RELATIONSHIP BETWEEN CHEMICAL COMPONENT OF CALCULI AND ITS PHYSICOCHEMICAL PROPERTIES, PROTEIN COMPOSITION AND BLOOD URIC ACID CONCENTRATION IN PATIENTS WITH UPPER URINARY CALCULI

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ABSTRACT

Objective*: This study explores the relationship between the chemical components of calculi and the physicochemical properties of urine, protein composition and blood uric acid concentration in patients with upper urinary calculi.*

Methods: *The observation group was comprised of 160 patients with upper urinary calculi who needed lithotripsy through ureteroscopy and percutaneous nephrolithotomy in our urology department from August 2018 to August 2019. For the control group, 30 healthy people were examined at our hospital. Then, 3 ml of fasting venous blood was collected from preoperative patients and the control group. The uric acid level in all patients was detected using uricase, and 24 h urine samples were collected from all subjects. Calcium, phosphorus, magnesium, potassium and uric acid in urine were detected using an automatic analyser, and albumin (mAlb), α1-microglobulin (α1-MG), β2-microglobulin (β2-MG) and transferrin (TRF) were detected using an automatic special protein analyser. Also, stone samples were collected from the patients after the operation, and the chemical components of calculi in all patients were detected by the type LIIR-20 infrared spectrometer. The relationship between the chemical components of calculi and the physicochemical properties of urine, protein composition and blood uric acid concentration in patients with upper urinary stones was investigated.*

Results: *A total of 160 patients with upper urinary calculi that were detected by an infrared spectrum analyser participated in the study: 83 patients (51.88%) had oxalate-based stones, 35 (21.88%) had phosphonate-based stones, 23 (14.38%) had uric acid-based stones and 19 (11.88%) had apatite-based stones. All patients with upper urinary calculi were divided into an oxalate group, phosphate group, uric acid group and apatite group according to the component of the calculi. Compared with the 24 h urine physicochemical properties of the control group, it was found that the pH of the urine in the phosphate group and apatite group was significantly higher than that in the control group, the urine pH of patients in the uric acid group and the oxalate group was significantly lower than that in the control group, and the difference was statistically significant ($P<0.05$). Also, the urine volume of patients in the phosphate group, apatite group, uric acid group and oxalate group was significantly lower than that in the control group, and the difference was statistically significant ($P<0.05$). Furthermore, the urine phosphate level in the phosphate group was significantly lower than that in the control group, and the difference was statistically significant ($P<0.05$). The urine magnesium in the uric acid group was also significantly higher than that in the control group, and it was significantly lower in the oxalate group than in the control group ($P<0.05$). The levels of β2-MG and α1-MG in the oxalate group were significantly higher than those in the control group, and the difference was statistically significant ($P<0.05$). There was no significant difference in urine protein levels in other groups compared with the control group ($P>0.05$). The levels of uric acid of urine and serum in the oxalate group and uric acid group were significantly higher than those in the control group, and the differences were statistically significant ($P<0.05$). There was no significant difference in the uric acid levels of urine and serum in other groups compared with the control group ($P>0.05$). The Pearson correlation analysis showed that β2-MG was positively correlated with oxalate and urinary calcium (all $P<0.05$) and negatively correlated with pH ($P<0.01$); uric acid was positively correlated with oxalate, urate and urinary calcium ($P<0.05$) and negatively correlated with pH ($P<0.05$); and α1-MG was positively correlated with oxalate and urinary calcium ($P<0.05$ or <0.01) and negatively correlated with pH ($P<0.05$).*

Conclusion: *Oxalate calculi are the most common type of upper urinary calculi. The abnormal expression of uric acid and urinary protein in patients with upper urinary calculi is significantly related to the chemical components of the calculi and the physicochemical properties of urine.*

Keywords: *Upper urinary calculi, chemical components of the calculi, urine physicochemical properties, protein composition, uric acid.*

DOI: 10.19193/0393-6384_2021_2_134

Received *March 15, 2020;* ***Accepted*** *October 20, 2020*

Introduction

A common disease in urology, upper urinary tract calculi include kidney and ureteral calculi[1]. The main symptoms are bladder irritation and haematuria and sometimes nausea and vomiting[2], and its degree is related to the calculi site, size and activity and whether there is injury, infection or obstruction[3]. The incidence of upper urinary tract calculi has been increasing each year, and China is one of the three highest incidence areas of calculi worldwide[4]. Statistics show that the incidence of urinary calculi in China is up to 2.5%, and 25% of patients need to be hospitalized for treatment[5]. The current treatment options for urinary calculi include the medical drug lysolithography, extracorporeal shock wave lithotripsy, endoscopic lithotripsy, and open surgical incision and stone removal. However, the results are unsatisfactory and the recurrence rate is relatively high due to a lack of understanding of the calculi formation cause[6].

