17 inseminated sheep were fertilized), from which 11 lambs were obtained (fertility 100%).

Thus, in winter, out of 153 treated females, 106 sheep were fertilized fruitfully (average fertility 69.3%), from which 109 lambs were obtained (average fertility 102.8%). There was no significant difference in fertility between lactating and dry ewes. The average number of lambs obtained per 1 treated sheep was 0.71.

Experiment 2. The second experiment was performed in April - May 2017. Two groups of sheep were formed.

The first group, consisting of lactating ewes (n = 58), was treated with the intravaginal injection of sponges impregnated with the AMOL preparation at a dose of 30 mg AS. The intravaginal exposure period of progestagens was 12 days.

The second group ewes (dry ewe, n = 45) were also intravaginally injected for 12 days with Syncro-part pessaries (France) containing 30 mg of active substance of fluogestone acetate (Flugestone acetatum).

After removing the sponges, all animals were intramuscularly injected one time: in the first group, PMS was administered at a dose of 600 units. (Syncro-part Pmsg 6000me France), in the second group - "Follimag" at a dose of 500 units.

In order to carefully establish the estrus-inducing effect of the drugs used in this experiment, after the administration of gonadotropin, sheep were controlled for the state of estrus. To do this, with the help of sheep-samplers with tied up aprons, sheep were selected in the estrus. For a more accurate determination of the estrus onset, the sample was carried out every 4 hours.

The following results were obtained.

In the first group of 58 sheep, during the first 20 hours after the PMS injection, no one ewe has been tupping. After sampling after 24 hours, 2 heads in estrus were selected, after 28 hours - 3 heads, after 32 hours - 6 heads, after 36 hours - 10 heads, after 40 hours - 11 heads, after 44 hours - 2 heads. Sampling after 48 hours and further till insemination, which was carried out 55-56 hours after the PMS administration, did not give positive results.

Therefore, in the first group of 58 lactating ewes, after progestagens + PMS treatment, 34 sheep were tupping within 44 hours (58.6%). At the same time, of these 34 animals, 27 sheep (79.4%) have been tupping in a period from 28 to 40 hours after the gonadotropin administration.

All animals in the estrus were inseminated with freshly obtained semen with a mobility of 7-8 points, a concentration of 2.6-2.8 billion/ml. Of the inseminated 34 heads, 22 sheep lambed (fertilization rate was 64.7%). 24 lambs were born (fertility was 109.1%).

In such a way, analyzing the results obtained in the first group of animals, we came to the conclusion that, out of 58 lactating ewes, after the estrus induction in spring, 22 heads were fruitfully inseminated (37.9%), with a yield of 0.41 lamb per 1 treated ewe.

In the second group of 45 sheep, within the first 24 hours after the PMS injection, no one single ewe was tupping. After sampling, after 28 hours, 3 blissom sheep were selected, after 32 hours - 3 heads, after 36 hours - 13 heads, after 40 hours - 2 heads, after 44 hours - 1 head. As in the first group, 48 hours later, before insemination, which was carried out 55-56 hours after the Follimag administration, the sample did not reveal blissom ewes.

In this group of 45 dry ewes, after treatment with progestogen + Follimag, 22 heads were tupping within 44 hours. (48.9%). At the same time, out of these selected 22 sheep in estrus, 21 animals (95.5%) have been tupping between 28 and 40 hours after administration of gonadotropin.

As in the first group, all the sheep in the estrus were inseminated with freshly obtained semen with a mobility of 8–9 points, a concentration of 2.8–3.0 billion/ml. Among 22 animals, 12 sheep lambed (fertilization rate was 54.5%). 14 lambs were born (fertility was 116.6%).

In such a manner, in the second group of animals of 45 dry ewes, after the estrus induction in the spring period, 12 heads were fruitfully inseminated (26.6%), with a yield of 0.31 lamb per 1 treated ewe.

