Accumulation of Radioiodine (I^{131}) in Thyroid Gland of the Hamster Embryo.* (19111)

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The correlation between the storage of colloid and functional activity has been the basis upon which many of the studies on the thyroid gland have been made both in the clinic and in the laboratory. This relation is also the basis upon which its initial function in the embryo can be established. Studies of this type are facilitated where radioactive iodine (I^{131}) is used as a tracer, since its presence in the colloid is easily demonstrated on a photographic emulsion. Recently, with the use of radioactive iodine, Hansborough and Khan(5) demonstrated that the thyroid gland of the chick embryo becomes functional toward the end of the 11th day of incubation and that the activity of the gland increases with the increase in the number of its follicles. Gorbman and Evans(4) found that radioactive iodine appeared in the colloid of the frog thyroid when the young tadpole reaches a length of 10 mm and also that the storage of colloid is correlated with the appearance of the thyroid follicles. These workers(3) also determined the age at which the thyroid of the fetal rat begins to function. They studied 20 fetuses of rats in the 15th to the 21st day of gestation. Of those studied, thyroids of none showed evidence of iodine storage before the 19th day of development. Coincident with the beginning of function of the thyroid in the 18-19-day-old rats, follicle differentiation occurred. In the human fetus, it has been observed(1) that the accumulation of radioactive iodine and the differentiation of follicles in the thyroid occur from the 14th to the 32nd week of development.

The purpose of this study is to establish the initial up-take of radioactive iodine by the thyroid of the embryo hamster and thus to reveal the age of the embryo at which the thyroid becomes functional.

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Materials and methods. Forty females were mated to 7 males (an average of 6 females to each male) and the time of copulation was recorded as the beginning of development, since it has been established that ovulation and fertilization follow within a few hours after mating(2). After copulation, the females were weighed and isolated in individual cages. Increase in weight was used as an index of pregnancy, particularly in the early stages of development, when observable size increase was negligible. With a No. 2 tuberculin syringe, the females of 8 to 15 days gestation (gestation period is 15 days 8 hours) were injected intraperitoneally with a solution of radioactive iodine (25 microcuries/cc) of from 50 to 100 microcuries, the amount of fluid not exceeding 2 cc per injection. A period of from 15-30 minutes was allowed after the last injection before the females were sacrificed and the embryos recovered. This was suggested by the work of Hertz, *et al.*(6) who found that the normal or hypoplastic thyroid of the rabbit collected a maximum of radioactive iodine within 15 minutes after injection and that for longer periods the amount collected was not greatly increased. The activity of the embryos was measured with the Geiger-Muller counter and whole embryos were fixed according to the method previously described(7). Similarly the ac-

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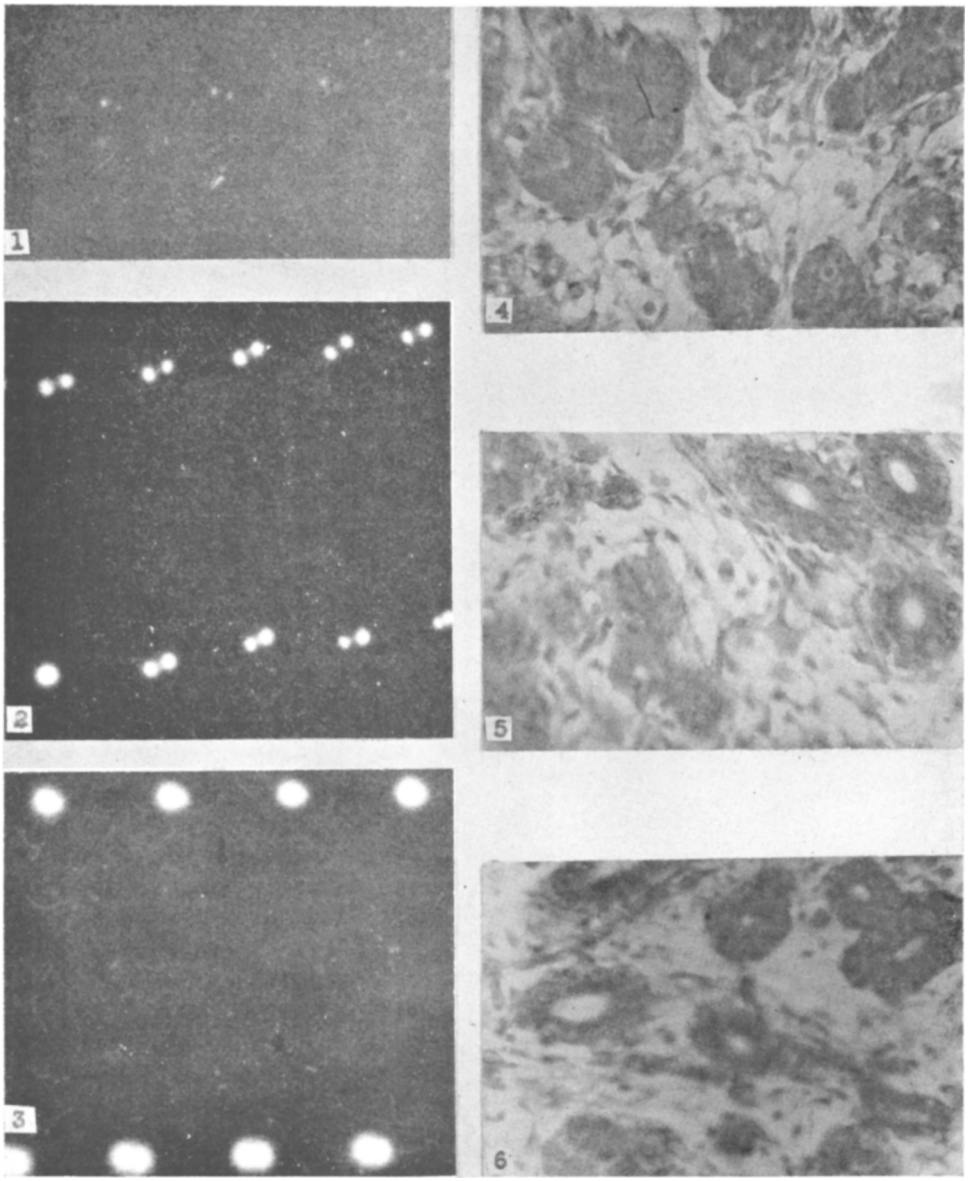

