INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY RESEARCH AND ANALYSIS

ISSN(print): 2643-9840, ISSN(online): 2643-9875

Volume 07 Issue 08 August 2024

DOI: 10.47191/ijmra/v7-i08-41, Impact Factor: 8.22

Page No. 3999-4006

The Role of Biochemistry in Understanding Kidney Stone Formation andUrinary Health

Athir Ihsan Shakir

Directorate of Salahaddin Education - Ministry of Education – Iraq

ABSTRACT: Renal illness caused by stones is the term for a solid substance concretion that usually develops inside the kidneys. A rising urological condition influences the well-being of people and affects about 12% of the world's population. An increased chance of ending stage of renal failure and they are closely related. There are several types of kidney stones. The CaC_2O_4 kidney calculi are the most common type. It appears at Randall plaque's renal papillary surfaces. (1)

Several physicochemical mechanisms, such as supersaturating, urinary stone nucleation, growth, aggregation, and retention components found in tubular cells, which aid in the complex process of formation of stones. An inequity between the elements that promote or inhibit the crystallization of urine influences these phases. Moreover, Scientists have noticed that renal injury of cells promotes particle retention reticular body. (2)

1. INTRODUCTION

1.1. An overview of Urinary tract stones

Stones in the urinary tract typically lodge in one kidney or more. The most prevalent type of kidney or bladder disease has afflicted people since 4000 B.C. and caused millennia of agony. Stopping the relapse of Kidney calculus is a serious health concern for people. A more profound understanding of the principles underlying the production of stones is essential to prevent the recurrence of stones. A higher chance of diabetes, vascular disease, end-stage renal failure, kidney stones and hypertension have been associated. Stones in the kidney have been suggested to be a systemic disease linked to metabolic syndrome. If nephrolithiasis is associated with nephrocalcinosis and represents 2–3% of patients with end-stage renal disease. (4)

2. THE URINARY TRACT AND RELATED STONES

The glomus shapes the pee filtrate, which at that point, goes towards the tubules where outflows or reabsorption changes its volume and creation. Whereas the proximal tub and gathering conduits handle miniature changes to pee structure, the proximal tubules handle most of the solute reabsorption. (5)

Pee that is 95% H2O, 2.5% urea, and 2.5% a blend of minerals, salts, chemicals, and proteins is concentrated by implies of the circle of Henle. Within the proximal tubules, basic supplements like proteins, amino acids, bicarbonate, calcium, phosphate, and potassium are reabsorbed and returned to the circulatory framework alongside glucose, cl, and H2O. Blood's salt and the destructive base agreement are controlled within the proximal tubule. Stone zones can differentiate, as shown. (6)

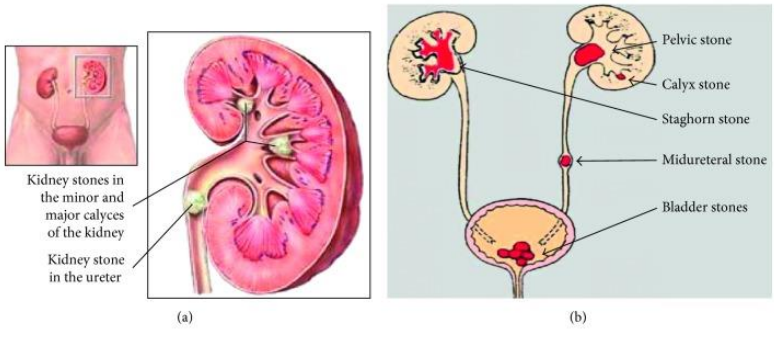

Fig (1): Urinary tract stones in the urinary system

3. TYPES OF KIDNEY STONES

3.1. Calcium stones: Oxalate and Phosphate

The awkward nature of the distinctive chemical composition of pee decides the chemical composition of kidney stones. The measure, frame, and chemical cosmetics (mineralogy) of stones change. Agreeing to varieties in mineral substance and etiology, kidney stones are regularly categorized into five categories as follows. Roughly eighty percent of all calculi within the pee are calcium stones, which are the foremost common sort of renal stones. Calcium phosphate (CaP, moreover known as apatite) (five percent), unadulterated oxalate of calcium (CaOx) (fifty percent), and a combination of both (45%) could be included within the rate of calcium stones. Brushite, also known as hydroxyapatite or calcium hydrogen phosphate, is the essential component of calcium stones. [7]

