Prevalence of Microscopic Calcium Oxalate crystalluria in Biochemically Normal Urine Samples

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Abstract

Background: Calcium oxalate is the major cause of human kidney stones in all over the globe. These are colorless, refractile, and envelope-shaped. Sometimes dumbbell-shaped or peanut-like forms are seen. They are soluble in dilute Acids. Consumption of some types of foods like tomatoes, spinach, cabbage, asparagus, etc. causes upsurge in its number. A large number are seen in ethylene glycol poisoning.

Method : This present research was aimed at investigating the prevalence of calcium oxalate crystalluria in biochemically normal urine samples came to Clinical pathology section of Central Diagnostic Laboratory, Dhiraj General Hospital for the period of two months from 1st December 2019 to 31st January 2020. The urine samples were centrifuged at 3000 rpm for 15 min. The supernatant were decanted and the sediments were viewed under microscope using high power objective lens with 40x magnification. Total 400 urine samples with normal urine protein and normal urine sugar were analyzed microscopically using an Olympus microscope 4000x. Results showed that calcium oxalate crystals had the prevalence rate of 13.26 %.

Conclusion: The study showed that this much of prevalence of COC was associated with food intake and the social life style.

Keywords: *Calcium oxalate crystalluria, Microscopy, Prevalence, Biochemical.*

Introduction

Calcium oxalate crystal is a chemical compound that forms an envelope shaped crystal, known in plants as raphides; a major constituent of human kidney stones.[1] Majority of the renal stones are the calcium stones, usually in form of calcium oxalate. This oxalate is a naturally occurring substance found in routine diet, includs some fruit , vegetable, nuts and chocolates. Human liver also produces oxalate, dietary factors, high doses of Vitamin D and several metabolic disorders can increase the amount of calcium oxalate in the urine.[2] Calcium oxalate crystals (COC) are the most frequently observe crystals in urine, and 75 % of renal calculi have calcium oxalate crystals as component. Calcium oxalate crystal can form at any pH, and have various microscopic morphologies. It is estimated that about half of the oxalate in urine comes from ascorbic acid (Vitamin C) which is a precursor to calcium oxalate. COC are also associated with ethylene glycol ingestion; another oxalate precursor is calcium carbonate (CaCO3), the chief element in egg shells.[3]The most common metabolic disorder that causes kidney stone in children is hypercalciuria, which causes extra calcium to collect in urine. Other more rare metabolic conditions involve breaking down of oxalate, a substance made in the body found in some foods.[4] These condition include hyperoxaluria, too much oxalate in urine oxalis characterized by deposits of oxalate and calcium in the body tissues.

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Calcium oxalate is found in rhubarb leaves in large quantity and in species of oxalis, Areacaceae taro, kiwifruits, tea leaves, agaves, alocasia and spinach in varying amount while insoluble calcium oxalate crystals are found in plant stem, root and leaves. Vegetable like okra, parsley, leeks, Swiss chard, collard and quinoa are among the most oxalate-dense vegetables.[5] Calcium oxalate is known to be a major constituent of human kidney stone. Even a small dose of calcium oxalate is enough to cause intense sensations of burning in the mouth and throat, swelling, and choking that could last for up to two weeks, and in greater doses it can cause severe digestive upset, breathing difficulties, coma or even death.[6] Urinalysis was the first laboratory test performed in medicine and has been used for quite a few years. Urinalysis continues to be a powerful tool in obtaining crucial information for diagnostic purposes in medicine.[7] Urine analysis is a test that evaluate a sample of urine, it is use to detect and assess a wide range of disorder, such as UTI, renal disease and diabetes. It involves a number of tests to detect and measure various compound that passes through the urine.[8] For example bilirubin pigment in bile may indicate liver diseases. Hemoglobin; iron-containing pigment in red blood cells may indicate injury of the urinary tract, anemia or infection. Cells from the lining of the urinary tract (epithelial cells), crystals may indicate metabolic disease.[9]

These are colorless, refractile, and envelope-shaped. Sometimes dumbbell-shaped or peanut-like forms are seen. [10]

Dihydrates may appear at pH 6 or in neutral urine. Their classic form is that of a small, colorless octahedron that resembles an envelope. Dumbbell shapes and ovoid forms may occur. Longer forms occur in calcium oxalate monohydrate. COCs are insoluble in acetic acid. Oxalate crystals in large numbers may reflect severe chronic kidney disease or ethylene glycol or methoxyflurane toxicity. Oxaluria has come into eminence as a reflection of the increased absorption of oxalates from food following small bowel disease and resection, notably for Crohn's disease. Oxaluria may also be present in genetically susceptible persons following large doses of ascorbic acid. [11]

This present study was carried out in order to determine the prevalence of microscopic calcium oxalate crystals in the urine with normal biochemical parameters at Dhiraj General Hospital.

