Diabetogenic Effects of Growth Hormone

The Role of the Adrenals in Nitrogen Loss

O. H. Gaebler, M.D., Ph.D., Rachel Glovinsky, M.S., Trieste Vitti, M.S., and Thomas G. Maskaleris, Detroit

Houssay and Penhos[1] cite numerous studies in which anterior pituitary preparations produced diabetogenic effects in the absence of the adrenals. In hypophysectomized, adrenalectomized, partially depancreatized dogs, they observed large increases in blood sugar after somatotropin or prolactin administration, and small increases after corticotropin. Hydrocortisone also produced diabetic hyperglycemia in their animals.

Early studies by Gaebler and Robinson[2] support the contention of Houssay and Penhos that adrenalectomy does not abolish diabetogenic effects of growth hormone. Table 6 of the paper cited[2] includes two experiments in which crude growth hormone was given to a dog with pancreas, adrenals, thyroid, and parathyroids removed. Lipemia, hyperglycemia, and increase in glucosuria all occurred in the first of these experiments, while in the second there was some increase in blood sugar. None the less, there was no loss of nitrogen, such as growth hormone elicited[3] in depancreatized dogs with adrenals intact; in fact, some storage of nitrogen was induced in one of the two depancreatized-adrenalectomized animals, and both of them tolerated the growth hormone preparation much better than depancreatized ones. Thus the alleviating effects of adrenalectomy, so well established by Long and Lukens in cats,[4] and by Long in rats,[5] were again confirmed in dogs.

In the present study, we have investigated the nitrogen storing action and diabetogenic effect of a growth hormone preparation that is virtually free of corticotropin, and contains far less thyrotropic hormone than previous preparations. We have also attempted to reproduce the diabetogenic effects of growth hormone with corticotropin and hydrocortisone under our experimental conditions, and have carried out experiments to determine whether prolonged pretreatment of depancreatized dogs with hydrocortisone mitigates effects of growth hormone in a manner analogous to that observed by de Bodo and associates in hypophysectomized dogs.[6]

From the Edsel B. Ford Institute for Medical Research, Henry Ford Hospital, Detroit, Michigan.

MATERIALS AND METHODS

Normal and depancreatized adult bitches were employed. Dog 63, also used in a previous study,[7] was depancreatized Jan. 19, 1954, and is in excellent condition during the fifth postoperative year. Its daily insulin requirement was 36 units during most of this long history, occasionally dropping to 32 or even 28 units for periods of varying duration. Dog 73, depancreatized May 19, 1958, requires only half as much insulin as dog 63. The food intake of the two animals, given in footnotes under the tables, is similar.

The basal diet had the following composition, in percentages: casein, 36.2; cracker meal, 36.2; corn oil, 19.6; yeast, 4.0; and salt[8] mixture, 4.0. All animals received two drops of haliver oil daily, and 200 mg. of a mixture of vitamins and calcium phosphate, containing the following daily allowances of vitamins in milligrams: thiamine hydrochloride, 0.75; riboflavin, 1.5; nicotinic acid, 7.5; pyridoxine hydrochloride, 0.6; calcium pantothenate, 3.0.

Normal dogs were fed once daily. Depancreatized ones received half of the daily ration at 9 a.m., and the remainder at 3 p.m. In addition to the above supplement, they were fed 500 mg. of choline with the morning meal, and 7.5 gm. of pancreatin with each meal. As a rule, one third of the total daily dose of insulin consisted of protamine insulin, injected at 3 p.m., and the remainder of plain insulin, injected in equal amounts after the morning and afternoon meals.

Urine periods of all animals were terminated by catheterizing and washing the bladder, just before the morning feeding.

The corticotropin preparation, Acthar, Gel (Armour), was injected subcutaneously, and hydrocortisone acetate (Sharpe & Dohme) intramuscularly. One half of the total daily dose indicated in the tables was given at 9 a.m., and the remainder at 3 p.m. The bovine growth hormone preparation, Endocrine Study Section Lot R-50109, List No. 916, was dissolved in water and injected subcutaneously.

