Reduction of Urinary Sodium and Potassium of Diabetic Rats Produced by Hypophyseal Growth Hormone.* (18835)

CONSTANTINE M. GLAFKIDES AND LESLIE L. BENNETT.

From Division of Physiology and Institute of Experimental Biology, University of California, Berkeley.

Growth hormone is known to produce hyperglycemia and glycosuria in normal cats (1) and dogs(2), and to intensify the glycosuria of diabetic rats(3). Growth hormone also reduces urinary nitrogen excretion and promotes gain in weight and deposition of body protein in normal rats(4). Reduction of urinary nitrogen excretion in diabetic rats occurs even when the glycosuria is increased (3). One would anticipate that the deposition of increased amounts of body protein and gain in weight under the influence of growth hormone would be associated with a positive sodium and potassium balance. The negative sodium and potassium balance that is associated with severe, uncontrolled diabetes is well recognized. Insofar as the authors are aware, no observations regarding the effects of growth hormone upon sodium and potassium balance have been reported. It is the purpose of this paper to report the fact that the administration of growth hormone to diabetic rats reduces the urinary excretion of both sodium and potassium even in the face of an increase in glycosuria.

Methods. The diabetic rats used were male animals of the Long-Evans strain that were partially depancreatized when they were from 25 to 32 days old. They were used in the experiments when they were from 120 to 150 days of age, and had an established glycosuria. Each rat was maintained in an individual metabolism cage and 24-hour urine specimens were collected daily for determination of glucose, nitrogen, sodium, and potassium. Glucose was determined as total reducing substances by the Somogyi procedure, nitrogen by the micro-Kjeldahl method, and sodium and potassium by the Perkin-Elmer flame photometer, model 52A, with a lithium internal standard. Four groups of animals with 4 or 5 animals in each group were studied. Two of the groups were maintained on the regular stock diet and two groups received the same diet with an additional 600 mg of potassium chloride added to the diet daily. In most instances the food intake was constant throughout the period of observation, the method previously described(3) being used to control the food intake. In two of the groups during growth hormone injection, some of the animals reduced their food intake slightly, but the maximum reduction in food consumption was 700 mg per rat per day for the treatment period and the total food intake remained in excess of 20 g per rat per day. The growth hormone was administered twice daily intraperitoneally and was prepared by Dr. C. H. Li, according to the previously published method(5).†

Results and discussion. The data for urinary sodium are summarized in Table I. It will be seen that in all groups growth hormone produced a reduction of urinary sodium and that the greatest reduction occurred in Groups I and IV where there was minimal or no reduction respectively in food consumption during the treatment period. The data for the glycosuria are not shown in the table, but the average glycosuria increased in all groups except Group I. One rat in Group I had a considerable reduction in glycosuria while

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TABLE I. Effect of Growth Hormone on Urinary Sodium (Sodium Expressed as mg/Rat/Day).

Group		Control period mg of Na	3 mg GH per day mg of Na	Change	p§	6 mg GH per day mg of Na	Change	p
I	Stock diet, 5 rats (20 g food daily)	52 ± 2‖ (10)*	44† ± 4 (3)	—8	.05			
II	Stock diet, 4 rats (20 g food daily)	69 ± 2 (10)	66 ± 1 (3)	—3	.50	63 ± 4 (2)	—6	.20
III	High KCl diet, 5 rats (21 g food daily)	57 ± 1 (10)	51‡ ± 4 (3)	—6	.10			
IV	High KCl diet, 5 rats (21 g food daily)	76 ± 1 (10)	65 ± 3 (3)	—11	<.01	64 ± 3 (2)	—12	<.01

* Fig. in parentheses indicates No. of days of observation.
† Food consumption reduced by 0.4 g per day during this period.
‡ Food consumption reduced by 0.7 g per day during this period.
§ From Fisher table of t.

‖ Std dev. $\sigma = \sqrt{\frac{\Sigma d^2}{n(n-1)}}$

TABLE II. Effect of Growth Hormone on Urinary Potassium (Potassium Expressed in mg/Rat/Day*)

Group		Control period mg of K	3 mg GH per day mg of K	Change	p	6 mg GH per day mg of K	Change	p
I	Stock diet, 5 rats	58 ± 2 (10)	42 ± 3 (3)	—16	<.01			
II	Stock diet, 4 rats	82 ± 1 (10)	68 ± 5 (3)	—14	<.01	67 ± 5 (2)	—15	<.01
III	High KCl diet, 5 rats	382 ± 4 (10)	353 ± 12 (3)	—29	<.01			
IV	High KCl diet, 5 rats	386 ± 2 (10)	366 ± 5 (3)	—20	<.01	368 ± 9 (2)	—18	<.01

