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Relationship of Enhanced Liver Fibrosis Score with Pediatric Nonalcoholic Fatty Liver Disease Histology and Response to Vitamin E or Metformin

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Abstract

Objectives—To study the diagnostic performance of the enhanced liver fibrosis score (ELF) for detecting different stages of fibrosis and its usefulness in detecting histologic response to vitamin E or metformin in children with nonalcoholic fatty liver disease who participated in the Vitamin E or Metformin for the Treatment Of NAFLD In Children (TONIC) trial.

Study design—ELF was measured at baseline and weeks 24, 48, and 96 on sera from 166 TONIC participants. Associations between ELF with baseline and end of trial (EOT) fibrosis stages and other histologic features were assessed using χ^2 tests and logistic regression models.

Results—ELF was significantly associated with severity of fibrosis at baseline and EOT. ELF areas under the curve for discriminating patients with clinically significant and advanced fibrosis were 0.70 (95% CI, 0.60–0.80) and 0.79 (95% CI, 0.69–0.89), respectively. A 1-unit decrease in ELF at EOT was associated with overall histologic improvement (OR, 1.86; 95% CI, 1.11–3.14; $P = .02$), resolution of steatohepatitis (OR, 1.88; 95% CI, 1.09–3.25; $P = .02$), improvement in steatosis grade (OR, 1.76; 95% CI, 1.06–2.82; $P = .03$), and hepatocellular ballooning (OR, 1.79; 95% CI, 1.06–3.00; $P = .03$), but not with improvement in fibrosis stage (OR, 1.26; 95% CI, 0.78–2.03; $P = .34$).

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Conclusions—ELF was associated with fibrosis stage in children who participated in TONIC. Although not associated with improvement in fibrosis, a decrease in ELF at EOT was associated with Nonalcoholic Steatohepatitis resolution and improvement in nonalcoholic fatty liver disease histology. ELF may be a useful noninvasive test to monitor treatment response in children with nonalcoholic fatty liver disease.

Nonalcoholic fatty liver disease (NAFLD) incidence and prevalence have been steadily increasing in the US along with the increasing rates of obesity.[1–5] Although there is a paucity of data on the natural history of NAFLD in children, it is clear that some children experience NASH progression to cirrhosis and end stage liver disease even requiring liver transplantation during childhood.[6,7] The progression of NAFLD is driven by propagation of hepatic fibrosis.[8–10]

Although liver biopsy is considered the gold standard for the diagnosis and staging of NAFLD, the risks, costs, sampling errors, and variability in interpretation are problematic and decrease enthusiasm for widespread use for NAFLD staging and monitoring response to therapy.[5,11] There is an unmet need for noninvasive methods to assess severity and progression of fibrosis and monitor response to interventions in children with NAFLD.[12]

The enhanced liver fibrosis score (ELF) was developed and validated as a serum-based biomarker for fibrosis in adult patients with NAFLD.[13,14] It is generated from an algorithm that incorporates measurements of tissue inhibitor of metalloproteinases-1, amino-terminal propeptide of type III procollagen (PIIINP), and hyaluronic acid. In 2 prior studies from the same Italian center, ELF showed excellent area under the receiver operating characteristic curve (AUROC) (>90%) for discriminating any stage of fibrosis, or clinically significant or advanced fibrosis in children and adolescents with NAFLD and elevated liver enzymes.[15,16] There are no available multicenter data on ELF performance in discriminating different stages of fibrosis in US children with NAFLD or as a tool to monitor their response to therapy.

The Vitamin E or Metformin for the Treatment Of NAFLD In Children (TONIC) trial was a multicenter, randomized controlled trial that compared the efficacy and safety of vitamin E and metformin in children with biopsy-proven NAFLD (NCT00063635). These patients had per-protocol liver biopsies at both baseline and end of trial (EOT) (after 96 weeks). In this study, we aimed to assess the association of ELF with stages of fibrosis in children enrolled in the TONIC trial, its diagnostic performance for detecting the different stages of fibrosis, and its usefulness in monitoring histologic response to therapy in the TONIC trial. Because PIIINP has previously been shown to be a biomarker of inflammation in adults with NASH, we investigated its performance here in children with biopsy-proven NAFLD as a secondary objective of this study.[17,18]

Methods

The TONIC trial was conducted by the NASH Clinical Research Network (NASH CRN). The design, methods, and results of this trial were published previously.[19,20] This randomized, double-blind, placebo-controlled trial was conducted at 10 university clinical research centers and included 173 children with biopsy-confirmed NAFLD. Participants

were randomized to receive vitamin E (800 IU daily, 58 patients), metformin (1000 mg/day, 57 patients), or placebo (58 patients) for 96 weeks. Eligibility criteria included persistently elevated alanine aminotransferase (ALT), defined as of ALT of more than 60 U/L for 1–6 months before randomization, and evidence of NAFLD on liver biopsy. Biopsies were interpreted by site pathologists to determine eligibility and later read centrally by NASH CRN pathologists masked to treatment assignment. The central read was performed by the NASH CRN Pathology Committee according to the NASH CRN scoring system and was used in the analysis.[21] Clinically significant fibrosis was defined as fibrosis stage 2 or higher and advanced fibrosis was defined as fibrosis stage 3 or higher.

For our study, outcome measures were assessed using change from baseline to week 96 of treatment for the following: NASH and borderline NASH improving to not NASH (resolution of NASH), NAFLD activity score (NAS) improvement of 2 or more points and no worsening of fibrosis (histologic improvement), improvement of 1 or more points in NAS, and in individual histologic scores including hepatocellular ballooning, fibrosis, steatosis, and lobular inflammation.

