The Impact of Calculi Weight and Frequency of Calculi Formation on their Compositions

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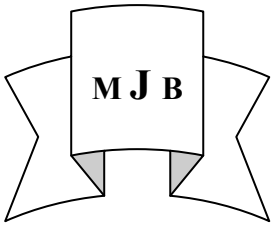

Abstract

The present investigation was conducted to evaluate the effect of calculi weight and the frequency of calculi formation in their compositions. The present investigation was conducted to evaluate the urinary calculi compositions quantitatively. Fifty-two calculi were obtained from 38 males and 14 females afflicted with urolithaisis, The calculi were subjected to qualitative analysis, and sought for calcium, uric acid, phosphorus, oxalate, magnesium and ammonium ion contents. Small calculi were demonstrated to be rich in calcium oxalate, while large calculi were rich in calcium phosphate. The frequency of calculi formation was demonstrated to influence the calculi compositions. Thus, calculi of non-stone formers were exhibited to be rich in calcium oxalate, while those of stone formers were rich in uric acid.

الخلاصة

لغرض تقيم كلاً من وزن الحصى وتأثير تكرار الحصى على المكونات. تم تصميم الدراسة الحالية لتقدير مكونات حصى المجاري البولية تقديراً كمياً. . تم تحليل الحصى بطرائق التحليل النوعي والكمي واستخدمت البيانات لحساب كلاً من الكالسيوم و حامض اليوريك و الفوسفات و الاوكزالات و المغنيسيوم وايون الامونيوم. تضمنت الدراسة ٥٢ حصاةً أخِّذت من ٣٨ رجل و ١٤ امرأة مصابين بحصى المجاري البولية. أظهر تأثير وزن الحصى على المكونات أن اوكزالات الكالسيوم هي المكون الأكثر في الحصى الصغيرة بينما كانت الحصى الكبيرة الوزن غنية بفوسفات الكالسيوم. . ولبيان تأثير تكرار الحصى على المكونات فقد بينت نتائج تحليل حصى المرضى المصابين لأول مرة محتواها العالي بأوكزالات الكالسيوم بينما كانت حصى المصابين لأكثر من مرة غنية بحامض اليوريك.

Introduction

rinary calculi have been found Uaround for centuries. Many notable individuals have suffered with urinary calculi, namely Louis Napoleon III (Emperor of France), Benjamin Franklin (American inventor), and Lyndon Johnson (American president) [1].

Urinary stones are hard masses of crystals formed in the kidney or urinary tract [1, 2].They have afflicted human kind since antiquity [3]. It would be interesting to know that first evidence of urinary stones dates back to 4800 b.c., when a bladder stones discovered in Egyptian mummy at El Amarh-Egypt [1, 4]. At the end of 18th century, as soon as modern chemistry was created, dedicated physicians tried to apply it to medicine. The first step was made by Scheele (1776) in Sweden who identified uric acid in calculi and during the following thirty years Fourcroy and Vauqueline in Paris and Wollaston, Porson and Prout in London identified the various salt, uric

acid, urate, various phosphate oxalate calcium, ammonium, cystine and magnesium forming current calculi[5].

The calculus is frequently composed of nucleus surrounded by the layers. Uroliths can be found in kidneys, ureters, bladder or urethra. They vary considerably in size from small "gravel like" stones to large "staghorn"' calculi. The calculi may stay in the position in which are formed, or migrate down the urinary tract producing symptomes along the way [6].

The knowledge of composition of urinary stones is important because uorlithiasis is a recurrent disease in many people and preventive measure must be based on such information. Uric acid and cystine stones are relatively insoluble in acidic urine and quite soluble in alkaline urine phosphate stones are relatively insoluble in alkaline urine and acidification may be theoretically practical in some cases [7].

Most stones consist of calcium, combined with either oxalate, phosphate (or both) and these types of stones account the majority of calculi in the population [8]. Ten percent of stones are formed of uric acid and often associated with gout [9-11]. Cystine stones represent 1 % to 2 % of urinary calculi [12], it is clinically important source of stones disease in children accounting for 6 %to 8 % of pediatric urinary calculi [13-15]. However, the formation of urinary calculi composed of xanthine is very rare occurance and xanthineuria first identified and named in 1817 by Marcent during the examination of urinary calculus given to him by Babington of London [16].