RESULTS

Data on nitrogen storage in five experiments on three normal bitches are presented in table 1. To calculate storage, we determine the average daily urine nitrogen output during a four- to eight-day control period, and integrate subsequent daily balances until the urine nitrogen returns to or exceeds its control value, whether this point precedes, coincides with, or follows termination of growth hormone injections. The amounts stored were as usual quite variable (table 1), but compare favorably with those reported previously from this laboratory.[7,9] In one of our studies (table 1 of reference 7), 5 mg. doses of growth hormone injected daily for six days induced storage of only 3.0 to 5.45 gm. of nitrogen. In an earlier series,[9] the amounts of nitrogen were larger than in the present one, since the period of injection was ten days instead of four. It is of interest, however, that the maximal fall in urine nitrogen in table 1 of the present study is similar to that which we have calculated from original data of the former one.[9] For the previous experiments (table 1 of reference 9) in which nitrogen storage was 11.76, 11.2, 18.78, 16.7, and 9.6 gm., the maximal reductions of urine nitrogen below the control values were respectively 36, 28, 34, 31, and 34 per cent.

TABLE 1

Nitrogen storage experiments with bovine growth hormone in normal dogs

Date 1957	Dog no.	Growth hormone mg. x days	Nitrogen stored* gm.	Reduction of N output† per cent	Increase in weight† kg.
1-28	57	5 x 4	6.09	44.3	0.69
10-21	57	5 x 4	5.95	27.4	0.36
1-14	64	5 x 4	9.74	32.3	0.66
11-23	64	5 x 4	13.95	35.9	0.79
11-13	70	2.5 x 6	9.79	31.1	0.63

* Total amount per period.
† Maximal values per day during period.

When all our data are considered, the effect of growth hormone on nitrogen storage appears to vary less in intensity than in duration. Variability of duration accounts for many large differences in total storage, and suggests that the speed with which the homeostatic mechanism comes into play in our adult animals may be quite as important as the dose or potency of the growth hormone, in determining the total amount of nitrogen stored. Whatever the basis for variability may be, it is evident that one can not determine whether the present preparation is more potent than former ones with respect to induction of nitrogen storage. Our data do establish that storage was induced regularly, to at least the usual extent, and that the findings in depancreatized dogs, which we will now consider, are not due to peculiarities of the present lot of growth hormone. Discrepancies between nitrogen storage and other criteria of activity have been observed with some preparations.[10]

In the first experiment shown in table 2, a depancreatized dog received 7.5 mg. of growth hormone on two successive days. Since we were aware that the diabetogenic effect appears during the second twenty-four-hour period following an injection, the dose of insulin was increased from 36 to 48 units on the first day of growth hormone therapy, and to 72 units on the second day. Nitrogen storage occurred, but failed to continue, although the doses of insulin on the following three days were still large. This experiment is an almost

TABLE 2

Experiments with growth hormone, with or without additional insulin, in depancreatized dog 63

	Date 1956—57	Insulin units/day	Growth hormone mg./day	Weight kg.	Urine sugar gm./day	Urine nitrogen gm./day
I.	12/26—12/31	36	0	17.02	< 2.3	12.61 ± 0.73
	1/1	48	7.5	17.27	< 2.0	10.66
	1/2	72	7.5	17.44	25.3	9.16
	1/3	60	0	17.33	40.1	12.00
	1/4	56	0	17.16	22.2	12.67
	1/5	48	0	17.21	5.8	12.30
	1/6—1/13	36	0	17.21	< 2.3	12.98 ± 0.85
II.	4/18—4/22	36	0	19.59	< 1.6	12.58 ± 0.37
	4/23	36	7.5	19.82	19.7	12.64
	4/24	36	7.5	18.91	75.2	14.51
	4/25	36	0	18.74	100.0	13.51

The daily nitrogen intake was 13.98 gm. during both experiments.
The daily ration consisted of 240 gm. of basal ration plus 50 gm. of sucrose during both experiments.
Numbers preceded by ± signs are standard deviations in this and subsequent tables.

exact repetition, with the present growth hormone preparation, of one carried out in the same animal with another purified preparation (table 4, experiment 1, of reference 7).

