TECHNICAL PAPERS

Alleviation of Experimental Diabetes in Man by Administration of Reduced Glutathione (GSH): Metabolic Implications[1]

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It has been reported from this laboratory (*1, 3*) that a state of metabolism similar to that observed in clinical diabetes mellitus can be produced in normal men and women by the administration of suitable amounts of purified preparations of pituitary adrenocorticotropic hormone (ACTH). The condition produced is characterized by sustained glycosuria and hyperglycemia, glucose tolerance curves characteristic of the diabetic state, and relative resistance to the hypoglycemic effect of exogenous insulin. During administration of ACTH the developing diabetic state is accompanied by a decreasing concentration of blood glutathione (*2*). Because of a temporally related upheaval of purine metabolism (the precise nature of which remains unknown) which is manifested by a sustained increase of urinary uric acid, we have hypothesized that a purine metabolite, exerting an alloxan-like effect, is responsible for reducing the intracellular availability of free sulfhydryl (—SH) groups, which are necessary for normal activity of many enzyme systems; that the combination of decreased intracellular concentrations of —SH and increased intracellular concentrations of purine metabolites impairs functional activity of the insulin-producing cells of the pancreas (the product of which is a protein rich in cystine) and also interferes with peripheral glucose utilization by inhibition of those enzyme systems which require free —SH groups.

In addition to hyperglycemia, and contributing to the degree of glycosuria, is the effect of ACTH in decreasing renal tubular reabsorption of glucose. This easily followed phenomenon can be assigned with reasonable justification to inhibition of an enzyme within the tubular epithelium.

An extensive metabolic balance study (30 days) was made upon a normal young man to determine whether or not large amounts of GSH administered intravenously would reverse (despite continued ACTH administration) an already established hyperglycemia and glycosuria. After an 8-day base-line period, ACTH[3] (68 mg/day, Armour Standard) was given in equal doses every 6 hrs for the following 6 days. On the fourth day of ACTH, pure reduced glutathione (buffered to neutrality and put in solution and into sealed glass containers under nitrogen) was given intravenously in the following amounts: 8 gm at 8 a. m., 4 gm at 1 p. m. and 4 gm at 2 p. m. On the sixth day of ACTH, 4 gm more of GSH were given at 2 p. m. The post-ACTH balance study was continued for the next 16 days. In addition to 24-hr balances throughout, blood specimens were obtained hourly from 8 a. m. to 4 p. m. daily, and urine specimens were collected every 2 hrs from 8 a. m. to 4 p. m. for 11 consecutive days which included the 6-day ACTH period. A large number of determinations other than those relating to carbohydrate metabolism were made upon these samples.

TABLE 1

Day of ACTH	1	2	3	GSH* (4)	5	6
Fasting blood sugar (mg/100 cc)	71	73	108	124	134	148
Blood sugar—average of 9 a. m., 2 p. m., 3 p. m. (mg/100 cc) .	115	165	184	*157*	204	193†
Urine sugar—8 a. m.–4 p. m. (gms)	1.0	5.1	14.7	*4.0*	16.2	24.3

* GSH: administered at 8 a. m., 1 p. m., and 2 p. m. on day No. 4.

† Values for this average are 198 at 9 a. m.; 201 at 2 p. m.; and 179 at 3 p. m. GSH administered once on this day at 2 p. m.

Renal glycosuria occurred on the first day of ACTH. Hyperglycemia became evident on the second day and continued with increasing intensity through the entire ACTH period *except during the intervals when GSH was administered.* The reversal was dramatic but transitory, lasting 1–2 hrs after each injection of GSH. Most striking were (1) sharp elevation of the renal threshold for glucose and (2) a fall of the blood sugar level. These phenomena resulted in glucose-free urine during each hour immediately following administration of GSH. Table 1 summarizes the influence of GSH

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[4] Both blood and urine glucose were determined by the Somogyi procedure as modified by Nelson (*5*).

