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Remnant cholesterol predicts the development of type 2 diabetes in patients with nonalcoholic fatty liver disease

Liping Yang[1†], Hangkai Huang[2†], Zejun Wang[3*] and Chengfu Xu[2*]

Abstract

Background The predictive value of serum remnant cholesterol in cardiovascular disease has been widely recognized. This value has also been explored in metabolic disorders such as type 2 diabetes mellitus and nonalcoholic fatty liver disease (NAFLD). However, whether remnant cholesterol can predict the risk of incident diabetes in NAFLD patients remains unclear.

Methods This study included adults who underwent health examinations from 2004 to 2015. NAFLD was diagnosed via abdominal ultrasonography with the exclusion of other causes of chronic liver disease. Cox proportional hazards regression analyses were performed to investigate the associations between baseline remnant cholesterol and diabetes risk in NAFLD patients and NAFLD-free participants.

Results A total of 15,464 participants were included in this study, and 2,741 adults had NAFLD. During the 93,537 person-years of follow-up, 233 cases and 150 cases of incident diabetes were recorded among NAFLD patients and NAFLD-free individuals, respectively. Compared with the first quartile, the fourth quartile of remnant cholesterol was positively associated with the risk of diabetes in NAFLD patients (HR: 1.68, 95% CI: 1.13 – 2.51; $P < 0.001$). However, this association was not significant in NAFLD-free individuals.

Conclusions High remnant cholesterol was associated with an increased risk of incident diabetes in NAFLD patients but not in NAFLD-free individuals.

Keywords Nonalcoholic fatty liver disease, Type 2 diabetes mellitus, Remnant cholesterol, Prospective study

Introduction

Nonalcoholic fatty liver disease (NAFLD) has become a global epidemic, particularly affecting populations with type 2 diabetes mellitus and obesity [1]. The global prevalence of NAFLD was 25% in 2016 and increased to 32% in 2022 [2]. NAFLD can potentially evolve from simple steatosis to steatohepatitis, fibrosis, and cirrhosis. The severity of liver histological injury in patients with NAFLD is closely related to the risk of cardiovascular disease [3] and intrahepatic and extrahepatic cancers [4]. Among NAFLD individuals without cirrhosis and those with cirrhosis, the incidence rate of hepatocellular carcinoma has

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increased from 1.25 per 1000 person-years to 14.46 per 1000 person-years [5]. The burden of end-stage liver disease becomes greater in patients with NAFLD, which is positively related to age and the course of diabetes [6].

The incremental burden of NAFLD was propelled by the substantial increase in the prevalence of diabetes [7]. Globally, the number of adults afflicted with diabetes has quadrupled in the past three decades [8]. The complications of diabetes are very common, with macrovascular complications occurring in 27% of diabetes patients and microvascular complications occurring in half of diabetes patients [9]. Patients with diabetes have a shortened life expectancy and impaired quality of life [10]. Accumulating evidence suggests that NAFLD promotes the development of diabetes [11] and that the risk of diabetes becomes more pronounced as the severity of hepatic steatosis and fibrosis increases [12]. While NAFLD is widely recognized to promote the development of diabetes, it is important to note that the pathogenesis of NAFLD is highly heterogeneous. Not all individuals with NAFLD exhibit insulin resistance or an elevated risk of cardiometabolic diseases. Recent studies highlight that NAFLD encompasses distinct subtypes, some of which are more strongly linked to metabolic dysfunction, whereas others may arise from nonmetabolic factors such as genetic predisposition or environmental influences [13, 14]. This variability underscores the need for further stratification of NAFLD populations to better understand the relationship between remnant cholesterol and diabetes risk. Our findings specifically suggest that remnant cholesterol may serve as a predictive marker for diabetes in NAFLD patients with underlying metabolic dysregulation, but its utility may vary across NAFLD subtypes.

Typical dyslipidemia is common in both NAFLD and diabetes [1]. However, in recent years, increasing attention has been given to nonconventional lipid parameters, such as remnant cholesterol [15]. In large cohort and genetic studies, serum remnant cholesterol has been reported to predict the adverse outcomes of cardiovascular diseases, even among patients who have achieved the optimal goals for conventional lipids recommended by the guidelines [16–18]. Remnant cholesterol is the cholesterol content of triglyceride-rich lipoproteins, which consist of intestinally derived chylomicrons and hepatically derived very low-density lipoprotein-cholesterols [19]. During the formation of atherosclerotic plaques, triglycerides are metabolized by macrophages, whereas cholesterols are not metabolized and accumulate gradually [20]. Therefore, remnant cholesterol, rather than triglyceride levels in triglyceride-rich lipoproteins, is more critical in atherogenesis. Notably, some epidemiological studies indicate a positive association between remnant cholesterol and NAFLD [21] as well as diabetes [22]. In cross-sectional studies, an independent relationship was identified for serum remnant cholesterol and the risk of NAFLD [23]. Observational studies have also revealed that remnant cholesterol is associated with the new onset of diabetes [24] and its macrovascular [25] and microvascular complications [26]. However, whether remnant cholesterol can predict the risk of incident diabetes in patients with NAFLD remains unclear.

In this study, we aimed to explore the association between serum remnant cholesterol and the risk of developing diabetes in the NAFLD population.

Methods

Study design

Data were obtained from the NAGALA study conducted at Murakami Memorial Hospital (Gifu, Japan) in 1994. This longitudinal study enrolled subjects from the general population who participated in health examinations from 2004 to 2015. A total of 15,464 participants were included according to the following exclusion criteria: (i) had viral hepatitis or other known liver disease other than NAFLD; (ii) were taking any agents at baseline; (iii) had diabetes or impaired fasting glucose at baseline; and (iv) had incomplete covariate data (Fig. 1). The NAGALA study was approved by the Murakami Memorial Hospital Institutional Ethics Review Committee (IRB2018-09-01), and informed consent was provided by all participants. This study was a secondary analysis of the NAGALA study and was approved by the Hospital Ethics Committee of the First Affiliated Hospital, Zhejiang University School of Medicine (IIT20221299).

