Effect of metabolic acidosis on insulin action and secretion in uremia

ROBERT H.K. MAK

Division of Nephrology, Department of Pediatrics, Oregon Health Sciences University, Portland, Oregon, USA

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Background. Metabolic acidosis affects both vitamin D and insulin metabolism. Vitamin D is important in modulation of both insulin secretion and insulin sensitivity in uremia. The present study examines the effect of correction of metabolic acidosis on insulin action and secretion as well as 1,25 vitamin D₃ concentrations in uremic patients.

Methods. Eight patients (age 18 ± 1 year) on maintenance hemodialysis with metabolic acidosis were studied before and after two weeks of oral sodium bicarbonate (NaHCO₃) treatment to correct the acidosis. To control for the effect of additional sodium, they were also studied after two weeks of an equivalent amount of oral sodium chloride (NaCl). Controls consisted of 7 healthy controls (age 19 ± 1 year). Insulin sensitivity was measured by the hyperinsulinemic euglycemic clamp technique. Insulin secretion was measured by the hyperglycemic clamp technique.

Results. Oral NaHCO₃ treatment led to significant increases in venous pH and serum bicarbonate concentrations but no significant change in intact parathyroid hormone (PTH) concentrations. Circulating 1,25 dihydroxyvitamin [(OH)₂]D₃ were significantly lower than control values initially and increased significantly after treatment. Oral NaCl did not change any of the biochemical parameters. Before treatment of acidosis, uremic patients had lower insulin sensitivity (insulin resistance) during constant hyperinsulinemia and lower insulin secretion during constant hyperglycemia compared with controls. Following two weeks of NaHCO₃ treatment there were significant increases in insulin sensitivity and insulin secretion, although the values did not normalize. There were no changes in insulin sensitivity or insulin secretion following two weeks of NaCl.

Conclusion. Treatment of metabolic acidosis increased both insulin sensitivity and insulin secretion in patients with uremia. This was accompanied by an increase in the circulating levels of 1,25(OH)₂D₃ but no change in those of parathyroid hormone.

Reduced tissue sensitivity to the hypoglycemic action of insulin is present almost universally in patients with moderate to severe uremia. Defects in insulin secretion have also been reported in these patients. The etiology of insulin abnormalities in CRF is probably multifactorial but not clearly understood [1, 2]. Metabolic acidosis is a frequent complication of uremia and may contribute to insulin resistance. Correction of metabolic acidosis in rats with uremia partially corrects insulin resistance [3]. NaHCO₃ treatment of patients, with metabolic acidosis from chronic renal failure not on dialysis, increases insulin sensitivity [4]. The mechanism by which metabolic acidosis affects insulin sensitivity in uremia is not clear. Metabolic acidosis may contribute to vitamin D deficiency in uremia [5]. Administration of 1,25 dihydroxyvitamin D₃ [1,25(OH)₂D₃] improves insulin sensitivity [6] and insulin secretion [7] in vitamin D deficient uremic patients on dialysis. The present study examines the effect of correction of metabolic acidosis on insulin metabolism as well as circulating levels of 1,25(OH)₂D₃ in patients with uremia on hemodialysis (HD).

METHODS

Eight patients (age 18 ± 1 year; 3 males and 5 females) with uremia on maintenance HD were studied. The underlying diagnoses included renal dysplasia, obstructive uropathy, reflux nephropathy, prune belly syndrome and unknown etiology. They had been stabilized on maintenance HD for at least six months prior to the study. Their blood pressure levels were controlled on medications and stabilized for at least two months before the study. None of the patients were obese. Their body wts were within 10% of the ideal weight for their height.

