SCIENTIFIC ARTICLES

# Chemical composition of kidney stones obtained from a cohort of Sri Lankan patients

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## Abstract

### Introduction

Evaluating the composition of kidney stones is important for the treatment and prevention of nephrolithiasis.

### Objectives

The aims of our study were to determine the composition of the renal stones in a cohort of Sri Lankan patients and to compare the composition of staghorn calculi with non-staghorn calculi.

### Methods

Kidney stones removed from adult patientswere analysed using chemical methods. Patients residing in different climate zones of Sri Lanka were analysed separately and comparisons made.

### Results

A total of 50 renal stones from adults (male:female = 2.6:1) were included in the study. There were 21 staghorn calculi. The male to female ratio among patients with staghorn calculi was 3.2:1. There was no statistically significant difference in distribution of staghorn calculi and non-staghorn calculi among males (p=0.17) and among females (p=0.06). Oxalate stones constituted 86% of renal stones. Ten percent were mixed stones. Phosphate and struvite calculi contributed 2% each. Even among staghorn calculi, 76% were oxalate stones. There was no statistically significant difference between contents of staghorn and non-staghorn calculi. The composition of stones in three climatic zones of Sri Lanka was similar except for phosphorus.

### Conclusions

The majority of renal stones (86%) in Sri Lanka are calcium oxalate stones.Contrary to the traditional view based on studies done in the western world, only 5% of staghorn calculi removed from patients in Sri Lanka are struvite or infection stones.

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## Introduction

Urinary stone is a rock particle or a crystal in the urinary tract formed by substances in urine. These have plagued humans since the beginning of recorded history. Archaeologists have uncovered them in mummified remains of Egyptians which are estimated to be 7000 years old [1]. In the western world about 5-10% of the population develop urinary tract stones [2]. The incidence is rising in women and children [3]. India, Pakistan and Southern China comprise an important part of the stone belt in Asia [4].Monotonous diet based on rice, water quality and hot climate which contribute to this high incidence are operable in Sri Lanka too, making it likely to be a part of the stone belt.

Stones are commoner in the young and the effects of absenteeism from work due to stones on economy and family is great. Furthermore, urinary calculi are the commonest cause of obstructive uropathy and renal impairment in Sri Lanka [5]. Hence it is important to effectively treat these stones to prevent serious morbidity and even mortality.

The composition of stones has a direct impact on treatment, secondary prevention and prognostication [6]. Calcium oxalate monohydrate stones are hard and resistant to extracorporeal shock wave lithotripsy [7]. Struvite calculi are associated with more infective complications postoperatively. Though treatment of renal stones is its removal, the primary cause that produced the stone should be known and corrected in order to prevent recurrences. The analysis of the stone helps to predict the underlying metabolic abnormality [8].

The composition of stones varies widely among populations [4,9]. Even in the same country stone composition can change according to the geographical locality [10]. Therefore it is essential to know the pattern of calculi in a country and region when planning treatment. A study done in Sri Lanka has shown that the stone composition of Sri Lankan staghorn calculi are likely to be different to that described in literature which are based on studies done in the western world [11]. Hence there is a need to study the composition of kidney stones in Sri Lanka. However there is a paucity of published data about composition of renal stones especially those of staghorn calculi in Sri Lankan patients.

The objectives of our study were to determine the composition of renal stones in a cohort of Sri Lankan patients and to compare the composition of staghorn calculi with non-staghorn calculi.

**Materials and methods**

This was a prospective study. The patients who underwent surgery to remove renal stones in a urology unit of a tertiary care hospital from 1 June 2012 to 31 May 2013 were included in the study. The demographic data including age, gender, place of residence during the last ten years, operative details and stone morphology were recorded prospectively.

Chemical methods were used to perform stone analysis. Calculi were thoroughly washed with tap water to remove attached debris. Then they were rinsed with deionised water and air dried for two weeks in a plastic container. Once the calculi were dry, they were weighed and then grounded to fine powder using mortar and pestle. These powdered calculi were used for qualitative and quantitative analysis.

Qualitative analysis was done to determine the presence of ammonium ion, carbonic ion, and cystine. Detection of ammonia was done using the Nessler's reagent [12] and carbonate by using HCl [13]. Cystine was detected using the method described by Hodgkinson [14].