The use of archived biosamples stored at the National Institute of Diabetes and Digestive and Kidney Diseases Biorepository was permitted under the original informed consent participants and their parents or guardians provided before enrollment in TONIC. The serum samples used in our study were obtained from blood drawn from fasting participants, which was collected into serum separator tubes, allowed to clot for at least 30 minutes at room temperature, and centrifuged at 1800×*g* for 15 minutes at 4 °C. Aliquots of 0.5 mL of serum were immediately frozen at −80 °C. Processing was completed within 2 hours, and samples were free of hemolysis.

Of 173 pediatric patients with NAFLD who participated in TONIC, 166 had serum samples available from baseline, week 24, week 48, and week 96 for the current study. ELF (Siemens Healthcare Diagnostics Inc) and the individual components of ELF were measured on an Advia Centaur XP (Siemens Healthcare Diagnostics Inc) according to manufacturer instructions.[22]

Statistical Analyses

The cross-sectional distribution of the continuous raw ELF score at baseline, 24, 48, and 96 weeks, change from baseline in ELF and NAS at 96 weeks, and ELF scores within levels of NAS, were graphically assessed using histograms overlaid with normal and/or kernel-based distributional curves. ELF and PIIINP scores by histologic features at baseline and EOT were tabulated for the overall cohort along with their corresponding means and SDs. The univariate differences in ELF and PIIINP scores within histologic features at baseline and EOT were assessed using nonparametric Kruskal-Wallis tests. The distribution of ELF score across baseline stages of fibrosis was assessed graphically using boxplots and analytically using linear regression models.

Associations and unadjusted discriminatory performance of baseline biomarkers with fibrosis stages were assessed within a logistic regression framework and ORs and AUROC reported along with their corresponding 95% CIs per unit SD change.

Histologic improvement over 96 weeks per unit decrease in the ELF score was assessed using a logistic regression model and predictive performance was again assessed using AUROC from participants with available paired histology measures, adjusted for treatment group. Fibrosis improvement per unit decrease in ELF components and their predictive performance was assessed as discussed elsewhere in this article and adjusted for baseline biomarker level and treatment group.

ELF changes over time were visualized graphically and assessed within a linear mixed model framework to account for correlation between successive patient measurements. *P* values reported are nominal. Measures of variability including SD and 95% CIs were reported. All analyses were performed using SAS software version 9.4 (SAS Institute Inc) at a 5% significance level.

Results

TONIC Trial

The results of the TONIC trial were published previously.[20] Participants were mostly White (74%), male (81%), of Hispanic ethnicity (61%) with a mean age of 13.1 ± 2.4 years, a body mass index (BMI) of 34 ± 6 kg/m^2, ALT of 123 ± 65 U/L, triglycerides of 153 ± 100 mg/dL, and Homeostatic Model Assessment for Insulin Resistance of 9.2 ± 11.6. The majority of patients (83%) had NASH at baseline biopsy (borderline or suspicious in 42%, definite in 41%) (Table I).

The proportion of TONIC participants treated with vitamin E or metformin with a sustained decrease in ALT was not different compared with placebo. Resolution of NASH, when present, was more frequent in the vitamin E group. Some histologic features like hepatocellular ballooning or NAS improved more in the vitamin E group, and ballooning improved more in the metformin group.[20]

Cross-sectional Association Between ELF, Fibrosis Stage, and Other Histologic Subphenotypes

Fibrosis of any stage was detected in 76% and 60% of the participants at baseline and EOT liver biopsy, respectively (Table I). ELF at baseline and EOT (96 weeks) was significantly associated with fibrosis stage ($P < .05$). At baseline, the ELF (mean ± SD) was 8.51 ± 0.57 in participants without fibrosis (F0), 8.49 ± 0.63 in those with mild fibrosis (F1), 8.77 ± 0.76 in those with moderate fibrosis (F2), and 9.28 ± 0.69 in those with bridging fibrosis (F3) ($P < .0001$) (Table I and Figure 1 [available at www.jpeds.com]). Histograms of ELF score by study visit are shown in (Figure 2, A–D; available at www.jpeds.com).

ELF was also significantly associated with steatosis grade, lobular inflammation, hepatocyte ballooning, Mallory-Denk bodies, and a diagnosis of steatohepatitis at baseline and EOT (Table I). ELF was associated with portal inflammation grade only at EOT (Table I).

Performance of ELF and Its Individual Components for Discrimination of Different Fibrosis Stages at Baseline

Only tissue inhibitor of metalloproteinases-1 (OR, 1.63; 95% CI 1.08–2.45; $P = .02$) significantly discriminated patients who had any stage of fibrosis (AUROC, 0.63; 95% CI 0.54–0.72) (Table II). ELF, hyaluronic acid, and PIIINP had AUROC 0.63–0.70 for discriminating patients with clinically significant fibrosis. ELF, hyaluronic acid, and tissue inhibitor of metalloproteinases-1 had AUROC 0.76–0.79 for discriminating patients with advanced fibrosis. PIIINP did not significantly discriminate participants with advanced fibrosis (OR, 1.45; 95% CI 0.98–2.14; $P = .06$). Overall, the ELF score had a higher AUROC than any of its individual components for detection of significant and advanced fibrosis (Table II).

Association of Changes in ELF with Histologic Response at EOT

Changes in ELF in response to therapy received in each study arm were evaluated. There were no significant changes in mean ELF at the 24-, 48-, or 96-week specific time points from baseline with vitamin E or metformin vs placebo (Figure 3, A). ELF did not significantly decrease over the study period in participants who achieved fibrosis improvement (Figure 3, B). However, ELF showed significant improvement over the study period in participants who achieved NAS-based histologic improvement and NASH resolution (Figure 3, C–D).

