Resolution of fatty liver and weight loss: Independent associations with changes in serum lipids and apolipoproteins

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ABSTRACT

Background and aims: It is uncertain whether resolution of fatty liver can improve cardiovascular disease risk factors, independently of changes in body mass index (BMI). Our aim was: to test whether resolution of fatty liver is associated with improvements in components of the lipid profile, independently of changes in BMI; and to quantify and compare the magnitude of benefit of resolution of liver fat, and decreases in BMI on the lipid profile.

Methods: 36,195 subjects with fatty liver were studied. Persistence/resolution of fatty liver was determined by ultrasound at follow up (Mean=4.93 years). Total cholesterol, triglycerides, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, and apolipoproteins were measured at baseline and follow up. Regression modelling was undertaken to test the independence of associations between change in fatty liver status, or change in BMI, with any change in lipid profile concentrations between baseline and follow up.

Results: Mean (SD) age was 36.3±6.6 and 39.8±8.7 years (men and women, respectively). Resolution of fatty liver occurred in 7,086, and persisted in 29,109 subjects. Mean±SD weight change was -3.2±4.3 (~1 kg/m^2 decrease in BMI) with resolution of, and +0.5±3.5 kgs with persistence of, fatty liver, respectively. Both resolution of fatty liver and decrease in BMI were independently associated with improvements in all components of the lipid profile and there was a similar magnitude of benefit associated with resolution of fatty liver, or 1 kg/m^2 decrease in BMI.

Conclusions: Resolution of fatty liver improves the lipid profile, independently of weight loss.

Keywords: BMI, obesity, non alcoholic fatty liver disease, fatty liver, lipid, insulin resistance, metabolic syndrome

1. Introduction

Recent evidence shows that non alcoholic fatty liver disease (NAFLD) is an independent cardiovascular disease (CVD) risk factor [1-3]. Several different mechanisms have been proposed that may explain the association between NAFLD and CVD and these include increased inflammation, altered paracrine cell signalling, increased angiogenesis and secretion of hepatokines, as well as altered redox status, hemostasis and lipoprotein profiles [1,2]. Accumulation of liver fat is used to diagnose NAFLD [4] and hepatic fat accumulation is strongly associated with dyslipidaemia, specifically, hypertriglyceridemia, and a low HDL-C concentration [5]. It has also been shown that patients with NAFLD have markedly higher plasma apolipoprotein B to apolipoprotein A1 ratios and smaller LDL particle sizes, and these changes were independent of obesity or steatohepatitis [6]. These data suggest that in NAFLD the severity of liver disease does not affect dyslipidaemia and the best predictor of atherogenic dyslipidaemia is liver fat accumulation and insulin resistance [6].

Obesity is a risk factor for increased all cause mortality [7-10]. Obesity frequently occurs with NAFLD, diseases associated with NAFLD (e.g. CVD and type 2 diabetes [3,11-16]), and the greater the BMI, the greater the risk of type 2 diabetes and CVD [17]. In obese subjects, it is recognised that a 5-10% weight loss produces a clinically meaningful benefit on obesity-related outcomes [17,18], and when patients lose as little as 5% of body weight, there is an even greater decrease in hepatic fat [19]. However, within any obese group of subjects, there are subjects who have a more metabolically healthy profile (so called metabolically healthy obesity) which is important because 5-10% weight loss does not improve obesity-related outcomes to the same extent in all subjects [20].

Although it is well known that weight loss produces an improvement in certain

components of the lipid profile such as atherogenic dyslipidaemia [21,22] and decreases liver fat [23], it is uncertain whether resolution of fatty liver in NAFLD improves the lipid profile, independently of any decrease in body mass index (BMI). Since weight loss does not improve obesity-related outcomes to the same extent in all individuals, our aim was therefore to test whether resolution of fatty liver was associated with improvements in components of the lipid profile, independently of changes in BMI. Additionally, we aimed to quantify the magnitude of benefit on the lipid profile components, of resolution of liver fat, versus decreases in BMI.

