

A Contribution to the Formation Mechanism of Calcium Oxalate Urinary Calculi

II. In Vitro Experiments Concerning the Theory of the Formation of Whewellite and Weddellite Urinary Calculi

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Received: August 28, 1975

Summary. In vitro investigations of the formation of Whewellite or Weddellite are described. By means of different precipitation models the influence of cationic minerals on the formation of Weddellite could be observed. The possible conversion of Weddellite into Whewellite in vivo is demonstrated by in vitro experiments. A theory of the formation of Weddellite or Whewellite urinary calculi is developed on the basis of the results obtained.

Key words: Urinary calculi - Weddellite - Whewellite - Formation mechanism.

A previous paper (5) dealt with the determination of the urinary constituents responsible for the formation and stabilisation of Weddellite. The high stability of the synthetic Weddellite precipitates after precipitation from solutions with physiological ion ratios is in contrast to the low incidence (20 to 30%) of Weddellite urinary calculi.

In the present paper the conditions present during the formation of calcium oxalate calculi are simulated in an improved model. On the basis of these experiments a theory of the formation of Whewellite and Weddellite urinary calculi is developed.

MATERIAL AND METHOD

Synthetic ion solutions corresponding to the varying cationic relations of urine were prepared (Table 1). Calcium oxalate was precipitated from these solutions under different conditions.

Immediate Precipitation of Calcium Oxalate

2 mmol of ammonium or sodium oxalate in 2 ml of distilled water were added at one time to 20 ml of an 0.1 M CaCl₂ solution. The further procedure was as described in a previous paper (5).

Fractionated Precipitation of Calcium Oxalate

Simulating in vivo conditions, the sodium oxalate was not added at one time to the calcium solutions (Fig. 1), but in ten 0.2 ml portions.

After each addition the precipitates were removed by centrifugation, washed with distilled water, alcohol, and ether and analysed by infrared spectroscopy.

The stability of these precipitates was tested in dry conditions at 38°C and 110°C and at 38°C under physiological conditions.

In addition, 20 Weddellite urinary calculi were powdered and treated with 5 different synthetic solutions in order to check the stability at 38°C for 6 days in a closed system (Table 1). The same urinary calculi were also treated in a flow system. The calculous material was brought into contact with the synthetic solutions at 38°C by dropwise addition and continuous overflow.

RESULTS

Weddellite precipitates which were stable under dry conditions were always obtained from solutions II and III following precipitation with sodium oxalate. If these precipitates as well

Table 1. Testing of the stabilising effect of various solutions on 20 Weddellite urinary calculi (6 days, 37°C) (1 = 8 mmol/l; Me = Ni, Co, Mn, Cu)

Solution	Ca	Mg	Zn	Me	Urea	Creatinine	Average conversion into Whewellite (%)
I	1	10 ⁻¹	-	-	-	-	70
II	1	10 ⁻¹	10 ⁻³	10 ⁻⁵	-	-	60
III	1	10 ⁻¹	10 ⁻³	10 ⁻⁵	35	19	40
IV	1	3 x 10 ⁻¹	10 ⁻³	10 ⁻⁵	-	-	10
V	1	3 x 10 ⁻¹	10 ⁻³	10 ⁻⁵	35	19	0

Molar Ion Ratios

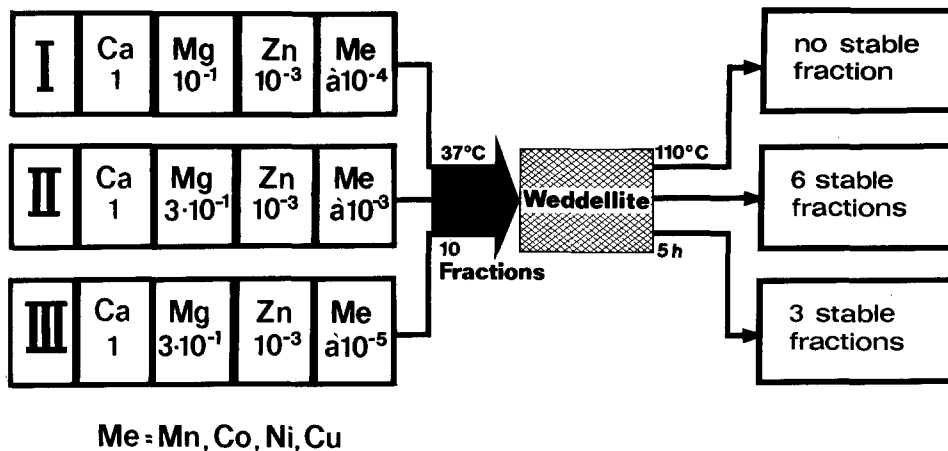


Fig. 1. Fractionated precipitation of calcium oxalate from synthetic ion mixture

as the Weddellite urinary calculous material, finely crushed in a mortar, were treated at 38°C with distilled water in a closed system, a 100% conversion into Whewellite could be observed within 6 days.

