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Hepascore: An Accurate Validated Predictor of Liver Fibrosis in Chronic Hepatitis C Infection

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Background: **Staging hepatic fibrosis by liver biopsy guides prognosis and treatment of hepatitis C, but is invasive and expensive. We sought to create an algorithm of serum markers that accurately and reliably predict liver fibrosis stage among hepatitis C patients.**

Methods: **Ten biochemical markers were measured at time of liver biopsy in 117 untreated hepatitis C patients (training set). Multivariate logistic regression and ROC curve analyses were used to create a predictive model for significant fibrosis (METAVIR F2, F3, and F4), advanced fibrosis (F3 and F4), and cirrhosis (F4). The model was validated in 104 patients from other institutions.**

Results: **A model (Hepascore) of bilirubin, γ-glutamyltransferase, hyaluronic acid, α_2-macroglobulin, age, and sex produced areas under the ROC curves (AUCs) of 0.85, 0.96, and 0.94 for significant fibrosis, advanced fibrosis, and cirrhosis, respectively. In the training set, a score ≥0.5 (range, 0.0–1.0) was 92% specific and 67% sensitive for significant fibrosis, a score <0.5 was 81% specific and 95% sensitive for advanced fibrosis, and a score <0.84 was 84% specific and 71% sensitive for cirrhosis. Among the validation set, the AUC for significant fibrosis, advanced fibrosis, and cirrhosis were 0.82, 0.90, and 0.89, respectively. A score ≥0.5 provided a specificity and sensitivity of 89% and 63% for significant fibrosis, whereas scores <0.5 had 74% specificity and 88% sensitivity for advanced fibrosis.**

Conclusions: **A model of 4 serum markers plus age and sex provides clinically useful information regarding different fibrosis stages among hepatitis C patients.**

Chronic hepatitis C infection is common with an estimated worldwide prevalence of 3% *(1)*. Death, hospitalization, and liver transplantation from hepatitis C have increased dramatically in the past decade *(2)*. Liver fibrosis is the main determinant of hepatitis C virus–related morbidity and mortality *(3)*. In addition, the stage of fibrosis is prognostic and provides information on the likelihood of disease progression and response to treatment *(4, 5)*. The presence of significant fibrosis (equivalent to METAVIR F2 or greater) on liver biopsy is widely accepted as an indication to commence treatment *(6–8)*. The presence of cirrhosis also has implications regarding screening for hepatocellular carcinoma and esophageal varices *(7)*.

Liver biopsy is currently the gold standard for staging fibrosis, but it has well-documented complications of pain, bleeding, and rarely, death *(9, 10)*. Liver biopsy is also expensive, as are the costs associated with treating its complications. In addition, inter- and intraobserver error may lead to incorrect staging *(4)*, as may sampling error in up to 33% of biopsies *(11)*.

Routinely measured serum markers, used either individually or in combination, have been examined as alternatives for staging fibrosis among hepatitis C patients. Platelet count, the ratio of aspartate aminotransferase

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(AST)[7] to alanine aminotransferase (ALT), or a combination of AST and platelet count are reliable predictors of cirrhosis *(12, 13)*, but their predictive value for mild or moderate fibrosis is insufficient to be of clinical utility *(13, 14)*. More complex models that include routinely available analytes such as cholesterol, γ-glutamyltransferase (GGT), platelet count, and prothrombin time have a high negative predictive value (NPV) for excluding significant hepatic fibrosis but a poor positive predictive value (PPV) and are applicable only to approximately one third of patients *(15)*. A recently reported model incorporating measures of insulin resistance and past alcohol intake reliably predicted significant fibrosis but was less accurate in excluding significant fibrosis *(16)*.