Therefore, it is important to find a test index to evaluate the type, component and size of calculi formation. Uric acid is one of the important components for the formation of uric acid calculi, and it is also involved in the formation of other components in calculi[7]. Studies have found that serum uric acid levels in patients with upper urinary calculi are significantly higher than those in the normal population, but the relevant mechanisms have not been reported in detail[8]. This study investigated the relationship between the chemical components of calculi in patients with upper urinary calculi and the physicochemical properties of urine, protein composition and blood uric acid concentration.

Information and methods

General information

Patients with upper urinary stones treated with ureteroscopy, lithotripsy and percutaneous nephrolithotomy in our hospital from August 2018 to August 2019 were recruited as the observation group. All patients in the study had a B-ultrasonography or CT examination of the urinary system, and calculi were found in one or more sites of the kidneys and ureters but the heart, liver and kidney functions were normal. Also, all participants and their families were informed and were required to provide consent. Patients were excluded if they had consciousness disorders, mental illness or the inability to take care of themselves; allergies; serious diseases in the heart, brain, liver or kidney; incomplete clinical data; diabetes, self-immune diseases, hyperuricemia or other diseases that can cause kidney damage; or were breastfeeding or pregnant.

There were 160 patients in the observation group, consisting of 110 males and 50 females, with an average age of 45.26±20.45 years and an average BMI value of 20.10±1.11 kg/m^2. In the same period, 30 healthy people who received a physical examination in our hospital were enrolled in the control group, which included 18 males and 12 females, with an average age of 45.33±19.76 years and an average BMI value of 20.11±1.09 kg/m^2. There were no statistically significant differences concerning age, gender and BMI between the two groups (P>0.05).

Observation indexes

Serum uric acid test: First, 3 ml of fasting venous blood was collected from patients and the control subjects before surgery and centrifuged at 3,000 r/min at room temperature.

Then, the serum was carefully separated and stored in a -80 °C refrigerator for testing to avoid repeated freeze-thaw cycles. Uric acid levels in all patients were detected using uricase.

24 h urine analysis

The 24 h urine specimens of all subjects were collected at 8 am on the same day and 8 am the next morning. Toluene was put into the urine at a ratio of 0.5 ml: 100 ml, and 20 ml urine after mixing well liquid inspection was collected. The levels of calcium, phosphorus and magnesium in the urine of all patients were determined using a fully automatic analyser. The potassium and sodium in the urine of all patients were detected by the ion-selective electrode method, and the uric acid level was found using the enzyme method. Albumin (mAlb), α1-microglobulin (α1-microglobulin, α1-MG) and β2-microglobulin (β2-microglobulin, β2) in the urine of all patients were detected through immunoturbidimetry and by using a fully automatic special protein analyser.

The chemical components of the calculi

The patients' calculi specimens were collected after the operation, and the type LIIR-20 infrared spectrum analyser was used to detect the chemical components of the calculi in all patients.

Statistical methods

The data in this study were analysed using the SPSS20.0 software package. All measurement data comparisons were expressed as ($\bar{x}$±s) and comparisons between groups were performed using the t-test. The enumeration data were expressed as percentages, and comparisons between groups were performed using the χ^2 test. The ranked data comparison was performed using the Ridit test. The relationships between the chemical constituents of the calculi and the physicochemical properties of urine, protein composition and blood uric acid concentration in patients with upper urinary calculi were found using the Pearson correlation analysis. Results are statistically significant at P<0.05.

Results

Analysis of calculi component in patients with upper urinary calculi

As shown in Table 1, a total of 160 patients with upper urinary calculi that were detected by an infrared spectrum analyser participated in the study: 83 patients (51.88%) had oxalate-based stones, 35 (21.88%) had phosphonate-based stones, 23 (14.38%) had uric acid-based stones and 19 (11.88%) had apatite-based stones.

Type	Male	Female	Total
Oxalate	56	27	83 (51.88%)
Phosphate	24	11	35 (21.88%)
Uric acid	16	7	23 (14.38%)
Apatite	14	5	19 (11.88%)

Table 1: Analysis of calculi component in patients with upper urinary calculi (case, %).

