Tokai J Exp Clin Med., Vol. 28, No. 2, pp. 65-70, 2003

Urinary Citrate in Kidney Stone Disease

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(Received December 9, 2002; Accepted February 28, 2003)

Backgrounds: **Hypocitraturia, or low urinary citrate excretion is known as a risk for nephrolithiasis. Though urinary citrate excretion is basically determined by acid-base balance, metabolic acidosis is not always manifest in urinary stone patients with hypocitraturia. From our stone clinic data, we estimated the incidence of hypocitraturia and addressed its causes in the absence of obvious acid-base imbalance.**
Methods: **We selected 310 stone patients in whom 24-hour urine chemistry was examined on regular diets on 2 or more occasions during follow-up. Totally, 1361 specimens were analyzed in them.**
Results: **In the male subjects, the average urinary citrate excretion was 450.9 ± 284.4 mg/day, whereas in the female, 536.5 ± 305.9 mg/day ($p < 0.0001$). Eventually, hypocitraturia was found in 119 of the 310 patients (38.4 %). Of 222 with calcium stones, 70 (31.5 %) had hypocitraturia. In 32 of those, potential causes of hypocitraturia were identified, but in the rest, no apparent cause was found. In the latter, the net gastrointestinal alkali absorption was calculated from the 24-hour urine chemical data, and it was lower in those with hypocitraturia than in the normal control (9.2 vs. 34.4).**
Conclusion: **It was suggested that defective gastrointestinal alkali absorption may be involved in hypocitraturia of calcium stone patients.**

Key words : Nephrolithiasis, Hypocitraturia (low urinary citrate excretion), Gastrointestinal alkali absorption, Calcium stones, Stone clinic

INTRODUCTION

Hypocitraturia is recognized as an important cause of nephrolithiasis, particularly calcium stones, but its definition varies among investigators. Hodgkinson found a gender distinction in control subjects and defined hypocitraturia as urinary excretion less than 200 mg/day in men, less than 400 mg/day in women [1].

Urinary citrate combines free calcium ion, thereby forming soluble calcium citrate and resulting in reduction of urinary supersaturation of calcium oxalate and phosphate. Once calcium oxalate and phosphate crystals are formed in the urine, citrate inhibits both crystal growth and aggregation that are presumed to be a prerequisite for stone formation [2]. Moreover, monosodium urate crystallization known as another risk for formation of calcium oxalate stones by heterogeneous nucleation, is also inhibited by urinary citrate. All these properties of citrate are considered to help prevent urinary stone formation.

In clinical practice at the stone clinic of community hospitals in Japan, however, a diagnosis of hypocitraturia should not motivate all urologists to prescribe citrate supplement because its use to correct lower urinary citrate excretion is not yet generally accepted due to lack of sufficient data from reliable, controlled clinical trials and therefore, it is not fully covered by the national health insurance policy. So, in this study, we reviewed the long-term follow-up data of metabolic studies filed at our stone clinic to estimate the incidence of hypocitraturia in our pa-

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tient population and to clarify its potential causes which can be targeted by oral citrate supplementation in future for treatment and prevention of nephrolithiasis.

MATERIALS AND METHODS

Based on the database from April 5, 1983 to December 4, 2000 of the Stone Clinic of Tokai University Tokyo Hospital, 310 patients were selected in whom multiple determinations of the volume, pH and chemical constituents of 24-hour urine samples had been done on their regular diets and in periods with no medication for stone disease (Table 1). Examined urinary constituents were stone-related substances: The sodium, chloride, potassium, creatinine, urea nitrogen and uric acid were determined with the serum multiple analyzer. The calcium, phosphate and magnesium were with the atomic absorption analysis. The oxalate was measured with the chromatography method and citrate with the enzyme method. Metabolic disorders potentially responsible for urinary stone formation were diagnosed according to the widely accepted criteria used by urinary stone researchers (Table 2) [3].

After metabolic disorders or risk factors for recurrent disease were established, specific medical treatments were instituted in 138 patients with citrate, allopurinol, thiazide or thiopuronin, but in this study, all their data obtained during the medically treated period were excluded.