In an effort to improve the accuracy of noninvasive methods for staging liver fibrosis, more sophisticated nonroutinely available biochemical markers associated with collagen and extracellular matrix deposition/degradation have been examined. Serum concentrations of hyaluronic acid, tissue inhibitor of matrix metalloproteinase-1 (TIMP-1), and matrix metalloproteinase-2 (MMP-2) correlate with liver fibrosis but by themselves have low predictive value for diagnosing significant fibrosis *(17, 18)*. The "Fibrotest", which combines multiple biochemical markers with age and sex, was accurate in detecting significant fibrosis in just under one half of patients from a center in France *(19)*. However, when applied to a population of hepatitis C patients from our institution, the Fibrotest was less accurate and had a PPV <80% *(20)*.

In an effort to create a predictive model with superior accuracy, we therefore examined a range of routine and novel biochemical markers. We specifically examined α_2-macroglobulin, hyaluronic acid, TIMP-1, and MMP-2 because these molecules are intimately involved in hepatic fibrogenesis *(18, 19, 21)*. In addition, we examined apolipoprotein A1, haptoglobin, and routinely available liver tests (bilirubin, GGT, ALT, and albumin) because these markers have been demonstrated to correlate with fibrosis stage among hepatitis C–infected patients *(19)*. The validity of the model was then tested in a separate population of patients from other Australian liver clinics.

Materials and Methods

PATIENTS

Patients were prospectively recruited from viral liver clinics in different tertiary referral centers; the training set was recruited from Sir Charles Gairdner Hospital (Perth, Australia) and the validation set from Westmead Hospital and Royal Prince Alfred Hospital (Sydney, Australia). All patients had detectable hepatitis C RNA at the time of evaluation and were treatment naive. Coexisting liver disease attributable to hepatitis B, hemochromatosis, α_1-antitrypsin deficiency, Wilson disease, and autoimmune or cholestatic liver diseases were excluded by standard clinical, laboratory, imaging, and histologic studies. No patient had HIV co-infection or had undergone liver transplantation. Liver biopsy was performed as part of the routine clinical care of these patients. Age, sex, and viral genotype were recorded at time of liver biopsy. The collection of serum was approved by the Sir Charles Gairdner Hospital Ethics Committee. Patients from the training set had the Fibrotest calculated as described previously *(20)*. All patients gave informed consent, and the study was carried out according to the principles of the Declaration of Helsinki.

BIOCHEMICAL MARKERS

Training set sera were analyzed for 10 candidate markers. Bilirubin, ALT, GGT, and albumin were all measured on fresh serum within 36 h of collection on an automated biochemistry analyzer (Hitachi 917; Roche Diagnostics). Other analyses were performed in batches with sera stored frozen at −20 °C. TIMP-1 and MMP-2 were measured by ELISA on a 96-well microplate (Biotrak; Amersham Biosciences). Hyaluronic acid was measured by an enzyme-linked protein-binding assay, also on a 96-well microplate (Corgenix). α_2-Macroglobulin, apolipoprotein-A1, and haptoglobin were all obtained by nephelometry (Immage; Beckman Coulter). All analyses were performed at a central laboratory, PathCentre in Perth.

The final predictive model was computed from the results obtained for the following 4 biochemical markers; bilirubin, GGT, α_2-macroglobulin, and hyaluronic acid. The in-house analytical CVs were 1.7% at a bilirubin concentration of 16 μmol/L, 2.7% at a GGT activity of 33 U/L, 2.8% at an α_2-macroglobulin concentration of 2.5 g/L, and 3.5% at a hyaluronic acid concentration of 50 μg/L.

