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Abstracts

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Author(s): **A. Hesse**
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(no abstract submitted)

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Title: INVESTIGATIONS OF THE STRUCTURE OF CALCIUM OXALATE CALCULI AND LITHOGENIC PARAMETERS IN URINE

Author(s): G. SCHUBERT (1), G. BRIEN
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Aim of the investigations was to clarify potential relations between the structure of urinary calcium oxalate calculi and lithogenic urinary parameters.
Calcium oxalate calculi were analysed using polarization microscopy and scanning electron microscopy. The variety of the known structural forms was reduced to four basic structure types (I-IV). Here, the formation of structure types I and II can be explained in terms of a primary crystallization of whewellite, and the formation of structure types III and IV can be explained in terms of primary crystallization of weddellite.
109 patients with calcium oxalate calculi were selected without significant previous medical metaphylaxis. Their structure type was determined using polarization microscopy. Calcium, magnesium, uric acid, oxalate, citrate and others were measured in 8 h urine collections. In addition part of patient population was tested for inhibitory index, relative supersaturation and glycosaminoglycanes.
The preliminary results show an increased concentration of calcium, magnesium and glycosaminoglycanes among the patient group with the structure types III and IV. In addition, the same patient group shows a decreased citrate concentration and higher values of the relative supersaturation in comparison to patients with structure types I and II.
These results indicate that calcium oxalate calculi with structure types III and IV are built in patients with higher metabolic activity. The predominant factor which determins the development of the calcium oxalate structure is the concentration of calcium in urine.
In future it may be possible to deduce informations on the pathogenetic situation of urinary calculi from the structure type of calcium oxalate calculi.

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Titel: PHASE COMPOSITION OF URINARY CALCULI

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From 1976 to 1982 and from 1985 to 1992, 1603 urinary calculi were analysed by x-ray diffraction, infrared spectroscopy and polarized microscopy.

The comparison of these two collectives shows partly significant differences with regard to the phase distribution of men to women.

In the first collective the ratio of men to women was 1.4 : 1. 66.2% of these stones (43.8% male, 22.4% female) consisted of oxalate phases (whewellite, weddellite, mixed oxalates), 7% (1.5% male, 5.5% female) of Ca-phosphate (apatite), 15.5% (5.4% male, 10.1% female) of Mg-phosphate (struvite) and 11.3% (8.2% male, 3.1% female) of uric acid.

The proportion of men to women in the second collective was 2.7 : 1. In this collective the phase compositions of the stones were 67.9% oxalate phases (53% male, 15% female), 14.1% Ca-phosphate (8.4% male, 5.7% female), 4% Mg-phosphate (1.5% male, 2.5% female) and 9.2% uric acid (7.5% male, 1.7% female).

Beside the comparison of the 4-class phase distribution to that of the first collective, this group is statistically evaluated referring to phase frequency and phase combination in connection with sex and age.

Another point of view of this collective was the mineralogical investigation of 53 urinary calculi (31 male, 22 female) from 31 (21 male, 10 female) recurrent stone formers.

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Title: **Analysis of thermally treated phosphate stones with x-ray diffraction**

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An exact quantitative analysis of phosphate stone material presents a lot of difficulties. A lot of suggestions have been made on how to deal with these. However, none of the methods applied has proved to be satisfactory.

According to ARMBRUSTER, DOSCH, ALTROCK and GEBHARDT and SEIFERT it is possible to improve the quantitative analysis of phosphate stones by submitting them to thermal treatment thus effecting crystal growth. However, the equations of chemical reactions demonstrate that the organic stone minerals are not stable at 900 C: CO2, H2O and NH3 are volatile matter contents, thus the equation final product and original material does not come out. Phosphate stone material was therefore submitted to thermal treatment and analyzed at lower temperatures and for a longer period. Results were compared to those abtained with 25 samples analyzed with the DTA (differential thermal analysis) method.

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Title: MINERAL COMPOSITION OF RENAL STONES FROM THE SUDAN

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Since Urolithiasis is still common in the Sudan, a knowledge of the composition of a stone yields a fundamental information concerning the pathogenesis of the disease and helps in the prevention of stone disease in this area. This study is concerned mainly with the quantitative mineral composition of stone analysed by IR-spectroscopy. This is the only series of stones from the Sudan analysed by this method, and compares the findings with those in other studies.

80 upper urinary tract stones from Sudanese patients (age: 33.25, SD: 16,57), 45 males (age: 32.20, SD: 16,94), 35 females (age: 34,6, SD: 16,23), were analysed by IR-spectroscopy. The distribution of stones materials was calculated on the basis of the three main components in each stone.

All stones are composed of maximal three components. The average percentage of components are Whewellite (46.0%), Weddellite (19,0%), Uric acid (10.2%), Uric acid dihydrate (1,1%), Carbonate apatite (11.8%), Ammonium urate (7.38%), Struvite (3.9%), Brushite (0.1%) and Cystine (0%). Greater differences are found in comparison of the present data with those of previous study from the Sudan analysed by quantitative chemical method. No differences are found in comparison of the present data with those in German materials.

Detailed physico-chemical quantitative analysis of stones may serve to identify different group of stone -formers and different type of stones.
Upper urinary tract stones in developing countries like the Sudan have the same composition as those in industrailised countries.

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Titel: **Comparative Infrared Spectroscopy and Scanning Electron Microscopy Investigation on Different Kinds of Encrustated Catheter Materials**

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Urinary tract infections (UTIs) are the most common nosocomial infections and most of them are catheter associated. Several studies have reported that some materials or coatings of catheters can decrease the rate of infection and others have pointed out, that the composition of encrustations with infection stone material although depends on the surface materials involved. This study examined different types of catheter materials concerning their tendency of encrustation, their alteration of materials and the functional consequences of it.

Encrustations on foley catheters (18 Charrier) of different pure or combined materials [all silicone (AS), silicone coated latex (SL), hydrogel coated latex (GL)], produced by simulated UTI in a standardized in vitro model, have been investigated by infrared spectroscopy (IR). Scanning electron microscopy (SEM), including energy dispensive X-ray micro analysis (μX-ray) and a photographic documentation have been established for the catheter before and after the encrustation process. Function tests had been performed to describe the catheters drainage capacity (DC) in ml/min of new and encrustated catheters as well as the relative decrease of their DC. Ten catheters of every kind were included in this study.

The IR resulted in 66.0% (AS), 60.5% (SL) and 71.0% (GL) struvite as the main encrustation material [variation coefficient (VC)=0.119]. The rest consisted of carbonate apatite and amorphous calcium phosphate in variable portions. There was no statistical difference in the composition of the encrustations between the tested catheter materials. The DC of the new catheter was highest for the AS with 14.13ml/min and significantly different to the others [6.04ml/min (SL), 6.58ml/min (GL)]. Additionally the AS appeared as the most homogenous one [VC=0.056; 0.115 (SL), 0.122 (GL)]. After the encrustation period the DC decreased for all catheters tested [12.75ml/min (AS), 5.54ml/min (SL), 5.68 ml/min (GL) with VC=0.114 to 0.209]. The relative decrease of DC ranged from 8.6% (SL) and 10.0% (AS) to 14.1% (GL) and was not significant different.

It could be shown, that - under conditions of simulated UTI- all tested kinds of catheters are encrustated with the same infection stone materials. However, the cathethers tested show very different results concerning their DC, probably caused by the greater lumen of the all silicone catheter. Furthermore, the low variation of the DC for the all silicone catheter indicates a higher quality standard of production of these kind of foley catheters.

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Title: **Rational evaluation of the stone forming patient**

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State of the art lecture
(no abstract submitted)

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Title: STUDIES IN ENTERIC HYPEROXALURIA

Author(s): R.A.L. Sutton, V.R. Walker
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Small bowel by-pass surgery for morbid obesity may be complicated by severe enteric hyperoxaluria and refractory calcium oxalate nephrolithiasis. We have applied newly developed HPLC assays for plasma and urinary oxalate and glycolate to the study of eight such patients. Baseline data included: (mean±SEM) 24 hour urinary oxalate 1471±275 μmol (normal <560), glycolate 524±67 μmol (normal <1400), calcium 2.38±0.28 mmol, magnesium 2.10±0.30 mmol, citrate 0.95±0.25 mmol. Plasma oxalate was above the normal range (<3.2 μmol/L) in 6 of 8 patients (mean 5.3±0.9). Plasma creatinine was modestly elevated in 4 patients (range 62-132 μmol/L). Plasma glycolate was normal in all patients (mean 4.7±0.4, normal <7.8 μmol/L). The fractional urinary excretion of oxalate based on a fasting urine sample and midpoint blood sample was 143±10% compared with a mean normal value of 92±8% ($p<0.001$). Fractional urinary excretion of glycolate was normal (59±12%). Oral calcium supplementation was given in an attempt to reduce oxaluria. At 3 g/day calcium supplement (given as calcium sandoz or calcium carbonate) mean 24 hour urinary oxalate decreased to 730±169 μmol and urinary calcium increased to 3.27±0.58 mmol, contributing to a decrease in the stone risk index (Tiselius) from 1171±105 to 711±151 ($p<0.05$). In conclusion, despite increased fractional urinary oxalate excretions which presumably reflect increased tubule oxalate secretion, plasma oxalate is commonly increased in these patients. As expected, indices of glycolate metabolism are normal, and calcium supplements effectively decrease urinary oxalate excretion while causing only minimal increases in urinary calcium in the low-normal range.

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Title: THE STOMACH - A NEW OXALATE ABSORPTION SITE IN MAN

Author(s): P. Flohr, K. Kleinschmidt, R.E. Hautmann
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In the etiology of calcium oxalate urolithiasis it has been generally accepted that hyperoxaluria is more important than has previously been thought.
Oxalate absorption appears to occur along the course of the entire gastrointestinal tract. The stomach as oxalate absorption site has not yet been considered.

Patients and methods:
After gastric administration of 5 mmol oxalate load, the intestinal absorption of oxalate was assessed indirectly from the increase in renal oxalate excretion, measured by ion-chromatography.
In 6 adult patients gastric emptying was blocked by an intrapyloric balloon after informed consent. All patients were on permanent gastric tube feeding for various reasons and the oxalate load with sodium oxalate with or without calcium, spinach or rhubarb was applied via the gastric tube.
Urinary oxalate was measured 2, 4 and 6 hours after gastric loading.

Results:
Our examination of the time course of gastric absorption and renal excretion following a single gastric load of oxalate shows that with increasing gastric loading time there is a linear increase in the urinary oxalate excretion.
After 2 hours loading 15 to 21 percent, after 4 hours 24 to 45 percent and after 6 hours 62 percent of the gastric oxalate appeared in the urine.

Conclusion:
Our data about these absorption kinetics suggest that the stomach is a critical site for intestinal oxalate absorption in an intact gastrointestinal tract. The stomach seems to be powerful oxalate absorption site in man.
This opens a new field for the discussion of etiology and pathogenesis of calcium oxalate stone formation.

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Title: DIETARY OXALATE AND OXALATE DEGRADING BACTERIA IN THE GUT IN THE PATHOGENESIS OF CALCIUM OXALATE UROLITHIASIS

Author(s): K. Kleinschmidt, A. Mahlmann, R. Hautmann
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Hugh amounts of dietary oxalate pass daily through the human gastrointestinal tract. Nutrition therefore represents the largest potential source of oxalate for the human body. Oxalate is absorbed via the intestinal mucosa by an ion exchange process. It is excreted unchanged into the urine. Just recently a new species of anaerobic bacteria, Oxalobacter formigenes, has been described. It is living exclusively from oxalate. The purpose of this study was to evaluate the function of oxalate degrading bacteria in the colon in preventing dietary oxalate from absorption and thus preventing calcium oxalate stoneformation.

31 selected patients with calcium oxalate urolithiasis of different activity were compared with 29 controls. During a five days hospital stay the study participants received an oxalate rich standard diet. Fecal samples were compared with regard to colony formind units of oxalate degrading bacteria and corresponding oxalate concentrations. Circadian urinary calcium and oxalate levels were measured. Oxalate was analyzed by ion chromatography.

In higly active stoneformers no oxalate degrading bacteria could be found. Oxalate concentrations after extraction from fecal samples were higher in stoneformers than in controls. Circadian urinary oxalate concentrations of stoneformers revealed in two fractions a significant difference of critical oxalate levels (500 umol/l) between stoneformers and controls ($p < 0.05$). We found a significant difference, too, in four fractions of the urinary calcium concentrations with higher levels in stoneformers ($p < 0.05$). We were not able to show a correlation between high numbers of colony forming units of oxalate degrading bacteria per g feces and corresponding low urinary oxalate concentrations.

From our data we conclude, that lack of intestinal oxalate degrading bacteria is not a predominant risk factor for calcium oxalate urolithiasis, but there is evidence, that these bacteria help to prevent dietary oxalate from intestinal absorption.

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Title: The significance of urease-splitting bacteria in upper urinary tract stones

Author(s): A. Heidenreich, R. Bonfig, W. L. Strohmaier, K. H. Bichler
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Infections with urease producing bacteria are claimed to be a prerequisite for the development of infection stones. Standard diagnostic procedures in stone disease and bacteriuria do not include determination of urease activity. Therefore a sensitive testing system would be important for the identification of urease-splitting bacteria since bacterial growth might appear in such low numbers that it may be missed. We tested the urine of 148 stone patients for ureolytic bacteria; correlation of urease activity, stone localisation, stone composition and urine cultures was performed.
148 pts. (100 males, 48 females) treated for upper urinary tract stones were studied. On the day of admission a mid-stream voided bladder urine was obtained for culture and determination of urease activity. Stone analysis was performed by X-ray diffractometry.
79 pts. (53%) were urease positive and 69 pts. (47%) were urease-negative. Significant bacteriuria with urease-splitting bacteria was shown in 23/79 (29%) and in 2/69 (2.9%); in 27/79 (34%) and 53/79 (76%) the number of bacteria was less than 10^4 and in 29/79 (37%) urease activity was positive in negative urine culture. Stone localisation revealed caliceal calculi in 26 (18%), staghorn calculi in 32 (22%), ureteral stones in 96 (65%) and bladder stones in 4 (3%). Urease positivity was determined in 15/26 (58%) caliceal stones, 24/32 staghorn calculi (75%) and in 38/96 (40%) ureteral stones.Stone analysis revealed calcium oxalate in 89 (60%), struvite/apatite in 14 (9.4%), uric acid in 16 (11%), cystine in 3 (2%), and mixed stones with struvite in 21 (14%). Urease activity was demonstrated in 38/89 (43%), in 11/14 (80%), in 10/16 (62%) and in 16/21 (80%) stones.
53% of the pts. were tested urease positive whereas a positive urine culture was obtained in only 17%. It appears that voided urine cultures do not always reflect bacteriology of the upper urinary tract and that detection of urease is possible below significant bacteruria. Since Proteus associated infection may cause asymptomatic colonization of the urinary tract with negative cultures we believe that testing for urease is a convenient and necessary completion of the bacteriologic-diagnostic measures. The urease test may also be useful in assessing the efficacy of antibiotic treatment in infection stones. 42% of calcium-oxalate stones were urease positive; the possible role of urease-splitting bacteria in pathogenesis of calcium stones will be discussed (dimnished GAG concentrations?).

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Title: Simple calculation of urinary titratable acidity.

Author(s): D.J. Kok
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One of the subjects of interest in calcium-salt stone formation is the metabolism of citrate which is related to the acid-base status of the body. In assessing the acid base status the renal Net Acid Excretion (NAE) is one of the parameters used. Renal NAE is calculated as titratable acidity plus urinary ammonia concentration minus urinary bicarbonate concentration. Titratable acidity (TA) is defined as the amount of base-equivalents needed to bring the urine back to its original pH of 7.4. The actual titration procedure is found here to be error-sensitive. The presence in the sample of uric acid crystals at the outset or the precipitation of calcium-phosphate during the titration may yield overestimates of the TA value of up to 25%. Furthermore storage of the samples, even under mineral oil, results in a decrease in measured TA over time due to the loss of bicarbonate. An alternative method has been developed in which TA is calculated using an adaptation of a computer-program for the calculation of Equilibria in the urine (Equil) and several urine components which are routinely measured in the evaluation of a stone patient. This method is shown to be insensitive to the errors found in the titration procedure. With the minimal use of the urine parameters pH, phosphate, citrate, bicarbonate and urate the TA can be calculated with a minimal accuracy of 95% using either the computer program or a nomogram developed by us. For urines with a pH value below 6.6 the positive contribution of bicarbonate as a buffer to the TA is almost equal to the bicarbonate concentration itself. Omittance of the bicarbonate does not affect the determination of NAE significantly in these urines and the relatively tedious measurement of carbon dioxide pressure can be omitted. In conclusion the calculation of TA proves a reliable and simple method.

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Title: NEW USES FOR SUPERSATURATION INDICES IN STONE PROPHYLAXIS

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Although most urines are supersaturated with calcium oxalate, stone formers tend to have higher supersaturations and it is accepted that decreasing their urinary supersaturation will decrease their risk of recurrent stone formation. This could, in principle, be achieved by increasing fluid output, decreasing calcium or oxalate excretion, or increasing citrate or magnesium excretion. What is not clear is the amount that these variables should be changed to bring about a more acceptable supersaturation.

Urine analyses from 40 patients attending a stone clinic were examined. For inclusion in this study, a pair of 24 hour samples on consecutive days, each with normal creatinine excretion, was required, with at least one of the pair of samples having a calcium oxalate supersaturation ratio (SS-CaOx) >5. When the volume, citrate or magnesium excretion or concentration was below a set limit, or calcium or oxalate excretion or concentration was above a set limit, then the amount by which this should be changed in order to reduce SS-CaOx to between 4.95 and 5.00 was calculated.

80 of the 100 samples had SS-CaOx >5. We found that this could, in theory, be relieved by increasing the average 24 hour volume from 1.7 to 2.8 litres. Reasonable limits were set for how far calcium or oxalate might be reduced, or citrate or magnesium increased, with the following results:

Urine abnormality	Samples which could realistically achieve SS-CaOx <5		Average mmol per day required to achieve SS-CaOx <5
High oxalate output or conc.	41/55	75%	0.31
High calcium output or conc.	62/64	97%	4.88
Low citrate output or conc.	6/31	19%	3.68
Low magnesium output or conc.	11/29	38%	7.55

A high SS-CaOx can nearly always be overcome by an increase in fluid output or decreasing calcium or oxalate when these are abnormally high. Increasing the citrate or magnesium on their own will not generally be sufficient and should usually be accompanied by other approaches. These are only general conclusions, ideally the appropriate calculations should be performed for each individual patient and therapy tailored accordingly.

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Title: **METABOLIC WORK-UP IN IDIOPATHIC CALCIUM STONE FORMERS - HOW MANY URINES AND HOW EARLY AFTER UROLOGICAL INTERVENTION ?**

Author(s): B.Hess, U.Strub, °D.Ackermann, Ph.Jaeger
Institution: Policlinic of Medicine & Dept. of Urology°, Univ. of Berne, Switzerland.