2. Materials and Methods

The study population consisted of individuals who participated in a comprehensive health screening program, at least twice, at Kangbuk Samsung Hospital, Seoul and Suwon, Korea from 2007 to 2014 (n = 259,011). Among these subjects, fatty liver was present at baseline in 67,138 subjects. Subjects were excluded if they were known to be treated with lipid lowering therapy (including statin treatment). Subjects were excluded if they were receiving treatment for diabetes, hypertension or CVD. Subjects were excluded: age<20 years; data recording the presence or absence of fatty liver at follow up were missing (n = 160); alcohol intake was >30g/day (men) or >20g/day (women); there was missing data, (n = 11,299 at baseline and n = 12,642 at follow up); HCV antibody status was positive (n = 34 at baseline and n = 69 at follow up); HBsAg status was positive (n = 840 at baseline and n = 1,753 at follow up); there was evidence of cancer (n = 682 at baseline and n = 1,675 at follow up; data on physical activity were missing (n = 974 at baseline and 2,119 at follow up; data on BMI were missing n = 5 at baseline and n = 73 at follow up; data recording blood pressure medication were missing (n = 4,901 at baseline and n = 9,298 at follow up; data on medication for treatment of diabetes were missing n = 1,711 at baseline and n = 4,471 at

follow up; and data recording lipid lowering therapy were missing n = 1,038 at baseline and n = 4,404 at follow up. After these exclusions the final number of subjects included in the analyses was n = 36,195. Mean follow up was 4.93 years, median 3.94 years (minimum 0.50 and maximum 12.65 years). The study was approved by the Institutional Review Board of Kangbuk Samsung Hospital and any requirement for informed consent was waived by the Board, because de-identified information was retrieved retrospectively.

2.1. Measurements

As part of the health screening program, individuals completed self-administered questionnaires, related to their medical and social history and medication use. Individuals were asked about duration of education (years), regular exercise, smoking history (never, former, or current) and alcohol consumption (grams, g/week). Trained staff also collected anthropometric measurements and vital statistics. Body weight was measured in light clothing with no shoes to the nearest 0.1 kilogram using a digital scale. Height was measured to the nearest 0.1 centimeter. Body mass index (BMI) was calculated as weight in kilograms divided by height in meters squared. Blood samples were collected after at least 10-hours of fasting and analyzed in the same core clinical laboratory. The core clinical laboratory has been accredited and participates annually in inspections and surveys by the Korean Association of Quality Assurance for Clinical Laboratories. Blood was drawn from participants after fasting for ≥10 hours and analyzed at the Laboratory Medicine Department at the Kangbuk Samsung Hospital. Serum levels of glucose, total cholesterol, triglycerides, low-density lipoprotein cholesterol (LDL-C), and high-density lipoprotein cholesterol (HDL-C) were measured using Bayer Reagent Packs (Bayer Diagnostics, Leverkusen, Germany) on an automated chemistry analyzer (Advia 1,650 Autoanalyzer; Bayer Diagnostics, Leverkusen, Germany). Apolipoprotein (apo) B and A1 concentrations were determined by rate nephelometry

(IMMAGE system; Beckman Coulter). Insulin was measured with an immunoradiometric assay (Biosource, Nivelle, Belgium) and HOMA-IR was calculated. High sensitivity-C reactive protein was analysed by particle-enhanced immunonephelometry with the BNIITM System (Dade Behring, Marburg, Germany) with a lower detection limit of 0.1 mg/L.

We assessed the weekly frequency of moderate- or vigorous-intensity physical activity which was assessed using the validated Korean version of the International Physical Activity Questionnaire Short Form (IPAQ-SF) [24]. The IPAQ-SF measures the frequency and duration of moderate to vigorous physical activity undertaken for more than 10 continuous minutes across all usual activities at work, home or during leisure for middle aged individuals during a seven-day period. Abdominal ultrasonography (Logic Q700 MR; GE, Milwaukee, WI, USA) was undertaken by clinical radiologists using a 3.5MHz probe for all subjects at baseline and after five years. The following images were undertaken; i) sagittal view of the right lobe of the liver and right kidney, ii) transverse view of the left lateral segment of the liver and spleen and iii) transverse view of the liver for altered echo texture. Fatty infiltration of the liver (fatty liver) was identified if there was an increase in echogenicity of the liver compared with the echogenicity of the renal cortex where the diaphragm and intrahepatic vessels appeared normal [25].

