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ORIGINAL ARTICLE

Observational Study

Cholecystectomy is independently associated with nonalcoholic fatty liver disease in an Asian population

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Data sharing: The technical appendix, statistical code and dataset are available from the corresponding author at messmd@chol.com. No additional data are available.

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Abstract

AIM: To investigate the relationship between gallstone disease and nonalcoholic fatty liver disease (NAFLD) in a large Asian population.

METHODS: A cross-sectional study including 17612 subjects recruited from general health check-ups at the Seoul National University Hospital, Healthcare System Gangnam Center between January 2010 and December 2010 was conducted. NAFLD and gallstone disease were diagnosed based on typical ultrasonographic findings. Subjects who were positive for hepatitis B or C, or who had a history of heavy alcohol consumption (> 30 g/d for men and > 20 g/d for women) or another type of hepatitis were excluded. Gallstone disease was defined as either the presence of gallstones or previous cholecystectomy, and these two entities (gallstones and cholecystectomy) were analyzed separately. Clinical parameters including body mass index, waist circumference, hypertension, diabetes, smoking status, and regular physical activity were reviewed. Laboratory parameters, including serum levels of gamma-glutamyl transpeptidase, alanine aminotransferase, aspartate aminotransferase, fasting glucose, fasting insulin, total cholesterol, triglycerides, and high-density lipoprotein, were also reviewed.

RESULTS: The mean age of the subjects was 48.5

± 11.3 years, and 49.3% were male. Approximately 30.3% and 6.1% of the subjects had NAFLD and gallstone disease, respectively. The prevalence of gallstone disease (8.3% *vs* 5.1%, $P < 0.001$), including both the presence of gallstones (5.5% *vs* 3.4%, $P < 0.001$) and a history of cholecystectomy (2.8% *vs* 1.7%, $P < 0.001$), was significantly increased in the NAFLD group. In the same manner, the prevalence of NAFLD increased with the presence of gallstone disease (41.3% *vs* 29.6%, $P < 0.001$). Multivariate regression analysis showed that cholecystectomy was associated with NAFLD (OR = 1.35, 95%CI: 1.03-1.77, $P = 0.028$). However, gallstones were not associated with NAFLD (OR = 1.15, 95%CI: 0.95-1.39, $P = 0.153$). The independent association between cholecystectomy and NAFLD was still significant after additional adjustment for insulin resistance (OR = 1.45, 95%CI: 1.01-2.08, $P = 0.045$).

CONCLUSION: This study shows that cholecystectomy, but not gallstones, is independently associated with NAFLD after adjustment for metabolic risk factors. These data suggest that cholecystectomy may be an independent risk factor for NAFLD.

Key words: Fatty liver; Hepatic steatosis; Gallbladder; Cholelithiasis; Gallbladder removal

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Core tip: The relationship between gallstone disease (gallstones and cholecystectomy, separately) and ultrasonographically diagnosed nonalcoholic fatty liver disease (NAFLD) was analyzed in a large Asian population. The prevalence of gallstone disease increased with the presence of NAFLD, and the prevalence of NAFLD increased with the presence of gallstone disease. Multivariate regression analysis showed that cholecystectomy was associated with NAFLD. However, gallstones were not associated with NAFLD. The independent association between cholecystectomy and NAFLD was still significant after additional adjustment for insulin resistance. This study showed that cholecystectomy, but not gallstones, is independently associated with NAFLD after adjustment for metabolic risk factors.

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INTRODUCTION

Nonalcoholic fatty liver disease (NAFLD) is one of the most common liver diseases, with a prevalence of 20%-35% in the general population[1,2]. NAFLD includes a spectrum of liver diseases, from simple steatosis to nonalcoholic steatohepatitis, cirrhosis, and hepatocellular carcinoma[3,4]. As obesity, type 2 diabetes, dyslipidemia, and insulin resistance are the underlying metabolic conditions that favor the occurrence of NAFLD, NAFLD is regarded as the hepatic manifestation of metabolic syndrome[5]. Gallstone disease is also common, and the prevalence of gallstones varies between 5% and 25%[6,7]. Increased age, female sex, obesity, metabolic syndrome, hypertriglyceridemia, diabetes, and insulin resistance are considered to be the major risk factors for gallstones[8,9].

