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Infrared Spectroscopy analysis of urinary tract stones collected from 4 community hospitals in Udorn Thani province

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การวิเคราะห์องค์ประกอบของก้อนนิ่วทางเดินปัสสาวะที่ได้จากโรงพยาบาลชุมชน 4 แห่ง ของจังหวัดอุดรธานี ด้วยวิธี Infrared Spectroscopy

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ได้ทำการวิเคราะห์องค์ประกอบของก้อนนิ่วทางเดินปัสสาวะจำนวน 114 ก้อนด้วยวิธี infrared spectroscopy ก้อนนิ่วเหล่านี้ได้จากการผ่าตัดนิ่วในโรงพยาบาลชุมชน 4 แห่งของจังหวัดอุดรธานี ระหว่างปี 2531 ถึง 2532 ประกอบด้วย นิ่วในผู้ใหญ่ 95 ราย (ชาย/หญิง = 2) และในเด็ก 25 ราย (ชาย/หญิง = 3) องค์ประกอบที่พบได้บ่อยที่สุดในนิ่วจากผู้ใหญ่คือแคลเซียมออคซาเลต ซึ่งพบในนิ่วจากทางเดินปัสสาวะส่วนบน (UUT) และส่วนล่าง (LUT) ร้อยละ 96 และ 80 ตามลำดับ ในกรณีของนิ่วจากเด็กนั้น พบแคลเซียมออคซาเลตในนิ่วจาก UUT ทุกก้อน (ร้อยละ 100) แต่นิ่วจาก LUT กลับพบว่า ยูริค แอซิด/ยูเรต เป็นองค์ประกอบที่พบได้บ่อยที่สุด (ร้อยละ 64) เมื่อทำการแบ่งก้อนนิ่ว ตามองค์ประกอบหลักสามารถแบ่งออกได้เป็น 4 ชนิด คือ แคลเซียมออคซาเลต แคลเซียมฟอสเฟต แมกนีเซียมแอมโมเนียมฟอสเฟต และยูริค แอซิด/ยูเรต นิ่วชนิดที่พบมากที่สุดในทั้ง UUT และ LUT ของผู้ใหญ่ และใน UUT ของเด็ก คือ แคลเซียมออคซาเลต ส่วนใน LUT ของเด็กชนิดที่พบมากที่สุด คือ ยูริคแอซิด/ยูเรต ข้อมูลการศึกษาของเราครั้งนี้แม้จะเป็นการศึกษาเชิงคุณภาพ แต่ผลที่ได้โดยทั่ว ๆ ไป ก็คล้ายกับการวิเคราะห์โดยวิธี wet chemical เชิงกึ่งคุณภาพ และปริมาณ (semi-quantitative) ซึ่งรายงานโดยคนอื่น

Abstract

A series of 114 urinary stones collected from 4 community hospitals were analyzed for chemical composition by infrared spectroscopy. These stones were surgically removed from 95 adults (male/female =2) and 25 children (boy/girl = 3) during the period of 1988. Calcium oxalate was the most frequent component found in adult stones of both upper (96%) and lower (80%) urinary tracts (UUT and LUT). Of the childhood stones, while calcium oxalate was seen in all 5 UUT stones (100%), uric acid/urate was the most common component of stones from LUT (64%). The calculi were classified according to their main components into 4 type; calcium oxalate, calcium phosphate, magnesium ammonium phosphate and uric acid/urate. The most predominant stone type found in adult UUT and LUT and in childhood UUT was calcium oxalate whereas uric acid/urate was the main type of childhood LUT stones. Although our data was obtained using a qualitative approach, in general, our observations agree with those semi-quantitative wet chemical methods reported by others in this region.