2.2. Statistical analyses

Statistical analyses were performed using STATA version 14.2 (StataCorp LP, College Station, TX, USA). Reported *p* values were two-tailed, and <0.05 were considered statistically significant. The distribution of continuous variables was evaluated and transformations were conducted for nonparametric variables. Baseline and follow up difference were evaluated using Kernel density plot, all variables are normally distributed, we conducted parametric tests (paired Student's t test). Analysis of variance (ANOVA) was undertaken to test for

between group differences. Multiple linear regression was undertaken to investigate the associations between change in lipid profile measurements (arithmetic difference between end of study concentration minus baseline concentration). Binary logistic regression modelling was undertaken to test the independence of associations between change in fatty liver status, and the change in lipid profile concentrations between baseline and follow up (total cholesterol, triglycerides, LDL-C, HDL-C, apo B-100 and apo A1 concentrations). Regression models were adjusted for age, sex, alcohol intake in grams, smoking status, educational attainment, exercise, baseline BMI (or waist circumference), change in alcohol, change in exercise, change in HOMA-IR and each component of the lipid profile at baseline. When testing associations between change in components (D) of the lipid profile and change in BMI, the model was adjusted for change in fatty liver status and when testing associations between change in components of the lipid profile and change in fatty liver status, the model was adjusted for change in BMI or change in waist circumference.

3. Results

36,195 subjects with fatty liver were studied. Mean (SD) age was 36.3±6.6 and 39.8±8.7 years (men and women, respectively). Fatty liver status at follow up was assessed by ultrasound (Mean = 4.93 years) and resolution of fatty liver occurred in 7,086 and persisted in 29,109 subjects. Mean±SD weight change was -3.2±4.3 and +0.5±3.5 kgs for groups with resolution and persistence of fatty liver respectively.

Table 1 describes the baseline and follow up characteristics of men in whom there was resolution of fatty liver at follow up examination (n = 5,470 subjects) and in whom fatty liver persisted at follow up examination (n = 25,521 subjects). **Table 2** describes the baseline and follow up characteristics of women in whom there was resolution of fatty liver at follow up

examination (n = 1,616 subjects) and in whom fatty liver persisted at follow up examination (n = 3,588 subjects). For both men and women, only in the group in whom there was resolution of fatty liver was there a marked improvements in all components of the lipid profile (total cholesterol, LDL-C, HDL-C, triglyceride, apolipoprotein-B and apolipoprotein A-I (For women, the improvement in LDL-C and Apo-B was non significant, although the changes were in the same direction as for men).

For both men and women, in the group in whom there was persistence of fatty liver, there a worsening in all components of the lipid profile (total cholesterol, LDL-C, HDL-C, triglyceride, apolipoprotein-B and apolipoprotein A-I. In the group in whom there was resolution of fatty liver there was also a ~1 kg/.m^2 decrease in BMI in men and women, compared with a ~0.2kg/m^2 increase in BMI in men and women. **Supplementary Tables 1** (men) and **2** (women) show the change in anthropometric and biochemical variables between baseline and follow up according to resolution or persistence of fatty liver and change in BMI between baseline and end of study. These data show that the most marked improvements in lipid variables between baseline and follow up, occurred in those subjects in whom there was both resolution of fatty liver and a decrease in BMI between baseline and follow up. **Supplementary Tables 3** (men) and **4** (women) show the change in biochemical and anthropometric measures according to decrease in BMI (BMI<0) and increase in BMI (BMI≥0). These data illustrate the beneficial effect of weight loss on components of the lipid profile (in contrast to the harmful impact of weight gain).

Next we tested whether the associations between improvements in the lipid variables and resolution of fatty liver, were independent of decreases in BMI and potential confounders. Since improvements in insulin sensitivity and resolution of fatty liver are co-linear variables, first in **Supplementary Table 5**, we show the change in lipid variables without adjustment for D-HOMA-IR, and then in **Table 3**, we show the change in lipid variables after adjustment for

D-HOMA-IR. As can be seen even after further adjustment for D-HOMA-IR (**Table 3**), there were significant and independent improvements in LDL-C, HDL-C, triglyceride, Apo-B and Apo-A-I concentrations, associated with resolution of fatty liver. Finally, to test the independence of associations between changes in BMI and changes in lipid variables, we investigated associations between change in BMI and change in lipid variables, adjusting for resolution of liver fat, as well as other potential confounders. **Supplementary Table 6** shows the model without adjustment for HOMA-IR, and **Table 4** shows the model after adjustment for HOMA-IR. These data illustrate that even after full adjustment (**Table 4**), that included adjustment for change in HOMA-IR, change in BMI was associated with a decrease in LDL-C, triglyceride and Apo-B and an increase in HDL-C and Apo A-I concentrations.