As mentioned above, gallstone disease and NAFLD are both prevalent in the general population and share the same risk factors, including obesity and insulin resistance. Therefore, several studies have investigated the association between gallstone disease and NAFLD and have demonstrated an independent association between them[8,10]. One study demonstrated a dose-dependent association between the severity of hepatic inflammation or fibrosis and the prevalence of gallstone disease[11]. On the contrary, another study showed no association between gallstone disease and the severity of fibrosis in NAFLD patients[12]. Recently, a population-based study using the National Health and Nutrition Examination Survey Ⅲ, evaluated gallstone disease by separating patients according to the presence of either gallstones or a history of cholecystectomy. This study revealed an association between cholecystectomy and NAFLD, but no association between gallstones and NAFLD, suggesting that cholecystectomy has metabolic consequences[13]. As discussed above, previous studies showed inconsistent results regarding the association between gallstone disease and NAFLD.

Therefore, the purpose of this study was to investigate the relationship between gallstone disease (including gallstones and cholecystectomy) and NAFLD in a large Asian population.

MATERIALS AND METHODS

Study population

Subjects who voluntarily visited the Seoul National University Hospital, Healthcare System Gangnam Center, for a health check-up between January 2010 and December 2010 were initially enrolled. Most of the screenees routinely underwent hepatic ultrasonography and blood sampling as part of their health care program. Of the 24550 initially enrolled subjects, patients with other causes of chronic liver disease were excluded as follows: 267 for hepatitis C (diagnosed by a positive hepatitis C antibody); 1186 for hepatitis B (diagnosed by a positive hepatitis B surface antigen); 3926 for excessive alcohol consumption (defined as > 30 g/d for men and > 20 g/d for women); and 105 for a history of other liver diseases (*e.g.*, Wilson's disease, autoimmune hepatitis, primary biliary cirrhosis, and hemochromatosis). We also excluded 697 subjects who had taken drugs that can cause fatty liver within the past year. Two subjects who were found to have gallbladder cancer on abdominal ultrasonography were also excluded. Additionally, 755 subjects who did not answer the questionnaire regarding alcohol drinking, smoking status, exercise, and past medical history were excluded. Therefore, 17612 subjects were finally included in this analysis. The study was approved by the Institutional Review Board of Seoul National University Hospital (H1309-019-518) and was performed according to the ethical guidelines of the 1975 Declaration of Helsinki and its later amendments. The need to obtain informed consent from the subjects was waived by the Institutional Review Board of Seoul National University Hospital. A corresponding author and all the co-authors had access to the full data of this study and reviewed and approved the manuscript.

Definition of NAFLD by ultrasonographic examination

Hepatic ultrasonography was performed by experienced radiologists. At the time of the procedure, the radiologists were blinded to the laboratory and clinical data of the subjects. Fatty liver was diagnosed by ultrasonographic findings (Acuson, Sequoia 512, Siemens, Mountain View, CA, United States), based on liver brightness, hepatorenal echo contrast, vascular blurring, and deep attenuation[14].

NAFLD was defined as the presence of fatty liver by ultrasonography without the presence of the following other possible causes of chronic liver disease: (1) excessive alcohol consumption (defined as > 30 g/d for men and > 20 g/d for women); (2) positivity for antibodies against the hepatitis C virus or the hepatitis B surface antigen; (3) other known causes of chronic liver disease; and (4) the use of drugs that can cause fatty liver.

Definition of gallstone disease

Gallstone disease was diagnosed by experienced radiologists using ultrasonography (Acuson, Sequoia 512, Siemens, Mountain View, CA, United States) after the subjects had fasted for at least 8 h. Gallstone disease was defined as the ultrasonographic presence of gallstones or absence of the gallbladder on ultrasonography due to a previous history of cholecystectomy. Gallstones were diagnosed based on the presence of movable hyper-echoic foci with acoustic shadows.

Clinical and laboratory assessments

Each subject answered a questionnaire regarding past medical history, including previous history of cholecystectomy. Anthropometric measurements and laboratory tests were performed on the same day. Waist circumference was measured by a trained nurse using a tape placed at the midpoint between the iliac crest and the lower costal margin. Height and weight were measured using a digital scale, and body mass index (BMI) was calculated using the following formula: BMI = weight (kg)/height squared (m^2). Systolic and diastolic blood pressures were checked twice, and the mean values of the two measurements were used. Hypertension was defined as the current use of anti-hypertensive drugs, a systolic blood pressure over 140 mmHg, or a diastolic blood pressure over 90 mmHg. The presence of diabetes was defined as the current use of anti-diabetic drugs or a fasting glucose level greater than or equal to 126 mg/dL. Current smokers were defined as subjects who had smoked at least 100 cigarettes in their lifetime and who smoked either every day or on some days during the previous year. Ex-smokers were defined as subjects who reported smoking at least 100 cigarettes in their lifetime and who had not smoked during the previous year. Regular physical activity was defined as regularly exercising more than once per week.

