Pituitary Role in the Estrogen Dependency of Experimental Mammary Cancer*

ABRAHAM STERENTAL,† J. MIGUEL DOMINGUEZ,‡
CORINNE WEISSMAN, AND OLOF H. PEARSON

(Departments of Pathology and Medicine, Western Reserve University School of Medicine, Cleveland, Ohio)

SUMMARY

Mammary tumors were induced in 50-day-old albino Sprague-Dawley rats by a single feeding of 20 mg. of 7,12-dimethylbenz[a]anthracene. Adrenalectomy-ovariectomy and hypophysectomy performed within 10–20 days after tumors were first detected resulted in tumor regression in all the animals. Estrogen administration reactivated tumor growth after adrenalectomy-ovariectomy but not after hypophysectomy. The estrogen unresponsiveness of tumors in hypophysectomized animals was not modified by thyroid and cortisone replacement therapy. These results indicate that the estrogen stimulation of this tumor is dependent upon the presence of the pituitary gland.

It has been demonstrated that oophorectomy induces regression of human breast cancer (1) and that estrogen administration exacerbates the disease previously improved by this ablative procedure (9). These observations indicate that the mechanism of ovariectomy-induced remissions involves estrogen deprivation. Hypophysectomy also produces breast cancer regression in women, but after hypophysectomy estrogen administration fails to reactivate the disease (8).

The studies reported here were designed to determine whether the growth of experimental, hormone-dependent mammary cancer in rats can be exacerbated by estrogen administration after remissions were induced by various endocrine ablative procedures. It has been shown that estrogens reactivate tumor growth after ovariectomy and adrenalectomy, whereas estrogens fail to exacerbate tumor growth after hypophysectomy-induced remissions in the rat.

MATERIALS AND METHODS

Ninety-four female albino rats of the Sprague-Dawley strain, 50 days old and weighing 145–155 gm., were treated with a single feeding of 20 mg. of 7,12-dimethylbenz[a]anthracene (DMBA) dissolved in 1 ml. of sesame oil (5). In six animals that received vehicle alone and remained as untreated controls, no tumors were detected during the length of the experiment. The animals were maintained on a Purina Laboratory Chow diet and tap water ad libitum. After adrenalectomy-ovariectomy tap water was replaced by normal saline.

Tumors were first detected 8–12 weeks after carcinogen administration, and growth was checked 3 times a week by measurement of its two greatest diameters from which the surface area was calculated. After tumor development six animals were left as intact controls.

Adrenalectomy-ovariectomy was successfully performed in fourteen animals and hypophysectomy in ten animals, bearing at least one tumor with a surface of one or more sq. cm. When almost complete tumor regression was induced by these procedures, eight animals of the adrenalectomized-ovariectomized group and seven of the hypophysectomized group received replacement treatment with estradiol benzoate, 5 μg. daily for 20–50 days. In addition to estrogen treatment, three of the hypophysectomized animals received physiological doses of L-thyroxine, 5 μg. a day (10), and cortisone, 0.5 mg. a day (2), for a period of 30 days. Two animals were treated twice with estradiol benzoate, first after tumor regression following adrenalectomy-ovariectomy, 5 μg. daily for 30 and
36 days, and later, after hypophysectomy, 5 \mu g. daily for 40 and 49 days, respectively. Completeness of ablative procedure was confirmed at necropsy, and microscopic studies of the tumors were done during each experimental stage.

Of the 692 animals fed with the carcinogen and not included in the study, 37 died of respiratory infection before tumors developed, seventeen died during surgical procedures, and eight developed either no tumors or small ones which were not considered suitable for adequate measurements of the changes induced by treatment. Estrogenic activity in the adrenalectomized-ovariectomized, hypophysectomized, and estrogen-treated animals were determined by vaginal smears.

RESULTS

Hormone dependence of DMBA-induced tumors.

Six animals fed with DMBA developed eight tumors that grew for periods of 30–65 days, reaching a mean surface area of from 2.5 to 25 sq. cm. Tumor size remained steady thereafter. Only one tumor had a partial spontaneous regression from 2.5 to 1 sq. cm. Seventy to 90 days after tumors were first detected, five of the six animals died of cachexia, tumor ulceration, and respiratory infection. Necropsy failed to show metastases, and microscopic studies of the tumors were described as “partially differentiated adenocarcinoma” (Fig. 1).

Of a group of nine animals bearing thirteen tumors with a mean surface area of 3.5 sq. cm., 10–20 days after tumors were first detected, six underwent adrenalectomy-ovariectomy, and three hypophysectomy. After the operation all tumors showed a steady regression for 5–35 days, reaching a mean surface of 0.25 sq. cm. Microscopic examination of these small tumors revealed them as “small foci of well differentiated adenocarcinoma, with varied degrees of replacement by connective tissue” (Figs. 2, 3).

