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## The Enhanced Liver Fibrosis (ELF) Blood Test

# Literature Compendium

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#### Siemens Healthineers Headquarters

Siemens Healthcare GmbH  
Henkestr. 127  
91052 Erlangen, Germany  
Phone: +49 9131 84-0  
[siemens-healthineers.com](https://www.siemens-healthineers.com)

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Siemens Healthcare Diagnostics Inc.  
Laboratory Diagnostics  
511 Benedict Avenue  
Tarrytown, NY 10591-5005  
USA  
Phone: +1 914-631-8000

## Introduction

The Enhanced Liver Fibrosis (ELF™) test\* is a noninvasive blood (serum) lab test designed to assess levels of three major components directly involved in liver matrix metabolism: hyaluronic acid (HA), procollagen III amino-terminal peptide (PIIINP), and tissue inhibitor of matrix metalloproteinase 1 (TIMP-1). The analytes are automatically measured, and the software calculates and reports a unitless numeric score. Increasing ELF scores are linked to both biopsy-proven fibrosis and prognosis for clinically significant outcomes (Figure 1).<sup>1</sup> The ELF score has been well-validated against biopsy-proven fibrosis across a range of chronic liver diseases (CLD) in both adult and pediatric populations.<sup>1-3</sup> The ELF test is serum-based and is available on the following immunoassay laboratory instruments: Atellica IM® Analyzers and ADVIA Centaur® Systems, broadly available worldwide in many labs offering routine diagnostic testing.

CLD is a leading cause of death worldwide, with disease burden rising significantly in many countries.<sup>4</sup> Common etiologies include nonalcoholic fatty liver disease (NAFLD), alcoholic liver disease (ALD), and viral hepatitis (VH). Disease progression is strongly linked to liver fibrosis.<sup>5-7</sup> Assessment of fibrosis has traditionally relied on biopsy and staging

systems used to define mild, moderate, or severe fibrosis or cirrhosis. Increasing recognition of the hazards and limitations of biopsy and histological staging has fueled pursuit of alternative methods. Advantages of the ELF test include ease of sample collection, automated analysis, reproducibility, and a near-linear correlation between ELF score thresholds and histological staging.<sup>1</sup> The ELF test is also highly prognostic and has been shown to outperform both simple markers and biopsy for outcomes.<sup>8,9</sup>

Liver fibrosis is biochemically complex but orchestrated primarily by activated hepatic stellate cells (HSCs). Activated HSCs produce components of the extracellular matrix (ECM). The ECM includes an array of proteins involved in scar formation, including fibronectin, laminin, collagens, hyaluronic acid (HA), and proteoglycans. Collagen types I, III, IV, and V are prominently expressed within the liver.<sup>10</sup> HA is an essential component of the ECM and is produced primarily by HSC.<sup>11</sup> The accumulation of deposited ECM progressively replaces the normal liver parenchyma, producing damage and scar tissue and ultimately disrupting hepatic architecture and function.

Fibrosis of the liver is a largely bidirectional process.<sup>5,12</sup> Both fibrosis and repair mechanisms have been linked to ECM-related pathways. Regression and repair are associated with upregulation of matrix metalloproteinases (MMPs), which are a family of zinc-dependent endopeptidases capable of degrading ECM deposition and therefore central to healing. Levels of MMPs are subject to inhibition by tissue inhibitors of metalloproteinases (TIMPs), a family of at least four proteins (TIMP 1–4) that bind MMPs. TIMP-1 overexpression hinders degradation and clearance of the fibrotic matrix, leading to increased levels of interstitial ECM and progressive fibrosis.<sup>13,14</sup> Additionally, low levels of TIMP-1 may promote hepatic stellate cell apoptosis.<sup>12</sup> By testing for direct markers associated with both ECM deposition and repair, the ELF test provides a direct measure for the assessment of fibrotic activity.

## Conclusion

The three direct markers of the ELF test provide complementary information, and the combined score outperforms both the individual markers and simple scores such as APRI or FIB-4.<sup>2,9</sup> The performance of the ELF test for liver fibrosis has been well-established in the scientific literature, and ease of testing and interpretation support routine clinical use as an alternative to invasive biopsy. This compendium highlights a small subset of the extensive number of ELF publications and serves as an introduction to the clinical utility of the ELF test.

Severity assessment (against biopsy-proven fibrosis) with the ELF blood test: the ELF scoring system<sup>1,15</sup>



≥9.8 associated with high risk of significant fibrosis.

\*The ELF test is not available for sale in the U.S. The ELF Testing Service is available from Siemens Healthcare Laboratory, a CLIA-certified laboratory located in Berkeley, CA.

†Not included in the Siemens Healthineers ADVIA Centaur® ELF test IFU.

## Glossary of Terms

ALD:	Alcoholic liver disease
APRI:	AST-to-platelet ratio index
AUROC:	Area under the receiver operating characteristic curve
CLD:	Chronic liver disease
ECM:	Extracellular matrix
ELF:	Enhanced Liver Fibrosis test
FIB-4:	Fibrosis-4 index
HA:	Hyaluronic acid
HBV:	Hepatitis B virus
HCC:	Hepatocellular carcinoma
HCV:	Hepatitis C virus
LFT:	Liver function test
LRE:	Liver-related event
MMP:	Matrix metalloproteinase
NAFLD:	Nonalcoholic fatty liver disease
NASH:	Nonalcoholic steatohepatitis
NIT:	Noninvasive test
PIIINP:	Procollagen III amino-terminal peptide
ROC:	Receiver operating characteristic curve
TIMP-1:	Tissue inhibitor of matrix metalloproteinase 1
VH:	Viral hepatitis

# The ELF Test Compared to Biopsy-proven Fibrosis

## Serum Markers Detect the Presence of Liver Fibrosis: A Cohort Study

Rosenberg WM, et al. Gastro. 2004;127:1704-13.

### Objective

Investigate a panel of matrix-constituent biomarkers and mediators of fibrosis present in serum in patients with CLD and biopsy-proven fibrosis.

### Methods

- A multicenter study with CLD patients from diverse etiologies, including VH, ALD, and NAFLD, who were scheduled to undergo biopsy and had elevated LFTs.
- Paired serum samples were collected at the time of biopsy. Biopsy samples were analyzed by a local pathologist and then reanalyzed by a central pathologist blinded to the initial results using both the Scheuer and Ishak scoring systems. A random subset was further analyzed by two independent expert pathologists using the same descriptors as the central pathologist.
- Nine serum biomarkers associated with liver fibrosis were selected and run as immunoassays (IAs) using an automated IA platform. Markers were evaluated alone and in conjunction. Algorithms for multiple combinations of markers were developed and assessed using a random subset of patients. An optimal algorithm was then selected and validated in the remaining patient cohort and performance compared to histologic fibrosis staging.
- The ability of the algorithm to detect significant levels of biopsy-proven fibrosis was assessed using three designators: significant fibrosis (upper three stages of scoring defined by the Scheuer system), histological distribution of the algorithm scores, and detection of cirrhosis.