Data collection

A detailed description of the data collection process has been published [27]. A standardized questionnaire was used to collect information on smoking, drinking, physical activity and medical history. Smoking status was categorized as nonsmoker, former smoker and current smoker. The average weekly ethanol intake was evaluated by the number and type of alcoholic beverages consumed in the previous month. Individuals were recognized as regular exercisers if they engaged in any type of physical activity more than once a week. Anthropometric features, including height, weight and blood pressure, were measured by experienced nurses. Venous blood samples were taken from the participants after an overnight fast to test their biochemical parameters. Remnant cholesterol was equal to total cholesterol minus HDL-C and minus LDL-C [18].

Diagnosis of NAFLD and diabetes

The definition of NAFLD was based on abdominal ultrasonography after excluding other causes of chronic liver disease, including viral hepatitis (defined by measurements of hepatitis B antigen and hepatitis C antibody),

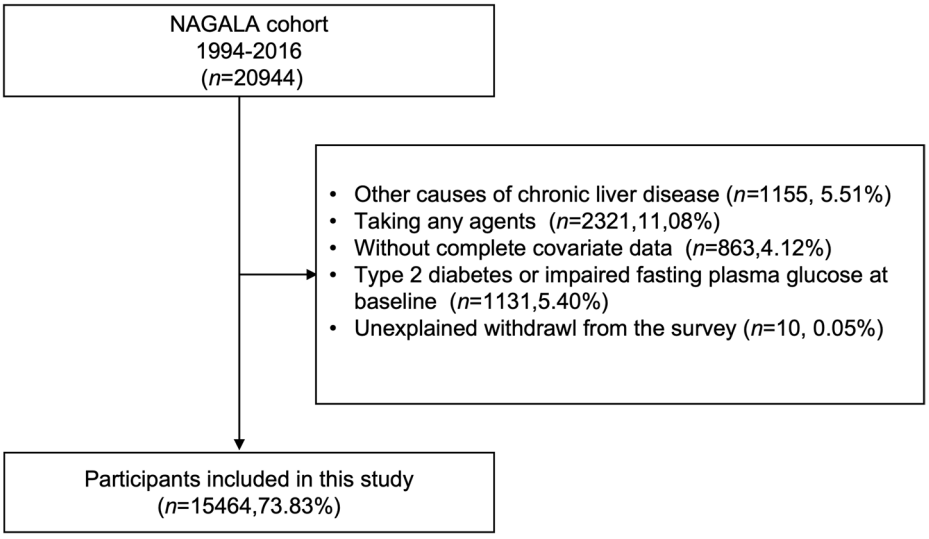

Fig. 1 Flowchart of this study

autoimmune liver diseases (e.g., primary biliary cholangitis), or other known chronic liver diseases in addition to NAFLD. The ultrasonographic presentation of fatty liver includes liver and kidney echo contrast, deep liver attenuation, liver brightness and vessel blur [28]. Diabetes was defined as fasting plasma glucose ≥ 7 mmol/L, HbA1c ≥ 6.5% or self-reported [29].

Statistical analysis

Continuous variables are presented as the means (standard deviations) or medians (interquartile ranges), and categorical variables are presented as percentages. We classified participants into different groups according to remnant cholesterol quartile. Comparisons between groups were performed via ANOVA, the Kruskal–Wallis rank-sum test or the chi-square test. Cox proportional hazards regression analyses were performed to assess the associations between remnant cholesterol quartile and diabetes risk with or without adjustment for potential confounders, including age, sex, BMI, systolic and diastolic blood pressure, smoking, alcohol intake, physical activity and LDL-C. This association was analyzed in the whole cohort, in NAFLD patients, and in NAFLD-free individuals. The proportional hazards assumption was tested via Schoenfeld residuals, with no significant violations observed (global test $P = 0.21$). Kaplan–Meier curves were drawn stratified by remnant cholesterol quartile, and the log-rank test was used for comparisons between groups.

To further explore the predictive value of remnant cholesterol for diabetes, the hazard ratio (HR) and 95% confidence interval (95% CI) were calculated at 4, 8, and 12 years of follow-up. We performed a sensitivity analysis excluding participants with a follow-up duration < 2 years ($n = 1,203$) to address potential surveillance bias in early diabetes detection. The multivariate Cox model was rerun using the same covariates as those used in the primary analysis. We also performed subgroup analyses stratified by age, sex, BMI, smoking status and physical activity. Multiplicative interaction analyses were performed by creating product terms between remnant cholesterol quartiles and stratification variables (age, sex, BMI, smoking status, and physical activity) in the fully adjusted model. The statistical significance of interactions was evaluated via likelihood ratio tests comparing models with and without the interaction terms. All the statistical analyses were performed via SPSS 26.0 software (SPSS Inc., Chicago, IL). $P < 0.05$ (2-tailed test) was considered statistically significant.

Results

Baseline characteristics

A total of 15,464 participants were included in this study, including 2,741 NAFLD individuals and 12,723 NAFLD-free individuals at baseline. Table 1 shows the clinical features of participants stratified by remnant cholesterol quartile. Adults in higher remnant cholesterol quartiles accounted for a greater proportion of drinkers and smokers, regardless of NAFLD status. In addition, they had higher BMIs; systolic and diastolic blood pressure; total cholesterol, LDL-C and fasting plasma glucose; and lower HDL-C levels.

Follow-up outcomes

During a follow-up of 93,537 person-years, 373 incident cases of diabetes were identified. Among them, 223 cases were recorded in patients with NAFLD. Participants in higher remnant cholesterol quartiles had an incrementally higher incidence rate of diabetes (Table 2).

