Am J Cancer Res 2023;13(2):475-484
www.ajcr.us /ISSN:2156-6976/ajcr0147777

Original Article

Prognostic and therapeutic value of serum lipids and a new IPI score system based on apolipoprotein A-I in diffuse large B-cell lymphoma

Tiantian Yu[1,3], Dan Luo[2], Cancan Luo[1], Zijun Y Xu-Monette[3], Li Yu[1]

[1]Department of Hematology, The Second Affiliated Hospital of Nanchang University, Nanchang, Jiangxi, China; [2]Department of Hematology, The First Affiliated Hospital of Jishou University, Jishou, Hunan, China; [3]Division of Hematopathology and Department of Pathology, Duke University Medical Center, Durham, NC, USA

Received November 15, 2022; Accepted January 10, 2023; Epub February 15, 2023; Published February 28, 2023

Abstract: Lipid metabolism is associated with lymphomagenesis and functions as a new therapeutic target in patients with lymphoma. Several serum lipids and lipoproteins have prognostic value in solid tumors; however, their value in diffuse large B-cell lymphoma (DLBCL) has been poorly described. We retrospectively analyzed and compared pre-treatment serum lipid and lipoprotein levels, including triacylglycerol (TG), low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), apolipoprotein A-I (ApoA-I), and apolipoprotein B (ApoB) between 105 DLBCL and 105 controls (no DLBCL). The prognostic significance of serum lipid and lipoprotein levels was determined using univariate and multivariate Cox proportional hazards models. The primary outcomes, overall survival (OS) and progression-free survival (PFS), were assessed by the Kaplan-Meier method. We combined the International Prognostic Index (IPI) with ApoA-I to build a nomogram model (IPI-A) to predict the OS and PFS of DLBCL. Serum TG, LDL-C, HDL-C, ApoA-I, and ApoB levels were significantly lower in the DLBCL patients than in controls and significantly increased after chemotherapy. Multivariate analyses showed that the ApoA-I level was an independent predictor of OS and PFS. In addition, our findings indicated that the prognostic index IPI-A significantly improves risk prediction over the traditional IPI score system. ApoA-I is an independent prognostic factor associated with poor OS and PFS in DLBCL patients. Our findings suggested that IPI-A is a prognostic index accurately used for risk assessment in patients with DLBCL.

Keywords: Diffuse large B-cell lymphoma, ApoA-I, prognosis, therapeutic value, new IPI score system

Introduction

Diffuse large B-cell lymphoma (DLBCL) accounts for almost 30% of all non-Hodgkin lymphomas (NHLs). The International Prognostic Index (IPI) has been widely used to predict the prognosis of DLBCL since 1993 [1]. However, the final survival outcomes differ in patients with identical IPI scores. Meaning that the IPI scoring system requires improvement. Molecular features of a lymphoma offer significant value in predictive and prognostic measures, but they have divergent results between different studies and treatment regimens. Various factors have a crucial impact on the prognosis of lymphoma and are identified as prognostic factors.

Over the past decade, unbalanced lipid metabolism has been established as an essential metabolic phenotype in the development of cancer and is suggested to be associated with cancer risk [2, 3]. In two population-based cohort studies including 27 cancer types in 116,728 individuals, low levels of high-density lipoprotein cholesterol (HDL-C) and/or apolipoprotein A-I (ApoA-I) were associated with an increased risk of several cancers, such as NHL and multiple myeloma [4]. Recently, the proliferation and progression of DLBCL cells have been recognized to be highly addicted to lipids metabolism, independent of their cell of origin [5]. One study showed that specific lipid and metabolic profiles, such as phosphatidylinositol and sphingomyelin fragments, were more common in resistant DLBCL [6]. Fatty acid synthase activity correlated positively with the PI3K-Akt-mTOR pathway, which can mediate protein synthesis and promote the oncogenic translation

Table 1. The clinical characteristics of patients with DLBCL

Variable	Total
Age, y	56.2 ± 14.5
Male, n (%)	60 (57.1)
IPI (> 2), n (%)	28 (26.7)
LDH, > ULN, n (%)	42 (40.0)
Ann Arbor stage III or IV, n (%)	61 (58.1)
ECOG PS (≥ 2), n (%)	7 (6.7)
Extranodal sites (≥ 2), n (%)	15 (14.3%)
B symptom, n (%)	11 (10.5)
Non-GCB, n (%)	69 (66)
Treatment contained R, n (%)	65 (62)
WBC count, 10^9/L	6.5 ± 3.9
RBC count, 10^{12}/L	4.1 ± 0.6
HGB, g/L	120.1 ± 22.8
PLT count, 10^9/L	209.0 (167.5-266.0)
TG, mmol/L	1.6 ± 1.0
LDL-C, mmol/L	2.6 ± 0.7
HDL-C, mmol/L	1.0 ± 0.3
ApoA-I, g/L	1.0 ± 0.4
ApoB, g/L	0.8 ± 0.2

Values are mean ± SD, median [IQR] for skewed variables, or n (%) for categorical variables. Abbreviations: ApoA-I, Apolipoprotein A-I; ApoB, Apolipoprotein B; DLBCL, Diffuse Large B Cell Lymphoma; ECOG, Eastern Cooperative Oncology Group; GCB, Germinal Center B-cell; HBG, Hemoglobin; HDL-C, High-Density Lipoprotein cholesterol; IPI, International Prognostic Index; LDH, Lactate Dehydrogenase; LDL-C, Low-Density Lipoprotein Cholesterol; PLT, Platelet; PS, Performance Status; R, Rituximab; RBC, Red Blood Cell; TG, triacylglycerol; ULN, Upper Limit of Normal; WBC, White Blood Cell.

of DLBCL [5]. Additionally, the excess fatty acids released by DLBCL cells with increased lipid metabolism in the microenvironment impairs natural killer cell function [7]. In population-based studies, statin use was associated with a lower risk of NHL [8] and a dose-related survival benefit prior to chemo/immunotherapy alone for newly diagnosed DLBCL patients [9]. Lipid metabolism is correlated to lymphomagenesis and is an attractive therapeutic target for lymphoma. However, the role of lipids in lymphoma development has not been explored in detail.

Herein, we conducted a retrospective study to examine the effect of serum lipids on the prognosis and treatment of DLBCL patients. Furthermore, we built a new risk scoring system to optimize the risk stratification of patients with DLBCL which may assist in future clinical trial designs.

Methods

Patients

Between October 2010 and January 2020, 105 eligible patients (60 men and 45 women; aged 18-83 years; median age, 57 years) were diagnosed with DLBCL at the Second Affiliated Hospital of Nanchang University in this retrospective study comprised the DLBCL group. The cases were defined as DLBCL according to the current World Health Organization diagnostic criteria. The overall survival (OS) and progression-free survival (PFS) times were also based on these definitions. In addition, 105 control participants without DLBCL were recruited from the Physical Examination Department of the Second Affiliated Hospital of Nanchang University as the control group. The demographic details of the patients are presented in **Table 1**. The last follow-up was performed in October 2021.

All the participants provided written informed consent. All the data were obtained following the Declaration of Helsinki. The Institute of Research Ethics Committee of the Second Affiliated Hospital of Nanchang University, Jiangxi, China approved this study.

