stroma cell nuclei after treatment with progesterone. In the animals receiving progesterone in addition to the estrogen the cellular changes in the tunica propria seen after progesterone alone were added to the typical estrogen effects.

In the androgen-treated animals the endometrium was more or less intermediate between that of the castrates and of the animals receiving progesterone. The tunica propria was slightly thickened and more fibrous than in the castrates; but the stromal nuclei did not become spherical and vesicular, nor were many mitoses in evidence. In the animals receiving both testosterone and progesterone the effects of both substances were seen in the stroma, although the progesterone characteristics were less marked than in animals receiving progesterone alone.

The tunica propria of the uteri of the few pregnant and lactating mice which have been examined also exhibited nuclei like those described in the castrates receiving progesterone. Apparently, therefore, both endogenous and injected progesterone elicit morphologic change in the uterus of the mouse. Moreover, the nuclear changes in the uterine tunica propria may constitute a specific response to progesterone in the mouse.

Summary. With or without concomitant treatment with estrogen, the daily injection of 0.25 or 0.5 mg of progesterone into mice ovariectomized 3 weeks previously evoked mitosis in all the tunics of the uterus. The nuclei of the cells of the uterine tunica propria became larger and transformed from the fusiform, pyknotic type into vesicular nuclei. The latter condition was also seen in pregnant and lactating mice. Neither estrogen alone nor androgen provoked this morphology.

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Effect of Desoxycorticosterone and Testosterone on Water and Chloride Metabolism.

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Selye, *et al.*,[1] showed that high doses of estrogens cause water retention in the rat and this has subsequently been confirmed in the dog by Thorn and Engel.[2] These latter investigators claimed,

1 Selye, H., Harlow, C. M., and Collip, J. B., *Endokrinol.*, 1936, **18**, 81.

2 Thorn, George W., and Engel, Lewis L., *J. Exp. Med.*, 1938, **68**, 299.

however, that such an antidiuretic effect is characteristic not only of the estrogens but also of other physiologically active steroid hormones such as the androgens and progesterone. They stated furthermore, that estrone, desoxycorticosterone acetate, corticosterone acetate and androgens decrease the urinary elimination of chlorides, thus the various active steroids appeared to have the same action on water and chloride metabolism irrespective of their specific physiological effects. The subsequent experiments of Selye and Dosne[3, 4] indicated, however, that desoxycorticosterone acetate (D.C.A.) and progesterone differ in their actions on blood chlorides inasmuch as the former elicits pronounced hypochloremia in the rat, while the latter exerts no such effect. Selye and Bassett[5] showed furthermore, that in the rat progesterone causes a pronounced increase in urine output especially after hypophysectomy. In view of these findings we decided to extend our studies and undertake a systematic investigation of the effect of testosterone and D.C.A. on water elimination and chloride metabolism both in the normal and in the hypophysectomized rat.

In our first experiment 24 female albino rats weighing 91-107 g were divided into 4 groups of 6. Group I was hypophysectomized and received D.C.A. in a peanut oil solution each ml of which contained 20 mg. During the first two days, 2 mg were administered once daily, then the same dose was given twice daily until the thirteenth day, after which 4 mg were injected twice daily subcutaneously, the total dose administered during the 21 days of the experiment being 112 mg per rat. Group II was hypophysectomized but received only as much peanut oil as had to be administered to the treated animals as a solvent for D.C.A. All hypophysectomies were performed on the same day on which the injections were started. Group III was given the same amount of D.C.A as Group I but was not hypophysectomized. Group IV acted as intact controls and received oil only.

All rats were kept in individual metabolism cages and the water intake and urine output were measured daily. The animals were given "purina fox chow" and ground oats *ad lib.* with a few leaves of celery throughout the experimental period except on 3 days on which the effects of changes in food and water intake were studied. Fig. 1, which gives the average water intake and urine output on

[3] Selye, Hans, and Dosne, Christiane, PROC. SOC. EXP. BIOL. AND MED., 1940, **44**, 165.

[4] Selye, Hans, and Dosne, Christiane, *Am. J. Physiol.*, in press.

[5] Selye, Hans, and Bassett, Lucy, PROC. SOC. EXP. BIOL. AND MED., 1940, **44**, 502.

the fifth day is representative of conditions as they existed throughout the experiment on days on which water and food were given *ad lib.* It will be seen that D.C.A. markedly increased both the water intake and diuresis but this effect was much more pronounced in the hypophysectomized than in the normal animals.

