
Original Articles

URINARY NET CHARGE IN HYPERCHLOREMIC METABOLIC ACIDOSIS**Seema Kumar, Meera Vaswani*, R.N. Srivastava and Arvind Bagga**

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This paper received the S.T. Achar Award at the 34th National Conference of Pediatrics held in Ahmedabad (1997).

Manuscript received: May 15, 1997; Initial review completed: July 8, 1997;

Revision accepted: September 1, 1997

Objective: (i) To examine the usefulness of urinary net charge ($UNa + UK - UCl$) in the evaluation of hyperchloremic metabolic acidosis secondary to diarrhea, distal RTA and proximal RTA and (ii) To characterize the type of distal RTA on the basis of the underlying defect.

Setting: Pediatrics division of a tertiary referral center. **Subjects:** Thirty four children with hyperchloremic metabolic acidosis secondary to diarrhea ($n=16$), distal RTA ($n=11$) and proximal RTA ($n=7$). Ten normal children with ammonium chloride induced acidosis were also studied. **Methods:** All subjects underwent urine collection of 30-60 minutes duration for measurement of Na, K, Cl, pH and pCO_2 . These measurements were also made on the blood samples collected at the midpoint of urine collection. The urinary net charge was calculated by subtracting Cl values from the sum of the Na and K. **Results:** Patients with proximal and distal RTA had a positive urine net charge. Patients with diarrhea and ammonium chloride induced acidosis showed negative urine net charge. Patients with diarrhea with extremely low urine sodium levels showed an inappropriately high urine pH despite persistent metabolic acidosis. All patients with distal RTA were found to have a secretory type of defect. **Conclusion:** Measurement of urine net charge is helpful in the initial evaluation of a patient with hyperchloremic metabolic acidosis.

Key words: Renal tubular acidosis, Diarrhea, Urine ammonium, Urine anion gap.

HYPERCHLOREMIC metabolic acidosis is a commonly encountered clinical problem in children. It is usually caused by gastrointestinal bicarbonate loss as in diarrhea, renal tubular acidosis (RTA), treatment with carbonic anhydrase inhibitors and exogenous acid loads. An increase in urinary ammonium excretion is an important feature of appropriate renal response to metabolic acidosis(1). Patients

with distal RTA do not show increased urinary ammonium excretion(2). Measurement of ammonium excretion therefore is useful in diagnosis of distal RTA(2). The estimation of urinary ammonium is however cumbersome and few laboratories perform this test routinely.

The urine net charge (anion gap) is a reliable, although not precise, quantitative index of ammonium excretion(3). The urine

net charge is calculated using urinary sodium, potassium and chloride concentration. Since the sum of urine anions and cations must be equal(3), therefore:

$$\text{Na} + \text{K} + \text{Ca} + \text{Mg} + \text{ammonium} = \text{Cl} + \text{sulfate} + \text{phosphate} + \text{other anions}$$

On a regular diet the amounts of calcium, magnesium, phosphate, sulfate and organic anions is fairly constant. The difference in excretion rates of these normally unmeasured anions and cations is close to 80 mEq/day(3), therefore:

$$\text{Na} + \text{K} + \text{ammonium} = \text{Cl} + 80$$

The urine net charge ($\text{UNa} + \text{UK} - \text{UCl}$), thus reflects the excretion of ammonium and becomes progressively negative as the rate of ammonium excretion increases. A negative net charge is found in patients with hyperchloremic metabolic acidosis secondary to diarrhea, and exogenous acid loading. A positive urine net charge indicates defective distal tubular urinary acidification (distal RTA)(2-4). The urine net charge has not been examined in patients with proximal RTA.

We have prospectively examined the usefulness of the urinary net charge estimation in patients with hyperchloremic metabolic acidosis due to diarrhea, RTA and exogenous acid load. Patients with distal RTA were further studied to characterize the underlying defect.