Statistical analysis

We performed all statistical analyses with IBM SPSS, version 26.0 (IBM Corp., Armonk, NY, USA), GraphPad Prism, version 5.0 (GraphPad Software, CA, USA), and the R programming language (R Core Team, Vienna, Austria). The chi-square test was conducted to compare categorical variables, and the Student's t-test was applied to continuous variables. After the five kinds of serum lipids were stratified by X-tile (Yale University, CT, USA), prognostic efficacy was analyzed using the Kaplan-Meier (KM) method (two-sided statistical tests). Univariate and multivariate logistic regression analyses were assessed to determine whether there were any statistical relationships between each independent variable and survival. Significant variables in the univariate analysis were measured in the multivariate analysis. The Cox proportional hazards regression model used the

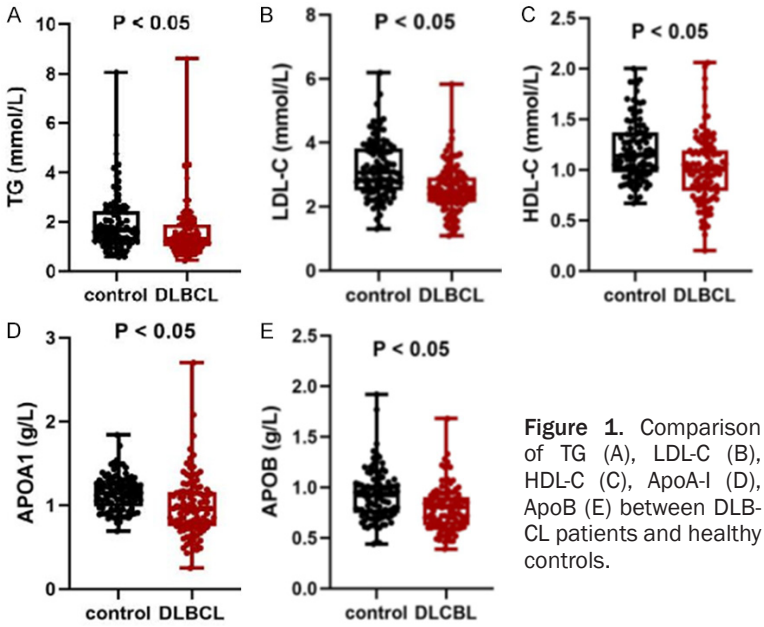

Figure 1. Comparison of TG (A), LDL-C (B), HDL-C (C), ApoA-I (D), ApoB (E) between DLBCL patients and healthy controls.

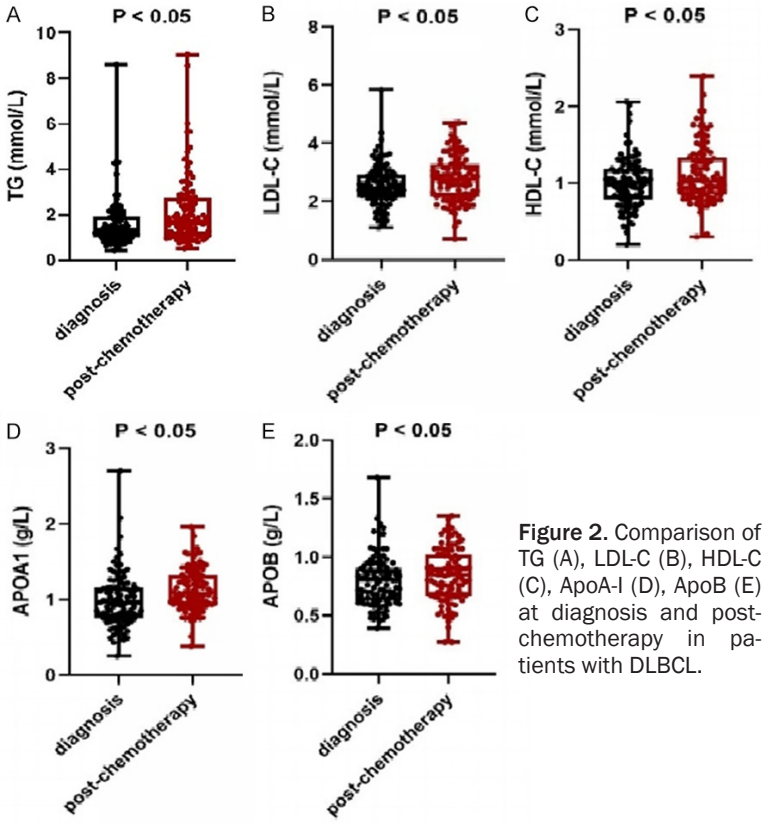

Figure 2. Comparison of TG (A), LDL-C (B), HDL-C (C), ApoA-I (D), ApoB (E) at diagnosis and post-chemotherapy in patients with DLBCL.

hazard ratio (HR) and 95% confidence interval (CI) to assess independent risk factors for serum lipids. Receiver operating characteristic (ROC) curves were performed to compare the IPI score and the newly established IPI-ApoA-I (IPI-A) score. Statistical significance was set at $P < 0.05$.

Results

The difference between the DLBCL cohort and the controls

The clinical characteristics and laboratory data of the 105 DLBCL patients are shown in **Table 1**. Of the patients, 60 were men (57%), and 45 were women (43%), with a total median age of 57 years (range 14-79 years). Of the 105 patients, 61 (58%) had advanced-stage disease (III-IV). The control group had similar age and sex characteristics. We compared the average levels of serum lipids of participants in the DLBCL group with those of the control group. Serum lipid levels, including triacylglycerol (TG) ($P < 0.05$), low-density lipoprotein cholesterol (LDL-C) ($P < 0.05$), HDL-C ($P < 0.05$), ApoA-I ($P < 0.05$), and apolipoprotein B (ApoB) ($P < 0.05$), were significantly lower in DLBCL patients than in the non-DLBCL and age-matched controls (**Figure 1**).

Post-chemotherapy serum lipid fluctuation

To understand the changes in lipid levels during treatment, we examined the lipid metabolism of patients with DLBCL after the fourth or sixth courses of chemotherapy. When the course of treatment was insufficient, the lipids after the last chemotherapy session were selected for comparison with the lipids at diagnosis. We found a significant increase in TG ($P < 0.05$), LDL-C ($P < 0.05$), HDL-C ($P < 0.05$), ApoA-I ($P < 0.05$), and ApoB ($P < 0.05$) levels after chemotherapy (**Figure 2**).

Table 2. The values of serum lipid levels at diagnosis in different therapeutic groups

Variable	Contained R (N = 65)			Treatments (N = 105)		
	CR	Non-CR	*P*-value	CR	Non-CR	*P*-value
TG (mmol/L)	1.43 (1.06-2.06)	1.41 (1.16-1.67)	0.714	1.33 (0.97-1.94)	1.30 (1.09-1.59)	0.868
LDL-C (mmol/L)	2.37 (2.14-3.08)	2.40 (2.12-2.81)	0.687	2.59 ± 0.77	2.43 ± 0.56	0.307
HDL-C (mmol/L)	1.02 (0.81-1.19)	0.83 (0.68-1.04)	0.039	1.02 (0.83-1.23)	0.87 (0.67-1.05)	0.053
ApoA-I (g/L)	1.03 (0.82-1.15)	0.76 (0.70-0.88)	0.008	1.03 (0.81-1.19)	0.78 (0.69-0.93)	0.001
ApoB (g/L)	0.81 (0.60-0.92)	0.73 (0.61-0.94)	0.769	0.80 ± 0.22	0.78 ± 0.20	0.746

Values are mean ± SD, median [IQR]. Abbreviations: ApoA-I, Apolipoprotein A-I; ApoB, Apolipoprotein B; CR, Complete Remission; HDL-C, High-Density Lipoprotein Cholesterol; LDL-C, Low-Density Lipoprotein Cholesterol; TG, triacylglycerol.

