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METHODS FOR ASSESSING THE STATE OF AGRICULTURAL ANIMAL MICROOBJECTS

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Abstract. *We carried out the methods` analysis for assessing the state of livestock micro-objects. It is showcased that in order to assess the state of embryos and determine the optimal biotropic parameters of the electromagnetic field for influencing embryos, in the technological process of animal reproduction, optoelectronic systems with optimal filtering of embryo image signals should be used.*

Key words: *optoelectronic systems, livestock micro-objects, electromagnetic field, state assessment*

Introduction.

The use of a low-energy electromagnetic field (EMF) for increasing animals' fertility is best used to influence micro-objects (embryos, ovules, sperm). This electromagnetic technology requires a mandatory micro-objects assessment state after exposure to EMF with optimal biotropic parameters [1, 2].

There are various ways to assess the ovules and embryos viability. Each of them is based on its own criteria characterizing certain structural or functional properties of a living object. Ovules and embryos are evaluated and selected: by morphological features; according to the stage of their development according to the actual age; in relation to test dyes; by osmotic reaction to solutions of different osmolarity; according to metabolic rate. The highest criterion for assessing the viability of an embryo is the birth of a normal offspring that has developed from it as a result of transplantation. It is this indicator that is usually used to determine the objectivity of a particular method for assessing embryos [3, 4].

Of all currently existing methods for assessing the ovules and embryos quality, the method of assessment by morphological features is most widely used in practice. The functional state of an object in this way is determined only by visible external signs and does not allow revealing hidden internal cellular defects that affect its vitality [5].

One of the most developing and informative methods is the determination of the qualitative and quantitative biological objects' characteristics by their image. This control method is especially important in biomaterial quality evaluation, since it implies remoteness, excludes the influence of external factors, and, ultimately, does not violate the vital functions of living biological objects.

All these methods, with the exception of the morphological one, are a load on sensitive diagnosed objects and involve a certain set of additional manipulations on embryos, such as, for example, washing them from the dye, etc.

Main text.

The most promising systems for assessing the state of animal microobjects are control systems based on optoelectronic systems (OES) [6, 7].

For optical images, which evaluate the quality of microbiological objects, the brightness function obtained in transmitted light is used, which determines the spatiotemporal distribution of light intensity, which is proportional to the square of the amplitude of the EMF light wave [8].

Hence, the brightness function describing the object image has the following form:

$$B = f(x, y, t),$$

where t – time, x, y – spatial coordinates.

The obtained embryo's image with this method on the photosensor is two-dimensional, and the brightness function of the static image does not depend on time. Hence, the assessment of the qualitative and quantitative characteristics of embryos, in order to determine their viability, is carried out according to a static image [9].

In real conditions, when using the visual control method, it is necessary to analyze the intensity distribution in the image of the edge of the object under study. As a result, such an assessment is made by the operator based on his experience and intuition [10].

The best samples of such systems at a high cost give an error in assessing small sizes of $\pm 0.5 \mu\text{m}$, which does not provide an evaluation of the belonging of embryos to a certain class (embryo quality).

In the case of expanding the functionality of the OES, when assessing the dynamic characteristics of microbiological objects (spermatozoa), it is necessary to take into account the time parameter for finding the motion parameters [11].

The use of OES with computer estimated parameters of living cells movement makes it possible to expand the functionality of the system and increase the information reliability on the effect of EMF on cells vital activity.

The method of living cell movement parameters computer estimation, based on the processing of a cell sample television image, makes it possible to build tracks of motile cells and determine their kinetic characteristics.

The disadvantage of this method is that the quality of information is determined by the contrast ratio of the image and the signal-to-noise ratio. To improve the information quality, when exposed to EMF on animal cells, one should strive to increase the signal-to-noise ratio. This problem can be solved based on the theory of optimal filtering [12].

In most cases, OES is used for tracking the relative movements of microbiological objects [13]. The task becomes much more complicated when it is necessary to provide tracking of objects that have a complex shape, with a multi-tone structure on an uneven background and changing geometric dimensions.

The article [14] describes devices for assessing the parameters of sperm movement, based on the measurement of Doppler frequency shifts that occur when

optical radiation is reflected from motile cells in the ejaculate. The principle of device's operation is based on the registration of laser light scattered by moving spermatozoa, which has a Doppler frequency shift.