### Results

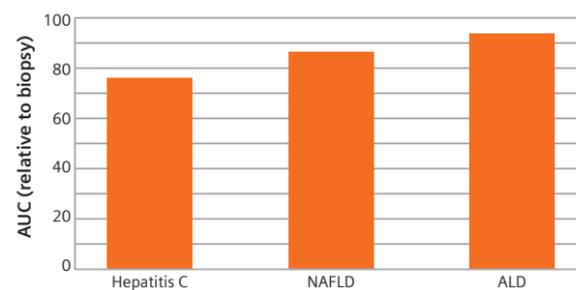
- Optimal performance results were found with algorithms that incorporated HA, PIIINP, and TIMP-1 (though other serum markers of fibrosis also performed well). Additional serum markers and hematological indices including platelet count and prothrombin time did not improve performance. Data for the discriminant performance for HA, PIIINP, and TIMP-1 are shown.
- ROC analysis showed good performance across the range of CLD, though AUROC varied by etiology. AUROC for the three most common forms of CLD is shown.

- Application of differing test thresholds showed the ability to exclude significant fibrosis with sensitivity in excess of 90% or to identify significant fibrosis with specificity in excess of 90%.

### Significance

- Though widely accepted, histologic staging provides discrete categorical values but does not describe a quantifiable variable such as the amount of fibrosis. Linearity in biopsy stages is often assumed, though it is widely recognized that stage 4 does not necessarily represent twice as much fibrosis as stage 2. Algorithm scores may better identify fibrosis as a continuous and quantifiable variable.
- Biopsy is invasive, carries risk, has sampling error, and may be suboptimal to detect and quantify significant fibrosis. Serum markers are less invasive, reproducible, and may be automated. Ease of sampling could support assessment over time and may be more acceptable to patients versus repeat biopsy.
- The strong correlation to biopsy-proven fibrosis suggests serum markers could be a useful alternative or addition to biopsy. The use of different thresholds offers significant clinical utility aiding a rule-out or rule-in for significant fibrosis.

Percent Area under the Curve (AUC) of ELF Test Relative to Biopsy



### Conclusion

"We have established that an algorithm combining serum markers of liver fibrosis can be used in a wide range of chronic liver diseases to identify patients who have little or no fibrosis, distinguishing them from those with clinically significant hepatic fibrosis."

## The Performance of Enhanced Liver Fibrosis (ELF) Test for the Staging of Liver Fibrosis: A Meta-analysis

Xie Q. PLoS One. 2014;9(4):e92772. doi: 10.1371.

### Objective

Perform a meta-analysis from the scientific literature to evaluate the diagnostic accuracy of the ELF test when using histopathology (biopsy) as a reference standard.

### Methods

- A literature search was performed using the terms cirrhosis, liver fibrosis, and enhanced liver fibrosis test or ELF test and limited to studies in humans and abstracts in English in 2013.
- Nineteen papers were identified, but 10 were excluded due to one or more of the following: unavailability of data on TP, FP, TN, or FN or missing data on histology, grading scores, or diagnostic accuracy. Nine papers were included in the analysis and included both European and Asian patient cohorts with a range of CLD.
- Statistical analysis included using a bivariate binomial model. This model assumed a binomial distribution in the number of TP and TN patient results and allowed the inclusion of covariates and random effects. The inherent association between sensitivity and specificity was modeled in a bivariate normal distribution by assuming random effects. Summary receiver operating characteristic (SROC) curves were constructed to express the test parameter results as the diagnostic odds ratio and to assess diagnostic threshold bias as a cause of between-study heterogeneity.

### Results

- AUROC analysis for the diagnostic threshold (cutoff) was 0.88 for the detection of significant fibrosis, 0.87 for severe fibrosis, and 0.88 for cirrhosis. Evidence supporting the diagnostic threshold (cutoff) bias as a source of heterogeneity was identified.
- Pooled sensitivity for the performance of the ELF test in the assessment of significant fibrosis was 83% and pooled specificity 73%. For the prediction of severe fibrosis, the pooled sensitivity value was 78% and pooled specificity 76%. Pooled sensitivity for the prediction of cirrhosis was 80% and pooled specificity 71%.

- Additional statistical analysis was performed on all three groupings for the pooled positive and negative odds ratios and the summary diagnostic odds ratio.

### Significance

- The good diagnostic accuracy of the ELF test was confirmed across studies for the detection of significant or severe fibrosis or cirrhosis, and strengths such as automation, reproducibility, and less invasiveness compared to biopsy were noted.
- In the three subgroups of this meta-analysis (significant fibrosis, severe fibrosis, and cirrhosis), the statistical analysis indicated at least 74% of patients could reasonably avoid biopsy.
- The results, coupled with the ELF test's reproducibility, support its use in clinical practice as a predictor of histological fibrosis.

Study	Disease Spectrum
Guha IN, et al.	CHC, CHB, PBC
Nobili V, et al.	NAFLD
Friedrich-Rust M, et al.	CHC, CLD, PBC
Parkes J, et al.	CHC
Kim BL, et al.	CHB
Wahl K, et al.	VH, AIH, Wilson's diseases, NAFLD, others
Guechot J, et al.	CHB, ALD, CHC, others
Lichtinghagen R, et al.	CHC
Wong GL.	CHB

### Conclusion

"The ELF test shows good performance and considerable diagnostic value for the prediction of histological fibrosis stage."

# The ELF Test as a Prognostic Tool

## Derivation and Performance of Standardized Enhanced Liver Fibrosis (ELF) Test Thresholds for the Detection and Prognosis of Liver Fibrosis

Day J, et al. J Appl Lab Med. 2019 Mar;3:815-26.

### Objective

Identify standardized thresholds for the ELF test for the detection of fibrosis severity and prognosis using data from a large prospective study.

### Methods

- Expert hepatologists were interviewed and asked to define clinically acceptable levels of test performance for the assessment of fibrosis in patients with CLD. Specifically, they were asked what proportion of patients with severe fibrosis or cirrhosis they would be willing to accept as misassigned for moderate or mild fibrosis. Additionally, the hepatologists also requested a highly specific value for the identification of cirrhosis.
- After identifying the sensitivities and specificities preferred by the clinical experts, data from the original ELF test patient cohort was analyzed for thresholds that would conform to the requested performance parameters.
- Linear regression analysis was used to generate and fit regression curves and straight lines for the ELF test relative to both Ishak and Scheuer staging (fibrosis assessment).
- Corresponding cutpoints identified for assessment were then investigated relative to outcomes. Thresholds identified for histological correlation were recalculated for prognosis. Use of the same thresholds for both fibrosis assessment and prognosis were explored.
- The prognostic performance of the ELF test at these cutpoints was assessed in the prediction of all-cause mortality or any liver-related event (LRE) postrecruitment.

