National Academy of Sciences of RA
Electronic Journal of
NATURAL SCIENCES

ՀՀ Գիտությունների Ազգային Ակադեմիա
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Национальная Академия наук РА
Электронный журнал
ЕСТЕСТВЕННЫЕ НАУКИ

2(25), 2015

Physiology *Ֆիզիոլոգիա* *Физиология*

NON-INVASIVE MONITORING OF EMBRYONIC DEVELOPMENT OF THE CHICK EMBRYO

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ABSTRACT

The results of studies on non-invasive evaluation of the features of embryonic development of the chick embryo using hardware complex "Bioscope" are presented. It is shown that signals of the device adequately reflect the basic stages of development of the embryo. We also show that the values of the "Bioscope"s spectral power signals of embryo, since the 8th day can predict the successful completion of the incubation process.

Keywords: *"Bioscope", embryonic development of the fetus*

INTRODUCTION

Searching of alternative methods for assessing the physiological state of biological systems led to the development of new hardware complex "Bioscope" [4] allowing to respond the non-contact approaching of biological systems (plants, laboratory animals, people). By reducing the distance between studied biological system and the "Bioscope" the characteristic oscillations in its signals are formed [5].

The device is simple in design, and its working principle is based on evaluation of the intensity of light scattered from the sensor - the glass plate covered by a thin opaque material. Structurally, "Bioscope" is optically and electrically isolated from the environment and the approaching of the various objects to the device obviously would not have to alter its readings. For inanimate objects with the ambient temperature, the device is insensitive. However it responds to the presence of a human at a distance of 5-6 meters.

Various biological objects affect the "Bioscope"s readings in different degrees, at the same time the apparatus signals depend on the changing of their physiological state [5].

Numerous experiments on the influence of stress, as well as the effect of number of pharmacological agents on animals have shown high sensitivity and specificity of "Bioscope"s signals to the change of physiological state of animals. The data are obtained that create the preconditions for the practical use of "Bioscope" for early prediction of infection and the beginning of the formation of pathological processes in the body [1, 2].

Thereby, it is interesting to study the possibilities of using the designed device to control the features of embryonic development of the chick embryo.

Unlike mammals the development of avian embryos occurs in two stages. The first beginning phase occurs in the mother's genital tract, and the final stage is outside the mother's body, under the hen or in an incubator. This paper presents data on the nature of change of "Bioscope"s signals for chick embryo during the 20-day incubation period.

MATERIALS AND METHODS

Experiments on remote registration of integrative state of 32 fertilized chicken eggs in the incubator were carried out using four-channel "Bioscope". All four devices have been integrated into the body of the incubator. During the incubation period (20 days), four times per day (at midnight, at 6 am, at12 pm and at 18 pm), was conducted the 30-minute recording of "Bioscope"s signals for different groups of embryos. Chicken eggs were installed at a distance of 2-3 mm from the "Bioscope" sensor. Generally, 320 registrations in different periods of incubation were conducted. *Fig. 1* shows an example of recording the "Bioscope"s signals of embryo in the first days of incubation.

In the studies a commercial incubator IB2NB was used. From the first to sixth days of incubation the temperature in incubator was maintained at 37.9 ^{0}C. Later, in the 15th day of incubation, it decreased to 36.8 ^{0}C, and further to 36.2 ^{0}C in 20th day. Every 6 hours until the last week of incubation the eggs were overturned. To analyze the obtained data the combined program developed in an environment of LabView was used.

For 30 minutes recording the band filtering was carried out in the frequency range of 0.1-10 osc/min, and the values of successive interpeaks time intervals of oscillation signals of "Bioscope" (BB-intervals) were determined. By the method of fast Fourier transform the spectral distributions for the initial signals were made and values of spectrum power density were calculated. Besides, the average oscillations frequency of recorded signals and the coefficient of variation of values of BB-intervals were calculated.

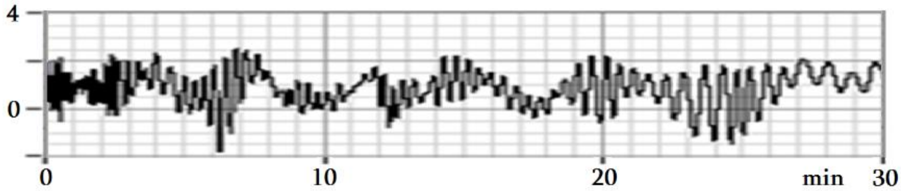

Fig. 1. Example of "Bioscope" signals for the chick embryo in the first days of incubation. On the X-axis – the time; ordinate - conditioned amplitude of "Bioscope" signal.