Most kidney stones contain calcium oxalate within the structure of CaOx monohydrate (COM, too known as weddellite, $CaC_2O_4 \cdot H_2O$) or Calcium Oxalate dihydrate (COD, moreover known as weddellite, $CaC_2O_4 \cdot 2H_2O$), or both together, which makes up more than sixty percentage of kidney stones. The foremost thermally steady sort of stone is called COM. In clinical stones, COM is seen more frequently than COD. [8]

3.2. Mg NH4 and $Po4^{3-}$ stones

Triple phosphate stones and malady stones are other names for struvite stones, which influence 10-15% of people. It happens to individuals who have constant urease-producing urinary tract contaminations; Proteus mirabilis is the first overwhelming pathogen, but Pseudomonas aeruginosa, Enterobacter, and Klebsiella pneumonia are less common afflictions. In organizing to portion or cleave urea into noticing salts and CO_2, urease is required. This increases urine's stomach settling agent pH (more frequently than not more noticeable than 7). Since phosphate is less dissolvable in stomach settling agent pH than in acidic pH, it quickens onto the insoluble NH things, coming around inside the creation of a sizable staghorn stone. Compared to men, women are more likely to set up somewhat stone. E. coli is not associated with struvite stones and is incapable of breaking urea. [9]

3.3. Stones that consist of uric acid

Almost three to ten percent of the full stone sorts are spoken to by this. Purine-rich diets, especially those that incorporate creature protein sources like meat, cause hyperuricosuria, moo sum of pee, and a moo pee pH (pH<5.05), which exasperates the advancement of uric corrosive stones. People who have gouty joint pain may have kidney stones. Uric corrosive kidney stones are more visited in guys than in ladies, and idiopathic nephrolithiasis is the foremost common reason for the situation. [9]

3.4. Cystine stones

Fair over two percent of all stone classifications are these stones. It could be a condition influencing the exchange of cystine and amino acids. Due to a transformation within the rBAT qualities on chromosome 2, it causes a tall level of cystinuria in pee excretions, an autosomal passive condition that impedes renal tubular admissions of cystine or spills cystine into pee. [10]

It causes cystine stones to create and cannot break down in pee. People who are homozygous for cystinuria excrete about six hundred millimoles of insoluble cystine each day. As it were a clinical sign of this cystine stone sickness is the arrangement of pee cystine. [12]

3.5. Stones that are induced by medications

This kind spoke to around one percent of the sorts of urinary tract stones. These stones are brought on by medicines counting Guaifenesin, Triamterene, Atazanavir, or Sulfate solutions. For illustration, kidney stones can happen in patients utilizing the protease protein inhibitor indinavir sulfate, which is endorsed to treat HIV disease. Such lithogenic solutions or their metabolites may settle on pre-existing renal calculi or deliver a nidus. Alternatively, these medicines may cause the generation of calculi by disturbing the digestion system of purines or CaC_2O_3. [13]

4. THE STRUCTURE OF RENAL STONES

Urinary stones can have two different chemical compositions: the nonchemical substance (the structure) and the mineral and uncrystallised stages. The natural base of urinary calculi is made up of macromolecules including lipids, proteins, carbohydrates, and glycosaminoglycans (GAGs), which can either promote or inhibit the processes leading to kidney calculus growing (Table 1). The primary ingredients of the stones structure are protein chains (64%), carbohydrates without an amino group (9.6%), carbohydrates with an amine group just like glucosamine (5%), liquids (9.99%), as well as non-organic cinereal (10.5%). This system works like an example for the renal stone construction, and the substrate of all calculi includes phosphate lipid (8.6%) of the whole lipid, that in hand makes up roughly 10.3% of all stones. [14] Phospholipids in cell membranes, which are a component of the natural structure, facilitate development of $CaPO_4^{-3}$ and CaC_2O_4 crystals. The primary ingredient in the structure of all forms of stones is albumin. [14]

