sium from uremic dogs by continuous circulation of blood through a cation exchange resin has been described. This method has been shown to reduce the extracellular potassium concentration by as much as 50% within 4 to 6 hours. However, it is necessary to replace calcium to prevent tetany when this method is used.

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Local Action of Adrenocortical Preparations on Secondary Sexual Organs.* (20695)

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The known local effects of adrenocortical steroids include histological alteration of the skin(1), suppression of hair growth(2,3) and of wound healing(4), and alleviation of inflammation resulting from a number of causes (5-7). Local effects on the reproductive organs have not been demonstrated. Three experiments will be described in this report which are concerned with (a) the local action of adrenocortical extract (ACE) on the vaginal mucosa, (b) the effect of intravaginal application of cortisone on stimulation of the vaginal mucosa elicited by subcutaneously injected estrone, and (c) the effect of direct application of cortisone on stimulation of the chick comb elicited by subcutaneously injected testosterone propionate (TP).

Procedure. For study of the local action of ACE† on the vaginal mucosa, 0.1 ml of hog adrenal extract (equivalent to 100 μg of cortisone as determined by the liver glycogen test) or of the solvent, 25% ethanol, was introduced daily into the vagina of adult Long-Evans rats which had been ovariectomized 8-13 days previously (Table I). Daily vaginal smears were made and the vaginae prepared for histologic study at the end of the experiment. For the study of the antagonism of cortisone to estrone, 16 young adult Long-Evans rats, which had been ovariectomized 90 days previously, were primed with 14 μg of estrone, half being given subcutaneously on each of 2 successive days. Only rats which showed cornification within 48 hours were used in the subsequent study. Vaginal smears were made 10 days after the last priming injection to ensure that the estrogenic effect had subsided. On the next day the rats were divided into 4 groups and treated as follows: (a) estrone subcutaneously, cortisone intravaginally; (b) estrone subcutaneously, saline vehicle intravaginally; (c) peanut oil subcutaneously, cortisone intravaginally; and (d) peanut oil subcutaneously, saline vehicle intravaginally. One-tenth ml of estrone (3 μg) or of peanut oil was injected subcutaneously on the 12th and 13th day after priming, 6 μg of estrone being just sufficient to produce full cornification in the primed rats as determined by previous experiments. Intravaginal injections of cortisone or saline vehicle were made with a short, blunted hypodermic needle to which a shield was attached. The cortisone was prepared by suspending 80 mg in a vehicle con-

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TABLE I. Effect of Daily Intravaginal Application of Adrenocortical Extract and Ethanol.

Treatment	No. of rats	% cornified	Mean No. days before cornified	Total days, Rx	Mean organ wt (mg) Uterus	Thymus	Adrenal
ACE	22	100	4	8	94 ± 4.1*†	266 ± 21.9	56 ± 5
25% ethanol	20	0	—	8	71 ± 8.1‡	327 ± 73	55 ± 4.1

* 5 rats. † Stand. dev. ‡ 4 rats.