LIVER BIOPSIES

Liver biopsies were obtained with an 18-gauge or larger needle with a minimum of 5 portal tracts and were routinely stained with hematoxylin–eosin and trichrome stains. Biopsies were interpreted according to the scoring schema developed by the METAVIR group *(22)* by 2 expert liver pathologists (B.deB., J.K.) who were blinded to patient clinical characteristics and serum measurements. Thirty biopsies were scored by both pathologists, and interobserver agreement was calculated by use of κ statistics. Fibrosis was scored on a 5-point scale: F0, no fibrosis; F1, portal fibrosis alone; F2, portal fibrosis with rare septae; F3, portal fibrosis with many septae; F4, cirrhosis. The presence of stage F2, F3, or F4 was termed "significant fibrosis", whereas the term "advanced fibrosis" was reserved for stage F3 or F4. Necro-inflammatory activity, based on assessment of piecemeal and lobular

[7] Nonstandard abbreviations: AST, aspartate aminotransferase; ALT, alanine aminotransferase; GGT, γ-glutamyltransferase; NPV, negative predictive value; PPV, positive predictive value; TIMP-1, tissue inhibitor of matrix metalloproteinase-1; MMP-2, matrix metalloproteinase-2; AUC, area under the curve; 95% CI, 95% confidence interval; and APRI, aspartate aminotransferase-to-platelet ratio index.

necrosis, was graded on a 4-point scale: A0, no activity; A1, mild; A2, moderate; A3, severe.

STATISTICAL ANALYSIS

Associations between each of the 10 biochemical markers and the presence or absence of significant fibrosis were assessed by logistic regression. In addition, the diagnostic accuracy of each biochemical marker was assessed by ROC curve analysis. All biochemical markers were combined with age and sex and entered into stepwise logistic regression analysis using a forward and a backward elimination procedure with a significance level of $P = 0.10$. The dependent variable was defined as significant fibrosis as detected by liver biopsy. Biochemical markers with a high area under the curve (AUC) or a high significance on univariate analysis were added to create different multivariable models. Models based on different marker combinations were then compared by ROC curve analysis to determine which was most accurate in detecting significant fibrosis. A single model with the fewest variables and the greatest AUC was selected and applied to the validation set. The logistic regression model consisted of:

$$y = \exp[-4.185818 - (0.0249 \times \text{age}) + (0.7464 \times \text{sex}) + (1.0039 \times \alpha_2\text{-macroglobulin}) + (0.0302 \times \text{hyaluronic acid}) + (0.0691 \times \text{bilirubin}) - (0.0012 \times \text{GGT})]$$

with age provided in years, male sex = 1, female sex = 0, α_2-macroglobulin in g/L, hyaluronate in μg/L, bilirubin in μmol/L, and GGT in U/L. The Hepascore was calculated from the following equation:

$$\frac{y}{1+y}$$

Sensitivity, specificity, PPV, and NPV for significant fibrosis, advanced fibrosis, and cirrhosis were determined for various cutoff points between 0 and 1 in the training and validation sets. The same Hepascore regression model was also used to calculate the accuracy for determining the combined endpoint of moderate to severe necro-inflammatory activity (A2 and A3) vs no or mild activity (A0 and A1). Clinical and demographic characteristics between the training and validation sets were compared by the Student *t*-test for continuous variables and χ^2 test or Fisher exact test for categorical variables. A P value <0.05 was considered significant. All statistical analyses were done with Stata, Ver. 8 (Stata Corporation)

Results

PATIENT CHARACTERISTICS

The demographic and biochemical characteristics of the training (n = 117) and validation sets (n = 104) were generally similar (Table 1), but the validation set had a lower proportion of patients with genotype 1 (48% vs 61%) and more genotype 4 patients (5% vs 0%). There was also a trend toward a greater proportion of patients in the validation set having significant fibrosis ($P = 0.05$) but not advanced fibrosis ($P = 0.4$). The median portal tract number was 9, and the median biopsy length was 13 mm. The interobserver agreement between pathologists was good ($\kappa = 0.56$) for METAVIR staging and for significant fibrosis ($\kappa = 0.72$).

Table 1. Clinical and laboratory features of the training and validation cohorts.

