largely prevented by the feeding of materials known to be high in vit. B_{12} activity(21) to the mothers or by injection of vit. B_{12} into the young(20). The results presented in Table I could therefore be interpreted as indicating that in the presence of PST the intestinal flora was altered so that microbiological synthesis of vit. B_{12} provided the maternal organism with a supply of this compound sufficient to prevent the appearance of the external symptoms of acute uremia in the newborn rats. Since absolutely specific methods for the determination of vit. B_{12} are not available at present a decision as to whether microbiological synthesis of vit. B_{12} was in fact responsible for the beneficial effects of PST reported here must be held in abeyance. The components of a ration may have a marked effect on gastrointestinal synthesis of vitamins(22). Thus, it has been reported(23) that the feces of rats maintained on an all plant ration were persistently negative for vit. B_{12} activity while those produced on a casein ration contained vit. B_{12} activity. In this connection it might be pointed out that the ceca of the rats consuming PST were in this experiment always found to be greatly enlarged compared to those of animals consuming the basal ration. This was observed even in 21-day-old weanling rats which had consumed the ration for only a few days. Baxter(17) has recorded a similar observation.

Summary. The feeding of 2% of phthalylsulfathiazole to rats maintained on a purified ration containing a commercial soybean protein and DL-methionine as the only source of amino acids produced no depression of growth; it supported better reproductive performance and no cases of acute uremia of the newborn were observed among about 200 rats. On the basal ration, acute uremia of the newborn occurred in 35% of the litters born.

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Effect of ACTH on Glycogenesis and Glycolysis in Hypophysectomized Rats. (18098)

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Abelin(1) showed that after feeding starved rats a high carbohydrate diet, an increase in liver glycogen accompanied a 25% reduction in adrenal cholesterol. He suggested that the rise in blood sugar may have stimulated the pituitary to secrete ACTH, which caused a release of adrenocortical hormone, and this, in turn, stimulated liver glycogenesis. Ingle(2) was able to demonstrate adrenal hypertrophy in rats which died as a result of an excessive carbohydrate diet. Similar results were reported by other workers (3,4). Hypophysectomy caused a decrease in the muscle glycogen of rats, but the glycogen level was restored to normal upon the administration of anterior pituitary extract; however, pure ACTH was without effect(5).

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Liver glycogen dropped slightly in hypophysectomized animals but was not increased by administering anterior pituitary extract or ACTH. Previously Russell and Craig(6) demonstrated that adrenocortical extracts increased liver glycogen.

The present study was undertaken in order to investigate the glycogenesis and glycolysis in the hypophysectomized rat and, then, to determine the effect of ACTH. An analysis of phosphorylated intermediates as well as acid-insoluble phosphorus components was conducted in the hope of gaining some insight into the mechanisms involved in energy turnover.

Methods. Sprague-Dawley rats were hypophysectomized when 40 days old and used 12 days later. The diet was Purina Dog Chow. Four hypophysectomized rats received 10 mg of a long-acting preparation of ACTH 24 hours before sacrifice. All animals received 2 cc of a saturated solution of glucose by stomach tube 3 hours before sacrifice.

Analyses for the phosphorylated intermediates were conducted according to the method outlined by LePage and Umbreit(7). The acid-insoluble phosphorus was determined by the method of Schneider(8). Anerobic glycolysis was measured by the method of LePage(9).

Results. Hypophysectomized rats showed a considerably decreased ability to synthesize liver glycogen from glucose, the inhibition being greater than 50% (Table I). In hypophysectomized rats treated with ACTH the liver glycogen content was essentially normal. Likewise, the glycogen level of the brain which was reduced in hypophysectomized rats was restored to normal by the administration of ACTH (Table II). The ATP content of the liver of normal and hypophysectomized animals treated with ACTH was considerably less than in the untreated hypophysectomized rats; whereas the reverse was true for ADP. In the former the ratio of ATP to ADP was 1 to 7, while in the latter the ratio was 10 to 1. Adenylic acid was about 50% lower in hypophysectomized animals than in the normals or ACTH-treated. In the brain the ATP con-

TABLE I. Phosphorylated Intermediates of the Liver of Normal and Hypophysectomized Rats With and Without ACTH.

The values are in μg/wet wt and represent an average of 2 rats, the determinations agreeing within 20%.

	Normal	Hypophysectomized	Hypophysectomized ACTH
Glycogen	48,000	18,000	44,000
Glucose-1-phosphate	120	160	140
Glucose-6-phosphate	1,380	1,330	2,000
Fructose-6-phosphate	170	180	150
Fructose-1,6-phosphate	25	28	138
Triose phosphorus	77	22	20
ATP	120	900	125
ADP	670	84	690
Adenylic acid	700	440	880
Total phosphorus	920	1,200	670
Inorganic phosphorus	240	470	265

TABLE II. Phosphorylated Intermediates of Brain of Normal and Hypophysectomized Rats With and Without ACTH.

Values are in μg/g wet wt and represent an average of 2 rats, the determinations agreeing within 20%.

	Normal	Hypophysectomized	Hypophysectomized ACTH
Glycogen	500	320	690
Glucose-1-phosphate	200	210	42
Glucose-6-phosphate	1,400	1,600	1,500
Fructose-6-phosphate	38	40	164
Fructose-1,6-phosphate	40	42	240
Triose phosphorus	10	10	17
ATP	600	450	590
ADP	10	110	236
Adenylic acid	580	480	465
Phosphocreatine	610	520	800
Total phosphorus	910	915	960
Inorganic phosphorus	320	470	425

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TABLE III. Acid-insoluble Phosphorus Components of Liver and Brain on Normal and Hypophysectomized Rats With and Without ACTH.

