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FORMATION OF URINARY CALCULI IN PAKISTANI PATIENTS FROM DIFFERENT AREAS

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ABSTRACT

A survey reveals that urinary tract calculi formation is very common in different areas of Pakistan. This may be due to many factors related to water mineral contents, diet composition and environmental factors. The aim of present study is to determine the chemical composition of urinary calculi in patients of different area of Pakistan of both sexes. The quantitative analyses were carried out by spectrophotometric method. The majority of stones were found to be calcium oxalate type stones with an overall male to female ratio 6:1.

Key-words: Urinary calculi, chemical composition, calcium oxalate, Pakistani patients

INTRODUCTION

Urinary stone formation is the tliird most common affliction known to mankind. Urolithiasis or calculogenesis is a general term rcfering to the formation of stones in urinary tract. Uroliths are polycryslalline masses composed of precipitated urinary salts or acid and are found in the collecting passages of kidney, ureters, bladder and urethra. These stones also contain organic matrix, mainly mucoproteins, which make up approximately 2-5% of the total stone weight (Binette and Binette, 1991]. Urolithiasis is common all over the world but in certain geographical areas the incidence is much higher, 'the global distribution of Urolithiasis divides very sharply into two types. In industrialized areas, upper urinary tract stones of calcium salts are quite common but in children both upper and lower tract stones are relatively less frequent. In underdeveloped areas bladder stones of ammonium hydrogen urate or uric acid admixed with calcium oxalate are more common in children (William and Thomas, 1992). But stones of any type in adults are comparatively low. The high incidence of upper tract stones which are reported to be increasing with a concomitant decrease in the number of lower urinary tract stones arc strongly correlated with the level of affluence and sedentary life of the populations in industrialized areas (Takaski, l986]. Apart from geographical variations, the incidence of stone formation also varies with the age and sex of the patients. In countries like Afghanistan, Pakistan, India and Thailand the age varies between 1 -30 years of both sexes. In Europe onset of disorder round about 30 years (IIughes and Norman, 1992). Crystallization and subsequent stone formation occurs either because of hyper excretion of certain agents resulting in the super saturation and precipitation of salts or acids in urine or because urine lacks those agents (inhibitors) which tend to inhibit crystallization, crystal growth and aggregation. A large number of substance including citrates, pyrophosphate, magnesium, glucosamine aminoglycans, acid peptides and glycoproteins have been ascribed inhibiting properties. The lithogenic substances contribute to urinary stone formation only when the balance between super saturation and inhibitory action becomes disturbed in urine and facilitates pH dependent precipitation (Cupisti *et al*., 1992). The aims of our studies were to analyze the urinary tract stones of patients belong to different areas of Pakistan and to find out the nature of calculi with respect to meir family back ground, drinking and eating habits.

Collection of samples

The samples were collected from kidney patients from different areas of Pakistan, mostly belonging to Quetta, Karachi, and Lahore. Samples (10) were collected from each city. The oldest age from 50 years and of youngest 1.5 years. Although majority of cases were of the age 1-30 years. The highest prevalence has been reported by different research workers in the age between 30-50 years. In this study there were 30 calculi samples, out of which only 5 were of females and 25 were of males.

Qualitative analysis

The samples were washed with distilled water .Weighed out the stones and described their shape and color .The samples were ground to a fine powder.

Carbonate test

Added dilute HCl to a small portion of the stone samples, effervasence showed the presence of carbonate.

Calcium test

A small amount of powder sample was dissolved in 1M HC1 and filtered 4 drops of sodium oxalate and 2 drops of ammonia were added to the 3 drops of stone extract solution. A white precipitate showed the presence of calcium.

Table 1. Compositions of stones.

Age (years)	Sex	Site	Compositions
50	M	Rt.Kidney	Calcium phosphorous oxalate
2.5	M	Bladder	Calcium phosphorous oxalate
50	F	Bladder	Calcium phosphorous urate
17	M	Bladder	Calcium phosphorous oxalate
3	M.	Bladder	Calcium phosphorous
5	M	Ureter	Calcium phosphorous
25	M	pelvis	Calcium phosphorous oxalate
7	M	Nepher.	Calcium phosphorous
2	F	Ureter	Calcium phosphorous
3	M	Bladder	Calcium phosphorous oxalate
15	M	Bladder	Calcium phosphorous
1.5	M	Bladder	Calcium phosphorous
4	M	Pelvis	Oxalate carbonate
27	F	Rt.Kidney	Calciumphosphorous oxalate
24	M	Rt.Kidney	Calcium oxalate
30	M	Nepherons	Calcium oxalate urate
20	M	Rt.Kidney	Calcium phosphorous
33	M	Ureter	Urate oxalate
13	M	Pelvis	Calcium phosphorous oxalate
7	M	Rt.Kidney	Calcium carbonate
20	F	Lt.Kidney	Calcium carbonate
29	F	Ureter	Calcium
12	M	Lt.Kidney	Calciumoxalate
9	M	Bladder	Calcium phosphorous urate
25	M	Lt.Kidney	Calcium
7	M	Bladder	Oxalate urate
35	M	Pelvis	Calcium oxalate
4	M	Lt.Kidney	Calcium phosphorous
25	M	Lt.Kidney	Oxalate phosphorous
20	m	Pelvis	Calcium oxalate

M = Male; F = Female; Rt. = Right; Lt. = Left

Uric acid test

A 5-10 mg of powder sample was added to 1ml of 0.1N sodium hydroxide and heated, centrifuged, add a drop of supernatant on spot plate, I drop of sodium carbonate and phosphotungstic acid were mixed, blue colour indicated the presence of uric acid (Freeman and Beeder, 1982).