* This table arranged as in Table I, and based on same animals; hence food consumption, etc. are the same as in Table I.

receiving growth hormone and, although the other rats had an increase, there was no enhancement of glycosuria for the group as a whole. The average daily increase in glycosuria was 550, 740, and 830 mg per rat per day in Groups II, III, and IV respectively during the administration of 3 mg of growth hormone per rat per day. In Groups II and IV during the administration of 6 mg of growth hormone per day, the average glycosuria increased 510 and 180 mg per day respectively. When the change in glycosuria was compared statistically on the basis of individual animals, the increase was significant in 7 out of 28 cases.

The data for urinary potassium are summarized in Table II. It will be seen that all groups showed a reduction in potassium excretion which in all cases was statistically significant, and it is to be remembered that there was no change in the food consumption during the treatment period in four of these six sets of observations. In all groups there was a marked reduction in urinary nitrogen varying from 73 to 152 mg per rat per day. When compared statistically on an individual animal basis, this was significant in 21 out of 28 cases. It seems very reasonable that the nitrogen and potassium retaining effects of growth hormone are related, in that both potassium and nitrogen are integral intracellular components of protoplasm.

The increase in glycosuria produced by withdrawal of insulin therapy in a diabetic animal is associated with an increase in the urinary excretion of nitrogen, sodium, and potassium. It is remarkable that the increase in glycosuria produced by growth hormone is associated with a reduction in the urinary excretion of nitrogen, sodium, and potassium. It seems unlikely that any contaminants of the growth hormone preparation used could have been responsible for these effects upon electrolyte metabolism. In our hands, ad-

renocorticotropic hormone causes sodium and potassium loss when it intensifies the glycosuria of diabetic rats, an effect opposite to that produced by growth hormone in these experiments when it enhanced the glycosuria. Posterior pituitary antidiuretic hormone might be expected to have effects upon electrolyte balance, but one would anticipate that they would be in the direction of sodium loss. The growth hormone preparation used in these experiments was assayed for antidiuretic activity by the Ham and Landis(6) procedure and found to be free of any antidiuretic activity whatsoever at a single dose level of 0.5 mg, a daily dose found to produce changes in sodium and potassium balance of normal animals (unpublished observations).

Conclusion. The administration of growth hormone to partially depancreatized diabetic rats produces a reduction in urinary nitrogen, sodium and potassium excretion, although there may be a concomitant increase in glycosuria.

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Circulating Antibodies in Vitamin-Deficiency States. Pteroylglutamic Acid, Niacin-Tryptophan, Vitamins B_{12}, A, and D Deficiencies.* (18836)

PETER P. LUDOVICI,† AND A. E. AXELROD.‡ (Introduced by Ralph R. Mellon.)

From the Institute of Pathology, Western Pennsylvania Hospital, and Department of Chemistry, University of Pittsburgh.

Previous work in this laboratory has demonstrated a severe impairment of antibody response in pyridoxine and pantothenic acid-deficient rats(1). Riboflavin(1), biotin and thiamine deficiencies(2) had considerably less effect. Later experiments proved conclusively that the absence of pantothenic acid *per se* and not the concomitant inanition was responsible for antibody impairment(3). Recently we have presented evidence which suggests that pantothenic acid and pyridoxine function directly in antibody synthesis(4).

In the present paper, the relationship between vitamins and antibodies has been extended further to include the effect of deficiencies of pteroylglutamic acid, niacin-tryptophan, vit. B_{12}, vit. A, and vit. D upon the antibody response of the rat to the antigenic stimulus of human erythrocytes.

Methods. Ninety-six male weanling albino rats of the Sprague-Dawley strain were distributed into 5 groups as indicated in Table I. The animals were housed individually in wide-meshed, screen-bottom cages and weighed weekly. The composition of the basal diets fed to each group is presented in Table II. These basal diets were fed *ad lib.* throughout the experimental period. Immunization was instituted in the vit. A, B_{12}, and D groups after 4 weeks, in the niacin-tryptophan group after 5 weeks, and in the pteroylglutamic acid group after 9 weeks on experiment. A 10% suspension of washed Group O, Rh positive human erythrocytes in physiological saline was injected intraperitoneally as antigen. An initial dosage of 0.5 ml of the erythrocyte suspension was followed by two 1 ml injections on alternate days. Five days after the final injection, the rats were bled from the

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† Present address: Department of Biological Science, University of Pittsburgh.

‡ Present address: Institute of Pathology, Western Reserve University, Cleveland, O.

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