Association Between Dyslipidemia And Kidney Stone Risk In Sudanese Patients

Rufayda O. Musa, Omer F. Idris

Abstract: This study is conducted to assess serum of lipid profile level (TC, TAG, HDL, LDL) in kidney stone patients and correlate between the distributions of the lipid profile level, it is descriptive study cross-sectional. The study was conduct during 1stMay to 30ty September 2014 and it was in Khartoum state hospital (Urology units; Alribat hospital, Military hospital and Ibn sina hospital). Hundred subjects were included in the study, sixty patients with kidney stone and forty healthy people considered control group. This study includes both gender, male were found to have high incidence (63%) compared with the female (37%). Moreover the incidence of disease was found to be higher in the patients above 40 years old. The results showed that the patients concentration of TC and LDL than control with Mean ± SD 180.66 ± 66.46 in TC and 173.20 ± 61.40 in LDL and lower concentration of TAG and HDL compare with control with Mean ± SD in TAG was 230.86 ± 103.87 and in HDL 24.65 ± 9.95, the results of TC, LDL and HDL were significant with P value of (0.00), with in case TAG the results were not significant (with P value of 0.549). It was noticed that gender has a pronounced effect in disease development. Specific alteration in patient's lipid profile may reflect on physicochemistry properties of urine, this will increase the risk of developing stones.

Keywords: Lipid profile; Kidney stones; Dyslipidemia; Stone nucleation Biochemical analysis; Statistical analysis.

Introduction

Kidney stones occur when there are too many waste products or not enough fluid to flush those waste products out [1]. Stones can be composed of either single substances or salts, which are two or more substances that join together. Uric acid and cystine are single substances. On the other hand, calcium stones and struvite stones are each composed of salts. Seventy five percent of the kidney stones that reach the stone analysis laboratory contain calcium oxalate as the predominant mineral [2]. When calcium combines with another mineral, insoluble crystals form which are commonly either calcium oxalate or calcium phosphate in composition. [3] About ten percent are composed of uric acid. It results from the metabolism of the purines, which are found in all animal protein and many seeds and plants. Therefore, all meat breaks down into uric acid. Ten percent are struvite, or infected stones, stones are among the most difficult and dangerous problems in stone disease because of the potential of life-threatening complications from infection. These stones are found mainly in women with recurring urinary infections, struvite stones are often called triple phosphate stones because they contain three different elements: magnesium, ammonium, and calcium. Struvite stones are caused by certain bacteria, the most common of which is called Proteus, Cystine stones result from an uncommon hereditary metabolic disorder and account for less than 1% of stone cases. [2] Prevalence of Stone type Calcium oxalate stones (with or without phosphate) was the most frequent (68% of stones). The remainders include uric acid 17%, infection stones 12% and pure calcium phosphate stones 3 % [4].Nucleation is the formation of a solid crystal phase in a solution. It is an essential step in renal stone formation the term super saturation refers to a solution that contains more of the dissolved material than could be dissolved by the solvent under normal circumstances. Crystal aggregation and attachment of crystals or aggregates to an alternative nidus such as renal epithelial cells are critical processes in stone formation [5].Specific alterations in the patient lipid profiles may portend unique aberrations in urine physicochemistry and stone risk [6]. Elevated BMI (body mass index), hypercholesterolemia, and hyperlipidemia, which are leading components of metabolic syndrome, may be associated with different types of urinary stone formation, [7]. The effects of acidic biopolymers and lipid membranes on nucleation, growth and aggregation of calcium oxalate (CaOx) crystals in an artificial urinary environment. [8]. [9] Noted that more than 30% of their cohort was characterized as dyslipidemic (defined by the use of a cholesterol lowering medication). Of these patients with dyslipidemia, nearly 70% had calcium oxalate stones and 15% had uric acid stones. Similarly found that total cholesterol levels were significantly higher in stone formers compared with patients who do not form stones. Stone disease is much more common in men than in women. The ratio, however, of male to female stone formers varies widely in populations, depending on diet and other factors, [2]. Baker et al, [4] found that 70% of all stones analyzed were from men. Men were at greater risk of producing calcium oxalate stones (73% were in men) and uric acid stones (79% were in men). Women were at greater risk of infection stones (58% occur in women).Also he found that the peak age for the development of calcium oxalate stones was between 50 and 60 years. Uric acid stones tended to occur in an older population with an average age of 60–65 years. Infection stones, however, occurred in younger people, most commonly in women between the ages of 20 and 55 years.