Laboratory examinations included serum gamma-glutamyl transpeptidase (GGT), alanine aminotransferase (ALT), aspartate aminotransferase (AST), total cholesterol, triglycerides, high-density lipoprotein (HDL), fasting glucose, HbA1c, fasting insulin, antibodies against the hepatitis C virus, and hepatitis B surface antigen. Blood sampling was performed before 10 am after an overnight fast. All the biochemical examinations were performed in the same laboratory according to standard laboratory methods. The homeostasis model assessment-estimated insulin resistance (HOMA-IR) was used to assess insulin resistance as follows: HOMA-IR = fasting plasma glucose (mmol/L) × fasting plasma insulin (μIU/mL)/22.5[15].

Statistical analysis

To compare the variables between subjects according to gallstone disease status (control, gallstones, cholecystectomy) and between subjects with and without NAFLD, the Student's *t*-test was used for continuous variables and the χ^2-test was used for categorical variables. Multivariate logistic regression analysis was performed including previously established risk factors and variables with a *P* value < 0.05 in the unadjusted analyses. SPSS 19 (SPSS Inc., Chicago, IL, United States) software was used. A two-tailed *P* value < 0.05 was considered statistically significant.

The statistical methods of this study were reviewed by Seung-sik Hwang from Inje University School of Medicine.

RESULTS

A total of 17612 individuals (8682 males and 8930 females, mean age, 48.5 years) were ultimately

Table 1 Baseline characteristics of control and nonalcoholic fatty liver disease subjects

	Control (*P* = 12275)	NAFLD (*P* = 5337)	*P* value
Age (yr)	47.4 ± 11.4	50.9 ± 10.5	< 0.001
Male	4839 (39.4)	3843 (72.0)	< 0.001
Waist circumference (cm)	80.3 ± 7.2	89.2 ± 6.9	< 0.001
Body mass index (kg/m^2)	22.0 ± 2.6	25.4 ± 2.7	< 0.001
Systolic blood pressure (mmHg)	111.3 ± 14.3	119.3 ± 13.2	< 0.001
Diastolic blood pressure (mmHg)	71.4 ± 11.0	78.1 ± 10.5	< 0.001
Hypertension	1570 (12.8)	1451 (27.2)	< 0.001
Diabetes	373 (3.0)	498 (9.3)	< 0.001
Gamma-glutamyl transpeptidase (IU/L)	24.7 ± 31.3	42.7 ± 39.8	< 0.001
Alanine aminotransferase (IU/L)	19.1 ± 16.1	33.0 ± 23.5	< 0.001
Aspartate aminotransferase (IU/L)	20.7 ± 11.5	25.6 ± 12.8	< 0.001
Total cholesterol (mg/dL)	191.6 ± 32.9	200.5 ± 34.9	< 0.001
Triglycerides (mg/dL)	85.9 ± 50.0	144.8 ± 84.1	< 0.001
HDL-cholesterol (mg/dL)	57.6 ± 12.3	48.9 ± 9.6	< 0.001
Fasting glucose (mg/dL)	92.4 ± 12.7	102.5 ± 19.8	< 0.001
HbA1c (%)	5.7 ± 0.4	6.0 ± 0.7	< 0.001
Fasting insulin, μIU/mL (*n* = 8622)	5.8 ± 3.4	9.6 ± 5.5	< 0.001
HOMA-IR index (*n* = 8622)	1.4 ± 1.0	2.5 ± 1.6	< 0.001
Gallstones	421 (3.4)	292 (5.5)	< 0.001
Cholecystectomy	207 (1.7)	149 (2.8)	< 0.001
Gallstone disease	628 (5.1)	441 (8.3)	< 0.001
Smoking			
Never-smoker	8300 (67.6)	2365 (44.3)	< 0.001
Current smoker	1419 (11.6)	1114 (20.9)	
Ex-smoker	2556 (20.8)	1858 (34.8)	
Regular physical activity	8112 (66.1)	3496 (65.5)	0.455

Data are presented as the mean ± SD or *n* (%). NAFLD: Nonalcoholic fatty liver disease; IR: Insulin resistance.

