Prediction of Liver Steatosis Applying a New Score in Subjects from the Brazilian Longitudinal Study of Adult Health

Hugo Perazzo, MD, PhD, Isabela Benseñor, MD, PhD,†*
José Geraldo Mill, MD, PhD,‡ Antônio G. Pacheco, MD, PhD,§
Maria de Jesus Mendes da Fonseca, MD, PhD,‖
Rosane Härter Griep, MD, PhD,¶ Paulo Lotufo, MD, PhD,†
and Dora Chor, MD, PhD‖

Goals: To develop a noninvasive algorithm for diagnosis of liver steatosis and to compare its diagnostic value with available predictive models.

Background: Liver steatosis represents the most frequent liver disease worldwide.

Study: This cross-sectional study analyzed data from the Brazilian Longitudinal Study of Adult Health (ELSA-Brasil). Patients were randomly divided into training (n = 6571) and validation (n = 3286) cohort. Abdominal ultrasound (US), used to grade steatosis, and overnight fasting blood tests were performed at the same day. Fatty Liver Index (FLI), Hepatic Steatosis Index, and Nonalcoholic Fatty Liver Disease-Liver Fat Score were calculated. A backward stepwise multivariate logistic regression analysis was used to develop the new predictive model, Steato-ELSA.

Results: In total, 9857 subjects [58% female, age = 51 (interquartile range, 45 to 58) years, body mass index = 26.4 (23.9 to 29.6) Kg/m^2] were included. Body mass index, waist circumference, homeostasis model of assessment of insulin resistance, transaminases, and triglycerides were independently associated with steatosis in the multivariate model (Hosmer-Lemeshow $P = 0.279$). In the validation cohort, the area under the receiver-operator characteristics (95% confidence interval) for prediction of mild and moderate steatosis were: (i) 0.768 (0.751-0.784) and 0.829 (0.810-0.848) for Steato-ELSA; (ii) 0.762 (0.745-0.779) and 0.819 (0.799-0.838) for Fatty Liver Index; (iii) 0.743 (0.727-0.761) and 0.800 (0.779-0.822) for Hepatic Steatosis Index; and (iv) 0.719 (0.701-0.737) and 0.769 (0.747-0.791) for Nonalcoholic Fatty Liver Disease-Liver Fat Score. Steato-ELSA performed significantly better than other models and yielded sensitivity (Se)/specificity (Sp) (95% confidence interval): (i) for mild steatosis (score ≥ 0.386): Se = 65.6% (63.0-68.3) and Sp = 73.7% (71.8-75.6); (ii) for moderate steatosis (score ≥ 0.403): Se = 83.5% (80.0-86.9) and Sp = 68.7% (67.0-70.4).

Conclusions: Steato-ELSA is an accurate and inexpensive tool that uses simple parameters to identify individuals at high risk of liver steatosis.

Key Words: liver steatosis, predictive model, noninvasive methods, fatty liver

(*J Clin Gastroenterol* 2018;00:000–000)

Liver steatosis, characterized by abnormal fat accumulation in hepatic parenchyma, represents the most frequent liver disease worldwide[1] and it has been associated with presence of metabolic features.[2,3] Liver biopsy has been used as the gold standard for diagnosis of liver steatosis. However, this invasive method has been challenged by limited feasibility and potential adverse events.[4] Noninvasive methods, as imaging techniques and serological markers, has been developed and validated in the last decade.[5] The advantage of serological tests relies on the combination of demographic and laboratory parameters to identify the presence of liver fat at low cost. Fatty Liver Index (FLI), Hepatic Steatosis Index (HSI), and Nonalcoholic Fatty Liver Disease-Liver Fat Score (NAFLD-LFS) are the most studied serological tests to detect liver steatosis.[6]

The Brazilian Longitudinal Study of Adult Health (ELSA-Brasil), a multicenter cohort study, is a unique opportunity to evaluate risk factors for liver steatosis and to propose an algorithm to detect liver fat based on anthropometric measures and blood tests.[7] Previously, we assessed the accuracy of FLI and HSI in a subsample (n = 195) of participants from the ELSA-Brasil.[8] The aims of the present study were to develop a new algorithm for diagnosis of liver steatosis and to compare its diagnostic value with available serological tests.

Received for publication October 10, 2017; accepted January 5, 2018.
From the *Laboratory of clinical research on STD/AIDS, Evandro Chagas National Institute of Infectious Disease (INI); §Scientific Computing Program (PROCC); ‖National School of Public Health; ¶Laboratory of Health and Environment Education, Oswaldo Cruz Institute, Oswaldo Cruz Foundation (FIOCRUZ), Rio de Janeiro; †Center for Clinical and Epidemiologic Research of the University of São Paulo, São Paulo; and ‡Department of Physiological Sciences, Federal University of Espírito Santo, Vitória, Brazil.
The ELSA-Brasil baseline study was supported by the Brazilian Ministry of Health (Science and Technology Department) and the Brazilian Ministry of Science and Technology (Financiadora de Estudos e Projetos and CNPq National Research Council) (grants: 01 06 0010.00-RS, 01 06 0212.00-BA, 01 06 0300.00-ES, 01 06 0278.00-MG, 01 06 0115.00-SP, 01 06 0071.00-RJ); the International Society for Infectious Diseases (ISID Research Grant 2016 to H.P.); FAPERJ (grant number E-26/201.471/2014 to A.G.P.); and Conselho Nacional de Desenvolvimento Científico e Tecnológico [grant number 305801/2015-5 to A.G.P., 405211/2016-3 to H.P.]. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

The authors declare that they have nothing to disclose.
Address correspondence to: Dora Chor, MD, PhD, National School of Public Health, Oswaldo Cruz Foundation (FIOCRUZ). R. Leopoldo Bulhões, 1480-CEP 21041-210-Manguinhos, Rio de Janeiro-RJ, Brazil (e-mail: dorachor@gmail.com).
Supplemental Digital Content is available for this article. Direct URL citations appear in the printed text and are provided in the HTML and PDF versions of this article on the journal's website, www.jcge.com.

DOI: 10.1097/MCG.0000000000001007

MATERIALS AND METHODS

ELSA-Brasil Study

The ELSA-Brasil study enrolled active or retired public servants [n = 15,105; aged 35 to 74 years; 46% male; 30% with body mass index (BMI) > 30 Kg/m^2] from 6 Brazilian cities: Belo Horizonte, Porto Alegre, Rio de Janeiro, São Paulo, Salvador, and Vitória.[9] The primary aim of ELSA-Brasil study was to investigate the incidence and determinants of chronic noncommunicable diseases during a long-term follow-up. In the baseline visit (2008 to 2010) participants were submitted to clinical evaluation and anthropometric measures, blood tests and abdominal ultrasound (US).[10,11] The study was approved by the Brazilian National Ethical Committee (CONEP) and Local Ethics Committees. All participants signed an informed consent upon enrollment in the ELSA-Brasil study.

Study Design

This cross-sectional study analyzed baseline data from the ELSA-Brasil. We excluded the participants without abdominal US (images not recorded or inadequate US images according to a senior US radiologist from a centralized analysis site),[8] those with abusive alcohol intake (> 210 g/wk for men and > 140 g/wk for women) or missing data for metabolic features. Subjects included in the present analysis were randomly divided into training cohort (two-third of the subjects) and validation cohort (one-third of the subjects) to build and to validate the new predictive model for detection of steatosis, respectively.

