Original Article

Assessment of Serum Cathepsin k and Lipid Profile in Chronic Coronary Syndrome Patients

Alyaa Mohammed Abdul Hasan, Mufeed Jalil Ewadh, Ameer Ahmed A. Aljubawii[1]

Department of Biochemistry, [1]Department of Medicine, College of Medicine, University of Babylon, Hilla, Iraq

Abstract

Background: The most affected public illness around the world in both industrialized and unindustrialized countries is chronic coronary syndrome (CCS). In a medical context, the measurement of lipid profile in the blood is considered as one of the most common diagnostic techniques. In addition, there is a correlation between increased level of cathepsin k (CatK) and CCS, and thus cathepsin is considered a useful biomarker for CCS. **Objective:** For the assessment of CatK, triglyceride (TG), cholesterol, low density lipoprotein (LDL), very-low-density lipoprotein (VLDL), and high density lipoprotein (HDL) level and to examine the probable relation of them with CCS in Babylon province. **Materials and Methods:** CatK, TG, cholesterol, LDL, VLDL, and HDL were estimated in 100 subjects; 50 patients with CCS and 50 healthy subjects participated in this study. Patients and control groups with an age ranged above 40 years. The CatK level was assessed by sandwich-ELISA technique whereas the level of TG, cholesterol, and HDL was assessed in serum by enzymatic colorimetric method. Also, cholesterol-LDL was measured by using Friedewald equation. **Results:** Serum level of CatK displayed a significant increase in CCS patients ($P \leq 0.01$) compared with control group, whereas serum cholesterol-HDL level significantly decreased ($P = 0.001$). Also serum levels of cholesterol and cholesterol-LDL a significant increase ($P \leq 0.001$), ($P = 0.00$) compared with control group. In contrast, the current study observed non-significant change in serum TG and VLDL ($P = 0.45$), ($P = 0.71$) respectively in CCS patients. **Conclusion:** Circulating CatK is a good biomarker for CCS disorders and that higher levels of CatK and lipid profile are closely related to the presence of CCS among CCS patients.

Keywords: CatK, cholesterol, chronic coronary syndrome, HDL, triglyceride

Introduction

One of the most common cardiovascular disorders impacting people worldwide is coronary artery disease (CAD). Lifestyle, environmental variables, and genetic factors all increase the chance of getting cardiovascular disease. It is the biggest cause of death in both industrialized and underdeveloped countries, according to research.[1,2] Chronic coronary syndrome (CCS) is a phrase used to describe CAD as having a chronic, progressive course that can be changed, stabilized, or improved with medication, lifestyle changes, and coronary revascularization. It has been used to replace the phrase "stable CAD," which was previously used.[3]

Cathepsin K (CatK), one of the most powerful mammalian collagenases, was initially discovered in macrophages. Cysteinyl cathepsins have been demonstrated to locate in lysosomes and endosomes and to work there to breakdown undesirable intracellular or endocytosed proteins. The ability of these vascular cells and macrophages to release CatK, which breaks down type I collagen and elastin, is consistent with these biochemical data. CatK deficiency has been found to lessen diet-induced atherosclerotic lesion formation. These findings revealed a link between catK levels and cardiovascular disease based on atherosclerosis.[4] Recent studies have shown that the cardiovascular cells and inflammatory cells produce the most significant and abundant protease, cysteinyl CatK, and that this has consequences for atherosclerosis-related

Access this article online

Quick Response Code:

Website: https://journals.lww.com/mjby

DOI: 10.4103/MJBL.MJBL_321_23

Address for correspondence: Mrs. Alyaa Mohammed Abdul Hasan, Department of Biochemistry, College of Medicine, University of Babylon, Hillah, Iraq.
E-mail: alyaamohammed1994@gmail.com

Submission: 18-Mar-2023 **Accepted:** 31-Mar-2023 **Published:** 17-Jul-2024

How to cite this article: Abdul Hasan AM, Ewadh MJ, Aljubawii AA. Assessment of serum cathepsin k and lipid profile in chronic coronary syndrome patients. Med J Babylon 2024;21:280-4.

cardiovascular illness. Animals and humans with injury-related neointimal lesions and atherosclerotic plaques had higher CatK protein levels than normal.[5]

In a medical context the measurement of lipid profile in the blood is considered as one of the most common diagnostic techniques.[6]

All cells in the body are capable of producing cholesterol, and the rate-limiting step is catalyzed by the enzyme 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA) reductase. When bound to statins, HMG-CoA reductase is inactivated, and transcription is decreased when intracellular cholesterol levels are high. Being a crucial component of cell membranes, cholesterol serves as a raw material for the production of bile acids and steroid hormones.[7]

