REPORT

of

Committee on Thesis

The undersigned, acting as a Committee of the Graduate School, have read the accompanying thesis submitted by Linwood Dickens Keyser for the degree of Master of Science in Pathology. They approve it as a thesis meeting the requirements of the Graduate School of the University of Minnesota, and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science in Pathology.

Wm. Carpenter MacCarty
Chairman

H. C. Mann

W. F. Braasch

E. T. Bell

4-21 6M

REPORT

of

COMMITTEE ON EXAMINATION

This is to cerfity that we the undersigned, as a Committee of the Graduate School, have given Linwood Dickens Keyser final oral examination for the degree of Master of Science. We recommend that the degree of Master of Science be conferred upon the candidate.

Minneapolis, Minnesota
November 4, 1921

W. Carpenter MacCarty
Chairman

E. T. Bell

4-21 6M

THESIS

THE ETIOLOGY OF URINARY LITHIASIS

A Review and an Experimental Study

Linwood Dickens Keyser, B.A.,M.D.

Submitted to the graduate faculty of the University of Minnesota in partial fulfillment of the requirements for the degree of Master of Science in Pathology.

November, 1921.

Historical.

The phenomenon of calculus formation in the urinary tract has attracted the attention of medical men from the earliest times. In consequence the theories advanced to explain the etiology of stone and the efforts made to treat and prevent the process have been legion. Hippocrates was the first ancient writer to venture an explanation of the disease. He felt that waters containing different varieties of mud and sand fostered the stone forming process but emphasized the importance of inflammatory changes in the kidneys and bladder as being essential factors also. Schepelmann feels that the Father of Medicine also appreciated the existence and importance of the mucoid binding substance in urinary concretions. Galen claimed a relationship between gout and stone and with him the long train of efforts to establish a "crystalline uric acid diathesis" as the forerunner of stone began. This concept held sway especially during the latter part of the nineteenth century and it was during this period that the use of lithium salts as a "uric acid solvent" was much in vogue in the therapy of both gout and stone.

Chemical Theories of Calculus Formation.

In 1856 Meckel expounded the doctrine of a "stone forming catarrh". According to this idea a low grade catarrhal inflammation of the kidneys leads to the precipitation of certain elements of the exudate together with certain of the urinary salts and thus the stone has its genesis. On the experimental side little was done until the middle of the last century. In 1857 Rainey demonstrated the fact that crystals of carbonate, oxalate,and phosphate of lime would assume different shapes depending upon whether they were precipitated from water or from a medium containing colloidal substances such as gelatin, albumin, gum acacia, and mucus. He suggested that the atypical crystals thus precipitated in the colloidal media might be related to

urinary concrements.

Ord and Shattock confirmed and extended the ideas of Rainey. Working with calcium oxalate, they showed that this substance usually crystallizes from water solutions as octahedra, but in colloidal media tabloid, dumb-bell, and spheroid forms were obtained. An examination of the nucleus and body layers of calculi of oxalate of lime showed these to be composed of atypical crystals, which in many respects resembled the forms produced in vitro. These crystals were such as might have been deposited in a colloidal medium and seemed to be fused together by an organic matrix. Fowler in 1906 studied a series of calcium oxalate calculi and came to similar conclusions.

Ebstein in 1884 carefully analyzed calculi and proved the existence therein of an organic matrix. Working with Nicolaier in 1891, he published an extensive series of feeding experiments, among other drugs using different derivatives of oxalic acid. These observers were surprised to find that one of these derivatives, the diamide of oxalic acid, commonly known as oxamide, would upon being fed to animals of different species, be excreted in the urinary stream and would form in many instances, concretions varying in size from yellow particles of sand to hard masses 1 cm. in diameter. Chemical analysis showed the stones to consist of oxamid plus an organic substance which Ebstein held to be albumin. Ebstein held that the desquamative catarrh and epithelial debris, produced by the toxic action of the oxamid upon the renal epithelium, furnished the colloidal material by means of which the stone forming crystals were precipitated. This we shall show later was not altogether a true explanation of the case.

Tuffier and Rosenbach repeated the work of Ebstein with positive results. Rosenbach found that sectioning, in part or entirely, the nerve or blood supply of the kidney on one side, or ligation of one ureter, would be

followed by a relative deficiency of excretion on the treated side, with an excess of excretion of the stone-forming material on the untreated side.

Schade of Kiel in 1909 advanced the idea that stones are formed by the clotting of fibrinogen in the urine, together with the simultaneous precipitation of urinary crystalloids. Fibrinogen, according to Schade, is an "irreversible colloid" or one which having passed to the state of clot or gel, cannot spontaneously return to the suspensoid phase, i.e. the state of colloidal solution. Schade was able to produce in vitro stony masses by mixing fibrinogen with freshly precipitated lime salts, and then clotting the mixture by the addition of calcium chloride. The weakest point in Schade's claim lies in the fact that "stone-forming" urine is not usually associated with fibrinogen in demonstrable quantity, while the rare instances of excessive fibrinogenuria, one of which has come under my observation in a case of adenocarcinoma of the kidney, (O'Conor) have not been associated with stone. However, there are rare cases of fibrin calculi, such as are reported by Gage and Beale, which might have such an etiology.

Finally, among other evidences of a so-called chemical etiology of stone are cited the occurrence of xanthin calculi in xanthinuria; of cystine calculi in cystinuria; the relatively infrequent occurrence of uric acid gravel in acute gout; of oxalate calculi in oxaluria; and of phosphatic calculi in phosphaturia.

On purely theoretical grounds, metabolic errors, in which deficiency in oxidation, neuroses, sedentary habits, and the like have been the chief factors accused (See Watson and Cunningham), have been assumed to account for concrement formation. Klotz would ascribe the process to calcification in necrobiotic tissue such as occurs in the degenerative tissues of organs of the body, while Young in Osler's system feels that concentration of crystals in the

collecting tubules, with subsequent erosion of the minute mass through a renal papilla serves to initiate the stone forming process.

Mechanical Theories

The so-called anatomic or mechanical theories of stone formation are based upon the relative frequency with which stones have been found associated with urinary stasis, diverticuli, stricture, prostatic hypertrophy, and other forms of obstruction. Foreign material, renal or extra-renal in origin, has also been supposed to cause stone, as a result of reduplication of surfaces and the increased opportunity for surface tension phenomena to come into play. The significance of such an association, which is clinically established, we will consider in more detail farther on.

Infectious Theories

The bacterial or infectious theory of stone formation is based on the demonstration of bacteria in stones and upon the fact that the kidney, associated with stone, is almost if not quite always an infected kidney. Especially has the clinical relationship between carbonate and phosphate calculi and urinary infection been emphasized. But we must bear in mind that the infection has been supposed to bring about the stone deposition by changing the reaction of the urine and not necessarily by other means. For this reason the uratic and oxalate stones have been considered usually as of non-bacterial origin.

With Rosenow's development of the doctrine of specific activity of bacteria, Dr. Charles H. Mayo has suggested that calculi are formed by the secondary invasion of a previously established low grade pyelonephritis by specific stone-forming bacteria. The experimental production of gallstones, of alkaline phosphatic cystitis, and recently of urinary concretions by Rosenow and Meisser, using specific bacteriologic methods, is a wonderful step forward and seems the best evidence to date that specific bacteria are related to the

process of stone formation.

The Physico-Chemical Features of Urinary Concrements.

A survey of the chemical constituents of urinary calculi has been interesting. Albarran divided calculi into those unaccompanied by infection, "primary calculi", and those which appeared clinically to be due to infection, "secondary calculi". This division, as we have seen, was really based more on the acidity or alkalinity than on the matter of infection. The "primary" calculi included uric acid, ammonium and sodium urate, calcium oxalate, which formed in highly acid urines; and also calcium phosphate (crystalline variety) and calcium carbonate (crystalline), which formed in alkaline urine, whose reaction was not due to infection, or in urines, faintly acid or neutral. The rare calculi of cystin, xanthin, indigo, urostealith and fibrin were also considered "primary".

"Secondary" calculi were found in alkaline urine, whose alkalinity was due to the conversion of urea into ammonium carbonate by so-called urea splitting organisms. These included therefore, ammonium magnesium phosphate and the calcium phosphates and carbonates of the amorphous varieties.

Occurrence of Different Chemical Types of Calculi

Clinically there is no uniformity of opinion as to the relative occurrence of the different chemical types of calculi. The work of Morris, Thompson, Kahn and others is conflicting. A careful review of the many articles given in the bibliography leads me to feel that the variation of opinion is due as much to the fact that no one observer analyzes a sufficient number of stones to draw conclusions, as to the discrepances in technique.

Calcium oxalate, uric acid and urates, phosphates and carbonates seem to be the most frequent constituents in the order named. Kahn would have us believe that calcium oxalate composes the bulk of most stones, with uric acid present for the most part in traces and seldom in amounts over ten per cent. A

few stones contain slight amounts of phosphorus according to this author. However his series included only sixteen calculi, all renal in origin, so that his analyses cannot be taken as holding true in a large series of stones.

Most authors consider the nucleus of the stone to consist of ammonium urate in infancy, of uric acid in young adults, and of calcium oxalate in older persons. The renal stones from patients of all ages tend to contain more oxalate while the bladder calculi are chiefly of uric acid and phosphate. Carbonatic stones, while common in lower animals, are rare in man, although minute amounts of carbonate are frequently found.

Many stones are layered, the different layers consisting not infrequently of different chemical constituents, either pure or mixed. A phosphate-carbonate layer may alternate with an oxalate-urate layer. This feature has been explained by a change in the reaction of the urine during the time of formation of the stone, the phosphate-carbonate layer coming down while the urine is alkaline, and the oxalate-urate layer being precipitated when the urine is acid. Layers of oxalate may vary with layers of urate and here again differences of H ion concentration or of colloidal relationship have been brought forth to explain the cause.

Factors Determining the Shapes Assumed by Calculi

The shape assumed by the calculus is supposed to be determined largely by the site of development of the stone. In my own experience this would seem a factor, but the oxalate "jack-stone" and "mulberry" calculi are difficult to explain on such a basis. Small calculi tend to lie in the minor renal calyces and when found there usually have the contour of the calyx. The shape of stag-horn calculi which ramify into the calyces and at times conform to the pelvis so as to simulate a pyelogram when studied roentgenographically, would support the view that the contour is determined by that of surrounding

structures.

The Microscopic Structure of Calculi

The structure of calculi can only be mentioned in passing. The works of Ord and Shattuck and of Fowler should be consulted for detail. All stones seem to be made up of crystals which are atypical microscopically in size and shape from the usual crystals of similar chemical composition found in voided urine. These crystals are fused together with a matrix of organic material, frequently pigmented in clefts and interstices. Most stones show a tendency to lamination and radial striation. Certain earthy phosphates precipitate from faintly acid or neutral urines and consist of large crystals macroscopically distinct and only partly fused together. These show no lamination. In pure phosphatic calculi firm amorphous material is fused together and the lamination is also absent. Young holds that the nucleus of urate and oxalate calculi is not laminated, but the outer body layers become concentrically striated as they are deposited.

Significant Clinical Features Associated with Calculi

A few clinical features associated with urinary calculi may be mentioned at this point. Stones occur more frequently in males than females, two to one in the Mayo Clinic series (Braasch). Recurrence occurs in only ten per cent of cases after operative removal, where fluouroscopy at the operating table and reray after operation shows that no fragments have been left behind. The development of stone in the remaining kidney following operative removal of a stone-forming kidney is extremely rare, while recurrence in damaged kidneys formerly the site of a large branched stone, is extremely common.

There is no great difference as to the side affected. The stones are bilateral in from ten per cent to fifty per cent of the cases, depending upon the series quoted. Stones are multiple in about one-third of the cases.

Conditions under which Uric acid, Urates, Oxalates and Phosphates are Deposited from Urine.

As the most common constituents of calculi are oxalates, urates, and phosphates, a review of the conditions in which these materials exist in the urine and the conditions under which they are precipitated appears to be warranted. Uric acid, calcium oxalate, calcium phosphate, and ammonium magnesium phosphate are in neutral distilled water entirely insoluble so far as any practical consideration goes. They are the most insoluble constituents of the urine and are present there in smaller quantities than other important crystalloids. Normally about 0.7 gm. of uric acid, 0.015 gm. of oxalic acid, 2.5 gm. of phosphoric acid, and 0.25 gm. of calcium are eliminated in twenty-four hours. There very insoluble substances are held in solution in urine to a far greater degree than they are in water. This property of urine to hold uric acid and calcium oxalate and phosphate in solution has been attributed by most physical chemists, among whom the names of Schade, Bechold and Lichtwitz, are prominent to the presence of so-called protective colloids (Schützkolloide). For instance Lichtwitz has shown that the extraction of colloidal material from the urine by means of benzin will result in the immediate precipitation of phosphates. (I have performed a similar experiment described below.) On dialyzing the urine against water the urinary crystalloids were found by Lichtwitz to pass into the water and be precipitated. The precipitate consisted chiefly of calcium oxalate.

Uric Acid and Urates

We know that uric acid is deposited in acid urine in several forms. It is supposed to be present in solution as the monosodium salt. Monosodium phosphate however, tends to take the sodium from sodium urate and be converted thereby to disodium phosphate. The uric acid thus formed is compara-

tively insoluble. Hence a high percentage of acid phosphates will tend to lower the solubility of uric acid while the neutral phosphates will tend to increase its solubility. Sodium chloride, urea, and the urinary colloids seem to increase the solubility of uric acid. Blatherwick has shown that uric acid is not deposited in a urine whose alkalinity is greater than Ph_7, whereas the acid urines deposit it in increasing amount as the acidity goes below Ph_7.

Oxalic Acid and Oxalates

Oxalic acid owes its presence in the urine according to most authors, to incomplete oxidation of uric acid or of carbohydrate. According to others it is formed in the stomach by carbohydrate fermentation. In excessive oxaluria, it is assumed that this deficiency of oxidation or fermentation assumes a pathological degree. In any crystalline oxaluria the limits of solubility of the calcium oxalate are reached and discrete octohedra separate and tend to be passed in large numbers. Striking is the fact that calculi are seldom seen under such conditions. The precipitation of calcium oxalate takes place best in faintly acid urine. It is deposited readily in alkaline urines also, but is supposed to be held in solution in urines of high acidity. Magnesium salts and acid sodium phosphate tend to increase its solubility to a questionable degree. A dietary or exogenous source of oxalic acid in the urine depends upon the ingestion of certain foods (e.g. rhubarb and spinach). It is doubtful in the extreme if such a diet could have any but a remote influence on calculus production, in the light of the work that follows.

Phosphates

Calcium and magnesium phosphates may be deposited in faintly acid or alkalin urine. The experiments of Lichwitz and of myself (cited later) would seem to show that colloidal material is necessary to carry them completely in solution. However, the urinary reaction is equally important and many

authors attribute the phosphaturia seen in debilitated persons to the lowered urinary acidity rather than to an excessive secretion of phosphates. The phosphates in urine are largely from exogenous or dietary sources. Only one to four per cent come from the disintegration of body tissues.

Calcium and Magnesium

The calcium and magnesium metabolism is of interest in relation to the "water theories" of stone. A few brief notes will suffice. The daily output of calcium is about 0.1 to 0.4 gm. expressed as CaO. By far the greater part of the calcium in health is excreted from the bowel. Intravenous injections and feeding of calcium salts do not materially increase the calcium content of the urine (Clark, Wells, Cushny), most of the metal being excreted through the intestine and little absorbed. In states of low calcium content of the blood and tissues, an excess absorption may be found to take place from the intestine, but with a normal calcium balance in the body an increased ingestion of calcium will not lead to a marked increase in the urine. An increased ingestion of magnesium salts leads to an increase of calcium in the urine and the converse is also said to hold true.

Magnesium is eliminated to the extent of 0.1 gm. to 0.3 gm. per day, expressed as MgO. Its amount depends on the diet. Fifty per cent of that ingested passes through the kidneys, the remainder through the intestines. Disturbances of magnesium metabolism are so far as I know, unrecognized in pathology.

Cystinuria and Cystine Calculi

Cystinuria is an error of metabolism which is inborn and hereditary (Wells, Hoffman, Garrod). Males seem to be affected by the disease twice as frequently as females. Cystin is the amino acid which contains most of the sulphur of the protein molecule. It is ordinarily destroyed in two

ways: in part by conversion to taurin and then to taurocholic acid, which is excreted in the bile; and in part by oxidation to sulphates, which are excreted in the urine. In cystinuria this mechanism seems to be interfered with, probably more in the oxidation to sulphate than in the conversion to taurin. In consequence, large quantities of cystin find their way to the urinary stream. Here they are carried in solution when the urine is alkaline, but an acid urine precipitates the cystin as colorless hexagonal plates. Cystinuria is frequently associated with cystin stones, in most instances the calculi being the first thing to attract attention towards the disease process. How many cases of cystinuria are unaccompanied by calculi is difficult to state. As cystin crystals are precipitated only in acid urine, many cases of alkaline cystinuria may be missed even if the urine is examined microscopically. The familiar occurrence of cystinuria may be one of the explanations of the relatively few cases of calculus in family groups (Poland, Marcet, Golding, Bird). The taking of ammonium carbonate to alkalinize the urine has been strongly recommended in cases of cystinuria as a means of dissolving the cystin and thus preventing the formation of stones.

The structure of cystin calculi is interesting. They seldom show lamination or atypical crystals. The deposit is never amorphous. Fused typical or slightly atypical, hexagonal crystals seem to be fused together. They may attain large size, may be pure in chemical composition, or mixed with other ingredients such as uric acid and oxalates.

Xanthinuria and Xanthin Calculi

Xanthin is a normal urinary constituent. It is the most abundant purine base present. It is nearly always soluble in normal individuals, the appearance of xanthin crystals in the urine being considered an evidence of a pathological condition. However, the nature of this crystalline xanthinuria

is a matter of total ignorance. The condition is very rare. Xanthin is often mixed with uric acid calculi and is rarely found as the chief constituent of stones. Rosenbloom collected six such cases from the literature and reported a seventh of his own.

Rarer Types of Calculi

Two cases of indigo calculi are on record. It is presumed that the indigo was derived from indican by oxidation.