Clinical Evaluation and Blood Tests

Clinical records included the measures of BMI, waist circumference (WC), blood pressure (the mean of 2 measurements) and alcohol consumption (quantified in g/wk and drinking patterns), and smoking (never, past, or current).[11] Blood tests were performed after an overnight fasting and analyzed in a central laboratory using an ADVIA 1200 and Centaur Siemens analyzer (Siemens, Illinois).[9,10] Serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) were measured by enzymatic and γ-glutamyltransferase by kinetic colorimetric assay. The upper limit of normal of aminotransferases values were 43 IU/L and 35 IU/L for ALT and AST, respectively, in men and 36 IU/L and 31 IU/L in women. Plasma levels of total and high-density lipoprotein-cholesterol and triglycerides were determined by enzymatic methods. Low-density lipoprotein-cholesterol were calculated using the Friedewald equation or measured by enzymatic colorimetric assay if triglycerides > 400 mg/dL. Glucose and glycated hemoglobin (HbA1c) were measured using hexokinase method and high-pressure liquid chromatography, respectively. Fasting insulin was determined by immunoenzymatic assay (ELISA). The homeostasis model assessment of insulin resistance (HOMA-IR) was calculated as HOMA-IR = [fasting glucose (mg/dL)×fasting insulin (μUI/mL)]/405[12] and insulin resistance was defined as HOMA-IR > 2.7 as previously validated.[13] Impaired glucose was defined as oral glucose tolerance test > 140 mg/dL and <200 mg/dL. Central obesity, dyslipidemia,[14] blood hypertension,[15] diabetes,[16] and metabolic syndrome were defined according to the International Diabetes Federation criteria.[17]

Abdominal US

Abdominal US were performed using a high-resolution B-mode scanner (SSA-790A, Aplio XG; Toshiba Medical System, Tokyo, Japan) and a convex array transducer (model PVT-375BT). Operators were previously trained in the University of São Paulo (ELSA-São Paulo site) before enrollment of the participants. Hepatic US images were recorded and analyzed by a centralized reading center at the ELSA-São Paulo site. The quality control of all abdominal US images was verified by a senior US radiologist, following a standardized protocol.

Liver steatosis was defined based on the evaluation of hepatic attenuation on B-mode images of abdominal US, loss of definition of the diaphragm and poor delineation of the intrahepatic architecture.[11] Absence of steatosis (grade 0) was defined as normal hepatic attenuation with complete diaphragm visualization. Presence of steatosis was classified as mild (grade 1: > 50% visualization of the diaphragm); moderate (grade 2: <50% visualization of the diaphragm); or severe (grade 3: poor or no visualization of the diaphragm) as previously validated.[8]

Liver Steatosis Predictive Models Previously Reported in the Literature

FLI includes BMI, WC, triglycerides and γ-glutamyltransferase; HSI uses transaminases, BMI and the presence of type-2 diabetes adjusted for gender and NAFLD-LFS includes fasting insulin, AST, ALT and presence of metabolic syndrome, and type-2 diabetes. These tests were calculated according to the following formulas:

$$\mathrm{FLI} = \frac{\left(e^{0.953\times \ln(\text{triglycerides, mg/dL}) + 0.139\times \mathrm{BMI} + 0.718\times \ln(\mathrm{GGT}) + 0.053\times \ln(\mathrm{WC}) - 15.745}\right)}{1+\left(e^{0.953\times \ln(\text{triglycerides, mg/dL}) + 0.139\times \mathrm{BMI} + 0.718\times \ln(\mathrm{GGT}) + 0.053\times \ln(\mathrm{WC}) - 15.745}\right)}\times 100^{18}.$$

$$\mathrm{HSI} = 8\times \mathrm{ALT/AST\ ratio} + \mathrm{BMI} + 2\ (\text{if type 2 diabetes}) + 2\ (\text{if female})^{19}.$$

$$\text{NAFLD-LFS} = -2.89 + 1.18\times(\text{metabolic syndrome} - \text{yes} = 1 / \text{no} = 0) + 0.45\times(\text{type 2 diabetes} - \text{yes} = 1/\text{no} = 0) + 0.15\times(\text{fasting insulin}, \mu\text{U/L}) + 0.04\times(\text{AST, U/L}) - 0.94\times(\text{AST/ALT})^{20}.$$

Presence of liver steatosis was defined as FLI ≥ 60[18], HSI ≥ 36[19] or NAFLD-LFS ≥ −0.640.[20]

Development of a New Predictive Model for Liver Steatosis Diagnosis

Parameters used for calculation of Steato-ELSA were identified in a backward stepwise multivariate logistic regression model, which included variables significantly associated with liver steatosis by abdominal US (grade ≥ 1) in univariate analysis from the training cohort. All predictors besides gender were evaluated as continuous variables in the final model. Regression coefficients (β) of variables independently associated with steatosis in the final model, as well

as its intercept, were entered in the logistic regression equation for calculation of this new test.

Statistical Analyses

Categorical variables were reported as absolute (n) and relative frequency (%) and continuous variables as median and interquartile range (IQR). χ^2 and Student t tests were used for comparisons of frequencies or means, respectively. A univariate logistic regression analysis was performed to estimate the odds ratios and confidence intervals (CIs) for the presence of steatosis. Backward stepwise multivariable logistic regression analysis was performed to build the Steato-ELSA. The severity of multicollinearity of variables entered into the final model was evaluated by the variance inflation factor (VIF)[21] and the final model goodness of fit was evaluated using the Hosmer-Lemeshow statistic.[22] Receiver-operator characteristic (ROC) curves analyses were performed and the optimal cutoff points were identified using the maximal Youden index. The diagnostic value of tests were assessed using sensitivity (Se), specificity (Sp), positive predictive value (PPV), negative predictive value (NPV), positive likelihood ratio (LR+), and area under the ROC (AUROC) that were compared using the empirical nonparametric method according to Delong et al.[23] Statistical analyses were performed using STATA (StataCorp LP, College Station, TX).

RESULTS

A total of 15,105 subjects were included in the ELSA-Brasil study between 2008 and 2010. For the present analysis we excluded subjects without abdominal US (n = 2486) or when this examination was inadequate for steatosis detection (n = 2116). In addition, 646 subjects were excluded due to abusive alcohol intake or missing data on metabolic features. Therefore, 9857 subjects [58% female; median (IQR) age = 51 (45 to 58) years, BMI = 26.4 (23.9 to 29.6) Kg/m^2, and ALT = 23 (18 to 32) IU/L; 38% with metabolic syndrome] were included (Fig. 1). Subjects included in the training (n = 6571) and in the validation (n = 3286) cohort had similar age and proportion of women/men, metabolic features, blood test results, and prevalence of steatosis according to abdominal US (Table 1). Prevalence of at least mild steatosis (grade $\geq$1) was 38% (n = 2476/6571) and 37% (n = 1227/3286) in training and validation cohorts, respectively. In addition, moderate steatosis (grade $\geq$2) was present in 14% (n = 940/6571) and 13% (n = 442/3286) of participants in the training and validation cohorts, respectively.

