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Morphological profile of the ovaries of high-yielding cows on day 0 of the induced sexual cycle

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Article's History: Received: 15.04.2023 Revised: 15.06.2023 Accepted: 10.07.2023 **Abstract**. An important part of reproduction management programmes on dairy farms is the detection of infertile cows and early re-insemination to achieve higher rates of pregnancy. The purpose of the study was to investigate the features of gonadal morphogenesis in high-yielding cows in the conditions of industrial keeping technology on day 0 of the sexual cycle *in vivo* in real time. Structural, comparative, and statistical methods were used, as well as biotechnical methods of palpation cyclic examination of ovaries with differential diagnosis of morphophysiological or pathomorphological

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indicators. The results of diagnostics of the ovaries of dairy cows (n=128) on day 0 of the induced cycle detected by step activity sensors showed a significant structural polymorphism of these organs of the reproductive system. It was found: from 66.67% to 30.77% of cows with a physiological course of the follicular phase of the cycle, i.e., on day 0, a preovulatory follicle or the beginning of ovulation on the ovaries (subject to a decrease in the number of cows suitable for insemination with increasing lactation duration from 50-75 days to 211-306 days (P<0.001). Hypogonadism was diagnosed in 19.45% of cows, and in 61.11% – ovarian dysfunctions with the manifestation of cystic follicular degeneration; 19.44% of cows at the time of the study had irreversible chronic adhesive processes of the genital tract tissues in the "ovary+oviduct" area (ovophoritis and ovosalpingitis in latent form), which allowed for pregnancy to occur, with a pronounced tendency to increase this gonadopathy with an increase in lactation (respectively: 50-75 days - 0.00%; 76-160 days - 22.22%; 161-210 days - 22.22%; 211 and more -33.33%; p<0.05). The trend of influence of subclinical disorders in the metabolism of cows of the experimental herd on the indicators of the biochemical composition of blood serum and the symptom of ovarian dysfunctions was investigated. Thus, the *in vivo* morphological and pathological profile of the ovaries of high-yielding cows on day 0 of the sexual cycle was established, which allowed optimising the artificial insemination regime and reduce economically nonprofitless sperm consumption. The results can be used in the practical work of veterinary medicine doctors, researchers, and applicants for higher education in veterinary and biological science

Keywords: dairy cows; sexual cycle; ovaries; differential palpation diagnostics; follicles; hypogonadism; ovary dysfunction; gonadopathy

INTRODUCTION

The efficiency of reproduction of breeding stock remains an urgent issue for all dairy enterprises in the world and tends to expand the problem of reproduction by increasing milk productivity due to the biological conflict between the genetically fixed ability of cows to produce a large amount of milk and the physiological needs of the cows. Commercial milk farms in the United States, Western Europe, and Ukraine annually cull 27-34% of cows, mainly due to infertility (García-Ispierto et al., 2019). These losses can only be eliminated by the intensive introduction of heifers into the herd. However, they are much more likely than cows to have problems with insemination and fertilisation after calving. Bardos et al. (2020) suggest that the shortage of repair livestock of the herd can be eliminated by modern biotechnical means (changing the sex of the calves towards the dominance of heifers, selection of embryos by sex).

According to Shebanin (2021), Sidashova *et al.* (2020), Borshch *et al.* (2020), in Ukraine, the main suppliers of commercial milk are large industrial complexes, where there is a high concentration of modern dairy cow breeds, which are characterised by a high genetic productivity potential.

According to modern biological concepts, the reproductive function pertains to healthy animals that exist in comfortable conditions of keeping. The cows' and heifers' reproductive system has physiological changes during the sexual cycle, the duration of which is on average 21 days and ranges from 16 to 28 days (Roman *et al.*, 2020). The day of growth of the dominant follicle and its ovulation with the release of the egg into the oviduct is considered to be day 0 of the cycle, or in a number of sources – day 1 (Denis-Robichaud *et al.*, 2018). A large number of resources in countries with developed cattle breeding have been devoted to the study of the specific features of reproductive ethology and physiology of cows in previous years, but due to significant changes in the industrial technology of milk production, rapid genetic progress of large-scale breeding of dairy cattle, the conditions of keeping and milking of modern dairy livestock have changed significantly. This could not fail to have an effect on the features of the reproductive function of cows, which began to form in the artificially created environment of industrial farms, which significantly differs from the natural environment which was common to cattle as a species due to long evolution (Ispada *et al.*, 2018; Danchuk *et al.*, 2020).

Modern technologies are designed for no grazing and tethered cows keeping in a confined space. From the perspective of a market economy, this is quite justified. However, they do not consider the biological features of cattle. As a result of separation from the natural environment, lack of living space, and excessive exploitation the productive life of cows is reduced to 1.5-2 lactations. Early culling of cows, regardless of their age and breeding value, occurs in 70-80% of cases due to irreversible pathologies of the reproduction organs. In the current conditions, the task is not only to preserve the livestock, but also to fully use its reproductive potential.