Fatty stones (Urostealiths) have been occasionally described. Their source and chemical composition is veiled in obscurity and I have found little literature on the subject. Horbaczewski analyzed one such specimen and found protein, fatty acids and neutral fats the chief constituents.

Cholesterol calculi have been found in rare instances (Wells). The cause is utterly unknown. Horbaczewski found one in a patient who had previously had cystin calculi.

Fibrin calculi are of infrequent occurrence. These seem to consist of alternating layers of fibrin and calcium phosphate. They are associated with repeated attacks of hematuria, the nature and cause of which is obscure. The review of the subject by Gage and Beale (See bibliography) is of interest. Possibly it is in such cases that Schade's hypothesis of calculi formation may find an application.

The Geographic Distribution of Urinary Calculi

No work of this kind would be complete without consideration of this much discussed subject. An analysis of the literature shows that the conceptions of the present day are largely based on the work of Hirsch in 1886, with several more recent contributions from Chinese and Indian sources. The data are chiefly drawn from hospital statistics, mortality records, and the experience of individual practitioners. Naturally data from well organized

hospitals with properly tabulated case records and autopsy protocols are most valuable. Mortality statistics are based chiefly on death certificate reports and are likely to vary with the training of physicians and the consequent liability to errors in diagnosis. The fact that many patients, perhaps the majority of them in civilized countries, do not die with calculi as a primary cause, also renders such data unreliable. However, on a large scale, mortality records may be of value. The experience of individual physicians contains so much of the personal factor that this seems to me the least reliable source of all.

According to most authors, calculus finds a high incidence uniformly in the following districts, viz. Holland, Syria, Lower Egypt, Persia, Italy, Morocco, and Algiers.

The rarity of the disease in Iceland, Norway, Sweden, Denmark, Finland, Northern Russia, Germany (except endemic), Ireland, East Indies, West Indies, East Africa, Central Africa, West Africa, Nubia, Tunis, Polynesia, Australia, British Guiana, Uraguay, Peru, and Nicaraugua has been emphasized by practically all writers, most of whom constantly quote Hirsch's observations.

Stone seems to be endemic in certain localities, often circumscribed within narrow geographic limits. Among such localities are the Canton province of China (extremely high), India (the Punjab and interior upland districts), Arabia, Germany (Altenberg has a high incidence within a radius of sixty miles, old Bavaria, and certain Alpine districts), England (Norfolk, Bristol, and other isolated areas), Scotland, Italy (especially in Brescia and Cremona), Mexico (Oaxaca), France, and Central Russia (a very high incidence in the region about Moscow).

The relationship of the soil and water to the incidence of stone has been studied only in a superficial manner. Areas where stone is of frequent occurrence and limestone abounds are the basins of the Don and Volga,

the eastern counties of England, Würtemberg, Italy (Brescia and Cremona districts), Syria, Bosnia and Herzegovinia.

Stone abounds in the following countries where limestone is not an abundant constituent of the soil, viz. Canton Province of China, the Island of Mauretius, Indian districts, the Duchy of Altenberg, and Lorraine.

Certain limestone areas notably free from stone are the West Indies, the Barbados, Western Switzerland, many parts of England and America, and the limestone districts of India. In this connection it is to be noted that in the Alpine districts where limestone and calculus are coincident, most of the natives drink rain water which is drawn from cisterns.

As the geographic data for the United States has not been carefully studied, Dr. Frederick L. Hoffman of the Prudential Insurance Company, has been kind enough to carry out an extensive investigation of this point. Three tables, which are based on mortality statistics, and which relate to the incidence of both biliary and urinary calculi are submitted herewith. A careful study of these tables tends to show an increase in the recorded death rate from urinary calculi within the last two decades. There is a certain definite variation between the states, a tendency to parallelism between biliary and urinary calculi deaths in many instances, and an increase in mortality with the advance of age. This is what might be expected when we consider the source of the statistics from a clinical point of view.

On the whole, a study of geographic data leaves me unsatisfied as to a geographic element in the etiology of stones. The tendency to calculus formation in limestone regions, in cold, damp, tropical or temperate countries is not clear cut. It does seem that the extremely cold regions of the far North are peculiarly exempt and that certain districts, especially noteworthy being China and India (mostly vesical), are more liable to this disease than

others. The vesical calculi of China and India seem to be mostly of the uratic and oxalate variety. Uric acid seems to predominate in Europe and oxalate in North America.

Race.- Racial differences seem to play little part. The Jews of Northern Germany, Christians in the Balkan States, and Italians in America are accredited with a high incidence. The Negro race has long been held to be exempt but the tables of Dr. Hoffman throw a new light upon this statement, as a careful study will show that the mortality in this country at least is about equal between the two races.

Heredity.- The familial incidence of cystin calculi has been mentioned. Some authors accredit uric acid calculi to a familial tendency but I fear their conclusions are drawn more from the hereditary nature of gout than of stone. On the whole there is little evidence that heredity is a factor of much importance.

Age.- The greatest discrepancies of opinion occur in regard to age. Some writers consider children almost exempt while others hold that the incidence is extremely high at this time of life. On the whole the greatest occurrence, as judged by time of operation, is in the second, third and fourth decades. Below ten years and above fifty years the incidence becomes much less. However, any age is liable to the disease. Thompson, reporting three thousand four hundred ninety-two operations for calculus (almost all vesical) in Canton Province, found forty-three percent in males under twenty years, forty-one percent from twenty to fifty years, and fourteen percent over fifty years. Only two percent of his cases occurred in females.

Hoffman's statistics corroborate the Mayo Clinic figures which show that males are affected more than females in a ratio of two to one.

Summary of Conclusions from an Analysis of the

Literature on the Etiology of Lithiasis.

1. The problem of calculus formation is one of chemical precipitation. We must endeavor to discover what mechanism causes urates, oxalates, phosphates etc. to be precipitated in such a way that a fused hard concretion arises, rather than that individual crystals continue to maintain their state of isolation in their passage through the urinary tract. Whatever mechanism be found at fault, I feel certain that the veracity of this statement will remain unaffected.

2. Differences of reaction as determined by the H ion concentration, and qualitative and quantitative changes in the colloidal materials of the urine (such as pigments, mucin, nebecula, albumin, and nucleoalbumin) have been shown to influence the nature of urinary sediments both chemically and physically. The microscopic and gross features of calculi would seem to show that an abnormal variation of these factors is at work in stone formation.

3. Race, heredity, occupation, diet, sex, trauma, geographic distribution, and occupation seem to offer little suggestion as to the etiology of concrements. If factors at all, they are probably of secondary importance.

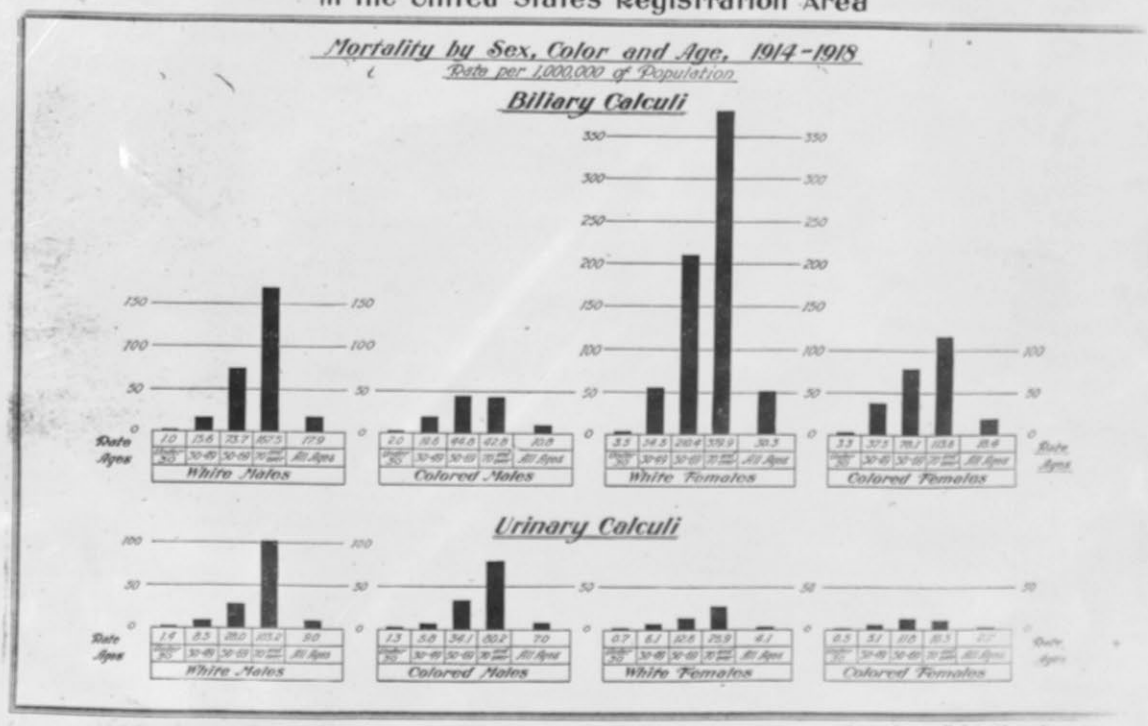
4. The three commonest diseases associated with a visible increase in the crystalline content of the urine, viz. gout, oxaluria and phosphaturia, are found far more frequently without accompanying calculus deposition than with it.

Xanthinuria and cystinuria are undoubtedly necessary to the formation of xanthin and cystin stones but how frequently xanthinuric and cystinuric patients escape calculus formation is problematic.

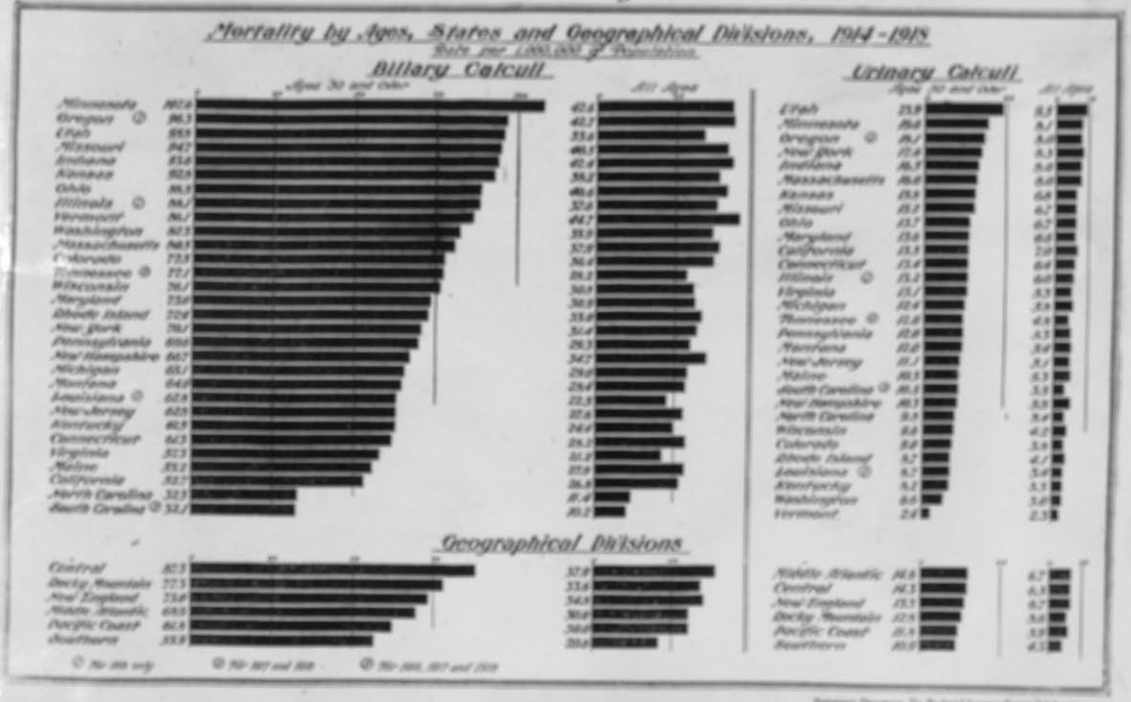
5. On the whole the clinical evidence rather points to a local mechanism at work in the kidney pelvis or in the bladder, as the cause

of stone. The work of Rosenow and Meisser, together with the frequent occurrence of demonstrable foci of infection in patients suffering from calculus, and the almost universal finding of infected kidneys or bladders associated with stone lend tremendous weight to the idea that a specific stone-forming infection is at work. There is little direct evidence that anatomic factors or stasis can initiate the stone forming process but their frequent association with calculus, makes it seem likely that the stone-forming mechanism may work to better advantage under such conditions.

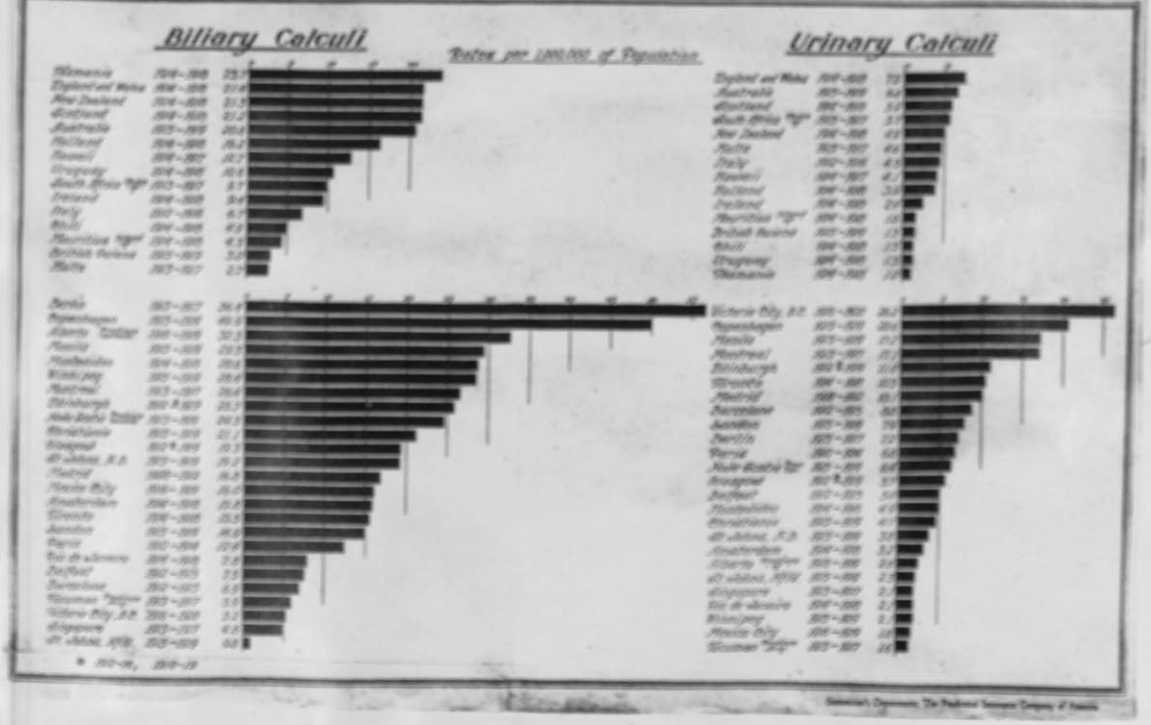
Mortality from Biliary and Urinary Calculi
In the United States Registration Area



Mortality from Biliary and Urinary Calculi
In the United States Registration States



Mortality from Biliary and Urinary Calculi
Foreign Countries and Cities



Experimental Studies.

At the outset it was decided to subject the several commoner theories of calculus formation to laboratory experimentation wherever possible, being guided by the preceding studies to a great degree. The magnitude of such a task is obvious and has necessarily prohibited my following up many details which, although important, did not seem matters of immediate expediency. To keep the experiments simple and uncomplicated, with the idea ever foremost to find new avenues of approach to the solution of the problem has been my aim. The following report will give an abstract of each type of experiment and its results with a typical protocol when necessary to give detail.

Series I. An effort was made to concentrate the calcium salts in rabbits' urine by means of administering massive doses of oxalate, chloride, lactate, and phosphate of calcium to normal rabbits, and also to animals, which had been subjected to a low grade toxic nephritis by the administration of such drugs as oxalic acid, bichloride of mercury, cantharides, and chloroform, in minute doses.

A. Seven rabbits were fed according to the following schedule.

Experiments 1 - 2 - 3 - 4 - 5 - 6 - 7.

Table I	Sodium Bicarbonate Oxalic Acid	1 gm.) 0.1 gm.)	orally by stomach tube
	Calcium Lactate Calcium Oxalate	0.1 gm.) 0.5 gm.)	suspension in glucose solution injected intramuscularly
Table II	Calcium Lactate Calcium Chloride	1 gm.) 1 gm.)	in solution orally by tube
	Calcium Lactate Calcium Oxalate	0.1 gm.) 0.5 gm.)	suspension in glucose solution injected intramuscularly
Table III	Sodium Benzoate Oxalic Acid	1 gm. - 0.1 gm.-	given orally in capsules given orally in solution by tube.
	Calcium Lactate Calcium Oxalate	0.1 gm.) 0.5 gm.)	suspension in glucose injected intramuscularly

Table IV	Calcium Chloride	1 gm.	)
	Calcium Lactate	1 cm.	) in solution orally by tube
	Hydrochloric Acid 20 c.c. of 1% sol.		)
	Calcium Lactate	0.1 gm.	) suspension in glucose
	Calcium Oxalate	0.5 gm.	) injected intramuscularly

Feeding according to Table I and Table II was carried out on alternate days for one week, the idea being to keep the urine alkaline. Then feeding according to Tables III and IV on alternate days was maintained, the purpose being to keep the urine acid.

Results:

1. The seven rabbits lived seventeen, twenty-seven, twenty-seven, fifty-four, thirty-three, nine and fourteen days, respectively.

2. Aside from an occasional calcium oxalate crystal, no visible crystalline increase in urinary sediments was noted.

3. The reaction of the urine was maintained with fair success.

4. A mild nephritis was found at autopsy consistently.

5. No evidence of lime salt deposit in the tissues or concrement formation was obtained.

B. Four normal rabbits were fed according to the following schedule: Experiments 8 - 9 - 10 - 11.

Table I	Bichloride of mercury	1 cc to 250 sol.	)
	Acid sodium phosphate	1 gm.	)
	Ammonium chloride	1 gm.	) orally by tube
	Magnesium oxide	1 gm.	)
	Calcium Lactate	0.1 gm.	) injected intramuscularly in sus-
	Calcium Phosphate	1 gm.	) pension in glucose solution.
Table II	Calcium Chloride	1 gm.	)
	Ammonium Chloride	1 gm.	) orally by tube in solution
	Hydrochloric Acid 20 cc of 1% sol.		) and suspension.
	Magnesium Oxide	1 gm.	)
	Calcium Lactate	0.1 gm.	) injected intramuscularly in
	Calcium Phosphate (neut.)	1 gm.	) suspension in dextrose solution.

