CLINICAL—LIVER

Association Between High-Normal Levels of Alanine Aminotransferase and Risk Factors for Atherogenesis

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BACKGROUND & AIMS: Liver disease has been associated with cardiovascular disorders, but little is known about the relationship between serum levels of alanine aminotransferase (ALT) and markers of atherogenesis. We investigated the relationship between low–normal and high–normal levels of ALT and an extended panel of cardiovascular risk factors among individuals with no known diseases in a primary care setting. **METHODS:** We performed a retrospective analysis of data collected from 6442 asymptomatic patients at wellness visits to a primary care setting in central Virginia from 2010 through 2011. Serum levels of ALT were compared with levels of lipids and lipoproteins, as well as metabolic, inflammatory, and coagulation-related factors associated with risk for cardiovascular disease. **RESULTS:** Serum levels of ALT were higher than 40 IU/L in 12% of subjects, and in the high–normal range (19–40 IU/L in women and 31–40 IU/L in men) in 25% of subjects. ALT level was associated with the apolipoprotein B level, concentration and particle size of very-low-density lipoproteins, concentration of low-density lipoprotein (LDL) particles (LDL-P), and percentages of small dense LDL (sdLDL) and sdLDL-cholesterol (sdLDL-C) ($P < .0001$ for all). A high–normal level of ALT was associated with higher levels of LDL-C, LDL-P, sdLDL-C, and sdLDL particles ($P < .001$ for all). These effects were independent of age, body mass index, and hyperinsulinemia. Increasing levels of ALT and fasting hyperinsulinemia (>12 μU/mL) synergized with increasing levels of triglycerides, very-low-density lipoprotein particles, LDL-P, sdLDL-C, and percentage of sdLDL-C. Levels of APOA1, high-density lipoprotein-cholesterol, and high-density lipoprotein-class 2 were associated inversely with serum level of ALT ($P < .0001$ for all). **CONCLUSIONS: In an analysis of asymptomatic individuals, increased serum levels of ALT (even high–normal levels) are associated with markers of cardiovascular disease.**

Keywords: Atherosclerosis; NAFLD; NASH; Heart Disease.

Chronic liver disease is widely prevalent in the general population.[1] Nonalcoholic fatty liver disease (NAFLD), viral hepatitis, and alcohol-induced liver disease are the most common causes of chronic liver disease. NAFLD affects about a third of the general population.[2,3] Chronic liver diseases often remain clinically silent until they become advanced and thus remain undetected for long periods of time. An increased alanine aminotransferase (ALT) level is a common laboratory marker for underlying chronic liver disease. The ALT level is associated with sex and body mass index (BMI), a risk factor for NAFLD, in the general population.[4] Also, a high-normal ALT level has been shown to be associated with increased liver-related mortality.[5] These have led to a proposal to redefine the upper limit of normal ALT level as 19 IU/L for women and 31 IU/l for men.[5]

The liver plays a central but often underappreciated role in lipoprotein biology. Lipoproteins are critically involved in atherogenesis and cardiovascular disease, the leading cause of death in the general population.[6] Triglycerides and cholesterol are packaged with apolipoprotein B (apoB) in very-low-density lipoproteins (VLDL) and exported from the liver. Triglycerides in VLDL are hydrolyzed by lipoprotein lipase in peripheral tissues, leaving behind cholesterol-enriched intermediate-density lipoproteins. These undergo additional modification, including action by hepatic lipases, to form low-density lipoproteins (LDLs) of varying size, containing mainly cholesterol. Small dense LDL (sdLDL) particles are highly atherogenic.[7,8] High-density lipoproteins (HDLs) include apolipoprotein A (apoA) and mediate reverse cholesterol

Abbreviations used in this paper: ALT, alanine aminotransferase; apoA, apolipoprotein A; apoB, apolipoprotein B; BMI, body mass index; HDL, high-density lipoprotein; HDL-C, high-density lipoprotein cholesterol; HDL^{INC}, Health Diagnostic Laboratories; HDL2-C, high-density lipoprotein subclass II cholesterol; LDL, low-density lipoprotein; LDL-C, low-density lipoprotein cholesterol; LDL-P, low-density lipoprotein particles; NAFLD, nonalcoholic fatty liver disease; sdLDL, small dense low-density lipoprotein; TSH, thyroid-stimulating hormone; VLDL, very-low-density lipoprotein; VLDL-P, very-low-density lipoprotein particle.

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transport by moving cholesterol from blood vessels back to the liver for clearance.

Historically, cardiovascular risk has been assessed by total, LDL, and HDL cholesterol and triglyceride levels. It is now appreciated that these do not fully capture cardiovascular risk and that lipoprotein particle size and characteristics along with inflammatory and metabolic markers more fully define the cardiovascular risk profile.[7,9–14] Specifically, sdLDL cholesterol has been shown to be particularly atherogenic whereas medium to large HDL particles are protective.