For a more objective assessment of the effectiveness of estrus induction in sheep in spring, we found it possible to summarize the data obtained for two groups and to derive the averages. Of 103 estrus-induced ewes, estrus was recorded in 56 animals (54.3%), of which, after insemination, 34 sheep lambed (fertility rate was 60.7%). 38 lambs were obtained (fertility was 111.8% for a sheep that gave birth, or 65.5% for an inseminated sheep). The estrus induction in the spring season resulted in 0.37 lamb per treated sheep.

Experiment 3. The third experiment was conducted in June-August 2017. As in the second experiment, 2 groups of sheep were formed.

The first group, consisting of lactating ewes (n=36), was treated with the intravaginal injection of sponges impregnated with the “AMOL” at a dose of 30 mg AS. The progestagens exposure period was 14 days.

The second group of ewes (dry ewe, n = 40) was also intravaginally injected for 14 days with pessaries Syncro-part (France) containing 30 mg of fluogestone acetate (Flugestone acetatum).

After sponge removal, all animals were injected intramuscularly with gonadotropin: in the first group, PMS at a dose of 600 units. (Syncro-part Pmsg 6000me France), in the second group - “Follimag” at a dose of 400 units.

In the third experiment, as in the second one, in order to accurately determine the stimulating effect of the used drugs after the gonadotropin administration, sheep were sampled into the estrus state with the help of sheep samplers with tied up aprons. To accurately determine the onset of estrus, sampling was performed every 4 hours.

In this experiment, the following results were obtained.

In the first group of sheep, during the first 20 hours after the PMS injection, no one single ewe was tupping. After sampling after 24 hours, 1 head in estrus was selected, after 28 hours - 2 heads, after 32 hours - 7 heads, after 36 hours - 6 heads, after 40 hours - 5 heads after 44 hours and then before insemination, which was carried out 55-56 hours after the PMS administration, ewes in estrus were not detected.

Thus, in the first group of 36 sheep, after estrus induction, within 40 hours, 21 heads were tupping (58.3%). At the same time of these 21 heads, in the time period from 28 to 40 hours, estrus was induced in 20 sheep (95.2%) after the PMS administration. All animals in estrus were inseminated with freshly obtained semen with a mobility of 8-8.5 points, a concentration of 2.5-3.0 billion/ml. Among 21 sheep, 15 animals lambed (fertility rate was 71.4%). 18 lambs were obtained (fertility was 120.0%). Consequently, in the first group of animals of 36 lactating ewes, after the induction of estrus in the summer season, 15 of them were fruitfully inseminated (41.6%), in this case, 0.5 lamb was obtained per 1 treated ewe.

In the second group, sheep in estrus were detected only 28 hours after the Follimag administration, when 2 heads in a state of estrus were selected. After 32 hours, 2 heads were selected, after 36 hours - 10 heads, after 40 hours - 12 heads, after 44 - 1 head. Sampling after 48 hours and further did not reveal any ewes in estrus. In this group of 40 dry ewes, after progestagens + Follimag treatment, 27 animals were tupping within 44 hours (67.5%). At the same time, 26 selected sheep in estrus (96.2%) were tupping in the time period from 28 to 44 hours after the Follimag use.

As in the first group, all the sheep in estrus were inseminated with freshly obtained semen, the quality of which corresponded to the minimum requirements of the Instruction. Of 27 sheep, 21 animals lambed (fertilization rate was 77.7%). 26 lambs were born (fertility was 123.8%).

Thus, in the second group of animals of 40 dry ewes, after the estrus induction in the summer, 21 animals were fruitfully inseminated (52.5%), 0.65 lamb was obtained per 1 treated ewe.

As in the second experiment, we think it possible to summarize the data obtained in two groups and to derive the averaged indicators of the effectiveness of estrus induction in sheep in the summer period. Of 76 estrus-induced ewes, estrus was detected in 48 heads (63.2%), of which, after insemination, 36 sheep lambed (fertilization rate - 75.0%). A total of 44 lambs were obtained (fertility - 122.2% in terms of lambed sheep, or 0.9 lamb per inseminated ewe) (figure 2). The induction of estrus in the summer season resulted in a yield of 0.58 lamb per treated sheep.

The summarized data for the three experiments are presented in table.