Table III	Sodium Bicarbonate	1-3 gn.	)
	Acid Sodium Phosphate	0.1 gn.	) orally by tube in solution
	Magnesium oxide	1 gn.	) and suspension
	Ammonium Chloride	1 gn.	)
	Bichloride of mercury, ½ c.c. of 1-250 solution - injected intramuscularly		
	Calcium Lactate	0.1 gn.	) injected intramuscularly in a
	Calcium Phosphate	1 gn.	) suspension in glucose solution
Table IV	Calcium Chloride	1 gn.	) orally in solution and
	Calcium Lactate	1 gn.	) suspension by tube
	Calcium Lactate	0.1 gn.	) injected intramuscularly as
	Calcium Phosphate	1 gn.	) suspension in dextrose solution

Feeding was carried out according to Tables I and II on alternate days for one week, the urine being maintained alkaline. Then feeding according to Tables III and IV was maintained on alternate days to the end of the experiment, the urine being (in most instances) kept acid to litmus.

Results:

1. The four rabbits lived twenty-four, sixteen, twenty and ten days. Two were killed by accidental intubation of fluid into the trachea.

2. A moderate degree of albuminuria was obtained. A toxic nephritis was consistently observed at autopsy.

3. No visible crystallin increase in calcium or "triple" phosphate was noted.

4. No concretions were found nor was deposition of calcium salts in any tissues noted.

Here, as may be seen by the tables, we endeavored to cause an increased deposit of "triple" phosphate by forcing the feeding of materials from which ammonium magnesium phosphate is derived. Bichloride poisoning is frequently associated with calcium deposit in the renal tissues. Therefore the use of this drug and the value of the negativity of this experiment is to be emphasized.

C. Five rabbits were placed on a feeding schedule as outlined under B. with the exception that 0.5 to 1 c.c. of chloroform was fed instead of Bichloride of Mercury at the places indicated in the tables. Chloroform was used because of its well-known action upon the renal epithelium. Experiments 16 - 17 - 18 - 19 - 20.

Results:

1. The five rabbits lived seventeen, eight, nineteen, fourteen and thirteen days, respectively.

2. The liver and renal epithelium constantly showed signs of degenerative changes.

3. The visible crystalline content of the urine was not noticeably affected.

4. No concretions or deposits of calcium salts were noted in any tissues.

D. Two rabbits were placed on the feeding schedule outlined under B. with the exception that 1/10 minim of Tincture of Cantharides was substituted at the places where Bichloride of Mercury is indicated in those tables. Tincture of Cantharides was used because of its well-known effect in causing congestion and irritation of the kidney. Experiments 21 and 22.

Results:

1. The rabbits lived twenty-one, and ten days respectively.

2. At autopsy the kidneys and livers of both animals showed degenerative changes in the epithelium.

3. No concretions or deposits of lime salts were found in any tissues.

E. Controls.

(A) Three rabbits were fed on the schedule outlined under

A. with the exception that no oxalic acid was given at the places indicated in those tables.

Results:

1. The animals lived five weeks, seven weeks and six weeks respectively.

2. No visible increase in the crystalline content of the urine was noted at any time.

3. No deposit of lime salts in the tissues or concretions were found.

(B) Controls. Three animals were fed on the schedule outlined under B. with the exception that no Bichloride of Mercury was given at the points indicated in the tables. Controls 16 - 17 - 18. This is essentially the condition of experimentation outlined under A.

Results:

1. The rabbits lived fourteen, sixteen and twenty days, respectively.

2. No visible increase in the visible crystalline content of the urine was noted at any time.

3. No concretions or deposits of lime salts were found in the tissues.

Conclusions from Series I.

1. The forcing of lime salts by mouth and intramuscularly in large doses is unassociated with microscopically apparent increase in the crystalline content of the urine of rabbits.

2. This is good negative evidence that an increased ingestion of lime salts in "hard" water or that the factor of lime containing soils would per se predispose to concrement formation.

3. These experiments corroborate and are in entire harmony with the views of present day pharmacology that an increased ingestion of calcium salts with a normal calcium balance will not lead to a significant increase of this metal in the urine.

Series II. An effort was next made to increase the visible crystalline content of the urine by other means. Ebstein had shown that uric acid on oral administration is destroyed in the metabolism of rabbits and dogs and it had long been known that oxalic acid upon being administered, would cause the appearance of calcium oxalate in the urine. Therefore in different groups of animals, oxalic acid, sodium oxalate and ammonium oxalate were administered in sublethal dosage.

A. Experiments 25 and 26.

Two rabbits were fed oxalic acid 0.4 gm. in solution daily by a stomach tube. Acetamide, 0.4 was given at each feeding. The use of acetamide was suggested because of the fact that oxamide or diamido-oxalic acid had been shown to produce concretions on feeding. We wished to include the amide grouping in the dietary to exclude the possibility of this radical having any bearing on the process of concrement formation.

Results:

1. The two rabbits lived twenty-four and thirty-eight days respectively.

2. A visible precipitation of a moderate number of calcium oxalate was noted daily in the urine. These crystals were always single, of the perfect octahedron type, and showed no tendency to coalescence or fusion.

3. Experiment 25. At autopsy there were found deposits of white material in the straight tubules in the renal pyramid. These proved to be made up of calcium oxalate crystals when scraped with the knife and examined

microscopically. These crystals were of the octahedron type and showed no tendency to fusion. In Experiment 26, no such deposit was found.

4. Both animals showed a slight vascular injection of the kidneys and epithelial degeneration in the tubules.

5. No deposit of concretions was noted.

B. Experiments 13 and 14.

Two rabbits were fed ammonium oxalate - 0.5 gm. daily.

Results:

1. The rabbit in Experiment 13 lived forty-seven days. That in Experiment 14 lived ten days.

2. An occasional calcium oxalate octahedron was noted in the urine.

3. Aside from a slight nephritis, no changes were noted at autopsy. No concretions, crystalline infarcts, or deposits were found.

Experiment 27.

A rabbit was fed ammonium oxalate - 1 gm. and acetamide,- 1 gm., daily by stomach tube. The animal died after fifteen days.

Results:

1. At autopsy a subacute tubulo-glomerular nephritis was found.

2. There was no evidence of deposition of crystals or concretions.

3. An occasional calcium oxalate crystal was noted in the urine. These were perfect isolated octahedrons in type.

C. Experiment 15.

A rabbit was fed sodium oxalate, 0.5 gm. every two days. The animal died after forty-six days.

Results:

1. A moderate number of calcium oxalate crystals were noted

daily. These were perfect octahedra and showed no tendency to fusion.

2. At autopsy the kidneys were found scarred and contracted with nephritis.

3. No deposition of calcium salts or concretions were observed.

Series III Inasmuch as the oxaluria produced under Series II was not apparently intense, it was decided to try feeding an ester of oxalic acid and an alcohol in the hope that it would be less toxic, as it would probably contain the oxalate ion in less concentration than the soluble salts of this acid. A few preliminary experiments were carried out with ethyl oxalate but this drug was found to kill the animals almost immediately when given intramuscularly in doses of $\frac{1}{2}$ c.c. or more. Butyl oxalate was next chosen for trial and was injected intramuscularly into thirteen rabbits in doses of $\frac{1}{2}$ to 1 c.c. daily. In several instances sodium oxalate was simultaneously fed by capsules in doses of 0.4 gm. daily. Experiments 28 - 31 - 32 - 33.

Results:

1. The rabbits lived from four days to nine days.

2. An intense oxaluria was consistently developed.

3. In all the experiments but one, Experiment 28, the calcium oxalate crystals were perfect octahedra showing no tendency to coalescence or fusion.

4. Experiment 28 - Butyl oxalate, 1 c.c. and sodium oxalate, 0.4 gm. given daily. Died spontaneously after six days. A marked oxaluria with crystals of dumb-bells and atypical octahedra was noted. At autopsy a marked distention of the intestines with injection of the mucosa was noted. The bladder was distended and contained about twenty small smooth white stones about 2 mm. by 1 mm. These proved to be calcium oxalate on chemical examination. The bladder was markedly injected. As this was the first experiment of the

series, no cultures were taken, and no sections were made of the tissues. The kidneys appeared slightly injected but otherwise normal. An effort to repeat the experiment proved futile.

Conclusions: The association of an atypical crystalloid form of oxaluria with formation of concretions is noted in one instance, while the remaining animals consistently showed a typical octahedron form of calcium oxalate in the urine with no concrement formation. This merely suggests that a change in crystalline form may be related to concrement formation.

Series IV. The Result of Placing an Organic Nucleus Into the Renal Pelvis of the Rabbit.

The presence of organic nuclei such as fragments of epithelium, disintegrating tissue, etc. having been observed in many calculi, it was decided to determine the effect of placing pieces of tissue into the pelvis of the rabbit's kidney. The following technique was carried out in Experiments 102 - 103 - 104 - 105.

Under ether anaesthesia and aseptic technique, the left kidney was exposed through a lumbar incision and a nephrotomy was done. A wedge shaped portion of the renal pyramid, not quite large enough to completely fill the cavity of the pelvis was excised. This was gently placed in the renal pelvis and the kidney carefully sutured with No. 00 catgut. The incision was closed in anatomic layers. The animals lived from six to one hundred twenty-one days, being sacrificed after the longer periods. The piece of tissue was found lying in a pool of urine and under-going disintegration in all cases. In Experiment 102, when the animal was sacrificed after one hundred twenty-one days, the pelvis was found clear, no trace of the tissue being seen. There was no evidence of calcareous deposit and the kidney seemed normal aside from operative scar. A culture from the kidney was negative for bacterial growth.

In view of this negative result, bits of muscle and fascia from the lumbar region, were introduced into the left renal pelvis of several rabbits, according to the technique above outlined, through a nephrotomy wound. Kidney and incision were closed. Several such animals are still living. One was sacrificed one hundred twenty-two days after operation (Experiment 108). Fig. XX. Here the disintegrating muscle tissue was found to be impregnated with lime salts, lying in a pool of purulent material, in a kidney which showed a moderate degree of hydronephrosis. A positive culture of a gram positive bacillus was grown from the pus. One half of the muscle tissue was firm and stony while the other half was soft and gritty.

We await with interest the outcome of further autopsies on animals of this series when the muscle tissue has been allowed to remain in situ for a longer period.

Conclusions: No conclusions can be drawn from a single experiment, but it would seem that foreign bodies or organic nuclei in themselves will not initiate stone formation, but if retained long enough they may form by irritation a locus minoris resistentiae for bacterial invasion. Under such conditions they become impregnated with crystalline material and this may develop into a true calculus. It is interesting to note that in the cases where bits of renal pyramid were inclosed in the pelvis, no infection was determined by culture. This was also the case in several animals which died within a week after the insertion of the muscle tissue into the kidney pelvis.

It is to be noted that the filling of the pelvis with bits of muscle tissue is equivalent to introducing stasis of high degree and also to increasing many times the number of surface reduplications in the pelvis. Hence it would seem that the failure to get a deposit of crystalline material in the absence of infection is a strong point against a purely mechanical theory

of stone formation.

Series V. A Study of the Formation of Oxamide Calculi.

The formation of concretions in the urinary tract after the administration of oxamide to animals had apparently been regarded as a medical curiosity since the time of Ebstein. Little or no mention of the process occurs in current literature, especially in this country. The fact that oxamide would form concrements had been confirmed in Mann's laboratory by J. R. McVay who had fed two dogs on oxamide over a period of several months. Hence it was decided to study this phenomenon in more detail in the hope that from observations on the development of this artificial concrement we might be able to gain some insight into the genesis of calculi seen clinically.

A. Fourteen rabbits (Experiments 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68) were fed oxamide orally in capsules in doses of 0.4 gm. to 0.8 gm. daily.

Five dogs were fed oxamide in doses of 2 gm. daily by the same method. Dogs D-740, D-741, E-189, E-207, E-260, E-337).

Results.	Duration Life.	Oxamide Fed.	Cause of Death.	Sediment, Concretions and Location
1. Expt. No.56	10 da.	8 gm.	Coccidiosis Nephritis	Two min.concretions rt. pelvis. Sand in left. Fig. I.
No.57	10 da.	8 gm.	Nephritis.	Few particles sand lt. pelvis. Stone in Bladder. Fig. XV.
No.58	17 da.	11 gm.	Nephritis.	Min.concretions in both pelves Fig. IV.
No.59	7 da.	4.8 gm.	Nephritis	Fine sand in rt. Left clear.
No.60	18 da.	12.8 gm.	Nephritis	Fine sand in calices both sides.
No.61	26 da.	13.4 gm.	Nephritis	No deposit.
No.62	17 da.	9.6 gm.	Nephritis.	Rt.clear. Few flecks of sand on left.
No.63	26 da.	9.6 gm.	Pneumonia	No deposit.
No.64	10 da.	8 gm.	Pneumonia	Few flecks rt. Left clear.
No.65	41 da.	28 gm.	Accidental	Fine sand both kidneys, slight amount.
No.66	7 da.	10 gm.	Accidental	No deposit noted.
No.67	20 da.	28 gm.	Ether.	Small stone rt. ureter. Marked sediment rt. Left slight deposit.
No.68	20 da.	28 gm.	Ether	Few flecks rt. Left clear.
No.69	25 da.	20 gm.	Pneumonia	Rt. clear. Left infarcts in pyramids. Few small stones.
Dog D-740	9 mo. 5 da.	130 gm.	Ether	No deposit noted. Not fed 3 mo. before death.
D-741	5 mo.10 da.	140 gm.	Accidental	Stones rt. kidney. Lt. clear. Hydronephrosis.
D-260	7 mo.	324 gm.	Nephritis.	Stones both kidneys and ureters.
E-207	36 da.	40 gm.	Pneumonia	Small stones both kidneys.
E-189	14 da.	20 gm.	Pneumonia	Infarcts rt. kidney; few small stones rt. Left clear.

The above outline shows the duration of life and the total amount of oxamide fed in each experiment, as well as the finding of deposits at autopsy when present.

It will be noted that after an interval varying from a few days to several months there appeared irregularly in the kidneys, ureters, or bladders of about 75 percent of the animals an amount of yellow crystalline material varying from traces of fine sand to concretions as large as 1 cm. in diameter. Definite concrements of sufficient magnitude to consider stones occurred in about 50 percent of the animals fed. Smaller sized deposits were the rule. The deposit was frequently bilateral, with no preference as to side.

2. The larger concretions showed definite radial striations and concentric markings suggestive of deposition in layers.

3. The deposits were more frequently multiple than single.

4. Five animals (about 25 percent) showed no deposit after long feeding.

5. The passage of sand and smaller concrements was frequently noted, being found in all animals whose 24 hour specimen was collected by means of metabolism cages.

6. Cultures from the kidneys and the urine showed the process to be consistently sterile.

7. The animals frequently lost weight, especially on long feeding. All showed a variable degree of toxic degenerative change in the epithelium of the liver and kidneys with an increase in the vascular injection of these organs. The liver especially was found firm, with an increase in fibrous tissue in the experiments of longer duration. No intracellular crystalline deposits or stones in the parenchyma were noted. Albuminuria without pus, casts, or red cells was a consistent finding. In several instances an atypical reduction of Fehling's solution was observed, but a true glycosuria was never demonstrated.

8. The reaction of the urine as roughly determined by litmus seemed to affect the process in no way, some of the animals putting out an acid and others an alkaline urine. Three rabbits (Expts. 78-79-80) were fed sodium

benzoate daily in doses of 1 gm. with the oxamide. The urine was usually acid. Two of these showed a moderate amount of oxamide deposition. Four rabbits (Expts. 81-82-83-84) were fed magnesium oxide 1 gm. daily with the oximide. The urine was consistently alkaline. One of these animals developed definite concretions in the kidney and others showed traces of sand deposition.

9. Extending the duration of feeding, increasing the daily dosage of oxamide to as much as 10 gm. (Expts. 76-77-67-68); varying the fluid intake in 24 hours (Expts. 69-70-71-72-73-74-75), seemed to produce within wide limits no quantitative change in the amount of oxamide deposition found in the urinary tract at autopsy. On complete water starvation the excretion of oxamide became less as the animal approached death. A toxic nephritis produced by bichloride of mercury (Expts. 85 and 86) likewise had no apparent effect on the process.

The Crystallography of Oxamide Calculi

B. As we have noted Rainey, Ord and others had suggested a relationship between calculi and abnormal crystalline forms which had occurred when the crystalline material was deposited from gelatinous or colloidal media. Accordingly a study of the crystalline morphology of the oxamide passed in the urine and a study of the structure of the oxamide stones became a matter of much moment. Within 24 to 36 hours after the feeding of oxamid to animals there appeared in the voided urine abnormal crystals. These were pigmented in varying degree, the color ranging from a light brown to a brownish black. The crystals were doubly refractile in the dark field when the brownish black color was found to be really a deep orange. The morphology of the crystals can be readily appreciated by referring to Figs. 2-3-5-6-7-8-9-10-11-12-13 and 14. Many of the crystals were isolated and single. Others were clumped together somewhat after the manner of agglutinated blood cells. However, dotting the

field in numbers varying with the specimen were masses of crystals in every degree of coalescence and fusion. There were in the different specimens a number of morphologic varieties of crystals. All seemed to be developed from a simple cross-like form (Fig. 3 and 4). There were crosses with smooth arms and crosses with rough pointed arms; crosses with the interbrachial spaces roughened and partly filled out; squared forms which seemed to be crosses with the interbrachial spaces entirely filled; and finally spheres, rough and smooth on their surfaces. These seemed to be a rounded variety of the squared form and the end product of development of the cross. The perfect smooth sphere appeared umbilicated and had radial striations proceeding from its center (Fig. 11-12). The size of the crystals varied from microscopic dust to larger crystals which attained 40 to 50 micra in diameter.