INTRODUCTION

A number of metabolic and environmental factors have been described as the causes of calculi formation in the urinary tract (1). One approach to understanding these disorders, from which the patients suffer, is the analysis of chemical compoition of the formed stones. Knowledge of urolith chemistry is a prerequisite underlying any effective treatment or measure to prevent recurrence (2,3). Numerous physical and chemical techniques have been used for the analysis of stone composition. Among them wet chemical analysis, the oldest technique, is still widely used due to its straight forward protocol and since it does not require expensive specialized equipments (4). Unfortunately chemical methods can only determine radicals and ions, and therefore cannot differentiate between similar crystalline entities (e.g.uric acid vs. uric acid dihydrate or apatite vs. brushite or whewellite vs. weddellite). Furthermore, these methods require a relatively large quantity of test materials. Infrared (IR) spectroscopy is a technique for urolith analysis. Although there is a wide divergence of expert opinion on the value of IR spectroscopy for urolith analysis (5,6), some modern laboratories are now analyzing calculi by the IR apectroscopy rather than by the classical wet chemical technique (5,7,8). The advantages of the IR spectroscopy are greater speed and specificity, uniform sensitivity to all components, and decreased quantities of test substances (7-10).

In 1983 we reported the results of stone analysis using the classical wet chemical method (11). Since an IR spectrophotometer is now available in our laboratory, the objective of this paper is to examine the chemical composition of urinary stones by IR spectroscopy. The stone samples were obtained from community hospitals.

Materials and Methods

Urinary tract stones were collected from patients undergoing surgery in 4 community hospitals (Pen, Sang Com, Nam Som and Ban Dung) of Udorn Thani province during the period 1988 to 1989. One hundred and fourteen stones from patients with complete hospital records were analyzed in this study.

Prior to analysis, the stones were washed with distilled water to remove blood and attached tissues and oven-dried at 70 °C overnight. These stones were then cut approximately at the middle with a fine saw. The powder resulting from the cutting process was often sufficient for the IR analysis.

IR spectra were obtained using the KBr pellet technique. These were made by mixing the calculi powder of about 1 percent with KBr, grinding together in an Agate mortar and pressing the resulting mixture into disks (with a force of $2.36x10^{-7}N$) The disks were run on Shimadzu IR spectrophotometer, model IR-460 (Shimadzu Corporation, Spectrophotometric Instruments Plant, Analytical Instruments Division, Kyoto, Japan) which covered the range 4000 to 500 cm^{-1}. Various principles of interpreting IR spectra of stones have been suggested (7,10). In our case, spectra of the unknowns were compared with spectra of the known pure substances as described by Corns (8).

Results

The patients were comprised as follows: 95 adults (16 years and above ;60 males and 35 females) and 19 children (15 years and below; 14 boys and 5 girls). The majority of stones found among children were in the lower urinary tract (LUT) whereas stones of adult were predominantly located in the upper urinary tract (UUT) (Table 1).

Calcium oxalate was the most frequent mineral component found in stones of adult both from UUT (96%) and LUT (80%) as shown in Table 2. In the case of children, although all 5 UUT stones (100%) examined contained calcium oxalate, the most common component of LUT stones was uric acid/urate (78%) (Table 3). Second to calcium oxalate was hydroxy apatite, a form of calcium phosphate, which were detected in 70%, 68% and 40% of stones from UUT, and LUT of adults, and UUT of children, respectively. However, carbonate apatite and magnesium ammonium phosphate hexahydrate, were observed only in amall number of stones from both groups of the subjects. Taking the number of components in a stone into account, most of adult stones had a mixture of 2 components : calcium oxalate and hydroxy apatite (Table 4). Stones in childhood, however, were composed mainly of a single component: calcium oxalate for UUT and uric acid for LUT.

