Research article JHEP|Reports

Enhanced liver Fibrosis® test predicts liver-related outcomes in the general population

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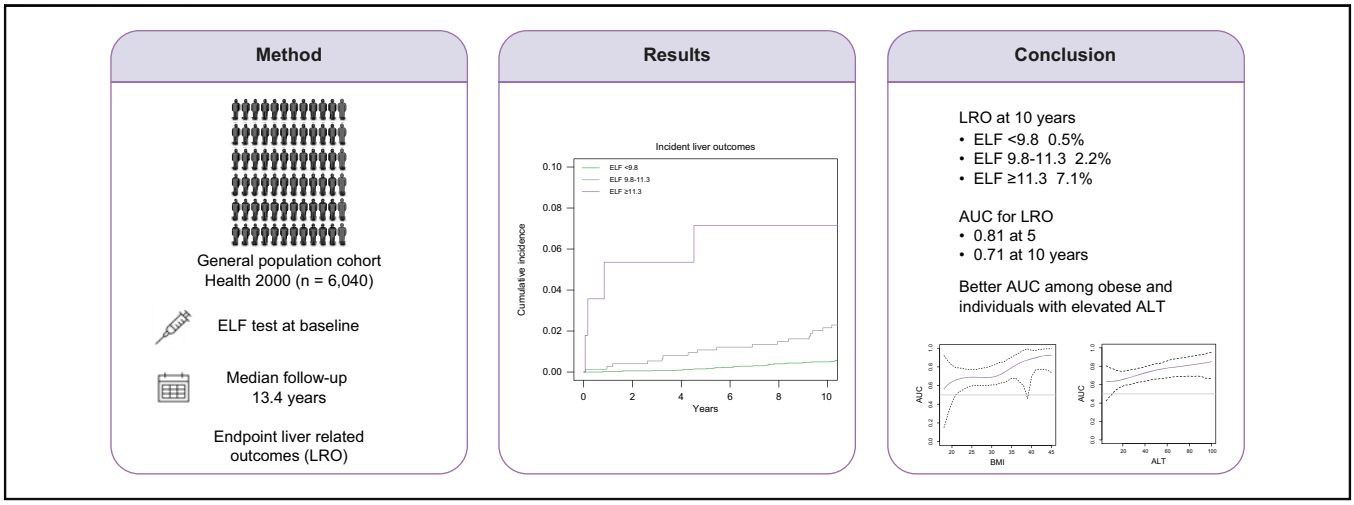
Graphical abstract



Highlights

- Liver fibrosis is the most important predictor of liver-related outcomes.
- In previous studies ELF had good discrimination for advanced liver fibrosis detection.
- ELF predicts liver-related outcomes in selected patient populations.
- In our large general population cohort study, ELF predicted liver-related outcomes.
- Predictive performance improved in the presence of obesity, diabetes, or raised ALT.

Impact and implications

The Enhanced Liver Fibrosis test exhibits good performance for predicting liver-related outcomes (hospitalisation, liver cancer, or liver-related death) in the general population, especially in those with risk factors.

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Enhanced liver Fibrosis® test predicts liver-related outcomes in the general population

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Background & Aims: The Enhanced Liver Fibrosis® (ELF) test exhibits good discriminative performance in detecting advanced liver fibrosis and in predicting liver-related outcomes in patients with specific liver diseases, but large population-based studies are missing. We analysed the predictive performance of the ELF test in a general population cohort.
Methods: Data were sourced from the Health 2000 study, a Finnish population-based health examination survey conducted in 2000–2001. Subjects with baseline liver disease were excluded. The ELF test was performed on blood samples collected at baseline. Data were linked with national healthcare registers for liver-related outcomes (hospitalisation, cancer, and death).
Results: The cohort comprised 6,040 individuals (mean age 52.7. 45.6% men) with 67 liver-related outcomes during a median 13.1-year follow-up. ELF predicted liver outcomes (unadjusted hazards ratio 2.70, 95% CI 2.16–3.38). with 5- and 10-year AUCs of 0.81 (95% CI 0.71–0.91) and 0.71 (95% CI 0.63–0.79) by competing-risk methodology. The 10-year risks for liver outcomes increased from 0.5% at ELF <9.8 to 7.1% at ELF ≥11.3, being higher among men than women at any given ELF level. Among individuals with body mass index ≥30 kg/m^2, diabetes, or alanine aminotransferase >40 U/L. Five-year AUCs for ELF were 0.85, 0.87, and 0.88, respectively. The predictive ability of the ELF test decreased with time: the 10-year AUCs were 0.78, 0.69, and 0.82, respectively.
Conclusions: The ELF test shows good discriminative performance in predicting liver-related outcomes in a large general population cohort and appears particularly useful for predicting 5-year outcomes in persons with risk factors.
Impact and implications: The Enhanced Liver Fibrosis test exhibits good performance for predicting liver-related outcomes (hospitalisation, liver cancer, or liver-related death) in the general population, especially in those with risk factors.

Introduction

The increasing prevalence and incidence of chronic liver disease presents a major health problem worldwide.[1–3] Non-alcoholic fatty liver disease (NAFLD) and alcohol-related liver disease (ArLD) are the leading causes for advanced liver diseases including liver cirrhosis, HCC and liver-related deaths in Western countries, whereas the role of hepatitis C is decreasing.[4–6] Overall, less than 5% of individuals with NAFLD are expected to develop advanced liver disease during a 20-year follow-up.[7] Nonetheless, because liver cirrhosis tends to develop without symptoms, detection of liver cirrhosis is frequently delayed until the patient develops decompensated disease.[8]

Previous studies have clearly shown that the stage of liver fibrosis is the best predictor of liver-related outcomes in both NAFLD[9–13] and ArLD.[14,15] Therefore. there is an urgent need for accessible instruments for early detection of advanced liver fibrosis in primary care.