A model predicting improvement in histology per unit decrease in ELF after 96 weeks showed that one unit decrease was significantly associated with overall histologic improvement (OR, 1.86; 95% CI, 1.11, 3.14; $P = .02$), resolution of steatohepatitis (OR, 1.88; 95% CI, 1.09–3.25; $P = .02$), improvement in steatosis grade (OR, 1.76; 95% CI, 1.06–2.82; $P = .03$), improvement in hepatocellular ballooning (OR, 1.79; 95% CI, 1.06–3.00; $P = .03$), and improvement in NAS (OR, 1.97; 95% CI, 1.16–3.36; $P = .01$), but not with improvement in fibrosis stage or lobular inflammation (Table III). None of the individual components of ELF significantly predicted improvement in fibrosis (Table IV; available at www.jpeds.com). Figure 4 (available at www.jpeds.com) shows histograms of ELF and NAS changes, as well as ELF changes per NAS changes at EOT.

Association of PIIINP with Baseline Histology and EOT Histologic Response

At baseline, the PIIINP level was significantly associated with fibrosis stage and Mallory-Denk bodies but no other NAFLD histologic features (Table V; available at www.jpeds.com). PIIINP level (mean ± SD) was 16.01 ± 6.74 ng/mL in participants with F0, 16.01 ± 6.74 ng/mL in those with F1, 20.35 ± 10.18 ng/mL in those with F2, and 20.11 ± 6.76 ng/mL in those with F3 ($P = .01$). At EOT, PIIINP was significantly associated with portal inflammation grade but not with fibrosis or other NAFLD histologic features (Table V). Change in PIIINP during the study did not correlate with any of the major histologic end points (Figure 5; available at www.jpeds.com).

Discussion

In this well-characterized pediatric cohort with biopsy-proven NAFLD, ELF was significantly associated with fibrosis stage and other NAFLD histologic features at baseline and EOT. ELF showed a significant decrease over the study period in patients who experienced important histologic outcomes, NASH resolution and NAS-based histologic improvement. In accordance with the histologic analysis findings at EOT of the TONIC trial, where neither vitamin E nor metformin resulted in significant improvement in fibrosis, ELF did not show a significant decrease with fibrosis improvement.

In contrast with multiple studies in the adult population, there are no data currently available on the usefulness of ELF for monitoring response to drugs used to treat NAFLD or NASH in children.[13,14,23–25] A prior study of 39 obese children with suspected NAFLD reported a median decrease of −0.39 ELF units with lifestyle intervention resulting in minimal weight loss.[26]

Accurate noninvasive tests are needed to assess disease activity and fibrosis in children with this common chronic liver disease. In contrast with the extensive development of noninvasive markers of fibrosis in adults with NAFLD, this research remains considerably more limited in pediatric NAFLD.[12,16,27–31] There are few data on the use of ELF as a noninvasive marker for fibrosis in this population.[12,15,16] In an analysis of 400 healthy volunteers including 32 individuals under 20 years of age, the mean ELF score was 7.95 in males and 7.80 in females; 9.80 was proposed as a general cutoff for moderate fibrosis and 11.3 for advanced fibrosis.[32] In contrast, the present study of children with NAFLD ages 8–18 years, ELF was 8.51 in participants without fibrosis (F0), 8.77 in those with moderate fibrosis (F2), and 9.28 in those with bridging fibrosis (F3). Given the paucity of data in children and the variability of ELF cutoffs used in different studies, more studies are needed to establish the optimal ELF cutoffs for discriminating different stages of hepatic fibrosis in children with NAFLD. Although ELF outperformed its individual components for discrimination of clinically significant and advanced fibrosis at baseline, it demonstrated only fair performance in this cohort. The AUROCs for ELF for discriminating different stages of fibrosis in children and adolescents with NAFLD in our study were lower than previously reported in the 2 prior studies from an Italian center.[15,16] Population differences may have influenced performance of ELF in these different studies. Although the Italian studies included mostly male children (56%−66%) with NASH (67%−69%), the mean BMI for those participants (approximately 25 kg/m^2) and the prevalence of clinically significant or advanced fibrosis (≥F2. 13%−15%) were markedly lower than in the TONIC study (mean BMI of 34 kg/m^2 and ≥F2 32%, respectively). In addition, most patients in our study (61%) were of Hispanic ethnicity and recruited from multiple centers in the US. This study demonstrated that the excellent performance of ELF for discriminating fibrosis stages in the Italian studies may not apply to typical pediatric NAFLD patients in the US.

A lack of significant improvement in ELF in this cohort was in line with lack of significant improvement in histologic fibrosis with either vitamin E or metformin. These findings are similar to the findings of our post hoc analysis of the adult PIVENS trial, in which neither vitamin E nor pioglitazone resulted in significant improvement in fibrosis in adults with

NASH.[18,33] ELF did not correlate with fibrosis improvement in PIVENS, but did correlate significantly with the NAS-based histologic improvement.[18] A possible explanation is that the histologic staging of fibrosis is too insensitive to pick up subtle changes in fibrosis that are detectable using ELF. This notion is plausible, given that ELF generates a score that is a continuous variable with a coefficient of variation of 3%−8% and histologic staging generates a categorical variable score with well-documented variation in excess of 20%.[11,32,34]

In adult patients with NASH in the PIVENS trial, PIIINP showed much more dynamic changes in relation to changes in fibrosis and other histologic end points.[18] In this analysis of TONIC participants, PIIINP showed a significant association only with fibrosis stage at baseline, but did not correlate with histologic improvement, NASH resolution, or other histologic end points. ELF was better than its individual components in discriminating children with clinically significant and advanced fibrosis and showed more dynamic changes with improvement in different histologic end points (except for fibrosis) than PIIINP. The differences in ELF performance between children and adults with NAFLD studies are likely related to factors influencing ELF performance such as sex and age.[32]

Because the TONIC trial included patients with NAFLD with a persistent elevation in ALT who did not have diabetes or cirrhosis, our findings on ELF performance may not be generalizable to NAFLD patients with normal ALT, diabetes, or cirrhosis. The relatively poor performance of ELF in discriminating children with no or mild fibrosis (AUROC of 0.60 and 0.70, respectively) may be a limiting factor for ELF use in general pediatric practice, where most children have less severe NAFLD compared with children enrolled in this NASH CRN study performed at tertiary care centers. Although TONIC was completed and published in 2011, ELF was measured on serum samples in 2012 and 2013, even though this current post hoc data analysis was done later. This study has several strengths. The population was enriched in children with a higher BMI and clinically significant fibrosis, adding value to the analysis. The pathology assessment was centrally performed by the NASH CRN Pathology Committee. The most unique feature of this study was that changes in ELF were assessed longitudinally in relation to histology and in response to therapeutic interventions.

