Interrelationship between Fatty Liver and Insulin Resistance in the Development of Type 2 Diabetes

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Context: Although fatty liver and insulin resistance are known to be associated, the relationship between the two in the development of type 2 diabetes mellitus (T2DM) is unclear.

Objective: We investigated the 5-yr risk of developing T2DM in individuals diagnosed with fatty liver using ultrasound and stratified by insulin sensitivity using quartiles of fasting insulin concentration.

Design and Methods: We examined the clinical and laboratory data of 11,091 Koreans who had a medical evaluation including fasting insulin concentration and abdominal ultrasound at baseline and had a follow-up after 5 yr.

Results: At baseline, 27% of the population had fatty liver. Almost half (47%) of the individuals with fatty liver had baseline insulin concentration in the highest quartile compared with 17% in those without fatty liver ($P < 0.001$). Regardless of baseline insulin concentration, individuals with fatty liver had significantly ($P < 0.001$) more baseline clinical and metabolic abnormalities, including higher glucose and triglyceride concentration and lower high-density lipoprotein cholesterol concentration. In addition, regardless of baseline insulin concentration, individuals with fatty liver had a significantly increased risk for incident T2DM compared with those without fatty liver [crude odds ratio, 5.05 (95% confidence interval, 2.08–12.29) in the lowest insulin quartile and 6.34 (3.58–11.21) in the highest quartile]. In individuals in the highest insulin quartile, the odds ratio for developing T2DM remained significant even after multivariate adjustment including baseline glucose concentration [2.42 (1.23–4.75)].

Conclusion: Although associated with insulin resistance, fatty liver diagnosed by ultrasound appears to independently increase the risk of T2DM. **(*J Clin Endocrinol Metab* 96: 1093–1097, 2011)**

Three recent studies have demonstrated that the baseline presence of fatty liver by ultrasound predicts the development of type 2 diabetes mellitus (T2DM) at 4–5 yr (1–3). However, at baseline, individuals with fatty liver were generally heavier, more hypertensive, and more likely to have metabolic abnormalities including higher glucose and triglyceride and lower high-density lipoprotein cholesterol (HDL-C) concentration. These changes are characteristic of insulin resistance (4), a well-recognized risk factor for the development of T2DM (5). Furthermore, insulin resistance has also been associated with fatty liver (6, 7). Therefore, identification of fatty liver may represent a surrogate marker of insulin resistance, and not an independent predictor of T2DM. As a corollary, it is possible that insulin resistance, not fatty liver *per se*, increased the risk for T2DM. Unfortunately, none of the previous studies (1–3) showing that fatty liver predicted the development of T2DM contained any estimates of insulin resistance (*e.g.* fasting insulin concentration). Thus, their results do not permit an understanding of the inter-

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Abbreviations: BMI, Body mass index; HDL-C, high-density lipoprotein cholesterol; OR, odds ratio; T2DM, type 2 diabetes mellitus.

relationship between fatty liver and insulin resistance in the development of T2DM.

The purpose of this study was to evaluate the relationship between baseline diagnosis of fatty liver and fasting insulin concentration in predicting the development of T2DM in 11,091 relatively healthy Koreans. We hypothesized that the increased T2DM risk associated with fatty liver will be mediated by insulin resistance.

Subjects and Methods

Subjects

The study population consisted of individuals who had a comprehensive health examination at baseline (2003) and were reexamined 5 yr later (2008) at Kangbuk Samsung Hospital, College of Medicine, Sungkyunkwan University. Initially 15,613 individuals were identified. Among these participants, 416 were excluded for having T2DM at baseline. Individuals were also excluded for missing the following variables: baseline insulin concentration (n = 1321), baseline serology for hepatitis B (n = 22) or C (n = 1), and follow-up glucose concentration (n = 1). The remaining individuals were excluded for having positive hepatitis B surface antigen (n = 669) and hepatic C antibody (n = 12). To minimize the confounding effects of alcohol on fatty liver, we also excluded 2080 individuals for reporting daily alcohol intake of at least 20 g. After these exclusions, 11,091 were eligible for the study.

