Peer Review File

Article information: Available at http://dx.doi.org/10.21037/atm-20-2444

Reviewer Comments:

The authors probed that non-invasive markers of liver fibrosis, γ-GTP to platelet ratio and Fibrosis index, were able to predict subsequent esophageal varices rupture within 1 and 2 years in patients with compensated liver cirrhosis, focusing on those with initial small EV without red-color sign (RCS), without use of non-selective beta-blockers (NSBB) or endoscopic variceal ligation (EVL). The study cohort was large enough to provide reliable conclusions. But, I should make several questions and wait for your answer.

Comments 1:

In Results section, the authors included 8310 compensated liver cirrhosis patients. However, how did the authors exclude decompensated cirrhosis patients? How to exclude patients with ascites and hepatic encephalopathy? Did the authors exclude ascites and hepatic encephalopathy using ICD-10?

Reply 1:

Thanks for the comment. The definition of decompensated liver cirrhosis depends on whether the patient had variceal bleeding, ascites or hepatic encephalopathy according to the AASLD guideline[1]. Therefore, we excluded patients with ascites, hepatic encephalopathy, or initial EV bleeding using ICD-9 or ICD-10 diagnostic codes or by means of abdominal ultrasound, abdominal computed tomography or EGD reports.

Code for ascites: 789.5 (ICD-9); R18, K70.31, K71.51 (ICD-10)

Code for hepatic encephalopathy: 348.3, 572.2 (ICD-9); G93.4 (ICD-10)

Code for EV bleeding: 456.20,530.82,456.0 (ICD-9); I85.11, I85.01 (ICD-10)

Change in the text:

We have modified our text as advised in the material and method section (Please see patient selection on page 9-10; and diagnostic criteria section on page 11).

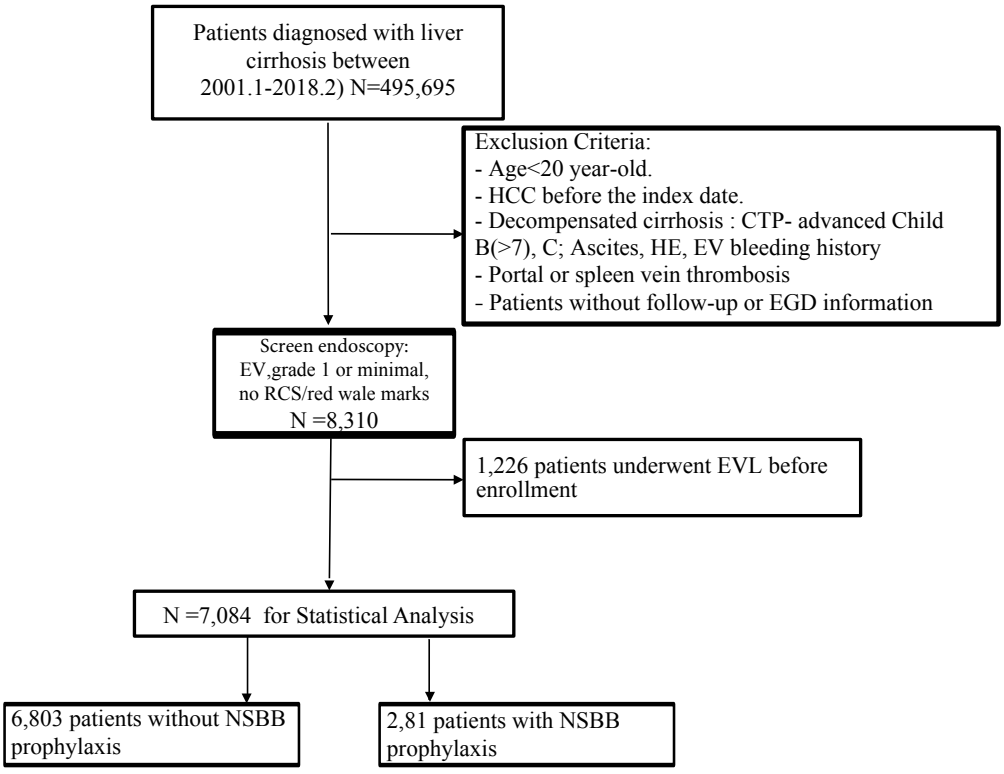
Comment 2.

In Materials and Methods, Child B and C cirrhotic patients should be excluded. However, patients with Child score 7 were included in the major cohort in Table 1.

