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Relative effect of urinary calcium and oxalate on saturation of calcium oxalate
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Relative effect of urinary calcium and oxalate on saturation of calcium oxalate.

Background. The study compared the effect of urinary calcium with that of oxalate on urinary saturation [relative saturation ratio (RSR)] of calcium oxalate.

Methods. A retrospective data analysis was conducted on urinary stone risk analysis from 667 patients with predominantly calcium oxalate stones. Urinary RSR of calcium oxalate was individually calculated using Equil 2. A "theoretical" curve of the relationship between urinary RSR of calcium oxalate and concentration of calcium or oxalate was obtained at two stability constants for calcium oxalate complex, while varying calcium or oxalate and using group mean values for urinary constituents.

Results. At the stability constant of 7.07×10^3, the increase in RSR of calcium oxalate was less marked with calcium than with oxalate. However, at the stability constant of 2.746×10^3 from the Equil 2 that is considered the "gold standard," calcium and oxalate were equally effective in increasing RSR of calcium oxalate. The above theoretical curves (relating RSR with calcium or oxalate) were closely approximated by the actual curves constructed with data from individual urine samples. Urinary saturation of calcium oxalate was equally dependent on urinary concentrations of calcium and oxalate ($r = 0.75$ unadjusted and 0.57 adjusted for variables, and $P < 0.0001$ for calcium; $r = 0.73$ unadjusted and 0.60 adjusted, $P < 0.0001$ for oxalate).

Conclusion. Among calcium oxalate stone-formers, urinary calcium is equally effective as urinary oxalate in increasing RSR of calcium oxalate.

A topic of considerable interest and continuing debate in nephrolithiasis field concerns the relative importance of urinary calcium and oxalate in the formation of calcium oxalate stones. The basis of this controversy is the so-called "calcium oxalate interaction," involving interaction between calcium and oxalate in the urine [1]. In the urinary environment, calcium forms a soluble complex with oxalate, reducing both the ionized calcium and oxalate [2]. Since the amount of oxalate (in milliequivalent amounts) in urine is normally much less than that of calcium, it has been suggested that a proportionately much greater decline in ionized oxalate would occur than in ionized calcium as a result of calcium oxalate complex formation. Thus, according to this scheme, a rise in calcium concentration would be offset by a decline in ionized oxalate concentration, blunting the increase in urinary saturation of calcium oxalate. Conversely, a rise in oxalate concentration would cause a proportionally smaller decline in ionized calcium concentration, causing a more prominent increase in urinary saturation of calcium oxalate.

Thus, in 1972, Nordin, Peacock, and Wilkinson [2] reported that the rise in urinary calcium concentration was less effective in increasing urinary saturation of calcium oxalate, compared with urinary oxalate concentration. Even at high calcium concentration, the saturation of calcium oxalate did not exceed the theoretic formation product (the limit of metastability above which precipitation of calcium oxalate takes place), whereas the formation product was exceeded at high oxalate concentration. This concept, invoking a greater pathogenetic role of urinary oxalate over calcium, has persisted through today [3–5].

It should be apparent, however, that the degree of calcium oxalate interaction in urine depends on the stability constant for the formation of soluble calcium oxalate complex used in the calculation of calcium oxalate saturation. Higher the stability constant, a more soluble complex would form and blunt the effect of calcium on calcium oxalate saturation. In 1976, Robertson et al [6] acknowledged that the stability constant employed by them in 1972 [2] had been too high, and changed it to a much lower value (from 7.07×10^3 to 1.9×10^3). However, they had not reexamined the effect of calcium or oxalate concentration on the saturation of calcium oxalate.

Key words: calcium oxalate saturation, urinary calcium, oxalate.

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The stability constant is now acknowledged to be 2.746×10^3, as first reported by Finlayson, Roth, and DuBois [7]; this is the value used in the Equil 2 program [8], considered the gold standard for calculating urinary saturation of stone-forming salts. In 1982, we used the two stability constants to calculate saturation of calcium oxalate in a synthetic solution simulating urinary environment [9]. We found that the ability of calcium concentration to increase the saturation of calcium oxalate was much more marked when the lower stability constant of Finlayson, Roth, and DuBois [7] was used instead of the value originally used by Nordin, Peacock, and Wilkinson [2].