Label of polypeptide	Part in distillation			
	Nuclei formation	**Growing**	**Accumulation**	**Cell keeping**
Calcium-binding protein	-	-	-	=
UMOD	+	=	-/+	=
Bone/sialoprotein I	-	-	-	-/+
Plasma protein	+	=	-	=
uF1	-	-	-	=
Alpha1microglobulin	=	=	-	=
S100Alpha	=	-	-	=
Interalphainhibitor	-	-	-	-
Ulinastatin	-	-	-	-
Reg protein	=	-	=	=
Crypt dins	non	pro	pro	=
PCYT1B	non	in	non	=
Myelo-peroxidase	=	+	+	=
Nucleolar protein	non	non	non	+
HistonelysineNmethyltransferase	=	-	-	=
K IR	non	in	in	=
PolypeptideWnt2	=	-	-	=
Fetuin-A	pro	in	non	=
Calcineurin A	non	non	non	-
Hyaluronate	non	non	non	+
CDS	non	in	in	=
Polysaccharide H S	non	in	non	=
TFF-3	non	in	non	=
CCL2	non	non	non	+
A2	non	non	non	+
CD-44	=	=	=	+
Gamma-carboxyglutamic acid	=	-	=	-
H 1 B	non	pro	non	non
Clg	non	non	in	in
Fibrous protein	pro	non	non	non
Glycosaminoglycan	-	-	-	-
Citric acid	non	in	non	non
Pyro-phosphate	=	-	=	=
Mg^{+2}	=	-	=	=

(- refers to inhibition, + refers to promotion, = refers to non-effectiveness)

5. CRYSTALS FORMATION

The initial stage of kidney stone development involves the nucleus, also known as nidus, forming from supersaturated urine that is held within the kidneys. Total particles, ions, and atoms inside an overwhelmed fluid begin to form tiny clusters, which separate once the entire free power within the group is lower compared to that of the fluid. [13]

Calcium oxalate crystals, for instance, are created when charged soluble molecules like calcium as well as oxalate interact to become insoluble. Either a fixed particle mechanism or a free particle mechanism can generate nucleation in the kidney. When promoters outnumber inhibitors in supersaturated fluids, nucleation begins. [13]

Crystallization can happen at a smaller biochemical stress than what is needed to generate the first nucleus if one has been formed as well as if it has been attached. Within the procedure of nucleus generation known as heterogeneous nucleation, epithelial cells that are already present, urine casts, red blood cells, and various crystals in urine can all function as nucleating centers. Through promoting varying nucleation and crystal accumulation, the organic structure, mucopolysaccharide, functions as an agent for binding. However, it is asserted that microorganisms create apatite structures that operate as a crystallization hub during the

creation of stones. The entire process amplifies stone formation. Current study focuses on the function of microorganisms that deconstruct $C_2O_4^{-2}$, like Oxalo-bacter formi-genes, in the development of CaC2O4 stones. [14]

Laboratory diagnosis

Laboratory diagnosis includes stone analysis, imaging studies, blood profiles, and a urine metabolic evaluation. Stone analysis plays a valuable role in the diagnosis of kidney stone patients, specifically in infrequently encountered kidney stones such as UA, cystine, infection-induced, drug-induced, and NH4+ urate stones. Imaging studies are valuable in the diagnosis of kidney stone disease. Despite numerous imaging methodologies, computed tomography is the most sensitive and specific mode of diagnosis (103). High fasting blood calcium, low phosphorus, and elevated PTH are suggestive of primary hyperparathyroidism. In that case, the patient must be considered for a noninvasive localization study followed by parathyroidectomy.

Normal serum calcium, low serum phosphorus, elevated 1,25(OH)2D, and normal PTH are suggestive of renal phosphorus leak. The finding of low serum potassium and low CO2 is suggestive of dRTA. Hyperuricemia and high serum triglycerides are encountered in patients with UA stones

Practical section

In this section, we presented several cases to describe examinations needed for ensuring the diagnosis of kidney stones. We examined the presence / absence of several parameters which are related with kidney stones. We conducted these examinations in two parts, for the first part of the examination we chose a sample consists of 16 volunteers from both genders and different age groups. Those participants had previously suffered from symptoms indicates to the probability of kidney stones existence. We applied different chemical & microscopic tests to the participants which are needed besides clinical symptoms for ensuring the presence of renal stones. In addition, for the second experiment we applied the same tests on one patient who had been diagnosed with kidney stones. We used the second part of the experiment to compare test values for the 16 participants with values related with kidney stones.