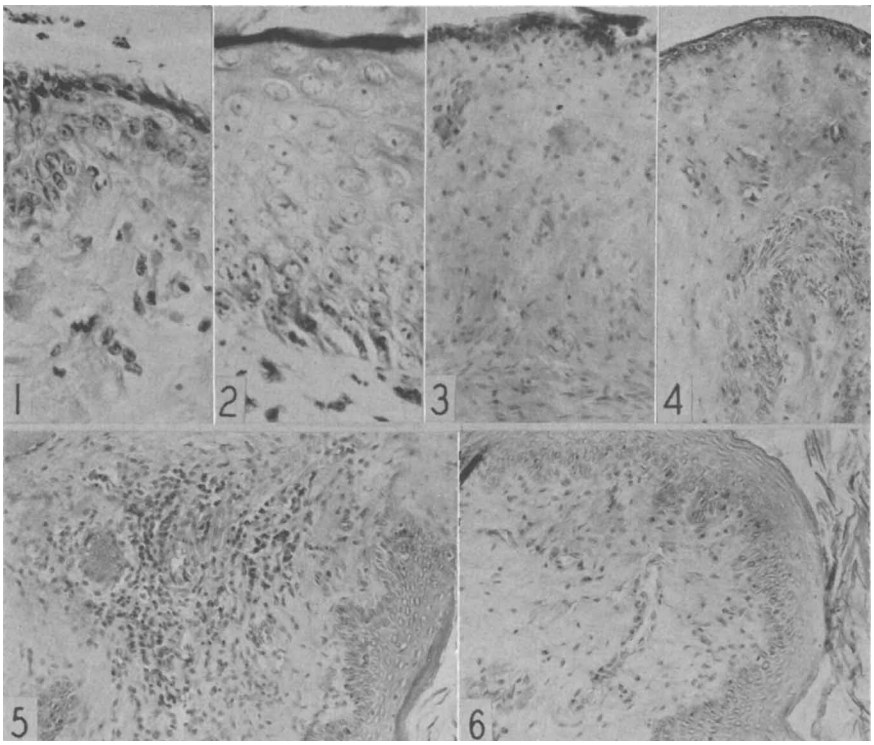
sisting of 2 drops of Tween 80 and 16 cc of 0.9% NaCl solution. One-tenth ml of cortisone or of the saline vehicle was administered 3 times daily from the 11th through the 14th day after priming. Vaginal smears were taken in the morning and evening of each day prior to the intravaginal treatment. All animals were sacrificed on the 15th day and the vaginae prepared for histologic study. For study of the effect of direct application of cortisone on stimulation of the chick's comb by TP, 2-3 day-old male Barred Rock chicks were treated as follows, the doses of TP and peanut oil being given subcutaneously once daily and those of cortisone and ethanol being applied directly to the comb thrice daily: (a) 10 chicks received 150 μg TP in sesame oil (.075 ml) and 40 μg cortisone (.02 ml) in 50% ethanol, (b) 10 received 150 μg TP and .02 ml 50% ethanol and (c) 9 received .075 ml sesame oil and .02 ml 50% ethanol. After 8 days the chicks were killed and the combs removed and weighed according to the method of Frank *et al.*(8).

Observations. (a) *ACE and vaginal mucosa.* After 8 days of treatment, the vaginal mucosa of all rats receiving ethanol intravaginally remained in the condition characteristic of ovariectomy as shown by diestrous vaginal smears and the histology of the vaginal mucosa (Fig. 1). The mucosae of all rats receiving ACE were cornified after an average of 4 days of treatment (Table I) and the epithelium was hyperplastic (Fig. 2). Weight of the adrenal glands was unchanged by treatment with ACE, but that of the thymus was reduced and of the uterus, increased.

(b) *Subcutaneous estrone and intravaginal cortisone.* The control group which received peanut oil subcutaneously and saline vehicle intravaginally showed no stimulation of the vaginal epithelium as determined by observation of vaginal smears or of the histologic sections. When peanut oil was given subcutaneously and cortisone intravaginally, no change from the ovariectomized condition was observed in the smears or histology of the vaginal epithelium. However, the concentration of cells in the lamina propria, especially of fibroblasts, was reduced and the connective tissue fibers were somewhat condensed (Fig. 4) as compared with rats which received saline intravaginally (Fig. 3). Intravaginal administration of saline vehicle did not modify the action of subcutaneously injected estrone, all rats showing full cornification 48 hours after the last injection of estrone (Fig. 5). Many mononuclear cells had infiltrated into the perivascular connective tissue of the lamina propria. Intravaginal application of cortisone failed to retard the cornification or hyperplasia of the vaginal epithelium induced by subcutaneously injected estrone but suppressed strikingly the perivascular infiltration of mononuclear cells into the lamina propria (Fig. 6).

(c) *Testosterone, cortisone, and chick's comb.* Table II demonstrates that the amount of TP injected caused a significant increase in weight of the comb and that treatment of the comb with cortisone suppressed this stimulation to a slight but statistically insignificant degree.

