# Predicting Hepatic Steatosis in a Racially and Ethnically Diverse Cohort of Adolescent Girls

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**Objective** To develop a risk assessment model for early detection of hepatic steatosis using common anthropometric and metabolic markers.

**Study design** This was a cross-sectional study of 134 adolescent and young adult females, age 11-22 years (mean 13.3 ± 2 years) from a middle school and clinics in Madison, Wisconsin. The ethnic distribution was 27% Hispanic and 73% non-Hispanic; the racial distribution was 64% Caucasian, 31% African-American, and 5% Asian, Fasting glucose, fasting insulin, alanine aminotransferase (ALT), body mass index (BMI), waist circumference (WC), and other metabolic markers were assessed. Hepatic fat was quantified using magnetic resonance imaging proton density fat fraction (MR-PDFF). Hepatic steatosis was defined as MR-PDFF >5.5%. Outcome measures were sensitivity, specificity, and positive predictive value (PPV) of BMI, WC, ALT, fasting insulin, and ethnicity as predictors of hepatic steatosis, individually and combined, in a risk assessment model. Classification and regression tree methodology was used to construct a decision tree for predicting hepatic steatosis.

**Results** MR-PDFF revealed hepatic steatosis in 16% of subjects (27% overweight, 3% nonoverweight). Hispanic ethnicity conferred an OR of 4.26 (95% CI, 1.65-11.04; $P$ = .003) for hepatic steatosis. BMI and ALT did not independently predict hepatic steatosis. A BMI >85% combined with ALT >65 U/L had 9% sensitivity, 100% specificity, and 100% PPV. Lowering the ALT value to 24 U/L increased the sensitivity to 68%, but reduced the PPV to 47%. A risk assessment model incorporating fasting insulin, total cholesterol, WC, and ethnicity increased sensitivity to 64%, specificity to 99% and PPV to 93%.

**Conclusion** A risk assessment model can increase specificity, sensitivity, and PPV for identifying the risk of hepatic steatosis and guide the efficient use of biopsy or imaging for early detection and intervention. *(J Pediatr 2014;165:319-25)*.

Nonalcoholic fatty liver disease (NAFLD) comprises a continuum extending from isolated hepatic steatosis to nonalcoholic steatohepatitis (NASH) to bridging fibrosis to cirrhosis.[1-3] The prevalence of NAFLD approaches 25% in overweight adolescent girls and ranges from 25% to 38% of all overweight children.[1,4,5] Studies have shown a higher prevalence of NAFLD in Hispanics and a lower prevalence in non-Hispanic blacks compared with non-Hispanic whites.[6-9] Even in non-overweight children, Hispanic ethnicity influences the risk for NAFLD.[10]

In both children and adults, NAFLD is strongly associated with metabolic syndrome and insulin resistance, which can lead to the development of NASH.[11-14] Hyperinsulinemia not only is more common in children with NAFLD, but also contributes to disease progression by facilitating intracellular accumulation of triglycerides and fatty acids in hepatocytes.[15,16] Accumulation of fatty acids in hepatocytes causes oxidative stress, activation of stellate cells, and hepatocellular injury and fibrosis.[17] Importantly, up to 68% of children and adolescents with NAFLD already have NASH at diagnosis.[5,18]

Early diagnosis is important, given that prognosis is significantly better when NAFLD is diagnosed before progression to NASH.[1,6] Although isolated hepatic steatosis is reversible with weight loss, the scarring and inflammation associated with NASH can lead to irreversible changes, including cirrhosis and end-stage liver disease.[19,20] Unfortunately, it is difficult to identify children with isolated steatosis and predicting which of these children will progress to NASH. Although

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| ALT | Alanine aminotransferase | MR-PDFF | Magnetic resonance imaging proton density fat fraction |
| AST | Aspartate aminotransferase | | |
| BMI | Body mass index | NAFLD | Nonalcoholic fatty liver disease |
| CART | Classification and regression tree | NASH | Nonalcoholic steatohepatitis |
| HDL | High-density lipoprotein | NPV | Negative predictive value |
| HgbA1c | Hemoglobin A1c | PPV | Positive predictive value |
| HOMA-IR | Homeostatic model assessment of insulin resistance | ROC | Receiver operating characteristic |
| | | WC | Waist circumference |
| MRI | Magnetic resonance imaging | | |

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elevated liver transaminase values (alanine aminotransferase [ALT] and aspartate aminotransferase [AST]) signify hepatocellular injury, liver enzymes are often normal in obese children despite evidence of hepatic steatosis on biopsy.[21,22] Multiple studies of pediatric NAFLD have shown that ALT correlates poorly or not at all with early steatosis,[1,4,15,21,23] and that the degree of elevation does not the predict severity or presence of NASH.[24] In addition, the National Health and Nutrition Examination Survey 1999-2004 survey found that ALT values vary by sex, race, and ethnicity, further limiting the utility of ALT as a screening tool for NAFLD in adolescents.[23] The lack of correlation between ALT and NAFLD has led to significant confusion among health care providers about screening for NAFLD in overweight and obese children.[25] Both the American Academy of Pediatrics[26] and the Endocrine Society[27] recommend using ALT to screen for NAFLD in this group; however, there is insufficient evidence on which to recommend the use of ALT for screening in overweight children or adults.[28]