### Results

- Clinician consensus for acceptable test sensitivity in low-risk patients was 80–85%, with the view that these patients could undergo repeat testing to aid assessment of progression. An 80% sensitivity was opted for in the detection of cirrhosis. An additional threshold that would identify cirrhosis with greater specificity and minimize inappropriate referral of patients with mild or moderate fibrosis was requested by the clinicians and identified as  $\leq 5\%$  (i.e., high specificity to minimize referral of patients without advanced disease).
- A near-linear correlation between ELF scores and biopsy staging was observed.
- The ability of the ELF test to identify the different binary categorizations from the biopsy results was calculated. Sensitivities and specificities were identified across the reporting range for the ELF test and showed a positive correlation between a rising ELF score and biopsy-proven fibrosis.
- Straight-line-fit analysis across the curves revealed an association with a change in ELF value and a change in histological staging relative to F1–F3 (Ishak) or S2–S5 (Scheuer).
- Evaluation of the prognostic performance relative to the initial ELF score was assessed up to 7 years for LRE in patients grouped by low to high ELF score threshold values. LREs and relative risk of death were significantly elevated in patients with ELF scores  $>9.8$ . Hazard ratios for patients with ELF scores  $\geq 11.3$  for LREs more than doubled compared to ELF scores falling between 9.80 and 11.29.

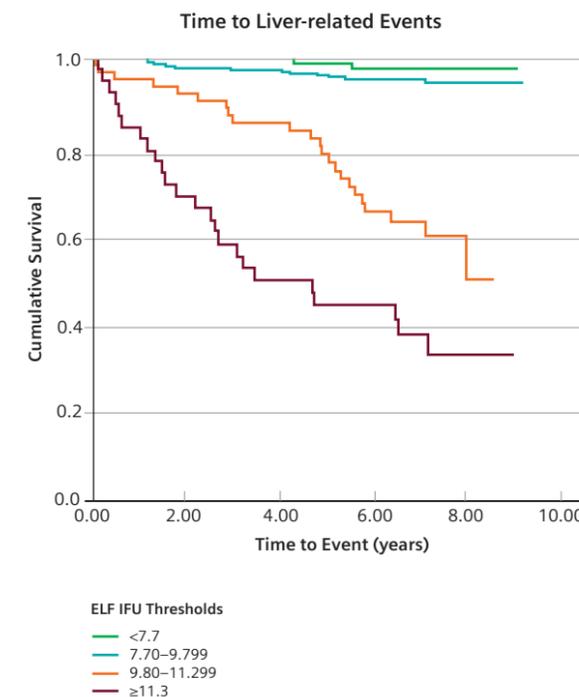
### Significance

- ELF thresholds correlated to biopsy-proven fibrosis can be useful as a surrogate measure for the identification of significant fibrosis.
- Three ELF score thresholds corresponding to values for fibrosis assessment were also prognostic. Use of the ELF score identified four categories of risk for liver-related outcomes, supporting clinical management and decision making.
- Changes in ELF score within a threshold range can be clinically meaningful, supporting the value of the ELF score as a continuous variable versus simple correlation to a discrete histological stage.
- A highly specific cutpoint for cirrhosis was identified.

### Conclusion

“Using data derived from a large prospective study and the opinions of expert hepatologists, we have identified standard thresholds for the Enhanced Liver Fibrosis test. These thresholds can be used to detect liver fibrosis of different degrees of severity and determine the prognosis of chronic liver disease.”

“These thresholds should prove useful in both interpreting and explaining test results and when considering the relationship of ELF score to Ishak stage in the context of monitoring.”



## The Enhanced Liver Fibrosis Score Is Associated with Clinical Outcomes and Disease Progression in Patients with Chronic Liver Disease

Irvine KM, et al. Liver Int. 2016;36:370-7.

### Objective

Investigate the ELF test as a predictor of liver-related outcomes or progression to fibrosis, and compare the performance of the ELF test to liver biopsy and to simple scores (APRI and FIB-4).

### Methods

- CLD patients with ELF scores and paired biopsies were followed for a median of 6.1 years for liver-related outcomes or disease progression.
- Prognostic performance of the ELF score was compared to simple scores (APRI and FIB-4).
- A change in ELF score relative to disease progression was investigated.

### Results

- Patients with ELF scores  $\geq 9.8$  had a significantly higher risk of a liver-related outcome than patients with ELF scores  $< 9.8$ .
- Simple scores performed significantly less well compared to ELF score for the prediction of LRE.
- A unit increase in ELF score was associated with greater than a doubling of risk for an LRE.
- More than half of patients without advanced fibrosis on biopsy at recruitment but with ELF scores  $\geq 9.8$  showed clear evidence of progression to advanced fibrosis within 6 years. In contrast, patients with ELF scores  $< 9.8$  at recruitment were unlikely to progress.

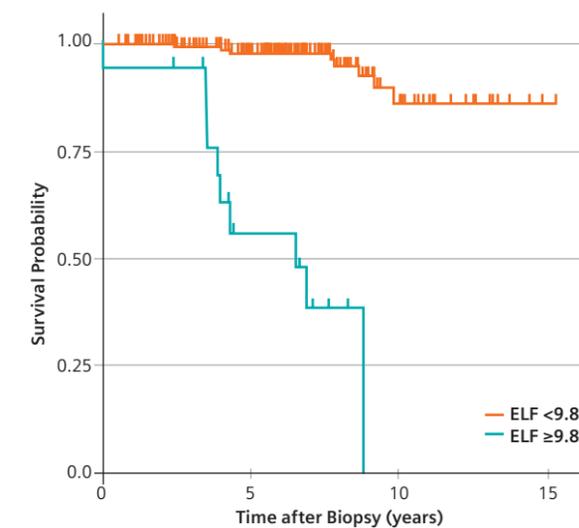
### Significance

- The ELF score is a clinically useful tool for patient risk stratification and outperforms biopsy as a predictor of disease progression.
- Compared to biopsy, ELF more readily supports the increasing clinical demand for identification and risk stratification of CLD patients.
- ELF is a more sensitive predictor of patients likely to experience advanced disease or an LRE than both biopsy and simple markers.
- A unit change in ELF score significantly increased risk of both disease progression and an LRE, supporting utility for comparing ELF test values over time.

### Conclusion

"The ELF score is a valuable tool for risk stratification of patients with chronic liver disease."

Survival of Patients without Advanced Fibrosis (biopsy) at Recruitment and ELF Scores  $< 9.8$  or  $> 9.8$



## Enhanced Liver Fibrosis Score as a Predictor of Hepatocellular Carcinoma

Loo WM, et al. Clin Chem. 2018;64:1404-5.

### Objective

Investigate the ELF test as a noninvasive test to identify patients at risk of developing hepatocellular carcinoma (HCC), especially patients with NAFLD.

