# ORIGINAL ARTICLES—LIVER, PANCREAS, AND BILIARY TRACT

## Independent Association Between Nonalcoholic Fatty Liver Disease and Cardiovascular Disease in the US Population

MARIA STEPANOVA*,‡ and ZOBAIR M. YOUNOSSI*,‡

*Center for Liver Diseases, Department of Medicine, Inova Fairfax Hospital, Falls Church; and ‡Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church, Virginia

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**BACKGROUND & AIMS:** Nonalcoholic fatty liver disease (NAFLD) and cardiovascular diseases (CVDs) have common metabolic risk factors. Despite reports from clinical studies, the association between NAFLD, cardiovascular disease, and cardiovascular mortality are not clear at the population level. **METHODS:** We analyzed data from the National Health and Nutrition Examination Survey III, conducted from 1988 to 1994, and compared hepatic ultrasound and mortality data. Participants were classified into those with NAFLD (moderate or severe hepatic steatosis, based on ultrasound analysis, without any evidence of other liver disease; n = 2492) and those without (absence of NAFLD or any other chronic liver diseases: controls). The prevalence of CVD was compared between subjects with and without NAFLD. Additional comparisons were made between NAFLD patients who had increased levels of liver enzymes and those who had normal levels. Independent predictors of CVD and cardiovascular mortality also were studied. **RESULTS:** During the follow-up period (median, 171 mo), 12.21% of the National Health and Nutrition Examination Survey III participants died; cardiovascular mortality was 3.76%. Regardless of whether levels of liver enzymes were increased or not, individuals with NAFLD were older, predominantly male, more likely to be Hispanic, and less likely to be African American than controls. They also had a higher prevalence of all components of metabolic syndrome and CVD. Regardless of levels of liver enzymes, NAFLD was associated independently with CVD, after adjusting for major demographic, clinical, and metabolic confounders (odds ratio, 1.23; 95% confidence interval, 1.04–1.44). The independent association of NAFLD with cardiovascular mortality was not statistically significant. **CONCLUSIONS: NAFLD is associated independently with an increased risk of CVD. However, NAFLD did not increase cardiovascular mortality over a 14-year period.**

*Keywords:* Population Study; Heart Disease; Obesity; Liver Disease.

The relationship between nonalcoholic fatty liver disease (NAFLD) and cardiovascular diseases (CVDs) has long been suspected.[1–3] In fact, NAFLD and CVD often are considered as 2 different manifestations of metabolic syndrome and share the same risk factors such as visceral obesity, type II diabetes, dyslipidemia, and insulin resistance.[4] Indeed, a number of studies have shown a higher prevalence of atherosclerosis in individuals with NAFLD.[5] Possible molecular mechanisms include the increased production of inflammatory cytokines, often observed in the setting of NAFLD, possibly accompanied by abnormal levels of lipoproteins, endothelial dysfunction, and oxidative stress.[6–8] Furthermore, some studies confirmed that NAFLD is associated with an increased risk of CVD, even after adjustment for major cardiovascular and metabolic risk factors.[9]

Both tertiary-care[10] and population-based studies[11] have shown that CVD is either the most common, or is among the 2 most common causes of death for individuals with NAFLD. However, the suggested independent association of NAFLD with cardiovascular mortality (CVM)[12] has not been proven in other studies using population-based data.[13–15] It is important to note, however, that population-based findings reporting an association of NAFLD and CVD/CVM published to date used the clinical definition of NAFLD using liver enzyme levels and laboratory tests only. Although valuable, this approach can result in a smaller and probably biased sample of patients presumed to have NAFLD. Thus, the suspected association of NAFLD with CVM at the population level requires validation using a more established approach for the diagnosis of NAFLD.

The aim of our study was to examine the association of radiologically proven NAFLD with cardiovascular events, including cardiovascular disease and cardiovascular mortality, using recent population-based data.

## Methods

### *Study Population*

For this study, we used the National Health and Nutrition Examination Survey III (NHANES III) conducted between 1988 and 1994. The survey data were collected by the US National Center for Health Statistics (NCHS) of the Centers for Disease Control and Prevention via household interviews, phys-

*Abbreviations used in this paper:* CI, confidence interval; CVD, cardiovascular disease; CVM, cardiovascular mortality; HCV, hepatitis C virus; MI, myocardial infarction; NAFLD, nonalcoholic fatty liver disease; NHANES, National Health and Nutrition Examination Survey; NCHS, US National Center for Health Statistics; OR, odds ratio.



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ical examination, and laboratory tests, and contains information describing the health and nutritional status of the US population as of the time of the data collection. In addition, the data collections with hepatic ultrasound data for NHANES III participants were released recently. Therefore, the NHANES III participants included in this study were required to have the hepatic ultrasound data together with the relevant demographic, clinical, and examination data available. Participants with data insufficient for ruling in/out NAFLD and CVD were excluded from the study.