Since IR analysis is primarily a qualitative procedure, the peak heights of spectra have only and approximate to concentration (12). Thus classification of our calculi would differ from those wet chemical methods described by Hodgkinson (13). In this report the calculi were divided into 4 types according to their main components: calcium oxalate, calcium phosphate, magnesium ammonium phosphate and uric acid/urate. The results showed that calcium oxalate stones were the predominant type in both UUT (76%) and LUT (64%) of adults (Table 4). This was also the case for childhood UUT stones (80%) as shown in Table 5. Of stone from childhood LUT, however the most common type was uric acid/urate (64%). The second most abundant type fo stones was calcium phosphate which was found common to both locations of the adult urinay tract. In the case of magnesium ammonium phosphate stones, this could be regarded as a minor stone type observed in this study since only a few of them were presented. Cystine and brushite were not detected in any stone studied.

DISCUSSION

Knowledge of the chemical nature of a stone is an important guide to success in management of the disease (2,3). Information on chemical analysis of stones from patients in the northeast region of Thailand is still limited and not up to date. Sakornmonkol et al (14) presented the first qualitative wet chemical method in an analysis of stones from Ubol provincial hospital in 1962 and by means of optical crystallography, X-ray diffraction photography and Geiger Muller counter X-ray diffractometer, Gershoff et al (15) reported the analysis of stone collected from Khon Kaen provincial hospital in 1963. More update data was studied by Prasongwatana and co-workers (11) using the conventional wet chemical method. IR spectroscopy has been claimed to have more advantages in many aspects over the wet chemical method in analysis of stone composition (7-10). The technique needs only a small amount of test sample, the result is more uniformly sensitive to all components and has greater reproducibility. All in all, it permits a positive identification of most of the components present in analyzed calculi.

Data from many epidemiological studies (16-18) suggest that the pattern of urinary tract stone disease in this region now has changed, with a steadily increasing prevalence of UUT stones, as is the case in many in dustrialized countries (19,20). Since all of these studies were carried out in central or provincial hospitals one might argue that this change of stone pattern is probably due to the referral system which is usually in the direction of community to provincial hospitals. Our present data from 4 community hospitals clearly showed that, for adult, the stones from UUT were three times more prevalence than from LUT (Table 1). This finding was similar to that reported by Raiyawa and co-workers who also studied urinary stone disease in a nearby community hospital (18) Chutikorn and associates (29) reported experiences at Ubol provincial hospital during the period 1956 to 1962. Of the children with calculi, about 91 percent had LUT stones. In this report we found that as much as one-third of childhood stones were from UUT (Table 1). Thus the childhood stone pattern seemed to change in the same manner as that of adult's. This finding was similar to those observed in northern Thailand (21).

Our data on the composition of childhood stones demonstrated that uric acid/urate was the major component of the stones from UUT and that they existed more commonly in the pure state (Table 5). The findings confirmed previous studies by others although different techniques were employed (11,15,20). High prevalence of uric acid/urate stone in childhood was a unique feature of endemic bladder stone in the Middle and the Far East. The stone was once also common in Britain during the late-18th-and early – 9th-century and then disappeared during the World War I (20). The disappearance of the disease is said to be due to a variety of factors but mainly to the improvement of nutrition, with increased dietary phosphate and protein intake (20,22). Our findings suggest that the problen of an unbalanced diet as a cause of LUT stones, similar to the past in area studied, still existed. Taking childhood UUT stones into account, calcium oxalate was the main component and the most frequent type of stone found. This composition is, infact, comparable to that of adult UUT stones suggestion that factors underlying stone formation are probably similar for UUT stones of both adults and children.

The chemical composition of adult stones resembled data obtained from Ubol (20) and Khon Kaen (11,15). Adult stones consisted mainly of calcium oxalate mixed with a variable amount of calcium phosphate in the form of hydroxy apatite (Table 4). Furthermore, stones containing calcium oxalate were the most common single component stones found in this investigation. The results were also similar to reports from elsewhere (23,24). Magnesium ammonium phosphate hexahydrate and carbonate apatite calculi are reported to occur only in the presence of infection (2). These types of stone found less frequently than as reported by Prasongwatana and co-workers (11). Since our study was not a case selection, the lower prevalence of infective stones should be partly due to a less sensitivity of IR spectroscopy technique in determining of these 2 components (7,8,12,25).

