Mechanism of Induction of Ovarian Tumors by X-Rays*

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Tumors have developed in ovaries grafted into the spleen or pancreas of gonadectomized mice or rats (1, 8, 11 to 14). Such tumors did not appear when animals bearing such grafts possessed a functional gonad in the normal position, or if adhesions formed between the spleen and body wall so that venous drainage was into the systemic circulation, or if estrogenic hormone was administered (14). Estrogenic hormone produced by the ovary in the splenic or pancreatic position drains directly into the portal venous system, and in these species is to a great extent inactivated by the liver before reaching the systemic circulation (9).

The gonadotrophic content of the pituitary gland of gonadectomized rodents is elevated (3, 4).

TABLE 1

DATA ON DEVELOPMENT OF OVARIAN TUMORS IN BAGG ALBINO MICE FOLLOWING RADIATION WITH 200 r OF X-RAYS

No. of mice	Method of radiation with 200 r	No. with tumors in radiated ovaries
4	Local to each ovary	4
2	Local to one ovary—2d ovary extirpated	2
15	Local to one ovary—2d ovary untreated	0
24	Whole body	24

Mice possessing ovarian tissue whose secretion passes directly into the portal venous system are physiologically castrate. Consequently it has been postulated that the stimulus responsible for the genesis of tumors in grafted ovaries is gonadotrophic hormone, which may be secreted in increased quantity by the pituitary gland if this organ is not inhibited by gonadal secretion.

Ovarian tumors may be induced in mice by whole body exposure to x-rays. These tumors are of the same histological types as those which develop in ovaries grafted into the spleen, that is, tubular adenomas, granulosa cell tumors, and luteomas (6, 7). Radiation with x-rays may reduce estrogenic homone output of the ovaries, thereby affecting the gonadotrophic output of the pituitary gland. Thus, the mechanism of induction of tumors might then be the same as in the splenic grafts, the gonadotrophic hormone acting upon the remaining ovarian tissue of reduced estrogenic secretory activity.

The purpose of these experiments was to determine whether x-rays *per se* induce ovarian tumors in mice in the presence of normal ovarian function.

MATERIALS AND METHODS

The Bagg albino stock of mice is very susceptible to the induction of ovarian tumors by x-rays. Practically all females receiving 200 r by whole body radiation at 8 to 10 weeks of age possess bilateral ovarian tumors if they survive 16 months post-irradiation (10). Females of this stock were placed on experiment in the following groups (Table 1):

I. Each ovary of 4 Bagg albino females received 200 r by contact radiation.
II. One ovary was radiated with 200 r, the other extirpated in 2 mice.
III. One ovary was radiated, the other untreated in 15 mice.

All animals were 8 to 10 weeks of age at the time of radiation. Originally the groups of mice were larger, but those developing pneumonia were sacrificed early and excluded from consideration.

In radiating the animals the entire mouse was shielded by lead except for a tiny aperture through which the ovary to be radiated was drawn. The ovary was placed in position on the lead plate covering the mouse at the time of radiation. When both ovaries were radiated the operation was done in 2 stages with a week's interval between exposures. The ovaries were radiated with a Phillips Contact Therapy Machine (45 KV, 2 ma., 1 mm., aluminum filter, 19 mm. target skin distance, 1500 r per minute). The period of exposure at 1500 r per minute was 8 seconds.

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All the mice except one were maintained as virgins, and vaginal smears were examined 12 and 16 months after radiation (Table 2). Between 16 and 17 months after radiation the animals were autopsied. Sections of the ovaries, adrenals, reproductive tracts, and submaxillary glands were studied.

OBSERVATIONS

Ovarian tumors were present in all of 24 Bagg albino females within 16 months following whole body radiation of mice 8 to 10 weeks of age with 200 r of x-rays. Larger doses of x-radiation produced similar results. Microscopically these tumors varied in structure, but were usually composed of surface epithelial downgrowths and proliferations of granulosa cells among which were nests of luteal cells (Fig. 2). Varying portions of the ovary contained cells filled with ceroid pigment (Fig. 1). This pigment may be deposited within either tumor cells or normal ovarian luteal cells.

Grossly, ovaries containing tumors were larger than normal, varying from approximately 3 mm. to over one cm. in diameter, and were usually yellow in color. In some instances the ovary contained cysts in addition to the yellow tumor masses. Cysts were filled with either clear fluid or blood. An animal died occasionally from hemorrhage into the peritoneal cavity.

