sponsiveness to noradrenaline but not to adrenaline. The hypotensive response to chlorpromazine was the same in normal and cold-adapted rats and adrenaline did not oppose this effect in both groups. However noradrenaline, which normally slightly antagonized the hypotension of chlorpromazine, was much more effective in that respect when tested on cold-adapted animals. Cold adapted animals were known to be more sensitive to the metabolic effect of noradrenaline; the present study indicates that this increased responsiveness to noradrenaline also exists for its cardiovascular effect.

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Gonadotropic Antiserum Effects in Normal and Irradiated Female Mice.* (26027)

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Antisera to crude sheep pituitary extract with antigonadotropic properties inhibited development and differentiation of ovaries transplanted to the spleen of castrate mice and modified changes usually seen in mouse ovaries after irradiation(1,2). The effects of sera with pituitary antigonadotropic properties upon reproductive organs of normal animals have been differently recorded by others and experimental circumstances have varied (3-7). Therefore, it was deemed necessary to extend observations made on experimentally altered animals to normal young adults using the same materials and procedures. Particular attention was given to gonadotropic content of pituitary glands.

Materials and methods. Female mice of the MFl strain‡ 60-70 days of age at time of first injection were used. Two-tenths ml of antigonadotropic serum was injected subcutaneously daily 5 times a week for 6 weeks. Controls received normal rabbit serum, sheep heart antiserum, or saline; others were not injected. Some mice were x-rayed with injection starting the day after total body exposure to 175 r.§ Details of the method of preparation and assay of antiserum have been given (1,2.8). Briefly, the antiserum is developed by injection of sheep pituitary extract into rabbits until a gonadotropic antagonistic effect appears in the serum. The effect is demonstrable when pituitary extract (the antigen) and antigonadotropic serum (the antibody) are injected simultaneously, but separately, into immature female mice. Preparations comparable to those used in this study inhibit endogenous gonadotropins(9) and are aspecific(10). Thyrotropic activity is measurable,‖ but there appear to be negligible amounts of lactogenic activity. And, on the basis of short term assays, there is little adrenocorticotropic or growth hormone ac-

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† The author was assisted by Rosemarie Tuercke and Edith Reismann.

‡ Purchased from Manor Farms, Staatsburg, N. Y.

§ Physical factors have been given in detail in (2).

‖ Preliminary studies do not indicate that I^{131} uptake after injection of pituitary extract is altered.

TABLE I. Effect of Antigonadotropic Serum on Irradiated and Non-Irradiated Female Mice.

Treatment	No. animals	Body wt (g)	Ovarian wt (mg)	Uterine wt (mg)	Splenic wt (mg)	Thymic wt (mg)	"Wheel"* cells (%)
1. a. None	22	30.5	13.7	99.5	217.9	53.3	20
b. X-ray alone	10	29.0	7.4	84.2	187.9	56.0	
2. a. Saline	19	30.3	10.3	100.9	188.9	49.1	41
b. X-ray + saline	19	29.3	7.0	80.9	244.2	54.2	
3. a. Normal serum	20	30.6	9.2	96.0	315.2	53.1	54
b. X-ray + normal serum	21	30.9	5.9	74.1	308.1	53.2	
4. a. Sheep heart antiserum	9	31.4	10.5	76.6	318.2	43.5	50
b. X-ray + sheep heart antiserum	9	31.2	6.5	61.1	278.6	48.6	
5. a. Antigonadotropic serum	29	32.2	8.6	61.3	375.0	51.1	89
b. X-ray + antigonadotropic serum	27	33.0	3.1	40.1	359.0	55.2	

Probability (P)†	Body wt	Ovarian wt	Uterine wt	Splenic wt
5a *vs* all controls		.4	.001	
5a " normal serum	.05	.5	.02	.1
5a " saline	"	.2	.01	
5a " no treatment		.001	.001	
5b " all controls		.001	.001	
5b " normal serum	.05	"	"	.1
5b " saline	.001	"	"	
5b " no treatment		"	"	

* % ovaries with "wheel" cells indicative of gonadotropic deficiency.

† P values based on SD $= \sqrt{\frac{\Sigma d^2}{N-1}}$.

tivity(11). At autopsy, 6 weeks after irradiation on the third and fourth day after last injection, pituitary glands were removed and frozen until used for assay. Other organs were fixed in Bouin's solution and prepared for histological study by the paraffin method. Hematoxylin and eosin and periodic acid-Schiff and hematoxylin stains were used. Vaginal smears were taken by saline lavage daily 6 days each week during the injection period. The animals were housed in groups of 5 in cages measuring 5x6x12 inches and constantly supplied with water and a mixture of Rockland Farms Mouse Diet and Purina Chow pellets. Four series of 20 mice each were treated at different times between September and January. For assay, single pituitary glands were homogenized in 1.5 ml saline and injected into C57 Black females¶ 22-24 days of age. One-quarter ml of the solution was given twice daily for 3 days with autopsy following on the morning of the fifth day, when ovaries and uteri were weighed.