The study was approved by the institutional review board at Kangbuk Samsung Hospital. Informed consent requirement was waived because personal identifying information was not accessed.

Measurements

The health examination consisted of a full medical history, comprehensive blood test evaluation, and abdominal ultrasound. Participants' height and weight were measured barefoot and in light clothing. Body mass index (BMI) was calculated as weight in kilograms divided by height in meters squared. Questionnaires were used to ascertain information regarding years of education and frequency of exercise (none, less than once a week, at least once a week), smoking (never, past, current), and alcohol consumption (both quantity and frequency). Grams of alcohol consumption were calculated by multiplying frequency by amount as previously reported (8).

Laboratory examinations were obtained after an overnight fast. Fasting plasma glucose, alanine aminotransferase, aspartate aminotransferase, triglyceride, and HDL-C concentrations were measured using Bayer Reagent Packs on an automated chemistry analyzer (Advia 1650 Autoanalyzer; Bayer Diagnostics, Leverkusen, Germany) (8, 9). Insulin concentration was measured with an immunoradiometric assay (Biosource, Nivelle, Belgium) with an intra- and interassay coefficient of variation of 2.1–4.5% and 4.7–12.2%, respectively. Insulin quartiles were used as a surrogate measurement of insulin sensitivity. Quartile 1 had the lowest fasting insulin concentration, and quartile 4 had the highest.

Abdominal ultrasonography (Logic Q700 MR; GE, Milwaukee, WI) was performed on all participants. Fatty liver was diagnosed based on known standard criteria (10).

Statistical analysis

Continuous variables were expressed as mean ± SD or median (interquartile range) if not normally distributed. Continuous variables were compared using the independent *t* test. Nonparametric variables were log-transformed before analyses. Categorical variables were expressed as percentage and compared using the χ^2 test. We used logistic regression to determine the odds ratio (OR) of developing T2DM as a function of fatty liver in the total population and in individuals stratified by quartiles of fasting insulin concentration (which was used as a surrogate measure of insulin resistance). We conducted both unadjusted and adjusted analyses. Adjustments were made for the following variables: age, gender, BMI, education (<16 yr, ≥ 16 yr), smoking status (never/past, current), exercise frequency (less than once a week, or at least once a week), and alcohol intake (grams/day). Because baseline glucose concentration is a strong predictor of T2DM (11), we also performed a separate analysis that adjusted for all previous variables and also baseline glucose concentration.

All statistical analyses were performed using SAS version 9.1.3 (SAS Institute, Inc., Cary, NC).

Results

At baseline, 27% of the population had evidence of fatty liver by ultrasound. As seen in Table 1, all baseline characteristics were significantly different between individuals with fatty liver compared with those without fatty liver. Although the study population had more men than women, 88% of those with fatty liver were male. Almost two thirds (63%) of the individuals with fatty liver were also overweight or obese compared with 19% in the individuals without fatty liver. They also had higher blood pressure and had more metabolic abnormalities including higher glucose, insulin, and triglyceride concentration and lower HDL-C concentration. Almost half (47%) of the individuals with baseline fatty liver were in the upper quartile of fasting insulin concentration compared with 17% in those without fatty liver.

Out of a total of 11,091 individuals, 174 (1.6%) developed T2DM at 5 yr. When separated by fatty-liver status as shown in Table 2, only 0.7% of individuals without baseline fatty liver developed T2DM compared with 4% of individuals with fatty liver. Even after adjustment for multiple variables including baseline glucose concentration, the OR of developing T2DM was 2-fold higher in individuals with baseline fatty liver.

