Suppression by Ergocornine and Iproniazid of Carcinogen-Induced Mammary Tumors in Rats; Effects on Serum and Pituitary Prolactin Levels[1] (35076)

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Many drugs have been demonstrated to alter secretion of pituitary hormones, and thereby modify functions regulated by these hormones. Ergocornine was reported to inhibit deciduoma formation (1), pseudopregnancy (2), early pregnancy (2), and lactation (3) in rats, and it was suggested that this drug inhibits pituitary prolactin secretion (4). Iproniazid injections inhibited mammary secretion in rats (5), suggesting that it may also depress prolactin secretion. Dopamine has recently been reported to profoundly reduce pituitary prolactin release *in vitro* (6, 7). In view of the importance of prolactin for mammary tumorigenesis (8, 9), it was of interest to determine the effects of these three drugs on carcinogen-induced mammary tumors and on serum and pituitary prolactin values in rats.

Materials and Methods. Sprague-Dawley female rats, 50 days old, were given a single intravenous injection of 5 mg of 7, 12-dimethylbenz(a)anthracene (DMBA) dissolved in a fat emulsion.[4] The rats were observed for appearance of mammary tumors beginning 3 weeks after injection and every 5 days thereafter. The two major diameters of each tumor were measured by calipers, and tumor size was expressed as the geometrical mean of the two diameters. When the largest tumor of each rat approximated 1 cm in diameter, the rats were treated daily with doses of the three drugs as shown in Table I. Only the growth of the initial tumors were measured. The amount of each drug dissolved in 0.9% saline was as follows: ergocornine (Sandoz, Ltd., Basel, Switzerland),[4] 3 mg/ml; iproniazid phosphate (Hoffman-La Roche Inc., Nutley, N. J.), 37.5 mg/ml; dopamine (Mann Res. Lab., New York), 0.5 mg/ml. Ergocornine was given daily for only an average of 15 days because of the limited supply of this drug, whereas iproniazid and dopamine were given daily for an average of 25 days. Two groups of rats were injected with 0.9% saline daily and served as controls.

Estrous cycles were followed by daily vaginal smears beginning about 7 days before the end of the experiments. On the day of estrus of the second cycle of each rat, the rats were killed by guillotine, and blood was collected immediately from the severed trunk. The anterior pituitary, adrenals and ovaries were removed and weighed. The serum and pituitary glands were kept at −20° until assayed for prolactin by radioimmunoassay (10). Average total growth rate of tumors was calculated as percentage increase in tumor size during the injection period. Significance of differences between groups was determined by Student's *t* test.

Results. The mammary tumors of all groups grew at about the same rate before injection of drugs (Fig. 1). Tumor growth in the rats given ergocornine and iproniazid was

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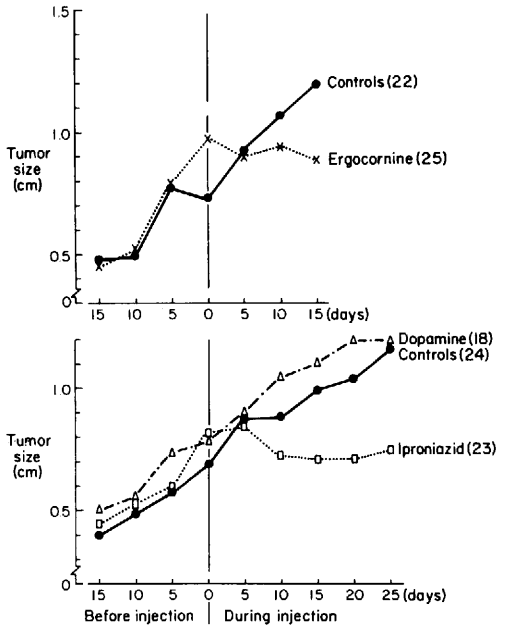

FIG. 1. Increase in average tumor size before and after daily injections of drugs. Total number of initial tumors measured is indicated in parentheses.

completely suppressed during injection, whereas the tumors of the two control groups and of the group given dopamine continued to grow at a similar rate. The average number of tumors per rat increased about 2-fold in the two control groups and in the rats given dopamine (Table I), whereas there was no significant change in average number of tumors per rat in the groups injected with ergocornine or iproniazid. Serum and pituitary concentrations of prolactin were significantly lower ($p<.01$) in the rats treated with ergocornine than in the controls and rats given the other drugs. Iproniazid elevated serum prolactin values above the controls, but because of the large standard error this rise was of doubtful statistical significance.

Table II shows that none of the drugs altered body weights significantly. Ergocornine produced an increase in the average weight of the ovaries, whereas iproniazid elicited small but significant reductions in the weights of the pituitary and adrenals. None of the drugs appeared to alter the estrous cycles.

Discussion. These results demonstrate that

TABLE I. Effects of Drugs on Development and Growth of DMBA-Induced Mammary Tumors, and on Serum and Pituitary Prolactin Levels.