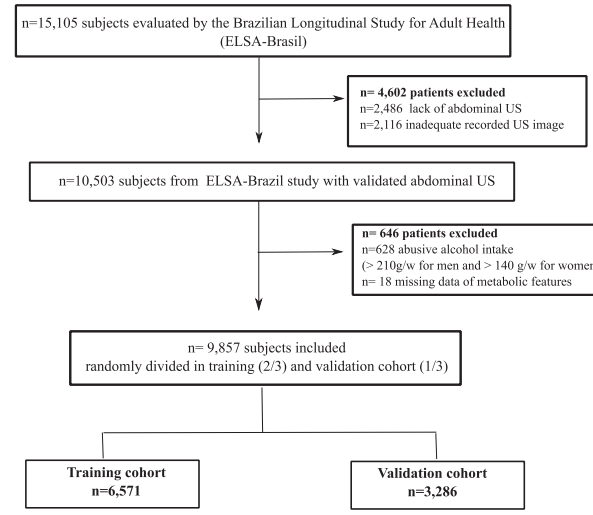

FIGURE 1. Study flow chart of patient's recruitment. ELSA-Brasil indicates Brazilian Longitudinal Study of Adult Health; US, ultrasound.

Factors Independently Associated With Steatosis and Included in the Steato-ELSA Formula

In the training cohort, subjects with steatosis (grade $\geq$1) were older and more frequently male and had higher alcohol consumption (g/wk), BMI, WC, HOMA-IR, liver enzymes, and lipid parameters compared with those without steatosis ($P < 0.001$ for all). The final logistic regression model that defined parameters for the Steato-ELSA's formula included [odds ratio (95% CI)]: BMI, [per Kg/m^2, 1.09 (1.06-1.11), $P < 0.0001$]; WC, [per cm, 1.03 (1.02-1.04), $P < 0.0001$]; HOMA-IR, [per unit, 1.06 (1.03-1.09), $P < 0.0001$]; ALT, [per IU/L, 1.02 (1.01-1.03), $P < 0.0001$]; AST, [per IU/L, 0.98 (0.97–0.99), $P = 0.008$]; and triglycerides [per mg/dL, 1.01 (1.01-1.01), $P < 0.0001$] (Hosmer-Lemeshow statistic $P = 0.279$, mean VIF = 2.76) (Table 2). Using this model, Steato-ELSA was calculated according to the following formula:

$$\frac{2.17828^{[(0.0823\times \text{BMI})+(0.0337\times \text{WC})+(0.0596\times \text{HOMA-IR})+(0.0036\times \text{triglycerides in mg/dL})+(0.0173\times \text{ALT})-(0.0124\times \text{AST})-6.6434]}}{1+2.17828^{[(0.0823\times \text{BMI})+(0.0337\times \text{WC})+(0.0596\times \text{HOMA-IR})+(0.0036\times \text{triglycerides in mg/dL})+(0.0173\times \text{ALT})-(0.0124\times \text{AST})-6.6434]}}.$$

In the training cohort, the median (IQR) Steato-ELSA value was 0.260 (0.169 to 0.399) in individuals without steatosis; 0.415 (0.284 to 0.573) in subjects with grade 1 steatosis; 0.588 (0.430 to 0.738) with grade 2 steatosis, and 0.714 (0.524 to 0.824) with grade 3 steatosis. In the validation cohort, median (IQR) Steato-ELSA values were 0.260 (0.168 to 0.395), 0.410 (0.277 to 0.577), 0.602 (0.442 to 0.737), and 0.685 (0.600 to 0.841) for subjects without steatosis, and with grades 1, 2, and 3 steatosis, respectively (Fig. 2). This predictive model was correlated [Spearman rho (P value)] with the presence of mild steatosis [rho = 0.44 ($P < 0.001$) for both cohorts] and moderate steatosis [0.38 ($P < 0.001$) for training and 0.40 ($P < 0.001$) for validation cohort].

Diagnostic Accuracy of Predictive Models for Liver Steatosis Diagnosis

In the training cohort, AUROCs (95% CI) of Steato-ELSA, FLI, HSI, and NAFLD-LFS for prediction of mild steatosis were 0.759 (0.747-0.771), 0.750 (0.738-0.762), 0.730 (0.717-0.742), and 0.711 (0.698-0.724) ($P < 0.0001$), respectively. In 2-by-2 comparisons, the AUROC of Steato-ELSA was significantly higher than FLI ($P = 0.0013$), HSI ($P < 0.0001$), and NAFLD-LFS ($P < 0.0001$). For prediction of moderate steatosis, the AUROCs (95% CI) were 0.820 (0.807-0.834) for Steato-ELSA, 0.808 (0.794-0.822) for FLI,

TABLE 1. Characteristics of Participants Included in Training and Validation Cohorts

Characteristics	Training Group (N = 6571)	Validation Group (N = 3286)	*P*
Male gender	2772 (42)	1382 (42)	0.903
Age (y)	51 (45-58)	51 (45-58)	0.553
Ethnicity/skin color			0.302
White	3592 (55)	1859 (57)	
African-Black	987 (15)	488 (15)	
Other	1992 (30)	939 (28)	
Alcohol consumption (g/wk)	0 (0-42)	0 (0-42)	0.942
Metabolic features			
BMI (Kg/m^2)	26.5 (23.9-29.7)	26.4 (23.8-29.6)	0.450
Waist circunference (cm)	89.6 (81.3-98.1)	89.5 (81.5-98.1)	0.589
Central obesity	4070 (62)	1999 (61)	0.288
Impared glucose tolerance	1285 (20)	680 (21)	0.271
Type-2 diabetes	1227 (19)	581 (18)	0.271
Dyslipidemia	2564 (39)	1350 (41)	0.057
Hypertension	2839 (43)	1465 (45)	0.204
Metabolic syndrome	2507 (38)	1270 (39)	0.633
HOMA-IR	1.69 (0.96-2.99)	1.69 (0.94-3.00)	0.825
Biochemistry			
ALT (IU/L)	23 (18-32)	23 (18-32)	0.866
AST (IU/L)	23 (20-28)	23 (20-28)	0.576
GGT (IU/L)	25 (18-39)	25 (17-39)	0.457
Fasting glucose (mg/dL)	104 (98-113)	104 (98-113)	0.435
HbA1c (%)	5.3 (5.0-5.8)	5.3 (4.9-5.8)	0.376
Insulinemia (mUI/L)	6.4 (3.8-10.8)	6.5 (3.7-10.9)	0.752
Triglycerides (mg/dL)	111 (80-160)	114 (81-161)	0.557
Total cholesterol (mg/dL)	210 (185-237)	210 (185-238)	0.371
LDL cholesterol (mg/dL)	128 (107-151)	128 (107-151)	0.699
HDL cholesterol (mg/dL)	54 (46-65)	55 (47-65)	0.663
Fatty liver grade [n (%)]			0.699
Grade 0	4095 (62)	2059 (63)	
Grade 1	1536 (24)	785 (24)	
Grade 2	800 (12)	376 (11)	
Grade 3	140 (2)	66 (2)	
Liver steatosis tests			
Fatty Liver Index	41 (18-70)	40 (18-70)	0.632
Hepatic Steatosis Index	38 (34-42)	37 (34-42)	0.891
NAFLD Liver Fat score	−1.139 (−2.104-0.070)	−1.136 (−2.100-0.125)	0.618

Data expressed as absolute (%) or median (interquartile range).
ALT indicates alanine transaminase; AST, aspartate transaminase; BMI, body mass index; GGT, γ-glutamyltransferase; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; LDL, low-density lipoprotein; NAFLD, Nonalcoholic Fatty Liver Disease.