According to Ong *et al.* (2022), a long-term study of the sexual cycle of cows in countries with a developed dairy cattle breeding has contributed to a significant improvement in reproductive biotechniqes including methodological approaches to the selection of cows and heifers for insemination. However, indicators of the size and morphological features of the gonads of the sexual cycle in cows or heifers remain a factor limiting the number of animals suitable for biotechnical procedures. At the same time, sources focus on improving the hormonal treatment schemes of animals.

Hansen (2019), Rodrigues *et al.* (2019) studied ovarian pathology in cows as the main cause of reduced fertility in dairy cattle and factors conducing to ovarian cysts. Monget *et al.* (2019) noted that the occurrence of cysts is caused by insufficient secretion of luteinising hormone during the arousal phase of the sexual cycle due to the inability of the hypothalamic-pituitary system to respond to estrogenic stimulation through positive feedback mechanisms.

Based on the researches, Kim (2018) found that follicular cysts are transient formations after loss of functional activity that undergo reverse development and are replaced by a new dominant ovarian structure that ovulates or transforms into a new cyst (the phenomenon of cyst change). The role of inflammatory processes in the tissues of the reproductive organs of cows in the disruption of ovarian function and cystogenesis was determined.

The breeding progress of dairy herds depends on the biological low fertility of cows and the duration of pregnancy, in addition, it is limited by the number and quality of gametes and the efficiency of reproduction of the herd. The purpose of this study was to investigate the features of gonad morphogenesis of highly productive cows in the conditions of industrial keeping technology on day 0 of the sexual cycle *in vivo* in real time.

MATERIALS AND METHODS

The study was performed in 2021-2022 on the number of dairy cows (imported purebred Danish Holsteins of 1-2 lactation), which belonged to a dairy farm that was a part of one of the agricultural holdings of Ukraine. The livestock was kept in a large farm in light hangar-type buildings that met modern zoohygienic requirements (climate control equipment). Dairy cows were kept in sections (100-120 animal units) with individual places for rest (equipped with rubber mats with an additional provision of sawdust bedding), for three times milking in an automated milking parlour with individual productivity accounting (electronic database "DairyPlan"). The animals received fully mixed fodder all year round, the content and ratio of feed nutrients corresponded to zootechnical standards for highly productive cows in accordance with the physiological state of the animals (9,450 kg of milk for 305 days of lactation).

Detection of cows for artificial insemination was carried out using the computer program for monitoring animal behaviour ("AfiFarm") using sensors attached to the hind limb of the animal. Considering a number of indicators of the step activity of cows, an integrated parameter corresponding to day 0 of the sexual cycle of cows was displayed on the monitor – an indicator of sexual arousal (heat) in the graphic image of the "reproduction windows", which showed the beginning, duration, and end of the period of sexual arousal of the cows, and the

optimal time of fertilisation (insemination for a single spermodose into the cow's uterine cervix). The "reproduction window" had duration of 8 hours, which allowed the staff to perform artificial insemination for cows indicated by sensors at a comfortable time of the working day between milking individual groups of the herd.

To optimise the schedule of artificial insemination of groups of animals kept in different buildings, an additional study of detected cows on day 0 of the induced sexual cycle before insemination was performed by differential palpation diagnostics of morphofunctional and morphometric features of the ovaries, using a modified cyclic technique provided in sourses (Sidashova, 2017; Sidashova, 2021). Induction of cows` sexual activity in was performed according to the standard OvSyng hormone synchronisation protocol (day 0 – Ovarelin 2.0 ml intramuscularly; day 7 – Enzaprost 5.0 ml intramuscularly; after 56 hours – Ovarelin 2.0 ml intramuscularly; after 16 hours – artificial insemination on day 10 day of the scheme).

The study used comparative structural, statistical, and biotechnical methods. The test samples were the cows' ovaries of on day 0 of the induced sexual cycle, which were examined in vivo using a differential palpation technique, considering the indicators of computer tracking of the step activity of cows. During the rectal palpation of the ovaries we revealed the cows is in libido for artificial insemination according to the following parameters, which are given in detail in the previous studies of the authors: the size of the gonads conformity in length-width-thickness of ovomorphometry and morphological formations of the gonad to the species norm, cm; the presence of signs of a preovulatory dominant follicle or ovulated follicle on one or both ovaries for typical tactile symptoms, which is shown in our previous studies (Roman et al., 2020). In the presence of symptoms of a violation of the ovarian follicular layer, characteristic signs of pathologies were noted: degenerative changes in the follicles (single or multiple follicular cysts).

Separately, the hypotrophy of the ovaries was noted, when their size and palpatory structure did not correspond to the species reference indicators, which characterised hypogonadism of both ovaries. The hypotrophy of one of the gonads, while compliancing to the normal morphology of the second, was determined to be the temporary inverse phenomenon of functional asymmetry (Roman *et al.*, 2020). Differential palpation diagnostics determined irreversible tissue changes in the ovaries and oviducts that were characteristic of animals with chronic symptomatic infertility that occurred due to long-term gynecological diseases, often latent (chronic oophoritis, salpingitis, ovosalpingitis (Roman *et al.*, 2020).

