ORIGINAL ARTICLE

Interleukin 6 concentration elevation as a risk of carotid intima-media thickness in chronic kidney disease patients with dialysis

Riri Andri Muzasti[1], Herman Hariman[2], Elvita Rahmi Daulay[3]

[1]Division of Nephrology and Hypertension, Department of Internal Medicine, [2]Department of Clinical Pathology, [3]Department of Radiology; Universitas Sumatera Utara, Medan, Indonesia

ABSTRACT

Aim To investigate the relationship between IL-6 concentration and the risk of carotid artery calcification in chronic kidney disease patients with dialysis (CKD-5D).

Methods This analytic observational cross-sectional study includes 95 clinically stable patients who underwent regular haemodialysis for at least three months at Rasyida Renal Hospital Medan, Indonesia. Serum IL-6 level was measured using the enzyme-linked immunosorbent assay (ELISA). Carotid artery calcification was determined by measuring Carotid Intima-Media Thickness (CIMT) using Real-Time B-mode ultrasound.

Results There were 53 males (55.8%) of the total samples with the mean duration of haemodialysis of 81.28±67.40 months. Ultrasound examination showed that 28 samples (29.5%) had carotid artery calcification. Statistical test significantly showed that patients with IL-6 ≥81.1 pg/mL were more likely to have carotid artery calcification with an increased risk of 12.92 times (95% CI: 5.54-30.12) compared to the group of patients who had IL-6 level <81.1 pg/mL (p <0.001).

Conclusion This study proves that a high level of IL-6 can increase the risk of carotid artery calcification in CKD-5D patients.

Key words: chronic kidney disease, interleukin-6, carotid intima-media thickness

Corresponding author:
Riri Andri Muzasti
Division of Nephrology and Hypertension.
Department of Internal Medicine,
Faculty of Medicine,
Universitas Sumatera Utara
Dr. Mansyur 5 Medan, Indonesia
Phone: +62 81 260 556 872;
Fax: +62 8161 821 6264;
E-mail: riri.andri@usu.ac.id
ORCID ID: https://orcid.org/ 0000-0001-7834-0740

Original submission:
20 March 2020;
Revised submission:
04 June 2020;
Accepted:
27 June 2020
doi: 10.17392/1172-20

Med Glas (Zenica) 2020; 17(2): 346-351

INTRODUCTION

Cardiovascular disease is a primary cause of morbidity and mortality in patients with chronic kidney diseases (CKD), and it accounts for approximately 39% of deaths among those on dialysis (1). One of the causes is vascular calcification that can occur both in tunica intima and media, which later contributes to the occurrence of myocardial ischemia, arrhythmia, and stroke (2,3). This vascular calcification has become a strong independent predictor of mortality in the general population (4) and dialysis population (5-7).

Chronic kidney disease is associated with a chronic inflammatory state characterized by increased levels of proinflammatory cytokines (8). As one of the proinflammatory cytokines, IL-6 is known as a central regulator of the inflammatory process and plays a crucial role in the induction of immune effectors and acute phase responses. IL-6 is also strongly associated with morbidity and a strong predictor of cardiovascular mortality in patients undergoing haemodialysis (9,10).

The aim of this study is to investigate the relationship between IL-6 concentration and carotid artery calcification in CKD-5D patients.

PATIENTS AND METHODS

Patients and study design

This study was an analytic observational with a cross-sectional design. Ninety-five clinically stable patients who underwent regular haemodialysis for at least three months at Rasyida Renal Hospital (Medan, Indonesia) and were willing to conduct laboratory and ultrasonography examination, which was proven by signing an informed consent, were included in this study. The patients with incomplete medical record data were excluded from the study.

The investigation was approved by the Health Research Ethical Committee of the Medical School of Universitas Sumatera Utara/H. Adam Malik General Hospital

Methods

The patients were interviewed to determine history of previous illness and how long they had been undergoing haemodialysis.

Standard laboratory methods examined serum phosphate and calcium level. The IL-6 analysis was performed by measuring serum level based on the ELISA technique. Ultrasonography (USG) examination was performed by a radiologist who did not know the patient's clinical condition. The presence of carotid artery calcification was defined if the thickness or distance between the intima-media tunica as measured by real-time B-mode ultrasound was >1 mm. Levels of calcium and phosphate were measured using the multiplication of serum level of phosphate and calcium (Ca x P).