During the interval between the two experiments in table 2, the diet and insulin dosage were constant, and the animal gained more than 2 kg. in weight. In the second experiment, when 7.5 mg. of growth hormone was given, with the insulin dose constant, nitrogen loss occurred, and glucose output rose to 100 gm. during the second twenty-four-hour period following the second injection. The effect of the present preparation on insulin requirement in the first experiment, and on glucosuria in the second one, clearly indicate that it, like previous preparations, is extremely diabetogenic; in fact, it is dangerous to inject 7.5 mg. daily for more than two days. Evidently the extremely low content of corticotropin and thyrotropin has not diminished the diabetogenic effect. Another point of interest, which we have confirmed many times, is that failure to induce storage of nitrogen with growth hormone in depancreatized dogs, receiving a constant dose of insulin, occurs in animals which can gain weight and store nitrogen spontaneously under the prevailing conditions of food intake and insulin dosage.

Attempts to duplicate the diabetogenic effect of growth hormone with 20 units of corticotropin daily are presented in table 3. In the first experiment, some loss of weight and of nitrogen occurred, but glucosuria was negligible. To bring the animal nearer the threshold of glucosuria, the basal ration was increased, and supplemented with sucrose as indicated in the footnote, while the insulin dose was left unchanged. Under these conditions, corticotropin induced more nitrogen loss than before, as well as glucosuria up to 36 gm. per day. Although 20 units of corticotropin is a large daily dose, the resulting glucosuria did not compare with that elicited by growth hormone in the second experiment of table 2, particularly if one considers that insulin dosage was 36 units in the experiment with growth hormone as against 32 in the one with corticotropin.

Experiments in which two depancreatized dogs received 15 mg. of hydrocortisone daily are presented in table 4. Neither nitrogen loss nor glucosuria occurred in dog 63. In dog 73, some nitrogen loss took place but significant glucosuria was absent. In two normal dogs which received 15 mg. of hydrocortisone daily for six and eight days, the average increase in urine nitrogen during the period of treatment was 1.55 and 1.49 gm. respectively, while the maximal increase was 2.4 gm. in both animals. The relative susceptibility of normal and depancreatized dogs to induction of nitrogen loss is being investigated further. The data in table 4 indicate quite clearly that no glucosuria comparable with that induced by growth hormone follows daily injection of 15 mg. of hydrocortisone. Results for 20 units of corticotropin daily were the same in dog 73 (table 4) as in dog 63 (table 3). When the dose of hydrocortisone administered to dog 63 was increased to 30 mg. daily (table 5), a decided increase in urine nitrogen occurred, but glucosuria was still very moderate.

For reasons indicated in the introduction, we also investigated the effect of prolonged administration of hydrocortisone upon the diabetogenicity of growth hormone. In the first experiment, 15 mg. of hydrocortisone were administered to dog 63 daily for eleven days. Data for the first nine days appear in line 2 of table 4. Not shown in the table is an experiment on the tenth day, when 5 mg. of growth hormone, Armour M10810, were injected. Urine glucose increased to 48 gm. on that day, and to 108 gm. on the day following. Since it was very evident that no reduction of diabetogenicity had occurred, we performed the more extensive experiment presented in table 6, using growth hormone supplied by the Endocrine Study Section. At the start of the experiment, the minimal daily insulin requirement of the animal was somewhat more than 28 units, as indicated by average daily excretion of 10.2 gm. of glucose (table

TABLE 3

Experiments with corticotropin in depancreatized dog 63

	Date 1956	Corticotropin units/day	Weight kg.	Urine sugar gm./day	Urine nitrogen gm./day
I.	3/29—4/3	0	15.49	< 2.0	12.00±0.49
	4/4	20	15.40	< 2.0	12.76
	4/5	20	15.40	4.2	13.81
	4/6	20	15.34	4.0	13.24
	4/7	20	15.22	4.8	11.92
	4/8—4/13	0	15.70	< 1.0	10.37±0.53
II.	8/24—9/1	0	16.37	< 3.7	11.46±0.37
	9/2	20	16.08	< 1.0	13.39
	9/3	20	15.80	30.0	17.34
	9/4	20	15.06	36.2	17.22
	9/5	20	15.68	26.0	17.24
	9/6—9/9	0	15.74	15.8	12.05±0.70
	9/10—9/16	0	16.03	< 7.2	11.57±1.08