Figure 2 – Breeding, estimation of growth and development of lambs got from ewes with induced estrus in the summer period

Indicators of the effectiveness of estrus induction in "Lacaune" dairy sheep breed in the anestrous period by the seasons

Season	Treated sheep, heads	were tupping		Of them lambed		Number of lambs		Obtained lambs per 1 treated sheep
		heads	%	heads	%	heads	%	
Winter (February-March)	153	–*	–*	106	69.3	109	102.8	0.71
Spring (April-May)	103	54	54.3	34	60.7	38	111.8	0.37
Summer (June-August)	76	48	63.2	36	75.0	44	122.2	0.58
*Sampling of ewes in estrus was not carried out. Frontal insemination of all sheep was used 55 hours after finishing the treatment with progestagens.								

Comparison of data shows that the greatest fertilization rate (75.0%) in sheep with induced estrus was established in the summer period. In the winter and spring seasons, this figure was 69.3% and 60.7%, respectively.

Conclusions. The obtained results indicate that in the anestrous season (from February to August) in the climatic conditions of the Krasnodar Krai, with high efficiency, it is possible to induce estrus in the sheep of the Lacaune dairy breed. In this case, the object of estrus induction can be both lactating and dry females. At the same time, some of the resulted indicators, in our opinion, require more careful analysis in order to make possible adjustments in terms and estrus induction scheme in sheep.

The best fertilization rate of sheep during induced estrus was found in the summer period. The smaller value of this indicator was recorded in the winter period and the smallest - in the spring. However, it should be noted that in the winter experiment, no sheep in estrus were sampled, the frontal insemination was applied 55 hours after the administration of the gonadotropic hormone. In this regard, it is difficult to judge the effectiveness of estrus induction. At the same time, the main resulting indicator of our experiments in the winter period is 0.71 lamb per treated sheep. It can be assumed that a sufficiently high rate of fruitful insemination is connected with the fact that the winter group of animals could get a part of the sheep that were before the very end of the estrous season, in which, apparently, the natural hormonal background allowed to sensibilize the sexual centers with endogenous progesterone, and the administration of exogenous progesterone (vaginal pessaries) coincided with the preovulatory phase of the estrus cycle, which led to the manifestation of estrus and successful fertilization.

In a similar way, it is possible to explain the high fertilization rate of females (69.3%) in the summer experiment (June-August). It could also happen that sheep with regenerating natural hormonal background (especially in August) fell into this group, which, after saturation of the body with exogenous progesterone followed by stimulation of folliculogenesis in the ovaries, showed a full estrus which was confirmed by obtaining 0.58 lamb per one treated sheep.

The smallest number of lambs (0.37 per one treated ewe) was obtained in the spring (April-May) treatment period. Apparently, out of the estrous season, the function of the pituitary and ovaries under the influence of unfavorable environmental factors (prolonged light day, insolation) noticeably weakens and even under the influence of exogenous gonadotropic hormone (PMS or Follimag), the developing ovarian follicles do not reach ovulation maturity and undergo partial luteinization without rupture and release of the egg. It may also happen that during this period so-called abortive ovulation when the selected eggs are defective and cannot be fertilized.

It is believed that in lactating females, the effectiveness of estrus induction and fertilization rate is lower than in dry (non-lactating) animals [6]. In our experiments, this pattern is quite clearly seen only during hormonal treatment in the summer.

Obviously, the obtained data are preliminary, and their analysis is not exhaustive. Moreover, the reproductive indicators obtained in our experiments are significantly lower than in the experiments of other authors [6]. In part, this can be explained by the fact that the data on fertility in these authors are given not by lambing, but by the early diagnosis of pregnancy of ewe by the ultrasound method, i.e. without taking into account possible fetal mortality. It is also possible the impact that the experiments were conducted in other climatic conditions and with other sheep breeds using various induction schemes [11-15]. For a more complete analysis and justification of the laws obtained, further research is needed, first of all, of the natural hormonal background in sheep of different age groups and physiological state in the context of the seasons of the year. Selective laparoscopy (using endoscopic equipment) to assess the state of the internal genital organs in treated animals could be a good diagnostic test to determine the effectiveness of estrus induction.

