Association of Clinical Characteristics with the Proportion of the HDL-Cholesterol Subclasses HDL-2b and HDL-3 Among Patients Undergoing Hemodialysis

Jin-Bor Chen (chenjb1019@gmail.com)
Kaohsiung Chang Gung Memorial Hospital and Chang Gung University college of Medicine
https://orcid.org/0000-0003-4007-1455

Wen-Chin Lee
Division of Nephrology, Kaohsiung Chang Gung Memorial Hospital and Chang Gung University college of Medicine

Sin-Hua Moi
Institute of Biotechnology and Chemical Engineering, I-Shou University

Cheng-Hong Yang
Department of Electronic Technology, National Kaohsiung University of Science and Technology

Research

Keywords: diabetes, HDL cholesterol, HDL-2b, HDL-3, hemodialysis, hs-CRP

DOI: https://doi.org/10.21203/rs.3.rs-30553/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License.
Read Full License
Abstract

**Background:** Altered high-density lipoprotein cholesterol (HDL-C) composition in patients with chronic kidney disease is common. However, reports on the distribution of HDL-C subclasses in patients undergoing hemodialysis (HD) are limited.

**Objective:** We aimed to compare the two main HDL-C subclasses, HDL-2b and HDL-3, in two cohorts of HD patients and healthy individuals and examine their associations with clinical characteristics.

**Methods:** A total of 164 prevalent HD patients and 71 healthy individuals in one hospital-facilitated outpatient clinic were enrolled from May 2019 to July 2019. The HDL-2b and HDL-3 proportions were measured and statistical analysis was performed.

**Results:** The mean ages of HD patients and healthy individuals were 63 and 49.9 years, respectively. HD patients showed lower HDL-2b and HDL-3 proportions compared with those of healthy individuals (23.6% vs. 31.2%, \( P < 0.001 \); 31.7% vs. 33.6%, \( P = 0.137 \), respectively). The HDL-2b proportion was significantly higher with a high-sensitivity C-reactive protein (hs-CRP) levels of <3 mg/L compared with hs-CRP \( \geq 3 \) mg/L in the HD cohort (\( P = 0.005 \)). HDL-3 proportion was lower with a hs-CRP level of <3 mg/L compared with hs-CRP \( \geq 3 \) mg/L in the HD cohort (\( P = 0.022 \)). Sex and diabetes did not influence the HDL-2b and HDL-3 proportions in the HD cohort.

**Conclusions:** HD patients had lower HDL-2b and HDL-3 proportions than those of healthy individuals. The distribution of the HDL-2b and HDL-3 subclasses in HD patients is influenced by proinflammatory status, not by sex and diabetic status.

Introduction

Patients with chronic kidney disease (CKD) are at high risk for cardiovascular diseases (CVD)[1, 2]. In particular, CVD confers to high overall mortality in the dialysis population[3, 4]. High-density lipoprotein cholesterol (HDL-C) has been well recognized as an independent predictor of CVD in the general population[5, 6]. Interestingly, several conditions are free from this association, for example, drug intervention to elevate the HDL-C levels, refined HDL-C by genetic effect, and specific higher HDL-C levels[7–9].

In the CKD population, dyslipidemia has been considered as a major risk factor for CVD. Screening for dyslipidemia in patients with CKD is supported by clinical guidelines[10–12]. Although decreased HDL-C levels and its impaired composition and function are generally documented in the CKD population, the association of HDL-C with CVD risk is still controversial[13–17].

HDL is composed of several subclass particles, varying in size, density, chemical composition, and physicochemical properties[18]. Generally, HDL can be divided into the following subclasses by gel electrophoresis coupled with immunoblotting: HDL-2a, HDL-2b (large size, cholesterol rich), HDL-3a, HDL-
3b, HDL-3c, preβ₁-HDL (small size), and preβ₂-HDL[19]. In prior studies, HDL subclasses contribute to different effects on atherosclerosis. An increased large particle level, HDL-2b, exerts an antiatherogenic effect. In contrast, increased smaller particle levels, HDL-3 and preβ₁-HDL, are positively associated with CVD[18, 20–22]. Due to complex technique and instrument requirement, the measurement of HDL subclasses is not widely applied in the past years. Recently, a microfluidic chip-based technique is developed. This fast, easy-to-operate technique is successfully applied to measure the HDL subclasses in the epidemiological studies[23, 24].