The follow-up method at our stone clinic is as follows, especially for recurrent stone patients. At the first visit after lithotripsy, a plain radiography is taken. Thereafter, if the patient is stone-free, but is still considered to need regular checkup because of a history of recurrent stones or a request to have metabolic study, he or she is instructed to collect a 24-hour urine sample on regular diets for determination of the stone-related urinary constituents. Renal tubular acidosis (RTA) is suspected when the urinary pH of each voiding specimen of the day showed a narrow range, fixed around 6.0, and not less than 5.5. The diagnosis is confirmed by providing oral ammonium chloride load (0.1 g/kg daily for 3 consecutive days) and is established when the urinary pH at the end of Day 3 is not lowered to 5.5 or less, even if other biochemical data, i.e. serum chloride and blood pH, are not in the criteria of metabolic acidosis [4].

In select patients with hypocitraturia in whom its cause had not been identified, we measured the net gastrointestinal (G-I) alkaliabsorption according to the following

Table 1 Patients and urine materials

Patients' gender (%)	
Male	246 (79)
Female	64 (21)
Mean age (years) ± S.D.	50.1 ± 12.5
Mean 24-hour urine samples examined per patient	4.3 ± 3.1
Total urine samples examined	1361

Table 2 Diagnostic criteria of various metabolic aberrations frequently encountered in stone clinic investigation

Hypercalciuria: urinary calcium: male $>$ 300 mg/day, female $>$ 250 mg/day
Hyperoxaluria: urinary oxalate $>$ 50 mg/day
Hyperuricosuria: urinary uric acid: male $>$ 800 mg/day, female $>$ 750 mg/day
Hypocitraturia: urinary citrate $<$ 320 mg/day

formula advocated by Oh *et al.*, [5] and compared with that obtained from a control group of 14 healthy volunteers: net G-I absorption of alkali = (Na + K + Ca + Mg) − (Cl + 1.8 P), all the molecules excreted in 24-hour urine samples in mEq, except for P in mmole.

RESULTS

A total of 1361 samples of 24-hour urine collection were examined in 310 subjects. The average daily urinary citrate was 450.9 ± 284.4 mg in the male and 536.5 ± 305.9 mg in the female ($p < 0.0001$). Hypocitraturia was found in 119 (38.4 %) patients. Of 310 patients, 122 had recurrent stones and had the average citrate excretion of 452.7 ± 208.7 mg, while 188 with the first stone episode had almost the same citrate level of 467.0 ± 222.5 mg as those with recurrent episodes. In uric acid stone patients, the urinary citrate level was significantly reduced as compared with calcium stone patients (Table 3). In 222 calcium stone patients, hypocitraturia was seen in 84 (37.8 %). However, there was no correlation between the urinary citrate and other pertinent urinary constituents (Table 4).

We compared the hypocitraturia group with the normal urinary citrate group matched as to the age and the number of 24-hour urine specimens examined (Table 5).

Excretion of urea nitrogen and potassium was significantly reduced in the hypocitraturia group. Of 222 calcium stone patients, 70 (31.5 %) had hypocitraturia and in this group, we investigated causes of hypocitraturia. In 32, various potential causes were identified, while in 38, no apparent cause was revealed (Table 6). In 36 of the 38 patients with

Table 3 Urinary Citrate Excretion in Patients with Various Stone Compositions

Stone analysis	Mean ± SD	No. cases
Calcium oxalate	469 ± 313 (mg)	110
Calcium oxalate/phosphate	434 ± 253	93
Calcium oxalate/urate	501 ± 224	9
Calcium phosphate	408 ± 240	10
Uric acid	393 ± 275	21
Cystine	590 ± 326	3
Struvite	587 ± 228	2
Unknown	528 ± 306	62

Table 4 Urinary Citrate and Other Constituents Excretion in Calcium Stone Patients