All of these varieties of crystal forms were found to some degree in states of fusion, but in this connection several features were noted early and were confirmed by a lengthy daily study of urinary sediments from many animals.

1. There was a tendency for all of the forms to coalesce and fuse, but this tendency was greater the more nearly the crystal approached the spheroidal form. Likewise the smaller the crystal and the lighter its pigmentation the more likely was it to be found fused with other crystals.

2. The converse was also true. The closer the crystal approached the cross form, the larger it was, and the greater its depth of pigmentation, the greater were its chances of being found isolated and unfused with other crystals. This seemed a very significant point.

3. Smaller concretions passed with the urine as well as those found in the kidney were invariably found to be made up of crystals of the type just described. In the larger concretions the individuality of the cryst-

als was found to be in some degree lost, but even here on crushing the stone, particles resembling crosses and spherical forms were observed.

4. Every animal, fed on oxamide, passed in the urine almost if not quite all of the different morphologic varieties of crystal types, but there was a great tendency for one or the other of them to predominate. Depending upon this to some degree was the likelihood of finding calculi in the kidneys at autopsy. Putting this in another way, it was noted that animals with a urine, in which small, light pigmented, smooth or rough spheres predominated, would show larger particles of sand in their 24 hour specimens, and the deposits found at autopsy in these animals would be of the larger, harder type. However, the animal with a predominance of large, dark brown spheres or crosses alone (associated with few spheres in the microscopic field), would pass only finer particles of sand and would present only fine grains of sand or smaller soft concretions in the urinary tract at autopsy.

Hence we see that there seemed to be some relationship between the type of crystal and the size and kind of calculus formed.

C. The chemistry of oxamid - Its reactions in Urine - The Chemistry of Oxamide Stones.

A rather superficial study of the chemistry of oxamide and its reactions in normal animal and human urine in vitro have given some results which appear to me particularly enlightening in their relationship to oxamide stone formation. Oxamide is the diamide of oxalic, having the formula $\overset{CONH_2}{\underset{CONH_2}{|}}$. While insoluble in cold water it goes into solution slowly in boiling water and is gradually hydrolized by further boiling to ammonium oxamate and later to ammonium oxalate. Acids or alkalins hasten this hydrolysis. As prepared synthetically by treating an oxalate ester with ammonia it consists of small, irregularly rhomboidal fissured, translucent, colorless crystals which at times

show splitting and roughened ends. (Fig. 21).

Oxamide stones, as has been noted, show all degrees of structure from fused aggregations of crystals, individual units of which may be made out on crushing and examining under the microscope, to more solid completely fused forms where the identity of the individual crystals is almost, if not quite, lost.

These concretions give the reactions of oxamide, yielding ammonium and oxalic acid ions on prolonged boiling in water or upon being dissolved in an acid or alkali. They also contain an organic pigment material the identity of which I have not yet been able to establish.

Synthetic oxamide upon being heated with human urine is taken up in solution at 90°-100° C. in an average quantity of 50 mg. to 10 c.c. of urine. Part of it is undoubtedly hydrolized and is not reprecipitated on cooling. However, some of it is reprecipitated and this precipitate upon being examined microscopically gives a surprising result. The crystals have become pigmented and have assumed the forms which we have noted in the crystals passed in the urine of animals fed on oxamid. Also there is to be noted a corresponding visible diminution in the pigmentation of the urine from which the crystals have been precipitated. All types of oxamide stone forming crystals have been noted under such conditions from the simple cross to the sphere.

Another striking observation was this: if we take the artificially prepared stone-forming crystals or the sediment from oxamide-fed rabbit's urine and boil this with water, a clear pigmented solution is finally obtained. If this solution, while hot, is filtered through a heated charcoal filter several times, the pigment and possibly other organic colloids are removed by a kind of fractional adsorption, some material being removed in each filtration. If this is done ten to fifteen times, the precipitate obtained on cooling

comes to assume an entirely altered form. The crystals are colorless and approach very nearly the form of oxamide crystals prepared synthetically.

Finally, we must emphasize an experiment in vitro which seemed to settle the question as to the process of oxamide stone formation. By repeated precipitation of increments of oxamide from human or animal urine into the same test tube over a period of days, fusion of crystals has been obtained and in vitro I have been able to produce small concretions which microscopically and chemically, so far as can be judged by simple tests, resemble small oxamide concretions found in the urinary tract of animals fed with the drug.

Conclusions from Series V: We may repeat in the test tube the same process that takes place in the urinary tract of the animal, and further we can extract by a sort of fractional adsorption the pigment material from the crystals deposited by oxamide fed animals, under which conditions a reversion to a simpler crystalline form takes place. Whether the pigment alone is responsible for the change, or whether there are other organic materials present, I cannot say. However, one conclusion can be drawn and held as established. The oxamide concretion is the product of a normal colloidal mechanism of the urine. In other words oxamide being secreted into the urine of animals is so precipitated that a loose physico-chemical union with organic materials (probably normally present) in the urine takes place. As a result of this unstable union the crystalline form is changed and individual crystals tend to coalesce with one another and become fused together. I have seen this process in every stage, both in the test tube and in the urine of oxamide fed animals. It speaks loudly for a colloidal precipitation mechanism in the formation of concrements anywhere in nature. The action of normal urine is specific so far as causing fusion of oxamide crystals is concerned. This is certainly not the case for crystals of oxalates, urates, etc. passed normally by human beings. The urinary

colloidal machinery tends to keep these crystals separate, unfused, and isolated under normal conditions. But oxamide is a crystalline material foreign to the normal urinary tract. It is the only abnormal or normal crystalloid of which I know that can be concentrated in the urine in massive amounts over an extended period of time by feeding. Our oxalate experiments most nearly approached this, but never did we obtain oxaluria in any degree comparable with our oxamiduria. Once by forcing the dosage of butyl oxalate we did obtain abnormal crystals and concretions. But this isolated experiment could not be repeated. That the sole factor is not the intensity of the oxamiduria is shown by the fact that the animals formed concrements in short time and small dosage and within wide limits the dosage or duration of feeding seemed to play little part. Hence we must conceive of the urine as being unable to handle oxamide excretion. The very colloidal factors that enable it to carry the normal uric acid, oxalates, phosphates, etc. in solution may cause it to throw the foreign crystalloid oxamide from solution in the form of fusing crystals.

This conception of oxamide stone formation differs entirely from that of Ebstein, who thought that the oxamide became held up in the cells of the straight tubules of the kidney and that the deposition of oxamide around these dying cells was the origin of the stone. Personally, I have not seen the intracellular deposits of oxamide in the microscopic sections either with fresh frozen tissue, with frozen fixed tissue, or with paraffin sections. Never in the voided urine have I noted the epithelial debris impregnated with oxamide. Ebstein probably paid more attention to his toxic nephropathy than to his oxamid crystalline masses in urine itself and to this I attribute the difference in our results.

The deductions I have made for oxamide concrement formation are obviously entirely in harmony with the work of Rainey, Ord and Shattock, Fowler,

Schade, and with our clinical experience with urinary, biliary and other concrements.

Series VI. A study of Anatomic or Mechanical Factors in Concrement Production.

Having established the type of mechanism fundamentally at fault in the production of oxamide calculi and by analogy and in accord with other observations, assuming that an abnormal colloidal mechanism in the urine is at work primarily in the formation of concrements seen clinically, it became a matter of immediate interest to see how anatomical or mechanical factors would alter the picture. We thus might gain some insight into the reason why stasis, diverticuli, organic nuclei, and reduplication of surfaces are so frequently associated with stone, but are as often, if not more often, found unassociated. The results in this connection were striking and I feel that they will explain fully the role of the so-called anatomic factors.
It will be remembered, first of all, that only 50 percent of our animals presented stones at autopsy, while 25 percent of them showed no trace of oxamide deposit. Two animals fed for so long a period as 26 days showed no deposit. Another, fed for six months and then having the feeding omitted, died three months later. No oxamide deposit was found. It should also be remembered that undoubtedly most of the small fusion masses of oxamide crystals are washed on in the urinary stream and voided with the urine, a fact which our collected 24 hour specimens clearly revealed. Also every animal fed on oxamide presented, at some time or other, sand in the urine while fusing crystals were consistent, the degree of fusion depending on the crystal type predominant. What then causes the retention of the precipitate that is to grow to the larger stone ? Obviously we think at once of stasis, diverticuli and surface reduplication as great accessory factors in holding back the fusing crystals.

I offer the following observations in proof of this conception, so far as the oxamide calculus goes:

1. The first site of deposition of oxamide in the kidney of the dog or rabbit was invariably in the ramifications of the calices along the line of attachment of the pelvis to the renal sinus. In the rabbit and dog there is a tiny pocket behind the cusp of the free margin of the calyx (See Fig. 19) and in these pockets the concretions were usually found to lie. Here is a site of reduplicated surface and a point where the normal urinary stream and the musculature of the pelvis probably exerts less force in washing crystalline material onward. The tendency of smaller urinary concrements in human beings to lie in the minor calices should be remembered in this connection.

2. The effect of traumatizing the renal pelvis on the deposition of oxamide concrements. (Expts. 91-92) Technique: Under ether anesthesia and with aseptic technique the left kidney of the rabbit was delivered thru a lumbar incision and the renal pelvis exposed. With a fine needle twenty punctures of the pelvis were made in different areas, care being taken to pierce thru and thru each time. The bleeding was controlled, the kidney replaced in situ, and the wound closed. Upon feeding oxamide to two animals thus treated it was found in both that there was an excess of deposit of oxamide and larger concretions on the left, or traumatized side, than upon the untreated right side, where no deposition happened to occur in either instance. Here we had roughened the pelvis, produced a minute reduplication of surfaces with microscopic diverticuli, in all probability. Cultures from the kidneys of both of these animals were entirely negative.

3. The effect of low grade stasis and slight hydronephrosis upon the deposition of oxamide concrements. To determine the effect of hydronephrosis and stasis alone, a rubber band was placed about the left ureter

in such a manner as to constrict it slightly (Experiment 93). The animal was fed on oxamide gm. .8 daily for eight days. This animal showed acute hydronephrosis on the left side at autopsy with no oxamide deposit at all. This was also found to take place in several preliminary experiments, which were done in trying to establish the technique. Hence I concluded that I had constricted the ureter on the left side to such an extent as to interfere with the pressure equilibrium of urinary secretion, so that neither the oxamide nor other material had been secreted upon that side.

I then modified the technique as follows: A left abdominal incision was made, the left ureter exposed by packing off the intestines, the peritoneum stripped away, and a rubber band was placed around the ureter so as to fit it snugly. No effort was made to constrict the ureter. The periureteritis and swelling, which took place in consequence of the presence of the foreign body (rubber band), was depended upon to constrict the ureter slightly. Upon feeding oxamide to rabbits thus treated (Experiments 94-95-96), we consistently obtained stones and a marked increase in deposition of sand on the treated side in a few days' time. On the untreated right side only a few flecks of oxamide were found in any instance. In Figure 18 the animal had been fed only nine days. The treated left side is filled with sand and minute concretions. The untreated right side is clear. This is a striking demonstration of the role which stasis plays in the retention of fusing oxamide crystals and in the growth of oxamide stones.

4. <u>The effect of foreign bodies upon the deposition of oxamide calculi</u>. Experiment 97. The left kidney was opened with a nephrotomy incision. A piece of blood vessel silk was placed upon the cut surface, running from the tip of the pyramid to the cortex. The free end was allowed to remain in the kidney pelvis. The animal was fed oxamide 0.8 gm. daily for 29 days. At

autopsy the left kidney was filled with several small stones and the thread was encrusted with a deposit of oxamide. The right untreated kidney also had several deposits in its calices, but there was much less here than on the left.

Experiments 88-89-90. Technique: Under ether anesthesia the left kidney was exposed thru a lumbar incision and a nephrotomy made. Bits of muscle and fascia were placed in the renal pelvis and the incision closed. Three rabbits thus treated were fed on oxamide. They lived 30 days, 9 days, and 54 days respectively. In every instance the tissue became encrusted with oxamide deposit, and in Experiment 90 the animal developed a concretion which entirely filled the renal pelvis and measured $\frac{1}{2}$ x 1 cm. This concrement lay in a pool of pus from which a gram positive staphylococcus was grown on culture. The rabbits which lived for 9 days and 30 days showed a negative culture, from the kidneys and urine. Hence we conclude that the presence of infection in Experiment 90, where the animal lived 54 days, was due to secondary invasion of bacteria to the locus minoris resistentiae produced by the presence of the foreign body. See Fig. 16 and Fig. 17.

Summary of Conclusions Concerning the Formation of Oxamide Calculi as Deduced from Series V and VI.

1. Oxamide, on being fed to animals, is specifically precipitated from the urine by a normal colloidal mechanism, in such a way that the unit crystals tend to fuse in loose chemico-physical union with certain of these colloids.

2. The concretions, formed as the result of this fusion, have the general physical features of calculi seen clinically.

3. The process is sterile at least at the beginning.

4. A considerable amount of the material thus deposited is

passed on with the urine, but an occasional fragment is retained in the calices or at points, where the surface tension between the minute fragment and the wall of the urinary tract epithelium is great enough to resist the force of the urinary stream.

5. Reduplication of surfaces, diverticuli, stasis, hydronephrosis of such a degree as to not markedly upset the preseure equilibrium of urinary secretion, and organic nuclei greatly enhance the chances of retention of the fusing crystals, and in this manner foster the formation of larger stones.

6. Oxamide concretions while sterile at the outset probably act after a time as foreign bodies and invite bacterial invasion which may in turn cause an increase in concrement growth.

Series VII. The Introduction of Inflammatory Exudates into the Urinary Stream by means of Experimental Renal Infection.

The influence of such Exudates upon the Deposition of Calcium Oxalate and the Production of Small Concretions by such Methods.

The mechanism of oxamide stone formation having been established at least in principle, it was felt that an analogous process must take place in calculi, seen clinically in human beings. In normal human and animal urine, when the saturation limit for oxamide (0.02 to 0.03 mg. per 100 c.c.; see footnote) is reached, this foreign crystalloid is precipitated as fusing crystals. On the other hand, we recall that the saturation limit for the normally present oxalates, urates, carbonates, etc. being reached, we have a deposition of crystals which normally do not tend to fuse. I shall speak of

---

N.B. In Expts. 67-68 the oxamide output was determined by hydrolysis to oxalic acid and quantitatively by means of a modification of the Salkowski-Autenreith tecnique for this material. The 24 hour urines contained from 0.02 to 0.03 gm. oxamide (as oxalic acid) per 100 c.c. urine after the sediment had been removed by filtration.

this later when considering the defensive mechanism of the urinary tract against stone formation.

What if abnormal colloidal material such as that contained in pus or inflammatory exudates be introduced? Will the normal precipitating mechanism of the urine be disturbed, and under such conditions what will the result of concentrating oxalates in the urine as we did in Series II.? Inflammatory exudates contain amino acids, albumin, nucleoprotein, albumoses, proteoses, peptones, enzymes, lecithin, fat, cholesterol, purine bodies, held for the most part in colloidal solution. Most of these materials are present in traces or not at all in normal urine.

Technique. Experiments 35-36-37-38-39-40-41-42-43-44-45-46-47-49: Minute doses of colon bacilli (Both 24 hour and old cultures) were injected directly into the left kidney by means of needle puncture, the kidney being located by palpation under anesthesia. Sodium oxalate 0.4 gm. was fed daily in thirteen instances. In Experiment 49, butyl oxalate 1/2 c.c. was given daily. The fourteen rabbits thus treated developed suppurative pyelonephritis consistently. This caused their deaths after a period varying from several days to two weeks. Three animals of the series (Experiments 35-36-44) showed evidence of atypical crystalline deposit in the calices. The largest of these deposits was about 0.5 mil. No evidence of crystalline deposit was found in the kidneys of the remaining animals. The crystalline morphology of the calcium oxalate excreted has not yet been determined under such conditions. We expect to carry forward this phase of the work now.

In Experiment 49 where butyl oxalate was fed after injection of a stool culture from the rabbit into its own left kidney a number of atypical crystalline particles were seen in the urine. We found, however, no concretions.

Series VIII. The Production of a Minute Concrement by Means of Trauma and Infection.

One experiment yielded a positive result, which although not entirely clear, should be reported, I feel, as a matter of fact. Cushing traumatized the gallbladder of a dog and soon thereafter introduced into a vein an attenuated culture of typhoid bacilli. An acute cholecystitis ensued and minute concrements were found in the gallbladder. This suggested a similar experiment upon the kidney.

Experiment 50: A left nephrotomy was made thru a flank incision under anesthesia and aseptic conditions. The renal pelvis was thoroughly traumatized by pinching with mosquito forceps, the kidney and wound being then closed. One c.c. of a twenty-four hour culture of colon bacilli, which had been obtained from a calculus removed clinically, was injected into the ear vein of the animal. After eight days the rabbit died. There was acute pyelonephritis and at autopsy several small doubly refractile crystalline bodies about 1/2 mm. in size were found in the calices. They were not associated with the line of suture. Repeated efforts to repeat this process (Experiments 51, 52, 53, 54) have proved futile. Whether the strain of bacterium, which was associated with the concrement in Experiment 50, was that of a specific stone producing bacillus, which lost its power in subculture, or whether the concretions were encrustations upon necrotic tissue or not, is an open question. I feel that no conclusion can be drawn from a single experiment which cannot be repeated.

Conclusions from Series VII and VIII.

1. Inflammatory exudates are possibly associated experimentally with a change in morphology of calcium oxalate crystals when deposited in urines containing such exudates. Several small concretions made up of atypical fused

crystals have been observed under such conditions.

2. Further experimentation along this line offers a hopeful avenue of approach.

Summary of Conclusions From the Foregoing Experimental Study.

1. I have endeavored to point out the difficulty with which calcium salts, oxalates, urates, and other normal urinary crystalline deposits are increased to any appreciable degree by various methods of administration. The only marked increase in visible crystals has been attained by feeding oxamide, a material foreign to the urinary stream of animals and man. From this it would seem that an exogenous or dietary increase in crystalline materials would not produce a corresponding pathological increase of these materials in the urine. On this basis, oxalurias, phosphaturias, xanthinurias, cystinurias, and excessive uric acid output must be more of endogenous origin than dietary. Some defect of metabolism rather than of diet is at work.