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АНЭСТРАЛЬДЫ КЕЗЕҢДЕ СҮТТІ ҚОЙЛАРДЫҢ ЭСТРУС ИНДУКЦИЯСЫ

Аннотация. Зерттеудің мақсаты әртүрлі гормоналды препараттарды қолдану арқылы анестралдық кезеңде сүт қойларында сүтқоректілерді индукциялау мүмкіндігін зерттеу және биотехнологиялық әдістердің тиімділігін анықтау болды.

Жыныстық күйіт ынталандыру жөніндегі эксперименттер Қырым аймағы Краснодар өлкесінің. Николаев М.И. Шаруа (фермер) шаруашылығында өтті. Зерттеудің нысаны 2,5 жылдан 5 жылға дейінгі кезеңде жалпы 332 басы бар сүт тұқымының «Лакон» ("Lacaune") лактация кезеңінде және суалу кезеңіндегі қойлары болды.

Краснодар өлкесінің климаттық жағдайында анэстральдық маусымда (ақпаннан тамызға дейін) жеткілікті жоғары тиімділікпен (54,3-63,2%) сүт қойларында «Лакон» күйіт табуға болады деп көрсетілген. Бұл жағдайда эструсты ынталандыратын зат сүтті және суалу кезеңіндегі аналық болуы мүмкін.. Жазғы кезеңде индуцирленген күйіт кезде қойлардағы жоғары өнімділік (75,0%) белгіленді. Қысқы және көктемгі мезгілдерде бұл көрсеткіш тиісінше 69,3% және 60,7% құрады. Күйітті ынталандырудың ең тиімділігі жазғы кезеңде қойлардың гормоналды ынталандыруы кезінде анықталды, ол бір қойға ең көп қозы (0,71) алынды. Көктемгі-қысқы мезгілдерде бұл көрсеткіш тиісінше 0,37 және 0,58 құрады.

Мерзімді кезеңде эструстың гормоналды индукциясының тиімді технологиясы жыл бойына ритмді сүтті өндіруге сүт қойларын жоспарлы көбейтуге мүмкіндік береді.

Түйін сөздер: сүт аналықтары, анэстральдық кезеңі, эстрогендік цикл, гормондық терапия, эструс индукциясы, прогестогендер, ұрықтандыру, өнімділік.

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ИНДУКЦИЯ ЭСТРУСА У МОЛОЧНЫХ ОВЕЦ В АНЭСТРАЛЬНЫЙ ПЕРИОД

Аннотация. Целью исследования являлось изучение возможности индукции эструса у молочных овец в анэстральный период применением различных гормональных препаратов и выяснение эффективности этого биотехнологического приема.

Эксперименты по стимуляции половой охоты проведены в КФХ Николаев М.И. Крымского района Краснодарского края. Объектом исследований были лактирующие и сухостойные овцы молочной породы «Лакон» ("Lacaune") общей численностью 332 гол. в возрасте от 2,5 до 5 лет.

Показано, что в анэстральный сезон (с февраля по август) в природно-климатических условиях Краснодарского края с достаточно высокой эффективностью (54,3-63,2%) можно индуцировать половую охоту у овец молочной породы «Лакон». При этом объектом стимулирования эструса могут быть как лактирующие, так и сухостойные самки. Высокая оплодотворяемость (75,0%) у овец с индуцированной охотой установлена в летний период. В зимний и весенний сезоны этот показатель составил соответственно 69,3% и 60,7%. Наибольшая эффективность вызывания половой охоты установлена при гормональной стимуляции овец в летний период, когда было получено наибольшее число ягнят (0,71) на одну обработанную овцематку. В весенний и зимний сезоны этот показатель составил соответственно 0,37 и 0,58.

Эффективная технология гормонального индуцирования эструса в неполовой сезон позволяет проводить плановое размножение молочных овец в течение всего года для круглогодового ритмичного получения молока.

Ключевые слова: молочные овцематки, анэстральный период, эстральный цикл, гормональная терапия, индукция эструса, прогестогены, осеменение, плодовитость.

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