These patients had moderate metabolic acidosis detected on entry into the study, and they underwent the initial metabolic studies while being acidic. Four of them were then treated first with oral NaHCO₃ (3 mEq/kg/day) for two weeks and restudied. They then were treated with an equivalent amount of NaCl for another two weeks and studied at a third time point. The other four patients were first treated with NaCl and then with NaHCO₃, and were studied in the same way. Other medications included dihydrotachysterol, calcium carbonate, antihypertensives in
the form of long acting nifedipine, water-soluble vitamins (Nephrovitea; R & D Laboratories, Inc., Marina Del Rey, CA, USA) and intravenous erythropoietin during HD sessions. All the medications (other than NaHCO₃ or NaCl) were continued and there was no dosage changes during the study. Their carbohydrate intake was 4.13 ± 0.21 g/kg/day and their body wts were stable for at least two months prior to the studies. Their dietary intakes of sodium (2 g/day), potassium (2 g/day) and phosphorus (800 mg) were restricted but the intake of protein was not. Dietary protein, phosphorus and sodium intakes were monitored by a three day recall every two weeks during the study and did not change. None of the patients had a personal history or family history of diabetes mellitus. All patients had Tanner puberty scores of 5. Controls consisted of 7 healthy subjects (age 19 ± 1 years; 3 males and 4 females) consuming regular weight maintaining diets and taking no medications. None of the patients had diabetes mellitus, nor was there a family history of diabetes mellitus. All patients and controls were free from infections at the time of the studies. The clinical studies were performed at Childrens’ Hospital of Los Angeles and were approved by the Committee for Clinical Investigations (local institutional review board). The purpose and potential risks of the study was carefully explained to all patients and subjects, and written informed consent was obtained before their participation.

Euglycemic clamp studies

All studies were started about 9 a.m. after an overnight fast of about 10 to 12 hours just before a scheduled HD session. The patients sat comfortably in a reclining chair and were not allowed to eat or drink apart from water during the studies. Insulin sensitivity was measured by the hyperinsulinemic euglycemic clamp technique [8]. Two intravenous lines were started in contralateral arms. One intravenous line was inserted in a vein on the dorsum of the hand, kept patent by a slow intravenous infusion of normal saline and used for blood sampling. This hand was placed in a heated box (65°C) to arterialize the blood [9]. An indwelling catheter was placed in a vein in the opposite arm for infusion of glucose and insulin. After obtaining at least three fasting serum samples for glucose and insulin concentration, a prime-continuous infusion of insulin was given intravenously at 40 mU/m² body surface area/min of to acutely raise and maintain the serum insulin concentrations at a plateau of around 100 μU/ml for 120 minutes. Serum glucose concentration was measured at five minute intervals and a variable infusion of 20% dextrose was adjusted to maintain the glucose concentration at fasting levels. Serum was also obtained every ten minutes for measurement of insulin concentrations. Under steady state conditions of euglycemia and hyperinsulinemia during the euglycemic clamp study, the rate of glucose infusion provides an index of insulin-stimulated glucose metabolism and is used as an index of insulin sensitivity (mg/m²/min).

Hyperclycemic clamp studies

Insulin secretion was measured by the hyperglycemic clamp technique as previously described [8]. Patient preparation was similar to the euglycemic clamp. A priming dose of 20% dextrose was given to acutely raise blood glucose concentration at 125 mg/dl (6.9 mmol/liter) above fasting glucose concentrations. Constant hyperglycemia at this level was then maintained for 120 minutes by varying the infusion of 20% dextrose but no insulin. In the first 10 minutes of the study, blood was taken at two minute intervals for measurement of serum glucose and insulin. The mean concentration of serum insulin in the first 10 minutes is an index of early insulin secretion (Ie) μU/ml in response to hyperglycemia. Thereafter, blood was taken every five minutes for measurement of serum glucose and every 10 minutes for measurement of serum insulin concentrations. The mean serum insulin concentration during the hyperglycemic clamp study, at ten minute intervals from 20 to 120 minutes, is an index of late insulin secretion (I) μU/ml under steady state conditions of constant hyperglycemia. Ie provides an index of insulin release from performed stores in the pancreas, whereas I provides an index of ongoing insulin synthesis and release.