In this report, we show that the conclusions from aforementioned analysis done in a synthetic medium [9] apply to whole urine collected from human beings suffering from stones. Data were derived from our kidney stone registry [10], comprised of 667 patients with predominantly calcium oxalate stones in whom detailed measurements of urinary constituents were available for the calculation of urinary saturation of calcium oxalate by Equil 2 program [8].

METHODS

Patient data

A total of 2209 patients with stones were included in the stone registry, whereby their data obtained during baseline ambulatory evaluation were entered onto the computer database. Patients who satisfied the following criteria were selected: (*1*) stone retrieved within 6 months of evaluation showing on analysis >70% calcium oxalate in composition; (*2*) one or two 24-hour urine samples collected during a random diet analyzed for a full set of stone risk factors [11], including calcium, oxalate, citrate, magnesium, phosphorus, uric acid, sodium, potassium, pH and total volume; (*3*) relative saturation ratio (RSR) of calcium oxalate calculated by the Equil 2 program [8]; (*4*) body weight and age; (*5*) stone formation rate or the number of stones formed (passed or removed) during 3 years preceding baseline evaluation, determined from history, review of patient data, and report of referring physician for stone passage, procedures for removal of stones and appearance on x-ray films; (*6*) number of stone surgeries or procedures (extracorporeal shock wave lithotripsy, endoscopic and ureteroscopic approaches, basket removal, and open surgery) for the removal of stones performed during preceding 3 years; and (*7*) adult patients ≥ 20 years old of either gender. None of the patients took drugs for the control of stone formation, calcium supplements, or pharmacologic doses of vitamin D preparations for at least 2 weeks prior to baseline evaluation. Patients with primary hyperparathyroidism, distal renal tubular acidosis, enteric or primary hyperoxaluria, infection of the urinary tract with urea-splitting organisms, or cystinuria were excluded. Some patients evaluated in the 1970s were not included in this report, since they did not have analysis of urinary uric acid or citrate. Those without accurate stone analysis were not considered here. There were a total of 667 patients (197 women and 470 men) who satisfied all of the above criteria; they comprised the data set for this report.

Table 1. Baseline presentation

	Mean ± SD	Median (range)
Age *years*	43.3 ± 11.9	42 (21–78)
Weight *kg*	79.7 ± 18.8	78.2 (38.2–204.5)
Urinary calcium *mg/L*	159 ± 86	147 (9–677)
Urinary oxalate *mg/L*	21.1 ± 10.5	19.3 (2.0–121.1)
Urinary citrate *mg/L*	329 ± 223	292 (4–2550)
Urinary magnesium *mg/L*	63.3 ± 30.4	57.9 (5.9–274.0)
Urinary phosphorus *mg/L*	605 ± 255	568 (136–1890)
Urinary uric acid *mg/L*	378 ± 154	364 (77–1502)
Urinary sodium *mEq/L*	110 ± 46	107 (13–245)
Urinary potassium *mEq/L*	32.2 ± 13.5	30.2 (5.7–83.2)
Urinary pH	6.00 ± 0.37	6.01 (4.98–7.25)
Urinary total volume *mL/day*	1761 ± 805	1593 (288–6905)
Relative saturation ratio, calcium oxalate	8.91 ± 4.41	8.21 (0.57–37.4)
Relative saturation ratio, Brushite	2.59 ± 1.86	2.19 (0.06–12.09)

For all parameters, the number of subjects evaluated was 667. To convert mg to mmol, divide by 40 for calcium, 88 for oxalate, 189 for citrate, 24.32 for magnesium, 31 for phosphorus, and 168 for uric acid.

Urinary stone risk factors were analyzed by previously described methods [11]. RSR represented the ratio of activity product and the thermodynamic solubility product of calcium oxalate. A value of 1 represented saturation, >1 supersaturation, and <1 undersaturation [10]. Among those with two random urine collections, the mean values were utilized in this analysis.