In summary, ELF was significantly associated with fibrosis and other NAFLD histologic features in children who participated in the TONIC clinical trial. Although not associated with improvement in fibrosis, a decrease in ELF was associated with NASH resolution and improvement in other NAFLD histologic lesions. ELF may be a useful noninvasive marker to monitor response to treatment in children with NAFLD.

Funding and Conflicts of Interest Disclosure

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Funding and disclosure information is available at www.jpeds.com.

Glossary

ALT	Alanine aminotransferase
AUROC	Area under the receiver operating characteristic curve
BMI	Body mass index
ELF	Enhanced liver fibrosis score
EOT	End of trial
NAFLD	Nonalcoholic fatty liver disease
NAS	NAFLD activity score
NASH	Nonalcoholic Steatohepatitis
NASH CRN	Nonalcoholic Steatohepatitis Clinical Research Network
PIIINP	Amino-terminal propeptide of type III procollagen
TONIC	Vitamin E or Metformin for the Treatment Of NAFLD In Children

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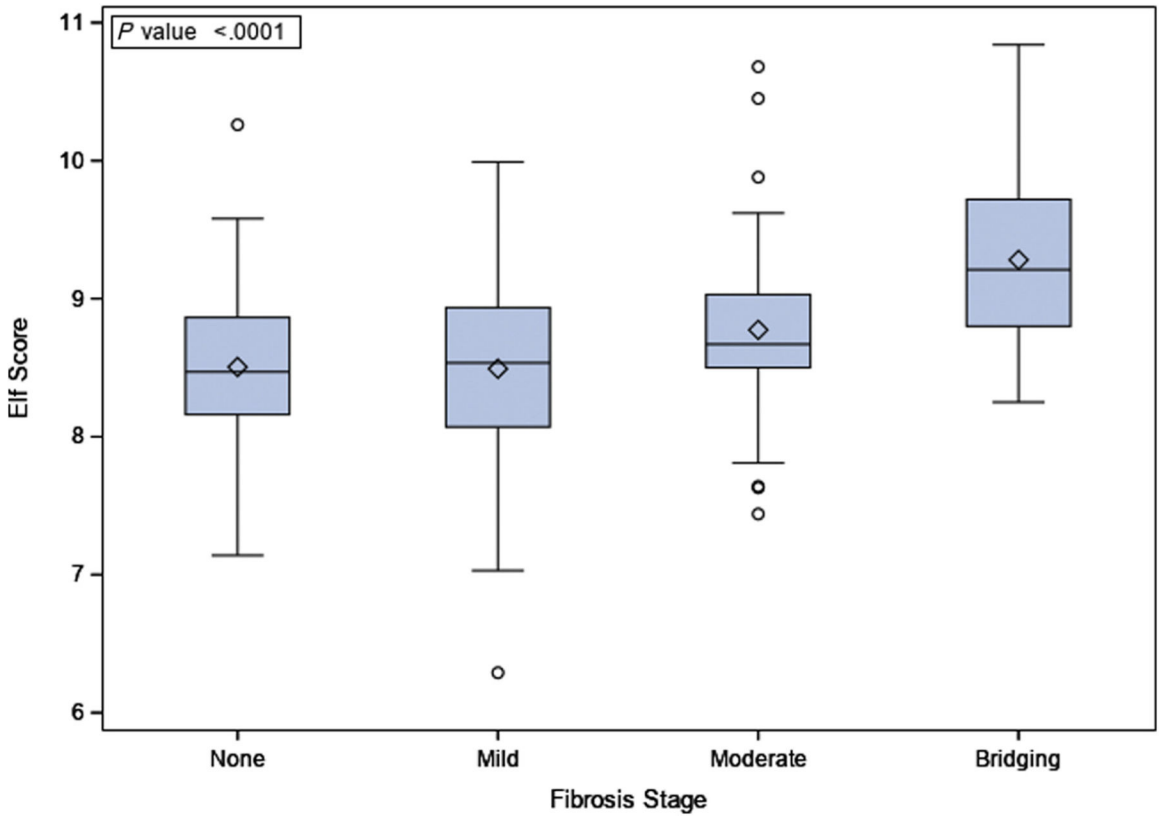

Figure 1.
ELF score and baseline fibrosis stages.