Determining fibrosis stage by liver biopsy is invasive and unsuitable for population-level screening.[16] Imaging techniques such as transient elastography are limited by cost and availability. This has led to wide interest in blood-based fibrosis markers as initial screening tests at the population level.[17] Indirect blood indices such as Fibrosis-4 (FIB-4) and AST to Platelet Ratio Index (APRI) have limited ability to detect advanced fibrosis and sub-optimal performance in predicting liver-related outcomes in the community.

The Enhanced Liver Fibrosis® (ELF) test is based on three serum markers of matrix turnover: tissue inhibitor of matrix metalloproteinase-1 (TIMP-1), hyaluronic acid (HA), and amino terminal peptide of pro-collagen III (PIIINP). It was developed in a mixed hepatitis C-dominated patient sample,[18] but has since been widely validated.[19–21] In NAFLD and ArLD, the ELF test shows high diagnostic accuracy for advanced liver fibrosis with AUC values of 0.83 and 0.92, respectively.[20,21]

The ELF test also predicts liver-related outcomes in patients with chronic liver disease (mostly HCV),[22–24] NAFLD with advanced fibrosis or compensated cirrhosis,[25,26] ArLD,[15] primary

Keywords: ELF test; Non-invasive liver fibrosis test; Prognosis; General population; Liver fibrosis; Cirrhosis.
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sclerosing cholangitis,[27,28] and primary biliary cholangitis,[29,30] in some studies even outperforming liver biopsy in this regard. The ELF test has been recommended by the National Institute for Health and Care Excellence (NICE)[31] and EASL[17] as a screening tool for liver fibrosis. Nonetheless, large cohort studies on the predictive performance of the ELF test for liver-related outcomes in an unselected general population are lacking.

In a large health-examination survey representative of the adult general population with linked electronic healthcare registry data, we analysed the predictive performance of the ELF test for liver-related outcomes.

Participants and methods

Health 2000

The study cohort was formed from the Health 2000 study, a multidisciplinary epidemiologic survey performed in Finland in 2000–2001, coordinated by the Finnish Institute for Health and Welfare (previously known as National Public Health Institute). Its use of a regional two-stage stratified cluster sampling procedure ensured that the sample was representative of the entire Finnish population. The Health 2000 study protocol is described in detail elsewhere.[32] Briefly, baseline data were gathered via structured interviews carried out by telephone and at home, and by questionnaires, clinical measurements, blood tests, and clinical examination by a physician. The Epidemiology Ethics Committee of the Helsinki and Uusimaa Hospital Region approved the Health 2000 Study protocol, and all participants provided signed informed consent. The Health 2000 sample collection was transferred to THL Biobank in 2015 after approval of the Coordinating Ethics Committee of the Helsinki and Uusimaa Hospital District.[33]

Of the original sample of 8,028 adults aged 30 years or more, 6,986 were interviewed at their homes, and 6,354 participated in a proper health examination at the study centres. Among them, 6,082 subjects, nearly 80% of individuals of the original sample (79.6 % men, 79.7% women) had complete data including blood samples for ELF analyses. We further excluded 42 individuals with a registry record of liver disease at or before baseline (International Classification of Disease [ICD-10]: K70–K77 or C22.0). The final study cohort thus comprised 6,040 individuals.

Anthropometric measurements, blood pressure measurements, and laboratory tests were obtained as previously described.[32] Average weekly alcohol use in grams of ethanol was assessed by a quantity–frequency questionnaire as described.[32] Alcohol risk use was defined as >210 g/week for men and >140 g/week for women. Binge-drinking was defined as five or more alcoholic drinks per occasion (equal to 60 g of pure ethanol). Exercise was assessed by asking how often the subject performs leisure-time physical exercise for at least 20–30 min so that he/she is at least slightly out of breath and sweaty. Smoking was categorised as current, former, or never smoker.

Diabetes was defined as fasting glucose ≥7.0 mmol/L or HbA1C ≥48 mmol/mol[34] or a previous diagnosis of diabetes or prescription of medication for diabetes. Metabolic syndrome was defined as the presence of at least three of the following components: waist circumference ≥94 cm for men and ≥80 cm for women; serum triglycerides ≥1.7 mmol/L; serum HDL-C <1.0 mmol/L for males or <1.3 mmol/L for females; blood pressure ≥130/85 mmHg or anti-hypertensive medication; and fasting serum glucose ≥5.6 mmol/l or diabetes medication.[35]

The Fatty Liver Index (FLI) was calculated based on BMI, waist circumference, serum triglycerides, and gamma-glutamyltransferase as previously described.[36] NAFLD was defined as an FLI ≥60 with no alcohol risk use.

ELF test

Serum samples were stored at -70 °C. Assays of HA, PIIINP, and TIMP-1 were determined from frozen serum samples in accordance with the manufacturer's instructions (Siemens Healthcare Diagnostics Inc., Tarrytown, NY) using the ADVIA Centaur XPT analyzer (Siemens Healthcare Diagnostics Inc.) at the Biomarkers Team, Finnish Institute for Health and Welfare (Helsinki, Finland). In previous studies ELF score and its components remain stable up to 25 years of storage frozen[37,38] The ELF test result is calculated using the following formula: 2.278 + 0.851 ln(CHA) + 0.751 ln(CPIIINP) + 0.394 ln(CTIMP1); concentrations are in ng/ml. During the measurements, the inter-assay coefficients of variation (CV%) of the ELF score determinations were 0.8% (at the mean level of 7.28 mg/L), 0.4% (at the mean level of 9.15 mg/L), and 0.4% (at the mean level of 11.1 mg/L) according to 3 × 81 control samples analysed at the beginning and end of the daily analysis series.

