

# New prognostic score based on galectin-3 has similar performance to model for end-stage liver disease and sodium score in patients with stable decompensated cirrhosis

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## Abstract

**Background** Galectin-3 (gal-3) has been proposed as a marker of established renal impairment, with predictive value in stable decompensated cirrhosis.

**Methods** 150 stable decompensated patients were assessed in 2 transplant centers. Patients' renal function was assessed using <sup>51</sup>Chromium-EDTA ("true" glomerular filtration rate). We measured basic laboratory variables and gal-3 in serum samples. Factors associated with patients' outcomes were determined.

**Results** Our patients were followed up for 12 months (range 1-48, interquartile range [IQR] 6, 95% confidence interval [CI] 10-13.5) and their mean prognostic scores were Child-Turcotte-Pugh (CTP) 7±2 and model for end-stage liver disease and sodium (MELD-Na) 15±6. Median gal-3 levels were 22 ng/mL. In a multivariate analysis of 94 patients (training group), gal-3 (hazard ratio [HR] 1.026, 95% confidence interval [CI] 1.011-1.041; P=0.003) and serum sodium (HR 1.032, 95%CI 1.006-1.062; P=0.05) were the only factors independently associated with patients' outcomes. Kaplan-Meier analysis using the median gal-3 values revealed different times of survival (log-rank P=0.006). We derived a new prognostic score, (0.026) × serum gal-3+ (-0.079) × serum sodium, with very good discriminative accuracy for the outcome (area under the curve [AUC] 0.71, 95%CI 0.63-0.88), similar to that of the MELD-Na score (AUC 0.69, 95%CI 0.67-0.89; P=0.73), while its diagnostic accuracy was validated in the remaining 56 decompensated patients (AUC 0.81, 95%CI 0.65-0.97).

**Conclusions** Gal-3 proved to be an accurate and plausible biomarker of renal dysfunction in patients with decompensated cirrhosis. A new prognostic model incorporating gal-3 and sodium was derived, with very good discriminative accuracy for the outcome.

**Keywords** Decompensated cirrhosis, galectin-3, renal function, liver cirrhosis, prognostic scores

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## Introduction

Cirrhosis is a major cause of morbidity and mortality and prognostic scores are widely used to predict survival and the need for transplantation [1]. Prognostic scores represent a quantitative estimation of the "reserve" in terms of liver function. Therefore, they could define important issues related to life expectancy and therapeutic options [2]. The model for end-stage liver disease (MELD) score is a useful prognostic tool for end-stage liver disease that takes into account renal function, showing its importance in cirrhosis [3]. Thus, accurate assessment of renal function has become more important for therapeutic decision-making and for evaluating prognosis. As serum creatinine is influenced by several extrarenal factors, measurement of the "true" glomerular filtration rate (GFR) proved to be a robust and significant prognostic factor in patients with decompensated cirrhosis [4]. However, there are

limitations to the routine use of “true” GFR, because a nuclear medicine laboratory is necessary and it is more expensive [4,5].

Alternatively, we have suggested in a previous study that galectin-3 (gal-3), a member of the lectin family, could be a trustworthy marker of established chronic kidney disease, with predictive ability in stable decompensated cirrhosis [6]. Gal-3 is known to play a critical role in the development of several chronic conditions, including cancer, fibrosis, and chronic inflammation [7]. As for the liver, gal-3 proved to be a good marker of fibrosis in cirrhosis [8], reflecting implications in profibrotic pathways [9]. Moreover, high gal-3 expression has been associated with renal fibrosis and impaired renal function [10]. Thus, it seems reasonable to define gal-3 as a new diagnostic and prognostic biomarker in hepatic diseases [7].

In general, several factors have been evaluated for their prognostic significance in a very large number of studies, indicating there is uncertainty about the available prognostic models in decompensated cirrhosis [11]. Among these, serum sodium (Na) concentration was an important predictor of survival among candidates for liver transplantation (LT) [12]. From this point of view, researchers introduced prognostic scores that incorporate sodium, such as the MELD-Na score, a good predictor of waiting list survival, and this score is used now for stratification of LT candidates on the waiting list [13].

Our study aimed to evaluate the prognostic role of gal-3 as a marker of renal dysfunction in decompensated cirrhosis and to confirm our previous findings. Moreover, we intended to investigate all important factors associated with our patients' outcomes and, if possible, to develop a new prognostic score.