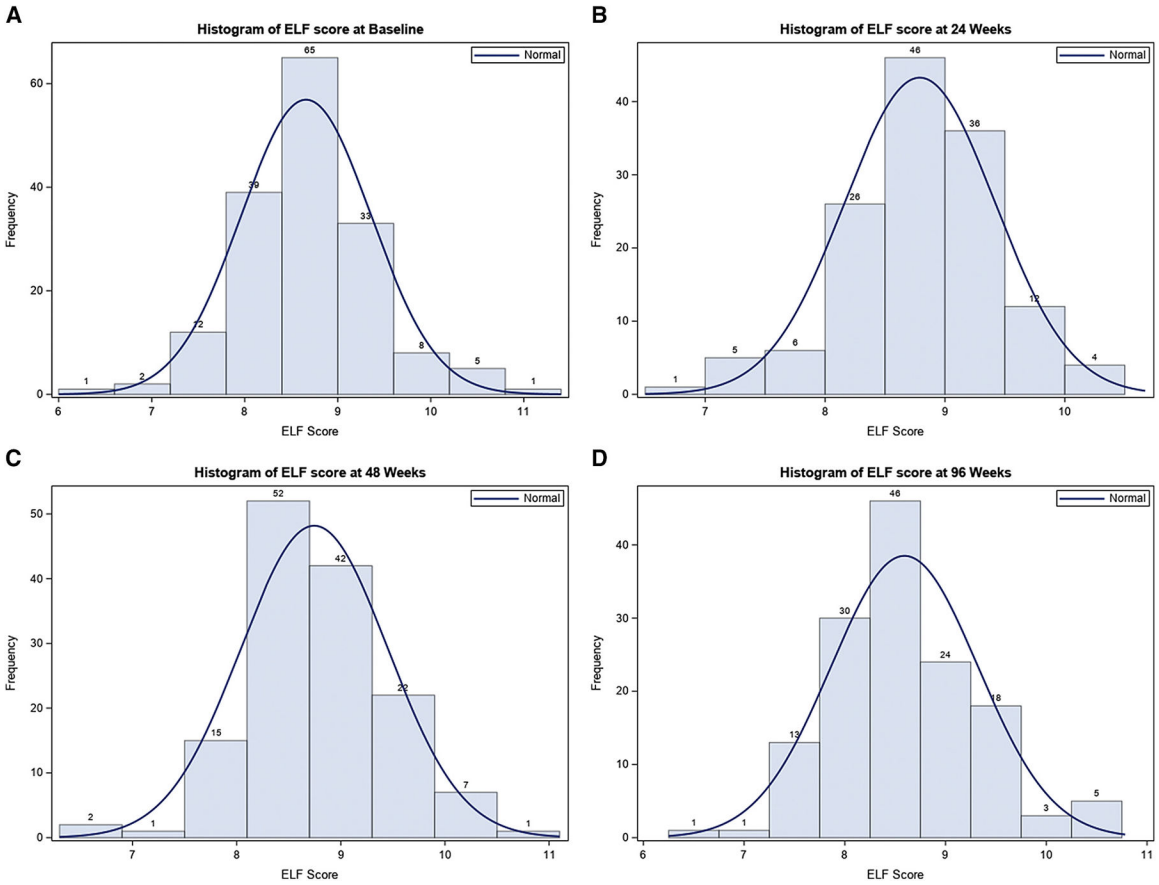

Figure 2.

Histograms of ELF score by study visit. **A,** Histogram of ELF score at baseline. **B,** Histogram of ELF score at 24 weeks. **C,** Histogram of ELF score at 48 weeks. **D,** Histogram of ELF score at 96 weeks.

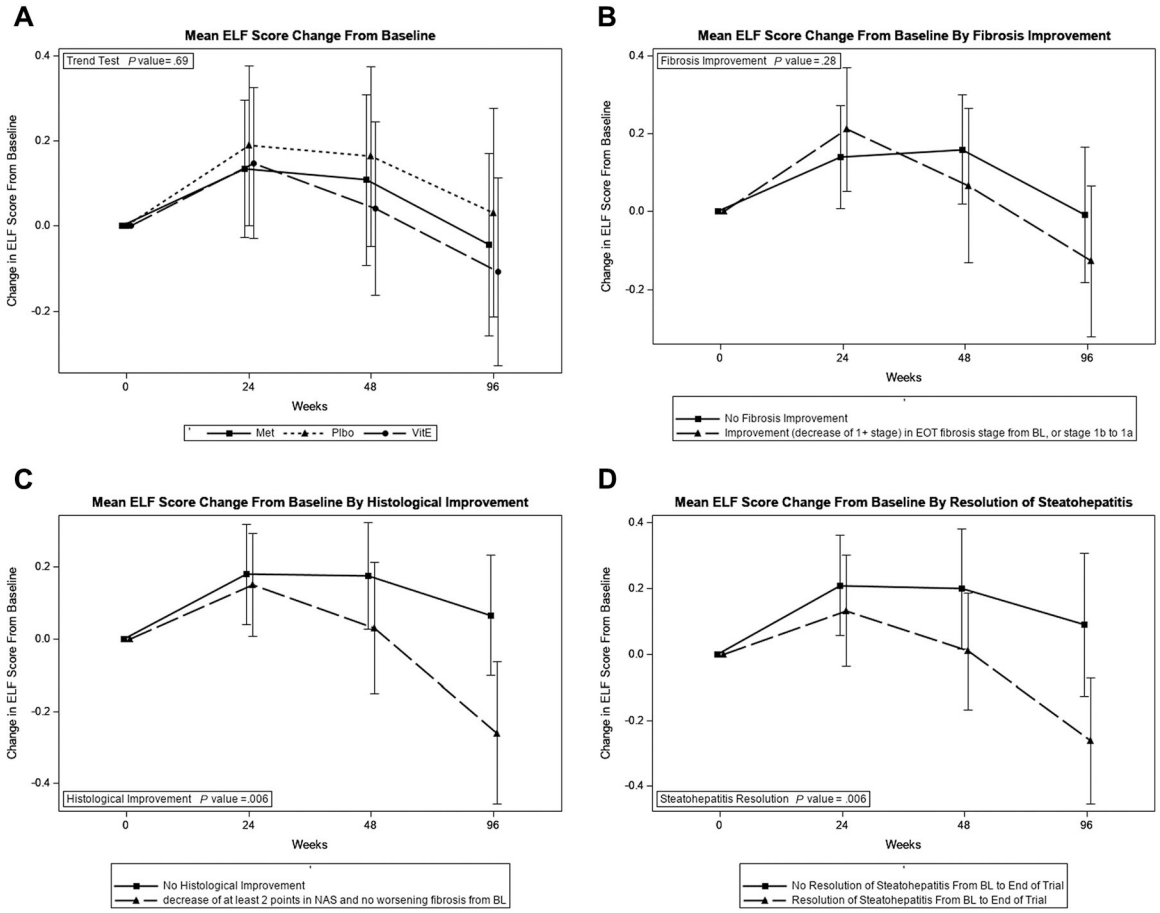

Figure 3.