All the data obtained were recorded in a specially developed accounting form, followed by biometric processing of generalised data and determination of the level of probability according to the Student's t-test. Experiments conducted on animals do not contradict the current legislation of Ukraine (Article 26 of the Law of Ukraine 5456-VI of 16.10.2012 "On the protection of animals from cruelty") and the "General ethical principles of animal experiments" adopted by the First National Congress on Bioethics (2001) and international bioethical standards (materials of the IV European Convention for the protection of vertebrates used for experimental and other purposes, Strasbourg, 1985) (Festing & Wilkinson, 2007; Simmonds, 2017; Kabene & Baadel, 2019).

RESULTS AND DISCUSSION

The examined group of high-yielding unfertilised cows (n=128) was structured according to the duration of

lactation: in the first subgroup there were animals from 50 to 75 days after calving that had not yet been inseminated; in the second and third (76-160 and 161-210 days, respectively) cows that already had a history of 1 to 3 ineffective inseminations, in the third (from 211 to 306 days of lactation) – cows with numerous infertile inseminations. Preliminary vaginal and ultrasound examination of the genitals of these groups of cows did not reveal obvious signs of acute or subacute inflammation of the genitals, but all livestock had clinical signs of damage to mucous membranes by chronic infectious rhinotracheitis-pustular vulvovaginitis of mixed etiology (PVV-IRT). Table 1 shows the dynamics of increasing the number of pathological changes in the ovaries of unfertilised cows with an increase in the lactation period.

Table 1. Morphological profile of the ovarian status of Holstein cows on day 0 of the induced sexual cycle with different lactation durations, n=128

Indicators	Lactation period, days				
	50-75	76-160	161-210	211 or more	
Number of cows examined, units	24	44	34	26	
Average lactation period, days (m±m)	56.63±1.99**	121.52±4.24**	182.82±2.28**	262.27±5.36**	
Number of cows with morphofunctional ovarian state on day 0 of the cycle, units	16	26	16	8	
%	66.67	59.09	47.06	30.77	
Number of cows with detected gonadopathies, units	8	18	18	18	
%	33.33	40.91	52.94	69.23	

Note: ** – *P*<0.001

Source: compiled by the authors

If by day 75 after calving, 66.67% of cows had a dominant preovulatory or already ovulating follicle on one of the ovaries on day 0 of the induced sexual cycle, which met the requirements for artificial insemination of the cows, then with each subsequent period the

number of animals with the functional state of the ovaries significantly decreased, which is graphically shown in Figure 1. After cows reach 160 lactation days, the ratio of cows with morphofunctional state of ovaries to pathologies becomes critically low.



Figure 1. Effect of increasing the lactation period on the prevalence of ovarian pathologies in high-yielding unfertilised cows, n=128

The identified situation indicated the need for a more detailed diagnosis of the causes of longterm infertility in highly productive cows that did not have obvious clinical symptoms of gynecological diseases. Differential palpation diagnostics *in vivo* allowed structuring the nature of anatomical and morphological changes in the gonads and palpatory state of the anatomical area "ovary+oviduct" in accordance with the lateral topography, which is shown in Table 2.

1	Lactation period, days			
Indicators	50-75	76-160	161-210	211 or more
Number of cows examined, units	24	44	34	26
Among them, gonadopathies were detected, units/100%	8	18	18	18
Structured nature of gonadopathies:				
• Bilateral hypogonadism, units	0	6	7	1
%	0.00	33.34	38.89	5.56
• Follicular cystic degenerations, units	8	8	7	11
%	100.00	44.44	38.89	61.11
Chronic adhesive processes "ovary+oviduct", units	0	4	4	6
%	0.00	22.22*	22.22*	33.33*

 Table 2. Results of detailed palpation diagnosis of gonadopathies on day 0 of the sexual cycle of high-yielding cows for different lactation periods, n=128

Note: * – P<0.01

Source: compiled by the authors

By differential palpation diagnostics of the sexual organs of cows within the anatomical area "ovary+oviduct", cases of irreversible morphological failures in tissues caused by chronic inflammatory adhesive processes with symptoms of chronic oophoritis and ovosalpingitis were established, which were the consequences of prolonged inflammation of the mucous membranes of the reproductive organs, which were usually externally asymptomatic and detected only by ineffective multiple inseminations. Palpation diagnostics of these gynecological pathologies reveal them already in a form unsuitable for rehabilitation of the reproductive function of cows, and other instrumental methods are generally unsuitable for diagnosis in vivo (Sidashova et al., 2020). The study showed that with the growth of lactation, the number of detected oophoritis and salpingitis increased from 22.22% to 33.33% with a highly significant correlation for other indicators of gonadopathies (P<0.01). The established trend required timely differential diagnosis of unfertilised cows to prevent sperm production costs for infertile inseminations.