Subjects and Methods

Thirty four patients with hyperchloremic metabolic acidosis between the ages of 6 months to 14 yrs and 10 normal children given an exogenous ammonium chloride load were studied. Sixteen patients had hyperchloremic metabolic acidosis due to acute diarrhea, 11 had distal RTA and 7 proximal RTA. Informed parental consent was obtained. The study was approved by the Ethics Committee of the Institute.

Hyperchloremic metabolic acidosis was defined as blood pH less than 7.35, bicarbonate less than 18 mEq/l and a normal plasma anion gap (8-16 mEq/l)(5). Blood samples were taken at the time of admission before any therapy was started, and urine parameters measured on the first voided urine sample. Children with diarrhea and suspected acute tubular necrosis whose urine output and blood levels of urea and creatinine did not normalize following correction of dehydration were excluded. All subjects with suspected RTA underwent urine collection of 30 to 60 minutes duration for measurement of Na, K and Cl excretion, pH and pCO_2 . Blood pH, pCO_2 , Na, K and Cl levels were obtained at the midpoint of the urine collection.

Renal Tubular Acidosis

Diagnostic criteria for characterizing the type of RTA are shown in *Table 1*. Proximal RTA was diagnosed if the urine pH was less than 5.5 in the presence of metabolic acidosis and the fractional excretion of bicarbonate was greater than 15%. Distal RTA was diagnosed if the urine pH was greater than 5.5 in the presence of metabolic acidosis and the fractional excretion of bicarbonate was less than 15%(6).

Patients with distal RTA underwent the furosemide test(7) and bicarbonate loading test(7,8) to identify the underlying defect. Furosemide was administered in a dose of 2 mg/kg intravenously followed by measurement of the urine pH at 30, 60 and 120 min(7). In the bicarbonate loading test, sodium bicarbonate was administered orally in a dose of 3-4 mEq/kg to increase the serum bicarbonate to 22-26 mEq/l. This was followed by calculation of the urine to blood pCO_2 gradient and the fractional excretion of bicarbonate.

TABLE I—Diagnostic Criteria for Types of RTA.

Diagnosis	Criteria
Proximal RTA	Urine pH <5.5 in the presence of metabolic acidosis; FEHCO ₃ >15%.
Distal RTA	Urine pH >5.5 in the presence of metabolic acidosis; FEHCO ₃ <15%.
(a) Secretory RTA	(U-B) pCO ₂ <10 mm Hg; normal or low serum potassium.
(b) Voltage dependent RTA	Hyperkalemia (serum K >5.5 mEq/l); urine pH >5.5 despite furosemide administration
(c) Backleak RTA	Hypokalemia (K <3.5 mEq/l); reduction in urine pH below 5.5 following furosemide; (U-B) pCO ₂ >20 mm Hg in alkaline urine

FEHCO₃ (fractional excretion of bicarbonate) $\frac{(\text{urine bicarbonate} \times \text{serum creatinine})}{(\text{serum bicarbonate} \times \text{urine creatinine})}$

(U-B) pCO₂ = urine to blood carbon dioxide tension gradient

Ammonium Chloride Loading Test(4)

Ten normal children convalescing from non-renal disorders served as controls. Ammonium chloride 0.1 g/kg body weight was administered orally for three days. Three to four hours after the last dose, blood and urine samples were obtained for pH and electrolytes.

Plasma and urine levels of Na, K and Cl were measured using ion selective electrodes (AVL 985-S, Radiometer, Copenhagen). Urinary net charge was determined by the formula: UNa + UK - UCl. Plasma pH, pCO₂ and bicarbonate were measured on the AVL-99S blood gas analyzer (Radiometer). Urine pH was measured using a pH paper. Statistical analysis was performed using Kruskal Wallis test.

Results

The blood and urine levels of pH and electrolytes in patients with diarrhea, distal RTA and proximal RTA are shown in Tables II & III, respectively.

