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# The relation between orthophosphate and pyrophosphate in normal subjects and in patients with urolithiasis

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**Summary.** In calcium lithiasis, inhibitors have a significant effect in reducing the crystallization process. This work evaluated orthophosphate in a group of patients with calcium oxalate lithiasis, and in a control group. The study of orthophosphate and pyrophosphate, showed differences between stone formers and the control group. These results could be attributed to a failure in the renal transformation of orthophosphate into pyrophosphate.

**Key words:** Orthophosphate and pyrophosphate – Calcium oxalate urolithiasis

## Introduction

Pyrophosphate is a possible inhibitor in calcium oxalate lithiasis. Pyrophosphate (PPi) is produced intracellularly by a large number of reactions, the most typical involving the removal of PPi from a nucleotide triphosphate [6]. The PPi produced from these reactions is hydrolysed within the cell to orthophosphate by intracellular pyrophosphateses. The major route of removal is by hydrolysis, less than 5% being excreted in the urine. Ingestion of orthophosphate increases the urinary excretion of PPi [2, 13], although it is probable that little PPi is absorbed from the diet. The fact that orthophosphate increases PPi excretion suggests a renal mechanism which results in the rapid clearance of PPi without a change in the blood concentration [14]. According to several authors [11, 15] the success of orthophosphate in preventing renal stone disease may be due to its tendency to increase urinary PPi.

Therefore the relationship of orthophosphate and pyrophosphate both in normal and in stone former's urine was studied.

## Material and methods

The subjects consisted of 24 men and women with no past history of urinary stone disease and 38 who had suffered one or more episodes of urolithiasis. In all cases the stones were composed of pure calcium oxalate alone or in combination with calcium phosphate.

All subjects were submitted to metabolic evaluation including, measurement of calcium, oxalate, phosphate and pyrophosphate. All subjects had normal renal function, none had detectable proteinuria or urinary tract infection. The urolithiasis group were divided into stone formers without metabolic alteration (A), and with metabolic alteration (B) (hyperoxaluria and/or hypercalciuria).

Calcium, phosphate and oxalate were evaluated by conventional methods and the pyrophosphate assay was performed by a method previously described [4]. The statistical study was done with Student's test, correlation coefficient and linear regression.

## Results

This paper reports the results of orthophosphate determination and their relationship with pyrophosphate; the results of PPi were reported previously [4].

The orthophosphate concentration, mean values with standard deviations, both for renal stone forming and normal subjects are shown in Table 1. There were no significant differences in orthophosphate concentration between normal subjects and stone formers (with or without metabolic alterations), but there were differences between stone formers with metabolic alteration and stone formers without metabolic alteration ($P < 0.01$).

We therefore considered the relationship between PPi and orthophosphate. If a renal mechanism transformed orthophosphate into pyrophosphate this would be significant in that PPi is considered as a inhibitor of calcium stone formation. Thus, Fig. 1 shows the representation of orthophosphate vs pyrophosphate as a mean ± standard deviation in the three groups considered. Figure 2, gives the relationship between these parameters from the experimental results, which can be adjusted to a straight line.

**Table 1.** Diuresis, orthophosphate and pyrophosphate concentration in urine of 24 h in healthy subjects and stone formers

| | Mean values | | | |
|---|---|---|---|---|
| | *N* | Diuresis | Orthophosphate (as phosphorous) (mg/100 ml) | PPi (µmol/l) |
| Healthy subjects (C) | 24 | 1,278 ± 553<br>1,278 ± 113 | 61,28 ± 20,67<br>61,28 ± 4,22 | 17,56 ± 11,98<br>17,56 ± 2,44 |
| Stone formers without alteration (A) | 20 | 1,674 ± 615<br>1,674 ± 137 | 61,57 ± 19,23<br>61,57 ± 4,30 | 11,23 ± 11,97<br>11,23 ± 2,67 |
| Stone formers with alteration (B) | 15 | 2,137 ± 866<br>2,137 ± 224 | 55,23 ± 20,58<br>55,23 ± 5,31 | 10,82 ± 6,85<br>10,82 ± 1,76 |

Values are expressed as the mean ± standard deviation and the mean ± s.e.m.