## Patients and methods

We evaluated a cohort of consecutive adult patients with decompensated cirrhosis admitted for pre-LT assessment in 2 transplant centers (“Hippokraton” General Hospital of Thessaloniki and “Laiko” General Hospital of Athens) between 2015 and 2019. Our patients were studied during a follow-up period, until a significant clinical outcome was recorded (death or LT), or until the end of the study. Decompensated cirrhosis was defined as a history of ascites, variceal bleeding or encephalopathy in patients with cirrhosis. We excluded patients who underwent LT before their admission. Patients were stable regarding their chronic liver disease, i.e., they had no active variceal bleeding, encephalopathy or infection, such as spontaneous bacterial peritonitis (SBP), during the last month before their admission. Detailed clinical evaluation, laboratory measurements (white blood cells, C-reactive protein, procalcitonin, blood cultures and ascitic fluid paracentesis) and radiological exams (chest X-ray, upper abdominal ultrasound) whenever necessary, were performed in order to exclude patients with clinical or subclinical infection. Since all were LT candidates, patients older than 67 years were also excluded.

On admission, several demographic and clinical characteristics were prospectively recorded for each patient, such as age, sex, cause of cirrhosis, previous complications of cirrhosis (i.e., variceal bleeding, encephalopathy or SBP),

medication administered for the liver disease (duration and dosage), vital signs (blood pressure, pulse rate) and concomitant extra-hepatic diseases (e.g., diabetes mellitus, coronary artery disease). In addition, the following laboratory variables were evaluated: hematocrit, white blood count, platelet count, creatinine, urea, electrolytes (sodium, potassium, magnesium, calcium, phosphate), aspartate and alanine aminotransferases, alkaline phosphatase,  $\gamma$ -glutamyl transpeptidase, bilirubin (total and direct), protein, albumin, lactate dehydrogenase, as well as clotting profile (prothrombin time, international normalized ratio [INR], activated partial thromboplastin time), ferritin and lipidemic profile. We evaluated the severity of liver disease and the prognosis of our patients by calculating the Child-Turcotte-Pugh (CTP) [14] and MELD-Na [15] scores. Presence of hepatocellular carcinoma (HCC) was assessed. Finally, we measured levels of gal-3 in serum samples using the Abbott Architect i1000SR Analyzer, which applies a method of chemiluminescent microparticle immunoassay to determine gal-3 values (upper limit of normal, based on the biochemical laboratory: 11.7 ng/mL). According to the manufacturer, reagents and analyzers for gal-3 estimation are available worldwide, in Europe and USA with FDA approval, with a low cost, about €10 per measurement. According to our protocol, patients underwent further assessment of their renal function before being entered in the LT list. In particular, their “true” GFR was assessed using  $^{51}\text{Chromium-EDTA}$  ( $^{51}\text{Cr-EDTA}$ ) [16], together with estimated GFR (eGFR) using the creatinine-based 4-variable Modification of the Diet in Renal Disease (MDRD) formula [17], to assess presence of chronic kidney disease [18].

Only patients with full demographic and laboratory data were included in the study. The study protocol was approved by our Institutional Review Board and conformed to the ethical guidelines of the 2013 Declaration of Helsinki.

## Statistical analysis

Continuous variables with normal distribution were presented as mean $\pm$ standard deviation, or as median with interquartile range (IQR) if non-normally distributed, and comparisons of means was performed using Student's *t* or Mann-Whitney *U* tests, as appropriate. Categorical variables were expressed as frequencies or percentages and the chi-square test was used for comparisons. Logistic regression analysis was carried out to identify factors associated with the outcome (survival, death or LT). Variables found significant in the univariate analysis were included in the multivariate model. The multivariate analysis used a Cox proportional hazard model with backward elimination of variables according to likelihood ratio criteria, starting with all variables with  $P < 0.05$  in univariate models in order to derive a new prognostic score for patients' outcomes. Collinearity was assessed with the tolerance and the variance inflation factor (VIF). The discriminatory ability of the prognostic score to predict the outcome (survival vs. death or LT) of patients with decompensated cirrhosis was evaluated using the area (AUC)

under the receiver operating characteristic curve (ROC). This has the true-positive and false-positive rates on the vertical and horizontal axes, respectively. As the AUC approaches 1.0, the model approaches 100% sensitivity and specificity [19]. A P-value <0.05 was considered statistically significant. To test the calibration (i.e., the degree of correspondence between predicted and observed mortality), a simplified goodness-of-fit (GOF) method for the Cox proportional hazards model proposed by May and Hosmer was used. The patients' survival according to gal-3 levels was calculated using Kaplan-Meier analysis and compared with the log-rank sum test. Statistical analysis was conducted using SPSS (IBM SPSS version 26.0 for Windows, Armonk, NY, USA) and MedCalc for Windows (MedCalc Software, Mariakerke, Belgium) software. Patients included had full demographic and laboratory data available. Only few missing values were handled by the SPSS analysis, which estimated summary statistics and imputed missing values using statistical algorithms.