Changes in ELF score in the TONIC trial. **A,** Mean ELF score change from baseline. **B,** Mean ELF score change from baseline by fibrosis improvement. **C,** Mean ELF score change from baseline by histologic improvement. **D,** Mean ELF score change from baseline by resolution of steatohepatitis.

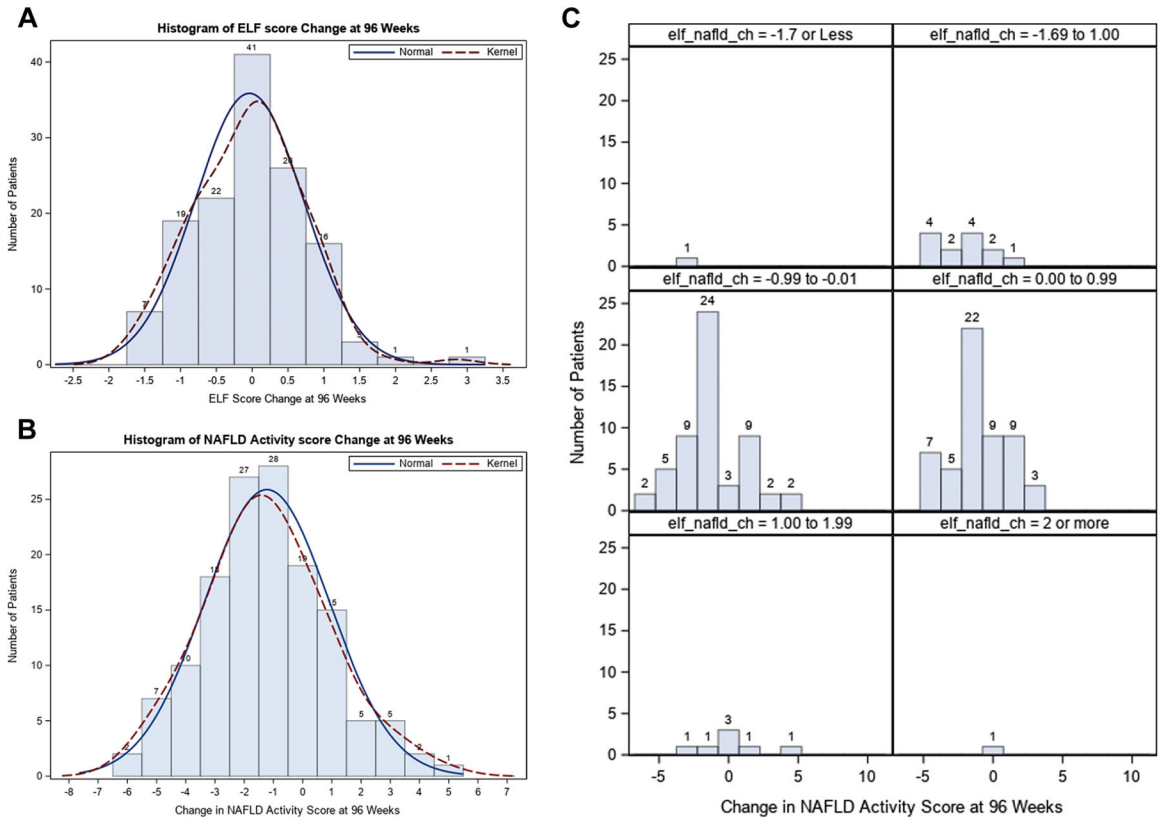

Figure 4.

Change in ELF and NAS at 96 weeks. **A,** Histogram of ELF score change at 96 weeks. **B,** Change in NAS at 96 weeks. **C,** ELF changes per changes in NAS.