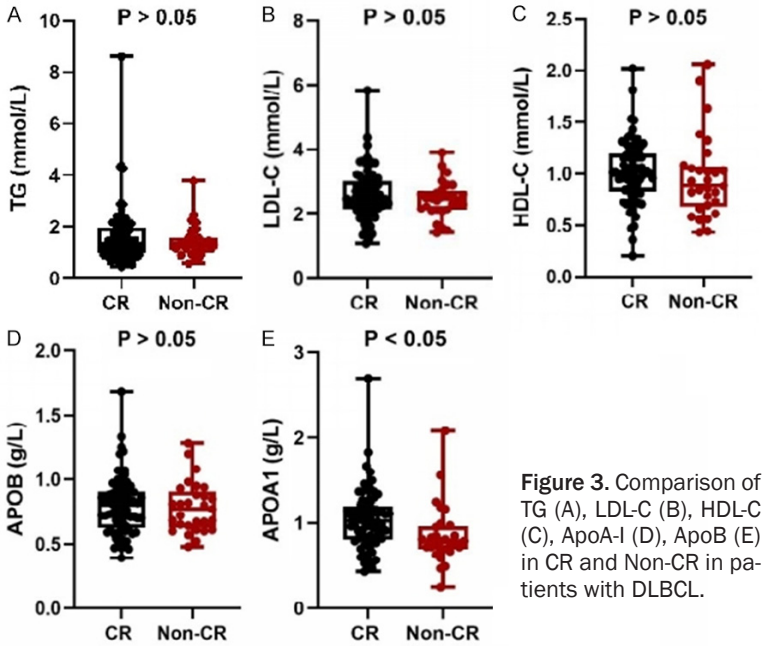

Figure 3. Comparison of TG (A), LDL-C (B), HDL-C (C), ApoA-I (D), ApoB (E) in CR and Non-CR in patients with DLBCL.

Relationship between serum lipid levels and curative effect

The relationship between lipids and the treatment's curative effect in patients with DLBCL was further analyzed. Patients with complete response (CR) exhibited a significantly higher ApoA-I level (P = 0.001) at diagnosis than the non-CR group (**Table 2**; **Figure 3**). Of the 65 patients who received rituximab-based chemotherapy, 47 (72%) achieved CR and 18 (28%) did not. Higher levels of ApoA-I and HDL-C in patients with DLBCL were associated with CR (HDL-C, P = 0.039; ApoA-I, P = 0.008) (**Table 2**). ApoA-I levels were significantly higher in the CR group than the non-CR group, regardless of whether the patients received rituximab, showing that ApoA-I is related to therapeutic efficacy and has value in predicting efficacy.

Association between serum lipid levels and survival in DLBCL cohort

The associations between survival and lipid metabolism parameters were analyzed. The X-tile program was used to determine the optimal cut-off values for TG, LDL-C, HDL-C, ApoA-I, and ApoB for OS, which were 1.43 g/L, 1.74 mmol/L, 0.61 mmol/L, 0.81 g/L, and 0.76 g/L, respectively (**Figures 4A** and S1). The KM curves revealed that patients with DLBCL and high levels of TG, LDL-C, HDL-C, and ApoB had better OS but not PFS (Figure S2) than those with low levels of the serum lipids. There is a significant concern that these patients with high levels of ApoA-I had markedly increased survival, both in OS and PFS (P < 0.001; **Figure 4B**). Patients with ApoA-I > 0.81 g/L had a median PFS of 36.5 months and a median OS of 40.5 months, whereas the median PFS and OS were 13 and 26 months, respectively, for patients with ApoA-I ≤ 0.81 g/L. The Cox proportional hazards model for the OS is shown in **Table 3**. In the univariate analysis, age (P = 0.023), Ann Arbor stage (P = 0.009), lactate dehydrogenase (LDH) (P = 0.026), extranodal sites of disease (P = 0.008), TG (P = 0.038), LDL-C (P = 0.007), HDL-C (P = 0.012), ApoA-I (P < 0.001), and ApoB (P = 0.026) were significantly associated with OS. In addition to the known prognostic factors, including age, Ann Arbor stage, extranodal sites of disease, and LDH, lipids were included in the multivariate Cox proportional hazards regres-

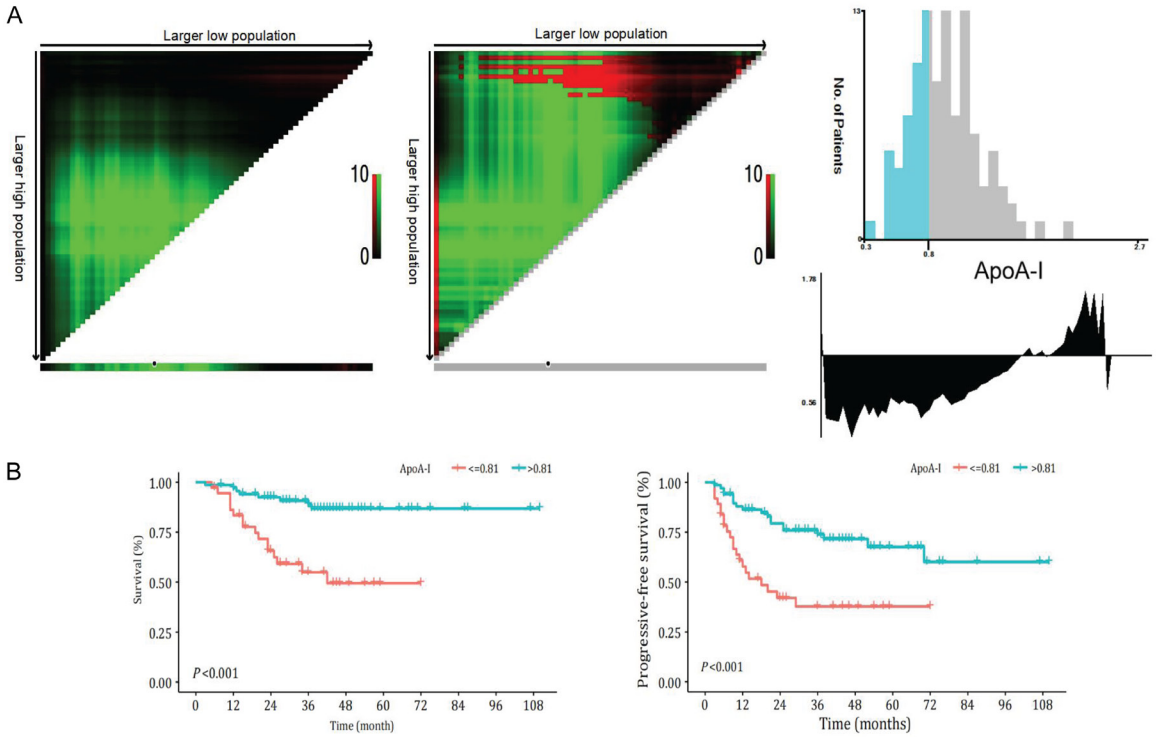

Figure 4. (A) X-Tile analysis of OS according to ApoA-I. The estimated optimal cut-off point of ApoA-I was 0.81 g/L, (B) Survival curves of ApoA-I in patients with DLBCL. The high level of ApoA-I was associated with better OS and PFS of DLBCL patients.