### Validation of the new prognostic score

The derived prognostic score was validated in 56 consecutive adult patients with decompensated cirrhosis admitted for LT assessment during 2017-2019, using the same exclusion and inclusion criteria. Comparisons between the training and validation sample were performed using Student's *t* or Mann-Whitney *U* tests, as appropriate, for continuous variables, and chi square-tests for categorical variables.

### Results

One hundred fifty patients (100 male, age 53±11 years) were included in the study. There were 90 patients from Thessaloniki transplant center and 60 patients from Athens. Viral hepatitis was the cause of cirrhosis in 51 patients (34%). On admission, CTP and MELD-Na scores were 7±2 and 15±6, respectively. Mean values of "true" GFR were 82±28 mL/min/1.73 m<sup>2</sup>. Mean eGFR based on the MDRD formula was 89±27 mL/min. The remaining patients' characteristics are presented in Table 1. The median follow-up period for our cohort was 12 months (range 1-48, IQR 6, 95% confidence interval [CI] 10-13.5). Thirty-one patients (21%) had gal-3 levels within the normal range, while 119 (79%) had values higher than 11.7 ng/mL. Median serum gal-3 levels were 22 ng/mL (range 4.9-96.5, IQR 18.3), showing no difference between men and women; 74 patients (49%) had gal-3 levels lower than the median value and 76 (51%) had values ≥22 ng/mL. Interestingly, patients with gal-3 <22 ng/mL, compared to those with gal-3 ≥22 ng/mL, had significantly lower MELD-Na (13±5 vs. 16±6, P<0.001) and CTP (7±2 vs. 9±3, P=0.03) scores. In addition, patients with MELD-Na scores <15, compared to those with ≥15, had significantly lower gal-3 levels (21±9 vs. 30±12 ng/mL, P<0.001). Similarly, patients with a CTP score <8, compared to those with ≥8, had significantly lower gal-3 levels (23±10 vs. 28±12 ng/mL, P=0.04).

**Table 1** Baseline clinical and laboratory characteristics of 150 patients with stable decompensated cirrhosis

Variable	Patients, n=150
Age (mean±SD, years)	53±11
Sex, male n, (%)	100 (67)
Etiology of cirrhosis, n, (%)	
Viral hepatitis	51 (34)
Alcohol	36 (24)
NASH	22 (15)
Other	41 (27)
Hepatocellular carcinoma, n (%)	20 (13)
History of complications, n, (%)	
GI bleeding	38 (25)
Encephalopathy	42 (28)
SBP	15 (10)
Total bilirubin (median, range, IQR, mg/dL)	1.98 (0.24-40.5, 2.5)
Albumin (median, range, g/dL)	3.6 (1.9-6.5)
Creatinine (median, range, IQR, mg/dL)	0.87 (0.49-3.02, 0.3)
"true" GFR by <sup>51</sup> Chromium-EDTA (mean±SD, mL/min)	82±28
MDRD-estimated GFR (mean±SD, mL/min)	89±27
Galectin-3 (median, range, IQR, ng/mL)	22 (4.9-96.5, 18.3)
Heart rate (/min, mean±SD)	75±17
CTP score (mean±SD)	7±2
MELD-Na score, (mean±SD)	15±6

NASH, nonalcoholic steatohepatitis; GI, gastrointestinal; SBP, spontaneous bacterial peritonitis; GFR, glomerular filtration rate; CTP, Child-Turcotte-Pugh score; MELD, model for end-stage liver disease; Na, serum sodium; MDRD, modification of the diet in renal disease formula; IQR, interquartile range; SD, standard deviation

### Patients' features associated with their renal function (univariate and multivariate analysis; Table 2)

We divided our patients into 2 groups based on their relatively preserved renal function: those with "true" GFR ≥60 mL/min (n=111, 74%) and the remaining 39 patients with "true" GFR <60 mL/min (26%). The former group of patients were younger (51±11 vs. 59±9 years, P=0.001) and had significantly lower gal-3 levels, serum creatinine levels, CTP scores and MELD-Na scores than the latter group of patients: 19 (4.9-66, IQR 13) vs. 33 (7.1-96.5, IQR 21.6) ng/mL, P<0.001; 0.8 (0.5-1.3, IQR 0.2) vs. 1.1 (0.6-3, IQR 0.5) mg/dL, P<0.001; 7 (5-12, IQR 3) vs. 8 (6-13, IQR 2), P=0.048; and 13 (6-26, IQR 7) vs. 15 (8-32, IQR 8), P=0.04; respectively. On multivariate analysis, including all significant factors from the univariate analysis, the only factors independently associated with renal function were gal-3 (odds ratio [OR] 0.96, 95%CI 0.93-0.98; P=0.009) and age (OR 0.92, 95%CI 0.86-0.97; P=0.007). Accordingly, we examined the ability of gal-3 and age to discriminate patients based on their preserved renal function. ROC analysis showed that they had good performance in predicting renal impairment, i.e., "true" GFR <60 mL/min: for gal-3 AUC 0.77, 95%CI 0.67-0.86, best cutoff value 21 ng/mL, sensitivity 82%, specificity 41%,