All powdered renal calculi were analysed quantitatively for oxalic acid, calcium, phosphorous, uric acid and magnesium. Oxalic acid estimation was performed using the titrimatric method [14]. Estimation of calcium was done by the spectrophotometric method [15]. Inorganic phosphorous was determined by phosphomolybdate method of Fiske & Subbarow [16] modified by King [17]. Phosphotungstate reduction method was used to estimate the uric acid amount [18]. Magnesium estimation was performed using commercially purchased reagent MAGNESIUM liquicolor from Human®.

After estimating the percentage composition of the stones, classification described by Abdel-Halim et al. was used to classify the stones [19]. Stones containing more than 20% uric acid were classified as uric acid stones and those containing more than 40% oxalate were considered as oxalate stones. Stones containing more than 10% phosphate with less than 20% uric acid and less than 40% oxalate were phosphate stones. Stones containing more than 3% magnesium were considered as infection (struvite) stones [19].

Sri Lanka is divided into three climate zones according to the average daily temperature and annual rainfall [20] (Figure). Those are dry zone (average daily temperature 300C and average annual rainfall of 1250 mm to 1900 mm), intermediate zone (average daily temperature 240C - 260C and average annual rainfall of 1900 mm to 2500 mm) and wet zone (average daily temperature 130C - 270C and average annual rainfall of 2500 mm to 5000 mm). The dry zone includes Jaffna, Killinochchi, Vavuniya, Trincomalee, Mullaithivu, Mannar, Anuradhapura, Polonnaruwa, Batticaloa, Ampara, Puttalam and Hambanthota districts. Kurunegala, Matale, Badulla and Moneragala districts are classified under the intermediate zone. The remaining nine districts – Colombo, Gampaha, Kalutara, Galle, Matara, Ratnapura, Kegalle, Kandy and Nuwera-Eliya are included in the wet zone. The study participants' resident climate zone was determined according to his/her residence during the previous 10 years.

Statistical software Minitab 16 was used for the analysis. P value $< 0.05$ was considered significant. Distribution pattern of stone contents were skewed. This was confirmed by the Anderson-Darling normality test.

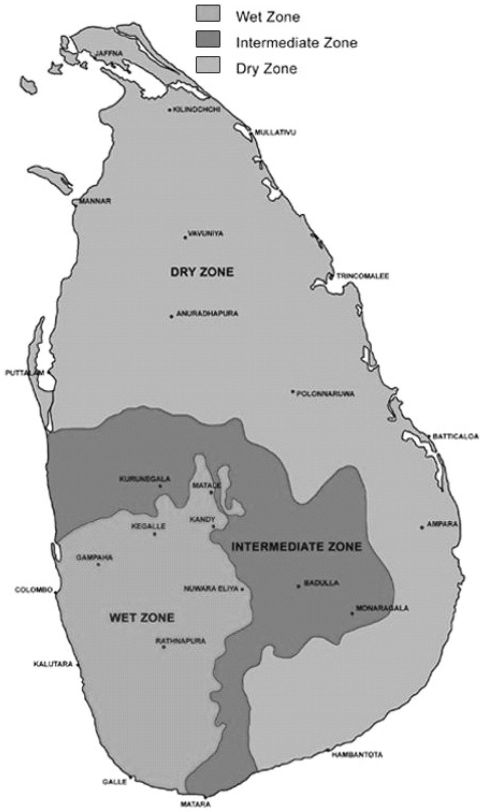


Figure 1. Climatic zones of Sri Lanka

Therefore non-parametric tests were used for statistical analysis. Median was used to compare the composition of calculi.

Approval for the study was obtained from the Ethics Committee of the Colombo South Teaching Hospital and informed written consent was obtained from participants.

**Results**

There were 50 stone samples collected during the study period. Average age was 49.8 years (range: 26 – 76 years). The commonest age group who had surgery for stones was 41 – 60 years (Table 1). There were 36 men. Male to female ratio was 2.6:1.