Table 1 Baseline characteristics of the study population

		Participants without NAFLD at baseline ($n=12723$)				Participants with NAFLD at baseline ($n=2741$)			
		Quartile of remnant cholesterol				Quartile of remnant cholesterol			
Variables	All	Q1 ($n=2965$)	Q2 ($n=3190$)	Q3 ($n=3263$)	Q4 ($n=3305$)	Q1 ($n=733$)	Q2 ($n=624$)	Q3 ($n=684$)	Q4 ($n=700$)
Age (years)	42 (37 – 50)	39 (35 – 45)	41 (36 – 49)	43 (37 – 51)	46 (39 – 53)	44 (38 – 51)	45 (39 – 51)	44 (38 – 51)	43 (38 – 50)
Gender (male, %)	45.5	75.6	59.2	46.0	27.7	27.7	20.8	13.5	12.5
Body mass index (kg/m^2)	21.79 (19.89 – 23.92)	20.04 (18.65 – 21.63)	20.84 (19.27 – 22.49)	21.35 (19.78 – 23.10)	22.48 (20.91 – 24.18)	24.59 (22.70 – 26.67)	24.87 (23.21 – 27.17)	25.23 (23.63 – 27.33)	25.65 (23.88 – 27.63)
Drinkers (%)	23.7	12.8	21.4	25.1	33.9	18.8	21.8	24.6	30.1
Smoking status (%)									
Never	58.4	78.0	68.2	57.2	44.0	54.3	47.4	40.2	36.7
Past smoker	19.1	11.5	15.9	19.1	22.9	24.6	28.7	27.6	25.4
Current smoker	22.5	10.5	16.0	23.8	33.2	21.1	23.9	32.2	37.9
Physical activity (%)									
Nonregular exerciser	82.5	82.7	79.8	82.4	82.6	84.4	83.8	86.0	87.1
Regular exerciser	17.5	17.3	20.2	17.6	17.4	15.6	16.2	14.0	12.9
Systolic blood pressure (mmHg)	113 (104 – 124)	106 (99 – 115)	110 (102 – 120)	113 (103 – 123)	117 (108 – 127)	120 (111 – 130)	121 (113 – 132)	123 (114 – 133)	126 (116 – 135)
Diastolic blood pressure (mmHg)	71 (64 – 78)	66 (61 – 72)	69 (62 – 75)	71 (64 – 77)	74 (68 – 81)	75 (69 – 82)	77 (71 – 84)	78 (72 – 85)	80 (73 – 87)
Total cholesterol (mmol/L)	5.07 (4.50 – 5.66)	4.60 (4.14 – 5.12)	4.89 (4.40 – 5.43)	5.09 (4.58 – 5.66)	5.40 (4.86 – 5.97)	5.07 (4.47 – 5.63)	5.30 (4.78 – 5.82)	5.53 (4.99 – 6.13)	5.73 (5.22 – 6.31)
HDL-C (mmol/L)	1.41 (1.16 – 1.71)	1.66 (1.45 – 1.93)	1.58 (1.34 – 1.84)	1.45 (1.24 – 1.71)	1.24 (1.05 – 1.45)	1.32 (1.15 – 1.53)	1.18 (1.04 – 1.37)	1.11 (0.98 – 1.27)	1.00 (0.88 – 1.16)
LDL-C (mmol/L)	3,43 (2.88 – 4.02)	2.83 (2.45 – 3.26)	3.17 (2.73 – 3.63)	3.45 (2.98 – 3.96)	3.83 (3.33 – 4.39)	3.55 (3.07 – 4.09)	3.88 (3.41 – 4.35)	4.11 (3.61 – 4.64)	4.22 (3.65 – 4.77)
Fasting plasma glucose (mmol/L)	5.16 (4.89 – 5.44)	4.94 (4.72 – 5.16)	5.05 (4.78 – 5.33)	5.11 (4.88 – 5.44)	5.27 (5.00 – 5.55)	5.33 (5.11 – 5.61)	5.38 (5.16 – 5.66)	5.38 (5.16 – 5.66)	5.50 (5.22 – 5.72)
Glycated hemoglobin A1c (%)	5.20 (5.00 – 5.40)	5.10 (4.90 – 5.40)	5.10 (4.90 – 5.40)	5.10 (4.90 – 5.40)	5.20 (4.90 – 5.40)	5.30 (5.10 – 5.50)	5.25 (5.10 – 5.54)	5.25 (5.04 – 5.50)	5.35 (5.10 – 5.50)
Remnant cholesterol (mmol/L)	0.14 (0.10 – 0.22)	0.07 (0.06 – 0.08)	0.11 (0.10 – 0.12)	0.16 (0.14 – 0.17)	0.25 (0.22 – 0.32)	0.14 (0.11 – 0.16)	0.21 (0.19 – 0.23)	0.30 (0.27 – 0.32)	0.47 (0.40 – 0.57)

Data are presented as median (interquartile range) or as proportions

HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol

Table 2 Hazard ratios for type 2 diabetes based on quartiles of remnant cholesterol at baseline

	Events/person-year	Multivariate model	
		Model 1	Model 2
All participants			
Q1	53/21,087	REF	REF
Q2	71/22,665	0.68 (0.97 – 1.38)	0.93 (0.65 – 1.34)
Q3	90/24,562	0.68 (0.96 – 1.35)	0.84 (0.59 – 1.19)
Q4	159/25,223	1.24 (0.90 – 1.70)	0.99 (0.71 – 1.38)
P for trend		< 0.001	0.013
Participants without NAFLD at baseline			
Q1	13/16,824	REF	REF
Q2	26/18,943	1.21 (0.62 – 2.37)	1.19 (0.61 – 2.34)
Q3	37/20,380	1.26 (0.66 – 2.42)	1.18 (0.61 – 2.28)
Q4	74/20,934	1.85 (0.99 – 3.46)	1.62 (0.84 – 3.14)
P for trend		0.013	0.022
Participants with NAFLD at baseline			
Q1	40/4262	REF	REF
Q2	45/3722	1.21 (0.79 – 1.86)	1.21 (0.79 – 1.85)
Q3	53/4182	1.28 (0.84 – 1.94)	1.21 (0.80 – 1.84)
Q4	85/4289	1.98 (1.35 – 2.91)***	1.68 (1.13 – 2.51)**
P for trend		< 0.001	0.001

Model 1 was adjusted for age, sex and BMI at baseline

Model 2 was additionally adjusted for systolic blood pressure, diastolic blood pressure, smoking, alcohol intake, physical activity and low-density lipoprotein-cholesterol

* $P < 0.05$, ** $P < 0.005$, *** $P < 0.001$

We investigated the predictive value of remnant cholesterol for diabetes in the whole cohort. No significant association was found between baseline remnant cholesterol and incident diabetes risk (HR: 0.99, 95% CI: 0.71 – 1.38). To assess the effect of NAFLD status on this association, we divided all participants into groups with or without NAFLD. According to the fully adjusted model, compared with patients in the first quartile of remnant cholesterol, NAFLD patients in the fourth quartile presented a 1.68-fold greater risk of developing diabetes (HR: 1.68, 95% CI: 1.13 – 2.51). In non-NAFLD individuals, the hazard ratio (HR) (95% CI) for incident diabetes was 4.17 (2.31 – 7.52) according to the univariate model. When further adjusting for potential confounding factors, no significant associations were observed (HR: 1.62, 95% CI: 0.84 – 3.14). These results indicated that the predictive value of remnant cholesterol for diabetes was found only in NAFLD patients but not in non-NAFLD individuals.