Statistical analysis

The characteristic of regular haemodialysis patient data was expressed as a percentage for categorical data. Numerical data were expressed as mean ± standard deviation (SD) if normally distributed and expressed as median (min-max) if the data distribution were not normally distributed. The normality test was done using Kolmogorov Smirnov. The $\chi 2$ or Fisher's Exact test was used to compare proportions between the two groups with variable data categories. The logistic regression test was used to explain the association between risk factors and carotid calcification. The $p < 0.05$ was considered significant. To determine the cut-off value of the IL-6 level as a predictor of the calcification carotid artery, the receiver operating characteristics (ROC) curve was used to obtain the area under the curve (AUC) value.

RESULTS

There were 53 (55.8%) males of the total samples, the mean duration of haemodialysis (HD) was 81.28±67.40 months. The median level of calcium, phosphate and calcium phosphate was 9.80 mg/dL, 5.50 mg/dL and 53.76 mg^2/ dL^2. Through the ELISA method, the mean IL-6 level was 97.95±117.93 pg/mL.

The results of the USG examination showed that most patients did not experience carotid artery calcification, 67 (70.5%) (Table 1).

Total area under the curve was 95.9% with $p<0.001$ (95% CI: 91.2% - 100.0%), meaning that the IL-6 level had an excellent diagnostic value (> 90%) (Figure 1).

Based on the analysis with the ROC curve, the cut-off value of IL-6 level can also be determined, which has the highest combination of sensitivity and specificity. The curve showed that the IL-6 level with a value of 81.1 had the highest combinati-

Table 1. Characteristics of the patients with chronic kidney disease and dialysis (CKD-5D)

Variable	No (%) of patients	Mean ± SD	Median (min-max)
Gender			
Male	53 (55.8)		
Female	42 (44.2)		
Duration of HD (months)		81.28 ± 67.40	62.00 (43-676)
<60	38 (40.0)		
≥60	57 (60.00)		
Frequency of HD (hours/week)			
10	72 (75.8)		
12	23 (24.2)		
Calcium (mg/dL)		9.72 ± 0.74	9.80 (8.0-10.9)
≤9.5	38 (40.0)		
>9.5	57 (60.0)		
Phosphate (mg/dL)		5.43 ± 0.64	5.50 (4.0-6.8)
≤5.5	50 (52.6)		
>5.5	45 (47.4)		
Ca x P (mg2/dL2)		52.98 ± 9.08	53.76 (33.20-69.36)
<55	53 (55.8)		
≥55	42 (44.2)		
IL-6 (pg/mL)		97.95 ± 117.93	70.10 (25.4-898.0)
Calcification of carotid artery			
Absence	67 (70.5)		
Presence	28 (29.5)		

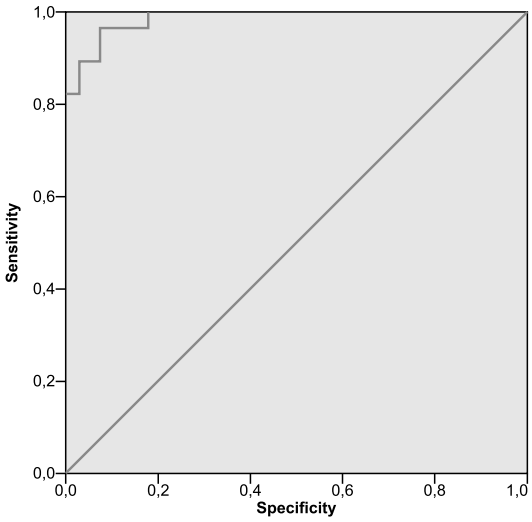

Figure 1. ROC curve of IL-6 level as a predictor of calcification carotid artery

on of sensitivity (96.4%) and specificity (92.5%). The higher IL-6 level above the cut-off value, the higher the risk of calcification. The cut-off point value of IL-6 ≥81.1 had an excellent diagnostic value and accuracy (Table 2, Figure 2).