Statistical methods

Objective of this retrospective data analysis was to seek factors that determine urinary RSR of calcium oxalate. Spearman correlation coefficients were determined in order to evaluate the association between RSR of calcium oxalate and urinary concentrations of calcium, oxalate, citrate, magnesium, phosphorus, uric acid, sodium, potassium, and urinary pH, total volume, age, and weight. Adjusted Spearman correlation coefficients were obtained after correcting for above urinary analytes. The factor being tested for association with RSR calcium oxalate was omitted as a partial variable.

RESULTS

Baseline presentation

Baseline data are presented in Table 1. Urinary calcium concentration varied widely (9 to 677 mg/L) as did urinary oxalate concentration (2.0 to 121.1 mg/L). Daily urinary calcium excretion was high at 280 mg/day (normal

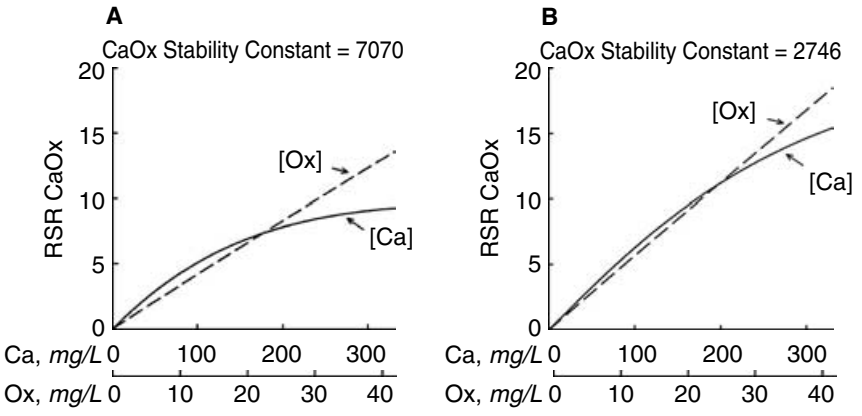

Fig. 1. Theoretic relationship between saturation of calcium oxalate [relative saturation ratio (RSR CaOx)] and concentration of calcium (Ca) or oxalate (Ox). RSR of calcium oxalate was calculated from mean values for urinary constituents derived from 667 patients with predominantly calcium oxalate stones, while calcium and oxalate were mathematically adjusted. The stability constant of calcium oxalate complex was assumed to be 7.07×10^3 (*A*), whereas it was considered to be 2.746×10^3 (*B*). For convenience, the same value was used as the thermodynamic solubility product in calculating RSR (A) and (B). To convert mg to mmol, divide by 40 for calcium, and by 88 for oxalate.

<250 mg/day on a random diet in our laboratory [11]), and urinary citrate was within normal limits at 579 mg/day (mean normal 640 mg/day). Mean values for other urinary risk factors were within normal limits.

"Theoretic" relationship between RSR of calcium oxalate and calcium or oxalate concentration using different stability constants

By using the mean values for stone risk factors from 667 patients shown in Table 1, RSR of calcium oxalate was calculated using Equil 2 while using stability constant for calcium oxalate complex of 7.07×10^3 (from Nordin, Peacock, and Wilkinson [2]) or 2.746×10^3 (from Finlayson, Roth, and DuBois [7]). For each stability constant, calcium concentration was varied from 0 to mean + 2 SD, while oxalate concentration was kept constant at the group mean. Conversely, oxalate concentration was varied from 0 to mean + 2 SD, while calcium concentration was kept constant at the mean value.

When the higher stability constant was used, RSR of calcium oxalate progressively rose with increasing oxalate concentration, whereas it reached a plateau at high calcium concentrations at a level much lower than that of corresponding oxalate concentrations (Fig. 1A). In contrast, when the lower stability constant [7] was used as in Equil 2 [8], the rise in RSR with increasing oxalate concentration was similar to that of increasing calcium concentration (Fig. 2B). The two curves departed only at high concentrations, with RSR from oxalate being about 15% higher than RSR from calcium, owing to the appearance of a plateau in the calcium curve. We next examined how this theoretic relationship compares with the actual data.