0.787 (0.761-0.792) for HSI and 0.776 (0.761-0.792) for NAFLD ($P < 0.0001$) (Table 3). The AUROC of Steato-ELSA was significantly higher than FLI, HSI, and NAFLD-LFS ($P < 0.0001$ in 2-by-2 comparisons). The maximal Youden index was applied to determine the optimal cutoff for Steato-ELSA for presence of mild (grade ≥ 1) and moderate (grade ≥ 2) steatosis. Considering the presence of mild steatosis, Steato-ELSA values > 0.386 correctly classified 70.3% of subjects with Se = 65.0% (95% CI, 64.1-67.8), Sp = 72.9% (71.5-74.2), PPV = 59.5%, NPV = 78.0%, and LR+ = 2.4. For presence of moderate steatosis, Steato-ELSA > 0.403 correctly classified 69.8% of cases and yielded Se = 81.0% (78.4-83.5), Sp = 68.0% (66.7-69.2), PPV = 29.7%, NPV = 95.5%, and LR+ = 2.53 (Table 4).

In the validation cohort, the AUROCs (95% CI) for prediction of mild and moderate steatosis were: (i) 0.768 (0.751-0.784) and 0.829 (0.810-0.848) for Steato-ELSA; (ii) 0.762 (0.745-0.779) and 0.819 (0.799-0.838) for FLI; (iii) 0.743 (0.727-0.761) and 0.800 (0.779-0.822) for HSI; and (iv) 0.719 (0.701-0.737) and 0.769 (0.747-0.791) for NAFLD-LFS (Table 3). The AUROCs of Steato-ELSA were significantly higher than FLI, his, and NAFLD-LFS for diagnosis of mild steatosis ($P < 0.0001$ for all comparisons) and moderate steatosis ($P < 0.0001$ for all comparisons). In addition, AUROCs (95% CI) for prediction of moderate steatosis were significantly better in white compared with black subjects [0.847 (0.822-0.871) vs. 0.798 (0.745-0.851), $P < 0.0001$] and in women compared with men [0.840 (0.813-0.867) vs. 0.809 (0.780-0.838), $P < 0.0001$]. Supplementary Table1 (Supplemental Digital Content 1, http://links.lww.com/JCG/A383) summarizes accuracy of non-invasive tests according to race and gender. The optimal cutoffs previously determined for Steato-ELSA were used and yielded: (i) Se = 65.6% (63.0-68.3), Sp = 73.7% (71.8-75.6), PPV = 59.8%, NPV = 78.2%, and LR+ = 2.49 for diagnosis of mild steatosis; and (ii) Se = 83.5% (80.0-86.9), Sp = 68.7% (67.0-70.4), PPV = 29.3%, NPV = 96.4%, and LR+ = 2.67 for moderate steatosis (Table 4). AUROCs of Steato-ELSA for detection of steatosis (grade ≥ 1) and moderate steatosis (grade ≥ 2) in training and validation cohorts are plotted in Figure 3.

DISCUSSION

This study highlighted the diagnostic accuracy of serological tests and developed a new predictive model for

TABLE 2. Characteristics of Participants and Multivariate Regression Model to Predict Presence of Steatosis (Grade ≥1) in the Training Cohort

	Training Cohort (N = 6571)						
	Univariate Analysis				Multivariate Analysis		
Characteristics	No Steatosis (N = 4095)	Steatosis (N = 2476)	OR (95% CI)	*P*	OR (95% CI)	β (SE)	*P*
Male gender	1560 (38)	1212 (49)	1.56 (1.41-1.72)	< 0.0001			
Age (y)	50 (44-58)	52 (46-59)	1.02 (1.01-1.02)	< 0.0001			
Alcohol consumption (g/wk)	0 (0-42)	0 (0-50)	1.01 (1.01-1.01)	< 0.0001			
Metabolic features							
BMI (Kg/m^2)	25.3 (23.0-27.9)	28.6 (25.9-32.1)	1.20 (1.19-1.22)	< 0.0001	1.09 (1.06-1.11)	0.0823 (0.0115)	< 0.0001
Waist circunference (cm)	86 (79-94)	96 (89-104)	1.08 (1.07-1.08)	< 0.0001	1.03 (1.02-1.04)	0.0337 (0.0046)	< 0.0001
HOMA-IR	1.41 (0.81-2.36)	2.45 (1.38-3.95)	1.33 (1.29-1.36)	< 0.0001	1.06 (1.03-1.09)	0.0596 (0.0136)	< 0.0001
Biochemistry							
ALT (IU/L)	22 (17-29)	27 (20-37)	1.03 (1.02-1.04)	< 0.0001	1.02 (1.01-1.03)	0.0173 (0.0030)	< 0.0001
AST (IU/L)	23 (19-27)	24 (20-29)	1.02 (1.01-1.03)	< 0.0001	0.98 (0.97-0.99)	−0.0124 (0.0047)	0.008
GGT (IU/L)	23 (16-35)	30 (21-45)	1.01 (1.01-1.01)	< 0.0001			
Triglycerides (mg/dL)	100 (73-139)	133 (96-191)	1.01 (1.01-1.01)	< 0.0001	1.01 (1.01-1.01)	0.0036 (0.0004)	< 0.0001
Total cholesterol (mg/dL)	207 (183-235)	214 (189-242)	1.01 (1.01-1.01)	< 0.0001			
LDL cholesterol (mg/dL)	127 (106-149)	130 (108-154)	1.01 (1.01-1.01)	< 0.0001			
HDL cholesterol (mg/dL)	56 (48-67)	52 (44-61)	0.98 (0.97-0.99)	< 0.0001			
Constant					0.0013 (0.0003)	−6.6434 (0.2629)	

Variance inflation factor values of the final model: mean = 2.76; waist circumference = 4.09; BMI = 3.65; ALT = 3.22; AST = 2.96; HOMA-IR = 1.28; triglycerides = 1.12.

β indicates regression coefficient; ALT, alanine transaminase; AST, aspartate transaminase; BMI, body mass index; CI, confidence interval; GGT, γ-glutamyltransferase; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; LDL, low-density lipoprotein; OR, odds ratio.

diagnosisi of liver steatosis. Steato-ELSA can predict the presence of steatosis using clinical (BMI and WC) and laboratory data (glucose, insulinemia, tryglicerides, and transaminases). Persons with only hepatic steatosis are thought to have a benign long-term prognosis. However, 20% to 30% of those with simple steatosis develop nonalcoholic steatohepatitis (NASH), which may further progress to cirrhosis and its complications.[24] Identification of high-risk subjects for presence of hepatic steatosis is essential to prevent development of liver-related complications.[25] Steato-ELSA was developed and validated in a large sample size of subjects from Brazil, the largest