	Training set (n = 117)	Validation set (n = 104)	*P*
Mean (SD) age, years	40 (9)	41 (9)	0.5
Female, n (%)	38 (32%)	28 (27%)	0.4
Genotype,[a] n (%)			0.03
1	67 (61%)	50 (48%)	
2/3	45 (39%)	48 (47%)	
4	0	5 (5%)	
Mean (SD) ALT, U/L	124 (90)	131 (99)	0.5
Mean (SD) bilirubin, μmol/L	12 (8)	12 (5)	0.5
Mean (SD) albumin, g/L	42 (4)	42 (3)	0.7
Stage,[b] n (%)			0.03
F0	23 (19%)	17 (16%)	
F1	43 (37%)	28 (27%)	
F2	29 (25%)	35 (34%)	
F3	15 (13%)	7 (7%)	
F4	7 (6%)	17 (16%)	
Significant fibrosis (F2–F4)	51 (44%)	59 (57%)	0.05
Advanced fibrosis (F3 and F4)	22 (19%)	24 (23%)	0.4
Necro-inflammatory activity, n (%)			0.001
Grade A0	20 (17%)	8 (8%)	
Grade A1	75 (64%)	63 (60%)	
Grade A2	13 (11%)	32 (31%)	
Grade A3	9 (8%)	1 (1%)	
Significant activity (A2 and A3)	22 (19%)	33 (32%)	0.03

[a] Genotype not available for 5 patients in the training set and 1 patient in the validation set.

[b] Histology scored according to METAVIR *(22)*.

MARKERS ASSOCIATED WITH SIGNIFICANT FIBROSIS

Univariate logistic regression analysis of the variables tested in the training set revealed that age, sex, albumin, hyaluronic acid, α_2-macroglobulin, and TIMP-1 were associated with significant fibrosis (Table 2).

PREDICTIVE MODEL

Biochemical markers assessed in the training set were combined with age and sex in logistic regression analyses to create several models that were predictive of significant fibrosis. The optimal multivariable model was considered as having the largest AUC by ROC analysis. This model (Hepascore) consisted of age, sex, bilirubin, GGT, hyaluronic acid, and α_2-macroglobulin (Table 3), which provided a high AUC [95% confidence interval (CI)] for the prediction of significant fibrosis [0.852 (0.778–0.926)] as

Table 2. Association of age, sex, and serum biochemical markers with significant fibrosis in the training cohort (n = 117).

Variable[a]	Stage F0/F1 (n = 66)	Stage F2–F4 (n = 51)	*P*, univariate analysis
Age, years	38.7	41.9	0.03
Sex (female), %	38.6	21.7	0.03
ALT, U/L	123.4	125.3	0.9
GGT, U/L	78.1	111.4	0.1
Bilirubin, μmol/L	10.8	13.0	0.2
Albumin, g/L	42.5	40.4	<0.001
Haptoglobin, g/L	1.0	0.9	0.4
Hyaluronic acid, μg/L	20.7	107.3	<0.001
Apolipoprotein A1, g/L	1.7	1.6	0.1
α_2-Macroglobulin, g/L	2.3	3.3	<0.001
TIMP-1, μg/L	880	1404	0.002
MMP-2, μg/L	731	830	0.1

[a] All variables except sex (female) are given as the mean.

well as for advanced fibrosis [0.957 (0.918–0.995)] and cirrhosis [0.938 (0.872–1.000)], as shown in Fig. 1. In comparison, the Fibrotest results in the training set provided AUC values for significant fibrosis, advanced fibrosis, and cirrhosis of 0.793 (0.706–0.880), 0.906 (0.833–0.979), and 0.966 (0.918–1.000), respectively.

The Hepascore (range, 0.0–1.0) increased significantly (P <0.001) as fibrosis stage increased (Fig. 2). A central cutoff point of 0.5 among the training set predicted significant fibrosis (F2–F4) with a sensitivity of 67% (95% CI, 58.1%–75.2%) and a specificity of 92% (87.6%–97.2%). When we applied the same cutoff point of 0.5 for the prediction of advanced fibrosis (F3 and F4), sensitivity was 95% (91.7%–99.2%) and specificity was 81% (74.0%–88.2%). When a cutoff point of 0.84 was applied for detection of cirrhosis (F4), it provided a 71% (63.2%–79.6%) sensitivity and an 84% (76.9%–90.3%) specificity.