When both ovaries were radiated individually with 200 r of x-rays by contact radiation, no other portion of the body being exposed to radiation, bilateral tumors appeared (Table 1). If one ovary was radiated in the same manner, and the other ovary removed, tumor development resulted (Table 1). In all of 15 cases where a single ovary was radiated with 200 r and the other ovary untreated, no tumor was induced in the radiated ovary (Table 1). Such ovaries presented a characteristic appearance 16 months post-radiation (Figs. 3 and 4). They were smaller than the non-irradiated ovaries. Microscopically they were composed of a cortex of anovular follicles (Figs. 3 and 4), and a medulla containing stromal cells, some of which contained ceroid pigment. Surface epithelial downgrowths were not present. The follicles appeared to be those which had persisted following destruction of the ovum by radiation. There was no evidence of new formation of anovular follicles from the surface epithelium.

One of the non-irradiated ovaries (in animals in which the other ovary was radiated) contained a granulosa cell tumor (Figs. 7 and 8). Of particular interest is the fact that although this ovary was almost completely replaced by tumor within it, at least two fully developed ovarian follicles were present. A second non-irradiated ovary contained large hemorrhagic cysts, and in a third epithelial downgrowths were present, and granulosa cell proliferation was evident immediately beneath the surface epithelium (Figs. 9 and 10). The remaining non-irradiated ovaries contained a few well developed ovarian follicles, with a considerable portion of the ovary being composed of stromal cells, many containing ceroid pigment (Fig. 5). No anovular follicles were present in the cortex (Fig. 6).

The incidence of spontaneous ovarian tumors in the Bagg albino stock is very low. No ovarian tumors were seen in any of 17 females beyond 17 months of age.

Data on vaginal smears of test animals are given in Table 2. Twelve months post-irradiation the vaginal smears of animals whose ovaries had been radiated by contact radiation were castrate, whereas 12 of 15 of those in which only a single ovary had been radiated were undergoing cycles. Sixteen months after radiation cycles were still evident in 7 of 15. At this age cycles are disappearing in intact, untreated Bagg albino females.

TABLE 2

DATA ON VAGINAL SMEARS OF BAGG ALBINO MICE RECEIVING 200 r OF X-RADIATION

No. of mice	Method of radiation with 200 r	Age post-irradiation	Number cycling	Number castrate
4	Local to each ovary (one week apart)	12 months	0	4
2	Local to one ovary—other extirpated	" "	0	2
15	Local to one ovary—other untreated	" "	12	3
4	Local to each ovary (one week apart)	16 months	0	4
2	Local to one ovary—other extirpated	" "	0	2
15	Local to one ovary—other untreated	" "	7	8

DISCUSSION

X-radiation of a single ovary did not induce ovarian tumors if the second ovary was not radiated. This strongly suggests that the mechanism of induction of ovarian tumors is similar to that operating in the development of tumors within ovaries grafted into the spleen, where the presence of a functioning ovary in the normal position inhibits tumor formation in the grafted ovary. When all of the ovarian tissue was radiated, the estrogenic secretion of the ovary was largely abolished within at least a few months post-irradiation, although some Bagg albino mice bear litters following radiation with 200 r. Estrous cycles have been reported in mice made anovular by roentgen irradiation (2). Furthermore, ovarian tumors have been induced in mice receiving only 87 r of x-rays (5). Complete withdrawal of ovarian estrogenic