Biochemical analysis

Serum glucose concentration was measured by the glucose oxidase method using a Yellow Springs 23 AM glucose analyzer (Yellow Springs Instruments, OH, USA). Serum immunoreactive insulin concentration was measured by double antibody radioimmunoassay (Pharmacia, Upplasa, Sweden). The intraassay and interassay coefficients of variation for serum insulin were 5.2 and 8.2%, respectively. Intact serum parathyroid hormone (PTH) was measured by an immunoradiometric assay (Nichols, San Juan Capistrano, CA, USA). The intraassay and interassay coefficients of variation for serum PTH were 7.2 and 9.3%, respectively. Serum 1,25(OH)₂D₃ was measured by a radioreceptor assay (Nichols) [10]. This assay is specific for both 1,25(OH)₂D₃ and 1,25(OH)₂D₂. It involves a preliminary extraction and purification of serum using a single column containing a C₁₈OH activated matrix. Once purified, the material was quantitated in a radioreceptor assay using a calf thymus receptor and tritiated 1,25(OH)₂D₃. Separation of bound from free 1,25(OH)₂D₃ was achieved by incubation with dextran coated charcoal. The intraassay and interassay coefficients of variation for serum 1,25(OH)₂D₃ were 8.2 and 9.9%, respectively. Serum biochemistry was measured by standard methods on multichannel autoanalyzers.

Statistical analysis

All values are expressed as mean ± SEM. The data were tested for normality using the chi square method. Student t-tests for paired and unpaired observations and the least
square method for linear regression were used for analysis of the results. Statistical significance was recognized at the 5% level.

RESULTS

Serum biochemical data of the patients at baseline (while acidic) and during treatment with NaHCO₃ and NaCl are presented in Table 1. Arterial pH and serum HCO₃ increased significantly during NaHCO₃ treatment ($P < 0.01$ in both cases) but did not change during NaCl treatment. Fasting serum glucose decreased and fasting serum insulin increased following NaHCO₃ treatment, but did not change during NaCl treatment.

Hyperglycemic clamp studies

Mean serum insulin concentrations during the euglycemic clamp studies, in patients on hemodialysis at baseline (while acidic), after two weeks of NaHCO₃ or after two weeks of NaCl or in controls (Fig. 1). The bottom panel represents the mean serum insulin concentrations during the euglycemic clamps. *$P < 0.01$ vs. control; # $P < 0.01$ vs. acidic.

**DISCUSSION**

Metabolic acidosis is a common complication of uremia and may contribute to the pathogenesis of insulin resistance. DeFronzo and Beckles studied ammonium chloride induced metabolic acidosis in human subjects with normal
renal function and found impaired tissue sensitivity to both endogenous and exogenous insulin [11]. Correction of metabolic acidosis in rats with chronic uremia partially corrects insulin resistance [3]. Jenkins et al assessed carbohydrate tolerance after correction of acidosis in patients with chronic renal failure using a crude one-hour glucose infusion methodology with measurement of glucose and insulin concentrations, and did not find any changes [12]. However, Reaich et al, using the euglycemic clamp technique (as in the present study), showed that NaHCO₃ treatment of patients with metabolic acidosis from chronic renal failure significantly increased insulin sensitivity [4]. The latter authors suggested that the use of the euglycemic clamp permitted greater sensitivity in the detection of changes in insulin-mediated glucose uptake. No previous study has examined the effect of acidosis on insulin secretion.

The present study showed that while acidic, uremic patients on HD had lower insulin sensitivity (insulin resistance) during constant hyperinsulinemia and lower insulin secretion during constant hyperglycemia compared with controls. Following two weeks of NaHCO₃ treatment, there were significant increases in insulin sensitivity and insulin secretion, although the values did not normalize. To control for the effect of additional sodium, they were also studied after two weeks of an equivalent amount of oral sodium chloride. There were no changes in insulin sensitivity or insulin secretion following two weeks of NaCl. The NaHCO₃ and NaCl studies were also randomized so that the cumulative effects of HD, which is also known to improve insulin sensitivity, can be ruled out [13, 14]. Also, these patients were treated with erythropoietin for their moderate anemia. Correction of anemia by erythropoietin is known to ameliorate insulin resistance in HD patients [15]. However, since hematocrit values did not change during the study and since there are randomized control studies with NaCl, the cumulative effects of erythropoietin on insulin metabolism are unlikely to be important. Thus, in the present study, treatment of metabolic acidosis increased both insulin sensitivity and insulin secretion in patients with end-stage renal disease on maintenance HD. However, the mechanism by which metabolic acidosis affects insulin metabolism in uremia is not clear.