**Table 2** Clinical and laboratory characteristics of 150 patients, including those with “true” GFR  $\geq 60$  mL/min and  $< 60$  mL/min

Variables	Patients with “true” GFR $\geq 60$ mL/min (n=111, 74%)	Patients with “true” GFR $< 60$ mL/min (n=39, 26%)	P-value
Age (mean $\pm$ SD, years)	51 $\pm$ 11	59 $\pm$ 9	0.001
Sex, male n, (%)	80 (72)	20 (51)	0.018
Total bilirubin (median, range, IQR, mg/dL)	2.1 (0.39-33, 2.2)	1.9 (0.24-40.5, 3.4)	0.6
Albumin (median, range, IQR, g/dL)	3.6 (2.2-6, 1)	3.4 (1.9-5.1, 1)	0.15
Etiology of cirrhosis, n, (%)			0.84
Viral hepatitis	41 (37)	10 (25)	
Alcohol	25 (23)	11 (28)	
NASH	16 (14)	6 (15)	
History of complications, n, (%)			0.45
GI bleeding	31 (28)	7 (18)	
Encephalopathy	33 (29)	9 (24)	
SBP	10 (9)	5 (13)	
Creatinine (median, range, IQR, mg/dL)	0.8 (0.5-1.3, 0.2)	1.1 (0.6-3, 0.5)	$< 0.001$
MDRD-estimated GFR (mean $\pm$ SD, mL/min)	99 $\pm$ 22	59 $\pm$ 17	$< 0.001$
Galectin-3 (median, range, IQR, ng/mL)	19 (4.9-66, 13)	33 (7.1-96.5, 21.6)	$< 0.001$
CTP score (median, range, IQR)	7 (5-12, 3)	8 (6-13, 2)	0.048
MELD-Na score (median, range, IQR)	13 (6-26, 7)	15 (8-32, 8)	0.04

NASH, nonalcoholic steatohepatitis; GI, gastrointestinal; SBP, spontaneous bacterial peritonitis; GFR, glomerular filtration rate; CTP, Child-Turcotte-Pugh score; MELD, model for end-stage liver disease; Na, serum sodium; MDRD, modification of the diet in renal disease formula; IQR, interquartile range; SD, standard deviation

positive predictive value (PPV) 52%, negative predictive value (NPV) 88%; for age AUC 0.67, 95%CI 0.57-0.77, best cutoff value 51 years, sensitivity 82%, specificity 40%, PPV 41%, NPV 86%. The results were similar when HCC patients (n=20) were excluded from the analysis.

### Training group: factors associated with mortality in multivariate Cox proportional hazard regression analysis

In the training group (n=94), as in the total cohort, patients with gal-3 levels  $< 22$  ng/mL (group 1) compared to those with gal-3  $\geq 22$  ng/mL (group 2) had significantly lower MELD-Na scores (13 $\pm$ 4 vs. 16 $\pm$ 6, P=0.015), while those patients with MELD-Na scored  $< 15$  compared to those with MELD-Na  $\geq 15$  had significantly lower gal-3 levels (21 $\pm$ 10 vs. 31 $\pm$ 13 ng/mL, P $< 0.001$ ). We conducted Cox regression analysis to detect factors associated with our patients' outcomes. Gal-3 proved to be significant risk factor associated with our patients' negative outcomes (hazard ratio [HR] 1.028, 95%CI 1.014-1.042; P $< 0.001$ ). Total bilirubin (HR 1.077, 95%CI 1.015-1.14; P=0.013), serum sodium (HR 0.88, 95%CI 0.79-0.97; P=0.016) and MELD-Na score (HR 1.13, 95%CI 1.048-1.23; P=0.001) were also significant risk factors (Table 3). However, in the multivariate analysis, the only factors independently associated with our patients' outcomes were gal-3 (HR 1.026, 95%CI 1.011-1.041; P=0.003) and serum sodium (HR 1.032, 95%CI 1.006-1.062; P=0.05). Kaplan-Meier analysis, using the median gal-3 value (22 ng/mL), revealed different times