### Methods

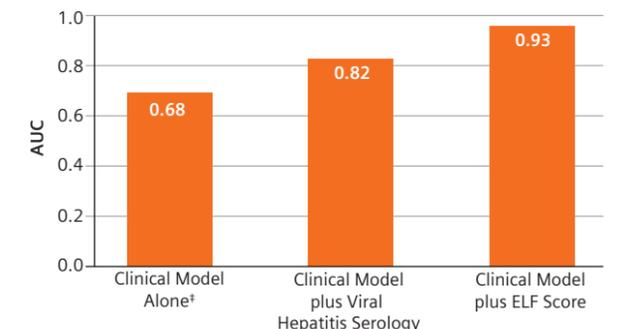
- A large population-based cohort of Chinese patients had serum samples collected and stored for an average of 14 years.
- Patients were followed for the development of HCC and matched for cases and controls.
- ELF scores were evaluated from the archived serum samples in a subset of HCC patients.
- ROC analysis included models incorporating parameters such as body mass index (BMI), alcohol use, viral hepatitis, and ELF score.
- ROC analysis and the Youden index were used to derive the optimal ELF score for diagnostic discrimination between HCC cases and controls.
- Risk analysis was calculated in the various clinical models (including with and without viral hepatitis).

### Results

- A clinical parameters-only model ROC analysis that included age, sex, dialect group, BMI, alcohol consumption, and a history of diabetes yielded only a moderate AUC value.
- AUC improved with the addition of viral hepatitis serology data.
- Addition of the ELF score to the clinical model further improved the AUC to  $> 0.9$ .
- An ELF score  $\geq 9.89$  was identified as highly associated with increased risk for the development of HCC.
- Risk estimates for patients negative for viral hepatitis serology and with ELF scores  $\geq 9.89$  were substantially higher, suggesting increased robustness in predicting non-viral-related HCC.

### Significance

- ELF testing can be highly useful in aiding identification of patients more likely to progress to HCC.
- An ELF score  $\geq 9.89$  is substantially associated with risk of HCC in patients with risk factors for CLD, including patients with viral hepatitis.
- The increased robustness for predicting risk in patients without viral hepatitis has implications for use in targeted populations such as NAFLD patients.



†Age, sex, dialect group, body mass index (BMI), alcohol consumption, and history of diabetes.

### Conclusion

The study demonstrated the potential utility of the ELF score as a predictor of HCC with high diagnostic sensitivity, specificity, and accuracy.

# The ELF Test Compared to Imaging

## Evaluation of Transient Elastography (TE), Acoustic Radiation Force Impulse Imaging (ARFI), and Enhanced Liver Function (ELF) Score for Detection of Fibrosis in Morbidly Obese Patients

Karlas T, et al. PLoS One. 2015;10:e0141649.

### Objective

Evaluate the performance of the ELF test compared to transient elastography (FIBROSCAN) for the detection of fibrosis in bariatric patients.

### Methods

- Patients with an indication for bariatric surgery were evaluated for liver fat and hepatic volume using MR.
- Patients scheduled for bariatric surgery were placed on a low-energy diet prior to surgery, and measurements were obtained before and after the 2-week diet.
- TE (M and XL probe), ARFI, and the ELF test were performed at day -14 and day -1 and compared with intraoperative liver biopsies (NAS staging) performed on day 0.
- TE and ARFI cutoff values were applied from published studies to estimate the risk of significant fibrosis or cirrhosis.
- The diagnostic performances of TE, ARFI, and the ELF score for detection of significant liver fibrosis (>F2) were evaluated.
- The cutpoint for the ELF score for significant fibrosis was derived using a nonbariatric cohort consisting of patients with biopsy-proven NAFLD.

### Results

- Both the ELF test and ARFI had significantly higher rates of valid results compared to TE. No failures were reported for the ELF test.
- Patients with significant hepatocellular inflammation had higher median TE and ARFI values. In comparison, no impact on ELF scores was observed in patients with steatohepatitis.
- In contrast to other published data, both TE and ARFI showed an overestimation of fibrosis in many patients (compared to biopsy-proven fibrosis). The authors speculate that this could be associated with the higher average BMI in their study cohort as well as a high prevalence of nonalcoholic steatohepatitis (NASH).

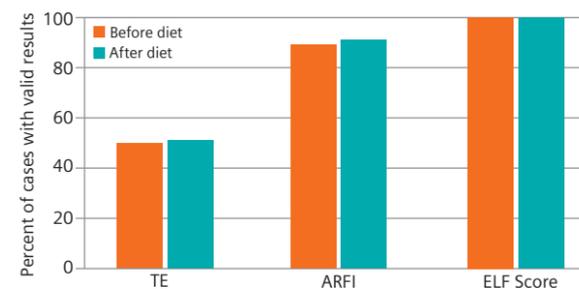
- The ELF score had a higher specificity for exclusion of fibrosis than either TE or ARFI.
- The ELF test cutoff derived from the nonbariatric control group correctly classified the majority of cases, including patients with advanced fibrosis.

### Significance

- Valid data acquisition using imaging for elastography in morbidly obese patients and patients with steatohepatitis can be challenging: Factors such as liver stiffness associated with high inflammation and probe-to-capsule distances affect performance.
- In contrast to the imaging modalities used in this study, the ELF test was not affected by liver stiffness, measuring depth, or steatohepatitis.
- The ELF test more accurately excluded fibrosis than did imaging, and it did not exhibit the failure rate observed for imaging (especially TE).
- Compared to both imaging modalities, the ELF test more accurately identified biopsy-proven significant fibrosis.

### Conclusion

"In bariatric patients, performance of TE and ARFI was poor and did not improve after weight loss. The ELF score correctly classified the majority of cases and should be further evaluated."



# Accuracy of the Enhanced Liver Fibrosis Test versus FibroTest, Elastography, and Indirect Markers in Detection of Advanced Fibrosis in Patients with Alcoholic Liver Disease

Thiele M, et al. Gastroenterology. 2018;154:1369-79.

### Objective

Compare the accuracy of the ELF test, FIBROTEST (FT), six indirect markers of fibrosis, and liver stiffness assessed using both transient elastography (TE) and 2-dimensional shear-wave elastography (2D-SWE) in the detection of advanced (>F3) liver fibrosis.

### Methods

- Patients with a significant risk of fibrogenic ALD were recruited from both primary and secondary care clinics.
- The ELF test, biopsy, FT, indirect markers, TE, and 2D-SWE were performed on the recruited patients (exclusion criteria were applied prior to testing).
- Imaging tests employed a highly experienced operator to maximize success of data acquisition.
- An ELF test value of 10.5 was used (adapted from the National Institute of Health and Care Excellence Guidelines<sup>16</sup> for the detection of advanced fibrosis).
- Performance of the noninvasive tests (NITs) was compared to biopsy and evaluated for the detection of significant fibrosis.