Lipoproteins carry cholesterol in blood plasma because of its hydrophobicity. As previously indicated, an important risk factor and cause of cardiovascular disease is the increased serum high density lipoprotein (LDL) cholesterol.[8]

Coronary heart disease (CHD) and high density lipoprotein (HDL) cholesterol have the opposite relationships,[9] Despite the fact that it has never been demonstrated, increasing HDL reduces the risk of cardiovascular disease in people.[10]

A group of glycerol and three fatty acids are listed in the carbon hydroxyl groups of glycerol to form a triglyceride (TG). Each TG may comprise three different fatty acids or a blend of different fatty acids.[11]

Very-low-density lipoprotein (VLDL) and chylomicrons, respectively, carry TGs from the liver and integuments to the peripheral tissues, where they are used to supply energy to the tissues.[12]

Because TGs do not accumulate in foam cells, the association of plasma TGs and Atherosclerotic Cardiovascular Disease these may be due to the remaining lipoproteins. The residues can accumulate in the arterials lining, where it can be ingested by macrophages, promoting the formation of foam cells, and ultimately, the formation of a fatty line that leads to the development of plaque.[13,14]

In accordance with anti-atherosclerotic properties such reverse cholesterol transport and lipid homeostasis, high density lipoprotein has been demonstrated to guard against atherosclerosis and other vascular disorders.[15]

In this study, we aimed to review the recent discoveries on the association between CatK and lipid profile with CCS.

Materials and Methods

Study design

Case–control study is the design of this revision.

Patients and control

This revision study comprised 100 subjects. 50 patients with CCS participated in this study. The complete history from all patients was recorded, which include: age, body mass index, gender, family history, and smoking behaviors.

The second group comprises 50 apparently healthy persons (control group).

Patients with pregnancy, concomitant acute or chronic inflammatory disorders and patients with malignancy were excluded from the revision.

Patients and control groups were aged above 40 years. For the statistical analysis, SPSS version 20.0 was used. All the results as mean ± SD, with significant effect recorded when P values less than 0.05.

1. Determination of serum CatK concentration.
Enzyme-linked immunosorbent assay is what this kit is for (ELISA). Human CTSK antibody (Elabscience Company, Wuhan, China) has been pre-coated on the plate. The sample's CTSK is introduced, and it binds to the antibodies that have been coated on the wells. Human CTSK antibody that has been biotinylated is then added, and it binds to CTSK in the sample. The biotinylated CTSK antibody is then bound by the addition of streptavidin-HRP. After incubation, a washing process removes any unbound streptavidin-HRP. The amount of Human CTSK is then correlated with the development of color in the substrate solution. By adding an acidic stop solution, the process is stopped, and absorbance is measured at 450 nm.
2. Total cholesterol concentration in serum were measured by enzymatic colorimetric method.
3. TG was measured by Fossati and Prencipe method related with Trinder reaction.
4. HDL-cholesterol was measured by magnesium chloride and phosphotungestic acid act on precipitating HDL, chylomicron and VLDL were extracted from the samples. Total cholesterol reagent was used to test the HDL-cholesterol that was found in the supernatant after centrifugation.
5. LDL-cholesterol was measured by using the Friedewald equation as follows.

$$LDL_{cholesterol}\,(mmol/L)=T._{cholesterol}\text{-}HDL_{cholesterol}\text{-}VLDL_{cholesterol}\left(\frac{Triglyceride}{2.22}\right)$$

Ethical approval

Based on the ethical standards laid out in the Helsinki Declaration, this study was approved. Before a sample was taken, it was done with the patients' verbal and analytical consent. According to document number 4, a local ethics committee evaluated and approved the study protocol, subject information, and consent form on July 6, 2022 to obtain this approval.

Results

The study consisted of 100 adults designated into two groups:

1. Adults having CCS (n = 50)
2. Adults as control group (n = 50)

Age

The discrepancy in age (as mean) showed no significant different among control and CCSs, demonstrated in Table 1, as shown in Figure 2.

Body mass index

The mean ± standard deviation (SD) of body mass index (BMI) for control and group CCS were 28.0 ± 3.8 and 28.9 ± 3.0, respectively. The result of the study has shown significant difference in BMI in patients and control group (P > 0.09), as shown in Table 1, as shown in Figure 3.