Discussion. The demonstration of estrogenic activity in ACE supports previous work in which estrone was isolated from the adrenal gland(9), and subcutaneous injection of ACE stimulated uterine growth(10,11) and inhibited secretion of gonadotrophin by the anterior hypophysis(12). Since most of the adrenocortical activity of our extract was due to the presence of the most potent growth inhibitor of the known adrenocortical compounds (17-hydroxycorticosterone(13)), it is surprising that the capacity of estrogen to



All figures illustrate the vaginal mucosa.

FIG. 1. One-tenth ml ethanol intravag. daily for 8 days.
FIG. 2. " " ACE for 4 days.
FIG. 3. Peanut oil subcut. and saline intravag.
FIG. 4. " " " " cortisone " .
FIG. 5. Estrone " " saline " .
FIG. 6. " " " cortisone " .

stimulate hyperplasia of the vaginal epithelium was not over-ridden. The presence of significant estrogenic activity in ACE must be taken into account in comparing the action of these preparations with that of any pure adrenocortical compound.

The failure of cortisone to stimulate the vaginal mucosa by local action supports the finding that no estrogenic activity is observed when this hormone is administered subcutaneously(14). The failure of cortisone to inhibit stimulation of the vaginal epithelium by subcutaneously injected estrogen and of the adrenocortical compounds in ACE to over-ride estrogenic activity present in the extract is due undoubtedly to the extreme sensitivity of the vaginal epithelium to the growth-stimulating action of estrogen. On the contrary others(14,15) have shown that estrogenic stimulation of the uterus may be antagonized by cortisone when it is administered *systemically*. The inhibition by cortisone of cellular infiltration of mononuclear cells into connective tissue of the lamina propria illustrates an action of the hormone which has been demonstrated under many circumstances(6).

The failure of local cortisone to inhibit growth of the chick's comb is in agreement with the results of Schiller *et al.*(16). Their experiment was repeated because they applied cortisone when dissolved in corn oil. Previous studies in this laboratory(17) showed that when adrenocortical compounds are dissolved in oils their growth-inhibiting capacity as determined by inhibition in rate of hair growth and of wound healing is relatively weak.

Summary. Under conditions of these ex-

TABLE II. The Effect of Subcutaneous TP and Local Cortisone on Weight of Chick's Comb.

Group	Mean body wt (g)	Mean comb wt (mg)	Comb wt (mg) / Body wt (g)
TP + cortisone	71 ± 8.1*	62.6 ± 12.9 P<.3†	.86 ± .15 P<.5
TP + ethanol	78 ± 8.5	73.6 ± 25.6	.94 ± .30
Ses. oil + ethanol	73 ± 11.4	23.3 ± 36.6	.31 ± .06

* Stand. dev. † Fisher student t test.

periments adrenocortical steroids failed to suppress the growth-stimulating action of sex hormones on secondary reproductive organs as revealed by hyperplasia and cornification of the vaginal epithelium which resulted from direct application of ACE, by failure of locally applied cortisone to suppress estrogenic stimulation of the vagina and of androgenic stimulation of the chick's comb.

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Comparison of *in vitro* Serum Protein Binding of Thyroxin and Triiodothyronine.* (20696)

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(Introduced by Ovid O. Meyer.)

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It has been observed that 3:5:3′ L triiodothyronine disappears more rapidly than L thyroxin from blood following intravenous administration(1). It is possible that this observation may be explained by a difference in serum protein binding of these two amino acids inasmuch as thyroxin, but not triiodothyronine, has been shown to associate with a specific alpha globulin *in vivo*(2).

The purpose of this paper is to report on the *in vitro* binding of L thyroxin and 3:5:3′ L triiodothyronine by serum proteins.

Methods and materials. The radioactive amino acids† employed in these experiments were of sufficiently high specific activity to permit the use of quantities that were approxi-

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† The radiothyroxin‡ used in these studies contained on chromatographic analysis nearly 100% of its radioactivity as thyroxin. The radio triiodothyronine‡ was found to contain approximately 5% of its radioactivity as thyroxin.

‡ Obtained from Abbott Laboratories.