The patients who had IL-6 level ≥81.1 pg/mL were experiencing more vascular calcification compared to those with IL-6 level ≤81.1pg/mL (p<0.001) (Table 3). The risk of carotid artery calcification increased 12.92 times (95% CI: 5.54-30.12) if the patient had IL-6 level ≥81.1 pg/mL compared to the group of patients who had IL-6 level <81.1 pg/mL (p <0.001).

Table 2. Diagnostic values of IL-6

Cut-off	Sensitivity	Specificity	PLR	NLR	PPV	NPV	Accuracy
IL-6 (≥81.1 pg/mL)	96.4%	92.5%	12.9	0.04	84.4%	98.4%	98.6%

level for carotid artery calcification PLR, positive likelihood ratio (LR+); NLR, negative likelihood ratio (LR-); PPV, positive predictive value; NPV, negative predictive value;

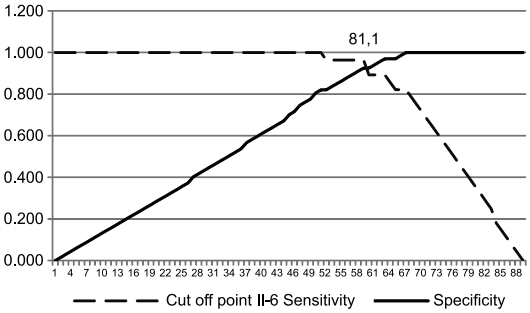

Figure 2. The cutting point value of IL-6 level as a predictor of carotid artery calcification based on the ROC curve

Table 3. Characteristics of HD patients based on carotid artery calcification

Variable	Carotid artery calcification		PR	95% CI	p
	Presence (n=28)	Absence (n=67)			
IL-6 (pg/mL)			12.92	5.54-30.12	<0.001
< 81.1	1 (1.6)	62 (98.4)			
≥ 81.1	27 (84.4)	5 (15.6)			
Calcium (mg/dL)			1.10	0.66-1.86	0.713
≤9.5	12 (31.6)	26 (68.4)			
>9.5	16 (28.1)	41 (71.9)			
Phosphate (mg/dL)			1.03	0.68-1.55	0.906
≤5.5	15 (30.0)	35 (70.0)			
>5.5	13 (28.9)	32 (71.1)			
Duration of HD (months)			1.02	0.71-1.45	0.927
<60	11 (28.9)	27 (71.7)			
≥60	17 (29.8)	40 (70.2)			

PR, Prevalence Ratio, CI, Confidence Interval

DISCUSSION

There are many risk factors that can cause kidney function to decline, such as genetic components, sex, age, diabetes, and hypertension (11). Sex has traditionally been seen as an essential factor influencing the development of kidney disease (12). Regardless of its etiology, females tend to progress more slowly to end-stage kidney disease (13). Our study shows that the prevalence of patients undergoing haemodialysis was commonly found in males (55.8%). These results are in line with the study of Lumtergul et al. in CKD patients. They found that most of the dialysis and non-dialysis patients in Thailand were male with the percentage of 55.4% and 53.5% respectively (14). The same results were obtained by Nakayama et al. (15) and Ok et al. (16).

Carotid artery calcification has been recognized as a risk factor for cardiovascular events in patients with end-stage renal disease (15). Traditional risk factors alone, such as hypertension and dyslipidaemia, do not sufficiently contribute to the high calcification burden in the dialysis patient (17,18). Hyperphosphatemia and increased level of calcium x phosphate products, which is caused by reduced renal phosphate excretion, were also identified as an essential cause of accelerated arterial calcification in this group of patients (4, 19-22). Our study and the study conducted by Barreto et al. (23) found the mean phosphate levels and calcium-phosphate multiplication still within the target range for dialysis patients determined by the Kidney Disease Outcomes Quality Initiative (KDOQI), which are 8.4-9.5 mg/L for calcium, 3.5-5.5 mg/dL for phosphate and <55 mg^2/dL^2 for calcium-phosphate multiplication (24). Its target range was achieved probably because almost all patients have taken the phosphate binders drugs.