MATERIALS AND METHODS

Spectrophotometer or colorimeter measuring at 500 nm.
Matched cuvettes 1.0 cm. light path.
Using other general laboratory equipments.
Test procedure for quantitative determination of uric acid:
1_ assay conditions
Wavelength (500 nm.)
Cuvette (1 cm. light path)
Temperature (37 ◦C)
2_ adjust the instrument to zero blank of reagent.
4_ mix and incubate for 5 minutes at 37◦C or 10 minutes at room temperature (15-25◦C).
5_ read the absorbance of the samples and calibrator, against the blank. The color is stable at least 5 minutes.

Calculation:

Sample – Blank / Standard – Blank * 6 (Std. conc.) = mg/dL uric acid in sample
Procedure for Creatinine determination:
1_ Bring reagents and samples to room temperature.
2_ Set the photometer to 0 absorbance with working reagent.
3_ Pipette into labeled tubes
4_ Mix gently at room temperature and start stopwatch.
5_ Record absorbance at 500 nm after 30 second (A1) and after 2 min. (A2) of the sample or standard addition.
Standard concentration = 2 mg/dl
Concentration (sample) = A2- A1 (sample) / A2-A1
Standard * Concentration.

RESULTS & DISCUSSION

For the first part of tests the results were recorded for chemical and microscopic tests on the 16 participants. The observations we noted due to tests for those volunteers, many have high concentrations of creatinine in their blood, which might be a sign of kidney stones. In addition, in urine tests for some participants, we found a positive presence of epithelial cells which is a sign of

inflammation (kidney stones could cause inflammation because of the injury in urinary tissues). Another clinical significance in others is that there is a presence of amorphous crystals in urine, which leads to high concentrations of uric acid in urine. (Positive sign for kidney stones case). For the rest, we noticed in the urine test that the range of RBCs and Pus is abnormal, which indicates an injury in renal tissues causing inflammation and the injury is from a possible presence of kidney stones. In the second part of the experiment we applied chemical and microscopic tests on blood and urine samples on a 27 years old young lady who already had the condition. These samples were taken from a pregnant female who had diabetic type2 and suffered from severe pain attacks frequently. She was diagnosed with kidney stones via ultrasound. The tests were applied in the early morning for two following days. She was diagnosed with (Hyperuricemia) after conducting the necessary tests on the patient's blood and urine samples. Accordingly, the blood serum test was repeated for two consecutive days to confirm the amount of uric acid, and it was performed in the morning. It was noted on the first day that the amount of uric acid was 9. The next day, the test was repeated under the same conditions and it was found that the amount of uric acid was 8.7. We were able to detect the existence of kidney stones in many participants by comparing with reference values and illness parameters which were emerged in the second part of the examination. The results for both sections of experiment are down below.

Patient	**Chemical test (serum samples)**	**Microscopic test (urine samples)**
Female – 34 years old	Uric acid → 7.3 Creatinine →3 high	RBC → 5-9 Pus → 9-13 high Epithelial cells →++++ Amorphous →+++
Male – 24 years old	Uric acid →5 Creatinine → 1.4	RBC → 3-6 Pus → 4-6 Epithelial cells → ++ Amorphous → ++
Male – 29 years old	Uric acid → 9.3 high Creatinine → 4 high	RBC → 7-9 high Pus → 10-12 high Amorphous → +++
Male – 43 years old	Uric acid → 7.1 Creatinine →3	RBC → 3-5 Pus → 4-6 Epithelial cells → ++ Amorphous →+++
Male – 32 years old	Uric acid → 5 Creatinine → 1.3	RBC → 3-5 Pus → 2-4 Epithelial cells → ++++ Amorphous → ++
Female – 38 years old	Uric acid → 6 Creatinine → 4.3 high	RBC → 9-15 high Pus → 5-6 Epithelial cells → +++ Amorphous → +++
Male – 26 years old	Uric acid → 5 Creatinine → 2	RBC → 3-5 Pus → 6-9 Amorphous → ++
Female – 34 years old	Uric acid → 5.1 Creatinine → 2	RBC → 9-13 high Pus→ 6-8 Epithelial cells → ++++ Amorphous →++
Female – 29 years old	Uric acid → 6.5 Creatinine→ 1.5	RBC → 5-10 high Pus →9-15 high Epithelial cells → +++ Amorphous → ++