Metabolic acidosis is known to alter vitamin D metabolism. Lu et al showed that acute correction of metabolic acidosis by intravenous NaHCO₃ infusion significantly increased serum 1,25(OH)₂D₃ concentrations without changes in plasma ionized calcium, potassium, magnesium, phosphorus and 25 hydroxyvitamin D₃ concentrations in patients with moderate chronic renal failure not on dialysis [5]. The present study confirms these results in patients with end-stage renal disease on maintenance hemodialysis. There were no simultaneous changes in oral vitamin D or phosphorus binder dosages, dietary phosphorus intakes or PTH concentrations, so that the 1,25(OH)₂D₃ changes were most likely a result of correction of acidosis. 1,25(OH)₂D₃ concentrations in the patients at the baseline acidic state were about 1/3 of control values and rose significantly after correction of metabolic acidosis but did not normalize. This may reflect the low renal reserve in these patients in terms of their remaining 1-α hydroxylase capacity. It is interesting that the insulin parameters follow this general trend: (1) they were fairly low to begin with (less than 50% of control values); and (2) they increased significantly following correction of metabolic acidosis but
did not normalize. These results suggest the mechanism of improvement in insulin abnormalities following correction of acidosis may be related to an increase in circulating 1,25(OH)_2D_3 concentrations. Indeed, there is evidence that either vitamin D deficiency and/or secondary hyperparathyroidism may contribute to the defects in glucose-stimulated insulin secretion and sensitivity in patients with end-stage renal disease. Pharmacological doses of intravenous 1,25(OH)_2D_3, given acutely, corrected glucose intolerance and increased insulin secretion in 1,25(OH)_2D_3-deficient hyperparathyroid hemodialysis patients without changing serum PTH concentrations [7]. Furthermore, Kautzky-Willer et al reported normalization of insulin sensitivity in patients on hemodialysis after 12 weeks of intravenous 1,25(OH)_2D_3 therapy [6]. The relationship between 1,25(OH)_2D_3 and insulin in uremic patients with acidosis and 1,25(OH)_2D_3 deficiency in the present study may be confirmatory of other studies of patients with uremia and 1,25(OH)_2D_3 deficiency alone [6, 7]. Alternatively, the observed changes in 1,25(OH)_2D_3 and insulin following correction of acidosis in uremic patients may be unrelated and not causal.

Parathyroid hormone is also known to impact insulin metabolism in uremia. Treatment of hyperparathyroidism in uremic patients, either surgically [16] or medically [17], led to the correction of glucose intolerance, an increase in insulin secretion and no change in insulin sensitivity. Akmal et al examined the effect of hyperparathyroidism on insulin metabolism in uremia by parathyroidectomy in a uremic dog model and came to the same conclusion [18]. There was a significant decrease in parathyroid hormone concentrations accompanying correction of insulin resistance in patients treated with intravenous 1,25(OH)_2D_3 in the study by Kautzky-Willer et al, so the role of secondary hyperparathyroidism cannot be ruled out [6]. In the present study, the improvements in insulin action and secretion occurred without changes in parathyroid hormone concentrations. However, this does not rule out the role of parathyroid hormone action. Metabolic acidosis inhibits the renal actions of parathyroid hormone [19] including the stimulation of renal 1α-hydroxylase [20]. The relief of acidosis may allow such actions to be increased without changes in levels of the hormone. Indeed, the increase in 1,25(OH)_2D_3 concentrations in the present study is likely to be due to the increased action of parathyroid hormone on renal 1α-hydroxylase following correction of metabolic acidosis. Whether this applies to the action of parathyroid hormone on insulin metabolism following correction of acidosis in uremia remains to be determined.

In summary, correction of metabolic acidosis increases insulin sensitivity and insulin secretion in uremic patients on HD. This is accompanied by an increase in the circulating levels of 1,25(OH)_2D_3 but no change in those of parathyroid hormone. Further research is needed to better define the pathophysiology.

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Correspondence to Robert Mak, M.D. Ph.D., Department of Pediatrics, Mailcode NRC5, Oregon Health Sciences University, 3181 S.W. Sam Jackson Park Road, Portland, Oregon 97201-3098, USA.

E-mail: makr@ohsu.edu

REFERENCES