K–M analysis revealed that individuals with higher remnant cholesterol levels had a greater probability of developing diabetes, regardless of NAFLD status (log-rank $P < 0.001$, Fig. 2).

We further explored the relationship between remnant cholesterol and incident diabetes in NAFLD patients stratified by follow-up time (Table 3). Compared with the lowest quartile, the highest quartile of remnant cholesterol was positively associated with diabetes risk after 4 years of follow-up. This association remained significant when the follow-up time was extended to 8 years and 12 years. These findings suggest that the role of remnant cholesterol in predicting the development of diabetes in patients with NAFLD has started to emerge at a relatively early stage. After 2919 participants with less than 2 years of follow-up were excluded, the association between remnant cholesterol and diabetes in NAFLD patients remained robust (Supplementary Table S1).

For subgroup analyses, we classified NAFLD participants into different groups according to age, sex, BMI, smoking status and physical activity (Table 4). We observed that the positive association between the fourth quartile of remnant cholesterol and diabetes was significant in individuals aged ≥ 40 years (HR: 1.79, 95% CI:

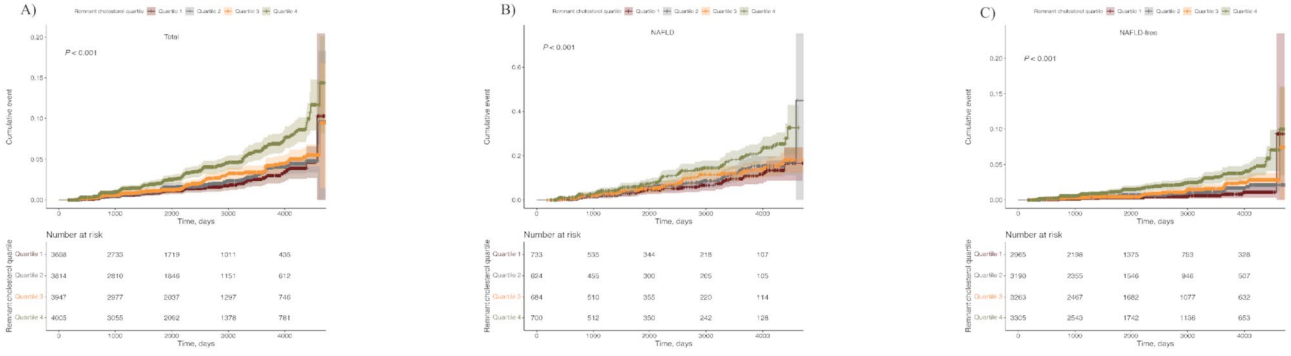

Fig. 2 Kaplan–Meier curves of quartiles of remnant cholesterol to illustrate the development of type 2 diabetes. The log-rank *P* value indicates the between-group differences in event probabilities. (**A**) All individuals; (**B**) NAFLD patients; and (**C**) non-NAFLD individuals

Table 3 The association between quartiles of remnant cholesterol and type 2 diabetes in NAFLD individuals stratified by follow-up time

	Events/person-year	Multivariate model	
		Model 1	Model 2
4 years of follow-up			
Q1	14/638	REF	REF
Q2	13/535	0.91 (0.42 – 1.94)	0.98 (0.46 – 2.12)
Q3	19/543	1.26 (0.62 – 1.57)	1.20 (0.57 – 2.50)
Q4	26/519	1.07 (1.07 – 4.01)*	2.06 (1.01 – 4.20)*
P for trend		0.007	0.016
8 years of follow-up			
Q1	28/1797	REF	REF
Q2	30/1413	1.19 (0.76 – 1.99)	1.21 (0.71 – 2.05)
Q3	45/1744	1.31 (0.81 – 2.10)	1.23 (0.75 – 2.01)
Q4	61/1578	2.22 (1.42 – 3.46)***	1.88 (1.16 – 3.06)**
P for trend		< 0.001	0.005
12 years of follow-up			
Q1	39/3847	REF	REF
Q2	44/3259	1.23 (0.80 – 1.90)	1.23 (0.80 – 1.91)
Q3	53/3585	1.33 (0.87 – 2.02)	1.24 (0.80 – 1.91)
Q4	83/3767	1.95 (1.32 – 2.89)***	1.78 (1.18 – 2.69)**
P for trend		< 0.001	0.003

Model 1 was adjusted for age, sex and BMI at baseline

Model 2 was additionally adjusted for systolic blood pressure, diastolic blood pressure, smoking, alcohol intake, physical activity and low-density lipoprotein-cholesterol

* $P < 0.05$, ** $P < 0.005$, *** $P < 0.001$

1.10 – 2.91), females (HR: 1.72, 95% CI: 1.09 – 2.70), those with a BMI < 25 kg/m^2 (HR: 2.10, 95% CI: 1.10 – 4.03), past smokers (HR: 2.42, 95% CI: 1.08 – 5.45) and non-regular exercisers (HR: 1.55, 95% CI: 1.01 – 2.39) but not in their counterparts. We further conducted multiplicative interaction analysis and found that the interaction of these conditions was not significant in the association of remnant cholesterol with diabetes.

Discussion

In this study, we found that there was a positive association between baseline remnant cholesterol and incident diabetes in the NAFLD population. First, an incrementally greater incidence of diabetes was observed in adults with higher remnant cholesterol levels. Second, serum remnant cholesterol was positively associated with the risk of incident diabetes in NAFLD patients. Third, the cumulative incidence risk of diabetes was statistically significant at 4, 8 and 12 years of follow-up.