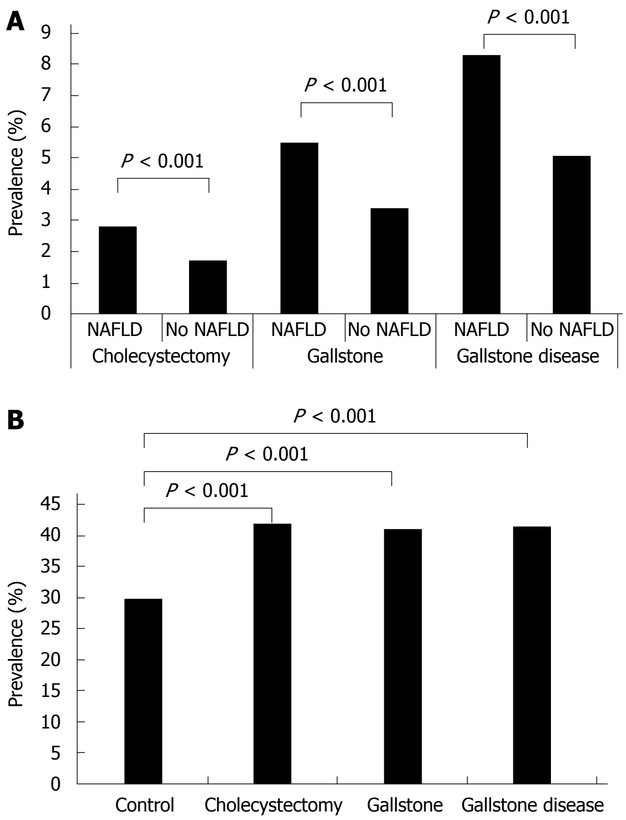

Figure 1 Prevalence of disease in subjects with or without nonalcoholic fatty liver disease. A: Prevalence of cholecystectomy, gallstones, and gallstone disease in subjects with and without nonalcoholic fatty liver disease (NAFLD). Cholecystectomy, gallstones, and gallstone disease were more commonly observed in subjects with NAFLD compared with subjects without NAFLD (*P* < 0.001 for all); B: The prevalence of NAFLD in the control, cholecystectomy, gallstone, and gallstone disease groups. NAFLD was significantly more likely in the cholecystectomy, gallstone and gallstone disease groups compared with the control group (all *P* < 0.001). NAFLD: Nonalcoholic fatty liver disease.

analyzed in this study. Of these, 5337 (30.3%) had NAFLD. Table 1 shows the baseline characteristics of the controls and subjects with NAFLD. The following factors were significantly associated with NAFLD: increased age; male sex; larger waist circumference; higher BMI; higher blood pressure; the presence of hypertension and diabetes; elevated levels of GGT, ALT, AST, cholesterol, triglycerides, fasting glucose, HbA1c, and HOMA-IR; and lower levels of HDL cholesterol (all *P* < 0.001). The presence of gallstones (5.5% *vs* 3.4%, *P* < 0.001) and a history of cholecystectomy (2.8% *vs* 1.7%, *P* < 0.001) were both significantly increased in the NAFLD group (Figure 1A).

Table 2 shows the baseline characteristics of the subjects according to their gallstone disease status (control, gallstones, cholecystectomy, and gallstone disease). Approximately 6.1% of subjects (*n* = 1069) had gallstone disease. Compared with the control group, subjects in the gallstone disease group were older. They also had larger waist circumference, a higher BMI, and elevated levels of GGT, ALT, AST, triglycerides, fasting glucose, HbA1c, and HOMA-IR, as well as lower levels of HDL cholesterol (all *P* < 0.001). There were more subjects with diabetes and hypertension in the gallstone disease group (*P* < 0.001). Figure 1B shows that the rate of NAFLD was increased in the gallstone (41.0% *vs* 29.6%, *P* < 0.001), cholecystectomy (41.9% *vs* 29.6%, *P* < 0.001), and gallstone disease (41.3% *vs* 29.6%, *P* < 0.001) groups.

When we analyzed the association between gallstone disease and NAFLD, gallstone disease (OR = 1.67, 95%CI: 1.47-1.90, *P* < 0.001), cholecystectomy (OR = 1.67, 95%CI: 1.35-2.07, *P* < 0.001), and gallstones (OR = 1.65, 95%CI: 1.42-1.92, *P* < 0.001) were significantly associated with NAFLD in the unadjusted analyses. Gallstone disease was independently associated with NAFLD after adjustment for age and sex (OR = 1.47, 95%CI: 1.28-1.68, *P* < 0.001). Cholecystectomy (OR = 1.46, 95%CI: 1.16-1.83, *P* = 0.001) and gallstones (OR = 1.46, 95%CI: 1.24-1.72, *P* < 0.001) were also independently associated with NAFLD in the age- and sex-adjusted model. In the multivariate model, after adjusting for other covariates such as BMI, smoking, physical activity, hypertension, diabetes, total cholesterol,

Table 2 Baseline characteristics of the subjects according to gallstone disease status