MODEL VALIDATION

The 4-marker model was applied to the 104 patients in the validation set and provided AUCs of 0.820 (95% CI, 0737–0.902) for significant fibrosis, 0.903 (0.835–0.971) for advanced fibrosis, and 0.891 (0.805–0.976) for cirrhosis (Fig. 2).

Table 3. Multiple logistic regression model for the prediction of significant fibrosis.

Variable	Coefficient	SE	*P*	Odds ratio (95% CI)
Age (years)	−0.02	0.03	0.44	0.98 (0.92–1.04)
Sex (female)	0.75	1.13	0.16	2.11 (0.74–6.05)
α_2-Macroglobulin (g/L)	1.00	0.78	0.0001	2.73 (1.56–4.79)
Hyaluronic acid (μg/L)	0.03	0.01	0.005	1.03 (1.01–1.05)
Bilirubin (μmol/L)	0.07	0.04	0.09	1.07 (0.99–1.16)
GGT (U/L)	-0.01	0.01	0.59	1.00 (0.99–1.03)

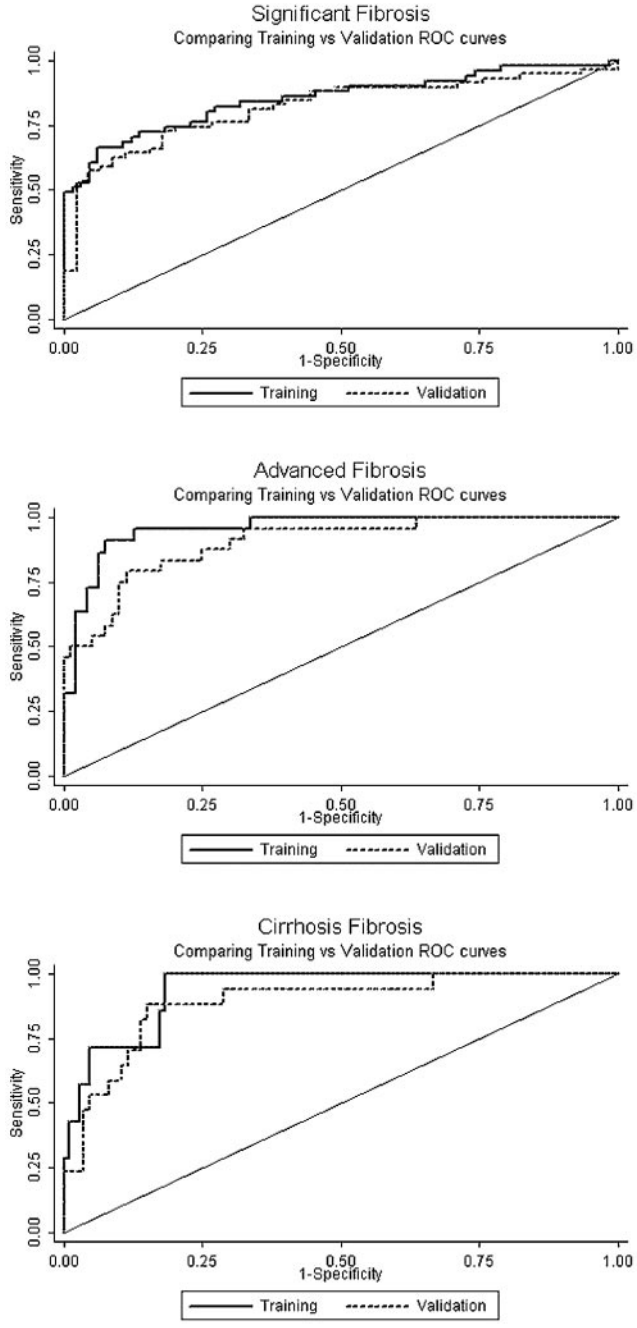

Fig. 1. Hepascore ROC curves for training and validation sets for significant fibrosis (F2–F4; *top*), advanced fibrosis (F3 and F4; *middle*), and cirrhosis (F4; *bottom*).

