Hypoglycemia in Compensated Chronic Renal Insufficiency

Substrate Limitation of Gluconeogenesis

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SUMMARY

A woman with diabetes mellitus, compensated chronic renal insufficiency, and multiple episodes of fasting hypoglycemia was studied. On fasting, despite appropriate insulin, glucagon and normal lactate levels, this patient developed early, profound hypoalaninemia followed by hypoglycemia. Glucose and alanine turnover rates were estimated after an overnight fast by the primed injection—continuous infusion of isotopically labeled alanine and glucose. Glucose production (mean ± S.E.) was 36.4 ± 2.8 mg./kg. hr. (normal 129.0 ± 10.1 mg./kg. hr., n = 6). Glucose production from alanine was 3.1 ± 0.7 mg./kg. hr. (normal 18.0 ± 1.0 mg./kg. hr.). Alanine turnover was 241 ± 11 μmoles/kg. hr. (normal 488 ± 48 μmoles/kg. hr.). These data suggest that an inadequate delivery of an important gluconeogenic substrate—alanine—can exert a significant rate limitation on glucose production thereby resulting in hypoglycemia during fasting. DIABETES *23*:982-86, December, 1974.

The appearance of glucose intolerance[1] as well as clinical amelioration of the diabetic state[2] have been associated with progressive deterioration of renal function. Recently, unexplained spontaneous hypoglycemia has been reported in patients with chronic renal insufficiency.[3-7] We have studied such a patient with diabetes mellitus, chronic renal insufficiency and fasting hypoglycemia. On the basis of these studies, a pathophysiologic mechanism for the development of fasting hypoglycemia due to a substrate limitation of glyconeogenesis is proposed.

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CASE REPORT

A fifty-seven-year-old woman (BH 1-0-73-19009) was found unarousable and brought to Barnes Hospital on July 1, 1973.

Diabetes mellitus was discovered in 1953 and insulin was subsequently prescribed. Retinal microaneurysms, hemorrhages, preretinal neovascularization and absent ankle jerks were noted in 1968. At that time the endogenous creatinine clearance was 31 ml. per minute, the urinary protein 3.0 gm. per twenty-four hours and the serum albumin 2.6 gm. per dl. Fasting plasma glucose concentrations ranged from 216 to 344 mg./dl. Insulin therapy was discontinued and tolazamide, 250 mg. daily, was prescribed. In August of 1969 fasting plasma glucoses ranged from 156 to 176 mg./dl. Anorexia, nausea and generalized weakness led to hospitalization in August of 1971. On admission the plasma glucose concentration was 46 mg./dl. but thirty-six hours later the fasting glucose level was 83 mg./dl. The creatinine clearance was 8 ml. per minute. Tolazamide was discontinued. Oral hypoglycemic agents were not prescribed thereafter. The fasting plasma glucose was 70 mg./dl in June of 1972.

The morning of July 1, 1973 the patient could not be aroused and was brought to Barnes Hospital. The plasma glucose concentration was 55 mg./dl; after intravenous glucose injection the patient awoke. A firm, nontender 5 x 3 cm. right submandibular mass was present. There was no hepatomegaly. The endogenous creatinine clearance was 3 ml. per minute. The total serum bilirubin was 0.4 mg./dl, the albumin 2.6 gm./dl and the serum glutamic oxaloacetic transaminase (SGOT) 20 mU/ml. The hematocrit was 29 per cent. There was 2+ proteinuria. Chest films revealed only mild cardiomegaly. The serum thyroxine concentration and an 0800 plasma cortisol level were normal. On the eighth hospital day the patient was fasted. After twenty-eight hours the patient became somnolent, blood was drawn and the symptoms cleared after intravenous glucose. The serum glucose determined by a cupric neocuproine AutoAnalyzer technic[8] was 65 mg./dl but was 33 mg./dl using a glucose oxidase method.[9] The submandibular mass was incised and drained a small amount of thick green pus. Routine bacterial and fungal cultures were sterile. However, *Mycobacterium tuberculosis* was subsequently isolated. After further diagnostic studies (see below) and supportive management, including peritoneal dialysis and the initiation of rifampin and isoniazid therapy, the patient was discharged on a multiple feeding dietary regimen. Ten months after discharge she remained free of symptomatic hypoglycemia.