Outcome data

Follow-up data were obtained from several national registers through linkage using the unique personal identity code assigned to all Finnish residents. Data for hospitalisations were obtained from the Care Register for Health Care (HILMO), which covers all hospitalisations in Finland since 1969. One or several ICD diagnoses are assigned to each hospitalisation at discharge; these diagnosis codes are systematically recorded in the HILMO register. Data for malignancies were obtained from the Finnish Cancer Registry, with nationwide cancer records since 1953. Vital status and cause-of-death data were obtained from Statistics Finland. In Finland, each person who dies is by law assigned a cause of death (in accordance with the ICD) on the official death certificate, issued by the treating physician based on medical or autopsy evidence or based on forensic evidence when necessary; the death codes are then verified by medical experts at the register and recorded according to systematic coding principles. Data reporting to all these registries is mandatory by law and general quality is consistent and virtually 100% complete.[39]

Follow-up was until December 2015. The primary outcome was fatal and non-fatal advanced liver disease requiring hospital admission or causing liver cancer, or liver-related death defined in line with a recent consensus paper[40] and the ICD codes presented in Table S1.

Statistical methods

For comparing groups. we used X^2 or Mann–Whitney tests as appropriate. We analysed cumulative incidences of liver-related outcomes using the Aalen–Johansen non-parametric cumulative incidence function, considering death without liver-related outcomes as a competing-risk event. Here we used previously recommended ELF cut-offs: ≤7.7 for ruling out fibrosis, ≥9.8 for ruling in advanced fibrosis or cirrhosis, and ≥11.3 for ruling in cirrhosis. We also analysed the ELF cut-off of 10.51 recommended by the NICE guidelines.[31] We used Fine and Gray competing-risk regression models to estimate the 10-year absolute risk of liver outcomes over the full spectrum of the ELF

Table 1. Baseline demographics. Data presented as mean (SD), median (IQR) or n (%).

	All individuals	Men	Women	*p* value
	6,040	2,755 (45.6)	3,285 (54.4)	
Age, years	52.7 (14.9)	51.5 (14.0)	53.7 (15.5)	<0.001
Body mass index (kg/m^2)	26.9 (4.7)	27.1 (4.10)	26.8 (5.1)	0.019
Waist circumference (cm)	92.7 (13.3)	97.8 (11.5)	88.4 (13.2)	<0.001
Waist–hip ratio	0.91 (0.08)	0.97 (0.06)	0.86 (0.06)	<0.001
Systolic blood pressure (mmHg)	135 (21.1)	136 (19.0)	134 (22.7)	0.001
Diastolic blood pressure (mmHg)	81.7 (11.1)	84.1 (10.9)	79.7 (10.8)	<0.001
Diabetes	596 (9.9)	289 (10.5)	307 (9.3)	0.149
Metabolic syndrome	2,706 (44.8)	1,321 (47.9)	1,385 (42.2)	<0.001
Alcohol use				<0.001
Lifetime abstainer	950 (16.4)	195 (7.3)	755 (24.2)	
Current abstainer	281 (4.9)	175 (6.6)	106 (3.4)	
Alcohol user	4,547 (78.7)	2,291 (86.1)	2256 (72.4)	
Alcohol consumption (g/week)	20.0 (0.0–80.9)	60.9 (7.0–162.0)	7.0 (0.0–38.9)	<0.001
Binge drinking frequency*	0 (0–7)	3 (0–18)	0 (0–2)	<0.001
Exercise†				0.134
At least two times a week	3,455 (58.8)	1,550 (57.5)	1,905 (60.0)	
Two to four times a month	1,643 (28.0)	775 (28.7)	868 (27.3)	
Less often	774 (13.2)	372 (13.8)	402 (12.7)	
Smoking status, n (%)				<0.001
Current	1,593 (26.5)	902 (32.9)	691 (21.1)	
Former	1,313 (21.8)	858 (31.3)	455 (13.9)	
Never	3,109 (51.7)	985 (35.9)	2,124 (65.0)	
Alanine transaminase (U/L)	20 (15–29)	26 (19–37)	17 (13–23)	<0.001
Aspartate transaminase (U/L)	25 (22–31)	28 (24–33)	24 (21–28)	<0.001
Gamma-glutamyltransferase (U/L)	24 (17–38)	23 (17–37)	24 (17–39)	0.378
Glycated haemoglobin (%)	5.3 (0.7)	5.3 (0.7)	5.4 (0.7)	<0.001
HOMA-IR‡	2.5 (5.4)	2.7 (7.1)	2.3 (3.3)	0.001
Fatty Liver Index	46.7 (27.0)	51.5 (25.3)	42.6 (27.8)	<0.001

* The number of times during the last 12 months that the subject had consumed five or more drinks per day.
† Exercise 20–30 min slightly out of breath and sweaty.
‡ Homeostasis model assessment for insulin resistance.

score, separately for men and women, and adjusted for age. The association between the ELF score and liver-related outcomes was tested using Cox regression analyses with the ELF score as a single covariate (continuous or categorical) and time to first liver event as the outcome. A possible non-linear association was analysed using restricted cubic splines. The proportional-hazards assumption of the Cox models was checked using Schoenfeld residuals, and no violation was detected. The predictive performance of the ELF score in terms of discrimination was assessed by Harrell's C-statistic, and, based on cause-specific Cox regression models with death without liver-related outcomes as a competing-risk event, by the time-dependent AUC metric.[41,42] We quantified overall predictive ability by Royston and Sauerbrei's R^2_D.[43]