There were 21 staghorn calculi. The average age of patients with staghorn calculi was 51.2 years while the average age of non-staghorn calculi was 49 years. The male to female ratio among patients with staghorn calculi was 3.2:1 while the corresponding value for non-staghorn calculi was 2.2:1. There was no statistically significant difference in distribution of staghorn calculi and non-staghorn calculi among males and females ($p>0.05$) (Table 2).

| Age group | Number (%) |
|---|---|
| 21-40 years | 11 (22%) |
| 41-60 years | 29 (58%) |
| 61-80 years | 10 (20%) |
| Total | 50 (100%) |

Table 1. Age distribution of patients

| | Male | Female | Total | *p* value |
|---|---|---|---|---|
| Staghorn calculi | 16 | 5 | 21 | 0.001 |
| Non-staghorn calculi | 20 | 9 | 29 | 0.001 |
| Total | 36 | 14 | 50 | |
| *p* value | 0.171 | 0.057 | | |

Table 2 - Gender distribution among staghorn and non-staghorn calculi

According to the results, all calculi had oxalic acid and uric acid which were the commonest components in calculi. Calcium and phosphorous were next common components followed by magnesium. Ammonium ion was detected in 59.5% renal calculi. None of the calculi contained carbonate or cystine.

Composition of renal calculi according to gender is given in table 3. The Mann Whitney test was used for the comparison of median values of the different components of calculi according to the gender. There was no statistically significant difference between males and females except in oxalic acid ($p<0.05$).

Kruskal-Wallis test was used for the comparison of median values of the different components of the calculi among the different age groups and the differences were not statistically significant (Table 4).

Distribution of the type of calculi is given in table 5. Accordingly 86% were oxalate stones. Even among staghorn calculi the majority (76%) were oxalate stones

(Table 5). Struvite or infection stones contributed to 5% of staghorn calculi only. Distribution of oxalate stones among staghorn calculi and non-staghorn renal stones were compared (Table 6). There was no statistically significant difference (p>0.05).

There were 32 patients from the wet zone, 12 from the dry zone and 6 from the intermediate zone. Kruskal-Wallis test was used to compare the median values of different components of calculi obtained from patients living in the three climate zones (Table 7). There was no statistically significant difference (p> 0.05) in the composition between the three zones except phosphorus (p<0.05).

| Analyte | Male | Female | p value |
|---|---|---|---|
| Oxalic acid (mg%) | 52.93 | 49.67 | 0.01 |
| Calcium (mg%) | 16.97 | 16.25 | 0.37 |
| Phosphorous (mg%) | 0.24 | 0.31 | 0.12 |
| Uric acid (mg%) | 0.07 | 0.10 | 0.47 |
| Magnesium (mg%) | 0.17 | 0.13 | 0.85 |

Table 3. Median value of different components of calculi

| Analyte | 21-40 years | 41-60 years | 61-80 years | p value |
|---|---|---|---|---|
| Oxalic acid (mg%) | 52.03 | 52.09 | 51.64 | 0.85 |
| Calcium (mg%) | 16.61 | 16.96 | 17.16 | 0.84 |
| Phosphorous (mg%) | 0.31 | 0.24 | 0.28 | 0.57 |
| Uric acid (mg%) | 0.12 | 0.07 | 0.06 | 0.15 |
| Magnesium (mg%) | 0.17 | 0.17 | 0.14 | 0.32 |

Table 4. Median value of components of calculi according to

| Type | Number (%) |
|---|---|
| Oxalate | 43 (86%) |
| Phosphate | 01 ( 2%) |
| Mixed | 05 (10%) |
| Struvite | 01 ( 2%) |
| Total | 50 (100%) |

Table 5. Different types of stones according to the composition

| Type | Staghorn calculi (%) | Non-staghorn calculi(%) |
|---|---|---|
| Oxalate | 16 (76%) | 27 (93%) |
| Phosphate | -- | 01 (3.5%) |
| Mixed | 04 (19%) | 01 (3.5%) |
| Struvite | 01 ( 5%) | – |
| Total | 21 (100%) | 29 (100%) |

Table 6. Different types of stones among staghorn and non-staghorn calculi

| Analyte | Dry zone | Intermediate zone | Wet zone | p value |
|---|---|---|---|---|
| Oxalic acid (mg%) | 51.52 | 55.69 | 51.75 | 0.23 |
| Calcium (mg%) | 17.19 | 16.84 | 16.96 | 0.95 |
| Phosphourous (mg%) | 0.23 | 0.17 | 0.31 | 0.04 |
| Uric acid (mg%) | 0.10 | 0.10 | 0.07 | 0.26 |
| Magnsium (mg%) | 0.14 | 0.20 | 0.16 | 0.66 |