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Material and methods

Ethical consideration

This study was approved term the committed of Alneelain University Faculty of post Graduate Studies Department of Biochemistry and Molecular Biology, all individual shared in the study was informed by the aim of the study and verbal informed console was obtained from each subject.

Blood sample and processing

Three ml venous bloods were taken from kidney stone patient, another three ml from controls group. The samples were collected in tube (Plain tube). Serum was separated from RBC by centrifugation at 2000 rpm for 10 minutes. Two ml serum was use for estimation of TC, TAG and HDL and by using calculation to give LDL. The samples were stored at -20oc. The process of measurement was conducted in biochemistry lab.

Cholesterol measurement

Principle of the method

Free and esterifies cholesterol in the sample originates, by means of coupled reaction described below, a colored complex that can be measured by spectrophotometer.

1- Cholesterol ester + H2O $\xrightarrow{\text{Cholesterol esterase}}$ Cholesterol + Fatty acid
2- Cholesterol+1/2O2 +H2O $\xrightarrow{\text{Cholesterol oxidase}}$ Cholestenone +H2O2
3- 2H2O2 +Aminoantipyrine+Phenol $\xrightarrow{\text{Peroxidase}}$ Quinoneimine + 4H2O

Procedure

Reagents brought to room temperature, 10 µl was added into 1 ml labeled test tube (standard, samples and blank) then mixed thoroughly and incubated for 10 min at room temperature. The absorbance of standard and samples were measured at 520 nm against blank using Biosystem BTS-310 spectrophotometer.

Triglyceride measurement

Principle of the method

Triglycerides in the sample originates, by means of the coupled reaction described below, a coloured complex that can be measured by spectrophotometry

1- Triglycerides + H2O $\xrightarrow{\text{lipase}}$ glycerol + fatty acids
2- Glycerol + ATP $\xrightarrow{\text{glycerol kinase}}$ glycerol - 3 - P + ADP
3- glycerol - 3 – P + O2 $\xrightarrow{\text{G - 3 - P – oxidase}}$ Dihidroxyacetone – P + H2O2
4- 2 H2O2 + 4 – Aminoantipyrine + 4 -Chlorophenol $\xrightarrow{\text{Peroxidase}}$ Quinoneimine +4H2O

Procedure

Reagents brought to room temperature, 10 µl was added into 1 ml labeled test tube (standard, samples and blank) then mixed thoroughly and incubated for 10 min at room temperature. The absorbance of standard and samples were measured at 520 nm against blank using Biosystem BTS-310 spectrophotometer.

High-density lipoprotein measurement

Principle of the method

Very low density lipoproteins (VLDL) and low density lipoproteins (LDL) in the sample precipitate with phosophotungstate and magnesium ions. The supernmatant contains high density lipoproteins (HDL). The HDL cholesterol is then spectrophotometrically measured by means of the coupled reaction describrd below.

Cholesterol ester + H2O $\xrightarrow{\text{Cholesterol esterase}}$ Cholesterol + Fatty acid
Cholesterol+1/2O2 +H2O $\xrightarrow{\text{Cholesterol oxidase}}$ Cholestenone +H2O2
2H2O2 +Aminoantipyrine+Phenol $\xrightarrow{\text{Peroxidase}}$ Quinoneimine + 4H2O

Precipitation Procedure

Sample (100 µl) was added to 250 µl precipitation reagents for HDL, then mixed thoroughly and was incubated for 10 minutes at room temperature, the mixture was centrifuged at 4000 r.p.m for 10 minutes then supernatant carefully collected and used for estimation of HDL.