Reply 2:

Thanks for the comment. In this study, we excluded patients with decompensated cirrhosis who had ascites, hepatic encephalopathy or esophageal variceal bleeding history. Therefore, some early Child B (B7) patients could also be enrolled (according to a previous study by Francesca Romana Ponziani et al. They defined Child A and B7 as early stage liver cirrhosis. World J Gastroenterol. 2013 Jun 7; 19(21): 3255–3262). We correct the mistake in the text and in the flowchart as follows:

Figure 1. Enrollment flowchart



Change in the text: We have modified our Figure 1 and text as advised(please see page 10, line 1-4).

Comment 3.

In Results, the authors stated that "The HR in EVB for patients not taking NSBB was significantly lower than that in patients taking NSBB." However, data in Table 4 strongly suggested that liver fibrosis in NSBB patients were more progressed that that of non-NSBB patients according to total bilirubin, albumin, platelet count, GPR, Fib-4 index. The baseline characteristics were too different between NSBB and non-NSBB patients to lead a conclusion.

Reply 3:

Thanks for the comment. We have calculated p-values comparing the non-NSBB group to the NSBB group in Table 4A as shown below. There were no significant differences between the non-NSBB and NSBB patients according to total bilirubin, albumin, platelet count, creatinine, INR, and Fib-4 index. In addition, there were no significant differences in the baseline CTP, MELD, PALBI, APRI, Log score, King's score, and Forn's index between the two groups. Age, sex, and Fibrosis index (FI) were also matched between the two groups because we wanted to test whether FI could differentiate patients with higher risk of EV bleeding and mortality. However, the Hb, WBC, r-GT, GPR, GAR, and AST/ALT ratio were significant different between the two groups. Therefore, we would mention in the text that the baseline characteristics were similar between NSBB and non-NSBB patients according to the majority of the important chronic liver disease scores such as CTP, MELD, PALBI, APRI, Log score, King's score, Fib-4 and Forn's index. However, the limitation is that a few important liver fibrosis scores such as GPR, GAR, and AST/ALT ratio were significant different between the two groups. There is a need for further prospective studies that match all these scores in order to compare between the two groups.

Table 4A. Selected patients with matched sex, age and FI score from the major and the minor group respectively for EV bleeding analysis

A.

Variables	**Non-NSBB (matched, n=183)**	**NSBB (matched, n=183)**	***p*-value**
Parameter			
Male (n, %)	148 (80.87)	148 (80.87)	1.00
Age (years, mean ± SD)	54.68 ± 11.71	54.68 ± 11.71	1.00
Etiology, n, (%)			<0.01
HBV	80 (43.71)	76 (41.53)	
HCV	51 (27.87)	29 (15.85)	

Alcohol	26 (14.21)	48 (26.23)	
Others	26 (14.21)	30 (16.39)	
Follow-up duration (months, mean ± SD)			
To EV bleeding	10.46 ± 3.78	3.97 ± 3.57	<0.01
Outcome			
Esophageal variceal bleeding in 1 year (n, %)	22 (12.02)	160 (87.43)	<0.01
Baseline laboratory value (mean ± SD)			
AST (U/L)	89.14 ± 88.74	95.19 ± 94.44	0.53
ALT (U/L)	58.24 ± 60.64	51.29 ± 49.73	0.23
Cr (mg/dL)	1.39 ± 1.64	1.48 ± 1.75	0.62
Na (mEq/L)	136.50 ± 9.61	136.80 ± 5.73	0.72
K (mEq/L)	3.93 ± 0.56	4.09 ± 1.44	0.18
Bilirubin-total (mg/dL)	3.46 ± 5.55	2.85 ± 3.34	0.20
Albumin (g/dL)	3.21 ± 0.71	3.10 ± 0.64	0.12
PT-INR	1.36 ± 0.34	1.35 ± 0.28	0.89
Hb (g/dL)	10.91 ± 2.54	10.08 ± 2.57	<0.01
WBC (×1000/μL)	6.31 ± 3.77	7.23 ± 4.37	0.03
Platelet (×1000/μL)	107.20 ± 57.99	118.00 ± 70.84	0.11
r-GT	172.50 ± 269.30	279.30 ± 380.30	0.01
Cholesterol	151.40 ± 48.76	161.00 ± 49.88	0.17
Prognostic systems (mean ± SD)			
CTP score	6.80 ± 1.67	6.89 ± 1.37	0.61
MELD score	14.65 ± 6.63	14.43 ± 5.19	0.74
MELD-Na score	16.49 ± 8.80	15.54 ± 6.41	0.27
PALBI score	-2.54 ± 0.73	-2.51 ± 0.69	0.74
PALBI grade 1 (n, %)	100 (54.64)	92 (50.27)	0.41
PALBI grade 2 (n, %)	39 (21.31)	50 (27.32)	
PALBI grade 3 (n, %)	44 (24.04)	41 (22.40)	
Spleen diameter	5.97 ± 1.15	6.11 ± 1.19	0.39
GUCI	4.90 ± 7.98	4.54 ± 5.94	0.64
Gamma-glutamyl transpeptidase-to-platelet ratio (GPR)	2.68 ± 3.62	3.88 ± 5.26	0.04
Gamma-glutamyl transpeptidase-to-albumin ratio (GAR)	5.41 ± 8.17	9.00 ± 11.47	<0.01
AST/ALT ratio	1.87 ± 1.43	2.36 ± 2.23	0.01
AST to platelet ratio index (APRI)	3.27 ± 3.81	3.26 ± 4.03	0.98