Table 3. Univariate and multivariate Cox hazards analysis for OS in DLBCL

Parameters		Univariate analysis		Multivariate analysis	
		HR (95% CI)	*P*-value	HR (95% CI)	*P*-value
Age, y	≥ 60 vs < 60	2.67 (1.14, 6.24)	0.023		
Sex	female vs male	0.67 (0.29, 1.56)	0.351		
LDH	> ULN vs ≤ ULN	2.72 (1.12, 6.57)	0.026		
Ann Arbor stage	III or IV vs I or II	3.10 (1.32, 7.27)	0.009		
ECOG performance status	> 1 vs ≤ 1	2.41 (0.72, 8.09)	0.155		
No. of extranodal sites of disease	> 1 vs ≤ 1	4.30 (1.47, 12.61)	0.008		
B symptom	Yes vs No	2.39 (0.81, 7.02)	0.113		
GCB	Yes vs No	1.19 (0.52, 2.72)	0.679		
TG, mmol/L	> 1.43 vs ≤ 1.43	0.35 (0.13, 0.94)	0.038	0.41 (0.14, 1.17)	0.096
LDL-C, mmol/L	> 1.74 vs ≤ 1.74	0.28 (0.11, 0.71)	0.007	0.25 (0.06, 1.11)	0.068
HDL-C, mmol/L	> 0.61 vs ≤ 0.61	0.31 (0.12, 0.77)	0.012	0.29 (0.08, 0.99)	0.048
ApoA-I, g/L	> 0.81 vs ≤ 0.81	0.21 (0.09, 0.49)	< 0.001	0.25 (0.09, 0.73)	0.011
ApoB, g/L	> 0.76 vs ≤ 0.76	0.37 (0.15, 0.88)	0.026	0.44 (0.17, 1.14)	0.090

Abbreviations: ApoA-I, Apolipoprotein A-I; ApoB, Apolipoprotein B; DLBCL, Diffuse Large B Cell Lymphoma; ECOG, Eastern Cooperative Oncology Group; GCB, Germinal Center B-cell; HDL-C, High-Density Lipoprotein Cholesterol; LDH, Lactate Dehydrogenase; LDL-C, Low-Density Lipoprotein Cholesterol; TG, triacylglycerol; ULN, Upper Limit of Normal.

sion analysis. Multivariate analysis showed that HDL-C (P = 0.048) and ApoA-I (P = 0.011) were independent prognostic factors for patients with DLBCL. In the PFS analyses, the Ann Arbor stage (P < 0.008), extranodal sites of disease (P = 0.01), and ApoA-I levels (P = 0.001) were detected as statistically significant in the univariate analysis. Multivariate analyses

Table 4. Univariate and multivariate Cox hazards analysis for PFS in DLBCL

Parameters		Univariate analysis		Multivariate analysis	
		HR (95% CI)	*P*-value	HR (95% CI)	*P*-value
Age, y	≥ 60 vs < 60	1.07 (0.57, 2.01)	0.831		
Sex	female vs male	1.08 (0.58, 2.01)	0.818		
LDH	> ULN vs ≤ ULN	1.64 (0.88, 3.05)	0.120		
Ann Arbor stage	III or IV vs I or II	2.63 (1.28, 5.40)	0.008		
ECOG performance status	> 1 vs ≤ 1	2.39 (0.85, 6.76)	0.100		
No. of extranodal sites of disease	> 1 vs ≤ 1	2.60 (1.25, 5.37)	0.010		
B symptom	Yes vs No	1.33 (0.47, 3.74)	0.593		
GCB	Yes vs No	0.65 (0.32, 1.32)	0.232		
TG, mmol/L	> 1.43 vs ≤ 1.43	0.92 (0.49, 1.74)	0.805	0.81 (0.40, 1.62)	0.550
LDL-C, mmol/L	> 1.74 vs ≤ 1.74	0.51 (0.21, 1.21)	0.126	0.18 (0.05, 0.60)	0.006
HDL-C, mmol/L	> 0.61 vs ≤ 0.61	0.48 (0.20, 1.15)	0.101	0.51 (0.18, 1.47)	0.211
ApoA-I, g/L	> 0.81 vs ≤ 0.81	0.33 (0.18, 0.62)	0.001	0.42 (0.19, 0.94)	0.034
ApoB, g/L	> 0.76 vs ≤ 0.76	0.59 (0.31, 1.11)	0.102	0.56 (0.27, 1.15)	0.112

Abbreviations: ApoA-I, Apolipoprotein A-I; ApoB, Apolipoprotein B; DLBCL, Diffuse Large B Cell Lymphoma; ECOG, Eastern Cooperative Oncology Group; GCB, Germinal Center B-cell; HDL-C, High-Density Lipoprotein Cholesterol; LDH, Lactate Dehydrogenase; LDL-C, Low-Density Lipoprotein Cholesterol; TG, triacylglycerol; ULN, Upper Limit of Normal.

identified LDL-C (P = 0.006) and ApoA-I (P = 0.034) as also statistically significant (**Table 4**). The above data suggest that ApoA-I levels act as an independent prognostic factor for OS and PFS.

The new risk scoring system for patients with DLBCL

To optimize the DLBCL risk stratification, a new risk score system was explored and established based on the multivariate Cox analysis of OS and PFS. ApoA-I was included in the IPI to form a new score system, which we named IPI-A. We compared our the IPI-A system with the IPI system using a ROC curve. Considering the overall distribution of the sample's survival, we evaluated the model's 1-year, 3-year, and 5-year predictive effects. The new prognostic index IPI-A of OS was more advanced, with an area under the curve (AUC) of 0.896 at 1 year, 0.883 at 3 years, and 0.870 at 5 years (**Figure 5A**), compared with the IPI of OS with an AUC of 0.868 at 1 year, 0.833 at 3 years, and 0.826 at 5 years. (**Figure 5B**). In addition, IPI-A exhibited better PFS (IPI-A, 1 year: AUC = 0.745, 3 years: AUC = 0.827, and 5 years: AUC = 0.763; IPI, 1 year: AUC = 0.709, 3 years: AUC = 0.808, and 5 years: AUC = 0.744) (**Figure 5C**, **5D**). We combined the IPI (age, LDH, ECOG Performance Status Scale, Ann Arbor stage, and extranodal sites of disease) with ApoA-I to construct a nomogram model to predict the OS and PFS of patients with DLBCL (**Figure 6A**, **6B**). Each factor had an accompanying score corresponding to the points at the top of the nomogram. The modeling results were evaluated using a calibration plot in Figure S3. The calibration plot for predicting 1-, 3-, 5-year OS and PFS were assessed with a bootstrapped sample, which showed the relationship between predicted risk and observed incidence. The prediction close to slope 1 and the intercept close to 0 indicates a good calibration. Thus our results showed that ApoA-I had an impact on survival prediction.

Discussion

Our study is the first to investigate the prognostic value of serum lipids and ApoA-I levels in patients with DLBCL. These results suggested that IPI-A may be a reliable tool for predicting DLBCL patients, and therapeutic strategies targeting ApoA-I might hold promise for improving DLBCL treatment.