FIG. 1. 1. Autoradiograph of sections through the thyroid of a 13-day embryo. 2. Autoradiograph of sections through the thyroid of a 14-day embryo. 3. Autoradiograph of sections through the thyroid of a 15-day embryo. 4. Microphotograph of the sections shown in 1. Note small follicles surrounded by vascular connective tissue. 5. Microphotograph of the sections shown in 2. 6. Microphotograph of the sections shown in 3. Note the increase in size and number of the follicles shown in 5 and 6.

tivity of the thyroids of the mothers was checked. Dental x-ray films were exposed to sectioned embryos for from 6 to 8 days, depending upon the activity of the iodine. After autoradiographs were made, sections were stained in Delafield's hematoxylin and eosin for microscopic study.

Observations and conclusions. Autoradiographs of embryos in the 8th to the 12th day of development showed no evidence of iodine accumulation. The first indication of the collection of radioactive iodine was observed in autoradiographs of 13-day embryos. Further study showed that iodine accumulation

increased with the age of the embryo, as indicated by the increase in blackening on the x-ray film. These observations were supported by measurements with the Geiger-Muller counter; embryos of 8 to 12 days development gave no significant measurements. However, those of 13 days and older gave an increasing number of counts.

Microscopic study of sections of the thyroid of embryos from the 9th and 10th day of development revealed the presence of epithelial cells in irregular cords, but no discrete follicles. Thyroids of embryos of the 11th and 12th day contained a few groups of cells arranged in circular patches (young follicles), with a few discrete follicles in the 12-day embryos. There is a progressive increase in the number of follicles from the 12th through the 15th day of development with an increase in the quantity of colloid present.

Since it is assumed by many workers that the storage of colloid in the follicles indicates the function of the thyroid, then we may also assume that the initial appearance of colloid indicates the onset of thyroid function. If these assumptions are correct, then our observations support the conclusion that the thyroid of the embryo hamster begins to function on the 13th day of development, a short time after the first appearance of follicles.

It has been demonstrated for the embryos of many animals, including the human fetus (see introduction) that the storage of colloid is correlated with the differentiation of follicles in the gland, the quantity depending upon the degree of differentiation and the number of follicles present and these, in turn, are correlated with the age of the embryo. Our findings through autoradiographic and microscopic study support these conclusions for the thyroid gland of the golden hamster.

Summary. (1) Fifty to 100 microcuries of radioactive iodine in the form of NaI were injected into the peritoneal cavity of the gestating hamster on the 8th to the 15th day after copulation. (2) Observations made on embryos of from 8 to 15 days of development by means of autoradiographic study and tests with the Geiger-Muller counter show that radioactive iodine begins to accumulate in the thyroid on the 13th day of development. (3) The accumulation of radioactive iodine is coincident with the appearance of discrete follicles, which first appear between the 12th and 13th day of development. (4) The quantity of iodine-containing colloid increases with the number of follicles present and the age of the embryo.

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Cross-Immunity Tests in Animals with Epidemic Keratoconjunctivitis and St. Louis Encephalitis Viruses. (19112)

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Recent investigations disclosed that it is impossible to differentiate the viruses of epidemic keratoconjunctivitis (EK) and St. Louis encephalitis (SLE) by serological means(1,2). A hint that the two viruses may be different comes from the fact that EK caused a fatal encephalitis in guinea pigs and rabbits while SLE engendered only a non-fatal pyrexia in guinea pigs and nothing in rabbits. However, no distinct differences could be demonstrated by cross-immunity tests in animals. This report presents the data on the infectivity tests as well as the results of the cross-immunity tests.

Materials and methods. Viruses. The 2 strains of epidemic keratoconjunctivitis virus

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