but is subject to considerable variation within certain tumors. These variations, reflecting differences in uptake of radioactive phosphate ion by various portions of the tumor, may be correlated with certain histologic features in the stained sections. Quantitative microdensitometric measurements have been made, and relative P^{32} uptake determined from these data. An attempt is being made to correlate radioautographic features, especially in the gliomas, with histologic appearance and with prognosis.

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Estrogenic and Gonadotrophic Hormone Inhibiting Activity of Some Adrenal Cortical Substances.* (17886)

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The secretion of estrogens by the adrenal cortex has been observed clinically(1-3), and steroids possessing female sex hormone activity have been isolated from adrenal cortical extracts. Englehart(4,5) and Shirrmeister (6) obtained uterine growth with cortical extracts, and Beall(7) isolated estrone from ox adrenals. Desoxycorticosterone acetate has also been found to have estrogenic activity. Hoffman(8) and Gallardo(9) report uterine growth, and van Heuverswyn *et al.* (10) found mammary duct growth with this substance. In view of the administration of commercial adrenal cortical extracts (Lipo Adrenal Cortex, Adrenal Cortex Extract), desoxycorticosterone acetate (DCA), and 11-Dehydro-17-hydroxycorticosterone - 21 - acetate (Merck and Co., Inc.) in large amounts in experimental procedures, it is of practical importance to determine the doses of these substances which cause inhibition of the pituitary gonadotrophic hormone secretion and which result in uterine stimulation.

An ovariectomized immature female rat united in parabiosis with an intact littermate provides an assay technic by which a compound may be tested for both its activity in inhibiting pituitary gonadotrophic hormone secretion and in causing uterine growth. An estrogenic substance administered in an adequate dose to the ovariectomized rat prevents the post-castration hypersecretion of the gonadotrophic hormone and the resultant ovarian hypertrophy in the co-parabiont. The uterine hypertrophy of the castrate rat serves as an index of the estrogenic activity of the injected substance.

Procedures. Thirty-day-old female littermate rats of the Sprague-Dawley strain weighing between 65 and 75 g were joined in parabiosis according to the method of Bunster and Meyer(11) except that metal skin clips were used instead of silk sutures in closing the skin incisions. The right partners were ovariectomized at the time of parabiosis. One group of parabionts was left uninjected

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TABLE I.
The Uterine Stimulating and Gonadotrophic Hormone Inhibiting Activity of Alpha-estradiol, Desoxycorticosterone Acetate, Compound E, Lipo Adrenal Cortex, and Adrenal Cortex Extract.

Compound	Dose (per day)	No. of pairs	Ovaries* (mg)	Uterus† (mg)
None	—	23	160 (110-230)	52 (32-90)
Alpha-estradiol	.0032 gamma	2	146	47
	.0065 "	2	122	60
	.009 "	2	16	45
	.012 "	3	25	51
	.025 "	3	25	135
	.050 "	2	29	168
DCA	250 gamma	2	133	80
	425 "	5	35	67
	500 "	2	20	77
	1000 "	2	23	123
	2000 "	1	35	110
Compound E	500 gamma	2	250	70
	2000 "	2	195	53
Lipo adrenal cortex	0.5 rat unit (.0125 cc)‡	2	170	45
	1 " " (.025 cc)	3	22	65
	2 " " (.05 cc)	4	20	117
	4 " " (.10 cc)	2	23	175
Adrenal cortex extr.	0.625 rat unit (.25 cc)‡	1	125	43
	2 " " (.8 cc)	3	118	52
	4 " " (1.6 cc)	3	65	45

* Ovaries of the left, intact partner.
† Uterus of the right, ovariectomized, injected partner.
‡ Original volume of the extract.

to serve as controls. In the remaining pairs, DCA, Lipo Adrenal Cortex, Adrenal Cortex Extracts, Compound E, and alpha-estradiol were assayed by subcutaneous administration to the ovariectomized partner daily for 10 days, beginning with the day of operation. DCA, Lipo Adrenal Cortex, and alpha-estradiol were dissolved in corn oil, and Compound E suspended in normal saline, so that a volume of 0.1 cc was injected daily. The Adrenal Cortex Extract was injected in relation to rat units without adjusting for volume. The animals were killed on the 11th day after parabiosis, and uterine and ovarian weights were determined.