Daily treatment	No. of rats	Av no. tumors per rat		Av total tumor growth rate (%)	Serum prolactin conen (mμg/ml)	AP prolactin conen (μg/mg AP)
		Initial	Final			
Controls, saline, 15 days	8	2.9 ± 0.5[a]	5.3 ± 1.3	59.3 ± 13.8 (22)[b]	58.5 ± 9.9	2.1 ± 0.2
Ergocornine, 0.6 mg, 15 days	9	2.8 ± 0.8	3.6 ± 0.8	2.8 ± 11.6[c] (25)	27.8 ± 4.2[c]	1.3 ± 0.1[c]
Controls, saline, 25 days	12	2.0 ± 0.4	5.8 ± 0.9	102.3 ± 15.9 (24)	64.4 ± 9.4	2.0 ± 0.2
Iproniazid, 7.5 mg, 25 days	12	2.4 ± 0.3	2.9 ± 0.6	3.4 ± 29.3[c] (23)	109.6 ± 28.1	2.5 ± 0.3
Dopamine, 0.1 mg, 25 days	9	2.3 ± 0.3	4.4 ± 0.5	69.0 ± 20.6	85.4 ± 13.8	2.9 ± 0.3

[a] Mean ± standard error of mean.
[b] ( ) Number of initial tumors measured.
[c] Significantly different from controls, $p<.01$.

TABLE II. Effects of Drugs on Body and Endocrine Organ Weights.

Treatment	Body wt (g)		% Change in body weight	Anterior pituitary (mg)	Adrenals (mg)	Ovaries (mg)
	Initial	Final				
Controls, saline	283 ± 5[a]	286 ± 5	+1.1	14.1 ± 0.8	77.7 ± 3.0	84.3 ± 2.6
Ergocornine, 0.6 mg	277 ± 5	269 ± 8	−3.2	12.1 ± 0.7	76.2 ± 4.5	120.4 ± 4.9[b]
Controls, saline	286 ± 4	291 ± 5	+1.8	14.1 ± 0.7	79.8 ± 2.9	88.2 ± 3.9
Iproniazid, 7.5 mg	277 ± 6	276 ± 5	+0.1	11.4 ± 0.5[b]	68.7 ± 3.1[b]	80.4 ± 5.2
Dopamine, 0.1 mg	281 ± 5	290 ± 5	+3.3	13.9 ± 0.3	72.9 ± 3.4	82.4 ± 4.0

[a] Mean ± standard error of mean.

[b] Significantly different from controls, $p < .01$.

ergocornine and iproniazid injections completely inhibited development and growth of DMBA-induced mammary tumors, whereas dopamine had no effect. Ergocornine significantly lowered both serum and pituitary prolactin concentrations, indicating that this drug depressed secretion of prolactin by the pituitary. This confirms previous indications that ergocornine inhibits prolactin secretion (1–4), and is believed to account for the cessation of mammary tumor growth in the present study. We have recently confirmed that ergocornine can significantly lower serum prolactin levels in rats without altering the estrous cycle (Cassel and Wuttke, unpublished). In agreement with the present results in rats, Yanai and Nagasawa (personal communication) recently found that ergocornine injections suppressed formation of hyperplastic alveolar nodules in C_3H/He mice and reduced pituitary levels of prolactin. Hyperplastic alveolar nodules are believed to represent the preneoplastic state of mammary tumorigenesis in mice, and prolactin is considered to be essential for development of these nodules (11).

Inhibition of mammary tumorigenesis and growth in the rats given iproniazid does not appear to be due to reduced prolactin secretion. On the contrary, serum prolactin levels in the iproniazid-treated rats were elevated, although this increase may not be significant. Iproniazid previously was found to inhibit mammary secretion in rats, but neither pituitary nor blood prolactin was assayed (5). Shubik (12) reported that isoniazid, a derivative of iproniazid, inhibited development and growth of spontaneous mammary tumors in Swiss and C_3H mice, whereas development of lung adenomas was enhanced. The mechanism(s) by which iproniazid inhibited mammary tumor growth in rats is not clear. Dopamine had no effect on mammary tumor development or growth in these rats, and produced no significant change in serum or pituitary prolactin levels. Recent studies in our laboratory have shown that injections of dopamine into rats have no effect on serum prolactin levels (13).

Summary. Mammary tumors were induced in Sprague–Dawley rats by a single intravenous injection of 7,12-dimethylbenz (*a*)anthracene. After these tumors reached about 1 cm in diameter, the rats were injected for 15 days with ergocornine or for 25 days with iproniazid or dopamine. Erogocornine and iproniazid completely suppressed mammary tumor growth and prevented development of new tumors, whereas in the control and dopamine-treated rats, growth of initial tumors increased by about 59–102% and total number of tumors about doubled. Radioimmunoassays revealed that ergocornine significantly reduced serum and pituitary prolactin levels, whereas iproniazid and dopamine had little or no effect on prolactin levels. Ergocornine is believed to inhibit mammary tumor growth by depressing secretion of pituitary prolactin, but the mechanism(s) by which iproniazid inhibited mammary tumor growth is not clear.

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