The nitrogen intake was 12.81 gm. daily during the first (I) corticotropin experiment and 13.98 during the second (II). During the first experiment, the daily ration consisted of 220 gm. of basal diet; during the second of 240 gm. of basal diet plus 50 gm. of sucrose. Insulin dosage was 32 units throughout.

TABLE 4

Experiments with hydrocortisone and corticotropin in depancreatized dogs

Dog	Days	Hydrocortisone mg./day	Corticotropin units/day	Weight kg.	Urine sugar gm./day	Urine nitrogen gm./day
63	1-6	0	0	16.0	< 2.7	14.47 ± 0.80
	7-15	15	0	15.9	< 6.1	14.18 ± 0.97
73	1-6	0	0	15.4	< 1.0	10.79 ± 0.33
	7-12	15	0	15.0	6.8	12.80 ± 1.59
	13-20	0	0	15.7	< 1.0	10.60 ± 0.66
	21-26	0	20	15.5	8.3	14.97 ± 0.99
	27-32	0	0	16.1	< 1.0	10.86 ± 0.63

Daily insulin doses, food intake, and nitrogen intake were 32 units, 250 gm., and 14.56 gm. respectively for dog 63; and 16 units, 220 gm., 12.81 gm., for dog 73.

TABLE 5

Experiments with hydrocortisone in depancreatized dog 63

Date 1956	Hydrocortisone mg./day	Weight kg.	Urine sugar gm./day	Urine nitrogen gm./day
3/15—20	0	15.90	< 1.3	11.48 ± 0.57
3/21	30	15.86	< 1.0	12.60
3/22	30	15.86	< 1.0	13.68
3/23	30	15.51	4.0	13.52
3/24	30	15.56	9.1	15.60
3/25	30	15.56	12.7	13.40
3/26	30	15.56	9.5	12.91
3/27	0	15.40	17.8	14.35
3/28	0	15.34	8.6	12.78
3/29—4/3	0	15.49	< 2.0	12.00 ± 0.49

The daily insulin dosage, food intake, and nitrogen intake were 32 units, 220 gm., and 12.81 gm. respectively.

TABLE 6

Induction of polyuria without glucosuria in a prolonged experiment with hydrocortisone

Days	Insulin units/day	Hydrocortisone mg./day	Growth hormone mg./day	Urine sugar gm./day	Water intake ml./day
1-4	28	0	0	10.2	1,247
5-15	28	15	0	25.3	1,898
16	32	15	0	20.0	1,990
17-24	36	15	0	3.3	1,718
25-37	36	30	0	<6.0	2,684*
38	36	30	7.5	17.5	3,400
39	36	30	7.5	53.8	3,900

Dog 63 was used in this experiment. The diet consisted of 240 gm. of basal ration.
* Highest value 3,450 ml.

6). This average value rose to 25.3 gm. when 15 mg. of hydrocortisone was injected daily for eleven days. The glucosuria was overcome by increasing the dose of insulin first to 32, then to 36 units; it did not return when the daily dose of hydrocortisone was increased to 30 mg. Polydipsia and polyuria, however, became extreme. On the thirty-fourth and thirty-fifth days of hydrocortisone injections, 7.5 mg. doses of growth hormone induced the usual severe and dangerous condition. During the night of the last day for which data are given in table 6, the animal vomited. On the following day, it refused half of its food, but still excreted 46.3 gm. of glucose, although the usual 36 units of insulin were given, and both growth hormone and hydrocortisone were discontinued.

