Relation of Mammary Tumors to Mammotropes.* II. Hormone Responsiveness of 3-methylcholanthrene Induced Mammary Carcinomas. (25622)

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Mammary tumors (MT) can be readily induced in rats by oral administration of 3-methylcholanthrene (3-MCA). Many of these tumors can be inhibited by androgens, ovariectomy, or hypophysectomy and stimulated by estrogens(1). Experiments point to a cell of the pituitary, named mammotrope (Mt), the hormones of which appear to be the main stimulants of the mammary gland. Estrogens appear to exert their effect indirectly by stimulating this cell (*cf.* 2,3).

The present study was undertaken to determine the role of Mt, *i.e.,* mammotropic hormone (MtH), on growth of primary MT, induced in 33 rats with 3-MCA. All tumors were biopsied after which 13 rats were ovariectomized and 20 hypophysectomized. Each rat was studied individually and progression of tumor growth was charted. When, following ovariectomy or hypophysectomy the tumors decreased in size markedly, a functional mammotropic tumor (MtT) providing MtH, was grafted on some of the animals, while others were left untreated to serve as controls. Typical experiments are illustrated in Fig. 1-3.

Materials and methods. Tumor bearing rats of the same group(4) were matched in trios; one was hypophysectomized, one ovariectomized and one left untreated as control. Operation was performed by standard surgical procedure. Hypophysectomized animals were given Huggins' Special Diet,† *ad lib.* No supplementary hormones were given. Two strains of MtT were used (for supplying endogenous MtH): F4 (estrogen induced in $F/_{Fu}$ rats) and W2 (spontaneous in $W/_{Fu}$ rats). F4 grew faster than W2. MtH secretion was greater by F4, as indicated by degree of mammary gland stimulation of tumor bearing hosts. Growth of both strains was greatly stimulated by estrogens. In our terminology, such tumors are designated as functional autonomous responsive tumors. Size of MT was measured immediately before treatment and thereafter every 2 to 5 weeks. Average diameters were calculated from the 3 dimensions.

Results. In the sample experiment illustrated in Fig. 1, one rat (B) was ovariectomized when she had 8 MT, the mean diameter of which was charted. Following ovariectomy all 8 MT decreased in size. Thirty-three days following ovariectomy, MtT was grafted on this rat (to supply MtH) and this caused resumption of growth of MT. Another rat (C) had 4 MT. Hypophysectomy caused decrease in size of all 4 tumors. Upon graft of MtT all 4 tumors resumed growth, and 6 new MT appeared. The tumor of the untreated control (A) grew progressively and 2 more tumors developed on this rat at 46 days.

Fig. 2 and 3 present similar trios. Fig. 2 shows an apparently complete regression of tumor following hypophysectomy (F) but MtT caused not only reappearance of this MT, but 11 new tumors appeared on this rat (Fig. 4). In the experiment illustrated in Fig. 3 hypophysectomy was performed when the rat had 2 fairly large MT. Both decreased in size after hypophysectomy and resumed growth following an MtT graft.

The Figures also illustrate the parallelism between rate of growth of MT and MtT; *e.g.,* in Fig. 3 (H) MtT growth plateaued and so did that of MT.

Altogether, ovariectomy was performed on 13 rats bearing 50 tumors of which 36 (72%) decreased in size and 14 (28%) continued to grow with no apparent restraint (Table I). Responsive and nonresponsive tumors ap-

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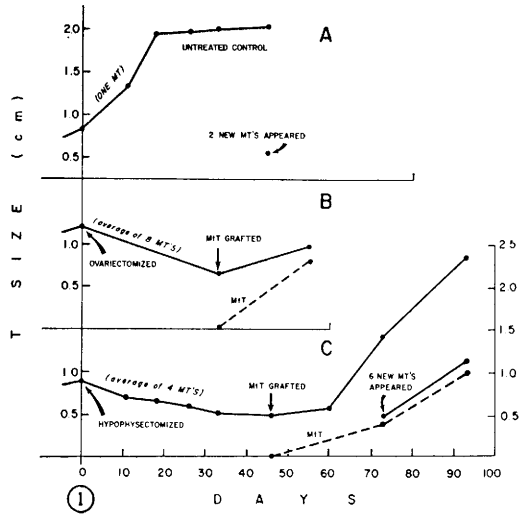
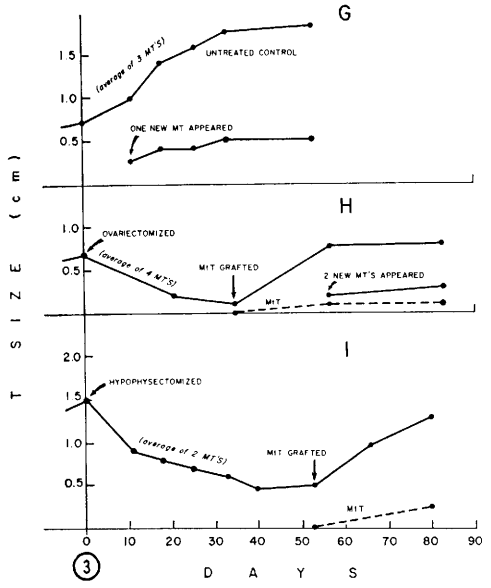
† Obtained from Nutritional Biochemicals Corp., Cleveland, O.





peared at random in the same animal.