Procedure

Reagents brought to room temperature, 100 µl was added into 1 ml labeled test tube (standard, samples and blank) then mixed thoroughly and incubated for 30min at (16-25oC) or for 10 min at 37oC ,The absorbance of standard and samples were measured at 520 nm against blank using Biosystem BTS-310 spectrophotometer.

Low-density lipoprotein calculation

The result of LDL calculated according to the fowling formula

$$\frac{(\text{Triglyceride} + \text{HDL}) - \text{Cholesterol}}{5} = \text{LDL mg/ dL.}$$

RESULTS AND DISCUSSION

The minimum and the maximum age of the participants were 1-90 years old. This study included both gender, the male and the female in the case group reported (63%) and (37%) respectively. where the control group include (58%) male and (43%) female. The mean ± SD of patient's ages was 41.75± 2.03 and for the control was 42.95±1.67.

Table 1. *The distribution of renal stone in gender*

Table 1. *Show the distribution of renal stone in gender shows that males (63%) had higher incidence of forming renal stone than females*

Gender	Frequency	Percentage
Male	38	63%
Female	22	37%

The result in Table1 showed that majority (63%) of male had higher incidence of forming renal stone than females (37%).

Table 2. *The descriptive analysis of lipid profile concentration.*

		Case	Control
TC mg/dl	Mean ± SD	180.66 ± 66.46	104.15 ± 44.61
	Median(Minimum- Maximum)	(20 - 334)	(8 - 213)
TAG mg/dl	Mean ± SD	230.86 ± 103.87	243.45 ± 100.67
	Median(Minimum- Maximum)	(74 - 588)	(75 - 546)
HDL mg/dl	Mean ± SD	24.65 ± 9.95	50.07 ± 21.08
	Median(Minimum- Maximum)	(9 - 48)	(8 - 88)
LDL mg/dl	Mean ± SD	173.20 ± 61.40	111.87 ± 45.14
	Median(Minimum- Maximum)	(38 - 397)	(23 - 197)

***SD (Stander Deviation)**

Table 2. *Show the descriptive analysis of lipid profile concentration.*

The result in Table1 showed that the comparison in Mean ± SD and Median(Minimum- Maximum) between case (patients with kidney stone disease) and control, the patients have high total cholesterol and LDL concentration compare with control but the in other hand it showed also the patients had low concentration in TAG and HDL.

Table 3. *comparison of the mean, and stander error of mean between the cases and the controls and the significant differences*

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Parameters	Control group 40 individuals	Case group 60 individuals	P value
Triacylglycerol	243 ± 15	230 ± 13	0.549
Cholesterol	104 ± 7	180 ± 8*	0.000*
LDL	111 ± 7	173 ± 7*	0.000*
HDL	50 ± 3	24 ± 1*	0.000*

Mean ± Std. Error;* = Significant differences; P < 0.05 consider to be significant

Used SPSS 16 .Independent t-test and one way a nova test there are significal difference in cholesterol, LDL and HDL between the patients with kidney stone disease and controls (people without kidney stone disease), but there no significal difference in triacylglycerols.

Figure1. *The frequency of age group distribution of renal stone.*

Figure1. *Shows the frequency of age group distribution in renal stone patients, (above 40) age group had high incidence 58%.*

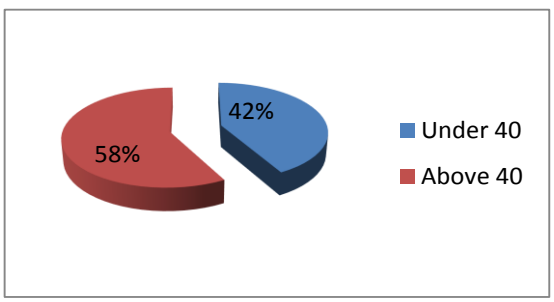

Figure1. Showed that the frequency of age group distribution in renal stone patients, (above 40) age group had high incidence 58%. The results of our study, indicated that the male had high incidence of kidney stones than female, it is similar to other studies that also reported occurrence of kidney stone high in male than female [2,4] have also reported occurrence of kidney stones is more in male than female. It was observed from our result high incidence of stone disease occur in the group (above forty years), [4] also reported in their study that kidney stones are uncommon before the age of 20 years. There are significal diffrence in cholestrol, LDL and HDL between the patients with kidney stone disease and controls, but there no significal diffrence in triacylglycerols.