"Actual" relationship between urinary RSR of calcium oxalate and urinary calcium or oxalate concentration

Individual RSR of calcium oxalate calculated by Equil 2 (with stability constant of 2.746×10^3) from all 667 patients were utilized to examine the determinants of RSR of calcium oxalate. Without adjustment for variables, RSR calcium oxalate was highly and significantly correlated with urinary concentration of calcium as well as that of oxalate ($r = 0.75$ for calcium and 0.73 for oxalate, $P < 0.0001$). RSR calcium oxalate was significantly and positively correlated with urinary concentration of citrate, magnesium, phosphorus, uric acid, sodium, and potassium, though less robust ($r < 0.6$) than with urinary concentrations of calcium and oxalate. It was significantly and inversely correlated with urinary pH and total volume ($r < -0.5$).

After adjustment for variables, the significant correlations between RSR calcium oxalate versus calcium and oxalate concentrations remained ($r = 0.57$ for calcium and 0.60 for oxalate, $P < 0.0001$). Though not robust ($r = -0.2$ to -0.4), there was a significant negative correlation between RSR calcium oxalate versus citrate, magnesium, phosphorus, and sodium.

In Figure 2A, mean RSR of calcium oxalate was calculated for each 25 mg/L interval of urinary calcium concentration over the range of 0% to 96% of values from 667 patients. Actual values closely resembled the theoretical curve obtained by using the lower stability constant (from Fig. 1B). In Figure 2B, mean RSR of calcium oxalate was calculated for each 5 mg/L interval of urinary oxalate concentration over the range of 0% to 96% of values from 667 patients. The actually derived values approximated the theoretical curve (Fig. 1B).

When drawn to the same scale of 0% to 96% range of urinary calcium and oxalate concentrations from 667 patients, a plot of RSR of calcium oxalate against urinary calcium concentration (in 25 mg/L intervals) was virtually indistinguishable from the plot of RSR against urinary oxalate concentration (in 5 mg/L intervals) (Fig. 3).

DISCUSSION

This retrospective data analysis was undertaken in order to critically appraise the long-held view that urinary

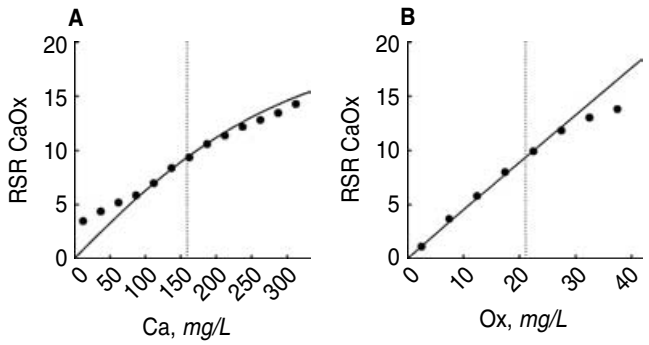

Fig. 2. Actual relationship between saturation of calcium oxalate and calcium or oxalate concentration. Relative saturation ratio (RSR) of calcium oxalate was individually, and the mean value derived for each 25 mg/L interval (or 0.625 mmol/L) of urinary calcium concentration and each 5 mg/L (0.057 mmol/L) interval of urinary oxalate concentration over a range of 0% to 96% from 667 patients. Each dot represents group mean at each interval. The curves from Figure 1B are drawn to show approximation of actual with theoretic data. (*A*) The relationship between RSR of calcium oxalate and calcium concentration. (*B*) The relationship of RSR with oxalate concentration.