After they had been given the advice of increasing fluid intake by the treating urologists, renal stone formers referred for metabolic evaluation routinely collected three 24-h urines (U1, U2 & U3) while on a free-choice diet; all medication interfering with calcium phosphate metabolism was discontinued for at least 2 weeks. To account for individual dietary variations, U1 had to be collected during the week, U2 on a weekend, and U3 again mid-week. No patient was studied prior to 2 months after urological intervention. Excretion rates in U1, U2 and U3 were compared with mean values of all three 24-h urines (U_{1-3}), taken as "gold-standard", as well as with normal 24-h urine excretions, obtained in healthy volunteers: volume (Vol) > 1200 ml, calcium (Ca) < 9.0 mmol (men)/< 7.5 mmol (women), oxalate (Ox) < 450 µmol, uric (UA) < 5000 µmol (men)/< 4000 µmol (women), and citrate (Cit) > 1.70 mmol. All values are means ± SEM.

Among our recurrent idiopathic calcium stone formers (**RCSF**), 66 strictly followed the protocol. The mean **time lag** between collections of U1 and U3 (T_{3-1}) was 62 days (range 4-243). Excretions of creatinine, Ca, Ox and UA did not vary significantly between collections; $\mathbf{U_{Cit} \times V}$ tended to be lower in U2 (2.87 ± 0.18 vs. 2.97 ± 0.16 mmol/d in U_{1-3}, p = 0.128). Compared with U_{1-3}, **Vol** was higher in U1 (2128 ± 103 vs. 2008 ± 96 ml, p = 0.002), but lower in U2 (1913 ± 101 ml, p = 0.036). There was an inverse correlation between the change of Vol (Vol_{3-1}) and T_{3-1} (R = 0.37, p = 0.004), i.e. the later U3 was collected after U1, the lower was Vol. $\mathbf{U_{Urea} \times V}$ was lower in U2 (390 ± 19 vs 412 ± 17 mmol/d in U_{1-3}, p = 0.012), suggesting lower protein consumption on weekends. The **diagnostic yield** was best in U3 where 67.2% of urines had 1 or more abnormalities, compared with 48.9% in U1 (p = 0.05, Chi-square).

In **conclusion**, metabolic work-up of idiopathic RCSF appears to be of greatest diagnostic value if two 24-h urines are collected, one on a weekend (potential higher yield for low volume and hypocitraturia), and one as late as possible after urological intervention when the "stone clinic effect" is already vanishing (more abnormalities of urine chemistry).

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Title: LIMITATIONS OF METABOLIC EVALUATION IN CALCIUM UROLITHIASIS

Author(s): K.Höbarth, J.Hofbauer
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For years, urinalysis for metabolic factors thought to inhibit or promote calcium crystallization has been considered to enable effective medical and dietary interventions in patients with calcium urolithiasis. However, the results in terms of recurrence rates are contradictory, so that the clinical value of the diagnosed 'metabolic disorders' appears to be limited.

134 calcium stone formers (54 with recurrent, 80 with single episodes) followed up for a mean of 72 months were retrospectively analyzed for metabolic disorders such as hypercalciuria (HCU), hyperparathyroidism (HPT), renal tubular acidosis (RTA), hyperphosphaturia (HPU), hyperuricuria (HUCU), hyperuricemia (HUA) and hypocitraturia (HCI). Collections of 24-hour urine, with and without calcium load, were routinely analyzed for volume, pH, creatinine, calcium, uric acid, phosphorus and citrate. Serum levels were obtained of calcium, phosphorus, uric acid, creatinine and electrolytes.

Metabolic disorders were found in 70% of recurrent stone formers (HCU 59%, HCI 41%, HUCU 35%, HUA 18%, HPT 3.7%, RTA 1.8%, HPU 1.8%) and in 73% of patients with a single stone episode (HCU 59%, HUCU 37%, HCI 29%, HUA 9%). Based on these findings, recurrent stone formers received either drug therapy (allopurinol, thiazide, orthophosphate, alkali citrate; 70%) or dietary recommendations (30%) for a mean of 45.7 months. An overall recurrence rate of 67% over the observation period documents the failure of this strategy.

Since single and recurrent stone formers exhibited metabolic disorders to the same extent, urinalysis in calcium urolithiasis can be confined to excluding HPT and RTA. Excretion of calcium, uric acid or citrate says nothing about an increased risk of recurrent stone formation. The effect of correcting these 'metabolic disorders' diagnosed by urinary findings tends to be cosmetic rather than clinical.

Inhibitors and promotors of crystallization

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Title: **In-vitro-systems for the investigation of cristallization**

Author(s): **J.M. Baumann**
Institution: **Dept. of Urology, Regionalspital, Biel-Bienne, Switzerland**

State of the art lecture
(no abstract submitted)

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Title: **Clinical significance of inhibitors and promotors**

Author(s): **P.O. Schwille**
Institution: **Dept. of Experimental Surgery, University of Erlangen, Germany**

State of the art lecture
(no abstract submitted)

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Title: CRYSTALLIZATION OF CALCIUM OXALATE EVALUATED BY MICROSCOPY AND IMAGE ANALYSIS - PRELIMINARY RESULTS IN UNDILUTED URINE OF CONTROLS AND RENAL CALCIUM STONE PATIENTS

Author(s): J. Fan, P.O. Schwille, M. Manoharan, A. Schmiedl
Institution: Departments of Surgery and Urology, University of Erlangen, Germany

Healthy subjects and patients with idiopathic recurrent calcium urolithiasis (RCU) differ with respect to risk factors for calcium oxalate (CaOx) stone formation, such as CaOx urine supersaturation, crystal growth, aggregation, and possibly CaOx nucleation the contribution of which is less well understood. Available test procedures are elaborate and suffer from disadvantages such as poor practicability.

Methods: An easy-to-handle closed system, consisting of 0.5 - 1.0 ml undiluted urine agitated at 37 °C, was set up; this urine served as reservoir for evaluation by light microscopy and computerized image analysis of 25 µl aliquots in a blood cell counting chamber, in terms of oxalate concentration tolerated until spontaneous nucleation occurred, crystal growth during the subsequent 30 minutes, and development of at least two CaOx crystal aggregates per field. Working hypotheses, to be confirmed or rejected, were: 1) nucleation depends on initially present promoters (such as CaOx supersaturation) and inhibitors (such as citrate); 2) crystal size (syn. growth) and aggregation time depend on residual pools of promoters and inhibitors; 3) differences between male controls and male age-matched RCU patients (mean age 31 years) are significant.

Results: The test is specific for CaOx crystallization (mostly weddellite), sensitive (detection of crystals >2 µm), practicable for multiple samples, uses generally available technology, shows favorable cost-benefit ratio, allows to characterize controls and untreated RCU (table), recognizes effects of anti-stone medication by alkali and alkaline earth citrates (1). At nucleation, solution depletion (oxalate, calcium) is greater in RCU, being commensurable with more crystals formed in these than in controls. Tolerated oxalate concentration mainly reflects CaOx supersaturation, whereas growth and aggregation appear related to crystal number and size, thereby sufficing the rules of surface-controlled processes.

	n	pH	[Ca][1]	[Ox]	[Cit]	[Mg]	Cit/Ca[2]	Mg/Ca	T-Ox-N[3]	CD-30[4]	CA[5]	RSP-CaOx[6]
Controls[7]	18	5.83	3.56	0.13	1.56	3.15	0.48	1.19	0.57	4.43	52.5	1.7
RCU[7]	12	5.32[b]	7.52[b]	0.19[a]	1.53	4.09[a]	0.22[b]	0.67[b]	0.33[b]	6.19[b]	36.3[b]	2.7[b]

[1]: mmol l^{-1}; [2]: molar ratio; [3]: total oxalate at nucleation (µmol ml^{-1}); [4]: maximal crystal diameter (µm); [5]: crystal aggregation (minutes post-nucleation); [6]: relative supersaturation product (Δ G); [7]: mean values are given; [a]: p<0.05; [b]: p<0.01 vs Controls

Conclusions: Existing sophisticated technology for evaluation of CaOx nucleation, growth, aggregation, may be supplemented by simplified test systems. Based on comparative data from control and RCU individuals, recommendation of the present technique for broader use in clinical laboratories may be justified. The true physico-chemical interactions and associated reactions await in-depth clarification.

References: (1) Herrmann U, Schwille PO, Fan J, Manoharan M (1993); Manuscript in preparation

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Title: **ONE TIME-ONE ASSAY MEASUREMENTS OF HOMOGENEOUS NUCLEATION, GROWTH AND AGGREGATION OF CALCIUM OXALATE CRYSTALS.**

Author(s): U.Meinhardt, Ph.Jaeger, B.Hess
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Solutions of $CaCl_2$ (1.0 ml) and $K_2C_2O_4$ (1.0 ml), buffered at pH 5.7 (10 mM Na acetate) and containing 200 mM NaCl, were Millipore-filtered and added into a spectrophotometric cuvette (37° C, constant magnetic stirring of 500 rpm). Optical density was monitored at 620 nm (OD_{620}). The time from addition of the second solution until the first detectable increment of OD_{620} from zero baseline was taken as a measure of nucleation (T_n). Subsequently, the maximum slope of the increase of OD_{620} over time reflected growth (S_g), since it was mainly due to growing crystals. Once equilibrium, i.e. saturation, had been reached, OD_{620} steadily decreased, although total crystal mass was constant; the maximum decrease of OD_{620} over time (S_a) is a measure of aggregation (Hess B. et al., Am.J.Physiol.257: F99-F106, 1989). Total time for 1 experiment was 25 min. All experiments were performed at least in triplicate, values are mean ± SD. Various Ca/Ox ratios as well as effects of citrate (Cit) and albumin (ALB) were tested. Results were as follows:

	[Ca++]/[Ox--]		
	5.0 mM/0.75 mM	**5.0 mM/0.5 mM**	2.5 mM/0.5 mM
T_n (s)	45 ± 3	59 ± 7	329 ± 25
S_g (s^{-1})	13.6 ± 2.1	5.6 ± 0.8	2.6 ± 0.5
S_a (s^{-1})	-1.00 ± 0.13	-0.44 ± 0.06	-0.29 ± 0.04

At 5 mM Ca and 0.5 mM Ox (control), increasing [**Cit**] (0.5, 1.5 and 2.5 mM, respectively) progressively raised nucleation time (T_n 63 ± 5, 102 ± 28 and 153 ± 4 s, respectively) and lowered the rates of crystal growth (S_g 2.9 ± 0.3, 1.3 ± 0.2 and 0.8 ± 0.2 s^{-1}, respectively) and aggregation (S_a -0.26 ± 0.03, -0.22 ± 0.04 and -0.15 ± 0.05 s^{-1}, respectively). **ALB** at 2×10^{-7} M inhibited nucleation (T_n 73 ± 7 s, $p < 0.01$ vs. control), had no influence on growth, but inhibited aggregation (S_a -0.13 ± 0.03 s^{-1}, $p < 0.001$ vs control). In **conclusion**, highly supersaturated CaOx solutions allow for in vitro measurements of homogeneous nucleation, growth and aggregation of CaOx crystals in a single experiment. Under these supersaturated conditions, Cit reduces rates of nucleation, growth and aggregation of calcium oxalate crystals, whereas ALB slows nucleation and aggregation down.

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Title: Computer controlled and automatized measurement of the crystallization of Ca-salts in native whole urine

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Measurement of crystallization of Ca-salts by ion selective electrodes has turned out to be a very sensitive method. The known difficulties of handling these electrodes in native whole urine have been eliminated by commercially available calcium analyzers which clean and calibrate the electrodes after each measurement. However they do not allow kinetic tests.

In order to study also kinetics we developed a special sampler and a PC software. At intervals given by the computer program the sampler brings a thermostated crystallization chamber with continuous magnetic stirring to the analyzer (AVL). The computer controls the sampler as well as the analyzer, stores results and plots crystal growth versus time.

Results are obtained within 30 minutes. The measurement of calciumoxalate monohydrate seeded crystallization in native whole urine showed a coefficient of variation of 6,41%.

The method can also be used for calcium phosphate and to study nucleation.

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Title: THERMODYNAMIC AND KINETIC EFFECTS ON CRYSTAL GROWTH AS EVALUATED BY THE GEL CRYSTALLIZATION METHOD (GCM)

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The crystal growth of calcium oxalate in urine is governed by a series of parameters like supersaturation, ionic strength and concentrations of different inhibitors. Corresponding effects may be of thermodynamic (influence on supersaturation) or kinetic origin (influence by adsorption on crystal surfaces). Often, both of them act simultaneously. Calculation of relative supersaturation in urine, which is often used to estimate the risk of stone formation, reflects thermodynamic effects only. Therefore, it is of interest to gain information on the mechanisms underlying any reduction of crystal growth relevant in the genesis or therapy of urolithiasis.
Method: In this work, the microphotometric GCM (*Achilles; Scanning Microscopy 5, 1001-1017 (1991)*) was used to investigate the influence of relevant parameters on the relative crystal growth rate (Vcr) of calcium oxalate (COM). The first time, Ca was used as the component contained in the gel matrix. The following conditions were applied. Gel phase: 0.1 ml of 0.5 wt% agar-agar per well of a microplate; 8 mM $CaCl_2$, 0.2 mM Na_2Ox to produce seed crystals, and 10 mM MES-pH-buffer at pH 6. Measuring solutions were composed as simple as possible. They contained NaCl (50-400 mM), oxalate (0.1-1.0 mM) and pH-buffer. Effectors were investigated at 200 mM NaCl and 1 mM Na_2Ox.
Results: At first, the effect of total oxalate (Ox_T) on Vcr was quantified at different ionic strength (I; adjusted by NaCl). If expressed in terms of relative supersaturation, the dependence of Vcr on I disappeared nearly in correspondence with the thermodynamic character of this effect. The well-known inhibition by citrate and Mg could be accounted for by mixed thermodynamic and kinetic contributions. Quantitative differentiation was possible by comparison of theoretically calculated and experimentally determined crystal growth kinetics. The drastic inhibiting effect of polyphosphate on Vcr was of purely kinetic nature. Chondroitin-4-sulfate (CS) did not show significant inhibition if applied in physiological concentration. A mixed thermodynamic (binding of Ca) and kinetic effect by CS could be observed at higher concentration (>0.02 mM).
Conclusion: The results demonstrate that the GCM is applicable to the efficient and precise microdetermination of relative crystal growth rates of sparingly soluble crystal phases independently of the distribution of their phase components. Taking into regard complex chemical interaction in the gel/solution system, thermodynamic and kinetic contributions of growth effectors may be quantitatively differentiated.

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Title: QUANTIFICATION OF CRYSTAL GROWTH OF CALCIUM OXALATE BY A NEW FLOW MODEL OF CRYSTALLIZATION IN GELS (FMCG)

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It is well known that urinary stone formation occurs in a fixed, gel-like state from a flow of supersaturated urine. We developed a new model of crystallization in gels (FCMG) which simulates these physiological conditions as far as possible. The present paper reports on the application of this dynamic constant composition technique to the quantification of crystal growth of calcium oxalate monohydrate (COM) in a gel matrix.
Method: Artificial urine supersaturated with respect to COM was prepared by mixing of two suitably composed solutions. The mixture was conducted through channels containing a 1-wt% agar-agar (1.0 ml gel per channel) with COM-seed crystals (0.5 mM) which had been generated in the sol state by a pre-precipitation process. A flow of 0.5 ml/min per channel was maintained for 4 hours simultaneously in each out of 12 channels at 37°C. Relative crystal growth rates (Vcr) of COM were quantified in gel by discontinuous hourly measurement of scattered light intensity using a micro-photometer equipped with scanning stage adapted for microtiter plates.
Results: Experiments on the effects on crystal growth were carried out varying the following parameters in artificial urine: calcium (1-12 mM), oxalate (0.1-0.6 mM), citrate (0.5-0.8 mM), magnesium (0.75-12 mM), pH (4.5-6.5), and dilution (2.0-0.5).
Crystal growth kinetics (plots of growth rate as a function of the parameters under regard) were shown in terms of normalized quantities. From them, a series of interesting results could be derived. For instance, variation of normalized total oxalate concentration resulted in a much more pronounced alteration of Vcr than a comparable variation of total calcium. This underlines the predominant role of oxalate with respect to the risk of stone formation.
Variations of normalized concentration of citrate caused a similar effect than compatible alterations of magnesium on the Vcr.
Decreasing the pH of the system from 6.0 to 4.5 , Vcr increased from 1 to about 1.8.
Dilution or concentration of all components of the urine at the same time (simulation of diuresis) could be shown to result in considerable change of Vcr, which was quite similar to the effect of oxalate.
Conclusions: Applying the flow model of crystallizatin in gels (FCMG) described above the effects of all essential urinary parameters on one of the most essential processes of calcium stone formation may be quantified and directly compared.

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Title: IN-VITRO FORMATION OF SPHERULITES OF CALCIUM PHOSPHATE IN GEL - A NEW APPROACH TO THE STUDY OF CALCIUM UROLITHIASIS

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Calcium phosphate (CaP) has been found in more than 70% of urinary stones consisting predominantly of calcium oxalate (CaOx; *Leusmann et al.*). Furthermore, it is known that hydroxyapatite (HAP) acts as a heterogeneous nucleator for CaOx in-vivo. Therefore, CaP might play a crucial role with respect to the formation of Ca-stones in general. In-vivo, CaP often has the shape of spherulites, which could be demonstrated in the core of urinary stones as well as in sections of human kidneys (e.g. *Blaschke et al.*).
In a new flow model of crystallization, spherulites of CaP could be generated in a gel matrix of agar-agar (1 wt%) at 37°C from a supersaturated, metastable solution continuously flowing over the gel surface. Final concentrations were: 200 mM NaCl, 10 mM Na_2HPO_4, 10 mM NaH_2PO_4, 1.5-2 mM $CaCl_2$, pH 6.8-6.9. During a flow of 6 hours, spherulites reached a diameter of more than 100 µm.
Scanning electron microscopy revealed that the particles consisted of a likely microcrystalline core, which was partly surrounded by a well crystallized shell showing radially oriented sheetlike structures. Investigations on the kind of the different crystal phases involved are being in progress.
In experiments on different effectors of the process of CaP formation, phosphatidylcholine (PC) could be demonstrated to act as a very effective nucleator of CaP spherulites in the gel system used. At a physiological concentration of 20 mg/l in the solution mentioned above, PC caused massive formation of small, agglomerated particles of spheroid structure. No such effect could be found with phosphatidyl serine. Furthermore, we could show that the nucleating action of PC could be inhibited by human serum albumin (HSA). Nearly 100% inhibition of the nucleation by PC was accomplished at a concentration of HSA of 100 mg/l.
Conclusions: The flow model of crystallization used in this work has been proven to be of high physiological relevance. The CaP spherulites generated in the gel resemble strongly those formed in-vivo. Phospholipids, which have been demonstrated by others to be constituents of all mineralizing tissues with CaP crystal phases, could recently be shown to occur also in human urinary stones (*Khan et al.*). The technique applied here allows a quite new approach to basic problems of urinary stone formation.