Platelet count to spleen diameter (PC/SD)	18.78 ± 10.70	20.29 ± 13.39	0.37
Fibrosis-4-index (FIB-4)	7.75 ± 6.52	8.53 ± 8.91	0.34
Fibrosis index (FI)	3.72 ± 0.95	3.72 ± 0.95	0.99
King's Score	89.47 ± 172.65	80.07 ± 99.28	0.54
Log score	3.09 ± 2.86	3.53 ± 3.48	0.20
Lok index	0.84 ± 0.18	0.88 ± 0.17	0.07
Portal vein size	1.14 ± 0.25	1.15 ± 0.26	0.93
Forn's index	10.77 ± 1.82	10.81 ± 1.72	0.88

Change in the text: We have modified our Table 4A and text as advised (please see result section: Comparing between the matched non-NSBB group and the NSBB group for EV bleeding analysis on page 17, line 10-13, and in the fourth limitation part of discussion section on page 26-27)

Comment 4.

In Table 4A, the authors should calculate p values comparing non-NSBB and NSBB.

Reply 4:

Thanks for the comment. We have calculated p-values comparing non-NSBB and NSBB in Table 4A as shown above (please see Table 4A in **Reply 3**).

Change in the text: We have calculated p-values comparing non-NSBB and NSBB in Table 4A.

Comment 5.

In Table 4B, the authors should spell out 'LB' and 'UB'. In Cox proportional hazard model analysis, HR, 95% confidence interval and p values should be described.

Reply 5:

Thanks for the comment. We had changed the table setting: deleted the lower and upper bound (LB and UB) of 95% CI into 95% confidence interval. We supposed the hazard ratio (HR) for EVB in patients taking NSBB was 1.000 (the reference group) so the HR for EVB in patients with non-NSBB was calculated as shown below.

Table 4B. Hazard ratio for EVB between the above patients in the no NSBB and the NSBB group respectively

Variable	Hazard Ratio	95% Confidence Interval	P-value
No NSBB	0.054	0.034-0.087	<0.001
NSBB	1.000		

Change in the text: We have modified Table 4B as the above mentioned.

Comment 6.

In Table 3, which parameters were subject to multivariate analysis? Did the authors evaluate GPR and FI alone in the multivariate analysis without including age, gender or etiology?

Reply 6:

Thanks for the comment. In Table 3, all the parameters listed in univariate Cox regression were carried into the initial multivariable analysis. However, after the stepwise Cox regression analysis, non-significant factors like age, gender or etiology were excluded based on stepwise model selection. Therefore, there were only GPR and Fibrosis index retained in our final multivariate model.

Table 3. Univariable and multivariable Cox regression analysis for prediction of EV bleeding within 1 year

Variables	**Univariate**			**Multivariate (stepwise)**		
	crude HR	95% CI	*p* value	Adjusted HR	95% CI	*p* value
Age	0.99	0.98-0.99	<0.01			
Sex						
Male	1.00					
Female	0.81	0.67-0.98	0.03			
Etiology						
HBV	0.66	0.52-0.84	<0.01			
HCV	0.83	0.65-1.06	0.14			
Alcohol	1.26	0.96-1.66	0.09			
Others	1.00					
Prognostic systems						
CTP score	1.24	1.16-1.31	<0.01			
MELD score	1.04	1.02-1.05	<0.01			
MELD-Na score	1.03	1.01-1.03	<0.01			
PALBI score	1.55	1.36-1.75	<0.01			
Spleen diameter	1.13	1.03-1.22	0.01			
GUCI	1.00	0.99-1.00	0.47			
Gamma-glutamyl transpeptidase-to-platelet	1.03	1.01-1.04	<0.01	1.05	1.03-1.67	< .001

ratio (GPR)						
Gamma-glutamyl transpeptidase-to-albumin ratio (GAR)	1.00	0.99-1.00	0.47			
AST/ALT	1.15	1.11-1.19	<0.01			
AST to platelet ratio index (APRI)	1.00	1.00-1.01	0.06			
Platelet count to spleen diameter (PC/SD)	0.99	0.97-0.99	0.00			
Fibrosis-4-index (FIB-4)	1.00	1.00-1.01	0.01			
Fibrosis index (FI)	1.38	1.26-1.50	<0.01	1.48	1.21-1.83	< .001
King's Score	1.00	1.00-1.00	0.59			
Lok score	1.04	1.02-1.05	<0.01			
Lok index	6.92	3.89-12.28	<0.01			
Portal vein size	2.42	1.35-4.33	<0.01			
Forn's index	1.10	1.03-1.16	<0.01			