2. Of the four methods by which artificial concretions have been obtained only the feeding with oxamide gave consistent results. There were suggestive features in connection with the other experiments which pointed to some abnormal precipitation mechanism being at work.

3. Oxamide, a crystalloid foreign to the urinary tract finds no mechanism present, on being excreted, to isolate its crystals and keep them separate as morphologic entities. Rather it is precipitated with colloidal material of normal urine in such a way that fusion of crystals and concrement formation ensues.

4. Mechanical factors are effective, at least in the case of oxamide, in promoting retention and growth of the stones, but they are not essential to the process.

5. The fact that pus and exudates of bacterial origin are,

aside from nephritis, the most commonly known means by which abnormal or pathological colloidal material may gain entry to the urinary tract makes it seem most likely that these constitute the mechanism by which our normal urinary precipitation apparatus is upset. As a consequence of this disturbance there is a change in crystalline morphology,and the crystals, combined with colloidal material, no longer remain isolated, but fuse. These fusion bodies are for the greater part passed with the urine, but many are retained and other crystals deposited on them add to the mass, so that a concrement arises. It must be emphasized that while this explanation is to my mind logical it cannot be held as proved for calculi seen in human beings.

Resume'

It is far from my intention to add another theory of calculus formation to the many that have been proposed. The work has only begun and all one can hope to do is to arouse interest in a field of research which has received little attention for a number of years. However, a careful analysis of the literature, and my own experiments, as well as those of Rosenow and Meisser, have suggested to my mind several fundamental ideas, which seem logical and consistent with clinical facts.

A Protective Mechanism against Stone Formation

Let us for a moment consider our problem from another point of view, namely, why do not all animals form stones in the urinary tract ? The human being puts out on an average, 2 to 4 gms. daily of chemicals, which yield precipitates, which are practically insoluble in water. What is it that prevents these insoluble materials from precipitating from solution and becoming encrusted upon the walls of the urinary tract, or clogging it with concretions, just as pipes carrying lime water of excessive hardness become covered with calcium carbonate scale ? Obviously Nature has provided a defensive mechanism against such deposits in animal excretory organs, more especially in the urinary tract.

1. Protective Colloids

The ability of the urine to hold in solution a moderate quantity of material, which is practically insoluble in water, constitutes our first line of defense against calculus formation. This property of the urine is related as we have seen to the H ion concentration and to the protective colloids (Shützkolloide), as emphasized by Lichtwitz.

An Experiment to Demonstrate the Effect of Urinary Colloids on Holding Water-Insoluble Material in Solution.

Recently I have performed a simple experiment to show the influence of the urinary colloids in holding in solution material which is not soluble in water.

Take 100 c.c. of urine of high color. This must be clear and a small specimen should not give a cloud on being brought to boiling temperature. Filter the specimen at room temperature through a charcoal filter. The filtrate is almost colorless compared with the unfiltered specimen. Pigment and other colloidal materials have been removed by the adsorptive action of the charcoal. Now heat the faintly colored filtrate gently. A cloud of variable intensity appears, due to the precipitation of phosphates. This cloud clears on adding a few drops of 1.5% acetic acid.

Just as Lichtwitz by extracting urinary colloids with benzin was able to precipitate phosphates from solution, here also by adsorption we have removed colloidal material from the urine and in consequence phosphates have been precipitated, showing that the presence of colloids in urine is necessary to maintain phosphates in the state of solution.

2. The Precipitation of Crystals as Isolated Units.

Hence the urinary "Shutzkolloide" are the first factors which prevent stone formation in the urinary tract. The second factor of stone prevention, lies in the ability of the urine when it becomes supersaturated, to deposit its crystalline material in isolated, morphologically complete crystal units, which have no tendency to fusion or coalescence.

3. The Form and Activity of the Urinary Tract.

The third natural protection against stone formation may be found in the anatomy of the urinary tract. The calices lead to the pelvis

with open, unobstructed orifices; the pelvis is funnel-shaped and actively contractile, as are also the ureter and bladder, all tending to push any foreign body forward with the urine. It is remarkable that so early a writer as Aretaeus was familiar with this natural defense for he says, "Nature therefore did well in forming the cavity of the kidneys oblong, and of equal size with the ureters, and even a little larger so that if a stone formed above, it might have ready passage to the bladder". The work of Wislocki and O'Conor on the introduction of small glass beads into the renal pelvis is interesting in this connection. So if we should have in spite of our first two mechanisms, viz. the protective colloid, and isolation of crystals, a formation of small deposits of fused crystals, then this third active extruding factor would come into play. This idea is borne out by the frequent passage of small multiple stones, which is seen in many clinical cases.

If we think of the biliary tract in this connection, we remember that it is not such an open system. The inactivity of its contractions, its sinuses of Luska, valves of Heister, tortuous cystic duct, ampulla of Vater, and sphincter of Oddi, all offer obstruction to the extrusion of crystalline deposits. This may be a significant factor in explaining the greater frequency of gallstones over urinary concretions.

4. Natural Defense against Over-Concentration of Urinary Crystalloids.

As we have seen, dietary considerations seem to be of little practical significance in stone formation. Otherwise most people who live on similar diets would have stone, if the diet were one which favored it. This we know is not true. Nature has again provided against such a catastrophe; as we have seen, the crystalline content of the urine is not markedly affected by exogenous causes.

Except in the case of xanthin and cystin calculi, no metabolic

error has ever been demonstrated to be consistently associated with lithiasis. In gout, oxaluria, and phosphaturia, our natural defenses against stone formation are actively at work. As a result stone is the exception in such cases and not the rule.

Cystinuria and xanthuria present unusual crystals in the urinary stream. Their similarity to our artificial oxaniduria in this respect is noteworthy. Here again we may conceive that the urinary colloids and precipitating mechanism is inadequate to properly handle cystin and xanthin, and as a result a tendency to stone formation takes place.

Calculi due to Abnormal Colloidal Matter in Urine

I can find then, no other reasonable explanation for the formation of calculus than that the process is due to a disturbance of the normal colloidal mechanism of the urine, either in holding water-insoluble materials in solution or in precipitating crystals as isolated entities. This disturbance may be due to quantitative or qualitative change in the colloids normally present, or to the entrance of abnormal colloids to the urine, either from the blood stream or from the products of local disease in the kidney. The later origin seems most likely.

Specific Exudates from Specific Infection the Source of Stone-Forming Colloids.

Bacterial infection seems to offer a most plausible source of the abnormal colloidal material necessary for stone formation. Inflammatory exudates contain albumin, nucleoprotein, lecithin, fats, fatty acids, and other colloids, which are foreign to the normal urinary tract. We must emphasize here, one point. Not every abnormal colloid introduced into the urinary stream will be associated with atypical crystalline deposits or stone. Were this the case, diabetes, nephritis, and pyelitis would be consistently

associated with stone. This leads us again to but one conclusion. The stone-forming colloid must be specific, and if due to bacteria, the bacteria which produce it must be specific.

Thus we go back to the doctrine of Mechel von Hemsbach who conceived of a "stone forming catarrh". The demonstration of the elective action of bacteria in different diseases and especially in those of the kidney, the formation of gall-stones, of alkaline phosphatic cystitis, and within the past few months, of renal concretions by Rosenow and his co-workers, almost add the final word to this idea of concrement formation.

The Role of the Mechanical Factors

We see in stasis, surface reduplication, organic nuclei and diverticula, conditions, which do not cause stone, but which, if the cause of stone be present, will greatly favor stone growth. This has been demonstrated to be the role of mechanical factors in the case of oxanid and all our clinical evidence supports such a view for calculi in human beings.

A Clinical Application of this Conception of Calculus Formation.

The clinical application of this conception of calculus formation will, I believe, strengthen the position I have taken. The stone-forming kidney becomes easily understood. Here there is an infected kidney which pours abnormal colloids into the urine. These are constantly causing precipitation of crystalloids and fusion of the crystals precipitated. We readily see why the removal of such a kidney is attended by so little recurrence of stone; why the recurrence of bladder stone is so frequent; why bad teeth, and tonsilitis, conditions of lowered nutrition, states of inactivity, alcohol, carbohydrate fermentation in the stomach, and other conditions of lowered bodily resistance, are accused by different authors of playing a part in the process of lithiasis.

We do encounter difficulty when we attempt to explain the high

incidence of stone in India and China, the association of uric acid gravel and uric acid infarct in children, the alleged association of stone with previous injuries to the vertebral column and spinal cord, and other similar more obscure features of lithiasis. But here again we may find that certain physical, climatic, geographic, or physiological differences at times lower the resistance to stone-forming infections, and this would be our tentative explanation.

Attention should be called also to the occasional instances of spontaneous disintegration of calculi. Englisch collected over one hundred such cases which he considered authentic and a number of others which were questionable. This phenomenon has been explained as being due to a re-solution of the organic framework or matrix of the stone. Rosenbach showed that human kidney stones placed in to the renal pelvis of animals, would diminish in size. This suggests that solution of the organic matrix may take place under unusual circumstances.

Conclusion.

We are just beginning to get light on this most mysterious process and much remains to be done. We must look to the biochemist and physical chemist to lead in the study of the complexities of urinary solution and the states of materials held in this solution. As yet our knowledge of the urinary colloids and pigments is very slight in a true physico-chemical sense. In trying to deal with things of this order we immediately find ourselves on the borderline of scientific knowledge.

On the clinical side we must endeavor to find out what bacteria are associated with the process, and whether or not there are specific strains for each chemical in the urinary calculus forming group. We must inquire why one patient has an oxalate and another a uratic or phosphatic calculus; why one individual has a single small stone for years, while another passes many

of such size, and still another presents himself with both kidneys filled with branching stones and no history of ever having passed even gravel.

These are the broader aspects of the problem and serve to impress us with its magnitude. However, I am more than ever inclined to believe that a proper cooperation of biochemists, bacteriologists, and urologists will ultimately clear up these points and bring the problem of urinary lithiasis to its proper solution.

Fig. I.-Expt. 56-Rabbit- Fed Oxamide 0.8 gm. daily for 10 days. Minute Concrements are Seen in Both Kidneys.

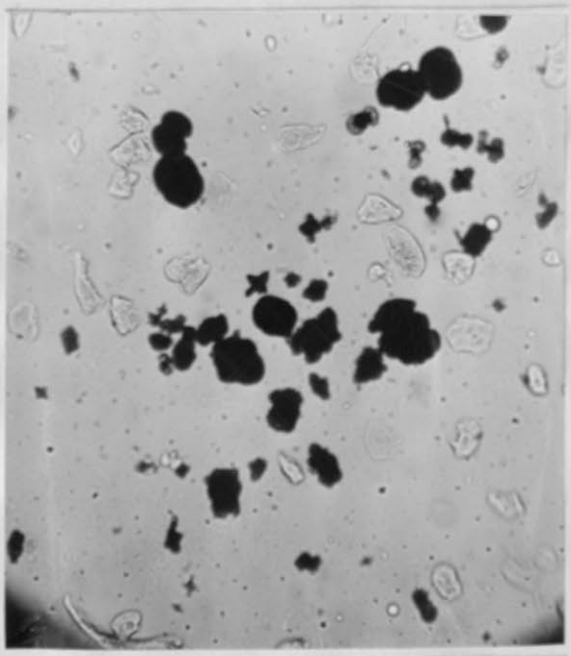


Fig.II.-Expt. 56- Rabbit Fed Oxamide. Urinary Sediment- Crosses-Dark Pigmented Spheres- Agglomerations are Composed of Fused Small Spheres.

7-20 2M

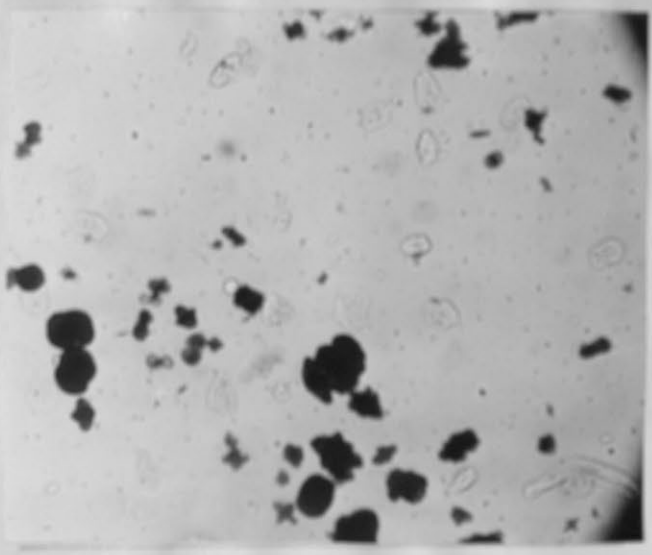


Fig. III.-Expt. 56- Rabbit Fed Oxamide. Urinary Sediment-Shows Tendency of Large Black, Spherical Crystals to Remain Isolated or to Fuse in Twos or Threes. Rougher Agglomerations Made up of Smaller Spheres and Crosses. These are Beginning Concretions.

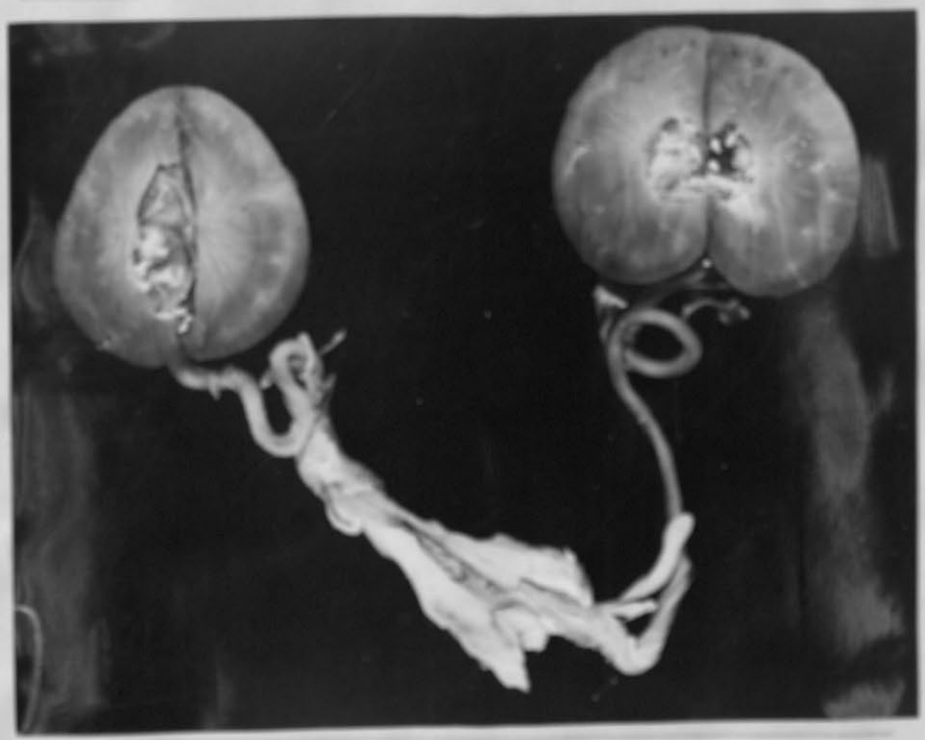


Fig. IV.- Expt. 58- Rabbit Fed Oxamid Daily for 17 da. Bilateral Lithiasis- Note that Larger Masses Lie behind the Free Margin Where the Calicesare Attached to the Renal Parenchyma.

Fig. V.- Expt. 61.- Rabbit Fed Oxamide- Note the Different Phases of Crystalline Morphology Present in this Sediment and in that Shown in the Next Two Figures, viz. Simples Crosses, Crosses with the Interbrachial Spaces Partly Filled, and Square Forms.

There is Little Tendency to Fusion of Crystals. This Animal had No Deposit in Kidneys or Bladder at Autopsy.

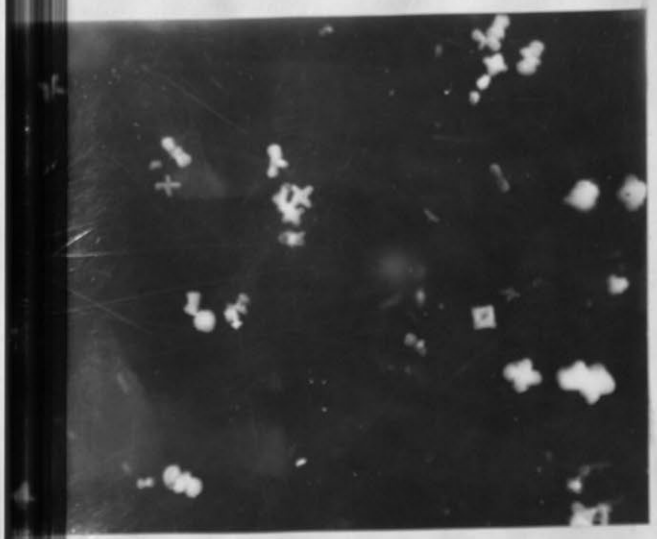


Fig. VI.- Expt. 61- Rabbit Fed Oxamide. Urinary Sediment.

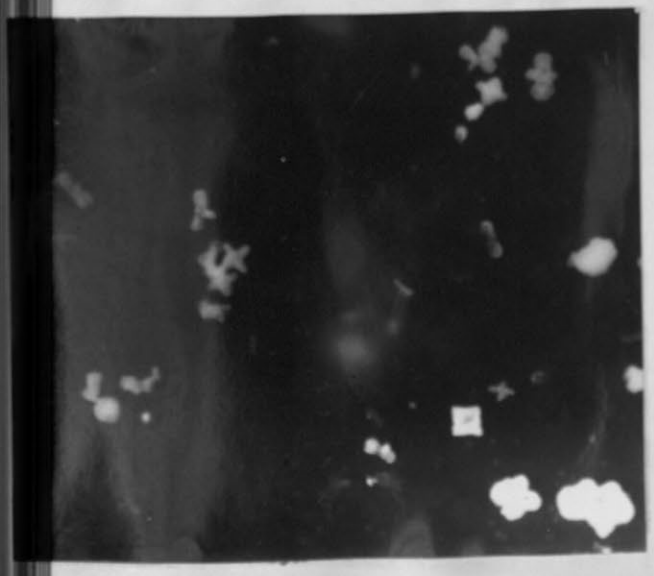


Fig. VII.- Expt. 61- Rabbit Fed Oxamide. Urinary Sediment.