The predictive performance of the ELF score was assessed for the entire follow-up, and with follow-up truncated at 5 or 10 years. We calculated sensitivity, specificity, positive and negative predictive values (PPV and NPV), and likelihood ratios (LR+ and LR-) for liver outcomes at 5 and 10 years of follow-up using various ELF cut-offs. Subgroup analyses were performed using

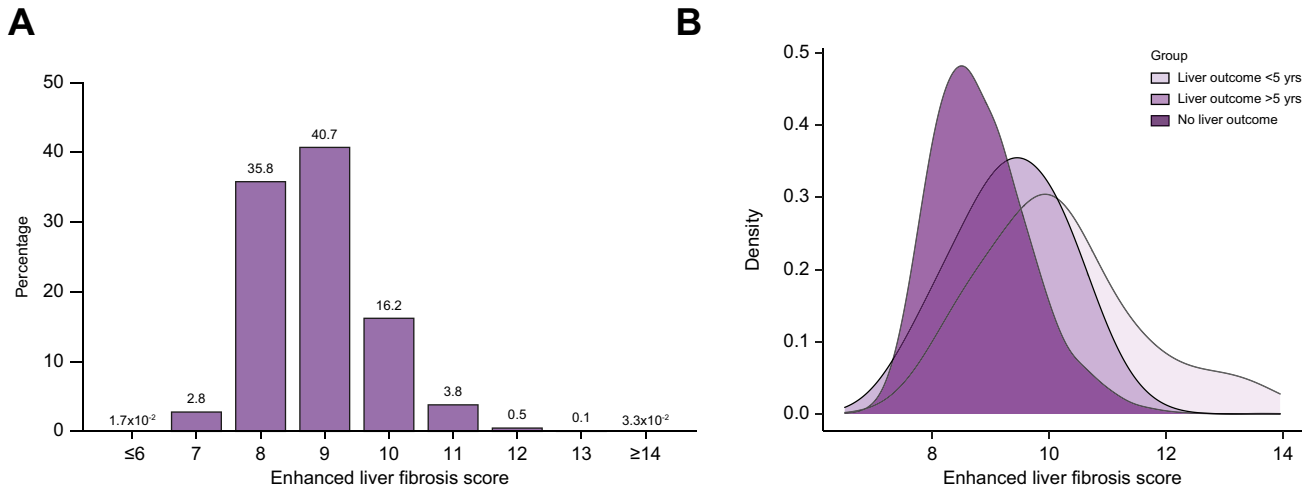

Fig. 1. Distribution of the Enhanced Liver Fibrosis test. (A) overall distribution and (B) according to development of liver-related outcomes during follow-up.

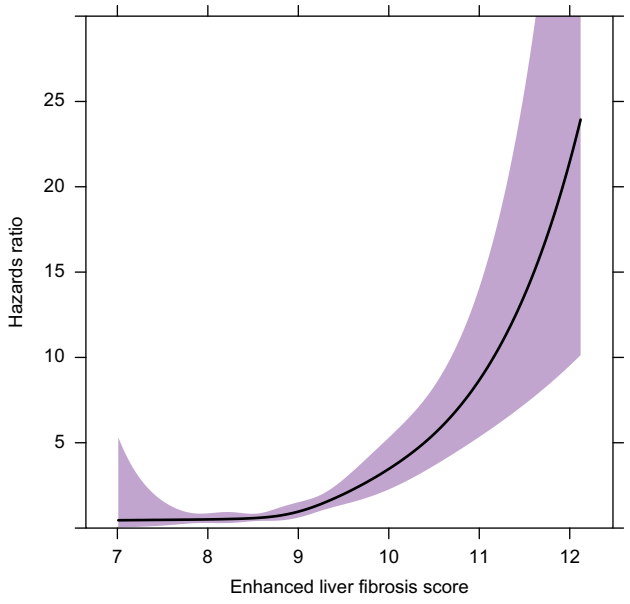

Fig. 2. Non-linear association between the Enhanced Liver Fibrosis test and liver-related outcomes. Cox regression, and performance measures. AUC values were calculated using competing-risk methodology.

the following baseline variables: median age, sex, diabetes, BMI (18.5–24.9, 25–29.9, ≥30 kg/m^2), alcohol risk use, NAFLD, and alanine aminotransferase (ALT) >40 IU/L.

We assessed calibration by comparing the predicted risk of liver-related outcomes with the observed risk using calibration plots when accounting for the competing risk of death without liver disease. Data were analysed with R software version 3.6.1 using the packages survival, rms, car, tableone, pec, CPE, survAUC, timeROC, survcomp, and riskRegression (R Foundation for Statistical Computing, Vienna, Austria).

Results

The study cohort comprised 6,040 individuals, with 46% men. mean age 52.7 years, mean BMI 26.9 kg/m^2, and a prevalence of diabetes of 10% (Table 1). Compared with women, men had larger waist circumference (97.8 cm *vs.* 88.4 cm) and greater prevalence of metabolic syndrome (47.9% *vs.* 42.2%) and consumed more alcohol and were less often never smokers (35.9% *vs.* 65.0%) (Table 1).

During a median follow-up of 13.1 years (IQR 12.9–13.2 years) (72,387.5 person-years), there were 67 first liver-related outcomes. Among the 67 cases, there were seven cases of alcoholic hepatitis (ICD-10 K70.1), and one acute liver failure (ICD-10 K72.0).

The mean ELF score at baseline was 8.85 (SD 0.86, median 8.74, IQR 8.22–9.36) (Fig. 1). Men had slightly higher scores (mean 8.88. SD 0.80, median 8.79, IQR 8.30–9.34) compared with women (mean 8.82, SD 0.90, median 8.68, IQR 8.16–9.37). ELF scores were lower among individuals with no liver event during follow-up (mean 8.84, SD 0.85, median 8.73, IQR 8.22–9.34) compared with those with a liver event within 5 years (mean 10.2, SD 1.35, median 10.0, IQR 9.39–10.6) or after 5 years (mean 9.34, SD 0.92, median 9.40, IQR 8.72–9.99) (Fig. 1).