Male – 54 years old	Uric acid → 8 high Creatinine → 3 high	RBC → 5-10 high Pus →13-15 high Epithelial cells → +++ Amorphous →+++
Female – 39 years old	Uric acid →5 Creatinine→ 1.5	RBC → 3-5 Pus→ 4-6 Epithelial cells →++
Female – 53 years old	Uric acid 7.3 Creatinine 4 high	RBC 7-10 high Pus 6-13 high Epithelial cells +++ Amorphous ++
Male – 19 years old	Uric acid →6.4 Creatinine →1.3	RBC →5-9 high Pus → 4-8 Epithelial cells → +++ Amorphous → +
Female – 40 years old	Creatinine → 3 high Uric acid → 5.1	RBC → 3-5 Pus → 15-20 high Epithelial cells → ++ Amorphous → +++
Male – 33 years old	Creatinine → 5 high Uric acid → 4.6	RBC → 5-10 high Pus → 9-13 high Amorphous → +++ Mucus → +
Female – 22 years old	Creatinine → 8 high Uric acid → 5.8	RBC → 5-9 high Pus → 4-6 Epithelial cells → +++ Amorphous → ++ Mucus → +++

The results as shown in the schedule for a young woman compared to reference values

Measured concentration of uric acid in blood serum	**Reference values for uric acid**
Day 1: 9 mg/dl High Day 2: 8.7 mg/dl High	Serum or plasma Women: 2.5 – 6.8 mg/dl Men: 3.6 – 7.7 mg/dl Urine 250 – 750 mg/24h
Microscopic test for a urine sample	Reference values
Day 1: Epithelial cells: 8-13 RBCs: 6-8 High Pus: 20-25 High Amorphous: + Mucus: few	RBC in urine: 4 cells/HPF Pus in urine: Male: 4 cells/HPF Female: 5-7 cells/HPF

Day 2: Epithelial cells: 6-10 RBCs: 4-7 Pus: 15-20 High Amorphous: +++ Mucus: ++	

Reference values:

Reference (normal) values
Creatinine in serum: Men (0.6-1.1) Mg/dl Women (0.5-0.9) Mg/dl
Uric acid in serum: Women: 2.5 – 6.8 mg/dl Men: 3.6 – 7.7 mg/dl
RBC in urine: 4 cells/HPF
Pus in urine: Male: 4 cells/HPF Female: 5-7 cells/HPF

CONCLUSIONS:

In conclusion, we found a graded association between episodes of kidney stones and the risk of adverse renal outcomes. Further research should be aimed at determining the mechanisms explaining this association and assessing the optimal way to prevent kidney stones in the general population, especially young women.