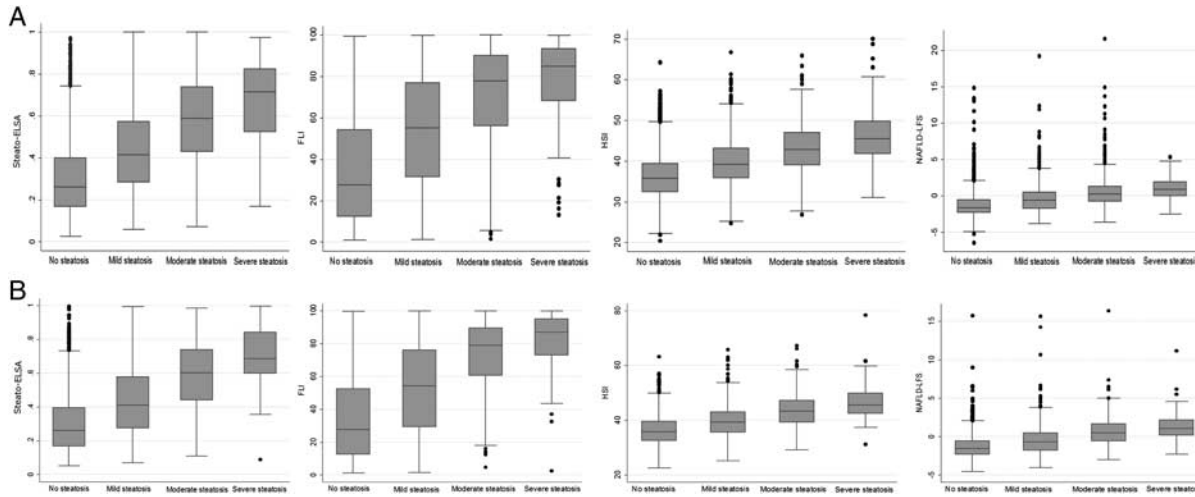

FIGURE 2. Box plots representing the relationship between Steato-ELSA, FLI, HSI, and NAFLD-LFS and the grade of steatosis by abdominal ultrasound in the training (A) and validation (B) cohorts. FLI indicates Fatty Liver Index; HSI, Hepatic Steatosis Index; NAFLD-LFS, Nonalcoholic Fatty Liver Disease-Liver Fat Score.

TABLE 3. AUROCs of Predictive Models for Detection of Mild and Moderate Steatosis in Training and Validation Cohorts

	Training Cohort (N = 6571)	Validation Cohort (N = 3286)
Predictive Model	**AUROC (95% CI) Presence of Mild Steatosis (Prevalence = 38%)**	**AUROC (95% CI) Presence of Mild Steatosis (Prevalence = 37%)**
Steato-ELSA	0.759 (0.747-0.771)*	0.768 (0.751-0.784)†
FLI	0.750 (0.738-0.762)‡	0.762 (0.745-0.779)§
HSI	0.730 (0.717-0.742)‖	0.743 (0.727-0.761)¶
NAFLD-LFS	0.711 (0.698-0.724)	0.719 (0.701-0.737)
P	< 0.0001	< 0.0001
	Moderate Steatosis (Prevalence = 14%)	**Moderate Steatosis (Prevalence = 13%)**
Steato-ELSA	0.820 (0.807-0.834)#	0.829 (0.810-0.848)**
FLI	0.808 (0.794-0.822)††	0.819 (0.799-0.838)‡‡
HSI	0.787 (0.761-0.792)§§	0.800 (0.779-0.822)
NAFLD-LFS	0.776 (0.761-0.792)	0.769 (0.747-0.791)
P	< 0.0001	< 0.0001

Training cohort: *higher than FLI ($P = 0.0013$); HSI ($P < 0.0001$), and NAFLD-LFS ($P < 0.0001$).
†Higher than HSI ($P < 0.0001$) and NAFLD-LFS ($P < 0.0001$).
‡Higher than NAFLD-LFS ($P = 0.0002$).
§Higher than FLI ($P < 0.0001$); HSI ($P < 0.0001$), and NAFLD-LFS ($P < 0.0001$).
‖Higher than HSI ($P = 0.0001$) and NAFLD-LFS ($P < 0.0001$).
¶Higher than NAFLD-LFS ($P = 0.0066$).
Validation cohort: #higher than FLI ($P < 0.0001$); HSI ($P < 0.0001$), and NAFLD-LFS ($P < 0.0001$).
**Higher than HSI ($P = 0.0028$) and NAFLD-LFS ($P < 0.0001$).
††Higher than NAFLD-LFS ($P = 0.0017$).
‡‡Higher than FLI ($P < 0.0001$), HSI ($P < 0.0001$), and NAFLD-LFS ($P < 0.0001$).
§§Higher than HSI ($P = 0.0132$) and NAFLD-LFS ($P = 0.0003$).
AUROC indicates area under the receiver-operator curve; CI, confidence interval; FLI, Fatty Liver Index; HSI, Hepatic Steatosis Index; NAFLD-LFS, Nonalcoholic Fatty Liver Disease-Liver Fat Score.

country in Latin American, and used landmarks of insulin resistance in this formula, such as HOMA-IR and obesity. Therefore, this new predictive model can be used to identify this subset of high-risk individuals that should be further evaluated for presence of advanced fibrosis and referred for modification of lifestyle habits. In the present study, Steato-ELSA performed significantly better than FLI, HSI, and NAFLD-LFS in predicting mild and moderate steatosis in both training and validation cohort.

FLI and HSI were validated using abdominal US and NAFLD-LFS using magnetic resonance spectroscopy (MRS) as the reference. FLI was developed in a sample of individuals with suspected liver disease (n = 216) and matched controls (n = 280) in Italy. For diagnosis of steatosis, AUROC was 0.84 (95% CI, 0.81-0.87) and FLI ≥ 60 had a high specificity (87%).[18] HSI was validated in Korean subjects (5362 individuals with steatosis and 5362 age-matched and gender-matched controls) submitted to a medical check-up. HSI had an AUROC of 0.812 (95% CI, 0.801-0.824) and values > 36 had a specificity of 92% for detection of steatosis.[19] Finally, NAFLD-LFS was developed in a sample of 470 subjects (57% with metabolic syndrome) with chronically elevated transaminases in Finland. The AUROC of NAFLD-LFS was 0.86 (95% CI, 0.80-0.92) and values > −0.640 predicted steatosis with a sensitivity of 86% and specificity of 71%.[20]

In the present study, Steato-ELSA performed significantly better than other models for diagnosis of mild and moderate steatosis in training and validation cohorts. This might be due to use of HOMA-IR, since insulin resistance is key to the pathogenesis of liver fat accumulation. Besides, potential collinearity for obesity assessment, BMI, and WC were entered in the formula. BMI has been limited by factors such as body size and body fat distribution[26] and WC has been observed to be a stronger predictor of obesity-related risk factors than BMI.[27] In addition, there was no severe collinearity (VIF values > 5) between both variables in the final multivariate model (Table 2). The accuracy of FLI, HSI, and NAFLD-LFS were lower in Brazilian subjects compared with the studies that described these predictive models.[18–20] Differences in population characteristics or methods for determination of steatosis might explain those discrepancies. Xia et al[28] reported the impact of ethnicity on the accuracy of FLI, HSI, and NAFLD-LS for predicting liver steatosis using MRS as the gold standard: AUROCs were 0.77 in Chinese subjects (n = 3548) and ranged from 0.72 to 0.81 in Finnish people (n = 572). Using US as the reference, the AUROCs (95% CI) of FLI for detection of steatosis were 0.827 (0.822-0.831) in Taiwanese individuals (n = 29,797),[29] 0.813 (0.797-0.830) in Netherlands (n = 2652),[30] and 0.78 (0.74-0.81) in the United States (n = 5869).[31] In a sensitivity analysis according to ethnicity and gender, Steato-ELSA seems to perform better in White than African-Black and female than male subjects. In addition, Steato-ELSA performed significantly better than HSI and NAFLD-LFS despite differences in gender or ethnicity (Supplementary Table1, Supplemental Digital Content 1, http://links.lww.com/JCG/A383). Ruhl and Everhart[31] proposed a FLI score adapted for multiethnic North-American population adding fasting insulin, glucose and ethnicity to this calculation, which performed slightly better than FLI [0.80 (0.77-0.83)]. FLI [0.76 (0.69-0.83)] was validated in 195 Brazilian individuals[8] and more recently, FLI [0.86 (0.72-0.99)] and HSI [0.75 (0.58-0.91)] were validated in patients with type 1 diabetes in Latvia.[32]