	Control (n = 16543)	Gallstone disease (n = 1069)	Gallstone disease (n = 1069)	
			Gallstones (n = 713)	Cholecystectomy (n = 356)
Age (yr)	48.1 ± 11.1	54.5 ± 11.4[a]	53.8 ± 11.2[b]	55.8 ± 11.5[c]
Male	8122 (49.1)	560 (52.4)[a]	377 (52.9)[b]	183 (51.4)
Waist circumference (cm)	82.8 ± 8.2	86.4 ± 8.5[a]	86.3 ± 8.2[b]	86.3 ± 7.7[c]
Body mass index (kg/m^2)	23.0 ± 3.1	24.1 ± 3.2[a]	24.0 ± 3.1[b]	23.9 ± 2.9[c]
Systolic blood pressure (mmHg)	113.5 ± 14.4	116.3 ± 15.2[a]	117.0 ± 15.0[b]	118.4 ± 14.5[c]
Diastolic blood pressure (mmHg)	73.3 ± 11.3	75.2 ± 11.4[a]	75.2 ± 11.2[b]	75.2 ± 10.6[c]
Hypertension	2710 (16.4)	311 (29.1)[a]	205 (28.8)[b]	106 (29.8)[c]
Diabetes	771 (4.7)	100 (9.4)[a]	67 (9.4)[b]	33 (9.3)[c]
Gamma-glutamyl transpeptidase (IU/L)	29.8 ± 32.9	34.6 ± 48.8[a]	35.7 ± 58.8[b]	38.0 ± 75.0[c]
Alanine aminotransferase (IU/L)	23.1 ± 19.8	25.9 ± 18.6[a]	26.0 ± 17.9[b]	26.1 ± 16.4[c]
Aspartate aminotransferase (IU/L)	22.1 ± 12.2	23.2 ± 11.1[a]	23.6 ± 10.7[b]	24.5 ± 9.9[c]
Total cholesterol (mg/dL)	194.4 ± 33.7	193.4 ± 33.7	192.5 ± 33.6	190.6 ± 33.4[c]
Triglycerides (mg/dL)	103.2 ± 68.0	111.1 ± 62.3[a]	112.9 ± 65.9[b]	116.4 ± 72.5[c]
HDL-cholesterol (mg/dL)	55.1 ± 12.2	53.2 ± 12.1[a]	53.3 ± 12.2[b]	53.7 ± 12.6[c]
Fasting glucose (mg/dL)	95.2 ± 15.5	99.6 ± 20.4[a]	99.6 ± 20.8[b]	99.5 ± 17.0[c]
HbA1c (%)	5.8 ± 0.5	5.9 ± 0.7[a]	5.9 ± 0.6[b]	6.0 ± 0.5[c]
Fasting insulin, μIU/mL (n = 8622)	6.9 ± 4.4	8.3 ± 6.2[a]	8.3 ± 6.0[b]	8.3 ± 5.7[c]
HOMA- IR index (n = 8622)	1.7 ± 1.3	2.1 ± 1.7[a]	2.1 ± 1.7[b]	2.2 ± 1.7[c]
NAFLD	4896 (29.6)	441 (41.3)[a]	292 (41.0)[b]	149 (41.9)[c]
Smoking				
Never-smoker	10042 (60.7)	623 (58.3)[a]	419 (58.8)	204 (57.3)[c]
Current smoker	2398 (14.5)	135 (12.6)[a]	95 (13.3)	40 (11.2)[c]
Ex-smoker	4103 (24.8)	311 (29.1)[a]	199 (27.9)	112 (31.5)[c]
Regular physical activity	10835 (65.5)	773 (72.3)[a]	521 (73.1)[b]	252 (70.8)[c]

Data are presented as the mean ± SD or n (%). [a]P < 0.05, control *vs* gallstone disease; [b]P < 0.05, control *vs* gallstone; [c]P < 0.05, control *vs* cholecystectomy. NAFLD: Nonalcoholic fatty liver disease; GSD: Gallstone disease; IR: Insulin resistance.

triglycerides, and HDL cholesterol, in addition to age and sex, subjects with gallstone disease had an increased risk of NAFLD (OR = 1.22, 95%CI: 1.04-1.43, P = 0.016). When cholecystectomy and gallstones were analyzed separately, cholecystectomy was independently associated with NAFLD in the multivariate model (OR = 1.35, 95%CI: 1.03-1.77, P = 0.028). In other words, subjects who underwent cholecystectomy had a 35% higher risk of NAFLD compared with subjects who had not undergone cholecystectomy. However, the presence of gallstones was not independently associated with NAFLD after adjusting for known metabolic risk factors (OR = 1.15, 95%CI: 0.95-1.39, P = 0.153) (Table 3). Similarly, when waist circumference, which is a surrogate marker for visceral obesity, was additionally accounted for in the multivariate analysis, cholecystectomy, but not gallstones, was independently associated with NAFLD (data not shown).