ANALYTICAL METHODS

Alanine,[10] β-hydroxybutyrate,[11] glucose[12] and lactate[13] were assayed fluorometrically in neutralized perchloric acid extracts of plasma using enzymatic methods. Insulin[14] and glucagon[15] were determined by double antibody radioimmunoassay. Turnover rates of plasma glucose and alanine were estimated by standard isotope dilution methods using a primed injection—continuous five-hour infusion of 50 μCi of glucose-2-^{3}H and 50 μCi of alanine-^{14}C (U). Plasma glucose specific activity was determined by the method of Kreisberg et al.[16] and radioactivity in plasma alanine was measured in the total alanine fraction collected by high speed, high resolution liquid chromatography.[17] Alanine recovery by this procedure ranged between 94 and 103 per cent. Alanine turnover and glucose appearance and disappearance rates were calculated according to standard formulas. The rate of appearance of glucose derived from alanine was determined from the carbon-14 precursor-product relationships previously described for lactate.[16]

RESULTS

On the eleventh hospital day, the patient was transferred to the Washington University School of Medicine Clinical Research Center for diagnostic evaluation. Caloric intake was not less than 1200 calories daily, consisting of a minimum of 50 gm. protein and 150 gm. carbohydrate for six days prior to the performance of the metabolic studies. An intravenous glucose tolerance test showed a k value for glucose disappearance of 0.5 per cent per minute. During a 75 gm. oral glucose tolerance test, the peak plasma glucose was 262 mg./dl. Peak serum insulin levels exceeded 90 μUnits/ml. during both glucose tolerance studies. There were also elevations of basal insulin levels, ranging from 21 to 29 μUnits/ml. after an overnight fast. These values may have resulted from the presence of measurable levels of insulin antibodies in this patient despite her lack of exposure to exogenous insulin for more than five years.

In view of the patient's apparent inability to maintain normoglycemia, a fasting study was initiated. Venous blood levels of glucose, β-hydroxybutyrate, lactate, alanine, glucagon and insulin were determined every four hours (figures 1 and 2). Plasma glucose fell continuously from 89 mg./dl at the beginning of the fast, to 33 mg./dl at the sixtieth hour of fasting, at which time the study was terminated. Serum insulin levels declined in parallel with the decline in plasma glucose, from 25 μUnits/ml. initially, to less than 9 μUnits/ml. at the time of hypoglycemia. Plasma glucagon was elevated after an overnight fast, as observed in other patients with chronic renal disease,[18] and these elevations persisted in this patient throughout the fasting period.

Circulating levels of various gluconeogenic substrates were determined during the fast. There were no observable increases in venous blood lactate levels throughout the study even at the point of hypoglycemia. Levels of β-hydroxybutyrate increased rapidly, reaching a concentration of 3mM at the fifty-seventh hour. At the beginning of the fast, the plasma concentration of alanine was normal (0.3mM). However, plasma alanine levels fell precipitously with fasting to 0.09 mM, at forty-eight hours. The abrupt fall in alanine levels on day 1 of the fast preceded by several hours a similar decline in plasma glucose.

In order to evaluate the relationships between hypoalaninemia and hypoglycemia, simultaneous glucose-2-^{3}H and alanine-^{14}C (U) turnover rates were determined during the first day of the fast (table 1). Glucose production (inflow) and utilization (outflow) rates were determined hourly during the second through fourth hours of the infusion and half-hourly during the final hour. Glucose production averaged 36.4 ± 1.5 mg./kg. per hour (mean ± S.E.) and glucose utilization was 36.9 ± 1.5 mg./kg. per hour, markedly reduced from the glucose inflow-outflow rates of 129.4 ± 10.1 mg./kg. per hour obtained in six normal controls. Alanine turnover determined from the mean specific activity plateau during the final three hours of infusion was 241 ± 10.7 μmoles/kg. per hour, substantially less than the mean normal value of 488 ± 48 μmoles-kg. per hour. Although Steele's equations for substrate inflow and outflow[19] were originally derived for glucose, they have been subsequently applied to lactate kinetics by others.[16] It has been our experience that these equations apply equally well to alanine when appropriate corrections for the apparent distribution of alanine in total body water are included.[20,21] In the present patient, the average alanine inflow of 238 ± 8 (mean ± S.E.) and outflow 248 ± 15 μmoles/kg. per hour for the final three-hourly intervals are virtually identical to the turnover rate determined from the specific activity plateau. Glucose production from alanine was 3.1 ± 0.7 mg./kg. per hour (normal 18.0 ± 1.0 mg./kg. per hour).