(during its effective period) upon glycosuria and hyperglycemia produced by ACTH.[4]

The effect of GSH in simultaneously reversing ACTH-induced hyperglycemia and renal glycosuria seems all the more striking since sudden cessation of glycosuria would be expected to result in further increase of hyperglycemia were the rate of utilization of glucose not significantly increased or the supply to the blood significantly decreased. Since a severe degree of negative nitrogen balance remained uninfluenced by GSH, decreased glyconeogenesis from protein was probably not a factor. That hepatic glycogenolysis may have been inhibited remains as a possibility to explain the fall of blood sugar.

Since GSH inactivates insulin *in vitro*, such an effect *in vivo* should have raised the blood sugar level. If this occurred to any degree, its effect was minor as compared with those forces which produced an actual fall of circulating blood sugar.

Also observed were other metabolic reversals of major significance. Among the most striking was a profound change in the number and character of the circulating white blood cells. This remarkable change, which lasted 1–2 hrs, was opposite in direction to the effects which we have observed repeatedly in normal people receiving ACTH for a number of days (*4*). These findings are being reported in detail elsewhere.

Not all of the metabolic effects of increased adrenal corticoid activity were reversed by administration of GSH. This indicates that reversal of those metabolic effects which responded to GSH was not by virtue of a block in steroid production. It seems likely that the changes are due to improved performance of those systems which require free sulfhydryl groups for their normal function. This appears to be the case with respect to (1) renal tubular reabsorption of glucose, (2) the systems involved in utilization of glucose, and (3) the systems responsible for production and release of white blood cells.

It is believed that the results reported give added weight to our hypothesis. The forces at work in the early stages of the development of human diabetes may be very similar to those described in this and in our previous studies. Of somewhat broader significance are the implications with respect to abnormalities of metabolism generally. Hormonal control of enzymatic processes, mediated via independent effects of the same hormone upon other systems, appears to be a fertile field for future investigations. But it implies the necessity of establishing technics which can be applied in the intact animal or man.

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Production of Acute Gouty Arthritis by Adrenocorticotropin[1]

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It has long been known that attacks of acute gouty arthritis can be precipitated by exposure to nonspecific stresses such as trauma, infection, operation, chilling, foreign protein therapy, X-radiation, and many other stimuli. These stresses have been shown to produce a syndrome of increased adrenal cortical activity (*5*) and associated biochemical and histologic changes, commonly referred to as "the alarm reaction" (*4*).

It is likely that all forms of nonspecific stress call forth increased adrenal cortical activity through a common pathway. This pathway is increased secretion of pituitary adrenocorticotropin (ACTH), which in turn stimulates the adrenal cortex (*4*).

ACTH has therefore been given to four patients with gout and has precipitated three attacks of acute gouty arthritis in five trials. The entire syndrome of electrolyte and water changes observed to occur with the acute attack of gout (*7*) has also been reproduced by the administration of ACTH.

In addition, since it was shown that the attack of gout follows a phase of relatively decreased adrenal cortical activity, the administration of ACTH to two patients during an attack of acute gouty arthritis produced a prompt disappearance of the acute arthritis. This aspect of the action of ACTH resembles that of colchicine, which relieves the acute attack of gout and is a potent stimulus to increased adrenal activity (*2*).

Three of the four patients were studied on a metabolic ward and received diets of known composition. After a contral period of four days, 150 mg (equivalent to Armour standard) ACTH[3] was given by intramuscular injection each day in divided doses for four days.

Control serum uric acid was elevated in all patients. During the administration of ACTH, there was an increase in urinary uric acid and nitrogen and a decrease in sodium and chloride excretion. Three patients had glycosuria of one to three gm daily. In two patients, there was a significant increase in glomerular filtration rate, renal plasma flow, and uric acid clearance (*1*). The circulating eosinophils disappeared while ACTH was being given.

For three days following completion of the ACTH injections, there was an excretion of sodium and chloride considerably greater than the control values while the uric acid excretion returned to the control value. The

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