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Title: **STUDY OF INHIBITORS OF CALCIUM OXALATE CRYSTAL GROWTH**

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The nucleation and crystal growth of calcium oxalate (CaOx) were studied at pH 5.5, at 37°C using turbidimetric measurements at 620 nm of suspensions produced by mixing calcium chloride and sodium oxalate (initial conditions for Ca, 4 $.10^{-3}$ M and Ox, 0.5 $.10^{-3}$ M). CaOx crystallization kinetics were defined by the induction time and then by the slope of turbidities as a function of time during the interval corresponding to a correlation coefficient $r^2 > 0.99$. Requiring only a small amount of material, the presently described technique is quick, reproducible and can be easily performed to study inhibitors of CaOx crystallization, by comparing the rate of crystal growth in the presence and absence of inhibitors. The effects on CaOx crystal growth of citric acid, tartaric acid, pyrophosphate (PPi), chondroïtin sulfate (CS) and Tamm-Horsfall glycoprotein (THP) isolated from urine of healthy subjects were examined. The majority of these compounds were inhibitors able to decrease turbidity slope at concentrations close to physiological urinary concentrations, except for THP, which had any effect on crystal growth in the range of concentrations tested (2.5 $.10^{-8}$ to 2.5 $.10^{-9}$ M). The order of inhibitory activity towards CaOx crystal growth was found to be CS > PPi > citric acid and isocitric acid > tartaric acid, according to the litterature. The inhibitors' behaviour regarding the medium composition was studied without any assumptions about the possible mechanisms of action. The measurements of ionized calcium before and after crystallization, as well as the observation of crystals using Scanning Electron Microscope allow us to formulate some hypothesis about the mechanism of action of the inhibitors tested. Citric acid exerts its overall effect through ion pairing, but an inhibition by surface phenomena is added to complexation. For isocitric acid and tartaric acid the mechanism is in part attributable to ion pairing, in part to crystal adsorption. For PPi and CS the latter mechanism is responsible for the inhibition effect.

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Title: INDUCTION OF CRYSTALLIZATION OF SPECIFIC CALCIUM OXALATE HYDRATES IN MICELLAR SOLUTIONS OF SURFACTANTS

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Calcium oxalate dihydrate, CaC204. (2+x)H20 (COD, x<0,5) is difficult to obtain from aqueous solutions but readily crystallizes from urine and artificial urine. In order to contribute to the understanding of this phenomenon, the influence of an anionic (sodium dodecyl sulphate, SDS), a nonionic (octaethylene glycol mono-n-hexadecyl ether, C16EO8), and a cationic surfactant (dodecyl ammonium chloride, DDACI) on the crystallization of calcium oxalate has been investigated under precipitation conditions similar to those in artificial urine (excesscalcium chloride, 0.3 M NaCl, 37 C). The kinetics of precipitation of calcium oxalate was followed by Coulter counter, and solid phases were characterized by polarized light microscopy, thermal analysis and X-ray diffraction powder patterns. Under the precipitation conditions employed, the thermodynamically stable monohydrate, CaC204H2O (COM) was the predominant crystalline form. SDS micelles retarded precipitation of COM but induced the crystallization of COD instead. It is shown that the effect is related to the critical micellar concentration (CMC) of the surfactant. Thus in the controlls and at c(SDS)<CMC >98mass% of COM and < 2mass% od COD crystallized while at c(SDS)>CMC the precipitates contained >85mass % COD. In contrast, DDACl, although strongly inhibiting COM crystallization did not induce the formation of COD. In the presence of micellar solutions of C16EO8 significant enhancement of the precipitation of COM was evidenced by higher initial precipitation rates and higher precipitate volumes and numbers of particles as compared to the controls. It may be assumed that the micelles of SDS an C16EO8 influence crystallization of calcium oxalate at a very early stage of solid phase formation, most probably at the nucleation stage. Natural surfactants present in urines (such as bile acids, etc.) may - by similar mechanismus - be involved in the induction of calcium oxalate crystalluria and urolithiasis.

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Title: THE INFLUENCE OF SURFACE ACTIVE AGENTS ON THE MORPHOLOGY OF CALCIUM OXALATE HYDRATES. A MOLECULAR RECOGNITION APPROACH.

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The monoclinic calcium oxalate monohydrate, CaC204H2O (COM crystallizes as hexagonal platelets while the tetragonal dihydrate, CaC204. (2+x)H2O (COD, x<0,5)appears in the form of octahedral bipyramids. However in crystalluria the same crystals assume very different morphologies. In order to explain this phenomenon calcium oxalate was crystallized in the presence of three different surfactants, the cationic sodium dodecyl sulphate, SDS, the anionic dodecyl ammonium chloride, DDACl and the nonionic octaethylene glycol mono-n-hexadecyl ether, C16EO8. The precipitation conditions were similar to those in artificial urine (excess calcium chloride, 0,3 M NaCl, 37 C). Crystals were indentified by X-ray diffraction powder patterns and changes in morphology were studied by scanning electron microscopy.

All three surfactants affected the morphology of COM crystals, but each influenced it in a different way. This shows that each of the surfactants recognized different crystal faces which has been explained by consideration of the ionic structure of the affected face(s). The morphology of COD crystals which formed in the presence of SDS micelles was also strongly but nonspecifically affected.

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Title: EFFECTS OF A NEW SEMI-SYNTHETIC POLYSACCHARIDE(G872) ON CALCIUM OXALATE CRYSTALLIZATION IN A WHOLE URINE MEDIUM

Author(s): Boevé ER,Cao LC,de Bruijn WC,Robertson WG and Schröder FH
Institution: Dept. of Urology, Erasmus University, The Netherlands

The effects of a new semi-synthetic polysaccharide(G872) that have been prepared by the chemical modification of extracts from seaweed on calcium oxalate (CaOx) crystallization in a ultrafiltered urine medium (WM<5000 Da) were investigated using particle size analysis and zeta meter. In the same study the activities of G872 in single concentration (2 mg/l) were compared with those of another semi-synthetic similar compound, sodium pentosan polysulfate(SPP) that has been proposed by others as a potential drug for the treatment of CaOx stone-formation. In the present study both compounds showed a strong inhibitory effects on CaOx crystallization induced in the medium by giving a standard oxalate load. The dynamic alterations of crystal distribution in total volume and in total number suggested that G872 exhibited its inhibitory effect by making the growing sites and aggregation sites on the CaOx crystal surface. The order in values of zeta potential at 20 min of incubation after giving a mount of standard oxalate load into the pooled urines with or without GAG is:

G872(-37.0±6.5mv) > SPP(-28.0±3.3mv) > Control(-21.0±6.6mv)

From the kinetic alterations of the zeta potential of the crystals, we speculate that the inhibitory activity of theses compounds could have something to do, not only, with their affinity to crystal surface, but also with their multidimensional structure in the medium. Finding and synthesizing new protective materials for stabilizing urinary colloidal system could be of a new approach for preventing stone formation and stone recurrence.

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Title: EFFECT OF TWO NEW POLYSACCHARIDES ON THE ZETA POTENTIAL OF CALCIUM PHOSPHATE CRYSTAL AND ON GROWTH AND AGGLOMERATION

Author(s): Boevé ER,Cao LC,de Bruijn WC,Robertson WG and Schröder FH
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The effects of a new semi-synthetic polysaccharides (G871,G872) on the crystal growth and agglomeration of calcium oxalate monohydrate (COM) crystals, have been reported (See J Urol 147:1643-1646,1992). In the present study, the effects of the same materials on the crystallization of calcium phosphates including calcium hydroxyapatite (HAP) and brushite (DCPD) were studied in artificial urine in vitro. A modified constant composition model and a seeded crystal growth based on particle size analysis were used to independently measure the rate of crystal growth and the degree of crystal agglomeration. The zeta potential on the interface between a crystal and its surrounding solution was measured using a Coulter DELSA 440 doppler electrophoretic light scattering analyzer. The previously reported heparin-like compound, sodium pentosan polysulphate (SPP). was studied as a reference inhibitor of crystallization using the same systems. All three substances have a concentration-dependent effect on the zeta potential and on the crystal growth rate of HAP in artificial urine. SPP had no the dose-response effect either on seeded DCPD crystal growth and agglomeration, or on the zeta potential of the crystals. In contrast, G872 showed a 50% reduction of DCPD crystal agglomeration at 3.2 mg/L and G871 at 6.0 mg/L. The effect of G871 and G872 on crystal growth is similar. G872 had a significant effect than either G871 or SPP on all the measured parameter. It is suggested that semi-synthetic polysaccharides such as those reported in this study can be of use in the prevention of calcium urolithiasis, not only in calcium oxalate, but also in calcium phosphate.

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Title: CLINICAL SIGNIFICANCE OF CITRATE EXCRETION IN RENAL LITHIASIS IN A LATINAMERICAN COMMUNITY.

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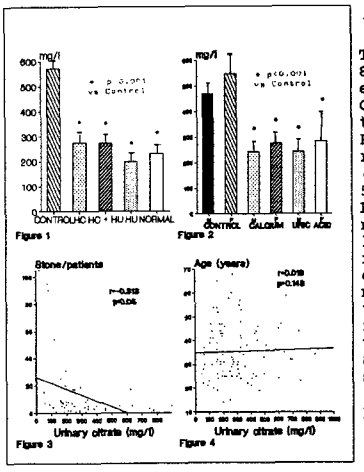
Urinary citrate is a known crystal inhibitor of calcium phosphate and oxalate. We report values of urinary citrate and its clinical implications in renal lithiasis patients (RL).
MATERIAL AND METHODS. 106 consecutive RL, age 33.2±2 years(14 to 68), 40 men and 66 women were submited to metabolic evaluation.



Type of stone were determined in 83 RL (calcium 65,uric acid 14, estruvite 3 and cystine 1). Citrate was measured by enzymatic method, Boehringer Mannheim RESULTS. RL showed a marked reduction in citrate excretion (286± 18 mg/l vs control group 523±49 mg/l p<0.001). 55.6% of RL had hypocitraturia (<320 mg/l). Main metabolic abnormalities in calcium RL were:hypercalciuria 41%,hyperuricosuria 15%,both 16%, and "normal" 12%. Citrate excretion in these abnormalities is shown in fig 1. In fig 2 citrate excretion according to type of stone and sex is shown. Fig 3 and 4 show correlation between citrate excretion and total of stones per patient or age respectively

CONCLUSION. Our population of RL showed a marked reduction in urinary citrate excretion, independently of the type of stone passed, age, sex or metabolic disturbance. Levels of citrate are negatively correlated with total of stones per patient, suggesting that low urinary citrate is associated with more aggressive stone disease.

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Title: URINARY MAGNESIUM EXCRETION (uMg) IN CALCIUM NEPHROLITHIASIS (CN).

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Mg is an inhibitor of calcium-oxalate (Ca-Ox) crystals formation, due to its complexation with Ox. In rats, Mg depletion leads to hyperoxaluria by enhancing both the endogenous synthesis and intestinal absorption of Ox. Clinical data on uMg in CN are controversial because the groups of examined patients (pts) were not homogeneous among previous studies. The aim of this study was to evaluate uMg in subgroups of CN pts.

159 subjects were studied: 40 normals (M 23, F 17) and 119 CN pts, 26 (M 13, F 19) of which with medullary sponge kidney (MSK), and 93 (M 74, F 19) with idiopathic CN. All subjects underwent a 24 hour urine collection, while on a free diet, in order to evaluate uCa, uMg, uOx, urinary sodium (uNa) and creatinine (uCr). The pts with idiopathic CN were classified in two groups: a) with hypercalciuria (uCa ≥4 mg/kg/day) (n = 38; M 30, F 8) and b) with normal uCa (n = 55; M 44, F 11).

uMg/Cr ratio, the most reliable index of Mg excretion, was lower in normocalciurics than in normals, hypercalciurics and MSK pts ($p < 0.001$ in all cases). uMg was not correlated with uOx in the four groups; it was directly correlated with uCa in all groups, except hypercalciurics, confirming an impaired Ca tubular reabsorption in such pts.

In conclusion, low uMg was detected only in normocalciuric stone formers, in whom, therefore, Mg supplements are suggested as a prophylactic approach. Hyperoxaluria,secondary to Mg depletion, was not confirmed in this clinical study, probably because a Mg depletion in humans is rare, being dietary Mg ubiquitarious.

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Title: Do urates play a major role in calcium oxalate precipitation?

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The role of uric acid and urates in the precipitation of calcium oxalate monohydrates (COM) is still unclear. Heterogeneous nucleation has been investigated by the constant composition method. $Ca(HU)_2$, NaHU and COM were used as seed crystals; the pH was kept konstant at pH 7,0 by means of a PIPES buffer.
Homogeneous precipitation was studied in non-equilibrium systems by using the "complex acidification" method. The concentrations were chosen such that precipitation of COM and $Ca(HU)_2$ would start at pH 7. By this method pure Ca-salts (COM and $Ca(HU)_2$) as well as mixed precipitates were prepared. All precipitates were characterized by x-ray powder diffraction and IR spectroscopy.

Results: COM crystallisation from supersaturated solution could be induced by all seed crystals. The induction period was very short for the addition of COM but lasted several hours using $Ca(HU)_2$ or NaHU.
Coprecipitation by the complex acidifications approach showed that at physiological $n(HU^-):n(C_2O_4^{=})$ ratios the precipitates contained Ca, oxalate and urate but should x-ray diffraction diagrams different from those of COM and $Ca(HU)_2$ indicating that a new, though unidentified phase had been formed with a very low degree of cristallinity.

Our experiments indicate that urates do not play a major role in COM lithiasis.

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Title: **URINARY COPPER AND PHOSPHORUS CONCENTRATIONS: ARE THEY LITHOGENIC FACTORS IN KIDNEY STONE FORMATION?**

Author(s): **A. Rodgers, L. Barbour, M. Pougnet, C. Lombard, R. Ryall**
Institution: **Department of Chemistry, University of Cape Town, South Africa**

Since the presence of any element in a kidney stone suggests that it may have fulfilled some determinant role in the stone's pathogenesis, the present study was undertaken to establish whether differences exist between stone formers and controls with respect to their excretion of various elements.

Nineteen male calcium oxalate stone formers and twenty healthy male controls participated in the study. Early morning urines were collected on 3 consecutive mornings (Mon, Tues, Wed) to compensate for random daily fluctuations in urine composition. The concentrations of several elements were determined by inductively coupled plasma atomic emission spectroscopy and graphite furnace atomic absorption spectroscopy. Data were treated using multivariate statistical methods.

The results showed that the urinary concentrations of several elements varied on a daily basis. However only Cu and P values were significantly different *on all three days*: Cu concentrations were significantly higher in the stone formers while P values were significantly lower.

It is interesting to speculate how these two elements might be determinant factors in urolithiasis. Cu occurrs in approximately 80% of stones analysed for its presence and its concentration in such calculi is approximately 1000 times greater than in urine, suggesting that it fulfills an active promotory role in stone formation. Indeed *in vitro* studies have shown that Cu promotes calcium oxalate dihydrate formation. Another possibility is that Cu could become incorporated into the gross architecture of a stone by virtue of it being a structural component of protein molecules which are released into the urine in response to cellular injury caused by the growing stone and which themselves then bind to the crystal surfaces. Phosphorus is a major constituent of renal calculi where its concentration far exceeds that of urine. Although it is reasonable to assume that an abnormally high renal excretion of phosphorus would increase the likelihood of phosphate precipitation our results show that stone formers have significantly *lower* values. Similar results have been reported by other workers. It has been hypothesized that low urinary phosphate excretion is indicative of high urinary Ca excretion. Conversely, it has been shown that urinary Ca decreases during P supplementation. We suggest that the lower urinary concentrations of P in stone formers might also be indicative of lower concentrations of pyrophosphate, a recognised inhibitor of calcium oxalate crystallization.

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Title: **THE RELATIONSHIP BETWEEN URATE SEEDS AND CALCIUM OXALATE CRYSTALLIZATION: EPITAXIS OR ENDOCRYSTALLOSIS?**

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The process of epitaxy has long been invoked to explain empirical clinical evidence suggesting the existence of a relationship between hyperuricosuria and calcium oxalate (CaOx) stone formation. However, although crystals of uric acid (UA) and sodium urate (NaU) have been shown to induce the deposition of CaOx from inorganic solutions, a similar effect has not been unequivocally demonstrated to occur in human urine. The aim of this study was to determine whether seed crystals of UA or NaU promote the deposition of CaOx on their surface in undiluted human urine.

24-hour urine specimens were collected from healthy men, pooled, centrifuged (10,000xg) and filtered (0.22µm). The metastable limit with respect to CaOx was determined by titration with oxalate. A portion of the sample was retained as control: the remainder was divided into three aliquots and seeds of UA, NaU or CaOx were added to give a final concentration of 6mg/100mL. Crystallization of CaOx was then induced by the addition of 15µmoles/100mL of oxalate in excess of the measured metastable limit, followed by incubation in a shaking water bath for 2 hours at 37°C. The number, size and volume (corrected to account for the contribution of seed crystals) of crystalline particles were determined using a Coulter Counter, and the crystals were examined by scanning electron microscopy. The precipitation of CaOx was independently determined in parallel experiments by ^{14}C-oxalate deposition. Results were compared using the Wilcoxon signed rank test.

Nucleation of CaOx crystals occurred in all the urine samples. In all cases, the addition of seed crystals increased the corrected particle volume deposited at 120min in comparison with a control containing no seeds; UA seeds increased the corrected volume by 13.6%, NaU by 56.8%, and CaOx by 206.5%. However, the ^{14}C-oxalate data demonstrated that these increases were not a result of a promotion of CaOx deposition, except in the case of the CaOx seeds. The deposition of CaOx relative to the control was 1.4% ($P<0.05$) in the presence of UA seeds, 5.2% ($P<0.01$)with NaU seeds and 54.0% ($P<0.001$) with CaOx. Scanning electron microscopic examination of the precipitated crystals showed that large aggregates of CaOx were formed in the presence of CaOx seeds, which themselves were not visible, presumably because they had been enveloped by deposition of CaOx upon them. In contrast, urate seeds were clearly visible, being scattered like barnacles upon the surface of the CaOx crystals. Many had been engulfed by the CaOx growth front, and were evident as protuberances upon the surface of the CaOx crystals, an observation which would explain the disparity between the Coulter Counter and ^{14}C oxalate data. It was notable that the overall size of the crystalline particles was markedly reduced in the presence of the NaU seeds, suggesting that their binding to CaOx crystals had decreased the tendency of the latter to aggregate.

It was concluded that, since seed crystals of NaU and UA do not promote CaOx deposition to a physiologically significant degree in urine, epitaxy is unlikely to contribute to stone formation. However, binding of NaU and UA crystals to, and their subsequent enclosure ("endocrystallosis") within actively growing CaOx crystals would occur *in vivo*, thereby increasing the overall volume of crystalline matter precipitated and explaining the occurrence of mixed urate/oxalate stones.

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Title: THE IMPACT OF SIALIC ACID ON STONE FORMATION

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Sialic acid (acylneuraminic acids) forms part of the uromucoid, which acts as a promotor of stone formation, and is also observed in stone matrix. Moreover, these substances appear to ficilitate binding of cationic compounds to macromolecules and cells. In spite of these interesting facts, an impact of these substances on stone formation has not yet been sufficiently verified. In the present study, sialic acids were investigated in 24-hour urine collections of 103 stone formers and 35 healthy individuals as well as in cell culture supernatants derived from non-stone-forming kidneys.