Liver steatosis can be detected based on hepatic parenchyma echogenicity or attenuation of the US wave leading to loss of definition of diaphragm and blurring vascular margins.[5] The accuracy of US for detection of steatosis (> 10% hepatocytes) was validated [0.93 (95% CI,

TABLE 4. Diagnostic Value of Predictive Models for Detection of Mild and Moderate Steatosis in Training and Validation Cohorts

Predictive Models (Cutoff)	Training Cohort (N = 6571)						Validation Cohort (N = 3286)					
	CC (%)	Sensitivity [% (95% CI)]	Specitivity [% (95% CI)]	PPV (%)	NPV (%)	LR+	CC (%)	Sensitivity [% (95% CI)]	Specificity [% (95% CI)]	PPV (%)	NPV (%)	LR+
	Presence of Mild Steatosis (Prevalence = 38%)						Presence of Mild Steatosis (Prevalence = 37%)					
Steato-ELSA ≥ 0.386*	70.3	65.0 (64.1-67.8)	72.9 (71.5-74.2)	59.5	78.0	2.43	70.7	65.6 (63.0-68.3)	73.7 (71.8-75.6)	59.8	78.2	2.49
FLI ≥ 43*	68.7	72.5 (70.7-74.3)	66.4 (65.0-67.9)	56.6	80.0	2.16	68.5	71.8 (69.3-73.4)	66.5 (64.5-68.6)	56.1	79.8	2.15
FLI ≥ 60[18]	70.4	55.7 (53.8-57.7)	79.2 (78.0-80.5)	61.9	74.7	2.68	71.1	55.6 (52.9-58.4)	80.2 (78.5-80.0)	62.7	75.2	2.82
HSI ≥ 38.6*	68.1	64.5 (62.6-66.3)	70.2 (68.8-71.6)	56.7	76.5	2.17	68.4	65.2 (62.5-67.9)	70.3 (68.3-72.3)	56.7	77.2	2.19
HSI ≥ 36[19]	62.1	80.5 (78.9-82.0)	51.0 (49.5-52.6)	49.8	81.2	1.64	62.6	80.6 (78.4-82.9)	51.9 (49.8-54.1)	50.0	81.8	1.68
NAFLD-LFS ≥ −0.720*	68.0	60.7 (58.1-62.7)	72.5 (71.1-78.3)	57.1	75.3	2.21	68.0	60.6 (57.8-63.3)	72.4 (70.5-74.3)	56.7	75.5	2.20
NAFLD-LFS ≥ −0.640[20]	68.1	60.3 (58.4-62.3)	72.8 (71.4-74.2)	57.3	75.2	2.22	68.1	60.2 (57.5-63.0)	72.9 (70.9-74.8)	56.9	75.4	2.22
	Moderate Steatosis (Prevalence = 14%)						Moderate Steatosis (Prevalence = 13%)					
Steato-ELSA ≥ 0.403*	69.8	81.0 (78.4-83.5)	68.0 (66.7-69.2)	29.7	95.5	2.53	70.7	83.5 (80.0-86.9)	68.7 (67.0-70.4)	29.3	96.4	2.67
FLI ≥ 57.5*	71.5	76.3 (73.6-79.0)	70.7 (69.5-71.9)	30.3	94.7	2.61	72.7	79.6 (75.8-83.3)	71.7 (70.0-73.3)	30.3	95.8	2.80
FLI ≥ 60[18]	72.9	73.9 (71.1-76.7)	72.7 (71.6-73.9)	31.2	94.4	2.71	74.1	77.2 (73.2-81.1)	73.7 (72.1-75.3)	31.3	95.4	2.93
HSI ≥ 39.7*	70.4	74.5 (71.7-77.3)	69.7 (68.5-70.9)	29.1	94.2	2.46	70.1	76.0 (72.0-80.0)	69.1 (67.4-70.8)	27.7	94.9	2.46
HSI ≥ 36[19]	50.6	89.8 (87.9-91.7)	44.0 (42.7-45.3)	21.1	96.3	1.60	51.7	94.3 (92.2-96.5)	45.1 (43.3-46.9)	21.1	98.1	1.72
NAFLD-LFS ≥ −0.360*	72.1	67.5 (64.5-70.4)	72.9 (71.7-74.1)	29.4	93.1	2.49	73.1	72.2 (68.0-76.3)	73.2 (71.6-74.9)	29.5	94.4	2.70
NAFLD-LFS ≥ −0.640[20]	67.4	74.8 (72.0-77.6)	66.2 (64.9-67.4)	27.0	94.0	2.21	68.2	78.7 (74.9-82.6)	66.6 (64.9-68.3)	26.8	95.3	2.36

*Cutoff based on the maximum Youden index (Se+Sp-1).

CC indicates corrected classified; FLI, Fatty Liver Index; HSI, Hepatic Steatosis Index; LR+, positive likelihood ratio; NAFLD-LFS, NAFLD Fat Liver Score; NPV, negative predictive value; PPV, positive predictive value.