Material and Method

This was retrospective cross sectional study, conducted to get the prevalence of calcium oxalate crystalluria in biochemically normal urine samples came to Clinical pathology section of Central Diagnostic Laboratory, Dhiraj General Hospital (Sumandeep University) for the period of two months from 1st December 2019 to 31st January 2020.

The material used were Microscope, centrifuge, urine sample bottles, glass slides, dropping pipette, ethanol, water bath, etc. All glass wares were sterilized prior to use and after use.

Urine has a long rich history as a source for measuring health, well-being and remains an important tool for clinical diagnosis. The clinical information obtained from a urine specimen is influenced by the collection method, timing and handling. A vast assortment of collection and transport containers for urine specimens are available. Determining which urine collection method should be used depending on the type of laboratory test ordered. Randomly selected specimens were used for this research since it is the easiest to obtain and it is readily available.

10 mL of the urine sample collected were centrifuged at 3000 rpm for 15 min. The supernatant were decanted and the sediments were viewed under microscope using highest power objective lens 400x. The number of calcium oxalate crystals and other microscopic parameters were reported .[12]

Photographic shots were made to portrayed. Raw data obtained were expressed as mean ± SD and %. Significance differences between means were compared using software. Statically significance was achieved at p value of ≤ 0.05.

Results And Discussion

Urine is sometimes supersaturated with calcium oxalate crystals (COC), which means that it contains oxalate crystals that formed spontaneously. COC exist

in monohydrate and dehydrate forms which can be differentiated by the shape of their respective crystals. The shape of COC is octahedral, and they can grow at any pH naturally in normal urine. Calcium oxalate monohydrate crystals vary in shape such as prisms, dumbbells, spindles, ovals or picket fences.[6] In this present study, the shapes of these crystals are mainly octahedral (Figure 1 and 2 (10x and 40x). COC is known to be major components of human kidney stones. [1]

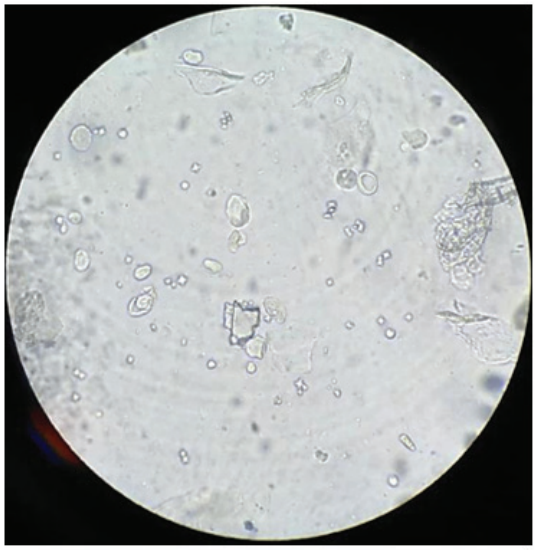

FIGURE:1: COC seen under low power. 10x

FIGURE:2: COC seen under high power. 40x

The presence of these crystals in urine were seen in 53 samples out of 400 cases studied. The distribution of these crystals in urine samples investigated was age dependent as adult patients had the highest percentage prevalence rate. It is possible that the social and food habits of the students must have contributed greatly to this anomaly. In their diet history, majority of them were vegetarian by their diet (89.9%).