Diagnostic studies have shown that cows with a technologically appropriate lactation term (from 50 to 75 days) among ovarian pathologies had only signs of follicular cysts in the form of large single (rarely double) follicular cysts, anovulatory follicles, or small multiple cystic follicles on one or both ovaries (in 33% of animals, which was presented in detail in the studies (Sidashova et al., 2021). With the extension of the lactation period, cows began to show signs of hypogonadism more often (18%), that is, hypotrophy in the tissues of both ovaries, which is logical, given the amount of nutrients that the lactating animal gives with milk daily. With an growth in the lactation period, there was an increase in signs of cystic degeneration in the ovarian follicular layer (or 42% of 26 animals of the last subgroup, which corresponded to 61.11% of cases of all pathologies in the subgroup) (Fig. 2). Due to the significant influence of paratypical (economic) factors on the level of reproduction of the dairy herd, the indicators of hypogonadism and follicular cysticity among the surveyed livestock did not have a significant correlation (P>0.05).



Figure 2. Prevalence of ovarian dysfunctions and ovarian pathologies in high-yielding unfertilised cows with different duration of the lactation period, n=62

Background examination of the biochemical composition of the blood serum of dairy cows of the experimental herd (Table 3) indicated the presence of subclinical signs of metabolic disorders in the body of lactating animals, which are very sensitive to an imbalance of nutrients due to a high level of metabolism and react significantly to a violation of the balance of the internal environment.

Indicators	Lactation period of more than	lim		
	40 days	min	max	Reference values
Total protein, g/l	74.40±1.17**	72	78	55-75
Albumin, g/l	36.00±0.55**	34	38	30-35.5
Globulin, g/l	38.40±1.36**	34	42	30-35
Protein coefficient, units.	0.94±0.04**	0.9	1.1	0.6-1.1
Urea, mmol/l	6.96±0.41**	6.2	8.5	2.8-5.8
Creatinine, mmol/l	100.00±11.81	86	116	45-140
AAT, u/l	100.00±11.48*	64	127	10-50
ALT, u/l	22.20±4.33*	7	33	10-40
AAT/ALT, units	6.50±2.71**	2.5	17	1.0-3.4
Alkaline phosphotase, u/l	78.06±8.76**	60.4	110.8	20-150
Glucose, mmol/l	2.12±0.06**	2.0	2.3	2.5-4.2
Calcium, mmol/l	1.88±0.04**	1.8	2.0	2.43-3.10
Carotene, mcg%	119.20±5.69*	100	131	275-965
Lipoproteins total, mg%	1,035.40±71.01**	833	1,254	400-800

Table 3. Indicators of the biochemical composition of blood serum of high-yielding dairy cows, n=8

Note: * - P<0.01, ** - P<0.001

Source: compiled by the authors

A generalised review of the data of morphometry and pathomorphology of the ovaries of dairy unfertilised cows for different lactation terms demonstrated the existing relationship between the functional or pathological state of the gonads and the content of individual nutrients in the blood serum, and, accordingly, in the daily diet of livestock. Numerous subclinical disorders in the metabolism of dairy cows, according to literature sources, could provoke an exacerbation of gynecological pathologies and dysfunctions of the sexual cycle, which resulted in numerous non-fertile inseminations. At the same time, the duration of the infertile period significantly worsened the morphofunctional characteristics of ovogenesis: in the period up to 75 days after calving, the number of cows with a functional state of ovarian morphology was twice as high as the number of animals with of various gonadopathies of various types, and in the subgroup of cows after 211 days of lactation, on the contrary, the number of cows with gonadal dysfunctions and pathologies was twice as high as the number of animals with a morphofunctional state of ovaries.

The established regularities of the development of ovarian morphogenesis during lactation indicate the need to increase the attention of veterinary medicine doctors to the differential diagnosis of the pathology of the reproductive organs of cows in the first half of lactation, and especially in the postpartum period, which is a resource for increasing the profitability of dairy production.

The reproduction of genetic resources in cattle breeding is based on the biological characteristics of cattle breeding. A retrospective analysis of the literature sources on ovarian morphogenesis has shown that not all aspects of this complex and multifaceted problem are fully revealed. The specifics of the morphological and pathomorphological profile of the ovaries of high-yielding cows on day 0 of the sexual cycle are insufficiently studied.

Thus, there is no consensus on the nature of the topography of physiological ovulation in single or multiple females. All these anatomical and physiological patterns directly affect on reproduction of productive animals. In addition, the likely influence of morphogenesis on the development of ovarian pathologies can be very important in solving issues of reproduction of not only farm animals, but also rare species. The key organs of the reproductive system, namely paired gonads, are the anatomical and physiological basis of technological processes in dairy and meat cattle breeding, so the understanding of ovarian morphogenesis and ways of its regulation determines the efficiency and the profitability of the livestock.