Of the 32 eggs only 10 chicks were hatched. In line with this, all the data were divided into two groups - for hatched chicks, and embryos the development of which turned out to be incomplete. Subsequently, for all daily registrations the averaging of groups of "hatched" and "not "hatched" embryos was performed. The significance of differences in performance of different experimental groups was evaluated using Student's t test at a significance level of $p < 0.05$.

RESULTS AND DISCUSSION

As seen from the Table 1, in the first day of incubation the initial values of calculated indicators are not statistically differ for hatched and hatched embryos groups.

Table 1. Mean values of statistical indicators of "Bioscope" s signals for chick embryo in the first days of incubation.

Embryos groups	Average frequency of "Bioscope" s oscillation signals (1/min)	Coefficient of variation of VV-intervals (%)	The spectrum power density of "Bioscope"s signals
Hatched chickens	2.3 ± 0.2	51.8 ± 3.1	0.221 ± 0.032
Chickens not hatched	2.3 ± 0.1	54.5 ± 2.0	0. 282 ± 0.044

Fig. 2 shows the variation of the oscillation frequency for the hatched and not hatched embryos. According to *Fig. 2*, from 4th to 8th and from 15th to 18th days of incubation, it is observed an increase in the oscillation frequency for hatched embryos in comparison to the signals frequencies for not hatched embryos.

A B

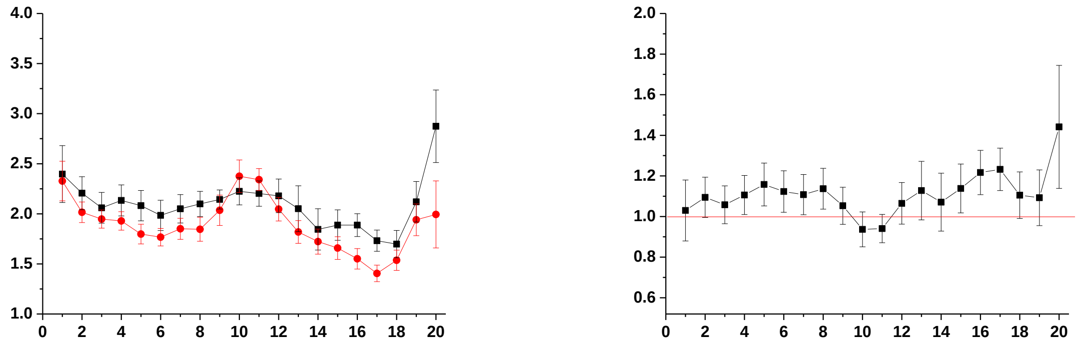

Fig. 2. The nature of the change of the oscillation frequency signals of "Bioscope" for chick embryo during incubation period. A–absolute values of frequencies "Bioscope"s signals for hatched (black) and unhatched (red) embryos; B – ratio of oscillation frequencies of "Bioscope" signals for hatched and unhatched embryos. The abscissa – days of incubation, the vertical axis on the A - oscillation frequency (osc/min)

During incubation, a very interesting dynamics takes place in the values of the coefficients of variation for the hatched and unhatched chick embryos (*Fig. 3A*). In both cases an antiphase cyclic increase and decrease of their values occurs (*Fig. 3B*). At this, the first cycle lasts 7 days, the second - from the 7th to the 11th day, and the third cycle begins from the 12th day to the 18th. The fourth final cycle includes the last days of incubation.

The calculations revealed a pronounced difference in the character of the change of spectral power density of "Bioscope" signals for hatched and unhatched chick embryos (*Fig. 4*). For unhatched embryos during incubation the spectral power remains almost unchanged. In contrast, for hatched embryos a monotonic decrease of the values of spectral power occurs.

A B

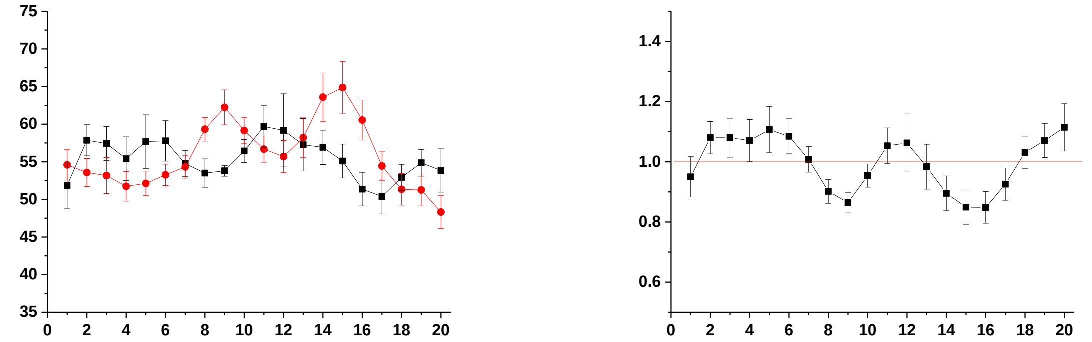

Fig. 3. The character of the change of the variation coefficients for intervals of interpeak signals of "Bioscope" for chick embryo during incubation. A - absolute values of the variation coefficients of BB-intervals for the hatched (black) and unhatched (red) embryos; B – ratio of values of the variation coefficients of BB-intervals for the hatched and unhatched embryos. On the abscissa axis - days of incubation, the ordinate on *Fig. 3A* - coefficient of variation in percent.

