ORIGINAL ARTICLES

Hyaluronic acid predicts hepatic fibrosis in children with nonalcoholic fatty liver disease

VALERIO NOBILI, ANNA ALISI, GIULIANO TORRE, RITA DE VITO, ANDREA PIETROBATTISTA, GIUSEPPE MORINO, JEAN DE VILLE DE GOYET, GIORGIO BEDOGNI, and MASSIMO PINZANI

ROME, FLORENCE, TRIESTE, AND MILAN, ITALY

Nonalcoholic fatty liver disease (NAFLD) is the most common chronic liver disease in children and adolescents, and it may progress to liver fibrosis and cirrhosis. Liver biopsy, which is the recognized gold standard for the diagnosis of hepatic fibrosis, is invasive. Thus, there has been increasing interest in the development of noninvasive markers. Hyaluronic acid (HA) has been shown to be a good marker of liver fibrosis in adults. In the current study, we evaluated the association of HA with liver fibrosis in 100 consecutive children with biopsy-proven NAFLD. In all, 65% of the children had liver fibrosis. Using proportional-odds ordinal logistic regression, we found that values of HA ≥ 1200 ng/mL made the absence of fibrosis (F0) unlikely (7%, 95% confidence interval (CI): 1% to 14%), whereas values of HA ≥ 2100 ng/mL made F2, F3, or F4 fibrosis likely (89%, 95% CI: 75% to 100%). Our study shows that HA is a predictor of fibrosis in children with NAFLD followed at a tertiary care center. Additional studies are needed to test whether HA can be employed to predict liver fibrosis in pediatric populations with similar and lower prevalence of liver fibrosis. (Translational Research 2010;156:229–234)

Abbreviations: ALT = aspartate aminotransferase; AST = aspartate aminotransferase; BMI = body mass index; CI = confidence interval; ECM = extracellular matrix; GGT = gamma glutamyl transferase; HA = hyaluronic acid; HOMA-R = homeostasis model assessment of insulin resistance; NAFLD = nonalcoholic fatty liver disease; NASH = nonalcoholic steatohepatitis; NPV = negative predictive value; OR = odds ratio; PPV = positive predictive value; ROC = receiver operating characteristic curve; SDS = standard deviation score

Parallel to the current pandemic of overweight and obesity, nonalcoholic fatty liver disease (NAFLD) has become the most common chronic liver disease in children and adolescents in Western countries.[1,2] The prevalence of pediatric NAFLD is 3% to 10% in normal-weight subjects and reaches a value of 80% in obese individuals.[3,4] These data are alarming because, even if the long-term course of pediatric NAFLD is not yet known, some evidence exists of a possible course toward cirrhosis and liver failure, resulting in an increased need for liver transplantation.[5,6]

Therefore, the early detection of NAFLD in children and adolescents is necessary to prevent the development

From the Liver Unit; Pathology Department; Dietetics Unit; Liver and Intestine Transplant Unit, ''Bambino Gesù'' Children's Hospital and Research Institute, Rome, Italy; Clinical Epidemiology Unit, Liver Research Center, Basovizza, Trieste, Italy; Department of Maternal and Pediatric Sciences, University of Milan, Fondazione IRCCS Cà Granda—Ospedale Maggiore Policlinico, Milan, Italy; Department of Internal Medicine, ''DENOThe'' Center for Research, High Education and Transfer, University of Florence, Florence, Italy.

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Reprint requests: Valerio Nobili, MD, Liver Unit, ''Bambino Gesù'' Children's Hospital and Research Institute, Square S. Onofrio 4, 00165 Rome, Italy; e-mail: nobili66@yahoo.it.

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AT A GLANCE COMMENTARY

Nobili V, et al.

Background

Recently, hyaluronic acid (HA) has been shown to be one of the best performing direct markers of liver fibrosis in adults.