Previous studies have reported a positive relationship between serum remnant cholesterol and NAFLD and diabetes [30, 31]. A cross-sectional study of 1,162 adolescents revealed that the first quartile of remnant cholesterol was associated with a 55% lower risk of NAFLD in females and an 85% lower risk in males than the fourth quartile was [23]. Another cross-sectional study involving 5,156 adults demonstrated that the highest quartile of remnant cholesterol was associated with a 1.714-fold greater risk of NAFLD [21]. More recently, a nonlinear correlation between remnant cholesterol and NAFLD was observed in a cross-sectional study of 3,370 adults [32]. The positive association was further validated in a cohort study of 31,662 person-years of follow-up [31]. In addition, a large cohort of 0.58 million Koreans reported that those in the fourth quartile of remnant cholesterol had almost twice the risk of developing diabetes [22]. For the complications of diabetes, adults in the third tertile of remnant cholesterol were 82% more likely to develop diabetic nephropathy [26] and 62% more likely to have a major adverse cardiovascular event than their counterparts were [33]. However, thus far, data on the association between remnant cholesterol and diabetes in patients with NAFLD are still limited. Diabetes is one of the most common extrahepatic complications of NAFLD, and reducing diabetes risk in these patients is highly challenging [34]. In this study, we revealed for the first time that NAFLD patients with higher remnant cholesterol levels were more likely to develop diabetes. These findings provide a rationale for screening remnant cholesterol to identify NAFLD patients who may be at increased risk of diabetes early. Furthermore, this risk could be targeted with remnant cholesterol-lowering agents such as angiopoietin-like 3 and apolipoprotein C III [35], which warrants investigation in the future. ANGPTL3 is a glycoprotein produced by the liver that inhibits lipoprotein lipase and endothelial lipase [36]. Apo C II is a cofactor for lipoprotein lipase, a plasma enzyme that hydrolyzes triglycerides [37]. ANGPTL3 and Apo C II are novel targets for lowering atherosclerotic risk in patients who tolerate lipid-lowering therapy [38].

A positive association between remnant cholesterol and diabetes was observed in NAFLD patients but not in NAFLD-free individuals. Remnant cholesterol is derived from an endogenous pathway through the liver and an exogenous pathway through the small intestine [39]. In the endogenous pathway, VLDL is produced in hepatocytes and secreted into the plasma. In the exogenous pathway, chylomicron from dietary fat also enters the circulation. A portion of the triglyceride content of both VLDL and chylomicron was hydrolyzed by lipoprotein

Table 4 Subgroup analyses of the association between remnant lipoprotein cholesterol and risk of incident type 2 diabetes in NAFLD individuals

	Events/person-year	Multivariate model		*P* for interaction
		Model 1	Model 2	
Stratified by age				0.634
Age < 40 years				
Q1	11/1272	REF	REF	
Q2	6/996	0.60 (0.22 – 1.64)	0.53 (0.194 – 1.47)	
Q3	9/1178	0.85 (0.35 – 2.07)	0.72 (0.289 – 1.77)	
Q4	20/20,934	1.69 (0.77 – 3.71)	1.21 (0.523 – 2.78)	
P for trend		0.048	0.255	
Age ≥ 40 years				
Q1	29/2990	REF	REF	
Q2	39/2726	1.46 (0.90 – 2.36)	1.37 (0.84 – 2.25)	
Q3	44/3004	1.43 (0.89 – 2.30)	1.26 (0.76 – 2.06)	
Q4	65/3082	2.08 (1.33 – 3.25)***	1.79 (1.10 – 2.91)**	
P for trend		0.001	0.020	
Stratified by gender				0.627
Male				
Q1	12/1110	REF	REF	
Q2	12/681	1.50 (0.67 – 3.35)	1.30 (0.56 – 3.03)	
Q3	10/497	1.66 (0.71 – 3.88)	1.30 (0.53 – 3.22)	
Q4	7/283	2.02 (0.78 – 5.24)	1.33 (0.48 – 3.75)	
P for trend		0.139	0.626	
Female				
Q1	28/3152	REF	REF	
Q2	33/3040	1.11 (0.67 – 1.84)	1.09 (0.66 – 1.82)	
Q3	43/3684	1.19 (0.74 – 1.91)	1.09 (0.66 – 1.77)	
Q4	78/4006	1.90 (1.23 – 2.93)**	1.72 (1.09 – 2.70)**	
P for trend		< 0.001	0.001	
Stratified by BMI				0.707
BMI < 25 kg/m^2				
Q1	18/2436	REF	REF	
Q2	18/1987	1.20 (0.62 – 2.30)	1.19 (0.61 – 2.32)	
Q3	16/2067	1.06 (0.54 – 2.11)	1.03 (0.51 – 2.10)	
Q4	27/1795	2.23 (1.20 – 4.17)*	2.10 (1.10 – 4.03)*	
P for trend		0.008	0.016	
BMI ≥ 25 kg/m^2				
Q1	22/1826	REF	REF	
Q2	27/1735	1.22 (0.69 – 2.16)	1.17 (0.66 – 2.08)	
Q3	37/2114	1.43 (0.84 – 2.44)	1.16 (0.67 – 2.03)	
Q4	58/2493	1.93 (1.17 – 3.19)**	1.57 (0.92 – 2.67)	
P for trend		0.004	0.061	
Stratified by smoking status				
Never				
Q1	16/2214	REF	REF	
Q2	25/1716	1.75 (0.92 – 3.30)	1.75 (0.91 – 3.36)	
Q3	18/1742	1.35 (0.68 – 2.67)	1.39 (0.68 – 2.82)	
Q4	20/1516	1.74 (0.88 – 3.44)	1.76 (0.86 – 3.62)	
P for trend		0.253	0.282	
Past smoker				0.401
Q1	9/1165	REF	REF	
Q2	8/1074	0.94 (0.36 – 2.45)	0.97 (0.37 – 2.55)	
Q3	11/1121	1.30 (0.54 – 3.13)	1.39 (0.56 – 3.44)	
Q4	22/1203	2.22 (1.01 – 4.86)*	2.42 (1.08 – 5.45)*	

Table 4 (continued)