The type of bacteria in urinary tract should be determined by culture than by stain alone because of need for a ascertaining certain characteristics of bacteria present, since the role of these bacteria in stone formation was recognized as early as 1901 [17].

The current investigation was conducted to analyze the urinary calculi quantitatively and assess the contribution made by factors such as weight and frequency of calculi formation.

Material and Methods

Patients and collection of specimens

Fifty two calculi obtained from patients afflicted with urolithiasis**.** They were 38 males and 14 females. All of them were attended the hospitals of Al Forat AL–Awsat province. Stones were obtained by surgery, passed spontaneously or from records. The calculi were washed to remove any dried blood or other matter, dried in an incubator at 37 Cºand weighed. They were pulverized in a mortar, a sample of the powdered stone was weighed, dried overnight in an oven at 105 Cº and weighed again to determine the water content. The residue after drying was divided into aliquots for chemical analyses. The calculi were subjected to quantitative analysis to determine their compositions.

Chemicals and apparatus

All laboratory chemicals and reagents were of analar grade**.** Potassium permanganate, Sulphuric acid, Sodium hydroxide and Sodium hypochlorite, were obtained from Merk Company, EDTA was obtained from BDH Company, Nitric acid from Fluka Company. Sodium tungstate from Fisher Company, Polyvinyl alcohol and Titan yellow, Ammonium sulphate, and Phenol were obtained from BDH Company, Disodium hydrogen orthophosphate from Fluka Company, Unico UV-2100 spectrophotometer, United products and instruments U.S.A., Julabo Water bath**,** super-mixer**,** pH meter, HANA

from Italy Company were used during our study.

Preparation of aliquots

Two aliquots of stone powder were weighed; the first contained 40 mg of such powder. It was mixed with 8 ml of concentrated nitric acid and used for determination of calcium, magnesium, phosphate by conventional methods. The second contained 100 mg of stone powder, it was mixed with 20 ml of EDTA (5 %) adjusted at pH 7.8 with NaOH. It was used for estimation of uric acid [18].

Methods

Calcium concentration was measured in stones of subjects according to the method of Hodgkinson [19]). Uric acid concentration was measured according to the enzymatic colorimetric method. Uric acid concentration was calculated from the standard curve constructed in the range of 1-6 mg/dl. Phosphorus concentration was measured in stones according to the method of Henry[20]. Phosphorus concentration was calculated from the standard curve constructed in the range of 0.5-5 mg/dl. Oxalate concentration was estimated according to the permanganate titration method [21]. The concentration of oxalate found in the stones was calculated from the volume of the equivalent potassium permanganate solution. Magnesium concentration was determined according to the titan yellow method [22]. Magnesium concentration was estimated from the standard curve constructed in the range of 0.5-2 mg/dl. Ammonium ion concentration was estimated according to the McCullough's modified method, which was based on the indophenol reaction of ammonium ion[23].

Statistical analysis

The results were expressed as mean ± SD and analyzed statistically. The differences between the results of location urinary calculi were assessed by student's t-test. Significant variation was considered when p value less than 0.05. The linear regression analyses were applied for analysis of the data.

Results and Discussion

To perceive the relevance of the calculi weight with their composition, they were classified into two groups, the first (13 calculi) contained those of weight less than 1g, it was considered as a group of small calculi. The second group (30 calculi) consisted of these of weights equal to 1g or more, it was regarded as a group of large calculi. The data were analyzed by using the student t test and linear regression analysis. Table 1 shows that calcium oxalate content was significantly ($p < 0.01$) increased in small calculi when compared with those of large calculi. Calcium phosphate content was significantly ($p < 0.05$) increased in large calculi as compared with those of small calculi. While magnesium ammonium phosphate and uric acid compositions did not show any variation during a comparable analysis.

Urinary calculi can vary in size and weight, and can be composed of a number of different constituents [24]. A number of statistically differences were observed between large and small calculi.

The results of the present study revealed a significant increase of calcium oxalate in small calculi as compared with large calculi. This observation may indicate that most calculi may start as an aggregate of calcium oxalate crystals [25). It is of interest in this sense that urine is usually supersaturated with respect to calcium oxalate [26]. Moreover calcium oxalate crystals is the most commonly found in freshly voided urine [27].

