REGULAR FEATURES OF THE DIURNAL RHYTHM IN MITOTIC CELL DIVISION OF THE OCULAR SAC AND PRESUMPTIVE ECTODERM DURING THE PERIOD OF THEIR CONTACT WITH EACH OTHER IN THE CHICK EMBRYO

(UDC 611.84-013-018.825)

A. A. Derdiyashchenko

Department of Experimental Embryology (Head—Professor O. E. Vyazov)
and Laboratory of Histophysiology (Head—Candidate of Biological Science V. N. Dobrokhotov)
Institute of Experimental Biology (Director—I. N. Maiskii) of the Academy
of Medical Sciences of the USSR, Moscow
(Presented by Active Member of the Academy of Medical Sciences, N. N. Zhukov-Verezhnikov)
Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 60, No. 9,
pp. 109-111, September, 1965
Original article submitted December 10, 1964

Diurnal periodicity in mitotic activity of tissues has been investigated mainly in adult organisms [1-4]. Similar periodicity in the tissues of embryos has received much less study. The present work deals with cell division in the optic vesicle and presumptive ectoderm of the embryo at one and the same stage of development but at various times of the day, with a view to determining whether any diurnal periodicity of mitotic activity exists in these tissues and, if it does exist, what its nature is.

EXPERIMENTAL

Hen eggs (Russian White breed) were placed in an incubator at 2 h intervals throughout the 24 h period beginning at 12:00 noon. In the incubator they were maintained under constant conditions: temperature 37.5°C humidity 70%. After 48 h the eggs were removed, opened and the embryo from each egg removed. The embryo was fixed in Bouin's fluid, embedded in paraffin wax and histological sections (thickness 7 μ) prepared by generally accepted methods. The preparations were stained with Carazzi's hematoxylin and eosin. The specimens selected for histological investigation were those which gave a monotypic picture of contact between the optic vessel and the presumptive ectoderm. Counts of the number of mitoses were carried out on 5 embryos for each particular time of the day—a total of 60 embryos. Mitoses counts were made from middle sections (using 3 consecutive sections with intervals of 1-2 sections) through the optic rudiment in the region where the somewhat thickened central portion of the ectoderm overlaid the optic vesicle. The regions investigated comprised: 1) that part of the optic vesicle from the point of its contact with the ectoderm to the midline; and 2) those parts of the ectoderm lying on both sides of the optic vesicle and extending from the point of contact with the optic vesicle to their points of origin. The outlines of the optic vesicle and the presumptive ectoderm, both in section, were drawn onto paper with the help of a camera lucida (objective 40 ×, ocular 7 ×). Afterwards counts of cell numbers and mitoses were made under a binocular microscope (objective 90 ×, ocular 7 ×). Mitotic coefficients were calculated in order to evaluate mitotic activity. All experimental results were subjected to statistical analysis using the Fisher-Student method.

Table of Mitotic Activity in the Optic Vesicle and Presumptive Ectoderm

Hour of day	Mitotic coefficient (%)	
	optic vesicle	ectoderm
2	88,2	66,0
4	104,2	60,0
6	89,1	58,2
8	108,9	64,6
10	96,6	38,6
12	119,1	84,1
14	92,9	58,9
16	118,6	90,3
18	121,9	76,0
20	143,8	108,2
22	116,2	85,7
24	118,4	103,2

RESULTS

The results of our counts of mitotic activity in the optic vesicle and presumptive ectoderm are given in the table.

The minimum amount of cell division activity in the optic vesicle occurred at 2:00 a.m., when the mitotic coefficient was 88.2%. Mitotic activity then gradually increased and during the afternoon (2:00 p.m.-6:00 p.m.) it reached a level higher than it had reached at night or in the morning. From 4:00 p.m. until 8:00 p.m. the mitotic activity of the cells increased rapidly. At 8:00 p.m. it reached a maximum with a mitotic coefficient of 143.8%.

Nevertheless, the difference in level of mitotic activity at 8:00 p.m. and at 6:00 p.m. and 10:00 p.m. was not found to be statistically significant. Precisely the same lack of statistical significance applies in relation to the difference between activity at 8:00 p.m. and the total activity between 10:00 p.m. and midnight ($P = 0.2$). The value for the number of mitoses at 8:00 p.m. does, however, appear to be significantly different, or very nearly significantly different, from the values of 2:00 p.m. and 2:00 a.m. ($P = 0.02$; 0.005 respectively). The curve for mitotic activity in the presumptive ectoderm is basically similar to that based upon observations of mitosis in the optic vesicle, but the former curve attains lower levels than does the latter. The mean daily mitotic activity of cells in the optic vesicle is 108.2%, whereas in the presumptive ectoderm, it is 73.5%.

The least mitotic activity in the cells of the presumptive ectoderm was observed at 10:00 a.m., when the mitotic coefficient equalled 38.6%. This level of mitotic activity is not statistically different from the level at 8:00 a.m. or 12:00 noon. However, in comparison with the aggregate mitotic activity over the period from 8:00 a.m. to 12:00 noon, the minimum activity at 10:00 a.m. is very nearly significantly different ($P = 0.04$). The total mitotic activity over the period from 8:00 a.m. to 10:00 a.m. is not, however, significantly different statistically from the total activity over the period from 10:00 a.m. to 12:00 noon ($P = 0.059$).

During the afternoon from 2:00 p.m. onwards, the mitotic activity of the ectodermal cells gradually increases, attaining a maximum at 8:00 p.m., as does the division of optic vesicle cells. However, the difference in mitotic activity of ectodermal cells at 8:00 p.m. is not statistically different from that at 6:00 p.m. and 10:00 p.m.

In this way, the results of our experimental observations have revealed the existence of a definite diurnal rhythm of mitotic activity in the cells of chick optic vesicle and chick presumptive ectoderm at one particular stage of development. The maximum mitotic activity in both embryonic tissues has been noticed at 8:00 p.m. We are therefore able to confirm the existence of a diurnal rhythm in the cell division process during early growth stages, as is known to occur in the adult organism. This rhythm is characterized particularly by peak mitotic activity in the evening hours.

LITERATURE CITED

1. M. T. Gololobova, Byull. éksper. biol., 10 (1960), p. 118.
2. V. N. Dobrokhotov, Vestn. AMN SSSR, 7 (1963), p. 50.
3. L. P. Kosichenko, Byull. éksper. biol., 6 (1960), p. 98.
4. L. V. Sokolova, Byull. éksper. biol., 7 (1964), p. 98.

All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.