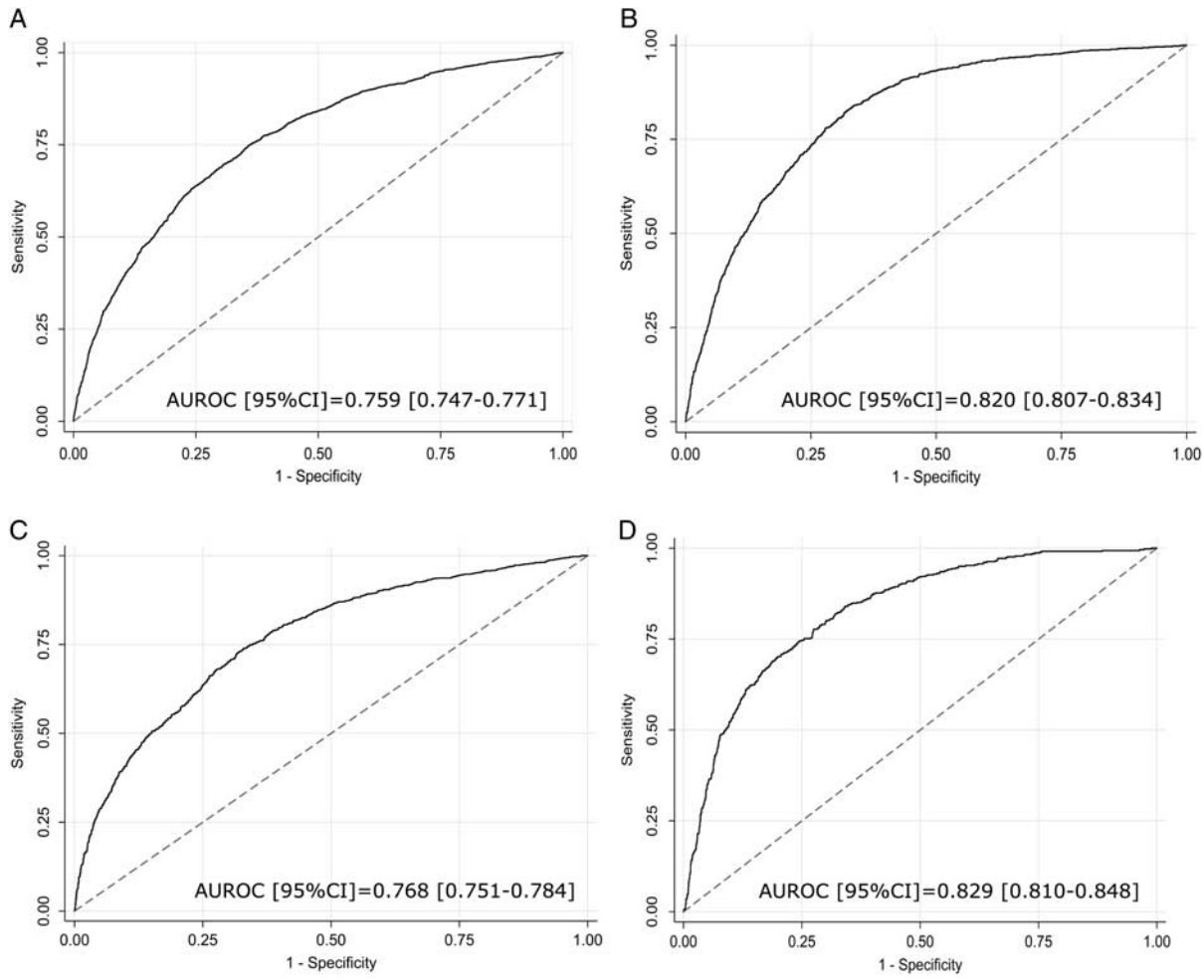

FIGURE 3. AUROC of Steato-ELSA for prediction of presence of steatosis (grade ≥1) (A) and moderate steatosis (grade ≥2) (B) in the training cohort (n = 6571). AUROC of Steato-ELSA for prediction of presence of steatosis (grade ≥1) (C) and moderate steatosis (grade ≥2) (D) in the validation cohort (n = 3286). AUROC indicates area under the receiver-operator characteristic; CI, confidence interval.

0.91-0.95)] in a meta-analysis (n = 2815 participants) which included 34 studies that used liver biopsy as the reference.[33] We previously validated the analysis of liver attenuation by US for steatosis detection using 64 channel high-resolution computed tomography as the gold standard [0.84 (95% CI, 0.77-0.90)].[8] However, this method has a considerable interobserver variability and sensitivity might be reduced in presence of <30% of fat infiltration.[34] In a study (n = 324) which liver biopsy was the gold standard, FLI, HSI, and NAFLD-LFS yielded AUROCs of 0.83 (0.72-0.91), 0.81 (0.71-0.88), and 0.80 (0.69-0.88), respectively, for steatosis detection (>5% hepatocytes). However, these noninvasive tests were less accurate for detection of moderate/severe steatosis (>33%).[35]

Differences in prevalence of steatosis grades might impact in diagnostic test's performance, known as spectrum bias.[36] FLI and HSI were developed in case-control studies, whereas NAFLD-LFS and Steato-ELSA in cross-sectional studies with prospective inclusion of subjects. Prevalence of steatosis was extremely variable in the studies that validated the diagnostic value of noninvasive tests for detection of steatosis using abdominal US as the reference. In the present study the prevalence of mild (grade ≥1) and moderate (grade ≥2) steatosis were 38% (n = 3703) and 14% (n = 1382), respectively. We are aware that fatty liver is strongly associated with insulin resistance,[37] but the need of insulinemia might be a concern for Steato-ELSA calculation. In absence of fasting insulin, Steato-ELSA without HOMA-IR might be calculated (Supplementary Tables 2 and 3, Supplemental Digital Content 1, http://links.lww.com/JCG/A383).

A major limitation of this study was the absence of liver biopsy to assess the diagnostic performance of predictive models for detection of steatosis. However, it might be unethical to perform liver biopsy in subjects without suspicion of liver disease as ELSA-Brasil population. MRS could be used to replace liver biopsy as the gold standard. However, this method is costly and not widely available despite its excellent sensitivity and high reproducibility.[38] We acknowledge that US has lower sensitivity in presence of mild fat infiltration, but abnormalities in hepatic echogenicity or attenuation of US waves were validated to detect steatosis.[33] Other potential criticisms should be the limited sample size of subjects with severe steatosis (n = 206/9587), the lack of evaluation of the impact of liver fibrosis on noninvasive tests' performance and the absence of screening

for chronic liver disease in the study population. The AUROCs (95% CI) of Steato-ELSA for severe steatosis (grade = 3) were 0.848 (0.820-0.875) and 0.869 (0.831-0.907) in the training and validation cohort, respectively. Subjects included in the analysis did not have known chronic liver diseases. In addition, the very low prevalence (< 1%) of necroinflammatory activity (ALT levels > 3× upper limit of normal) and advanced fibrosis (METAVIR F3F4) using a noninvasive test (FIB-4 ≥ 3.25) minimized the potential bias of lacking screening of chronic viral hepatitis in the ELSA-Brasil study. We acknowledge a potential selection bias by exclusion of 5248 subjects who were significantly older, more frequently male and had higher BMI, WC liver enzymes and lipid parameters compared with those included in the study (Supplementary Table 4, Supplemental Digital Content 1, http://links.lww.com/JCG/A383). The strengths of our study include large size of study population and the use of a centralized laboratory for blood tests and a centralized and blinded reading center for analysis of recorded hepatic images to determine steatosis degrees. Despite the fact that this is not a population-based study, we included 9587 subjects from 6 Brazilian centers.

In conclusion, available predictive models using serological parameters and the new proposed Steato-ELSA are accurate and inexpensive tools for identifying individuals at high risk of liver steatosis. These noninvasive tests can be used to select individuals for screening of liver fibrosis related to NAFLD or to be referred for modification of lifestyle habits. Even though Steato-ELSA seems to be an accurate predictive model for detection of liver steatosis, this noninvasive test must be further validated in studies including different populations with different chronic liver diseases.

ACKNOWLEDGMENTS

The authors thank the ELSA-Brasil participants who agreed to take part in this study and the ELSA-Brasil research team for their contributions.