In men and women, in the group with resolution of fatty liver, all measured components of the lipid profile improved. For example, the improvement in triglyceride concentration was a decrease in ~27 mg/dL (~17%) in men and a decrease in ~19 mg/dL (~16%) in women from baseline, and for both sexes, the decrease in BMI at follow up was ~1kg/m^2 (which represented ~ 3kg weight loss). In the regression model shown in **Table 4**, the B coefficient for change in triglyceride, indicates that for a 1 kg/m^2 decrease in BMI (for men and women combined), there was a 14.3 mg/dL decrease in triglyceride concentration. For comparison, with resolution of fatty liver (Table 3), (for men and women combined), there was a 9.2 mg/dL decrease in triglyceride concentration. For Apo A-I, the improvement was slightly greater with resolution in fatty liver, than with a decrease of 1 kg/m^2 in BMI; for the other measured components of the lipid profile, the improvements associated with decrease in BMI were slightly greater than the improvements associated with resolution in fatty liver.

In this cohort waist circumference was only available in 22,440 subjects (~60%) of the subjects. However, to test whether resolution of fatty liver was independently associated with changes in the lipid parameters after adjustment for baseline waist circumference, change in

waist circumference and the same co-variates as shown in Tables 3 and 4, we repeated the regression models shown in Tables 3 and 4, replacing BMI with waist circumference. Resolution of fatty liver was independently associated with similar changes in each of the lipid parameters after adjusting for waist circumference and change in waist circumference (**Supplementary Table 7**). In men, there was a more marked decrease in serum triglyceride concentration (14.27 mg/dL) with resolution of fatty liver, after adjusting for waist circumference (**Supplementary Table 7)**; compared with the change in serum triglyceride concentration (9.19 mg/dL) with resolution of fatty liver, after adjusting for BMI (**Table 3**). In contrast, in women, the decrease in serum triglyceride concentration (10.38 mg/dL) was remarkably similar with resolution of fatty liver, after adjusting for waist circumference (**Supplementary Table 7)**; compared with the change in serum triglyceride concentration (10.03 mg/dL) with resolution of fatty liver, after adjusting for BMI (**Table 3**).

Next, we investigated the change in lipid parameters per unit change (1cm) in waist circumference (**Supplementary Table 8)**, adjusting for resolution of fatty liver and the same covariates as shown in **Table 4**. These data were very similar to those shown for **Table 4**, although the magnitude of the change in lipid parameters per 1 cm decrease in waist circumference was much smaller than the magnitude of the change in lipid parameters per 1kg/m^2 decrease in BMI. For both men and women a 5cm decrease in waist circumference was associated with similar decreases in serum triglyceride concentrations as a 1kg/m^2 decrease in BMI.

4. Discussion

Our novel results obtained in 36,195 men and women with existing fatty liver at baseline show that resolution of fatty liver (that occurred in almost 7,086 people), was associated with improvements in lipid variables (LDL-C, triglyceride and Apo-B, HDL-C and Apo A-I

concentrations) independently of any decrease in BMI, or other potential confounders. Mean±SD weight change was -3.2±4.3 kgs in subjects in whom there was resolution of fatty liver, and there was a small amount of weight gain i.e. +0.5±3.5 kgs in subjects in whom there was persistence of fatty liver at follow up. Importantly, we have been able to adjust for a comprehensive range of covariates and potential confounders. Specifically, we have adjusted for age, alcohol intake, smoking status, educational status, exercise, BMI and concentration of each lipid variable at baseline; and importantly we also adjusted for changes in exercise, alcohol intake, BMI, and HOMA-IR, between baseline and follow up examination. Although waist circumference was only available in ~60% of this cohort, we have also verified our results (showing that resolution of fatty liver is associated with changes in the lipid parameters, independently of weight loss), by adjusting for baseline waist circumference and change in waist circumference. Adjusting for waist circumference did not attenuate the strength of the association between resolution of fatty liver and change in lipid profile parameters, which we had observed after adjusting for BMI. In men, after adjustment for waist circumference, the association between resolution of fatty liver and decrease in serum triglyceride concentration was strengthened (compared with the association between resolution of fatty liver and decrease in serum triglyceride concentration, after adjustment for BMI).

