

1-29-1990

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### Recommended Citation

Deganello, Sergio and Di Franco, Livia (1990) "Crystal Growth of Calcium Oxalate Monohydrate and Calcium Carbonate from Dilute Solutions," *Scanning Microscopy*. Vol. 4 : No. 1 , Article 17.

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CRYSTAL GROWTH OF CALCIUM OXALATE MONOHYDRATE AND  
CALCIUM CARBONATE FROM DILUTE SOLUTIONS

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(Received for publication October 12, 1989, and in revised form January 29, 1990)

Abstract

By controlling evaporation, calcium carbonate (calcite) is precipitated together with calcium oxalate monohydrate from equimolar ( $1 \times 10^{-5}$  M) solutions of calcium chloride and sodium oxalate under decreasing ( $37.5^{\circ}$ - $32.7^{\circ}$ C) and increasing temperature ( $22^{\circ}$ - $42^{\circ}$ ,  $22^{\circ}$ - $45^{\circ}$ ,  $22^{\circ}$ - $50^{\circ}$ C), and initial pH's of 6.6 and 6, respectively. If, however, the pH of the solutions is, respectively, 8 and 8.5 and the temperature is  $32^{\circ}$ C, oxalate breaks down to carbonate and calcium precipitates solely as calcite. This process materializes as the pH of both solutions initially adjusts to about 7.5. Both the quality and the size of the crystals of calcite and calcium oxalate monohydrate varied markedly. Euhedral crystals of calcite, measuring in excess of 200 micrometers in cross-section, and grown at  $37.5^{\circ}$ C were remarkably ordered as shown by the lack of twinning, streaks and/or diffuseness around the diffracts, which is evidenced by single-crystal x-ray diffraction analysis. In contrast, calcium oxalate monohydrate was almost always disordered due to twinning along the (101) plane and never exceeded 100-150 micrometers along a coordinate axis.

Key words: Precipitation, Calcium oxalate monohydrate, Calcite, pH, Evaporation, Carbonate artifacts, Urinary calculi.

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Introduction

Calcium oxalate monohydrate (COM) and dihydrate (COD) are common constituents of the alteration patinas found between the calcitic substrate and the external degradation layers of carbonate artifacts (Cipriani and Franchi, 1958; Franzini et al., 1983; Dal Monte et al., 1987). Although the paragenesis of COM and COD in those findings has long been controversial, recent unpublished evidence by Deganello and Montana indicates that the destabilization of oxalate to calcium carbonate occurs with a low energy of activation at a solid-liquid interface. As a consequence, one would therefore assume that, under suitable conditions, it should be possible to crystallize calcite from dilute solutions which usually precipitate only COM and/or COD. Here we present preliminary data which support this hypothesis. A more comprehensive report will be presented at a later date.

Materials and Methods

Crystallization was induced by using highly diluted solutions of reagent-grade equimolar calcium chloride (Fisher Scientific) and sodium oxalate (Baker and Adamson), initially at  $1 \times 10^{-5}$  M concentration. Water was doubly distilled. The acidity was adjusted with 0.1 N HCl and NaOH. In all instances the solutions were unbuffered in order to avoid influencing crystallization with additives. The pH of the solutions was measured at room temperature before and after crystallization. All the experiments were carried out in triplicate and tested for reproducibility at least twice.

Experiments at decreasing temperature

Five ml of calcium chloride were added to 5 ml sodium oxalate using an infusion pump (Harvard, model 600-910) operating at 0.5 ml/min. The starting pH was 6.6. The solution was then transferred to a 50 x 22 mm vial which was tightly covered with five layers of gauze (mesh size: approximately 2.5 x 1.1 mm). The vial was immersed in a water bath equilibrated at  $37.5^{\circ}$ C. After 24 hours the temperature (T) was lowered by a predetermined amount once a day. This resulted in decreases of about  $0.3^{\circ}$ C/day for the first 11 days and of about  $0.5^{\circ}$ C/day until the temperature was  $32.7^{\circ}$ C when the experiment was completed.