There is scare information on the influence of functional insufficiency on the intensity and nature of morphofunctional changes in the gonads. The influence of feeding and environmental factors on the biochemical composition of blood in different phases of the sexual cycle, both in normal and gynecological pathology, is not fully disclosed. The differential method used in this study for diagnosing the physiological or pathological state of the ovaries highlights the features of gonad morphogenesis of cows in industrial dairy cattle; the importance of this method is emphasised in data by Borshch et al. (2020). Based on the results obtained, differentiation of diagnostic indicators of morphofunctional ovarian structures in cows (n=423) that were synchronised prevented a significant level of ineffective artificial inseminations (pregnancy of experimental cows was 2.1 times higher). Similar results were obtained by Ault (2019).

Researches by Denis-Robichaud *et al.* (2018), indicate the need for monitoring the indicators of automatic sensors of sexual activity detection systems in cows, which increases the number of effective inseminations conducted. In addition, this research expands the methodological database of such studies, which can be implemented in real production conditions.

Studies by Cavallari de Castro *et al.* (2019) and Pascottini (2020) on the impact of chronic or acute and subacute gynecological diseases in lactating cows on fertility indicators emphasise versatile approaches to the diagnosis of pathologies of the reproductive system of cows. Results obtained by Appiah *et al.* (2020) indicate that the above data are debatable, which is explained by the impossibility of creating similar survey conditions at work, which are characterised by a variety of paratypical factors. Probably, the lack of a methodologically developed protocol for comparing indicators of ovarian pathomorphology of cows against the background of their anatomical species polymorphism causes inconsistency in the results of research in different sources.

Nowadays, researchers continue to discuss the main factors that provoke cystic follicle degeneration in dairy cows (Ong *et al.*, 2021), which indicates a lack of knowledge of the phenomenon. At the same time, there are reports in the literature about a steady trend towards an increase in the number of cystic ovarian diseases in cows, but practical experts state that there is no effective scheme for the prevention and therapy of animals with ovarian dysfunction (Nowicki *et al.*, 2017).

Cystic ovarian disease is polyetiological disease and has a common pathology; it is registered in 3.8-6.5% of infertile cows. The frequency of cysts is influenced by a number of factors: breed, stable maintenance, number of calving, duration of previous and stage of current lactation. Thus, ovarian cysts were registered 1.7 times more often in Ayrshire cows (6.5% vs. 3.8%) than in black-and-white cows (Monget *et al.*, 2020). Based on the conducted experimental studies, it was established that follicular cysts were the most common pathologies of the ovaries. They were registered in 33.33-42.31% of high-yielding cows and positively correlated with the duration of lactation. The results obtained do not differ from the data of Gonzalez Moreno *et al.* (2020).

Bomko *et al.* (2018), Grymak *et al.* (2020) indicate a significant influence on the phenomenon of ovarian dysfunctions of deviations in the optimal ratio of nutrients in the diet, provoking metabolic disorders in various body systems of dairy cows, including hormonal imbalance.

According to Mazur et al. (2020), glucogenesis is continuous in ruminants, unlike monogastric animals. Their need for glucose by 90% or more is provided by gluconeogenesis, which occurs mainly in the liver. These processes are actively regulated by the endocrine system, so there is hyperproduction of glucagon, somatotropin, and hypoproduction of insulin. This condition of the endocrine glands enhances lipomobilisation, lipolysis, proteolysis, ketogenesis and contributes to the development of liver dystrophy. With a long and massive intake of lipids in the liver, there is a deficiency of oxaloacetic acid, the source of which is glucose. The concentration of ketone bodies increases in blood serum (ketonuria), urine, and milk, which is expressed by a significant increase of triacylglycerols in the blood of such animals, rapid fat infiltration of haepatocytes and provokes the course of lipomobilisation syndrome, which is characterised by fatty liver dystrophy and other pathologies associated with reduced fertility in highly productive young cows (Malashka *et al.*, 2021).

A decrease in the functional state of the reproductive system during the lactation can also be affected by low levels of calcium (which is important for the contractile activity of the uterus), the content of which was on average 60-70% of the reference values in cows. At the same time, we established that the deficiency of precursors of fat-soluble vitamins had a noticeable negative effect on the secretion of sex hormones in cows.

Based on the experimental data, the carotene content in the blood serum of cows was 2.5-9.0 times less than the need, which was a prerequisite for an increase in the symptoms of ovarian hypotrophy, especially in the middle of lactation (33.34-38.89% of hypogonadism among all detected gonadopathies). The results obtained do not differ from the data of research by Rodriguez *et al.* (2020). Bomko *et al.* (2018) showed that optimising the composition of the diet of dairy of high-yielding cows for a number of ingredients, in particular, Zinc, Manganese, and Cobalt, and improving the balance of the ratio of nutrients in the diet led to an increase in the level of fertility of livestock.