Current population-based prevalence of NAFLD is approximately 30-40% in men and 15-20% in women [26] and is even higher in people with type 2 diabetes mellitus (T2DM), occurring in up to 70% of this group of patients [27]. Recent evidence shows that NAFLD is an independent risk factor for CVD [3] and NAFLD increases risk of incident CVD by approximately ~60% [3]. Given that NAFLD is very common in patients with obesity [28] and is a strong risk factor for CVD, it is very important to investigate the effects of resolution of fatty liver on improvements in CVD risk factors. The relationship between decrease in liver

fat content and weight loss is variable between individuals, and therefore it is important to test the independent effects of both resolution of fatty liver and decrease in weight, in studying the effects of both, on change in lipid profile. It has been postulated that the large variability in the change in liver fat with weight loss, may be due to variable changes in cardiorespiratory fitness [29]. In this study, the authors showed that cardiorespiratory fitness, independently of total adiposity, body fat distribution and exercise intensity, determined liver fat content in humans, suggesting that fitness and liver fat are causally related to each other. Furthermore, measurement of fitness at baseline predicted the effectiveness of a lifestyle intervention to decrease hepatic steatosis. Taken together with our results, these data emphasise to clinicians that weight loss and resolution of fatty liver are different, and have independent beneficial effects on the lipid profile. Thus in subjects with fatty liver we reason that clinicians should emphasise to patients that there should be a dual focus on both decreasing body fat and ameliorating liver fat. Since some patients have great difficulty in losing body fat but can increase their fitness (that may independently ameliorate liver fat [30]), our data lend supporting evidence with which to provide encouragement to these patients. It is also interesting to note that there was a slighter higher alcohol intake in the group with resolution of fatty liver than in subjects with persisting fatty liver. For example, for men, the baseline group comparisons for persisting fatty liver, versus resolution of fatty liver group was 8.58 vs. 8.86 g/day, (p-value=0.011). At the end of the study, group comparison for alcohol intake was 8.26 vs. 8.67 g/day, p-value<0.001, for persisting fatty liver versus resolution of fatty liver. For women, the daily alcohol intake was much lower than in men. However, the baseline group comparison for persisting versus resolution of fatty liver was 1.75 vs. 1.88 d/day, p-value=0.19; and the end of study group comparison was 1.92 vs. 2.28 g/day, p-value<0.001, for persisting fatty liver versus resolution of fatty liver, respectively. We adjusted the regression models for change in alcohol intake, and therefore we do not consider that alcohol

intake confounds our data, although, it is plausible that the slightly higher alcohol intake in the group with resolution of fatty liver represents an unmeasured potential difference in lifestyle between groups.

Besides showing that resolution of fatty liver is associated with favourable changes in the lipid profile, our results show that decrease in BMI was independently associated with improvements in the lipid profile. Therefore, these data emphasise that both resolution of fatty liver and weight loss are having beneficial, independent effects on all measured components of the lipid profile. The magnitude of weight loss in subjects with resolution of fatty liver was ~3kg for both men and women, which was approximately a decrease in BMI of ~1 kg/m^2 for both sexes. The results of regression modelling shown in Tables 3 and 4, illustrate that the effects of a 1 kg/m^2 decrease in BMI was very similar to the effect of resolution of fatty liver. For example, for change in triglyceride concentration, a 1 kg/m^2 decrease in BMI, was associated with a 14.3 mg/dL decrease in triglyceride concentration and for resolution of fatty liver, the associated change in triglyceride concentration was 9.2 mg/dL between baseline and follow up.