Increased ALT level recently has been associated with cardiovascular disease, apoB and apoB:apoA1 ratio, and LDL and VLDL particle size.[15] However, the relationship between ALT sdLDL-cholesterol (sdLDL-C), percentage of sdLDL-C (% sdLDL-C), HDL subclass II cholesterol (HDL2-C), and lipoprotein (a) mass were not studied. The levels and impact of other known cardiovascular risk factors that often are present concomitantly such as vitamin D, thyroxine-stimulating hormone (TSH), homocysteine, and phospholipase A_2, also not were evaluated. Furthermore, the potential interactions between ALT and these factors in driving the risk profile are unknown. Importantly, the relationship of high-normal ALT level to cardiovascular risk factor profile remains unclear.

We hypothesized that, in a general population of apparently healthy individuals, both a high-normal and a high ALT level would have a poorer cardiovascular risk factor laboratory profile than those with a low-normal ALT level independent of BMI and insulin resistance. We therefore performed a study with the following aims: (1) to define the relationship of ALT with lipoprotein size and characteristics, inflammatory risk factors, and metabolic risk factors in an asymptomatic ambulatory population in a community-based primary care setting, (2) to determine if this relationship was independent of associated BMI and insulin resistance, and (3) to determine whether the impact of ALT was additive or synergistic with hyperinsulinemia, a marker of insulin resistance.

Materials and Methods

A retrospective analysis of a cohort of asymptomatic subjects without any known acute or chronic medical illness undergoing an annual wellness visit in a community-based primary care setting throughout central Virginia was performed. The advanced cardiovascular profile ordered by primary care physicians as part of this assessment was analyzed by Health Diagnostics Laboratories (HDL^{INC}) (Richmond, VA), a commercial diagnostic laboratory. All tests were performed using well-established methods. HDL^{INC} maintains de-identified data on all the samples collected including laboratory results, International Classification of Diseases, 9th revision codes, and basic demographic information. Patient level data were extracted from the de-identified database. The data were provided to the investigators by HDL^{INC}, under a materials transfer agreement with the investigators' institution and analyzed entirely by the lead and senior investigators who are fully responsible for the data and conclusions. The article was written in its entirety by the investigators. Given the anonymous nature of the data set, this study met criteria for exemption from a full institutional review board review.

The Study Population

The inclusion criteria included asymptomatic subjects undergoing a wellness visit with their primary health care provider in a community-based, ambulatory clinical setting. Only data from adults were obtained for this analysis because the number of children receiving such tests was too low for meaningful analysis. Data from individuals with known heart disease, type 2 diabetes, hypertension, dyslipidemia, and chronic liver disease were excluded. These exclusions were selected to avoid their potential confounding effects, especially the use of lipid-lowering agents (eg, statins), on the end points of interest and to allow easier assessment of the linkage between ALT and cardiovascular risk parameters. Also, those with an ALT level greater than 100 IU/L were excluded because some of these subjects could have had acute liver injury, which was not the focus of this study.

Serum ALT levels were classified into the following 3 groups: (1) low-normal ALT level: less than 19 IU/L in women and less than 31 IU/L in men; (2) high-normal ALT level: between 19 and 40 IU/L in women and between 31 and 40 IU/L in men; and (3) increased ALT level: greater than 40 IU/L in either men or women.

Laboratory-Based Cardiovascular Risk Factors Measured

The specific tests performed and the literature supporting their linkage to cardiovascular risks are listed later.

Lipids, lipoprotein characteristics, and subparticles. A description of lipoprotein measurements is described further in the Supplementary Materials and Methods section.

LDLs. LDL-C, LDL particle concentration (LDL-P) and size, sdLDL-C, sdLDL particle concentration, % sdLDL-C, and apoB were measured.[7,8,10,14]

HDLs. HDL-cholesterol (HDL-C), HDL particle and concentration, HDL2-C, apoA1) were measured.[12,16]

VLDLs. Serum triglycerides, VLDL particle size, and VLDL particle (VLDL-P) concentration were measured.[17]

Miscellaneous. Total cholesterol, lipoprotein (a) mass and cholesterol concentration, apoB:apoA1 ratio (calculated value), and desmosterol: total cholesterol ratio were measured.[17,18]

Insulin resistance-related markers. Insulin resistance was quantitated by measuring serum concentrations of fasting insulin, glucose, free fatty acids, and hemoglobin A1c. A fasting insulin level greater than 12 mIU/mL was taken as the principal parameter for analysis of the linkage between insulin resistance and ALT.[19] Insulin levels contribute principally to variability in models of insulin resistance such as the homeostatic model and are a validated marker of insulin resistance in nondiabetic individuals.[20]

Inflammatory markers. These included serum fibrinogen, high-sensitivity C-reactive protein, myeloperoxidase, and lipoprotein-associated phospholipase A_2.[11,21,22]

Metabolic. These included serum folate, red cell folate, homocysteine, vitamin B12, vitamin D, and TSH.[22,23]