24 h urine physical and chemical properties of all subjects

All patients with upper urinary calculi were divided into an oxalate group, phosphate group, uric acid group or apatite group according to the composition of the stones. Compared with the 24 h urine physicochemical properties of the control group, it was found that the pH of the urine in the phosphate and apatite groups was significantly higher, the urine pH of patients in the uric acid and oxalate groups was significantly lower, and the difference was statistically significant ($P<0.05$). Also, the urine volume of patients in the phosphate, apatite, uric acid and oxalate groups was significantly lower than that in the control group, and the difference was statistically significant ($P<0.05$). Furthermore, the urine phosphate level in the phosphate group was significantly lower than that in the control group, and the difference was statistically significant ($P<0.05$). Finally, the urine magnesium in the uric acid group was significantly higher than that in the control group, but it was significantly lower in the oxalate group ($P<0.05$). For a summary of the results, see Table 2.

Urine protein analysis of all subjects

The levels of β2-MG and α1-MG in the oxalate group were significantly higher than those in the control group, and the difference was statistically significant ($P<0.05$). There was no significant difference in urine protein levels in other groups compared with the control group ($P>0.05$), as shown in Table 3.

Group	Case	pH value	Urine volume	Urine calcium	Urine phosphorus	Urine Magnesium
The control group	30	6.18±0.46	1960±330	4.18±0.33	14.26±0.51	3.58±0.41
Oxalate group	83	5.36±0.31^a	1540±260^a	7.65±0.33^a	13.98±0.48	3.01±0.25^a
Phosphate group	35	6.98±0.77^a	1200±300^a	7.99±0.56^a	10.46±0.63^a	3.45±0.52
Uric acid group	23	4.95±0.45^a	1310±430^a	4.26±0.45	13.66±0.56	3.74±0.38^a
Apatite group	19	6.86±0.69^a	1950±550^a	4.17±0.65	13.75±0.72	3.65±0.55

Table 2: 24 h urine physical and chemical properties of all subjects ($\bar{x}\pm s$).

Note: The superscript a means $P<0.05$ compared with the control group.

Group	Case	α_1-MG	β_2-MG	TRF	mAlb
The control group	30	1.05±0.58	0.19±0.46	0.13±0.06	1.27±0.95
Oxalate group	83	1.56±0.43^a	0.58±0.05^a	0.13±0.04	1.32±0.32
Phosphate group	35	1.11±0.78	0.21±0.13	0.13±0.10	1.34±0.82
Uric acid group	23	1.09±0.45	0.19±0.12	0.10±0.06	1.26±0.52
Apatite group	19	1.05±0.43	0.19±0.13	0.13±0.014	1.29±0.85

Table 3: Urine protein analysis of all subjects ($\bar{x}\pm s$).

Note: The superscript a means $P<0.05$ compared with the control group.

Analysis of uric acid levels in serum and urine of all subjects

As shown in Table 4, the levels of uric acid of urine and serum in the oxalate and uric acid groups were significantly higher than those in the control group, and the differences were statistically significant ($P<0.05$). There was no significant difference in uric acid levels in urine and serum in other groups compared with the control group ($P>0.05$).

Group	Case	Uric acid	
		Serum	Urine
The control group	30	2.23±0.43	3.06±0.38
Oxalate group	83	2.53±0.24^a	3.49±0.24^a
Phosphate group	35	2.21±0.21	2.95±0.43
Uric acid group	23	3.46±0.41^a	4.13±0.35^a
Apatite group	19	2.16±0.21	3.01±0.41

Table 4: Analysis of uric acid levels in serum and urine of all subjects ($\bar{x}\pm s$).

Note: The superscript a means $P<0.05$ compared with the control group.

Correlative analysis of chemical components of calculi and its physicochemical properties, protein composition and blood uric acid concentration

The Pearson correlation analysis showed that β2-MG was positively correlated with oxalate and calcium (P<0.05) and negatively correlated with pH (P<0.01); uric acid was positively correlated with oxalate, urate and calcium (all P<0.05) and negatively correlated with pH (P<0.05); and α1-MG was positively correlated with oxalate and urinary calcium (P<0.05 or <0.01) and negatively correlated with pH (P<0.05).