of survival for our patients (median survival: 48 months, 95%CI 19-76 vs. 10 months, 95%CI 5-25; log-rank P=0.006) (Fig. 1A). In addition, when HCC patients (n=20) were excluded, Kaplan-Meier analysis showed that patients with gal-3 higher than 22 ng/mL compared to those with less than 22 ng/mL had significantly worse outcomes (log-rank P=0.001) (Fig. 1B). There was no collinearity between the 2 independent variables, gal-3 and serum bilirubin, as assessed by tolerance and VIF, with values near 1 for both factors (tolerance 0.96; VIF 1.05).

Based on the coefficients of these 2 independent variables, a new prognostic model was derived:

new prognostic score = (0.026)  $\times$  serum gal-3 + (-0.079)  $\times$  serum sodium.

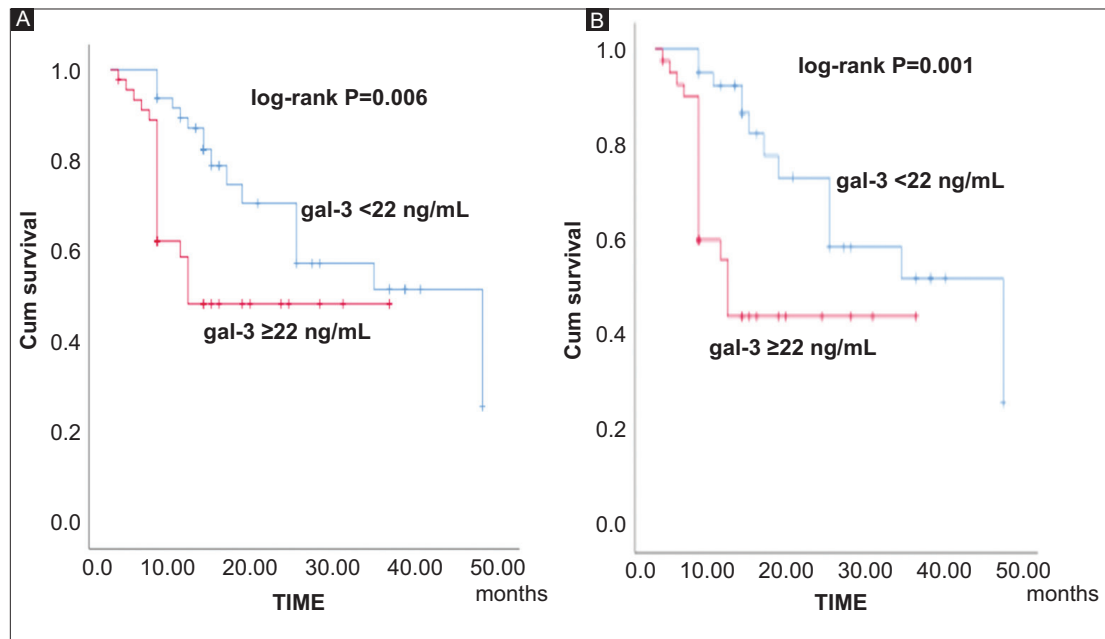
Based on the AUC, the new prognostic score had very good discriminative accuracy for the outcome (AUC 0.71, 95%CI 0.63-0.88), whilst MELD-Na score had similar performance (AUC 0.69, 95%CI 0.67-0.89; P=0.73) (Fig. 2A). Table 4 shows the sensitivity, specificity, PPV, NPV and diagnostic accuracy at the cutoff point giving the best Youden index for each scoring system.