Currently, there are a number of OES based on the use of statistical and structural methods of image recognition [15]. Automatic construction of structural descriptions of images can be considered as image segmentation and feature measurement.

The article [16] reviews a computer method for estimating the parameters of sperm movement, based on the processing of a television image of cells with further statistical analysis. To implement the computer method following equipment was used: R-11 microscope; thermostatic table; camera (photocathode size 320x240 pixels); block of synchronization and pairing; personal computer (CIRIX 6x86 PR-200, ASUS V264 video adapter).

The processing of a video fragment in order to form tracks occurs by sequential addition of frames, which leads to a loss of information quality. In addition to this disadvantage, there are others, such as: a long processing time for video frames, 4 ... 5 minutes., A long time for obtaining histograms due to manual retrieval of information.

In article [17], an automated device was proposed for measuring the kinetic characteristics of livestock cells motility. The measurement method is based on computer analysis of a television image of a cell sample.

However, this method does not allow to unambiguously restore of the studied objects' coordinates and motion parameters, because when determining motion parameters, objects are not identified. Using this method, it is impossible to determine the individual characteristics of cell movement parameters. This method cannot be used for morphological studies of cells. Computer technology for processing and storing information about the studied animal cells has become widespread. Diagnostic systems of this type are united under the general name CASA. CASA systems have been used to study potential reproductive ability [18], poor fertility, the relationship between morphology and cell motility.

CASA-system consists of: a thermal table; video cameras; video recorder; monitors for analog and digital images visualization; computer system with equipment for digitization and image processing; keyboard and printer for data input and output.

It is noted in article [19] that the main disadvantage of the considered systems is that CASA systems either do not carry out simultaneous measurements of the mobility and morphological features parameters of the same cell, or do not allow analyzing a large number of cells as well as require a large amount of memory and high processing ability of the PC.

Summary and conclusions.

The analysis performed shows that in order to create electromagnetic technology with animal embryos, optoelectronic systems must provide the following functions: create conditions for the movement of embryos; determine the total number of embryos in the field of view; determine the percentage of mobile, immobile and locally swinging embryos; carry out simultaneous measurement of movement and

morphological features of the same embryo; image single embryos at different magnifications using a single microscope objective.

References:

1. Mykhailova, L., Kozak, O., Kosulina, N., Potapsky, P., Cherenkov, A. (2018). Determining the parameters of the acoustic system for the primary treatment of wool. *Eastern-European Journal of Enterprise Technologies* this link is disabled, 2018, 3(5-93), 61–68. DOI: 10.15587/1729-4061.2018.133710
2. A., V. Mandra, N., G. Kosulina, E., A. Chuguj, N. V. Titova. (2020). Analiz biofizicheskogo dejstviya elektromagnitnogo polya dlya povysheniya reproduktivnosti osetrovyyh ryb. *Materiali X mizhnarodnoyi naukovo-praktichnij internet-konferenciyi «Suchasnij ruh nauki», Dnipro, 1, 733–37.*
3. M, S. Sorokin, A, D. Cherenkov, N, G. Kosulina. (2009). Analiz vozmozhnosti primeneniya elektromagnitnogo polya dlya uvelicheniya vyhoda kriokonservirovannyh spermiev. *Visnik Harkivskogo nacionalnogo tehnicnogo universitetu silskogo gospodarstva im. Petra Vasilenka. Vip. 86. Problemi energozabezpechennya ta energozberezhennya v APK Ukrayini, Harkiv: HNTUSG, 86, 70–72.*
4. Casimova I. A., Kosulina N. G., Cherenkov A. D. (2010). Rezultaty eksperimentalnyh issledovanij c embrionami zhivotnyh. *Visnik Harkivskogo derzhavnogo tehnicnogo universitetu silskogo gospodarstva. «Problemi energozabezpechennya ta energozberezhennya v APK Ukrayini», Harkiv: HNTUSG, 2010.101, 1, 109–112.*
5. Shigimaga V. A., Kosulina N. G., Chorna M. A., Borodai I. I. (2022). Cell conductivity as a probability process of membrane electroporation. *Provodimost kletki kak veroyatnostnyj process elektroporacii membrany. International periodic scientific journal MODERN SCIENTIFIC RESEARCHES, 16(1), 71–84. DOI: 10.30889/2523-4692.2021-16-01-022*
6. M, S. Sorokin, N, G. Kosulina. (2009) Teoreticheskoe opredelenie parametrov impulsnogo EMP dlya povysheniya oplodotvorennosti spermiev zhivotnyh / *Vostochno-evropejskij zhurnal peredovyh tehnologij, 6/2(42), 47–49.*
7. Kosulina N. / Kosulina N. G. (2022). Electrophysical parameters of materials and bioobjects, methods and means of their measurement in electrotechnological processes. *Elektrofizicheskie parametry materialov i bioobektov, metody i sredstva ih izmereniya. SWorldJournal (Svishtov, Bulgaria). Issue 15, Part 1. September 2022, 14–20. DOI: 10.30888/2663-5712.2022-15-01-030*
8. Katys, G. P., Prezhu, V. L., Rotar, S. L. (1991). *Metody i vychislitelnye sredstva obrabotki izobrazhenij, pod red. L.D. Bahraha, Kishinev: Shtiinca, 209.*
9. Galaj, M. V., Lisenko, G. M., Solovjov, V. V. (2000). *Principi ta metodologiya avtomatichnogo rozpiznavannya ob'yektiv za yih fizichnimi vlastivostyami, Poltava: ASMI, 339.*
10. Lysikiewicz, A., Enhorning G. (1983). Tricolor photography for assessment of spermatozoa motility. *Am J. Obstet Gynecol, 145, 229–233.*
11. Kosulina, N. G. (2013). Biofizicheskij analiz vozdejstviya informacionnogo elektromagnitnogo polya na biologicheskie obekty. *Visnik Harkivskogo*

nacionalnogo tehničnogo universitetu silskogo gospodarstva im. Petra Vasilenka. Problemi energozabezpečennya ta energozberezhennya v APK Ukrajini, 141, 86–87.

12. Gutkin. L. S. (1992). Teoriya optimalnyh metodov radiopriema pri fluktuacionnyh pomegah. M.: Sov. radio, 232.

13. Casimova, I. A., Kosulina, N. G., Cherenkov, A. D. (2010). Rezultaty eksperimentalnyh issledovanij c embrionami zhivotnyh. Visnik Harkivskogo derzhavnogo tehničnogo universitetu silskogo gospodarstva. «Problemi energozabezpečennya ta energozberezhennya v APK Ukrajini». Harkiv: HNTUSG, 101 (1), 109–112.

14. Jouannet, P. Volochine B., Deguent P. (1997). Light scattering determination of various characteristics parameters of spermatozoa motility in a series of human sperm. *Andrologia*, 36–49.

15. N., G. Kosulina, O., D. Cherenkov, V, G. Gorpichenko, O., G. Avrunin. (2013). Opredelenie optimalnyh biotropnyh parametrov elektromagnitnogo polya s pomoshyu kompyuternoj obrabotki. Visnik Harkivskogo nacionalnogo tehničnogo universitetu silskogo gospodarstva im. Petra Vasilenka. Problemi energozabezpečennya ta energozberezhennya v APK Ukrajini, 141, 102–104.

16. Strelkov, A. I., Ostashko, F. I. (2000). Kompyuternyj metod formirovaniya trekov dvizheniya spermiev pri ocenke kachestva. *Medicinskaya tehnika*, Moskva, 1, 34–36.

17. Zubec, M. V, Megel, Yu. Ye., Putyatin, V. P. (1998). Pat. № 21719. UA, Sposib viznachennya ruhomosti spermiv silskogospodarskih tvarin. A61 D 19/02. publikaciya 30.04.98, – Byul. № 2.

18. Centers for Disease Control Vietnam Experience Study: Health Status of Vietnam Veterans. (1988). II. Physical Health, *JAMA*, 1988, 259, 2708–2714.

19. Boyers, S. P., Davis, R. O., Katz, D. F. (1989). Automated semen analysis *Curr. Probl. Obstet. Gynecol. Fertil*, 12(5). 165–200.

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