There are strengths and limitations of our study that need to be considered. We have studied a single population of Koreans largely comprising one ethnic group and consequently, our results cannot be extrapolated to other ethnic groups. Subjects with a known diagnosis of diabetes, hypertension, CVD, cancer, or dyslipidaemia were excluded. Subjects were also excluded if they were taking medication for these conditions (including all lipid lowering therapies). Consequently, changes in the lipid profile between baseline and follow up are not due to these conditions or to the effects of lipid lowering medication. We have been able to adjust for multiple key potential confounders that are known to modify liver fat such as physical activity, alcohol, BMI (and also change in these variables between baseline and follow up. We have assessed the presence of fatty liver using abdominal ultrasonography at

baseline and follow up. Whilst the sensitivity of ultrasound for detecting fatty liver is limited to identification of >25% fat infiltration [31], and the detection of liver fat can be affected by severe obesity, in our predominantly single ethnic group population, there were very few severely obese subjects. The mean BMI for men and women with fatty liver at baseline was between 24.6-26.1 kg/m^2, and the SDs for both groups were ~3kg/m^2. Consequently there were very few subjects with morbid obesity in whom assessment of fatty liver status by ultrasound would be unreliable because of excess body fat. Although we acknowledge that it is possible that subjects with very low levels of liver fat at follow up would not have been identified by ultrasound, any misclassification bias in the 'resolution of fatty liver group', would only act to attenuate the strength of the associations we have observed between resolution of fatty liver and improvements in lipid profile components. Additionally, it is plausible that different lifestyle factors occurred in the group with resolution of fatty liver but it was not possible to study lifestyle factors with this study design. We were also unable in this study design to address the underlying mechanisms or to identify the genetic (and epigenetic) factors that possibly define the subset of patients that showed resolution of fatty liver, as this was beyond the scope of this study and such data are not available in this cohort.

We have not undertaken a randomized placebo-controlled trial (RCT) to test the effect of decreasing liver fat on CVD risk factors, which would have been the gold standard approach to addressing the effect of resolution of fatty liver on change in the lipid profile. Whilst we are not able to elucidate why there was resolution of fatty liver in subjects, there was resolution of fatty liver in 7086 subjects, and we have opportunistically assessed the improvements in lipid profile components associated with resolution of fatty liver over ~4 years of follow up.

5. Conclusion

In conclusion, our novel results add to existing evidence by showing that resolution of fatty liver improves all the measured components of the lipid profile (total cholesterol, triglycerides, LDL-C cholesterol, HDL-C, Apo B-100 and Apo A-I), independently of weight loss. With resolution of fatty liver, the decrease in triglyceride concentration was~17% in men and ~16% in women, and the decrease in BMI was ~1kg/m^2 (which represented ~ 3kg weight loss for both sexes. The independent effects of resolution of fatty liver and weight loss were very similar on each of the measured components of the lipid profile.

Conflict of interest

The authors declared they do not have anything to disclose regarding conflict of interest with respect to this manuscript.

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Author contributions

K.S contributed to the hypothesis. K.S. wrote the methods and contributed to discussion. M.L analyzed the data. J.L., S.L., J.K., S.W. and C.D.B. wrote the introduction, results and discussion, K.S. is the guarantor for the article.

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Tables

Table 1

Baseline and follow up data for men according to resolution or persistent fatty liver (men) (n = 30,991)

	Persistent fatty liver at follow up			Resolution fatty liver at follow up		
	Baseline (n = 25,521)	Follow up (n = 25,521)	*p*	Baseline (n = 5,470)	Follow up (n = 5,470)	*p*
Age (years)	36.30±6.55	41.24±6.84	<0.001	37.00±7.01	42.34±7.29	<0.001
BMI (kg/m^2)	26.01±2.63	26.20±2.74	<0.001	25.39±2.49	24.37±2.27	<0.001
Systolic BP (mmHg)	117.31±11.62	116.58±11.95	<0.001	117.05±11.74	113.83±11.72	<0.001
Diastolic BP (mmHg)	76.42±8.82	76.77±9.45	<0.001	76.12±8.78	74.55±9.19	<0.001
Education			<0.001			0.084
≤high school	1,869(7.32)	2,043(8.01)		451(8.24)	482(8.81)	
Higher education (%)(>High school)	15.712(61.56)	20,235(79.29)		3,269(59.76)	4,302(78.65)	
Unknown	7,940(31.11)	3,243(12.71)		1,750(31.99)	686(12.54)	
Smoking			<0.001			<0.001
Never/former	14,606(57.23)	16,340(64.03)		3,338(61.02)	3,574(65.34)	
Current	10,580(41.46)	8,624(33.79)		2,089(38.19)	1,776(32.47)	