The AUCs for the training and validation sets were not significantly different for significant fibrosis (P = 0.6), advanced fibrosis (P = 0.2), or cirrhosis (P = 0.4).

Among the validation cohort, 42 of 104 (40%) had a score ≥0.5. A cutoff point of 0.5 gave a sensitivity of 63% (95% CI, 53.4%–72.0%) and a specificity of 89% (82.9%–94.9%) for the presence of significant fibrosis (F2 to F4); therefore, 37 of 42 (88%) patients with a score ≥0.5 had

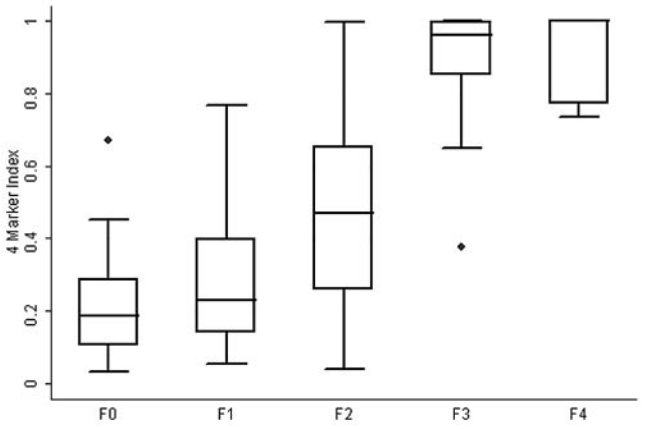

Fig. 2. Box plots of Hepascore according to fibrosis stage in the training set (n = 117).

Hepascores ranged from 0.0 to 1.0. Fibrosis was staged according to METAVIR. The *line inside* each *box* represents the median, the *upper* and *lower limits* of the *boxes* represent the 25th and 75th percentiles, respectively. The *whiskers* are the 25th and 75th percentile ± (1.53 × interquartile range). ● represent outliers.

significant fibrosis (Fig. 3). A score <0.5 was observed in the remaining 62 (60%) patients, which excluded advanced fibrosis (F3 and F4) with a sensitivity of 88% (81.1%–93.9%) and a specificity of 74% (65.3%–82.2%). A cutoff point of 0.84 yielded a sensitivity of 71% (61.8%–79.4%) and a specificity of 89% (82.4%–94.6%) for predicting cirrhosis (F4).

HEPASCORE AND NECRO-INFLAMMATORY ACTIVITY

The Hepascore model was accurate in excluding moderate to severe necro-inflammatory activity (A2 and A3), providing an AUC of 0.707 (95% CI, 0.579–0.835) in the training set, with 59% (50.2%–68.0%) sensitivity and 73% (64.6%–80.7%) specificity.

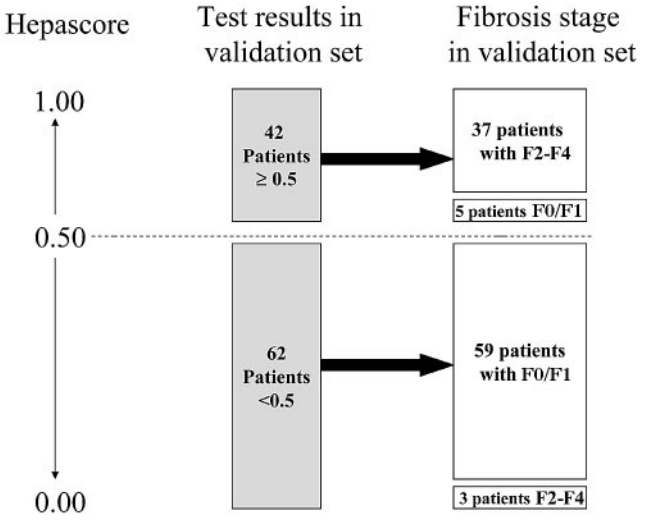

Fig. 3. Application of Hepascore fibrosis model to the validation set (n = 104).