Translational Significance

Here, we demonstrated the association of serum HA levels with liver fibrosis in children with biopsy-proven NAFLD, suggesting that it may allow a simple and efficient screening of patients at risk of progressive liver disease needing subsequent investigation. The translational impact of our research is that HA assessment might come into clinical practice for the management of children with suspected NASH, contributing to simplify and improve the management of untreated patients, as well as those included in a therapeutic plan.

of advanced liver disease, both in pediatric age as well as later in life.[7-9] The progression of NAFLD toward liver fibrosis and cirrhosis strongly depends on the presence of a necroinflammatory and fibrogenic milieu defined as nonalcoholic steatohepatitis (NASH).[10,11] The distinction between simple fatty liver and NASH, and the exclusion of competing causes of chronic liver disease, is based on the histopathologic evaluation of liver tissue. However, because liver biopsy is invasive, painful, and expensive, there has been increasing interest in the development of noninvasive markers for the diagnosis of NASH and liver fibrosis.[12]

Many noninvasive markers of liver fibrosis have been proposed so far.[13] Although all available markers have suboptimal diagnostic accuracy, they may reduce the need for liver biopsy when used alone or in combination.[14] These findings are more relevant for the pediatric setting, in which liver biopsy is perceived as more risky than in adults.[15] Among the proposed markers, some reflect alterations of hepatic function but not of extracellular matrix (ECM) metabolism and, therefore, are labeled ''indirect markers.'' Those markers directly linked to modifications in ECM turnover during fibrogenesis are instead defined ''direct markers.''[16-19]

Among direct markers, hyaluronic acid (HA) is one of the best predictors of liver fibrosis in adults.[20] HA is a glycosaminoglycan synthesized by ECM-producing cells, including activated hepatic stellate cells. The circulating levels of HA might reflect not only the stage of disease but also ECM metabolism and, to some extent, inflammatory activity within the liver.[20] Recent studies performed in patients with chronic viral hepatitis C either before[21,22] or after[23] liver transplantation have reinforced the notion that HA is a low-cost and accurate marker for the staging of liver fibrosis. A recent study showed that serum HA is a marker of fibrosis in unselected children undergoing liver biopsy,[24] but the possibility of employing HA for the prediction of liver fibrosis in children with NAFLD has not been tested so far.

In the current study, we evaluated the association of serum HA levels with the degree of liver fibrosis in children with NAFLD, aiming to determine the diagnostic performance of HA as a single, low-cost, and easily available marker of hepatic fibrosis suitable for everyday clinical practice.

METHODS

Patients. This cross-sectional study involved 100 consecutive children and adolescents (68 males and 32 females) with biopsy-proven NAFLD referred to the Liver Unit of the ''Bambino Gesù'' Children's Hospital and Research Institute between May 2006 and November 2009. Exclusion criteria were (1) excessive alcohol intake ($\geq$20 g/day), (2) hepatitis A, B, C, D, E, or G or cytomegalovirus or Epstein-Barr virus infection, (3) autoimmune liver disease, (4) metabolic liver disease, (5) celiac disease, (6) Wilson's disease, (7) alpha-1-antitrypsin deficiency, (8) total parenteral nutrition, and (9) use of steatogenic drugs. The study protocol was conducted in accordance with the Declaration of Helsinki and was approved by the Ethics Committee of the ''Bambino Gesù'' Children's Hospital. Informed consent was obtained from each patient and/or at least 1 legal guardian.

Anthropometric and laboratory measurements. Weight and height were measured and body mass index (BMI) was calculated and converted to standard deviation scores (SDSs) using the CDC 2000 reference data.[25] Alanine transaminase (ALT), aspartate transaminase (AST), and gamma-glutamyl-transferase (GGT) were measured using standard methods as described elsewhere.[26] Glucose was measured using standard laboratory methods and insulin by radioimmunoassay (Myria Technogenetics, Milan, Italy). The homeostasis model assessment index of insulin resistance (HOMA-R) was calculated as (fasting insulin [μU/mL] $\times$ fasting glucose [mmol/L]/22.5). Serum HA was collected at the time of liver biopsy and immediately stored at –80 °C. HA was then measured using an enzyme-linked binding protein assay (Hyaluronan; R&D Systems, Minneapolis, Minn) and is reported as ng/mL.