A study conducted by Barreto et al. took 2.97 pg/mL as the cut-off IL-6 (23), while the study conducted by Kato et al. dividing the IL-6 cut-off into three groups consisting of ≤1.1 pg/mL, 1-1-2pg/mL and >2 pg/mL (25). The IL-6 cut-off by Honda et al. was 8.1 pg/mL (sensitivity 63.3%, specificity 73.3%) (26). In this study, the IL-6 cut-off obtained was 81.1 pg/mL (sensitivity 96.4%, specificity 92.5%). This cut-off difference can be due to the characteristics of the sample and the cut-off method used.

The prevalence of vascular calcification varies greatly from 60-100% depending on the location of the examination and the diagnostic method used and, in the area where the study was conducted (27). London et al. reported that 68% of 202 patients undergoing haemodialysis in France had arterial calcifications determined by radiography and echocardiography (28). A study conducted by Nakayama et al. in 135 patients undergoing routine haemodialysis at hospitals in Japan, the prevalence of carotid artery calcification was 71% (15). This study found that the prevalence of carotid artery calcification was 29.5%. This difference in prevalence can be due to sample characteristics such as genetic, demographic, lifestyle differences, as well as the distribution and number of samples.

Systemic inflammation is commonly found in patients with chronic kidney disease undergoing routine haemodialysis (29). The exact cause is unknown. However, it is suspected that every time a patient undergoes haemodialysis, the formation of reactive oxygen species (ROS) occurs, which plays an essential role in endothelial dysfunction and atherogenesis, which is modulated by IL-6 (31). A study conducted by Beberashvili et al. showed chronic inflammation characterized by elevated levels of IL-6 correlated with all-cause mortality in stable chronic haemodialysis patients (32). A study conducted by Maddhumathi et al. showed that a median plasma IL-6 level in 206 patients undergoing haemodialysis was 7.9 pg/mL (ranging from 0.1 to 90.2 pg/mL), and was found to be higher in patients with vascular disease (9). In this study, from the 95 patients, we found that the proportion of patients with IL-6 ≥81.1 pg/mL was more likely to have carotid artery calcification with an increased risk of 12.92 times compared to the group of patients who had IL-6 levels <81.1 pg/mL. Similar results were also obtained by Kato et al. (25) and Krasniak et al. (33).

In this study, we found that the mean of the duration of haemodialysis was 81.28 ± 67.40 months and showed no significant relationship between the duration of haemodialysis and the occurrence of carotid artery calcification. It may be because the process of vascular calcification can occur at any time and started at a younger age (34). Even the study conducted by Nitta et al. with the mean duration of haemodialysis about 7.7 ± 5.8 years also showed no significant results between the duration of haemodialysis and the occurrence of vascular calcification (35).

In conclusion, this study proves that high level of IL-6 can increase the risk of carotid artery calcification in CKD-5D patients.

ACKNOWLEDGEMENTS

Universitas Sumatera Utara financially supported this work under Talenta research implementation contracts 2019.

FUNDING

This work was supported by Universitas Sumatera Utara under Talenta research implementation contracts 2019, number: 4167/UN5.1.R/PPM/2019

TRANSPARENCY DECLARATION

Conflict of interest: None to declare.