The same result was obtained when ionic solutions with physiological concentration ratios of calcium, sodium, and potassium (1:20:6) were used. But the same materials were not changed under the same conditions if Ca-Mg solutions (1:1 and 1:0.3) were employed. The same applies to solutions which additionally contained physiological concentrations of trace elements (Zn, Ni, Co, Mn). Urea and creatinine also delay the conversion of Weddellite into Whewellite. Table 1 shows the results obtained.

Weddellite precipitates were always obtained by immediate precipitation from a physiologi-

cal ion model solution at 37°C. If precipitation was fractionated, i. e. if the same quantity of sodium oxalate solution was divided into 10 equal portions and added dropwise to the same solution, only the first three precipitation fractions yielded temperature-stable (110°C) Weddellite and all other fractions were converted into Whewellite. More or less stable fractions were obtained depending on the concentration of the additional cations. Figure 1 is a schematic representation of the results obtained. In stability tests of the individual fractions under physiological conditions at 37°C, all unstable fractions were fully converted into Whewellite. The stable fractions showed a differential conversion analogous to the reaction of the Weddellite urinary calculous material shown in Table 1.



Fig. 2 Part of a calcium oxalate urinary calculus. The points consist of Weddellite, the base of Whewellite.

DISCUSSION

Hypothesis Concerning the Formation of Weddellite or Whewellite Urinary Calculi

The ion concentration ratios in urine collected over 24 hours are frequently such that calcium oxalate can be precipitated at 37°C by the addition of a small amount of oxalate and Weddellite is formed instead of the thermodynamically stable Whewellite. Weddellite is stabilised by the incorporation of additional cations. Calcium oxalate crystals (Weddellite) are found in the urine sediment as small refracting octahedrons. Rarely crystal shapes typical of Whewellite are seen, such as small plates, dumb-bells or hour-glass forms. These observations agree with the experimental results which suggest that certain ions may support the formation of Weddellite. Magnesium shows a particular stabilising effect.

The formation of two hydrate crystal shapes in calcium oxalate calculi has induced other authors to carry out detailed studies (1, 2, 3, 4, 6, 7, 8, 9, 10, 11, 12).

From our experimental findings we conclude that Weddellite crystals are formed primarily in most cases and excreted without aggregation. However, if the renal epithelium has been damaged the conditions exist for epitaxial growth on the tissue. Gebhardt describes the epitaxial deposition of calcium phosphate on collagen after depolymerisation of the surrounding mucopolysaccharides (3).

The microlite deposited on the tissue now

continues to grow depending upon the urine composition and maturation of the primarily formed crystals and conversion into Whewellite may occur at the same time. This is clearly shown in thin ground sections. Prien and Frondel (11), Cifuentes et al. (2), and Szabo (12) also describe the frequent observation of Weddellite crystal layers on the surface of calcium oxalate urinary calculi. Under these layers there are loose structures which under the polarising microscope show neither typical Weddellite nor Whewellite forms. In 1957, Hösl (6) also observed crystals on ground sections with the outward appearance of Weddellite which were changed inside into Whewellite.

Our experiments show that the conversion of Weddellite into Whewellite depends mainly on the concentrations of the additional cations. Low concentrations during the day when urine output is high may initiate Whewellite formation. The stability tests with solutions of differing ionic composition carried out on finely crushed Weddellite calculus material (corresponding to the initially porous surface of a growing calculus) show that Weddellite can be converted into Whewellite within a few days if there are not enough stabilising substances present. If there is sufficient magnesium, this conversion can be prevented. With immediate precipitation the formation of crystals is influenced by a constant concentration of additional ions. The absorption or incorporation of these ions into the crystal structure results in uniform stabilisation. Under the conditions of fractionated precipitation the solution be-

comes depleted in stabilising ions, and depending on the initial concentrations sufficient stabilising factors to produce stable Weddellite are present in only some fractions.

Depending upon the variation in the excretion of calcium and oxalate ions with food intake the crystallisation conditions in urine lie between the experimental extremes of immediate and fractionated precipitation.

The shape of the so-called mulberry calculi is due to the primary formation and rapid conversion of the sharp-edged Weddellite crystals into Whewellite. Figure 2 shows a calcium oxalate calculus the points of which consist of Weddellite, whereas the basal layers have already been converted into Whewellite. The present theory does not account for the very hard, smooth, frequently dark-brown to black coloured Whewellite calculi and further analysis is required.

Acknowledgement. We are particularly grateful to Mrs. G. Beck for technical assistance.

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