A subgroup analysis was conducted in 8622 subjects in whom fasting insulin was examined. The baseline characteristics of subjects with or without fasting insulin measurements are shown in Table 4. In the group whose fasting insulin levels were checked, there were more older subjects and male subjects with hypertension or diabetes. Gallstone disease was associated with NAFLD (at a marginal level of significance) after adjusting for insulin resistance in addition to metabolic risk factors (OR = 1.25, 95%CI: 1.00-1.55, P = 0.052) (Table 5). Cholecystectomy was independently associated with NAFLD in the multivariate model (OR = 1.45, 95%CI: 1.01-2.08, P = 0.045), but gallstones were not (OR = 1.14, 95%CI: 0.87-1.50, P = 0.336).

DISCUSSION

This study demonstrated that gallstone disease is associated with NAFLD, independent of well-established, common metabolic risk factors. This association was mainly attributable to a history of cholecystectomy, not the presence of gallstones. Subjects who underwent cholecystectomy had a 35% higher prevalence of NAFLD. However, gallstones were not independently associated with NAFLD.

The association between gallstone disease and NAFLD has been evaluated in several studies; however, most previous studies did not differentiate between gallstones and cholecystectomy[8,11,16]. Therefore, the effect of cholecystectomy itself has not been fully investigated. An *in vivo* study suggested that cholecystectomy has metabolic consequences by demonstrating that cholecystectomized mice had increased levels of hepatic and serum triglycerides and very low-density lipoprotein[17]. Recently, Ruhl *et al*[13] reported an independent association between NAFLD and cholecystectomy, but not between NAFLD and gallstones, indicating cholecystectomy *per se* is a risk factor for NAFLD in the United States. However, it is hard to apply this result directly to Asian populations

Table 3 Univariate and multivariate analyses for the presence of nonalcoholic fatty liver disease according to gallstone disease status

Variable	Univariate model		Age, sex-adjusted model		Multivariate model[1]	
	OR (95%CI)	*P* value	OR (95%CI)	*P* value	OR (95%CI)	*P* value
Gallstone disease						
Control (*n* = 16543)	1		1		1	
Gallstone disease (*n* = 1069)	1.67 (1.47-1.90)	< 0.001	1.47 (1.28-1.68)	< 0.001	1.22 (1.04-1.43)	0.016
Cholecystectomy						
Control	1		1		1	
Cholecystectomy (*n* = 356)	1.67 (1.35-2.07)	< 0.001	1.46 (1.16-1.83)	0.001	1.35 (1.03-1.77)	0.028
Gallstones						
Control	1		1		1	
Gallstones (*n* = 713)	1.65 (1.42-1.92)	< 0.001	1.46 (1.24-1.72)	< 0.001	1.15 (0.95-1.39)	0.153

[1]The multivariate model was adjusted for age, sex, hypertension, diabetes, body mass index, smoking, physical activity, total cholesterol, triglycerides and HDL cholesterol. NAFLD: Nonalcoholic fatty liver disease; OR: Odds ratio.

Table 4 Comparison of characteristics between subjects who underwent fasting insulin testing and those who did not

	Fasting insulin absent (*n* = 8990)	Fasting insulin available (*n* = 8622)	*P* value
Age (yr)	45.8 ± 11.8	51.2 ± 9.9	< 0.001
Male	4808 (46.5)	4748 (55.1)	< 0.001
Waist circumference (cm)	82.8 ± 8.3	83.3 ± 8.1	< 0.001
Body mass index (kg/m^2)	23.0 ± 3.1	23.1 ± 3.0	0.167
Systolic blood pressure (mmHg)	113.2 ± 14.2	114.3 ± 14.6	< 0.001
Diastolic blood pressure (mmHg)	73.3 ± 11.3	73.9 ± 11.3	0.020
Hypertension	1300 (14.5)	1721 (20.0)	< 0.001
Diabetes	356 (4.0)	515 (6.0)	< 0.001
Gamma-glutamyl transpeptidase (IU/L)	30.1 ± 30.7	30.3 ± 39.0	0.732
Alanine aminotransferase (IU/L)	23.1 ± 19.4	23.5 ± 20.0	0.142
Aspartate aminotransferase (IU/L)	21.8 ± 12.4	22.6 ± 11.8	< 0.001
Total cholesterol (mg/dL)	193.0 ± 33.3	195.6 ± 34.1	< 0.001
Triglycerides (mg/dL)	105.0 ± 70.2	102.5 ± 65.5	0.017
HDL-cholesterol (mg/dL)	55.0 ± 12.0	55.0 ± 12.5	0.693
Fasting glucose (mg/dL)	95.4 ± 14.9	95.6 ± 16.9	0.606
HbA1c (%)	5.77 ± 0.51	5.84 ± 0.56	< 0.001
NAFLD	2619 (29.1)	2718 (31.5)	0.001
Gallstone	336 (3.7)	377 (4.4)	0.033
Cholecystectomy	160 (1.8)	196 (2.3)	0.020
Gallstone disease	496 (5.5)	573 (6.6)	0.002
Smoking			< 0.001
Never-smoker	5300 (59.0)	5365 (62.2)	
Current smoker	1440 (16.0)	1093 (12.7)	
Ex-smoker	2250 (25.0)	2164 (25.1)	
Regular physical activity	5559 (61.8)	6049 (70.2)	< 0.001