On the seventh day, food and water was withheld for a period of 24 hours. At the end of this time the normal controls excreted 3.0 ml, the D.C.A.-treated intact animals 7 ml, the hypophysectomized controls 3.5 ml and the hypophysectomized D.C.A.-treated 4 ml of urine on the average. This indicates that in the absence of food and water, the action of D.C.A. was not so obvious as otherwise, especially in the hypophysectomized group; yet the hormone

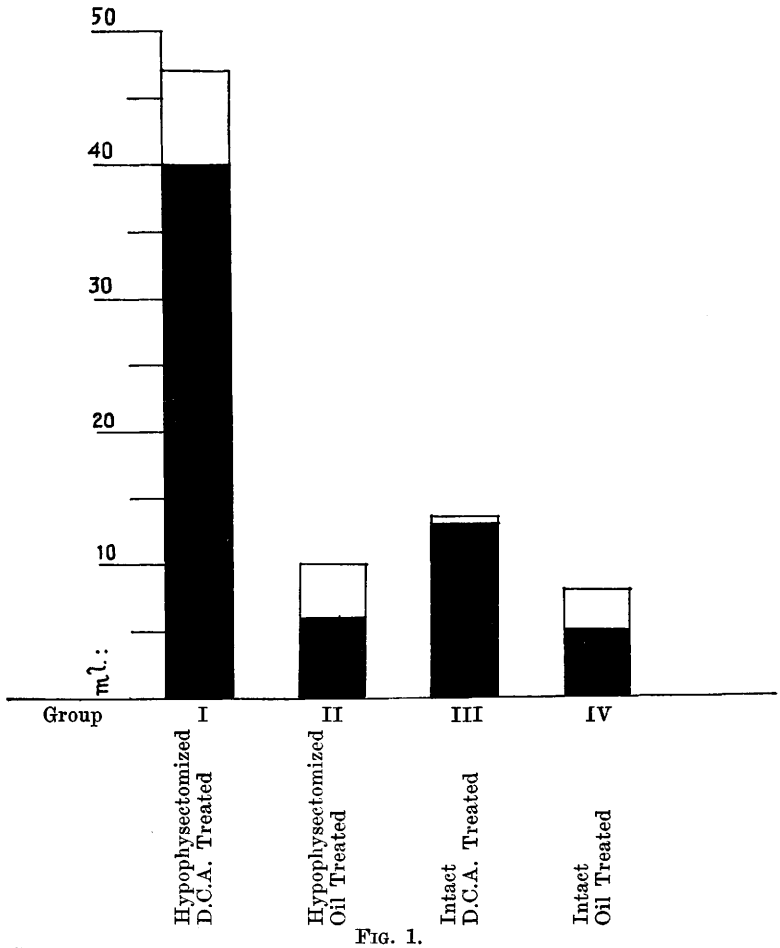

FIG. 1.

Graph illustrating the average urine output of normal and hypophysectomized rats treated with D.C.A. and receiving food and water *ad lib.*

retained some of its diuretic action so that this effect cannot be regarded as a mere consequence of increased water intake. On the twenty-first day the animals were fasted but received 10 ml of water by stomach tube in order to determine whether ingested water is excreted more rapidly by the D.C.A.-treated than by the normal animals. This proved to be the case both in the intact and in the hypophysectomized series so that it may safely be concluded that the action of D.C.A. is not merely due to increased voluntary water ingestion.

The effect of D.C.A. on whole blood and urine chlorides was determined several times during the experiment by the Van Slyke method and Cl is expressed in Table I as NaCl. The figures which appear in brackets in the table indicate the range of the variations in each group. The results given in this table as a representative example were obtained on the 11th day after a 24-hour fasting period so as to avoid post-absorptive variations. Water was allowed *ad lib.* during this period.

As Fig. 1 and Table I indicate, D.C.A. caused marked diuresis both in the normal and in the hypophysectomized animal but this effect was particularly marked in the latter. In some of the D.C.A.-treated hypophysectomized animals, this diuresis may well be compared with that of severe diabetes insipidus. Only average values are given here in order to save space, but we may mention as an example of a particularly severe case that in one animal whose body weight decreased to 90 g after hypophysectomy the urine output reached 85 ml/24 hours on certain days, that is to say, the animal voided almost the equivalent of its own body weight in a single day. The blood chlorides were consistently and markedly decreased by D.C.A. in the intact animals throughout the experiment irrespective of the water intake. No such significant decrease could be obtained, however, after hypophysectomy. The urinary

TABLE I.
Effect of D.C.A. on Urine Volume and Blood and Urine Chlorides of Fasted Normal and Hypophysectomized Rats Receiving Water *ad lib.*