Serum lipid levels are demonstrated to be associated with certain metabolic states and multiple disorders, such as advanced cancer [10, 11]. Lipids analysis is a promising direction in pathogenesis, mechanisms and clinical treatment of tumors [12, 13]. ApoA-I, the main component of HDL produced in the liver [14],

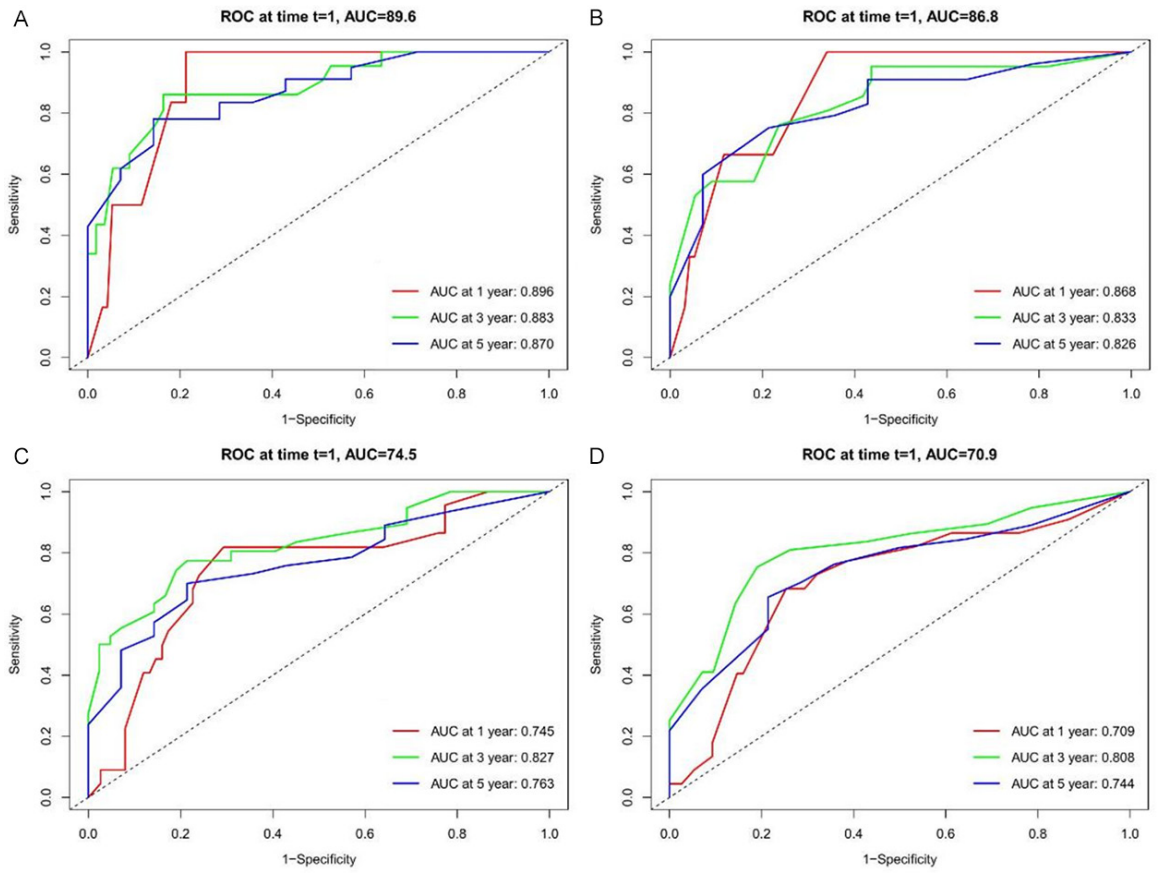

Figure 5. ROC curves and AUC values. (A, B) Construction and evaluation of the IPI-A (A) and IPI (B) of OS with each AUC of the 1-, 3- and 5-year predictive effect above. (C, D) The AUC values of ROC predicted 1-, 3- and 5-year PFS rates of IPI-A (C) and IPI (D) indexes.

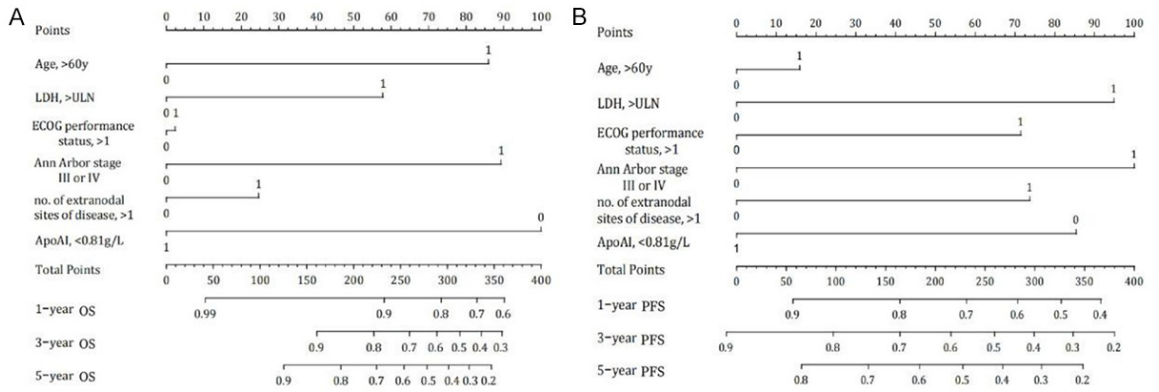

Figure 6. Construction of nomogram model. A. Nomogram predicting 1-, 3- and 5-year OS for patients with IPI-A. B. Nomogram predicting 1-, 3- and 5-year PFS for patients with IPI-A. The nomogram was applied by adding up the points identified on the points scale for each variable to a total points amount. Finally, beneath the total points, the probability of 1-, 3- or 5-year survival was projected on the bottom scales.

plays several well-documented cardioprotective functions and anti-tumorigenic activities [15]. Besides mediating cholesterol trafficking, it is involved in the innate humoral immune response and has anti-inflammatory potential as well as anti-oxidant capacity [16]. Several studies reported that alterations in ApoA-I can have a wide variety of effects in many kinds of

tumors [17, 18] and that serum ApoA-I levels may be a potential clinical prognostic marker [19, 20]. Zamanian-Daryoush et al. [21] demonstrated that the pharmacological delivery of ApoA-I was usually functional in terms of reducing tumor burden and retarding metastasis in preclinical studies. A retrospective study showed that ApoA has prognostic value and is a potential therapy for patients with chronic lymphocytic leukemia [22]. There have been no studies regarding ApoA-I on DLBCL. Our study found that patients with low ApoA-I more likely to present with stage III or IV disease and extranodal sites (Table S1). The low levels of ApoA-I were significantly associated with more severe clinical presentation and worse PFS and OS in DLBCL patients. Furthermore, patients who achieved CR after chemotherapy were significantly associated with higher ApoA-I levels. Our study showed that ApoA-I plays an essential role in DLBCL.

Imbalance in cholesterol metabolism and homeostasis, resulting in reduced cholesterol efflux from the tumor cells and increased cholesterol influx into the tumor cells, is a hallmark characteristic of cancer with lipid metabolism dependence. Several possible mechanisms could underly the part of ApoA-I in inhibiting tumor progression. First, ApoA-I may participate in tumor development by regulating lipid metabolism [23]. Second, decreased ApoA-I can lead to extravasation of cholesterol efflux [24] and monocyte subsets [25, 26]. The former may induce the binding of macrophages and phospholipids, which play an anti-inflammatory role in the tumor microenvironment [26, 27]. ApoA-I binds lysophosphatidic acid (LPA), an activator of tumor proliferation, which can inhibit LPA-induced tumor formation [18]. Conversely, the inflammatory response reduces ApoA-I synthesis and secretion, and low serum ApoA-I concentration indirectly causes increased cytokine production and induces a robust immune response [14, 27]. Supplementation with ApoA-I mimetics weakens the pro-inflammatory effects of endotoxins (lipopolysaccharides), cyclooxygenase 2, and the production of bioactive lipids [28]. Third, ApoA-I may suppress granulocytic polymorphonuclear neutrophils (PMN) myeloid-derived suppressor cell (MDSC) differentiation and inhibit the accumulation of PMN-MDSCs in tumor tissue. ApoA-I weakens the immunosuppressive function of MDSCs and regulates the transcription 3 signaling pathway in PMN-MDSCs [29]. Mangaraj et al. [30] reported that low ApoA-I concentrations were found in acute lymphoblastic leukemia with more pro-inflammatory and pro-angiogenic activity. Our study's findings showed a statistical difference in ApoA-I levels between pre-therapy and post-therapy, which supports previous research. Thus, ApoA-I may be a potential therapy for DLBCL and may improve therapeutic efficacy. The suppressive effect and mechanism of ApoA-I in DLBCL deserve further study.