Urine constituents	Mean ± SD	Correlation coefficient	P value
Citrate (mg)	453 ± 285		
Urea nitrogen (mg)	9792 ± 2984	− 0.082	0.273
Creatinine (mg)	1276 ± 427	− 0.047	0.529
Uric acid (mg)	659 ± 200	− 0.004	0.959
Sodium (meq)	223 ± 81	− 0.019	0.800
Chloride (meq)	225 ± 80	− 0.110	0.137
Potassium (meq)	52 ± 20	− 0.075	0.313
Calcium (mg)	239 ± 104	0.044	0.551
Magnesium (mg)	94 ± 38	0.390	0.602
Phosphate (mg)	826 ± 276	− 0.032	0.664
Oxalate (mg)	47 ± 23	− 0.018	0.805
Volume (ml)	2018 ± 652	0.164	0.026
pH	5.7 ± 0.8	− 0.040	0.594

Table 5 24-hour urine examination in subjects with low and normal citrate excretion

	Subjects with low urinary citrate	Subjects with normal urinary citrate	P value
No. male subjects	61	123	
No. female	9	29	
Age	46.5 ± 11.7	51.0 ± 12.3	0.0110
No. examination	4.4 ± 3.3	4.4 ± 3.1	
Citrate (mg)	242 ± 64	553 ± 190	< 0.0001
Volume (ml)	1903 ± 562	1970 ± 494	0.368
pH	5.7 ± 0.8	5.7 ± 0.8	
Urea nitrogen (mg)	8353 ± 2079	10402 ± 2423	< 0.0001
Creatinine	1173 ± 332	1282 ± 318	0.0205
Uric acid	602 ± 163	667 ± 149	0.0056
Sodium	197 ± 60	229 ± 63	0.0060
Potassium	41 ± 12	54 ± 15	< 0.001
Calcium	212 ± 75	246 ± 92	0.0700
Magnesium	82 ± 28	95 ± 32	0.0052
Phosphate	726 ± 219	853 ± 230	0.0010
Oxalate	42 ± 18	49 ± 17	0.0199

Table 6 Causes of Hypocitraturia Identified in 70 Calcium Stone Patients

Renal Tubular Acidosis	13
Medullary Sponge Kidney	6
Urinary Tract Infection	6
Hyperuricemia	3
Chronic diarrhea	2
Hyperaldosteronism	1
Unknown	39
Total	70

Table 7 Gastrointestinal Alkali Absorption in Subjects with Hypocitraturia of Unknown Etiology

		Hypocitraturia	Control	p values
Urinary citrate (mg/day)		270 ± 50	499 ± 124	0.0001
No. patients	Male	34	12	
	Female	2	2	
Age		47 ± 12	49 ± 17	
G-I alkali absorption		9.2 ± 2.5	34.4 ± 9.0	0.0001

hypocitraturia of unknown etiology, the net gastrointestinal alkali absorption calculated according to the Oh's formula was significantly lowered to 9.2 on average, whereas in the normal control group, it was 34.4 (Table 7).

DISCUSSION

A significant proportion of patients with nephrolithiasis have a low urinary citrate excretion in the absence of a known cause such as renal tubular acidosis, chronic bowel disease complicated by chronic diarrhea and in-

testinal alkali loss or hypokalemia inducing intracellular acidosis. The most important determinant of renal tubular reabsorption of citate is acid-base balance. Systemic acidosis increases citrate reabsorption from the renal tubules because of an increased demand of the body (resulting in a lower urinary citrate excretion), and conversely, alkalosis or alkali-loading from the GI tract decreases citrate reabsorption (thus increasing urinary citrate excretion) [6, 7].