Table 2 also shows the incidence of T2DM in individuals stratified by baseline fasting insulin concentration. With the exception of quartile 2, the OR of developing T2DM was significantly increased in individuals with baseline fatty liver similar to the total population. Therefore, regardless of being in the lowest or highest quartile of fasting insulin concentration, the identification of fatty liver at baseline significantly increased the OR of devel-

TABLE 1. Baseline characteristics

	No fatty liver	Fatty liver	*P*
n	8120	2971	
Age (yr)	40.7 ± 6.3	41.4 ± 6.0	<0.001
No. of males (%)	4626 (57%)	2610 (88%)	<0.001
BMI (kg/m^2)	22.8 ± 2.5	25.9 ± 2.4	<0.001
BMI class, no. (%)			<0.001
Normal (<25 kg/m^2)	6569 (81%)	1093 (37%)	
Overweight (25-29.9 kg/m^2)	1487 (18%)	1722 (58%)	
Obese (≥30 kg/m^2)	48 (1%)	152 (5%)	
Education, no. (%)			<0.001
≤12 yr	2154 (28%)	559 (20%)	
12-16 yr	727 (9%)	206 (7%)	
≥16 yr	4913 (63%)	2065 (73%)	
Exercise, no. (%)			<0.001
None	2376 (30%)	805 (28%)	
<1 time/wk	2680 (34%)	1225 (42%)	
≥1 time/wk	2884 (36%)	878 (30%)	
Smokers, no. (%)			
Never	4895 (62%)	1171 (40%)	<0.001
Past	1207 (15%)	747 (26%)	
Current	1810 (23%)	982 (34%)	
Alcohol (g/d)	5 ± 6	7 ± 6	<0.001
Systolic blood pressure (mm Hg)	112 ± 13	118 ± 13	<0.001
Diastolic blood pressure (mm Hg)	73 ± 10	77 ± 10	<0.001
Heart rate (beats/min)	66 ± 9	68 ± 9	<0.001
Triglyceride (mg/dl)	98 [73, 136]	166 [122, 225]	<0.001
HDL-C (mg/dl)	55 [48, 63]	49 [44, 55]	<0.001
Fasting glucose (mg/dl)	92 ± 8	96 ± 9	<0.001
Fasting insulin (μIU/ml)	6.2 [5.2, 7.7]	8.2 [6.4, 10.1]	<0.001
Insulin quartile, no. (%)			<0.001
Quartile 1 (<5.39 μIU/ml)	2468 (30%)	307 (10%)	
Quartile 2 (5.39-6.61 μIU/ml)	2262 (28%)	511 (17%)	
Quartile 3 (6.62-8.50 μIU/ml)	2002 (25%)	768 (26%)	
Quartile 4 (≥8.51 μIU/ml)	1388 (17%)	1385 (47%)	
Alanine aminotransferase (IU/liter)	20 [16, 26]	36 [26, 50]	<0.001
Aspartate aminotransferase (IU/liter)	22 [19, 26]	27 [23, 33]	<0.001

Data are expressed as mean ± SD or median [interquartile range], unless indicated otherwise. To covert triglyceride from mg/dl to mmol/liter, multiply by 0.0113; HDL-C from mg/dl to mmol/liter, 0.0259; glucose from mg/dl to mmol/liter, 0.0555; insulin from μIU/ml to pmol/liter, 6.945.

oping T2DM at 5 yr. In individuals in quartiles 3 and 4, the OR remained significantly increased even after adjustment for baseline fasting glucose.

To better understand the role of fatty liver in increasing T2DM risk independent of insulin resistance, we compared baseline characteristics in individuals with and without fatty liver in the lowest (quartile 1) and highest (quartile 4) insulin quartiles (Supplemental Table 1, published on The Endocrine Society's Journals Online web site at http://jcem.endojournals.org). The purpose of this analysis was to isolate differences between individuals by fatty liver status within the same insulin quartile. As with the whole population shown in Table 1, all baseline characteristics were different between individuals with and with-

TABLE 2. OR for T2DM at 5-yr follow-up

	T2DM [no./total no. (%)]		OR (95% confidence interval)		
	No fatty liver	Fatty liver	Unadjusted	Adjusted[a]	Adjusted[a] + baseline glucose
All	54/8120 (0.7%)	120/2971 (4%)	6.29 (4.55-8.69)	3.24 (2.19-4.78)	2.05 (1.35-3.12)
Insulin					
Quartile 1	13/2468 (0.5%)	8/307 (2.6%)	5.05 (2.08-12.29)	3.47 (1.23-9.79)	1.96 (0.63-6.13)
Quartile 2	16/2262 (0.7%)	6/511 (1.2%)	1.67 (0.65-4.28)	1.34 (0.46-3.87)	0.71 (0.22-2.26)
Quartile 3	11/2002 (0.6%)	22/768 (2.9%)	5.34 (2.58-11.06)	3.74 (1.59-8.84)	2.92 (1.12-7.62)
Quartile 4	14/1388 (1.0%)	84/1385 (6.1%)	6.34 (3.58-11.21)	3.31 (1.76-6.20)	2.42 (1.23-4.75)