Indexes	β_2-MG		Uric acid		α_1-MG	
	r	*P*	*r*	*P*	*r*	*P*
Oxalate	0.569	0.023	0.425	0.016	0.058	0.002
Urate	0.336	0.056	0.385	0.022	0.521	0.058
pH value	-0.265	0.001	-0.163	0.041	-0.263	0.021
Urine calcium	0.482	0.043	0.263	0.002	0.165	0.015

Table 5: Correlative analysis of chemical components of calculi and its physicochemical properties, protein composition and blood uric acid concentration.

Discussion

Urolithiasis is one of the most common diseases in urology. The incidence of urolithiasis has increased each year, and the recurrence rate is relatively high[9]. Statistics indicate that the recurrence rate of urinary tract calculi in China is as high as 50%[10]. Urinary lithiasis in China occurs mostly in middle-aged and elderly people and mainly in females[11]. Urolithiasis includes upper urinary tract calculi and lower urinary tract calculi according to the position of the calculi. Due to large amounts of protein intake and changes in diet in recent years, upper urinary calculi are more common[12]. As previously mentioned, a total of 160 patients with upper urinary calculi that were detected by an infrared spectrum analyser participated in the study: 83 patients (51.88%) had oxalate-based stones, 35 (21.88%) had phosphonate-based stones, 23 (14.38%) had uric acid-based stones and 19 (11.88) had apatite-based stones. Thus, oxalate calculi account for the highest proportion of the cases.

All patients with upper urinary calculi were divided into the oxalate, phosphate, uric acid or apatite group according to the composition of the calculi. Compared with the 24 h urine physical and chemical properties of the control group, it was found that the urine pH, urine volume and urine magnesium levels were significantly lower in the other groups, urine calcium levels were significantly higher, and the differences were statistically significant (P<0.05). Calcium is one of the important constituents of oxalate calculi, and the pH value in urine is relatively low, which can reduce the solubility of oxalate and cause the oxalate crystals to precipitate out in the urine, thus leading to the formation of calcium oxalate calculi. Urinary volume and urinary phosphorus levels in the carbonate group were significantly lower than those in the control group, the pH and urinary calcium levels were significantly higher, and the differences were statistically significant (P<0.05), suggesting that the increase of calcium level in urine is closely related to the formation of carbonate and that the higher the pH value in urine, the lower the solubility of phosphorus carbonate, thereby accelerating the formation of carbonate calculi. Patients in the uric acid group had lower urine pH and higher levels of urinary phosphorus, indicating that uric acidification is one of the main reasons for the formation of uric acid calculi, and alkalized urine reduces the formation of uric acid calculi.

α1-MG is a glycoprotein, which is mainly synthesized in the liver and lymphoid tissues. A pathological increase is seen in chronic renal failure such as renal tubular damage[13]. β2-MG is a β light chain of human leukocyte antigen molecules, and its increase can indicate the occurrence of various renal dysfunctions such as acute and chronic nephritis, renal failure, kidney tumours and renal transplant rejection[14]. In this study, the levels of β2-MG and α1-MG in the urine of patients in the oxalate group were significantly higher than those in the control group, and the difference was statistically significant (P<0.05), suggesting that patients with oxalate calculi are more prone to kidney damage.

Uric acid is the final product of purine metabolism and slightly soluble in water, and easily forms crystals. Normal human urine is comprised mainly of urea and contains a small amount of uric acid, which is weakly acidic[15]. In this study, the levels of uric acid in the urine of patients and serum in the oxalate and uric acid groups were significantly higher than those in the control group, and the differences were statistically significant (P<0.05), showing that the increase of uric acid level is closely related to the formation of oxalate calculi and uric acid calculi.

To further understand the chemical components of calculi and the relationships between the physicochemical properties of urine, its protein composition and blood uric acid, a Pearson correlation analysis showed that β2-MG was positively correlated with oxalate and urinary calcium (all P<0.05) and negatively correlated with pH (P<0.01); uric acid was positively correlated with oxalate, urate and urinary calcium (P<0.05) and negatively correlated with pH

(P<0.05); α1-MG was positively correlated with oxalate and urinary calcium (P<0.05 or <0.01) and negatively correlated with pH (P<0.05); and abnormal uric acid and urinary protein levels are closely related to the formation of upper urinary calculi.

In conclusion, oxalate calculi are the most common type of upper urinary calculi. Patients with upper urinary calculi have abnormal levels of uric acid and urinary proteins, which have a relationship with the chemical components of calculi and the physicochemical properties of urine.

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Acknowledgement:
This study was supported by the Clinical Research Plan of SHDC (No.16CR4006A).

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