Data are presented as the mean ± SD or *n* (%). NAFLD: Nonalcoholic fatty liver disease; IR: Insulin resistance.

because the prevalence of and risk factors for gallstone disease vary across ethnicities[9,18,19]. In general, BMI, one of the risk factors for gallstones, is lower in Asians compared with Western populations; however, Asians have a higher risk of visceral obesity than Caucasian populations with the same BMI[20,21]. Thus, differences in general obesity, as assessed by BMI and visceral obesity, may have some effect on the association between gallstone disease and NAFLD. To date, studies evaluating the association between gallstone disease and NAFLD in Asian populations are scarce. This is the largest study confirming the independent association between cholecystectomy, but not gallstones, and NAFLD in an Asian population. This study supports the idea that cholecystectomy may have some effect on the development of NAFLD.

There are several possible mechanisms for this relationship: (1) Because the gallbladder regulates bile acid homeostasis, alterations in bile acid metabolism after cholecystectomy may alter glucose and lipid metabolism, causing NAFLD[22,23]. Bile acids exercise their action by binding to nuclear receptors, such as the farnesoid X receptor and TGR5, leading to gene expression changes in the liver[24,25]. The farnesoid X receptor plays an important role not only in maintaining cholesterol and bile acid homeostasis, but also in the regulation of many metabolic enzymes and transporters[26]. TGR5 also plays crucial roles in lipid metabolism, glucose homeostasis, and energy expenditure[27]. Thus, it can be inferred that

Table 5 Univariate and multivariate analyses for the presence of nonalcoholic fatty liver disease according to gallstone disease status in subjects in whom fasting insulin testing was performed

Variable	Univariate model		Age, sex-adjusted model		Multivariate model 1[1]		Multivariate model 2[2]	
	OR (95%CI)	*P* value	OR (95%CI)	*P* value	OR (95%CI)	*P* value	OR (95%CI)	*P* value
Gallstone disease								
Control (n = 8049)	1		1		1		1	
Gallstone disease (n = 573)	1.67 (1.41-1.99)	< 0.001	1.51 (1.26-1.82)	< 0.001	1.27 (1.03-1.58)	0.029	1.25 (1.00-1.55)	0.052
Cholecystectomy								
Control	1		1		1		1	
Cholecystectomy (n = 196)	1.76 (1.32-2.35)	< 0.001	1.59 (1.17-2.16)	0.003	1.45 (1.01-2.07)	0.044	1.45 (1.01-2.08)	0.045
Gallstones								
Control	1		1		1		1	
Gallstones (n = 377)	1.63 (1.32-2.01)	< 0.001	1.48 (1.18-1.85)	0.001	1.18 (0.91-1.54)	0.21	1.14 (0.87-1.50)	0.336

[1]The multivariate model 1 was adjusted for age, sex, hypertension, diabetes, body mass index, smoking, physical activity, total cholesterol, triglycerides and high-density lipoprotein cholesterol; [2]The multivariate model 2 was adjusted for multivariate model 1 in addition to insulin resistance as assessed by HOMA-IR. NAFLD: Nonalcoholic fatty liver disease; IR: Insulin resistance; OR: Odds ratio; HOMA-IR: Homeostasis model assessment-estimated insulin resistance.