Urinary citrate excretion is reported to be correlated to the urinary volume, calcium, magnesium excretion and GI-alkali load. Hess *et al.* also reported that with respect to urinary citrate, normal citrate excretion is associated with an increased excretion of urea nitrogen, phosphate, calcium, uric acid, sodium and magnesium, whereas low urinary citrate tends to be associated with a decreased excretion of those substances [6]. In this study, the characteristics of urinary citrate excretion were compatible with the findings of Hess *et al.* (Table 5). When limited to calcium stone patients, the urinary citrate excretion seemed to be independent of the excretion of other stone-related substances (Table 5). However, with regard to the correlation between urinary citrate to urinary urea in the 2 groups, it appears inconsistent that the urinary urea level was significantly higher in the normal urinary citrate group than in the low group, taking into consideration that the urinary urea excretion should be proportional to protein consumption of a subject. Theoretically, a high intake of protein and consequent high amino acid-load may enhance acidosis tendency and promote compensatory renal tubular absorption of citrate, resulting in low urinary citrate. On the other hand, as Hess *et al.* reported, most patients with high urinary calcium excrete more urea, uric acid and sulfate, as compared to ones with normal calcium excretion [6]. Thus, it may follow that the normal citrate group in our study included some hypercalciuric patients with a concurrent high urinary urea and dietary-induced high citrate excretion that are frequently seen in those patients, because the mean urinary calcium rate was also higher in the normal citrate group than in the low citrate group.

In this study, potential causes of hypocitraturia were identified only in 32 of the examined patients with calcium stones (32/70, 45.7 %, Table 6). Among them, 13 had renal tubular acidosis (RTA). A clinical diagnosis of RTA was based on the fasting morning urinary pH and oral ammonium chloride tests. If we had suspected RTA more frequently, especially in stone-clinic patients with persistent alkaline urine, its incidence might have been increased because the pertinent diagnostic tests could be done on an outpatient basis. In 6 of the 13 patients with RTA, medullary sponge kidney was diagnosed concomitantly. Medullary sponge kidney is characteristically demonstrated as a structure of bundles of linear striation or blush-like patterns localized in the medullary portion of minor calyces on intravenous pyelography [8]. This condition is frequently associated with hypercalciuria, acidification defects and consequent hypocitraturia, and urinary stasis in the ectatic, cystic lesions of collecting tubules, all of which will enhance urinary stone formation. While nephrolithiasis or nephrocalcinosis is a common complication of RTA and/or medullary sponge kidney, those stones are small, multiple and commonly localized in the collecting ducts, but not in the calyces so that it is not easy to treat them efficiently with extracorporeal shock-wave lithotripsy (ESWL) [9]. In these cases, we believe that oral citrate medication will be an optimal treatment when they have significant hypocitraturia.

We failed to identify clear causes in 38 subjects of the low urinary citrate group. The hypocitraturia of indeterminate causes or idiopathic hypocitraturia may be secondary to intrinsic renal defects (dysfunction of the sodium-citrate co-transport or disordered intracellular citrate regulation etc.), inappropriate intestinal citrate or alkali absorption, or a normal physiologic response to animal protein-rich diets. In this study, 36 had a significantly low gastrointestinal (GI) alkali absorption compared with the control (Table 7, $p = 0.0001$). In them, a decrease in gastrointestinal absorption of citrate might have contributed to their hypocitraturia. Cowley *et al.* suggested that impaired GI absorption of citrate or alkali was not uncommon in hypocitraturic patients with recurrent calcium stone disease [10]. However, they did not mention which might play a major role, inappropriate GI absorption of alkali or a low intake of alkaline substances. Sakhee *et al.* stated that the net GI alkali absorption

was a factor in urinary citrate excretion similarly in the normal subjects as well as in the stone-forming patients without RTA [11]. They suggested that reduced citrate excretion was largely dietary in origin as a result of low net alkali absorption from a probable relative deficiency of vegetables and fruits or a relative excess of animal proteins. In our studies, 24-hour urine specimens were collected under their regular diets. Although we had no detailed information about their dietary components, when considering that average Japanese diets are rich in alkaline foods, the low GI alkaline absorption seen in the hypocitraturic subjects in our study was unlikely to be of dietary origin. More recently, He Y *et al.* reported that the Na +/ dicarboxylate co-transporter 1 (hNaDC1), one of sodium-citrate co-transporters, was expressed highly in the kidneys of stone formers with hypocitraturia, and concluded that the upregulation of hNaDC1 mRNA induced by intake of protein-rich foods may be an important cause of hypocitraturia [12].

The efficacy of citrate medication in recurrent stone patients with idiopathic hypocitraturia are being studied at our stone clinic.

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