Thus, the study of the morphology and function of the ovaries of cows and heifers remains at the level of comparing the morphometric parameters of the extracted organs (*in vitro*) or (*in vivo*) using hardware methods (ultrasound scanning) or palpation, which does not allow drawing conclusions about the quantitative and structural patterns of differentiation of generative and hormonal functions of the gonads. All current instructive and methodological guidances are based on generalised and averaged data on the functional activity of the ovaries as a pair of equivalent gonads.

CONCLUSIONS

Studies were conducted *in vivo* to establish morphological and pathomorphological profile of the ovaries of high-yielding cows on day 0 of the sexual cycle to optimise the timing of artificial insemination of cows and reduce economically nonprofitless costs of bull sperm.

Differential palpation diagnostics of high-yielding lactating cows *in vivo* revealed the actual morphological and functional structures of the ovaries on day 0 of the sexual cycle, which was characterised by significant polymorphism, associated both with the specifics of functional morphogenesis of gonads of cows in conditions of industrial keeping (66.67-30.77% of the examined animals) and the presence of reproductive pathology of various etiologies (33.33-69.23% of the studied cows at different lactation periods, respectively). sexual cycle of the lactation period, namely: on days 50-75, 66.67% of dairy cows had physiological morpho-functional ovarian structures; and on days 76-160, 161-210, and 211 or more – 59.09%; 47.06%, and 30.77%, respectively (P<0.001).

Based on the conducted experiments, it was established that follicular cystic degenerations were the most common pathologies of the ovaries of of high-yielding cows. Signs of ovarian cysts grew with an increase in the lactation term: from 33.33% at the beginning of lactation to 42.31% in the period after 211 days.

Differential palpapatory examination *in vivo* allowed diagnosing an increase in the number of chronic irreversible pathologies in of high-yielding non-inseminated cows in the ovary-oviduct area, manifested by adhesive inflammatory processes in the form of chronic ovosalpingitis (P<0.05). An influence of subclinical disorders in the metabolism of high-yielding cows on the development of ovarian dysfunctions was revealed. Identification of the causes and prevention of ovarian dysfunction is a prospect for further study.

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CONFLICT OF INTEREST

It was experimentally established that the morphofunctional state of the ovaries on day 0 of the induced

REFERENCES

- [1] Appiah, M.O., Wang, J., & Lu, W. (2020). Microflora in the reproductive tract of cattle: A review. *Agriculture*, 10(6), article number 232. doi: 10.3390/agriculture10060232.
- [2] Ault, T.B., Clemmons, B.A., Reese, S.T., Dantas, F.G., Franco, G.A., Smith, T.P.L., Edwards, J.L., Myer, Ph.R., & Pohler, K.G. (2019). Uterine and vaginal bacterial community diversity prior to artificial insemination between pregnant and nonpregnant postpartum cows. *Journal of Animal Science*, 97, 4298-4304. doi: 10.1093/jas/skz210.
- [3] Bardos, J., Fiorentino, D., Longman, R.E., & Paidas, M. (2020). Immunological role of the maternal uterine microbiome in pregnancy: Pregnancies pathologies and alterated microbiota. *Frontiers in Immunology*, 10, 2823-2823. doi: 10.3389/fimmu.2019.02823.
- [4] Bomko, V., Kropyvka, Yu., Bomko, L., Chernyuk, S., Kropyvka, S., & Gutyj, B. (2018). Effect of mixed ligand complexes of Zinc, Manganese, and Cobalt on the Manganese balance in high-yielding cows during first 100-days lactation. *Ukrainian Journal of Ecology*, 8(1), 420-425. doi: 10.15421/2018_230.
- [5] Borshch, O.O., Gutyj, B.V., Sobolev, O.I., Borshch, O.V., Ruban, S.Yu., Bilkevich, V.V., Dutka, V.R., Chernenko, O.M., Zhelavskyi, M.M., & Nahirniak, T. (2020). Adaptation strategy of different cow genotypes to the voluntary milking system. Ukrainian Journal of Ecology, 10(1), 145-150. doi: 10.15421/2020_23.
- [6] Cavallari de Castro, F., Leal, C.L.V., Roth, Z., & Hansen, P.J. (2019). Effects of melatonin on production of reactive oxygen species and developmental competence of bovine oocytes exposed to heat shock and oxidative stress during *in vitro* maturation. *Zygote*, 27(3), 180-186. doi: 10.1017/S0967199419000236.
- [7] Danchuk, O.V., Karposvkii, V.I., Tomchuk, V.A., Zhurenro, O.M., Bobrts'ka, O.M., & Trokoz, V.O. (2020). Temperament in cattle: A method of evaluation and main characteristics. *Neurophysiology*, 52, 73-79. doi: 10.1007/s11062-020-09853-6.
- [8] Denis-Robichaud, J., Cerri, R.L.A., Jones-Bitton, A., & LeBlanc, S.J. (2018). Performance of automated activity monitoring systems used in combination with timed artificial insemination compared to timed artificial insemination only in early lactation in dairy cows. *Journal of Dairy Science*, 101(1), 624-636. doi: 10.3168/ jds.2016-12256.
- [9] Festing, S., & Wilkinson, R. (2007). The ethics of animal research. Talking point on the use of animals in scientific research. EMBO Reports, 8, 526-530. doi: 10.1038/sj.embor.7400993.
- [10] García-Ispierto, I., De Rensis, F., Pérez-Salas, J., Nunes, J., Pradés, B., Serrano-Pérez, B., & López-Gatius, F. (2019). The GnRH analogue dephereline given in a fixed-time AI protocol. *Research in Veterinary Science*, 122, 170-174. <u>doi: 10.1016/j.rvsc.2018.11.020</u>.

