decrease the tubular secretion of penicillin and raise its concentration in the plasma after injection. The results reported indicate that cinchoninic acid derivatives which decrease the rate of excretion of phenol red will also block the renal tubular secretion of penicillin.

Summary. Similar to carinamide, 3-hydroxy-2-phenylcinchoninic acid causes higher concentrations of penicillin in the plasma when injected before the administration of the penicillin. Both substances also decrease the rate of excretion of phenol red. The activities of the two drugs are roughly of the same order of magnitude. This increase in concentration of penicillin in the plasma and decrease in rate of excretion of phenol red is attributed to a blocking of the renal tubular secretory mechanism.

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Desoxycorticosterone Acetate and Adenohypophyseal Content of Adrenocorticotrophic Hormone.* (18012)

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Twenty-four hours after adrenalectomy the content of adrenocorticotrophic hormone (ACTH) in the rat adenohypophysis is reduced to one-fifth of normal(1). Sayers and Cheng(1) favor the view that this marked depletion of pituitary ACTH is a consequence of rapid and uninhibited discharge of trophin in response to the absence of circulating cortical hormone. The data of the present paper are compatible with this interpretation. They demonstrate the desoxycorticosterone acetate (DCA) prevents the marked depletion of ACTH which normally occurs after adrenalectomy, and that DCA increases the concentration of ACTH in the adenohypophysis of the intact rat.

Methods. The donor rats were male animals obtained from the Sprague-Dawley farm. The body weights of these animals are presented in Table I. DCA pellets (six-15 mg pellets per rat) were implanted 72 hours before sacrifice and adrenalectomy was performed 24 hours before sacrifice. The animals were given free access to food and water at all times except those in Experiments 3 and 4, from whom food was withdrawn 24 hours previous to sacrifice. The animals were anesthetized with sodium pentobarbital, exsanguinated by severing the abdominal aorta and the pituitary dissected away from the sella. The anterior lobes were carefully freed of posterior pituitary tissue, frozen and lyophilized. The concentration of ACTH in the dried tissue was determined in hypophysectomized recipient rats by the method of Burns *et al.*(2).

A previous study(2) has demonstrated that each mg of dried adenohypophysis from untreated intact rats contains the biological equivalent of 80 μg of an ACTH standard (La-1-A†). This value was employed in the calculation of pituitary ACTH content. The weight of the pituitary tissue multiplied by ACTH concentration gives total hormone content per pituitary. For example, in Experiment 1 the content of ACTH in each pituitary of the intact untreated controls was obtained by multiplying 1.35 mg, the average weight of each pituitary, by 80 μg per mg, the concentration of ACTH in the tissue.

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† Kindly supplied by Armour Laboratories.

TABLE I.
Effect of DCA on Pituitary Content of ACTH.

Exp. No.	Donor rats	No. donor rats	Avg body wt of donor rats, g	No. recipient rats	b	P	s	λ	M	Antilog M	S_M	Antilog (M — S_M) to antilog (M + S_M)	Pituitary dry wt, mg	ACTH per pituitary μg of La-1-A
1	intact DCA pellets	6	277	9	254	.2	42.7	.168	.132	1.36	±.090	1.10-1.67	1.25	136 (110-167)
	"	6	302	9	151		31.1	.206					1.35	108
2	" " "	6	328	12	73	.05-.02	23.9	.328	.154	1.42	±.101	1.13-1.80	1.57	179 (142-226)
	"	6	342	12	117		22.4	.191					1.57	126
3	sham adrenex	3	330	8	130	.7	29.7	.228	$\bar{1}$.861	.73	±.097	.58- .91	1.17	68 (54-85)
	intact	7	267	13	149		30.0	.201					1.19	95
4	adrenex	10	256	11	136	.7	24.4	.180	$\bar{1}$.306	.20	±.083	.17- .25	.98	16 (13-20)
	intact	7	267	13	149		30.0	.201					1.19	95
5	adrenex	6	297	9	171	.7	25.9	.151	$\bar{1}$.414	.26	±.084	.21- .31	1.16	24 (20-29)
	intact	6	302	9	151		31.1	.206					1.35	108
6	adrenex " "	6	289	9	180	.6	22.1	.123	$\bar{1}$.627	.42	±.078	.35- .51	1.19	40 (33-49)
	intact	6	302	9	151		31.1	.206					1.35	108
7	adrenex	6	238	11	191	.4	31.4	.165	$\bar{1}$.188	.15	±.130	.11- .21	1.08	18 (13-25)
	intact " "	6	247	10	103		8.6	.084					1.08	120
8	adrenex " "	6	253	8	75	.3	2.9	.038	$\bar{1}$.635	.43	±.059	.38- .50	1.15	55 (49-64)
	intact " "	6	247	10	103		8.6	.084					1.08	120

b = slope of the log dose response curve; P = probability factor measuring the significance of the difference between the slopes; s = standard deviation of all of the individual responses about this curve (a straight line fitted by the method of least squares); λ = s/b, a measure of the accuracy of the assay method; M = logarithm of the ratio of the potencies; S_M = standard error of M; antilog (M — S_M) to antilog (M + S_M) is the range of estimate for one standard error.

