Reports

Cultures of Gonads of Mammalian Embryos

Since the explanation of sex reversal in the freemartin was given by Lillie (*1*), there have been a number of attempts to investigate sex reversal experimentally, but, until recently, results with mammalian forms have been rather meager. Grafts of embryonic mammalian gonads to adult hosts of the opposite sex (*2*) have given negative or nearly negative results. Hormones of adults, when given to pregnant animals, have produced marked effects on the fetal reproductive tracts but inconsistent effects on the gonads. Recently MacIntyre (*3*), by grafting embryonic rat gonads in pairs to castrated adult hosts, has demonstrated that, when an ovary is grown with an embryonic testis, differentiation of the ovary is suppressed, while the testis is unaffected. Similar experiments on the rabbit by one of us (*4*) have produced the same result.

Wolff and his coworkers (*5*) have cultured embryonic duck gonads together in male-female pairs. In these experiments the ovary proved to be the dominant gonad, and the testis was suppressed.

It was felt that such cultures of embryonic mammalian gonads might be of some value as a control on the grafting experiments, inasmuch as cultures provide an environment more nearly free of extraneous hormones than adult hosts.

We therefore undertook a series of cultures of embryonic mammalian gonads. Both rabbit and rat embryos were used. The cultures were grown in hollow ground slides. We used plasma clots and embryonic extract as a culture medium after attempts with synthetic media failed. The embryonic extract was prepared from the anterior (gonad-free) halves of 11-day chick embryos, the plasma from heparinized blood of castrated rabbits. Gonads for culturing were obtained from embryos of the desired age and were grown in cultures for 4 to 18 days.

The 56 cultures were distributed as follows: male-female combinations, 17; male-male, 8; female-female, 10; single male, 9; single female, 8. There were also four cultures of gonads recovered at indifferent stages. Rabbits and rats gave the same results. No heteroplastic combinations were attempted.

Embryonic testes usually grew and differentiated well in all combinations. The seminiferous tubules were well formed and contained spermatogonia, some of which were degenerating, as they normally do. The differentiation was never equal to that occurring in a similar period of normal development but, in most cases, was well advanced over the differentiation that had occurred at the time of explanation.

Embryonic ovaries, on the other hand, only grew and differentiated in 19 of 35 cultures. When cultured alone or in combination with other ovaries, those that did grow produced some cortical differentiation. There were cell nests, occasional structures suggestive of primordial follicles, some proliferation of interstitial cells, and the retention of a cuboidal epithelium, which might be interpreted as a germinal epithelium. When explanted with testes, the ovaries which grew were always retarded in development in comparison with those cultured in the absence of the male gonad. None of them developed cell nests or other indications of cortical structures. In three instances there were indications of a masculinizing effect, as shown by the development of structures suggestive of testicular cords in medullary portions of the cultures. MacIntyre has suggested that such cords may in fact be "converted follicles of cortical or secondary sex cord origin." In either case, differentiation of the ovary was altered when the ovary was cultured with a developing testis. The differentiation of the testis, on the other hand, was not affected by the presence of the ovary.

These experiments (*6*) appear to support the findings obtained by grafting embryonic gonads. As the embryonic ovary is not affected in its development when grafted to the adult male, even if grown in the host testis (*7*), we suggest that it is probable that the embryonic testis produces a substance or "hormone" capable of modifying ovarian development and that this substance is not identical with the adult testicular hormone.

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References and Notes

1. F. R. Lillie, *Science* 43, 611 (1916).
2. A. Buyse, *J. Exptl. Zool.* 70, 1 (1935); C. R. Moore and D. Price, *ibid.* 90, 229 (1942); E. A. Holyoke, *Anat. Record* 103, 675 (1949).
3. M. N. MacIntyre, *Anat. Record* 124, 27 (1956).
4. E. A. Holyoke, *ibid.* 124, 307 (1956).
5. E. Wolff and K. Haffen, *Arch. Anat. Microscop.* 41, 184 (1952).
6. This study was supported by research grants from the National Institutes of Health, U.S. Public Health Service.
7. E. A. Holyoke, *Anat. Record* 127, 470 (1957).

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Ultraviolet Mitigation of X-ray Lethality in Dividing Yeast Cells

Abstract. The lethal effect of x-rays on dividing yeast cells can be decreased by small ultraviolet exposures delivered before or after x-ray exposure. This mitigating action can be decreased by exposure to visible light concomitant with photoreactivation of ultraviolet lethality. The results suggest considerable overlap between x-ray and ultraviolet lethality sites in dividing cells.

In the course of a study of the molecular and anatomical nature of the sites sensitive to lethal irradiation in dividing yeast cells, different combinations and permutations of x-rays, ultraviolet (UV), and visible light (VL) were employed. Depending upon the radiation and the sequence of administration, both coupling and uncoupling effects were observed. One such effect is the ability of UV either to protect or to reactivate x-rayed dividing yeast cells (that is, cells in division when irradiated).

Although the results to be described were obtained with a nonrespiring strain of haploid *Saccharomyces cerevisiae,* SC-7(ρ), qualitatively similar results were obtained with the parental, respiring diploid strain, SC-6, and the respiring haploid strain SC-7 from which SC-7(ρ) was derived. The mitigating effects were largest in SC-7(ρ), comparable in SC-6, and small but present in

Instructions for preparing reports. Begin the report with an abstract of from 45 to 55 words. The abstract should *not* repeat phrases employed in the title. It should work with the title to give the reader a summary of the results presented in the report proper. (Since this requirement has only recently gone into effect, not all reports that are now being published as yet observe it.)

Type manuscripts double-spaced and submit one ribbon copy and one carbon copy.

Limit the report proper to the equivalent of 1200 words. This space includes that occupied by illustrative material as well as by the references and notes.

Limit illustrative material to *one* 2-column figure (that is, a figure whose width equals two columns of text) or to *one* 2-column table or to *two* 1-column illustrations, which may consist of two figures or two tables or one of each.

For further details see "Suggestions to Contributors" [*Science* 125, 16 (1957)].