- [11] Gonzalez Moreno, C., Torres Luque, A., Oliszewski, R., Rosa, R, & Otero, M.C. (2020). Characterization of native *Escherichia coli* populations from bovine vagina of healthy heifers and cows with postpartum uterine disease. *PLoS ONE*, 15, article number e0228294. doi: 10.1371/journal.pone.0228294.
- [12] Grymak, Y., Skoromna, O., Stadnytska, O., Sobolev, O., Gutyj, B., Shalovylo, S., Hachak, Y., Grabovska, O., Bushueva, I., Denys, G., Hudyma, V., Pakholkiv, N., Jarochovich, I., Nahirniak, T., Pavliv, O., Farionik, T., & Bratyuk, V. (2020). Influence of "Thireomagnile" and "Thyrioton" preparations on the antioxidant status of pregnant cows. *Ukrainian Journal of Ecology*, 10(1), 122-126. doi: 10.15421/2020_19.
- [13] Hansen, P.J. (2019). Reproductive physiology of the heat-stressed dairy cow: Implications for fertility and assisted reproduction. *Animal Reproduction*, 16(3), 497-507. doi: 10.21451/1984-3143-AR2019-0053.
- [14] Ispada, J., Rodrigues, T.A., Risolia, P.H.B., Lima, R.S., Gonçalves, D.R., Rettori, D., Nichi, M., Feitosa, W.B., & Paula-Lopes, F.F. (2018). Astaxanthin counteracts the effects of heat shock on the maturation of bovine oocytes. *Reproduction, Fertility and Development*, 30(9), 1169-1179. doi: 10.1071/RD17271.
- [15] Kabene, S., & Baadel, S. (2019). Bioethics: A look at animal testing in medicine and cosmetics in the UK. *Journal of Medical Ethics and History of Medicine*, 12, article number 15. <u>doi: 10.18502/jmehm.v12i15.1875</u>.
- [16] Kim, SJ. (2018). Patterns of ovarian changes associated with surge mode secretion of gonadotropin in dairy cows with cyclic estrous cycle. *Journal of Embryo Transfer*, 33(4), 297-304. <u>doi: 10.12750/JET.2018.33.4.297</u>.
- [17] Malashka, V.V., & Tumilovich, G.A. (2021). Ultrastructural organization of the liver in high-yielding cows with impaired substance metabolism. In *Actual problems of treatment and prevention of diseases of young people: Materials of international scientific and practical conference* (pp. 241-250). Vitebsk.
- [18] Mazur, N.P., Fedorovych, V.V., Fedorovych, E.I., Fedorovych, O.V., Bodnar, P.V., & Pakholkiv, N.I. (2020). Effect of morphological and biochemical blood composition on milk yield in Simmental breed cows of different production types. *Ukrainian Journal of Ecology*, 10(2), 61-67. doi: 10.15421/2020_110.
- [19] Monget, P., & Monniaux, D. (2019). <u>Growth factors and control of folliculogenesis</u>. *Journal of Reproduction and Fertility*, 49, 321-333.
- [20] Nowicki, A., Baranicki, W., Baryczka, A., & Janowski, T. (2017). Ovsyng protocol and modifications in the reproduction management of dairy cattle herds-an update. *Journal of Veterinary Research*, 61(3), 329-336. <u>doi: 10.1515/jvetres-2017-0043</u>.
- [21] Ong, C.T., Ross, E.M., Boe-Hansen, G., Turni, C., Hayes, B.J., Fordyce, G., & Tabor, A.E. (2021). Interrogating the bovine reproductive tract metagenomes using culture-independent approaches: A systematic review. *Animal Microbiome*, 3, article number 41. doi: 10.1186/s42523-021-00106-3.
- [22] Ong, C.T., Ross, E.M., Boe-Hansen, G., Turni, C., Hayes, B.J., Fordyce, G., & Tabor, A.E. (2022). Adaptive sampling during sequencing reveals the origins of the bovine reproductive tract microbiome across reproductive stages and sexes. *Scientific Reports*, 12, article number 15075. doi: 10.1038/s41598-022-19022-w.
- [23] Pascottini, O.B. (2020). Dynamics of uterine microbiota in postpartum dairy cows with clinical or subclinical endometritis. *Scientific Reports*, 10, article number 12353. <u>doi: 10.1038/s41598-020-69317-z</u>.
- [24] Rodrigues, T.A., Tuna, K.M., Alli, A.A., Tribulo, P., Hansen, P.J., Koh, J., & Paula-Lopes, F.F. (2019). Follicular fluid exosomes act on the bovine oocyte to improve oocyte competence to support development and survival to heat shock. *Reproduction, Fertility and Development*, 31(5), 888-897. doi: 10.1071/RD18450.
- [25] Roman, L., Broshkov, M., Popova, I. Hierdieva, A., Sidashova, S., Bogach, N., Ulizko, S., & Gutyj, B. (2020). Influence of ovarian follicular cysts on reproductive performance in the cattle of new Ukrainian red dairy breed. Ukrainian Journal of Ecology, 10(2), 426-434. doi: 10.15421/2020_119.
- [26] Roman, L., Sidashova, S., Danchuk, O., Popova, I., Levchenko, A., Chornyi, V., Bobritska, O., & Gutyj, B. (2020). Functional asymmetry in cattle ovaries and donor-recipients embryo. *Ukrainian Journal of Ecology*, 10(3), 139-146. doi: 10.15421/2020_147.
- [27] Shebanin, V.S. (2021). <u>Dual form of educational training of highly qualified specialists for the agricultural sector of Ukraine</u>. *Agro-Industrial Complex Economy*, 3, 5-14.
- [28] Sidashova, S.O., Gutyj, B.V., Khalak, V.I., & Humeny, O.G. (2020). Influence of complex action of probiotic and specific prophylaxis of associated mucosal diseases on some quantitative traits of dairy cattle performance. *Scientific Messenger of Lviv National University of Veterinary Medicine and Biotechnologies. Series: Veterinary sciences*, 22(97), 79-87. doi: 10.32718/nvlvet9714.
- [29] Sidashova, S.O., Popova, I.M., Travetsky, M.A., & Khotsenko, A.V. (2021). The morphological and functional state of the ovaries of dairy cows on the 0th day of the sexual cycle detected by the step activity sensors. In *Actual* problems of treatment and prevention of diseases of young children: Materials of the International scientific and practical contrence (pp. 137-141). Vitebsk.
- [30] Simmonds, R.C. (2017). Chapter 4. Bioethics and animal use in programs of research, teaching, and testing. In Weichbrod, R.H., Thompson, G.A.H., Norton, J.N. (Eds.). *Management of animal care and use programs in research, education, and testing.* (2nd ed) (pp. 1-28). Boca Raton: CRC Press, Taylor & Francis. <u>doi: 10.1201/9781315152189-4</u>.