The linear regression analysis of urinary (kidney and vesical) calculi compositions indicated significant ($r = -0.22$, $p < 0.05$, fig 1) negative correlation for calcium oxalate with weight of urinary calculi and significant ($r = 0.26$, $p < 0.05$, fig 2) positive correlation for uric acid with weight of kidney calculi (table 2).

Uric acid was found to be positively correlated with the weight of calculi. This result may belong to that most of the large calculi are vesical in their origin, in which uric acid is present in high amount[28,29].

Magnesium ammonium phosphate data of present study demonstrated comparable results in both large and small calculi. Two explanations may be applied to demonstrate the reason of this observation. The first the effect of anatomical site, i.e., whither the region of calculi is the kidney or the bladder. The second is the sex of patients, since magnesium ammonium phosphate is excessively produced in females and most of large calculi are vesical and produced by males [1].

To determine the effect of patient history's on the parameters of urinary calculi, patients were categorized into two groups. Group 1 contained those of non stone former (43 calculi). Group 2 consisted of stone formers calculi [9]. The statistical analysis of the results was carried out using the student t test. The results of urinary calculi constituents (table 3) demonstrated significant ($p < 0.05$) elevation for calcium and oxalate in non stone former patients group when compared with those of the stone formers. Uric acid was found to be raised significantly ($p < 0.005$) in calculi
of stone former patients when compared with those of non-stone formers.

The degree to which a stone former is able to reproduce the pattern of chemical composition of the initial calculus depends to a large extent upon whether the urine remains sterile[30].

In the present investigation a significant increases of calcium and oxalate were demonstrated in non stone formers as compared with stone former patients while stone former patients exhibited significant increase of uric acid comparable with that of non stone former patients. The most likely explanations for these data are that uric acid may decrease the activity of urine crystal inhibitors [31]. Increased uric acid content in patients may belong to the familial uric acid nehprolithiasis since it occurs at younger age and follows an autosomal dominant pattern of inheritance. Men and women are equally affected [32]. These results are consistent with those reported by Low and Stoller [33]. who reported that uric acid calculi tend to recur in anyone who does have this type of calculi.

Further examination of the staghorn calculi composition was carried out in order to verify the probable cause of the development of such calculi. Staghorn calculi from males and females were categorized in separated groups. They were examined for their components. The data were analyzed by using the student` t test. There was a significant ($p < 0.05$) raise of calcium phosphate in males staghorn calculi when compared with that of females. Other constituents did not exhibit significant variation (table 4).

The staghorn calculi analysis indicated that these calculi are produced in excess in females than males. They contained higher contents of magnesium ammonium phosphate, calcium oxalate and uric acid compared with those of males. It is believed that the predominant staghorn

calculi formation in female is related to urinary tract infection in these patients [34]. Urologists assume that most staghorn calculi are composed of struvite, while others stated that some calculi can assume staghorn configuration ; cystine, calcium oxalate monohydrate and uric acid which may grow to the point where they full the collecting system. Ligeman and others [35,36] showed some of staghorn calculi are composed of calcium oxalate, calcium phosphate or cystine.

Table 1 The effect of urinary calculi weight on their compositions.

Parameters	Subjects	Mean ± SD	Range	P value
Ca-oxalate	**Small** **large**	**40.16 ± 20.27** **21.29 ± 21.31**	**1.78 -73.10** **0.00- 71.00**	**<0.01**
Ca-phosphate	**Small** **large**	**20.87 ± 12.36** **29.67 ± 22.93**	**6.50- 49.52** **0.00- 77.00**	**<0.05**
Magnesium ammonium Phosphate	**Small** **large**	**5.77 ± 10.01** **13.00 ± 18.33**	**0.00- 30.97** **0.00- 73.00**	**N.S**
Uric acid	**Small** **large**	**20.29 ± 13.42** **31.64 ± 25.85**	**4.50- 53.40** **0.186- 95.23**	**N.S**

Table 2 The correlation of urinary calculi weight with calcium oxalate and uric acid contents.