Values are in $\mu g/g$ wet wt and represent an average of 2 rats, the determinations agreeing within 30%.

	Liver			Brain		
	Normal	Hypophysec.	Hypophysec. ACTH	Normal	Hypophysec.	Hypophysec. ACTH
Phospholipid P	1,170	1,500	1,250	1,700	1,850	1,800
Nucleic acid P	450	450	390	300	270	250
Phosphoprotein P	530	300	460	550	260	530

tent of the hypophysectomized rats was somewhat lower than in the other animals. The fructose-1,6-phosphate was significantly greater in the liver and brain of hypophysectomized rats given ACTH. Total and inorganic phosphorus were higher in the hypophysectomized rats than in the normals or hypophysectomized rats receiving ACTH.

The phospholipid and nucleic acid phosphorus content of hypophysectomized rats showed no significant variation from the normals (Table III). Phosphoprotein phosphorus, however, decreased up to 50% in the hypophysectomized rats as compared to the normals and hypophysectomized rats treated with ACTH.

A typical experiment of the anaerobic glycolysis of the brain of normal and hypophysectomized rats with and without ATH is represented in Fig. 1. It can be seen that the rate of glycolysis of hypophysectomized rats is over 20% less than the normal or hypophysectomized rats given ACTH.

Discussion. The relative inability of hypophysectomized rats to synthesize liver glycogen is in accord with the results of Abelin(1). That the adrenal cortex is specifically involved here is evident from the fact that ACTH restores the glycogenetic capacity of the liver of hypophysectomized rats to normal. It is interesting in this respect that liver ATP is converted to ADP during glycogen synthesis, and that when glycogen synthesis is impaired, as in the hypophysectomized animals, the ratio of ATP to ADP is high. Glycogenesis must be a form of stress in the liver enlisting high energy phosphate bonds in order to continue. It is not clear from these results whether or not the high ratio of ATP to ADP is antecedent to the reduced glycogenetic capacity of the liver of hypophysectomized rats; however, the decreased rate of glycolysis in the brain of hypophysectomized rats is suggestive of some interference with energy turnover. The high inorganic phosphorus and low adenylic acid content of untreated hypophysectomized animals are indicative of an interference with phosphorus utilization. Preliminary studies have revealed no inhibition of pyruvate, succinate, and glutamate oxidation in tissues of untreated hypophysectomized rats.

Summary. An analysis of the phosphorylated intermediates of the liver and brain of hypophysectomized rats with and without ACTH was made. The decreased ability of hypophysectomized rats to synthesize glycogen from glucose was restored to normal by ACTH. Phosphoprotein phosphorus levels also returned to normal with ACTH. The

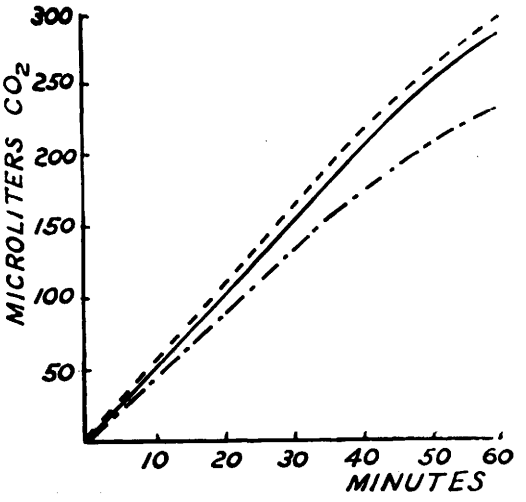

FIG. 1.
Anaerobic glycolysis of brain of hypophysectomized rat given ACTH (– – –), of normal (——), and of untreated hypophysectomized rat (–•–•).

anaerobic glycolysis of the brain of hypophysectomized rats was reduced 20% below that of normal rats but this was restored to normal by the administration of ACTH.

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Adrenal Medullary Hormones in Water Diuresis.* (18099)

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Recent studies have established the existence of nor-epinephrine both in the adrenal medulla and in natural U. S. P. epinephrine and it is now generally believed that this substance is normally secreted by the adrenal glands(1-4). Many workers(*e.g.* 5-8) have found nor-epinephrine to have biological and pharmacological properties different from those of epinephrine and it has become of general interest and importance to determine which of the many actions ascribed to adrenal medullary secretions are due to epinephrine, nor-epinephrine or both. The diuretic response to water offers an easy and convenient testing procedure for the investigation of one possible difference in the action of these compounds since it has been established that large doses of adrenalin (nor-epinephrine and epinephrine) greatly stimulate the flow of urine in water diuresis(9). The purpose of the experiments reported here, therefore, was to determine whether one or both of the adrenal medullary hormones was responsible for the augmentation of urine flow such as that obtained by Gaunt *et al.*(9). The results indicate that nor-epinephrine rather than epinephrine was the causative agent involved.

Methods. Four types of adrenal medullary hormone preparations† were employed. The adrenalin (Parke-Davis) was predominately l-epinephrine but contained small amounts of nor-epinephrine. The pure compounds used were dl-nor-epinephrine hydrochloride, l-epinephrine bitartrate and l-nor-epinephrine bitartrate monohydrate. Each of these was suspended in peanut oil and administered so that the volume of oil was kept constant (0.05 ml per 100 g body weight) although the amount of hormone varied from 3 to 30 μg per 100 g body weight. Female rats, weighing approximately 200 g were fasted 18 hours but allowed water *ad libitum*. They were placed in individual metabolism cages, injected subcutaneously with hormone, and given orally one dose of water (3 ml per 100 sq cm of body surface)(11) warmed to body temperature. Urine output was recorded at half-hour intervals for a period of 3 hours.

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