of vaccinia virus in roller tube cultures of chick fibroblasts. This resemblance is possibly another expression of the relation of pox viruses to those of fibroma and myxoma(8). Propagation of fibroma virus in tissue culture offers a means of studying the nature of this tumor-inducing virus, its effect on living cells, and its transformation into strains of greater or lesser virulence.

Summary. (1) A mammalian tumor-inducing virus, that of the Shope fibroma, has been propagated in tissue culture. (2) Two lines of fibroma virus were each carried through 14 successive passages in tissue cultures of cottontail testes. (3) One line of virus was carried up to 4 passages in similar cultures of domestic rabbit testes. (4) Growth of virus was accompanied by formation of acidophilic, intracytoplasmic inclusions and by vacuoles containing a substance with a ground glass appearance.

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Effect of Diethylstilbestrol on Plasma 17-hydroxycorticosteroid Levels in Humans.* (22599)

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Evidence to indicate a stimulatory effect of estrogens on the pituitary-adrenocortical system in animals has been reviewed by Roberts and Szego(1). In the guinea pig, Zondek and Burstein(2) found increased urinary corticosteroid excretion following estrogen administration. On the other hand, Vogt(3) has measured directly adrenocortical hormones in the adrenal venous blood of the rat and rabbit, and found diminution of secretion following administration of hexoestrol in the rat but not in the rabbit. In the human, Gemzell(4) found elevated plasma 17-hydroxycorticosteroids during pregnancy, and suggested that such elevations are due to adrenal activation from the increased estrogen which occurs during pregnancy.

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This study is concerned with plasma 17-hydroxycorticosteroid levels following diethylstilbestrol (stilboestrol) administration to human subjects, and was carried out because of the finding of markedly elevated levels in a patient with carcinoma of the prostate while on stilboestrol therapy.

Procedure. Plasma 17-hydroxycorticosteroids were determined by the method of Nelson and Samuels(5), blood being drawn between 8:30 and 9:30 a. m. In this laboratory levels in 30 normal adults of both sexes were 14.4 $\pm$ 6.2 (mean $\pm$ S.D.) μg per 100 ml plasma (range 3.3 to 28.4); these values are similar to those of Bliss *et al.*(6).

Seven men with carcinoma of the prostate, one male (E.W.) with Addison's disease of 12 years duration, and one female (F.P.) who had undergone subtotal adrenalectomy for Cushing's syndrome were studied. In the patients with carcinoma of the prostate, pre-

TABLE I. 17-Hydroxycorticosteroid Plasma Levels (μg/100 ml) in 7 Patients with Carcinoma of Prostate Treated with Stilboestrol.

Last previous stilboestrol	Before				Stilboestrol, 20 mg/day, on days 9-18				After		
	Day 1	4	8	Mean	11	15	18	Mean	22	25	29
14 days	21.4	15.5	20.5	(19.1)	30.8	33.4	14.7	(26.3)	27.4	26.3	29.4
2 years	14.7	10.1	7.5	(10.8)	33.0	14.0	30.5	(25.8)	30.4	15.2	13.4
34 days	11.4	11.7	9.9	(11.0)	54.3	44.5	47.5	(48.8)	36.7	20.2	17.9
8 "	23.7	28.3	25.6	(25.9)	36.1	41.2	34.4	(37.2)	35.6	21.6	22.3
7 "	29.4	19.8	—	(24.6)	—	41.9	39.6	(40.8)	41.5	26.4	22.8
7 "	22.4	22.4	29.4	(24.7)	40.2	30.2	34.8	(35.1)	13.4	20.0	13.4
30 "	22.9	22.0	25.1	(23.3)	48.6	39.8	62.5	(50.3)	—	—	—
			Mean	(19.9)			Mean	(37.8)			

vious treatment with stilboestrol was discontinued, and after an interval without this drug, 17-hydroxycorticosteroid blood levels were determined before, during, and after a 10-day course of stilboestrol, 20 mg per day orally in 2 doses. The same dose was given to the 2 other patients. For comparative purposes and at a later period the patient with subtotal adrenalectomy was tested with ACTH, using 25 i.u. in an intravenous infusion over a 6-hour period on 2 successive days, with blood samples taken at the beginning and end of the infusions.