In this study, we hypothesize that patients with hemodialysis (HD) have different proportions of the HDL subclasses compared with healthy individuals. Using the microfluidic chip-based technique, we measured the HDL-2b and HDL-3 subclass proportions in HD patients and correlated these with their demographic characteristics and proinflammatory status.

**Materials And Methods**

**Study design and participants**

Adult patients (> 18 years) who underwent maintenance HD thrice weekly for at least 3 months in the outpatient clinic in Kaohsiung Chang Gung Memorial Hospital in Taiwan from May 2019 to July 2019 were enrolled in this study. The exclusion criteria were as follows: ongoing treatment for malignancy, acute inflammatory diseases, hospitalization within 3 months, malnutrition defined by serum albumin level < 3.5 g/dL, and pregnancy. Healthy controls were recruited voluntarily in the outpatient clinic by posted protocol notification. All blood samples from HD patients in the fasting status and in mid-week (Wednesday and Thursday) were obtained. Informative patient data, including demographic profiles and laboratory parameters, were also collected.

**Analytic Parameters**

All blood samples for biochemistry measurement were obtained using commercial kits and an autoanalyzer (Hitachi 7600 – 210, Hitachi Ltd., Tokyo, Japan). Albumin levels were measured using the bromocresol green method. Intact parathyroid hormone level was measured using a chemiluminescence immunoassay (Siemens Healthcare Diagnostics Inc., USA). The high-sensitivity C-reactive protein (hs-CRP) level was assayed using the immunoturbidimetric method (Spectra East Laboratories, Rockleigh, NJ, USA).

The plasma total cholesterol, triglyceride, and HDL-C levels were determined enzymatically on the Eroset Hitachi 7600 – 210 analyzer. The low-density lipoprotein cholesterol (LDL-C) levels were calculated using to the Friedewald formula, which provides reliable values up to a triglyceride level of 4.0 mmol/L.

HDL-C subclass profiles were measured by electrophoresis of a microfluidic chip system. Briefly, serum samples, calibrator, and QC materials were diluted 1:50 in sample buffer in the presence of a mixture of
lipophilic fluorescent dyes and allowed to incubate for 5–15 min prior to loading on to chips. Separation was carried out in a microfluidic device (MICEP-30, Ardent BioMed). The entire procedure was performed in less than 1 h. The HDL-2b and HDL-3 subclasses were automatically calculated in line by a proprietary algorithm (Ardent BioMed LLC, Mt. View, California, USA).

Statistical analysis

The baseline demographic characteristics and laboratory measurements in HD patients and healthy controls are presented as frequency (percentage) and mean (standard deviation). The distribution difference was estimated using the independent two-sample t-test or chi-square test. The correlation between the HDL-2b and HDL-3 proportions and associated variables was estimated using the Pearson correlation test. A boxplot was used to illustrate the HDL-2b and HDL-3 subclass proportions in different subgroups, and the difference in these proportions between comparison groups was estimated using the independent two-sample t-test. All P values were two-sided, and P < 0.05 was considered statistically significant. All statistical analyses were performed using the R 3.6.3 software (R Core Team, 2020).