HR, hazard ratio; CI, confidence interval

Change in the text: We have modified Table 3 and our text as advised (please see the result section: prediction of EVB within 1 year and 2 years by Cox regression analyses, on page 15, line 11-14).

Comment 7.

In the minor cohort, how did the authors confirm that the aim of NSBB prescription was prophylaxis of esophageal varix rupture? Didn't the patients need NSBB for hypertension, chronic heart failure or tachycardia?

Reply 7:

Thanks for the comment. The NSBB chosen in the study were limited to propranolol (inderal) or carvediolol. Because this is a retrospective study analyzing data from CGRD, confirming the aim of NSBB prescription is used for prophylaxis of esophageal varix rupture is difficult. However, despite beta-adrenergic antagonists were once used as part of the cornerstone of the medical management of hypertension, acute coronary syndrome, and congestive heart failure, nowadays, the 2014 statement from the "American Society of Hypertension and the International Society of Hypertension" and Wiysonge CS et al. (Cochrane Database Syst Rev. 2012;11: CD002003) recommend that beta-blockers not be used as first-line therapy for hypertension, particularly in patients over age 60 years(Weber MA et al. J Hypertens. 2014;32(1):3). In addition,

major cardiac effects caused by beta blockade include the precipitation or worsening of congestive heart failure, and significant negative chronotropy. Furthermore, the reduction in cardiac output in patients with cirrhosis receiving propranolol might pose a detrimental effect to the heart especially during stress such as infection (Wong F et al. Hepatology. 2010;52:811–813). Patients with cirrhosis are also associated with hypotension (SÁNDOR BÁTKAI et al. Nat Med 2001 Jul;7(7):827-32) thus beta-blocker for hypertension are less often used. On the contrary, the use of non-selective beta-blocker therapy in the secondary prevention of variceal hemorrhage was first introduced in 1981[2]. Then the application for primary prevention of variceal hemorrhage in patients with known cirrhosis and large esophageal varices was expanded in subsequent studies[3]. Therefore, we speculate that the NSBB used in cirrhosis were mostly used for primary prevention or secondary prevention of variceal hemorrhage. However, we will mention this in the limitation section.

Change in the text:

We have modified our text as advised (please see the fifth limitation part on page 27, line 2~12).

Comment 8.

In Table 3, 11th raw, it seems that the authors calculated HR of HBV, HCV and Alcohol using Others as control. But, the definition and ingredients of Others are quite obscure and not suitable to control.

Reply 8.

Thanks for the comment. In our study, "Others" was mainly due to non-alcoholic steatohepatitis (NASH). Other etiologies included primary biliary cirrhosis, primary sclerosing cholangitis, Wilson's disease, autoimmune hepatitis or etiology unknown. They are not used as control but simply used as a reference group in the Cox regression (Suppose their HR=1.000 for reference). Even when we changed different etiologies for reference group, the results did not change.

Change in the text: We have modified Table 3.

Minor points:

1. In Table 3, the 5th raw, 'Male 1', should be deleted because gender is divalent.

Reply

Thanks for the comment. As the revised Table 3 shown above, the 1.00 is a HR and the male is used as a reference group for comparison.

Change in the text: We have modified Table 3.

1. Garcia-Tsao, G., et al., *Portal hypertensive bleeding in cirrhosis: Risk stratification, diagnosis, and management: 2016 practice guidance by the American Association for the study of liver diseases.* Hepatology, 2017. **65**(1): p. 310-335.
2. Lebrec, D., et al., *Propranolol for prevention of recurrent gastrointestinal bleeding in patients with cirrhosis: a controlled study.* NEJM,1981. **305**(23): p. 1371-1374.
3. Pascal, J.-P., P. Cales, and A.M.S.G.J.N.E.J.o. Medicine, *Propranolol in the prevention of first upper gastrointestinal tract hemorrhage in patients with cirrhosis of the liver and esophageal varices.* NEJM, 1987. **317**(14): p. 856-861.