Free sialic acid before and after acid hydrolysis was determined using the periodate acid and thiobarbituric acid reaction. To eliminate interference of other substances - e.g. 2-deoxyribose - in this assay, measurement was performed using two different wavelengths, and correction of values was calculated.

There was no statistical difference regarding the concentrations of total sialic acid between the two groups, although 24-hour urine excretion differed significantly ($p < 0.005$). Stone formers revealed lower concentrations of free and higher levels of bound sialic acids. The ratio of free to bound sialic acids in healthy individuals versus stone formers was 1.83 and 0.9, respectively. Sialic acids were also detectable in the cell culture supernatants, which demonstrates the capacity of renal cells to produce these substances.

As a preliminary explantation of our findings, we hypothesize that sialic acid are of importance in stone formation because (i) bound sialic acid increase the intrinsic viscosity of mucous secretions in the urogenital tract; (ii) the strong binding activity for calcium ions causes high local calcium accumulation on the cell surface which might act as nucleus for stone formation; and (iii) free sialic acid binds calcium ions, therefore reducing free calcium ions in the urine.

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Title: Structure and Expression of the mRNA Encoding Urinary Stone Protein (Osteopontin)

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The chemical nature of urinary stone protein is poorly understood. We have sequenced a cDNA of urinary calcium oxalate stone protein . cDNA sequences showed complete homology between urinary stone protein and human osteopontin. Osteopontin protein was detected by staining with Stains-All, demonstrating that only calcium oxalate and calcium phosphate stones consist of osteopontin protein. We used a technique of in situ hybridization to detect osteopontin mRNA in the kidney. In control rats, distal tubular cells were sporadically positive ,and proximal tubular cells and glomeruli were negative for osteopontin mRNA. In stone-forming rats, staining of distal tubular cells was remarkably increased, but proximal tubular cells and glomeruli were still negative. Northern blot analysis showed a significant increase of osteopontin mRNA in stone-forming rats in proportion to the dosage and the duration of the stone inducing drugs. These results show that osteopontin in the kidney is presumably involved in urinary stone formation as the stone matrix.

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Title: SEQUENCING OF URINARY STONE PROTEIN, WHICH IS IDENTICAL TO ALPHA-ONE ANTITRIPSIN

Author(s): Tohru UMEKAWA, Kenjiro KOHRI, Takanori YAMATE, Naoya AMASAKI, Takashi KURITA
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We have extracted and refined urinary stone proteins. The protein materials showed well difined three bands when analyzed by sodium dodecyl sulfate (SDS) gel electrophoresis, the amino acid sequence of them is the same as that human alpha-one antitripsin (From 22nd to 41st amino acids of NH_2-terminal). Alpha-one antitripsin is a most popular anti elastase which has been implicated in playing an important role in inflamatory lesions. The finding of alpha-one antitripsin (AT) opens up the possibility that proteolysis mighgt be involved in the stone forming process. It was presumably involved in stone formation as coprecipitating substance because equilibrium dialysis revealed that AT has no affinity for calcium.

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Title: **FTIR SPECTROSCOPIC INVESTIGATIONS OF TAMM-HORSFALL PROTEINS FROM RECURRENT STONE FORMERS AND HEALTHY PROBANDS**

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Institution:

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Tamm-Horsfall Protein (THP) has been tested for its effect on the precipitation of calcium oxalate by means of the Oxalate-Tolerance Test. In this test system THP from healthy probands inhibited the precipitation of calcium oxalate whereas recurrent stone formers' THP had no or even a promoting effect. To elucidate possible structural differences between the two functional different THPs we used Fourier transform infrared (FTIR) spectroscopy, which has become a powerful tool for the investigation of protein secondary structure by the amide I band (1700-1600 cm^{-1}). The analysis of proteins in aqueous solutions has proved to be extremely difficult due to the strong absorption of water in this region. Spectra were therefore obtained in D_2O solutions in the region between 1850 and 1300 cm^{-1}. The spectra of glycoproteins show a number of overlapping bands which may be assigned to vibrations of the protein and the carbohydrate moiety. The high degree of glycosylation of THP ($\approx$30%) made it difficult to analyse the protein secondary structure but spectra of THP from stone formers and healthy probands showed differences which could be explained by an altered nature of glycosylation. THP of healthy probands showed strong carboxylate vibrations which are assumed to result from a high degree of glycosylation with hexuronic acids whereas stone formers showed less or even no carboxylate vibrations in the spectrum.

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Title: **Influence of N-acetylglucosamine on urinary mucopolysaccaride and mucoprotein excretion in healthy volunteers**

Author(s): K.-H. Bichler, S. Kleinknecht, C. Schipke, C. Stahl, U. Speck
Institution: Dept. of Urology and Institut of Pathology, Eberhard-Karls-University Tübingen, Germany

There are several indications in the literature on clinical pictures accompanied by reduced excretion of inhibitors of lithogenesis and of protective substances of the efferent urinary tract such as complex carbohydrates, proteoglycans, glycoproteins and glycolipides, which might result from a diminished performance of the organism to synthesize these substances. Application of their basic components may lead to an increased production and excretion of mucoproteins (MP) and mucopolysaccharides (MPS). Accordingly, urinary glycosaminoglycan (GAG) excretion can increase after administration of the GAG-component Glycosamine sulfate.
The objective of the study on hand was to test whether N-acetylglucosamine has an effect on urinary MP and MPS excretion in healthy subjects. In a double-blind trial 5 healthy subjects were given 400 mg of N-acetylglucosamine 3 times a day for 10 days; 5 test persons were given placebo. 24h urines were collected prior to treatment, on day 7 and 10 of treatment and 7 days after treatment. We recorded urine volume, pH, creatinine, total protein, GAG and Tamm-Horsfall-Protein (THP-=uromucoid).
THP excretion increased markedly in 2 persons of the verum group, while in the other 3 persons, as in the control group, there was no consistent change of THP excretion. On the average, compared to initial values THP increased about 15% in the verum group. The other parameters tested were not influenced by application of N-acetylglucosamine.
Although urinary THP-excretion tendentiously increased in the verum group, for definite proof of an influence of N-acteylglucosamine on MP synthesis and excretion data from larger collectives should be obtained.

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Title: URINARY RISK FACTORS FOR STONES AND OESTROGENIC HORMONES

Author(s): A.Trinchieri,F.Rovera,G.Zanetti,A.Guarneri,F.Colombo
Institution: Istituto di Urologia,Univ.di Milano (Dir.Prof.E.Pisani)

Idiopathic calcium stone disease occurs mainly in men,the male/female ratio being about 4 to 1.
The lower risk of stones in women than in men may be attributable to the influence of oestrogenic hormones.In fact at least one urinary inhibitor,citrate,has been reported to be excreted in greater amounts during hormone administration.
The possible influence of oestrogenic hormones on the propensity to form calcium renal stones has been examined in female renal stone formers and the results are presented in this paper.
Observations were made on 12 premenopausal and 27 postmenopausal female renal stone formers.Furthermore we studied 5 patients using oral contraception.
Twentyfour hour urines were collected in order to measure potassium, sodium,calcium,magnesium,phosphate,oxalate,urate,citrate,glycosoaminoglycans (GAGs),creatinine and pH;in premenopausal women urine samples were collected during different phases of the menstrual cycle Specimens of venous blood were obtained over the same period from each patient to measure serum estradiol and progesterone.The urinary outputs of electrolytes were unrelated to normal oestrogenic and progestegenic cycle surges.Neither were any significant changes in postmenopausal women and in those using oral contraception.

	Oestr.	Prog.	Postmenopausal	Pill
Ca ur (mg/day)	216±89	203±87	214±34	174±101
Cit ur(mg/day)	367±215	437±239	392±151	401±295

In conclusion our results would suggest that the relatively low incidence of calcium renal stones in women,compared with men,is not attributable to the influence of oestrogenic hormones on the urinary risk factors measured in this study.A study of other substances contained in female urine should provide more definitive information.

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Title: **Steroid hormones, glucagon, and urolithiasis in the portacaval shunt rat**

Author(s): A. Heidenreich[1], U. Engelmann[2], R. Bonfig[1], K. H. Bichler[1]
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Following the publication of a reliable microsurgical technique for portacaval anastomosis (PCA) in rats numerous biological and biochemical consequences have been studied extensively. However, few studies investigating the pathogenesis of uric acid urolithiasis following PCA have mainly focused on chemical stone analysis. In previous studies we could demonstrate a sex dependant urolithiasis with formation of potassium-hydrogen-urate (PHU) stones predominately in male rats. The following study investigates the influence of testosterone, estradiol, and glucagon on stone formation in the PCA rat.

82 male and 71 female rats were studied after creation of a PCA. 73% of male rats and nil of the females developed uroithiasis. In 90% PHU stones were formed. Male rats showed a significant decrease of total and free testosterone (T) serum levels postoperatively with concentrations of 51.2+11.2 ng/ml and 0.03+0.001 pg/ml (preop.: 860.4+112.3 ng/ml and 10.9+1.2 pg/ml, resp.). Serum levels of estradiol (E) increased significantly from 0.89+0.1 pg/ml to 21.4+1.1 pg/ml (p 0.01). Female PCA rats showed a significant increase in free and total T levels from 0.45+0.09 pg/ml and 90.5+20.4 pg/ml to 49.6+5.7 pg/ml and 7124.3+146.9 pg/ml, resp. (p 0.01). E conentrations increased from 16.3+1.4 pg/ml to 45.9+5.7 pg/ml (p 0.01). Glucagon levels showed postoperative alterations with an increase in males from 94.2+9.2 pg/ml to 744.2+2262.1 pg/ml (p 0.01) and from 137.2+5.5 pg/ml to 369.7+56.9 pg/ml in females. There was a remarkable correlation between glucagon plsma levels and stone formation: male stone forming rats had significant higher plasma levels than non-stone forming male rats, female PCA rats and sham operated rats. The median glucagon concentration in the stone forming group was 733.9 pg/ml compared to 358.2 pg/ml (p 0.01) in the non stone forming group.

Our findings suggest that hormonal alterations might contribute to sex dependant stone formation in the PCA rat. Low androgen levelsmight interfere with biosynthesis of urinary inhibitors of uric acid lithiasis causing a lack of glycosaminoglycanes. Similar changes in stone formation rate have been observed in male cats after castration. In humans the sex dependant excretion of citrate is well known. An interesting correlation between stone formation and glucagon was discovered. Stone forming male rats revealed significantly higher hormone levels compared to non stone forming animals. Glucagon might influence urinary uric acid excretion by inhibition of tubular reabsorption.

Calcium metabolism and related problems

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Title: **Calcium metabolism and urolithiasis**

Author(s): **Ph. Jaeger**
Institution: **Poliklinic of Medicine, University of Bern, Switzerland**

State of the art lecture
(no abstract submitted)

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Title: **Renal tubular calcium transport**

Author(s): **J.H. Dirks**
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State of the art lecture
(no abstract submitted)

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Title: INFLUENCE OF ORAL PROTEIN INTAKE ON THE CALCIUM EXCRETION AND THE GLOMERULAR FILTRATION RATE

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A number of epidemiological studies suggest a relation between the daily protein intake and the risk of upper tract stone formation [Urol Int 37: 394-399, 1982]. The reason for this correlation is unknown. On the other hand a high urinary calcium excretion is known to predispose to stone formation. Therefore the aim of our study was to investigate the influence of the oral protein intake on the calcium excretion. Since the glomerular filtration rate is highly dependant on the daily protein intake, endogenous creatinine clearance and inulin clearance were further parameters of our study. 19 healthy subjects were investigated eating for 1 week a protein content of 80 g/d, following a period of 2 weeks with 30 g/d and 200 g/d respectively. During the 80g/d period the calcium excretion showed a mean value of 4.51±0.42 mmol/l. During the 30g/d period it decreased to 2.64±0.35 mmol/l and increased again to 6.62±0.75 mmol/l during the 200 g/d period. The time course of creatinine- and inulin clearance was parallel to the one of the calcium excretion [80g/d period: 121.5±5.76 ml/min; 30 g/d period: 105±4.66 ml/min; 200g/d period: 144±7.29 ml/min]. Between the calcium excretion and the creatinine clearance a highly significant correlation was found. Beside a direct influence of the glomerular filtration rate on the calcium excretion a common protein induced mechanism has to be discussed as a possible reason for the observed phenomenon.

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Title: ON THE CALCIURIC RESPONSE TO ACID LOADING

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The purpose of the present study was to evaluate the calciuric response to acute acid loading in healthy subjects and renal calcium stone formers during a standardized metabolic regime.

Material and methods. Ten consecutive male idiopathic calcium stone formers (ICSF) and 10 healthy men matched for age and body mass index underwent two ammonium chloride (NH_4Cl) loading studies using 2 mmol and 4 mmol NH_4Cl (oral) per kg body mass. The loading studies were preceded by an eight hour fasting period, and for 24 hours prior to the fasting periods the participants were on a standardized calcium-fixed diet. One hour before and 5 hours after the administration of NH_4Cl urine was collected for measurement of pH, Ca and creatinine. Venous and arterialized capillary blood samples were collected hourly for measurement of pH, bicarbonate, base excess, Ca^{2+}, and creatinine. 150 ml of demineralized water was given every hour during the test.

Results. The total urinary calcium excretion (5 hours) after the administration of 2 mmol NH_4Cl/kg body mass was 2.33 (±0.5) mmol in ICSF compared to 1.80 (±0.3) mmol in healthy subjects ($p=0.05$). After the administration of 4 mmol NH_4Cl/kg body mass the total urinary calcium excretion (5 hours) was 2.85 (±0.6) mmol in ICSF and 1.72 (±0.4) in healthy controls ($p=0.01$). There were no differences in blood acid base status, serum Ca^{2+}, serum albumin, and serum creatinine during NH_4Cl loading between the two groups.

Discussion and Conclusion. The calciuric response to acute acid loading was more pronounced in ICSF compared to healthy subjects. The calciuric response was dose-dependent in ISCF, whereas the calciuria in the healthy subjects did not increase by increasing the acid load. It is concluded that increased urinary calcium excretion in response to acid loading may be a contributory cause of hypercalciuria in patients with calcium nephrolithiasis. It is conceivable, therefore, that an increased stone risk due to a high intake of animal protein at least in part may arise via such a pathophysiological sequence.

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Title: CALCIUM BINDING BY CEREAL BRANS UNDER SIMULATED GASTROINTESTINAL pH-CONDITIONS: SIGNIFICANCE OF PHYTIC ACID

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Controlled intake of bran is thought to be an alternative therapy in the methaphylaxis of urolithiasis to decrease renal calcium excretion, if absorptive hypercalciuria is diagnosed. Phytic acid (PA), amounted in cereal brans up to 6%, is discussed as decisive factor for gastrointestinal binding of calcium, leading to a reduction of renal calcium excretion. However, in vivo studies reported on different efficacy of various brans and gave no evidence for a distinct correlation between PA content and effectiviness in decreasing renal calcium.

Therefore, calcium binding capability by cereal brans (rye-, fine wheat and heat treated coarse wheat bran) was investigated in vitro, simulating a gastro -> small intestinal pH-course. Incubation was performed in a $CaCl_2$-solution (0,625mmol Ca/ g bran and 50ml), starting with pH 2.2 resp. 3.0 and 4.0 for 0.5/1/1.5/3 h, each time followed by buffering steps to pH 6 and than pH 8. Calcium binding was determined using an isotope (^{45}Ca) labelling-technique. Simultaneously, solubility profile of PA and inorganic phosphate (inorg. PO_4) was examined.

For all brans about 80% of PA content was released in the $CaCl_2$-solution at pH 2.2. Starting with pH 3 and pH 4, resp., solubility of PA from coarse wheat bran was slightly increased. In contrast, for rye- and fine wheat bran a reduction of first dissolved PA occurred, rising with incubation time and higher pH (rye: max. reduction= 100% ; fine wheat: max. reduction= 80%). At the same time, content of dissolved inorg. PO_4 increased and corresponded directly to PA reduction. Buffering to pH 6 resulted in a complete remove of still existing dissolved PA, whereas the content of dissolved inorg. PO_4 remained nearly unchanged. Setting the pH to 8.0 inorg. PO_4 became almost insoluble. Calcium binding by all brans was low at gastric pH-values (2.2/3/4). Buffering to pH 6 led to a significant but variable increase of binding values, which was obviously correlated to the difference in solubility of PA between gastric pH and pH 6. Maximal reduction of binding at pH 6, caused by "break down" of PA, accounted 80% for rye bran and 60% in case of fine wheat bran. Buffering to pH 8, calcium binding by all brans increased significantly. Rise of binding was correlated to the difference in solubility of inorg. PO_4 between pH 6 and pH 8.

The results indicate that content of PA in a cereal bran is of great influence on its calcium binding capability. However, calcium binding at duodenal pH 6 (where in vivo absorption of calcium mainly takes place) depends chiefly on the quantity of still existing dissolved PA in the transitional phase between gastric and duodenal pH. In vivo studies have shown, that endogenous phytase found in bran is capable to hydrolyze phosphate ester of PA during gastrointestinal passage. Our in vitro investigation show, that the effect of endogenous phytase depends on incubation-pH and -time while enzyme activity basically varies within origin and processing of the bran. Therefore, calcium binding capability of cereal brans at duodenal pH can be strongly affected by phytase action which is influenced either by extrinsic factors and either by present gastric conditions. High content of PA in bran combined with a complete deactivation of endogenous phytase is possibly the most important prerequisite for a successful use of cereal brans in the metaphylaxis of urolithiasis accompanied by absorptive hypercalciuria.

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Title: Bone mineral density and calcium metabolism in stone patients

Author(s): K.H. Bichler, A. Heidenreich, W.L. Strohmaier, S. Kleinknecht, M. Kalchthaler
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The pathogenesis of hypercalciuria (hyperc.) has been attributed to increased intestinal absorption or increased renal loss of calcium. Therapy consists of low calcium diet or application of oral thiazides. However, the role of the skeleton in hyperc. and the possibility of bone derrangement as consequence of therapy remains controversial. In our study we assessed the vertebral bone mineral density (BMD) in patients (pts.) with hyperc. just before initiation of specific therapy and during follow-up in order to evaluate the role of bone metabolism of urolithiasis.
24 pts. with hyperc. and 13 normocalciuric pts. were studied. According to the results of a PAK-test 7 pts. were classified as renal hyperc., 12 as absorptive (abs.) and 5 pts. as idiopathic hyperc.. Each pt. had 2 serum and 2 24-hrs. urine collections for evaluation of calcium, phosphorous, osteocalcin, Vit.-D, parathormone and calcitonin. Pts. with abs. hyperc. were set on a low calcium diet, pts. with renal hyperc. received oral thiazides 2x100mg/d.
Results of BMD demonstrates significantly less bone mineral contents in renal hypercaliuria when compared to the control group. Results of BMD of pts. under therapy shows a bone mineral content of 90% and 88% in renal and abs. hyperc., resp. (ns). However, in 4 pts. with abs. hyperc. BMD shows a significant decrease of mineral content from normal values to 72%. A significant increase of mineral content to 110% could be demonstrated in 4 pts. with renal hyperc. under therapy. Pts. with abs. hyperc. revealed significant higher levels of 1,25-Vit.D; no correlation could be found between BMD results and serum parameters of calcium metabolism.
Our study suggests that BMD might be a useful tool in monitoring pts. with hyperc.. We could demonstrated that 60% of pts. with renal hypercaliuria present with osteoporosis due to renal calcium loss. Nevertheless bone derangement might successfully be treated by calcium sparing therapy. On the other hand pts. with abs. hyperc. are at risk of osseous calcium mobilisation during a low calcium diet. In these cases BMD demonstrates a significant decrease of bone mineral content indicating that severe changes of bone metabolism might occur and the diet has to be quit in order to prevent osteoporosis. In our opinion BMD is useful in monitoring all pts. with hyperc. prior to or under therapy.