The presence of calcium phosphate stones, mainly hydroxy apatite but often with some calcium oxalate would suggest renal tubular acidosis or hyperparathyroidism (26). Study of blood and urine biochemistry of renal stone-formers, Sriboonlue and co-workers have shown that hyperparathyroidism was very unlikely to be a contributing factor for the formation of this stone type (27). On the other hand, distal renal tubular acidosis was reported to be endemic in northeast Thailand (28). Our data showed that most calcium phosphate calculi of adults had hydroxyapatite as the main component and were always mixed with calcium oxalate (Table 4). Since the indication of infection in adult urinary tract (the presence of carbonate apatite and magnesium ammonium phosphate haxahydrate as the outcomes of infection) was rare, the association of calcium phosphate stones with renal tubular acidosis was very likely. We suggest that this ralationship should have been further investigated in order to achieve a better understanding of the disease. Furthermore, our data suggests that stones composed of cystine and brushite are not a health problem of people in the northeast. Similar observation have also been made by Gershoff and co-workers (15) and Prasongwatana and co-workers (11).

Table 1 Demographic data

	Adult	children
Number of patients	95	19
Age in years (X ± SD)	40.8 ± 15.2	7.7 ± 3.6
Male : female	60:35	14:5
Upper urinary tract (UUT) stones	70 (73%)	5 (26%)
Lower urinary tract (LUT) stones	25 (27%)	14 (74%)

Table 2 Number of adult stones containing each chemical component

Chemical Names	No. of Stones (%)	
	UUT (n=70)	LUT (n=25)
1. Calcium oxalate	67 (96)	20 (80)
2. Hydroxy apatite	48 (70)	17 (68)
3. Carbonate apatite	1 (1)	1 (4)
4. Magnesium ammonium phosphate hexahydrate	2 (3)	-
5. Uric acid/urate	5 (7)	4 (16)

Table 3 Number of childhood stones containing each chemical component

Chemical Names	No. of Stones (%)	
	UUT (n=5)	LUT (n=14)
1. Calcium oxalate	5 (100)	4 (28)
2. Hydroxy apatite	2 (40)	-
3. Carbonate apatite	-	-
4. Magnesium ammonium phosphate hexahydrate	-	2 (14)
5. Uric acid/urate	1 (20)	11 (78)

Table 4 Types of adult stone classifield according to main components

Stone Types	Location of Stones			
	UUT (n=70)		LUT (n=25)	
1. Calcium oxalate (CaOx), total	53	(76%)	16	(64%)
CaOx only	15		3	
CaOx + HAp*	35		13	
CaOx + U	2		-	
CaOx + HAp + U	2		-	
2. Calcium Phosphate (Cap), total	13	(18%)	5	(20%)
HAp + CaOx	12		4	
CAp**	-		1	
CAp + CaOx	1		-	
3. Magnesium ammonium phosphate hexahydrate (MAP)	2	(3%)		
4. Uric acid/Urate (U), total	2	(3%)	4	(16%)
U	-		3	
U + CaOx	2		1	

**HAp = hydroxy apatite*

***CAp = carbonate apatite*

Table 5 Types of childhood stone classified according to their main components.

Stone Types	Location of Stones			
	UUT (n=5)		LUT (n=14)	
1. Calcium oxalate (CaOx), total	4	(80%)	4	(29%)
CaOx only	2		2	
CaOx + HAp*	1		-	
CaOx + U	1		2	
2. Calcium phosphate (CaP), total	1	(20%)	-	
HAp + CaOx	1		-	
3. Magnesium ammonium phosphate	-		1	(7%)
4. Uric acid/urate (U), total	-		9	(64%)
U	-		8	
U + MAP	-		1	

**HAp = hydroxy apatite*

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