Results. The blood levels found are presented in Table I. In the case of each of the 7 patients with carcinoma of the prostate the mean 17-hydroxycorticosteroid plasma level before stilboestrol and the mean value while taking this drug were calculated. In each of these patients when the mean 17-hydroxycorticosteroid value before stilboestrol is compared with the mean value while taking this drug, a definite increase during the latter period seems obvious. When the means of the means for the group were compared, the 17-hydroxycorticosteroid plasma levels were significantly higher during stilboestrol therapy than before this medication was given ($P = < .01$). One male with long-standing Addison's disease and maintained on 10 mg oral cortisone acetate twice daily, showed control plasma values of zero when tested on day one and day 3 (the last cortisone was always taken 12 hrs before the blood sample). Stilboestrol 20 mg/day was begun orally on day 3, and 17-hydroxycorticosteroid values of 0.7, 4.5 and 3.8 μg% were found on days 5, 7 and 10 respectively. In the patient with subtotal adrenalectomy, control values were 14.2 and 16.0 μg% while values of 36.9, 35.9, and 40.3 μg% were seen on days 3, 6, and 8 of stilboestrol therapy. The values found on the 2-day ACTH test were 12.4 μg% (before), 18.9 (after) on day one; and 20.2 (before), 21.9 (after) on day 2.

Discussion. The increased levels of plasma 17-hydroxycorticosteroids found in the patients with carcinoma of the prostate while taking stilboestrol might be attributed to (1) interference of the drug with the Porter-Silber color reaction, (2) measurement of a metabolite of stilboestrol, (3) stimulation of the pituitary-adrenal system, (4) interference with normal pathways for removal of adrenal steroids from the blood.

It appears improbable that there was interference with the color reaction since 40 μg of crystalline stilboestrol in chloroform when run through the chromatographic and color developing procedures resulted in a negligible absorption in the range of 410 mμ. Although unlikely because of the benzenoid character of stilboestrol, the possibility exists that some metabolic product of stilboestrol was responsible for the elevated values found. Evidence against this is the small rise produced in the blood levels of the patient with Addison's disease after 7 days on stilboestrol. If a stilboestrol metabolite were produced and measured, a rise similar in magnitude to that seen in the other patients might be expected since all were on the same dose of stilboestrol. Other than male patients with absolute Addison's disease, suitable control patients to test the hypothesis of pituitary-adrenal stimulation would have been males having com-

plete adrenalectomy with no remaining aberrant adrenal tissue, or males with complete anterior pituitary failure (as from hypophysectomy), whose adrenals were not atrophic and would therefore be capable of responding to stimulation. In lieu of such patients, one normally menstruating female (F.P.) who had had subtotal adrenalectomy because of Cushing's syndrome was studied after stilboestrol as well as after ACTH administration. The results were difficult to interpret since they showed normal control levels, poor response to ACTH, and elevated levels after stilboestrol.

The elevated plasma 17-hydroxycorticosteroid values observed following stilboestrol therapy may indeed indicate pituitary-adrenal stimulation. This would be consistent with the evidence in certain animals which indicates pituitary-adrenal activation by estrogens(1,2), but not with the findings of Vogt (3) using hexoestrol in rats. It would support the suggestion of Gemzell(4) that the elevated plasma 17-hydroxycorticosteroid levels found in pregnancy are due to estrogen stimulation of the pituitary-adrenal system. The poor response to ACTH in the patient with subtotal adrenalectomy is not consistent with this theory. The results in this individual suggest that the adrenal remnant was functioning maximally at all times and could hardly be stimulated further by an effect of stilboestrol on the anterior pituitary.

Another hypothesis which would explain the findings in the patients with carcinoma of the prostate is that stilboestrol may interfere with the metabolic pathway normally employed in the removal of 17-hydroxycorticosteroids from the blood, with the resulting accumulation reflected in elevated plasma levels. The data obtained on the patient with subtotal adrenalectomy seem to fit best with this hypothesis; also this hypothesis could account for the results in the patient with Addison's disease. It cannot be concluded from the present data whether stilboestrol stimulated the pituitary-adrenal system in these patients, or whether it blocked a metabolic pathway for disposal of 17-hydroxycorticosteroids.

Summary. Seven patients with carcinoma of the prostate showed a significant rise in plasma 17-hydroxycorticosteroid levels during treatment with stilboestrol as compared with pretreatment control levels. In a male with Addison's disease such a rise was not seen, while in a female with subtotal adrenalectomy a greater rise in these blood levels occurred following stilboestrol than following ACTH administration. Several mechanisms to explain these observations are discussed.

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