Results. Organ weight data are recorded in Table I. Ovaries and uteri of mice given antiserum weighed less than controls within irradiated or non-irradiated groups; they weighed most after saline was injected or when no injection was made. As a rule, the weights of ovaries and uteri from animals which had received either normal rabbit serum or sheep heart antiserum were intermediate. Ovaries and uteri of irradiated mice weighed less than those of non-irradiated mice. Differences between ovarian weights of controls after normal rabbit serum when compared with those of antihormone injected females were significant only in case of irradiation. Uterine weight differences, however, were significant throughout.

Essentially similar results were obtained by Rowlands(6) who concluded that persistent corpora lutea in mouse ovaries were probably responsible for maintaining ovarian weights after antihormone treatment. In this

¶ Purchased from Rockland Farms, New City, N. Y.

TABLE II. Effect of Antigonadotropic Serum and Irradiation on Pituitary Gland Gonadotropic Potency.

Material inj. into assay mice	Treatment of donor mice	Assay mice: No. animals	Body wt (g)	Ovarian wt (mg)	Uterine wt (mg)
Nothing (control)	——	34	11.3	3.0	6.0
Sheep pituitary extract*	——	6	13.6	5.9	22.6
Pituitary from donor mice	Castration	28	12.9	3.8	14.9
	No treatment	17	11.3	2.7	7.4
	X-ray + no treatment	8	12.3	3.4	20.8
	Saline	15	11.5	2.7	8.7
	X-ray + saline	17	11.5	4.3	18.6
	Sheep heart antiserum	8	10.8	3.0	8.2
	X-ray + sheep heart antiserum	9	10.2	4.1	20.9
	Normal serum	19	10.9	3.2	8.4
	X-ray + normal serum	21	11.9	4.0	16.9
	Antigonadotropic serum	28	11.9	3.6	15.0
	X-ray + antigonadotropic serum	24	10.6	3.8	13.3

* Equivalent of 25 mg of acetone dried pituitary glands.

study, 11% (4/37) of the antihormone treated non-irradiated ovaries had corpora lutea, whereas 52% (13/25) of ovaries of mice treated with normal serum had corpora lutea. They were present in 90% (27/30) of uninjected mice. Apparently normal follicles with large antra were present in about 80% of ovaries of controls and in 64% of ovaries from mice given antihormone but not irradiated. After irradiation less than 35% of ovaries, irrespective of treatment, had an occasional abnormally small corpus luteum or Graffian follicle.

Cytological differences were evident in ovarian interstitial tissue. Cells with coarsely granular hyperchromatic nuclei were more frequently encountered in ovaries from mice given antihormone (Table I). In addition, the number of these cells per section and intensity of development of hyperchromasia was greater after injection of antihormone. These cells resembled the "wheel" cells which, according to Selye *et al.*(12), appear shortly after hypophysectomy. Essentially similar results were suggested after examination of a few irradiated mouse ovaries confirming earlier studies(2).

On the basis of criteria established by Hooker and Forbes(13) for the mouse uterus, 53% (10/19) of the uteri from non-irradiated groups given antihormone showed slight or no evidence of estrogenic and/or progesterone-like effects. About 20% (4/14) of the uteri in normal serum-treated groups were similarly unstimulated. Uteri of irradiated mice presented much the same response.

Irradiation as well as injection of gonadotropic antiserum increased the gonadotropic potency of pituitary glands to the same degree as in glands from castrate mice. Other injected materials had no effect (Table II). Mandl and Zuckermann(14) reported increased gonadotropic activity within the pituitary glands of irradiated rats, and Levine and Witschi(15) demonstrated increased circulation of gonadotropic hormones after rats were exposed to x-rays. A similar effect on circulating hormones in parabiotic mice after irradiation was shown by Vermande-Van Eck and Chang(16). Meyer *et al.*(18) showed that injection of an antigonadotropic serum under conditions somewhat comparable to those used in this study, increased the gonadotropic content of pituitary glands of rats. The present study indicates that, as in rats, there is enhanced storage of gonadotropic hormones in pituitary glands of mice after antihormone and after irradiation. It would appear that the well known disparity in ultimate outcome of effects of irradiation upon ovaries in these forms does not lie with initial response of the pituitary gland to altered ovarian function.