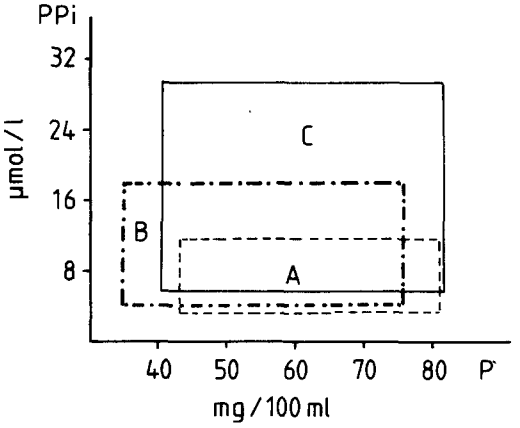


**Fig. 1.** Representation of the Pyrophosphate vs Orthophosphate (as phosphorous) as the mean ± standard deviation. A = Stone formers without metabolic alteration. B = Stone formers with metabolic alteration. C = Healthy subjects

## Discussion

Data from several authors [1, 9, 10] showed that urine from stone formers inhibits heterogeneous nucleation less than normal urine, Bauman et al. [1] attributed this to an increase in calcium phosphate nucleation caused by a decrease in PPi excretion and Pak and Galosy [10] suggested increases in both the nucleation of calcium phosphate and calcium oxalate in stone formers. PPi was proposed as an inhibitor of calcium stone formation which prevented crystallisation by adsorption to active growth sites on the seed crystal surface in trace concentrations [3, 7, 8, 12, 16]. In this aspect, in recent studies [5] we demonstrated that PPi acted as an active inhibitor on the heterogeneous calcium phosphate nucleation of calcium oxalate.

From the results obtained in our study it can be observed that there were no important differences (Fig. 1), in the concentration range of orthophosphate between groups A, B and C. Nevertheless, the PPi concentration was significantly lower in the stone formers than in the control group [4]. It was interesting that in the stone former group without metabolic alteration (A) the PPi concentration hardly varied while the orthophosphates concentration did (Fig. 2c). In the control group (C) (Fig. 2a) there was a relationship between PPi and orthophosphate that can be adjusted to a straight line with a positive slope, which seems to indicate the tendency of PPi to increase with the orthophosphate concentration. In the stone former group with metabolic alteration (B) the relationship between PPi and orthophosphate also can be adjusted

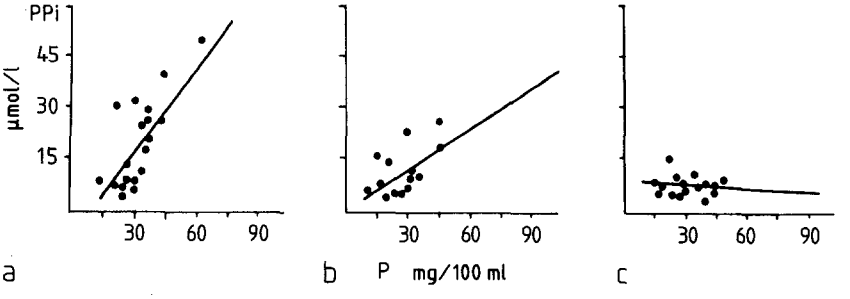


**Fig. 2a–c.** Relationship between urinary Pyrophosphate and Orthophosphate (as phosphorous) concentration. **a** Healthy subjects. N = 24; y = 0.406 × −7.345; r = 0.702. **b** Stone formers with metabolic alteration. N = 15; y = 0.179 × +0.925; r = 0.538. **c** Stone formers without metabolic alteration. N = 20; y = 0.029 × −9.024; r = 0.721

to a straight line (Fig. 2b), but with a positive slope which was lower than in the control group.

The results observed in the control group (C) are in accordance with Russell and Fleisch [13, 14] which found that there was a relation between both urinary orthophosphate and urinary PPi. This fact is probably due to a renal transformation of orthophosphate into PPi. The results from the stone former groups (A and B) could be attributed to a failure in this renal mechanism, which would be a lot more serious for the stone former group without metabolic alteration (A), than for the stone former group with metabolic alteration (B).

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