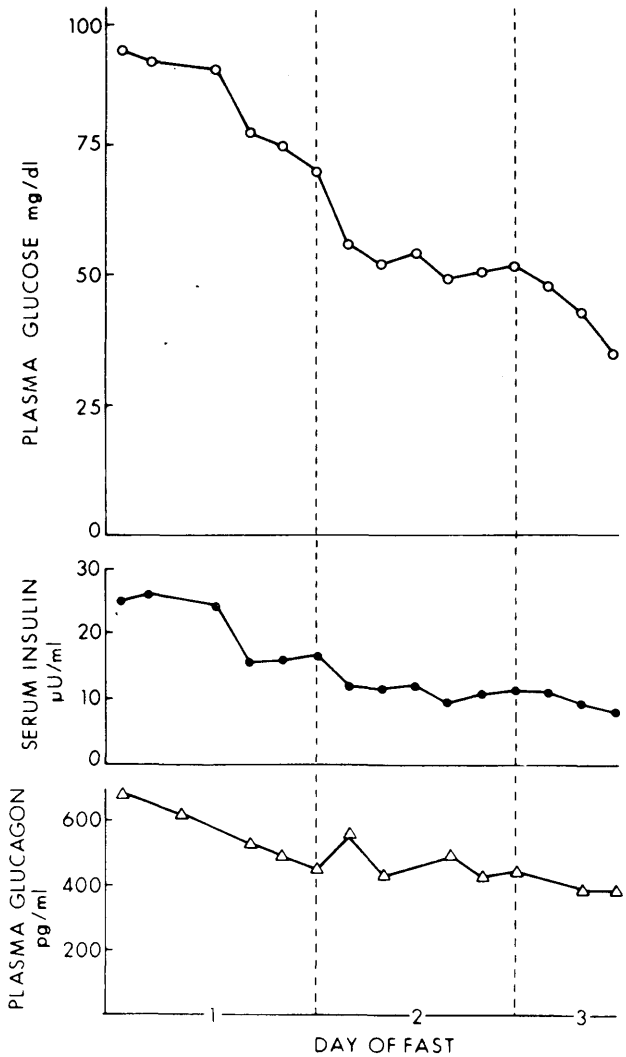

FIG. 1. Metabolic responses to fasting in a patient with spontaneous hypoglycemia and chronic renal disease. Fasting was initiated at midnight following a bedtime meal. Vertical dashed lines indicate the twenty-four hour intervals between successive midnights. Glucose was determined enzymatically on neutralized perchloric acid extracts of plasma. Insulin and glucagon were measured in serum and plasma, respectively.

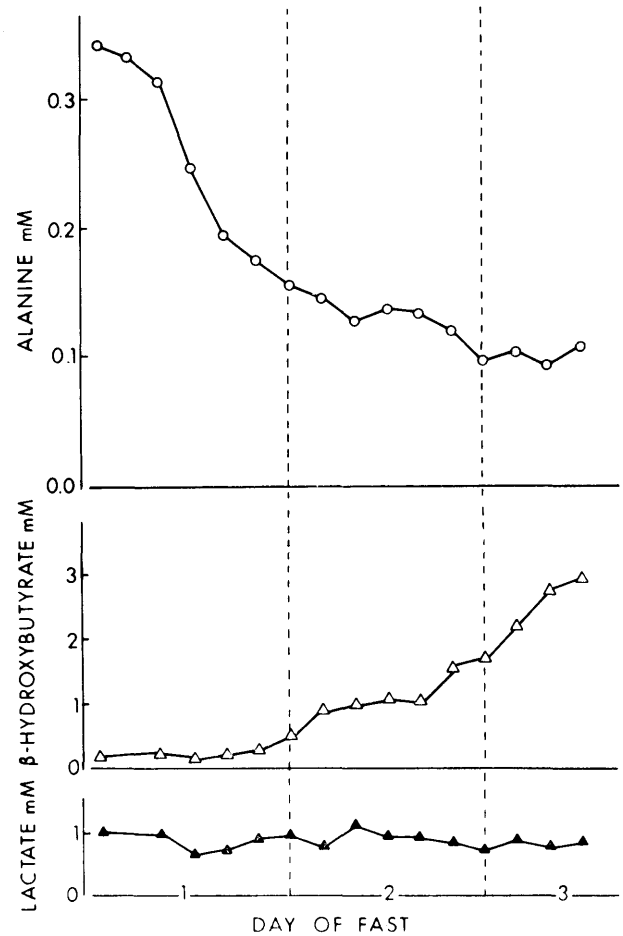

FIG. 2. Metabolic responses to fasting in a patient with spontaneous hypoglycemia and chronic renal disease. Fasting was initiated at midnight following a bedtime meal. Vertical dashed lines indicate the twenty-four hour intervals between successive midnights. Alanine, β-hydroxybutyrate and lactate were determined enzymatically on neutralized perchloric acid extracts of plasma.