Unknown	335(1.31)	557(2.18)		43(0.79)	120(2.19)	
Alcohol intake (g/day)	8.58±7.30	8.26±7.29	<0.001	8.86±7.30	8.67±7.43	0.041
Insulin (IU/mL)	7.83±4.31	9.42±5.26	<0.001	6.41±3.35	6.30±3.10	0.182
Glucose (mg/dl)	96.02±11.88	99.63±16.92	<0.001	96.03±13.08	97.14±15.61	<0.001
Total cholesterol (mg/dl)	207.71±33.61	209.05±34.01	<0.001	204.09±33.01	198.23±32.60	<0.001
LDL-C (mg/dl)	129.29±29.40	135.50±30.52	<0.001	125.62±29.16	126.60±29.68	0.004
HDL-C (mg/dl)	47.78±9.17	46.88±9.82	<0.001	49.32±9.77	52.18±11.62	<0.001
Triglycerides (mg/dl)	175.30±100.71	178.69±107.53	<0.001	158.57±88.05	131.55±74.48	<0.001
Apolipoprotein (A)	132.17±19.11	135.98±18.96	<0.001	133.73±19.72	140.76±20.86	<0.001
Apolipoprotein (B)	105.47±22.37	111.34±22.60	<0.001	101.82±21.73	101.07±21.82	0.022
HOMA IR	1.90±1.16	2.33±1.54	<0.001	1.54±0.87	1.50±0.81	0.078
Hs-CRP (mg/L)	0.14±0.28	0.14±0.31	0.153	0.12±0.27	0.13±0.52	0.35
WBC count (10^3/ul)	6.54±1.50	6.47±1.54	<0.001	6.31±1.49	6.04±1.57	<0.001
Exercise (≥1 times per week)	11,835(46.37)	11,184(43.82)	<0.001	2,634(48.15)	3,060(55.94)	<0.001

BMI, body mass index; BP, blood pressure; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, homeostatic model assessment-insulin resistance; Hs-CRP, high-sensitivity C-reactive protein.

Table 2

Baseline and follow up data for men according to resolution or persistent fatty liver (women) (n = 5,204)

	Persistent fatty liver at follow up			Resolution fatty liver at follow up		
	Baseline (n = 3,588)	Follow up (n = 3,588)	*p*	Baseline (n = 1,616)	Follow up (n = 1,616)	*p*
Age (years)	39.75±8.71	44.13±8.53	<0.001	37.27±7.75	41.95±7.73	<0.001
BMI (kg/m2)	25.84±3.26	26.14±3.48	<0.001	24.55±3.16	23.43±2.82	<0.001
Systolic BP (mmHg)	112.21±13.24	112.09±14.43	0.554	109.53±12.99	106.14±13.53	<0.001
Diastolic BP (mmHg)	71.86±9.32	71.75±10.14	0.537	70.10±9.33	68.10±9.60	<0.001
Education			0.258			<0.001
≤High school	984(27.42)	1,014(28.26)		371(22.96)	429(26.55)	
Higher education (%)(>High school)	1,431(39.88)	1,872(52.17)		724(44.8)	968(59.9)	
Unknown	1,173(32.69)	702(19.57)		521(32.24)	219(13.55)	
Smoking			0.007			0.091
Never/former	3,346(93.26)	3,291(91.72)		1,511(93.5)	1,488(92.08)	