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Title: BONE MINERAL DENSITY IN YOUNG ADULT, NORMOCALCAEMIC PATIENTS WITH CALCIUM UROLITHIASIS

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The data about bone mineral content in patients with urolithiasis are scanty.
Bone mineral density (BMD) was determined in lumbar spine and femoral neck with a dual energy x-ray absorptiometry technique (Lunar DPX) in 32 normocalcaemic, recurrent calcium stone formers, aged 20-40 years.
Each subject collected a 24-urine sample on free diet. Morning blood samples for determining serum calcium, phosphate and PTH levels were taken after overnight fasting. The patients were assigned as hypercalciuric when daily calcium excretion exceeded 7.5 mmol in males and 6.25 mmol in females.
BMD results were expressed as Z-score (SD from mean BMD of age and sex matched controls).
The results showed that marked osteopenia is not a rare finding in stone formers with hypercalciuria.
Thus BMD measurement should be included in diagnostic protocols of these patients.

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Title: **W.L. Strohmaier, K.-H. Bichler**

Author(s): **Cellular aspects of stone formation**
Institution: **Dept. of Urology, Eberhard-Karls-University Tübingen**

State of the art lecture
(no abstract submitted)

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Title: **Erythrocyte Ca Transports in Idiopathic Hypercalciuria**

Author(s): F. Francesca, G. Vezzoli, M.C. Reina, M. Grasso, P Casirati, T.Stellato, P.Bollini, P. Rigatti, G. Bianchi.
Institution: Department of Nephrology and Department of Urology, H S Raffaele, University of Milan.

In order to evaluate cellular Ca metabolism in the patients with idiopathic hypercalciuria (IH), we studied plasma membrane Ca pump (Ca-ATPase) and passive influx of bivalent cations in the erythrocytes (RBC) of 15 hypercalciuric patients with Ca-oxalate nephrolithiasis and in 20 normocalciuric subjects with Ca-oxalate nephrolitiasis (UCa<0.1 mM/kg body weight per day). Ca-ATPase was measured at Vmax as ATP hidrolysed by erythrocyte membranes in the presence of free Ca 10 μM and Calmodulin 1 μg/ml. Ca influx was evaluated as Sr influx into RBC incubated in isotonic medium with Sr 3 mM and Vanadate 3 mM (Ca-ATPase inhibitor). Electrolytes were determined in plasma and urine; PTH was measured in plasma; the intestinal Ca absorption was evaluated as absorbed fraction of an oral load of stable Sr (2.65 mg per kg of body weight) after 1 and 4 hours.

Our results showed a significatively higher Ca-ATPase activity in the patients with IH (54±4.5 vs 43±3.8 nmoles ATP split / min per mg of proteins [mv±SE]; p<0.05). Sr influx was not different in the two groups (4.6±0.6 vs 4.7±0.6 μmoles / l RBC per min). Intestinal Sr absorption was higher in the patients with IH at the first hour (16.9±1.3% vs 13.5±1%; p<0.05), but not at the fourth hour (20±1.6% vs 19±1.2%). Moreover the patients with IH showed Na, K and P excretion more elevated than in normocalciuric subjects.

These results suggest that IH is associated with increased values of Ca-ATPase activity in erythrocyte plasma membrane. As erythrocyte Sr influx, a marker of Ca influx, is not different in normo and hypercalciuric subjects, we suggest that the defect of Ca-ATPase in IH may be primitive and genetically determined.

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Title: PLASMA MEMBRANE CALCIUM PUMP KINETIC IN IDIOPATHIC HYPERCALCIURIC RENAL LITHIASIS (HRL).

Author(s): Alvarez Arroyo MV, Traba ML, Rapado A.
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Introduction. Hypercalciuria is the most common metabolic cause associated with renal lithiasis(RL). Different mechanisms could explain the cause of elevated urinary calcium excretion: renal, absorptive and resorptive. A genetic factor associated to an enzymatic fault in calcium transport has been proposed in HRL.

Objective. To study plasma membrane Ca^{2+}-Mg^{2+} ATPase in human erythrocyte as this defect could be common to different cells in RL patients but not in controls.

Material and methods. Fifteen healthy persons, fourteen normocalciuric RL (NRL) and 33 HRL patients matched for age and sex have been studied. In controls and patients we measured Ca^{2+}-Mg^{2+} ATPase kinetic in intact erythrocyte' cells and a metabolic study in 24-hour urine and blood after fasting was assessed. Comparison of mean values were analyzed using "t" Student's test for unpaired samples.

Results.

	Control(I)	NRL(II)	HRL (III)
Vmax (mmol/l cell/ h)	5.87± 0.51	6.17 ± 0.37	4.50 ± 0.33**
Kca (mmol/l cell)	77.4 ± 12.8	79.3 ± 9.23	38.2 ± 4.63**

* I vs III: p<0.05; ** I vs III: p<0.05.

Conclusions. Ca^{2+}-Mg^{2+} ATPase in HRL patients showed a higher affinity for calcium transport than controls and NRL. This anomaly in cell calcium transport could be implicated in hypercalciuria.

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Title: RELATIONSHIP BETWEEN INTRACELLULAR CALCIUM AND PLASMA MEMBRANE CALCIUM PUMP IN PATIENTS WITH IDIOPATHIC HYPERCALCIURIC RENAL LITHIASIS (HRL).

Author(s): Alvarez Arroyo MV, Traba ML, Rapado A.
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Introduction. The hormonal component of mineral metabolism, in particular calcium, magnesium and phosphorus, and its relation to hypercalciuria has been studied for a better understanding of kidney stone formation. But the cellular component of calcium metabolism is less known.

Objective. To study the relationship between intracellular calcium in platelets and plasma membrane erythrocyte Ca^{2+}-Mg^{2+} ATPase kinetic in HRL patients.

Material and methods. Twenty HRL patients have been studied. After overnight fasting plasma membrane erythrocyte[1] Ca^{2+}-Mg^{2+} kinetic and intracellular calcium in platelets were measured[2]. Comparison of mean values were analyzed using Student's t test for unpaired samples. Correlation between different variables were calculated by lineal regression analysis.

Results. A significant positive correlation (r= 0.48; p<0.01) was found between intracellular calcium in platelets(139 ± 12.8 nM) and Vmax of erythrocyte plasma Ca^{2+}-Mg^{2+} ATPase(4.53 ±0.44 mmol/L cell/h).

Conclusions. These results suggest that ATP-dependent uptake of calcium must be regulated by intracellular calcium in HRL patients.If a defect in cell calcium transport is implicated in hypercalciuria, the intracellular calcium should be related to in the elevated urinary calcium excretion.

1 J Hypertens 1990; 8:285-293.
2 BBRC 1991; 178(3):884-891.
This work was supported with a FISS grant N° 89/0799.

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Title: ABNORMAL Na-K-2Cl ERYTHROCYTIC COTRANSPORT AND FUROSEMIDE NATRIURETIC RESPONSE IN IDIOPATHIC CALCIUM NEPHROLITHIASIS

Author(s): B.Baggio, G.Gambaro, F.Marchini, M.Vincenti, G.Ceolotto, A.Semplicini.
Institution: Division of Nephrology and Institute of Clinical Medicine, University of Padova, Italy.

Anomalous transmembrane anion transport has been observed in erythrocytes of idiopatic calcium nephrolithiasic patients. Since biochemical and physiological correlations exist between the anion carrier and the Na-K-2Cl cotransport, we investigated the furosemide sensitive sodium transport in stone formers.
In 11 idiopatic calcium-oxalate renal stone formers with abnormal erythrocyte self-exchange of oxalate and 9 healthy controls we evaluated the furosemide sensitive sodium efflux from sodium loaded red blood cells, and the natriuretic response to acute i.v. furosemide administration.
Abnormal kinetic properties of erythrocyte Na-K-2Cl cotransport were observed in stone formers in respect to controls [Vmax, median 1.17, range (Ø.51-1.47) vs. Ø.57 (Ø.25-1.Ø8) mmol per liter of erythrocytes per hour, p= Ø.Ø1; Km 22.9 (12.Ø-48.Ø) vs. 13.Ø (5.4-18.5) mmol per liter of erythrocytes, p= Ø.Ø1). The furosemide administration showed a different response in the two groups: the fractional excretion of sodium increased in patients to 5.9 (4.2-7.5)% and in controls to 7.5 (6.8-9.5) (p= Ø.ØØ7)].
Calciuria was directly correlated with Km of the red cell Na-K-2Cl cotransport (r= Ø.73).
The abnormal kinetic properties of Na-K-2Cl cotransport may be relevant for stone formations, hampering renal calcium reabsorption in the distal nephron, and so determining critical physico-chemical conditions for Calcium-Oxalate crystallization.

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Title: INFLUENCE OF NIFEDIPINE ON RENAL FUNCTION AND URINE COMPOSITION IN NEPHROCALCINOTIC RATS

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Previous own investigations showed that the calcium antagonist nifedipine could limit a nephrocalcinosis induced by cholesterol diet in rats. This study was performed to obtain further insights into the effects of nifedipine on stone prevention, renal function and urine composition in nephrocalcinotic rats. Male Wistar rats were assigned to one of the following collectives: 1. cholesterol diet (n = 22), 2. cholesterol diet plus nifedipine (n = 22), 3. control (n = 6). Cholesterol diet (Altromin, Lage/Lippe, Germany) was fed for 4 weeks, nifedipine was administered to group 2 for 4 weeks by gavage (50 mg/kg/24h). During weeks 1 and 4, 5 rats of each group were housed in metabolic cages for urine collection. The urine of each animal was pooled in weekly portions. Calcium, magnesium, phosphate, citrate and creatinine were determined in the urine. The kidneys of 4 animals of group 1 and 2 were perfused and removed for histology after 1,2,3 and 4 weeks respectively. Clearance studies (inulin, calcium, magnesium, phosphate) were performed (n = 6/group) after four weeks. Cholesterol diet induced a marked nephrocalcinosis which could be limited significantly by nifedipine (calcification index (week 4) 1.75 +- 0.5 vs. 0.75 +- 0.5). The sequential histological examinations showed that concrement formation started already after 1 week in group 1, whereas the first concrements in group 2 were observed only after 3 weeks. Cholesterol diet induced an increased excretion of calcium and phosphate, citrate and and magnesium were reduced. The concomitant application of nifedipine resulted in a higher excretion of calcium, magnesium and citrate when compared to the cholesterol group. The inulin clearance was decreased in the latter group. This fall could be limited by nifedipine. The fractional excretion (FE) of calcium, phosphate (pi) and magnesium was increased by cholesterol diet. Nifedipine further increased the FE Ca and Mg, FE pi was decreased when compared to group 1. Our results show that nifedipine limits nephrocalcinosis and renal functional deterioration in rats fed cholesterol diet. Mechanisms to be discussed are effects on renal hemodynamics, tubular cells and an increased excretion of inhibitors as magnesium and citrate.

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Title: ASSOCIATION OF TAMM HORSFALL PROTEIN WITH RENAL CALCIUM OXALATE DEPOSITS IN RATS

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Although extensive research has been done to determine the normal physiological function of Tamm Horsfall protein (THP) and its involvement, if any, in urolithiasis, its exact role remains controversial. It has been suggested that in contrast to normal THP, stone former's THP is functionally different and may promote crystal aggregation and consequently stone formation. The purpose of this study was to investigate the involvement of THP in the deposition of calcium oxalate (CaOx) crystal aggregates in a rat model of urolithiasis. Ethylene Glycol (up to 2%), a precursor of oxalate, and 2.5% ammonium chloride were administered to male Sprague Dawley rats in drinking water. At approximately 5 weeks, kidneys were removed and processed for histology. THP was localized on the sections by protein A-gold (PAG) labelling and immunoperoxidase methods. The non-specific binding was blocked by 1% bovine serum albumin followed by incubation with a polyclonal antibody to rat THP (kindly provided by Dr. John Hoyer). The bound antibody was then visualized either by incubation with PAG followed by a silver intensification procedure or by a peroxidase labelled second antibody. Both methods showed excellent specific labelling. In addition to its normal pattern of distribution restricted to the thick ascending limb of the loop of Henle, THP was present closely associated with the crystal deposits in the papilla. The crystals appeared to be embedded in this dark brown/black material in the tubular lumen. In some samples, although the crystals were lost during processing, THP was still present on the same spot. This is direct evidence that THP is involved in CaOx crystal deposition in this model of urolithiasis.

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Title: CELLULAR MEMBRANE AS HETEROGENEOUS NUCLEATOR OF CALCIUM OXALATE CRYSTALS

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The nucleation of various urinary crystals is probably heterogeneous but the nature of catalytic surfaces involved is not known. Since 1. urine is constantly supersaturated with respect to calcium oxalate, 2. there is a continuous sloughing of membrane material into the urine from regenerating cells of the renal tubular epithelium, 3. cellular membranes and their components have been identified in urinary stone matrix, and 4. membranes are involved in both physiological and pathological calcification processes, we are investigating the possibility of cellular membranes playing a role in the nucleation of urinary calcium oxalate crystals.

Proximal tubular brush border membrane (BBM), isolated from the rat kidney cortex, was incubated at 37°C and a pH of 6.5 in a metastable calcium oxalate (CaOx) solution made by mixing calcium chloride and postassium oxalate. The relative supersaturation of the solution was maintained at 6 or 10 or 12, by simultaneous addition of calcium and oxalate solution using constant-composition method. The rate of crystallization was determined by following the rate at which calcium and oxalate were added to the reaction chamber. At various time intervals an aliquot was filtered through a 0.2µm nucleopore filter. The crystals were identified by x-ray diffraction and scanning electron microscopy. Control experiments were done without the addition of brush border membranes in the solution.

In the presence of brush border membrane addition of calcium and oxalate started much earlier and rate of addition was significantly higher than in the control. As expected, lag time decreased with the increasing supersaturation. Few crystals were seen in the control experiments. Plenty of CaOx crystals were seen in experiments with the brush border. Crystals were identified as CaOx monohydrate. The results indicate that cell membrane fragments are capable of initiating crystallization of calcium oxalate at supersaturations prevalent in the mammalian urine.

Urological stone management – state of the art

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Title: **Extracorporeal shock wave lithotripsy**

Author(s): **D.M. Wilbert**
Institution: **Dept. of Urology, Eberhard-Karls-University Tübingen**

State of the art lecture
(no abstract submitted)

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Title: **Ureterorenoscopy - technique, indication and limitation**

Author(s): **P.N. Rao**
Institution: **University Hospital of South Manchester, Manchester, United Kingdom**

State of the art lecture
(no abstract submitted)

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Title: **Percutaneous nephrolithotomy**
Author(s): **P. Alken**
Institution: **Department of Urology, Clinic of Mannheim, Germany**

State of the art lecture
(no abstract submitted)

Metaphylaxis

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Title: **Metaphylaxis: calcium stones**
Author(s): **M. Butz**
Institution: **Dept. of Urology, St. Josephskrankenhaus, Paderborn, Germany**

State of the art lecture
(no abstract submitted)

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Title: **Metaphylaxis: organic stones**
Author(s): **D. Ackermann**
Institution: **Department of Urology, University of Bern, Switzerland**

State of the art lecture
(no abstract submitted)

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Title: NIFEDIPINE AND METHYLPREDNISOLONE IN FACILITATING URETERAL STONE PASSAGE: A RANDOMISED, DOUBLE-BLIND, PLACEBO-CONTROLLED STUDY.

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The expulsive medical therapy of a partially obstructive ureteral stone is not well established. The three factors that appear to be most useful in facilitating stone passage are an increase in hydrostatic pressure proximal to calculus, the resolution of edema and the relaxation of the ureter in the vicinity of the stone. We studied 86 consecutive stone patients with unilateral ureteral partial obstruction confirmed in each case by intravenous pyelography, who complained of renal colic. Patients were randomly treated, for 45 days in the maximum time, under double-blind condition with methylprednisolone 16 mg/day plus nifedipine 40 mg/day (group A: 13 F, 30 M; mean (SD) 45±14 y.) and with methylprednisolone 16 mg/day plus placebo (group B: 18 F, 25 M; mean age 43±14). They all received 2 liters/day of low in mineral content water (S. Carlo).

Drop-outs were 4 in group A and 6 in group B. In group A the stone passage without surgical manipulation was obtained in 34/39 (87%), while in group B the same result was obtained in 24/37 (65%), p=0.021 (Fisher's exact test).

In group A the mean period for the stone passage was 11.2±7.5 days, while in group B it was 16.4±11.0 days, p=0.036 (Student's T test).

No difference was present in the stone size (diameter 6.9±2.7 mm group A vs 5.8±2.2 group B, NS) and weight (116.7±239.4 mg group A vs 60.5±53.4 group B, NS).

We conclude that nifedipine is effective in facilitating the passage of the stone in partially ureteral obstructive lithiasis.

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Title: HOW MUCH SHOULD STONE FORMERS DRINK?

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It is customary to advise idiopathic stone formers to drink large quantities of fluid to reduce the risk of recurrence. How much should be consumed, and the effect of this upon solute excretion is not known.

We therefore investigated 25 recurrent stone formers who were already following standard dietary advice. Three 24 hour urine collections were made whilst established upon their dietary regimen. Seven days later three further 24 hour urine collections were made on consecutive days whilst being advised to drink at least 5 litres of fluid per day. The urine samples were analysed for calcium, oxalate, and other electrolytes. Urinary supersaturation ratios were calculated using the computer program EQUIL-II.

It was found that increasing fluid intake increased total calcium excretion from 6.45 to 7.51 mmol/day (p=.016) and total oxalate excretion increased from 0.39 to 0.51 mmol/day (p=.036). Both calcium and oxalate concentrations fell markedly (p<.001) as the 24 hour volume increased from 2.23 to 4.44 litres. The overall effect was a significant reduction in the calcium oxalate supersaturation ratio from 6.0 to 3.1.

Whilst some patients remained hypercalciuric or hyperoxaluric by definition, the dilutional effect of the large fluid intake was such that these abnormalities became insigificant in respect of the risk of stone formation. This emphasises the importance of concentration values rather than total excretion. It appears that the beneficial effects of increased fluid intake may not become apparent until daily output exceeds 3 litres.