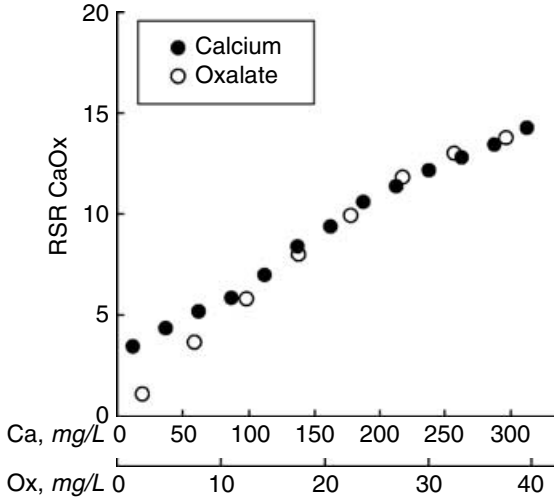

Fig. 3. Approximation of the actual relationship between relative saturation ratio (RSR) of calcium oxalate and calcium concentration by the actual relationship between RSR and oxalate concentration. Group mean RSRs of calcium oxalate for each 25 mg/L intervals of calcium concentration from 667 patients are shown (•), and group mean RSRs for each 5 mg/L intervals of oxalate are depicted (○).

oxalate is more important than urinary calcium owing to the formation of soluble calcium oxalate complex [2–5]. In urine samples obtained from 667 patients with predominantly calcium oxalate stones (>70% on stone composition) on a random diet, urinary saturation of calcium oxalate was calculated using Equil 2 program [8]. We found that urinary calcium concentration was equally effective as urinary oxalate concentration in increasing urinary saturation of calcium oxalate.

The persisting belief that urinary saturation of calcium oxalate is much more dependent on urinary oxalate than on calcium was derived by using stability constant of 7.07×10^3 that probably exaggerated the formation of calcium oxalate complex [2]. The same authors of that report [2] later conceded that the constant used earlier was too high [6], and suggested a value of 1.9×10^3. The correctness of the latter value was espoused from the close agreement between calculated and measured values of ionized calcium concentration [6]. In this study, we employed the widely accepted stability constant of 2.746×10^3, a value utilized in Equil 2, a computer program considered to be the gold standard for estimating urinary saturation of stone forming salts [8]. This constant was derived experimentally by Finlayson, Roth, and DuBois [7] from solubility studies conducted at body temperature. A similar value was obtained experimentally by another renowned group in physical solution chemistry [12].

From theoretic grounds, the formation of calcium oxalate complex should lower the ionized concentration of calcium and oxalate, and thereby reduce the saturation of calcium oxalate. Since urinary calcium is proportionately much higher than urinary oxalate, the fractional decline in ionized oxalate should be greater than that of ionized calcium. Thus, a rise in urinary calcium concentration is expected to produce a less prominent increase in urinary saturation of calcium oxalate than a rise in urinary oxalate concentration. The use of a high stability constant would exaggerate complex formation, cause a proportionately greater fractional decline in ionized oxalate, and further blunt the effect of calcium on urinary saturation of calcium oxalate. Thus, our failure to show a more prominent effect of oxalate over calcium on saturation of calcium oxalate might have resulted from the use of lower stability constant.

In this study, we tested the above hypothesis incorporating the following unique features. First, we utilized stone risk factors from 24-hour random urine samples, collected from 667 patients with predominantly calcium oxalate stones (>70% on stone analysis). Thus, the data were derived in actual urine samples rather than synthetic medium [9]. The use of mean values for urinary analytes from a large number of patients with idiopathic calcium oxalate nephrolithiasis made the analysis clinically more relevant.

Second, we constructed theoretic curves depicting dependence of saturation of calcium oxalate on calcium or oxalate concentration, by taking the mean values for urinary constituents derived from 667 patients, while arbitrarily varying calcium or oxalate concentration. With Equil 2 program, we calculated the saturation of calcium oxalate using stability constant of calcium oxalate complex of 7.07×10^3 as in the original report by Nordin, Peacock, and Wilkinson [2], and a value of 2.746×10^3 introduced by Finlayson, Roth, and DuBois [7] and contained in Equil 2. When the higher stability constant was used, we found qualitatively the same result as reported

by Nordin, Peacock, and Wilkinson [2]. A rise in calcium concentration produced a much less prominent increase in the saturation of calcium oxalate, compared with a rise in urinary oxalate. However, when a more accurate stability constant of Finlayson, Roth, and DuBois [7] was employed, urinary calcium was equally effective as urinary oxalate in increasing the urinary saturation of calcium oxalate, confirming a similar analysis performed earlier in a synthetic medium [9].