A point of considerable interest in the experiment shown in table 6 was the occurrence, as the result of hydrocortisone therapy alone, of a condition which we have encountered in a number of experiments with growth hormone in depancreatized dogs. The animals become wildly excited, and, if water is available ad libitum, alternately drink and vomit, and are presently in very critical condition. This disturbance occurred during the thirteen-day period preceding administration of growth hormone. Although water intake was restricted sufficiently to prevent vomiting, the average intake was 2,684 ml. per day, and the highest value was 3,450 ml. Glucose output was negligible—less than 6 gm. per day.

DISCUSSION

Present findings support the idea that nitrogen loss caused by growth hormone in depancreatized dogs may be mediated by the adrenals. In our early experiments, adrenalectomy either abolished the loss, or permitted some nitrogen storage to occur;[2] in the present ones, nitrogen loss was readily induced with corticotropin in depancreatized dogs receiving constant amounts of food and insulin (tables 3 and 4). Since 7.5 mg. doses of the present growth hormone preparation contain only 0.45 milliunit (U.S.P.) of corticotropin, this exogenous source can not be implicated; it is, however, quite conceivable that injection of growth hormone could lead to discharge of endogenous corticotropin in depancreatized dogs that are not hypophysectomized. The very limited

glucosuria in our experiments with corticotropin and hydrocortisone presents no problem, if one accepts the well-supported view[11,12] that the effect of these hormones is one of accelerating gluconeogenesis. The amount of protein corresponding to the small increases in urine nitrogen would yield very little glucose.

The role of the adrenals in the induction of glucosuria by growth hormone in depancreatized dogs is more obscure. Adrenalectomy diminishes[2] but does not abolish this diabetogenic effect.[1,2] The amount of glucose excreted is quite out of proportion to the nitrogen loss. For example, in the experiment shown in table 2, excretion of 100 gm. of glucose was accompanied by a rise of about 1 gm. in urine nitrogen. Since no comparable glucosuria was induced by injected corticotropin or hydrocortisone, it would be illogical to ascribe this outpouring of glucose to a discharge of endogenous corticotropin brought about by injection of growth hormone. It appears more likely that the adrenals become involved as a result of the diabetogenic effect, and further complicate it. McArthur and co-workers[13,14] have presented evidence that insulin withdrawal brings about adrenal stimulation in some way. The doses of insulin required to correct the disturbance of carbohydrate metabolism that growth hormone causes in depancreatized dogs are so large that continuance of the usual maintenance dose may be analogous to withdrawal of insulin from a depancreatized dog not treated with growth hormone.

Scow[15] observed that whether the dose of insulin was 1 unit or 3 units per day, hypophysectomized depancreatized rats, tube-fed 7 gm. of food per day, responded to growth hormone equally well with respect to gains in weight and storage of nitrogen. A daily insulin dose of 0.3 unit was not enough to keep the animals alive and well. He believes that additional insulin is necessary for induction of maximal nitrogen storage in depancreatized cats,[16] or of any storage in depancreatized dogs[2,7] because growth hormone is more diabetogenic in these species than in rats. The role he ascribes to insulin is essentially one of controlling the exacerbation of diabetes. In the discussion section of a previous paper,[7] the present writer has stated his reasons for believing that counteraction of the unfavorable effect of growth hormone on carbohydrate utilization (a less inclusive term than exacerbation of diabetes) may not be the only role of insulin in this connection.

Spontaneous growth of an animal can be initiated or stopped by regulating the energy intake, if the diet is complete. Hormonal induction of nitrogen storage, on the other hand, is a process for which energy is drawn from sources other than surplus. Crude growth hormone preparations induced nitrogen storage in intact dogs despite very large calorigenic effects,[17] with food intake constant. Even a negative energy balance did not prevent this.[18] Growth hormone also diminished protein catabolism in fasting rats[19] and mice.[20] In depancreatized dogs, suitable combinations of purified growth hormone and insulin induce storage of nitrogen despite a rise in glucose output to 80 gm. per day; other combinations of dosages fail to induce nitrogen storage despite absence of glucosuria.[7] In all the above instances in which the matter has been investigated, increased oxidation of fat, and substitution of water for fat in tissue, removed any element of mystery so far as energetics are concerned. The findings may not, however, be entirely understood until more knowledge concerning the action of insulin becomes available from other types of experiments. Present suggestions concerning the role of this hormone in connection with nitrogen storage have the logical basis that protein synthesis requires energy. One can not overlook the fact that it also requires substrate. It seems possible that studies on the effect of insulin and other hormones on amino acid transfer[21] and retention[22] will provide important information in this connection.