Patients with Diarrhea

The mean age of these patients was 11.9

±7.1 months (range 6-24 months). Ten patients had severe dehydration and six moderate dehydration. The mean plasma pH was 7.17 ± 0.84 and bicarbonate 10.0 ± 3.2 mEq/l. Twelve patients had a urine pH of 5.5 or less while 4 showed urine pH of 6 despite metabolic acidosis. The mean urinary Na in the former group of patients was 55.8 ± 35.7 mEq/l (range 20.5 - 116.4 mEq/l) compared to 3.0 ± 1.8 mEq/l (range 1.2 - 5.4 mEq/l) in the latter. The urinary net charge was negative in all patients. The mean urinary net charge was -38.4 ± 28.3 mEq/l.

Patients with Distal RTA

The mean plasma pH was 7.27 ± 0.04 and bicarbonate 13.2 ± 2.7 mEq/l. The urinary pH was more than 6 in all patients and did not fall following furosemide administration. The urinary net charge was positive in all patients (mean 26.5 ± 14.9 mEq/l). The presence of high urine pH with no response to furosemide administration and urine to blood pCO₂ gradient of < 10 mm Hg suggested a diagnosis of secretory defect in these patients.

Patients with Proximal RTA

Of 7 patients, 2 had isolated proximal RTA and 5 had Fanconi syndrome. Proximal RTA was considered primary in 6 patients and secondary to Lowe syndrome in one. The mean plasma pH was 7.28 ± 0.03 and bicarbonate 14.2 ± 1.3 mEq/l. The urine pH was 5.5 or less in 6 patients; one with Lowe's syndrome however had a pH of 6.5. The urinary net charge was positive in all cases, the mean level being 23.1 ± 14.5 mEq/l.

Normal Subjects

After three days of ammonium chloride administration all normal subjects had urinary pH below 5.5. The urinary net charge was negative in all with a mean of -25.9 ± 19.5 mEq/l.

The levels of blood pH, bicarbonate and anion gap were comparable in patients of proximal and distal RTA (*Table II*). Patients with diarrhea, however, showed significantly lower blood pH and bicarbonate as compared to patients with proximal and distal RTA and normal controls ($p < 0.01$). The urinary pH of patients with distal RTA was significantly different from the pH of patients with diarrhea, proximal RTA and normal controls (*Table III*; $p < 0.001$). The urinary net charge in patients with proximal RTA and distal RTA was significantly different from that in patients with diarrhea and normal controls ($p < 0.001$). There was however, no significant difference in the urine net charge between patients with proximal and distal RTA.

Discussion

There is a significant negative correlation between the urine net charge and the ammonium excretion(3). In the present study patients with distal RTA showed a positive urine net charge. However, the urinary net charge was negative in those

with diarrhea and ammonium chloride induced acidosis. These findings are similar to those reported previously(4).

All patients with proximal RTA showed positive urine net charge. The metabolic acidosis in proximal RTA is chiefly due to proximal tubular bicarbonate wasting. The urinary ammonium excretion is reported to be increased in these patients(9-11). However, a recent study showed that the ammonium excretion was inappropriately low(12). The positive urine net charge amongst our patients with proximal RTA supports the hypothesis of reduced ammonium excretion. The precise reasons for reduced ammonium excretion in proximal RTA are not clear. However, it is suggested that the increased cytosolic pH in proximal tubules leads to decreased ammonia synthesis(13).

Urine pH is used as the initial test to differentiate various causes of hyperchloremic metabolic acidosis. Patients with diarrhea and proximal RTA show a low urine pH; the pH is inappropriately high in distal RTA. Patients with protracted diarrhea and chronic metabolic acidosis may, however, occasionally show a high urine pH despite increased urine ammonium excretion(14). This is considered secondary to a larger rise in ammonia availability than in hydrogen ion secretion(15).

Four of our patients with diarrhea showed a urine pH of 6 despite metabolic acidosis. All of them had low levels of urinary sodium, presumably secondary to its avid reabsorption in proximal tubules and the loop of Henle. Adequate delivery of sodium to the distal tubules is important for hydrogen ion secretion(16). Our findings of high urine pH in presence of low urinary sodium confirm those reported previously(4,16).