Group	I	II	III	IV
Treatment	Hypophysectomized D.C.A.	Hypophysectomized Oil	Intact D.C.A.	Intact Oil
Urine in 24 hr (in ml)	16.8 (14.5-21.5)	4.1 (2.5-5.5)	20.7 (14.5-28.5)	9.9 (4.0-21.0)
Chlorides/100 ml Urine	693 (410-1290)	916 (700-1230)	750 (585-880)	470 (410-585)
Total Urine Chlorides, 24 hr	116 (73.8-187)	37.5 (17.5-51.4)	155 (84.8-199.5)	46.5 (18.8-86.1)
Chlorides/100 ml Blood	423 (386-468)	433 (398-468)	362 (346-374)	441 (416-468)

excretion of chlorides per 24 hours was always greatly increased by D.C.A. irrespective of the presence or absence of the hypophysis. This was due mainly to the increase in urine volume and not to an increased chloride concentration, although on certain days (Table I) the concentration of the chlorides in the urine was also increased at least in intact rats. It will be seen that although in the hypophysectomized animals, D.C.A. failed to decrease the blood chloride concentration, it still increased the total chloride output. The chloride concentration of the urine on the other hand, was not significantly influenced by this steroid. D.C.A. causes marked diuresis even in the hypophysectomized animal in which it does not decrease the blood chlorides. This is in agreement with the finding that progesterone causes polyuria without significantly influencing the blood chloride concentration[5] and indicates that the diuretic effect of these steroids is not dependent upon changes in blood chlorides. The increase in urinary chloride elimination and decrease in blood chlorides reported in this communication are difficult to reconcile with the finding that this same compound causes a rise in blood sodium and a decrease in the urinary sodium excretion.[6, 7] Further experiments will have to show whether these changes in Na and Cl metabolism can be produced with the same dose and at the same time. If this should prove to be the case, it is evident that at least a considerable part of the blood Na and urinary Cl of D.C.A.-treated rats is not present in the form of NaCl.

In connection with these experiments, we should also like to mention that determinations performed on all animals on the last day of the experiment indicate that D.C.A. causes a noteworthy increase in the circulating blood volume both in the hypophysectomized and in the intact animals.

We also studied the actions of testosterone (not esterified) administered in exactly the same manner and dosage as D.C.A. was given in the above experiments. We omit a detailed description of these experiments since testosterone proved to have no effect on diuresis or chloride excretion either in intact or hypophysectomized rats even though such high dosages had been administered. The only significant change observed was a slight decrease in the whole blood chlorides, a fact which is in agreement with the previous observations of Selye and Dosne.[8] These observations clearly indi-

[6] Hartman, Frank A., Spoor, H. J., and Lewis, L. A., *Science*, 1939, **89**, 204.

[7] Wells, B. B., and Kendall, E. C., *Proc. Staff Meet. of the Mayo Clin.*, 1940, **15**, 133.

[8] Selye, Hans, et Dosne, Christiane, *Annales d'Endocrinol.*, in press.

cate that there are significant qualitative differences in the action of the various steroid hormones on electrolyte and water metabolism. Since previous observations indicated that androgens, progesterone and D.C.A. cause kidney enlargement due to hypertrophy of the proximal and distal convoluted tubules,[9, 10] we were at first inclined to regard the diuretic action of progesterone and D.C.A. as a consequence of this kidney change. However, the fact that androgens, which are most active in causing renal enlargement, do not elicit diuresis clearly indicates that there is no correlation between the kidney enlarging and the diuretic action of steroids. In conclusion, it may also be mentioned that progesterone, D.C.A., and testosterone all cause significant kidney enlargement even in the hypophysectomized rat but in the latter this effect was much less pronounced than in intact animals. The mechanism of the renal effect will be discussed elsewhere. Suffice it to state here that unlike the mammogenic action of the steroids[11] the renal effect is not entirely mediated by the hypophysis nor is it completely independent of this gland as are the actions of steroids on vagina, oviduct and uterus.[11]

Summary. In the rat, desoxycorticosterone acetate (D.C.A.) causes marked diuresis, accompanied by increased chloride excretion, but not by a constant increase in the chloride concentration of the urine. The diuretic effect is much more pronounced in the hypophysectomized than in the intact rat. Unlike in the intact animal, D.C.A. fails to decrease the blood chlorides in the hypophysectomized rat. The hyperchloruria and polyuria cannot be considered, therefore, as the consequence of the hypochloremia.

The kidney hypertrophy elicited by progesterone, D.C.A., and androgens is likewise not the cause of the polyuria since testosterone —which is most active among the steroids as far as kidney enlargement is concerned—does not influence diuresis. The kidney-enlarging action of all three above mentioned steroids is considerably decreased but not prevented by hypophysectomy.

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9 Selye, Hans, *Montreal Physiol. Soc. Meet.*, 1939.

10 Selye, Hans, *J. Urol.*, 1939, **42**, 637.

11 Selye, Hans, *Anat. Rec.*, in press.