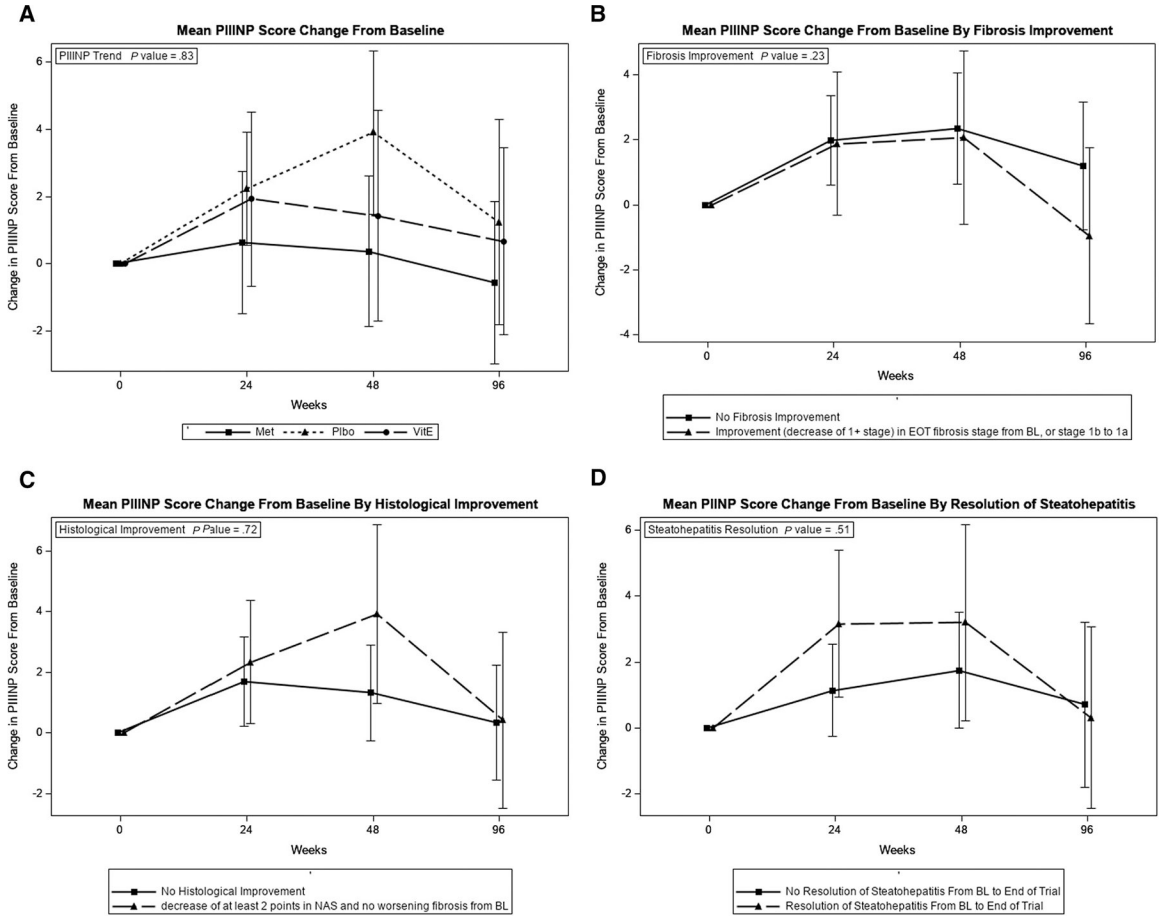

Figure 5.

Changes in PIIINP and histologic end points in the TONIC trial. **A,** Mean PIIINP score change from baseline. **B,** Mean PIIINP score change from baseline by fibrosis improvement. **C,** Mean PIIINP score change from baseline by histologic improvement. **D,** Mean PIIINP score change from baseline by resolution of steatohepatitis.

Table I.

Distribution of ELF by histologic features at baseline and EOT

Variable labels	Overall, baseline (n = 166)	ELF score at baseline	*P* value*	Overall EOT (n = 141)	Elf score at EOT	*P* value*
Fibrosis stage			<.0001			.003
None	40 (24.2)	8.51 ± 0.57		55 (39.6)	8.36 ± 0.55	
Mild	72 (43.6)	8.49 ± 0.63		42 (30.2)	8.61 ± 0.79	
Moderate	29 (17.6)	8.77 ± 0.76		22 (15.8)	8.67 ± 0.78	
Bridging	24 (14.5)	9.28 ± 0.69		20 (14.4)	9.13 ± 0.77	
Steatosis grade			.024			<.001
≤33%	44 (26.5)	8.51 ± 0.74		68 (48.2)	8.36 ± 0.61	
34%−66%	52 (31.3)	8.55 ± 0.54		37 (26.2)	8.79 ± 0.80	
≥66	70 (42.2)	8.84 ± 0.74		36 (25.5)	8.29 ± 0.76	
Lobular inflammation			.044			<.001
<2 foci	75 (45.2)	8.51 ± 0.59		95 (67.4)	8.46 ± 0.73	
≥2 foci	91 (54.8)	8.78 ± 0.76		46 (32.6)	8.86 ± 0.67	
Hepatocellular ballooning			.004			.01
None	69 (41.6)	8.47 ± 0.62		83 (59.7)	8.47 ± 0.71	
Few	57 (34.3)	8.70 ± 0.68		33 (23.7)	8.59 ± 0.67	
Many	40 (24.1)	8.92 ± 0.77		23 (16.5)	9.06 ± 0.76	
Portal inflammation			.352			.04
None	13 (7.8)	8.43 ± 0.50		20 (14.4)	8.41 ± 0.63	
Mild	136 (81.9)	8.67 ± 0.73		101 (72.7)	8.56 ± 0.72	
More than mild	17 (10.2)	8.76 ± 0.59		18 (12.9)	9.01 ± 0.81	
Mallory-Denk bodies			.005			<.001
Absent/rare	160 (96.4)	8.62 ± 0.67		130 (93.5)	8.53 ± 0.70	
Many	6 (3.6)	9.65 ± 0.85		9 (6.5)	9.56 ± 0.62	
Steatohepatitis diagnosis			.005			.02
NAFLD, not NASH	29 (17.5)	8.26 ± 0.63		62 (44.0)	8.39 ± 0.60	
Borderline, suspicious	69 (41.6)	8.67 ± 0.60		42 (29.8)	8.63 ± 0.78	
Definite NASH	68 (41.0)	8.82 ± 0.76		37 (26.2)	8.88 ± 0.78	

Values are number (%) or mean ± SD. Baseline and EOT.

* *P* values for continuous variables are based on nonparametric Kruskal Wallis tests.

Table II.

Comparison of the baseline values of serum hyaluronic acid, PIIINP, TIMP-1, and ELF for discrimination of fibrosis stages

Biomarker performances by baseline fibrosis*	OR (95% CI)†	*P* value†	AUROC (95% CI)‡
Any fibrosis			
HA (per SD ng/mL)	1.26 (0.80–1.99)	.32	0.57 (0.47–0.67)
PIIINP (per SD ng/mL)	1.32 (0.88–1.97)	.18	0.55 (0.45–0.65)
TIMP-1 (per SD ng/mL)	1.63 (1.08–2.45)	.02	0.63 (0.54–0.72)
ELF (per SD score)	1.36 (0.93–1.97)	.11	0.60 (0.50–0.70)
Clinically significant fibrosis			
HA (per SD ng/mL)	2.15 (1.34–3.46)	.002	0.64 (0.54–0.74)
PIIINP (per SD ng/mL)	1.81 (1.26–2.60)	.002	0.66 (0.57–0.75)
TIMP-1 (per SD ng/mL)	1.57 (1.12–2.21)	.01	0.63 (0.53–0.72)
ELF (per SD score)	2.28 (1.53–3.39)	<.0001	0.70 (0.60–0.80)
Advanced fibrosis			
HA (per SD ng/mL)	1.98 (1.31–3.01)	.001	0.77 (0.66–0.88)
PIIINP (per SD ng/mL)	1.45 (0.98–2.14)	.06	0.65 (0.53–0.76)
TIMP-1 (per SD ng/mL)	2.86 (1.74–4.72)	<.0001	0.76 (0.64–0.88)
ELF (per SD score)	3.03 (1.82–5.05)	<.0001	0.79 (0.69–0.89)

HA, hyaluronic acid; *TIMP-1*, tissue inhibitor of metalloproteinases-1.