Measurement of urine pH after admin-

TABLE II—Blood pH and Electrolytes in Patients with Hyperchloremic Metabolic Acidosis

Group	pH	HCO ₃	Cl	Na	K
Diarrhea (n=16)	7.17±0.84 (7.03-7.32)	10.0±3.2 (5.9-16.7)	115.6±11.2 (96-140.3)	136.5±11.2 (120-160)	3.6±1.2 (1.6-5.7)
Distal RTA (n=11)	7.27±0.04 (7.21-7.33)	13.2±2.7 (10-18)	114.8±3.0 (109.8-118)	139.7±3.0 (135-145)	3.5±0.9 (2.2-4.9)
Proximal RTA (n=7)	7.28±0.03 (7.23-7.34)	14.2±1.3 (12.4-15.9)	113.9±4.8 (108.6-120)	139.4±6.1 (132-148)	3.9±0.8 (2.8-5.3)
Controls (n=10)	7.30±0.03 (7.22-7.33)	15.9±2.0 (12.5-17.6)	109.7±5.1 (102.4-116.4)	138.3±4.2 (132-144.5)	4.5±0.6 (3.7-5.6)

Values are expressed as mean±SD.
Figures in parenthesis indicate range.

TABLE III—Urine pH, Electrolytes and Anion gap in Patients with Hyperchloremic Metabolic Acidosis.

Group	pH	Cl	Na	K	AG
Diarrhea (n=16)	5.0(n=10) 5.5(n=2) 6.0(n=4)	92.4±37.9 (16.1-141.2)	46.6±38.6 (1.2-116.4)	11.4±11.9 (2.3-51.4)	-38.4±28.3 (-88.1-6.4)
Distal RTA (n=11)	6.5 (n=3) 7.0(n=8)	69.9±50.6 (22.4-156)	72.0±38.3 (29.3-149.5)	31.06±29.1 (2.4-88.8)	26.5±14.9 (4.6-54.4)
Proximal RTA (n=7)	5.0(n=2) 5.5(n=4) 6.5(n=1)	78.6±33.8 (36.3-134.5)	88.3±41.6 (35.8-157)	13.51±8.4 (5-28)	23.1±14.5 (1.9-46.4)
Controls (n=10)	5.0(n=7) 5.5(n=3)	73.8±14.6 (55.1-96.2)	37.5±12.2 (25-64)	10.3±5.1 (5.7-24)	-25.9±19.5 (-63.4- -8.3)

Values are expressed as mean±SD.
Figures in parenthesis indicate range.
AG-Anion gap.

istration of furosemide or sodium sulphate helps in characterization of the defect in distal RTA(7). These measures cause an increase in the delivery of sodium to the distal tubules, enhanced transepithelial voltage and increased secretion of hydrogen ions. Patients with secretory defect do not lower urine pH below 5.5 following administration of furosemide or sodium sulphate, unlike those with backleak or voltage dependent distal RTA(7). The presence of a persistently elevated urine pH with no response to furosemide administration, low

urine to blood CO₂ gradient and hypokalemia suggested the presence of secretory type of distal RTA in all our patients.

The differentiation of acute diarrhea from RTA, the two chief causes of hyperchloremic metabolic acidosis in children, is usually straightforward. Gastrointestinal symptoms are however frequent in RTA and occasionally distinction between gastrointestinal bicarbonate losses and defects of renal tubular acidification may be difficult. While urine pH is usually

proposed as the initial test to differentiate various causes of metabolic acidosis, the results can occasionally be fallacious. Patients with proximal RTA and symptomatic acidosis often show urine pH <5.5. On the other hand the urinary pH may occasionally be high in patients with acute diarrheal dehydration and chronic diarrhea.

This study confirms the importance of the measurement of urine net charge in the evaluation of patients with unexplained hyperchloremic metabolic acidosis. A positive urinary net charge, indicative of impaired urinary ammonium excretion, is seen in patients of both proximal and distal RTA. The finding of a negative urinary net charge suggests that renal acidification is not impaired, and should prompt a search for an extrarenal cause of bicarbonate loss.

Acknowledgement

This study was supported by a research grant from the All India Institute of Medical Sciences.

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