There is a link between distribution of lipid profile level and kidney stone risk especially in high level of cholesterol and LDL and in low level of HDL, but there is no relation in case of triacylglycerols level [6] reported that the specific alterations in the patients lipid profiles may portend unique aberrations in urine physicochemistry and stone risk, [7] hypercholesterolemia may be associated with different types of urinary stone formation. [8] Suggested in his study that the effects of acidic biopolymers and lipid membranes on nucleation, growth and aggregation of calcium oxalate (CaOx) crystals, indicate that the of calcium oxalate stones (with or without phosphate) were the most frequent (68% of stones) and [4] reported that 70% of all stones analyzed were from men. Men were at greater risk of producing calcium oxalate stones (73% were in men).

CONCLUSION

This study was designed to investigate the differences between serum TC, TAG, HDL and LDL level in 60 patients suffering from kidney stone disease and 40 normal individual, the indepented sample t-test showed significal difference in TC, HDL and LDL concentration (P < 0.05), The cholesterol may have role in increase the viscosity of urine that leads to increase the nucleation process of stone formation, the analysis of serum lipid profile can be helpful for understanding the risk of stones formation and this knowledge may be helpful in the eliminating the cause leading to further recurrence of stone formation.

RECOMMENDATION

To prevent renal stone formation, decrease the fat diet or any dietary food or habits that may increase the cholesterol and LDL level and decrease the HDL level and take atorvastatin medication or any lowering cholesterol treatment when

needed.

REFERENCES

[1] Rodman, J.S., Seidman, C and Jones, R (1996). No More Kidney Stones. Somerset, NJ: John Wiley and Sons, Inc. p. 3-8.

[2] John S. Rodman, R. Ernest Sosa, Cynthia Seidman, Rory Jones (2007). No More Kidney Stone, United States, Hoboken, New Jersey, page 20-30.

[3] Raymond Ko (2009). Kidney Stone Clinic Complex Stone & Laser Endourologist Laparoendoscopic Surgeon, Kidney Stone Clinic Nepean, Westmead, and Macquarie Street ,www.stoneclinic.com.au ,page 1-4

[4] Baker PW, Coyle P, Bais R, Rofe AM (1993). Influence of season, age and sex on renal stone formation in South Australia. Med J Aust.159,390–392.

[5] Doddametikurke Ramegowda Basavaraj, Chandra Shekhar Biyani ,Anthony J. Browning , Jon J. Cartledge (2007). The Role of Urinary Kidney Stone Inhibitors and Promoters in the Pathogenesis of Calcium Containing Renal Stones.2007 Published by Elsevier B.V. on behalf of European Association of Urology and European Board of Urology doi:10.1016/j.eeus.2007.03.002. www.sciencedirect.com.

[6] Torricelli FC, De SK, Gebreselassie S, Li I, Sarkissian C, Monga M (2014). Dyslipidemia and kidney stone risk; J Urol. 2014 Mar;191(3):667-72. doi: 10.1016/j.juro.2013.09.022. Epub 2013 Sep 18

[7] Inci M, Demirtas A, Sarli B (2012). Association between body mass index, lipid profiles, and types of urinary stones. Ren Fail;34(9):1140–3.)

[8] Timing: Fiscal (2001). Project Start 15-JUL-; Project End 30-JUN-2005, Principal Investigator & Institution: Gower, Laurie A. Assistant Professor; Materials Science and Engr; University of Florida Gainesville, FL 32611

[9] Kadlec AO, Greco K, Fridirici ZC (2012). Metabolic syndrome and urinary stone composition: what factors matter most Urology;80(4):805–10.)