Hypophysectomy was performed on 20 rats bearing 59 tumors (Table II), of which 51 (86%) were inhibited. Thus, hypophysectomy was slightly more effective than ovariectomy. All tumors of all 11 rats which were inhibited by hypophysectomy resumed progressive growth following receipt of an MtT graft. In hypophysectomized rats which did not receive MtT grafts, most MT (about 54%) remained minute (measuring a few mm across) and about 37% could not be found at autopsy. About 9% of the tumors resumed

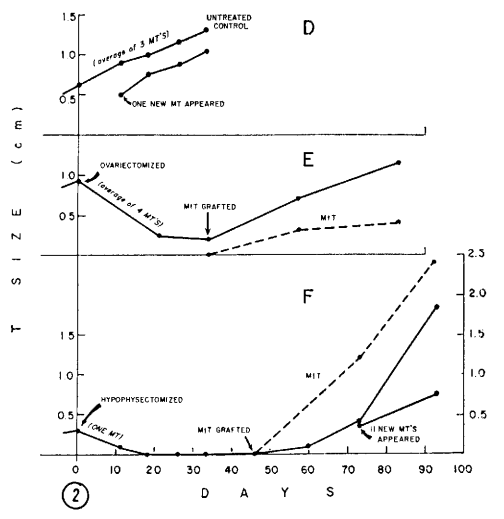


FIG. 1. Effect of ovariectomy (B) and hypophysectomy (C) on 3-MCA induced primary mammary carcinoma. Charts show decrease in tumor size following operative procedures and resumption of growth following grafts of mammotropic tumors (supplying MtH).

FIG. 2. Trio similar to that of Fig. 1. Apparent complete regression of mammary carcinoma following hypophysectomy with reappearance of this and other latent tumors following MtT graft.

FIG. 3. Trio similar to those in Fig. 1 and 2 illustrating further parallelism between MT and MtT growth rates.

progressive growth in hypophysectomized rats but at a much slower rate. No new tumors appeared in hypophysectomized rats, while this was a common event in untreated controls.

Comments. These observations are in accordance with those in man on the influence

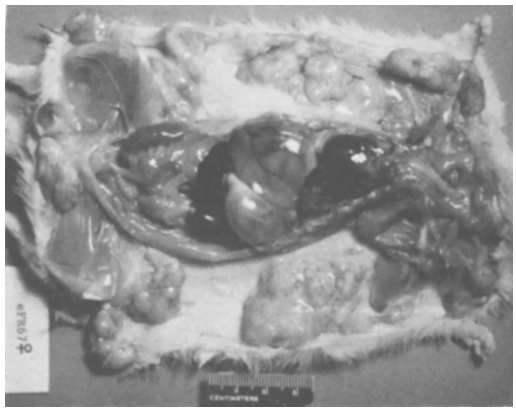


FIG. 4. Hypophysectomized rat (Fig. 2, F) with 12 macroscopic tumors appearing after MtH stimulation.

TABLE I. Effect of Ovariectomy and Mammotropic Tumor Grafts on 3-MCA Induced (Original) Tumors.

Group	No. of rats	No. of tumors	Inhibition by ovariectomy	MtT graft* No. of rats†
			%	
F-I	1	8	8 (100)	
III	5	19	11 (57)	3
IV	4	11	10 (91)	3
W-IV	4	11	17 (58)	2
Total	13	50	36 (72)	8

* Following effective ovariectomy.
† All were stimulated.

of ovariectomy and hypophysectomy on growth of mammary tumors(5,6). Rat tumors appeared more responsive to hormonal procedures than human tumors. This may be either a species characteristic or due to an earlier institution of operative procedures. Resumption of tumor growth in hypophysectomized and ovariectomized rats following grafts of functional MtT (*i.e.* MtH) is in line with our supposition that MtH secreted by Mt is the most direct stimulus of MT and estrogens act indirectly by stimulating this cell(2,3). The effectiveness of hypophysectomy in women is generally attributed to deprivation of several hormones, mainly gonadotropins, growth hormone and prolactin. Experiments in our laboratory attribute an indirect role of gonadotropin, acting through enhancement of estrogen production and link somatotropin and prolactin to the same cell, named mammotrope. Whether this cell discharges one or 2 hormones remains to be determined. Other hormones play only a minor, "permissive" non-specific function (*cf.* 2). Corticoids markedly enhance secretion but not growth of the mammary gland(2,7).

TABLE II. Effect of Hypophysectomy and Mammotropic Tumor Graft on 3-MCA Induced Tumors.

Group	No. of rats	No. of tumors	Inhibition by hypex.	MtT graft* No. of rats†
			%	
F-I	2	6	4 (67)	
III	5	9	8 (89)	4
IV	7	16	13 (81)	7
W-IV	6	28	26 (93)	
Total	20	59	51 (86)	11

* Following effective hypophysectomy.
† All were stimulated.

Summary. 1. About 72% of primary 3-MCA induced mammary carcinomas were inhibited to varying degrees by ovariectomy and 86% by hypophysectomy. 2. Most inhibited tumors remained minute; a few resumed progressive growth and a moderate number (about 37% in the hypophysectomized series) could not be identified at autopsy. 3. Grafts of functional mammotropic tumors (providing a steady supply of mammotropin) on hypophysectomized animals, whose mammary tumors have markedly decreased in size following ovariectomy or hypophysectomy, caused not only resumption of progressive tumor growth, but also appearance of numerous new mammary tumors. Thus MtH can unfold latent neoplastic potentialities.

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