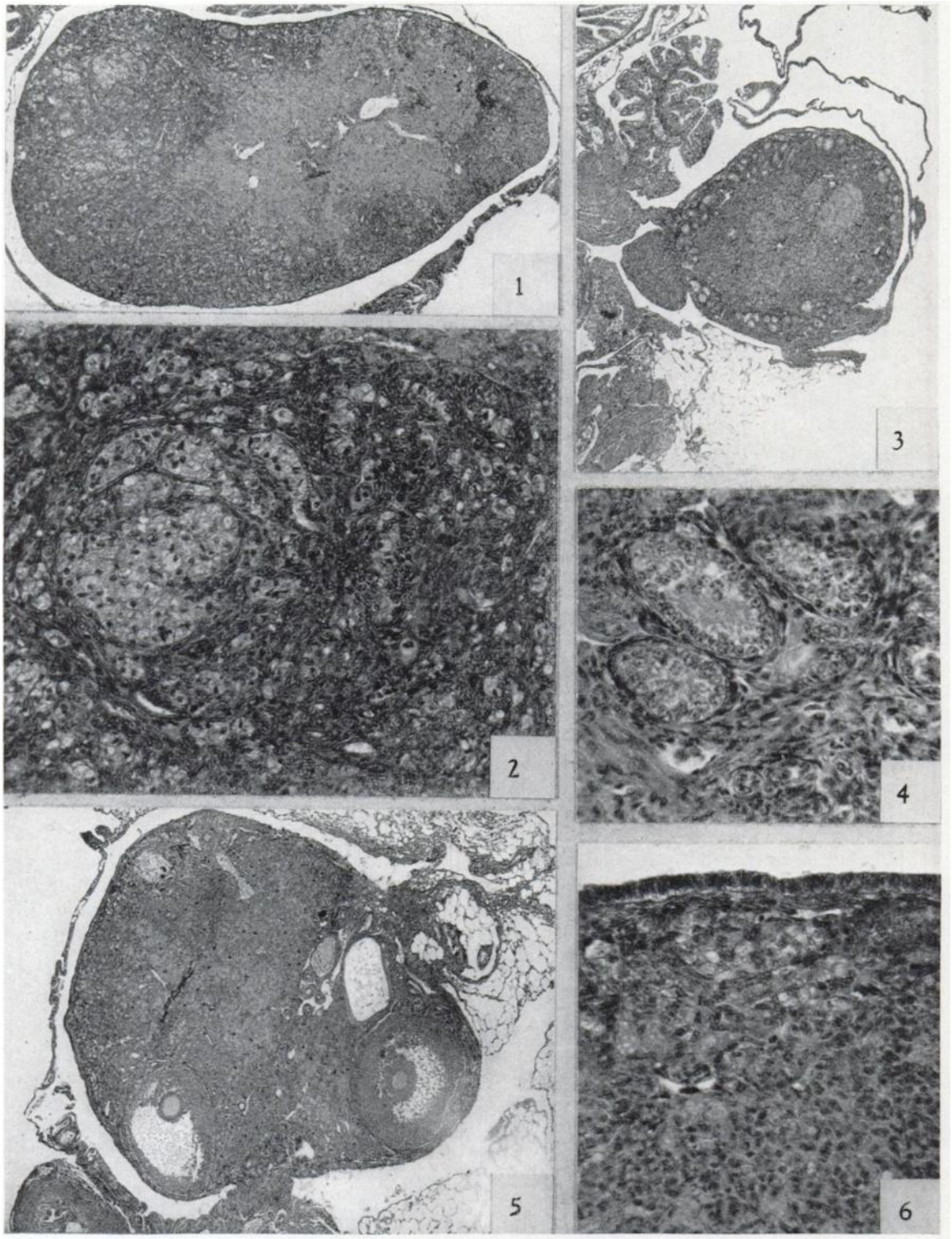

FIG. 1.—Ovary of Bagg albino female; both ovaries radiated with 200 r of x-rays by contact radiation 16 months prior to autopsy. Most of cortex invaded by granulosa cell tumor shown in Figure 2. Central portion contains cells with ceroid pigment. Ovary grossly enlarged. Mag. ×40.

FIG. 2.—High power view of tumorous portion of ovary illustrated in Figure 1. Dark cells are granulosa cells; those with more abundant pale cytoplasm are luteal cells. Mag. ×70.

FIG. 3.—Ovary of Bagg albino female radiated with 200 r by contact radiation 16 months before autopsy. Other ovary not radiated, and shown in Figure 5. Cortex composed of anovular follicles. Central portion has fibroblastic stroma, some of the cells containing ceroid pigment. Tumors did not develop in radiated ovaries if the second ovary was not radiated and remained functional. Grossly this ovary was smaller than the non-radiated ovary. Mag. ×40.

FIG. 4.—Anovular follicles from the cortex of the ovary shown in Figure 3. Mag. ×150.

FIG. 5.—Non-irradiated ovary from animal whose other ovary, shown in Figure 3, was radiated with 200 r 16 months before autopsy. Mature follicles still present. Mag. ×40.

FIG. 6.—Cortex of non-irradiated ovary shown in Figure 5. No anovular follicles present as in radiated ovary illustrated in Figure 3. Mag. ×150.