[a] Adjusted for age, gender, BMI, alcohol (grams per day), education (<16 yr, ≥ 16 yr), smoking (never or past, current), and exercise (<1 time/wk, ≥1 time/wk).

out fatty liver, regardless of baseline fasting insulin concentration. Therefore, compared with individuals in the same insulin quartile without fatty liver, individuals with fatty liver were heavier, had higher blood pressure, and had more unfavorable metabolic characteristics.

Interestingly, clinical characteristics in individuals in insulin quartile 1 with fatty liver were all significantly different compared with individuals in quartile 4 without fatty liver ($P < 0.001$), with the exception of baseline fasting glucose concentration ($P = 0.41$). Instead of having a better metabolic profile based on insulin classification, individuals in insulin quartile 1 with fatty liver were heavier, had higher blood pressure, and had worse metabolic characteristics compared with individuals in quartile 4 without fatty liver. Therefore, despite being in the lowest insulin quartile, the 307 individuals in quartile 1 with fatty liver appeared more insulin resistant by clinical characteristics than the 1388 individuals in insulin quartile 4 without fatty liver.

Discussion

Our study shows in a large population of relatively healthy individuals that identifying fatty liver by ultrasound strongly predicts the development of T2DM in 5 yr. In addition, our findings reveal a complex relationship between baseline fatty liver and fasting insulin concentration. As expected, individuals with fatty liver were more likely to have higher insulin concentration compared with those without fatty liver, and nearly half of those with fatty liver were in the top insulin quartile. However, the presence of fatty liver also identified individuals with worse clinical and metabolic profile and greater risk for developing T2DM regardless of baseline fasting insulin concentration. Therefore, the presence of fatty liver disease, although associated with insulin resistance, likely has an independent effect on the risk of developing T2DM.

Our results add to the growing evidence that having fatty liver predicts the development of T2DM (1–3). Our study, however, is the only study to have measured baseline insulin concentration and to have evaluated the risk of developing T2DM among individuals stratified by insulin concentration. Contrary to our hypothesis, the presence of fatty liver increased the risk of developing T2DM, regardless of baseline fasting insulin concentration. There are likely two explanations for this finding. First, fasting insulin concentration may not accurately reflect insulin action for all individuals. Although fasting insulin concentration is significantly associated with insulin resistance, it reflects less than 40% of the variability in insulin resistance as measured by a direct technique (12). In this study, individuals with baseline fatty liver had more stigmata of insulin resistance (higher glucose and triglyceride and lower HDL-C concentration) regardless of being in the lowest or highest quartile of fasting insulin. In addition, in the small group of individuals (2.8% of the population) in insulin quartile 1 with fatty liver, the metabolic profile was not only worse compared with counterparts in the same insulin quartile without fatty liver, but worse compared with individuals in quartile 4 without fatty liver. Therefore, the presence of fatty liver may better indicate the presence or severity of insulin resistance than fasting insulin concentration. Second, the presence of fatty liver may not only identify insulin-resistant individuals but may also independently increase the risk of T2DM. The liver secretes many factors, known as hepatokines, which may modulate the risk of T2DM (7). For example, plasminogen-activator inhibitor-1 is an inflammatory marker that is secreted by the liver and predicts the development of T2DM (13). Recently, Ardigò *et al.* (14) found that plasma concentration of plasminogen-activator inhibitor-1 was only elevated in individuals with both evidence of insulin resistance and ultrasound-diagnosed fatty liver, and not insulin resistance alone. Therefore, it is conceivable that the presence of fatty liver, through hepatokines, may modulate the risk of T2DM in insulin-resistant individuals.