REFERENCES

1. Bethesda, MD. The United States Renal Data System. 2018 USRDS annual data report: Epidemiology of kidney disease in the United States. Bethesda, MD: National Institutes of Health: National Institute of Diabetes and Digestive and Kidney Diseases, 2018.
2. Benz K, Hilgers KF, Daniel C, Amann K. Vascular calcification in chronic kidney disease: the role of inflammation. Intl J Nephrol 2018; 2018:4310379.
3. Floege, Jürgen; Ketteler, Markus. Vascular calcification in patients with end-stage renal disease. Nephrol Dial Transplant 2004; 19(Suppl 5):V59-66.
4. Wilson PW, Kauppila LI, O'Donnell CJ, Kiel DP, Hannan M, Polak JM, Cupples LA. Abdominal aortic calcific deposits are an important predictor of vascular morbidity and mortality. Circulation 2001;103: 1529–34.
5. Blacher J, Guerin AP, Pannier B, Marchais SJ, London GM. Arterial calcifications, arterial stiffness, and cardiovascular risk in end-stage renal disease. Hypertension 2001; 38:938–42.
6. London GM, Guerin AP, Marchais SJ, Metivier F, Pannier B, Adda H. Arterial media calcification in end-stage renal disease: impact on all-cause and cardiovascular mortality. Nephrol Dial Transplant 2003; 18:1731–40.
7. Okuno S, Ishimura E, Kitatani K, Fujino Y, Kohno K, Maeno Y, Maekawa K, Yamakawa T, Imanishi Y, Inaba M, Nishizawa Y. The presence of abdominal aortic calcification is significantly associated with all-cause and cardiovascular mortality in maintenance hemodialysis patients. Am J Kidney Dis 2007; 49:417–25.
8. Stenvinkel P, Alvestrand A. Inflammation in end-stage renal disease: sources, consequences, and therapy. Semin Dial 2002; 15:329-37.
9. Madhumathi Rao et al. Plasma interleukin-6 predicts cardiovascular mortality in hemodialysis patients. Am Kidney Dis 2005; 45.2:324-33.
10. Pecoits-Filho R, Lindholm B, Axelsson J, Stenvinkel P. Update on Interleukin -6 and its role in chronic renal failure. Nephrol Dial Transplant 2003; 18:1042-5.
11. Kazancioglu R. Risk factors for chronic kidney disease: an update. Kidney Int Suppl 2013; 368-71.
12. Baylis, Chris. Sexual dimorphism in the aging kidney: differences in the nitric oxide system. Nat Reviews Nephrol 2009; 5: 384-96.
13. Neugarten J, Acharya Anjali, Silbiger, Sharon R. Effect of gender on the progression of nondiabetic renal disease: a meta-analysis. J Am Soc Nephrol 2000; 11:319-29.
14. Lumlertgul D, Kantachuvesiri S, Apichaiyingyurd S, Treamtrakanpon W, Rattanasompattikul M, Gojaseni P, Thanakitcharu P, Trakarnvanich T, Poonvivatchaikarn U, Vareesangthip K, Impact-CKD investigators. Prevalence of and predictive factor for abdominal aortic calcification in Thai chronic kidney disease patients. Ther Apher Dial 2017; 21:611-9.
15. Masaru N, Ura Y, Nagata M, Okada Y, Sumida Y, Nishida K, Ikeda H, Kaizu Y. Carotid artery calcification at the initiation of hemodialysis is a risk factor for cardiovascular events in patients with end-stage renal disease: a cohort study. BMC Nephrol 2011:12:56.
16. Ok E, Asci G, Toz H, Ok ES, Kircelli F, Yilmaz M, Hur E, Demirci MS, Demirci C, Duman S, Basci A, Adam SM, Isik IO, Zengin M, Suleymanlar G, Yilmaz ME, Ozkahya M, Turkish Online Haemodiafiltration Study. Mortality and cardiovascular events in online haemodiafiltration (OL-HDF) compared with high-flux dialysis: results from the Turkish OL-HDF Study. Nephrol Dial Transplant 2013; 28:192-202.
17. Longenecker JC, Coresh J, Powe NR, Levey AS, Fink NE, Martin A, Klag MJ. Traditional cardiovascular disease risk factors in dialysis patients compared with the general population: the CHOICE Study. J Am Soc Nephrol 2002; 13:1918–27.
18. Sarnak MJ, Coronado BE, Greene T, Wang SR, Kusek JW, Beck GJ, Levey AS. Cardiovascular disease risk factors in chronic renal insufficiency. Clin Nephrol 2002; 57:327–35.
19. Goodman WG, Goldin J, Kuizon BD, Yoon C, Gales B, Sider D, Wang Y, Chung J, Emerick A, Greaser L, Elashoff RM, Salusky IB. Coronary artery calcification in young adults with end-stage renal disease who are undergoing dialysis. N Engl J Med 2000; 342:1478–83.
20. Chertow GM, Raggi P, Chasan-Taber S, Bommer J, Holzer H, Burke SK. Determinants of progressive vascular calcification in hemodialysis patients. Nephrol Dial Transplant 2004; 19:1489–96.
21. Raggi P, Boulay A, Chasan-Taber S, Amin N, Dillon M, Burke SK, Chertow GM. Cardiac calcification in adult hemodialysis patients. A link between end-stage renal disease and cardiovascular disease? J Am Coll Cardiol 2002; 20:39:695–701.
22. Asmus HG, Braun J, Krause R, Brunkhorst R, Holzer H, Schulz W, Neumayer HH, Raggi P, Bommer J. Two-year comparison of sevelamer and calcium carbonate effects on cardiovascular calcification and bone density. Nephrol Dial Transplant 2005; 20:1653–61.
23. Barreto DV, Barreto FC, Liabeuf S, Temmar M, Lemke H-D, Tribouilloy C, Choukroun G, Vanholder R, A Massy ZA, European Uremic Toxin Work Group (EUTox). Plasma interleukin-6 is independently associated with mortality in both hemodialysis and pre-dialysis patients with chronic kidney disease. Kidney Int 2010; 77:550-6.
24. NKF KDOQI Guidelines. KDOQI Clinical Practice Guidelines for Bone Metabolism and Disease in Chronic Kidney Disease. 2003 http://kidneyfoundation.cachefly.net/professionals/KDOQI/guidelines_bone/guide6.htm (28 January 2020)
25. Akihiko K, Odamaki M, Takita T, Maruyama Y, Kumagai H, Hishida A. Association between interleukin-6 and carotid atherosclerosis in hemodialysis patients. Kidney Int 2002; 61:1143-52.
26. Hirokazu H, Qureshi AR, Heimbürger O, Barany P, Wang K, Pecoits-Filho R, Stenvinkel P, Lindholm B. Serum albumin, C-reactive protein, interleukin 6, and fetuin as predictors of malnutrition, cardiovascular disease, and mortality in patients with ESRD. Am J Kidney Dis 2006, 47:139-48.