Discussion

After assessing a set of 10 potential biochemical markers of fibrosis, we developed a predictive model (Hepascore) consisting of age, sex, and 4 serum markers: bilirubin, GGT, α_2-macroglobulin, and hyaluronic acid. The Hepascore accurately predicted different degrees of fibrosis among patients with chronic hepatitis C infection, and its performance was confirmed in an independent validation set of patients from separate institutions. The Hepascore provided information for all patients: a score ≥0.5 was 89%–92% specific for the presence of significant fibrosis (METAVIR ≥ F2); and a score <0.5 was 88%–95% sensitive for the absence of advanced fibrosis (METAVIR ≥ F3). Thus, in our 2 cohorts, a Hepascore ≥0.5 provided high PPVs (87% and 88%) for the presence of significant fibrosis, a Hepascore <0.5 provided NPVs of 95% and 98% for advanced fibrosis, and a Hepascore <0.84 provided NPVs of 94% and 98% for cirrhosis. It should be noted, however, that predictive values of a diagnostic test vary according to the underlying prevalence of the condition. Therefore, because treatment is generally recommended when significant fibrosis is present *(8)*, patients with a Hepascore ≥0.5 may be considered for antiviral therapy without the requirement for liver biopsy. In addition, the exclusion of advanced fibrosis among patients who have a Hepascore <0.5 may be particularly useful in providing prognostic information for patients who are reluctant to undergo biopsy or among elderly patients who are unlikely to develop liver-related morbidity or mortality in the absence of advanced fibrosis *(3)*. Finally, a score >0.84 is 84%–89% specific for the presence of cirrhosis. This may be useful to avoid liver biopsy in patients in whom occult cirrhosis is suspected or to guide management decisions regarding variceal and cancer screening and patient follow-up *(6)*.

Bilirubin, GGT, hyaluronic acid, and α_2-macroglobulin are rational candidates to provide useful information in determining liver fibrosis stage. As fibrosis progresses, bilirubin increases as a result of reduced hepatic excretion and less enterohepatic circulation attributable to portal systemic shunting *(23)*. GGT has previously been found to be correlated with liver fibrosis among patients infected with hepatitis B and C *(15, 19, 24)*. Liver injury increases hyaluronic acid production by hepatic stellate cells and decreases its clearance by sinusoidal endothelial cells *(25)*. α_2-Macroglobulin is a protease inhibitor whose concentrations increase with stellate cell activation and liver fibrosis *(21)*.

Many published studies have also developed panels of serum markers applicable to fibrosis prediction in hepatitis C–infected patients *(13, 15, 16, 19, 26, 27)*; however, our study has several unique features. By recruiting all eligible consecutive patients undergoing liver biopsy, we were able to avoid potential selection bias resulting from recruiting patients enrolled in treatment trials. In addition, our model was robust and remained accurate with an AUC of 0.82 for the prediction of significant fibrosis

when validated in a different population of patients with a different distribution of fibrosis. In contrast, the accuracy of other predictive models tends to decrease when applied to other populations.

The Fibrotest, a predictive model developed in France, had a reported AUC of 0.83–0.85, although predictive values for the 5-marker index were not described *(19)*. When examined in different populations, the AUC varied appreciably, from 0.73 to 0.85 *(28, 29)*. We were able to directly compare the accuracy of the Hepascore and the Fibrotest in the training set, but unfortunately, a lack of sera prevented a similar comparison in the validation group. The Hepascore provided a higher (although not significantly) AUC for the detection of significant and advanced fibrosis in the training set. An obvious advantage of the Hepascore is that it is published and freely available.