REFERENCES

1. Bellentani S. The epidemiology of non-alcoholic fatty liver disease. *Liver Int*. 2017;37(suppl 1):81–84.
2. Bhala N, Jouness RI, Bugianesi E. Epidemiology and natural history of patients with NAFLD. *Curr Pharm Des*. 2013;19: 5169–5176.
3. Niriella MA, Pathmeswaran A, De Silva ST, et al. Incidence and risk factors for non-alcoholic fatty liver disease: a 7-year follow-up study among urban, adult Sri Lankans. *Liver Int*. 2017;37:1715–1722.
4. Bedossa P, Dargere D, Paradis V. Sampling variability of liver fibrosis in chronic hepatitis C. *Hepatology*. 2003;38:1449–1457.
5. Castera L, Vilgrain V, Angulo P. Noninvasive evaluation of NAFLD. *Nat Rev Gastroenterol Hepatol*. 2013;10:666–675.
6. Stern C, Castera L. Non-invasive diagnosis of hepatic steatosis. *Hepatol Int*. 2017;11:70–78.
7. Schmidt MI, Duncan BB, Mill JG, et al. Cohort profile: longitudinal study of adult health (ELSA-Brasil). *Int J Epidemiol*. 2015;44:68–75.
8. Goulart AC, Oliveira IR, Alencar AP, et al. Diagnostic accuracy of a noninvasive hepatic ultrasound score for non-alcoholic fatty liver disease (NAFLD) in the Brazilian Longitudinal Study of Adult Health (ELSA-Brasil). *Sao Paulo Med J*. 2015;133:115–124.
9. Aquino EM, Barreto SM, Bensenor IM, et al. Brazilian Longitudinal Study of Adult Health (ELSA-Brasil): objectives and design. *Am J Epidemiol*. 2012;175:315–324.
10. Fedeli LG, Vidigal PG, Leite CM, et al. Logistics of collection and transportation of biological samples and the organization of the central laboratory in the ELSA-Brasil. *Rev Saude Publica*. 2013;47(suppl 2):63–71.
11. Mill JG, Pinto K, Griep RH, et al. Medical assessments and measurements in ELSA-Brasil. *Rev Saude Publica*. 2013;47 (suppl 2): 54–62.
12. Matthews DR, Hosker JP, Rudenski AS, et al. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia*. 1985;28:412–419.
13. Bugianesi E, Pagotto U, Manini R, et al. Plasma adiponectin in nonalcoholic fatty liver is related to hepatic insulin resistance and hepatic fat content, not to liver disease severity. *J Clin Endocrinol Metab*. 2005;90:3498–3504.
14. Lotufo PA, Santos RD, Figueiredo RM, et al. Prevalence, awareness, treatment, and control of high low-density lipoprotein cholesterol in Brazil: baseline of the Brazilian Longitudinal Study of Adult Health (ELSA-Brasil). *J Clin Lipidol*. 2016;10:568–576.
15. Chor D, Pinho Ribeiro AL, Sa Carvalho M, et al. Prevalence, awareness, treatment and influence of socioeconomic variables on control of high blood pressure: results of the ELSA-Brasil Study. *PLoS One*. 2015;10:e0127382.
16. Schmidt MI, Hoffmann JF, de Fatima Sander Diniz M, et al. High prevalence of diabetes and intermediate hyperglycemia—The Brazilian Longitudinal Study of Adult Health (ELSA-Brasil). *Diabetol Metab Syndr*. 2014;6:123.
17. Alberti KG, Zimmet P, Shaw J. Metabolic syndrome—a new world-wide definition. A consensus statement from the International Diabetes Federation. *Diabet Med*. 2006;23: 469–480.
18. Bedogni G, Bellentani S, Miglioli L, et al. The fatty liver index: a simple and accurate predictor of hepatic steatosis in the general population. *BMC Gastroenterol*. 2006;6:33.
19. Lee JH, Kim D, Kim HJ, et al. Hepatic steatosis index: a simple screening tool reflecting nonalcoholic fatty liver disease. *Dig Liver Dis*. 2010;42:503–508.
20. Kotronen A, Peltonen M, Hakkarainen A, et al. Prediction of non-alcoholic fatty liver disease and liver fat using metabolic and genetic factors. *Gastroenterology*. 2009;137:865–872.
21. Akinwande MO, Dikko HG, Samson A. Variance inflation factor: as a condition for the inclusion of suppressor variable(s) in regression analysis. *Open J Stat*. 2015;5:754–767.
22. Hosmer DW, Lemeshow S. *Applied Logistic Regression*. New York: Wiley; 2000.
23. DeLong ER, DeLong DM, Clarke-Pearson DL. Comparing the areas under two or more correlated receiver operating characteristic curves: a nonparametric approach. *Biometrics*. 1988;44:837–845.
24. Tsai E, Lee TP. Diagnosis and evaluation of nonalcoholic fatty liver disease/nonalcoholic steatohepatitis, including noninvasive biomarkers and transient elastography. *Clin Liver Dis*. 2018;22: 73–92.
25. Chalasani N, Younossi Z, Lavine JE, et al. The diagnosis and management of nonalcoholic fatty liver disease: practice guidance from the American Association for the Study of Liver Diseases. *Hepatology*. 2018;67:328–357.
26. Kyle UG, Schutz Y, Dupertuis YM, et al. Body composition interpretation. Contributions of the fat-free mass index and the body fat mass index. *Nutrition*. 2003;19:597–604.
27. Dalton M, Cameron AJ, Zimmet PZ, et al. Waist circumference, waist-hip ratio and body mass index and their correlation with cardiovascular disease risk factors in Australian adults. *J Intern Med*. 2003;254:555–563.
28. Xia MF, Yki-Jarvinen H, Bian H, et al. Influence of ethnicity on the accuracy of non-invasive scores predicting non-alcoholic fatty liver disease. *PLoS One*. 2016;11:e0160526.
29. Yang BL, Wu WC, Fang KC, et al. External validation of fatty liver index for identifying ultrasonographic fatty liver in a large-scale cross-sectional study in Taiwan. *PLoS One*. 2015;10: e0120443.

30. Koehler EM, Schouten JN, Hansen BE, et al. External validation of the fatty liver index for identifying nonalcoholic fatty liver disease in a population-based study. *Clin Gastroenterol Hepatol*. 2013;11:1201–1204.
31. Ruhl CE, Everhart JE. Fatty liver indices in the multiethnic United States National Health and Nutrition Examination Survey. *Aliment Pharmacol Ther*. 2015;41:65–76.
32. Sviklane L, Olmane E, Dzerve Z, et al. Fatty liver index and hepatic steatosis index predict non-alcoholic fatty liver disease in type 1 diabetes. *J Gastroenterol Hepatol*. 2018;33:270–276.
33. Hernaez R, Lazo M, Bonekamp S, et al. Diagnostic accuracy and reliability of ultrasonography for the detection of fatty liver: a meta-analysis. *Hepatology*. 2011;54:1082–1090.
34. Venkatesh SK, Hennedige T, Johnson GB, et al. Imaging patterns and focal lesions in fatty liver: a pictorial review. *Abdom Radiol (NY)*. 2017;42:1374–1392.
35. Fedchuk L, Nascimbeni F, Pais R, et al. Performance and limitations of steatosis biomarkers in patients with nonalcoholic fatty liver disease. *Aliment Pharmacol Ther*. 2014;40:1209–1222.
36. Poynard T, de Ledinghen V, Zarski JP, et al. Fibrotest and fibroscan performances revisited in patients with chronic hepatitis C. Impact of the spectrum effect and the applicability rate. *Clin Res Hepatol Gastroenterol*. 2011;35:720–730.
37. Cruz MA, Cruz JF, Macena LB, et al. Association of the nonalcoholic hepatic steatosis and its degrees with the values of liver enzymes and homeostasis model assessment-insulin resistance index. *Gastroenterology Res*. 2015;8:260–264.
38. Tyagi A, Yeganeh O, Levin Y, et al. Intra- and inter-examination repeatability of magnetic resonance spectroscopy, magnitude-based MRI, and complex-based MRI for estimation of hepatic proton density fat fraction in overweight and obese children and adults. *Abdom Imaging*. 2015;40:3070–3077.