Results

Characteristics of participants

Participants were screening by inclusion and exclusion criteria. Finally, a total of 164 participants with HD and 71 healthy controls were included in the analyses (Fig. 1). The mean ages of participants with HD and healthy controls were 63 and 49.9 years, respectively. The male-to-female percentages in the HD cohort and healthy controls were 48.8–51.2% and 31.0–69.0%, respectively. In the comparison of main laboratory parameters, HD patients showed a significantly lower HDL-2b subclass proportion compared with healthy controls (23.6% vs. 31.2%, P < 0.001) as well as lower total cholesterol (152 vs. 189 mg/dL, P < 0.001), HDL-C (44.5 vs. 59.8 mg/dL, P < 0.001), and LDL-C (88.1 vs. 112.1 mg/dL, P < 0.001) levels. The HDL-3 subclass proportion was not significantly different between the HD patients and healthy controls (31.7% vs. 33.6%, P = 0.137) (Table 1).
| Variables                          | HD (n = 164) | Control (n = 71) | P     |
|-----------------------------------|-------------|-----------------|-------|
| Age (years)                       | 63.1 ± 12.3 | 49.9 ± 10.6     | < 0.001 |
| Sex                               |             |                 | 0.017 |
| female                            | 84 (51.2%)  | 49 (69.0%)      |       |
| male                              | 80 (48.8%)  | 22 (31.0%)      |       |
| BMI (kg/m$^2$)                    | 23.5 ± 12.5 | 23.3 ± 4.1      | 0.925 |
| Dialysis vintage (years)          | 10.48 ± 7.53| -               |       |
| Diabetes                          | 27 (16.5%)  | N/A             |       |
| CVA                               | 3 (1.8%)    | N/A             |       |
| CAD                               | 6 (3.7%)    | N/A             |       |
| Antihypertensive                  | 66 (40.2%)  | -               |       |
| Lipid-lowering drugs              | 24 (14.6%)  | -               |       |
| Etiology of kidney failure        |             |                 |       |
| Primary kidney disease            | 61 (37.2%)  | N/A             |       |
| Systemic disease                  | 79 (48.2%)  | N/A             |       |
| Unknown                           | 24 (14.6%)  | N/A             |       |
| Laboratory measurements           |             |                 |       |
| Total Cholesterol (mg/dL)         | 152 (94–314)| 189 (135–310)   | < 0.001|
| Triglyceride (mg/dL)              | 108 (30–994)| 80.5 (18–237)   | < 0.001|
| HDL-C (mg/dL)                     | 44.5 ± 15.4 | 59.8 ± 14.0     | < 0.001|
| HDL-2b (%)                        | 23.6 ± 9.1  | 31.2 ± 6.7      | < 0.001|
| HDL-3 (%)                         | 31.7 ± 11   | 33.6 ± 7.5      | 0.137 |

Abbreviations: BMI, body mass index; CVA, cerebral vascular accident; CAD, coronary artery disease; HDL, high-density lipoprotein; LDL, low-density lipoprotein; VLDL, very low-density lipoprotein; C, cholesterol; T, total; PTH, parathyroid hormone; BUN, blood urea nitrogen; Cr, creatinine; hs-CRP, high-sensitivity C-reactive protein.
| Variables           | HD (n = 164) | Control (n = 71) | P       |
|---------------------|--------------|-----------------|---------|
| LDL-C (mg/dL)       | 88.1 ± 35.0  | 112.1 ± 30.8    | < 0.001 |
| Intact-PTH (pg/ml)  | 268.2 (2.4–2819.9) | 53.6 (20.9–90.2) | < 0.001 |
| Albumin (g/dL)      | 3.9 ± 0.3    | 4.2 ± 0.8       | 0.075   |
| hs-CRP (mg/L)       | 6.9 ± 13.0   | 2.7 ± 5.0       | 0.016   |
| Hemoglobin (g/dL)   | 10.7 ± 1.2   | 12.6 ± 1.7      | < 0.001 |
| Total leukocytes (10^9/L) | 6.3 ± 2.4   | 6.1 ± 1.7       | 0.567   |
| BUN (mg/dL)         | 69 (32–151)  | 13 (7–46)       | < 0.001 |
| Cr (mg/dL)          | 10.4 ± 2.3   | 0.7 ± 0.2       | < 0.001 |

Abbreviations: BMI, body mass index; CVA, cerebral vascular accident; CAD, coronary artery disease; HDL, high-density lipoprotein; LDL, low-density lipoprotein; VLDL, very low-density lipoprotein; C, cholesterol; T, total; PTH, parathyroid hormone; BUN, blood urea nitrogen; Cr, creatinine; hs-CRP, high-sensitivity C-reactive protein.

Pearson correlation analysis on study parameters

The results from the Pearson correlation test showed that the HDL-2b subclass proportion was significantly negatively correlated with age (r = -0.26, P < 0.001), HDL-3 subclass proportion (r = -0.54, P < 0.001), triglyceride (r = -0.44, P < 0.001), and total leukocyte count (r = -0.16, P < 0.05). On the other hand, it was significantly positively correlated with HDL-C (r = 0.68, P < 0.001), total cholesterol (r = 0.21, P < 0.01), and hemoglobin (r = 0.16, P < 0.05). Similarly, the HDL-3 subclass proportion was significantly negatively correlated with the HDL-2b subclass proportion (r = -0.54, P < 0.001) and HDL-C (r = -0.52, P < 0.001) and positively correlated with triglyceride (r = 0.51, P < 0.001), hemoglobin (r = 0.14, P < 0.05), and total leukocyte count (r = 0.19, P < 0.01) (Table 2).
Table 2
Pearson correlation analysis.