Another novel model (fibrosis probability index) incorporated measures of insulin resistance as well as age, total cholesterol, AST, and past alcohol intake. This model was accurate in predicting significant fibrosis with an AUC of 0.84 in the index patients, although this decreased to 0.77 when examined in the validation set *(16)*. The Hepascore maintained its accuracy in a different patient population, with an AUC of 0.82. Wai et al. *(13)* proposed a simple and elegant model of AST-to-platelet ratio index (APRI), which predicted bridging fibrosis as determined by the Ishak scoring system, with an AUC of 0.80–0.88. We found the AUC for advanced fibrosis (portal fibrosis with many septae) to be 0.90–0.96 with our model. APRI had lower accuracy (AUC of 0.74) for determining more subtle grades of fibrosis (portal fibrosis with rare septae) compared with the Hepascore (AUC of 0.82–0.85) *(30, 31)*. Another model, developed by Forns et al. *(15)*, included the routinely measured variables of GGT, cholesterol, platelet count, and prothrombin time in combination with age. This model could be applied to approximately one third of their patients and had an AUC of 0.81–0.86 for predicting significant fibrosis *(16)*. In contrast to the Hepascore, no information regarding advanced fibrosis or cirrhosis is provided by this model. A model recently published by Patel et al. *(26)* also incorporates hyaluronic acid and α_2-macroglobulin. This commercially available model had modest predictive values between 71% and 79% and thus may not be accurate enough to guide clinical decision-making. Similarly, an algorithm incorporating hyaluronic acid, TIMP-1, and N-terminal of propeptide of type III collagen, examined in a large number of patients with chronic liver disease, had an AUC of 0.78 for significant fibrosis from all etiologies and an AUC of 0.77 when limited to patients with hepatitis C *(27)*.

Although the Hepascore was robust during testing in another population, further validation in community-based patients is required before it can be applied outside of tertiary referral centers. We have published the model defining Hepascore to allow other investigators to obtain further validation data. Longitudinal studies are also necessary to determine whether the model is responsive to fibrosis change in the same individual over time. In addition, the number of patients with cirrhosis in our study was relatively small, limiting conclusions regarding the accuracy of the test in this population.

It should be noted that although the mean (SD) Hepascore values were significantly higher for patients with F2 fibrosis compared with those with F1 [0.49 (0.29) vs 0.28 (0.18), respectively; $P < 0.001$], there was considerable overlap between these categories (Fig. 2). This may in part reflect variability in pathologist interpretation, with poor interobserver agreement previously noted in the scoring of these stages *(32)*. In addition, the METAVIR staging system may not reflect a linear increase in fibrosis. In particular, the increase in the degree of fibrosis between F1 (enlarged portal tract) and F2 (enlarged portal tract with rare septae) may not be as great as the increase between F2 and F3 (enlarged portal tract with numerous septae). Indeed, in early-stage disease, there is poor correlation between degree of liver fibrosis as detected by digital image analysis and staging by a pathologist *(33)*. Because serum markers are likely to reflect the quantity of fibrotic matrix/tissue, they may correlate better with fibrosis as detected by image analysis than stage as determined by a pathologist.

Of the 4 markers, GGT and bilirubin are measured routinely. α_2-Macroglobulin is available to any laboratory with a nephelometer, and a hyaluronic acid assay is available commercially (Corgenix Inc.) and requires only a microplate colorimetric reader. It is therefore less costly and more convenient to perform these assays than a liver biopsy.

In conclusion, the Hepascore—a combination of bilirubin, GGT, hyaluronic acid, and α_2-macroglobulin together with age and sex—was accurate and reliable in predicting different stages of fibrosis among hepatitis C patients. The entire range of scores allows accurate estimation of particular fibrosis stages and provides clinically relevant information for hepatitis C–infected patients.

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