SUMMARY

1. A highly purified bovine growth hormone preparation was tested in normal and depancreatized bitches. In normal animals, its nitrogen storing action was similar to that of other purified growth hormone preparations; in depancreatized animals, the diabetogenic effects were very severe, despite absence of noteworthy amounts of corticotropin or thyrotropic hormone.

2. Induction of nitrogen storage in the depancreatized dog with the present growth hormone preparation did not occur unless the dose of insulin was greatly increased, thus confirming our previous findings. The role of insulin in this connection is briefly discussed.

3. The nitrogen loss produced by growth hormone in depancreatized dogs, which was prevented by adrenalectomy in earlier experiments,[2] is readily reproduced with corticotropin or hydrocortisone; it may therefore be mediated by the adrenals.

4. Unfavorable effects of growth hormone on carbohydrate utilization in depancreatized dogs, which were reduced but not prevented by adrenalectomy in earlier experiments,[2] could not be reproduced with corticotropin or hydrocortisone.

5. Prolonged pretreatment of a depancreatized dog with hydrocortisone did not prevent occurrence of severe glucosuria when growth hormone was also given. It did cause wild excitement and polydipsia, without noteworthy glucosuria.

SUMMARIO IN INTERLINGUA

Effectos Diabetogene De Hormon De Crescentia—Le Rolo Del Corpores Adrenal In Le Perdita De Nitrogeno

1. Un purificatissime hormon de crescentia bovin esseva testate in normal e dispancreatisate canes-femina. In animales normal, le effecto del hormon super le immagasinage de nitrogeno esseva simile a illo de altere purificate preparatos de hormon de crescentia. In animales dispancreatisate, le effectos diabetogene esseva severissime, in despecto del absentia de quantitates notabile de corticotropina e de hormon thyrotropic.

2. In canes dispancreatisate, le presente preparato de hormon de crescentia non induceva immagasinage de nitrogeno, excepte quando le dose de insulina esseva grandemente augmentate. Isto confirmava nostre previe constatationes. Le rolo de insulina in iste connexion es brevemente discutite.

3. Le perdita de nitrogeno producite per hormon de crescentia in canes dispancreatisate—le qual esseva prevenite in previe experimentos per adrenalectomia—es facilemente reproducite per medio de corticotropina o hydrocortisona. Il es possibile, per consequente, que illo es mediate per le adrenales.

4. Adverse effectos de hormon de crescentia super le utilisation de hydrato de carbon in canes dispancreatisate —le quales esseva reducite ben que non prevenite in previe experimentos per adrenalectomia—non poteva esser reproducite per medio de corticotropina o hydrocortisona.

5. Le prolongate pretractamento de canes dispancreatisate con hydrocortisona non preveniva le occurrentia de glucosuria sever quando hormon de crescentia esseva etiam administrate. Illo causava un stato de grande excitation e polydipsia, sin grados notabile de glucosuria.

ACKNOWLEDGMENT

This study was supported by grant A-1362, National Institutes of Health. We are also indebted to the Michigan Chapter, Arthritis and Rheumatism Foundation, for supporting the predoctoral fellowship of one of the authors. The growth hormone used was a gift of the Endocrine Study Section, National Institutes of Health, and Armour and Company.

REFERENCES

[1] Houssay, B. A., and Penhos, J. C.: Diabetogenic action of pituitary hormones on adrenalectomized hypophysectomized dogs. Endocrinology *59*:637-41, 1956.

[2] Gaebler, O. H., and Robinson, A. R.: Effects of the pancreas and the adrenals upon production of nitrogen storage with pituitary preparations. Endocrinology *30*:627-34, 1942.

[3] Gaebler, O. H., and Galbraith, H. W.: Effects of anterior pituitary preparations in experimental pancreatic diabetes. Endocrinology *28*:171-78, 1941.