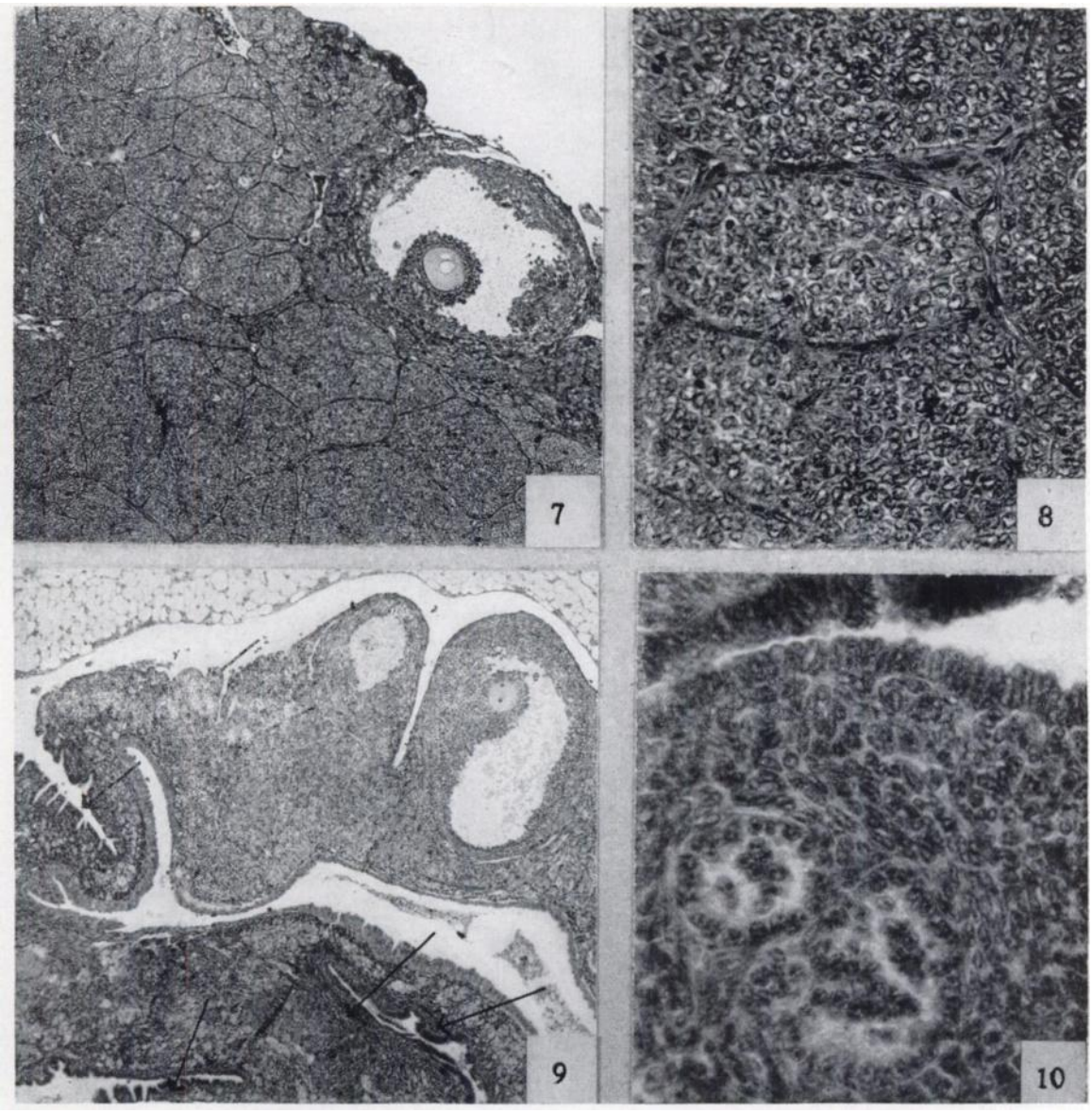

FIG. 7.—Granulosa cell tumor in non-irradiated ovary of a Bagg albino female whose other ovary was radiated with 200 r of x-rays by contact radiation 16 months before autopsy. Except for two mature follicles enlarged ovary is composed of cords of granulosa cells. Mag. ×60.

FIG. 8.—High power view of granulosa cell cords shown in Figure 7. Mag. ×180.

FIG. 9.—Non-irradiated ovary of animal whose other ovary received 200 r of x-rays 16 months before autopsy. Arrows indicate areas of granulosa cell proliferation and surface epithelial downgrowth. Mature follicles are present. Mag. ×60.

FIG. 10.—High power view of area of granulosa cell proliferation in ovary illustrated in Figure 9. Mag. ×180.

secretion by radiation is thus not essential for the induction of ovarian tumors.

The development of ovarian tumors or tumor-like proliferations in the second non-irradiated ovary, 3 of 15, may be significant. To determine whether reduction in estrogenic secretion resulting from radiation of a single ovary plays a role in the genesis of such tumors, the effect of extirpation of a single ovary on tumor formation in the remaining ovary is being tested.

SUMMARY

X-rays did not induce tumor development in a radiated ovary of the Bagg albino stock if the animal's second ovary remained unirradiated and functional. Contact radiation of a single ovary induced ovarian tumor development only if the second ovary was extirpated. Bilateral contact radiation resulted in tumor development. Although a second functioning ovary inhibited ovarian tumor development following unilateral contact radiation, a granulosa cell tumor developed spontaneously in a non-irradiated ovary containing two fully developed ovarian follicles.

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