Table 1: The general characteristics of patients and controls according to study

Study variables	Patients (N = 50) Mean ± SD	Controls (N = 50) Mean ± SD	P value
Age (years)	55.43 ± 5.17	53.13 ± 5.09	0.00
BMI (kg/m^2)	28.9 ± 3.0	28.0 ± 3.8	0.09

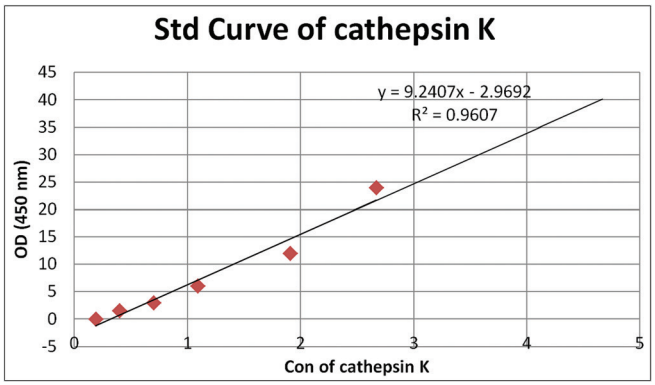

Figure 1: Standard curve of CatK

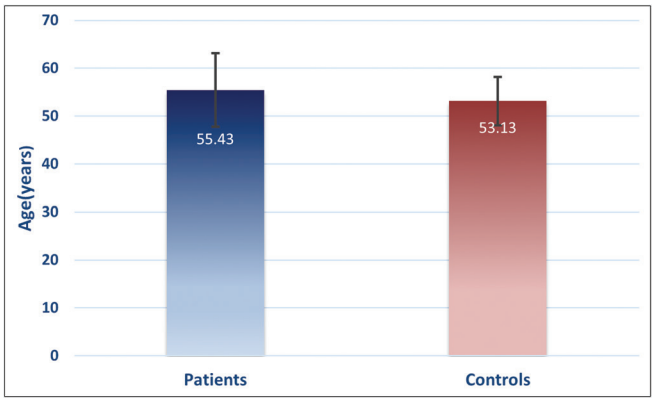

Figure 2: The mean differences of age according to study group (P < 0.001*)

Gender

Among fifty patients with CCS who contributed to this study, there were 28 men and 22 women, and this represents 56% and 44% of patients, respectively, as shown in Figure 4.

Table 2 and Figure 1 show increased concentration of CatK in CCS patients compared with the control (P value < 0.01) with significant mean differences between them.

In contrast, the current study observed non-significant change of TG, VLDL (P value ≤ 0.45), (P value ≤ 0.71), in CCS patients while increase concentrations of cholesterol, LDL in CCS patients compared with the control (P value < 0.001), (P value ≤ 0.00) with significant mean differences between them, as in Table 2.

Discussion

The onset of CCS may be influenced by a number of variables, such as age or sex, family history, smoking, obesity, diabetes, and changes in lifestyle.[16,17]

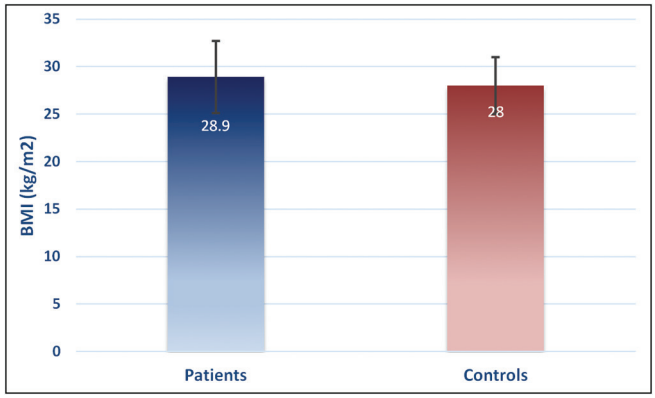

Figure 3: The mean differences of BMI according to study group (P < 0.001*)