Each of the pituitaries of intact animals treated with DCA had an ACTH content equal to 1.25 mg, the average weight of each pituitary, times 80 μg per mg multiplied by 1.36, the ratio of potencies of the DCA-treated to the untreated animals.

Results. The assay data of the various experiments are presented in Table I. For a detailed discussion of the statistics the reader is referred to the paper of Sayers *et al.*(3). Suffice it to point out here that the antilogarithm of M gives an estimate of the ratio of potencies of the assay pairs. The calculation antilogarithm $(M - S_M)$ to antilogarithm $(M + S_M)$ is the range of the estimate for $P = 0.67$.

In Exp. 1 and 2 (Table I) the influence of DCA upon the concentration of ACTH in the adenohypophysis of the intact rat was determined. In both experiments DCA induced a small but significant increase in both the concentration and content of adenohypophyseal ACTH. In Exp. 2, for an unexplained reason, the slopes derived from the data of the two members of the assay pair appear to be different. The P value for the significance of the difference of the slopes is 0.05 to 0.02.

Sham adrenalectomy (Exp. 3), performed 24 hours before sacrifice, resulted in a significant reduction (27%) in the concentration of pituitary ACTH. Bilateral adrenalectomy induced a very marked depletion of adenohypophyseal stores of ACTH. In Exp. 4, an 80% and in Exp. 5, a 74% reduction in hormone concentration occurred 24 hours after this operation.

Exp. 5 and 6 were conducted simultaneously. The concentration of ACTH in the pituitaries of the untreated adrenalectomized animals of Exp. 5 and that of the DCA-treated animals of Exp. 6 were each compared with the ACTH stores in the same untreated intact controls. DCA appreciably inhibited the marked reduction in pituitary concentration of ACTH which normally follows adrenalectomy. However, the steroid did not restore the value to that of intact sham-operated animals.

In Exp. 7 and 8, ACTH stores in untreated and DCA-treated adrenalectomized animals were compared with those in DCA-treated intact rats. The results confirm those of Exp. 5 and 6. DCA actually increased pituitary ACTH content in adrenalectomized rats to a value not significantly less than that in sham-operated intact animals.

Discussion. DCA induces adrenal cortical atrophy(4-8) and prevents the reduction in adrenal ascorbic acid which normally occurs when the rat is subjected to acute stress(9). These observations suggest that the steroid inhibits discharge of ACTH from the adenohypophysis. The data of the present report demonstrate that DCA increases the ACTH content of the pituitary in intact rats and inhibits the depletion of pituitary stores of ACTH which normally follows adrenalectomy. The results are compatible with the concept that DCA depresses the rate of discharge of ACTH from the adenohypophysis. However, they do not constitute proof of such a concept. It is to be emphasized that pituitary content of hormone is not necessarily related to rate of discharge of hormone. (For an analysis of this problem, see the paper by Sayers and Cheng(1).) However, it has been demonstrated by Taylor *et al.*(10) that the blood of patients with untreated Addison's disease, but not that of normal subjects, contains detectable quantities of ACTH.

DCA in the doses employed in these experiments allows some depletion of pituitary ACTH stores in adrenalectomized rats even when the comparison is made against the hormone content of the pituitaries of sham-operated animals. It would be of interest to know whether larger doses of DCA bring about complete inhibition of pituitary loss of

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ACTH. Additional studies employing various doses of both DCA and cortisone are contemplated.

The metabolic and toxic actions of DCA in the intact organism are a result, in part at least, of its inhibitory effect upon pituitary adrenocorticotrophic activity. The steroid induces insulin hypersensitivity(11). Furthermore, the elevation of electroshock threshold and hypernatremia, normally produced by DCA, are prevented by the simultaneous administration of ACTH(12).

In addition to a pituitary inhibitory effect, it is quite probable that DCA induces certain changes characteristic of adrenocortical insufficiency by competing with cortisone-like compounds for strategic loci in peripheral effector cells. This aspect of the problem is undergoing study in our laboratory at the present time.

The data of the present report have an important bearing on the mechanism of action of DCA in inducing pathological changes in experimental animals similar to those observed in the collagen diseases in man. It has been suggested by Cheng and Sayers(11) and by Woodbury *et al.*(12) that this toxic action of DCA is a result, in part, of a steroid hormone imbalance, namely, an excess of DCA (exogenous synthetic steroid) and a deficiency of the secretion of the adrenal cortex. The recent demonstration of Woodbury *et al.*(13) that ACTH given simultaneously with DCA can inhibit the development of pathological changes, which are normally induced by the synthetic steroid when administered alone, lends strong support to this interpretation.

Summary. DCA pellets increase the concentration of ACTH in the adenohypophysis of the intact rat. The steroid inhibits the marked depletion of pituitary stores of ACTH which normally occurs 24 hours after adrenalectomy.

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Cortisone Acetate and Terramycin in Polyarthritis of Rats.*† (18013)

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The L_4 strain of pleuropneumonia-like organism will produce in rats an arthritis which has been used as a method of evaluating various therapeutic agents(1-5). Pleuropneumonia-like organisms are evidently species specific as far as pathogenicity is concerned so that human strains cannot be used in animals. Dienes and his associates

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