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Title: HOW SHOULD STONE PATIENTS MONITOR THE DEGREE OF URINARY DILUTION ?

Author(s): D. Ackermann[1], A. Weber[1], B. Hess[2], R. Takkinen[2], Ph. Jaeger[2]
Institution: Department of Urology[1] and Policlinic of Medicine[2], University of Berne, CH

The increase of daily urinary volume in the prophylaxis of stone disease is an important measure. Patients' compliance seems to be improved when they are told how to monitor the degree of dilution in single urine portions which might be achieved by measuring specific gravity and / or electrical conductivity. The aim of this study was to compare different methods of measurements of specific gravity (dip-sticks, hydrometer) and electrical conductivity (Urimho (R)) with respect to reliability and feasibility. These methods were chosen since they are recommended for patients' use.

In fifty-four 24-hour urine specimen, urinary specific gravity was measured with dip-sticks (Combur 10 (R)) and hydrometer (Zylometer (R)) and plotted against the results obtained with a refractometer (Reichert (R)) which was taken as a reference. A highly significant linear correlation was found between all these measurements (R ≥ 0.95). For measuring electrical conductivity, the Urimho (R) device was used, which displays five values corresponding to increasing levels of electrical conductivity. Additional thirty-one urine specimen were analysed with the Urimho (R) device, and the results were compared with specific gravity obtained refractometrically. At Urimho (R) values 1 and 2, ten of eleven urines had a specific gravity lower than 1.010 g / ml. Over the observed range of Urimho (R) values 1 to 5, linear regression analysis revealed a significant correlation with specific gravity (R = 0.85).

Dip-sticks and Urimho (R) allow direct measurement in the urinary stream. For hydrometers, several milliliters of urine are necessary. The reading of the results is most convenient with the Urimho (R) device. For the dip-sticks, patients need a good color vision. Hydrometers should be placed on a horizontal support for exact reading. For the use of the Urimho (R), some technical understanding is requested since the device has to be calibrated periodically. The costs (purchase and upkeep) for 500 measurements amount to 620.- DM with dip-sticks (Combur 10 (R)), 260.- DM with the Urimho (R) device, and 10.- DM with a hydrometer (Zylometer (R)).

Hydrometer, dip-sticks, and the Urimho (R) device give reliable estimates of urinary dilution. All three are handy. The hydrometer might be the least convenient for reading; however, it is by far the cheapest approach.

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Title: EFFICACY OF METAPHYLAXIS IN OUTPATIENTS WITH RECURRENT CALCIUM OXALATE UROLITHIASIS - The Bonn Urolithiasis Post-Episode Care Program -

Author(s): A. Nolde, O. Scharrel, A. Hesse, W. Vahlensieck
Institution: Division of Experimental Urology, Department of Urology, University of Bonn

During 3.5 years (June 1988 - October 1991) 1356 patients with recurrent stone formation participated in the Bonn Urolithiasis Post-Episode Care Program. This program was initiated in order to prevent relapses by propagation of specific diagnostic measures (standard program) and a special metaphylaxis in particular with regard to feeding. Up to now **238 calcium oxalate** stone formers (162 men, 76 women; mean age 52 years) were observed for an average of 15 months (12-18 months).

In close co-operation with the attending physician check-ups were made in 4 months intervals.

Therapy included general as well as specific recommendations concerning diet, fluid intake and drugs depending on the analysis of stone composition, 24-h urines (standard program: Ca, Mg, uric acid, oxalic acid, citric acid, phosphate), serum parameters, nutrition records and accompanying diseases.

Specific physicochemical disturbances have been recognized in the beginning of our investigations including low urine volume (46 %), hypercalciuria (56 %), hyperuricosuria (59 %), hyperoxaluria (26 %) and hypocitraturia (59 %). More than 80 % of the patients showed 2 or more abnormal urine findings. The average stone passage rate previous to participation in the program amounted to 8.1 stones in men and 5.7 stones in women.

Patients with at least four check-ups (observation period 12-18 months) showed a significant improvement in the excretion of lithogenic parameters (calcium, oxalate, uric acid), the inhibitor citric acid and urine volume. In addition there was a significant decrease in supersaturation index of calcium oxalate and uric acid. The average stone loss rate 12 months previous to the start of the program amounted to 1.46 stones in women and 1.54 stones in men. This rate decreased to 0.73 stones in women and 0.77 stones in men.

From these results we conclude that by following individual dietary guidelines and medical therapy risk of stone formation can be reduced significantly. Therefore metaphylaxis should follow all forms of stone removal or disintegration.

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Title: CITRATE TREATMENT IN CALCIUM NEPHROLITHIASIS

Author(s): A.Trinchieri,F.Rovera,A.Guarneri,G.Zanetti,F.Colombo
Institution: Istituto di Urologia,Univ.di Milano.Direttore Prof.E.Pisani

Several workers using treatment with alkaline citrate,either as potassium citrate or as potassium/sodium citrate,have reported the control of stone formation in the majority of the patients.
In this paper we have attempted to assess the long-term effectiveness of potassium/sodium citrate and the mechanisms involved in this effectiveness.
We selected 38 patients with idiopathic calcium stones for treatment with potassium/sodium citrate (5-10 g/daily).Before treatment venous blood and 24 hour urine samples were obtained for determination of pH,potassium,sodium,calcium,phosphate,urate,oxalate(urine),citrate (urine) and creatinine.The investigations at follow up visits included urological examination,measurement of blood and urine levels of electrolytes,urine culture and X-ray examination.
After 2 years of follow up 19 patients dropped out (50%).A total of 19 patients were treated for 24 months or more.Urinary pH,potassium, sodium and citrate significantly increased.

Table I - Clinical results

	Hypocitraturic patients	Normocitraturic pts.
Pretreatment (stones/pt/yr)	1.15	0.72
Treatment (stones/pt/year)	0.22	0.27
Free of recurrences (2 years)	66%	43%

In conclusion our study with potassium/sodium citrate have confirmed it to be capable of increasing the urinary excretion of citrate.
It may be a first choice for calcium stone formers with hypocitraturia.

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Title: ORAL MAGNESIUM-CITRATE LOAD IN HEALTHY MALES - ACUTE EFFECTS OF THREE PREPARATIONS ON ACID-BASE AND MINERAL HOMEOSTASIS, AND URINE PARAMETERS OF RENAL STONE FORMATION

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In idiopathic recurrent calcium urolithiasis metaphylaxis with magnesium seems to be efficient via formation of magnesium-oxalate complex in the gastrointestinal tract and urine (1). It was shown that magnesium-citrate was the superior form of supplement with respect to magnesium bioavailability and increase of citraturia (2). We tested effects on urinary biochemistry and supersaturation, when magnesium-citrate was given alone or together with different alkali salts.
Participants and Procedures: Five male healthy volunteers, age 25 - 31 years, were studied on an ambulatory protocol following an 12 -15 h overnight fast; mildly hydrated with oral aqua dest. the participants underwent three consecutive 2 h periods between 8:00 a.m. and 2:00 p.m., for collection of blood and urine (basal, i.e. before oral load at 10:00 a.m.; period 1, 2 after that). Load types were: BR - breakfast (2 rolls, 20 g butter, 20 g honey); BR + I - breakfast and neutral Mg_3Cit_2 (Boehringer, Ingelheim; FRG); BR + II - breakfast and acidic MgHCit (Sigma, Deisenhofen; FRG) combined with potassium-citrate, potassium-bicarbonate, sodium-bicarbonate, citric acid; BR + III - breakfast and Mg_3Cit_2 combined with potassium-citrate, potassium-bicarbonate, sodium-bicarbonate, citric acid. Each load was dissolved in aqua dest., supplying an equal amount of net base (45.4 mEq) and only slightly differing amounts of citrate (45.4 - 53.0 mEq), whereas magnesium supply varied (I: 45.4 mEq; II: 15.1 mEq; III: 19.5 mEq). Routine methods were used for analytes in urine and serum.
Results (table): With all three magnesium preparations there were insignificant increases of urinary magnesium, calcium, but with the two, alkali salt supplemented, preparations urinary citrate, pH were elevated. By contrast, neutral magnesium reduces oxaluria and CaOx supersaturation and prevents HAP supersaturation from rising significantly, whereas the other two preparations are less anti-oxaluric and additionally tend to increase HAP supersaturation due to the higher pH. The best control of crystallization in terms of crystal growth and aggregation (3) was observed after administration of magnesium-citrate in combination with alkali ions ($p < 0.05$ vs BR; data to be shown).

Median change from basal [(1-0)+(2-0)] in urine (U) and serum (S)

Code	U-pH[a]	U-Ci[1a]	U-Mg[1]	U-Ox[1]	U-Ca[1]	U-CaOx[2]	U-HAP[2]	S-Mg[3]*	S-Ca[3]**	S-PTH[4]
BR	0.58	118	4.5	11	39	2.38	-0.11	-0.02	0.27	-10.0
BR + I	0.92	225	116	2.3	96	-1.20	4.16	0.13	0.53	- 8.2
BR + II	2.52[b]	547[c]	14	6.1	40	-0.37	9.10	0.06	0.10	- 5.2
BR + III	2.50	682[c]	33	6.4	110	-0.82	8.85	0.11	0.72	-31.4

1: mg g Cr^{-1}; 2: Δ G (EQUIL-fit); 3: mg dl^{-1}; 4: pg ml^{-1}; *: Total magnesium; **: Total calcium, corrected for serum total protein; a: $p < 0.05$ (ANOVA); b: $p < 0.05$ vs BR, c: $p < 0.01$ vs BR (paired t- or U-test)

Conclusions: 1) Acute magnesium citrate load results in changes of numerous acid-base, mineral and physico-chemical variables none of which appears deleterious when occuring over the long term; 2) considering documented complexor and inhibitor effects of magnesium, and additional alkalizing citrate effects, anti-stone medication by one of the studied preparations should allow tailoring to individual stone patients.

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Title: RESIDUAL FRAGMENTS AFTER ESWL IN STERILE CALCIUM AND INFECTION NEPHROLITHIASIC PATIENTS AND EFFECT OF CITRATE THERAPY

Author(s): E.Cicerello,F.Merlo,L.Maccatrozzo,G.Gambaro,B.Baggio,G.Anselmo
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Residual stone fragments after ESWL are common; their natural history (clearance, growth, aggregation) is incompletely known, although it is claimed that they constitute a risk in term of new lithogenesis and persistent infection of the urinary tract. We carried out a 12 month follow-up study addressing this issue; moreover, the hypothesis that alkaline citrate therapy improves residual stone fragment clearance was verified. Fourty sterile calcium and thirty struvite/carbonate apatite stone formers with residual fragments after ESWL (diameter lower than 5 mm) were consecutively enroled and randomly allocated in a citrate therapy (6-8 g/day) or in a control (igienic measures only) branch. Infection stone formers, in both branches, received also adequate antibiotic therapy during the 12 month follow-up period. Stone free patients were at 6 months 21% and at 12 months 32% in the sterile group, while in the infection group they were 27% and 40% respectively. In untreated sterile calcium stone formers where clearance of residual fragments were not achieved, these grew or reaggregated in a big percentage of cases. Citrate therapy significantly improved the stone clearance rate in both sterile (stone free patients were at 6 months 65% and at 12 months 75%) and infection (71% and 86% respectively) patients, and prevented residual fragments growth or reaggregation in subjects where clearance was not achieved. In conclusion, these data show that, in the natural history of residual stone fragments, their growth and persistence are quite common, and, finally, citrate significantly ameliorated the outcome of these residual fragments increasing their clearance rate and reducing their growth or agglomeration both in calcium oxalate and especially in infection stone patients.

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Title: ON THE EFFECTS OF CALCIUM-MINERAL-WATERS ON THE INTESTINAL OXALATE-ABSORPTION.

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The use of calcium containing mineral waters in patinets suffering from relapsing calcium-oxalate urolithiasis is discussed controversially. Possibly the inhibition of the intestinal oxalate-absorption can be seen as a favorable effect. Therefore we tested the renal oxalate excretion after a standardized oxalate-rich test meal (oxalate-content: 680 mg) in two groups of healthy test persons (n=2x10). Simultaneously 500 ml of different mineral waters or tap water were administered (calcium-concentrations see below). 12 hours before and 6 hours after the test the food and fluid intake were standardized. Among others the urinary oxalate excretion was measured. In the first four hours after the test meal the urinary oxalate excretion amounted to:

	Ca-content of the water (mg/l)	Urinary oxalate excretion (x±sM)	Probability of error (ANOVA)
Test I	40	0,164 ± 0,017	p<0,02
	122	0,105 ± 0,018	
	312	0,096 ± 0,016	
Test II	40	0,209 ± 0,027	p<0,01
	155	0,146 ± 0,013	
	261	0,112 ± 0,019	

In is concluded that the calcium-containing mineral waters inhibit the intestinal oxalate-absorption and may be favorable for patients with oxalate stone formation, at least if they are administered during oxalate-containing meals.

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Title: OXALATE DEPLETED FOOD FOR HYPEROXALURIC PATIENTS : USE OF AN ENZYME FROM BANANA FRUIT PEELS

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Hyperoxaluria resulting from an increased intestinal absorption of oxalate is the principal abnormality in enteric hyperoxaluria. Patients suffering from this disorder are often advised to restrict the intake of calcium and oxalate rich foods. However, realistically, an ambulatory patient would be often tempted to eat chocolate, nuts, and leafy vegetables which are rich sources of oxalate. In the present study, we demonstrate for the first time, the feasibility of using an enzyme, banana oxalate oxidase, to deplete the oxalate content of such oxalate-rich food materials, rendering them stone-risk free dietary items.

The occurrence of the enzyme, oxalate oxidase (EC 1.2.3.4), that degrades oxalate into carbon dioxide and hydrogen peroxide was identified in banana fruit peels. Purified preparation of banana oxalate oxidase, when directly added to a 10% suspension of cocoa powder or to homogenates of spinach and amaranthus leaves, effectively eliminated 80% of their oxalate content within 15 min. at room temperature. Similarly, a 5% suspension of cocoa powder, when passed through a column packed with alginate-oxalate oxidase beads, was freed 85% of its oxalate content within 5 min. Coimmobilization of catalase with banana oxalate oxidase greatly enhanced the efficacy of the enzyme-beeds in decomposing oxalate into CO_2 and H_2O_2 and reduced the poisoning effect of hydrogen peroxide on oxalate oxidase by immediately removing it from the medium. The regimen of reduced intake of oxalate, usually recommended to enteric hyperoxaluric patients, is difficult to adhere, since many vegetables and nuts are good sources of oxalate. The above technology developed for the preparation of oxalate-depleted diets for hyperoxaluric patients, is simple to follow and does not require buffering or addition of other exogenous chemicals for the reaction to occur. The process can be scaled up in bioreactors of appropriate design.

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Title: ENZYME THERAPY FOR HYPEROXALURIA : A POSSIBILITY ?

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Hyperoxaluria develops in man consequent to accumulation of oxalate as a metabolic end product or in genetic disorders where excess formation of oxalate is induced by the inadvertent action of lactate dehydrogenase on glyoxylate. Therefore, the feasibility of either degrading the preformed oxalate or preventing the oxidation of glyoxylate to oxalate by enzyme therapy has been examined in rats in vivo. In the first approach, the enzyme oxalate oxidase (oxalate:oxygen oxidoreductase, EC 1.2.3.4) that degrades oxalate into CO_2 and H_2O_2 was identified in the banana fruit peels and purified to homogeneity. This enzyme, active only in the acidic range, was found to be nonactive at the physiological pH of 7.4 of rat. However, its functional viability in the rat was ensured by complexing with its antibody raised in rats and then covalently linking it to ethylene maleic anhydride (EMA). These treatments effected a shift in its pH profile, rendering it partially active at pH 7.4. Animals implanted with minidialysis bags containing immobilized oxalate oxidase and catalase in the peritoneal cavities effectively degraded intraperitoneally injected [1-^{14}C]oxalate, in contrast to control animals.

Another approach examined the feasibility of limiting the availability of glyoxylate pool for lactate dehydrogenase by administering to rats exogenous glyoxylate reductase (EC 1.1.1.26), which can metabolically compete for glyoxylate. Prior to the administration of the enzyme, miniosmotic pumps (Alza Corpn.), primed to deliver sodium glyoxylate continuously for a week at a steady rate, were implanted in the peritoneal cavities of rats. Glyoxylate reductase, isolated from spinach, was covalently linked to monomethoxy polyethylene glycol (PEG) and administered intraperitoneally to rats. Determination of urinary oxalate showed that animals administered immobilized glyoxylate reductase excreted lesser amount of newly synthesised oxalate from glyoxylate compared with control animals. The demonstration of suppressing the formation of oxalate and degrading the preformed oxalate in rats, in vivo, using exogenous enzymes, points out for the first time the potential of developing newer techniques of enzyme therapy for long term treatment of hyperoxaluria.

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Title: LONG-TERM RESULTS ON TREATMENT OF CYSTINURIA WITH ASCORBIC ACID AND LOW-PROTEIN DIET

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In the University Clinic of Marburg, 16 patients with cystinuria including cystine stone formation (14 adults, 2 children; age 5-52 years) have been treated from 1982 up to now. The mean cystine excretion rate was 3736 umol/d (range: 1075-7370), the mean time of clinical observation was 70 months (range: 12-120).

The following treatment has been applied:

1. High fluid intake (excretion volume: >4 l/d); 2. concurrent restriction of dietary protein with sulfur-containing amino acids; 3. alkalinization of urine by Na-K-citrate (Oxalyt U (R)) and 4. oral application of 5g ascorbic acid/d.

Results: The restriction of sulfur-containing protein, as registere by current protocols on the diet and analytical control of daily urinary cystine excretion, accounted for 30-50% of cystine reduction. During alkalinization, formation of precipitation of calcium phosphate could be successfully avoided by high fluid intake.

During application of ascorbic acid, no siginificant increase of urinary oxalate or formation of calcium oxalate stones could be observed.

Among the 13 patients being evaluable, there were 2 noncompliers having recurrent cystine stone formation, which could be due to uncomplete diuresis, alkalinization and/or diet restriction. There has been no evidence of recurrent formation of cystine stones in the remaining 11 patients. 8 of them have been completely free of stones after ESWL, URS, irrigation resp. open surgery (mean duration of treatment: 84,4 months; range: 48-129), and 3 have had residual concrements mean duration of treatment: 39,4 months; range: 12-58).

Conclusions: The treatment combined with ascorbic acid as applied here has been shown to be as effective as other treatments (e.g. by mercaptopropionylglycine or D-penicillamine) thereby practically free of side effects. Therefore, our conception should be especially recommendable for pregnants and children. Furthermore medical costs are lower than those of other therapies.