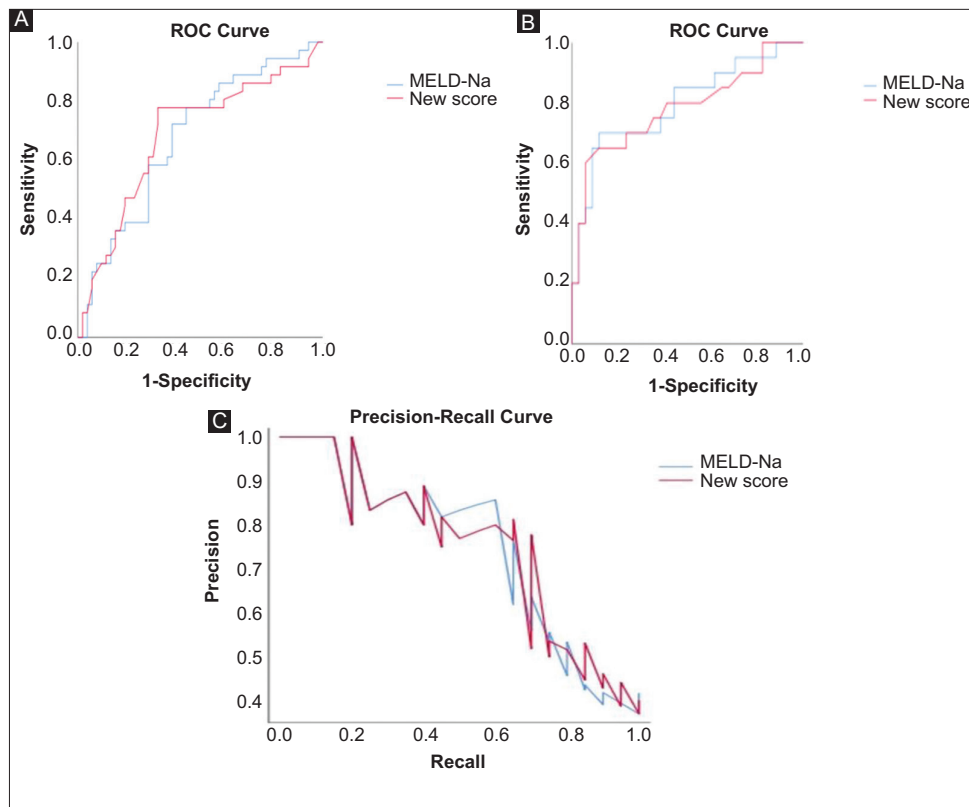
The best cutoff point for the new prognostic score was -9.07, giving 86% sensitivity, 37% specificity, 48% PPV and 87% NPV (for MELD-Na score the best cutoff point was 12, giving 77% sensitivity, 55% specificity, 57% PPV and 78% NPV). When we excluded patients who underwent LT, the performance of both new prognostic score and MELD-Na score was improved (AUC 0.84, 95%CI 0.71-0.96; AUC 0.86, 95%CI 0.76-0.97, respectively).

**Table 3** Clinical and laboratory characteristics of 94 patients (training group) with stable decompensated cirrhosis associated with the outcome (univariate analysis)

Variables	Hazard ratio	P-value	95% Confidence Interval	
			Lower	Upper
Age (mean±SD, years)	0.99	0.78	0.97	1.03
Sex, male n, (%)	0.87	0.72	0.42	1.8
Total bilirubin (median, range, mg/dL)	1.077	0.013	1.015	1.14
Albumin (median, range, g/dL)	0.8	0.72	0.51	1.2
Etiology of cirrhosis, n, (%)				
Viral hepatitis	1.20	0.55	0.80	1.45
Alcohol	1.22	0.41	0.75	2.13
NASH	0.92	0.37	0.65	1.42
History of complications, n, (%)				
GI bleeding	0.45	0.47	0.09	1.24
Encephalopathy	1.10	0.66	0.62	1.89
SBP	0.33	0.28	0.07	2.11
Creatinine (median, range, mg/dL)	1.22	0.64	0.52	2.8
“true” GFR by <sup>51</sup> Chromium-EDTA (mean±SD, mL/min)	0.99	0.69	0.98	1.016
MDRD-estimated GFR (mean±SD, mL/min)	0.98	0.36	0.97	1.02
Galectin-3 (median, range, ng/mL)	1.028	<0.001	1.014	1.042
Na (mean±SD, mmol/L)	0.88	0.016	0.79	0.97
CTP score (median, range)	1.06	0.56	0.88	1.24
MELD-Na score, (mean±SD)	1.13	0.001	1.048	1.23

NASH, nonalcoholic steatohepatitis; GI, gastrointestinal; SBP, spontaneous bacterial peritonitis; GFR, glomerular filtration rate; CTP, Child-Turcotte-Pugh score; MELD, model for end-stage liver disease; Na, serum sodium; MDRD, modification of the diet in renal disease formula; SD, standard deviation

**Figure 1** Kaplan-Meier curves showing difference in survival among decompensated patients based on median galectin-3 (gal-3) levels in the training group: (A) whole cohort, (B) excluding hepatocellular carcinoma patients



**Figure 2** Receiver operating characteristic (ROC) analysis. (a) ROC curve showing the discriminatory ability of the herein introduced prognostic score to predict decompensated patients' outcome (area under the curve (AUC) 0.71, 95% confidence interval [CI] 0.63-0.88) (model for end-stage liver disease [MELD] and sodium [Na]: AUC 0.69, 95%CI 0.67-0.89). (b) ROC analysis in the validation group showing the discriminative accuracy for mortality of the new prognostic score (AUC 0.81, 95%CI 0.65-0.97) (MELD-Na: AUC 0.81, 95%CI 0.68-0.95). (c) Precision-recall curves in the validation group of the new prognostic score and MELD-Na score (AUC 0.79 and 0.78, respectively)