Calcium oxalate

The 5-10 mg powdered calculi was transferred in a test tube, add 0.1 N I ml NaOH and warmed in 60°C water bath for 5 minutes. Centrifuged, decant off the supernatant, added a drop of concentrated HCl and 0.5 ml distilled water boiled gently for a few seconds, cooled, centrifuged and to 0.3 ml of this supernatant added potassium acetate reagent to pH 3-4. Precepitation indicated the presence of calcium oxalate (Hamernyik, 1977).

Triple phosphate

Heated a little powder with 10% cone. potassium hydroxide solution. Evalution of ammonia showed the

presence of triple phosphate.

Ammonium urate

The test done for triple phosphate also showed the presence of ammonium urate. To distinguish between them Nesseler's Reagent was used, which showed that only 6 calculi had triple phosphate and 4 had ammonium urate (Andersen, 1961).

QUANTITATIVE ANALYSIS

Estimation of phosphorous by spectrophotometrically

For the calibration curve of phosphorous determination 0.038 mg/ml was prepared, 0.4343 g potassium hydrogen phosphate in 100ml water. The series of standard were prepared. Then 2.5 ml sodium molybadate solution and 1ml hydrazine solutions were added, absorbance noted at the filter of 830 nm. The concentration of stone samples were determined by calibration curve.

Estimation of calcium and oxalate

A 2 ml HC1 extract of stone solution, 2 ml of 4% ammonium oxalate solution was added or 2 ml of 4% calcium chloride was added (in case of oxalate estimation), centrifuged and washed with 2% ammonia solution. The precipitate was dissolved in 2 ml 0.1M sulphuric acid. A blank was made by taking 2ml 0.1M sulphuric acid in a tube. Both tubes were placed in hot water bath for 30 minutes and titrated with 0.1M potassium permanganate solution till pink colour appeared (Galon *et al*., 1996).

Experimental design

Detailed chemical evaluation regarding the formation of renal calculi was done / in 30 patients from hospitals of Pakistan. Detailed history was obtained f including family background, drinking habits, types of vegetables used and source of water supply. In this work attempts had been made to analyze the urinary calculi constituents qualitatively and quantitatively as possible by different methods. The major constituents like phosphates, oxalates and calcium have been analyzed quantitatively. While other constituents like uric acid, ammonium, magnesium, carbonates, xanthine and cystine were analyzed qualitatively.

RESULTS AND DISCUSSION

Table 1 shows the composition of different samples analyzed. The samples collected from kidney patients from different areas of Pakistan, mostly belonging to Quetta, Karachi, and Lahore, 10 samples from each city. The oldest age from 50 years and of youngest 1.5 years. Although majority of cases were of the age 1-30 years. The highest prevalence has been reported by different research workers is the age between 30-50 years. In this study there were 30 calculi samples out of which only 5 were of females and 25 were of males. Among 30 patients 21 had multiple stones (male-18 and female -3) while 9 had single stone (male-7 and female-2). Ureter stones were found in 3 patients (male-2 and female-1). Patients with kidney stones were 8 (male-6 and female-2). Bladder stones were also 8 (male-7 and female-1). In hilly areas frequent use of vegetable, milk products and protein rich food (meat, egg etc.) is common. The analysis results showed that mostly stones contain Calcium phosphate and Calcium oxalate, uric acid stones were also present to some extent. Other constituents were also present in the samples analyzed. The male to female ratio is 6:1. It is suggested that the difference in the two sexes lies in the emptying mechanism at the bladder neck (Anderson, 1961). The patients with positive case family history were prone to develop stones. This suggests that some faulty dietary and fluid in take habits are common in these families. The presence of Calcium oxalate calculi in most of the patients may be due to the single cereal based diet and tea which is high in oxalate. The chemical composition of urinary calculi show geographical, age and urinary tract related variations (Galon *et al*., 1996). Sensitive components of food like proteins, vitamins and trace elements are prone to many possible interactions in the food itself or in the body such interactions have major influence on the formation of different calculi. It is reported that the great number of interactions between macro and micro nutrients and naturally occurring antinutrients such as enzyme inhibitors vitamins antagonist saponines and polyphenoles under certain unfavorable food combinations increase the tendency to form urinary calculi (Bodo and Maksay, 20021. Among people of Pakistan self-medication is common. They use to take protein extracts ioi common diseases. The disadvantage is that, people used them without precautions and limits, the excessive intake may cause different interactions in the food. There are many different types ofcomplexation of amino acids with bio available trace metal or micron utrients occurs (Roat-Malone, 2002). Taking certain precautionary measures can reduce these

interactions. Our studies showed that high protein diet has positive interaction with unfavorable food components. At stomach pH, which is very low near about two, at this pH complexation does not occur. The intestinal pH is alkaline, most of the interactions occur at intestinal pH, if the nature of food is alkaline most of the calcium gel precipitated do not absorb therefore such types of foods have ability to form calcium oxalate stones.

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