### Results

- The ELF score and indirect serum indices were successfully measured in all patients. FT failed in a small number of patients (associated with low haptoglobin or high apolipoprotein A1). In comparison, higher rates of either unreliable or invalid results were observed for the imaging modalities than for blood-based testing.
- The ELF score outperformed all six indirect serum indices of fibrosis, but no statistically different performance was noted for the ELF score compared to FT. No added diagnostic value was observed by combining the ELF score with either FT or routine serum indices.
- Neither TE nor 2D-SWE were superior to either the ELF test or FT in the intention to diagnose patients with and without advanced fibrosis (though TE and 2D-SWE did differ from the ELF test and FT in the per-protocol analysis).
- Elastography was superior to serum markers in the detection of severe fibrosis on a per-protocol basis only when failures and unreliable results were excluded.

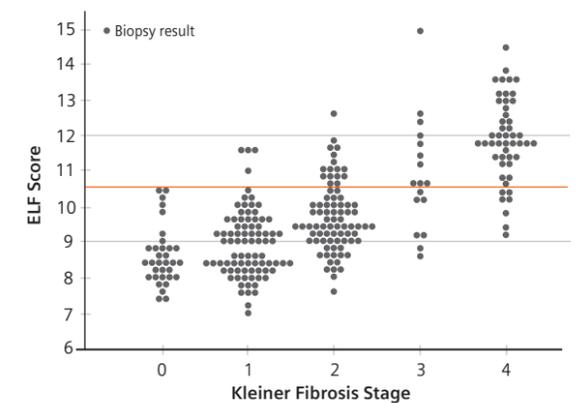
- An analysis of patients in the primary care cohort showed that initial testing with simple markers (Forn's index) excluded nearly half of the patients for significant fibrosis. The remaining patients were tested with the ELF test and referred if the ELF score was  $\geq 10.5$ . This approach prevented unnecessary referral in the majority of primary care patients while identifying patients at greater risk.

### Significance

- The ELF test was found to be an efficient diagnostic tool to assess patients with alcohol overuse in both primary and secondary healthcare settings.
- Use of a 10.5 cutoff value was not statistically different from FIBROTEST or elastography in intention-to-diagnose, suggesting the ELF test could be used in lieu of alternative NIT methodologies.
- Though imaging slightly outperformed blood-based testing in the detection of severe fibrosis, factors such as failure rate and accessibility can limit utility of imaging modalities in both the primary and secondary care settings.
- Use of simple markers followed by ELF testing in patients who cannot be excluded with simple markers could be readily incorporated in both primary and secondary care settings and could greatly minimize referral rates.

### Conclusion

"In a prospective, direct comparison of tests, the ELF test and FIBROTEST identified advanced liver fibrosis in alcoholic patients from primary and secondary care with high diagnostic accuracy (AUROC values of 0.90 or higher using biopsy as reference)."



# Health Economics and the ELF Test: Pathway to Improve Clinical Identification and Reduce Healthcare Burden in Both Adults and Children with NAFLD

## Prospective Evaluation of a Primary Care Referral Pathway for Patients with Non-alcoholic Fatty Liver Disease

Srivastava A, et al. J Hepatol. 2019 Apr 6. pii: S0168-8278(19)30227-2. doi: 10.1016/j.jhep.2019.03.033.

### Objective

Develop a pathway for the risk stratification of patients with NAFLD (initially evaluated in a primary care setting) using blood tests to both improve detection of advanced fibrosis/cirrhosis for specialist referral and limit unnecessary referrals.

### Methods

- A large prospective longitudinal cohort of NAFLD patients seen in a primary care (community) setting were either assigned for assessment of advanced fibrosis using a 2-step screening approach ("NAFLD Pathway") or managed using standard of care (SOC).
- Patients in the NAFLD Pathway were assessed for risk of significant fibrosis ( $\geq$ Kleiner F3) using blood-based testing (FIB-4 and the ELF test).
- Initial testing used FIB-4 for the rule-in or rule-out of advanced fibrosis, followed by assessment with the ELF test for patients with indeterminate FIB-4 values.
- Patients at high risk by either FIB-4 or the ELF test were referred for secondary assessment and care by a hepatology specialist.
- Fibrosis determination for the purpose of appropriate/inappropriate referral included history and physical, blood tests, imaging for liver elastography, and liver histology when available.
- NAFLD Pathway performance was evaluated after 2 years.
- Before-and-after analysis was performed and results compared to controls for the increased detection of advanced fibrosis/cirrhosis and for any reduction in unnecessary referrals.

### Results

- Most of the NAFLD patients had FIB-4 values of  $<1.30$  and remained in primary care. Management included annual liver function testing and reassessment for advanced fibrosis after 3 to 5 years.
- Patients with FIB-4  $>3.25$  (a small minority) were stratified as high risk and recommended for referral to a specialist.
- Patients with indeterminate FIB-4 values ( $\geq 1.30$  and  $<3.25$ ) were reflexed for testing with the ELF test.
- Patients with indeterminate FIB-4 and ELF scores of  $\geq 9.5$  were referred for assessment by a specialist.
- The NAFLD Pathway was significantly superior to SOC at selecting cases of advanced fibrosis and cirrhosis.
- A greater than 80% reduction in unnecessary referrals was observed for patients in the NAFLD Pathway.
- Detection of cirrhosis in patients referred from the NAFLD Pathway improved nearly 3-fold compared to that in patients receiving SOC.

### Significance

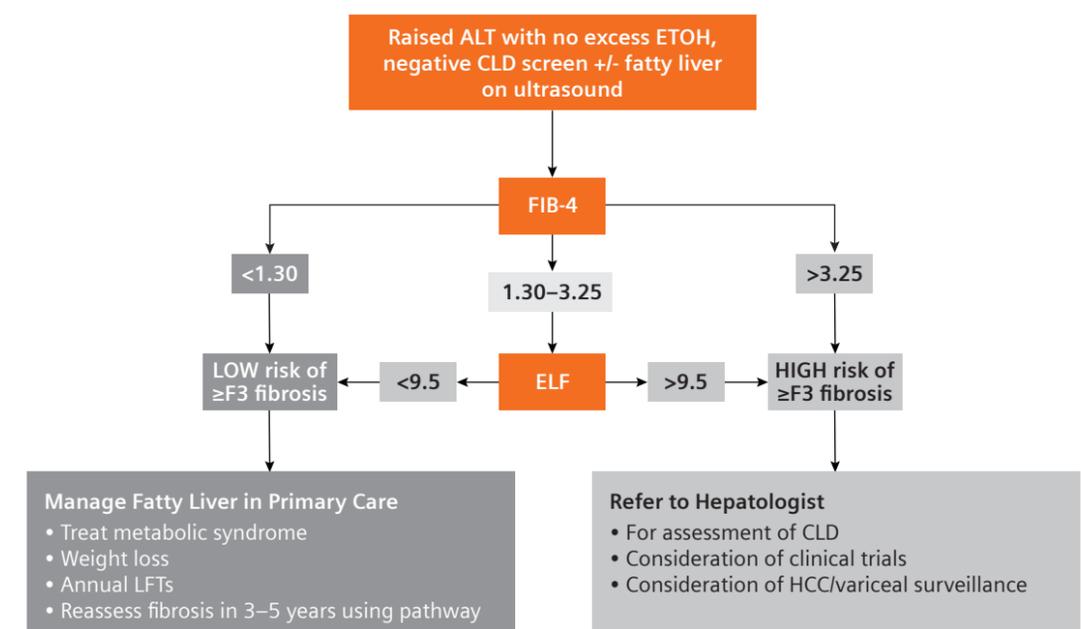
- NAFLD patients are typically seen initially in a primary care setting where most can be appropriately managed. The challenge is to accurately risk-stratify patients for the subset that could benefit from referral. Currently, limited guidance and tools exist for the primary care physician to enable appropriate referral. SOC produces a high rate of unnecessary referrals as well as missed cases of advanced fibrosis and cirrhosis, resulting in increased healthcare burden and unnecessary cost.
- Performing simple blood tests in a two-step approach allows a significant improvement in both detection of advanced disease and identification of low-risk patients who can be safely retained. It also provides a definitive pathway to referral for the physician.