For the past 30 years, the IPI remains the most powerful tool for risk stratification, assisting treatment regimen selection, and predicting the prognosis of DLBCL patients [31]. Patients with the same IPI score may respond differently to treatments and have different survival outcomes. Most importantly, the IPI cannot be used to identify patients at very high risk or discern biological heterogeneity [32]. The IPI was incorporated into our nomogram developed for predicting survival. Integrating the assessment of IPI-A might markedly improve the predictive power of IPI in patients with DLBCL. Our results showed that the IPI-A prediction model had a better clinical application value than the traditional IPI for survival rates. No statistically significant difference was observed between the IPI-A and IPI groups, which may be related to a limited DLBCL sample size. Our findings indicated that a convenient serological indicator (i.e., ApoA-I) incorporated into the IPI score seems more precise than the traditional IPI score in predicting OS and PFS. Thus, serum lipid markers could be applied for therapeutic monitoring and prognostic prediction in DLBCL patients.

Although these results suggested that ApoA-I is an independent predictor of outcomes and that the IPI-A score might be valuable for clinicians in treatment strategy selection and prognosis prediction for DLBCL, our studies also suffer from some limitations. First, a retrospective study has intrinsic limitations and biases in data collection. Secondly, the database used for training came from a single cancer center which could limit the generalizability of our study. Large-scale, multicenter randomized controlled studies are warranted to validate the causality and the prediction value.

Our study demonstrated that ApoA-I functions as an independent prognostic factor in DLBCL patients undergoing first-line therapy. We also introduced ApoA-I as a novel prognostic index to establish the IPI-A for risk prediction in DLBCL. Our results have a valuable understanding of DLBCL pathogenesis and far-reaching implications in clinical practice.

Acknowledgements

We thank the patients who participated in this study and Editage (www.editage.com) for its linguistic assistance during the preparation of this manuscript. LY is supported by the National Natural Science Foundation of China 81460030 and 81770221, and Leading Talent Foundation of Jiangxi Province 20225BCJ22001.

All the participants provided written informed consent.

Disclosure of conflict of interest

None.

Address correspondence to: Dr. Li Yu, Department of Hematology, The Second Affiliated Hospital of Nanchang University, Nanchang 330006, Jiangxi, China. Tel: +86-791-86300947; Fax: +86-919-668-7568; E-mail: ndefy02021@ncu.edu.cn; zengyulii@126.com

References

[1] Sehn LH and Salles G. Diffuse large B-cell lymphoma. N Engl J Med 2021; 384: 842-858.
[2] Beloribi-Djefaflia S, Vasseur S and Guillaumond F. Lipid metabolic reprogramming in cancer cells. Oncogenesis 2016; 5: e189.
[3] Zhao W, Guan J, Horswell R, Li W, Wang Y, Wu X and Hu G. HDL cholesterol and cancer risk among patients with type 2 diabetes. Diabetes Care 2014; 37: 3196-3203.
[4] Pedersen KM, Colak Y, Bojesen SE and Nordestgaard BG. Low high-density lipoprotein and increased risk of several cancers: 2 population-based cohort studies including 116,728 individuals. J Hematol Oncol 2020; 13: 129.
[5] Kapadia B, Nanaji NM, Bhalla K, Bhandary B, Lapidus R, Beheshti A, Evens AM and Gartenhaus RB. Fatty acid synthase induced S6Kinase facilitates USP11-eIF4B complex formation for sustained oncogenic translation in DLBCL. Nat Commun 2018; 9: 829.
[6] Barre FPY, Claes BSR, Dewez F, Peutz-Kootstra C, Munch-Petersen HF, Gronbaek K, Lund AH, Heeren RMA, Come C and Cillero-Pastor B. Specific lipid and metabolic profiles of R-CHOP-resistant diffuse large B-cell lymphoma elucidated by matrix-assisted laser desorption ionization mass spectrometry imaging and in vivo imaging. Anal Chem 2018; 90: 14198-14206.
[7] Kobayashi T, Lam PY, Jiang H, Bednarska K, Gloury R, Murigneux V, Tay J, Jacquelot N, Li R, Tuong ZK, Leggatt GR, Gandhi MK, Hill MM, Belz GT, Ngo S, Kallies A and Mattarollo SR. Increased lipid metabolism impairs NK cell function and mediates adaptation to the lymphoma environment. Blood 2020; 136: 3004-3017.
[8] Liebow M, Larson MC, Thompson CA, Nowakowski GS, Call TG, Macon WR, Kay NE, Habermann TM, Slager SL and Cerhan JR. Aspirin and other nonsteroidal anti-inflammatory drugs, statins and risk of non-Hodgkin lymphoma. Int J Cancer 2021; 149: 535-545.
[9] Smyth L, Blunt DN, Gatov E, Nagamuthu C, Croxford R, Mozessohn L and Cheung MC. Statin and cyclooxygenase-2 inhibitors improve survival in newly diagnosed diffuse large B-cell lymphoma: a large population-based study of 4913 subjects. Br J Haematol 2020; 191: 396-404.
[10] Salvadori G and Longo VD. Diet comparison suggests a lipid imbalance can slow tumour growth. Nature 2021; 599: 206-207.
[11] Qin WH, Yang ZS, Li M, Chen Y, Zhao XF, Qin YY, Song JQ, Wang BB, Yuan B, Cui XL, Shen F, He J, Bi YF, Ning G, Fu J and Wang HY. High serum levels of cholesterol increase antitumor functions of nature killer cells and reduce growth of liver tumors in mice. Gastroenterology 2020; 158: 1713-1727.
[12] Wang S, Liu Y, Feng Y, Zhang J, Swinnen J, Li Y and Ni Y. A review on curability of cancers: more efforts for novel therapeutic options are needed. Cancers (Basel) 2019; 11: 1782.
[13] Huang PT, Einav S and Asquith CRM. PIKfyve: a lipid kinase target for COVID-19, cancer and neurodegenerative disorders. Nat Rev Drug Discov 2021; 20: 730.
[14] Georgila K, Vyrla D and Drakos E. Apolipoprotein A-I (ApoA-I), immunity, inflammation and cancer. Cancers (Basel) 2019; 11: 1097.
[15] Walldius G, de Faire U, Alfredsson L, Leander K, Westerholm P, Malmstrom H, Ivert T and Hammar N. Long-term risk of a major cardiovascular event by apoB, apoA-1, and the apoB/apoA-1 ratio-experience from the Swedish AMORIS cohort: a cohort study. PLoS Med 2021; 18: e1003853.
[16] Catapano AL, Pirillo A, Bonacina F and Norata GD. HDL in innate and adaptive immunity. Cardiovasc Res 2014; 103: 372-383.
[17] Zhou Y and Luo G. Apolipoproteins, as the carrier proteins for lipids, are involved in the devel-