| Variables           | HDL-2b (%) | HDL-3 (%) |
|---------------------|------------|-----------|
| Age (years)         | -0.26**    | 0.01      |
| Total Cholesterol (mg/dL) | 0.21**   | -0.03     |
| Triglyceride (mg/dL) | -0.44***   | 0.51***   |
| HDL-C (mg/dL)       | 0.68***    | -0.52***  |
| HDL-2b (%)          | 1.00       | -0.54***  |
| HDL-3 (%)           | -0.54***   | 1.00      |
| LDL-C (mg/dL)       | 0.10       | 0.02      |
| Intact-PTH (pg/ml)  | < 0.001    | -0.03     |
| Albumin (g/dL)      | 0.05       | 0.13      |
| hs-CRP (mg/L)       | -0.11      | < 0.001   |
| Hemoglobin (g/dL)   | 0.16*      | 0.14*     |
| Total leukocytes (10^9/L) | -0.16*   | 0.19**    |

*p < 0.05, **p < 0.01, ***p < 0.001.

- Figure 1. Participants flow diagram.

Influence of sex on the HDL-2b and HDL-3 subclass proportions

Both sexes in the HD cohort demonstrated significantly lower HDL-2b and HDL-3 subclass proportions compared with healthy controls. On the other hand, the HDL-2b and HDL-3 subclass proportions were not different between sexes in the HD cohort (Fig. 2).

Associations of the hs-CRP levels with the HDL-2b and HDL-3 subclass proportions

We stratified the entire cohort with the cutoff hs-CRP level of 3 mg/L (normal range < 3 mg/L in the laboratory) and examined the associations with the HDL-2b and HDL-3 subclass proportions. The results showed that the HDL-2b (P = 0.031) and HDL-3 (P = 0.030) subclass proportions were significantly lower in the HD cohort under the hs-CRP level of < 3 mg/L compared with healthy controls. Similarly, the HDL-3 (P = 0.004) subclass proportion was significantly lower in the HD cohort under the hs-CRP level of ≥ 3 mg/L compared with healthy controls. However, this relationship was not noted in the HDL-2b (P = 0.274) subclass proportion (Fig. 3). We further examined the associations of the hs-CRP levels < 3 mg/L with the HDL-2b and HDL-3 subclass proportions in the HD cohort. The results showed that the HDL-2b (P = 0.005) subclass proportion was significantly lower under hs-CRP levels ≥ 3 mg/L compared
with those with hs-CRP levels < 3 mg/L. In contrast, HDL-3 ($P = 0.022$) subclasses revealed an opposite trend (Fig. 3 – 2).

Influence of diabetes on the HDL-2b and HDL-3 subclass proportions in the HD cohort

Diabetic patients with HD did not demonstrate significant differences in the HDL-2b and HDL-3 subclass proportions compared with nondiabetic HD patients (Fig. 4).

**Discussion**

The key findings in our study are that HD patients presented lower HDL-2b and HDL-3 subclass proportions compared with healthy controls. In HD patients, the HDL-2b and HDL-3 subclass proportions was influenced by proinflammatory status, not by sex differentiation and diabetes. Our findings indicate altered HDL composition in HD patients and potential utilization of the proportion of the HDL subclass analysis in clinical practice in these patients. We prefer to use proportions or ratio of HDL subclass analysis instead of a simple subclass measurement to generalize across the whole spectrum of the HDL levels[25, 26]. Moreover, the microfluidic chip-based technique is easy to be applied in the clinical investigation and could forward to cause-relationship investigation.