Current	90(2.51)	85(2.37)		49(3.03)	39(2.41)	
Unknown	152(4.24)	212(5.91)		56(3.47)	89(5.51)	
Alcohol intake (g/day)	1.75±3.32	1.92±3.20	0.001	1.88±3.45	2.28±3.39	<0.001
Insulin (IU/mL)	9.39±5.12	10.62±6.02	<0.001	7.41±4.31	6.73±4.23	<0.001
Glucose (mg/dl)	96.84±16.61	100.75±23.11	<0.001	94.88±14.27	93.89±12.96	<0.001
Total cholesterol (mg/dl)	203.95±35.93	207.60±35.66	<0.001	197.38±35.02	193.73±32.98	<0.001
LDL-C (mg/dl)	124.98±31.03	131.85±31.51	<0.001	118.45±29.86	118.06±29.17	0.541
HDL-C (mg/dl	52.75±11.07	52.15±11.86	<0.001	56.11±12.46	59.81±13.60	<0.001
Triglycerides (mg/dl)	143.09±89.19	145.87±87.64	0.036	119.42±66.53	100.08±50.99	<0.001
Apolipoprotein (A)	139.26±22.30	142.56±21.59	<0.001	141.54±22.95	150.11±24.63	<0.001
Apolipoprotein (B)	100.63±23.01	106.67±23.52	<0.001	92.16±22.24	91.24±21.31	0.116
HOMA IR	2.28±1.44	2.67±1.88	<0.001	1.76±1.15	1.56±1.07	<0.001
Hs-CRP (mg/L)	0.17±0.27	0.17±0.36	0.604	0.14±0.24	0.10±0.20	<0.001
WBC count (10^3/ul)	6.49±1.54	6.47±1.60	0.413	6.21±1.54	5.85±1.50	<0.001
Exercise (≥1 times per week)	1,205(33.58)	1,255(34.98)	0.165	520(32.18)	632(39.11)	<0.001

BMI, body mass index; BP, blood pressure; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, homeostatic model assessment-insulin resistance; Hs-CRP, high-sensitivity C-reactive protein.

Table 3

Multiple linear regression showing the change (between baseline and follow up) in lipid variables with persistent vs. resolution in fatty liver in men and women

	Men						Women					
	B Coefficients	SE	t	*p*	95% CIs		B Coefficients	SE	t	*p*	95% CIs	
D-LDL cholesterol	2.31	0.56	4.11	<0.001	1.21	3.41	4.61	1.17	3.92	<0.001	2.31	6.91
D-HDL cholesterol	-1.77	0.17	-10.47	<0.001	-2.11	-1.44	-2.17	0.47	-4.60	<0.001	-3.10	-1.25
D-Triglyceride	9.19	2.01	4.57	<0.001	5.25	13.14	10.03	3.30	3.04	0.002	3.57	16.50
D- Apolipoprotein (A)	-2.65	0.44	-6.06	<0.001	-3.50	-1.79	-4.46	1.10	-4.04	<0.001	-6.62	-2.29
D- Apolipoprotein (B)	2.21	0.43	5.16	<0.001	1.37	3.05	4.75	0.96	4.94	<0.001	2.87	6.64

Persistent fatty liver vs resolution of fatty liver between baseline and follow up, is a dichotomous exposure variable in the models. D = change in variable i.e. follow up minus baseline measurement. Adjustments; Age, alcohol, smoking, education, exercise, BMI, each lipid level at baseline, change in exercise, change in alcohol amount, change in BMI and change in HOMA-IR

LDL, low-density lipoprotein; HDL, high-density lipoprotein; SE, standard error; CIs, confidence intervals.

Table 4

Multiple linear regression showing the change (between baseline and follow up) in lipid variables by unit change in BMI (kg/m^2) in men and women

	Men						Women					
	B Coefficients	SE	t	*p*	95% CIs		B Coefficients	SE	t	*p*	95% CIs	
D-LDL cholesterol	3.76	0.18	20.96	<0.001	3.41	4.12	2.35	0.35	6.72	<0.001	1.67	3.04
D-HDL cholesterol	-1.59	0.05	-29.30	<0.001	-1.70	-1.48	-1.50	0.14	-10.71	<0.001	-1.78	-1.23
D-Triglyceride	14.31	0.64	22.24	<0.001	13.05	15.57	7.40	0.98	7.56	<0.001	5.48	9.32
D- Apolipoprotein (A)	-1.33	0.14	-9.68	<0.001	-1.60	-1.06	-1.26	0.32	-3.87	<0.001	-1.89	-0.62
D- Apolipoprotein (B)	3.51	0.13	26.09	<0.001	3.24	3.77	2.40	0.28	8.53	<0.001	1.85	2.95

Adjustments; Age, alcohol, smoking, education, exercise, baseline BMI, change in exercise, change in alcohol amount and resolution of fatty liver and each lipid level at baseline and change in HOMA-IR.

LDL, low-density lipoprotein; HDL, high-density lipoprotein; SE, standard error; CIs, confidence intervals.