By univariable Cox regression analysis, ELF as a continuous variable was significantly associated with liver-related outcomes (hazard ratio [HR] 2.70, 95% CI 2.16–3.38, p <0.001) (Table 2 and Fig. 2), with no significant nonlinearity observed for this association (p = 0.622). In a Cox model adjusted for age, sex, waist circumference, BMI, diabetes, weekly alcohol use, binge-drinking

Table 2. Cox regression with ELF as a covariate and time to the first event as the outcome overall and in various subgroups.

	n/N	HR (95% CI)	aHR* (95% CI)	AUC at 5 years (95% CI)	AUC at 10 years (95% CI)
ELF continuous	67/6040	2.70 (2.16–3.38)	3.34 (2.54–4.39)	0.81 (0.71–0.91)	0.71 (0.63–0.79)
ELF categorical					
<9.8	40/5243	Reference	Reference		
9.8–11.2	22/741	5.30 (3.14–8.94)	6.44 (3.37–12.29)		
≥11.3	5/56	24.24 (9.51–61.80)	24.37 (8.55–69.50)		
<10.51	57/5773	Reference	Reference		
≥10.51	10/267	6.09 (3.10–12.00)	4.84 (2.24–10.47)		
Cox regression in subgroups					
Age <51 yr	23/2971	3.43 (1.98–5.95)	2.69 (1.46–4.96)	0.85 (0.71–0.98)	0.72 (0.57–0.87)
Age ≥51 yr	44/3069	2.84 (2.11–3.80)	3.48 (2.60–4.67)	0.80 (0.66–0.94)	0.70 (0.59–0.80)
Men	50/2755	2.89 (2.23–3.76)	3.75 (2.78–5.06)	0.80 (0.68–0.92)	0.72 (0.62–0.82)
Women	17/3285	2.32 (1.47–3.66)	2.09 (1.09–4.00)	0.85 (0.65–100)	0.67 (0.50–0.84)
Diabetes	22/596	2.48 (1.67–3.69)	2.68 (1.80–3.98)	0.87 (0.77–0.97)	0.69 (0.54–0.84)
No diabetes	45/5444	2.35 (1.75–3.17)	3.22 (2.17–4.77)	0.75 (0.62–0.88)	0.68 (0.57–0.78)
BMI 18.5–24.9	20/2207	2.61 (1.76–3.88)	2.28 (1.26–4.11)	0.82 (0.69–0.94)	0.71 (0.60–0.83)
BMI 25–29.9	24/2436	2.17 (1.48–3.18)	3.26 (2.15–4.93)	0.82 (0.64–0.99)	0.69 (0.54–0.84)
BMI 30+	21/1344	3.99 (2.53–6.29)	4.73 (2.86–7.82)	0.85 (0.64–100)	0.78 (0.64–0.92)
ALCO risk†	29/705	2.88 (2.09–3.97)	2.60 (1.80–3.77)	0.80 (0.63–0.98)	0.71 (0.57–0.85)
Non-RISK ALCO	36/5067	2.88 (2.15–3.85)	3.88 (2.66–5.66)	0.87 (0.78–0.96)	0.75 (0.65–0.85)
NAFLD‡	16/1635	3.42 (2.07–5.65)	3.48 (1.87–6.47)	0.86 (0.65–100)	0.75 (0.61–0.89)
ALT >40 U/L	30/740	3.31 (2.42–4.53)	2.94 (1.96–4.43)	0.88 (0.81–0.95)	0.82 (0.72–0.92)

* Adjusted for age and sex.
† 210 g/week for men and >140 g/week for women.
‡ Fatty Live r Index ≥60 and non-risk alcohol drinker.

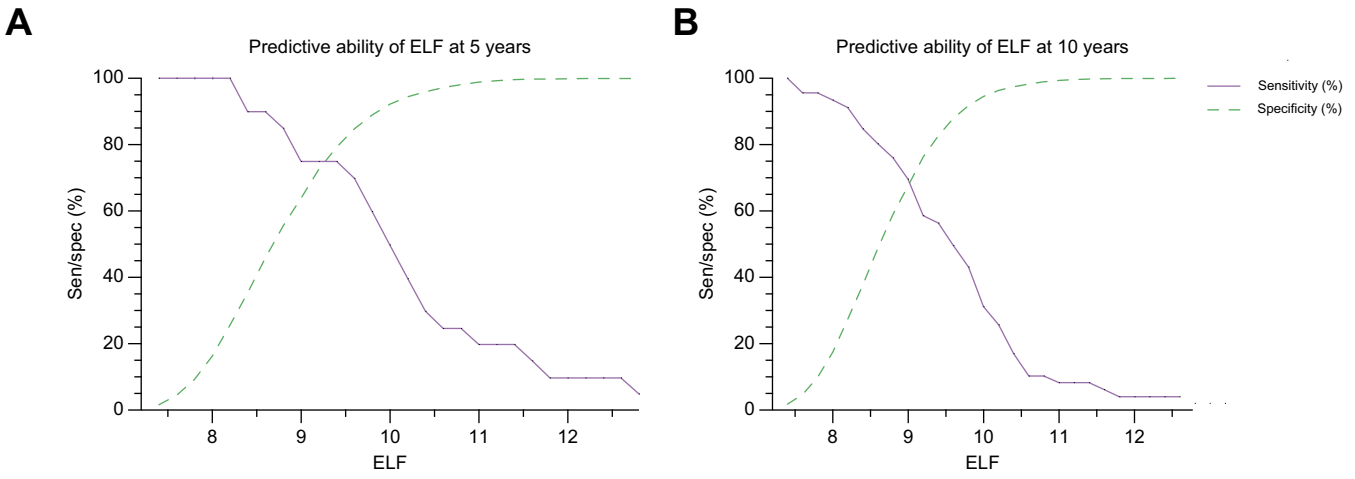

Fig. 3. Sensitivity and specificity of Enhanced Liver Fibrosis for predicting serious liver outcomes. (A) Five years. (B) Ten years.

frequency, and smoking status. ELF remained independently and strongly associated with liver events (HR 2.49, 95% CI 1.82–3.40, $p < 0.001$).