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Анотація. Важливою частиною програм управління відтворенням на молочних фермах є виявлення неплідних корів і раннє повторне осіменіння для досягнення вищих показників стільності. Метою дослідження було вивчення особливостей морфогенезу гонад високопродуктивних корів в умовах промислової технології утримання в 0-й день статевого циклу *in vivo* в режимі реального часу. Було використано структурно – порівняльний і статистичний методи, а також біотехнологічну методику пальпаторного циклічного обстеження яєчників з диференційною діагностикою морфофізіологічних або патоморфологічних показників. Результати діагностики яєчників дійних корів (n=128) в 0-й день індукованого циклу, виявленого датчиками крокової активності, показали значний структурний поліморфізм цих органів репродуктивної системи. Встановлено: від 66,67 % до 30,77 % самиць з фізіологічним перебігом фолікулярної фази циклу, тобто в 0-й день на яєчниках преовуляторний фолікул або початок овуляції (за умови зменшеня придатних для запліднення корів за ростом тривалості лактації від 50-75 днів до 211-306 діб (Р<0.001). Був діагностований гіпогонадизм у 19,45 % корів, а у 61,11 % – оваріальні дисфункції з проявом кістозних дегенерацій фолікулів; 19,44 % корів на момент дослідження мали незворотні хронічні злипливі процеси тканин статевого тракту у ділянці «яєчник+яйцепровід» (овофорити та овосальпінгіти у латентній формі), що унеможливлювало настання тільності, з вираженою тенденцією росту цієї гонадопатії за збільшення терміну лактації (відповідно: 50-75 діб – 0,00 %; 76-160 діб – 22,22 %; 161-210 діб – 22,22 %; 211 і більше – 33,33 %; Р<0.05). Було досліджено тенденцію впливу субклінічних відхилень у метаболізмі корів піддослідного стада на показники біохімічного складу сироватки крові та прояв оваріальних дисфункцій. Отже, було встановлено іп vivo морфологічний і патоморфологічний профіль стану яєчників високопродуктивних корів в 0-й день статевого циклу, що дозволило оптимізувати режим штучного осіменіння і зменшити економічно невиправдані витрати сперми. Результати досліджень можуть бути використані в практичній роботі фахівців ветеринарної медицини, наукових співробітників та здобувачів вищої освіти ветеринарного та біологічного профіля

Ключові слова: дійні корови; статевий цикл; яєчники; диференційна пальпаторна діагностика; фолікули; гіпогонадизм; оводисфункції; гонадопатії