	Events/person-year	Multivariate model		*P* for interaction
		Model 1	Model 2	
P for trend		0.013	0.008	
Current smoker				0.415
Q1	15/881	REF	REF	
Q2	12/931	0.76 (0.36 – 1.62)	0.60 (0.27 – 1.30)	
Q3	24/1318	0.98 (0.51 – 1.87)	0.76 (0.38 – 1.51)	
Q4	43/1570	1.44 (0.80 – 2.60)	1.13 (0.60 – 2.12)	
P for trend		0.047	0.138	
Stratified by physical activity				0.707
Nonregular exerciser				
Q1	38/3643	REF	REF	
Q2	35/3144	0.99 (0.63 – 1.58)	0.95 (0.60 – 1.52)	
Q3	50/3633	1.24 (0.81 – 1.91)	1.10 (0.70 – 1.71)	
Q4	78/3794	1.83 (1.22 – 2.73)**	1.55 (1.01 – 2.39)*	
P for trend		< 0.001	0.009	
Regular exerciser				
Q1	2/619	REF	REF	
Q2	10/578	4.86 (1.06 – 22.35)*	4.14 (0.87 – 19.67)	
Q3	3/549	1.47 (0.24 – 8.85)	1.13 (0.17 – 7.21)	
Q4	7/495	3.94 (0.81 – 19.21)	2.91 (0.52 – 16.19)	
P for trend		0.370	0.672	

Model 1 was adjusted for age, sex and BMI at baseline

Model 2 was additionally adjusted for systolic blood pressure, diastolic blood pressure, smoking, alcohol intake, physical activity and low-density lipoprotein-cholesterol

* $P < 0.05$, ** $P < 0.005$, *** $P < 0.001$

lipase. Lipoprotein lipase plays a key role in the metabolism of remnant cholesterol [40]. In animal models of NAFLD, the activity of lipoprotein lipase was reported to be inhibited [41]. We speculated that the reduced activity of lipoprotein lipase may be responsible for the positive association between remnant cholesterol and diabetes among NAFLD patients. The underlying mechanism remains unclear and needs to be explored in future studies.

The mechanisms by which remnant cholesterol is linked to diabetes remain unclear. There are several possible explanations. To date, insulin resistance has been considered the main mediator of this association [42]. Previous observational studies demonstrated that remnant cholesterol was closely related to insulin resistance, independent of traditional metabolic profiles [43]. Hepatic insulin resistance inhibits LDL receptor-related protein 1 from translocating to the hepatocellular membrane, which results in insufficient clearance of triglyceride-rich lipoproteins [44]. In addition, ectopic lipid deposition in pancreatic cells leads to β-cell dysfunction [45]. Second, high levels of remnant cholesterol indicate a proinflammatory status [24]. The overproduction of proinflammatory cytokines in the pancreas is one of the pathogeneses of diabetes [46]. Third, the metabolism of peripheral remnant cholesterol may be inhibited due to the reduced activity of lipoprotein lipase in diabetes [47]. The mechanisms by which NAFLD promotes the development of diabetes are multifaceted and may explain why elevated remnant cholesterol specifically exacerbates diabetes risk in this population. NAFLD is associated with systemic metabolic disturbances, including hepatic insulin resistance, chronic low-grade inflammation, and dysregulated lipid metabolism [48]. These alterations can impair insulin signaling in peripheral tissues (e.g., skeletal muscle and adipose tissue) while also promoting pancreatic β-cell dysfunction through lipotoxicity and oxidative stress [49].

This study has several limitations. First, this was an observational study and was unable to draw causal conclusions for the relationship between remnant cholesterol and diabetes among NAFLD patients. Second, serum levels of remnant cholesterol were only measured at baseline. While this approach is pragmatic for large cohort studies, it does not account for potential fluctuations in remnant cholesterol levels over time owing to lifestyle changes, medications, or disease progression. This limitation could bias our results in several ways: (1) if remnant cholesterol levels decreased in some participants during follow-up (e.g., through lifestyle interventions), we may have overestimated the true association between remnant cholesterol and diabetes risk; (2) conversely, if remnant cholesterol levels increased in others, we may have underestimated the strength of the association; and

(3) we cannot determine whether sustained high RC levels versus transient elevations confer different diabetes risks. However, prior studies suggest that remnant cholesterol levels remain relatively stable in untreated individuals, and our sensitivity analyses revealed consistent associations across different follow-up periods (4, 8, and 12 years), suggesting that baseline remnant cholesterol provides meaningful predictive value despite this limitation. Third, the serum levels of remnant cholesterol were calculated by total cholesterol, HDL-C and LDL-C. Previous studies reported that the calculation of remnant cholesterol may overestimate the level compared with direct measurement [50]. However, the calculation method was more affordable and convenient. The measured and calculated levels were also highly correlated [25]. Fourth, the values of remnant cholesterol in this study appeared to be lower than those reported in previous studies [22]. Fifth, in this study, type 2 diabetes was defined as HbA1c ≥ 6.5%, fasting plasma glucose ≥ 7 mmol/L or self-reported. This study was a secondary analysis of NAGALA data and adopted the definition of type 2 diabetes in the primary analysis [27]. We admit that we are unable to diagnose type 2 diabetes on the basis of plasma glucose and HbA1c levels in this population in Japan. Sixth, the prevalence of NAFLD was 18%, which was relatively low and can be explained by several key characteristics of our study population. We excluded individuals with diabetes and impaired fasting glucose. These exclusions systematically removed higher-risk individuals who would typically have a higher NAFLD incidence. In addition, only 23% of the included participants were overweight/obese (BMI ≥ 25 kg/m²). This low prevalence of NAFLD likely represents the "healthier spectrum" of middle-aged Japanese adults after excluding those with metabolic abnormalities.

In conclusion, high remnant cholesterol was associated with an increased risk of developing diabetes in patients with NAFLD but not in patients without NAFLD. The prognostic value of remnant cholesterol for long-term outcomes in NAFLD patients warrants further investigation.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s13098-025-01828-z.

Supplementary Material 1

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Author contributions
Conceptualization and study design: Liping Yang, Hangkai Huang. Statistical analysis: Liping Yang, Hangkai Huang. Manuscript drafting: Hangkai Huang. Writing- Reviewing and Editing: Zejun Wang, Chengfu Xu. Supervision: Chengfu Xu.

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Data availability
No datasets were generated or analysed during the current study.