cholecystectomy may alter the circulation of bile acid, the activation of bile acid receptors, and the downstream signaling pathways related to hepatic lipid and glucose metabolism, thereby contributing to the development of NAFLD; (2) Gallbladder-related hormonal effects represent another plausible explanation, whereby fibroblast growth factor 19 (FGF 19), which is secreted from the gallbladder mucosa and regulates the synthesis of bile salts, has a beneficial effect on the metabolic syndrome[28]. *In vitro* and *in vivo* studies have demonstrated the inhibitory effect of FGF-19 on hepatic fatty acid synthesis[29,30]. Lower serum FGF-19 levels were reported in NAFLD patients[31], and cholecystectomy reduces FGF-19 levels[32]. Therefore, it can be inferred that decreased FGF-19 levels after cholecystectomy may increase the hepatic triglyceride content, thereby exerting some effect on the development of NAFLD[13,33]. Although insulin resistance is a well-known risk factor for both gallstones and NAFLD, the association between NAFLD and cholecystectomy persisted with only a minimal change after additional adjustment for insulin resistance. This is similar to the results of previous studies[13,34]; and (3) There may be an association between pain or inflammatory symptoms, which are associated with gallbladder pathology, before cholecystectomy and the occurrence of NAFLD[35]. A recent population-based study[13] demonstrated that cholecystectomy in subjects with pain had a lower OR for NAFLD than cholecystectomy in subjects without pain. As we did not have any data concerning abdominal pain or biliary colic due to our study design, we could not evaluate the effect of pain on the association between cholecystectomy and NAFLD. Further prospective studies should be conducted to investigate the exact mechanism of the association between cholecystectomy and NAFLD.

In contrast to our study, several previous studies have demonstrated an independent association between gallstones and NAFLD[8,10,36]. This contradiction may be due to different definitions of NAFLD or insufficient adjustment for NAFLD risk factors. In other studies, NAFLD was defined by AST/ALT levels, which often underestimate and misclassify NAFLD. In our study, we defined NAFLD by ultrasonography and sufficiently adjusted for metabolic risk factors, including insulin resistance. Another plausible explanation is that ethnic differences may affect the association between gallstones and NAFLD. Ethnic differences are observed in the prevalence of gallstone disease; specifically, the prevalence of gallstones is reported to be as high as 60% to 70% in American Indians, 25% to 30% in Hispanic populations in Central and South America, and 10% to 15% in Caucasian adults in developed countries. A low prevalence is reported in African Americans and East Asians[9,37]. In Asia, the prevalence of gallstone disease was reported to be 3.2% in Japan, 3.1% in India, and 10.7% in Taiwan[10,38,39]. Similar to other Asian countries, the prevalence of gallstone disease in this Korean population was 6.1%.

This study has strengths compared with previous studies. First, the subjects in this study were representative of the general population, considering the nature of the health screenings. Thus, due to the sufficiently large sample size, we could provide more definitive evidence for an independent association between cholecystectomy itself and NAFLD, consistent with a previous report[13]. Second, this study confirmed the independent association between cholecystectomy and NAFLD in an Asian population, which may have different characteristics to a Western population.

This study also has several limitations. First, it was a cross-sectional study; thus, the temporal relationship between cholecystectomy and NAFLD could not be evaluated. Second, we diagnosed NAFLD by ultrasonography without histological confirmation, which is considered the gold standard for diagnosing NAFLD. However, histological diagnosis of NAFLD is difficult to accomplish in a large general population and creates the risk of certain complications. Third, insulin resistance was not evaluated in all patients due to the retrospective design of our study. Fourth, the type of cholecystectomy (open or laparoscopic) and the conversion rate of cholecystectomy (laparoscopic to open), which may have affected its association with NAFLD, were not reviewed in this study, because the previous history of cholecystectomy, not the type of cholecystectomy were only available due to our study design[40].

In conclusion, this study showed an independent association between previous cholecystectomy and NAFLD, independent of other established metabolic risk factors, in a large Asian population. However, gallstones were not independently associated with NAFLD. This result suggests that cholecystectomy may increase the risk of NAFLD. Further prospective studies are warranted to confirm these observations.

COMMENTS

Background

Nonalcoholic fatty liver disease (NAFLD) and gallstone disease are both prevalent diseases which share the same risk factors, including insulin resistance and obesity. However, the association between gallstone disease and NAFLD has not been definitively established.

Research frontiers

This study investigated the relationship between gallstone disease (presence of gallstones or previous cholecystectomy) and NAFLD in a large Asian population.

Innovations and breakthroughs

This study showed that cholecystectomy, but not gallstones, is independently associated with NAFLD after adjustment for other established metabolic risk factors in a large Asian population.

Applications

Clinicians may be more alert to the risk of NAFLD in patients with a history of cholecystectomy.

Terminology

Cholecystectomy is the surgical removal of the gallbladder for symptomatic gallstones or other gallbladder conditions.

Peer-review

This study is original and interesting and includes a large population. This study showed an independent association between cholecystectomy and NAFLD in the Asian population. Further prospective studies are warranted to confirm these associations.

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