- NAFLD prevalence is approaching epidemic levels in many countries. Unnecessary referrals could quickly overwhelm available specialist resources. Implementation of a NAFLD Pathway using simple blood tests with proven efficacy could increase clinical confidence in the decision to refer or retain patients within a primary care setting.
- Increased detection of cirrhosis could enable better management and monitoring for a reduction in liver-related events. It remains to be seen whether the use of the NAFLD Pathway produces a reduction in the incidence and complications of NAFLD cirrhosis.

### Conclusion

"The reduction in referrals to secondary care reduces strain on services that are confronting a rising prevalence of obesity and NAFLD as well as benefitting patients' experiences by avoiding unnecessary clinic appointments and investigations."

The Camden & Islington NAFLD Pathway



# Guidelines on the Management of Abnormal Liver Blood Tests

Newsome PN, et al. Gut. 2018;67:6-19.

## Objective

Update to guidelines written by the British Society of Gastroenterology (BSG) under the auspices of the liver section of the BSG for the management of abnormal liver blood tests, including NAFLD and ALD patients.

## Methods

- The recommendation for screening blood tests for potential liver disease included bilirubin, albumin, alanine aminotransferase (ALT), alkaline phosphatase (ALP), aspartate aminotransferase (AST), and  $\gamma$ -glutamyltransferase (GGT), together with a full blood count if not already performed within the previous 12 months.
- The authors recognized that hepatobiliary enzymes in isolation can convey information on ongoing liver injury while bilirubin, albumin, international normalized ratio (INR), and platelet count convey information on liver function (with platelets reflective of the level of fibrosis).
- While elevated AST/ALT levels could be predictive of liver disease, >95% of patients with an elevated level do not develop significant liver disease within 5 years of testing. The authors recognized the desirability of a pathway with greater specificity.
- Focusing on NAFLD separately, the authors developed a pathway in which initial screening consists of either FIB-4 or NAFLD fibrosis score. FIB-4 values of  $\leq 1.3$  support a rule-out and FIB-4 of  $> 3.25$  a rule-in. Age-specific cutoffs for patients over 65 years old are identified. Patients with indeterminate values are reflexed for testing with either the ELF test or imaging for liver elasticity (both are equivalent options).
- Patients with ELF values of  $\leq 9.5$  should be managed in primary care, with repeat assessment suggested every 2 to 5 years (depending on risk). Patients with ELF values of  $> 9.5$  are recommended for referral to a hepatology clinic for management of advanced fibrosis.

## Results

- Initial testing depends on risk for liver disease. Risk factors identified include evidence of viral hepatitis, suspected chronic liver disease (CLD), conditions associated with the development of CLD (such as inflammatory bowel disease), family history, and the use of hepatotoxic drugs. Distinct pathways are identified for ALD and NAFLD, both of which recommend a quantitative measure of fibrosis following establishment of NAFLD or excessive alcohol use. The ELF test or liver elasticity imaging (FIBROSCAN or ARFI) are recommended as quantitative measures.
- Second-line testing with the ELF test or imaging achieved a level 2b, grade B recommendation, meaning supporting evidence in the scientific literature is available, either as extrapolations from level 1 studies or consistency through level 2 or 3 studies.
- Differences in testing between adult versus pediatric populations are reviewed. The authors express the opinion that noninvasive markers of fibrosis have not been sufficiently validated in children for recommendation.