Given the relative insensitivity of ALT as a marker of NAFLD and lack of consensus on appropriate screening of overweight and obese children, pediatric NAFLD likely is underdiagnosed, particularly in the early stages.[22] Comprehensive NAFLD prediction scores have been proposed to improve early detection, but 2 existing pediatric NAFLD scores are based only on obese white children and do not address the effect of race and ethnicity on NAFLD risk.[29,30]

The objectives of the present study were to identify early hepatic steatosis using quantitative magnetic resonance imaging–derived proton density fat fraction (MR-PDFF), to correlate hepatic fat with metabolic disease in an ethnically and racially diverse group of adolescent girls, and to develop a prediction model to identify individuals at high risk for hepatic steatosis and guide the efficient use of a model for risk assessment to increase early identification.

## Methods

The study subjects were females who responded to a general invitation distributed to University of Wisconsin pediatric clinics and a local middle school to participate in the study. After obtaining informed written consent and assent, magnetic resonance imaging (MRI) safety screen and a brief survey of personal and family medical history, medication use, and self-identified race and ethnicity (based on National Institute of Health race and ethnicity criterion for subjects in clinical research) were obtained. Study entrance criteria were female sex and age 11-22 years. Exclusion criteria included a history of chronic disease affecting hepatic or renal function, including type 1 or type 2 diabetes mellitus, known liver disease, or other chronic illness; treatment with medications, including oral contraceptives, lipid-lowering or glucose metabolism-altering agents, or vitamin E supplement at a daily dose >100 IU; pregnancy; excess alcohol consumption, defined as an average of >1.5 drinks per day; and standard contraindications to MRI (eg, metallic implants, claustrophobia). A total of 136 subjects were enrolled in the study.

Height was measured using a stadiometer and recorded to the nearest 0.5 cm. Waist circumference (WC) was measured twice just above the iliac crest with Graham-Field cloth measuring tape, and the average was recorded to the nearest 1 mm. Weight was measured with the subject in light clothing without shoes on a beam balance platform scale to the nearest 0.1 kg. Body mass index (BMI) was then calculated. Tanner staging for breasts and pubic hair was self-reported.[31]

Fasting blood samples were analyzed for lipids (total cholesterol, high-density lipoprotein [HDL], low density lipoprotein-calculated, and triglycerides), AST, ALT, hemoglobin A1c (HgbA1c), glucose, and insulin. HgbA1c was measured by ion-exchange chromatography/spectrometry. AST and ALT were determined by nicotinamide adenine dinucleotide phosphate with pyridoxal-5 phosphate assay. Sex hormone–binding globulin was measured with a quantitative electrochemiluminescent immunoassay. Free testosterone was measured with a quantitative high-performance liquid chromatography–tandem mass spectrometry/electrochemiluminescent immunoassay. Adiponectin was analyzed by radioimmunoassay; leptin, by enzyme-linked immunosorbent assay. The homeostatic model of assessment–insulin resistance (HOMA-IR) was calculated as (fasting glucose [mg/dL] × fasting insulin [$\mu$U/mL]/405).

The presence of metabolic syndrome was identified using 2 different sets of criteria. The first of these, metabolic syndrome with impaired fasting glucose, requires the presence of at least 3 of the 5 criteria: fasting blood glucose $\geq$100 mg/dL, blood pressure >90th percentile for age/sex,[32] WC >90th percentile for age/sex,[33] HDL <40 mg/dL, and triglycerides >150 mg/dL.[34] The second set, metabolic syndrome with insulin resistance, substitutes HOMA-IR >4.0 for impaired fasting glucose.[35]

Quantitative MRI was performed at the Wisconsin Institute for Medical Research. The Human Subjects Committee of the University of Wisconsin approved all procedures. Single breath-holding MRI was performed over the entire liver using a clinical 3T scanner (MR750; GE Healthcare, Waukesha, Wisconsin) with a 32-channel phased-array body coil (Neocoil, Pewaukee, Wisconsin). MR-PDFF was determined using an investigational version of a chemical shift-encoded water–fat separation method (three-dimensional iterative decomposition of water and fat with echocardiographic asymmetry and least-squares estimation-spoiled gradient echocardiography).[36,37] Separated water-only and fat-only images, as well as hepatic MR-PDFF maps,[38] were created using an online reconstruction algorithm method that includes spectral modeling of fat[39] and corrects for eddy currents,[40] T1 bias,[41] T2* decay,[42] and noise-related bias.[41] Because all known confounders have been addressed, the resulting MR-PDFF map provides an accurate and fundamental measure of the fat concentration in tissue.[37]

Hepatic PDFF was determined by averaging MR-PDFF values measured from 9 regions of interest placed in each