A growing body of evidence has demonstrated the decreased HDL function as anti-inflammatory, antioxidant, and endothelial protection in patients with CKD and dialysis[27–30]. The mechanisms are complex, but altered HDL composition in CKD is thought to be one of the significant points[14, 15]. There are still rare reports in the literature about the HDL subclass distribution in patients with dialysis. Two reports have described that HD patients have a lower HDL-3 cholesterol level and do not have a decrease in the HDL-2 cholesterol level compared with healthy individuals[31, 32]. Another report observed contradictory results of increased HDL-3a and decreased HDL-2b subclass levels in patients with HD compared with healthy controls[33]. It is also reported that HD patients have decreased levels of both HDL-2 and HDL-3 cholesterol[34, 35]. The discrepancies in the aforementioned studies may have resulted from either the different methods for HDL subclass measurement or statistical comparison in different dialysis populations and relatively small case number. In our study, we used the microfluidic chip-based technique more easily to examine the proportional distribution of the HDL subclasses in HD patients. This technique has been successfully used to measure the HDL subclasses in the Prospective Cardiovascular Munster study[23]. Our participants with HD showed decreased proportions of both HDL-2b and HDL-3 compared with healthy individuals. This result is in accordance with previous recognition regarding the alteration of HDL composition in patients with CKD[13, 14, 16, 17]. The cause relationship of alteration in distributed proportions of the HDL-C subclasses on CVD prevalence in patients with CKD still warrants further investigation in a population-based, longitudinal observational study.

Interestingly, a difference was noted in the HDL-2 and HDL-3 cholesterol levels between sexes in the prior studies. The HDL-2 and HDL-3 cholesterol levels were lower in men compared with women in the general population and participants with rheumatoid arthritis and acute coronary syndrome[18, 36]. However, there was no difference in both sexes in participants with metabolic syndrome[26]. In our study, we found
that patients with HD showed lower HDL-2b and HDL-3 subclass proportions in both sexes compared with healthy individuals. We also examined the associations of HDL-C subclasses with various clinical parameters. However, there is only HDL-C showing strong association with HDL-2b and HDL-3 subclasses. Due to the lack of compared reports, above findings need to be clarified in the future.

CKD is a proinflammatory status. CRP is commonly used as a marker of systemic inflammation. Recently, an elevated hs-CRP level has been linked to malnutrition, inflammation, and atherosclerosis syndrome and is considered to be a risk factor for morbidity and mortality in patients with CKD[37–39]. For mechanism investigation, CRP/oxLDL/β2GPI complex-aggravated atherosclerosis by increasing lipid uptake has been reported in a diabetic mouse study[40]. In our study, we found that decreased HDL-2b and HDL-3 subclass proportions in HD patients in either low or high hs-CRP levels compared with healthy individuals. Interestingly, the HDL-2b and HDL-3 subclass proportions were influenced by the hs-CRP levels in HD patients. However, diabetic status did not influence the HDL-2b and HDL-3 subclass proportions in HD patients. Accordingly, proinflammatory status could suppress the HDL-2b and increase HDL-3 subclass proportions in patients with HD. However, the relationship between the severity of proinflammatory status and diabetes and HDL subclass distribution still needs to be determined in a future investigation.

Our study has several limitations. First, this was a small-sized, single-center study on an Asian HD population. The results may not be extrapolated to other ethnic HD populations. Second, our study only measured the HDL subclass proportion in HD patients in one time point. The longitudinal effect of HD on HDL subclass distribution cannot be obtained in our study. Third, our study participants were relatively clinically stable; therefore, associations between proinflammatory status and HDL subclass distribution cannot be stratified by wide-range hs-CRP levels. Despite the aforementioned limitations, the strengths of our study are that this is the first clinical study to examine the proportions of the HDL subclasses HDL-2b and HDL-3 in HD patients and there is availability of cause-specific data to fill knowledge gap about reasons for HDL subclass distribution in these patients. The clinical utility of HDL subclass distribution analysis would be further facilitated by spanning a full-range HDL-C population and determine the subclass distribution on adverse CVD event.

Conclusions

HD patients have lower HDL-2b and HDL-3 subclass proportions compared with healthy individuals. The distribution of the HDL-2b and HDL-3 subclasses is influenced by proinflammatory status, not by sex and diabetic status.

Declarations

Acknowledgments

Authors are grateful for the Miss Chiu-Hua Chen for her laboratory work.
Authors’ contributions

J.B.C. conceptualized the study. J.B.C. and W.C.L. wrote the methodology. S.H.M. performed the formal analysis. J.B.C. wrote and prepared the original draft. W.C.L. and J.B.C. wrote, reviewed, and edited the manuscript. C.H.Y provided supervision.

Funding

Authors are grateful for funding support in this work by the grant from Kaohsiung Chang Gung Memorial Hospital in Taiwan (document no: CMRPG8J0031).

Availability of data and materials

The data analyzed during the present study are available from the corresponding author on reasonable request.