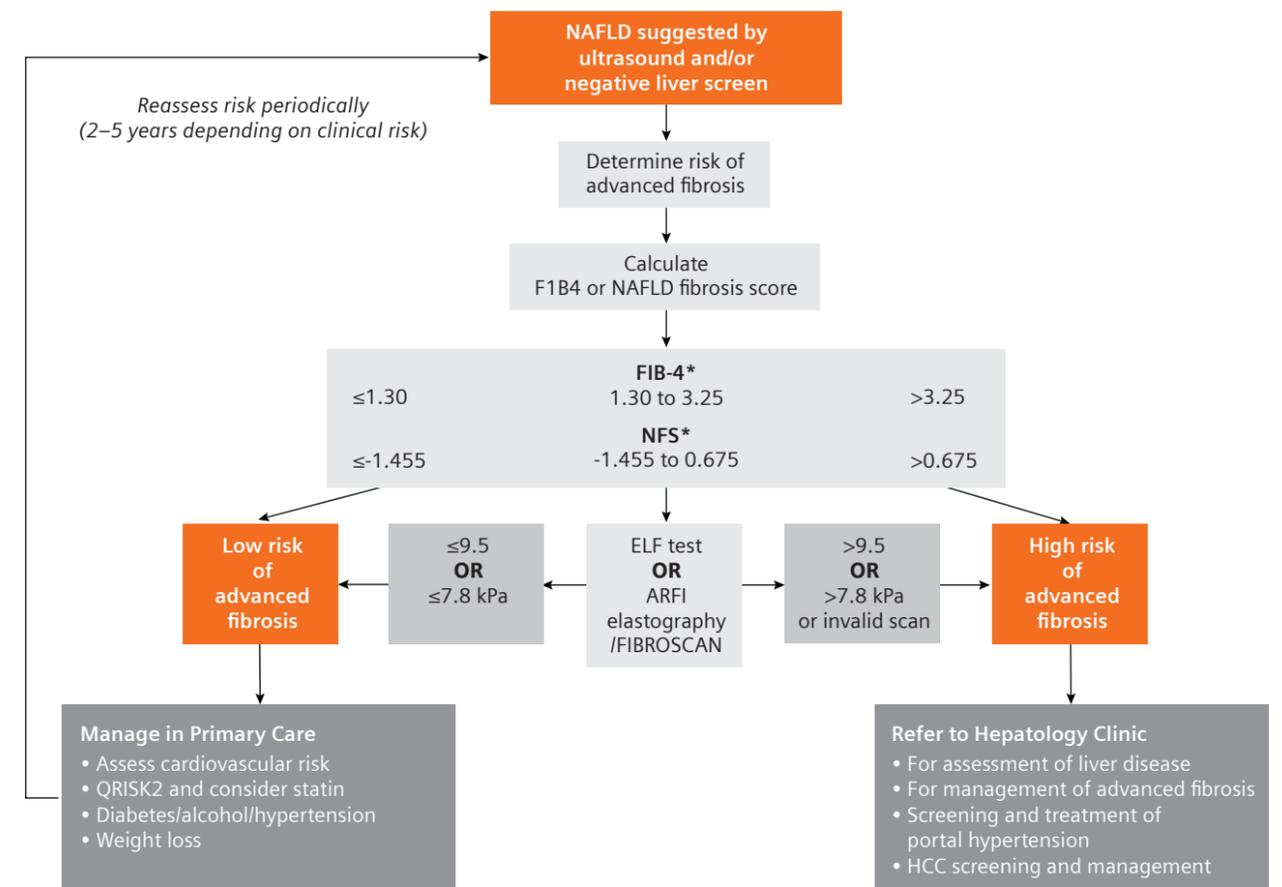
## Significance

- The use of a 2-step process for identifying patients at high risk of advanced liver fibrosis greatly improves specificity of CLD patient assessment. As both FIB-4 and the ELF test are blood-based tests, broad access to routine testing can be achieved.
- This pathway provides clear guidance for the management of NAFLD patients seen in a primary care setting where only a minority are likely to benefit from referral to a specialist. Risk assessment is critical both to improve patient outcomes and reduce unnecessary specialist patient burden.
- The FIB-4 and ELF test threshold values used are aligned with the recommendations published by Srivastava et al., providing additional confidence in the clinical cutpoints.

## Conclusion

“Those patients with indeterminate FIB-4 (1.3–3.25) or NFS scores (–1.455 to 0.675) should undergo further testing with a second-line test such as serum enhanced liver fibrosis (ELF).”

“Second-line testing requires a quantitative assessment of fibrosis with tests such as serum ELF measurements or Fibroscan/ARFI elastography.”



\*Higher cutoffs  $< 2.0$  and  $< 0.12$  should be used for patients aged over 65 years.

# Non-alcoholic Fatty Liver Disease: Assessment and Management

NICE guideline NG49. Methods, evidence and recommendations. July 2016.

## Objective

Define an algorithm for the assessment and monitoring of NAFLD in adults, children, and young people using noninvasive tests (NITs) to identify patients at high risk of advanced liver fibrosis.

## Methods

- NAFLD prevalence is estimated at 20%–30% in the UK, and 2%–3% of the population has NASH. As liver disease is typically silent until advanced cirrhosis, the authors recognized value for risk stratification and intervention in the subset of patients with progressive chronic liver disease (CLD).
- NAFLD should also be investigated in adults and children with type 2 diabetes or metabolic syndrome, owing to the increased incidence of CLD in these populations.
- The assessment pathway for patients with suspected NAFLD is currently unclear. Guidance is needed for patients in both primary and secondary care settings. As 80% of NAFLD patients have normal standard blood tests, alternative testing is needed.
- Diagnostic techniques for both identifying steatosis (at  $\geq 5\%$  and  $\geq 30\%$ ) and assessing fibrosis were included to identify high-risk NAFLD patients.
- Given the substantial number of NAFLD patients and the risks and limitations of biopsy, NITs for the assessment of liver fibrosis were explored as alternatives and testing algorithms developed for children and adults.
- Initially, questions were identified and full literature searches, critical appraisals, and evidence reviews completed. Questions focused on methods to identify NAFLD and NASH and which assessment tools are most accurate in identifying the severity or stage and for monitoring for advancing disease. Other questions focused on health economics and the clinical value and cost-effectiveness of intervention and treatment.
- Available NITs were ranked for performance (accuracy and cost-effectiveness), along with methods to initially identify NAFLD. Techniques included liver biopsy, MRI or MRS, ultrasound (presence or absence of steatosis only), the enhanced liver fibrosis (ELF) score, transient elastography, and the NAFLD fibrosis score, among others.

- Available evidence was assessed by an expert panel and draft guidance submitted for review. The Final Guideline, published following consultation on the draft, covers the assessment and management of NAFLD in adults, children, and young people.

- NIT performance parameters included accuracy in distinguishing NASH from NAFLD or simple steatosis (SS) as well as cases of advanced fibrosis. The algorithm could help identify the highest-risk patients for referral to a specialist.

## Results

- Techniques for identifying steatosis were numerically ranked for both performance and cost. Techniques were considered independently for the adult vs. pediatric populations.
- Testing for NAFLD was found to be cost-effective. Among the eight diagnostic tests compared for identifying NAFLD, the fatty liver index (FLI) ranked first for assessing diagnostic accuracy and cost, and ultrasound second.
- Of the 13 diagnostic testing strategies for the assessment (accuracy and cost) of significant fibrosis in NAFLD, the ELF test ranked number one.
- A cost-utility analysis that compared 17 strategies for testing adults with NAFLD for advanced fibrosis (with retesting every 2 years) found that the ELF test ranked first when compared to imaging and blood test options (both single and in combination).
- After assessing performance of both imaging and blood testing modalities, the ELF test was the recommended test in the algorithm for detection of advanced fibrosis in both adults and children with NAFLD. An ELF value of 10.51 was used for the high specificity.
- If the initial ELF value falls below 10.51, retesting with the ELF test is recommended every 3 years for adults and every 2 years for children and young people.