Table 7. Median values of chemical components of stones from different climate zones

## Discussion

The majority of stones (86%) in our study were calcium oxalate similar to that (93%) of India [4]. In contrast the percentage of oxalate stones in Jordan is 37.6% [21], in France 66% [22] and the USA 26% [23]. The percentage of struvite calculi varies from country to country. In our study there were only 2% struvite calculi which are also known as infection stones. In India it is 1.4% [4] while in Pakistan and Jordan the percentage is 17.4% [24] and 13.7% [21] respectively. In the USA struvite stones constitute 22% of the stones analysed [23]. We did not encounter uric acid stones while it is 0.93% in India [4] and 9.4% in Pakistan [24]. In Jordan it is 20.2% which may be related to high consumption of nuts which are a rich source of uric acid [21]. In the USA it is 5% [23]. The results show that distribution of different types of calculi in Sri Lanka is similar to that of India but differs significantly from that of the Western world and Middle Eastern countries. Identification of this pattern is important as it may determine the response to treatment by standard therapeutic manoeuvres [25] or by herbal extracts [26].

A staghorn calculus is a stone that occupies the renal pelvis and extends into at least two major calyceal systems [27]. Management of patients with staghorn calculi is a challenging problem. High incidence of intra-operative complications such as damage to the kidney and renal pelvis, frequent occurrence of residual stones and sepsis related complications are some of the problems associated with staghorn calculi. Staghorn calculi are commoner among females in the Western countries. However in our study staghorn calculi are commoner among men.

According to the literature originating from European countries, most (60%-75%) staghorn calculi are struvite or infection stones [28,29]. In our study such stones among staghorn calculi was only 5%. In India it is 4.02% [4]. The distribution of different types of calculi between staghorn and non-staghorn stones in our study was similar with no statistically significant difference. This shows that infection or struvite stones are not the common type of staghorn calculi in Sri Lanka as described in literature. This is so in India as well. Hence the aetiology of staghorn calculi differs significantly between the Western world and Sri Lanka. There are important clinical implications in this finding. The associated sepsis of struvite calculi destroys the kidney tissues and impairs the function of kidneys with longstanding staghorn calculi. Hence surgical removal is the treatment recommended for staghorn calculi [6]. Since infections stones are rare among staghorn calculi of Sri Lankan patients, in a subgroup of high risk patients with a short life expectancy, it may be possible to manage staghorn calculi expectantly rather than subjecting such patients to a major operation with potential serious complications. Such a subgroup of staghorn calculi can be identified by analysing urine pH and culture for bacteria [11]. It has been shown that damage to renal tissues by oxalate stones is minimal compared to struvite and brushite (phosphate) stones [30].

According to our study, the composition of renal stones in different climate zones of Sri Lanka was similar. Therefore the aetiological factors that play a role in causation of renal stones appear to be same throughout the country contrary to common beliefs. Since the pattern of calculi is similar to India, it may be that the monotonous diet mainly based on rice consumed by Sri Lankans could be a major determining factor for renal stones. The high acidic load resulting from rice consumption is implicated in formation of renal stones [4,31].

Different analytical methods are available to study components of renal stones. Infrared spectroscopy, X-ray diffraction crystallography and scanning electron microscopy are superior to the chemical method we used. This was a limitation of the study. However such facilities are not accessible to Sri Lankan clinicians and the method that is available and can be performed locally is chemical analysis. The stones that were analysed were obtained after surgical removal. Therefore the study sample consisted of large stones which warrant surgical methods for removal. This may be having a bearing on the overall composition of all stones.

## Conclusions

The common type of renal calculi in Sri Lankan patients is calcium oxalate. Staghorn calculi in Sri Lanka are commoner in men similar to other kidney stones. They consist mainly of calcium oxalate and are not struvite or infection stones. There is no significant difference in the composition of calculi in different climate zones of Sri

Lanka except for the phosphorous content.

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