Ethics approval and consents to participants

The protocol for the study was approved by the Committee on Human Research at Kaohsiung Chang Gung Memorial Hospital (IRB document: 201801486B0) in Taiwan and conducted in accordance with the principles of the Declaration of Helsinki. All participants signed informed consent to approve study initiation.

Consent for publication

Not applicable

Competing interests

All authors declared no conflict of interest in the manuscript preparation.

Author details

1 Division of Nephrology, Department of Internal Medicine, Kaohsiung Chang Gung Memorial Hospital and Chang Gung University College of Medicine, Kaohsiung,

2 Institute of Biotechnology and Chemical Engineering, I-Shou University, Kaohsiung, Taiwan

3 Department of Electronic Engineering, National Kaohsiung University of Science and Technology, Kaohsiung, Taiwan

References

1. 1. Sarnak MJ, Levey AS: Cardiovascular disease and chronic renal disease: a new paradigm. Am J Kidney Dis. 2000; 35:S117-131.
2. Sarnak MJ, Levey AS, Schoolwerth AC, Coresh J, Culleton B, Hamm LL, McCullough PA, Kasiske BL, Kelepouris E, Klag MJ, et al: Kidney disease as a risk factor for development of cardiovascular disease: a statement from the American Heart Association Councils on Kidney in Cardiovascular Disease, High Blood Pressure Research, Clinical Cardiology, and Epidemiology and Prevention. Circulation. 2003; 108:2154–2169.

3. Wen CP, Cheng TY, Tsai MK, Chang YC, Chan HT, Tsai SP, Chiang PH, Hsu CC, Sung PK, Hsu YH, Wen SF: All-cause mortality attributable to chronic kidney disease: a prospective cohort study based on 462 293 adults in Taiwan. Lancet. 2008; 371:2173–2182.

4. Collado S, Coll E, Nicolau C, Azqueta M, Pons M, Cruzado JM, de la Torre B, Deulofeu R, Mojal S, Pascual J, Cases A: Serum osteoprotegerin in prevalent hemodialysis patients: associations with mortality, atherosclerosis and cardiac function. BMC Nephrol. 2017; 18:290.

5. Gordon DJ, Probstfield JL, Garrison RJ, Neaton JD, Castelli WP, Knoke JD, Jacobs DR, Jr., Bangdiwala S, Tyroler HA: High-density lipoprotein cholesterol and cardiovascular disease. Four prospective American studies. Circulation. 1989; 79:8–15.

6. Gordon DJ, Rifkind BM: High-density lipoprotein—the clinical implications of recent studies. N Engl J Med. 1989; 321:1311–1316.

7. Boden WE, Probstfield JL, Anderson T, Chaitman BR, Desvignes-Nickens P, Koprowicz K, McBride R, Teo K, Weintraub W: Niacin in patients with low HDL cholesterol levels receiving intensive statin therapy. N Engl J Med. 2011; 365:2255–2267.

8. Voight BF, Peloso GM, Orho-Melander M, Frikke-Schmidt R, Barbalic M, Jensen MK, Hindy G, Holm H, Ding EL, Johnson T, et al: Plasma HDL cholesterol and risk of myocardial infarction: a mendelian randomisation study. Lancet. 2012; 380:572–580.

9. Madsen CM, Varbo A, Nordestgaard BG: Extreme high high-density lipoprotein cholesterol is paradoxically associated with high mortality in men and women: two prospective cohort studies. Eur Heart J. 2017; 38:2478–2486.

10. Anderson TJ, Gregoire J, Pearson GJ, Barry AR, Couture P, Dawes M, Francis GA, Genest J, Jr., Grover S, Gupta M, et al: 2016 Canadian Cardiovascular Society Guidelines for the Management of Dyslipidemia for the Prevention of Cardiovascular Disease in the Adult. Can J Cardiol. 2016; 32:1263–1282.

11. Authors/Task Force M, Catapano AL, Graham I, De Backer G, Wiklund O, Chapman MJ, Drexel H, Hoes AW, Jennings CS, Landmesser U, et al: 2016 ESC/EAS Guidelines for the Management of Dyslipidaemias: The Task Force for the Management of Dyslipidaemias of the European Society of Cardiology (ESC) and European Atherosclerosis Society (EAS) Developed with the special contribution of the European Association for Cardiovascular Prevention & Rehabilitation (EACPR). Atherosclerosis. 2016; 253:281–344.