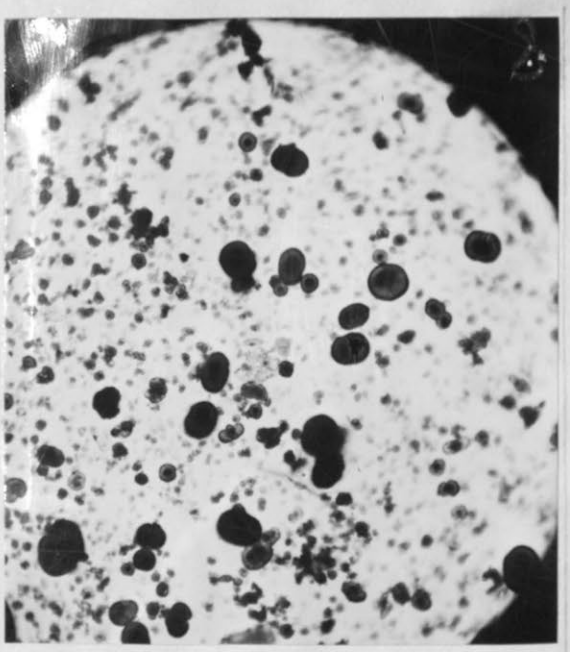


Fig. VIII.- Expt. 68.- Rabbit Fed Oxamide. Sediment from 24 hr. Urine. The Large, Darker, Smooth Spheres Tend to Remain Isolated or to Fuse in Groups of Two or Three.

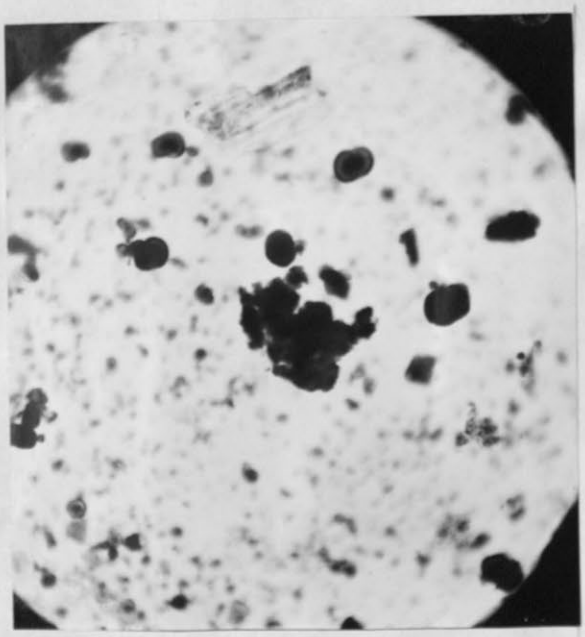


Fig. IX.- Expt. 68.- Rabbit Fed Oxamide. Sediment from 24 hr. Urine. A Small Agglomeration or Beginning Calculus Composed of Smaller, Rough Spheres.

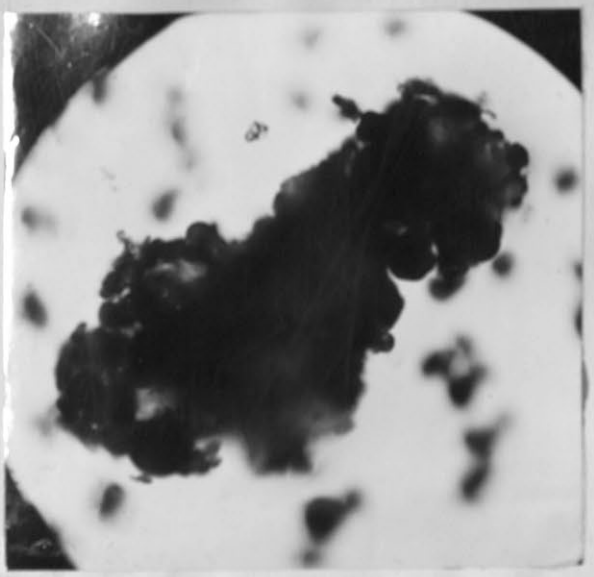


Fig. X.- Expt. 68.- Rabbit Fed Oxamide-Sediment from 24 hr. Urine. A Larger Agglomeration or Beginning Calculus Composed of Small Spheres and Crosses.

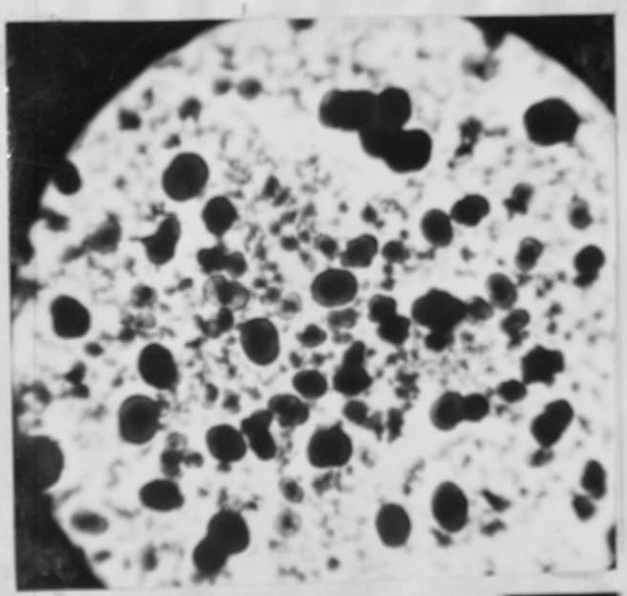


Fig. XI.- Expt. 68.- Rabbit Fed Oxamide. Sediment from 24 hr. Urine.

The Larger, Smooth Spheres Tend to Remain Isolated.

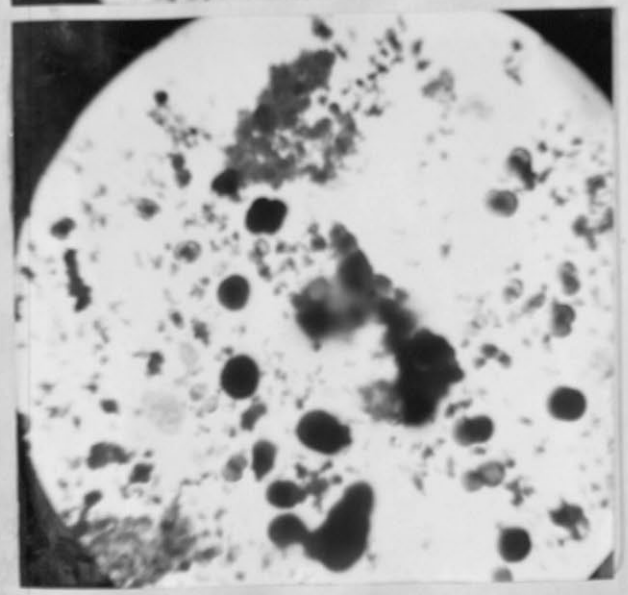


Fig. XII.- Expt. 68.-Rabbit Fed Oxamide. Sediment from 24 hr. Urine.

Note the Agglomerations of Small, Light Colored Crystals. The Larger, Dark Crystals Tend to Remain Isolated.

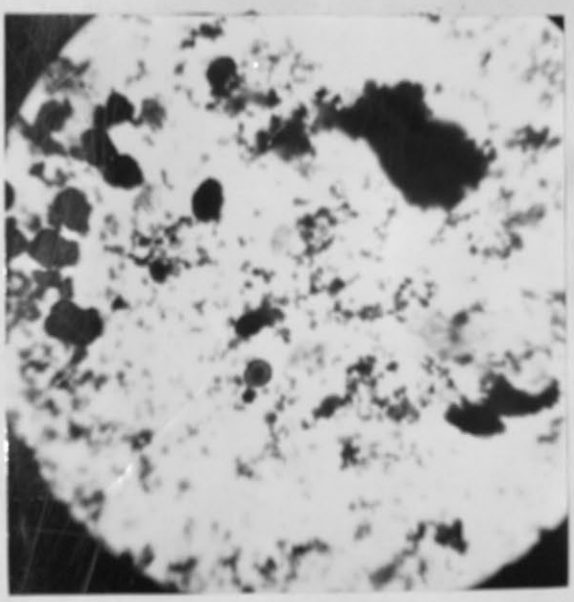
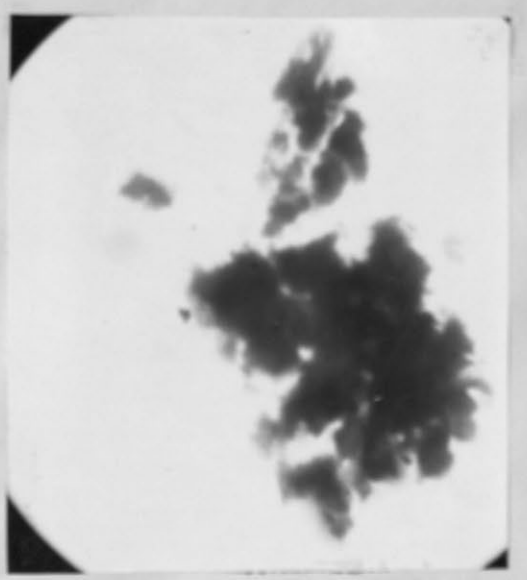


Fig. XIII.- Expt. 68- Rabbit Fed Oxamide. Sediment from 24 hr. Urine. A Small Concrement Made up of Minute, Lightly Pigmented Crystals.

Fig. XIV.- Expt. 68- Rabbit Fed Oxamide. Sediment from 24 hr. Urine. A Small Concrement Which has been Broken up. It is Found to Consist of Lighter Colored Crystals Which have Coalesced and Fused.



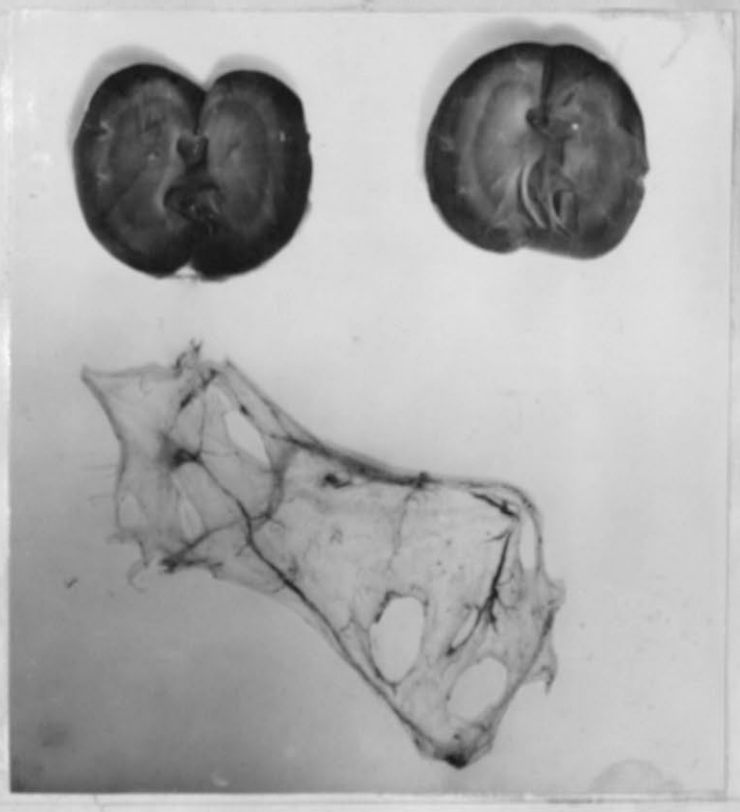


Fig. XV.- Expt. 57.- Rabbit Fed Oxamide.
Showing a Small Stone in the Bladder.
Minute Concretions of Oxamide in the Kidneys.

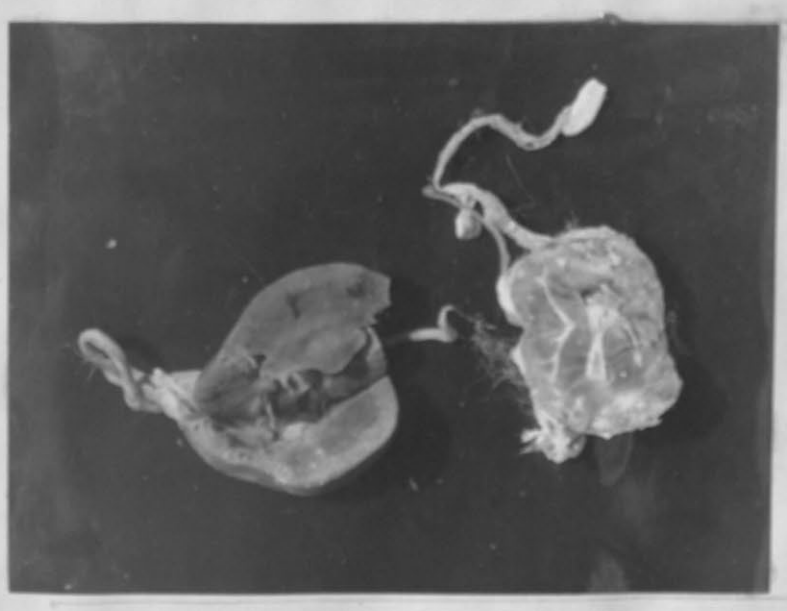


Fig. XVI.- Expt. 89.- Rabbit Fed Oxamide.
Showing Formation of Oxamide Calculus about an
Organic Nucleus or Foreign Body(Muscle Tissue),
Previously Placed in the Left Renal Pelvis.

Fig. XVII.- Expt. 90.- Rabbit Fed Oxamid. Showing the Formation of an Oxamide Calculus about an Organic Nucleus or Foreign Body(Muscle Tissue), Previously Placed in the Left Renal Pelvis.

The Right or Untreated Kidney is Clear of Deposit. A Striking Example of theRole Played by Stasis, Reduplication of Surfaces, and Minute Diverticula.

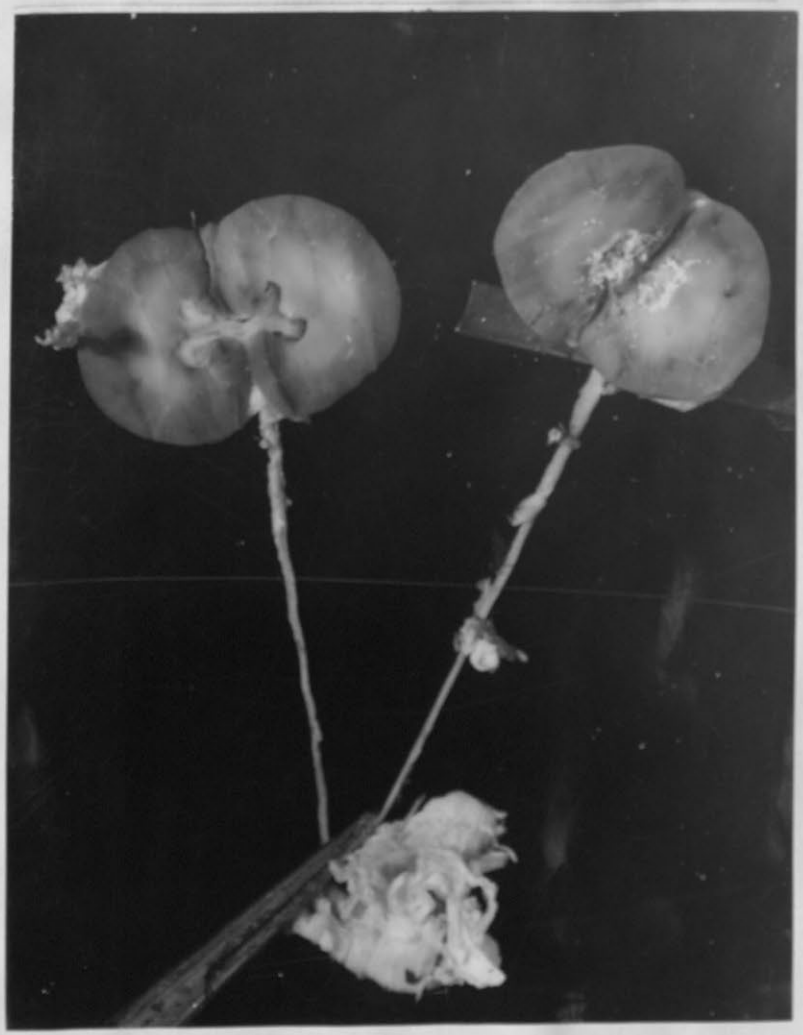


Fig. XVIII.- Expt. 96.- Rabbit Fed Oxamide for 9 Days. A Rubber Band was Previously Placed around the Left Ureter so as to Cause Subsequent Inflammatory Reaction and in this Manner Produce Slight Constriction.

Marked Deposit of Oxamide in Left Kidney as the Result of Low Grade Stasis. The Untreated Right Kidney is Clear.

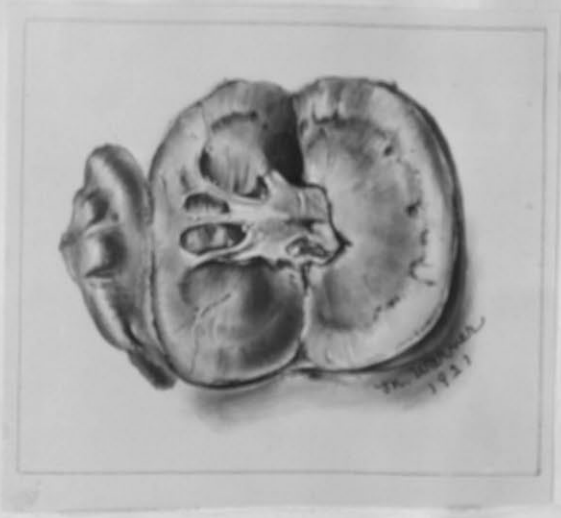


Fig. XIX.- Dog E 207.- Fed on Oxamide. Showing How the Calices are Attached to the Renal Parenchyma in the Dog and Rabbit in Such a Way as to Form Little Pockets behind the Cusps. These Pockets are the First Site of Oxamide Deposition.