In this study, the distribution of octahedral shape COC in urine may leads to blockage of the urinary tracts. This can lead to pain; most commonly beginning in the flank, groin or lower back. This pain is often known as renal colic and typically comes in waves lasting 20-60min. [13] Crystals found in urine may indicate an abnormality, however they may also be present in normal urine as an artifact .[14] There are few relatively health condition that require strict oxalate restriction. These conditions include absorptive hypercalciuria type II, enteric hypercalcuria, and primary hyperoxaluria, Dietary oxalates are usually restricted to 50 mg per day under these circumstances.[15] The frequency of these crystals and other microscopic parameters in the urine samples of these students calls for urgent medical attention. For example, pus cells in urine signifies that the body is fighting an infection in the lower or upper urinary tract, or it contains dead skin cells, bacteria or white blood cells. Pyuria for instance, mostly exist as result of urinary tract infection .[16]

Apart from COC; Triple phosphate, Urate, Billirubin crystals were also found while studying the data. The presence of intact red blood cell (RBC) in urine usually signifies source of blood loss in the lower part of the urinary tract such as urethra, bladder and ureter because gross haematuria may be related to trauma of the bladder, urethra, tumors or hemorrhage.[17] Patients with this in their urine may likely experience this disease if not checked properly. Epithelial cells in the urine can deliver some information of diagnostic use based on the type and the numbers of the cells.[2,17] Transition epithelial cells which lines the urethra and bladder are also commonly seen under microscopy.

Conclusion

This study showed that most of the urine samples collected randomly from patients with normal urine sugar and normal urine protein, has significant level of COC. However, after this study, it is recommended that persons with high level of COC in their urine should reduce the rate of consumption of oxalate rich foods such as spinach and they should take much fluid (water) each

day as this will help to dissolve the crystals in urines.

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Ethical Clearance: There were no ethical issues involved as the study was analytical and did not violate any ethical principles. Also this article has used only urine samples received in Laboratory and does not comprise any interventional studies on animal or human.

References

1. Francesco, V.R. Calcium oxalate in plants, formation and functions. Annual Review in Plant Biology, 2015, 56: 41-71.
2. Mayo, F.K. Foundation for Medical Education and Research. Mayo Family Health. 2015, 4:56.
3. Kevin, F. Urine calcium laboratory measurement and clinical utility. American Society for Clinical Pathology, 2010, 41 (11): 683- 686.
4. Michael, L. The truth about oxalate. Answers to frequently ask questions. Medicine Online. www.mediconline.com, 2012.
5. Assimos, A.G. Bilirubin reductase a major bilirubin excretion. Science, 2015, 208 (440): 145-151.
6. Martin, G., Matteo, G., Daniel, B., Jacob, Z., and Guillaum, C., Association between serum total bilirubin levels and functional dependence in the elderly. Internal Medicine Journal, 2012, 42(11): 119-207.
7. Echevery, G., Abdul, H.A., and Mayowa, A.A. Introduction to urinalysis in historical perspectives and clinical application. Methods in Molecular Biology. 2010, 641:112-120.
8. McPherson, R.A., and Ben, E.J. Microscopic examination of urine in clinical diagnosis and management: 2013, 35-42.
9. Stanley, C.G. Bladder varices rare cause of painless haematuria in idiophatic retroperitoneal fibrosis. Journal of urology, 2007, 73(1): 58-9.
10. Essentials of Clinical Pathology. First edition. Shirish M Kawthalkar .JAYPEE BROTHERS MEDICAL PUBLISHERS (P) LTD. Page 27-29.
11. HENRY'S Clinical Diagnosis AND Management BY Laboratory Methods. 23rd edition. Page 469-470.
12. Siamaki, A.H. A short guide to common heterotrophic flagellates of fresh water habitat based on the morphology of living organisms. Protists, 2014, 164 (6): 842-860.
13. Starkey, C., Jones, J. M., and Lee, D.W. Animal pigment bilirubin discovered in plants. Journal of the American chemical society, 2014, 131 (8): 28-30.
14. Lean, M. Effects of calcium and magnesium on urinary oxalate excretion after oxalate loads, Journal of urology, 2015, 23(2):43-51.
15. Freidig, D.M. Episodic gross haematuria in association with allergy symptoms in a child. Clinical Nephrology, 2011, 58(5): 389-392.
16. Sharib, C.W. Renal Calculus in children. Journal of urology, 2013, 107(2):306-307
17. Melisa, D.S., Ureteropelvic junction obstruction with concurrent renal pelvic calculi in pediatric patients a long term follow up. Intl Journal of endocrinology, 2014, 34(2):56- 60.