When excluding cases of alcoholic hepatitis and acute liver failure (total n = 8), ELF as a continuous variable remained significantly associated with liver-related outcomes both in unadjusted analysis (HR 2.92, 95% CI 2.31–3.68, p <0.001) and in analysis adjusted for age and sex (HR 3.54 95% CI 2.68-4.70, p <0.001).

Harrell's C-statistic as a measure of the ability of ELF to predict liver-related outcomes in the univariable Cox model was 0.82 at 5 years and 0.73 at 10 years (Fig. 2). Accounting for the competing risk of death without liver disease using cause-specific Cox regression, the time-dependent AUC was 0.81 (95% CI 0.71–0.91) at 5 years and 0.71 (95% CI 0.63–0.79) at 10 years (Table 2 and Fig. S1). Royston and Sauerbrei's R^2_D as a measure of the overall predictive value of ELF for liver-related outcomes was 0.80 (95% CI 0.64–0.90). Regarding 10-year risks for liver-related outcomes, ELF showed fairly good calibration for predictions up to 7.5% but overestimated the risk above that (Fig. S1).

Performance measures of ELF for predicting liver-related outcomes at different cut-offs are shown in Table S2. The PPV at 10 years increased above 5% at the ELF cut-off of 10.2. while the 10-year NPV remained over 99% at all cut-offs. The optimal cut-off points for maximising sensitivity and specificity of predictive ability of ELF at 5 and 10 years are 9.5 and 9.0, respectively (Fig. 3).

Based on the Aalen–Johansen non-parametric competing-risk method, cumulative incidences of liver-related outcomes among individuals with ELF <9.8 were 0.2% at 5 years and 0.5% at 10 years, with ELF 9.8–11.3, 1.1%, and 2.2%, and with ELF ≥11.3, 7.1%, and 7.1%, respectively (Fig. 4). With ELF ≥10.51, 5- and 10-year cumulative incidences were 2.2% and 2.6%, respectively.

Based on Fine–Gray competing-risk regression, age-adjusted absolute 5-year risks (Fig. 5) and 10-year risks for liver-related outcomes increased along the entire spectrum of ELF scores, being higher among men than among women (Fig. 5). Ten-year risks above 5% were seen at ELF scores above 10.5 among men and above 11.5 among women. Higher absolute risks were also noticed along the entire spectrum of ELF scores among individuals with either alcohol risk use or diabetes (Fig. 5).

In the various subgroups, AUC values ranged from 0.75 to 0.88 at 5 years and from 0.67 to 0.82 at 10 years (Table 2). Discriminative ability was excellent among individuals with an elevated ALT (>40 U/L) or diabetes, with AUCs at 5 years being 0.88 and 0.87, and at 10 years, 0.82 and 0.69, respectively. Covariate-specific AUC analysis further showed that the discriminative performance of ELF increased with increasing BMI and increasing ALT, but decreased among alcohol risk drinkers, and decreased at both ends of the age range (Fig. S2). The discriminative ability of ELF decreased with increasing follow-up time, and we found a substantial decrement after 5 years (Table S3): AUCs at 5 years and 6 years were 80.6 and 73.6, respectively. After excluding cases of alcoholic hepatitis and acute liver failure, the predictive ability of ELF increased slightly, AUC at 5 and 10 years were 0.84 (0.73–0.94) and 0.73 (0.65–0.81), respectively.

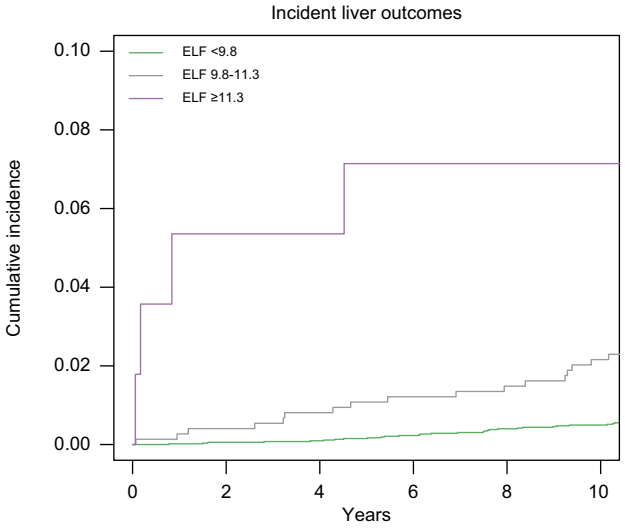

Fig. 4. Cumulative incidence of liver-related outcomes by Enhanced Liver Fibrosis category. Non-parametric Aalen–Johansen method, considering death without liver disease as a competing-risk event.