	Ca-oxalate		Uric acid	
	r	p	r	p
Weight of calculi	**- 0.22**	**0.05**	**0.26**	**0.05**

Table 3 Urinary calculi compositions in relevance to the frequency of calculi formation.

Parameters	Subjects	Mean ± SD	Range	P value
Calcium	NSF SF	27.67 ± 11.22 19.68 ± 10.92	2.85 – 42.25 1.30 – 36.00	<0.05
Oxalate	NSF SF	18.74 ± 13.43 10.93 ± 8.17	1.10 – 45.10 1.10 – 30.00	<0.05
Phosphate	NSF SF	13.69 ± 10.09 8.29 ± 2.35	0.55 – 41.50 4.41 – 11.50	N.S
Magnesium	NSF SF	8.73 ± 8.58 3.66 ± 2.82	0.05 – 28.50 1.77 – 6.90	N.S
Ammonium	NSF SF	1.69 ± 1.06 1.10 ± 0.47	0.24 – 4.21 0.41 – 2.00	N.S
Uric acid	NSF SF	24.14 ± 20.71 47.32 ± 28.84	0.186 – 95.23 11.51 – 98.72	<0.005

SF: Stone former, NSF: Non stone former

Table 4*:* The differences in staghorn calculi composition

Description of patients	Composition of calculi (mg/100 mg)			
	Ca-oxalate	Ca- phosphate	Uric acid	Magnesium Ammonium Phosphate
From men	10.69 ± 12.59	54.88 ±13.18*	26.85 ± 2.62	14
From women	18.03 ± 10.49	27.01± 11.94	39.82± 22.82	16.12 ± 1.91

*** p < 0.05**

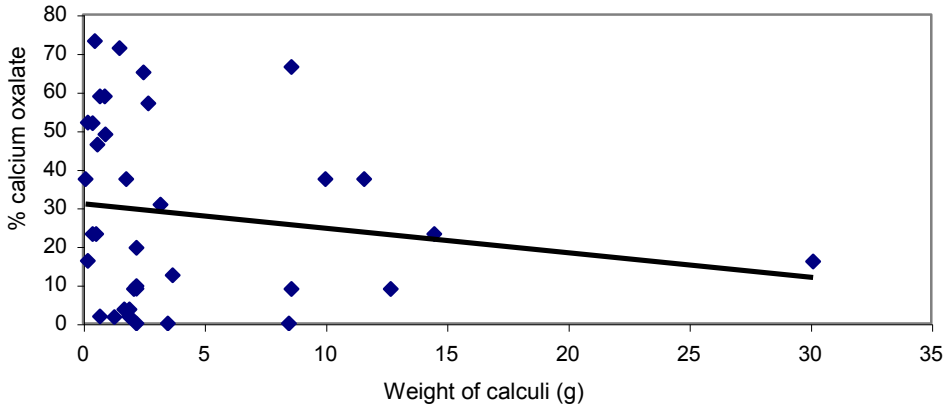

Figure 1 The correlation between weight of kidney and vesical calculi and calcium oxalate percentage.

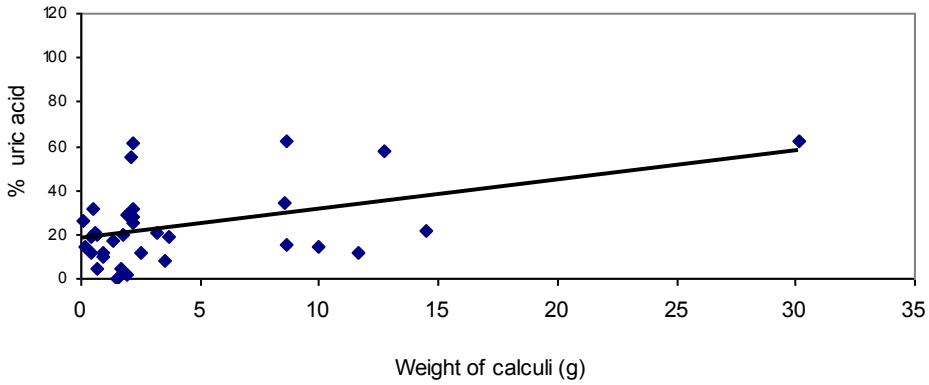

Figure 2 The correlation between weight of kidney calculi and uric acid percentage.

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