A B

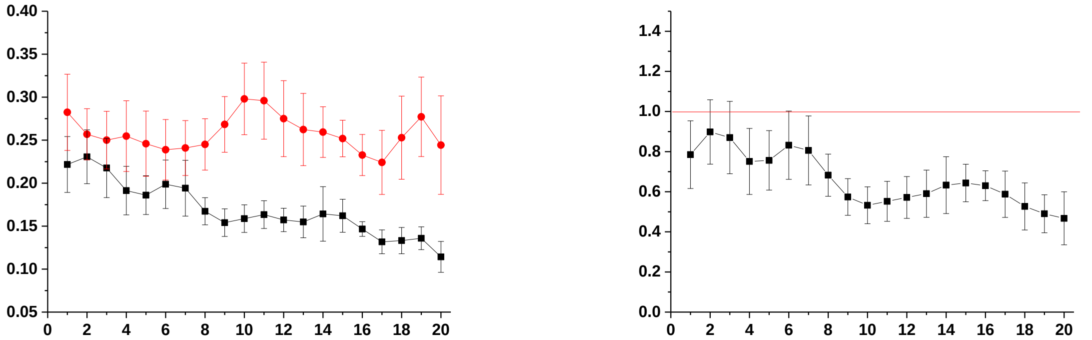

Fig. 4. The nature of the change in spectral power density of the "Bioscope"signals for chick embryo during incubation. A-the absolute value of power density for hatched (black) and unhatched (red) embryos; B – ratio of values of spectrum power density for hatched and unhatched embryos. The abscissa – days of incubation, the ordinate on A - spectral power density.

As it is known, from the first hours of incubation cycle, begins an intensive processes associated with the formation of various organs and systems of the future chicken organism. In the process of embryo development there are three periods [3]. The first, embryonic, lasts from the moment of fertilization up to 8 days. At this time a development of the skeleton occurs. Already at the second day a development of the cardiovascular system occurs. In the second (prefetal) period (from 8th to 12th days of incubation) the growth and specialization of the organism systems takes place. Third fetal period begins from the 12th day, while there is growth and development of organism which ends with the hatching. Calculations showed that for 4-8, 15-17, and the 20th days of incubation the oscillations frequency of the recorded signals for hatched embryos is higher than for unhatched.

More pronounced dynamics occurs in the values of the coefficients of variation for BB-intervals for the hatched and unhatched chicken's embryos. In both cases an antiphase cyclic increase and decrease of their values occurs. At this, the first cycle lasts 7 days, the second, from 7th to 11th day, and the third cycle begins from the 12th day to the 18th. The fourth and final cycle includes the last days of incubation.

The revealed by us cyclicity in the change of variation coefficients for BB-intervals fully correlates with the literature data which indicates that the signals of "Bioscope" adequately reflect the most important stages of embryo development.

Furthermore it is known that due to intensive stages of differentiation of the embryo at 6-11 days and 12-18 days there are periods of depression of embryo growth [3]. Interestingly, also in this case the days with minimal correlation coefficients in our experiments with hatched embryos occur precisely in these periods of incubation.

The calculations show a significant difference in the values of power density spectra of “Bioscope” signals for hatched and unhatched embryos. In unhatched embryos its value retains at a relatively constant level, for the hatched embryos to the end of the incubation cycle it is observed a monotonic decrease in the values of their spectral power. This fact allows already on the 8th day predict the successful completion of the incubation process.

CONCLUSIONS

This study showed that during the remote assessment of development of chick embryos in the process of incubation the most informative factor is the value of the spectrum power density of “Bioscope” signals and coefficients of variation of BB-intervals. The character of these changes for hatched embryos adequately reflects the known three stages of embryo development. It is also shown that the days with minimal relative correlation coefficients of BB-intervals of “Bioscope” signals coincide with periods of depressed growth of hatched embryos. Already starting from 8-9 days of incubation, the values of the spectrum density of 30-minutes registering of signals of “Bioscope” statistically significantly differ for hatched and not hatched embryos. This fact indicates the possibility of predicting the successful completion of the incubation process of chicken embryo since the 8th day, on the base of the values of spectral power of “Bioscope” signals.

This study demonstrates the possibility of using the hardware complex “Bioscope” for remote control of embryonic development of the chick embryo. The obtained results are of practical value and can be used to increase and optimize the output of chickens on poultry farms.

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