*Fibrosis categories defined as: Any fibrosis = F1, F2, F3, F4 vs F0; clinically significant fibrosis = F2, F3, F4 vs F0, F1; advanced fibrosis = F3, F4 vs F0, F1, F2.

†ORs and associated 95% CIs were determined from a logistic regression model of fibrosis on specified biomarker. The ORs shown in this table are standardized and represent the odds of fibrosis categories per SD change in the biomarker. *P* values (2-sided) were determined from a Wald test.

‡AUROC to assess the discriminatory power of distinguishing between fibrosis stages.

Table III.

Relative odds of improved histology per unit decrease in ELF over 96 weeks

Histologic changes over 96 weeks	Relative odds (95% CI) of improved histology per unit decrease in ELF over 96 weeks (n = 133)*	*P* value†	AUROC (95% CI)‡
Overall histologic improvement§	1.86 (1.11–3.14)	.02	0.62 (0.52–0.71)
Resolution of steatohepatitis¶	1.88 (1.09–3.25)	.02	0.63 (0.52–0.73)
Improvement of ≥1 point			
Fibrosis stage**	1.26 (0.78–2.03)	.34	0.54 (0.44–0.64)
Steatosis grade	1.76 (1.06–2.82)	.03	0.60 (0.51–0.70)
Lobular inflammation	1.55 (0.96–2.50)	.07	0.60 (0.50–0.69)
Hepatocellular ballooning	1.79 (1.06–3.00)	.03	0.62 (0.52–0.72)
NAS	1.97 (1.16–3.36)	.01	0.64 (0.54–0.74)

*Participants with paired ELF scores (baseline and 96 weeks) and paired histology were included in analyses.

†*P* value determined from logistic regression of change in histologic feature on change in ELF and assigned treatment group.

‡The AUROC is the predictive power for the performance of ELF change at 96 weeks.

§Overall histologic improvement response defined as a decrease in the NAS score by ≥2 points and no worsening of fibrosis.

¶Resolution of steatohepatitis was defined as a diagnosis of borderline or definite steatohepatitis at baseline and a diagnosis of not NAFLD or NAFLD only at 96 weeks; n = 77 with borderline/definite steatohepatitis at baseline; n = 51 with resolution of steatohepatitis.

**Fibrosis improvement defined as a decrease by one or more stage, with change from stage 1b to 1a also considered improvement.

Table IV.

Relative odds of fibrosis improvement per unit decrease in ELF components over 96 weeks

ELF components	Relative odds (95% CI) of fibrosis improvement per SD decrease in biomarker over 96 weeks (n = 133)*	*P* value†	AUROC (95% CI)
HA (ng/mL)	1.23 (0.78–1.95)	.38	0.58 (0.48–0.68)
PIIINP (ng/mL)	1.13 (0.78–1.63)	.51	0.53 (0.43–0.63)
TIMP-1 (ng/mL)	1.43 (0.93–2.19)	.10	0.58 (0.48–0.68)

HA, hyaluronic acid; *TIMP-1*, tissue inhibitor of metalloproteinases-1.

*Participants with paired ELF scores (baseline and 96 weeks) and paired histology were included in analyses.

†Adjusted for baseline biomarker and treatment group.

Table V.

Distribution of PIIINP by histologic feature at baseline and end of study

Histologic features	Baseline		EOT	
	Mean (SD)*	*P* value†	Mean (SD)*	*P* value†
Fibrosis stage		.01		.82
None	16.01 (6.74)		16.68 (7.39)	
Mild	16.01 (6.74)		18.00 (9.40)	
Moderate	20.35 (10.18)		16.75 (6.94)	
Bridging	20.11 (6.76)		18.11 (7.09)	
Steatosis grade		.09		.32
≤33%	15.92 (7.08)		15.99 (7.18)	
34%−66%	16.50 (5.94)		19.02 (8.66)	
>66%	18.83 (8.45)		17.73 (8.03)	
Lobular inflammation		.06		.53
<2 foci	16.05 (6.39)		17.11 (8.46)	
≥2 foci	18.38 (8.11)		17.48 (6.53)	
Hepatocellular ballooning		.09		.17
None	15.59 (5.81)		17.94 (8.27)	
Few	18.41 (7.46)		15.59 (8.08)	
Many	18.79 (9.32)		17.41 (5.91)	
Portal inflammation		.88		.01
None	16.02 (6.11)		16.79 (8.49)	
Mild	17.49 (7.84)		16.50 (7.54)	
More than mild	17.06 (4.97)		22.29 (7.78)	
Mallory-Denk bodies		.03		.76
Absent/rare	17.14 (7.47)		17.28 (8.04)	
Many	22.26 (5.23)		17.47 (5.62)	
Steatohepatitis diagnosis		.12		.30
NAFLD, not NASH	15.01 (6.80)		17.87 (7.72)	
Borderline, suspicious	16.81 (5.83)		16.29 (9.12)	
Definite NASH	18.84 (8.84)		17.22 (6.53)	

**P* values obtained using the nonparametric Kruskal-Wallis test.

†Units are nanograms per milliliter.