Typically vaginal cycling continued

throughout the period of observation regardless of treatment. However, the cycles of mice given antihormone were initiated consistently at a fixed time after the fifth injection. It was concluded that there was a release of increased amounts of gonadotropic hormone at these intervals which was responsible for initiation of a new cycle. There were no important differences in length or frequency of vaginal cycles among experimental groups. However, occurrence of vaginal smears with a preponderance of cornified cells was less frequent after antihormone.

Grossly the ovaries of assay mice which received one pituitary gland from donors irradiated, castrated, or treated with antihormone had much the same appearance. The follicles were somewhat cloudy and there was an occasional corpus luteum, whereas follicles of controls were clear, and there were no corpora lutea. Uteri of all assay mice were opaque and had no intraluminal fluid. Those which were stimulated by injection of pituitary glands from mice given antihormone, irradiated, or castrated had heightened epithelium and a slight submucosal edema. The stromal nuclei were fusiform with dense chromatin. Results with the method used here suggest that a single gland of a mouse irradiated with 175 r or given antihormone has sufficient FSH and LH to stimulate release of estrogen from the ovary of the recipient, but that there is slight but insufficient luteotropin present for fully developed luteinization and progesterone formation. In the immature rat Maddock *et al.*(17) concluded that thick-walled, opaque uteri devoid of fluid indicated the presence of both estrogen and progesterone.

Particular attention was given to selection of control materials, since the influence of exogenous factors upon gonadotropic storage by the pituitary gland is well known. For example inanition appears to increase gonadotropic content per milligram of gland(19, 20). Protein deficiency may or may not increase gonadotropic levels of pituitary glands, but in any event it does not prevent the expected increase in gonadotropic activity of the pituitary gland after gonadectomy(21,22). Vit. A deficiency has been shown to elevate gonadotropic potency of the pituitary gland (23), and some reports indicate a similar effect when Vit. E is deficient(24). Herlant (25) reported hypertrophy and hyperplasia of anterior lobe basophiles in rats induced by a wide variety of stressing agents. Nonspecific stress of sufficient magnitude to bring about atrophy of gonads and accessories frequently, if not invariably, induces thymic involution, adrenal hypertrophy, and body weight loss(26). It has been estimated that for each gram of body weight loss there is a 10% decrease in number of estrous cycles (27). In the present study mice which had received antihormone were slightly heavier than controls (Table I), and number of cycles could not be shown to differ among injected groups. Changes in thymic weights were not marked (Table I). Kidneys and thyroids of about one-half of the animals were studied histologically and appeared to be essentially normal. Adrenal glands did not reveal differences among variously treated animals, and measurement of width of the cortex was not pursued after preliminary trials proved unprofitable. Thus it may be noted that gonadotropic content of pituitary glands was not elevated in mice receiving heterologous normal serum or sheep heart antiserum as it was after antigonadotropic serum. There was no evidence of nonspecific stress in control animals.

Summary. Normal and irradiated female mice were given gonadotropic antiserum. Controls received normal rabbit serum, sheep heart antiserum, or normal saline solution. After antihormone serum treatment ovarian and uterine weights were below those of controls. Ovaries of mice after injection of antihormone serum had fewer corpora lutea than did controls, and interstitial tissue gave evidence of diminished gonadotropic stimulation as indicated by increased frequency of cells indicative of gonadotropic deficiency. Uteri lacking histological evidence of stimulation were found most frequently in animals given antihormone serum. Ovaries and uteri of irradiated animals responded in essentially similar manner to treatment, but the response was more extreme. Irradiation and gonadotropic antiserum increased gonadotropic po-

tency of pituitary glands; neither normal rabbit serum nor sheep heart antiserum had any effect. A pattern of vaginal cycling was observed after antihormone treatment which was not apparent in cycles of controls. No important differences were found in thymus weight or in adrenal or thyroid histology.

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Effects of Psychotropic Drugs on Limbic System of Cat.* (26028)

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There has been increasing evidence that the limbic system (rhinencephalon) is involved in emotional responses(1). This suggests that drugs which alter mood might produce changes in this part of the brain. The present paper describes effects on the septum, amygdala and hippocampus of the cat of 2 psychodepressant drugs (chlordiazepoxide† and meprobamate) and 2 psychostimulant agents (iproniazid and imipramine).

Methods. Thirty cats were prepared under ether anesthesia. The trachea was cannulated for artificial respiration and the left femoral vein for drug injection. Bipolar concentric electrodes were inserted with the aid of a Baltimore stereotaxic instrument in anterior and posterior portions of septum, amygdala and hippocampus. Recording electrodes were placed on frontal and occipital areas of cortex. All wound edges were infiltrated with procaine. The ether was withdrawn and the animals immobilized with decamethonium (C_{10}). Stimulation was carried out with a Grass S-4 stimulator, using the following parameters: frequency, 40 per sec.; pulse

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† Librium® formerly referred to as methaminodiazepoxide.