12. Li YH, Ueng KC, Jeng JS, Charning MJ, Lin TH, Chien KL, Wang CY, Chao TH, Liu PY, Su CH, et al: 2017 Taiwan lipid guidelines for high risk patients. J Formos Med Assoc. 2017; 116:217–248.
13. Hager MR, Narla AD, Tannock LR: Dyslipidemia in patients with chronic kidney disease. Rev Endocr Metab Disord. 2017; 18:29–40.

14. Holzer M, Birner-Gruenberger R, Stojakovic T, El-Gamal D, Binder V, Wadsack C, Heinemann A, Marsche G: Uremia alters HDL composition and function. J Am Soc Nephrol. 2011; 22:1631–1641.

15. Silbernagel G, Genser B, Drechsler C, Schamal H, Grammer TB, Stojakovic T, Krane V, Ritz E, Wanner C, Marz W: HDL cholesterol, apolipoproteins, and cardiovascular risk in hemodialysis patients. J Am Soc Nephrol. 2015; 26:484–492.

16. Chang TI, Streja E, Soohoo M, Ko GJ, Rhee CM, Kovesdy CP, Kashyap ML, Vaziri ND, Kalantar-Zadeh K, Moradi H: Increments in serum high-density lipoprotein cholesterol over time are not associated with improved outcomes in incident hemodialysis patients. J Clin Lipidol. 2018; 12:488–497.

17. Navaneethan SD, Schold JD, Walther CP, Arrigain S, Jolly SE, Virani SS, Winkelmayer WC, Nally JV, Jr.: High-density lipoprotein cholesterol and causes of death in chronic kidney disease. J Clin Lipidol. 2018; 12:1061–1071.e1067.

18. Tian L, Li C, Liu Y, Chen Y, Fu M: The value and distribution of high-density lipoprotein subclass in patients with acute coronary syndrome. PLoS One. 2014; 9:e85114.

19. Xu Y, Fu M: Alterations of HDL subclasses in hyperlipidemia. Clin Chim Acta. 2003; 332:95–102.

20. Kontush A, Chapman MJ: Antiatherogenic small, dense HDL–guardian angel of the arterial wall? Nat Clin Pract Cardiovasc Med. 2006; 3:144–153.

21. Asztalos BF, Cupples LA, Demissie S, Horvath KV, Cox CE, Batista MC, Schaefer EJ: High-density lipoprotein subpopulation profile and coronary heart disease prevalence in male participants of the Framingham Offspring Study. Arterioscler Thromb Vasc Biol. 2004; 24:2181–2187.

22. Cheung MC, Brown BG, Wolf AC, Albers JJ: Altered particle size distribution of apolipoprotein A-I-containing lipoproteins in subjects with coronary artery disease. J Lipid Res. 1991; 32:383–394.

23. Mueller O, Chang E, Deng D, Franz T, Jing D, Kincaid R, Konigshofer Y, Kratzmeier M, McNulty M, Qian H, et al: PROCAM Study: risk prediction for myocardial infarction using microfluidic high-density lipoprotein (HDL) subfractionation is independent of HDL cholesterol. Clin Chem Lab Med. 2008; 46:490–498.

24. Ma YH, Zhao L, Xian XD, Yang D, Huang W, Wang YH, Mueller O, Chang E, Königshofer Y, Van-Cleve M, et al: A case-control study on the relationship between HDL2b and non-alcoholic fatty liver disease in Chinese type 2 diabetic patients. Clin Chem Lab Med. 2009; 47:1067–1072.

25. Moriyama K, Negami M, Takahashi E: HDL2-cholesterol/HDL3-cholesterol ratio was associated with insulin resistance, high-molecular-weight adiponectin, and components for metabolic syndrome in Japanese. Diabetes Res Clin Pract. 2014; 106:360–365.

26. Yang HS, Hur M, Kim H, Kim SJ, Shin S, Di Somma S: HDL Subclass Analysis in Predicting Metabolic Syndrome in Koreans With High HDL Cholesterol Levels. Ann Lab Med. 2020; 40:297–305.