Third, we compared the theoretic curves described above, with the *actual* curves obtained from individual values obtained from 667 patients with predominantly calcium oxalate stones. In constructing the actual curves, mean RSR of calcium oxalate was calculated for each 25 mg/L interval of calcium and for each 5 mg/L interval of oxalate. Each interval therefore represented a composite of varying urinary composition from different patients with stones. In contrast, the theoretic curves were constructed while other urinary analytes were kept the same as the mean values for the whole group of patients with stones. Thus, the outcome from the actual curves could not have been predicted from the theoretic curves just because the same stability constant had been used. The theoretic and actual curves closely approximated each other. Moreover, the actual curves for calcium and oxalate, derived at varying intervals of calcium and oxalate from urine samples of a large group of well-characterized patients with calcium oxalate stones, were nearly identical.

Fourth and finally, in comparing calcium and oxalate curves, the upper limits for calcium and oxalate concentrations were not arbitrarily set, but represented experimentally derived 96 percentile value from our patients with stones. Thus, we were able to examine the effect of calcium and oxalate at urinary concentration ranges occurring in most patients with idiopathic calcium oxalate nephrolithiasis. In some reports [13], the upper limit of calcium was set at an unusually high value at which a flattening of calcium curves becomes more prominent.

We therefore believe that the prior reports [3] showing a greater dependence of the saturation of calcium oxalate on oxalate than on calcium concentration may have exaggerated the degree of calcium oxalate complexation. Using the state-of-the-art Equil 2 computer program [8] that utilized a lower value for the stability constant of calcium oxalate [7], the degree of increase in the saturation of calcium oxalate with rising calcium concentration was found to be equivalent to that obtained by a rise in oxalate, and more marked than in the earlier study [2] (that had used a higher value for the stability constant).

Our findings are not at variance with an early review by Finlayson [14] in which a much more prominent role of oxalate was claimed. Finlayson's view was based on a study by Marshall et al [15], who had examined diurnal variation in urinary composition and saturation of calcium oxalate calculated by using the higher stability constant for calcium oxalate. In recalculating the same data with the use of Equil 2 program, we found that the substitution of urinary oxalate from stone-formers into the urinary composition of normal subjects increased RSR of calcium oxalate by 28% (unpublished data of authors). Substitution of calcium resulted in a similar increase of 26%.

We are not casting doubt on the importance of urinary oxalate in stone formation. Rather, we are suggesting that urinary calcium exerts a similar effect as oxalate within the 4% to 96% range of calcium and oxalate concentrations encountered among patients with idiopathic calcium oxalate nephrolithiasis.

Nor are we disparaging the importance of oxalate in the formation of the stone nidus, suggested by many studies in animal models and cell systems. However, some of these studies employed high oxalate loads or exposure, creating a situation analogous to enteric hyperoxaluria in human beings, rather than idiopathic calcium oxalate nephrolithiasis, the target of our study. Relevant here is the recent work by Evan et al [16], who had obtained papillary biopsies in human beings intraoperatively. Among patients with idiopathic calcium oxalate nephrolithiasis, the nidus began as hydroxyapatite deposition in the basement membrane of the thin loop of Henle. The plaque formation was correlated with hypercalciuria, not with urinary oxalate. Among patients with enteric hyperoxaluria, the nidus began as hydroxyapatite crystals deposited within the lumen of distal collecting ducts.

CONCLUSION

Urinary calcium exerts a similar effect as urinary oxalate in increasing urinary saturation of calcium oxalate among patients with predominantly calcium oxalate stones. This assessment is based on a reasonably safe assumption that the stability constant for calcium oxalate complex utilized in the Equil 2 program is accurate.

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