In conclusion, we have shown that fatty liver, diagnosed by ultrasound, strongly predicts the future development of T2DM. Ultrasound detects moderate to severe steatosis [>30% fat on liver biopsy (15)]; therefore, our findings may not be applicable to those with milder steatosis. Nonetheless, fatty liver detectable by ultrasound identified individuals with worse metabolic profile and greater risk for T2DM, regardless of baseline fasting insulin concentration. Therefore, our findings suggest that fatty liver, although associated with insulin resistance, is also an independent predictor of T2DM.

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References

1. **Fan JG, Li F, Cai XB, Peng YD, Ao QH, Gao Y** 2007 Effects of nonalcoholic fatty liver disease on the development of metabolic disorders. J Gastroenterol Hepatol 22:1086–1091
2. **Kim CH, Park JY, Lee KU, Kim JH, Kim HK** 2008 Fatty liver is an independent risk factor for the development of type 2 diabetes in Korean adults. Diabet Med 25:476–481
3. **Yamada T, Fukatsu M, Suzuki S, Wada T, Yoshida T, Joh T** 2010 Fatty liver predicts impaired fasting glucose and type 2 diabetes mellitus in Japanese undergoing a health checkup. J Gastroenterol Hepatol 25:352–356
4. **Reaven GM** 1988 Banting lecture 1988. Role of insulin resistance in human disease. Diabetes 37:1595–1607
5. **Martin BC, Warram JH, Krolewski AS, Bergman RN, Soeldner JS, Kahn CR** 1992 Role of glucose and insulin resistance in development of type 2 diabetes mellitus: results of a 25-year follow-up study. Lancet 340:925–929
6. **Fabbrini E, Magkos F, Mohammed BS, Pietka T, Abumrad NA, Patterson BW, Okunade A, Klein S** 2009 Intrahepatic fat, not visceral fat, is linked with metabolic complications of obesity. Proc Natl Acad Sci USA 106:15430–15435
7. **Stefan N, Kantartzis K, Häring HU** 2008 Causes and metabolic consequences of fatty liver. Endocr Rev 29:939–960
8. **Sung KC, Kim SH, Reaven GM** 2007 Relationship among alcohol, body weight, and cardiovascular risk factors in 27,030 Korean men. Diabetes Care 30:2690–2694
9. **Sung KC, Ryan MC, Kim BS, Cho YK, Kim BI, Reaven GM** 2007 Relationships between estimates of adiposity, insulin resistance, and nonalcoholic fatty liver disease in a large group of nondiabetic Korean adults. Diabetes Care 30:2113–2118
10. **Bae JC, Cho YK, Lee WY, Seo HI, Rhee EJ, Park SE, Park CY, Oh KW, Sung KC, Kim BI** 2010 Impact of nonalcoholic fatty liver disease on insulin resistance in relation to HbA1c levels in nondiabetic subjects. Am J Gastroenterol 105:2389–2395
11. **Sung KC, Reaven GM, Kim SH** 2010 Utility of homeostasis model assessment of β-cell function in predicting diabetes in 12,924 healthy Koreans. Diabetes Care 33:200–202
12. **Kim SH, Abbasi F, Reaven GM** 2004 Impact of degree of obesity on surrogate estimates of insulin resistance. Diabetes Care 27:1998–2002
13. **Festa A, D'Agostino Jr R, Tracy RP, Haffner SM** 2002 Elevated levels of acute-phase proteins and plasminogen activator inhibitor-1 predict the development of type 2 diabetes: the insulin resistance atherosclerosis study. Diabetes 51:1131–1137
14. **Ardigò D, Franzini L, Valtueña S, Numeroso F, Piatti PM, Monti L, Reaven GM, Zavaroni I** 2010 The increase in plasma PAI-1 associated with insulin resistance may be mediated by the presence of hepatic steatosis. Atherosclerosis 208:240–245
15. **Saadeh S, Younossi ZM, Remer EM, Gramlich T, Ong JP, Hurley M, Mullen KD, Cooper JN, Sheridan MJ** 2002 The utility of radiological imaging in nonalcoholic fatty liver disease. Gastroenterology 123:745–750