Declarations

Ethics approval and consent to participate
The study was conducted in accordance with the Declaration of Helsinki and was approved by the Murakami Memorial Hospital Institutional Ethics Review Board (IRB2018-09-01).

Competing interests
The authors declare no competing interests.

Strength and limitations of this study
The strengths of this study include its longitudinal population-based design. Serum remnant cholesterol was positively associated with the risk of incident diabetes in NAFLD patients.

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References

1. Tilg H, Adolph TE, Dudek MKnolle P. Non-alcoholic fatty liver disease: the interplay between metabolism, microbes and immunity. Nat Metab. 2021;3(12):1596–607.
2. Riazi K, Azhari H, Charette JH, Underwood FE, King JA, Afshar EE, et al. The prevalence and incidence of NAFLD worldwide: a systematic review and meta-analysis. Lancet Gastroenterol Hepatol. 2022;7(9):851–61.
3. Toh JZK, Pan X-H, Tay PWL, Ng CH, Yong JN, Xiao J, et al. A Meta-Analysis on the global prevalence, risk factors and screening of coronary heart disease in nonalcoholic fatty liver disease. Clin Gastroenterol Hepatol. 2022;20(11):2462–e247310.
4. Orci LA, Sanduzzi-Zamparelli M, Caballol B, Sapena V, Colucci N, Torres F, et al. Incidence of hepatocellular carcinoma in patients with nonalcoholic fatty liver disease: A systematic review, Meta-analysis, and Meta-regression. Clin Gastroenterol Hepatol. 2022;20(2):283–e29210.
5. Thomas JA, Kendall BJ, Dalais C, Macdonald GAThrift AP. Hepatocellular and extrahepatic cancers in non-alcoholic fatty liver disease: A systematic review and meta-analysis. Eur J Cancer. 2022;173:250–62.
6. Zhang X, Wong GL-H, Yip TC-F, Cheung JTK, Tse Y-K, Hui VW-K, et al. Risk of liver-related events by age and diabetes duration in patients with diabetes and nonalcoholic fatty liver disease. Hepatology. 2022;76(5):1409–22.
7. Stefan NCusi K. A global view of the interplay between non-alcoholic fatty liver disease and diabetes. Lancet Diabetes Endocrinol. 2022;10(4):284–96.
8. Bullard KM, Cowie CC, Lessem SE, Saydah SH, Menke A, Geiss LS, et al. Prevalence of diagnosed diabetes in adults by diabetes Type - United states, 2016. MMWR Morb Mortal Wkly Rep. 2018;67(12):359–61.
9. Ma RCW. Epidemiology of diabetes and diabetic complications in China. Diabetologia. 2018;61(6):1249–60.
10. Tang TS, Yusuf FLA, Polonsky WHFisher L. Assessing quality of life in diabetes: II - Deconstructing measures into a simple framework. Diabetes Res Clin Pract. 2017;126:286–302.
11. Morrison AE, Zaccardi F, Khunti KDavies MJ. Causality between non-alcoholic fatty liver disease and risk of cardiovascular disease and type 2 diabetes: A meta-analysis with bias analysis. Liver Int. 2019;39(3):557–67.
12. Björkström K, Stål P, Hultcrantz RHagström H. Histologic scores for fat and fibrosis associate with development of type 2 diabetes in patients with non-alcoholic fatty liver disease. Clin Gastroenterol Hepatol. 2017;15(9):1461–8.
13. The Lancet Diabetes E. Redefining obesity: advancing care for better lives. Lancet Diabetes Endocrinol. 2025;13(2):75.
14. Stefan NTargher G. Clusters of metabolic dysfunction-associated steatotic liver disease for precision medicine. Nat Rev Gastroenterol Hepatol. 2025;22(4):226–7.