Experiments at increasing temperature

Each of nine vials (15 x 100 mm) was filled with 5 ml of calcium chloride and sodium oxalate and

**Table 1.** Results of crystallization experiments.\*

Experiment number	Number of layers of gauze	Time (h)	Concentration (M)	T <sub>i</sub> (°C)	T <sub>f</sub> (°C)	pH <sub>i</sub> (22°C)	pH <sub>f</sub> (22°C)	Phases
1	5	360	1x10 <sup>-5</sup>	37.5	32.5	6.6	**	COM, Cal
2	6	104	1x10 <sup>-5</sup>	36.5	50.0	6.0	7.7	COM, Cal, NaH
3	6	179	1x10 <sup>-5</sup>	34.0	45.0	6.0	8.0	Cal, COM, NaH
4	6	468	1x10 <sup>-5</sup>	34.0	42.0	6.0	8.2	Cal, COM(t), NaH
5	6	432	1x10 <sup>-5</sup>	32.0	32.0	4.0	4.2	-----
6	6	432	1x10 <sup>-5</sup>	32.0	32.0	8.5	7.1	Cal, Tet
7	6	432	1x10 <sup>-5</sup>	32.0	32.0	8.0	7.0	Cal, Tet

\*T<sub>i</sub> and T<sub>f</sub> apply to initial and final T; pH<sub>i</sub> and pH<sub>f</sub> are the pH's read at room temperature, respectively, at the beginning and end of the experiments. Cal, NaH and Tet stand for calcite, sodium bicarbonate and tetracosan. (t) refers to traces.

\*\* After 53 hours at 37°C the pH is 7.1 once experiment #1 is repeated under continuous pH monitoring.

then closed with six layers of gauze. The initial pH was 6. The first set of three vials was stabilized in a water bath for 48 hours at 36.5°C, after which the temperature was raised and held at 50°C for 56 hours. The second set of three vials was equilibrated to 34°C for 48 hours and the temperature then raised to 45°C for 132 hours. Finally, the third set of vials was equilibrated at 34°C (48 hours) and then reequilibrated to 42°C for 420 hours. In all cases, the temperature was increased in steps of about 0.5°C/10 minutes using a Lauda immersion circulator; the temperature variability in the water bath was less than ± 0.2°C.

#### Experiments at constant temperature and varying pH

Three solutions were prepared, each consisting of 5 ml of calcium chloride and 5 ml of sodium oxalate. Their pH's were, respectively, 4, 8 and 8.5. Each of these solution was transferred into separate vials (15 x 100 mm) which were sealed with six layers of gauze; the temperature was increased in steps of about 0.5°C/10 minutes to 32°C and kept at this value for 432 hours.

#### Analyses

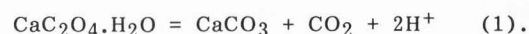
The precipitates were analyzed by plane and polarized light microscopy, scanning electron microscopy, and both powder and single-crystal x-ray diffraction. For the electron-microscopy work, an ETEC microscope, operating at 25 kV, was used. The samples were mounted on platinum grids, using a silver-based cement, and gold-coated by glow discharge under vacuum. For the powder diffraction studies, the patterns were generated directly from single crystals using a Gandolfi camera (114.6 mm) operated under vacuum (10<sup>-3</sup> torr) in Ni-filtered CuKα. The single-crystal work, instead, was carried out with a Buerger-precession camera using both unfiltered and Zr-filtered MoKα.