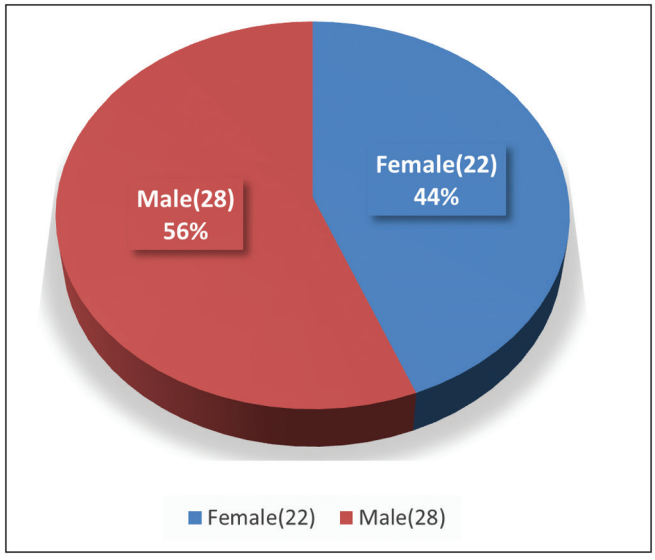

Figure 4: The distribution of gender in study group

Table 2: The mean differences of biochemical parameters rendering to study

Study variables	Study group	*N*	Mean ± SD	*P*-value
Cathepsin K (pmol/L)	CCS patients	50	9.29 ± 4.22	0.01*
	Control group	50	6.65 ± 2.40	
Cholesterol (mmol/L)	CCS patients	50	4.4 ± 0.55	<0.001*
	Control group	50	3.5 ± 0.80	
TG (mmol/L)	CCS patients	50	1.21 ± 0.36	0.45
	Control group	50	1.31 ± 0.80	
HDL (mmol/L)	CCS patients	50	0.90 ± 0.16	0.00
	Control group	50	1.20 ± 0.23	
LDL (mmol/L)	CCS patients		2.66 ± 0.50	0.00
	Control group		1.98 ± 0.67	
VLDL (mmol/L)	CCS patients		0.63 ± 0.46	0.71
	Control group		0.59 ± 0.36	

*P value ≤ 0.05 was significant

As shown in Table 2, there was significant difference in level of CatK and CCS prevalence between patients and control groups. The patients group displayed increase level CatK compare with control group. This study provides evidence that elevated levels of circulating CatK are independently associated with the prevalence of CAD after adjusting for conventional CAD risk factors including diabetes-related factors, blood pressure, dyslipidemia and BMI, excess inflammation is strongly associated with an increased risk of cardiovascular disease.[18]

Estimation of total cholesterol (TC) [Table 2] showed that results of the present study the mean value of TC was significantly in male patients and female patients groups (P-value ≤ 0.001) and SD of the patient's group (4.4 ± 0.55), SD of the control group (3.5 ± 0.80).

In our study of a group of patients with CAD, we have seen a significant increase in the incidence of CAD in people over 40 years of age due to increased lipid profile including cholesterol.[19]

Estimation of TG [Table 2] showed that results of the present study the mean value of TG was no significantly in male patients and female patients groups and (P-value ≤ 0.45).

In this study it was observed that the mean of TGs during fasting is normal in patients with CAD than the control group, which may be due to the healthy lifestyle or they have taken drugs.[20]

Estimation of HDL-C [Table 2] showed that the results of the present study the mean value of HDL-C was significantly in male patients and female patients groups (P-value ≤ 0.00) and SD of the patient's group (0.90 ± 0.16) SD of control group (1.20 ± 0.23), decreased in the mean of HDL-C level in patient groups.

Our results provide evidence that HDL-C patients with low HDL-C still have a significant risk of CHD.[21]

Estimation of LDL-C [Table 2] showed that the results of the present study the mean value of LDL-C was significantly in male patients and female patients groups (P-value ≤ 0.00) and SD of the patient's group (2.66 ± 0.50) SD of control group (1.98 ± 0.67), increased the mean LDL-C level in patient groups.

The present data suggest that elevated serum LDL-C concentration is a marker of elevated CHD risk.[22]

Estimation of VLDL [Table 2] showed that the results of the present study the mean value of VLDL was no significantly in male patients and female patients groups (P-value = 0.71) and SD of patients group (0.63 ± 0.46) SD of the control group (0.59 ± 0.36), normal in the mean of VLDL level in patient groups.

VLDL is the precursor of LDL, that have low content of cholesterol, but also contains some TGs, and these are also risk factors for CVD.[23] High levels of VLDL cholesterol can be associated with an increased risk of CAD, but in some cases, VLDL levels may remain normal even in the presence of CCS, which may be due to healthy lifestyle.

Conclusion

Our results showed that circulating CatK is a good biomarker for CCS disorders and that higher levels of CatK and lipid profile are closely related to the presence of CCS among CCS patients in Babylon province.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1. Malakar AK, Choudhury D, Halder B, Paul P, Uddin A, *et al*. A review on coronary artery disease, its risk factors, and therapeutics. J Cell Physiol 2019;234:16812-23.