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Title: CALCIUM-OXALATE STONE FORMATION UNDER LONG TIME HIGH DOSE THERAPY WITH ASCORBIC ACID IN A WOMEN WITH CYSTINURIA: A CASE REPORT

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Cystinuria was one of the first inborn errors of metabolism to be described in relation to urolithiasis. In drug treatment D-Penicillamine was used since CRAWHALL 1963 described a reduction in cystine excretion to normal. But there were seen a lot of clinical side effets up to death in agranulocytosis. Since 1969 α-mercaptopropionylglycine is available. The side effects under this therapy are rare but in long term trials tachyphylaxis was seen. High dosage ascorbic acid therapy was established as a new treatment in 1979 by ASPER. Oral intake of ascorbic acid shifts the redox potential of the urine to cause dose dependent conversion of poorly soluble cystine to soluble cysteine. One of the potential side effects is the conversion of ascorbic acid to oxalic acid and the subsequent development of hyperoxaluria.

Since 1982 a young woman with cystinuria was treated in our hospital with 5 gramms ascorbic acid per day. From 1982 until 1988 there were five stone periods and the analysis of all stones showed pure cystine. The cystine concentration in the urine ranged from 180 mg/l up to 470 mg/l. In 1986 a stone spontaneusly passed the ureter and the subsequent analysis showed a calcium-oxalate stone of 100 % Whewellit. During this period we found an increase of urine oxalate concentration of 35,4 mg/d. Ascorbic acid therapy was stopped and under further treatment with α-mercaptopropionylglycine the excretion of urine oxalate was normalized (11,8 mg/d).

SUMMARY: The treatment with high dose ascorbic acid is an alternative therapy with no toxic side effects especially in childhood and for young women during pregnancy.

We showed for the first time that this treatment can promote a calcium-oxalate stone formation but not only a hyperoxaluria as still published. Therefore the periodical measurement of the oxalate excretion under therapy seems to be indispensable.

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Title: **The treatment of urolithiasis at the health resort Marienbad**

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1. The choice of the mineral water cure, of the diatetic measures or the drugs resp. is made when we have knowledge of the chemical composition of the concrement and of the metabolic risk factors.
2. At Marienbad the patient undergoing a mineral water cure can select from a variety of mineral waters of different
Ca^{2+}, Mg^{2+} and HCO_3 contents.
However, a mineral water health cure of 2 to 4 weeks is only a short intermezzo within the stretch of time of a chronic disease. One has to instruct the patient about continuous regimen.
3. Risk factors can be determined by urinalysis (measuring Ca^{2+}, Mg^{2+}, oxalic and uric acid) when the patient is on a standard diet.
4. With hereditary cystinuria attention is directed to the patient's relatives (dispatching of urine specimens by mail). So far 270 families have been recorded.
5. For some years now a new area of indication has been the follow-up treatment after ESWL, particularly with residual desintegrates and residual lower calyceal stones.
In this case several methods are applied: massive dose drinking, shower bath, vibromassage, tramboline jumping, bedding on a tilting table.
It is intended to build up a sanatorium for specific therapy of ESWL and after-care.

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Title: Stone prevention. Why so little progress?

Author(s): G. Alan Rose
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In 1969 the 1st of the 4-yearly international stone symposia was held in Leeds; there were delegates from 61 centres in 14 countries. In 1971, the first of the Bonn-Vienna symposia on Urolithiasis was started by Professors Vahlenseick & Gasser. In about 1970, the NIH in Bethesda decided to fund research into stone research in a number of centres in the USA. These events stimulated research work in many parts of the world.

This explosion of research did produce certain advances. Thus, there have been great improvements in methods of analysis of stones, and of oxalate, citrate, mucoproteins, pyrophosphate, and other chemicals. The science of crystal formation and of inhibitors thereof has advanced considerably. Epidemiological studies have been rewarding. Many therapies have been proposed to prevent stone recurrences. Yet, we seem to have made little impact on actually preventing stone formation. This is in striking contrast with the dramatic progress made by our surgical colleagues, where the knife has been replaced by non-invasive technology. One must therefore question whether the money devoted to the medical side of stone research has been wisely spent.

A number of mistakes have been made over the years. First, it has been wrongly assumed by many that if two observations are correlated, they must be cause and effect, although this is not neccessarily so, and examples of this will be given. Second, it has become apparent over the years that for calcium oxalate stones, concentration of urinary oxalate is more significant than that of calcium Yet, many still devote their energies to studying calcium metabolism in greater and greater detail, ignoring the oxalate. Some continue to fast patients to see if the patients have "renal" or "absorptive" hypercalciuria, although this is flawed as will be shown. Although Finlayson, at the Bologna symposium (1987) made a plea to find ways of reducing urinary oxalate, a chemical useless to man, this has apparently fallen on deaf ears. Third, the studies on formation and inhibition of crystal formation were for many years carried out on simple inorganic solutions. Even after it was shown clearly that Tamm-Horsfall mucoprotein dramatically changed the crystallisation properties of urine, this was largely ignored for many years, and is still ignored by many.

In short, the large-scale funding of urinary stone research has enabled many to travel up blind alleys, and to be so immersed in their own work that they have often ignored the work of others.

Experimental ESWL

P1

Title: BIOLOGICAL EFFECTS OF SHOCK WAVES CHARACTERIZED IN VITRO USING SUSPENDED CELLS

Author(s): F. Brümmer, D. Suhr, U. Irmer and D.F. Hülser
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Fragmentation of renal and ureteral stones by extracorporeal generated shock waves is a clinical standard method. Little is known, however, about the interactions of shock waves with the calculi and the surrounding tissue. Clinical analyses revealed both renal and extrarenal tissue injury, including disruption of the vasculature, damage of the tubular epithelium and transient alterations in the renal functions as well as haematomas, petechial and duodenal bleedings and minor pulmonary hemorrhage. Experiments with animals were mostly used to document types and extent of tissue injury due to shock waves. Such studies, however, cannot precisely distinguish the role of various shock wave parameters (number of shocks, p^+, p^-, rise time, temperature, cavitation) causing tissue injury on the cellular level.
We, therefore, developed several standardized *in vitro* models: Here we report about two simple and well established models for studying biological effects of shock waves in relation to clinical observations: human erythrocytes and mouse leukemia L1210 cells. Erythrocyte analysis is based on a photometric measurement of released haemoglobin and the determination of the proportion of erythrocytes left intact. After shock wave treatment, L1210 cells were analysed by a double staining technique and flow cytometry which allowed a precise differentiation of viable, dead, and disrupted cells. On the ultrastructural level, this dose dependent cell damage is manifested by a transient increase in membrane permeability as well as alterations of cell shape, vacuolisation of the cytoplasm, perinuclear cisternae, swelling of mitochondria or rupture of the mitochondrial fine structure. A delayed proliferation for 72 hours after shock wave treatment indicates that possible long term effects must be considered. A closer view on the mechanisms of intracellular damage revealed the occurence of extra- and intracellular cavitation generated free radicals.
Under carefully controlled and constant experimental conditions, cells have different sensitivities to shock waves, but a general difference in shock wave resistance between normal and malignant can be excluded.

P2

Title: A TISSUE PHANTOM OF IMMOBILIZED AND VASCULARISED MULTICELL SPHEROIDS FROM KIDNEY EXPLAINS CLINICAL OBSERVATIONS.

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Shock waves used for extracorporeal shock wave lithotripsy produce side effects which can be classified as pain, skin petechiae or ecchimosis, hematuria, and renal injury. The tissue injuries observed in patients are mainly associated with disruption of the vasculature or damage of tubular epithelium. For a better understanding of the interaction of shock waves with tissue or cells, a vascularised tissue like cell culture model is necessary.
During the last years the culture of multicell tumor spheroids as a threedimensional model system has been further developed and standardized. These multicell tumor spheroids exhibit structures analogous to those observed in the original tumor and a histological organisation similar to that of solid tumors *in vivo*.
During shock wave treatment of multicell spheroid suspensions, considerable agitation of the aggregates can be observed resulting in strong shear forces and collisions which cause severe cell damage. This observation may explain, why cellular injuries *in vivo* are found in vasculature structures as well as in interstitial cavities.
Immobilizing multicell tumor spheroids in gelatine prevents significant dose-dependent cellular damage after shock wave treatment except after application of very high levels of acoustic energy. This is consistent with the clinical observation that solid, avascular tissue remains unaffected and that shock wave efficacy is significantly attenuated when impacted ureteral calculi were treated.
A tissue like phantom was constructed with immobilized and vascularised multicell spheroids growing from tumor cells or embryonic tissue, both originating from kidney. In these cases, we observed tubular structures inside the multicell spheroids which can easily be distinguished from the surrounding kidney tissue.
We have tested whether a correlation exists between shock wave parameters (p^+, p^-, rise time) and cellular damages. For this purpose, the spatial acoustic sound field of an electromagnetic generator was analyzed by a highly sophisticated detection device developed by Staudenraus and Eisenmenger, the fibre optic probe hydrophone. In the same area immobilized, vascularised kidney multicell spheroids were exposed to shock waves. Their cellular damage was correlated to the local shock wave parameters.
This approach gives new insights in the mechanisms of stone destruction and tissue damage and could lead to a higher fragmentation efficiency with reduced side effects.

P3

Title: SHOCK WAVE INDUCED VASCULAR LESIONS EVALUATED ON THE ISOLATED PERFUSED KIDNEY OF THE PIG

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Evaluation of shock wave (SW) induced vascular lesions in vivo is disadvantageous due to artefacts, i. e. respiratory movement, SW path in the body. Therefore we used the model of the isolated perfused kidney to examine location and type of lesions and its dose-dependency.

Material and Methods: 47 porcine kidneys were isolated, flushed and perfused by tyrode solution. SW were applied using the Modulith SL 20, Storz Medical. Number of SW (2-100), generator voltage (12 - 20 kV) and focus localization (capsule, cortex calix, pelvis) were variied. Vascular lesion was ducumanted by microangiography. Therefore BaSO4 containing dye was perfused during SW exposure. Furthermore the tissue was evaluated histologically.

Results: Dye paravasation was detected over the whole range of SW dosis. The cortical vessels mainly showed petechial paravasation compared to diffuse pattern originating from the arcuated arteries. The average diameter of cortical trauma correlated with generator voltage: 8-11 mm at 12 kV, 11-13 mm at 20 kV. Higher SW numbers increased the density of paravasation. Focussing of the capsule induced subcapsular paravasation. Histological examination revealed dose-dependent tubular cell necrosis, destruction of parenchyma with residual stroma or complete tissue defect as maximal lesion.

Discussion: The isolated perfused porcine kidney is a sufficient and easy to handel model to evaluate SW induced vascular lesion under standardized conditions. The high sensitivity revealed lesions even after lowest SW dosis. SW number, generator voltage and localization of the focus determine type and degree of the lesion.

P4

Title: Histopathologic effects of Extracorporeal shock wave lithotripsy on rabbit kidney.

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Despite the widespread clinical use of the lithotriptor the margin of safety for kidney during shock wave application is substantially unknown. Although a series of pilot studies have been performed, in laboratory animals, long-term follow-up is mandatory to establish the effect of ESWL and subsequent dose dependent changes on the kidneys.

An experimental study was performed in 45 rabbits, to define and compare the early and late complications of ESWL in rabbit kidneys. The animals were divided into three groups of 15 animals each that received 1000,1500 or 3000 shock waves respectively.at 15-20 kV. The rabbits in each group were killed and necropsy performed within 24 hours for the first 5 animals, 1 week for the second 5 animals and 2 months post ESWL for the last 5 animals. Dose dependent moderate damage (subcapsular hemorrhage,interstitial hemorrhage,capsular tension and perirenal hemorrhage) were noted in all kidneys at 24 hours after treatment. Evidence of permanant changes (some fibrosis, tubular and glomerular damage, chronic inflammatory alterations) was noted in long term follow-up. Complete necrosis of the treated kidney was not encountered in this study.

P5

Title: QUANTIFICATION OF RENAL PARENCHYMAL DAMAGE INDUCED BY EPL - AN ANIMAL STUDY

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Extracorporeal shock wave induced damage to the renal parenchyma was systematically evaluated in an animal study (wistar rats). The urinary excretion rate of the lysosomal enzyme N-acetyl-ß-D-glucosaminidase (NAG) was used as a sensitive and highly specific marker for functional kidney lesions. Its basic value expresses the turnover rate, its rise describes leakage of proximal tubular cells.

Materials and methods:
The left kidney of small wistar rats (300 g) were treated with a piezoelectric lithotripter (Piezolith 2500) in general anaesthesia and ultrasound localization. NAG enzymuria was measured in 24 hour urines before and immediately after EPL applied at different intensity grades and numbers of shock waves. The measurement was done according to the method of Maruhn et al., the values were calculated as U/g creatinine (determined colorimetrically, Jaffe's method).

Results:

No of rats	83	5	10	15	15
anaesthesia	no	yes	yes	yes	yes
shockwaves	0	0	1000	2000	3000
intensity	0	0	5	5	2
mean NAG	7,28	10,43	12,23	22,68	14,95
SEM	0,44	1,05	4,69	1,65	1,50

Discussion:
An increase in urinary NAG excretion was observed after EPL in all cases and exceeded the values post anaesthesia (control group).

Urinary NAG seems to be suitable for the rapid, simple, and noninvasive experimental (and clinical) recognition of renal functional lesions caused by different EPL treatment conditions.

P6

Title: EXPERIMENTAL EVIDENCE FOR PROTECTIVE EFFECTS OF NIFEDIPIN AGAINST SHOCK WAVE INDUCED RENAL DAMAGE

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Previous investigations demonstrated beneficial effects of verapamil on shock wave induced tubular dysfunction both in vitro and in vivo. The present study was initiated to investigate whether other calcium antagonists (e.g. nifedipine) can also limit shock wave induced renal damage. Male rats (n = 36) were randomly assigned to one of the following groups: 1. nifedipine (n = 18), 2. control (n = 18). Both groups were treated with 500 shock waves (Dornier/Philips MFL 5000) on each kidney (day 0). Nifedipine was given to group 1 for 5 days starting the day before shock wave exposure (SWE) (50 mg/kg/24h by gavage). Urine samples were collected on days -1, 0 (=immediately after SWE), 1, 7 and 28. To assess renal function, urine volume, osmolality, N-acetyl-ß-glucosaminidase (NAG), ß-2-microglobulin (ß2M) were measured. Creatinine clearance (CC) and fractional excretion of sodium (FE Na) were determined on day 1 in 6 rats of each groups. Kidneys were perfused and examined by histology in 6 rats of each groups on days 1,7,28. After SWE there was a temporary increase of urine volume, NAG, ß2M and a decrease of osmolality which was significantly less pronounced in nifedipine treated animals compared to the controls. CC and FE Na were less influenced in the verapamil group.
Histology revealed venous dilatation, tubular dilatation, epithelial swelling and vacuolisation and potocytosis. These effects were reduced by verapamil. Our biochemical and histological results demonstrate a protective effect of nifedipine against shock wave induced renal damage similar to that observed on verapamil treatment.

P7

Title: Pathogenesis of intrarenal bioeffects after extracorporeal lithotripsy and its clinical relevance

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In animal experiments the most important acute changes were intrarenal hematomas reported by many groups. Besides transitory tubular dysfunction occured. Clinically the intrarenal bleeding were difficult to detect in contrast to the neglectable subcapsular hematomas. In autopsies after extracorporeal shock wave lithotripsy (ESL) all patients documented intrarenal hematomas especially at the corticomedullary junction as seen in own animal investigations. On this borderline seperating two different tissue densities (cortex, medula) the arcuata vessels extend. Our scanning selectron microscopy (SEM) examinations documented the rupture of these vessels, especially the veins. They run in a circular path parallel to the renal outline and are mostly exposed to ESL. Their normal function is governed by water reabsorption, so they are extremly thin walled and thus highly damageable. Cavitation bubbles collapsing under jet formation is a suitable mechanism for vessel trauma and explain the accidental focal nature of the damage. It is also supported by the greater vulnerability when identical numbers of shock waves were applicated in double shot form in our experiments. Besides vulnerability seems also to be a question of vessel architecture because exposition of identical shock waves to ovary and uterus in our experiments failed to damage the tissue. In long term follow up, we saw the original regions of hematoma (NMR detected) developing to fibrosis with surrounding tubular necrosis and glomerular sclerosis. Clinically sonography and CT were not able to reveal most important primary damage, the intrarenal hematoma. The aim was to document the vessel rupture. With the aid of urinary marker proteins, especially alfa 2 macroglobulin (MG 720000), which can only be released by vessel ruptures, we were able to detect hematomas after ESL in patients. Vessel rupture may also be the reason for the observed proteinuria of IgG and albumin which led to the assumption of glomerular lesions, but what never could be documented in man or in own animal histology or SEM. Thus vessel rupture are the worst bioeffects. Clinically it seems advisable to tend to a smaller focus area, to be careful with double ESL and to look for adjuvant medical treatment (Ca++ antagonists) to reduce side effects.

P8

Title: INTRARENAL BIOEFFECTS IN PRE-DAMAGED KIDNEYS AFTER EXTRACORPOREAL LITHOTRIPSY

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With the aid of quantitative determination of highly specific urinary marker proteins we were able to detect intrarenal side effects and to differentiate hematuria as to a glomerular or postglomerular source. The kind and course of bioeffects in pre-damaged kidneys should be compared to normal kidneys.

Methods Urinary marker proteins alfa 2 macroglobulin (a2m), immunoglobulin G (IgG), albumin (alb), alfa 1 microglobulin (alm) and NAG were measured before and on the days 0,1,4,7,14 after piezolectric lithotripsy with 4000 shock waves to caliceal stones in 15 patients with known proteinuria (D.M., G.N., I.N.) (group I) and 15 control patients (group II).

Results after shock wave delivery

day	-1	0	1	4	7	14
			group I			
a2m/cr (mg/gm)	6.6±4.5	35.6±22.9*	6.7±4.9	7.6±6.7	6.7±3.9	5.5±3.2
IgG/cr (mg/gm)	93.6±72.4	346.2±245.3*	107.9±131.1	80.2±98.3	105.6±160.4	49.5±60.0
alb/cr (mg/gm)	572.5±402.3	1,935.2±1,099.9*	614.2±668.0	461.4±564.9	605.3±739.4	363.4±300.2
alm/cr (mg/gm)	23.9±11.9	33.7±27.0	29.6±33.4	32.8±58.6	26.5±31.1	16.8±8.1
NAG/cr (u/gm)	9.2±7.6	12.3±8.6	9.3±9.9	9.5±8.6	12.9±19.2	7.7±6.3
			group II			
a2m/cr (mg/gm)	4.6±5.1	19.7±17.9*	9.3±6.4	8.2±5.7	6.7±2.9	8.2±3.4
IgG/cr (mg/gm)	12.7±9.7	184.4±206.3"	41.3±38.7*	37.2±42.7	8.4±6.7	9.6±2.2
alb/cr (mg/gm)	36.5±52.4	1,228.2±1,067.2"	190.0±211.4*	132.3±204.5	51.0±48.3	28.3±23.2
alm/cr (mg/gm)	9.7±3.6	11.7±4.2	11.5±6.0	13.0±9.8	7.4±6.1	8.0±4.1
NAG/cr (u/gm)	2.4±1.4	7.1±416*	3.0±2.8	5.8±5.7	3.3±2.4	2.2±0.7

* p 0.05, " pc 0.005

In group I a2m, IgG and alb were stat. sig. enhanced only day 0 in contrast to group II, where elevation lasted two days for IgG and alb.-Ratio between high molecular a2m to alb for alb/cr>100mg/gm differentiated glomerular (<2.00) from postglomerular (>2.00) hematuria. The period of postglomerular hematuria (>2.00) did not differ in pre-damaged kidneys (group I, day 0:2.16, day 1:2.42, day 4:-) from normal kidneys (day 0:3.51, day 1:3.01, day 4-).