15. Tall AR, Thomas DG, Gonzalez-Cabodevilla AGGoldberg IJ. Addressing dyslipidemic risk beyond LDL-cholesterol. J Clin Invest. 2022;132(1):e148559.
16. Varbo A, Benn M, Tybjærg-Hansen ANordestgaard BG. Elevated remnant cholesterol causes both low-grade inflammation and ischemic heart disease, whereas elevated low-density lipoprotein cholesterol causes ischemic heart disease without inflammation. Circulation. 2013;128(12):1298–309.
17. Vallejo-Vaz AJ, Fayyad R, Boekholdt SM, Hovingh GK, Kastelein JJ, Melamed S, et al. Triglyceride-Rich lipoprotein cholesterol and risk of cardiovascular events among patients receiving Statin therapy in the TNT trial. Circulation. 2018;138(8):770–81.
18. Quispe R, Martin SS, Michos ED, Lamba I, Blumenthal RS, Saeed A, et al. Remnant cholesterol predicts cardiovascular disease beyond LDL and apob: a primary prevention study. Eur Heart J. 2021;42(42):4324–32.
19. Varbo ANordestgaard BG. Remnant lipoproteins. Curr Opin Lipidol. 2017;28(4):300–7.
20. Chait A, Ginsberg HN, Vaisar T, Heinecke JW, Goldberg IJBornfeldt KE. Remnants of the Triglyceride-Rich lipoproteins, diabetes, and cardiovascular disease. Diabetes. 2020;69(4):508–16.
21. Huang H, Guo Y, Liu Z, Zeng Y, Chen YXu C. Remnant cholesterol predicts Long-term mortality of patients with metabolic Dysfunction-associated fatty liver disease. J Clin Endocrinol Metab. 2022;107(8):e3295–303.
22. Huh JH, Roh E, Lee SJ, Ihm SH, Han KDKang JG. Remnant cholesterol is an independent predictor of type 2 diabetes: A nationwide Population-Based cohort study. Diabetes Care. 2022;46(2):305–12.
23. Chin J, Mori TA, Adams LA, Beilin LJ, Huang R-C, Olynyk JK, et al. Association between remnant lipoprotein cholesterol levels and non-alcoholic fatty liver disease in adolescents. JHEP Rep. 2020;2(6):100150.
24. Hu X, Liu Q, Guo X, Wang W, Yu B, Liang B, et al. The role of remnant cholesterol beyond low-density lipoprotein cholesterol in diabetes mellitus. Cardiovasc Diabetol. 2022;21(1):117.
25. Cao YX, Zhang HW, Jin JL, Liu HH, Zhang Y, Gao Y, et al. The longitudinal association of remnant cholesterol with cardiovascular outcomes in patients with diabetes and pre-diabetes. Cardiovasc Diabetol. 2020;19(1):104.
26. Wu Z, Yu S, Zhu Q, Li Z, Zhang H, Kang X, et al. Association of baseline and cumulative remnant cholesterol with incidence of diabetic nephropathy: A longitudinal cohort study. Diabetes Res Clin Pract. 2022;191:110079.
27. Okamura T, Hashimoto Y, Hamaguchi M, Obora A, Kojima TFukui M. Ectopic fat obesity presents the greatest risk for incident type 2 diabetes: a population-based longitudinal study. Int J Obes (Lond). 2019;43(1):139–48.
28. Hamaguchi M, Kojima T, Itoh Y, Harano Y, Fujii K, Nakajima T, et al. The severity of ultrasonographic findings in nonalcoholic fatty liver disease reflects the metabolic syndrome and visceral fat accumulation. Am J Gastroenterol. 2007;102(12):2708–15.
29. Basevi V, Di Mario S, Morciano C, Nonino FMagrini N. Comment on: American Diabetes Association. Standards of medical care in diabetes–2011. Diabetes Care. 2011;34(Suppl. 1):S11-S61. Diabetes Care. 2011;34(5):e53; author reply e54.
30. Xie G, Zhong Y, Yang SZou Y. Remnant cholesterol is an independent predictor of New-Onset diabetes: A Single-Center cohort study. Diabetes Metab Syndr Obes. 2021;14:4735–45.
31. Huang H, Xie J, Zeng Y, Liu Z, Miao M, Xu L, et al. Remnant cholesterol independently predicts the development of nonalcoholic fatty liver disease. J Clin Endocrinol Metab. 2023;108(11):2907–15.
32. Chen J, Su Y, Su XLuo F. Remnant cholesterol has a non-linear association with non-alcoholic fatty liver disease. Diabetes Res Clin Pract. 2023;201:110733.
33. Fu L, Tai S, Sun J, Zhang N, Zhou Y, Xing Z, et al. Remnant cholesterol and its Visit-to-Visit variability predict cardiovascular outcomes in patients with type 2 diabetes: findings from the ACCORD cohort. Diabetes Care. 2022;45(9):2136–43.
34. Hazlehurst JM, Woods C, Marjot T, Cobbold JFTomlinson JW. Non-alcoholic fatty liver disease and diabetes. Metabolism. 2016;65(8):1096–108.
35. Lévesque V, Poirier P, Després J-PAlméras N. Relation between a simple lifestyle risk score and established biological risk factors for cardiovascular disease. Am J Cardiol. 2017;120(11):1939–46.
36. Mohamed F, Mansfield BSRaal FJ. ANGPTL3 as a drug target in hyperlipidemia and atherosclerosis. Curr Atheroscler Rep. 2022;24(12):959–67.
37. Sakurai T, Sakurai A, Vaisman BL, Amar MJ, Liu C, Gordon SM, et al. Creation of Apolipoprotein C-II (ApoC-II) mutant mice and correction of their hypertriglyceridemia with an ApoC-II mimetic peptide. J Pharmacol Exp Ther. 2016;356(2):341–53.
38. Ruscica M, Zimetti F, Adorni MP, Sirtori CR, Lupo MGFerri N. Pharmacological aspects of ANGPTL3 and ANGPTL4 inhibitors: new therapeutic approaches for the treatment of atherogenic dyslipidemia. Pharmacol Res. 2020;153:104653.
39. Sandesara PB, Virani SS, Fazio SShapiro MD. The forgotten lipids: triglycerides, remnant cholesterol, and atherosclerotic cardiovascular disease risk. Endocr Rev. 2019;40(2):537–57.
40. Rosenson RS, Davidson MH, Hirsh BJ, Kathiresan SGaudet D. Genetics and causality of triglyceride-rich lipoproteins in atherosclerotic cardiovascular disease. J Am Coll Cardiol. 2014;64(23):2525–40.
41. Shimizu K, Nishimuta S, Fukumura Y, Michinaga S, Egusa Y, Hase T, et al. Liver-specific overexpression of lipoprotein lipase improves glucose metabolism in high-fat diet-fed mice. PLoS ONE. 2022;17(9):e0274297.
42. Shalaurova I, Connelly MA, Garvey WTOtvos JD. Lipoprotein insulin resistance index: a lipoprotein particle-derived measure of insulin resistance. Metab Syndr Relat Disord. 2014;12(8):422–9.
43. Funada J-i, Sekiya M, Otani T, Watanabe K, Sato MAkutsu H. The close relationship between postprandial remnant metabolism and insulin resistance. Atherosclerosis. 2004;172(1):151–4.
44. Taskinen MR. Diabetic dyslipidaemia: from basic research to clinical practice. Diabetologia. 2003;46(6):733–49.
45. Brunham LR, Kruit JK, Verchere CBHayden MR. Cholesterol in islet dysfunction and type 2 diabetes. J Clin Invest. 2008;118(2):403–8.
46. Böni-Schnetzler MMeier DT. Islet inflammation in type 2 diabetes. Semin Immunopathol. 2019;41(4):501–13.
47. Shirakawa T, Nakajima K, Yatsuzuka S-i, Shimomura Y, Kobayashi J, Machida T, et al. The role of Circulating lipoprotein lipase and adiponectin on the particle size of remnant lipoproteins in patients with diabetes mellitus and metabolic syndrome. Clin Chim Acta. 2015;440:123–32.
48. Targher G, Corey KE, Byrne CDRoden M. The complex link between NAFLD and type 2 diabetes mellitus - mechanisms and treatments. Nat Rev Gastroenterol Hepatol. 2021;18(9):599–612.
49. Stefan N, Schick F, Birkenfeld AL, Häring H-UWhite MF. The role of hepatokines in NAFLD. Cell Metab. 2023;35(2):236–52.
50. Chen J, Kuang J, Tang X, Mao L, Guo X, Luo Q, et al. Comparison of calculated remnant lipoprotein cholesterol levels with levels directly measured by nuclear magnetic resonance. Lipids Health Dis. 2020;19(1):132.

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