2. Zainal IG. Study the profile of some antioxidant markers in diabetic mellitus and non-diabetic patients with cardiovascular disease. Med J Babylon 2022;19:653.
3. Knuuti J, Wijns W, Saraste A, Capodanno D, Barbato E, Funck-Brentano C, *et al*. ESC Guidelines for the diagnosis and management of chronic coronary syndromes: The Task Force for the diagnosis and management of chronic coronary syndromes of the European Society of Cardiology (ESC). Eur Heart J 2020;41:407-77.
4. Cheng XW, Kikuchi R, Ishii H, Yoshikawa D, Hu L, Takahashi R, *et al*. Circulating cathepsin K as a potential novel biomarker of coronary artery disease. Atherosclerosis 2013;228:211-6.
5. Li X, Li Y, Jin J, Jin D, Cui L, Li X, *et al*. Increased serum cathepsin K in patients with coronary artery disease. Yonsei Med J 2014;55:912-9.
6. Knuuti BG, Nordestgaard J, Langsted BG, Mora A, Kolovou S, Baum G, *et al*. Fasting is not routinely required for determination of a lipid profile: Clinical and laboratory implications including flagging at desirable concentration cut-points—A joint consensus statement from the European Atherosclerosis Society and European Federa. Eur Heart J 2016;37:1944-58.
7. Rubin R, Strayer DS, Rubin E. Rubin's Pathology: Clinicopathologic Foundations of Medicine. Philadelphia, PA: Lippincott Williams & Wilkins; 2008.
8. Greenwood M, Meechan JG. Management of specific medical emergencies in dental practice. In: General Medicine and Surgery for Dental Practitioners. Cham: Springer; 2019. p. 205-19.
9. Roffi M, Patrono C, Collet, J-P, Mueller C, Valgimigli M, Andreotti F, *et al*. ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation. Kardiol Pol (Polish Hear J) 2015;73:1207-94.
10. Brieger D, Eagle KA, Goodman SG, Steg PG, Budaj A, White K, *et al*. Acute coronary syndromes without chest pain, an underdiagnosed and undertreated high-risk group: Insights from the Global Registry of Acute Coronary Events. Chest 2004;126:461-9.
11. Čulić V, Eterović D, Mirić D, Silić N. Symptom presentation of acute myocardial infarction: Influence of sex, age, and risk factors. Am Heart J 2002;144:1012-7.
12. Hausenloy DJ, Botker HE, Engstrom T, Erlinge D, Heusch G, Ibanez B, Garcia-Dorado D, *et al*. Targeting reperfusion injury in patients with ST-segment elevation myocardial infarction: trials and tribulations. Eur Heart J 2017;38:935-41.
13. Mehtaa PK, Weia J, Wenger NK. Ischemic heart disease in women: A focus on risk factors. Netherlands Hear J 2017;25:181-5.
14. Moesker MJ. Assessing the cost-effectiveness of point-of-care testing for primary care patients with symptoms suggestive of acute coronary syndrome: A threshold analysis. Acta Clin Bel 2014;70:S13-S14.
15. Berry SEE. Triacylglycerol structure and interesterification of palmitic and stearic acid-rich fats: an overview and implications for cardiovascular disease. Nutr Res Rev 2009;22:3-17.
16. Wei J, Wenger NK. Ischemic heart disease in women: A focus on risk Factors. Trends Cardiovasc Med 2015;25:140-51.
17. Mohammed NI, Al-ghanimi MKS. Assessment the risk factors of congenital heart disease among children below 5 years age in Babylon province. Med J Babylon 2022;19:554.
18. Webb TR, Erdmann J, Stirrups KE, Stitziel NO, Masca NGD, Jansen H, *et al*. Increased serum cathepsin K in patients with coronary artery disease. J Am Coll Cardiol 2017;69: 823-36.
19. Lasek-Bal A, Kopyta I, Warsz-Wianecka A, Puz P, Łabuz-Roszak B, Zaręba K. Risk factor profile in patients with stroke at a young age: Stroke risk factors in young patients. Neurol Res 2018;40: 595-601.
20. Manochehri M, Moghadam AJ. Studying the relation of postprandial triglyceride with coronary artery disease (CAD). Med Arch 2016;70:261-4.
21. Pellegrino D. Antioxidants and cardiovascular risk factors. Diseases 2016;4:11.
22. Chang C-T, Wang G-J, Kuo C-C, Hsieh J-Y, Lee A-S, Chang C-M, *et al*. Electronegative low-density lipoprotein increases coronary artery disease risk in uremia patients on maintenance hemodialysis. Medicine (Baltim) 2016;95:e2265.
23. Bradley RD, Oberg EB. Are additional lipid measures useful? Integr Med (Encinitas) 2008;7:18-23.