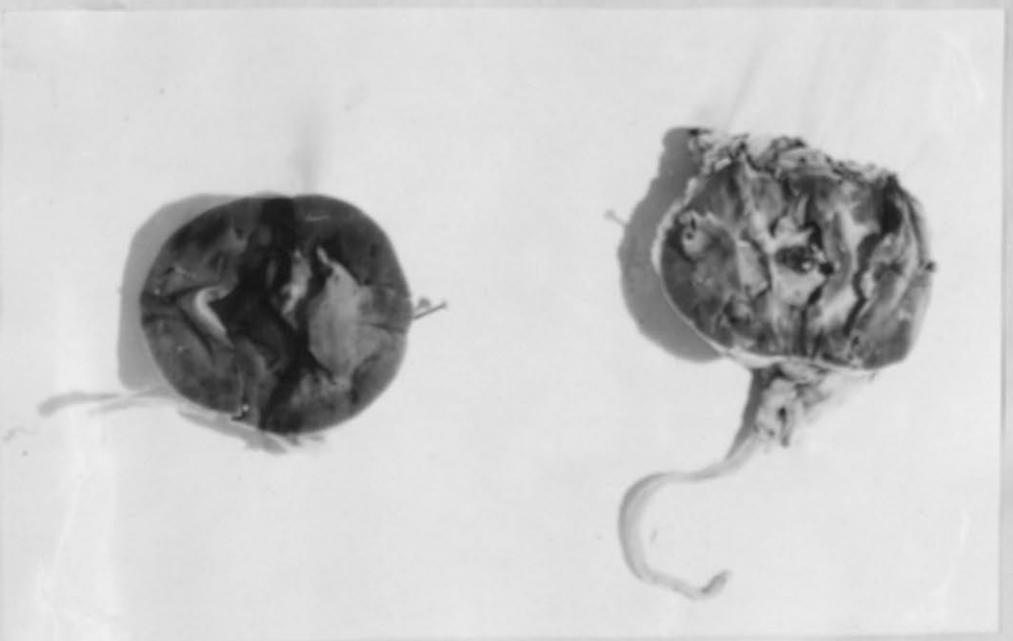


Fig. XX.- Expt. 108- Muscle Tissue Placed in Left Renal Pelvis has Become Impregnated with Crystalline Material After a Period of 4 Months. Presence of Infection was Noted in this Experiment.

The Rabbit was Fed Neither Oxamide nor Other Crystalline Material. Had the Usual Laboratory Diet.

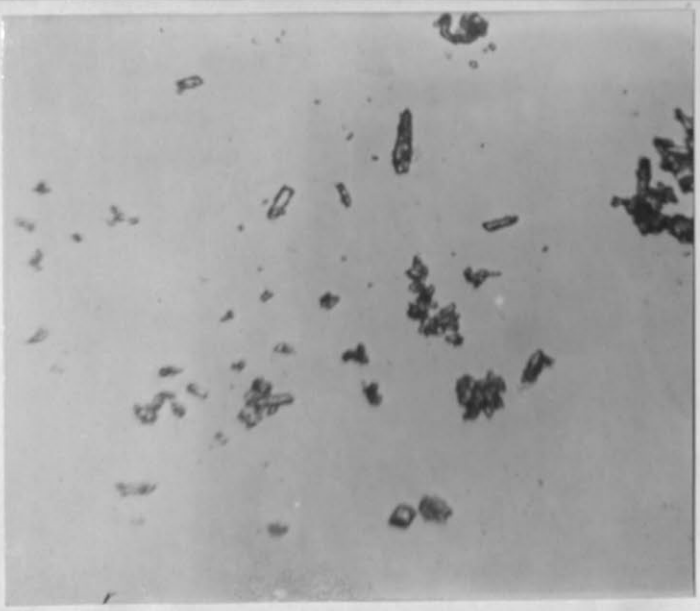


Fig. XXI.- Crystal of Pure Oxamide as Synthetically Prepared by Treating andOxalate Ester with Ammonia.

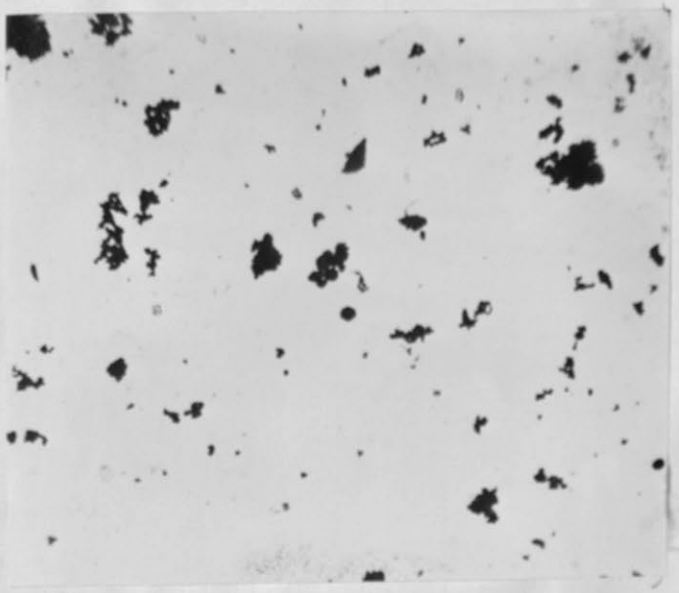


Fig. XXII.- Deposit Obtained When a Great Excess of Oxamide is Dissolved in Boiling Urine and Allowed to Precipitate from Solution on Cooling.

The Crystalline Morphology has Changed, The Crystals have become Pigmented, and Show Tendency to Fusion.

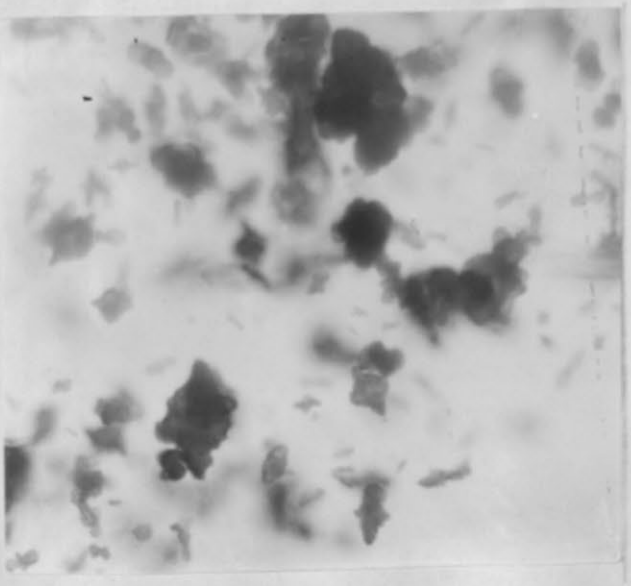


Fig. XXIII.- Another Illustration of Deposit Obtained When Synthetic Oxamide is Dissolved in Boiling Urine and Allowed to Precipitate Out on Cooling.

The Crystalline Morphology has Changed (Developing Cross Type), The Crystals are Pigmented, and Show Marked Tendency to Fuse into Small Agglomerations. This Sediment is Indistinguishable from that in the Urine of Oxamide-Fed Rabbits.

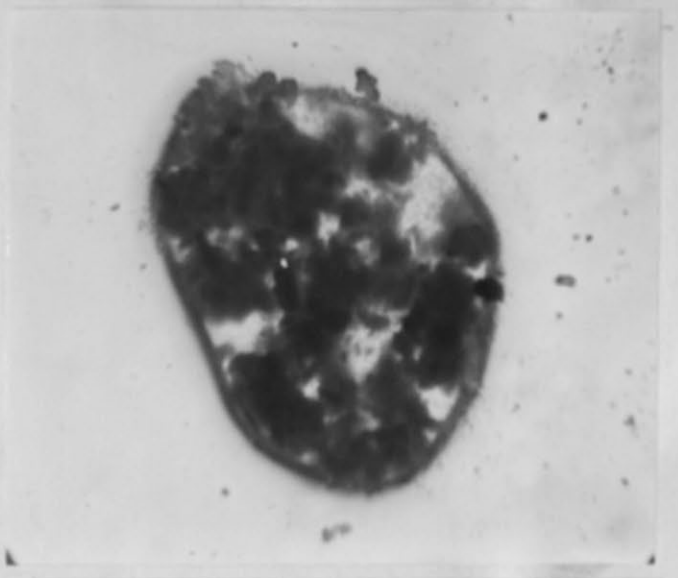


Fig. XXIV.- A Small Oxamide Concrement from A Rabbits Kidney. Crushed on a Slide to Show Component Crystals.

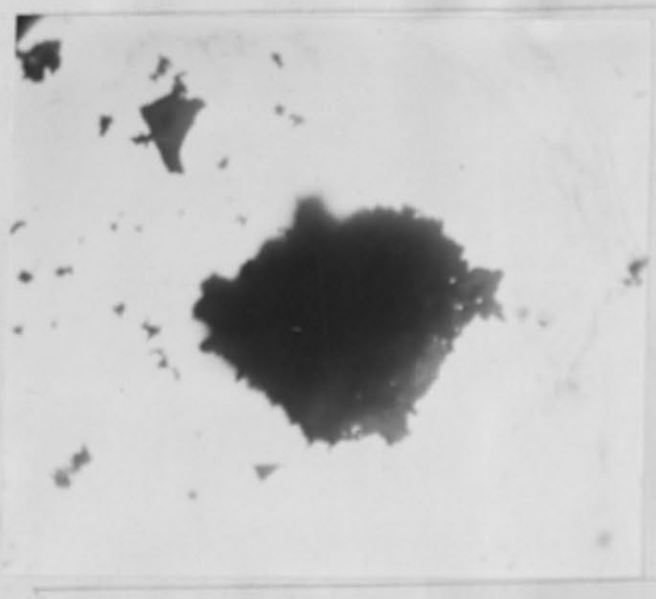


Fig. XXV.- A Small Oxamide Concrement. Formed in vitro by Repeatedly Precipitating Oxamide from Human Urine as Described in the Text.
This is a Dense Form and is Indistinguishable from Particles of Hard Crystalline Material Observed in the Kidneys and Voided Urine of Oxamide-Fed Animals.

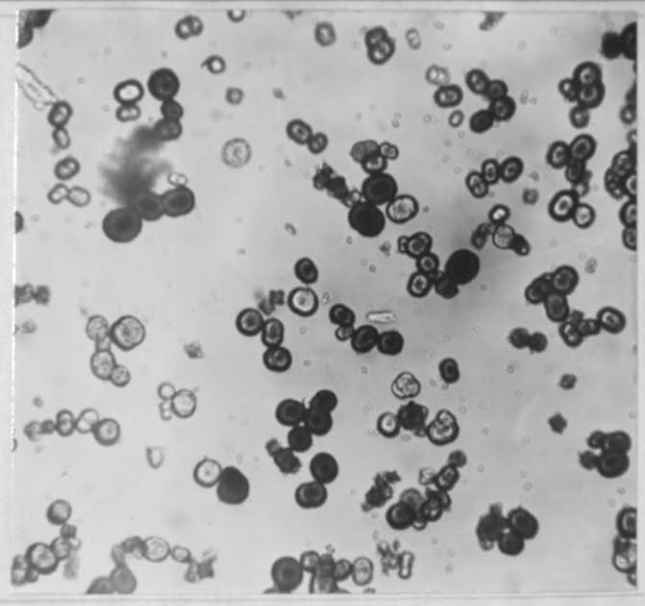


Fig. XXVI.- Oxamide Crystals Deposited in vitro from Urine in Which It was Dissolved by Boiling.

The Perfect Smooth Sphere is Shown Here. There is No Tendency to Fusion. Such Crystals and Such Behavior of this Type of Crystal with Regard to Fusion has been Observed Repeatedly in Oxamide -Fed Animals.

Fig. XXVII.- Fragments of Larger Oxamide Stone. Note the Concentric and Radial Striations.

This is a Physical Feature Characteristic of Most Stones Seen Clinically.

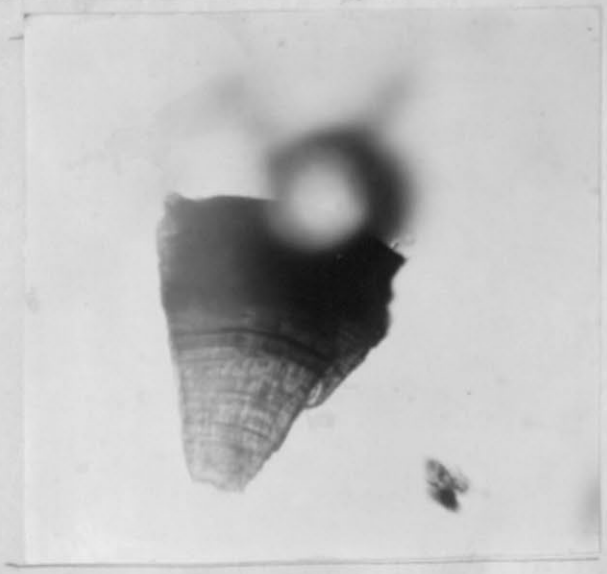


Fig. XXVIII.- Fragments of an Oxamide Stone. Showing Radial Markings and The Concentric Deposition in Layers.

BIBLIOGRAPHY

1. Abderhalden, E.:Handbuch der Biochemischen Arbeitsmethoden, Berlin u. Wien, Urban and Schwarzenburg, 1912, vol. v, 791.

2. Abderhalden, E. and Kankelheit, O.; Die Experimentelle Erzeugung von krystallinischen Infarkte in den Niere. Zeitschrift für Experimentelle Biochemie und Biophysik, xix, 191. Quoted from Chemical Abstracts, 1920, 561.

3. Adami, J.G.: The Principles of Pathology, Lea and Febiger, Phil., 1908,i, 864-868 and ii, 778.

4. Adams, Francis; The Genuine Works of Hippocrates, London, 1849, i, 200. The Seven Books of Paulus Aegineta, London, 1844, i, 548-552.

5. Adler, Alfred; Zur Aetiologie, Diagnostik, und Therapie der Nephrolithiasis. Wiener Klinische Wochenschrift, 1907, xx, No. 49, 1534-1539.

6. Austin, A.E.; The Uricolytic Enzyme in Animal Organs, Journal of Medical Research, 1906, xv, 314.

7. Baldwin, Helen;An Experimental Study of Oxaluria, with Special Reference to its Fermentative Origin. Journal of Experimental Medicine, 1900,v, 27-46.

8. Bechhold, H.: Colloids in Biology and Medicine. Bullowa's Translation. New York, Van Nostrand, 1919, 464 pp.

9. Bevan, A.D. and Smith, J.F.: The Diagnosis and Treatment of Kidney Stone. Surgery, Gynecology, and Obstetrics, 1908, vi, 675-691.

10. Blatherwick, N.R.: The Specific Role of Foods in Relation to the Composition of the Urine. Archives of Internal Medicine, 1914, xiv, 409-450.

11. Braasch, W. F.: Pyelography, Phil. Saunders, 1915, 323 pp.
Clinical Data of Nephrolithiasis. Surgery, Gynecology, and Obstetrics, 1917, xxiv, 8-14.
Lithiasis with Bilateral and Renal Involvement. Boston Medical and Surgical Journal, 1918, clxxviii, 292-298.

12. Braasch, W. F. and Moore, A.B.; Stones in the Ureter. Journal of the American Medical Association, 1915, lxv, 1234-1237.

13. Brown, T.R.; On the Relationship between the Variety of Microorganisms and the Composition of Stone in Calculous Pyelonephritis. Journal of the American Medical Association, 1901, xxxvi, 1395-1397.

14. Brugsch, Th.; Ueber Gicht, Gichtneire, und Uratsteindiathese. Münchener Medizinische Wochenschrift, 1908, lv Jahr., No. 47, 2463.

15. Bryan, R. C.; Stone in the Ureter, Surgery, Gynecology, and Obstetrics, 1911, xii, 134-151.

16. Bumpus, H. C., and Meisser, J.G.; Focal Infection and Selective Localization of Streptococci in Pyelonephritis. Study i. Archives of Internal Medicine, 1921, xxvii, 326.

17. Cabot, Hugh; Modern Urology. Philadelphia, Lea and Febiger, 1918,ii, 132-182. Stone in the Kidney and Ureter. Journal of the American Medical Association, 1915, lxv, 1233-1234.

18. Cabot, Hugh, and Crabtree, E. G.; Frequency of Recurrence of Stone in the Kidney after Operation. Surgery, Gynecology, and Obstetrics, 1915, xxi, 223-225.

19. Clark, G.W.; Effect of Hypodermic and Oral Administration of Calcium Salts on the Calcium Content of Rabbit's Blood. Journal of Biological Chemistry, 1920, xliii, No. i, 89-95.

20. Crenshaw, J. L.; Vesical Calculus. Journal of the American Medical Association, 1921, lxxvii, 1071-1075.

21. Cushing, Harvey; Observations upon the Origin of Gall Bladder Infections and upon the Experimental Formation of Gall Stones. Bulletin of the Johns Hopkins Hospital, 1899, x, 166-168.
Typhoidal Cholecystitis and Cholelithiasis. Discussion of a Possible Agglutinative Reaction in the Bile and its Relations to Stone Formation. Bulletin of the Johns Hopkins Hospital, 1898, ix, 91-95.

22. Cushny, A.R.; Textbook of Pharmacology and Therapeutics. 5th Ed. Phil., Lea and Febiger, 1913, pp. 571-577, 547-556, 560-561.
The Secretion of the Urine. London, Longmans, Green and Co., 1917, 241 pp.

23. Determeyer, D.; Die Losungsbedingungen der Harnsäure in Harn. Berliner Klinische Woohenschrift, 1907, 44 Jahrgang,No. 17, 544-545.

24. Ebstein, Wilhelm; Die Natur und Behandlung der Harnsteine, Wiesbaden, 1884. Quoted.

25. Ebstein, Wilhelm and Nicolaier, Arthur; Über die Experimentelle Erzeugung von Harnsteinen.
Wiesbaden, 1891. From Surgeon-General's Library.
Ueber die Ausscheidung der Harnsäure durch die Nieren. Archiv für pathologische Anatomie (Virchow), 1896, cxliii, 337-368.
Ueber die Wirkung der Oxalsäure und einiger ihrer Derivate auf die Nieren. Archiv für pathologische Anatomie (Virchow), 1897, cxlviii. 366.