No spontaneous death or tumor reactivation occurred in these groups for periods of 65–110 days following the operation, with the exception of one adrenalectomized-ovariectomized animal that, following a temporary remission, had a reactivation of the tumor which at necropsy was described as “foci of degenerating adenocarcinoma in a leiomyoma” (Fig. 4).

Chart 1 shows the surface area of individual tumors of intact, adrenalectomized-ovariectomized, and hypophysectomized animals.
Tumor response to estrogens after adrenalectomy-ovariectomy.—A group of eight animals bearing ten tumors was adrenalectomized-ovariectomized 10–20 days after tumors were first detected. For 10–40 days following the operation the mean surface area of the tumors decreased from 3.3 sq. cm. to 0.25 sq. cm. A week later the animals were given injections of estradiol benzoate, 5 μg/day for periods of 20–40 days, resulting in a tumor growth to a mean surface area of 5.6 sq. cm. Microscopic appearance of the tumors in this stage is similar to that of intact controls. Twenty to 40 days after estrogen withdrawal the tumors regressed to a mean surface area of 0.45 sq. cm. No tumor reactivation was observed for periods up to 70 days after estrogens were withdrawn, and animals were sacrificed for histologic studies of the tumors. Chart 2 shows the results of these experiments.

Tumor response to estrogens after hypophysectomy.—A group of seven animals bearing ten tumors was hypophysectomized 10–40 days after tumors were first detected. For 15–30 days following the operation the mean surface area decreased from 3 sq. cm. to 0.3 sq. cm. A week later the animals were treated with estradiol benzoate, 5 μg. daily for periods of 20–30 days. In addition to estrogens, three animals received cortisone, 0.5 mg., and T₄-thyroxine, 5 μg/day for 30 days. Neither estrogens alone nor estrogens with cortisone and thyroid resulted in an increase in tumor size. In this respect this tumor behaves like normal breast tissue (4). Microscopic studies of these tumors showed a similar pattern to that of the tumors that regressed after hypophysectomy or adrenalectomy-ovariectomy. In one animal tumor size increased during estrogen administration but at necropsy was found to be owing to intratumoral necrosis and hemorrhage. Chart 3 shows the results of these experiments.

Tumor response to estrogens after adrenalectomy-ovariectomy and after hypophysectomy.—Two animals bearing one tumor each of 3.6 and 2 sq. cm. of surface area were adrenalectomized-ovariectomized 22 and 14 days after tumors were first detected, resulting in tumor regression to a surface area of 0.25 sq. cm. Estradiol benzoate, 5 μg/day for periods of 30 and 30 days, induced tumor growth to surface areas of 9 and 2.25 sq. cm., respectively. After estrogen withdrawal the tumors regressed to 0.25 sq. cm. A similar course of estrogens following hypophysectomy 16 days after tumor regression failed to induce tumor growth. Chart 4 shows the results in each of these two animals.
DISCUSSION

DMBA mammary tumors are histologically and probably biologically malignant because, although they do not metastasize, they invade adjacent tissues and end the life of the host. A highly important characteristic of these tumors is their hormonal dependency, since the removal of the ovaries and adrenals or pituitary gland results in tumor regression. These observations confirm those of Huggins and Yang (6). The hormonal dependency resembles that of some human breast cancers, and, therefore, the evaluation of these tumors' response to changes in hormonal environment could result in useful clinical application.

The observations that removal of the ovaries and adrenals resulted in tumor regression and that estrogen administration reactivated tumor growth indicates that these tumors are estrogen-dependent. On the other hand, the failure of estrogens to reactivate the growth of tumors that had regressed after hypophysectomy indicates that estrogen dependency is conditioned by the presence of the pituitary gland. The pituitary factor or factors responsible for these effects appears to be independent of the secondary thyroid and adrenal insufficiency because thyroid and cortisone replacement did not return the tumor capacity to respond to estrogen stimulation. The pattern of estrogen response after ovariectomy-adrenalectomy and hypophysectomy, using the same animals as controls, reaffirms the concept that estrogen dependency is conditioned to the presence of the pituitary gland. A factor or factors similar to that produced by rat mammotropic pituitary tumors described by Furth and Clifton (3) and Kim and Furth (7) could be responsible for this phenomenon. Further studies are in progress to determine the mechanism of these experimental observations.

ACKNOWLEDGMENTS

The authors are indebted to Dr. Robert R. Kohn for performing the pathologic studies. We also wish to acknowledge the active participation of Alan B. Pearson.

REFERENCES

Pituitary Role in the Estrogen Dependency of Experimental Mammary Cancer


*Cancer Res* 1963;23:481-484.

**Updated version**  Access the most recent version of this article at: http://cancerres.aacrjournals.org/content/23/3/481

**E-mail alerts**  Sign up to receive free email-alerts related to this article or journal.

**Reprints and Subscriptions**  To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

**Permissions**  To request permission to re-use all or part of this article, contact the AACR Publications Department at permissions@aacr.org.