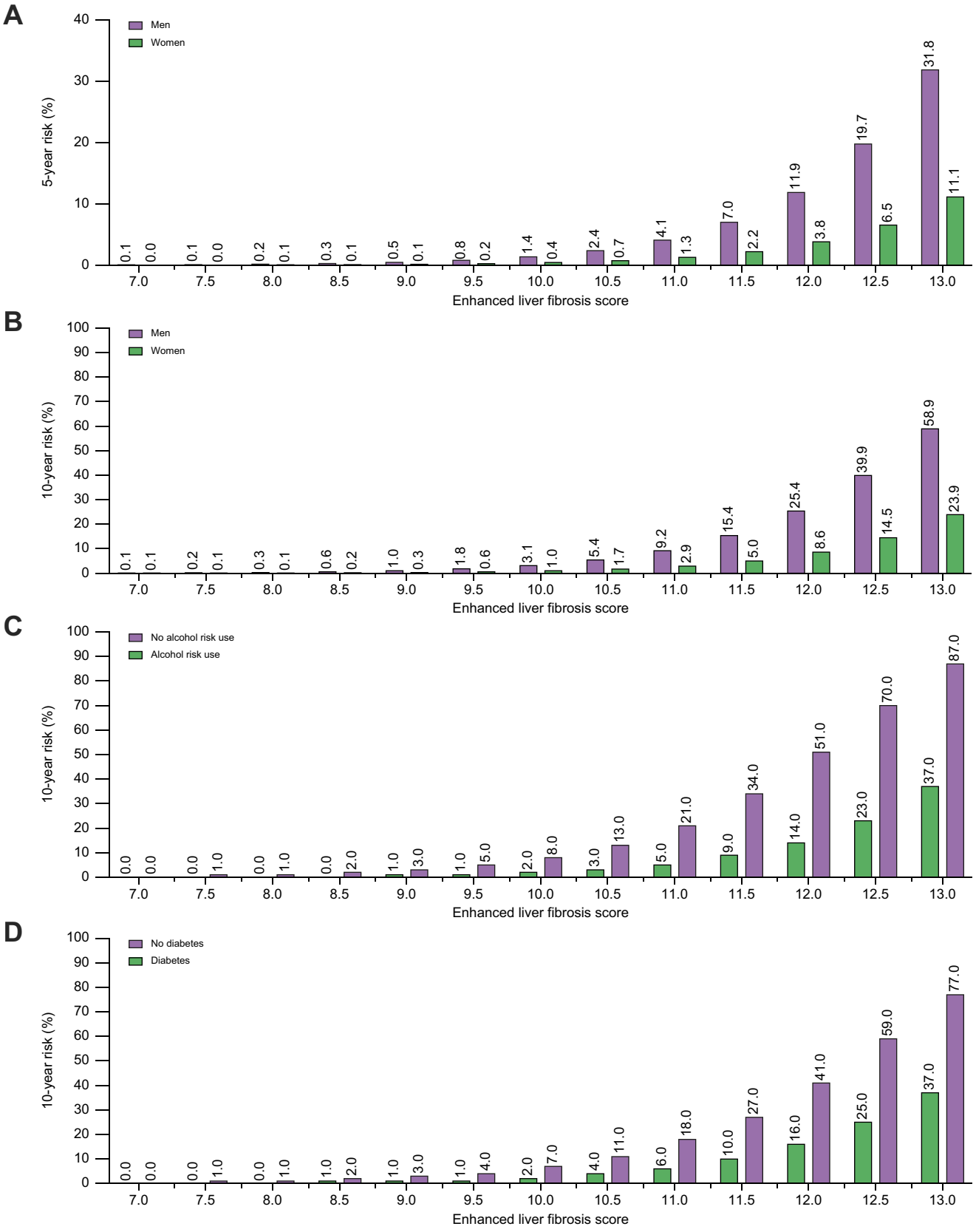

Fig. 5. Risks for liver-related outcomes by Enhanced Liver Fibrosis category at different time points separated by sex, alcohol risk use, and diabetes. (A) Absolute 5-year risks of liver-related outcomes according to the ELF score separately by sex. (B) Absolute 10-year risks of liver-related outcomes according to the ELF score separately by sex, (C) alcohol risk use (weekly >210 g men and >140 g women), (D) and diabetes. Analyses were made using Fine–Gray regression adjusted for age and sex.

Of the individual components of ELF, HA showed the highest C-statistic (0.73. 95% CI 0.67–0.79) but poorer model fit than ELF ($p < 0.001$, likelihood ratio test) (Table S4).

Discussion

In the present analysis of the ELF test in predicting liver-related clinical outcomes in an unselected general population cohort with long follow-up, we found that the ELF test had good discriminative performance with AUC values of 0.82 at 5 years and 0.73 at 10 years. The absolute risk for liver-related outcomes at 10 years increased from 0.5% for ELF test scores <9.8 to 7.1% for ELF ≥11.3. Discriminative performance at the 5-year perspective was excellent among individuals with ALT >40 U/L, diabetes, or obesity (AUC values 0.85–0.88). In addition, the ELF test emerged as an independent risk factor for liver-related outcomes in multivariable analysis.

When excluding eight patients with acute liver incidents, the discriminative capacity of ELF increased slightly at 5 years (AUC 0.82–0.83) and remained stable at 10 years (AUC 0.73). Liver histology is almost invariably associated with advanced liver fibrosis or cirrhosis among patients with alcohol hepatitis. In the Steroids or Pentoxifylline for Alcoholic Hepatitis (STOPAH) trial, the majority had advanced fibrosis or cirrhosis.[44]

In the unselected population, advanced liver fibrosis is a rare finding. In our cohort, the number of individuals with ELF values ≥9.8, >10.51, and ≥11.3 were 797 (13.2%), 260 (4.3%), and 56 (0.9%), respectively. Therefore, PPV at 10 years for liver-related outcome remained low, 4.7%. at the cut-off of ≥10.51 recommended by NICE. The prevalence of diabetes in our cohort was similar to that reported by recent guidelines,[34] 9.9%. In Health 2000, the prevalence of type I diabetes was 0.5%.[32] The prevalence of fatty liver disease defined by FLI ≥60 was about 30%. Similar high prevalence numbers have been detected before in European countries.[45]