[4] Long, C. N. H., and Lukens, F. D. W.: The effects of adrenalectomy and hypophysectomy upon experimental diabetes in the cat. J. Exper. Med. *63*:465-90, 1936.

[5] Long, C. N. H.: The influence of the pituitary and adrenal glands upon pancreatic diabetes. The Harvey Lectures *32*:194-228, 1936-1937.

[6] de Bodo, R. C., Sinkoff, M. W., Kiang, S. P., and Den, H.: Prevention of growth hormone-induced diabetes in hypophysectomized dogs by adrenocortical steroids. Proc. Soc. Exper. Biol. & Med. *81*:425-30, 1952.

[7] Gaebler, O. H., Liu, C. H., and Zuchlewski, A.: Effects of small daily doses of growth hormone on nitrogen output in normal and depancreatized dogs. Am. J. Physiol. *187*:357-60.

[8] Phillips, P. H., and Hart, E. B.: The effect of organic dietary constituents upon chronic fluorine toxicosis in the rat. J. Biol. Chem. *109*:657-63, 1935.

[9] Gaebler, O. H., Bartlett, P. D., and Sweeney, M. J.: Remarkable effectiveness of small daily doses of growth hormone in dogs. Am. J. Physiol. *165*:486-90, 1951.

[10] Bartlett, P. D., and Gaebler, O. H.: Anterior pituitary growth preparations: The relationship between nitrogen storage and other criteria of activity. Endocrinology *43*:329-35.

[11] Long, C. N. H., Katzin, B., and Fry, E. G.: The adrenal cortex and carbohydrate metabolism. Endocrinology *26*:309-44.

[12] Lewis, R. A., Kuhlman, D., Delbue, C., Koepf, G. F., and Thorn, G. W.: The effect of the adrenal cortex on carbohydrate metabolism. Endocrinology *27*:971-82, 1940.

[13] McArthur, J. W., Smart, G. A., MacLachlan, E. A., and associates: Studies concerning the role of the adrenal cortex in the pathologic physiology of diabetic acidosis. I. Temporal relations between the metabolic events of experimental diabetic acidosis and the level of adrenal cortical function. J. Clin. Invest. *33*:420-36, 1954.

[14] McArthur, J. W., Gautier, E., Swallow, K. A., and associates. Studies concerning the role of the adrenal cortex in the pathologic physiology of diabetic acidosis. II. The identification of adrenal-conditioned factors in the physiologic reaction to the stress of insulin deprivation. J. Clin. Invest. *33*:437-51, 1954.

[15] Scow, R. O.: Effect of growth hormone on growth in hypophysectomized-pancreatectomized rats. Endocrinology *61*: 582-86, 1957.

[16] Milman, A. E., DeMoor, P., and Lukens, F. D. W.: Relation of purified pituitary growth hormone and insulin in regulation of nitrogen balance. Am. J. Physiol. *166*:354-63, 1951.

[17] Gaebler, O. H.: Some effects of anterior pituitary extracts on nitrogen metabolism, water balance, and energy metabolism. J. Exper. Med. *57*:349-63, 1933.

[18] Gaebler, O. H.: Effects of thyroparathyroidectomy and carbohydrate intake on the action of anterior pituitary extracts. Am. J. Physiol. *110*:584-92, 1935.

[19] Long, C. N. H.: Diabetes mellitus in the light of our present knowledge of metabolism. Transactions & Studies of the College of Physicians of Philadelphia, 4 Ser., Vol. 7, No. 1, 21-46, 1939.

[20] Szego, C. M., and White, A.: The influence of growth hormone on fasting metabolism. Endocrinology *44*:150-66.

[21] Noall, M. W., Riggs, T. R., Walker, L. M., and Christensen, H. N.: Endocrine control of amino acid transfer. Science *126*:1002-05, 1957.

[22] Riggs, T. R., and Walker, L. M.: Hormonal action on the distribution of C^{14}-α-aminoisobutyric acid in rats. Fed. Proc. *17*:297, 1958.