opment of breast cancer. Clin Transl Oncol 2020; 22: 1952-1962.
[18] Su F, Kozak KR, Imaizumi S, Gao F, Amneus MW, Grijalva V, Ng C, Wagner A, Hough G, Farias-Eisner G, Anantharamaiah GM, Van Lenten BJ, Navab M, Fogelman AM, Reddy ST and Farias-Eisner R. Apolipoprotein A-I (apoA-I) and apoA-I mimetic peptides inhibit tumor development in a mouse model of ovarian cancer. Proc Natl Acad Sci U S A 2010; 107: 19997-20002.
[19] Ma XL, Gao XH, Gong ZJ, Wu J, Tian L, Zhang CY, Zhou Y, Sun YF, Hu B, Qiu SJ, Zhou J, Fan J, Guo W and Yang XR. Apolipoprotein A1: a novel serum biomarker for predicting the prognosis of hepatocellular carcinoma after curative resection. Oncotarget 2016; 7: 70654-70668.
[20] Mao M, Wang X, Sheng H, Liu Y, Zhang L, Dai S and Chi PD. A novel score based on serum apolipoprotein A-1 and C-reactive protein is a prognostic biomarker in hepatocellular carcinoma patients. BMC Cancer 2018; 18: 1178.
[21] Zamanian-Daryoush M, Lindner D, Tallant TC, Wang Z, Buffa J, Klipfell E, Parker Y, Hatala D, Parsons-Wingerter P, Rayman P, Yusufishaq MSS, Fisher EA, Smith JD, Finke J, DiDonato JA and Hazen SL. The cardioprotective protein apolipoprotein A1 promotes potent anti-tumorigenic effects. J Biol Chem 2013; 288: 21237-21252.
[22] Yun X, Sun X, Hu X, Zhang H, Yin Z, Zhang X, Liu M, Zhang Y and Wang X. Prognostic and therapeutic value of apolipoprotein a and a new risk scoring system based on apolipoprotein a and adenosine deaminase in chronic lymphocytic leukemia. Front Oncol 2021; 11: 698572.
[23] Liu D, Ding Z, Wu M, Xu W, Qian M, Du Q, Zhang L, Cui Y, Zheng J, Chang H, Huang C, Lin D and Wang Y. The apolipoprotein A-I mimetic peptide, D-4F, alleviates ox-LDL-induced oxidative stress and promotes endothelial repair through the eNOS/HO-1 pathway. J Mol Cell Cardiol 2017; 105: 77-88.
[24] Zhang T, Wang Q, Wang Y, Wang J, Su Y, Wang F and Wang G. AIBP and APOA-I synergistically inhibit intestinal tumor growth and metastasis by promoting cholesterol efflux. J Transl Med 2019; 17: 161.
[25] Patel VK, Williams H, Li SCH, Fletcher JP and Medbury HJ. Monocyte subset recruitment marker profile is inversely associated with blood ApoA1 levels. Front Immunol 2021; 12: 616305.
[26] Penson PE, Long DL, Howard G, Toth PP, Muntner P, Howard VJ, Safford MM, Jones SR, Martin SS, Mazidi M, Catapano AL and Banach M. Associations between very low concentrations of low density lipoprotein cholesterol, high sensitivity C-reactive protein, and health outcomes in the Reasons for Geographical and Racial Differences in Stroke (REGARDS) study. Eur Heart J 2018; 39: 3641-3653.
[27] Sathiyakumar V, Kapoor K, Jones SR, Banach M, Martin SS and Toth PP. Novel therapeutic targets for managing dyslipidemia. Trends Pharmacol Sci 2018; 39: 733-747.
[28] Daskou M, Sharma M, Mu W, Heymans R, Ritou E, Rezek V, Hamid P, Kossyvakis A, Sen RS, Grijalva V, Chattopadhyay A, Papesh J, Meriwether D, Kitchen SG, Fogelman AM, Reddy ST and Kelesidis T. ApoA-I mimetics favorably impact cyclooxygenase 2 and bioactive lipids that may contribute to cardiometabolic syndrome in chronic treated HIV. Metabolism 2021; 124: 154888.
[29] Peng M, Zhang Q, Liu Y, Guo X, Ju J, Xu L, Gao Y, Chen D, Mu D and Zhang R. Apolipoprotein A-I mimetic peptide L-4F suppresses granulocytic-myeloid-derived suppressor cells in mouse pancreatic cancer. Front Pharmacol 2020; 11: 576.
[30] Mangaraj M, Nanda R and Panda S. Apolipoprotein A-I: a molecule of diverse function. Indian J Clin Biochem 2016; 31: 253-259.
[31] International Non-Hodgkin's Lymphoma Prognostic Factors Project. A predictive model for aggressive non-Hodgkin's lymphoma. N Engl J Med 1993; 329: 987-994.
[32] Lossos IS and Morgensztern D. Prognostic biomarkers in diffuse large B-cell lymphoma. J Clin Oncol 2006; 24: 995-1007.