## Significance

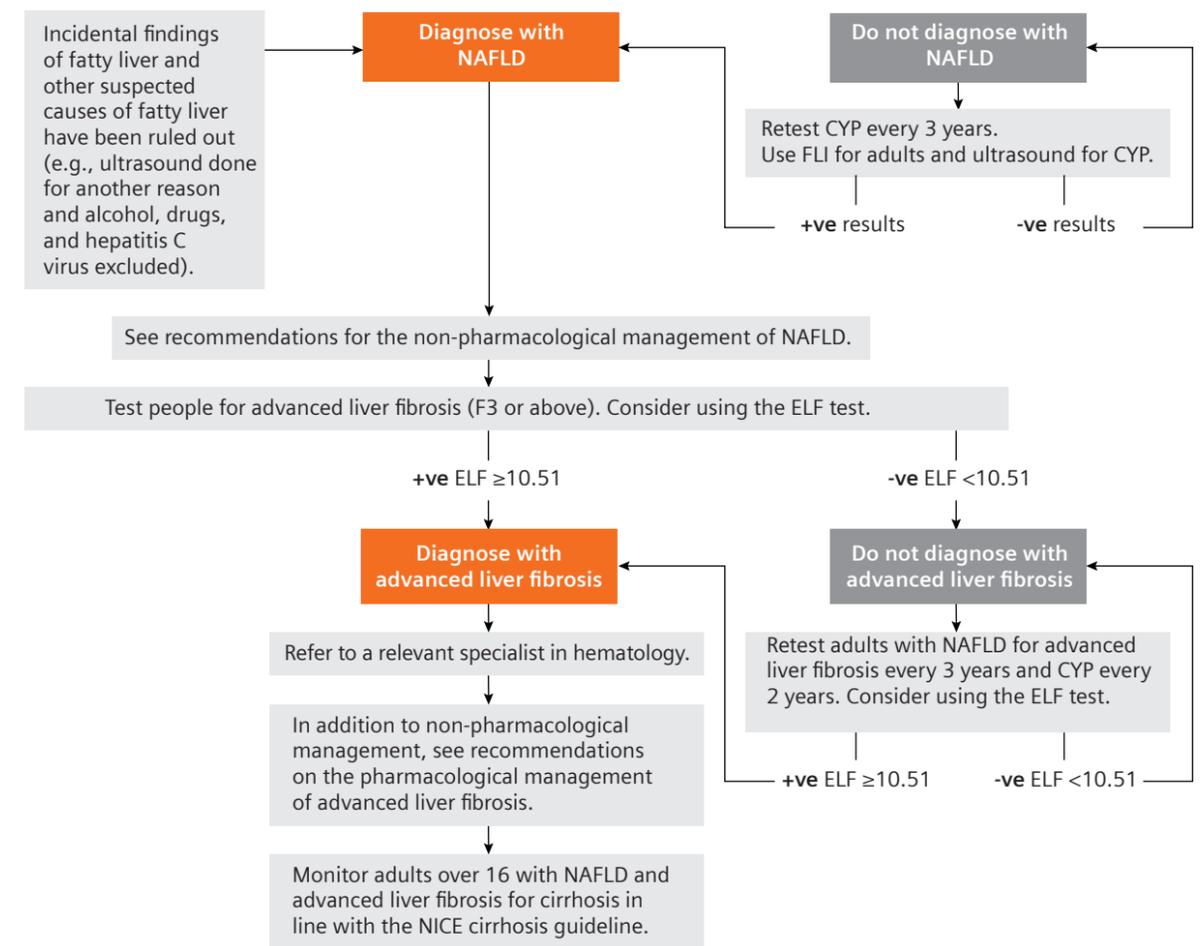
- This study included the assessment of many NITs and a combined performance and cost-analysis ranking. This is of value to clinicians confronted with multiple testing options who may be uncertain about what tests are most useful and what thresholds perform well.
- The ELF test ranked number one for both the pediatric and adult populations, and a common threshold of 10.51 was adopted for identifying advanced fibrosis.
- This pathway is similar to those proposed for Camden and Islington and the BSG Guidelines for NAFLD but includes single-step testing with the ELF test directly for advanced fibrosis. It used a higher cutpoint (10.51 vs. 9.5) based on data derived in part from a pediatric population.

- The recommendation for repeat testing with the ELF test supports detection of disease progression in patients initially stratified as lower risk.

## Conclusion<sup>17</sup>

“Consider using the enhanced liver fibrosis (ELF) test in people who have been diagnosed with NAFLD to test for advanced fibrosis.”

“Do not use routine liver blood tests to assess for advanced fibrosis in people with NAFLD.”



Be aware that NAFLD is a risk factor for type 2 diabetes, hypertension, and chronic kidney disease. Be aware that in people with type 2 diabetes, NAFLD is a risk factor for atrial fibrillation, myocardial infarction, ischemic stroke, and cardiovascular death.

# ELF in a Pediatric Population

## Performance of ELF Serum Markers in Predicting Fibrosis Stage in Pediatric Non-alcoholic Fatty Liver Disease

Nobili V, et al. Gastroenterology. 2009;136:160-7.

### Objective

Investigate the performance of the ELF test in assessing liver fibrosis in children and adolescents with NAFLD identified by biopsy.

### Methods

- Pediatric patients diagnosed with NAFLD and with elevated serum aminotransferases were recruited from a specialized tertiary referral center. NAFLD was confirmed by histopathology. All patients demonstrated evidence of insulin resistance.
- Paired serum and liver biopsy samples were obtained from each patient enrolled.
- ELF scores were obtained on all serum samples.
- Biopsy specimens were evaluated for NASH and fibrosis staging by a single liver pathologist blinded to the ELF score using a modified Brunt scoring system.
- The accuracy of the ELF test was evaluated against biopsy-proven fibrosis staging using AUROC analysis.

### Results

- ELF scores were associated with the discriminative stages of fibrosis (Brunt scoring).
- Combining simple markers with the ELF test did not provide additional value compared to the ELF test alone for the identification of any, significant, and advanced fibrosis.
- Analysis showed that 88% of patients could have correctly been spared biopsy if the ELF test had been used for the identification of significant fibrosis, while 12% would have had an indeterminate classification.
- Additional analysis explored the clinical utility of various single vs. two (high and low) ELF score thresholds as well as performance against different stages of fibrosis. In all scenarios evaluated, a significant majority of patients could have correctly been spared biopsy.
- A value of 10.51 supported the correct exclusion from advanced fibrosis ( $\geq 3$ ) in >95% of pediatric patients.

- The authors cautioned that additional pediatric studies were needed given the relatively low prevalence of advanced fibrosis in their study population.

### Significance

- NAFLD is increasing in prevalence in the pediatric population and will require accurate and cost-effective measures to assess the subset of high-risk patients with advancing fibrosis.
- Children are often perceived as a population for which, liver biopsy is needed to assess fibrosis, parents may be reluctant to agree to an invasive technique with its attendant risk. An NIT is of substantial appeal when assessing pediatric patients.
- The strong performance of the ELF test observed with AUROC analysis ( $\geq 0.90$ ) compared to biopsy-proven fibrosis in this pediatric population could be linked to the fact that children are less likely to have any significant extrahepatic sources of fibrosis such as organ fibrogenesis.

### Conclusion

“Taken together, the results of the study suggest that the ELF panel offers considerable promise in its ability to detect liver fibrosis in children and adolescents affected by NAFLD.”

Subjects in Each Cut		
Modified Brunt	(n = 112)	AUC
0 vs $\geq 1a$	37/75	0.92
$\leq 1a$ vs $\geq 1b$	45/67	0.92
$\leq 1b$ vs $\geq 1c$	51/61	0.90
$\leq 1c$ vs $\geq 2$	95/17	0.98
$\leq 2$ vs $\geq 3$	104/8	0.99

**NOTE:** Fibrosis severity was defined as follows: any fibrosis ( $\geq 1a$ ), moderate fibrosis-perisinusoidal ( $\geq 1b$ ), moderate fibrosis portal/periportal ( $\geq 1c$ ), significant fibrosis ( $\geq 2$ ), and advanced fibrosis ( $\geq 3$ ).

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