Results and discussion. The results of these experiments are found in Table I. Ovarian weights of less than 100 mg are considered to indicate that partial inhibition of gonadotrophic hormone occurred, and weights of less than 50 mg are evidence that the inhibition was almost complete. Uterine weights of more than 100 mg indicate definite uterine stimulation by the injected substance.

It is seen that alpha-estradiol inhibited the pituitary secretion of gonadotrophic hormone when administered at a dose less than that required for uterine stimulation. This relationship was found to be true for several other estrogens tested in parabionts and in single *immature* rats, and will be reported in detail elsewhere(12). The DCA followed the same pattern, inhibiting the pituitary gonadotrophic hormone secretion at 425 γ per day, and producing uterine growth at 1 mg per day. On this basis, the gonadotrophin inhibiting activity of 425 γ of DCA is equivalent to approximately 0.01 γ of alpha-estradiol.

Compound E failed to inhibit the gonadotrophin at 500 γ or at 2 mg per day. Since the 2 mg dose caused a marked emaciation in

12. Byrnes, W. W., and Meyer, R. K., unpublished data.

the injected animals, higher doses were not administered.

Lipo Adrenal Cortex was found to inhibit the gonadotrophic hormone secretion at 1 rat unit per day and to produce uterine stimulation at 2 rat units per day. The gonadotrophin inhibiting activity of 1 rat unit of Lipo Adrenal Cortex can be estimated as equivalent to approximately 0.01 γ of alpha-estradiol. Adrenal Cortex Extract produced partial inhibition of gonadotrophin secretion in daily doses of 4 rat units. Due to the large amount of fluid required, higher doses were not tried. We can conclude that Adrenal Cortex Extract has less than one-fourth the gonadotrophin inhibiting activity of the Lipo Adrenal Cortex. The gonadotrophic inhibiting activity of the Lipo Adrenal Cortex and the Adrenal Cortex Extract is not proportional to their adrenal cortical activity, and is probably due to estrogenic substances obtained from the adrenals in the extraction procedures. To the best of our knowledge, DCA has not been isolated from these adrenal preparations, and does not account for the gonadotrophin inhibiting activity of the Lipo Adrenal Cortex or the Adrenal Cortex Extract.

The possibility of interference with the pituitary-ovarian interrelationship should be considered in the administration of these hormones in experimental and therapeutic procedures.

Summary. Lipo Adrenal Cortex and desoxycorticosterone acetate were found to stimulate uterine growth and to inhibit pituitary gonadotrophic hormone secretion when administered to female rats in relatively small amounts. Adrenal Cortex Extract partially inhibited gonadotrophin secretion but was less than one fourth as effective as the Lipo Adrenal Cortex. Large doses of Compound E failed to inhibit the gonadotrophin secretion. The secretion of gonadotrophic hormone is inhibited in *immature rats* by steroids in amounts less than those which result in uterine growth.

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Effect of Folic Acid, Aminopterin and Vitamin K on Growth of Roots of *Allium cepa.** (17887)

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Previous reports have described the effect of colchicine on nuclear divisions in onion root tips(1) and in human tumors(2). The action of colchicine, acenaphthene and a combination of colchicine and x-ray treatments have also been reported(3). It seemed of interest to study the effect of folic acid (pteroylglutamic acid) and its antagonist aminopterin (4-amino pteroylglutamic acid) to determine the influence these agents have on the rate of growth of fundamental tissue since they have been used as theraputic agents in the treatment of cancer. Vitamin K substitute (2-methyl 1,4-naphtho-hydroquinone-diphosphoric acid ester, tetra sodium salt plus 6 H_2O) another agent that gained some popularity in the treatment of neoplasia was studied and compared with colchicine, folic acid and its antagonist in the light of recent claims that this substance combined with

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