**Table 4** Prediction of outcome by the new prognostic score in 94 patients with stable decompensated cirrhosis (training group)

	Cutoff point	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
New prognostic score	-9.07	86	37	48	87
MELD-Na score	12	77	55	57	78

PPV, positive predictive value; NPV, negative predictive value; MELD, model for end-stage liver disease; Na, serum sodium

### Validation of the new prognostic score

The validation cohort (n=56 patients) had similar follow up, as well as clinical and laboratory characteristics (including "true" GFR, gal-3, serum creatinine, bilirubin, INR and serum sodium) compared to patients in the training group. At the end of follow up (median time 6 months, range 1-36), 36 patients (72%) were still alive on the waiting list, whilst 14 (28%) patients had died (5 of sepsis, 1 of variceal bleeding, 4 of liver failure, and 4 of multi-organ failure) and 6 underwent LT. Based on ROC analysis, the new prognostic score confirmed its discriminative accuracy for mortality in the validation

group (AUC 0.81, 95%CI 0.65-0.97), similar to that of the MELD-Na score (AUC 0.81, 95%CI 0.68-0.95) (Fig. 2B), confirmed using Precision-recall curves (AUC 0.79 vs. 0.78) (Fig. 2C). Regarding the GOF, measured by the GOF method, the calibration of the new prognostic score in the validation group ( $\chi^2 = 4.2$ ,  $P=0.66$ ) was superior to the MELD-Na score ( $\chi^2 = 3.9$ ,  $P=0.52$ ), as well as based on Akaike information criterion (45.2 vs. 87.6).

### Discussion

To the best of our knowledge, this is the first study in which the discriminative ability of a new prognostic score based on gal-3 was evaluated in patients with stable decompensated cirrhosis. In our cohort, we found that gal-3 and serum sodium were the only independent factors associated with the outcome, while a new mathematical formula we derived, based on these 2 variables, had a very good performance, similar to that of the well-established MELD-Na score. Gal-3 is considered an important regulatory factor in profibrotic pathways and it has already been suggested as a noninvasive serum marker that reflects the severity of fibrosis in patients with chronic hepatitis C and nonalcoholic

steatohepatitis [9,20]. In a previous study, which included 100 patients with decompensated cirrhosis, we showed for the first time the significant impact of gal-3 on prognosis and renal function in this group of patients [6].

It is known that renal dysfunction is associated with poor survival in patients with chronic liver failure, as indicated by the inclusion of serum creatinine in the MELD-Na score. In fact, the superiority of the MELD-Na score, compared to the CTP score, is partly associated with the inclusion of serum creatinine in the MELD-Na score computation. However, serum creatinine is not considered an accurate index of renal function, because individuals with the same GFR may have different values of creatinine, due to differences in age, race and sex. In fact, the assessment of GFR would be the optimal method for the accurate evaluation of prognosis in patients with decompensated cirrhosis [4]. We have shown that “true” GFR measurement based on <sup>51</sup>Cr-EDTA was an independent factor for survival in patients with decompensated cirrhosis, while the incorporation of “true” GFR in the MELD-Na score in place of serum creatinine was associated with an improvement in its discriminative accuracy [4,21]. However, it should be mentioned that the methods for “true” GFR evaluation are time-consuming and costly, requiring special laboratory facilities [4]. Since accurate evaluation of renal function in patients with decompensated cirrhosis is crucial, different equations based on serum creatinine have been derived to provide an estimation of GFR (such as the MDRD formula), but these formulas have been reported to overestimate or underestimate “true” GFR in patients with decompensated cirrhosis [5,22]. Thus, new serum markers are needed in order to overcome all these limitations. In the present study, we were able to expand our previous findings regarding the association of gal-3 with “true” GFR, while evaluating 150 patients with stable decompensated cirrhosis. In fact, we showed in this larger cohort that gal-3 was again an independent factor (OR 0.96, 95%CI 0.93-0.98; P=0.009) for the presence of “true” GFR <60 mL/min, with very good discriminative ability (AUC 0.77, 95%CI 0.67-0.86). These new findings confirm the possibility of using gal-3 as an accurate and plausible biomarker of renal dysfunction in patients with decompensated cirrhosis, irrespectively of several baseline characteristics, such as underlying liver disease, HCC, diabetes mellitus, ascites, or administration of diuretics.