26. Englisch, J. E.; Ueber Spontane Zertrümmerung der Harnsteine in der Blase. 1905, lxxvi, 962-1056.

27. Frankenthal, Ludwig; Ueber Cystinurie und Cystinstine. Deutsche Zeitschrift für Chirurgie, 1914, cxxxi, 442-465.

28. Folin, Otto and Denis, W.; The Diagnostic Value of Uric Acid Determinations in Blood. Archives of Internal Medicine, 1915, xvi, 33-37.

29. Fowler, H.A.; The Microscopic Structure of Urinary Calculi of Oxalate of Lime, with Special Reference to the Spiculated Variety. Johns Hopkins Hospital Reports, 1906, xiii, 507-556.
Cystinuria with Formation of Calculi. Report of a Case. Johns Hopkins Hospital Reports, 1906, xiii, 557-569.

30. Fowler, O.S.; Ureteral Obstruction Causing Urinary Stasis. A New Etiology in Kidney Stones etc. Journal of the American Medical Association, 1914, lxii, 367-372.

31. Gage, Homer and Beale, Howard W.; Fibrinous Calculi in the Kidney, Annals of Surgery, 1908, xlviii, 378-387.

32. Garrod, Archibald E.; The Croonian Lectures on Inborn Errors of Metabolism. Lancet, 1908, ii, 1-7,73-79,142-148.

33. Golding Bird; quoted by Morris.

34. Guiteras, Ramon; Urology. New York, Appleton, 1912,i, 499-524 and ii, 83-94.

35. Harris, M. L.; Renal Calculi, Journal of the American Medical Association, 1900, xxxiv, 643-647.

36. Hawk, Philip B.; Practical Physiological Chemistry. Phil. Blakiston, 1919. Heller's Table for Chemical Examination of Urinary Calculi, p. 506.

37. Harting: On the Artificial Production of Some of the Principal Organic Calcareous Formation. Quarterly Journal of Microscopical Science, 1872, xii, 118-119.

38. Hirsch, August; Handbook of Geographical and Historical Pathology. London, New Sydenham Society, 1886, vol. iii, 456-492.

39. Holt, L. Emmet; The Diseases of Infancy and Childhood. New York, Appleton, 1917, pp. 646-666.

40. Horbaczewski; quoted by Wells.

41. Homans, John; Gall Stones around Silk Sutures, Twenty Months after Recovery from Cholecystectomy. Annals of Surgery, 1897, xxvi, 114-116.

42. Hunner, Guy L.; The Etiology of Ureteral Calculus. Surgery, Gynecology, and Obstetrics, 1918, xxvii, 252-270.

43. Hutyra, Franz and Marek, Josef ; Special Pathology and Therapeutics of the Diseases of Domestic Animals. 2nd American Edt. Chicago, Eger, 1920, pp. 1100-1104.

44. Judd, E. S.; Foreign Bodies in the Urinary Bladder. Collected Papers of the Mayo Clinic, 1915, vii, 364-372.

45. Johnsen; Ein durch Operationsentfernter Riesen-Nierenstein. Berliner Klinische Wochenschrift. 1906, xliii Jahrgang, No. 51, 1623-1625.

46. Kahn, Max; Study of the Chemistry of Renal Calculi. Archives of Internal Medicine, 1913, xi, 92-99.

47. Kahn, Max and Rosenbloom, Jacob; A Report of Some New Chemical Analyses of Urinary Calculi with Indications for Treatment. Journal of the American Medical Association, 1912, lix, 2252-2253.

48. Kaufmann, Eduard; Specielle Pathologische Anatomie, Berlin, Reiner, 1919, 872-875.

49. Keen, W. W. and Da Costa, J. C.; Surgery, Its Principles and Practice., Phil., Saunders, 1910, vol. iv, 233-238 and 335-371.

50. Kelly, Howard A. and Burnam, Curtis F.; Diseases of the Kidneys, Ureters and Bladder. New York, Appleton, vol. ii, 90-186.

51. Keyes, Jr., E. L.; Diseases of the Genito-Urinary Organs. New York, Appleton, 1911, 406-448.
Some Problems Concerning Renal and Ureteral Stone. Medical Record, 1919, xcvi, 774.
Problems Concerning Urinary Calculi. American Journal of the Medical Sciences, 1921, clxi, No. 3, 334-339.

52. Kidd, Frank; Urinary Surgery. London, Longmans, 1910, 70-110 and 254-267.

53. Klotz, Oskar; Studies upon Calcareous Degeneration. Journal of Experimental Medicine. 1905, vii, 633-675, and ibid. 1906, viii, 322-336.

54. Kühn, Walter; Ueber der Kalkinfarkt der Nierenpyramiden. Archiv für pathologische Anatomie (Virchow), 1918, ccxxv, 44-59.

55. Kumita; Experimentelle Untersuchung über die nach der Anwesenheit von Steinen auftratenden Veranderungen im Harnapparte. Mitteilungen aus der Grenzgebiete der Medizin und Chirurgie. 1909, xx, 565-604.

56. Kümmel, Hermann; Der Steinkrankheit der Nieren und der Harnleiter, Deutsche Medizinische Wochenschrift, 1908, xxxiv, Jahrgang, No. 13, 537-542.

57. Küster, E.; Bemerkungen über Nieren und Harnleiter Stine mit Demonstrationen. Berliner Klinische Wochenschrift, 1894, no. 35, pp. 813.
Deutsche Chirurgie- Die Chirurgie der Nieren, der Harnleiter und der Nebennieren. Stuttgart, Enke, 1896-1902. Die Steinkrankheit der Nieren, Nephrolithiasis, Calculosis Renalis. Capital X, pp. 393-465. - This probably the best review to the date.

58. Küster, William; Beiträge zur Kenntniss der Gallengarbstoffe. Ueber die Bildung von Gallensteinen. viii and ix. Hoppe Seyler's Zeitschrift für Physiologische Chemi, 1915 xciv, 136-171.

59. Legeu, Felix; De L'Anurie Calculeuse. Annales des Maladies des Organes Genito-Urinaires. 1895, xiii, 865-910.

60. Lenander, K. G. ; Ueber zwei glücklich opirirte Fälle von Nierensteinen. Deutsche Medizinische Wochenschrift, 1897, 23 Jahrgang, No. 22, 337-340 and No. 23, 360-362.

4-21 6M

61. Lichtwitz,L.; Untersuchungen über Kolloide im Urin. Über Beziehungen der Kolloide zur Löslichkeit der Harnsäure und harnsauren Salze. Hoppe-Seyler's Zeitschrift für Physiologische Chemie, 1910, lxiv, 144-157.
Über die Bedeutung der Kolloide für die Konkrementbildung und die Verkalkung. Deutsche Medizinische Wochenschrift, 1910, 36 Jahrgang, No. 15, 704-706.
Die Bildung der Harnsedimente und Harnsteine. Zeitschrift für Urologie, 1913, vii, 810-820.

62. Lichtwitz,L. and Rosenbach, Otto; Untersuchungen über Kolloide im Urin. Über Kolloide im normalen menschlichen Urin. Hoppe-Seyler's Zeitschrift für Physiologische Chemie, 1909, lxi, 112-118.

63. MacCallum, W. G.; Textbook of Pathology, Phil. Saunders, 1919, pp. 428-433.

64. MacNeider, William de B; Renal Function and the Associated Disturbance in the Acid-Base Equilibrium of the Blood in Certain Experimentally and Naturally Acquired Nephropathies. Archives of Internal Medicine, 1920, xxvi, 1-37.

65. Marcet; quoted by Morris.

66. Mathews, A. P.; Physiological Chemistry, New York, Wood, 1919, 2nd Ed., 681-766.

67. Mayo, C. H.; Stone in the Kidney. Annals of Surgery, 1920, lxxi, 123-127.

68. Mayo, W. J.; The Removal of Stones from the Kidney. Surgery, Gynecology, and Obstetrics, 1917, xxiv, 1-8.
Hematogenous Infections of the Kidney. Journal of the American Medical Association, 1919, lxxiii, 1023-1026.

69. Mayo, W. J., Braasch, W. F., and MacCarty, W.C.; Relation of Anomalous Renal Blood Vessels to Hydronephrosis. Journal of the American Medical Association, 1909, lii, 1383-1388.

70. Meckel von Hemsbach; quoted by Küster and others.

71. Meyer, Willy; Catheterism of the Ureters in the Male with the Help of the Ureter Cystoscope- A report of Seven Cases. Medical Record, 1897, li, 617.

72. Meyers, Victor C., Fine, Morris, S., and Lough, Walter G.; The Significance of the Uric Acid, Urea, and Creatinin of the Blood in Nephritis. Archives of Internal Medicine, 1916, xvii, 570-583.

73. Mingramm; Beitragzur Frage der Steinbildung in der Harnwegen nach Wirbelbruchen. Deutsche Zeitschrift fur Chirurgie, 1909, xcviii, 89-93.

74. Minor, A. S., and Denis, W.; Effects of Feeding with Calcium Salts on the Calcium Content of the Blood. Journal of Biological Chemistry, 1920, xli, 357.

4-21 6M

75. Morawitz, P., and Adrian, C.; Zür Kenntnis der Sogenannte Eiweissteine der Niere und über die Ausscheidung Membranoser Massen aus dem Uropoetischen System. Mitteilungen aus den Grenzgebieten der Medizin und Chirurgie, 1907, xvii, 579-588.

76. Morgan, J. H.; TheLettsonian Lectures on the Affections of the Urinary Apparatus in Children. Renal and Vesical Calculi. Lancet, 1898, i, 556-560.

77. Morris, Henry; Surgical Diseases of the Kidney and Ureter, London, Cassell, 1901, ii - 43-171.

78. Müller, Kurt; Über die Nephrolithiasis nach Ruchenmarks Verletzungen. Archiv für Klinische Chirurgie, 1895, 1, 601-614.

79. Neuberger, Max; History of Medicine. Translation by Playfair, Ernest. London, Frowde, 1910, vol. i, pp. 159 and 266.

80. Neuman, A.; Zystinsteine und Zystinurie. Deutsche Medizinische Wochenschrift, 1914, ii, No. 50, 2065-2066

81. Nevarro, M.; Lithiase rénal expérimentale avec hydronéphrose. Bulletins de la Societé Anatomique de Paris, 1893, lxviiie Année, 658.

82. Newman, David; Lectures on the Surgical Diseases of the Kidney. London, Longmans, Green, 1888, Lect. iv, 168-178.

83. Nicolaier, Arthur und Dorn, Max; Ueber die Wirkung von Cholincarbonsauren und ihrer Derivate auf die Ausscheidung der Harnsaure. Deutsche Archiv fur Klinische Medizin, 1908, xciii, 331-355.

84. O'Conor, Vincent J.; Fibrinuria; Occurrence in a Case of Carcinoma of the Kidney. American Journal of the Medical Sciences, 1920, clix,729-742.

85. Ord, W. M.; Some Experiments Relating to the Forms Assumed by Uric Acid. St. Thomas Hospital Reports, 1870, i, 335-348.
An Account of Some Experiments Relating to the Influence Exercised by Colloids upon the Forms of Inorganic Matter. St. Thomas Hospital Reports, 1871, ii, 1-22.
On "Molecular Coalescence" and on the Influence Exercised by Colloids upon the Forms of Inorganic Matter. Quarterly Journal of Microscopic Science, 1872, xii, 219-239.

86. Ord, W. M., and Shattock, Samuel G.; On the Microscopic Structure of Urinary Calculi of Oxalate of Lime. Transactions of the Pathological Society of London, 1895, xlvi, 91-132.

87. Packard, F. R.; An Analysis of Two Hundred and Twenty One Cases of Foreign Body Introduced into the Male Bladder per Urethram, with Report of a Recent Case. Annals of Surgery, 1897, xxv, 568-599.

88. Poland; quoted by Morris.

89. Posner, C.; Die Bildung der Harnsteine. Zeitschrift für Urologie, 1913, vii, 799-809.

90. Rainey, G.; Quoted by Ord and Shattock and others.

91. Roberts, A. E. ; Vesical Calculus in India, its Distribution and a Theory of its Cause. Lancet, 1895, i, 381-382.

92. Robinson, Byron; Landmarks in the Ureter. Annals of Surgery, 1902, xxxvi, 850-898.

93. Rosenbach; Kritischer und Experimentelle Beitrag zur Frage der Entstehung der Nierensteine. Mitteilungen aus der Grenzbebieten der Medizin und Chirurgie, 1911, xxii, 630-656.

94. Rosenbloom, Jacob; A Case of Zanthin Calculus. New York Medical Journal, 1915, ci, 120.
A Further Study of the Chemical Composition of Urinary Calculi. Journal of the American Medical Association, 1915, lxv, 161-162.

95. Rosenow, E. C.; Bacteriology of Cholecystitis and its Production by Injection of Streptococci. Journal of the American Medical Association, 1914, lxiii, 1835-1836.
The Etiology of Cholecystitis and Gall Stones and their Production by the Intravenous Injection of Bacteria. Collected Papers of the Mayo Clinic, 1916, viii, 222-252.
Elective Localization and Focal Infection from Oral Sepsis. Canadian Journal of Medicine and Surgery, 1921, i, 34.
Focal Infection and Elective Localization of Bacteria in Appendicitis, Ulcer of the Stomach, Cholecystitis, and Pancreatitis. Surgery, Gynecology, and Obstetrics, 1921, xxxiii, 19-26.

96. Schade,H.; Zur Genese der Gallensteine. Zeitschrift für Experimentelle Pathologie und Therapie, 1910-1911, viii, 92-124.
Beiträge zur Konkrementbildung. i. Zur Entstehung der Harnsteine. Münchener Medizinische Wochenschrift, 1909, 56 Jahrgang, i, 307 and 77-80.

97. Shattock, Samuel G.; Iridescence in Calculi. Transactions of the Pathological Society of London, 1886, xxxvii, 325-339.

98. Shepelmann, Emil; Historisches zur Kenntnis der Entstehung von Harnsteinen. Berliner Klinische Wochenschrift, 1911, xlviii Jahrgang, 525-526.

99. Schenck, Benjamin R.; The Symptoms, Diagnosis, and Surgical Treatment of Ureteral Calculus. Johns Hopkins Hospital Reports, 1902, x, 477-513.

100. Seefisch, G.; Ein Beitrag zur Frage der Steinbildung in den oberen Harnwegen nach Verletzung der Wirbelsäule. Deutsche Zeitschrift für Chirurgie, 1908, xciv, 426-433.

101. Studensky, N.; Zur Lehre von Bildung der Harnsteine. Deutsche Zeitschrift für Chirurgie, 1877, vii, 171-186.

102. Tenney, Benjamin; Some Conclusions Based on A Study of One Hundred and Thirty Four Cases of Calculus in the Ureter with a Report of Three New Cases. Boston Medical and Surgical Journal, 1904, cl, 115-121.

103. Thomson, Joseph Oscar; Urinary Calculus at the Canton Hospital, Canton, China. Based upon Three Thousand Five Hundred Operations. Surgery, Gynecology, and Obstetrics, 1921, xxxii, 44-55.

104. Thomson Walker, J. W.; Genito-Urinary Surgery, New York, Funk and Wagnals, 1914, 249-283.

105. Thorn, John Mills; A Case of Recurrence of Calculus in a Paraplegic Subject-Suprapubic Lithotomy, Twice Performed. Guy's Hospital Reports, 1897, Published 1900, vol. liv, 183-189.

106. Tuffier, M.; Lithiase urinaire artificelle. Bulletins de la Societe Anatomique de Paris, 1893, lxviiie Annee, 49-52.

107. Ulzmann; quoted by Shepelmann.

108. Wadsworth, Augustus B.; Nephrolithiasis, Purpura Hemorrhagica, and Streptococcus Infection. Report of A Case without Febrile Reaction. Journal of the American Medical Association, 1918, lxxi, 2052-2053.

109. Watson, Francis S. and Cunningham, John H.; Diseases and Surgery of the Genito-Urinary System. Philadelphia, Lea and Febiger, 1908, pp. 168-211.

110. Wells, H. Gideon; Pathological Calcification. Journal of Medical Research, 1906, xiv, 491-525.
Chemical Pathology, Philadelphia, Saunders, 1918, 3rd. ed. pp. 93-94, 277-280, 311-312, 435-467, 568-569, 582-584, 618-635.

111. Wislocki, George B. and O'Conor, Vincent J.; Experimental Observations upon the Ureters with Especial Reference to Peristalsis and Antiperistalsis. Bulletin of the Johns Hopkins Hospital, 1920, xxxi, 197-203.

112. Young, Hugh Hampton; In "Modern Medicine", edited by Osler, Sir William and McCrae, Thomas. Philadelphia, Lea and Febiger, 1914, Chapter on Urinary Calculi, vol. iii, 1007-1025.

113. Zuckerkandl, O.: Über Nierensteine. Archiv für klinische Chirurgie, 1908, lxxxvii, 461-489.

Additional References

114. Adams, Francis; The Extant Works of Aretaeus, the Cappadocian. London, Sydenham Society, 1856, pp. 340-343.

115. Hart, E. B., Steenbock, H., and Hoppert, C. A.; Dietary Factors influencing Calcium Assimilation. The Comparative Influence of Green and Dried Plant Tissue, Cabbage, Orange Juice, and Cod Liver Oil on Calcium Assimilation. Journal of Biological Chemistry, 1921, xlviii, 33-50.

116. Haskins, Howard D.; The Uric Acid Solvent Power of Normal Urine. Journal of Biological Chemistry, 1916, xxvi, 205-215.

117. Moore; quoted by Kahn.

118. Rowlands; quoted by Kahn.

119. Mackerell; quoted by Kahn.

120. Osborne, Thos. B., Mendel, Lafayette B., and Perry, Edna L.; The Incidence of Phosphatic Urinary Calculi in Rats fed on Experimental Rations. Journal of the American Medical Association, 1917, lxix, 32-33.

121. Thompson; quoted by Watson and Cunningham.

122. Shohl, A. T. Changes in Acidity or Alkalinity of the Urine Produced by B. Coli as Measured by the Final Hydrogen Ion Concentration. Journal of Urology, 1920, iv, 371-379.

4-21 6M