Discussion The extent of proteinuria did not support an enhancement of lithotripsy damage in preinjured kidneys. The course of intrarenal hematomas, detected by a2m, is comparable in both groups. Transient tubular dysfunction was comparable. The results support an identical pathophysiologic mechanism in lithotripsy damage.

P9

Title: **Extrcorporeal shock waves - more than lithotripsy of kidney and gall stones ?**

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Since the extracorporeal shock wave lithotripsy was introduced into medicine in 1980 for urolithiasis this technique has experienced a broad development. Gall stone lithotripsy, which was started in 1985, was the first use of shock waves outside the classic ESWL. Since then a number of experimental therapies have been investigated:

The initial succes of ESWL of gallstones led to the therapy of patients with chronic pancreatitis. While acute pancreatitis is a contraindication for shock wave therapy, 50 % of the patients with chronic pancreatitis, which were treated with shock waves, became painfree.

Another stone disease which can be treated with shock waves is sialolithiasis. After Iro first described the in vitro desintegration of salivary stones, some hundred human applications have been performed with a success rate above 50 %.

Using the known effect of membrane depolarisation the value of shock waves for reanimation was investigated, apparently this was not successful.

Various groups looked for the effects of shock waves on tumor cells. Although this has not yet clinical impact, some aspects need further investigation.

A relative to shock waves is the focussed extracorporeal piezoelectric pyrotherapy. Here thermonecrosis is induced in defined areas, the method has been used in humans for renal cell cancer or BPH.

Another complex of shock wave research is the effect of shock waves on collagenous and hard tissue. It could be shown that shock waves can enhance or reduce wound healing. In further studies we saw the osteogenetic effect of shock waves - based on the shock wave induced focal aseptic necrosis - in normal and fractured bone. This led to the therapy of pseudarthrosis. The therapy of other bone diseases including influences on the stability of endoprotheses of the hip joint or therapy of tendinosis like "tennis elbow" are under further investigation. However, the therapy of pseudarthrosis seems to become established and is now performed by various groups. The shock wave has revolutionized urologic stone therapy, but with shock waves the urologist has a tool, which may be helpful in a much broader area in medicine.

P10

Title: Efficiency of ESWL on Calcium-oxalate stones: Role of Copper,Iron Magnesium and Zinc concentrations on disintegration of the stones

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Clinical use of Extracorporeal shock wave lithotripsy (ESWL) in the noninvasive management of urinary calculi is highly effective and practical. But its results vary in relation to several factors including localization,size and the chemical composition of the stones. Morover, the anatomy of the kidneys treated is another contributing factor to the success rate of ESWL in upper urinary tract stones.

Currently, except cystine and struvite stones, the efficiency of ESWL has been proved and has become the most outstanding therapy for calculi in a great number of stone centers. In this study the efficiency of ESWL treatment in respect to the concentrations of 4 different trace elements (Cu,Fe,Mg and Zn) in the chemical composition of Calcium-oxalate monohydrate stones have been evaluated in 20 patients. Our results revealed a statistically significant (p 0.05) correlation between the concentrations of 4 trace elements in the chemical composition of Calcium-oxalate monohydrate stones and the fragility of them against ESWL therapy.

P11

Title: ARE DIURETICS ADVISABLE DURING EXTRACORPOREAL SHOCK WAVE LITHOTRIPSY?

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The protective effect that diuretics have against certain harmful agents acting on the kidney, particularly those capable of causing tubule damage, is a generally accepted fact.

Nowadays, when there are already over 2 million patients treated by means of extracorporeal shock wave lithotripsy (ESWL), with over 1.100 machines, no clear criteria have been published about the opportunity of using them during treatment of this kind.

Our results, obtained during an experimental study carried out with a piezoelectric lithotriptor (Piezolith 2.200), in which we bombarded the Left Kidney of 41 New Zealand rabbits with the mean dose used in treatment in humans (5.000 pulses), lead us to form the hypothesis that the use of diuretics is not advisable during the treatment, but in instead they should be used after treatment.

The reasoning used to arrive at this conclusion is based on the following facts:

- The harmful effect of shock waves arises basically through the effect of cavitation.
- Tissues are particularly sensitive to cavitation when submerged in liquids.
- The renal glomerulus may be considered to be an ideal model for studying the consequences of this physical phenomenon: a capillary tangled ball surrounded by liquid (ultrafiltrate in Bowman's space and covered by a thin fragile cell layer, the Bowman's capsule).
- During treatment with ESWL, there is tubule obstruction, which immediately causes a dilation of Bowman's space, due to accumulation of ultrafiltrate.
- This hyperdistension of Bowman's capsule of the glomerulus makes it particularly sensitive to cavitation, and signs of rupture can be seen, with consequent sclerosis of the glomerular ball.

These findings lead us to think that a relative dehydration may be a protective factor of the glomeruli during ESWL.

General Posters

GP1

Title: RENAL BLOOD FLOW AND URINARY CALCULUS DEVELOPMENT

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In animal experiments sex differences of the renal papillary blood flow are detectable. These distinctions are adjusted by means of the intervention in the angiotensin-conversion (L. Eloy et al., Pflügers Archiv 398 (1983) 253-58). The catalyzed enzyme of this process was discovered in increased concentrations in urinary stone patients (B. Baggio et al., J. Urol. 129 (1983) 1161-62). - Our own bloodtests will clarify whether equivalent sex differences also exist in angiotensin II-concentrations in the case of human beings, and which amounts are found in urinary stone patients with heterogenous renal stone paragenesis. For the present we have taken blood samples from respectively 10 control people of female and male sex and we have compared their angiotensin II-concentrations with the concentrations from 30 urinary stone patients (radioimmunoassay with previous extraction step-silica columns).

GP2

Title: SERUM PYRUVATE LEVELS IN IDIOPATHIC URIC ACID LITHIASIS

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Uric acid stones form because the urine becomes supersaturated with undissociated uric acid.The factors that lead to supersaturation of the urine with respect to undissociated uric acid are increased concentration of uric acid in the urine and undue acidity of the urine. Excessively acidic urine has been claimed to be prevalent in older patients,who may form uric acid stones,even in absence of hyperuricosuria.

We studied 56 patients older than 50 years with idiopathic stone disease.The patients were divided in 2 groups: uric acid stone formers (19M,5F) and calcium stone formers (25M,6F).Serum was analyzed for urate,pyruvate,glucose and creatinine;24 hour urine samples were analyzed for their urate and creatinine contents;fasting 2 hour samples were collected for determination of pH,titrable acidity and ammonium.The uric acid stone formers had significantly higher serum urate (5.3±1.5 mg/dl) and pyruvate (0.90±0.63 mg/dl) levels than the calcium stone formers (5.5±0.7 and 0.55±0.21).The mean urinary pH (5.1±2.8 vs 5.5±0.7) and the mean urate clearance (9.2±5 vs 5.5±2.8 ml/min) were significantly lower and the mean titrable acid/ammonium ratio (0./4±0.59 vs 0.39±0.23) was significantly higher in the uric acid stone group than in the calcium stone group.

The high serum pyruvate levels suggest a disorder of adenosine triphosphate (ATP) synthesis in relation with ageing,that may induce hyperuricaemia and deficiency in renal ammoniogenesis.A disorder of ATP synthesis might enhance uric acid production by increasing the conversion of hypoxanthine to xanthine and xanthine to uric acid and depress renal ammoniogenesis by increasing the concentration of Krebs cycle intermediates such as alpha-ketoglutarate.

GP3

Title: THE LONG-TERM STONE RECURRENCE RATE AND RENAL FUNCTION CHANGE IN THE UNINEPHRECTOMIZED UROLITHIASIS PATIENTS

Author(s): Y.H. Lee, W.C. Huang, L.S. Chang, M.T. Chen, Y.F. Yang, J.K. Huang
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INTRODUCTION

By using animal model, our previous study showed that uninephrectomy increase the vulnerability of the contralateral remnant kidney to urolithiasis and crystal deposition when the lithogenic risk factors are present. However, the clinical implications associated with uninephrectomy in human urinary calculi remain poorly explored.

MATERIALS AND METHODS

We evaluated the stone recurrence rate and long-term renal function change of 50 patients with a solitary kidney who had undergone uninephrectomy for urinary tract stone disease. They were evaluated 60 to 84 months after surgery (mean,70). The follow-up evaluation included detailed history taking, physical examination, blood and urine biochemistry, urinalysis, urine culture, intravenous pyelography, ultrasonography of kidney and I-131 ortho-iodohippurate renography.

Results

The overall stone recurrence rate in uninephrectomized urinary calculi patient was 30% (15/50). The mean time to develop recurrent stone was 31.1 months (6-74) and the mean episodes of recurrent stone were 2.1 times per patient (1-5). The renal fucntion of the remnant kidney in most patients were unchanged during the follow-up. However, of the 15 recurrent stone formers 2 developed anuria during the acute attack of renal colic and required percutaneous nephrostomy urinary diversion, 1 developed proteinuria (3g/day) and progressive renal failure 47 months after nephrectomy. The metabolic stone formers seemed more likely to develop recurrent stone than infection stone formers (37% vs. 13%) although no statistical significance was noted (P=0.198).

Conclusions

During a 5 to 7 years follow-up, 30% uninephrectomized urinary calculi patient developed recurrent stones. The long-term renal function remained stable in most patients. However, the renal function of these patients are vulnerable to stone recurrence.

GP4

Title: Treatment using ureteral catheter with a brush for the removal of stone pieces adhered to uretera mucous membrane

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Despite favorable lithotrispy achieved by extracorporeal shock wave lithotripsy (ESWL), we may encounter difficulties in the treatment of patients whose hydronephrosis and pyuria showed no improvement for a lengthy period because of smashed fine stone pieces adhered to the ureteral mucous membrane. The following are the favourable results obtained by the ureteral catheter with a brush which was newly developed by our hospital for the removal of stones under fluoroscopic control.

The subjects were 12 patients (8.4 months on the average after ESWL) whose hydronephrosis and pyuria were caused by residual stones after ESWL. An 8-Fr catheter with a brush fixed on a tip of a guide wire was insered in a location of stones before the brush was moved up and down severe times until the residual stones were pushed up into the kidney. In 9 of 11 patients (82%), the residual stones were successfully pushed up into the kidney, and a D-J catheter was indwelled in 8 of 9 patients and 7 patients (64%) became stone free in the kidney, ureter and bladder (KUB) 1 months after treatment.

GP5

Title: ESWL TREATMENT OF KIDNEY AND URETER STONES BY SIEMENS LITHOSTAR-PLUS

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This is a report on ESWL treatment using the Siemens Lithostar-Plus appliance.

Altogether, we treated 412 patients, 161 (39%) females and 251 (61%) males. The average age was 51.2 years for the female, 50.9 years for the male patients.

In most cases (347 = 84.2%), there was one stone; 48 patients (11.7%) had two and 17 (4.1%) more than two stones. The stones were localized on the right side in 182 (44.2%) and on the left side in 230 (55.8%) cases.

Localization in detail: 256 (62.1%) in the kidney with 16 (3.9%) in the upper, 36 (8.7%) in the middle and 46 (11.2%) in the lower calix plus 158 (38.4%) in the renal pelvis. The rest of the stones (156 cases = 37.9%) were found in the ureter with 59 (14.3%) in the upper, 23 (5.6%) in the middle and 74 (18.0%) in the lower part.

The total number of therapeutic sessions was 561, 97 patients (23.5%) needed more than one session.

In the great majority of the cases (93.9%), we used the undertable unit and localization by x-ray. Larger kidney stones (6.1%), which could be localized well sonographically and demanded higher amounts of energy, could be treated with the overtable unit. Respiratory shockwave triggering was necessary in 58.5% of the cases.

The total number of shockwaves was 2456 (average), the mean working voltage 17.8 kV. Average x-ray time was 0.26 min.

The stone(s) could be removed during the first session in 244 cases (59.2%). Another 74 patients needed a second session and, in some cases, three or more sessions. Altogether, 318 patients (77.2%) were cured completely.

Partial disintegration was observed with 76 patients (18.4%). In 18 cases (4.4%), finally, there was no disintegration at all, so that different measures became necessary, such as the Seitz loop, uretero-renoscopy or open surgery. Especially, stones in the lower part of the ureter turned out to be difficult ESWL cases.

GP6

Title: EXTRACORPOREAL SHOCK WAVE LITHOTRIPSY IN TUBULAR ACIDOSIS

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Though the distal renal tubular acidosis (RTA) is a rare cause of urolithiasis it belongs to the hardly curable diseases because of the formation of bilateral multiple calculi or nephrocalcinosis wich may frequently cause renal colic, occlusion and infection of urinary tract.

During the last 4 years we observed 5 women (between 20-45 years old) with RTA verified by ammonium chloride loading test. Nephrocalcinosis was diagnostized by X-rays in 3 cases, 2 patients had only big pelvic stones.

Patients requiring therapy for kidney stones were treated with ESWL (Siemens Lithostar) in 1-3 sessions. Taking care of patients the number and size of calculi were followed by radiograph. ESWL was necessary if the diameter of a pelvic stone had reached the 1 cm even though the patient had no complaint. Considering the great numbers of calculi the sighting of stones to be crashed was sometimes difficult.

According to our observations the complicated stone streets in these patients were rare despite of the big volume of fragments because the lumen of their ureter was large enough to pass big stones or fragments. Two patients with single stones became stone-free, in the other 3 cases the fragmentation of stones was successful. In case of RTA the most serious complication is the urinary infection caused by Proteus (break of alkalization would increase acidosis and hypercalciuria). For that very reason we disregarded the regular use of stents. Early diagnosis of Proteus infection is as important after ESWL as before it.

We belive that ESWL is the treatment of choise for stone-patients with RTA. ESWL monotherapy is more favourable than the ESWL+PCNL combination because of the predictably great numbers of treatments. If ESWL could have been planned it was alternately performed on the two kidneys. Our experiences are similar to those gained with ESWL of calculi in medullary sponge kidney.

GP7

Title: ESWL Monotherapy in Staghorn Calculi

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The Treatment of staghorn calculi is always a problem. Having more experience with ESWL, now we perform a shock wave treatment in staghorn calculi in a non dilated collecting system. Mostly we insert a d-j-stent before the treatment.

Since January 1987 at the Dept. of Urology University of Frankfurt Medical School 52 patients (mean age 54 years) with partial or complete staghorn calculi were treated according to this protocol. The treatment was performed either on the Dornier HM3 or on the Modulith SL 20 normally in analgosedation. For the complete stone desintegration a mean of 2,3 sessions was necessary (renge 1-6 sessions) with 2 800 shocks per session.

61 % of the patients suffered from urinary tract infection whereas only 30 % developed feaver after the treatments. In 11,5 % a percutaneous nephrostomy must be placed after the shockwave treatment depending on obstruction of the d-j-stent by desintegrated stone material or large " Steinstresse ".

We conclude that ESWL of partial or complete staghorn calculi using a d-j-stent is a step forward in the difficult therapy of staghorn calculi.

GP8

Title: FOLLOW-UP OF CHILDREN AFTER ESWL: STONE FREE / RECURRENCE RATE, BLOOD PRESSURE AND RENAL FUNCTION

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ESWL is the treatment of choice for 96 % of upper tract urinary stones in adults and has also proved to be save and highly effective in children.
However, there is some controversy regarding a potential risk for damaging the growing kidney and inducing hypertension.
Since 1984, in 56 children (30 boys and 26 girls, age range: 1.5 - 16 years, mean age: 8.8 years) 105 ESWL treatments were performed using a Dornier HM 3 lithotripter and a Storz modulith. This accounts for 1 % of ESWL treatments (10 250) during this time period. 9 (14 %) children had bilateral stones. 65 renoureteral units (RU) contained 13 staghorn stones (20 %), 22 renal pelvic stones (34 %), 22 caliceal stones (34 %), 4 upper ureter (6 %) and 7 lower ureter stones (10 %). Associated metabolic disorders were detected in 10/56 children (18 %) and anatomical anomalies in 10/56 children (18 %). 33/56 patients (59 %) had recurrent urinary tract infection. To prevent damage to infantile tissues, several precautions during ESWL were undertaken in children such as styropore foam shielding for lung protection, limitation of energy and number of shock waves per treatment and staging treatment in lager stones (mean number of treatments: 1.7 per RU). 18 treatments required auxiliary instrumentation. There were no complications related to the children's anatomy encountered.
28 children (50 %) were stone free at the time of hospital discharge. Three months after ESWL 90 % of the children were free of stones.
Follow-up period ranged from 9 months to 8 years (mean follow-up: 3.8 years). At the time of evaluation, KUB and renal ultrasound or IVP, blood pressures and MAG 3 or I^{123}-Hippuran isotope renograms were obtained. Stones recurred in 18 % of children who had become stone free after ESWL. In those 10 % of patients, who never became stone free, re-growth of residual fragments was encountered in 100 %. 9 children had during the follow-up period a repeat hospital admission for stone treatment: 4 children had ESWL because of the residual fragments and stone regrowth (1 horseshoe kidney, 1 previous pyelolithotomy), 1 child because of true stone recurrence. In 3 children with staghorn calculi and previous stone surgery in 2 cases, 3 partial nephrectomies were performed because of residual stones in a destructed collecting system; one additional nephrectomy was performed in a girl with cystinurica, who did not comply with stone metaphylaxis.
Blood pressures were normal at the time of ESWL in all children and remained at the age specific percentiles after ESWL. Global renal function remained stable. The only exception was the nephrectomy case with grossly reduced function of the ESWL-treated kidney with recurrent stone formation and renal obstruction.

GP9

Title: Nephrolithiasis In Children

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Urolithiasis in children are not very common. Normaly 2 % of all stones were produced by children. The reason for stone forming was investigated in our ESWL (HM_3 Dornier) patients.
Aetiologically, renal stones can be classified into two categories, metabolic (Ca-Ox, RTA, Cystine ...) or non metabolic disorders, if stone analysis is known. 0,9 % of our patients, who underwent ESWL were children. From 1.3.1985 - 31.10.1992 47 patients (o 30, o 17, 7 months - 14 à age) were treated. We did not know in every patient the stone analysis, because some children did not collect the urine.
Results: Most of the stones were ca-ox (n=21) followed by carbonat-apatit (n=8), cystine (n=2), mg-ammoniumphosphat (n=2) and 2,8 dihydroxyadenin (n=1). No uric acid stones were seen. Children have a better stone clearance rate than adults, and thats the reason while also in staghorn calculi ESWL monotherapy is possible. Auxiliary measurements were rarely necessary. 17/24 of our patients had become stone free. In case of cystine, 2,8 dihydroxyadenin and recurrent ca-ox stones further metabolic investigations were necessary. In case of infection stones examinations of the urinary tract and the urine are important.

List of contributors*

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