Although several cross-sectional studies have analysed the performance of the ELF test in discriminating advanced liver fibrosis,[20,23,46] longitudinal data on the predictive performance of the ELF test for liver-related outcomes are scarce. Studies involving mainly selected patients with NAFLD with advanced fibrosis or cirrhosis,[25,26] ArLD,[15] PSC,[27,28] or PBC[29,30] reported good discrimination of the ELF test for liver-related outcomes. The mean follow-up in these studies ranged from 1 to 9 years and involved a higher proportion of individuals with advanced liver fibrosis compared with that in our population-based study. A small case-control study (n = 120) based on the Singapore Chinese Health Study reported an AUC value of 0.89 for incident HCC for a model including ELF, age, sex, and dialect group.[47] Another population-based case-control study (n = 173) comprising post-menopausal women with risk factors for liver disease found an AUC of 0.58 for liver-related outcomes.[48]

Lack of data on platelet counts in our study prevented a head-to-head comparison of the ELF test with other non-invasive fibrosis tests such as FIB-4 or APRI. In cohorts of NAFLD and ArLD patients, the ELF test has previously been shown to outperform other non-invasive tests (NITs) in predicting liver-related outcomes.[15,26]

The strengths of our study include the large general population cohort, long follow-up, and linkage with reliable electronic healthcare registers for liver-related clinical outcomes. The national health care registers in Finland have virtually 100% coverage and negligible loss to follow-up.[39] The percentage of participants in the Health 2000 study with complete data and blood samples was high, almost 80 % (79.6 % men, 79.7 % women). No major differences could be seen between different socioeconomical groups, and the final cohort can be considered as representative of the whole Finnish population.[49]

Study limitations include an uncertainty in risk estimates when dealing with rare outcomes, and the inability to compare the performance of the ELF test with other similar NITs. There were 67 liver outcomes during the follow-up, that is 1.1% of the cohort. We acknowledge that reliance on registry linkage omits undiagnosed liver disease and less severe cases that may have been largely managed in primary care, but we specifically sought to examine complicated liver disease, not subclinical liver fibrosis.

We found that the discriminative performance of the ELF test decreased along with longer follow-up. This finding is inherent to any liver fibrosis test, including liver biopsy, because these tests only reflect the baseline disease status (stage of liver fibrosis), not the drivers of disease progression. Although fibrosis stage reflects short-term risks for clinical liver outcomes, long-term risks are more dependent on disease drivers such as harmful alcohol use and diabetes. It is likely therefore that repeated measurements of ELF would improve risk stratification. We noticed a substantial decrease in the discriminative ability of the ELF test after 5 years. To have an updated risk assessment for liver events, it would be logical to repeat ELF measurements at 5-year intervals.

Interestingly, performance of the ELF test tended to improve with increasing BMI. This contrasts with transient elastography, where reliable results may be more difficult to obtain in obese individuals.[50] Despite of introduction of the XL-probe, a failure rate of 5–10% has been reported among obese individuals,[51,52] and accuracy is lower among extremely obese people.[53] The ELF test could be particularly useful in obese individuals.

The risk for liver-related outcomes at any given ELF test value was higher among men than among women, in line with previous findings regarding other NITs.[54] Higher mean ELF values among males have been reported in a study with healthy blood donors.[55] This may call for sex-specific cut-offs of ELF test scores when evaluating future liver-related risks.

Despite the good discrimination of the ELF test, 60% (40/67) of the liver-related outcome events still occurred among individuals with an ELF value <9.8 at baseline. Therefore, ELF test alone cannot be used as an only method for assessing the risk for serious liver outcomes. The same issue was also observed with other NITs (*e.g.* FIB-4 and APRI) in previous population-based studies[54,56] and may reflect a need to incorporate data on risk factors and drivers of disease progression when making long-term predictions of incident clinical liver disease in the population.

In conclusion. a single ELF test provides good predictive value for liver-related outcomes in an unselected population cohort, especially among those with risk factors for liver disease, but discriminative performance decreases with longer follow-up. There are sex-related differences, with the risk for liver-related outcomes being higher among men at any given ELF level.

Abbreviations

aHR, adjusted hazard ratio; ALT, alanine aminotransferase; APRI, AST to Platelet Ratio Index; ArLD, alcohol-related liver disease; AST, aspartate aminotransferase; ELF, Enhanced Liver Fibrosis® test; FIB-4, Fibrosis-4; FLI, Fatty Liver Index; HA, hyaluronic acid; HCC, hepatocellular carcinoma; HR, hazard ratio; ICD, International Classification of Diseases; NAFLD, nonalcoholic fatty liver disease; NICE, National Institute for Health and Care Excellence; NITs, non-invasive tests; NPV, negative predictive value; PIIINP, amino terminal peptide of pro-collagen III; PPV, positive predictive value; TIMP-1, tissue inhibitor of matrix metalloproteinase-1.

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Conflicts of interest

The authors declare that they have no conflicts of interest regarding the content of this manuscript.

Please refer to the accompanying ICMJE disclosure forms for further details.

Authors' contributions

Conceptualisation: KS, MF, AJ, AL, FÅ. Investigation: AJ, AL. Data curation: AJ, AL, FÅ. Formal analysis: IE, TV, FÅ. Writing – original draft: KS. Writing – review & editing: KS, MF, AJ, IE, TV, AL, FÅ. Funding acquisition: FÅ; Supervision: FÅ.

Data availability statement

FINRISK and Health 2000 data are available from the THL Biobank based on a research application, as explained on the website of the THL biobank (https://thl.fi/en/web/thl-biobank/for-researchers).

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Supplementary data

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Author names in bold designate shared co-first authorship

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