27. Guillermo R-D, Griselda B, Graciela F, Adriana P, Fabian O, Miriam L, Martin O, Gustavo L, Mariana V-D, Soledad C, Mariano F, Carlos M, Marcelo O, Ricardo H, Carlos D, Rcardo G-M, Oscar V, Hernan T M. Prevalence of factor related to vascular calcification in patients with chronic kidney disease on dialysis. MedICINA (BAires) 2017; 77:3.
28. London GM, Guerin AP, Marchais SJ, Metivier F, Pannier B, Adda H. Arterial media calcification in end-stage renal disease: impact on all-cause and cardiovascular mortality. Nephrol Dial Transplant 2003, 18.9:1731- 40.
29. Lau WL, Kalantar-Zadeh K, Vaziri NDJN. The gut as a source of inflammation in chronic kidney disease. Nephron 2015; 130:92-8.
30. Spittle MA, Hoenich NA, Handelman GJ, Adhikarla R, Homel P, Levin NW. Oxidative stress and inflammation in hemodialysis patients. Am kidney dis 2001; 38:1408-13.
31. Yudkin JS, Kumari M, Mohamed-Ali V. Inflammation, obesity, stress, and coronary heart disease: is interleukin-6 the link. Atherosclerosis 2000; 148.2:209-14.
32. Beberashvili I, Sinuani I, Azar A, Yasur H, Shapiro G, Feldman L, Averbukh Z, Weissgarten J. IL-6 levels, nutritional status, and mortality in prevalent hemodialysis patients. Clin J Am Soc Nephrol 2011; 6:2253-63.
33. Krasniak A, Drozdz M, Pasowicz M, Chmiel G, Michalek M, Szumilak D, Podolec P, Klimeczek P, Konieczynska M, Muniak EW, Tracz W, Khoa TN, Souberbielle JC, Drueke TB, Sulowicz W. Factors involved in vascular calcification and atherosclerosis in maintenance hemodialysis patients. Nephrol Dial Transplant 2007; 22:515-21.
34. Jayalath RW, Mangan SH, Golledge J. Aortic calcification. Eur J Vasc Endovasc Surg 2005, 30:476-88.
35. Nitta K, Akiba T, Uchida K, Kawashima A, Yumura W, Kabaya T, Nihei H. The progression of vascular calcification and serum osteoprotegerin levels in patients on long-term hemodialysis. Am J Kidney Dis 2003; 42:303-9.