DISCUSSION

Although a number of diverse metabolic abnormalities have been described in patients with compensated chronic renal insufficiency, the development of spontaneous hypoglycemia has been recognized only recently, and its pathogenesis is unclear. All patients reported had normal adrenal, hepatic and thyroid function. In this patient, the appropriate decline in serum immunoreactive insulin levels together with the prompt development of prominent ketosis during the fasting study provide good evidence that there was not inappropriate hyperinsulinism. Glucagon insufficiency can be excluded in this patient since persistently elevated glucagon levels were observed throughout the starvation period. No evidence of hepatocellular dysfunction was observed, nor was there any abnormality of adrenal, pituitary or thyroid function.

A decreased rate of gluconeogenesis appears to be the principal phenomenon underlying the spontaneous hypoglycemia in this woman. The deficiency arises from neither an hepatic nor a renal defect, but rather from an inadequate availability of the primary gluconeogenic amino acid substrate—alanine. Five observations support this conclusion: First, the hypo-

TABLE 1

Turnover and precursor-product relationships of alanine and glucose

Glucose and alanine kinetics were estimated isotopically using a primed injection—continuous five hour infusion of glucose-2-^{3}H and alanine-^{14}C (U). Blood samples were obtained after 2, 3, 4, 4½ and 5 hours of infusion. Values shown for the patient are the means (± S.E.) for those determinations. Comparison values obtained in six normal weight [74.6 ± 2.7 kg. (mean ± S.E.)] controls were determined under identical conditions.

Subjects	Glucose production	Alanine turnover	Glucose production from alanine
	mg./kg./hr.	μmoles/kg./hr.	mg./kg./hr.
Controls	129.4 ± 10.1	488 ± 48	18.0 ± 1.0
Patient	36.4 ± 1.5	241 ± 10.7	3.1 ± 0.7

glycemia was relatively delayed in onset, becoming apparent at a time (fifty-seven hours) when the role of gluconeogenesis for the maintenance of blood glucose is paramount.[22] Second, blood lactate levels remained normal in this patient during fasting. Thus, it appears likely that, despite the low rates of glucose production, there was no hepatic defect in the metabolic clearance of gluconeogenic substrates. In contrast to this case, lactate accumulation has been observed in patients with absolute deficiencies of enzymes essential to the pathway of hepatic gluconeogenesis.[23] Third, alanine levels fell precipitously upon fasting to abnormally low levels not encountered in normals even upon prolonged fasting.[24] At the time of hypoglycemia, the patient's alanine level was less than one-half the level observed in comparably fasted normal controls and the observed fall in alanine levels preceded by several hours a similar decline in the concentration of plasma glucose. Fourth, isotopically measured alanine turnover and production rates were substantially reduced in this patient. Last and most important, the absolute rate of glucose production from alanine was markedly depressed prior to the appearance of hypoglycemia. It seems probable, therefore, that overall rates of glucose formation during the first day of fasting were not matched by appropriate rates of alanine production. As a consequence of this imbalance alanine levels decline, thereby imposing a considerable substrate limitation on the rate of gluconeogenesis. Alanine is the principal amino acid substrate for hepatic gluconeogenesis.[24] Thus alanine insufficiency alone could limit the overall rate of new, nonrecycled glucose formation. This conclusion is supported by studies in the perfused rat liver, in which alterations of alanine concentration have been shown to produce proportional changes in the rate of glucose formation.[25]

Previous studies have suggested that the hypoglycemia noted in patients with advanced renal disease may result from inadequate glycogenolysis.[3-6] Such a deficiency also may have contributed to the hypoglycemia in this patient. There was a significant reduction in the overall rate of glucose production even after an overnight fast. Since glycogenolysis is quantitatively most important to glucose homeostasis in the postabsorptive state,[26] it seems likely that some decrease in this process may account in part for the observed, low rate of glucose production. Nevertheless, in view of the late onset of hypoglycemia, and the decreased rate of glucose production from alanine, the importance of inadequate gluconeogenesis for the development of fasting hypoglycemia seems paramount in this patient.

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