REFERENCES

1) Giannossi L., Summa V. A review of pathological biomineral analysis techniques and classification schemes. In: Aydinalp C.,editor. *An Introduction to the Study of Mineralogy.* InTech, IMAA-CNR, Italy: InTechOpen; 2012. http://www.intechopen.com/books/ [Google Scholar]
2) Lopez M., Hoppe B. History, epidemiology and regional diversities of urolithiasis. *Pediatric Nephrology.* 2008;25(1):49–59. doi: 10.1007/s00467-008-0960-5. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
3) Mikawlrawng K., Kumar S., Vandana R. Current scenario of urolithiasis and the use of medicinal plants as antiurolithiatic agents in Manipur (North East India): a review. *International Journal of Herbal Medicine.* 2014;2(1):1–12. [Google Scholar]
4) Khan S. R., Pearle M. S., Robertson W. G., et al. Kidney stones. *Nature Reviews Disease Primers.* 2016;2:p. 16008. doi: 10.1038/nrdp.2016.8. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
5) Sigurjonsdottir V. K., L.Runolfsdottir H., Indridason O. S., et al. Impact of nephrolithiasis on kidney function. *BMC Nephrology.* 2015;16(1):p. 149. doi: 10.1186/s12882-015-0126-1. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
6) El-Zoghby Z. M., Lieske J. C., Foley R. N., et al. Urolithiasis and the risk of ESRD. *Clinical Journal of the American Society of Nephrology.* 2012;7(9):1409–1415. doi: 10.2215/cjn.03210312. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
7) Rule A. D., Roger V. L., Melton L. J., et al. Kidney stones associate with increased risk for myocardial infarction. *Journal of the American Society of Nephrology.* 2010;21(10):1641–1644. doi: 10.1681/asn.2010030253. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
8) Worcester E. M., Coe F. L. Nephrolithiasis. *Primary Care.* 2008;35(2):369–391. doi: 10.1016/j.pop.2008.01.005. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
9) Taylor E. N., Stampfer M. J., Curhan G. C. Obesity, weight gain and the risk of kidney stones. *Journal of the American Medical Association.* 2005;293(4):455–462. doi: 10.1001/jama.293.4.455. [PubMed] [CrossRef] [Google Scholar]
10) Courbebaisse M., Prot-Bertoye C., Bertocchio J., et al. Nephrolithiasis of adult: from mechanisms to preventive medical treatment. *Revue Medicale Internationale.* 2017;38(1):44–52. doi: 10.1016/j.revmed.2016.05.013. [PubMed] [CrossRef] [Google Scholar]

11) Kumar S. B. N., Kumar K. G., Srinivasa V., Bilal S. A review on urolithiasis. *International Journal of Universal Pharmacy and Life Sciences.* 2012;2(2):269–280. [Google Scholar]
12) Teichman J. M., Joel M. H. Acute renal colic from ureteral calculus. *New England Journal of Medicine.* 2004;350(7):684–693. doi: 10.1056/nejmcp030813. [PubMed] [CrossRef] [Google Scholar]
13) Knoll T. Epidemiology, pathogenesis and pathophysiology of urolithiasis. *European Urology Supplements.* 2010;9(12):802–806. doi: 10.1016/j.eursup.2010.11.006. [CrossRef] [Google Scholar]
14) Chauhan C. K., Joshi M. J., Vaidya A. D. B. Growth inhibition of struvite crystals in the presence of herbal extract *Commiphora wightii*. *Journal of Materials Science.* 2008;20(1):85–92. doi: 10.1007/s10856-008-3489-z. [PubMed] [CrossRef] [Google Scholar]
15) Moe O. W. Kidney stones: pathophysiology and medical management. *The Lancet.* 2006;367(9507):333–344. doi: 10.1016/s0140-6736(06)68071-9. [PubMed] [CrossRef] [Google Scholar]
16) Romero V., Akpinar H., Assimos D. G. Kidney stones: a global picture of prevalence, incidence, and associated risk factors. *Reviews in Urology.* 2010;12(2-3):e86–e96. [PMC free article] [PubMed] [Google Scholar]
17) Edvardsson V. O., Indridason O. S., Haraldsson G., Kjartansson O., Palsson R. Temporal trends in the incidence of kidney stone disease. *Kidney International.* 2013;83(1):146–152. doi: 10.1038/ki.2012.320. [PubMed] [CrossRef] [Google Scholar]
18) Afsar B., Kiremit M. C., Sag A. A., et al. The role of sodium intake in nephrolithiasis: epidemiology, pathogenesis, and future directions. *European Journal of Internal Medicine.* 2016;35:16–19. doi: 10.1016/j.ejim.2016.07.001. [PubMed] [CrossRef] [Google Scholar]
19) Robertson W. G., Heyburn P. J., Peacock M., Hanes F. A., Swaminathan R. The effect of high animal protein intake on the risk of calcium stone-formation in the urinary tract. *Clinical Science.* 1979;57(3):285–288. doi: 10.1042/cs0570285. [PubMed] [CrossRef] [Google Scholar]
20) Singh K. B., Sailo S. Understanding epidemiology and etiologic factors of urolithiasis: an overview. *Scientific Visualization.* 2013;13(4):169–174. [Google Scholar]