Interestingly, we found that gal-3 was also a significant risk factor associated with our patients’ prognosis. In the total cohort, as well as in the training group, patients with gal-3 levels lower than the median value (i.e., 22 ng/mL), compared to those with gal-3 ≥22 ng/mL, had significantly better MELD-Na scores, while patients with MELD-Na score <15, compared to those with MELD-Na ≥15, had significantly lower gal-3 levels. In addition, in the training group (n=94), gal-3 proved to be a significant risk factor associated with our patients’ negative outcomes (HR 1.028, 95%CI 1.014-1.042; P<0.001), while Kaplan-Meier analysis using the median value (i.e., gal-3 22 ng/mL) revealed different times of survival in the total cohort (log-rank P=0.006), a difference that persisted when HCC patients were excluded (log-rank P=0.001). In the multivariate analysis, the only factors

independently associated with our patients’ outcomes were gal-3 and serum sodium (HR per unit 1.026, 95%CI 1.011-1.041; P=0.003) and (HR per unit 1.032, 95%CI 1.006-1.062; P=0.05), respectively. In fact, based on the coefficients of these 2 independent variables (i.e., gal-3 and serum sodium), a new prognostic model was derived, which had very good discriminative accuracy for the outcome in the training group (AUC 0.71, 95%CI 0.63-0.88), similar to that of the MELD-Na score (AUC 0.69, 95%CI 0.67-0.89). The results were similar when the patients who underwent LT in the training group were excluded (AUC 0.84, 95%CI 0.71-0.96; AUC 0.86, 95%CI 0.76-0.97, respectively). Importantly, the performance of this new prognostic score (based on 2 variables, i.e., gal-3 and sodium) was confirmed in the validation cohort, since based on the AUC, its predictive ability for the outcome was very good (AUC 0.81, 95%CI 0.65-0.97), similar to that of the MELD-Na score (based on 4 variables, i.e., serum creatinine, INR, total bilirubin and serum sodium: AUC 0.81, 95%CI 0.68-0.95). It should be mentioned that serum sodium was a common component in both prognostic scores, reflecting the importance of hyponatremia in the outcome of patients with decompensated cirrhosis [23]. On the other hand, gal-3 was able to replace not only serum creatinine, as a marker of renal function, but also INR and bilirubin, achieving in combination with serum sodium similar discriminatory ability to the MELD-Na score regarding outcomes. Thus, it seems that the new prognostic score may represent a simple and attractive alternative for accurate assessment of outcomes in patients with stable decompensated cirrhosis. Interestingly, the new score, compared to the MELD-Na score, had better sensitivity (e.g., in the training cohort 86% vs. 77%, respectively) and NPV (87% vs. 78%, respectively).

We acknowledge our study has limitations (including the absence of a prior power calculation) and further studies are needed to establish the predictive utility of gal-3 regarding cirrhotic patients. Ideally, an independent validation cohort of patients could study our findings. However, we managed to find significant results for this marker and incorporated it in a new prognostic model. Among prognostic scores, the one introduced here is easily available, based only on 2 variables, gal-3 and sodium. Besides, gal-3 has already been used as a therapeutic target, reflecting liver, renal and cardiac dysfunction, and has proven its significance in everyday clinical practice. In addition, we are thinking of calculating this new score as a follow-up variable and evaluating the importance of its variance. This is an interesting topic that needs further study.

In conclusion, our findings confirmed the previously known use of gal-3 as biomarker of renal dysfunction in patients with decompensated cirrhosis. Our study has another strength, as the results were derived from a large cohort from 2 transplant centers in Greece. Furthermore, the newly introduced model, built from 2 prognostic markers, gal-3 and serum sodium, showed very good discriminative accuracy for the outcomes in the training group. This result was similar to the performance of the MELD-Na score and seems promising as an indicator of prognosis in decompensated patients.

## Summary Box

### What is already known:

- Renal dysfunction is associated with poor survival in patients with chronic liver failure
- Galectin-3 (gal-3) has been proposed as a marker of established renal impairment
- Serum sodium concentration is considered an important predictor of survival among candidates for liver transplantation

### What the new findings are:

- It was confirmed that gal-3 is an accurate biomarker of renal dysfunction in patients with decompensated cirrhosis, irrespectively of several baseline characteristics
- Gal-3 was established to be a significant risk factor associated with decompensated patients' prognosis, even when hepatocellular carcinoma patients were excluded
- A new prognostic model was derived that incorporated the prognostic importance of gal-3 and serum sodium and had very good discriminative accuracy for the outcome in stable decompensated cirrhosis

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