#### Results and Discussion

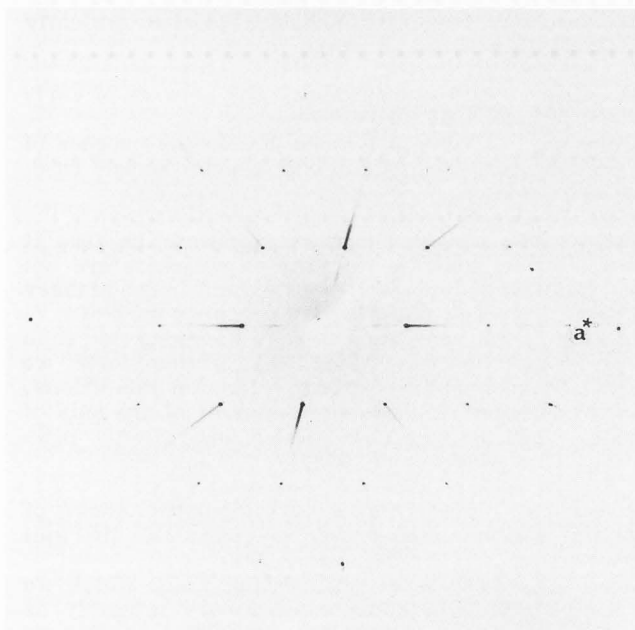
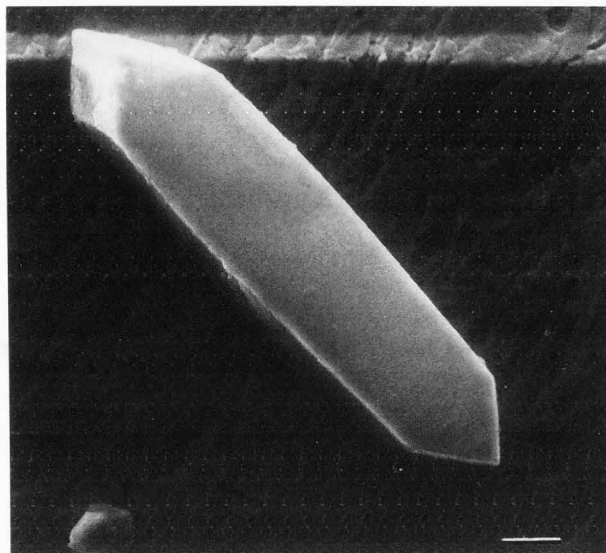
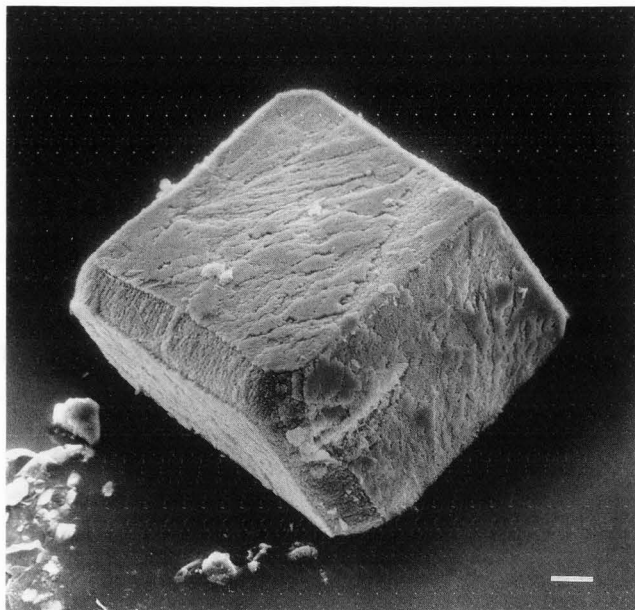
The results of the experiments are shown in Table 1. Calcite was detected in all growth baths with the exception of experiment #5 (pH 4), wherein no precipitate formed. COM occurred in experiments #1, 2, 3, 4 but not in #6 and 7. Traces of sodium bicarbonate (NaHCO<sub>3</sub>) and tetracosan (C<sub>24</sub>H<sub>50</sub>) were

also detected; the former in experiments #2, 3, and 4 and the latter in experiments #6 and 7. Both the quality and the size of the crystals of calcite and COM varied markedly. Euhedral crystals of calcite, measuring in excess of 200 micrometers in cross-section, were found in experiment #1; one such specimen is shown in the scanning electron micrograph of Fig. 1. This crystal was remarkably ordered as shown by the lack of twinning, streaks and/or diffuseness around the diffracta, which is evidenced (Fig. 2) by a [0kl] precession photograph taken according to the rhombic cell a = 6.42 Angstroms, α = 101.55°. In contrast, COM was almost always disordered due to twinning along the (101) plane; this face, together with the well developed (010) [P2<sub>1</sub>/n setting], conferred to COM its elongated prismatic habit. Never did any of the COM crystals exceed 150 micrometers along a coordinate axis.

The preceding results reflect the synergistic role that ionic concentration, pH, temperature, time and rate of evaporation, assume in the reaction:



This proceeds very slowly at room and moderate temperatures since oxalate is extremely persistent in water solutions (Latimer, 1952). However, as oxalate begins to break down to carbonate, the amount of CO<sub>2</sub> which is expelled from the solutions increases, thus causing the pH to rise. Such an effect is consistent with the results from experiments #1, 2, 3, 4. Specifically, the pH was measured to rise from 6.6 to 7.1 after 53 hours at 37°C in experiment #1 once this was repeated under continuous pH monitoring. Similarly, in experiments #2, 3, and 4, when measured at 22°C, the pH shifted from 6 to 7.7, 8.0 and 8.2, respectively, following warming to 50°, 45° and 42°C for 56, 132 and 420 hours. In agreement with such an increase in alkalinity, sodium bicarbonate precipitated. This phase was identified by powder diffraction analysis using the data provided by Zachariasen (1933). Furthermore, since the highest pH was observed in experiment #4 (this having the longest crystallization run) and this pH value did not coincide with the highest temperature, it would appear that time was an important factor in



**Figure 1 (above top).** A single crystal of calcite produced from experiment #1. Notice well developed morphology. Bar = 10 micrometers.