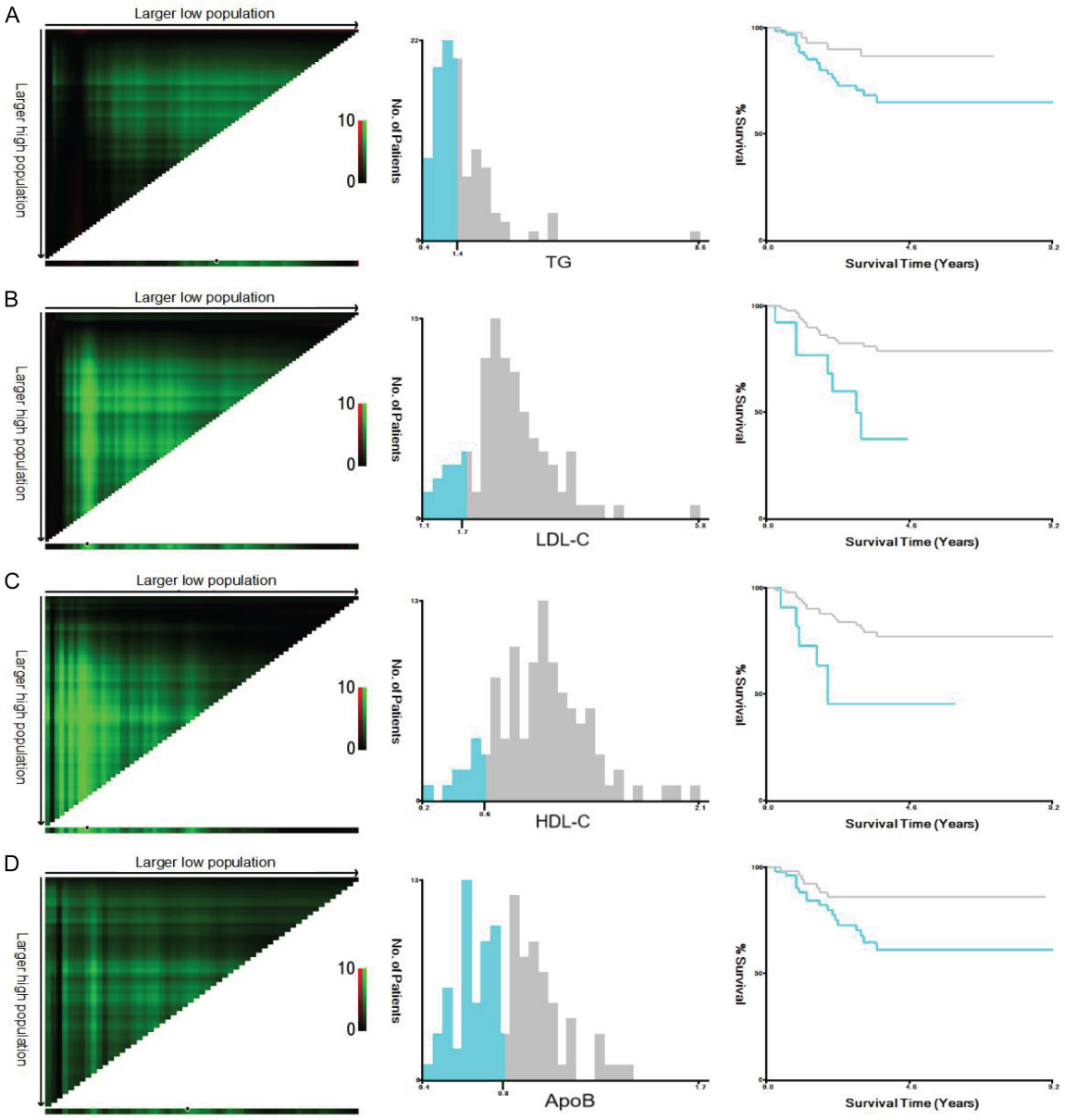

Figure S1. X-Tile analysis of OS according to TG, LDL, HDL and ApoB. A. The estimated optimal cut-off point of TG was 1.43 g/L; B. LDL was 1.74 mmol/L; C. HDL was 0.61 mmol/L; D. ApoB was 0.76 g/L. Abbreviation: TG, triacylglycerol; LDL-C, Low-density Lipoprotein Cholesterol; HDL-C, High-Density Lipoprotein Cholesterol; ApoA-I, Apolipoprotein A-I; ApoB, Apolipoprotein B.

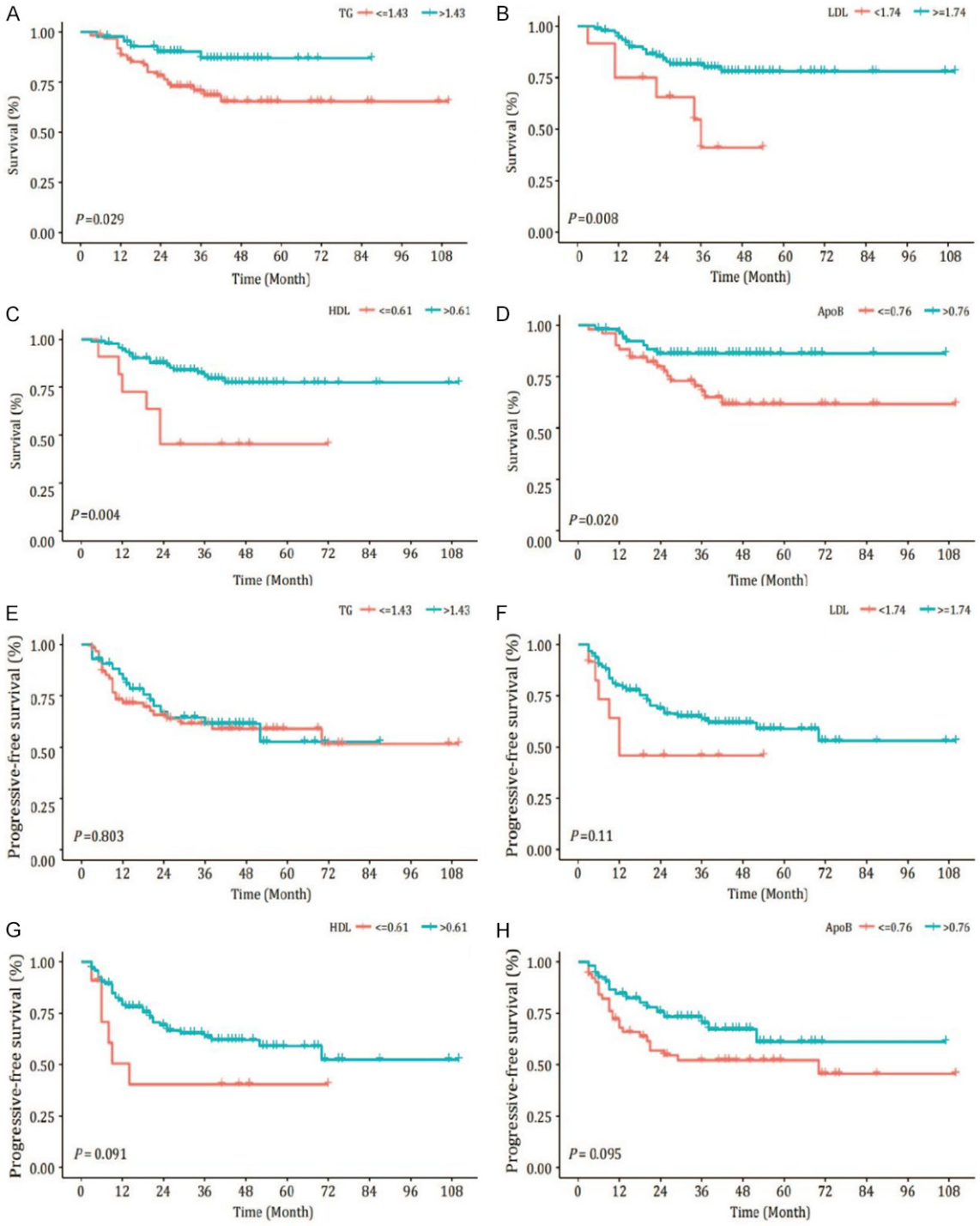

Figure S2. Kaplan-Meier curves for OS by TG levels (A), OS by LDL levels (B), OS by HDL levels (C), OS by ApoB levels (D), PFS by TG levels (E), PFS by LDL levels (F), PFS by HDL levels (G), PFS by ApoB levels (H). Abbreviation: TG, triacylglycerol; LDL-C, Low-Density Lipoprotein Cholesterol; HDL-C, High-Density Lipoprotein Cholesterol; ApoB, Apolipoprotein B.

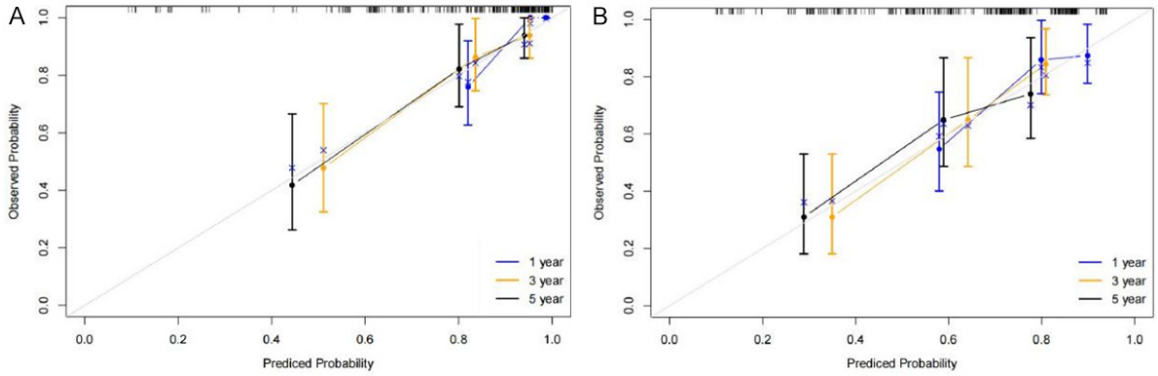

Figure S3. A. Calibration curve of IPI-A model in OS; B. Calibration curve of IPI-A model in PFS.

Table S1. The Ann Arbor stage and Extranodal sites of patients with different ApoA-I levels

Variable	ApoA-I ≤ 0.81	ApoA-I > 0.81	*P*-value
Ann Arbor stage III or IV, n (%)	28 (75.5)	33 (48.8)	0.007
Extranodal sites (≥ 2)	9 (24.3)	6 (8.8)	0.03