### *NAFLD Diagnosis*

For eligible NHANES III participants, which included adults between 20 and 74 years of age at the time of examination, the archived hepatobiliary ultrasound video images recently were re-reviewed by NCHS and the results of their assessment were published.[16] From the data collection, to establish the diagnosis of NAFLD, we used the parameter representing the presence of fat within the hepatic parenchyma, which was graded as normal-mild or moderate-severe. Quality control and quality assurance procedures were used by NCHS to standardize the readings from 3 ultrasound readers who had no access to any other clinical or laboratory data of the participants.[16]

In our study, subjects were considered to have NAFLD if moderate to severe hepatic steatosis was noted on the ultrasound in the absence of any other possible causes of chronic liver disease such as excessive alcohol use (defined as self-reported consumption of ≥20 g/d in men and ≥10 g/d in women during the year preceding the examination for NHANES III), iron overload (defined as transferrin saturation of ≥50%), or a positive hepatitis B surface antigen or anti-hepatitis C virus (HCV) test (anti-HCV by enzyme-linked immunosorbent assay and HCV RNA by polymerase chain reaction).

Furthermore, because abnormal level of aminotransferases could be indicative of a progressed form of NAFLD or nonalcoholic steatohepatitis, the individuals with a diagnosis of NAFLD were classified further into those with increased liver enzyme levels (defined as alanine aminotransferase level ≥40 U/L or aspartate aminotransferase level ≥37 U/L in men, alanine aminotransferase or aspartate aminotransferase ≥31 U/L in women) and those with normal liver enzyme levels.

Individuals without hepatic steatosis on the ultrasound and any other cause of chronic liver disease (as described earlier) were presumed to have no chronic liver disease and were used as the control cohort.

### *Other Definitions*

Four major race or ethnic groups included non-Hispanic whites, non-Hispanic blacks, Hispanics, and "other," which included Aleut, Eskimo, American Indian, Asian, or Pacific Islander. We also defined obesity as a body mass index greater than 30 and insulin resistance as a homeostasis of model assessment score of 3.0 or higher. In addition, diabetes mellitus and history of smoking were consistent, as we have reported previously.[11]

For the purpose of this study, we defined CVD as self-reported history of congestive heart failure, stroke, angina, or myocardial infarction (MI). Furthermore, family history of CVD was defined as a history of MI before the age of 50 in a parent or a sibling.

### *Mortality Follow-up Data*

For all eligible NHANES III participants, the mortality follow-up data were collected from a publicly available NHANES III–Linked Mortality File provided by NCHS. In addition to mortality status, the linked mortality file contains the length of the follow-up period between the time of examination for NHANES III and the reported death or the end of the follow-up period (whichever is earlier), as well as causes of death. In the most recent data collection, the NHANES III participants aged 17 years and older had mortality follow-up evaluation until December 31, 2006. Individuals without available mortality follow-up data were ineligible for the study.

The NHANES III Linked Mortality file uses the Underlying Cause of Death 113 code (UCOD113) to recode all deaths according to International Classification of Diseases 9th and 10th revision criteria. For the purpose of this study, we evaluated cardiovascular mortality (UCOD113 58–63, 67, 70–74), which covered causes of death such as ischemic heart diseases, heart failure, atherosclerosis, cerebrovascular diseases, aortic aneurysm, and other diseases of arteries, arterioles, and capillaries.

### *Statistical Analyses*

As recommended by the NHANES III Analytic Guidelines,[17] the sampling weights were used to account for nonresponse and unequal selection probabilities for certain categories of the population. Thus, after weighting on the basis of age, sex, level of education, and race or ethnic group, the distribution of participants was representative of that of the US population. In addition, stratum and sampling units accounted for the survey design effects using Taylor series linearization.

As a first step, individuals with NAFLD as well as its subgroups with increased and normal liver enzyme levels were compared separately with non-NAFLD controls. For the purpose of the study, continuous variables were compared between the cohorts using a *t* test for a contrasted mean, and the prevalence of various parameters, including demographic parameters, metabolic syndrome components, and CVD, was compared by the stratum-specific chi-square test for independence. *P* values of .05 or less were considered potentially significant.

At the next step, the presence of NAFLD or its subgroups was tested for independent association with CVD in the US population and certain subpopulations. Logistic regression was used to identify independent predictors of CVD, and all studied demographic, clinical, and laboratory parameters were used as potential confounders.

Finally, to identify risk factors independently associated with cardiovascular mortality, a Cox proportional hazard model was used. Proportional hazard assumption was controlled for all potential mortality predictors.

All analyses were run with SAS 9.1 and SUDAAN 10.0 (SAS Institute, Inc, Cary, NC). The protocol of the study was approved by the Inova Institutional Review Board.

## Results

Of the initial study population (20,050 adult participants from NHANES III), 2492 individuals (18.77% ± 0.76%)