**Figure 2 (above).** A  $h01$  precession photograph of the crystal in from Fig. 1. Notice absence of twinning and lack of streaking around the diffracta (Zr-filtered  $\text{MoK}\alpha$ ,  $\mu = 20^\circ$ , 40 kV, 20 mA, 12 hours).

**Figure 3 (top right).** A particularly well developed crystal of COM. The morphology of COM, although always governed by the preferential development of  $\{010\}$ 's and  $\{\bar{1}01\}$ 's, is rarely as well developed as the one shown in this micrograph. Bar = 10 micrometers.

determining the final acidity of the solutions. Such a conclusion is particularly important since evaporation never exceeded 8% of the original volume of the solutions.

Due to the high value of the initial pH's (8.0 and 8.5), reaction (1) proceeded far more rapidly in experiments #6 and 7 than in experiments #1, 2, 3, and 4 even with T as low as  $32^\circ\text{C}$ . Consequently oxalate readily began to break down to carbonate and calcium precipitated solely as calcium carbonate. The dynamics of these changes was rationalized once we repeated experiments #6 and 7 under continuous pH monitoring (8-72 hours) and, eventually, retested them for various values of T's ( $32^\circ$ - $55^\circ\text{C}$ ) and pH's (8.0, 8.5, 8.8, 9.4). Invariably, in all of these studies, after the experimental temperature was reached, the pH commenced to decrease, appearing to stabilize at about 7.5 after 72 hours. Depending on the temperature and rate of evaporation chosen, such a decrease took up to several hours to materialize and was attributed to the precipitation of calcium carbonate, in agreement with the findings of De Keyser and Dugueldre (1950) in the calcium chloride-sodium carbonate system. Consequently, it became clear that the additional decline in the value of the pH from 7.5 and 7.1 to 7.0, measured at the end of experiments # 6 and 7, must have taken place at a rate far slower than that which controlled its initial decrease from 8.5 and 8 to 7.5. In light of this, as well as the fact that alkanes precipitate out of dibasic carboxylic acids solely after these decompose to carbonate, it appears reasonable to suggest that tetracosan (approximately 85 % C and 15 % H) began crystallizing at a pH value comprehended between 7.5 and 7. Tetracosan was identified by powder diffraction analysis (Strunz and Contag, 1964; Powder diffraction file, fiche #28-2004, Amer. Soc. Test. Mat., Philadelphia); according to Maazee (1948), the polymorph detected here corresponds to the monoclinic II phase which is metastable at room temperature.

#### Acknowledgments

Sergio Deganello acknowledges grant DK-33949 from the National Institutes of Health.



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## Discussion with Reviewers

R. Tawashi: Would it be possible to grow single crystals of calcite larger than 200 micrometers using the technique described in this paper?

Authors: In theory it should be possible to grow larger crystals provided that the correct conditions (especially with respect to time, pH and rate of evaporation) are met. The question, however, remains as to whether or not this is the most efficient technique to do so.

R. Tawashi: What was the rate of evaporation in the experiments conducted at increasing temperature?

Authors: We estimate the rate of evaporation to be about 0.04 ml/day in experiments #2, 3 and 4.

A. Rodgers: Were the morphologies of the COM and COD crystals similar to those occurring in urine?

Authors: The morphology of COM tends to be determined by the preferential development of the {010}'s and {101}'s. These faces cause the crystals to assume an elongated prismatic habit (See Fig. 3). As to COD, this was occasionally detected in a series of experiments conducted under faster rates of evaporation and lower final temperatures (37°-39°C) than those reported in experiments 2, 3, and 4. The habit of these COD crystals is similar to that typically found in urine.

S.R. Khan: Did you determine the amounts of various precipitates formed?

Authors: No, we did not. We are in the process of expanding our studies and will report, in due time, on this question.

S.R. Khan: The temperature, pH and the time it took for precipitation in these experiments are not the conditions which one would expect in the urinary system. Are you planning experiments at 37°C?

Authors: We have been already carrying out such experiments for quite some time. In particular, we have chosen, as our initial reference conditions, those characterizing the thin segment of the loop of Henle. Preliminary data have been recently presented (Deganello et al, 1989).

G. Mandel: Does the pH dependent breakdown of oxalate explain the presence of calcite in gall stones?

Authors: This is an interesting point which we hope to be able to address with more authority as soon as we complete the above referred studies on the conditions governing crystallization in the loop of Henle.

## Additional Reference

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