27. Kalantar-Zadeh K, Kopple JD, Kamranpour N, Fogelman AM, Navab M: HDL-inflammatory index correlates with poor outcome in hemodialysis patients. Kidney Int. 2007; 72:1149–1156.
28. Shroff R, Speer T, Colin S, Charakida M, Zewinger S, Staels B, Chinetti-Gbaguidi G, Hettrich I, Rohrer L, O'Neill F, et al: HDL in children with CKD promotes endothelial dysfunction and an abnormal vascular phenotype. J Am Soc Nephrol. 2014; 25:2658–2668.

29. Jones PH, Nair R, Thakker KM: Prevalence of dyslipidemia and lipid goal attainment in statin-treated subjects from 3 data sources: a retrospective analysis. J Am Heart Assoc. 2012; 1:e001800.

30. Yamamoto S, Yancey PG, Ikizler TA, Jerome WG, Kaseda R, Cox B, Bian A, Shintani A, Fogo AB, Linton MF, et al: Dysfunctional high-density lipoprotein in patients on chronic hemodialysis. J Am Coll Cardiol. 2012; 60:2372–2379.

31. Schiavon R, Battaglia P, De Fanti E, Fasolin A, Biasioli S, Targa L, Guidi G: HDL3-related decreased serum paraoxonase (PON) activity in uremic patients: comparison with the PON1 allele polymorphism. Clin Chim Acta. 2002; 324:39–44.

32. Samouilidou EC, Karpouza AP, Kostopoulos V, Bakirtzi T, Pantelias K, Petras D, Tzanatou-Exarchou H, E JG: Lipid abnormalities and oxidized LDL in chronic kidney disease patients on hemodialysis and peritoneal dialysis. Ren Fail. 2012; 34:160–164.

33. Alabakovska SB, Todorova BB, Labudovic DD, Tosheska KN: LDL and HDL subclass distribution in patients with end-stage renal diseases. Clin Biochem. 2002; 35:211–216.

34. Piperi C, Kalofoutis C, Tzivras M, Troupis T, Skenderis A, Kalofoutis A: Effects of hemodialysis on serum lipids and phospholipids of end-stage renal failure patients. Mol Cell Biochem. 2004; 265:57–61.

35. Samouilidou E, Karpouza A, Grapsa E, Tzanatou-Exarchou H: Serum oxidized LDL is inversely associated with HDL2-cholesterol subclass in renal failure patients on hemodialysis. Nephron Clin Pract. 2010; 115:c289-294.

36. Arts E, Fransen J, Lemmers H, Stalnhoef A, Joosten L, van Riel P, Popa CD: High-density lipoprotein cholesterol subfractions HDL2 and HDL3 are reduced in women with rheumatoid arthritis and may augment the cardiovascular risk of women with RA: a cross-sectional study. Arthritis Res Ther. 2012; 14:R116.

37. Qureshi AR, Alvestrand A, Divino-Filho JC, Gutierrez A, Heimburger O, Lindholm B, Bergstrom J: Inflammation, malnutrition, and cardiac disease as predictors of mortality in hemodialysis patients. J Am Soc Nephrol. 2002; 13 Suppl 1:S28-36.

38. Iseki K, Tozawa M, Yoshi S, Fukiyama K: Serum C-reactive protein (CRP) and risk of death in chronic dialysis patients. Nephrol Dial Transplant. 1999; 14:1956–1960.

39. Su YJ, Liao SC, Cheng BC, Hwang JC, Chen JB: Increasing high-sensitive C-reactive protein level predicts peritonitis risk in chronic peritoneal dialysis patients. BMC Nephrol. 2013; 14:185.

40. Zhang R, Zhou SJ, Li CJ, Wang XN, Tang YZ, Chen R, Lv L, Zhao Q, Xing QL, Yu DM, Yu P: C-reactive protein/oxidised low-density lipoprotein/beta2-glycoprotein I complex promotes atherosclerosis in diabetic BALB/c mice via p38mitogen-activated protein kinase signal pathway. Lipids Health Dis. 2013; 12:42.
Figures

Figure 1

Participants flow diagram.
Figure 2
Proportion of HDL-2b and HDL-3 subclasses among male and female study cohorts

Figure 3
Proportion of HDL-2b and HDL-3 subclasses in study cohorts stratified by an hs-CRP concentration ≥ 3 mg/L.
Figure 4

Proportion of HDL-2b and HDL-3 subclasses in patients with hemodialysis stratified by an hs-CRP concentrations 3 mg/L.

Figure 5

Proportion of HDL-2b and HDL-3 subclasses in diabetic and non-diabetic patients with hemodialysis.