

STATE OF THE GAMETES AND CHARACTER OF MATURATION OF FOLLICULAR
OOCYTES IN NEONATALLY ANDROGENIZED RATS WITH LHRH-INDUCED OVULATION

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The causes of formation of anomalous gametes still remain largely unexplained. There is no doubt that the origins of this phenomenon go back into the pre-embryonic period of ontogeny, i.e., the time when the oocyte is developing inside the follicle. The role of periovulatory changes in the character and level of secretion of gonadotrophins and ovarian hormones in the formation of anomalous gametes has recently been demonstrated [2, 3, 5]. The importance of disturbances arising in the follicles, growth of which takes place against the background of prolonged disturbances of hormonal regulation of the reproductive process in the genesis of gametopathies has received less study. These problems have assumed special significance at the present time through the widespread use of hormonal methods of correcting human reproductive function and in the treatment of some forms of sterility. Hence the importance of a study of the conditions of formation and the state of gametes formed during induced ovulation in animals with a prolonged anovulatory state.

The aim of this investigation was to study these processes in neonatally androgenized rats after single and repeated injections of luteinizing hormone releasing hormone (LHRH).

EXPERIMENTAL METHOD

Experiments were carried out on 123 noninbred albino rats aged 5-7 months. The animals were androgenized neonatally by a single injection of 25 µg testosterone propionate on the 2nd day of life. The animals were kept under controlled conditions of illumination (10 h of daylight, 14 h of darkness) and on an ordinary diet. There were four series of experiments: I) control (18 rats); II) intact animals (38 rats) receiving a single intramuscular injection of 500 ng LHRH (from Agerst Laboratories, Canada) neonatally androgenized animals (36 and 31 rats respectively), with permanent estrus or with atresia of the vagina, into which LHRH was injected once or 6 times at intervals of 4 days. Ovulating oocytes were obtained by incision of the ampulla of the oviducts. Culture and analysis of the oocytes was carried out by the method described previously [1]. The results were subjected to statistical analysis by the Fisher-Student test.

EXPERIMENTAL RESULTS

Injection of LHRH into the control animals was accompanied by an increase in the number of rats with unovulating follicles. In the androgenized rats, after injection of LHRH a tendency was observed for the number of such animals to increase (16-26% in series I and II, 41-43% in androgenized rats). Single and repeated injections of LHRH into the control animals (series I) did not increase the number of ovulating oocytes (8.2 compared with 9.6 in rats not receiving LHRH). Meanwhile, in the androgenized animals, the number of ovulating oocytes was significantly less than in intact rats after injection of LHRH (7.8 and 5.6 per rat respectively in series III and IV). As in the previous experiments [1], the present investigations showed that single and repeated injections of LHRH into intact animals, while increasing the number of oocytes resuming meiosis, caused a decrease in the number of oocytes completing meiosis normally after culture for 42-46 h, and this also was accompanied by an increase in the number of chromosomal aberrations (16.6% compared with 6.2% in the control). Injection of

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LHRH into the androgenized rats was accompanied by similar changes in the parameters of culture of the follicular oocytes. The frequency of resumption of meiosis rose in this case to 84.6-90.7% compared with 66.2% in intact rats. An increase in the number of oocytes with evidence of degeneration of chromosomes at the metaphase II stage was observed after culture for 42-45 h. Finally, in the group of experimental rats receiving a single injection of LHRH the frequency of chromosomal aberrations reached 20% on account of an increase in the number of aneuploid (hypo- and hyperploid) ova.

Repeated injections of LHRH, while not significantly changing the parameters of maturation of the follicular oocytes by comparison with a single injection of LHRH, was however accompanied by significant changes in the degree of heterogeneity of the gametes formed in androgenized rats. Their cytogenetic analysis showed that stimulation of ovulation by LHRH is accompanied by a significant increase in the number of ovulating oocytes with chromosomal disturbances, mainly of degenerative character (41.3 and 61.3% compared with 26% in intact animals without injection or after a single injection of LHRH) in the androgenized animals. The frequency of chromosomal aberrations in the experimental animals, on the other hand, decreased under these circumstances along with an increase in the number of gametes with clear signs of disturbance of their structure (5.3% in intact rats after a single injection of LHRH, 2.7 and 0.7% in androgenized rats after single and repeated injections of LHRH respectively). Meanwhile, in intact and androgenized rats induction of ovulation by LHRH caused the appearance of immature ova among the ovulating oocytes in 1.2-1.4% of cases.

These results are evidence that changes in growth and maturation of the follicles, giving rise to corresponding changes in the character of maturation of the follicular oocytes, are accompanied by a tendency toward an increase in the number of chromosomal aberrations in the oocytes in culture. This tendency was manifested most clearly after induction of ovulation by a single injection in both intact and experimental rats. Whereas in the first case no increase was observed in the number of gametes with chromosomal disturbances compared with natural ovulation, in the group of androgenized rats the number of gametes with marked structural disturbances increased significantly. Repeated injections of LHRH increased the number of gametes with marked disturbances of chromosome structure in the population of ovulating even more. A decrease in the frequency of chromosomal aberrations in the population of ovulating oocytes in the experimental animals, associated with an increase in the number of oocytes with marked signs of degeneration, is apparently a regular feature.

It can accordingly be postulated that stimulation of ovulation by LHRH, while adversely affecting the population of follicular oocytes, as is demonstrated by data in the literature [4, 6], on the one hand has a more significant effect on the state of the follicles (and ova) in the ovaries of neonatally androgenized rats, and on the other hand, stimulates ovulation of a greater number of imperfect ova in animals with prolonged disturbance of reproductive function.

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